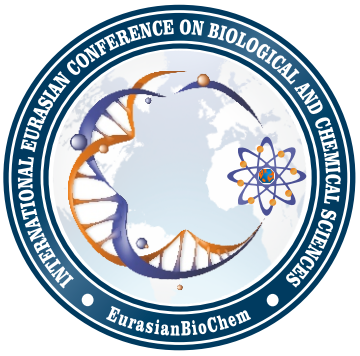


6th INTERNATIONAL EURASIAN CONFERENCE ON

BIOLOGICAL AND CHEMICAL SCIENCES

11 - 13 October 2023
Ankara / Turkey

(EurasianBioChem2023)

www.eurasianbiochem.org

PROCEEDING BOOK

(Abstracts and Full-Text Papers)

EurasianBioChem

ISBN :

1. ORAL PRESENTATIONS

DRAFT

1.1. ABSTRACTS

DRAFT

ORAL PRESENTATION

Doped manganese perovskites for room-temperature magnetic refrigeration applications: Synthesis and characterization

Nisrine El Hamouchi[1*], Sara Ait Bouzid[1], Mohammed Sajieddine[2,3], Victor Kuncser[4], Aurelian Catalin Galca[5], Nicusor Iacob[4], Monica Enculescu[5], Abdellatif Essoumhi[1]

[1]Equipe of Chemical Processes and Applied Materials, Polydisciplinary Faculty, Sultan Moulay Slimane University, B.P 523, 23000, Beni-Mellal, Morocco.
[2]Laboratory of Materials Physics, Faculty of Sciences and Techniques, Sultan Moulay Slimane University, B.P 523, 23000, Beni-Mellal, Morocco.
[3]National School of Applied Sciences, Sultan Moulay Slimane University, Khouribga, Morocco.
[4]Laboratory of Magnetism and Superconductiviy, National Institute of Materials Physics, Atomistilor 405A, 077125, Magurele, Romania
[5]Laboratory of Multifunctional Materials and Structures, National Institute of Materials Physics, Atomistilor 405A, 077125, Magurele, Romania

*Corresponding author e-mail: nisrineelhamouchi@gmail.com

Abstract

This study is primarily focused on investigating how the introduction of dopants influences the properties of a manganese perovskite material intended for use in room-temperature magnetic refrigeration applications. The synthesis of these materials employed the flash combustion technique, a highly regarded method renowned for its swift and precise synthesis process. After the synthesis process, the materials were subjected to analysis through X-ray Diffraction (XRD), Scanning Electron Microscopy (SEM), and magnetic analysis. XRD analysis played a key role in providing crucial information on the crystal structure, phase composition, and lattice parameters of both materials. Our investigation revealed that both compounds exhibited a rhombohedral $R\bar{3}c$ structure. SEM analysis, meanwhile, provided intrinsically detailed information on the structure, composition, and surface morphology of the samples, affirming the high homogeneity of both compounds. Magnetic analysis, using a superconducting quantum interference device (SQUID) magnetometer, facilitated the characterization of the materials' magnetic properties. It will cover the Curie temperatures, the nature of the phase transitions, their order, the variation of the magnetic entropy, and the magnetocaloric properties of the samples examined.

Keywords: Flash combustion, Magnetic refrigeration, Magnetocaloric materials.

DRAFT

ORAL PRESENTATION

Impact of sodium doping on the magnetic characteristics of manganite-based materials

Nisrine El Hamouchi[1*], Sara Ait Bouzid[1], Mohammed Sajieddine[2,3], El Kebir Hlil[4], Mohammed Mansori[5], Abdellatif Essoumhi[1]

[1]Equipe of Chemical Processes and Applied Materials, Polydisciplinary Faculty, Sultan Moulay Slimane University, B.P 523, 23000, Beni-Mellal, Morocco.
[2]Laboratory of Materials Physics, Faculty of Sciences and Techniques, Sultan Moulay Slimane University, B.P 523, 23000, Beni-Mellal, Morocco.
[3]National School of Applied Sciences, Sultan Moulay Slimane University, Khouribga, Morocco.
[4]Institut Néel, Univ. Grenoble Alpes, CNRS, Grenoble INP, 38000, Grenoble, France.
[5]Laboratory of Materials Chemistry and Environment, Faculty of Science and Technology, Cadi Ayyad University, Av. A. Khattabi B.P 549, 40000, Marrakech, Morocco.

*Corresponding author e-mail: nisrineelhamouchi@gmail.com

Abstract

Given their substantial magnetocaloric effect, substituted lanthanum manganites have attracted considerable attention and are considered an essential class of materials for magnetic refrigeration applications. This study investigates the thermal, structural, and magnetic characteristics of nanopowders composed of sodium-doped lanthanum manganite. The nanopowder was carefully prepared by the flash combustion method and then subjected to calcination. To understand its thermal properties, thermogravimetric and differential thermal analyses were carried out. In addition, X-ray diffraction analysis revealed that the nanomaterial exhibited a single-phase structure with no discernible secondary phase, adhering to the rhombohedral structure in the $R\bar{3}c$ space group. To further investigate the ferromagnetic-paramagnetic phase transition and unveil the magnetic entropy variations within the sample, we used SQUID (Superconducting Quantum Interference Device) magnetometry in our study.

Keywords: Magnetocaloric effect, Magnetic refrigeration.

DRAFT

ORAL PRESENTATION

Microbial contamination of *s*kin care product due to its using method

Anna Chałubek[1](https://orcid.org/0009-0004-2749-6832), Natalia Śmietańska[1] (https://orcid.org/0009-0004-2099-0977), Katarzyna Duda-Grychtoł[1] (https://orcid.org/0009-0008-7306-9611)

[1]Silesian College of Medicine in Katowice, Katowice, Poland

*Corresponding author e-mail: annachalubek@onet.pl

Abstract

Cosmetics are preparations that are used extensively in everyday life. During their use as well as their manufacture, contamination with potentially pathogenic microorganisms may occur, which may endanger human life and health. Factors affecting the microbiological purity of cosmetics include excessive heat, number of users of the cosmetic, length of use and type of packaging. Preservatives, which have an antimicrobial effect, have a significant impact on microbiological purity. The aim of the study was to evaluated what differences will be observed if face skin care product will be use by hand or via plastic spatula and what kind of bacteria and/or fungi will be isolated. Baird – Parker Agar, MacConkey Agar, Cetrimide Agar, Sabouraud Dextrose Agar and TSA Agar were used for *Staphylococcus aureus*, bacteria from *Enterobacteriaceae* family, *Pseudomonas aeruginosa*, *Candida albicans* and other mesophilic, aerobic bacteria isolation. The cream was applied twice a day for 4 weeks. Samples of skin care product were analysis three times: at the beginning, immediately after opening the products, during using (2 weeks after opening) and last time, after 4 weeks. Results showed that despite of different methods of using skin care product there was no significant differences in quality and quantity of microorganisms.

Key words: microorganisms, skin care products, contamination

DRAFT

ORAL PRESENTATION

In-silico predictions of novel isoxazole-piperazine hybrids as anticancer agents inhibiting liver cancer stem cells

Youness Moukhliss[1]*,Yassine Koubi[1], Marwa Alaqarbeh[2], Hamid Maghat[1], Abdelouahid Sbai[1], Mohammed Bouachrine[1,3], Tahar Lakhlifi[1]

[*1] University of Moulay Ismail, Faculty of Science, Department of Chemistry, Molecular Chemistry and Natural Substances Laboratory (MCNSL), Meknes, Morocco.
[2] The Hashemite University, Faculty of Science, Department of Chemistry, Zarqa, Jordan.
[3] Sultan Moulay Slimane University, EST Khenifra, Beni Mellal, Morocco

*Corresponding author e-mail: y.moukhliss@edu.umi.ac.ma

Abstract

This current research based on the computational examination of novel isoxazole derivatives to investigate a new anti-cancer agent. The study began with the construction of 3D-QSAR models, which resulted in 6 candidate agents that were more active than the marketed anti-cancer agent Sorafenib (SOR), which was chosen as a reference. Evaluation of the synthetic accessibility coefficient and Lipinski properties of 6 newly designed drug candidates indicate that only three of these compounds (P1-P3) meet the criteria. This drug behavior was related to the Lipinski and Veber rules as they can be synthesized. In-silico evaluation of ADMETox properties of these three compounds, shows satisfactory results. According to Molecular Docking, the drug candidate P2 is the most stable in the target receptors (PDB code: 1E31, 1QX3, and 2AR9) comparable to Sorafenib. The Molecular Dynamics study allowed us to confirm the previous results obtained by Molecular Docking. A computational analysis using the CaverDock software allows us to study the release of the drug candidate Pr2 and Sorafenib, through the different tunnels present in the receptors (PDB ID: 1E31, 1QX3, and 2AR9), as well as the corresponding energies during transport.

Keywords: 3D-QSAR, ADMETox, Molecular Docking, Molecular Dynamics, Metadynamics.

DRAFT

ORAL PRESENTATION

Study of environmental state and microplastics as potential pollutants in Albanian coast

Vanela Prifti[1*],Sonila Kane[2],Pranvera Lazo[2]

[1] National Environmental Agency, Sector of Sampling and Laboratory Analyses, Tirana, Albania.
[2] University of Tirana, Faculty of Natural Sciences, Department of Chemistry, Tirana, Albania.

*Corresponding Author: Vanela.Gjeci@akm.gov.al

Abstract

This research presents the monitoring data of seawater parameters linked to the seawater quality of Vlora Bay, Albania. Vlora Bay is the most attractive and frequented tourist area in the south of Albania. The chemical status of the coastal area has been determined in accordance with the Water Framework Directive. Three monitoring sites positioned at a distance of 10 m from the coastal line, one site positioned at the delta of the Vjosa River estuary discharging site, and a control site positioned 100 m away from the coastal line have been selected to assess the water quality of the Vlora Bay estuary. Seawater samples were collected in December 2022. Seawater quality is assessed based on physicochemical parameters such as water temperature, pH, alkalinity, salinity, total suspended solids, electric conductivity, dissolved oxygen, COD, and BOD_5, and nutrient content such as nitrite, nitrate, ammonia, total phosphorous, and orthophosphate. The observed data were compared with the Albanian Standard DCM 177, dated March 31, 2005, "The Permitted Values of Liquid Discharges and Zoning Criteria of the Water Receiving Environment." It revealed that the measured parameters did not go above the allowed limits. The water quality classification for the Vjosa estuary is done according to Water Framework Directive (WFD) 2000/60/EC of the European Parliament and the Council of October 23, 2000. The monitoring data, compared to the requirements of WFD for the nutrients, classifies the Vjosa estuary sample as in a good status (Class I).

Keywords: Seawater; physical-chemical parameters; nutrients content; chemical status; Water Framework Directive, Vlora Bay

DRAFT

ORAL PRESENTATION

Knowledge of Monkeypox virus and disease at the University of Tlemcen

Ilyes Zatla[1,*](ORCID: https://orcid.org/0000-0003-0476-8765), Lamia Boublenza[1] & Wafa Abid[1]

[1] Laboratory of Microbiology applied to the Food industry, Biomedical and the Environment, Faculty of Natural and Life Sciences, Earth and Universe Sciences. Department of Biology. University of Tlemcen, Algeria.

*Author for correspondence, E-mail: ilyes.zatla@aol.com, Tel: +213-540-315-422, Address: Les Oliviers, Kiffane, Tlemcen.

Abstract

A new emerging virus named Monkeypox Virus (MPXV) reappeared and captured all of the attention, while spreading panic among people by threatening their health, expecting at any moment a COVID-19 scenario, if not worse. Our study aims to evaluate knowledge about this virus and its disease and raise consciousness among the Faculty of Natural and Life Sciences and Earth and Universe Sciences at the University of Tlemcen. We launched an online web-based survey for a twenty days period that contained sociodemographic and perceptiveness questions about the emergent virus, its disease, and vaccination, and that was later on analyzed using SPSS and Excel. Results showed that most of our study's respondents were students that belonged to the Department of Biology, and the participants have proven their satisfactory level of knowledge about this emerging virus and its disease. Moreover, most participants showed a positive attitude towards the vaccine, considering it the best preventive means to fight against MPX disease. Although the MPXV may not become a pandemic, knowing the various ways that contribute to its spread is essential to avoid any possibility of a new outbreak, especially in Algeria.

Keywords: Emerging viruses, Monkeypox virus, Monkeypox disease, Survey, University of Tlemcen.

DRAFT

ORAL PRESENTATION

The excess molar volume of liquid mixtures 1,2-dichloroethane with ethers experimental study and application of flory's theory

BOUSSAID Linda[1*], BENSIRADJ Nour El Houda[2] , ZOUAGHI Nafila[3]

[*1]Crystallography-Thermodynamics laboratory. Faculty de chemistry. University of Science and Technology Houari Boumedienne .USTHB
[2]Laboratory of computationel Theoretical chemistry and photonics. University of Science and Technology Houari, Boumediene. USTHB
[3]Laboratory for the study and development of traetment and purification techniques water and environmental Management. Higher Normal School, Kouba.

[*]E-mail : linda.boussaid@hotmail.fr

Abstract

Knowledge of the physico-chemical properties of liquid materials in the industrial field, in general, and in that of the chemical industries, in particular, constitutes a prerequisite for the design of equipment, the resolution of specific problems (related to the techniques of purification and separation, risks in the transport of certain materials, etc.) and, consequently, at the production stage.
Chloroalkanes, ethers constitute chemical families having an industrial, theoretical and environmental interest. For example, these compounds are involved in various applications in the chemical and pharmaceutical industries. In addition, they contribute to the particular thermodynamic behavior (deviation from ideality, association, etc.) of certain mixtures which constitute a severe test for predictive theoretical models. Finally, due to the degradation of the environment in the world, a renewed interest is observed for ethers, because some of their physico-chemical properties could contribute to reducing pollution (ethers would be used as additives in fuels).
This work is a thermodynamic, experimental and theoretical study of the volumetric properties of liquid binary systems formed of compounds belonging to the chemical families of chloroalkanes, ethers, having an industrial, theoretical and environmental interest.
The experimental determination of the densities and excess volumes of the systems studied, at different temperatures located in the interval [278.15-333.15] K and at atmospheric pressure, using an Anton-Paar type DMA5000 vibrating tube densimeter. This contribution of experimental data, on the volumetric properties of liquid binary mixtures of 1,2-dichloroethane with an ether, supplemented by an application of the theoretical model of Prigogine-Flory-Patterson PFP, will probably contribute to the enrichment of the database. thermodynamics and the further development of Flory's theory in its Prigogine-Flory-Patterson (PFP) version, with a view to a better understanding of the thermodynamic behavior of these liquid binary mixtures.

Keywords: Volumetric Properties, Ethers, 1,2-Dichloroethane, Densimetry, Flory Patterson Theory

DRAFT

ORAL PRESENTATION

Analyzing kinetics and adsorption isotherms of anionic dye by a low coast material

Chergui Yamina[1] (https://orcid.org/0000-0003-3036-6452), Ouazani Fouzia [2], Iddou Abdelkader[3]

[1]Laboratoire des Ressources Naturelles Sahariennes. Faculté des sciences et de la technologie, Université Ahmed Draia – Adrar, Algeria
[2] Laboratory of Process Engineering, Materials and Environment, Faculty of Technology, University of Djillali Liabes, PO Box 89, Sidi Bel Abbes 22000 -Algeria.
[3] Higher School of Saharan Agriculture – Adrar, Algeria

chergui.amina@yahoo.fr

Abstract

The pollution of water by organic matter such as dyes requires the use of very efficient processes. Among the solutions proposed, adsorption techniques on less expensive adsorbent materials concretely from plant waste . In this work, we study the elimination of organic dye (Malachite Green) by an activated carbon. A series of experiments was carried out in order to study the influence on the adsorption capacity of certain p arameters such as the mass of the adsorbent, the pH, the contact time, the initial concentration of the dyes and the temperature. All the results obtained show that the adsorption kinetics of the dye on the material is well described by the pseudo second order model with a correlation coefficient $R2 > 0.99$. The adsorption isotherms of the adsorbent/adsorbate systems studied are satisfactorily described by the mathematical model of Langmuir and Freundlich. The pH of the solution has little influence on the bleaching performance. On the other hand, the thermodynamic study revealed that the adsorption is spontaneous and endothermic for the material used.

Keywords: Malachite Green, activated carbon, adsorption, adsorption isotherms, adsorption kinetics

DRAFT

ORAL PRESENTATION

Synthesis and Characterization of Amine Functionalized Graphene Oxide and its Preliminary Adsorption Study towards Methyl Orange Dye.

Mohd Haniff Wahid[1*] (*ORCID: https://orcid.org/ 0000-0002-2721-8697*),
Noor Nazihah Bahrudin[1] (*ORCID: https://orcid.org/ 0000-0001-9302-6741*)

[*]Universiti Putra Malaysia, Faculty of Science, Department of Chemistry, Serdang, Selangor, Malaysia.

*Corresponding author e-mail: mw_haniff@upm.edu.my

Abstract

Amine functionalized graphene oxide was successfully prepared using a facile technique. GO was first synthesized using the modified Hummers method, followed by chemical functionalization with dimethylamine (DMA). FTIR analysis of the end product ascertains the successful synthesis of GO as well as its functionalization with DMA. Typical characteristic peaks attributed to GO and DMA is clearly discernible in the spectrum. The resultant DMA modified GO is more stable as a suspension in water which further extends its potential applications. Preliminary adsorption test carried out at 10 ppm concentration of anionic dye i.e.; methyl orange (MO) showed that the modified GO is able to remove 12% compared to 7% removal achieved by pristine GO at 30 minutes contact time. Further studies will be conducted to determine the optimum adsorption condition for the highest removal of MO.

Keywords: Graphene oxide, dimethylamine, methyl orange.

DRAFT

ORAL PRESENTATION

Physicochemical analysis and microbiological quality of industrial and Algerian traditional cheese

Wafaa DIB[1,2*], Hadria GRAR[2,3], Aicha Lilia ANANI[1], Manel AOUFFEN[1], Fatouma Nadjoua ASLI[1], Djamel SAIDI[2,4], Omar KHEROUA[2], Samia ADDOU[2]

[1] Université des Sciences et de la Technologie d'Oran Mohamed Boudiaf, Faculté des Sciences de la Nature et de Vie, Département de Biotechnologie, Oran, Algérie.
[2] Laboratory of Physiology of Nutrition and Food Safety, University of Oran 1 Ahmed Ben Bella, Faculty of Science of Nature and Life, Department of Biology, Oran, Algeria.
[3] University of Mostaganem, Faculty of Science of Nature and Life, Department of Biology, Mostaganem, Algeria.
[4] Ecole Supérieure en Sciences Biologiques d'Oran, ex-IAP, BP 1042 Saim Mohamed, Oran, Algérie.

*Corresponding author e-mail: wafaa.dib@univ-usto.dz

Abstract

Cheese is a dairy food produced from coagulated milk. There is a wide variety of cheese around the world, each with its own characteristics and manufacturing methods. Our study focused on the characterization of three types of cheese [one traditional fresh cheese (Jben) and two industrial cheeses (T and P) by determining their physicochemical and microbiological properties. Physicochemical analyses indicated a pH value of 7.18 for the two type of industrial cheese. For the traditional cheese, the pH was acidic 4.47. Our results showed a high percentage of fat, dry matter and chloride for industrial cheese compared to traditional cheese: 19.5% > 6.83%, 39.75% > 20.61%, 1.01% > 0.47% respectively.
The microbiological study showed satisfactory microbiological quality in all three products with a total absence of pathogenic germs such as *E. coli*, *Listeria*, *Salmonella* and *Staphylococcus aureus.* In the light of these results, we conclude that all three type of cheese are of good microbiological and physicochemical quality.

Keywords: Raw milk, industrial cheese, traditional cheese, microbiological quality, physicochemical quality.

DRAFT

ORAL PRESENTATION

Phytochemical study and antimicrobial effect of the hydro-alcoholic extract of Calamintha nepeta from western Algeria

Chiali Fatima Zohra[1], Merzouk Hafida[2], Bahri Youcef[3]

[1]Laboratory of physiology physiopathology and biochemistry of nutrition, University of Tlemcen-department Biology –university of Mostaganem-Algéria.
[2]Laboratory of physiology physiopathology and biochemistry of nutrition, University of Tlemcen
[3]Laboratory of microbiology, University of Mostaganem

Abstract

Plants constitute an enormous reservoir of potential new molecules that can be the source of new active ingredients and lead to drugs. Our study consists in researching the antimicrobial activity of a plant, Calamintha nepeta belonging to the Lamiaceae family. Results & Discussion: The yield of Calamintha nepeta leaves in crude hydroethanolic extract is 12.33%. Phytochemical screening of Calamintha nepeta leaves show the presence of phenols, flavonoids, tannins, coumarins, saponins, alkaloids, proteins and terpenoids and the total absence of carbohydrates, quinones and anthraquinones. The content of polyphenols, flavonoids and tannins is 138.66mg/mL, 34.03mg/mL and 0.017mg/mL respectively. The sensitivity of the microbial strains tested to standard antimicrobial agents varies between 7 and 30 mm. The antibiotics that gave the largest zones of inhibition are: pristinamycin (PT15), chloramphenicol (C), nalidixic acid (NA), spiramicin (SP), gentamicin (GM) and neomycin (N). The hydroethanolic extract of Calamintha nepeta exerted an antimicrobial power against the microbial strains tested, the diameters of the zones of inhibition vary between 8 and 29 mm. The most sensitive strains are C. albicans ATCC 10231, P. mirabilis ATCC 35659 and Proteus vulgaris ATCC 6380 with inhibition zone diameters of 29, 19 and 17 mm respectively. The hydroethanolic extract of Calamintha nepeta exerted activity against the microbial strains tested, the values obtained range from 0.781 to 3.125 µl/mL for the CMI and from 3.125 to 50 µl/mL for the CMB and the CMF. Conclusion: Indeed, the hydroethanolic extract showed bacteriostatic activity on the majority of microbial strains tested. The hydroethanolic extract of Calamintha nepeta showed better activity against Gram-negative strains than Gram-positive strains. This result is important since gram-negative bacteria are more resistant to antimicrobial agents than gram-positive bacteria.

Keywords: Calamintha nepeta, hydroethanolic extract, nosocomial infections, antimicrobial activity

DRAFT

ORAL PRESENTATION

Chronic oxycarbonism in active and passive smokers in Algeria

Boudemagh Kenza [1], Belmahi Mohamed Habib[1]

[1]Constantine 3 University, Faculty of Medicine, Department of Pharmacy, Toxicology Laboratory, Algeria

Kenza.boudemagh@univ-constantine3.dz

Abstract

Smoking is a global scourge, leading to significant morbidity and mortality. In Algeria, tobacco kills 15,000 of its regular consumers, imposing a substantial burden in terms of diseases, disabilities, and suffering, with an extremely negative impact on family incomes and the national economy. One of the consequences of smoking is chronic oxycarbonism, which has multiple and potentially severe sequelae. Our study aimed to evaluate chronic oxycarbonism. among active and passive smokers. Carboxyhemoglobin levels were measured using the WOLFF method at the Ibn Badis Constantine University Hospital Toxicology Department from March to May 2023, involving 94 volunteers. The average carboxyhemoglobin concentration among active smokers in our study was 7.69% ± 4.02%. The highest carboxyhemoglobin values (3.10 ± 2.04%) were recorded among active smokers aged 60 to 85. For a tobacco consumption duration of more than 10 years, the average carboxyhemoglobin level was 8.54 ± 4.15%. The mean carboxyhemoglobin level among passive smokers in our study was 2.53% ± 1.81%. Interestingly, 42% of passive smokers studied had carboxyhemoglobin concentrations within normal limits despite their exposure to tobacco smoke. The fight against tobacco is a social responsibility of all healthcare personnel, including pharmacists. It is crucial to raise awareness among the population about the harmful effects of tobacco, particularly the issue of chronic oxycarbonism.

Keywords: Smoking, active smokers, passive smokers, oxycarbonism.

DRAFT

ORAL PRESENTATION

Aging-related changes in the diversity of scalp microbiome – preliminary studies

Karolina Oleś[1] (https://orcid.org/0009-0000-9708-5298),
Katarzyna Duda-Grychtoł[1] (https://orcid.org/0009-0008-7306-9611)

[1]Silesian College of Medicine, Health Department in Katowice, Poland

*Corresponding author e-mail: karolinainkaoles@gmail.com

Abstract

The scalp like other areas of the body is populated by microorganisms that make up the physiological flora. It is responsible for maintaining the proper functioning of the body. These microorganisms include bacteria, viruses, fungi or archaea. The physiological flora of the scalp can be disrupted as a result of various factors, which can lead to the development of conditions such as dandruff or seborrhea dermatitis. The aim of the study was microbiological diagnosis of the scalp and hair in subjects aged 20-40 years old. Material for the study was collected using contact plates for total bacterial count - Rodac Contac Test in 6 different individuals from three parts on the head. Colonies grown on microbiological media were characterized, subjected to Gram staining, and observed under a microscope. The results showed that there is a high morphological diversity of microorganisms isolated from the scalp from different individuals and collected from different sites. Gram-positive bacteria are more common in the physiological flora of the scalp. The most common form of bacteria are granular forms. The occurrence of candida is observed most abundantly in the middle part of the scalp, followed by the back and front. Fungal hyphae are present in subjects 4 and 6. In subjects 1 and 3, spore forms of bacteria are present.

Keywords: physiological flora of the scalp and hair, contact plates, Gram staining

DRAFT

ORAL PRESENTATION

Hepatotoxicity of Ecballium elaterium Following Traditional Therapeutic Use (Phytotherapy)

Mohamed Habib Belmahi, Kenza Boudemagh, Imane Mecheri

Toxicology Laboratory - Department of Pharmacy - Constantine3 University

habib.belmahi@univ-constantine3.dz

Abstract

Phytotherapy holds a significant place among the medicinal practices in our society. However, the underestimated risks associated with its use raise substantial health concerns. Hepatotoxicity is one of the major adverse effects of medicinal plants, given the physiological importance of the liver and the diversity of plants that can affect it. Our study aims to catalog several Algerian plants used in traditional therapy that carry hepatotoxic risks and contain various toxic compounds such as cucurbitacins. Our practical investigation focused on the plant "Ecballium elaterium." An epidemiological survey revealed widespread usage of this plant in two forms: the juice of the fruit and maceration. Administration to mice resulted in the development of hepatic lesions and disruptions in reduced glutathione (GSH) concentrations. This leads us to conclude that Ecballium elaterium indeed exhibits a genuine potential for hepatotoxicity.

Keywords: Ecballium elaterium, squirting cucumber, phytotherapy, traditional use, hepatotoxicity, oxidative stress.

DRAFT

ORAL PRESENTATION

Risk factors and biochemical complications of hypertension in pregnant women: Study from west Algeria

Hadria Grar[1,2*], Wafaa Dib[2,3], Latifa Benani[1], Nour El Islam Berrahil[1], Wahiba Rached[1], Hanane Gourine[2], Omar Kheroua[2], Djamel Saidi[4]

[1] University of Mostaganem Abdelhamid Ibn Badis, Faculty of Natural and Life Sciences, Department of Biology, Mostaganem, Algeria.
[2] University Oran 1 Ahmed Ben Bella, Faculty of Natural and Life Sciences, Department of Biology, Laboratory of Physiology of Nutrition and Food Safety, Oran, Algeria.
[3] University of Science and Technology USTO, Faculty of Natural and Life Sciences, Department of Biology, Oran, Algeria.
[4] Higher School of Biological Sciences of Oran (ESSBO), BP 1042 Saim Mohamed, Oran, Algeria.

*Corresponding author e-mail: ghadria@yahoo.fr

Abstract

Hypertension is the most common medical complication during pregnancy. It is one of the leading causes of maternal, fetal and neonatal morbidity and mortality. We aimed to investigate the risk factors for hypertension and its related complications in west Algerian pregnant women with the determination of their biochemical profile. Our population included 30 pregnant women (15 hypertensive women *vs.* 15 healthy women). A questionnaire containing 23 items was used to determine the main characteristics of the studied population. The biochemical profile was estimated by the determination of blood glucose, urea, creatinine, transaminases (TGO and TGP), prothrombin, quick time, TCK and proteinuria. In our study, 60 % of hypertensive pregnant women were in the age group of 30-40 years. Multiparity (73.33%), A blood group (60%), obesity (50%), stillbirth (40%) and diabetes (33.33%) were the main risk factors and complications observed among hypertensive women during pregnancy. Biochemical profile abnormalities were mainly presented by hyperglycemia, an increase in the level of TGO and TCK ($p<0.001$). During pregnancy, high blood pressure increased the risk of maternal and fetal morbidity and mortality.

Keywords: Biochemical profile, Complications, High blood pressure, Pregnancy, Risk factors

DRAFT

ORAL PRESENTATION

In silico investigation of effective antiviral compounds of natural origin for the treatment of COVID-19

Nabila Aoumeur [1,2*], Mebarka Ouassaf [2], Salah Belaidi [2]

[1*]Department HIS, Institute of Maintenance and Industrial Safety, University Mohamed Ben Ahmed Oran2, Algeria.
[2]Department of Chemistry, Faculty of Exact Sciences, Group of Computational and Medicinal Chemistry, LMCE Laboratory, Biskra University, Biskra 07000, Algeria.

*Corresponding author e-mail: aoumeur.nabila@yahoo.fr

Abstract

In molecular docking studies, a series of natural compounds investigated as inhibitors for the SARS-CoV-2 main protease (PDB ID: 6lu7) protein, the new strain of coronavirus, showed binding energy values with scores ranging from -7.99 to -7.62 kcal/mol. This study identified two compounds, h85 and h87, with high binding affinity, favorable pharmacokinetic characteristics, and minimal toxicity. Molecular dynamics (MD) simulation was also used to study the structural stability and dynamics of the two lead compounds inside the active site of 3CLpro; MD simulations revealed stable trajectories (RMSD and RMSF) and molecular properties with a rational interaction profile. It was established that these two lead molecules seemed to have the potential to function as attractive therapeutic candidates and would be of interest as starting points for SARS-CoV-2-targeting medicines.

Keywords: SARS-CoV-2, Natural compounds, Molecular docking, ADMET, Molecular dynamics.

DRAFT

ORAL PRESENTATION

Essential oils of three hemp (Cannabis sativa L.) cultivars in Rif Mountains (northern Morocco)

EL BAKALI Ismail [1*] (https://orcid.org/0000-0002-3013-534X)

[*1]Abdelmalek Essaâdi University, Faculty of Sciences, Department of Biology, Tetuan, Morocco

*Corresponding author e-mail: ismail.elbakali@gmail.com

Abstract

Hemp known botanically as Cannabis sativa L. (CS) is an important industrial crop. A huge literature was devoted to its biology owing to its numerous health-healing properties and biotechnological applications. However, little is known about essential oil (CSEO) chemical composition in the cultivars grown in Morocco. In this research work, we aimed at investigating CSEO chemical composition of three cultivars namely: 'Beldiya', 'Mexicana', and 'Critical Plus'. CSEOs were isolated from dried inflorescences using hydrodistillation and then subjected to chemical analysis using GC-MS. Our results showed that CSEOs yield and chemical composition varied significantly among cultivars. 'Critical Plus' showed the greatest record of CSEO yield (0.688%), while 'Mexicana' had the lowest value (0.330%). However, 'Critical Plus' presented the smallest values for the most of major compounds. Chemical composition showed an important phytochemical richness especially terpenoids. The most abundant compounds found in CSEOs were the following: β-caryophellene (13.39–25.32%), β-myercene (10.03–20.09), α-humulene (4.88–8.73%), caryophellene oxide (1.46–6.07%), decane (1.41–4.46%), and α-pinene (1.91–3.66%). The highest records of these compounds were reported in 'Beldiya' (β-caryophellene, caryophellene oxide, and decane) and 'Mexicana' (β-myercene, α-humulene, and α-pinene). Principal component analysis allowed a good separation of the three investigated cultivars based on their essential oil major compounds. It could be concluded that, the studied cultivars were caryophellene chemotypes, the local cultivar 'Beldiya' along with 'Mexicana' performed better in terms of essential oil yield and major phytochemicals over 'Critical Plus'.

Keywords:) Hemp cultivars, Essential oil, Terpenoids, Cultivars discrimination.

DRAFT

ORAL PRESENTATION

Worldwide potential insect vectors of *Xylella fastidiosa* and assessment of their importance with a focus on Morocco

Najat Haddad[1,2]* (*ORCID: https://orcid.org/0000-0001-7924-0928*), Moulay Chrif Smaili[1] (*ORCID: https://orcid.org/0000-0002-2390-6476*), Mohamed Afechtal[1] (*ORCID: https://orcid.org/0000-0003-4409-1226*), Rachid Benkirane[2] (*ORCID: https://orcid.org/0000-0002-2390-6476*)

[*1] National Institute of Agricultural Research, Regional Center of Agricultural Research of Kenitra, PO Box 257, 14000 Kenitra Morocco.
[*2] University Ibn Toufail, Faculty of Sciences, Laboratory of Plant, Animal and Agro-Industry Productions, Kenitra Morocco.

Corresponding author e-mail:* najat.haddad@uit.ac.ma

Abstract

Spittlebugs and sharpshooter leafhoppers, treehoppers and Cicadas are the main potential vectors of the bacterium *Xylella fastidiosa* (*Xf*) in the world. This study aims to give current data and analyze the occurrence and interaction between the potential insect vectors with *Xf* around the world, including America, Europe, Asia and North Africa, and in comparison, with Morocco. Citrus, olive, almond and grapevine, forest agroecosystems and oleander are considered the main susceptible host species to *Xf* and present in Morocco. This suggest the importance of the monitoring the potential insect vectors of this bacterium, including *P. tesselatus*, and the use the scientific tools to detect quickly the first bacteria inoculums of *Xf*, that could be introduced in the near future.

Key words: *Xylella fastidiosa*, potential vectors, *Philaneus tesselatus*, spittlebugs.

DRAFT

ORAL PRESENTATION

Starvation effect against ionizing radiation damages

Saloua Mabsor Zgandaoui[1,2*] (ORCID: *https://orcid.org/0009-0006-4601-9059*), Aboubaker El Hessni[2] (ORCID: *https://orcid.org/0000-0003-4890-7069*), Mohammed El Mzibri[1] (ORCID: *https://orcid.org/0000-0002-3148-1527*), Abdelghani Iddar[1] (ORCID: *https://orcid.org/0000-0001-6111-0607*), Adnane Moutaouakkil[1] (ORCID: *https://orcid.org/0000-0002-1762-881X*)

[1] National Center for Energy, Sciences and Nuclear Techniques (CNESTEN), Biotechnology and Biomolecules Engineering Unit, Rabat, Morocco.
[2] University Ibn Tofail, Faculty of Sciences, Department of Biology, Kenitra, Morocco.

[*] Corresponding author email: saloua.mabsor@uit.ac.ma

Abstract

The aim of this work was to highlight the effect of starvation against ionizing radiation damages using *Tetrahymena pyriformis* as a cellular model. Ionizing radiation has an indirect effect: the radiolysis of water which causes the generation of harmful molecules such as reactive oxygen species (ROS). Cellular detoxification processes involve the activation of antioxidant enzymes leading to the elimination of ROS. Numerous studies reveal that starvation has a beneficial impact on longevity, cell survival, and antioxidant effects. To evaluate the starvation effect on ionizing radiation damages, we exposed *T. pyriformis* cells, suspended in non-nutrient and nutrient media, to a cobalt source for 24h. Control cells were taken in the same conditions without irradiation. We monitored the cell number and the activity of some metabolic enzymes such as glyceraldehyde-3-phosphate dehydrogenase (GAPDH) and aldehyde dehydrogenase (ALDH) as well as some antioxidant markers such as catalase, superoxide dismutase and lipid peroxidation. Our results showed that the decrease in cell number after exposure to ionizing radiation was more important in non-starved cells than in starved ones. The decreases in the enzymatic activities of GAPDH and ALDH recorded after ionizing radiation exposure were greater in the case of non-starved cells compared to starved cells. The level of antioxidant markers remains relatively high after exposure to ionizing radiation in cells re-suspended in the non-nutrient medium compared to those re-suspended in the nutrient medium.

Keywords: Starvation, Ionizing radiation, *Tetrahymena pyriformis*, Metabolic enzymes, Antioxidant markers.

DRAFT

ORAL PRESENTATION

Biodiversity and Ecology of *Juniperus phoenicea* Litter Mesofauna in the Djebel Anoual Forest in the Tebessa Region

Hayat Benmaamar[1], Linda BouguessaCheriak[2], Slim Bouguessa[2], Faiza Marniche[3]

[1]Biological active molecules and applications laboratory, University of Tébessa
[2]Water and environment laboratory, University of Tébessa
[3]ENSV National School of Veterinary Sciences, El Harrache, Algeria

Corresponding author: hayatbenmaamar2022@gmail.com

Abstract

Juniperus phoenicea L.1753 is a tree or shrub adapted to the arid Mediterranean climate, it belongs to the cupressaceae family, the aim of our study is to contribute to the knowledge and identification of the meso-fauna of the litter of this species (*Juniperusphoenicea*) during the winter, spring and summer seasons of the year 2023 as well as its distribution in the different points selected sampling sites in the study site, The methodology consists of recovering *Juniperusphoenicea* litter from five (05) stations in the Djebel Anoual forest and placing it in the extractor device (Berlese-Tullgren) to extract the specimens, which are identified and then counted by the magnifying glass binocular, The results obtained showed the presence of differences in the composition of the mesofauna of the litter of this forest species during the three seasons (winter, summer and spring), thus in summer 14 species, 12 families and 05 orders of the single class of insects are inventoried, where the order *Diptera* and the family Muscidae are the most diverse taxa (28.57%, 21.43%), *Bradysia* sp. is the most represented species during this season, on the other hand, 40 species, 28 families, 15 classes, 04 orders are identified in winter where, *Sarcoptiforma* (*Arachnida*), Acaridae are the most diversified taxa with respectively 22.5%, 10%, 90.27%. Ceratophysellaarmata is the species the most represented during this season, Only 13 species, 09 families, 07 orders and 03 classes are noted in spring where *Entomobryomorpha* (16.67%) and *Poduromorpha* 16.67% are the most diverse orders as well as families and the most diverse family is *Sminthuridae* 15, 38%% and *Formicidae* 15.38%. and *entomobryidae* 15.38%. *Ceratophysellaarmata* is the most represented species during this season, so the seasonal factor affects the diversity and dominance of the litter mesofauna of this forest species.

Key words: *Juniperus phoenicea*, Djebel Anoual, mesofauna, litter

DRAFT

ORAL PRESENTATION

Earthworms affect the microbial community during vermicomposting

Lamia Yakkou[ab], Sofia Houida[ab], Maryam Chelkha[a], Serdar Bilen[b], Mohammed Raouane[a], Souad Amghar[a], Abdellatif El Harti[a].

[a]Research Team "Lumbricidae, Improving Soil Productivity and Environment (LAPSE)", Center "Eau, Ressources Naturelles, Environnement et Développement Durable (CERNE2D)" Mohammed V University, Ecole Normale Supérieure (ENS). Med Belhassan El Ouazani street. 5118, Takaddoum, Rabat, Morocco.
[b] Department of Soil Science and Plant Nutrition, Faculty of Agriculture, Ataturk University, 25000, Erzurum, Turkey.

Abstract

Earthworms, characterized as "ecosystem engineers", impact the soil microbial population and function by boosting micro-habitat, increasing the surface area of organic compounds, feeding and transporting microorganisms, and increasing the surface area of organic compounds. The multiscale holes produced by earthworm activities increase soil porosity and aeration, hence supporting the development and reproduction of microorganisms. In addition, earthworms transform complex organic components into microbe-accessible nutrients by feeding on, compacting, and mixing the soil. This increases decomposition and the cycling of key soil constituents such as carbon, nitrogen, and phosphorus, so enhancing the biological productivity of the soil. Vermicomposting is the process through which earthworms and microorganism's breakdown organic waste. Recent research emphasizes the significance of the earthworm gut as a significant influencer on microbial ecosystems. It has been demonstrated that the addition of these microbial communities to fresh organic matter modifies the activity level and functional diversity of microbial populations in vermicomposting systems. The involvement of microbial activity in the gut and cast of earthworms and soil is crucial for the decomposition of organic wastes. Therefore, our review paper examined how the microbial population changes during vermicomposting in order to provide a better understanding of the role of vermicompost as an additive to improve the quality of soil.

Keywords: Earthworm, microorganisms, vericompost, waste recycling

DRAFT

ORAL PRESENTATION

An Overview: The Botanical Extract Produced with SC -CO_2 Extraction for the Determination of Flavonoids and Terpenoids in Albanian Medicinal Plants

Jona KERI[1] (https://orcid.org/0000-0001-7606-341X), Joana MALO[2], Lorena MEMUSHAJ[3] (*https://orcid.org/0000-0003-1009-9289)*, Ina XHANGOLI[4] (https://orcid.org/0000-0001-6407-6408), Griselda ZACAJ[5], Lindita VRUSHI[6].

[1,2]Pharmacotherapeutic Research Center, Aldent University, Dibras Street no. 235, Tirana, Albania
[3,4] Pharmacy Department, Faculty of Medical Sciences, Aldent University, Dibras Street no. 235, Tirana, Albania
[5] Department of Medical Laboratory and Imaging Technology, Faculty of Technical Medical Sciences, Aldent University, Dibras Street no. 235, Tirana, Albania
[6]Lindita Vrushi – Esencial Manufactory, National Road Elbasan – Librazhd, km 2, Lizas Bridge, Elbasan 3014, Albania.

*Corresponding author e-mail: jona.keri@ual.edu.al

Abstract

Flavonoids are a group of substances with various phenolic structures found in foods such as fruits, vegetables, grains, flowers, tea, wine and other naturally occurring foods. These organic substances exhibit beneficial health properties, which is why researchers are working to isolate them from their natural sources, including medicinal plants. The aim of this study was to examine the presence of these compounds and terpenoids in medicinal plants that grow in Albania
After using the SC-CO_2 extraction technique, we were able to identify flavonoids present in approximately more than 10 distinct plants using our state-of-the-art method. With the support of GC-FID, the chemical profile was examined with a focus on identifying the 14 distinct terpenoids (carvacrol, citronellal, geraniol, linalool, linalyl acetate, piperitone, menthol and thymol) of these plants. With the use of HPLC, it was possible to identify the flavonoids. Mentha piperita (10.2 mg/g extract) is the medical plant with the lowest flavonoid content, while Cistus incanus (650 mg/g extract) and Melissa officinalis (480 mg/g extract) are the plants with the highest quantity. Citrus aurantium extract is the only plant with the highest concentration of terpenoids, containing 38.41% linalool, 4.95% geraniol and 11.7% linalyl acetate, followed by Origanum Vulgaris and Origanum heracleoticum, which contains higher concentrations of carvacrol at 76.94%.

Keywords: Albanian medicinal plants, flavonoids, terpenoids SC-CO_2 extraction

DRAFT

ORAL PRESENTATION

A new approach to preparation methodology of Polydopamine-Containing Systems

Cuneyt Erdinc Tas[1*] (*ORCID: https://orcid.org/0000-0001-8390-1434*)

[*1]Technological University of the Shannon, PRISM Research Institute, Centre for Polymer Sustainability, Athlone, Ireland.

*Corresponding author e-mail: cuneyt.tas@tus.ie

Abstract

Polydopamine (PDA) is an appealing material used for extensive scientific purposes, including light-to-thermal energy conversion, due to its numerous advantages. Properly integrating PDA into a composite system is vital to benefit from the PDA content to a maximum level. The PDA-containing systems can be produced by following the two main preparation methods: coating the surfaces of a targeted substrate with a PDA layer or preparing PDA nanoparticles in aqua media. In many different composite designs containing PDA chemistry, the emulsion/dispersion polymer latex systems should be considered from another perspective. From a general viewpoint, preparing hybrid systems possessing the PDA content with the polymer latex material, either the targeted composite product should be obtained after the water content is evaporated, followed by coating its surface with the PDA layer via a further reaction, or the PDA nanoparticles should be synthesized separately, and added in a polymer/water dispersion medium, producing the final product after the water part is evaporated. However, the composite systems designed with these methodologies are all in multicomponent and possibly inhomogeneous forms, which may need to be improved in fabrication, ecofriendliness, cost-effectiveness, and ease of applicability.

With this perspective, in this study group, a one-component, monolithic polymer matrix was produced by coating the surfaces of each polymer particle with the PDA layer while they were in the dispersion state. The new hybrid material preparation methodology was tested with polystyrene nanospheres synthesized by emulsion polymerization, and waterborne polyurethane systems. The comparative studies were evaluated in terms of the contribution of the method on the light-to-thermal energy conversion performance of PDA content in the hybrid materials as well as fundamental material characteristics and thermal energy storage.

Keywords: polydopamine, polystyrene, thermal energy storage, photothermal materials, polyurethane.

DRAFT

ORAL PRESENTATION

Immunomodulatory Properties of Mesenchymal Stem Cell

Fidan GAKHIYEVA[1-2] *(*ORCID: *0000-0002-0124-4694),* Ayla EKER SARIBOYACI[1-3*] (ORCID: *https://orcid.org/0000-0003-4536-9859*) Onur UYSAL[1-3] (ORCID: *https://orcid.org/0000-0001-6800-5607*), Sibel GÜNEŞ BAĞIŞ[1-3] (ORCID: *https://orcid.org/0000-0003-0846-1170*), Melih VELIPASAOĞLU[4] (ORCID: https://orcid.org/0000-0003-4070-3432), Burcugül ALTUĞ TASA[1] (ORCID:https://orcid.org/0000-0003-4460-8467), Merve Nur SOYKAN[1-2] (ORCID:https://orcid.org/0000-0003-1231-9791)

[1] Cellular Therapy and Stem Cell Production Application and Research Centre, ESTEM, Eskisehir Osmangazi University, Eskisehir, Turkey.

[2] Department of Stem Cell, Institute of Health Sciences, Eskisehir Osmangazi University, Eskisehir, Turkey.

[3] Department of Medical Laboratory Techniques, Vocational School of Health Services, Eskisehir, Turkey.

[4] Department of Gynecology and Obstetrics, Faculty of Medicine, Eskisehir Osmangazi University, Eskisehir, Turkey.

*Corresponding E-mail: aylaekersairiboyaci@yahoo.com

Abstract

Cellular therapy is one of the treatment methods for immunological diseases, and mesenchymal stem cells (MSCs), which are widely used in this therapy, are special cells that can self-renew without differentiation and differentiate into different cell types in the body. MSCs can be obtained from different tissues, including adult tissue and fetuses. In the literature, it has been shown that MSCs have a role in the regulation of the immune system and can be used in the treatment of immunological diseases. In immunological diseases, the sudden and acute increase in the level of pro-inflammatory cytokines in the circulation, which is called the cytokines storm, is mortal, as it happened during the COVID-19 pandemic. Preclinical studies with MSC paved the way for the clinical phase and the use of MSC for the treatment of GvHD. The aim of these preclinical studies is to analyze the immunosuppressive activity of hWJ-MSC on T cells. This study was planned in vitro, and instead of using high-cost autologous MSC as a high-tech product, MSC obtained from Wharton jelly of the umbilical cord, which is obtained from a fetal source and has stronger stemness because it is closer to the embryonic period, is used.In this study, the immunosuppressive effect of WJ-MSCs on T cells with respect to T cell anti- and pro-inflammatory cytokines, regulatory T cells (Tregs), some T cell marker gene expressions, T cell viability, proliferation, apoptosis, and cell cycle changes after co-culture have been examined. The initial findings of this study showed that hWJ-MSCs, as a universal and more potent source of MSCs allowing allogeneic applications, are at least as immunosuppressive as bone marrow-derived MSC.

Keywords: Wharton Jelly, MSCs, Immunosuppression, T cells.

Acknowledgment

This study was supported by Turkish Scientific and Technological Council (TUBİTAK 1004- Regenerative and Restorative Medicine Research and Applications) under the grant numbers of 20AG003 and 20AG031, Scientific Research Projects Coordination Unit under the grant number of 201846042 and TO-2022-2364.

DRAFT

ORAL PRESENTATION

Investigation of vascular damage in liver-cancer-on-chip model

Nigar GASIMZADE [1-2] (ORCID:https://orcid.org/0000-0001-6291-2337), Ceren OZEL[1-2] (ORCID: https://orcid.org/0009-0006-8189-6858), Özlem TOMSUK [1,3] (ORCID: https://orcid.org/0000-0001-7201-686X), Aliakbar EBRAHIMI[1] (ORCID:https://orcid.org/0000-0001-6437-7796), Hamed GHORBANPOOR[1,4] (ORCID:https://orcid.org/0000-0002-2665-8172), Zineb BENZAIT [1] (ORCID https://orcid.org/0000-0002-5329-6007). Nuran ABDULLAYEVA[1-2] (ORCID: https://orcid.org/0000-0002-9945-6686), Ayla Eker SARIBOYACI[1-2] (ORCID: https://orcid.org/0000-0003-4536-9859), Onur UYSAL[1-2] (ORCID: https://orcid.org/0000-0001-6800-5607), Hüseyin AVCI [1,5,6*] (ORCID: https://orcid.org/0000-0002-9638-3645).

[1] Cellular Therapy and Stem Cell Production Application and Research Center (ESTEM), Eskişehir Osmangazi University, Eskişehir, Turkey
[2] Department of Stem Cell, Eskişehir Osmangazi University, Eskişehir, Turkey
[3] Department of Mechanical Engineering, Middle East Technical University,Ankara 06800, Turkiye
[4] Department of Biomedical Engineering, Eskişehir Osmangazi University, Eskişehir, Turkey
[5] Department of Metallurgical and Materials Engineering, Eskişehir Osmangazi University Eskişehir, Turkey
[6] Translational Medicine Application and Research Center, Eskisehir Osmangazi University, Eskisehir, Turkey

*Corresponding author: havci@ogu.edu.tr

Abstract

The functional liver sinusoid consists of microvessels and can be modelled on a microscopic level in a three-dimensional (3D) *in vitro* environment using the liver on-chip model due to its early, quick, sensitive, and instant ability to detect clinical diagnoses of drug-induced injury, which is regarded as the most trustworthy microphysiological system in drug screening studies. Despite being successful against cancer drugs also impact healthy cells, which results in harmful consequences in numerous organs, including vascular tissue. Our study focused on the cytotoxicity and vascular damage effects of an anti-cancer drug on endothelial cells in a 3D microfluidic liver cancer model.

The PDMS-based microfluidic chip, which was fabricated by photolithography, consists of two microchannels: The parenchymal upper and the vascular bottom channels. Liver cancer cells were seeded on the upper side of the porous membrane, followed by seeding endothelial cells on the bottom side of the membrane for recapitulation of the liver sinusoidal capillary. The injury was created after administration of the anti-cancer drug to each microchannel at predetermined doses in our study, and then assessed by flow cytometry analysis with vascular damage biomarker and Live/Dead staining under fluorescence microscopy. According to Live/Dead images, the cell viability was lower in higher drug doses in 24 hours of chip culture. In addition, a higher vascular damage biomarker expression was detected after drug-treatment compared to the control (without drug treatment) in flow cytometry. The biomarker used in this study was found very effective to study vascular damage in chip systems such as tumor vasculature and other organ vasculature, which is shown for the first time in our chip system. In this study, liver cancer cell platforms, which are of great importance in both drug development and disease modelling, will enable the early detection of drug-related diseases such as cardiovascular damage.

Keywords: Drug-induced vascular injury, precision medicine. 3D vasculature model, liver-on-a-chip.

Acknowledgment
This study was supported by Turkish Scientific and Technological Council (TUBİTAK 1004- Regenerative and Restorative Medicine Research and Applications) under the grant numbers of 20AG003 and 20AG031 and Scientific Research Projects (BAP- the priority areas project (ONAP) under the grant numbers of TOA-2022-2307 and TYL-2023-2720 of Eskisehir Osmangazi University.

DRAFT

ORAL PRESENTATION

Synthesis of the Anthracene Sulfonyl Hydrazone Compound for Investigation of Biosensor Properties: Characterization, Electrochemical Measurements, and Theoretical Calculations.

Hilal Güler[1*] (*ORCID: https://orcid.org/0000-0003-2378-9174*), Servet Çete[1] (*ORCID: https://orcid.org/0000-0002-9316-2812*), Ümmühan Özdemir Özmen[1] ORCID: *https://orcid.org/* 0000-0001-9161-9367, Ayla Balaban Gündüzalp[1] ORCID: *https://orcid.org/* 0000-0002-5740-3674, and Deniz Akın Anakök[1] ORCID: https://orcid.org/0000-0002-7984-9159

[1]Gazi University, Science Faculty, Department of Chemistry, Ankara, TURKEY

*Corresponding author e-mail:hilal.guler1@gazi.edu.tr

Abstract

Thanks to the stability that conjugated double bonds in the structure of anthracene add to the molecule, their potential for use in sensors is very high. They are used in many different sensors, especially biosensors and chemosensors. Anthracene-like molecules with polycyclic structures (such as pyrene, and phthalocyanine) and their derivatives are known to be used for the immobilization of biomolecules on carbon nanotubes (CNT). Accordingly, anthracene derivatives have been used as binding agents for non-covalent immobilization. In this study, since the importance of S, N, and O atoms in compounds with known biosensor properties is emphasized in the literature, a new anthracene sulfonyl hydrazone compound containing sulfone and hydrazine was designed and anthracene ethanesulfonylhydrazone (Antesh) compound was synthesized. The excess hydrazine monohydrate used in the synthesis of sulfonylhydrazone compounds should be removed from the synthesized product due to its high cost and toxicity. The synthesis of Antesh has been handled in an environmentally friendly approach by using less amount of hydrazine monohydrate with solvent and mild reaction conditions suitable for green chemistry. The characterization of the compound (Antesh) was carried out by elemental analysis and spectroscopic methods (FT-IR, ^{1}H-NMR, and ^{13}C-NMR). Since Antesh is intended to accelerate electron transfer, electrochemical measurements by cyclic voltammetry and impedance spectroscopy were investigated. In this direction, within the scope of the project, graphene, gold nanoparticles, and transition metals were doped on the bare electrode surface together with Antesh, and a new interface was obtained. In addition, the molecular structure of Antesh was determined by computational studies (Gaussian 09 package program). The HOMO-LUMO energies, chemical reactivity descriptors, and molecular electrostatic potential (MEP) map of Antesh optimized by DFT/B3LYP method and 6-31+G(d,p) basis set were investigated.

K**eywords**: Sulfonyl hydrazone, Anthracene, Electrochemical measurements, Computational Studies

This work is supported by the Tubitak 123Z387 project

DRAFT

ORAL PRESENTATION

The Role of Fullerene C_{60} Nanoparticle against Lung Tissue Damage on Caspase-3, HO-1 and p53 Gene Expressions

Seda Beyaz[1]* *(ORCID: https://orcid.org/ 0000-0003-0436-8112)*, Abdullah Aslan[1] *(ORCID: https://orcid.org/0000-0002-6243-4221)*, Can Ali Agca[2] *(ORCID: https://orcid.org/ 0000-0002-0244-3767)*, Ibrahim Hanifi Ozercan[3] *(ORCID: https://orcid.org/ 0000-0002-8781-8838)*

[1]Firat University, Faculty of Science, Department of Biology-Molecular Biology and Genetics Program, Elazig, Turkiye
[2]Bingol University, Faculty of Science, Department of Molecular Biology and Genetics, Bingol, Turkiye
[3]Firat University, Medical School, Department of Pathology, Elazig, Turkiye

*Corresponding author: beyazseda23@gmail.com

Abstract
In this study, the treatment effect of fullerene C_{60} nanoparticle against lung tissue damage caused by 7,12-dimethylbenz[a]anthracene (DMBA) in Wistar albino female rats was investigated. The animal experiments part of this study was conducted in the Firat University Experimental Animal Research Center (FUDAM) with the permission of the Firat University Animal Experiments Ethics Committee dated 27.01.2021 and numbered 2021/02. In this study, 60 Wistar albino female rats (n = 60, 8 weeks old) were used. These rats were divided into 4 groups. Groups: (1) Control; (2) Fullerene C_{60}; (3) DMBA; (4) Fullerene C_{60} + DMBA. Expression levels of caspase-3, HO-1 and p53 proteins in lung tissue were determined by western blotting technique. Comparing the groups given Fullerene C_{60} to the groups given DMBA, it was found that the expression levels of the proteins caspase-3, HO-1, and p53 were much higher in the groups given Fullerene C_{60}. According to the results, it was determined that fullerene C_{60} has a therapeutic effect by reducing lung tissue damage.

Keywords: Caspase-3, HO-1, Lung, p53

Acknowledgements
This work was supported by Firat University Scientific Research Projects Unit (FUBAP) with FF. 20.07 project number. In addition, this study was supported by the Council of Higher Education (CoHE) 100/2000 Biotechnology priority field doctoral project and The Scientific and Technological Research Council of Turkey (TUBITAK) 2211/C program.

ORAL PRESENTATION

Development and efficacy of collagen suppressing peptide nanofibers

Deniz SEZLEV BILECEN (*https://orcid.org/0000-0002-5708-2276*)

Konya Food and Agriculture University, Dept of Molecular Biology and Genetics, Konya, Turkey

deniz.bilecen@gidatarim.edu.tr

Abstract

Peptide amphiphiles are monomers, having a hydrophobic core and an ionic segment thus have the ability to self-assemble into nanofiber structures. During their self-assembly process, they can encapsulate small molecules into their structure, becoming nano structured vehicles for cellular transport. Antisense oligonucleotides are 12-25 nucleotides long single stranded nucleic acid sequences which can inhibit target gene expression within the cells thus they are considered as nucleotide based therapeutic agents. One major drawback of antisense oligonucleotide delivery is their susceptibility to nucleases in the extracellular and intracellular environments. This obstacle might be overcome by using a vehicle for protection of the oligonucleotide in both sites. In this study, a ten amino-acid peptide with antifibrotic properties were modified to form the peptide amphiphiles (PA). These PAs were then used to form nanofibers which were used to encapsulate a GapmeR (antisense oligonucleotide) sequence, targeted towards a cytokine (Connective Tissue Growth Factor – CTGF) that is involved in excessive collagen synthesis. A variety of nanofiber formulations were studied, and the most stable form was administered to fibroblasts. The efficacy of the nanofibers was examined by tracking the collagen levels within the cells. The most stable nanofiber had the ratio of 100:1 and this nanofiber significantly reduced the collagen levels in the cells. In conclusion, the nanofibers may be an alternative therapeutic agent for the treatment of diseases with elevated extracellular matrix production is observed such as skin fibrosis.
Keywords: Nanofibers, antisense oligonucleotide, collagen

Acknowledgement: This study was funded by TUBITAK (Project number 120Z037).

DRAFT

ORAL PRESENTATION

Synthesis, Characterization and Determination of Antimicrobial Properties of New Intragenic Antimicrobial Peptide Derivative: GV-13

Güler Tuba Buğdacı[1,2*](*https://orcid.org/0009-0005-2518-025X*), Ruhane Tosunoğlu[1,2](*https://orcid.org/0009-0001-1715-0094*), Simay Aldağ[1,2] (*https://orcid.org/0009-0006-5282-1945*), Şeymanur Çobanoğlu[1,2] *(https://orcid.org/0000-0002-2805-0523)*, Ayşenur Yazıcı[1,2] (*https://orcid.org/0000-0002-3369-6791*), Serkan Örtücü[1,2](*https://orcid.org/0000-0002-3180-0444*), Mesut Taşkın[3](*https://orcid.org/0000-0002-9350-9628*)

[1]Erzurum Technical University, Faculty of Science, Molecular Biology and Genetics Department, Erzurum, Turkey.
[2]Erzurum Technical University, High Technology Research and Application Centre (YUTAM), Molecular Microbiology Laboratory, Erzurum, Turkey
[3]Atatürk University, Faculty of Science, Molecular Biology and Genetics Department, Erzurum, Turkey

*Corresponding author e-mail: guler.bugdaci48@erzurum.edu.tr

Abstract

Antibiotic resistance has become one of the most critical health problems today. New antimicrobial molecules should be developed due to the inadequacy of existing antibiotics. In this study, we aimed to determine for the first time the characterization and antimicrobial properties of synthetic antimicrobial peptide, GV-13, which we derived from human HS02 intragenic antimicrobial peptide by using database filtering technology. For this purpose, we first performed the solid phase peptide synthesis of the peptides, followed by their characterization by fouirer transform infrared (FTIR) and circular dichroism (CD) spectroscopy. Finally, the minimum inhibition value (MIC), motility and antibiofilm activity against Staphylococcus aureus (ATCC 43300) and Pseudomonas aeruginosa (PAO1) pathogen strains in vitro conditions were revealed. According to our results, it was revealed that GV-13, which we determined to have α-helix structure, exhibited MIC values of 64 and 128 µg/mL against S. aureus and P. aeruginosa, respectively. It provides biofilm inhibition and degradation of the formed biofilm at increasing concentrations. In line with these results, it has been determined that GV-13 is a potential antimicrobial peptide candidate and can be used against antibiotic resistance.

Keywords: Antimicrobial Peptides, Antibiotic Resistance, Database Filtering Technology, GV-13

This work is supported by TUBITAK Project 122Z741.

DRAFT

ORAL PRESENTATION

Synthesis of New Hydrazide-Hydrazone Derived from Etophenamate and Investigation of Their Antimicrobial Activity

Sude SARAL ÇAKMAK[1,3] (*ORCID: https://orcid.org/0000-0002-7342-8222)*, Burçak GÜRBÜZ[2] (*ORCID:* https://orcid.org/0000-0002-1002-8264), Pervin RAYAMAN[2] (*ORCID:* https://orcid.org/0000-0002-0487-8692), Sevgi KARAKUŞ[3] (*ORCID: https://orcid.org/0000-0002-7911-8372)*

[1]Marmara University, Institute of Health Sciences, Dragos, 34865 Kartal/Istanbul/Turkey
[2]Marmara University, Faculty of Pharmacy, Department of Pharmaceutical Microbiology, 34854 Basibuyuk, Maltepe/Istanbul/Turkey
[3]Marmara University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, 34854 Basibuyuk, Maltepe/Istanbul/Turkey

sudesaral@gmail.com

Abstract

Recently since the emerging resistant strains to antimicrobial agents, the development of original antimicrobial agents in pharmaceutical chemistry has gained great importance. Antibiotic resistance is accepted as one of the greatest threatens against health worldwide. Since 1950s to now it is known that hydrazide-hydrazones have antimicrobial, anticancer, antitubercular, anticonvulsant etc., biological spectrum and biological activity (Polovic et al., 2019). Hydrazone derivatives are used in treatment and involved in the structure of many drugs such as nifuroxazide, nitrofurazone, bisanthren, budralazine, and levosimendan. Etophenamate was chosen as the starting compound to synthesis of 16 novel hydrazide-hydrazones. All of these derivatives were investigated in order to determine their antimicrobial activities. The structures of the synthesized compounds were characterized by IR, ^{1}H-NMR, ^{13}C-NMR, mass spectroscopy and elemental analysis. In this study, the antimicrobial activity of 16 hydrazide-hydrazone derivatives to 4 standard bacterial strains (*Staphylococcus aureus* ATCC 25923, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853, *Enterococcus faecalis* ATCC 29212) and 4 standard yeast strains (*Candida albicans* ATCC 90028, *Candida glabrata* ATCC 90030, *Candida parapsilosis* ATCC 90018, *Candida tropicalis* KUEN 1021) was examined by the microdilution method in accordance with the Clinical and Laboratory Standards Institute (CLSI) standards, and the Minimal Inhibitory Concentration (MIC) values of these derivatives were determined. It was found that among these derivatives, **2c** (725-0.35µg/ml) and **2p** (1000-0.5µg/ml) possed antimicrobial activity against all of the 8 standard microorganism strains.

Keywords: Hydrazid-Hydrazone, Synthesis, Characterization, Antimicrobial activity

Acknowledgement: This work was supported by the The Scientific and Technical Research Council of Turkey (TÜBİTAK), project number: 121S745.

References:

*Polović, S.; Bilić, V.L.; Budimir, A.; Kontrec, D.; Galić, N.; Kosalec, I. *Acta Pharm*. **2019,** *69*(2), 277–285.
*Clinical and Laboratory Standarts Institue; Performance Standarts for Antimicrobial Susceptibility Testing. *CLSI document M100.* **2020,** *30th Ed*, January.

DRAFT

ORAL PRESENTATION

Investigation of the Effect of Different Extracts of *Fumaria officinalis* on Phenolic Content and Bioactivities

Bihter Şahin[1*], (*ORCID: https://*0000-0001-8657-052X), Merve Öner[1] (*ORCID: https://orcid.org/0009-0005-3562-6980)*, Mine Fındık[1] (*ORCID:* https://orcid.org/0009-0000-4311-2318), (*ORCID: https://*, Cansel Çakır[2] (*ORCID: https://*0000-0002-6175-9008), Yusuf Sıcak[3] (*ORCID: https://0000-0003-2339-5837*), Mehmet Öztürk[2] (*ORCID: https://orcid.org/0000-0001-8932-4535*)

[1] Bandırma Onyedi Eylül University, Susurluk Agriculture and Forest Vocational High School, Department of Chemistry and Chemical Processing Technologies, 10600, Balıkesir, Turkey
[2]Muğla Sıtkı Koçman University, Faculty of Science, Department of Chemistry, Muğla, Turkey
[3]Muğla Sıtkı Koçman University, Köyceğiz Vocational School, Department of Plant and Animal Production, Muğla, Turkey

* Corresponding author e-mail: bsahin@bandirma.edu.tr

Abstract

Fumaria officinalis which belong to the Fumariaceae family is represented by about 15 genera including the genus Fumaria, which encompasses about 40 species [1]. These species are herbaceous annuals [2] with a wide distribution in temperate areas of Europe and North Africa as well as in India, Pakistan, and West Asia [3,4]. It has been reported that the plant can be used for gastrointestinal disorders, as a laxative, anthelmintic, cholagogue or sedative as well as in the treatment of skin diseases, conjunctivitis and rashes, rheumatism, and high blood pressure [4]. The antioxidant activities of *Fumaria officinalis* aerial parts were determined by taking ethyl acetate (FO1), methanol (FO2) and aqueous ethanol (FO3) extracts, DPPH radical removal and ABTS cation removal activity, CUPRAC activity method, *β*-carotene-linoleic acid color bleaching activity methods, spectroscopic and *in vitro*. The phenolic content concentrations of the extracts in different polarities were determined by HPLC-DAD. Also, we tested anticholinesterase inhibition activity and tyrosinase inhibition activity in all extract. In conclusion, this study evaluated the bioactivities and chemical content of *Fumaria officinalis*, which grows in our country. It was determined that FO3 extract better results than other extracts in all antioxidant tests.

Keywords: *Fumaria officinalis*, Antioxidant Activity, Anticholinesterase Activity, Tyrosinase Activity, HPLC-DAD

Acknowledgement: This study was supported by TUBITAK 2209-A with the project numbered 1919B0122220690 within the scope of the University Students Research Projects Support Program in the 2022/2.

References:

1. Maiza-Benabdesselam, F., Khentache, S., Bougoffa, K., Chibane, M., Adach, S., Chapeleur, Y., Max, H., Laurain-Mattar, D., Antioxidant activities of alkaloid extracts of two Algerian species of Fumaria: *Fumaria capreolata* and *Fumaria bastardii*, Rec. Nat. Prod., 1, (2007), pp. 28-35.
2. Goetz, P., Ghedira, K., Le Jeune, R., *Fumaria officinalis* L. (Fumariaceae), *Phytothérapie*, 7 (2009), pp. 221-225.
3. Gilani,A.H., Rahman, A.U., Trends in ethnopharmacology, *J. Ethnopharmacol*., 100 (1–2) (2005), pp. 43-49.
4. Khamtache-Abderrahim, S., Lequart-Pillon, M., Gontier, E., Gaillard, I., Pilard, S., Mathiron, D., … Maiza-Benabdesselam, F., Isoquinoline alkaloid fractions of *Fumaria officinalis* : Characterization and evaluation of their antioxidant and antibacterial activities. *Industrial Crops and Products*, (2016), 94, 1001–1008. doi:10.1016/j.indcrop.2016.09.016.

DRAFT

ORAL PRESENTATION

Exploring the Theoretical Framework of Cobalt-Pentapyridyl Molecular Catalyst: A Deeper Dive into the EECC Water Reduction Mechanism

Ayas Kiser[1](*ORCID: https://orcid.org/0000-0001-8422-2687),*
Yeliz Gurdal[1*] (*ORCID: https://orcid.org/0000-0002-6245-891X)*

[1]Adana Alparslan Türkeş Science and Technology University, Faculty of Engineering, Department of Bioengineering, Adana, Turkiye.

*ygurdal@atu.edu.tr

Abstract

Natural sources of the remarkable energy carrier hydrogen (H_2) include biomass, water, and hydrocarbons. The most abundant energy source is solar energy, which plants use naturally to fuel their photosynthetic processes. Utilizing cobalt-based photocatalysts, researchers have recently studied photocatalytic water reduction, which was inspired by the natural photosynthesis mechanism. Different transition metals have been used to simulate the water reduction mechanism of photosynthesis. However, in terms of price/performance ratio, no appropriate solution could be discovered. In this context, we explore the water reduction, commonly known as H_2 production, process of Co-based polypyridyl catalyst possessing perfect octahedral coordination using Ab-initio Molecular Dynamics (AIMD), Density Functional Theory (DFT), and Free Energy Perturbation Theory. Our goal is to understand how the perfect coordination around the Co centrer affects the EECC reaction pathway. Two reduction and two protonation steps of the water reduction cycle have been simulated using AIMD simulations and allowed spin states as well as solvent response around the reaction center of the catalyst have been determined. Electronic structures of each intermediate steps have been further examined applying hybrid density functional framework of DFT. Marcus's theory of electron transfer has been used to compute the first and the second reduction free energies.

Keywords: H_2 production, water reduction, ab-initio molecular dynamics, density functional theory

This study has received funding from the TUBITAK under the 3501 Career Development Program (grant agreement No: 120Z240).

DRAFT

ORAL PRESENTATION

Phycobiliprotein content and growth profile of *Spirulina platensis* in airlift photobioreactor

Buse Dincoglu[1] (https://orcid.org/0009-0001-1305-6121), Ugur Tepe[1] (https://orcid.org/0000-0003-0054-8941) Bahar Aslanbay Guler[1] (https://orcid.org/0000-0002-0113-4823), Zeliha Demirel[1] (https://0000-0003-3675-7315), Esra Imamoglu[1*] (https://orcid.org/0000-0001-8759-7388)

[*1]Ege University, Faculty of Engineering, Department of Bioengineering, İzmir, Turkey.

*Corresponding author e-mail: esra.imamoglu@ege.edu.tr

Abstract

Microalgae are organisms that can easily adapt to different environmental conditions, allowing them to increase their biomass and produce metabolites. They are widely utilized in various industries due to their biochemical content and rapid growth rate. Cultivation of microalgae cells occurs in different systems at various scales and methods, including photobioreactors and open ponds. Among numerous microalgae, *Spirulina platensis* is a commonly preferred species in industry. It is one of the most important commercial producers of phycobiliproteins, which are characteristic bioactive materials found in blue-green algae. Phycocyanin produced from *S. platensis* also holds high added value. This study reports on the production of phycocyanin from *S. platensis* using an airlift photobioreactor. The airlift photobioreactor provides several advantages, including the absence of additional mixing components and the ability to replace O_2 and CO_2 with aeration gas and medium. The growth rate of *S. platensis* was found as 0.28 ± 0.01 day^{-1} and the doubling time was 2.47 ± 0.09 day. The phycobiliprotein contents were obtained as 1.03±0.001 mg/L for phycoerythrin, 10.37±0.08 mg/L for phycocyanin, 6.29±0.24 mg/L for allophycocyanin, and 17.71±1.76 mg/L for total phycobiliprotein. Given the benefits of an airlift photobioreactor, cultivating *S. platensis*, a commercial microalgae, in such a reactor can result in significant potential for producing biomass and bioactive material.

Keywords: *Spirulina platensis*, phycobiliprotein, phcocyanin, airlift photobioreactor

Acknowledgements
This study was a part of Cost Action CA20127 and the authors would like to thank the Scientific and Technological Research Council of Turkey (TUBITAK) with project number 121R100 for the financial support.

DRAFT

ORAL PRESENTATION

Pre-sowing treatment with seaweed extracts improve early growth of wheat

Hande MUTLU-DURAK[*1] (*ORCID: https://orcid.org/0000-0003-4351-5795*), Yağmur ARIKAN[1] (*ORCID: https://orcid.org/0000-0002-1252-2979)*, Ümit Barış KUTMAN (*ORCID: https://orcid.org/0000-0002-9158-0332*), Bahar YILDIZ KUTMAN[1]*(ORCID: https://orcid.org/0000-0002-7891-7527*)

[*1]Gebze Technical University, Institute of Biotechnology, Gebze, Kocaeli, Turkey.

*Corresponding author's e-mail: handemutlu@gtu.edu.tr

Abstract

To ensure better crop yields and produce vigor seedlings, each seed must be healthy and germinate quickly, which is very important for agricultural production. However, seeds may deteriorate over time and lose their germination capacity, viability, and vitality. In such cases, the pre-sowing application of biostimulants as growth-promoting agents is one of the most promising approaches in agricultural practices. With these applications, improved plant metabolism and performance can be observed both before and after germination. It is well recognized that seaweed-based formulations that contain rich bioactive compounds are used as biostimulants in crop production and the effects of pre-sowing treatment with seaweed extracts on germination, growth, and early plant development have been the subject of various studies in the literature. This study used two different application methods (seed or substrate applications) on wheat (*Triticum durum* cv. Saricanak-98) seeds to investigate the biostimulant effect of several extracts (hot water, alkali, acidic) from *Cystoseira barbata* (*C. barbata*), a brown seaweed species that is prevalent throughout the Mediterranean coasts. In this investigation, the effects of seaweed extracts on wheat seedling growth characteristics, root system morphology, and mineral concentrations were assessed. The results demonstrate that seaweed extracts can improve the performance of wheat seedlings and have a positive impact on several growth parameters. Moreover, the findings showed that seaweed extracts were more effective at enhancing seedling performance when added to the growing medium than when applied directly to seeds. *C. barbata* extracts can be used as promising agents in sustainable agriculture as they are highly efficient, eco-friendly, and safe products. Therefore, this research reveals the potential of our country's economic resources such as *C. barbata* to be transformed into high-value goods that can contribute to the country's economy.

Keywords: Biostimulant, *Cystoseira barbata*, Seaweed Extract, Seed Treatment, Wheat.

This study was supported by TUBITAK (Project number: 121Z215).

DRAFT

ORAL PRESENTATION

Enhancement of the activity and stability of *Porcine Pancreatic* lipase by physical adsorption immobilization onto Zeolitic Imidazolate Frame (ZIF-67)

Ozge Caglar[1*] (*ORCID: https://orcid.org/0000-0002-6595-1582*),
Elif Ozyilmaz[2] (*ORCID: https://orcid.org/0000-0003-4360-4165*)

[*1] Selcuk University, Institute of Sciences, Department of Chemistry, Konya, Turkey
[2] Selcuk University, Faculty of Science, Department of Biochemistry, Konya, Turkey

*Corresponding author e-mail: ozgecaglar07@gmail.com

Abstract

Zeolitic imidazolate frames (ZIFs), which are composed of metal sources such as Zn and Co and organic ligands such as imidazole or imidazole derivatives, are hybrid structured materials with unique features such as designable surface area, pore size, chemical and thermal stability. The free lipase is known as the most used in industrial biotechnology and microbiology. Although free lipases are frequently used in industry, their stability is poor and their conformational structures are disrupted when exposed to high temperatures. In order to overcome these disadvantages and improve a strong biocatalyst, immobilization of enzymes on solid supports is believed one of the most efficient methods to get better their activity, affinity, selectivity and stability. ZIFs are thought to be the perfect support materials to protect the conformational structure of enzymes against adverse environments and to enhanced their activity and stability due to their special properties. One of the strategies for immobilizing the enzyme on the surface of zeolites is adsorption. Adsorption-based techniques are superior to other methods due to their economical, high performance, easy applicability, suitability for a wide range of concentrations, and ease of installation. In this study, ZIF-67, a new support material was prepared for physical adsorption of *Porcine Pancreatic* lipase (PPL). The ZIF-67@PPL was characterized by using techniques such as SEM, XRD, and FT-IR. Furthermore, pH, thermal stability, storage time, and the reusability of immobilized lipase were determined and compared to free lipase. It was determined that ZIF-67@PPL still retained 61% of its activity after 5 times reuse. The Free PPL retained its catalytic activity of 25% at 60 °C after 120 min, while ZIF-67@PPL retained 49 % of its catalytic activity at the end of 120 min.

Keywords: MOF, ZIF-67, lipase, immobilization

Acknowledge: This research is supported by the Selcuk University Scientific Research Projects Foundation (SUBAP grant number 22401034) and TUBITAK 2211-C PhD scholarship.

DRAFT

ORAL PRESENTATION

Modulation of in vitro pollen germination and tube elongation of kiwi (*Actinidia deliciosa*) by spermidine

Melse Su Bilgili* (*ORCID: https://orcid.org/0000-0002-2062-7295*), Özkan Kilin (*ORCID: https://orcid.org/0000-0002-9283-4576)*, Aslıhan Çetinbaş-Genç (*ORCID: https://orcid.org/0000-0001-5125-9395*)

*[1]Marmara University, Faculty of Science, Biology Department, İstanbul, Türkiye.

*Corresponding author e-mail: melsesubilgili@gmail.com

Abstract

The aim of this study is to investigate whether possible to modulate the in vitro pollen germination and tube elongation of kiwi (*Actinidia deliciosa*) by spermidine treatment. For this purpose, pollen grains were germinated for 3 hours in pollen germination media containing spermidine at different concentrations (0.01 mM, 0.025 mM, 0.05 mM, 0.1 mM, 0.25 mM, 0.5 mM). First, pollen germination rates and pollen tube lengths were determined and then pollen germination rates and pollen tube lengths results were evaluated cumulatively. The results showed that spermidine treatments did not generate a significant change in pollen germination rates when compared to the control. Pollen tube lengths were significantly increased by 13,85% after 0,01 mM spermidine treatment. However, pollen tube lengths were significantly decreased by 24,60% after 0,025 mM, by 22,97% after 0,05 mM, by 24,62 after 0,1 mM, by 21,65% after 0,25 and by 25,92% after 0,5 mM spermidine treatment, compared to the control. According to cumulative stress response index results, 0,01 mM spermidine stimulated pollen germination and tube elongation while the other concentrations decreased. As a result, spermidine could be used for modulating the pollen germination and tube length and these findings could be useful for artificial pollination practices.

Keywords: kiwi, pollen germination, pollen tube, polyamine, spermidine

This work was supported by the 2209-A-Research Project Support Programme for Undergraduate Students with project identification number 1919B012201192.

DRAFT

ORAL PRESENTATION

Investigation of the presence of tetracycline antibiotic residues and total aerob mesophilic bacteria in some foods consumed in Ankara

Özdegül Baştürk[1*] *(ORCID: https://orcid.org/0000-0003-2554-9588)*, Burak Demirhan[2] *(ORCID: https://orcid.org/0000-0001-8551-1472)*, İnci Barut[2] *(ORCID: https://orcid.org/0000-0002-0289-1447)*, Buket Er Demirhan[2] *(ORCID: https://orcid.org/0000-0002-7938-6553)*

[*1]Gazi University, Institute of Health Sciences, Department of Food Analysis and Nutrition, Ankara, Turkey

[2]Gazi University, Faculty of Pharmacy, Department of Pharmaceutical Basic Sciences, Ankara, Turkey

*Corresponding author e-mail:ozdegul.basturk@gazi.edu.tr

Abstract

Antibiotics are used in poultry to promote growth, increase feed efficiency, and reduce diseases. One of the most used antibiotics on farms is tetracycline. However, excessive use of tetracycline in food-producing animals can lead to residues that may adversely affect public health. Regarding food safety and human health, it is significant to check for the presence of antibiotics in animal foods and to calculate the total aerob mesophilic bacteria (TAMB) counts. This study aimed to determine whether certain foods consumed in Ankara, Turkey, contained TAMB and tetracycline antibiotic residues. For this purpose, the enzyme-linked immunosorbent assay (ELISA) technique was used to analyze a total of 82 food samples (chicken meat, eggs, pasteurized milk and unpackaged raw milk). Additionally, the existence of TAMB was analyzed using classical culture methods. According to our findings, tetracycline was found in 9 (10.98%) of 82 food samples at concentrations ranging from 0.90-11.24 ppb (µg/kg, µg/L). Tetracycline was not detected in the egg samples. The mean value (±S.E) of positive chicken meat and milk samples for tetracycline were determined to be 1.59 µg/kg and 6.40±1.68 µg/L, respectively. The TAMB values of the chicken samples for the firms A, B, and C were 4.78±0.09, 5.68±0.17, and 4.38±0.11 log cfu/g, respectively. Additionally, TAMB scores for pasteurized milk samples from the firms F, G, H and raw milk were 3.40± 0.22; 4.09± 0.20; 3.11± 0.23; 6.64± 0.13 log cfu/mL, respectively. Our data showed that the levels of tetracycline detected in all samples were within the limits set by Turkish Food Codex. Finally, the present study highlights the importance of antibiotic detection and microbiological analysis not only for food control but also for human health.

Key words: Antibiotic, Food, Tetracycline, ELISA, Microbiological analysis

Acknowledgements: This research was supported by Gazi University Scientific Research Projects Foundation (Ankara, Turkey, Project No: TYL-2022-7515).

DRAFT

ORAL PRESENTATION

Green synthesis of ZnO nanoparticles using plant wastes: Antimicrobial and cytotoxic activities

Merve Çapkın Yurtsever[1*] (*ORCID: https://orcid.org/0000-0001-7874-4016*), Zeynep İyigündoğdu[1] (*ORCID: https://orcid.org/0000-0003-2067-4822*), Hüsnü Arda Yurtsever[2] (*ORCID: https://orcid.org/0000-0002-1920-8149*)

[1]Adana Aplarslan Türkeş Science and Technology University, Faculty of Engineering, Department of Bioengineering, Adana, Türkiye.
[2]Department of Materials Science and Engineering, Adana Alparslan Türkeş Science and Technology University, Adana, Türkiye

*Corresponding author e-mail: mcyurtsever@atu.edu.tr

Abstract

In recent years, biological molecules such as proteins, vitamins and phenols have been frequently used in the synthesis of metallic nanoparticles due to their reducing and stabilizing properties. The avocado seed, with its valuable components could be an important source for the food and/or pharmaceutical industry. Within the scope of this study, ZnO nanoparticles (ZnONPs) were synthesized at room temperature using MeOH-H_2O extract of avocado seed. Different concentrations of avocado seed 1, 3, and 5 mg/mL were used in the green synthesis of ZnONPs. Results of XRD, ATR-FTIR, DLS, SEM analysis, and UV-Vis spectroscopy showed that crystalline ZnONPs can be produced in the presence of avocado seed extract at all concentrations. The crystallite sizes of nanoparticles with hexagonal crystalline structures ranged from 17 to 20 nm. SEM images of ZnONP agglomerates revealed flower-like formations. ZnONPs-3 was found to have the most uniform size distribution according to DLS analysis. Minimal inhibitory concentrations were determined between 7.81 - 125 µg/mL, for ZnONPs. Human normal skin (BJ) and brain glioblastoma cancer (T98G) cell lines were used to investigate the cytotoxicity of ZnONPs-3. The IC50 concentrations for BJ and T98G cells were found to be 36.05 µg/mL and 23.60 µg/mL, respectively, at 24 hours. These numbers decreased to 31.07 µg/mL and 18.05 µg/mL at 48 hours. ZnONPs-3 showed toxicity against cancer cells at concentrations which were non-toxic for normal cells. Results of this study reveal that ZnONPs produced in the presence of avocado seed extract can be considered as a valuable active material that can be used in biomedical applications.

Keywords: Green synthesis, ZnO, antimicrobial, cytotoxicity

Acknowledgements: This work was supported by the Scientific Research Projects Unit of Adana Alparslan Türkeş Science and Technology University with project number 21103012.

DRAFT

ORAL PRESENTATION

Synthesis, aggregation behaviour, fluorescence, singlet oxygen generation and photodegradation studies of pyrazoline substituted axial silicon, alpha (α) and beta (β) zinc phthalocyanines

Halise Yalazan[1*] (*ORCID:https://orcid.org/0000-0003-1234-2721*), İpek Ömeroğlu[2] (*ORCID: https://orcid.org/0000-0002-7528-0911*), Gonca Çelik[1] (*ORCID:https://orcid.org/0000-0002-4634-3354)*, Halit Kantekin[1] (*ORCID:https://orcid.org/0000-0003-2625-2815*), Mahmut Durmuş[2] (*ORCID:https://orcid.org/0000-0003-0440-7345*)

[1]Karadeniz Technical University, Faculty of Sciences, Department of Chemistry, Trabzon, Turkey
[2]Gebze Technical University, Department of Chemistry, Gebze, Kocaeli, Turkey

haliseyalazan@hotmail.com

Abstract
Photodynamic therapy (PDT) is a clinically proven cancer treatment method with fewer side effects and a two-stage treatment designed to destroy cancerous or precancerous cells after mild activation of a photosensitizer (PS). Light-activated PSs of a specific wavelength are responsible for the conversion of molecular oxygen to cytotoxic singlet oxygen ($^1O_2$) in the targeted tissue [1,2]. Phthalocyanines (Pc), used as photoactive drugs in PDT, are second generation photosensitizers due to their chemical stability, sufficient fluorescent emissions and singlet oxygen activities [3]. In this study, pyrazoline substituted axial silicon, peripheral (β) and non-peripheral (α) zinc phthalocyanines were synthesized and their photophysicochemical properties were investigated. When the results are examined, it is thought that silicon phthalocyanine shows the highest singlet oxygen production in DMSO and can be a candidate photosensitizer for PDT [4].

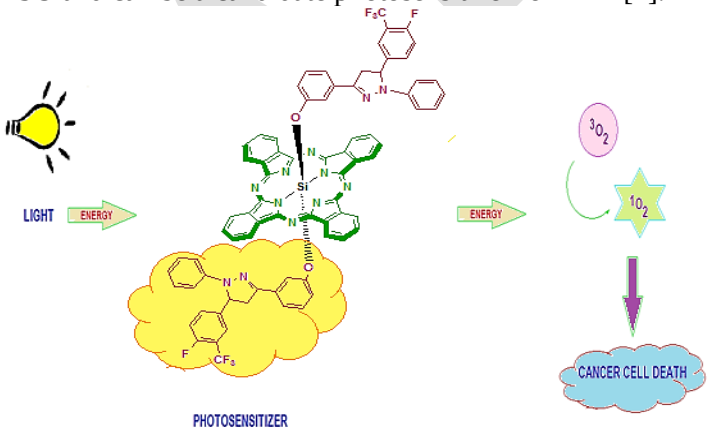


Figure 1. Silicon phthalocyanine compound, photosensitizing candidate for PDT

Keywords: Phthalocyanine, photodynamic therapy, pyrazoline, singlet oxygen

Acknowledgements: This work was supported by Office of Scientific Research Projects of Karadeniz Technical University. Project number: FDK-2021-9351.

References

J.F. Lovell, T.W. Liu, J. Chen, G. Zheng, Chem. Rev. 110 (2010) 2839–2857. https://doi.org/10.1021/cr900236h.
A. Sowa, A. H¨oing, U. Dobrindt, S.K. Knauer, A. Galstyan, J. Voskuhl, Chem.–A Eur. J. 27 (2021) 14672–14680. https://doi.org/10.1002/chem.202102255.
S.D. Ezquerra Riega, F. Valli, H.B. Rodríguez, J. Marino, L.P. Roguin, B. Lantaño, M. C. García Vior, Dyes and Pigments 201 (2022) 110110. https://doi.org/10.1016/j.dyepig.2022.110110.
H. Yalazan, İ. Ömeroğlu, G. Çelik, H. Kantekin, M. Durmuş, Inorganica Chimica Acta 551 (2023) 12180. https://doi.org/10.1016/j.ica.2023.121480.

ORAL PRESENTATION

Two-dimensional WSe_2 nanosheets-based electrochemical sensor for simultaneous detection of epinephrine and dopamine

Gamze Celık Cogal[1,2*] (*https://orcid.org/0000-0001-9064-0834*), Sadik Cogal[2,3] (*https://orcid.org/0000-0001-8904-1332*), Maria Omastová[3] (*https://orcid.org/0000-0003-0210-5861*)

[1]Suleyman Demirel University, Faculty of Arts and Science, Department of Chemistry, 32000, Isparta, Türkiye
[2]Polymer Institute, Slovak Academy of Sciences, Dubravska cesta 9, 84541 Bratislava, Slovakia
[3]Burdur Mehmet Akif Ersoy University, Faculty of Arts and Science, Department of Chemistry, 15030, Burdur, Türkiye
*Corresponding author e-mail: gamzec3557@gmail.com

Abstract

Transition metal dichalcogenides (TMDs) possess unique properties that make them highly suitable candidate materials for sensing applications. Tungsten diselenide (WSe_2), as a remarkable member of TMDs, has emerged as a promising material for the development of advanced electrochemical sensors [1-2]. WSe_2 has exceptional electronic, optical, and chemical properties, combined with its high surface-to-volume ratio and large specific surface area, contribute to its exceptional sensing performance. Furthermore, the facile functionalization of WSe_2 surfaces allows for tailored sensor designs, enabling the detection of a wide range of molecules. The electrochemical sensor design incorporates 2D-TMD nanomaterials as the sensing platform due to their large surface area, high electrical conductivity, and exceptional electrocatalytic properties [3]. The selective detection of dopamine (DA) and epinephrine (EPI) is of significant importance in clinical diagnostics and neurochemical research, as these neurotransmitters play crucial roles in various physiological processes and are associated with numerous neurological disorders. In this work, we focused on the preparation of 2D-WSe_2 nanosheets for application in electrochemical sensors for determination of EP and DA. WSe_2 nanosheets were prepared via a facile hydrothermal method. The surface morphology and composition of the WSe_2 were characterized by using X-ray diffraction (XRD), scanning electron microscopy (SEM) coupled with energy-dispersive X-ray spectroscopy (EDX), and transmission electron microscopy (TEM). The as-prepared WSe_2 were coated on a glassy carbon electrode (GCE) to fabricate an electrochemical sensor for simultaneous detection of DA and EP. Cyclic voltammetry (CV) and electrochemical impedance spectroscopy (EIS) methods were performed to determine the electrochemical behaviours of the WSe_2-modified electrodes. Performance parameters of the developed sensor were obtained from the differential pulse voltammetry (DPV) measurements. It was found that WSe_2-based electrochemical sensor exhibited wide linear detection ranges with low detection limits for DA and EP.

Keywords: Two-dimensional nanomaterials, transition metal dichalcogenides, tungsten diselenide, electrochemical sensor, dopamine, epinephrine.

Acknowledgements:
This work was partially funded by Project VEGA 02/0006/2023. G.C. C and M. O gratefully acknowledges the National Scholarship Programme of the Slovak Republic. This work has received funding from the European Union´s Horizon 2020 Research and Innovation Programme under the Programme SASPRO 2 COFUND Marie-Sklodowska-Curie grant agreement No. 945478.

References:

[1] I. Kim, S. W. Park, D. W. Kim, Carbon-coated tungsten diselenide nanosheets uniformly assembled on porous carbon cloth as flexible binder-free anodes for sodium-ion batteries with improved electrochemical performanceJournal of Alloys and Compounds 827 (2020) 154348.
[2] Tangal, Y., Coban, D., Cogal, S., A WSe2@poly(3,4-ethylenedioxythiophene) nanocomposite- based electrochemical sensor for simultaneous detection of dopamine and uric acid. J. Electrochem. Sci. Eng.12(6) (2022) 1251-1259.
[3] Divya, M., Surbhi, S., Nagaraj P.S., Soumen, B., Kakarla R. R., Tejraj, M.A., Advances in transition metal dichalcogenide-based two-dimensional nanomaterials. Materials Today Chemistry. 19 (2021) 100399.

DRAFT

ORAL PRESENTATION

Green synthesis of Ag-doped on CuO from pomegranate and its antibacterial and photocatalytic activities application

Nesrin Ankıt[1], Keziban Atacan[1*] (*ORCID:* https://orcid.org/0000-0003-3927-7245)

[1]Sakarya University of Applied Sciences, Akyazı Vocational School of Health Services, 54400 Sakarya, Turkey

*Corresponding author e-mail: kezibanatacan@subu.edu.tr; keziiiban@gmail.com

Abstract

In recent years, plants have been used extensively for nanoparticle (NP) synthesis due to the presence of many useful phytochemicals used as reducing and stabilizing agents in their extracts [1]. Green synthesis of nanoparticles using plant extracts is an emerging research area and has advantages over chemical synthesis as it reduces the elaborate process and can also afford large-scale production [2, 3]. Metal oxide nanoparticles have many notable applications such as cell line studies, antimicrobial and dye degradation [4-5]. Copper oxide (CuO) nanoparticles have outstanding properties that make them essential for a variety of applications such as sensors, catalysts, and super-strong materials [4,5]. Despite the high availability and application of CuO NPs in multiple organs/systems, few studies have been conducted to examine their immunotoxic effects. The toxicity of CuO NPs on human lymphocytes remains to be investigated [4,5]. Antimicrobial mechanisms of nanomaterials can be investigated to study the specific binding of microorganisms to their surfaces and their metabolism in microorganisms [6].
In this study, the first goal is the preparation of copper oxide nanoparticles using the green synthesis method using pomegranate fruit and the doping of silver nanoparticles on copper oxide nanoparticles. Secondly, the antibacterial effect of the prepared nanoparticles against both gram positive (*Staphylococcus aureus*) and gram negative (*Escherichia coli*) bacteria and the photocatalytic activity have been examined.

Keywords: green synthesis, CuO, antibacterial activity, photocatalytic.

Acknowledgment
The authors thank the Scientific and Technological Research Council of Türkiye (TÜBİTAK 2209-A, Project No. 1919B012216479) for financially supporting this study.

References
[1] Sajadia, S.M., Kolo, K., Abdullah, S.M., Hamad, S.M., Khalid, H.S., Yassein, A.T., Surfaces and Interfaces 13 (2018) 205–215.
[2] Suhad, A. Yasin, Jamal, A. Abbas, Ibtisam, A. Saeed, Idrees, H. Ahmed, Polymer Bulletin 77 (2020) 3473–3484.
[3] Kumar, PV, Shameem, U, Kollu, P, Kalyani, RL, Pammi, SV, BioNanoScience 5 (3) (2015) 135–139.
[4] Naika, HR., Lingaraju, K., Manjunath, K., Kumar, D., Nagaraju, G., Suresh, D., Nagabhushana, H., J Taibah Univ Sci 9(1) (2015) 7–12.
[5] Al-Dhabi, N., Valan A.M., Nanomaterials 8(7) (2018) 500.
[6] Ghosh, S., Ahmad, R., Zeyaullah, M., Khare, S.K., Front. Chem. 9 (2021) 1-19.

DRAFT

ORAL PRESENTATION

DNA binding and nuclease properties of magnesium (II) phthalocyanines

Gökçe Seyhan[1*] (*ORCID: https://orcid.org/0000-0002-8553-9093*), Burak Barut[1] (*ORCID: https://orcid.org/0000-0002-7441-8771*), Barış Kulein[1](*ORCID: https://orcid.org/0009-0004-5141-2299),* Turgut Keleş[2] (*ORCID: https://orcid.org/0000-0002-1911-8020*), Zekeriya Bıyıklıoğlu[2](*ORCID: https://orcid.org/0000-0001-5138-214X*)

[*1]Karadeniz Technical University, Faculty of Pharmacy, Department of Biochemistry, Trabzon, Türkiye.
[2]Karadeniz Technical University, Faculty of Science, Department of Chemistry, Trabzon, Türkiye.

*Corresponding author e-mail: gokceseyhan@ktu.edu.tr

Abstract

Photodynamic therapy (PDT) is an alternative cancer treatment method based on the dynamic interaction between a photosensitizer that provides selective destruction of the target tissue, light of a certain wavelength, and molecular oxygen, and the free radicals formed as a result of this interaction destroy the cancerous tissue. PDT is triggered by the application of photosensitizer that selectively accumulates in the tumor tissue, followed by exposure of light of a suitable wavelength to the applied area. It transfers energy from light to molecular oxygen to form reactive oxygen species, such as singlet oxygen, superoxide radical, hydroxyl radical and hydrogen peroxide. These cytotoxic photoproducts initiate a series of biochemical events that can cause damage and death in the target tissue. Phthalocyanines are one of the photosensitizers used in photodynamic therapy. In this project, the DNA interactions of water-soluble non-peripheral and peripheral tetra-({6-[3-(diethylammonium)phenoxy)hexyl}oxy substituted magnesium (II) phthalocyanine (n-DE-C6-MgQ and DE-C6-MgQ) compounds were investigated. This study aimed was to examine the therapeutic potential of the compounds against cancer. The ct-DNA binding properties of the compounds were investigated using UV-Vis spectroscopy (UV-Vis absorption and competitive ethidium bromide binding studies) and the electrophoretic method. Then, plasmid DNA nuclease activities of the compounds (hydrolytic cleavage, photocleavage, oxidative cleavage, oxidative photocleavage) were investigated using the electrophoretic method. The results showed that n-DE-C6-MgQ and DE-C6-MgQ interacted to ct-DNA via intercalation. In agarose electrophoresis studies indicated that the compounds did not show any nuclease effect without irradiation, whereas they had photonuclease activity with light irradiation via a singlet oxygen pathway. The results revealed that the non- n-DE-C6-MgQ had a stronger DNA photodamage potential than DE-C6-MgQ due to their solubility properties.

Keywords: DNA binding, magnesium, phthalocyanine, plasmid DNA.

This study was supported by The Scientific and Technological Research Council of Türkiye, TÜBİTAK (TÜBİTAK 2209-A program) (Project number: 1919B012104848)

DRAFT

ORAL PRESENTATION

The Investigation of Epistatic Associations of Two SNPs in the NRG1 and ERBB4 Genes with Schizophrenia

Mustafa Mert SÖZEN[1*] (ORCID: https://orcid.org/0000-0002-1672-2206),
Şükrü Kartalcı[2] (ORCID: https://orcid.org/0000-0003-2560-0355)

[1] Inonu University, Faculty of Science and Literature, Department of Molecular Biology and Genetics, Malatya, Türkiye
[2] Inonu University, Faculty of Medicine, Department of Psychiatry, Malatya, Türkiye.

* Corresponding author e-mail: mert.sozen@inonu.edu.tr

Abstract

The variants of NRG1 and ERBB4 genes are listed in the earliest genetic factors associated with schizophrenia. We analyzed the epistatic interactions between the alleles of NRG1 SNP rs10503929 and ERBB4 SNP rs1026882 to see if they have effects on schizophrenia. We screened the SNPs in a case-control group (96 schizophrenia patients and 100 controls) living in Malatya-Turkey. None of these SNPs were associated with the disease at single SNP levels. To find potential epistatic associations, we determined the combinations of SNP genotypes and predicted the epistatic structures in each individual. Following this, we compared two groups for the distributions of these epistatic structures.
We determined eight potential epistatic structures. Statistical tests indicated there were no significant differences between the two groups for the distribution of any structures we have determined. Even though one combination was seen in six patients, but only one normal control sample, these numbers were too low to make a reliable comment. Even if there is a positive association between the disease and this epistatic structure, our relatively small sample size and the limited geographic area may have prevented us from finding clear evidence. Screening new samples from more populations in larger groups may help to understand the connection between schizophrenia and these SNPs.

Keywords: Schizophrenia, Genetics, Association, NRG1, ERBB4, Neuregulin-1, Turkey

This study has been supported by The Unit of Scientific Research Projects at İnonu University. Project Number: FCD-2021-2552 to Mustafa Mert SÖZEN.

DRAFT

ORAL PRESENTATION

Investigation of mesenchymal stem cells on drug-induced liver injury by using liver on a chip system

Leman GULUZADE[1-2] (ORCID: https://orcid.org/ 0009-0000-3538-2561), Özlem TOMSUK[1,3] (ORCID: *https://orcid.org/* 0000-0001-7201-686X), Ceren ÖZEL[1-2] (ORCID: *https://orcid.org/*0009-0006-8189-6858), Nigar SEYIDOVA[1-2] (ORCID: *https://orcid.org/* 0009-0005-4533-2513), Aliakbar EBRAHİMİ[1] (ORCID: *https://orcid.org/*0000-0001-6437-7796), Hamed GHORBANPOOR[1,4] (ORCID: *https://orcid.org/*0000-0002-2665-8172), Zineb BENZAIT[1] (ORCID: *https://orcid.org/* 0000-0002-5329-6007), Nuran ABDULLAYEVA[1-2] (ORCID: *https://orcid.org/*0000-0002-9945-6686), Nigar GASİMZADE[1-2] (ORCID: *https://orcid.org/0000-0001-6291-2337*), Ayla Eker SARIBOYACI[1-2] (ORCID: *https://orcid.org/*0000-0003-4536-9859), Onur UYSAL[1-2] (ORCID: *https://orcid.org/*0000-0001-6800-5607), Ender YILDIRIM[3] (ORCID: *https://orcid.org/* 0000-0002-7969-2243), Hüseyin AVCI[1,5-6*] (ORCID: *https://orcid.org/*0000-0002-9638-3645)

[1]Cellular Therapy and Stem Cell Production Application and Research Center (ESTEM), Eskişehir Osmangazi University, Eskişehir, Türkiye
[2]Department of Stem Cell, Eskişehir Osmangazi University, Eskişehir, Türkiye
[3] Department of Mechanical Engineering, Middle East Technical University, Ankara 06800, Türkiye
[4]Department of Biomedical Engineering, Eskişehir Osmangazi University, Eskişehir, Türkiye
[5]Department of Metallurgical and Materials Engineering, Eskişehir Osmangazi University Eskişehir, Türkiye
[6]Translational Medicine Application and Research Center, Eskisehir Osmangazi University, Eskisehir, Türkiye

*Corresponding author: havci@ogu.edu.tr

Abstract

Drug-induced liver injury (DILI) is now a major medical issue that affects both the pharmaceutical business and the general public health on a global scale. DILI refers to both clinically visible and silent liver damage. In regenerative medicine, mesenchymal stem cells (MSCs) have been used for treatment for many years. On the other hand, the shortcomings of present tools utilized in cell cultures, animal models, and drug research have brought to light the need for a novel new strategy. However, there is a large gap between in vitro and in vivo approaches and their applications in clinical practice. Thus, organ-on-a chip technology (OoC) has been utilized recently as one of these methods. OoC microfluidic devices coated with live cells cultured under fluid media can recapitulate organ-level physiology. Therefore, in the present study, it is aimed to form a hepatotoxicity model on the liver on a chip platform and investigate the effect of MSCs. For this purpose, initially, static co-cultures are used to determine the effect of a hepatotoxic drug with the effect of mesenchymal stem cells including parenchymal and nonparenchymal liver cells. As a result, the MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) test was used to determine the effect of the hepatotoxic drug dosages on cell survival and to optimize the number of MSCs. Depending on the time and the quantity of MSCs, it has been found that MSCs therapy can be an effective in cytotoxic doses of the drug by using on a chip platform. As a result, a novel model of liver on a chip for examining the effects of hepatotoxicity and MSCs can be a therapeutic strategy for the liver treatment.

Keywords: Hepatotoxic drug, MTT, Mesenchymal Stem Cells, Liver on a chip.

Acknowledgment
This study was supported by Turkish Scientific and Technological Council (TUBİTAK 1004- Regenerative and Restorative Medicine Research and Applications) under the grant numbers of 20AG003, 20AG031 and 122C158 (TUBİTAK 2218- National Postdoctoral Research Fellowship Program) and Scientific Research Projects (BAP- the priority areas project (ONAP) under the grant number of TOA-2022-2307 of Eskisehir Osmangazi University.

DRAFT

ORAL PRESENTATION

A novel tool for regenerative medicine: OoC platforms meet MSCs

Nigar SEYIDOVA[1-2] (ORCID: *https://orcid.org/* 0009-0005-4533-2513), Özlem TOMSUK[1,3] (ORCID: *https://orcid.org/* 0000-0001-7201-686X), Ceren ÖZEL[1-2] (ORCID: *https://orcid.org/*0009-0006-8189-6858), Leman GULUZADE[1-2] (ORCID: https://orcid.org/ 0009-0000-3538-2561), Aliakbar EBRAHİMİ[1] (ORCID: *https://orcid.org/*0000-0001-6437-7796), Hamed GHORBANPOOR[1,4] (ORCID: *https://orcid.org/*0000-0002-2665-8172), Zineb BENZAIT[1] (ORCID: *https://orcid.org/* 0000-0002-5329-6007), Nuran ABDULLAYEVA[1-2] (ORCID: *https://orcid.org/*0000-0002-9945-6686), Nigar GASİMZADE[1-2] (ORCID: *https://orcid.org/0000-0001-6291-2337*), Ayla EKER SARIBOYACI[1-2] (ORCID: *https://orcid.org/*0000-0003-4536-9859), Onur UYSAL[1-2] (ORCID: *https://orcid.org/*0000-0001-6800-5607), Ender YILDIRIM[3] (ORCID: *https://orcid.org/* 0000-0002-7969-2243), Hüseyin AVCI[1,5-6*] (ORCID: *https://orcid.org/*0000-0002-9638-3645)

[1]Cellular Therapy and Stem Cell Production Application and Research Center (ESTEM), Eskişehir Osmangazi University, Eskişehir, Türkiye
[2]Department of Stem Cell, Eskişehir Osmangazi University, Eskişehir, Türkiye
[3] Department of Mechanical Engineering, Middle East Technical University, Ankara 06800, Türkiye
[4]Department of Biomedical Engineering, Eskişehir Osmangazi University, Eskişehir, Türkiye
[5]Department of Metallurgical and Materials Engineering, Eskişehir Osmangazi University Eskişehir, Türkiye
[6]Translational Medicine Application and Research Center, Eskisehir Osmangazi University, Eskisehir, Türkiye
*Corresponding author: havci@ogu.edu.tr

Abstract

Regenerative medicine is an interdisciplinary field and a modern branch of medicine that deals with the restoration of body tissues damaged by disease, injury or age-related changes. Mesenchymal stem cells (MSCs) are among the most promising stem cell groups in regenerative medicine due to their wide clinical use potential. MSCs play an important role in the repair of damaged tissues and suppression of inflammation due to their differentiation potential; growth factor, cytokine, chemokine secretion; migration to the injury site and immunomodulatory effects. In addition, MSCs have an antifibrotic effect on fibrotic processes such as liver cirrhosis as well as they have a strong anti-inflammatory effect. Through paracrine pathways in particular, they can differentiate into hepatocytes, stimulate liver regeneration, prevent liver fibrosis, and induce liver apoptosis. Thus, stem cell therapy has a therapeutic importance in liver diseases. On the other hand, today the technology known as "Organ-on-a-Chip (OoC)" is frequently utilized as a common tool to simulate microengineered and biomimetic systems that mimic the structural and functional properties of human tissue. Besides, culturing the stem cells alone or together with other cells and differentiating the stem cells in the desired cell types, which has attracted the attention of regenerative medicine, have begun to be studied in OoC systems. Therefore, in the present study, studies covering the effect of MSC treatment on drug-induced hepatotoxicity were reviewed using keywords such as "MSCs therapy for liver diseases; MSCs therapy in liver regeneration; DILI on liver-on-a-chip and MSC therapy in DILI" in search engines such as Pubmed, ScienceDirect, Google Academic and Scopus. Finally, it has been demonstrated by clinical studies that MSCs therapy has an important position in the treatment of liver diseases. The generation of MSCs therapy protocols, which specifically in regeneration of damaged cells and support the host tissue, can be facilitated by OoC platforms with increased output metrics.

Keywords: MSCs therapy, Regenerative medicine, Organ-on-a-chip (OoC), Drug-induced liver injury (DILI).

Acknowledement
This study was supported by Turkish Scientific and Technological Council (TUBİTAK 1004- Regenerative and Restorative Medicine Research and Applications) under the grant numbers of 20AG003, 20AG031 and 122C158 (TUBİTAK 2218- National Postdoctoral Research Fellowship Program) and Scientific Research Projects (BAP- the priority areas project (ONAP) under the grant number of TOA-2022-2307 of Eskisehir Osmangazi University.

DRAFT

ORAL PRESENTATION

Exploring anti-cancer drug activities by 3-dimensional liver cancer on a chip

Nuran ABDULLAYEVA[1-2] (ORCID: https://orcid.org/0000-0002-9945-6686), Özlem TOMSUK [1,3] (ORCID: https://orcid.org/0000-0001-7201-686X), Bahar DEMİR CEVİZLİDERE [1] (https://orcid.org/0000-0002-2444-8833), Ceren OZEL[1-2] (ORCID: https://orcid.org/0009-0006-8189-6858), Aliakbar EBRAHIMI[1] (ORCID:https://orcid.org/0000-0001-6437-7796), Hamed GHORBANPOOR[1,4] (ORCID:https://orcid.org/0000-0002-2665-8172), Zineb BENZAIT1 (ORCID: https://orcid.org/ 0000-0002-5329-6007), Nigar GASIMZADE [1-2] (ORCID:https://orcid.org/0000-0001-6291-2337), Ayla Eker SARIBOYACI[1-2] (ORCID: https://orcid.org/0000-0003-4536-9859), Onur UYSAL[1-2] (ORCID: https://orcid.org/0000-0001-6800-5607), Hüseyin AVCI [1,5,6*] (ORCID: https://orcid.org/0000-0002-9638-3645)

[1] Cellular Therapy and Stem Cell Production Application and Research Center (ESTEM), Eskişehir Osmangazi University, Eskişehir, Turkey
[2] Department of Stem Cell, Eskişehir Osmangazi University, Eskişehir, Turkey
[3] Department of Mechanical Engineering, Middle East Technical University,Ankara 06800, Turkiye
[4] Department of Biomedical Engineering, Eskişehir Osmangazi University, Eskişehir, Turkey
[5] Department of Metallurgical and Materials Engineering, Eskişehir Osmangazi University Eskişehir, Turkey
[6] Translational Medicine Application and Research Center, Eskisehir Osmangazi University, Eskisehir, Turkey

*Corresponding author: havci@ogu.edu.tr

Abstract

Organ on a chip (OOC) is a dynamic culturing system that mimics cells and tissues closer to their in-vivo environment. OOC devices are more ethical and cost-effective than animal models, which are also time-consuming and frequently fail to predict therapeutic effectiveness and toxicity in people. Furthermore, OOC devices that create a more realistic and regulated microenvironment are less expensive because they require fewer sample quantities, expensive reagents and equipment. Cancer-on-a-chip models based on microfluidics are also an effective tool for researching the tumor microenvironment and its involvement in metastasis, as well as fast assessing the efficacy of cancer medicines. Therefore, in this study, a liver cancer model was formed as a liver cancer on-a-chip platforms and the effect of a chemotherapeutic agent was investigated. For this purpose, WST-8 analysis was used to determine different doses of the chemo applied in static co-culture as a control to compare with the microfluidic setup. In order to mimic liver cancer on the chip model, both surfaces of the membrane integrated into the chip system were coated with extracellular matrix (ECM). Then, the liver cancer and endothelial cells were seeded on a coated membrane oppositely and incubated in a culture incubator at 37°C under an atmosphere of 5% CO_2. The cell viability and apoptotic effects of the drug on the cells were investigated by injecting the chemotherapeutic agent in both channels at a constant flow rate for 24 hours using a peristaltic pump. Afterward, the cell viability on the membrane was determined by the Live/Dead staining. Thus, it has been demonstrated that cell viability is reduced significantly with the effect of the chemotherapeutic agent at the administered dose. Finally, we demonstrated that liver cancer-on-chip can have a potential to investigate anti-cancer drug activities which are closer to *in vivo* models in a faster and a more accurately manner.

Keywords: Liver cancer-on-a-chip, chemotherapeutic, precision medicine, drug screening, 3-dimension model.

Acknowledgment
This study was supported by Turkish Scientific and Technological Council (TUBİTAK 1004- Regenerative and Restorative Medicine Research and Applications) under the grant numbers of 20AG003 and 20AG031 and Scientific Research Projects (BAP- the priority areas project (ONAP) under the grant number of TOA-2022-2307 of Eskisehir Osmangazi University.

DRAFT

ORAL PRESENTATION

Evaluation of cholinesterases inhibitory effects of some azole derivatives

Didem Akkaya[1*] (*ORCID: https://orcid.org/0000-0002-0711-951X*), Burak Barut[1] (*ORCID: https://orcid.org/0000-0002-7441-8771*), Suat Sarı[2] (*ORCID: https://orcid.org/0000-0002-8248-4218*), Arzu Özel[1](*ORCID: https://orcid.org/0000-0001-7381-5575*)

[*1]Karadeniz Technical University, Faculty of Pharmacy, Department of Biochemistry, Trabzon, Türkiye.
[2]Hacettepe University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, Ankara, Türkiye.

*Corresponding author e-mail: didemakkaya@ktu.edu.tr

Abstract

Alzheimer's disease (AD) is a neurological disorder that is usually seen in elderly individuals and causes memory loss, cognitive impairment, behavioral changes and loss of functionality over time. This disease occurs as a result of the death of brain cells over time and the deterioration of their communication. The cholinergic hypothesis is a theory that attempts to explain the effects of AD on acetylcholine, the neurotransmitter system. Acetylcholine is a chemical messenger (neurotransmitter) that enables communication between nerve cells. In AD, the nerve cells that produce acetylcholine in the brain are damaged, leading to decreased acetylcholine levels. According to the cholinergic hypothesis, cognitive symptoms of AD occur due to acetylcholine deficiency. The azole ring has a five-membered ring structure and contains one or more nitrogen atoms. Such compounds may have various biological activities in pharmaceutical, agricultural, chemical and other industrial fields. In this study, it was aimed to determine acetylcholinesterase (AChE) and butyrylcholinesterase (BuChE) inhibitory properties of some azole compounds were investigated using spectroscopic method. Moreover, Lineweaver-Burk and Dixon plots were performed to determine kinetic parameters (inhibitory type and constant (K_i) values) of the compounds against AChE and BuChE. Among tested compounds, **13** showed the best inhibitory effect on BuChE with an IC_{50} value of 18.41 ± 1.55 µM. Also, Compound **13** was more potent than galantamine (40.48 ± 0.06 µM). These results showed that compound **13** had an important potential for anti-AD drug due to their cholinesterase inhibitory properties.

Keywords: azoles, acetylcholinesterase, butyrylcholinesterase, kinetic analysis.

This work was supported by Office of Scientific Research Projects of Karadeniz Technical University. Project number: TSA-2022-10388.

DRAFT

ORAL PRESENTATION

The Role of lncRNA H19 in the Resistance of Ovarian Cancer Cells to Cisplatin Therapy

Ehsan SAYEDALI [1*] (0000-0003-1140-1216), Merih AKKAPULU[1] (0000-0002-5884-2986), Kadir ESER[2] (0000-0001-8666-6177), Halil ÇELİK[2] (0000-0003-2246-8707), Mehmet YALDIZ[3] (0000-0003-0640-4551), Ali Erdinç YALIN[1] (0000-0002-3351-6885), Emel SEZER[2] (0000-0001-9881-3533), Vehbi ERÇOLAK[2] (0000-0003-1014-1694), Serap YALIN[1](0000-0002-1286-2172)

[1]Department of Biochemistry, Faculty of Pharmacy, Mersin University, Mersin, Turkey
[2]Department of Medical Oncology, Faculty of Medicine, Mersin University, Mersin, Turkey
[3]Department of Pathology, Faculty of Medicine, Mersin University, Mersin, Turkey

*ehsanhasan66@yahoo.com

Abstract

Epithelial ovarian cancer, often diagnosed at an advanced stage, possesses one of the highest mortality rates among gynecological malignancies. The standard treatment regimen combines surgery with chemotherapy, mainly platinum-based therapies. Regrettably, the effectiveness of this approach is compromised by the emergence of chemoresistance. Recent research has illuminated the role of long non-coding RNAs (lncRNAs) in chemoresistance, offering potential insights into this clinical challenge. LncRNAs wield substantial influence over various aspects of cancer biology, including cell survival, apoptosis, and responsiveness to chemotherapy. Among these, lncRNA H19 emerges as a pivotal player in the initiation, progression, and recurrence of diverse human cancers. This study aimed to investigate the correlation between lncRNA H19 expression and cisplatin resistance in ovarian cancer cells, a widely employed platinum-based chemotherapy agent. The study cohort comprised ovarian cancer patients subjected to cisplatin treatment. Formalin-fixed, paraffin-embedded ovarian tissue specimens were collected from these individuals. Stratifying the samples into two categories based on treatment response, Group 1 represented patients exhibiting sensitivity to cisplatin therapy with subsequent recovery, while Group 2 encompassed patients who remained resistant to cisplatin treatment without improvement. RNA isolation was performed on all tissue samples using a specialized RNA isolation kit, followed by cDNA synthesis using a cDNA synthesis kit. The resulting cDNAs served as templates for precise quantification of lncRNA H19 gene expression through Real-Time PCR. The expression levels between the two groups were compared using the T-test and a regression model mediated by SPSS. In our comparative analysis of the resistant and sensitive groups, our investigation revealed a statistically significant increase in lncRNA H19 expression within the resistant group. This finding underscores the potential implication of lncRNA H19 in the development of cisplatin resistance in ovarian cancer, shedding light on a novel avenue for further exploration and therapeutic intervention.

Keywords: Ovarian cancer, cisplatin, chemotherapy, lncRNA H19, long noncoding RNA

Acknowledgement: This study was supported by the Research Fund of Mersin University with Project Number: 2022-1-TP2-4608

DRAFT

ORAL PRESENTATION

Investigation of Electrical Conductivity of Polyselenophene/MnO Hybrid Composites

Kevser Okcu,[1] Kemal Özdoğan,[1] Azra Hüner[2]

[1]Department of Physics, Yildiz Technical University, Esenler 34220, Istanbul, Turkey
[2]Department of Chemistry, Yildiz Technical University, Esenler 34220, Istanbul, Turkey

E-mail: kevserokcu1@gmail.com

Abstract

The polymer chain of conducting polymers contains alternating single and double bonds that present delocalized electrons that serve as charge carriers. Conducting polymers are considered promising semiconductor materials due to their distinct electrical properties, relative simplicity of processing, and mechanical flexibility. Hybrid composites of conductive polymers attract great attention from researchers due to their unique electrical and optical properties, as well as their many applications in sectors such as sensors, electronics, nanotechnology and biotechnology. Since these systems have a wide variety of applications, much research is being done on hybrid composites of conducting polymers with transition metal oxidesIn this study, polyselenophene (PSe), a conducting polymer, was synthesized by chemical oxidative polymerization. Manganese oxide (MnO) were synthesized from manganese salts. Then, PSe/MnO hybrid composites containing different amounts of MnO were synthesized by in situ oxidative polymerization. Their electrical conductivity was measured in the range of approximately 10^4-10^5 S/cm with the four-point probe technique. Characterization of PSe and PSe/MnO hybrid composites was performed by FTIR-ATR, UV-Vis spectroscopy, XRD and SEM.

Keywords: Polyselenophene, manganese oxide, electrical conductivity, composite.

Acknowledgements: This study was supported by Yildiz Technical University Scientific Research Projects Coordination Unit (Project No: FYL-2023-5633).

DRAFT

ORAL PRESENTATION

Synthesis and antimicrobial activity of some new thiosemicarbazone derivatives containing benzothiophen moiety

Emine Öztürk[1*] (*https://orcid.org/0000-0002-0466-5638*), Elif Subaşı[2] (*https://orcid.org/0000-0002-4235-9391*) Aslı Şahiner[3] (*https://orcid.org/0000-0001-5095-9461*), Betül Şen Yüksel[4] (*https://orcid.org/0000-0001-7846-8090)*

[*1]Dokuz Eylul University, The Graduate School of Natural and Applied Sciences, Department of Chemistry, Izmir, Turkiye
[2]Dokuz Eylul University, Faculty of Sciences, Department of Chemistry, Izmir, Turkiye
[3]Ege University, Faculty of Science, Department of Biology, Izmir, Turkiye
[4]Dokuz Eylul University, Faculty of Science, Department of Physics, Izmir, Turkiye

*emine.ozturk.mail@gmail.com

Abstract

The thiosemicarbazones (TSCs) are extensively investigated not only because of the interesting aspects of their binding and structure, but also because of the potential biological activities they acquire due to their ability to chelate metals. TSCs have numerous applications like antitumor, antibacterial, antiviral, antifungal, anticancer and other biological activities. Although there are studies in the literature on the activities of TSC ligands based on thiophenes and their complexes with transition metals, benzothiophene-based TSCs and their metal complexes have not been investigated sufficiently [1-4]. In this sense, TSC compounds and their antimicrobial activities were investigated in this study. A series of thiosemicarbazones (TSCs) (**L$^{1-5}$**) containing benzothiophen moiety were prepared with appropriate thiosemicarbazides. Structures of the newly synthesized compounds were confirmed by elemental analysis and spectral data from FT-IR, ^{1}H NMR, MALDI-TOF and single crystal X-Ray diffraction techniques. These compounds were researched for their antibacterial activity against Gram-positive bacteria (*Enterococcus faecalis* and *Staphylococcus aureus*), Gram-negative bacteria (*Escherichia coli, Pseudomonas aeruginosa* and *Serratia marcescens*) and antifungal activity against a yeast species *Candida albicans*.

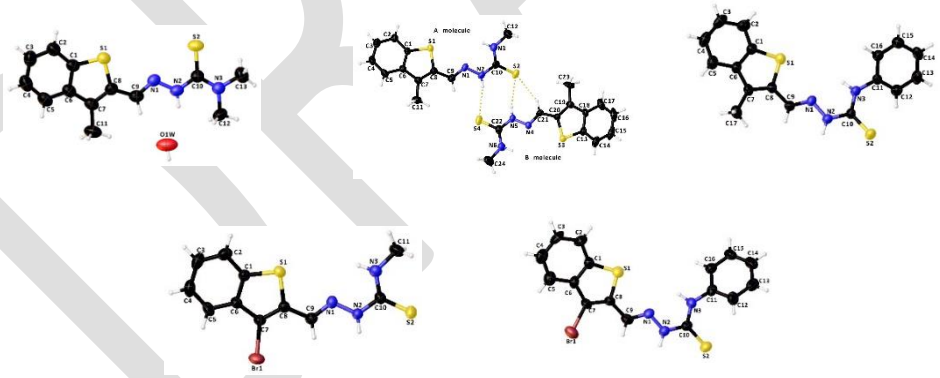


Figure: Single Crystal X-ray Structures of Thiosemicarbazones

Keywords: Benzothiophene, Thiosemicarbazone, Crystal Structure, Antibacterial activity, Antifungal activity

Acknowledgements: This study is supported within the scope of 100/2000 YÖK Doctoral Scholarship Program. The authors thank the Dokuz Eylül University Rectorate Scientific Research Projects Coordination Center for financial support for the 2019.KB.FEN.033 project.

References:

Kayed SF, Farina Y, Simpson J, et al. Inorganic Chemistry Communications. (2022), 110138.
Kayed SF, Farina Y, Simpson J, et al. In Prosiding Seminar Kimia Bersama UKM-ITB VIII. (2009), 9, 11.
Bao B, Bai S, Fan J, et al. Dyes and Pigments. (2009), 171, 107778.
Qiao Y, Che Y, Yu Y, et al, Dyes and Pigments. (2018), 156, 326.

ORAL PRESENTATION

Immobilization of Urease onto Titanium (IV) Oxide Nanoparticles with Different Spacer Arms

Özlem Alptekin[1]* (*ORCID: https://orcid.org/0000-0002-0458-7609*)

[1]Çukurova University, Faculty of Pharmacy, Department of Biochemistry, 01330, Sarıçam, Adana, Turkey

*oalptekin@cu.edu.tr

Abstract

In this study, urease was immobilized onto titanium (IV) oxide (TiO_2) nanoparticles (490 nm) carrier. Firstly, the carrier was modified with Piranha solution to generate terminal hydroxyl groups on the carrier surface (Alptekin, 2022). Then the carrier was treated with 3-aminopropyl triethoxysilane (APTES) solution (Tukel and Alptekin, 2004). After this step carrier was modified with different spacer arms such as glutaraldehyde (CA) Bis(3-aminopropyl)amine (CB), 1,2 Bis(3-aminopropylamino)ethane (CC), polyethyleneimine (CD), Bis(3-aminopropyl)amine and glutaraldehyde (CBA), 1,2 Bis(3-aminopropylamino)ethane and glutaraldehyde (CCA), and polyethyleneimine and glutaraldehyde (CDA). After various modifications were made to TiO_2 nanoparticles, urease was immobilized onto modified carriers. Urease samples immobilized to the surface of carriers CB, CC, and CD by the ionic adsorption method are expressed as IU_{CB}, IU_{CC} and IU_{CD}, and urease samples covalently immobilized to the surfaces of carriers CA, CBA, CCA, and CDA are expressed as IU_{CA}, IU_{CBA}, IU_{CCA}, and IU_{CDA}, respectively. In order to determine the high activity of immobilized urease preparation, immobilization was performed at a concentration of 1 mg/mL urease prepared in 50 mM HEPES buffer (pH 7.0) for 30, 60, 90, and 120 min immobilization time. The percentage of bound urease amount and the relative activities of the immobilized urease preparations were determined depending on the immobilization time.The bound amount of urease was found to be between 90% and 95%. The highest immobilized urease activity was obtained from IU_{CD}. Among all tested carriers, modified with polyethylenimin (CD) is a more suitable spacer arm for urease immobilization.

Keywords: Urease, Immobilization, Titanium (IV) oxide, Polyethyleneimine

References

1. Tükel S. S., Alptekin Ö. (2004). Immobilization and kinetics of catalase onto magnesium silicate. Process Biochemistry, 39(12), 2149-2155.
2. Alptekin Ö. (2022). Characteristics of immobilized urease onto modified zirconium (IV) oxide via glutaraldehyde: kinetic, stability, and operational stabilities in bioreactors. Chemical Papers, 76, 749-761.

This research was supported from Cukurova University Scientific Research Projects Unit (TSA-2022-14281)

DRAFT

ORAL PRESENTATION

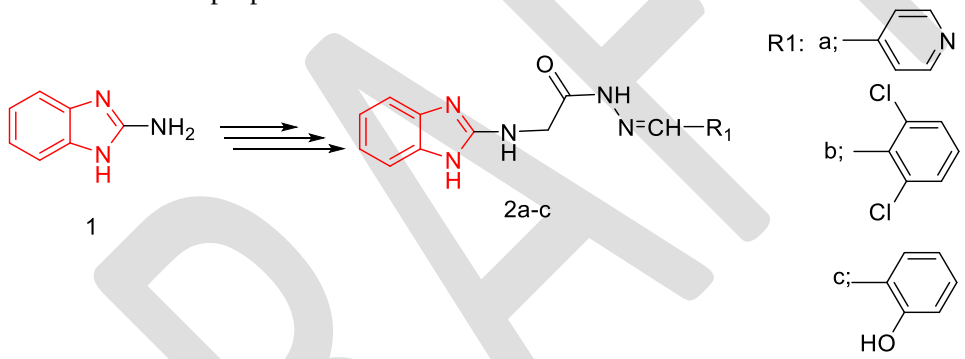
Synthesis of Benzimidazole Derivatives an of Their Anticancer Properties

Serpil Demirci[1*] (*https://orcid.org/0000-0002-6579-4273*)

[*1]Department of Medical Services and Techniques, Vocational High School of Health Services, Giresun University, Giresun, Türkiye

*Corresponding author e-mail: demirciserpil17@gmail.com

Abstract
In this study, new benzimidazole derived Schiff bases were synthesized. The anticancer activities of the synthesized compounds were investigated against HT29 (Human colon adenocarcinoma) and A549 (Human non-small cell lung cancer) cancer cell lines. i*n silico* analyzes of all compounds were performed.
The hybrid anticancer drug approach is an innovative synthetic strategy that involves either the merging or blending of hepatophoric moieties of different drugs in a new molecular structure or combining two or more potential anticancer pharmacophores directly through cleavable/non-cleavable linkages [1,2].
On the other hand, synthetic approaches and technological developments have been progressing, combinatorial chemistry, coupled with traditional chemistry and in silico drug design, is expected to lead to a number of new drug molecules in the future. So all synthesized compounds were confirmed anticancer activities. And we studied this molecules in silico properties.



Scheme 1. Synthesis of compounds 2a-c

Keywords: Benzimidazole, Anticancer activity, Schiff base

Acknowledgments
This study was supported by the Giresun University, Supported by the Scientific Research Projects Coordination Unit as project numbered FEN-BAP-A-240222-28.
I would like to thank Assoc. Prof. Arif Mermer and Ph.D. Tuğba Kul Köprülü for their contributions.

References
[1] S.N. Manjula, N. Malleshappa Noolvi, K. Vipan Parihar, S.A. Manohara Reddy, V. Ramani, A.K. Gadad, G. Singh, N. Gopalan Kutty, C. Mallikarjuna Rao, Synthesis and antitumor activity of optically active thiourea and their 2-aminobenzothiazole derivatives: A novel class of anticancer agents, European Journal of Medicinal Chemistry. 44 (2009) 2923–2929. doi:10.1016/j.ejmech.2008.12.002.
[2] J. Liu, S. Yang, X. Li, H. Fan, P. Bhadury, W. Xu, J. Wu, Z. Wang, Synthesis and Antiviral Bioactivity of Chiral Thioureas Containing Leucine and Phosphonate Moieties, Molecules. 15 (2010) 5112–5123.

DRAFT

ORAL PRESENTATION

Synthesis of Benzimidazole Derivatives as New Anticancer Compounds

Serpil Demirci[1*] (*https://orcid.org/0000-0002-6579-4273*)

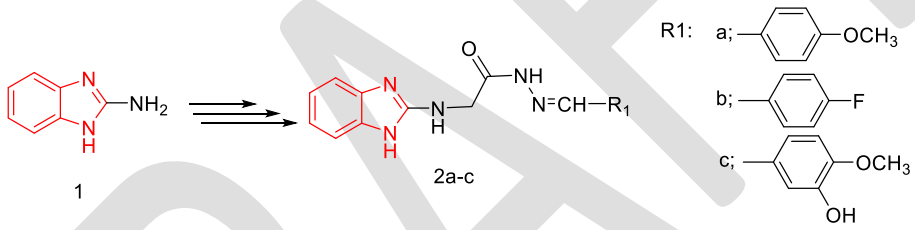
[*1]Department of Medical Services and Techniques, Vocational High School of Health Services, Giresun University, Giresun, Türkiye.

*Corresponding author e-mail: demirciserpil17@gmail.com

Abstract

In this study, novel benzimidazole-Schiff base derivatives starting from the commercially available 1*H*-benzo[d]imidazole-2-amine were synthesized. The anticancer activities of the synthesized compounds were investigated against HT29 (Human colon adenocarcinoma) and A549 (Human non-small cell lung cancer) cancer cell lines using xCELLigence real-time cell analysis. Additionaly, all compounds demonstrated favorable pharmacokinetics properties compared to Paclitaxel (reference compound).

Our approach in this study was to combine benzimidazole ring and *N*-acylhydrazone moiety, which are two important pharmacophores in medicinal chemistry. For this purpose, benzimidazole was converted to ester and hydrazide compounds. The anticancer activities of the synthesized molecules were investigated against HT29 and A549 cancer cell lines. all compounds demonstrated favorable pharmacokinetics properties compared to Paclitaxel (reference compound) [1,2]



Scheme 1. Synthesis of compounds 4a-c

Keywords: Benzimidazole, Anticancer activity, Schiff base

Acknowledgments

This study was supported by the Giresun University, Supported by the Scientific Research Projects Coordination Unit as project numbered FEN-BAP-A-240222-28. I would like to thank Assoc. Prof. Arif Mermer and Ph.D. Tuğba Kul Köprülü for their contributions.

References

[1] A. Sudhakar, History of Cancer, Ancient and Modern Treatment Methods, Journal of Cancer Science & Therapy. 01 (2009) i–iv. doi:10.4172/1948-5956.100000e2.

[2] J.B. Wright, The Chemistry of the Benzimidazoles., Chemical Reviews. 48 (1951) 397–541. doi:10.1021/cr60151a002.

DRAFT

ORAL PRESENTATION

Sublethal exposure to aclonifen ınduces alteration in biochemical indices in serum of juvenile *Oncorhynchus mykiss*

Reham AL HORANI[1*] (ORCID:0000-0002-6222-3918), Demet DOGAN[2] (ORCID: 0000-0001-6356-0679)

[1*]Department of Biochemistry Science and Technology, Graduate School of Natural and Applied Sciences, Gaziantep University, Turkey.
[2]Faculty of Arts and Sciences, Department of Biology, Gaziantep University, Turkey.

*Corresponding author e-mail: Rehamalhorani8@gmail.com

Abstract

Aclonifen, diphenylether herbicide, is used in the pre-emergence control of broad leaves and grass weeds throughout the world. Herbicides are introduced into aquatic environment during direct spraying or later through rainfall and air drift and pose thret to non-target organisms. Therefore this investigation was conducted to evaluate the toxic effects of aclonifen on oxidative stress biomarkers and biochemical indices of serum in juvenile *Oncorhynchus mykiss* following 24, 48, 72 and 96 hours of application to sublethal concentrations of 12.7, 63.5 and 127 µg/L. There was no visible change in protein level and ALP activity. Pesticide treatment caused prominent elevation in the levels of serum glucose level showing increase in liver glycogenolysis, the conversion of glycogen stored in hepatocytes to glucose, in order to compansate the high energy demand to handle the stresful situation. Marked induction was observed in the activities of ALT and AST reflecting hepatotoxic potential of aclonifen. Also determined decline in cholesterol level evidences the involvement of aclonifen in lipid metabolism. Considering the changes, it can be concluded that aclonifen has hepatotoxic potential and the application resulted in adaptive responses in juvenile *O. mykiss* as manifested in altered levels of glucose and cholesterol.

Keywords: Herbicide, Aclonefin, *O. mykiss*, biochemical indices

Acknowledgment: The authors would like to thank Gaziantep University Research Fund for financial assistance (Project Number: FEF.YLT.22.13).

ORAL PRESENTATION

Lipid peroxidation promoted inhibition of membrane-bound enzymes in tissues of juvenile *Oncorhynchus mykiss* following spirotetramate application

Esra COLAK[1*](ORCHID:0000-0002-4880-4169), Demet DOGAN[2] (0000-0001-6356-0679)

[1*]Department of Biology, Graduate School of Natural and Applied Sciences, Gaziantep University, Turkey.
[2]Faculty of Arts and Sciences, Department of Biology, Gaziantep University, Turkey.

*Corresponding author e-mail: Esra.colak1998@gmail.com

Abstract

Spirotetramat, an acetyl-CoA carboxylase inhibitör, is an insecticide used to increase crop productivity in agriculture. Data related to its sublethal effects on *Oncorhynchus mykiss* is unavailable. Therefore this study was performed to evaluate toxic effects of spirotetramat on brain, muscle, gill and kidney of juvenile *O. mykiss* following 24, 48, 72 and 96h of exposure to 0.0254, 0.0508 and 0.254 mg/L concentrations. Insecticide treatment caused marked decrease in AChE activity in muscle while there was no significant change in brain tissue. A remarkable decline in Na^+, K^+-ATPase activity was observed for gill and kidney tissues. An ascending trend was observed in TBARS levels in all tissues being more pronounced at high concentrations with prolonged exposure. Statistically significant and negative relationship was confirmed between TBARS, AChE and Na^+, K^+-ATPase in all tissues except for brain. The findings clearly indicates the anticholinesterase activity of spirotetramat and its disruptive effect on osmoregulation as demonstrated with the inhibition of AChE and Na+-K-ATPase, respectively. The direct action of spirotetramat and/or lipid peroxidation triggered loss of membrane integrity may be suggested as underlying mechanisms of this effect. Considering the ecologically relevant consequences of these markers on behavior, growth, reproduction and survival, outcomes of this investigation presents the risk spirotetramat may pose at ecosystem by causing adverse effects at population level.

Keywords: Spirotetramat, *O. mykiss,* neurotoxicity, osmoregulation and lipid peroxidation

Acknowledgements: This research is funded by The Scientific and Technological Research Council of Turkey (TUBITAK) under the project number of 122Z844. The study is also granted infrastructure project support from Gaziantep University Research Fund (Project Number: FEF.ALT.22.25).

ORAL PRESENTATION

In silico studies on possible Topoisomerase IIα inhibitors

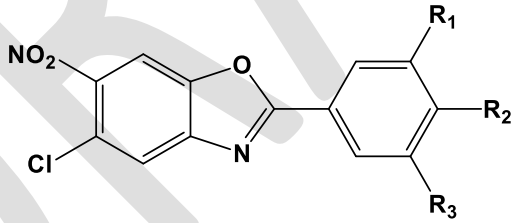
İrem Nur Çelik Civelek[1,2*] (*https://orcid.org/0000-0003-0213-3635*), İlkay Yıldız[2] (*https://orcid.org/0001-9526-0232*), [3]Serap Yılmaz Özgüven[3] *(https://orcid.org/0000-0002-6458-2658*)

[*1]Ankara University, Graduate School of Health Sciences, Ankara, Türkiye
[2] Ankara University, Faculty of Pharmacy, Department of Pharmaceutical Chemsitry Ankara, Türkiye
[3]Trakya University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, Edirne, Turkey

*Corresponding author e-mail: irmnur@hotmail.com

Abstract

Cancer is a group of fatal diseases characterized by the proliferation of cells that do not have a normal structure beyond their limits due to disruption of the cell cycle. According to World Health Organization data, it is expected that there will be 28.9 million cancer cases in 20 years. There are active pharmaceutical ingredients against many types of cancer, but most of them have side effects that limit their use. For this reason, the need for targeted treatment with minimal side effects at minimum doses is increasing day by day[1,2,3,4,5]. DNA toposomerases are one of the most important enzyme groups that manage topological problems in DNA structure. Inhibitors have been identified that act at several different steps of the topoisomerase II catalytic cycle, including DNA binding, ATP binding, ATP hydrolysis, and DNA cleavage. Etoposide is a podophyllotoxin derivative that inhibits DNA topoisomerase II, resulting in DNA chain breaks and induction of cytotoxic and apoptotic cell death[6]. It is used as a reference inhibitor compound into *in silico* studies of the molecule to be analyzed in many literatures. The benzoxazole ring is one of the important heterocyclic ring systems in drug design due to its structural similarity to adenine and guanine bases. Today, there are drugs that are derivatives of benzoxazole and its analogues and are actively used in treatment for various purposes. In our study, we examined *in silico* molecular modeling data and ADME/Tox properties of new 2-substituted phenyl-5-chloro-6-nitro-benzo[d]oxazole derivatives targeting the topoisomerase IIα (Topo IIα) enzyme. It is predicted that all of the compounds we designed will have good pharmacokinetic properties and therefore good bioavailability.



R_1: -H, -F, -OCH_3; **R_2:** n-C_4H_9, n-C_5H_{11}, n-C_6H_{13}; **R_3:** -H, -OCH_3

Figure. General formula of the designed benzoxazole compounds

Keywords: Benzoxazole, Drug Design, Molecular Docking, Anticancer, Topoisomerase IIα

Acknowledgments: We thank TÜSEB (Helath Institutes of Türkiye) for supporting this research as a 2022B02-22566 project.

Resources:

Andoh T, & Ishida R (1998) Catalytic inhibitors of DNA topoisomerase II, Biochim. Biophys. Acta 1400, 155–171.
Larsen AK, Escargueil AE, and Skladanowski A (2003) Catalytic topoisomerase II inhibitors in cancer therapy, Pharmacol. Ther 99, 167–181.
Fortune, J. M., & Osheroff, N. (1998). Merbarone inhibits the catalytic activity of human topoisomerase IIα by blocking DNA cleavage. Journal of Biological Chemistry, 273(28), 17643–17650. https://doi.org/10.1074/jbc.273.28.17643
Fortune, J. M., & Osheroff, N. (2000). Topoisomerase II as a Target for Anticancer Drugs: When Enzymes Stop Being Nice. Prog. Nucleic Acid Res. Mol. Biol 64, 221–253.
Minniti, E., Byl, J. A. W., Riccardi, L., Sissi, C., Rosini, M., de Vivo, M., Minarini, A., & Osheroff, N. (2017). Novel xanthone-polyamine conjugates as catalytic inhibitors of human topoisomerase IIα. Bioorganic and Medicinal Chemistry Letters, 27(20), 4687–4693. https://doi.org/10.1016/j.bmcl.2017.09.011
Montgomery, B., & Lin, D. W. (2009). Toxicities of chemotherapy for genitourinary malignancies. In Complications of Urologic Surgery: Expert Consult (pp. 117–123). Elsevier. https://doi.org/10.1016/b978-1-4160-4572-4.00010-8

DRAFT

ORAL PRESENTATION

Consumption of High-Fructose Corn Syrup Delays Puberty in Female Rats

Elif Dide Polat[*1] (*https://orcid.org/0009-0008-3025-2977*), Muhammed Enes Canakci[1] (*https://orcid.org/0009-0009-0708-8563*), Muhammed İkbal Bilik[1] (*https://orcid.org/0009-0000-4068-6367*), Mert Ali Horoz[2] (*https://orcid.org/0000-0002-4208-9512*), Ramazan Fazil Akkoc[2] (*https://orcid.org/0000-0002-0559-8932*), Ihsan Serhatlioglu[1] (*https://orcid.org/0000-0002-2384-7971*)

[1] Firat University, Faculty of Medicine, Department of Biophysics, Elazig, Turkey.
[2] Firat University, Faculty of Medicine, Department of Anatomy, Elazig, Turkey.

*Corresponding author e-mail: elifdidepolat@gmail.com

Abstract

High-fructose corn syrup (HFCS) consumption, which has increased, especially among children and adolescents, is recognized as a major contributor to the development of metabolic diseases. However, the effects of a fructose-rich diet during the prepubertal period on pubertal maturation in female rats have not been sufficiently investigated. The aim of this study was to investigate the effects of HFCS consumption on pubertal maturation. Sprague-Dawley female rats were used in the study. Rats were selected to have an average body weight of 35 ± 2 grams on the 21st day after weaning. Rats were divided into control and HFCS-55 groups (n = 10 in each group). Daily measurements of food and liquid intake, as well as body weights, were recorded. Vaginal opening was monitored daily, starting on the 30th day to determine the onset of puberty in rats from both the HFCS and control groups. Estrogen levels in serum samples after decapitation were measured using the ELISA method. When comparing changes in body weight, no significant difference was observed in the HFCS group. However, an increase in liquid consumption and a significant decrease in food consumption were observed in the HFCS group ($p<0.001$). When comparing the onset of puberty between the control and HFCS groups, it was found that HFCS application significantly delayed the onset of puberty ($p<0.05$), although it did not affect pubertal body weights. HFCS application was found to cause a non-significant increase in serum estrogen levels in female rats. Our findings suggest that HFCS consumption in prepubertal female rats is responsible for abnormalities that delay the onset of adolescence. Additional research is required to determine the underlying mechanismes via which HCFS influences the process of pubertal development.

Keywords: Fructose, female rat, puberty

Acknowledgment: This study was supported by TUBITAK (Project #1919B012111223).

DRAFT

ORAL PRESENTATION

Production, characterization and antioxidant activity evaluation of green synthesized iron oxide nanoparticles

Betül Şura Akalın[1*] (https://orcid.org/0009-0003-5244-050X) & Şevval Kızılkaya[1*] (https://orcid.org/0009-0000-5769-5382), Seçil Kaya[1] (*https://orcid.org/0000-0001-6614-6641*), Buşra Akgül[1] (*https://orcid.org/0000-0002-3566-8874*), Emrah Şefik Abamor[1] (https://orcid.org/0000-0002-9174-4528) Serap Derman[1] (*https://orcid.org/0000-0002-6662-6642*)

[*1]Yıldız Technical University, Faculty of Chemical and Metallurgical Engineering, Department of Bioengineering, Istanbul, Turkey.

*Corresponding author e-mail: betulsuraakalin@gmail.com / kizilkaya.sevval@gmail.com

Abstract

Nanotechnology is the efficient production of materials by controlling matter at the nanometer scale and taking advantage of new properties and phenomena developed at this scale. Today, the production of nanoparticles has been investigated for use in the effective treatment and diagnosis of many diseases, including cancer. Metallic nanoparticles are one of the easiest protocols to process and synthesize due to metal oxides found in large amounts in nature. Iron nanoparticles, which are member of metallic nanoparticles, are preferred due to their suitable electrical, magnetic, and optical properties, their ability to have various shapes and structures, and their toxicity limitations. The green synthesis method, which is environmentally friendly, cost-effective and ensures that nanoparticles remain stable in solution, has become a preferred method in recent years. Contained in plants; Substances such as phenolic acid, flavonoid, tannin, terpene, coumarin, lycopene, vitamin, carotenoid, and anthocyanin are natural agents that can reduce metal ions. In this study, iron oxide (Fe_2O_3) nanoparticles were synthesized by green synthesis, a biological method, using plant extract, which was discovered to have anti-inflammatory, antimicrobial, antioxidant and anticancer properties. The produced nanoparticles were physically and chemically characterized by Fourier transform infrared spectrophotometry, UV-Vis spectrophotometry and Zetasizer. Antioxidant activities were determined using the 2,2-Diphenyl-1-picrylhydrazyl (DPPH) method. As a result, it was confirmed that the iron nanoparticles were successfully synthesized with the color change during synthesis and the peaks obtained in FT-IR and UV-Vis spectrophotometers. In the continuation of the study, it is planned to use the cancer cell line and evaluate its cytotoxic activity on these cancer cells by the MTT method. It is aimed to develop the data obtained for use in cancer treatment.

Keywords: Nanotechnology, green synthesis, metal nanoparticles, iron nanoparticles, antioxidant activity

Acknowledgements: We appreciate the Scientific and Technological Research Council of Türkiye (TÜBİTAK) for 2209-B Undergraduate Research Project for Industry support, Assoc. Prof. Ömer Tahir Günkara for industrial consulting and Res. Asst. Kübra Sayarım for her valuable assistance during the project's writing phase.

DRAFT

ORAL PRESENTATION

Production of wound dressings including the metal complex of phenylboronic acid-functionalized 4,5-diazafluorene

Seçil Kaya[1*] (*https://orcid.org/0000-0001-6614-6641*), Caner Cebeci[2] (*https://orcid.org/0000-0002-9808-8279*), Hande Hançer[3] (*https://orcid.org/0000-0003-1971-1454*), Buşra Akgül[1] (*https://orcid.org/0000-0002-3566-8874*), Emrah Şefik Abamor[1] (*https://orcid.org/0000-0002-9174-4528)*, Tülin Özbek[3] (*https://orcid.org/0000-0001-6858-7045*), İbrahim Erden[2] (*https://orcid.org/0000-0002-3145-1280)*, Serap Derman[1] (*https://orcid.org/0000-0002-6662-6642*)

[*1]Yıldız Technical University, Faculty of Chemical and Metallurgical Engineering, Department of Bioengineering, Istanbul, Turkey.
[2]Yıldız Technical University, Faculty of Arts and Sciences, Department of Chemistry, Istanbul, Turkey.
[3]Yıldız Technical University, Faculty of Arts and Sciences, Department of Molecular Biology and Genetics, Istanbul, Turkey.

*Corresponding author e-mail: kayasecil@outlook.com.tr

Abstract

Wound healing is complex process consisting of hemostasis, inflammation, proliferation and re-modelling. Recently, innovative approaches in the field of wound care have led to the development of nanofiber-based wound dressings. Additionally, boronic acid based active substances are increasingly being utilized in wound dressings[1-2].
In this study, metal complexes were synthesized with a phenylboronic acid-functionalized 4,5-diazafluorene ligand[3]. The ligand and its metal complexes were characterized utilizing Fourier-transform infrared, mass, and UV-Vis spectrophotometry. Their free radical scavenging activities were determined using the 2,2-Diphenyl-1-picrylhydrazyl (DPPH) method. Then, the ligand and its metal complexes were loaded onto polymeric electrospun membranes. Experiments were carried out to determine the loading efficiency, release rates, and swelling properties of the wound dressing materials[3]. As a result, the successful synthesis of the ligand and its metal complexes was confirmed through spectroscopic analyses. It was observed that the formation of complexes between the ligand and metals increased antioxidant activity. In the release study, it was found that the substances were released slowly from the nanofibers over a period of 11-14 days. And, the swelling study demonstrates that the produced wound dressing may offer advantages in terms of usage for highly exudative wounds. Thus, it is believed that an antioxidant, and high-swelling capacity drug delivery system has been developed. In future studies, a detailed characterization of the active substance-loaded nanofibers is planned.

Keywords: wound dressing, electrospinning, 4,5-diazafluorene, boronic acid, metal complexes

Acknowledgements: We appreciate the Council of Higher Education (YÖK) for the 100/2000 Ph.D. scholarship, the Scientific and Technological Research Council of Türkiye (TÜBİTAK) for the BİDEB 2211-C scholarship and Health Institutes of Türkiye (TUSEB) for project support (project no: 21307).

References:

1) Ambekar, R. S., & Kandasubramanian, B. (2019). Advancements in nanofibers for wound dressing: A review. European Polymer Journal, 117, 304-336.
2) Dalisson, B., & Barralet, J. (2019). Bioinorganics and wound healing. Advanced Healthcare Materials, 8(18), 1900764.
3) Cebeci, C., Arslan, B. S., Güzel, E., Nebioğlu, M., Şişman, İ., & Erden, İ. (2021). 4, 5-Diazafluorene ligands and their ruthenium (II) complexes with boronic acid and catechol anchoring groups: design, synthesis and dye-sensitized solar cell applications. Journal of Coordination Chemistry, 74(9-10), 1366-1381.

DRAFT

ORAL PRESENTATION

The effect of hydroglycerolic extraction on biofunctional characteristics of yarrow (*Achillea millefolium* L.) flowers: An optimization study using simplex lattice mixture design approach

Beyza ÇİFTCİ[1*] (*ORCID: https://orcid.org/0000-0002-3080-0880)*, Kevser KARAMAN[2] (*ORCID: https://orcid.org/0000-0003-0729-6185*), Ali İrfan İLBAŞ[3] (*ORCID: https://orcid.org/0000-0001-9640-5237*), Mahmut KAPLAN[3] (*ORCID: https://orcid.org/0000-0002-6717-4115*)

[*1]Erciyes University, Graduate School of Natural and Applied Sciences, Department of Field Crops, Kayseri, Türkiye.
[2]Erciyes Unıversıty, Faculty of Agriculture, Department of Agricultural Biotechnology, Kayseri, Türkiye.
[3]Erciyes Unıversıty, Faculty of Agriculture, Department of Field Crops, Kayseri, Türkiye.

*Corresponding author e-mail: beyzacftc.58@gmail.com

Abstract

Yarrow is a flowering plant in the family of Asteraceae and it is considered one of the oldest medicinal plants. Yarrow extracts are used as a remedy for some diseases in phototherapy because of its high bioactivity. It contains many bioactive constituents and it shows high antioxidant capacity. In this research, hydroglycerolic extraction was performed to produce green extract and its some bioactive properties were characterized. For this purpose, three different solvents namely ethanol, glycerol and water were used in the extraction and simplex lattice mixture design was used to determine the effects of the sole or mixed solvent effects on the studied parameters such as total phenolic and flavonoid, antiradical and antioxidant activities. Total phenolic content of sole water, ethanol and glycerol extracts was determined as 12.81, 2.92 and 9.17 mg GAE/g, respectively while the total flavonoid content ranged between 3.76-19.35 mg QE/g. The linear effects of the used solvent were determined to be significant ($p<0.05$) on both total phenolic and flavonoid contents. The constructed regression equations for total phenolic and flavonoid contents showed good fitting ability with quite high coefficient of determination ($R2>0.850$). The solvent effects on studied antioxidant and antiradical activities were also significant. The optimization procedure of the mixture design revealed that the highest total phenolic content (19.64 mg GAE/g) would be obtained by the extraction of water, ethanol and glycerol mixture at 52.58, 6.35 and 41.04% while the minimum levels would be observed by sole ethanol (2.42 mg GAE/g). The recorded results showed that the yarrow could be extracted by the mixture of water and glycerol and hydroglycerolic extraction of yarrow samples showed superior bioactive properties.

Keywords: yarrow, glycerol, bioactivity, simplex lattice mixture design, optimization

Note: This study has been supported by Erciyes University Scientific Research Projects Unit with FBA-2020-10122 code.

DRAFT

ORAL PRESENTATION

Lyophilization of *Dicentrarchus labrax* and Investigation of the Effect of Ultrasonic Pretreatment

Merve Alp[1*] (0009-0002-9849-9825), Zehra Ozden Ozyalcin [1] (0000-0002-6662-5885), Azmi Seyhun Kipcak[1] (0000-0003-2068-6065)

[*1]Yıldız Technical University, Faculty of Chemical and Metallurgical, Chemical Engineering Department, Istanbul, Türkiye.

*Corresponding author e-mail: mervealp834@gmail.com

Abstract

It is known that seafood contains high protein, as well as various vitamins, minerals and healthy fats. Like many sea creatures, fish contain high levels of moisture. Problems such as microbial spoilage, taste and color changes may occur due to high moisture content until the fish are ready for consumption. For these reasons, various preservation methods are being developed in order to ensure a long shelf life, preserve its quality, taste and nutritional values, and deliver it to the consumer more safely. With the freeze drying method, drying occurs by direct sublimation of the moisture in the food. With this method, color change, loss of nutritional content and deformities in foods are prevented. In this study, sea bass (*Dicentrarchus labrax*) was dried by the lyophilization (freeze drying) method. Approximately 5 grams of sea bass samples, 1.5 cm thick, 1 cm long and 1 cm wide, were examined without pre-treatment and by applying ultrasonic pre-treatment, and the effect of pre-treatment on the drying behavior was examined. Sea bass, which contains approximately 70% moisture, was dried until it reached 5% moisture content. The drying process continued for 7 hours to ensure the desired humidity rate and weight measurements were taken every hour. Before starting the drying process, the moisture content for sea bass was calculated as 2.1778 kg water/kg dry matter. The lyophilization process continued until the samples reached a content of 0.0767 kg water/kg dry matter with unpretreated samples, 0.1354 kg water/kg dry matter with 1 minute ultrasonic pretreatment and 0.2034 kg water/kg dry matter with 5 minutes ultrasonic pretreatment. As a result of the data obtained, effective moisture diffusions were calculated as 3.90×10^{-10}, 3.32×10^{-10} and 2.99×10^{-10} m^2/s for unpretreatment, 1 min ultrasonic and 5 min ultrasonic, respectively.

Keywords: *Dicentrarchus labrax*, Drying, Freeze Drying, Effective Moisture Diffusion, Ultrasonic Pretreatment, Moisture Ratio

Acknowlegment
This study was supported by the project number FYL-2023-5828 of Yıldız Technical University Scientific Research Project Coordination.

DRAFT

ORAL PRESENTATION

Drying of Blue Mussels by Freeze Drying Technique and Investigating the Effect of Ultrasonic Pretreatment

Nurgul Alp[1*] (0009-0002-1528-7836), Zehra Ozden Ozyalcin [1] (0000-0002-6662-5885), Azmi Seyhun Kipcak[1] (0000-0003-2068-6065)

[*1]Yıldız Technical University, Faculty of Chemical and Metallurgical Engineering, Chemical Engineering Department, İstanbul, Türkiye.

*Corresponding author e-mail: alp.nurgul139@gmail.com

Abstract

Seafood has become an important part of a balanced and healthy diet with its diverse nutritional content such as high-quality proteins, omega-3 fatty acids, vitamins and minerals. They can deteriorate very easily due to the moisture they contain. For this reason, it is necessary to obtain quality products by extending the shelf life through various processes while preserving them. In recent years, studies on drying foods to ensure long shelf life and preserve nutritional values have been increasing. The drying process is carried out by removing the moisture contained in the food by using drying methods. In the freeze drying technique, the moisture in the frozen product is removed from the food by sublimation. In this study, blue mussels were dried with modern freeze drying (lyophilization) technique. Samples were prepared unpretreated and with ultrasonic pretreatment of 1 minute and 5 minutes, and the effect of pretreatment on the drying rate was examined. Samples with a moisture content of approximately 74% were subjected to freeze drying until a moisture content of 7% was reached. Before starting the drying process, the moisture content was calculated as 2.5577 kg water / kg dry matter for mussels. Samples were freeze-dried to a content of 0.1700 kg water/kg dry matter without a pretreatment, 0.1956 kg water/kg dry matter with 1 min ultrasonic pre-treatment, and 0.2069 kg water/kg dry matter with 5 min ultrasonic pre-treatment. The drying process was carried out in 420 minutes. Effective moisture diffusions were calculated as 3.79×10^{-10}, 3.72×10^{-10} and 3.72×10^{-10} m^2/s for unpretreated samples, 1 min ultrasonic and 5 min ultrasonic pretreatment, respectively.

Keywords: blue mussel, drying, lyophilization, effective moisture diffusion, ultrasonic process, drying rate

Acknowlegment
This study was supported by the project number FYL-2023-5830 of Yıldız Technical University Scientific Research Project Coordination.

DRAFT

ORAL PRESENTATION

Comparative Growth of Green Basil and Purple Basil in Deep Water Culture vs. Peat

Kübra Tutkun Akdeniz[1*] (*ORCID: https://orcid.org/0000-0002-6505-3555*),
Bahar Yıldız Kutman[1*] (*ORCID: https://orcid.org/0000-0002-7891-7527*.)

[*1]Gebze Technical University, Institute of Biotechnology, Kocaeli, Turkiye.

*Corresponding author e-mail: bykutman@gtu.edu.tr

Abstract

Basil (*Ocimum* L.) is a diverse genus in the Lamiaceae family, grown in home gardens as well as commercial farms. Next to lettuce, basil is one of the most commonly grown high-value leafy greens in soilless greenhouses and indoor farms. In addition to its use as a spice in both fresh and dry form, it serves as a source of essential oils for food, cosmetics, and pharmaceutical sectors. As a medicinal herb, it is prized for its rich essential oils and phenolic compounds. Phenolic acids in basil are valued for their potential to combat various health issues, including cardiovascular diseases. In this study, green and purple basil plants were grown for six weeks in peat or deep water culture under the same artificial light by using the Vahaa Smart Garden system. To assess the nutritional quality various mineral and metabolite concentrations were determined by ICP-OES and UV-Vis spectrophotometer. The fresh weights of green and purple basil grown in deep water culture were higher than those grown in peat, by approximately 100%. Chlorophyll and carotenoid concentrations of plants grown in Vahaa system were significantly higher than those grown in peat. Vitamin C concentrations of plants grown in deep water culture were also enhanced when compared to their peat grown counterparts. Protein concentration, antioxidant capacity, and total phenolic content of green and purple basils grown in deep water culture were dramatically higher than those grown in peat. Based on these results, it is documented that deep water culture can be a good growing technique both for high yielding and high-quality basil production.

Keywords: basil, purple basil, peat, nutritional quality, hydroponic, smart garden, deep water culture, soilless agriculture

This study was conducted as part of a university-industry partnership project between VAHAA DİKEY TARIM ÇÖZÜMLERİ VE TEKNOLOJİ A.Ş. and Gebze Technical University. This study was funded by The Scientific and Technological Research Council of Turkey (TUBITAK Project number: 130614).

DRAFT

ORAL PRESENTATION

Evaluation of biochar obtained via gasification of oak wood waste as a soil amendment and plant growth promoter

Elif Özlem Günçaldı[1,2] *(https://orcid.org/0000-0002-5165-0703)*,
Ümit Barış KUTMAN[1] *(https://orcid.org/0000-0002-9158-0332)*

[*1] Institute of Biotechnology, Gebze Technical University, Kocaeli, Turkey.
[2]GÜBRETAŞ R&D Center, Kocaeli, Turkey.

*eoguncaldi2018@gtu.edu.tr

Abstract

Biochar, a carbon-rich byproduct of pyrolysis, is recently attracting more interest in the context of sustainable agriculture due to its potential to improve soil quality and promote plant growth. It has been used in agriculture to manage soil acidity, improve soil fertility, increase organic matter and water availability, and enhance crop yield. The aim of this study was to investigate the effect of biochar application on the growth and nutrition of alfalfa (*Medicago sativa* cv. Sunter) and lettuce (*Lactuca sativa* var. *crispa* cv. Levistro). The biochar used to conduct the experiments was a byproduct of energy generation via the gasification process of waste oak wood biomass that was unsuitable for further uses. In the experiments, plants were grown in two different soils with different pH levels (acidic: 5.0; alkaline: 8.0) and two levels of biochar were applied in the form of powder or granules. The results of the study demonstrated that particle size and soil pH had a significant impact on how plants responded to growth stimuli. In acidic soil, biochar treatment in powder form significantly increased lettuce yield. In both soils, the highest alfalfa yield was obtained in soils treated with the higher level of biochar in powder form. Alfalfa and lettuce both had notable changes in their macro- and micronutrient concentrations. The results of these studies suggested that using biochar obtained as a byproduct of evaluating waste biomass as a feedstock for energy generation has considerable promise in terms of improving barren soils and enhancing agricultural yield in a sustainable manner.

Keywords: Biochar, plant growth promoter, lettuce, alfalfa, soil pH, plant nutrition.

This study was supported by TÜBİTAK 2244 - Industrial PhD Fellowship Program (Project number: 119C030) and GÜBRETAŞ.

DRAFT

ORAL PRESENTATION

Biodegradation of Microplastics with *Aspergillus flavus* and *Aspergillus versicolor*

Burcu Şimşek Uygun[1] (*ORCID: https://orcid.org/0000-0003-1211-4198*)
Semra Malkoç[1,2*] (*ORCID: https://orcid.org/0000-0002-8092-411X)*

[1] Eskişehir Technical University, Faculty of Engineering, Department of Environmental Engineering, Eskisehir, Turkey

[2] Eskişehir Technical University, Environmental Research Center (ÇEVMER), Eskişehir, Turkey

*Corresponding author e-mail: satik@eskisehir.edu.tr

Abstract

Microplastics (MPs) have indeed become a significant concern due to their widespread presence and potential adverse effects on both the environment and human health. This review sheds light on the critical aspects of MPs, including their sources, migration behavior, and the potential of bioremediation as an effective strategy for their removal. Microplastics originate from various sources, including fragmentation of larger plastics, microbeads in personal care products, fibers from textiles, industrial pellets, and microplastic-containing products. These sources contribute to the ubiquity of microplastics in terrestrial and aquatic environments. Microplastics originate from various sources, including fragmentation of larger plastics, microbeads in personal care products, fibers from textiles, industrial pellets, and microplastic-containing products. These sources contribute to the ubiquity of microplastics in terrestrial and aquatic environments.
In this study, the biological degradation process of *Aspergillus flavus* and *Aspergillus versicolor* fungi has been observed for three different types of microplastics, namely Polypropylene (PP), Polyethylene (PE), and Polystyrene (PS). As a result of the experiments, removal efficiencies of *A. flavus* and *A. versicolor* were calculated. Based on the data from the 10th week of using two different types of fungi, it was observed that *A. flavus* has a removal efficiency of 18.33% for PE, 6.83% for PP, and 1.90% for PS. *A. versicolor*, on the other hand, provided removal efficiencies of 6.68% for PE, 5.11% for PP, and 3.25% for PS. It was determined that *A. flavus* and *A. versicolor* were most effective in biodegrading microplastic polyethylene and least effective in microplastic Polystyrene.

Keywords: *Aspergillus versicolor, Aspergillus flavus,* Biodegradation, Polyethylene, Polystyrene, Polypropylene.

Acknowledgements
This work was financially supported by the Unit of the Scientific Research Projects of Eskişehir Technical University under grant no. [22ADP090].

DRAFT

ORAL PRESENTATION

Size-dependent potential toxicological effects of iron oxide nanoparticles (Fe_3O_4 NPs)

Azada Aliyeva[1] (ORCID: https://orcid.org/0009-0007-8592-7727), Ahmet Enes Bulut[1](ORCID: https://orcid.org/0009-0007-8805-3164), Buket Bakan[1*] (ORCID: https://orcid.org/0000-0002-4400-6776),

[*1]University of Atatürk, Faculty of Science, Department of Molecular Biology and Genetics, Erzurum, Türkiye

E-mail: *buketbakan@gmail.com

Abstract

In recent years, researchers have focused on nanomaterials in order to increase their widespread use mostly in medicine, imaging systems and medical fields for diagnostic and treatment of diseases. Nanoparticles can be designed by modified their physico-chemical properties depending on application areas. Due to their unique characteristic features such as size, morphology and large surface areas, it allows easy penetration through tissues and cells which requires a potential risk assessment of these materials. Iron Oxide nanoparticles (Fe_3O_4 NPs) have attracted attention as vaccine adjuvants, as well as their common use, especially in the biomedical field.

The aim of the study is to investigate hemolytic activity and irritation effects of iron oxide nanoparticles (Fe_3O_4 NPs) with dimensions of 18-28 nm and 50-100 nm by hemolysis and HET-CAM tests. As a result of the assays, there is no hemolytic activity in applied doses of different sizes iron oxide nanoparticles and also according to evaluation in terms of hemorrhage, lysis and coagulation parameters, no irritant effects were observed in both size ranges. Although this is a preliminary study shows that the particle does not have a toxic effect in both size ranges, its size-dependent mechanism needs to be elucidated with further in vitro tests.

Keywords: iron oxide nanoparticles (Fe_3O_4 NPs), hemolysis, HET-CAM

Acknowledgements: This study was supported by TUBITAK 2209-A - Research Project Support Programme for Undergraduate Students.

DRAFT

ORAL PRESENTATION

Determination of the contents of naringin, myricetin and rutin in *Centaurea kurdica* Reichardt (Asteraceae) by HPLC

Umut Çelikoğlu[1*] (*ORCID: https://orcid.org/0000-0003-0995-8154*),
Alevcan Kaplan[2] (*ORCID: https://orcid.org/0000-0001-6738-7527*)

[*1]Amasya University, Faculty of Arts and Sciences, Department of Chemistry, Amasya, Turkey
[2]Batman University, Sason Vocational School, Department of Plant and Animal Production, Batman, Turkey

*Corresponding author e-mail: umut.celikoglu@amasya.edu.tr

Abstract

Phenolics are a group of biologically active compounds containing one or more hydroxyl groups and one or more aromatic rings. Phenolic compounds and their subclasses are found in many plant species and belong to the class of secondary metabolites involved in defense against ultraviolet radiation or pathogens. HPLC is widely used for the analysis of these compounds. The species of genus *Centaurea* are important plant species used alone or with other plants for antidiabetic, antidiarrheal, antirheumatic, anti-inflammatory, cholagogue, choleretic, digestive, stomachic, diuretic, astringent, hypotensive, antipyretic, cytotoxic, antibacterial purposes. *Centaurea kurdica* Reichardt species is an endemic species and its Turkish name is Cottonthorn. In this study, as a result of HPLC separation of *C. kurdica* extract, the amounts of naringin, myricetin and rutin compounds (according to peak area and peak height) were found to be 975-610, 1920-1860, and 760-650 $\mu g_{(component)}/g_{(extract)}$, respectively. As a result of the studies conducted on *C. kurdica* methanol extract, the concentration of total phenolic components was calculated to be 25.635 $mg_{(GAE)}/g_{(extract)}$.

Keywords: *Centaurea kurdica* Reichardt, HPLC, myricetin, naringin, rutin

This study was supported by Amasya University Scientific Research Projects Coordination Unit with the project code FMB-BAP 22-0561.

DRAFT

ORAL PRESENTATION

Artificial Neural Network (ANN) for Estimating Growth Model of Turkish Crayfish (*Pontastacus leptodactylus*) in Yenice Reservoir (Çanakkale, Türkiye)

Semih Kale[1*] (*ORCID: https://orcid.org/0000-0001-5705-6935*),
Selçuk Berber[2] (*ORCID: https://orcid.org/0000-0003-1123-7217*)

[*1]Çanakkale Onsekiz Mart University, Faculty of Marine Sciences and Technology, Department of Fishing and Fish Processing Technology, 17020, Çanakkale, Türkiye.
[2]Çanakkale Onsekiz Mart University, Faculty of Marine Sciences and Technology, Department of Marine and Inland Water Sciences, 17020, Çanakkale, Türkiye.

*Corresponding author e-mail: semihkale@comu.edu.tr

Abstract

This study aimed to suggest an artificial neural network model as a new alternative method for estimating the growth model of the Turkish crayfish (*Pontastacus leptodactylus*) in Yenice Reservoir, Çanakkale, Türkiye. Specimens were collected monthly between July 2007 and June 2008 by using fyke nets (34 mm mesh size). Total length (TL) and total weight (TW) values were measured, and the relationship between TL and TW was presented with the equation of the traditional length-weight relationship (LWR) and artificial neural network (ANN). Subsequently, both the traditional LWR equation and the ANN approach were assessed for estimating the growth of Turkish crayfish in Yenice Reservoir. The results showed that artificial neural networks can be used as a novel approach for estimating the growth of Turkish crayfish. It can be clearly stated that the ANN approach would provide useful information for sustainable and successful fisheries management.

Keywords: Artificial neural networks, Crayfish, Length-Weight relationship, Growth models.

This study was financially supported by Çanakkale Onsekiz Mart University The Scientific Research Coordination Unit with project number: ÇOMÜBAP-FHD 2020-3273.

DRAFT

ORAL PRESENTATION

Determination of radiation absorption properties of composite used in dental applications by Monte Carlo method

Aycan ŞENGÜL[1,*] (https://orcid.org/0000-0003-4548-5403.) İskender AKKURT[2]*(https://orcid.org/0000-0002-5247-7850)*, Kadir AKGÜNGÖR [3](*https://orcid.org/0000-0003-1071-4405.*), Kadir GÜNOĞLU [4] (https://orcid.org/0000-0002-9008-9162)

[1*] Akdeniz Üniversitesi, Sağlık Hizmetleri Meslek Yüksek Okulu, Tıbbi Görüntüleme Teknikleri, Antalya,
[2]Süleyman Demirel Üniversitesi, Fen Bilimleri Fakültesi, Fizik Bölümü, Isparta,
[3] Dokuz Eylül Üniversitesi, Fen Bilimleri Fakültesi, Fizik Bölümü, Atom Ve Molekül Fiziği, İzmir
[4] Isparta Uygulamalı Bilimler Üniversitesi, Nükleer Teknoloji ve Radyasyon Güvenliği, Isparta,

*Corresponding author e-mail: aycansahin@akdeniz.edu.tr

Abstract

The purpose of this study is to compute and compare the linear attenuation coefficients of a composite material used in dental applications using the Monte Carlo method. Using Gamos 6.2 software, a simple geometry consisting of a focused point photon source (monoenergetic between 1 keV and 20 MeV) and NaI detector in parallel with the absorber material was used in the simulation part of the research. The experiment was carried out with a gamma spectrometer system containing NaI(Tl) and gamma rays with energies of 662, 1173, and 1332 keV. Additionally, parameters such as half value layer (HVL), tenth value layer (TVL), and mean free path (MFP) were determined, which are utilized for assessing a material's radiation absorption properties.
When the values obtained by both methods are compared to the theoretical values obtained by XCOM in the energy range of,1 keV-20 MeV, the results are found to be compatible ($R^2 = 0.93$). Uncertainties caused by poor geometry conditions in the experimental setup can account for acceptable differences between test and simulation results.
The Monte Carlo method has been demonstrated to be a viable option for calculating linear absorption coefficients at the desired gamma energy, particularly in the investigation of physically difficult to produce samples.

Keywords: Monte Carlo, linear attenuation coefficients, composites, NaI (Tl), gamma spectrometry, Gamos 6.2

DRAFT

ORAL PRESENTATION

Synthesis and Charcterization of Cationic Derivatives of Plant Oil Triglycerides

Gökhan ÇAYLI[1*] (*ORCID: https://orcid.org/..0000-0002-3395-5642*)

[*1]Istanbul University-Cerrahpaşa, Faculty of Engineering, Department of Engineering Sciences, Istanbul, Türkiye.

*Corresponding author e-mail: gokhan.cayli@iuc.edu.tr

Abstract

Plant oil triglycerides are one of the most important renewable resources. They can be easily obtained and modified to many versatile intermediates. One of the most used plant oil based intermediates is epoxidized plant oil triglycerides. Due to the strained structure of epoxide rings, carboxylic acid can be added. The product is a hydroxyl group containing ester. In this work, epoxidized soybean oil (ESO) was used as a starting material. That compound was then reacted with monochloroacetic acid (MCA). Reaction product (MCA-ESO) is a halogen containing triglyceride. When MCA-ESO met with tertiary amines such as, triethyl amine and pyridine, the reaction products was a quaternary ammine salts. Those compounds have potential antibacterial effect. Moreover, that can be used for waste water treatment for anion removal.

Keywords: Plant oil triglycerides, epoxidized soybean oil, monochloroacetic acid, quaternary amines.

DRAFT

ORAL PRESENTATION

The effect of the use of different stocking volumes on the development of blue crab (m.j. rathbun *callinectes sapidus*) broodstocks

Övgü GENCER[1*] (https://orcid.org/0000-0001-8403-1274)

[*1]Ege University Faculty of Fisheries, Aquaculture Department, 35040 Bornova-Izmir-TURKEY.

*Corresponding author e-mail: ovgu.gencer@ege.edu.tr

Abstract

Crabs are an aquatic product that has found a very high price in developed countries in terms of edible meat quality and economic value. Blue crab broodstocks are among the most important crab species that are commercially dec in the world and in Turkey. There have been no studies on the impact of *C. Sapidus* broodstocks on survival and growth and development. The aim of this study is to determine the effect of different stocking volumes on the survival and growth, development of blue crab (*C. sapidus*) broodstocks. Pearson correlation test has been used in the study. In the study, 0.70 liter tanks with three different stocking densities have been used, including 15, 10 and 5 main ones. The results showed that the mortality rate due to eating each other was high in the first session, while it has been found to be lower in the third session. There were significant differences in broodstock cultivation survival rates in three different sessions ($p<0.05$). Survival rates have been found to be lower at higher stocking densities. According to these results, it seems that increasing the broodstock density significantly reduces the survival rate. Statistical differences have been found in the development rates of broodstocks ($p<0.05$) and it has been noted that there is lower development at higher stocking densities. If it is desired to calculate the optimal stock density for mass production purposes, less density is advantageous from the point of view of both survival and growth rate.

Keywords: Blue crab broodstocks, stocking volume, development, survival rate

DRAFT

ORAL PRESENTATION

Design, synthesis and characterization of A novel series of isoxazole-containing thiourea

Nurcan BERBER[1*] (*ORCID: https://orcid.org/0000-0002-1595-585X*)

[*1]Çanakkale Onsekiz Mart University, Department of Pharmacy Services, Çanakkale, Turkey.

*Corresponding author e-mail: nberber@comu.edu.tr

Abstract

Cancer is one of the leading cause of deaths globally, and can be classified as a multifactorial disease, which is diligently orchestrated by a combination of genetic and environmental factors working together towards the progression of tumours. In this respect, one of the most constantly growing derivatives was the investigation of the antitumor properties of compounds containing isoxazole core structure. Cancer is one of the leading cause of deaths globally, and can be classified as a multifactorial disease, which is diligently orchestrated by a combination of genetic, epigenetic, and environmental factors working together towards the progression of tumours. In this study, to the synthesis of new derivative isoxazole compounds; It was started β-ketoester derivative. For this was reacted 4-bromoacetophenone with diethyl oxalate in the presence of ethanolic solution of sodium ethoxide. After, to the solution of β-ketoester was reacted with hydroxylamine hydrochloride to form the isoxazol ring. Afterwards, after the compound containing isoxazole was hydrated with hydrazine hydrate, it was reacted with five different thioisocyanate derivatives to obtain new derivative isoxazole-containing thiourea derivatives. The chemical structures of the synthesized compounds were ascertained with FTIR, ^{1} H NMR and ^{13} C NMR spectrum. (This work was supported by Çanakkale Onsekiz Mart University The Scientific Research Coordination Unit, Project number: FBA-2022-3950)

Keywords: Isoxazole, thiourea, β-ketoester

DRAFT

ORAL PRESENTATION

Insulinoma INS-1 cells undergo both cell death and cell proliferation with Dinutuximab beta treatment under different conditions

Ayse Karatug Kacar (ORCID: https://orcid.org/0000-0001-6032-470X)

Department of Biology, Faculty of Science, Istanbul University, Istanbul, Turkey

*Corresponding Author: Ayse Karatug Kacar, e-mail: akaratug@istanbul.edu.tr

Abstract

Insulinoma is a gastroenteropancreatic neuroendocrine tumor. This tumor occurs by the uncontrolled proliferation of pancreatic β cells. Dinutuximab beta is a monoclonal antibody. DB is used to treat neuroblastoma. This antibody binds to the GD2 receptor. Treatment with DB takes place through the destruction of tumor cells by their own immune system. The aim of this study is to investigate cell death and cell proliferation at the molecular level as a result of the effects of DB on pancreatic beta-cell tumors. Insulinoma INS-1 cells were used in the study. DB (Qarziba®) was obtained from EUSA Pharma and used in the experiments. Streptozotocin (STZ) was used to induce cell cytotoxicity. DB was applied to cells before or after cytotoxicity. Protein levels of PERK, CHOP, HSP90, p-c-Jun, p-Atf2, and p-Elk1 were analyzed by simple WES. While PERK, CHOP, and p-Elk1 levels decreased, HSP90 levels increased in DB application after cytotoxicity. CHOP, p-Akt-2, and p-c-Jun levels increased in the DB group compared to the control group. As a result, INS-1 cells go to cell death by means of Elk-1 transcription factor without ER stress in DB application after cytotoxicity. In contrast, cells proliferate with the effect of Atf2 and c-Jun transcription factors as a result of only DB application to cells. CHOP may also be effective in this proliferation. DB has shown positive results for the treatment of insulinoma as a result of *in vitro* experiments. The study should be supported by *in vivo* studies.

Keywords: Insulinoma, Dinutuximab beta, Cell Death, Cell Proliferation, MAPK signal pathway

DRAFT

ORAL PRESENTATION

Türkiye'deki SARS-CoV-2 Genomik Sürveyansı Stratejisi: Filogenetik Analiz Sonuçları

Süleyman Yalçın[*1] (*ORCID: https://orcid.org/0000-0003-1434-2717*), Gültekin Ünal[2] (*ORCID: https://orcid.org/0000-0002-8996-7028*)

[*1]Halk Sağlığı Genel Müdürlüğü, Mikrobiyoloji Referans Laboratuvarları ve Biyolojik Ürünler Daire Başkanlığı, Ulusal Moleküler Mikrobiyoloji Referans Laboratuvarı, Ankara, Türkiye
[2]Dünya Sağlık Örgütü, Türkiye Ülke Ofisi, Ankara, Türkiye

*Sorumlu yazar e-mail: suleyman.yalcin3@saglik.gov.tr

Özet

Coronavirüs (COVID19) hastalığına sebep olan Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2), 11 Mart 2020 tarihinde pandemi ilan edilmesine neden olmuştur. SARS-CoV-2'ye ait genetik değişikliklerinin ve virüsün moleküler yöntemler kullanılarak zaman içerisindeki gelişimini incelemek amacıyla dünyada genomik sürveyans çalışmaları geniş bir kullanım alanı bulmuştur. Bu kapsamda Türkiye'de de bir genomik sürveyans stratejisi oluşturulmuş ve hızlı bir şekilde uygulamaya konulmuştur. Bu çalışmada, genomik sürveyans kapsamında dizilemesi yapılan genomlardan yüksek kalitede olan 235 kadarı seçilerek metaveriler ile de ilişkilendirilerek filogenetik analizlerinin yapılması amaçlanmıştır.
Bu çalışmada, genomik dizilemesi Halk Sağlığı Genel Müdürlüğü, Ulusal Moleküler Mikrobiyoloji Referans Laboratuvarı'nda (UMMRL) gerçekleştirilen ve uluslararası bir platform olan GISAID veritabanına yüklenen 235 yüksek kaliteli (Coverage ve bütünlük) genom GISAID'den FASTA formatında indiridi. İndirilen 235 genom MEGA X programında MUSCLE (Gap open: -2,90, gap extend: 0, cluster method: UPGMA) algoritması kulanılarak hizalandı ve sonrasında da ağaç çizimi tamamlandı. Dış grup için referans genom NC_045512.2 (NCBI) kullanıldı. Çizilen filogenetik ağacın görsel açıdan düzenlenmesi Microreact (https://microreact.org/) aracı kullanılarak yapıldı. Filogenetik ağaç için kullanılan örneklere ait metaveriler UMMRL veritabanlarından elde edilmiş ve filogenetik ağaç verileri ile ilişkilendirilmiştir.
Yapılan hizalama ve ağaç çizimi sonucuna göre çalışma kapsamında yer alan genomlar ile ilgili SARS-CoV-2 virüsüne ait Türkiye'nin belirli bölgelerindeki güncel durumu kapsamlı şekilde değerlendirilebildi. Yapılan filogenetik analizler sayesinde etkenin bulaşma yolları, kaynak takibi ve halk sağlığı politikaları oluşturmak için gerekli diğer bilgilerin de elde edilebildiği ve genomik sürveyansa ait süreçlerinin daha da kapsamlı, hızlı ve sistematik olması gerektiği görüldü. Buna bir örnek olarak şehirlerarası virüs dolaşımı ve mutasyonel değişikliklerin izlenebildiği görüldü. Sonuç olarak filogenetik analizlerin metaveriler ile de birleştirilerek halk sağlığı politikalarının oluşturulması adına genomik sürveyans stratejilerinin ileride yaşanabilecek pandemilere de önlem olması açısından büyük önem arz ettiği ortaya konmuş oldu.

Anahtar Kelimeler: SARS-CoV-2, COVID19, Filogenetik Analiz, Genomik Sürveyans.

DRAFT

ORAL PRESENTATION

Tekstil Endüstrisi Atıksularının Ters Osmoz Membranları ile Arıtımı Sonucu Oluşan Konsantre Akımından Organik Madde Gideriminde Farklı Elektro-Fenton Proseslerin Etkisinin Araştırılması

Gamze Canbakal[1*](*ORCID:https://orcid.org/0009-0006-0713-0862*), Oyku Nur Bilgin[1](*ORCID: https://orcid.org/0000-0003-4178-0990*), Senem Yazici Guvenc[1](*ORCID: https://orcid.org/0000-0002-2877-0977),* Gamze Varank[1](*ORCID:https://orcid.org/0000-0003-3437-4505),* Emine Can Guven[1](*ORCID: https://orcid.org/0000-0002-3540-3235)*

[1]Yıldız Teknik Üniversitesi, İnşaat Fakültesi, Çevre Mühendisliği Bölümü, 34220, Esenler, İstanbul-Türkiye

*Sorumlu yazar e-mail:gamze.canbakal@std.yildiz.edu.tr

Özet

Tekstil endüstrisi boyama, durulama, ağartma, sonlandırma gibi ürünü işleme alan ve su tüketimi odaklı aşamalardan oluşmaktadır. Tüm bu proseslerden farklı karakterizasyonlarda ve yüksek hacimlerde atıksular meydana gelmektedir. Tekstil endüstrisi atıksuları, yüksek konsantrasyonlarda organik madde içeren, yüksek renkli, kompleks bileşenler, reaktif boya ve kimyasal kalıntıları ile büyük ölçüde değişen bileşenlere sahiptir. Alıcı ortama arıtılmadan deşarj edildiğinde sudaki organizmaların ihtiyaç duyduğu ışığın geçişine engel olan bu renkli sular, ekosistemde bir dengesizliğe yol açarak fotosentezi engelleyebilir. Bu nedenle toksisiteyi, kirliliği en aza indirmek ve çevreyi korumak için tekstil endüstrisi atıksularının deşarjdan önce arıtılması önemlidir. Bu tür atıksular toksisitesi ve dirençli kirleticiler içermesi nedeniyle konvansiyonel ve biyolojik arıtma prosesleri ile etkili şekilde arıtılamamaktadır. Yüksek hacimlerde ortaya çıkan bu atıksuların yeniden üretim aşamalarında kullanımı için geri kazanımını hedefleyen ters osmoz (TO) membran sistemler tercih edilmektedir. Ancak, membran sistemlerde ortaya çıkan konsantre akımı yüksek miktarda kirlilik yüküne sahip ve düşük hacimli bir atıksu olup, konvansiyonel yöntemler ile arıtımı mümkün değildir. Bu çalışmada, toksik içeriği ve kirlilik konsantrasyonu yüksek ve renkli olan ters osmoz konsantresi atıksularından organik madde gideriminde farklı türde Elektro-Fenton (EF) proseslerin performansları incelenmiştir. Bu amaçla, belirli koşullar altında Peroksi-Koagülasyon (PK), Elektrokimyasal-Peroksidasyon (EPO), Fered-Fenton (FF) ve EF prosesleri uygulanarak kimyasal oksijen ihtiyacı (KOİ) giderim verimleri karşılaştırılmıştır. Fenton reaktiflerinin dışarıdan ilavesi ya da yerinde üretimine bağlı olarak değişen bu prosesler PK, EPO, FF ve EF için sırasıyla KOİ giderim verimleri %70.4, %78.9, %72.3 ve %55.6 elde edilmiştir. KOİ giderim verimlerine bağlı olarak birinci mertebeden reaksiyon kinetikleri hesaplanmış ve PK, EPO, FF ve EF prosesler için sırasıyla 0.0132, 0.0168, 0.0143 ve 0.0085 1/dk olarak belirlenmiştir. Tüm bu elektrokimyasal proseslerin organik madde gideriminde etkin bir yöntem olduğu görülmüştür. Proses şartlarının optimizasyonu ile TO konsantre atıksularının üretim proseslerinde yeniden kullanımı mümkün olabilir.

Anahtar Kelimeler: Tekstil Endüstrisi Atıksuları, Membran Konsantresi, Peroksi-Koagülasyon, Elektrokimyasal-Peroksidasyon, Fered-Fenton, Elektro-Fenton.

DRAFT

ORAL PRESENTATION

Preparation and characterization of decellularized tendon extracellular matrix-halloysite biocomposite scaffold for bone tissue engineering applications

Miray Yıldırım (https://orcid.org/0009-0006-7182-1123), Aysel Koç Demir (https://orcid.org/0000-0001-9870-9947)

Ankara Üniversitesi, Fen Fakültesi, Kimya Bölümü, Ankara, Türkiye.

*Corresponding author e-mail: aykoc@science.ankara.edu.tr

Abstract

The aim of bone tissue engineering (BTE) is to treat bone defects resulting from various bone diseases, tumors or fracture with alternative approaches. The basic principle of BTE approach is to design and develop three-dimensional, functional biological systems specific to the targeted tissue and/or organ by combining the cell, biomaterial and various bioactive molecules. One of the most important factors that ensure successful achievement of the goal is the development of the most ideal biocompatible and biodegradable three-dimensional (3D) biomaterials that will mimic the "artificial culture medium" for cells. In recent years, natural extracellular matrix (ECM) structures obtained from different tissues or organs and halloysite (HNT) have been started to be used in tissue engineering applications due to their biological advantages. In light of this information, the aim of this study is to develop and characterize bone tissue engineering scaffolds by using dECM and HNT and to evaluate their usability for bone tissue engineering the feasibility of the prepared scaffolds for bone tissue engineering applications. For this purpose, the dt-ECM structure was obtained by decellularization of bovine tendon tissue. Following the decellulation process, the dt-ECM was digested with pepsin solution. In the next step, different concentrations of HNT were added separately to the pepsin-digested and neutralized dtECM pre-gel solution, and the suspensions were mixed at 4 ^{o}C by using a mechanical stirrer until a homogeneous solution was obtained. In the last step, the obtained composite suspensions were poured into the molds and kept at 37 ^{o}C to complete gelation. Nanocomposite scaffolds were obtained by freezing and then lyophilizing the gelled samples at -80 ^{o}C.While the physical and chemical properties of the obtained scaffolds were examined by different analysis methods, the potential for use in biological applications was evaluated by blood compatibility and in-vitro cytotoxicity tests. The obtained results showed that the composite scaffold as a prospective candidate for bone tissue engineering.

Keywords: Bone tissue engineering, Halloysit, Extracellular matrix, Scaffold.

DRAFT

ORAL PRESENTATION

Gelincik çiçeği (*Papaver rhoaes* L.) ekstraktının renk maddesi olarak kullanım olanakları

Gülce Bedis KAYNARCA[1*] (*ORCID: https://orcid.org/0000-0001-7896-457X*), Şeyda Yanardağ Karabulut[1] (*ORCID: https://orcid.org/0000-0002-9649-5874*), Hacı Ali Güleç[2] (*ORCID: https://orcid.org/0000-0002-9525-6206*), Deniz Damla Altan Kamer[3] (*ORCID: https://orcid.org/0000-0002-9119-5979*)

[*1]Kırklareli Üniversitesi, Mühendislik Fakültesi, Gıda Mühendisliği Bölümü, Kırklareli, Türkiye
[2]Trakya Üniversite, Fakülte, Gıda Mühendisliği Bölüm, Edirne, Türkiye
[3]Tekirdağ Namık Kemal Üniversitesi, Ziraat Fakültesi, Gıda Mühendisliği Bölümü, Tekirdağ, Türkiye

*Sorumlu yazar e-mail: b.gulcebedis@klu.edu.tr

Özet

Paketli gıdalarda tüketiciyi ilk cezbeden özellikler renk ve görünüştür. Bu nedenle, renk maddeleri hem tüketici beğenisine uygun olmalı hem de yapay renk maddeleri gibi kanserojenik, alerjik reaksiyonlar ve nörolojik etkiler gibi olumsuz sağlık problemlerine neden olmamalıdır. Gıdalarda kullanılan renk maddeleri asıl amaçlarının yanı sıra ürünleri tatlandırmak, korumak ve fonksiyonel özelliklerini geliştirmek amacıyla da kullanılabilmektedir.
Gelincik (*Papaver rhoeas* L.) *Papaveraceae* familyasında bulunan, dünyanın pek çok yerinde yetişen, tek yıllık otsu bir bitkidir ve geleneksel olarak kırmızı taç yaprakları gelincik şerbeti yapımında kullanılmaktadır. Gelincik bitkisinin antikarsinojenik, antiinflamatuvar, antimutajenik ve antigenotoksik etkilere sahip olduğu bilinmekte ve öksürük, uyku problemleri ve anksiyete durumlarında kullanılabileceği ileri sürülmektedir.
Literatürde daha çok gelincik bitkisinin medikal alanda kullanımı üzerine çalışmalara rastlanmıştır. Bu araştırma ile gelinciğin kırmızı taç yapraklarından elde edilen ekstraktın renk maddesi olarak değerlendirilebilme potansiyeli ve bu ekstraktın fizikokimyasal özellikleri ve fonksiyonel özellikleri tespit edilmeye çalışılmıştır. Etanol ile ekstrakte edilen ekstraktın pH, % kuru madde ve % kül değerleri sırasıyla 0.58 ± 0.01, 1.03 ± 0.12 ve 0.11 ± 0.00 olarak belirlenmiştir. Toplam fenolik madde içeriği 3772 ± 70.71 mg gallik asit eşdeğeri (GAE)/ L olarak tespit edilirken toplam antosiyanin içeriği 1427.76 ± 47.23 mg siyanidin-3-glukozit/ L olarak bulunmuştur. Ekstraktın termal stabilitesini belirlemek amacıyla sitrat tamponu ile hazırlanan örnekler 1 saat boyunca 60, 80 ve 100°C sıcaklığa maruz bırakılmış bunun sonucunda total antosiyanin içeriklerinde sırasıyla %4.88, 6.37 ve 15.49 oranında bir azalma meydana gelmiştir. Oksidatif stabiliteyi belirlemek için 9.31mM, 18.61mM ve 27.92 mM hidrojen peroksit ile muamelem edilen ekstraktların toplam antosiyanin içerikleri ise sırasıyla %11.21, 12.90 ve 16.28 oranında azalma göstermiştir. Ekstraktın pH'a bağlı renk değişimi incelendiğinde kırmızı rengi pH 5' e kadar koruyabildiği sonucuna varılmıştır. Ekstrakt sodyum, potasyum, fosfor ve demir mineralleri bakımından oldukça zengin bulunmuştur. Ayrıca bu ekstraktının ayırt edici düzeyde kuarsetin ve prokateşik asit içerdiği tespit edilmiştir.

Anahtar Kelimeler: Gelincik çiçeği (*Papaver rhoaes* L.), renk maddesi, fenolik profil, mineral madde.

DRAFT

ORAL PRESENTATION

Zr-MOF ile tetrasiklin antibiyotiğinin sulu ortamdan giderimi

Tuğba Alp Arıcı[1*] *(ORCID: https://orcid.org/0000-0003-3927-9849)*, Mürsel Arıcı[2] *(ORCID: https://orcid.org/0000-0001-8527-1587)*

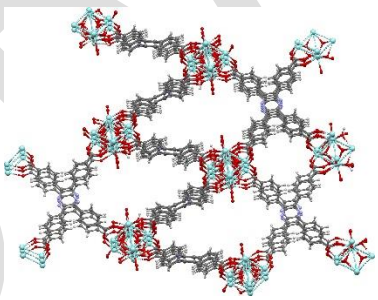
[*1]Kütahya Dumlupınar Üniversitesi, Emet Meslek Yüksekokulu, Kimya Teknolojisi Programı, 43700, Kütahya, Türkiye
[2]Eskişehir Osmangazi Üniversitesi, Fen Fakültesi, Kimya Bölümü, Eskişehir, Türkiye

*Sorumlu yazar e-mail: tugba.alp@dpu.edu.tr

Özet

Antibiyotikler, son yıllarda tıp, hayvan hastalıklarının önlenmesi, su ürünleri yetiştiriciliği ve hayvancılık alanlarında yaygın olarak kullanılmaktadır. Tetrasiklinler (TC), dünya çapında en çok üretilen ikinci antibiyotik grubu olarak bilinmektedir [1]. Bu antibiyotik türü bakteriyel enfeksiyonlarla savaşmak için tarım ve hayvancılıkta yaygın olarak kullanılmaktadır. TC'nin çevreye salınımının ana kaynağının (%69-86) insan ve hayvanların idrarı ve dışkısı olduğu bildirilmektedir [2]. TC'nin sularda yüksek derişimlerde bulunması bu ortamlardaki sucul canlıların yaşamında toksik etki oluşturmakta, bakteri ve mikroorganizmaların bu antibiyotiğe karşı direnç kazanmalarına sebep olmaktadır. Sulu ortamdan farmasötik kirleticilerin gideriminde pek çok geleneksel arıtım yöntemi kullanılmasına rağmen adsorpsiyon, ekonomik, kolay uygulanabilir oluşu ve toksik yan ürün oluşturmama özelliği nedeniyle oldukça avantajlıdır. Metal organik kafes yapıların (MOF) bu yöntemde, geniş yüzey alanı, termal kararlılığı, ultra yüksek gözenekliliği ve ayarlanabilir gözenek boyutları gibi dikkate değer avantajları nedeniyle adsorpsiyon yetenekleri incelenmektedir.
Bu çalışmada, amid grubu içeren Zr-MOF, $[Zr_6O_4(OH)_8(H_2O)_4(TCPP)_4]\cdot 9DMF\cdot 3.5H_2O$, yapısı sentezlenmiş ve karakterize edilmiştir [3]. Zr-MOF sulu ortamdan TC gideriminde adsorban olarak kullanılmıştır. Adsorpsiyona, pH, adsorban miktarı ve temas süresinin etkileri araştırılarak optimum adsorpsiyon koşulları belirlenmiştir. MOF'un üç boyutlu (3D) yapısı Şekil 1'de verilmiştir. MOF kullanılarak TC'nin %84 verimle sulu ortamdan uzaklaştığı sonucuna ulaşılmıştır.

Anahtar Kelimeler: Adsorpsiyon, Kinetik, Tetrasiklin, Zr-MOF



Şekil 1. Zr-MOF'un 3D yapısı

Kaynaklar

[1] S. Qian, L. Qiao, W. Xu, K. Jiang, Y. Wang, H. Lin Talanta, 2019, 194, 598–603.
[2] A. Abbasnia, A. Zarei, M. Yeganeh, H. R. Sobhi, M. Gholami, A. Esrafili, Inorg. Chem. Commun. 2022, 145; 109959
[3] J. Liu, Y. Ye, X. Sun, B. Liu, G. Li, Z. Liang and Y. Liu, J. Mater. Chem. A, 2019, 16833-16841.

DRAFT

ORAL PRESENTATION

Zeytinyağının oleuropeini alzheimer hastalığı için umut olabilir mi?

Murat ÇELEBİ[1] (*ORCID: https://orcid.org/0000-0002-1769-2512*), Çağla ÇELEBİ[2] (*ORCID: https://orcid.org/0000-0001-6253-7346*), Elif AKSÖZ[3*] (*ORCID: https://orcid.org/0000-0002-4827-804X*)

[*1]Balıkesir Üniversitesi, Savaştepe Meslek Yüksekokulu, Veterinerlik Bölümü, Balıkesir, Türkiye
[2]Balıkesir Üniversitesi, Veteriner Fakültesi, Farmakoloji ve Toksikoloji Anabilim Dalı, Balıkesir, Türkiye
[3]Balıkesir Üniversitesi, Tıp Fakültesi, Tıbbi Farmakoloji Anabilim Dalı, Balıkesir, Türkiye

*Sorumlu yazar e-mail: aksoz@balikesir.edu.tr

Özet

Alzheimer hastalığı (AH) 65 yaş ve üzerindeki insanlarda görülen ilerleyici ve ölümcül demans vakalarının dörtte üçünden sorumludur. Dünya çapında yaklaşık 50 milyon insan demansdan mustariptir. 2050 yılına kadar bu sayının 152 milyona çıkması öngörülmektedir. Maalesef, günümüzde halen AH'nin tedavisi tamamen semptomatik olarak yapılmaktadır. Ayrıca, güncel tedavide kullanılan kolinesteraz inhibitörü ilaçların yan etkilerinin fazlalığı hastalar için ciddi sorunlar yaratmaktadır. AH'nin oluşması ve ilerleyişinde oksidatif stres ve nöroinflamasyonun etkili olduğu düşünülmektedir. Zeytinyağı bileşenleri içerisinde büyük yer kaplayan oleuropein, güçlü antioksidan etkileri ile bilinmektedir. Bir çalışmada oleuropein ile 7 günlük tedavinin reaktif oksijen türlerinin seviyelerini düşürdüğü, lipid peroksidayonu azalttığı, süperoksit dismutaz ve katalaz aktivitelerini arttırdığı gösterilmiştir. Ayrıca oleuropein, oksidatif stres nedeniyle oluşan heterosiklik aminlere bağlı DNA hasarına karşı da koruma sağlamaktadır. Ek olarak eozinofil ve makrofaj infiltrasyonunu önleyerek IL-4'e maruz kalan epitel hücrelerinde inflamasyonu azalttığı da bildilmiştir. Oleuropeinin nöroprotektif rolü için, apoptoz ve otofajinin indüklenmesi, proinflamatuar sitokinlerin ve kemokinlerin gereksiz salınımını azaltarak nöroinflamasyonu önleme başta olmak üzere çeşitli mekanizmalar öne sürülmektedir. Düzenli oleuropein alımı AH, Parkinson, inme, depresyon, anksiyete, epilepsi gibi nöral bozuklukların azalmasıyla ilişkili görülmektedir. Ancak randomize kontrollü çalışmalarla kanıtlanmış veriler bulunmamaktadır. Oleuropeinin metabolik yollardaki etkileri, biyoyararlanımı ve gen ekspresyonu gibi farklı konulardaki etkinliklerinin belirlenmesi bilişsel bozulmayla olan savaşımızda yeni ve kuvvetli bir savunma hattı olabilir. Ülkemizde bolca bulunan oleuropeinden zengin, zeytin ve zeytinyağı ürünlerinin korunma ve tedavide etkili bir strateji haline gelebilmesi için daha fazla araştırma yapılması, ülkemize de bir katma değer olarak katkı sağlayacaktır.

Anahtar Kelimeler: Alzheimer, antioksidan, oleuropein.

DRAFT

ORAL PRESENTATION

Irisin levels in schizophrenia and psychosis: Unraveling the hormonal link

*Erhan DINCER ORCID:(0000-0002-4060-8067, [2]Sermin ALGÜL ORCID:(0000-0003-2489-3619)

[*1] Bitlis Eren University, Faculty of Health Sciences, Department of Physiotherapy and Rehabilitation, Bitlis, Turkey
[2] Van Yüzüncü Yıl University, Faculty of Medicine, Department of Basic Medical Sciences, Van, Turkey

* Corresponding author e-mail: edincer@beu.edu.tr

Abstract

Introduction: Irisin, recently identified as a hormone classified as both a myokine and an adipokine, has emerged as a significant factor in promoting the conversion of white adipose tissue to brown adipose tissue. This process, known as "browning," is associated with increased energy expenditure and calorie burning. Moreover, irisin has been linked to several beneficial outcomes of regular exercise, contributing to improved metabolism and overall well-being. Despite the growing understanding of irisin's role in metabolism and its potential therapeutic implications, the exact pathophysiology of schizophrenia and psychosis remains incompletely understood, particularly concerning their connections with hormonal regulation. Further investigations and research are needed to unravel the intricate interplay between irisin and the development or progression of these complex psychiatric conditions. Gaining a deeper comprehension of these relationships could open the door to novel therapeutic strategies and shed light on the potential impact of irisin in managing these disorders.
Objective: The main objective of this study was to investigate potential differences in irisin hormone levels between individuals diagnosed with schizophrenia, psychosis, and healthy individuals without these conditions. Additionally, the study aimed to evaluate the potential of irisin hormone as a diagnostic marker for individuals with schizophrenia and psychosis.
Materials and Methods: The study included a total of 80 participants, consisting of 30 individuals diagnosed with schizophrenia, 20 with psychosis, and 30 healthy individuals. Blood samples were collected from all participants between 08:00-10:00 in the morning after an overnight fasting period. The levels of irisin in the collected blood samples were measured using the ELISA method with commercially available kits.
Results: The study identified significant variations in irisin hormone levels among the groups analyzed. Specifically, the irisin level in schizophrenia patients (544±28 pg/ml) was notably lower than that in both psychosis patients (643±23 pg/ml) and the control group (735±28 pg/ml) ($p<0.05$). Additionally, the irisin level in psychotic patients (643±23 pg/ml) was found to be significantly lower than in the control group (735±28 pg/ml) ($p<0.05$).
Conclusion: Based on these findings, it is evident that irisin hormone plays a critical role in the pathophysiological mechanisms of schizophrenia and psychosis. Furthermore, it is suggested that irisin holds substantial potential as a noteworthy diagnostic marker for these disorders, offering possibilities for prognostic value. Consequently, irisin emerges as a subject of considerable interest in the pursuit of a deeper understanding and enhanced management of schizophrenia and psychosis. Nevertheless, to gain a more comprehensive comprehension of the precise role of irisin hormone in the treatment processes of these conditions, further research and clinical studies are warranted.

Keywords: Schizophrenia, Psychosis, Irisin, ELISA, Hormone

DRAFT

ORAL PRESENTATION

Development of a novel pro-angiogenic and antimicrobial synthetic skin graft

Serkan Dikici[1*](*ORCID: https://orcid.org/0000-0001-9933-5254)*

[*1]Izmir Institute of Technology, Faculty of Engineering, Bioengineering Department, Izmir, Turkey

*Corresponding author e-mail: serkandikici@iyte.edu.tr

Abstract

Every year, between 9.1 and 26.1 million diabetic patients worldwide face foot ulcers that do not heal on their own. Approximately 15-25% of all diabetic patients develop ulcers that do not necessarily heal on their own throughout their lives and require bioactive treatments. Similarly, when examining deep burns, which constitute a significant part of the wounds that do not heal completely, approximately 50000 of the 1.1 million burns requiring medical intervention each year in the USA require hospitalization, and 20000 of them are suffering from non-self-healing major burns, which cover up to 25% of their total body surfaces and waiting for treatment.

Biomaterials and tissue engineering offers promising approaches for healing wounds that cannot follow the ordinary wound healing process and do not heal on their own. Non-healing wounds could be acute wounds, such as multilayer wounds and deep burns, as well as chronic wounds, such as diabetic ulcers or venous ulcers. The treatment of such difficult-to-heal or non-self-healing wounds requires more than the use of traditional wound dressings that only provide moisture, control exudate and odour, and protect the wound from external factors. Skin substitutes, one of the most important clinical approaches of biomaterials science and tissue engineering, play a key role in the treatment of these wounds.

In the scope of this study, a bioactive and antimicrobial bilayer polycaprolactone (PCL) skin substitute was developed combining electrospinning and emulsion templating. 2-deoxy-D-ribose (2dDR) was incorporated into PCL electrospun layer to induce angiogenesis whereas PCL PolyHIPE layer was conjugated with antimicrobial peptide sequences to control wound infection. To date, we have manufactured and characterised both layers individually *in vitro* and successfully combined them together. This study was financially supported by a grant from The Scientific and Technological Research Council of Türkiye (TÜBİTAK) (Grant No. 221S594).

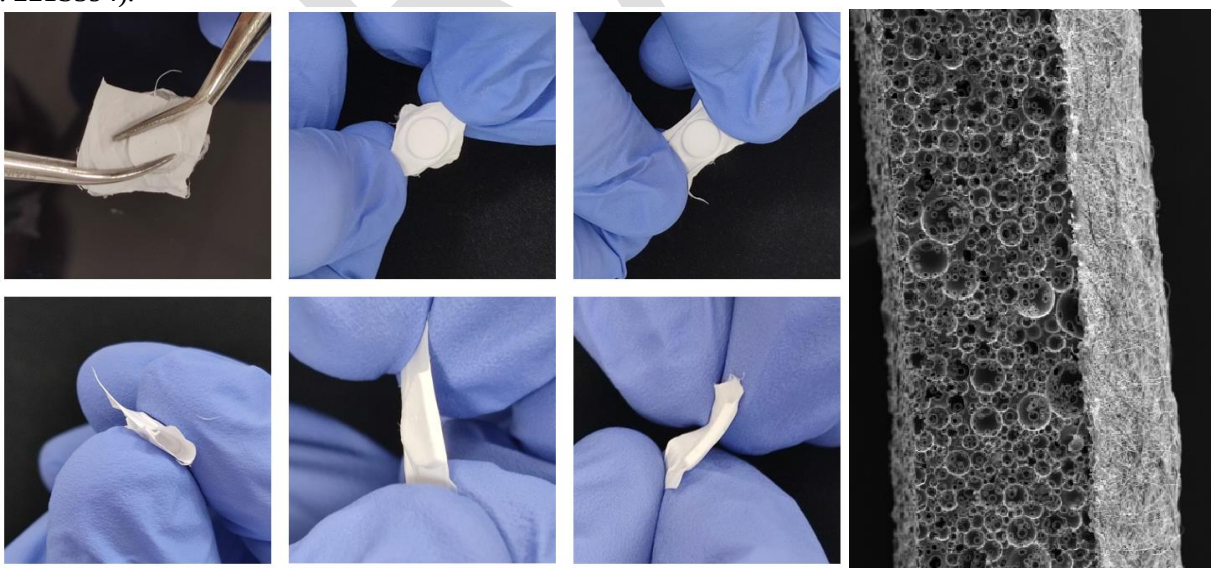


Figure 1. Macro images of the bilayer synthetic skin graft

Keywords: biomaterials, tissue engineering, decellularization, parsley, vascular graft, blood vessel

ORAL PRESENTATION

Development of bioactive wound dressings from 2-deoxy-D-ribose (2dDR) loaded decellularized plant leaves

Serkan Dikici[1*] (*ORCID: https://orcid.org/0000-0001-9933-5254)*

[*1]Izmir Institute of Technology, Faculty of Engineering, Bioengineering Department, Izmir, Turkey

*Corresponding author e-mail: serkandikici@iyte.edu.tr

Abstract

Wound treatment requires the use of dressing materials that will ensure a safe barrier for the coverage of wounds as well as the preservation of the moist environment. Traditional wound dressings are cost-effective and provide some of the abovementioned properties. However, they lack bioactivity and do not contribute to the wound healing process. A bioactive wound dressing is required to manage challenging wounds.
2-deoxy-D-ribose (2dDR) is a novel, cost-effective and safe-to-use pro-angiogenic agent to promote angiogenesis and accelerate wound healing. It can easily be introduced into biomaterials and will be eventually released to show its effect. We have recently discovered its potential to promote angiogenesis and accelerate wound healing when released from several biomaterials in vitro and in vivo.
Lately, the use of decellularized biological constructs has become an emerging strategy to produce physiologically compatible tissue engineering scaffolds for use in both preclinical studies and clinical applications. The main principle of decellularization can be summarized as the removal of the native genetic material of the tissue or organ while preserving the basic ECM elements. The use of decellularized plant tissues as TE scaffolds is a promising alternative to animally sourced tissues and organs. Plant tissues are readily available, cost-effective, and clinically safe. Thus, turning plants into clinical biomaterials brought a new and unique perspective into the field of tissue engineering.
Accordingly, in this study, we developed a bioactive wound dressing from 2dDR-loaded decellularized spinach leaves and investigated its potential to promote angiogenesis in vitro. Our results demonstrated that a cellulose-based biomaterial can be developed from spinach leaves which can then be used as a potential drug delivery system. It showed great potential as a bioactive dressing to promote angiogenesis in vitro when it is loaded with 2dDR. This study was supported by IZTECH Research Foundation (Grant no: 2021İYTE-1-0057).

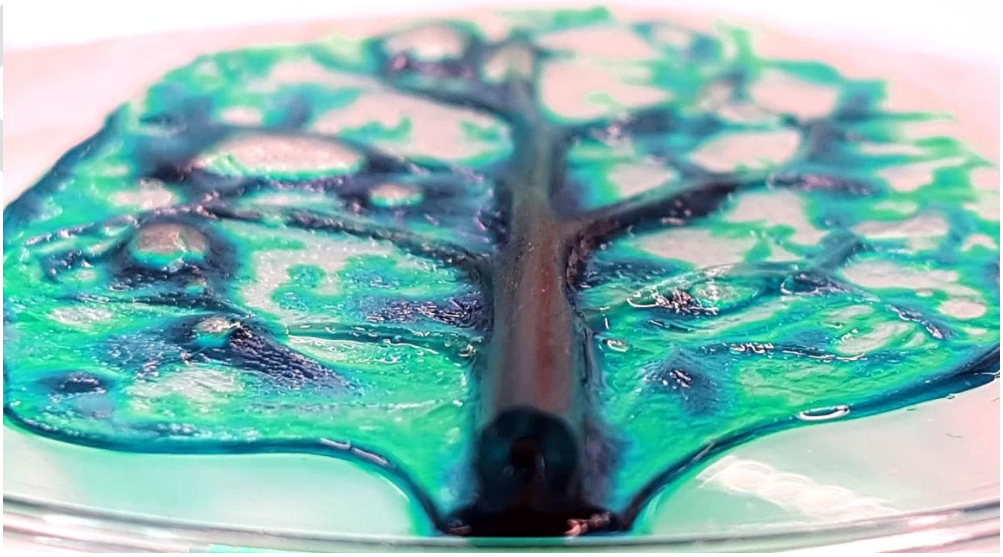


Figure 1. The native vascular architecture of a decellularized baby spinach leaf. The patency was shown by injecting a blue food dye from the petiole.

Keywords: biomaterials, tissue engineering, decellularization, spinach leaves, angiogenesis, wound healing

ORAL PRESENTATION

Which one is a healthier option for parsley: eating it or using it as a tissue-engineered vascular graft?

Serkan Dikici[1*], Merve Çevik[2] (*ORCID: https://orcid.org/0000-0001-9933-5254)*

[*1]Izmir Institute of Technology, Faculty of Engineering, Bioengineering Department, Izmir, Turkey
[2]Izmir Institute of Technology, Department of Biotechnology and Bioengineering

*Corresponding author e-mail: serkandikici@iyte.edu.tr

Abstract

With 17.9 million deaths annually, cardiovascular diseases (CVDs) are the most common cause of death worldwide. Over 80% of CVD deaths are caused by strokes or heart attacks, and 1/3 of these deaths are among adults below the age of 70.
A harmed blood vessel must be replaced/bypassed or repaired to eliminate the blockage. Vascular grafts play a significant part in today's care of a variety of clinical problems. For small-diameter arterial grafts, autologous tissue, such as the internal thoracic artery or saphenous vein, is frequently the initial option. However, earlier operations and other medical problems may constrain the availability of a patient's own vessels, and their collection may be linked with high morbidity. Synthetic grafts have demonstrated good long-term outcomes and patency for managing the aorta and other big arteries, but their usage for bypassing small-diameter vessels has been linked to worse-than-ideal outcomes.
In this study, we successfully developed a tissue-engineered vascular graft using decellularized parsley stems, characterized it, demonstrated its mechanical durability under flow conditions, and assessed its cytotoxicity. Then, we evaluated the potential of decellularized parsley stems to support fibroblast and endothelial cell growth in the lumen structure to create a potential tissue-engineered vascular graft. From the clinical point-of-view, to shorten the time of vascularization upon implantation, we loaded parsley stems with a pro-angiogenic agent (2-deoxy-D-ribose (2dDR)) and showed its release over 48 hours to promote vascularization. Finally, we sutured decellularized parsley tubes to an artery isolated from a goat, ex vivo and evaluated its mechanical durability under flow conditions. This study was financially supported by IZTECH Research Foundation (Grant no: 2022İYTE-3-0002).

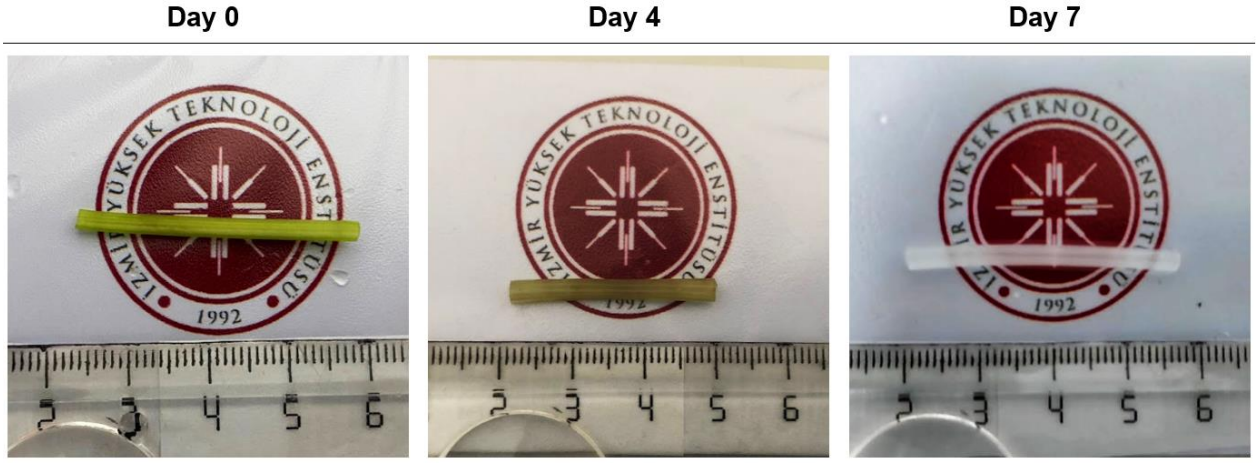


Figure 1. Macro photos showing the colour loss of parsley stems over 7 days

Keywords: biomaterials, tissue engineering, decellularization, parsley, vascular graft, blood vessel

ORAL PRESENTATION

Biogas, hydrochar and biochemical production from spent coffee grounds with biorefinery approach integrating hydrothermal carbonization and anaerobic digestion

Berrak Fidan[1,2] *(ORCID:https://orcid.org/0009-0000-0359-5255)*, Tülay Güngören-Madenoğlu[1*] *(ORCID: https://orcid.org/0000-0001-6014-9956)*, Nalan Kabay[1] *(ORCID: https://orcid.org/0000-0003-2306-830X)*, Levent Ballice[1] (*ORCID: https://orcid.org/0000-0002-3137-1352)*

[1]Ege University, Faculty of Engineering, Department of Chemical Engineering, Bornova-Izmir, Turkey.
[2]Ege University, Graduate School of Natural and Applied Sciences, Chemical Engineering Division, Bornova-Izmir, Turkey.

*Corresponding author e-mail: tulay.madenoglu@ege.edu.tr

Abstract

As a biorefinery approach, combining thermochemical (hydrothermal carbonization, HTC) and biological (anaerobic digestion, AD) processes for valorization of spent coffee grounds (SCGs) can pave the new ways for renewable energy production, when the annual coffee consumption in the world and its lignocellulosic wastes were considered. The aim of this study was to produce biofuel and biochemicals via biorefinery approach involving AD of the hydrochars obtained after HTC of SCGs, and investigate the optimum conditions for maximizing methane-rich biogas yield. HTC was conducted at 180, 220 and 260°C with 1 h residence time and biomass to water ratio as 0.2. Hydrochar efficiencies were found as 57.46%, 47.48% and 44.90% for 180, 220 and 260°C, respectively. While waste sludge (WS) obtained from AD tanks of a yeast plant was used as inoculum, SCGs, hydrochar and process water formed during HTC were used as substrate in AD at mesophilic condition (37°C). Within 50 days of AD and with VS ratio of 3.6, maximum cumulative biogas and biomethane content was reached to 182.8 mL/g TVS and 125.1 mL CH_4/g TVS by using hydrochars obtained at 180°C, while it was 278.6 mL/g TVS and 180.5 mL CH_4/g TVS for SCG, respectively. Carboxylic acids (acetic acid, propionic acid, butyric-isobutyric acids, glycolic acid and formic acid), furfurals (MF, 5-HMF), phenols (phenol and o-m-p-cresols) and ketones (3-methyl-2-cyclopenten-1-one and 2-cyclopenten-1-one) analyzed in liquid phase that obtained at the end of integrated process. Volatile fatty acid (VFA) content in the liquid phase obtained at the end of AD of SCGs and hydrochars was found suitable for a stable AD, while it was found unfavorable for process water samples because of unbalanced C/N ratio. Hydrochars were characterized for surface morphology, thermal degradation properties, functional groups, elemental composition by SEM, SDT (TGA+DSC), FT-IR, elemental analyzer (ICP-OES), respectively.

Keywords: Biorefinery, Biogas, Hydrochar, Hydrothermal Carbonization, Anaerobic Digestion

DRAFT

ORAL PRESENTATION

Investigation of the antioxidant and antibacterial effects of fermented *Cornus Mas* and *Rubus Sanctus* fruits

Derya Ünal[1] (https://orcid.org/0000-0001-8322-7400), Tuba Sevimoglu[2,*] (https://orcid.org/0000-0003-4563-3154)

[1]Department of Bioengineering, Üsküdar University, İstanbul, Türkiye
[2]Department of Bioengineering, University of Health Sciences, İstanbul, Türkiye

*Corresponding Author e-mail: tuba.sevimoglu@sbu.edu.tr

Abstract

In this study antioxidant and antibacterial activities of fermented *Cornus Mas* and *Rubus Sanctus* berries collected from province of Bartın in the Western Black Sea region of Türkiye were analyzed. Prior to fermentation with *Saccharomyces cerevisiae,* the fruits were tested for 58 pesticides such as Dicloran and Quintozene and none of the pesticides were detected. The presence of ascorbic acid in the fruits, which is a nutrient needed by the body, was also detected by FTIR. Then the pesticide free berries were crushed, and the samples were fermented separately. Testing after the fermentation process revealed the samples contained ethyl alcohol. Antioxidant activities of fermented samples were analyzed using CUPRAC, DPPH and Folin Ciocalteu methods. The results suggest high antioxidant contents of the fermented samples. Evaluation of antimicrobial activity was done through disk diffusion method using *Pseudomonas aeruginosa and Staphylococcus aureus* suggesting that these samples do not suppress these bacteria for the studied concentrations. Furthermore, the growth of *Candida albicans* was examined immediately, demonstrating that the fermented samples do not show antifungal effects. The reason for these shortcomings could be inadequate concentration levels. The antioxidant content of these fermented fruits is intended to contribute to human health.

Keywords: *Cornus Mas*, *Rubus Sanctus*, Fermentation, Antioxidant, Antibacterial

DRAFT

ORAL PRESENTATION

The key roles of mTOR and p53 during cell fate decision: Senescence or quiescence?

Menderes Yusuf Terzi[1*] (*https://orcid.org/0000-0001-8478-0451*)

[*1]Hatay Mustafa Kemal University, Faculty of Medicine, Department of Medical Biology, Hatay, Türkiye.

*Corresponding author e-mail: menderesyusufterzi@gmail.com / myterzi@mku.edu.tr

Abstract

The researchers can use the terms “senescence” and “quiescence” interchangeably in their studies although they are two distinct cellular states in which altered expression/activation levels of common molecular players play pivotal roles. Among which, mechanistic target of rapamycin (mTOR) and tumor suppressor protein p53 are the key molecules that are accounted for during the decision of a cell’s fate. The presence or absence of mTOR determines whether the cell undergoes normal cell cycle and cell division. If mTOR presents, in this case the activation status of p53 decides further cell pathway; while maximal activation of p53, which inhibits mTOR, leads the cell to quiescence, the partial activation of p53, which keeps mTOR in active state, results in senescence. In the big picture, the blockade of mTOR causes nutritional and growth insufficiency which brings about quiescence however, cellular aging and severe DNA damage cause senescence. In this perspective, senescence is a detrimental process and senescent cells eventually enter into apoptosis whereas quiescent cells can stay at cell cycle arrest as until the growth conditions are optimized. On the other hand, the sustained or prolonged quiescence can lead to degeneration of cells which transforms quiescence into senescence phenotype by abnormal cellular hypertrophy and apoptosis.

Keywords: p53, mTOR, Senescence, Quiescence.

DRAFT

ORAL PRESENTATION

Synthesis and Characterization of Fluorine-Contained Dental Composites

Sedef Kaptan Usul[1*] , Ayşe Aslan[1,2]

[1]Bioengineering Department, Gebze Technical University, Kocaeli
[2]Institute of Energy Technologies, Gebze Technical University, Kocaeli

Abstract

In this study, dental composite series were designed with new-generation antimicrobial, fluorine-containing silica nanoparticles. Silica nanoparticles provided the advantage of long-term use of the composite thanks to their mechanical strength. The fluorine element interacted with silica nanoparticles giving the dental composite anti-microbial properties. The synthesized functional nanoparticles were homogeneously distributed in the resin and dental filling material by UV polymerization. The silica nanoparticles with fluorine-containing content were characterized by FTIR spectroscopy to confirm their binding to BisGMA/TEGDMA resin. Scanning electron microscopy analysis results showed homogeneous distribution of fluorinated nanoparticles in the dental composite. X-ray diffraction studies have shown how the amorphous character of the materials changes with the addition of fluorine-containing silica nanoparticles to the structure. The thermal properties of the materials were determined by TGA. Chemical stability, water absorption, and dissolution experiments of fluorine-containing silica-based dental materials were also carried out. In addition, the antimicrobial properties of dental materials were determined by MIC tests using bacteria and fungi.

Key Words: Fluorine, Silica, Dental composites

DRAFT

ORAL PRESENTATION

Biyopriming tekniği ve stres toleransı üzerine etkisi

Ayşe Feyza Tufan Dülger[1*] (*https://orcid.org/0000-0003-4779-6811*)

[*1]Ondokuz Mayıs Üniversitesi, Ziraat Fakültesi, Tarımsal Biyoteknoloji Bölümü, Samsun, Türkiye

*Sorumlu yazar e-mail: feyzadulger@omu.edu.tr

Özet

Başarılı bir tarımsal üretimde, tohumdan yüksek verimin elde edilmesi tohumun hızlı ve üniform bir şekilde çimlenmesine ve güçlü fide gelişimi göstermesine bağlıdır. Çeşitli biyotik/abiyotik stres faktörlerine veya doğrudan tohum kalite ve yapısına bağlı olarak tohumun çimlenmesi sırasında sorunlar yaşanabilmektedir. Tohumların ekilmesinden önce tohumlara yapılan çeşitli uygulamalar priming olarak adlandırılmaktadır. Temel olarak priming, tohumda çimlenme için gerekli metabolik aktiviteyi başlatacak, ancak tohumun tam çimlenmeye geçişine imkân tanımayacak seviyedeki kontrollü su alımıdır. Priming uygulamasından sonra tohumlar yıkanmakta ve ardından kurutulmaktadır. Bu şekilde, priming, çimlenme öncesi metabolizmayı harekete geçirmektedir. Bu durum, DNA onarım yolaklarını ve reaktif oksijen türlerini süpürme sistemlerini aktive eder ve ayrıca genom bütünlüğünün korunmasına yardımcı olur. Çimlenme ile ilgili birçok aktivitenin başlatılmasını da kolaylaştırır. Priming yaklaşımlarından biri olan biyopriming tohumların faydalı mikroorganizmalar ve biyokontrol ajanlarıyla aşılanması yoluyla biyotik/abiyotik streslere tolerans sağlayan bir yaklaşımdır. Biyopriming sırasında, mikroorganizmalardan gelen uyarı algılanır ve bitkide fizyolojik, transkripsiyonel, metabolik ve epigenetik değişiklikler meydana gelir. Bu durum, stres toleransı ile sonuçlanan daha yüksek savunma tepkilerine neden olur. Biyopriming uygulamalarında yaygın olarak *Trichoderma, Enterobacter, Pseudomonas* ve *Bacillus* cinslerine ait türler kullanılmaktadır. Biyopriming tekniğinde kullanılan bazı mikroorganizmalar ayrıca bitki büyümesini doğrudan ve dolaylı olarak teşvik edebilir. Biyopriming'in diğer priming uygulamalarına kıyasla üstün olan özelliği, tohumun hem çimlenme hızı ve üniformitesi üzerine daha yoğun bir etki göstermesi hem de toprak ve tohum kökenli patojenlerin baskılanmasını sağlamasıdır. Tohumlar şekil, boyut ve tohum kabuğunun doğası bakımından farklılık gösterdikleri için biyopriming uygulamasında, kullanılan tohuma göre mikrobiyal konsantrasyon ve sürenin optimize edilmesi önemlidir. Strese toleransta biyopriming yaklaşımı düşük konsantrasyonda aşılamanın yeterli olması, ekonomik olması ve çevre dostu bir yaklaşım olmasıyla ön plana çıkmaktadır. Bu derlemede, biyopriming tekniğinin bitkide yol açtığı fizyolojik ve moleküler değişiklikler ile stres toleransını iyileştirmesindeki rolüne değinilecektir.

Anahtar Kelimeler: Tohum, biyopriming, abiyotik stres, biyotik stres

DRAFT

ORAL PRESENTATION

The impact of antitranspirants on grain yield and photosynthetic characteristics of two wheat cultivars during water deficit stress

Somayyeh Razzaghi[1](0000-0002-8028-452X), Mohammad Rezaei[2](0000-0002-3684-2053), Fatma Nur Kılıç[3](0000-0003-3498-2455)
[1,3]Department of Soil Science and Plant Nutrition, Agricultural Faculty, Erciyes University, Kayseri, Turkey
[2]Seed and Plant Improvement Research Department, West Azerbaijan Agricultural and Natural Resources Research Center, AREEO, Urmia, Iran
Corresponding author E-mail address: srazzaghi@erciyes.edu.tr

Abstract

This study presents the results of a split-plot experiment conducted at Miandoab Agriculture Research Station, West Azarbaijan, during the 2015-2016 years. The study aimed to assess the effects of antitranspirants on several physiological traits of wheat, including photosynthesis, total chlorophyll content (chlorophyll *a* and chlorophyll *b*), grain protein percentage, grain yield, transpiration rate, and photosynthetic water use efficiency, under water deficit stress conditions. The experiment utilized a randomized complete block design with three replications. The main plots were subjected to different irrigation treatments, involving complete irrigation, irrigation withholding at heading stages, and irrigation withholding at flowering stages. Within each main plot, subplots were allocated for foliar applications of distinct antitranspirants, namely Prometrin, Kaolin, Tmatrom, Chitosan, and a control group. Additionally, two wheat cultivars, Zarin and Mihan, were evaluated to study cultivar-specific responses. The findings revealed significant effects of the antitranspirants on various physiological characteristics of wheat under drought stress. Specifically, the application of prometrin during irrigation withholding at the flowering stage resulted in increased photosynthesis at 2 µmol CO2 m^{-2} s^{-1} compared to the control. Chitosan treatment at irrigation withholding during the heading stage exhibited the highest total chlorophyll content (4.69 µg/g FW), while chitosan treatment also demonstrated the highest chlorophyll content (2.42 µg/g FW). In the Zarin cultivar, chitosan application during irrigation withholding at the heading stage showed the highest chlorophyll *b* content. Furthermore, chitosan treatment during irrigation withholding at the flowering stage significantly increased the protein content (12.27 mg/g DW) and enhanced the yield by 868 kg/ha in the Mihan cultivar compared to the control. Notably, the lowest transpiration rate was observed during the irrigation withholding stage (3.83 mM H2O m-2 s-1). Overall, the results highlighted the importance of considering the timing of stress and the type of cultivar in assessing the effects of antitranspirants. Chitosan, in particular, showed promising potential for improving the photosynthetic properties of wheat (Mihan cultivar) and enhancing seed yield under drought stress conditions.

Keywords: Low irrigation, Chitosan, Chlorophyll, Protein, Wheat Cultivars, Grain Yield

DRAFT

ORAL PRESENTATION

Combined treatment of sprouting and high intensity ultrasound to produce functionalized hemp protein

Gülşah Karabulut[1*]

[*1] Department of Food Engineering, Sakarya University, 54187, Sakarya / Turkiye

*Corresponding author e-mail: gkarabulut@sakarya.edu.tr

Abstract

In recent years, hemp protein has garnered significant attention for its exceptional nutritional content and high digestibility. However, its industrial applications have been limited due to poor solubility attributes, similar to other plant-based protein sources. To address this issue, a biotechnological approach using sprouting (1, 3, 5 days) was employed to enhance the physicochemical, functional, and bioactive properties of hemp protein. Additionally, high-intensity ultrasound was applied in combination with these treatments for further modifications. The results of the study showed promising outcomes. Protein recovery in the defatted flour increased by an impressive 42% in germinated isolates compared to the untreated isolate. Notably, the particle sizes of the isolates were reduced by a remarkable 83%, leading to a significant increase of 133% in solubility for the sprouted and ultrasonicated isolates. The improved functionality of the modified isolates was also evident in their enhanced water, oil absorption capacities, and emulsion activities, which saw increases of 103%, 194%, and 300%, respectively, when compared to the untreated isolate. Overall, the combination of sprouting and ultrasonication proved to be promising, eco-friendly, and safe biomodification techniques for effectively improving the functionality of hemp protein. These modifications resulted in clean-labeled proteins that can be utilized as bioactive agents in the food industry, offering a valuable alternative for various applications.

Keywords: Biotechnology, functional, sprouting, proteolysis, hemp protein.

DRAFT

ORAL PRESENTATION

Evaluation of the Cytotoxic Effects of Epigallocatechin Gallate and Cetuximab in Lung Cancer Cells

Ayse Erdogan[1]* (ORCID: https://orcid.org/0000-0003-0843-173X)

*[1]Alanya Alaaddin Keykubat University, Faculty of Engineering, Genetic and Bioengineering Department, Alanya, Antalya, Turkey.

*Corresponding author e-mail:ayse.erdogan@alanya.edu.tr

Abstract

Lung cancer is the most common type of cancer all over the world. Chemotherapy is one of the most effective therapeutic modalities to treat a variety of tumors, including lung cancer. Due to drug resistance and serious side effects of chemotherapy, great difficulties are encountered in getting a response from the treatment. Therefore, it is very important to find new therapeutic agents of natural origin that will increase the therapeutic efficacy of targeted chemotherapeutics such as cetuximab. Therefore, to reduce the side effects of cancer drugs, attention has been paid to plant-derived molecules that are non-toxic to humans and have therapeutic effects and biological activities. Epigallocatechin-3-gallate (EGCG) is a compound, one of the most prominent catechins in green tea leaves, that attracts a lot of attention in medical research.

In our study, 3-(4,5-dimethylthiazol-2- yl)-2,5-diphenyltetrazolium bromide (MTT) and Lactate dehydrogenase (LDH) activity tests were performed to demonstrate the cytotoxic effect of cetuximab alone and also with EGCG in cells. It was observed that cell viability of A-549 cells exposed to different concentrations of cetuximab and EGCG for 48 hours decreased depending on the concentration increase. It was determined that the most effective combination concentrations were IC_{20} cetuximab and IC_{10} EGCG by calculating the cell viability and combination index (CI) value in cells where cetuximab ($<IC_{50}$) and EGCG ($<IC_{50}$) were administered together.

Lactate dehydrogenase (LDH) is the enzyme that converts lactic acid to pyruvic acid in the presence of nicotinamide adenine dinucleotide (NAD^+). Lactic acid occurs with disruption of cell integrity. EGCG and cetuximab were applied to the cells and it was found that the LDH activity increased compared to the control.

Keywords: Cetuximab, combined therapy, epigallocatechin-3-gallate

DRAFT

ORAL PRESENTATION

Design and synthesis of a new chemiluminogenic sensor

Ömer SONKAYA[1*] (*ORCID: https://orcid.org/ 0000-0001-9432-8338*)

[*1] Aksaray University, Technical Sciences Vocational School, Department of Chemistry, 68100, Aksaray, Turkey.

*Corresponding author e-mail: omersonkaya@gmail.com

Abstract

Cancer is a large group of diseases that can grow abnormally and spread beyond its normal limits to other parts of the body. Various methods are used in the treatment of cancer. Surgery is very effective in removing tumors. However, these are limited to low-stage tumors and cannot be adequately applied to high-stage tumors. However, even with chemotherapy, there is no guarantee of recovery. Photodynamic therapy (PDT) is a successful and clinically approved treatment method used for the treatment of many diseases from past to present. Although there are FDA-approved drugs used in PDT, it is not at the desired level and there has been an increasing interest in new PDT agents recently. The number of efficient organic photosensitizers has been limited. Therefore, there is a strong demand for the development of new photosensitizers. The research in this area is mainly focused on organic dyes that absorb light in the far red or near infrared region of the electromagnetic spectrum, where the light tissue penetration is relatively high.
Within the scope of this study, the synthesis and characterization of fluorescence-based compound that will enable cancer treatment were carried out. On the one hand, a chemiluminescent unit that will be sensitive to reactive oxygen species (ROT) in the design; On the other hand, compounds that simultaneously contain fluorescent dyes suitable for bioimaging and FDT with their superior properties were synthesized. The structures of the synthesized fluorescent dyes were elucidated by spectroscopic methods and their potential as photosensitizing agents was determined.

Keywords: Near Infrared Zone, Chemilumingic Theraneustic, Luminescence, Photothermal Therapy, Photodynamic Therapy, Singlet oxygen

DRAFT

ORAL PRESENTATION

Kepek otu bitkisinden (*Paronychia sp.*) Tip I ribozom inaktive edici proteinin (RIP I) saflaştırılması ve karakterizasyonu

Ali ZEYTÜNLÜOĞLU[1*] (*ORCID: https://orcid.org/0000-0002-2534-7241)*
Figen ZİHNİOĞLU[2] (*ORCID: https://orcid.org/0000-0001-8216-7004*)

[*1] University of Pamukkale, Denizli Vocational School of Technical Sciences, Department of Chemistry and Chemical Processing Technologies, Denizli, Türkiye.
[2] University of Ege, Faculty of Science, Department of Biochemistry, İzmir, Türkiye.

*Corresponding author e-mail: azeytun@pau.edu.tr

Özet

Kanser; kalp damar hastalıklarından sonra tüm dünyada sayısı giderek artan, bazen sinsice bazense belirgin semptomlar halinde kendini gösteren çağımızın en amansız ölümcül hastalıklarından biridir. Son yıllarda; kanser tedavilerinde hedefe yönelik ilaçların olumlu etkilerinin tespit edilmesi, bu alanda yapılan araştırmalara hız kazandırmıştır.

Ribozom inaktive edici proteinler (RIP'ler); toksine dayalı kanser tedavilerinde kullanılmakta olan önemli bir sitotoksik ajan grubudur. Ribozom inaktive edici proteinlerin antikorlar ile oluşturulan immunokonjugatları, endositozla hücre içine alınmakta ve kanser hücresinin ölümüne neden olmaktadırlar. Bu grup içerisinde özellikle tip I RIP'lerin; daha düşük toksisite göstermeleri, direkt olarak hedef hücreye bağlanmaları, daha fazla stabil olmaları, hazırlanmalarının daha güvenli oluşu, çok fazla sayıda aktif konjugat oluşturabilme yetenekleri toksine dayalı kanser terapilerinde, onlara yüksek kullanılabilirlik potansiyeli sunmaktadır.

Bu çalışma kapsamında; kepek otu olarak bilenen Paronychia sp. (Illecebraceae) bitkisi tohumlarından, kanser tedavilerinde kullanılabilecek bir ribozom inaktive edici proteinin saflaştırılması gerçekleştirildi. Ribozom aktive edici proteinin saflaştırılması işlemlerinde; amonyum sülfat çöktürmesi, katyon değişim ve jel filtrasyon kromatografisi adımları uygulandı. Ribozom inaktive edici protein % 1,6 verimle 66 kat saflaştırıldı Saflaştırma adımlarının etkinliği N-glukosidaz aktivitesi ile değerlendirildi. Saflaştırılan proteinin karakterizasyonunda; SDS-PAGE ile molekül kütlesi 5.2 kDa olarak belirlendi. Yapı analizi MALDI-TOF-MS ile gerçekleştirilerek uzunluğu 7 ila 13 arasında değişen 6 farklı peptid dizisi tanımlandı. Ribonükleaz aktivitesi 9,04 U/mg olarak tespit edildi.

Keywords: Kanser, ribozom inaktive edici protein, saflaştırma, *Paronychia*

DRAFT

ORAL PRESENTATION

Evaluation of antimicrobial and antioxidant activities of propolis samples prepared with different solvents

Emine Sönmez* (*ORCID: https://orcid.org/0000-0003-4418-5599*)

* Düzce University, Beekeeping Research Development and Aplication Centre, 81620 Düzce, Turkey.

*Corresponding author e-mail:eminesonmez@duzce.edu.tr

Abstract

Due to its natural bioactive components, propolis has been used in the treatment of many diseases since ancient times. Since porpolis has a complex structure, it is necessary to extract bioactive substances from it. The extraction technique and solvent used in the extraction process are very important. In this report, antioxidant and antimicrobial activity rates of propolis samples collected from Gümüşhane were tested using two different solvents (ethanol and glycol). Total Phenolic (TP) – Flavonoid (TF) contents of the samples were calculated with using Folin-Ciocalteu method and colorimetric aluminum chloride assay respectively. Antioxidant activity was measured by the FRAP method. The antimicrobial activity levels of the samples were tested against 6 different microorganisms using the agar well diffusion method. TP and TF values of ethanolic extraction (72.04 mg GAE/mL-39.41 mg QE/mL) were significantly higher than glycolic extraction (52.15 mg GAE/mL-27.23 mg QE/mL). Stronger antioxidant activity was obtained from ethanolic extract (484.15 mM Troloks/mL). Although the zone diameters obtained from the agar well method varied between 7 and 17 mm, it was observed that ethanolic extraction was more effective against selected microorganisms. The most susceptible strain was *Staphylococcus epidermidis* for both extraction. As a result, it was determined that both antioxidant and antimicrobial effects of ethanol, which is the most widely used in propolis extraction, were higher than glycol, but the biological effect of multiple alcohol derivative glycol was undeniably important. In conclusion it is thought that testing different extraction methods and solvents of propolis, which is promising for drug discoveries that can be used in the treatment of some infectious diseases, will contribute to the science of pharmacology.

Keywords: Propolis, solvent, antioxidant, antimicrobial.

DRAFT

ORAL PRESENTATION

The Effect of Lead on Some Ecophysiological and Molecular Parameters of *Cucurbita moschata* Duch.

Kübra SEVGİ[1*] (*ORCID: https://orcid.org/0000-0001-8382-2162*), Sema LEBLEBİCİ[2] (*ORCID: https://orcid.org/0000-0002-3762-6408*)

[*1]Bilecik Seyh Edebali University, Graduate Education Institute, Department of Molecular Biology, Bilecik, Turkey.
[2] Bilecik Seyh Edebali University, Faculty of Science, Department of Molecular Biology and Genetics, Bilecik, Turkey.

*Corresponding author e-mail: kubrassevgi@gmail.com

Abstract

Nowadays, rapid industrialization and urbanization have caused a serious increase in heavy metal pollution, which is one of the important ecological problems. Due to the their sessile nature, plants are among the most affected organisms by heavy metal pollution. Heavy metal stress negatively affects the growth and development of plants, leads to losses in crop yield and quality, and threatens the life of other living things through the food chain. Some heavy metals are essential to plants at low levels, while others are toxic even at low levels. Lead, one of the most toxic heavy metals, is not necessary for the growth of plants and has toxic effects on plants even at low concentrations. On the other hand, plants have developed effective defense mechanisms to survive in areas contaminated with heavy metals.
In this study, the ecophysiological and molecular responses of the pumpkin plant exposed to two different lead concentrations (25 and 50 mM Pb) were investigated. With some exceptions, it was determined that the ecological parameters of stem and root increased at 25 mM Pb concentration compared to control, while these ecological parameters decreased at 50 mM Pb concentration. It was observed that chlorophyll *a*, *b* and total chlorophyll amounts increased in both applied lead concentrations compared to the control, but MDA content increased only at 50 mM Pb concentration. It was noted that SOD and CAT activities and gene expressions of these enzymes increased at both lead concentrations compared to the control, while APX activity and gene expression increased only at 50 mM Pb concentration. The results show that pumpkin is highly tolerant to lead.

Keywords: Heavy metal stress, lead, pumpkin, antioxidant enzyme, gene expression

DRAFT

ORAL PRESENTATION

Chemistry of Michael adducts of nitroolefins: Unexpected *N*-acetylation products

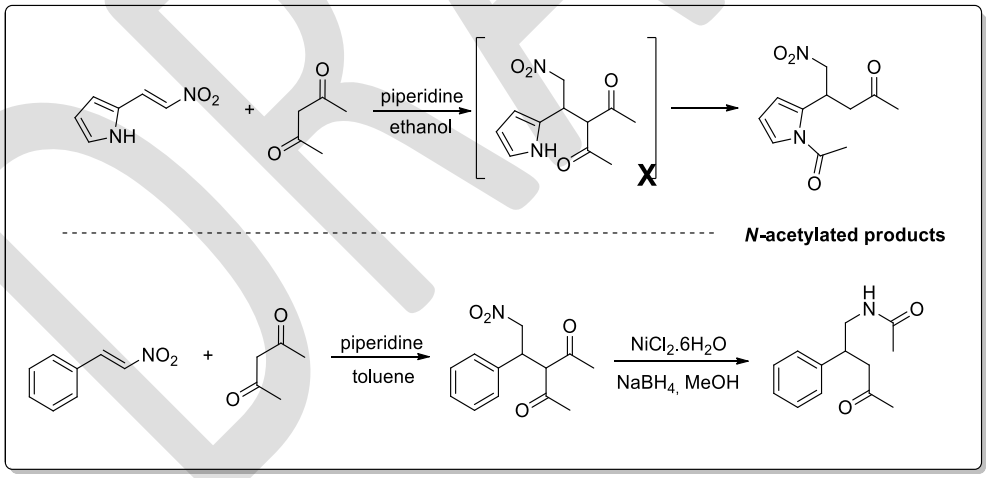
Seda Çınar[1,2*] (*ORCID: https://orcid.org/ 0000-0001-6591-7141*)

[*1]Ankara Hacı Bayram Veli University, Polatlı Faculty of Science and Letters, Chemistry Department, Ankara, Türkiye
[2]Hacettepe University, Faculty of Science, Chemistry Department, Ankara, Türkiye

*Corresponding author e-mail: seda.cinar@hbv.edu.tr, sedacnr@gmail.com

Abstract

N-acetylation reactions are widely used in synthetic chemistry, biology, pharmaceutical industries, and drug research. *N*-Acetylated products of organic reactions with acetylation reagents contain an amide bond, which is crucial in medicinal chemistry. Acetylation is generally accomplished with acetic anhydride or acetyl chloride. Beyond these relatively disadvantageous reagents, selective acetylation reactions including thioacids, in situ formed ketene intermediate, ethyl acetate, or *N*-acetyltransferase have been reported. These works provided intermolecularly acetylation reaction mechanisms via two individual reagents or substances.
Michael addition reaction is one of the most useful methods to form carbon-carbon bonds. Active methylene and nitroolefin compounds are commonly used in this method due to their conversion abilities to other functional groups. This functionality enables the formation of biologically active heterocyclic compounds. A typical study for the further reactions of addition products is the intramolecular or intermolecular cyclization reactions. In these reactions, generally, pyrrole, lactam, pyrolizine, pyrazole, and isoxazole derivatives were obtained. Change in starting materials or catalysts can result unexpectedly by means of product formation.
In this study, Michael addition of acetylacetone as an active methylene compound to nitroolefins was investigated. Then, with the addition products in hand, their chemistry in the presence of catalysts was studied and as an interesting outcome, *N*-acetylated products were obtained. Structures of the isolated compounds were clarified using ^{1}H NMR, ^{13}C NMR, and HR-MS techniques.



Keywords: Nitroolefins, *N*-acetylation, Reduction

DRAFT

ORAL PRESENTATION

Methylene blue adsorption on sodium dodecyl sulfate modified chalcopyrite

Maha Abobakr Yahya Almezgagi[1] (*ORCID: https://orcid.org/0000-0002-6253-6826*),
Gülcihan Güzel Kaya[1*] (*ORCID: https://orcid.org/0000-0003-2753-7724*),
Hüseyin Deveci[1] (*ORCID: https://orcid.org/0000-0002-1103-7234*)

[1]Konya Technical University, Faculty of Engineering and Natural Sciences, Department of Chemical Engineering, Konya, Turkey.

*Corresponding author e-mail: ggkaya@ktun.edu.tr

Abstract

$CuFeS_2$ known as chalcopyrite is one of the most abundant minerals in nature. $CuFeS_2$ consists of tetragonal structure in which Fe and Cu ions coordinate with S ions in the crystal lattice [1]. Due to its benefits, $CuFeS_2$ is widely used as a precursor for Cu smelting, an anode material, an adsorbent for heavy metal removal, and so on [2, 3]. In this study, $CuFeS_2$ modified with sodium dodecyl sulfate was utilized as an adsorbent for methylene blue adsorption from aqueous solutions. First, adsorption parameters including pH, contact time, adsorbent content and temperature were optimized. No important effect of pH and adsorbent content on the methylene blue adsorption was observed. While the methylene blue removal was about 60% at room temperature, a decrease in the methylene blue removal was determined with increasing temperature. In the optimum conditions, methylene blue adsorption isotherms and kinetics were examined. It was clear that the adsorption data were well fitted to Freundlich isotherm model and pseudo-second-order kinetic model. In the light of the results, the chalcopyrite can be evaluated as a potential adsorbent for dye adsorption processes.

Keywords: Chalcopyrite, surface modification, methylene blue adsorption, adsorption kinetics

[1] Mikhlin YL, Tomashevich YV, Asanov IP, Okotrub AV, Varnek VA, Vyalikh DV. Spectroscopic and electrochemical characterization of the surface layers of chalcopyrite ($CuFeS_2$) reacted in acidic solutions. Applied Surface Science 2004; 225 (1): 395-409. doi: https://doi.org/10.1016/j.apsusc.2003.10.030.
[2] Ding Z, Yang C, Zhang H, Mei J, Wang J, Yang S. New utilizations of natural $CuFeS_2$ as the raw material of Cu smelting for recovering Hg^0 from Cu smelting flue gas. Fuel 2023; 341: 126997. doi: https://doi.org/10.1016/j.fuel.2022.126997.
[3] Zhou J, Jiang F, Li S, Xu Z, Sun W, Ji X, et al. $CuFeS_2$ as an anode material with an enhanced electrochemical performance for lithium-ion batteries fabricated from natural ore chalcopyrite. Journal of Solid State Electrochemistry 2019; 23 (7): 1991-2000. doi: 10.1007/s10008-019-04284-8.

DRAFT

ORAL PRESENTATION

Assessing the environmental impact of functional foods

Abuzer Çelekli[1,2] (*ORCID: https://orcid.org/0000-0002-2448-4957*),
Özgür Eren Zariç[1*] (*ORCID: https://orcid.org/0000-0001-5293-871X*)

[1]Gaziantep University, Environmental Research Center (GÜÇAMER), Gaziantep, Turkey.

*Corresponding author e-mail: ozgurerenzaric@gmail.com

Abstract

The emergence and significance of functional foods have drawn attention to their production's ecological consequences. Integrating sustainability in the production process of these foods, known for their positive effects on human health, is critical. A mixed-method approach was employed, utilizing quantitative analysis of industry data and qualitative evaluation through an extensive literature review. The study encompassed various stages, including raw material sourcing, production, processing, packaging, transportation, and consumer behavior. Findings indicate that functional food production may require higher energy and water resources, but advantages were observed in waste management and transportation stages. The study reveals a multifaceted relationship between functional food production and environmental sustainability. Innovative technologies and environmentally friendly methods present potential solutions to mitigate adverse effects. Collaboration within the industry and with regulators, researchers, and consumers is imperative for a holistic approach to environmental stewardship. This research underscores the importance of considering the environmental impacts in the burgeoning field of functional foods, highlighting their increasing significance in human nutrition and health. It calls for targeted strategies, guided by the complexity and variability observed, emphasizing the need for future research, effective policy formulation, and industry-wide sustainable practices. The proliferation of functional health foods is necessary, and continued research is required to facilitate their widespread adoption. The findings provide a basis for further exploration and a pathway towards an environmentally responsible approach in the production of functional foods.

Keywords: Environmental impact, Functional foods, Sustainable production.

DRAFT

ORAL PRESENTATION

Catalytic effect of Zr loaded ZSM-5 doped Beta zeolite catalyst on the transalkylation of 1-methylnaphthalene with toluene for 2,6-DMN and p-xylene synthesis

Abdul Aziz ABDULLAH[1*] (*ORCID: https://orcid.org/0000-0003-1860-0664*),
Ali KARADUMAN[2] (*ORCID: https://orcid.org/0000-0003-1061-8288)*

[*1]Ankara University, Department of Chemical Engineering, Tandoğan 06100 Ankara, Türkiye.
[2]Ankara University, Department of Chemical Engineering, Tandoğan 06100 Ankara, Türkiye.

*Corresponding author e-mail: AbdullahAbdulAziz999@yahoo.com

Abstract

2,6-Dimethylnaphthalene (2,6-DMN) serves as a crucial intermediate in the synthesis of Polyethylene naphthalate (PEN) and offers promising applications due to its favourable thermal, mechanical, and gas barrier properties. Unfortunately, the expensive multi-step industrial synthesis of 2,6-DMN limits PEN production and its potential applications. Meanwhile, p-xylene, constituting over 80% of the global xylene demand, holds significant importance as a material for terephthalic acid production.

The primary objective of the study was to address the challenges in 2,6-DMN synthesis while concurrently producing valuable p-xylene. To achieve this, unmodified, and Zr-impregnated ZSM-5 doped Beta zeolite catalysts were prepared and utilized in the transalkylation of 1-Methylnaphthalene with toluene. The prepared zeolite catalysts were characterized through BET, SEM, FTIR, Py-FTIR, XRD, and XRF techniques. The catalytic studies were conducted at atmospheric pressure in a fixed-bed continuous flow reactor at varying WHSV (Weight Hourly Space Velocity) between 1 – 3 hr^{-1}, and temperatures ranging from 400 – 500°C, employing three different metal loadings.

The findings revealed that metal impregnation improved zeolite's physical properties and acidity. In the catalytic tests, zeolite activity, in terms of conversion, yield, and selectivity, generally increased with increasing temperatures but decreased with increasing WHSV. Notably, Zr impregnation led to substantial improvements in both 2,6-DMN and p-xylene yield and selectivity. The Zr-loaded catalysts demonstrated enhanced performance, yielding 3.8% 2,6-DMN and 11.7% p-xylene compared to 1.83% and 10.7%, respectively on unmodified zeolite catalysts. Furthermore, the Zr impregnation increased the 2,6-DMN/2,7-DMN ratio in unmodified zeolites from 1.6 to 1.9. The study provides insight that demonstrates that Zr loading enhances catalyst properties by offering alternative pathways to achieve higher yields and selectivity.

Keywords: Transalkylation, 1-Methylnaphthalene, Zeolite catalyst, Toluene, 2,6-DMN

DRAFT

ORAL PRESENTATION

Lathyrus annuus L., *L. hierosolymitanus* Boiss. ve *L. hirsutus* L. türlerinin toplam fenolik ve flavonoid içerikleri ve antioksidan özellikleri

Bekir YILDIRIM[1*] (*ORCID: https://orcid.org/0000-0003-2378-7697*)

[1]Burdur Mehmet Akif Ersoy Üniversitesi, Burdur Gıda Tarım ve Hayvancılık Meslek Yüksekokulu, Bitkisel ve Hayvansal Üretim Bölümü, Burdur, Türkiye

*Sorumlu yazar e-mail: bytr33@yahoo.com

Özet

Fabaceae familyasında bulunan *Lathyrus* L. ekonomik açıdan önemli bir bitki cinsidir. Dünyada 200'den fazla taksonu yayılış gösteren cins, Türkiye'de tür, alt tür ve varyete seviyesinde 79 taksonla temsil edilmektedir. Cinsin bazı türleri dünyanın değişik yerlerinde hayvan yemi veya insan gıdası olarak değerlendirilmektedir. Bazı *Lathyrus* türlerinin güçlü antioksidan etkileri olan fenolik bileşiklerin kaynağı olabileceği belirtilmiştir. Bitkilerin her yerinde bulunabilen sekonder metabolitler olan fenolik bileşikler çiçeklere renk vermenin yanında bitkileri UV ışınlar, böcekler, otçullar ve patojenlere karşı da korur. Araştırmalar insan diyetinde en çok bulunan antioksidanlar olduğu düşünülen fenolik bileşiklerin birçok hastalığı önlemede etkili olduğunu göstermiştir. Son zamanlarda biyoaktif bileşikler ve biyoaktivite konularında çalışılan bitki cinslerinden birisi de *Lathyrus*'tur. Bu çalışmanın materyalleri de *L. annuus* L., *L. hierosolymitanus* Boiss., ve *L. hirsutus* L. türleridir. Örnekler çiçeklenme döneminde doğal habitatlarından toplanmış, tekniğine uygun olarak kurutulmuş, öğütülerek toz haline getirilmiş ve değişik çözücüler (aseton, etanol, su) kullanılarak ekstreleri hazırlanmıştır. Daha sonra ekstrelerin toplam fenolik ve flavonoid içerikleri (TPC, TFC) belirlenmiş, serbest radikal (DPPH: 2,2-diphenyl-1-picrylhydrazyl, ABTS: 2,2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)) süpürme ve indirgeyici güç (RP: reducing power) testleri uygulanarak antioksidan özellikleri ortaya konulmuştur. En yüksek TPC değeri aseton ekstreleri için *L. hierosolymitanus*'ta, etanol ve su ektreleri için *L. hirsutus*'ta, en yüksek TFC değeri ise aseton ekstreleri için *L. hirsutus*'ta, etanol ve su ekstreleri için ise *L. hierosolymitanus*'ta tespit edilmiştir. TPC ve TFC değerleri bakımından en düşük içerikler aseton ve etanol ekstreleri için *L. annuus*'ta, su ekstreleri için sırasıyla *L. hierosolymitanus* ve *L. hirsutus*'ta belirlenmiştir. En iyi antioksidan aktivite gösteren tür; DPPH radikali süpürme testlerinde aseton, etanol ve su ekstreleri için sırasıyla *L. hierosolymitanus, L. hirsutus* ve *L. annuus*; ABTS radikali süpürme testlerinde ise aseton ve su ekstreleri için *L. hierosolymitanus*, etanol ekstreleri için ise *L. annuus* olmuştur. İndirgeyici güç testlerinde aseton ekstrelerinde *L. hirsutus*, etanol ve su ekstrelerinde ise *L. hierosolymitanus* en yüksek antioksidan aktivitiye sahiptir.

Anahtar Kelimeler: ABTS, Antioksidan aktivite, DPPH, *Lathyrus*, Toplam fenolik içerik

DRAFT

ORAL PRESENTATION

Synthesis, characterization and *in silico* studies of some new compounds bearing thiadiazole ring

Sena DEMİRARAN[1,2*] (ORCID: https://orcid.org/0000-0001-8030-3471),
Fatih TOK[3] (ORCID: https://orcid.org/0000-0002-4569-008X)

[*1]Duzce University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, Düzce, Türkiye.
[2]Marmara University, Institute of Health Sciences, İstanbul, Türkiye.
[3]Marmara University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, İstanbul, Türkiye.

*Corresponding author e-mail: demirrnsena@gmail.com

Abstract

Thiadiazoles are one of the most preferred heterocyclic structures in new drug discovery studies. They exhibit many pharmacological and biological effects such as antioxidant, anti-inflammatory, anticancer, antidiabetic, antimicrobial. The pharmacophore structure responsible for the biological effect is -N=C-S- and plays an important role in the interaction with biological membranes. There are many molecules with thiadiazole structure such as acetazolamide, methazolamide, sulfamethizole, megazol and cefazolin, which are drugs on the market or in phase studies. In this study, the synthesis of compounds with 1,3,4-thiadiazole structure was carried out in three steps. In the first step, hydrazide synthesis was carried out from the starting material containing ester structure on pyridine ring. Then this hydrazide structure was reacted with various aromatic or aliphatic isothiocyanates to obtain thiosemicarbazides. In the last step, 1,3,4-thiadiazole structures were obtained in high yields by cyclization of thiosemicarbazides in acidic medium. It was aimed to elucidate the structures of the synthesized compounds by various spectral methods such as IR, ^{1}H-NMR, ^{13}C-NMR and elemental analysis.
After the structural characterization of the compounds was completed, it was aimed to examine the in silico ADME properties. The physicochemical and pharmacokinetic properties of the synthesized compounds were investigated by SwissAdme software. It was found that the compounds were in full compliance with the rules such as Lipinski, Veber, Egan, Muegge and Ghose, which are expressed as drug-like properties. In addition, it was predicted that the passage of the compounds through the gastrointestinal system was high and the blood brain barrier passage was weak. The low blood brain barrier passage is a positive feature in terms of the lack of central side effects of the compounds. The solubility of the compounds was found to be moderately. Therefore, the compounds were found to have high drug candidate properties.

Keywords: Thiosemicarbazide, 1,3,4-thiadiazole, synthesis, ADME.

DRAFT

ORAL PRESENTATION

Development and application of a new electrochemical herbicide sensor based on molecular imprinting

Sezen İrem Aslaner[1*] (*ORCID: https://orcid.org/0000-0001-8163-7078*),
Ayça Demirel ÖZEL[1] (*ORCID: https://orcid.org/0000-0002-7129-0370*)

[1]Ankara University, Faculty of Science, Department of Chemistry, Ankara, Turkey.

*Corresponding author e-mail: iremkaftanoglu@gmail.com

Abstract

Pesticides have shown great benefits in increasing agricultural and industrial production. Rapid population growth and industrialization have increased the demand for the use of pesticides as a necessity in terms of obtaining food products and other agricultural resources to meet the world population [1]. With the development of intensive agricultural practices, widely use of pesticides emerged environmental pollution that has gained great interest as a new task of current research for chemists.
Glyphosate (Gly) as an herbicide, is used to reduce the effects of weeds and pests. Nonetheless, Gly exposure can cause permanent damage to human health [2]. Therefore, it is important to monitor the environment and reduce public health risks. Many time-consuming, expensive instrumental analysis methods requiring experienced staff have been proposed [3]. Based on previously reported studies, the preparation of pesticide sensors by molecular imprinting technology which offers advantages such as ease of fabrication and use, high sensitivity and selectivity, rapid response, and low cost, has been studied with increasing demand in recent years [4]. Among them, molecularly imprinted polymer based Gly sensors are very few. Therefore, it is aimed to prepare a new molecularly imprinted polypyrrole Gly sensor with some modifications compared to similar studies.
Gly imprinted polypyrrole films were electrochemically synthesized on the Pt plate electrode at optimum pH 5.0 PBS buffer media in the presence of functional monomer, template molecule and $LiClO_4$ as supporting electrolyte by cyclic voltammetry in a potential range of -0.1- +1.4 V with a scan rate of 50 mV/s. The selectivity of the electrode was tested in the presence of chlorpyrifos, aldicarb, phosmet, aminomethylphosphonic acid and glufosinate. LOD (1.98×10^{-6} fM) and the linear working range (4.98×10^{-4}-0.26 fM) were calculated by DPV. The proposed MIP sensor was successfully employed in tap water and orange juice samples.

Keywords: Electrochemical sensors, Molecular imprinting, Polypyrrole, Pesticide.

References:
[1] F. Ganjeizadeh Rohani, A. Mohadesi, and M. Ansari, *ChemistrySelect*, **2019**, 4(42), 12236-12244.
[2] O. D. Ogunbiyi, D. O. Akamo, E. E. Oluwasanmi, J. Adebanjo, B. A. Isafiade, T.J. Ogunbiyi, and P. O. Oladoye, *Groundwater for Sustainable Development*, **2023**, 100961.
[3] X. Ren, H. Zeng, Q. Zhang, H. Cai, and W. Yang, *Int. J. Electrochem. Sci.*, **2022**, 17(12), 221292.
[4] P. Leepheng, D. Limthin, W. Homchan, S. Suramitr, and D. Phromyothin, *Jpn J. Appl. Phys.*, **2020**, 59(SI), SIIJ09.

DRAFT

ORAL PRESENTATION

GENZPRO Development Kit Used in GENZPRO© Mini Chip Auto Device Working with Capillary ELISA Technique, Providing Development of Rapid Protein Tests

Umut Ağyüz[*] (*ORCID: https://orcid.org/0000-0002-1242-0454*), Caner Demir[1] (*ORCID: https://orcid.org/0000-0001-9460-2333*), Aybüke Üretmen[1] (*ORCID: https://orcid.org/0009-0009-2681-6001*)

[*1]Genz Biotechnology, İstanbul, Turkey.

*umut@genzbio.com

Abstract

GENZPRO© Minicihip Auto is an automatic device that works with the capillary ELISA technique developed by our company. Capillary ELISA works by transporting the sample through a capillary and reacting with the analyte. In this method, proteins are immobilized inside a capillary tube. The capillary ELISA technique has been optimized to achieve faster results compared to traditional ELISA methods. It also requires less sample and reagent usage and can be integrated with automated instruments, making the analysis process easier.
GENZPRO© Minicihip Auto automatic device is a portable device with 7-inch touch screen for user control and fully automatic testing. It reads real-time colorimetric signal change at wavelengths between 450nm and 650nm. It works with a 120-volt power supply.
The GENZPRO Development Kit is a test chip with chemically treated active bonding surfaces, the surfaces of which have been specially modified by chemical and physical methods. The desired proteins can be attached to these chips and the test can be performed on the GENZPRO© Minicihip Auto automated instrument. The test is completed in approximately 20 minutes.
For the preparation of test chips, the "GENZPRO Development Kit" is used, which includes the chips and the necessary solutions. First, the necessary tools and auxiliary laboratory equipment are prepared. Then, the solutions required for coating are prepared. Coating and blocking processes are applied to the test chips, respectively.
With the Genzpro Development Kit, It is aimed to develop rapid quantitative protein tests with low cost, low limit of detection value. Like this, Within the scope of their studies, researchers will be able to develop original tests of the protein they want to detect in the analyte and accelerate their academic research with affordable budgets with an innovative platform with high innovation and technology value.

Keywords: Capillary ELISA, Molecular Diagnosis, POC

DRAFT

ORAL PRESENTATION

Green synthesis of zinc oxide and manganese oxide nanoparticles from *Brassica oleracea* and *Triticum monococcum* extracts and their hybrid advanced oxidation applications of Rhodamine B dye removal

Şennur Merve YAKUT[1](*ORCID: https://orcid.org/ 0000-0001-9190-4061*),
Gamze DOĞDU YÜCETÜRK[2*](*ORCID: https://orcid.org/0000-0002-0278-8503*)

[1] Nevşehir Hacı Bektaş Veli University, Faculty of Engineering and Architecture, Department of Environmental Engineering, Nevşehir, Türkiye.
[*2]Bolu Abant İzzet Baysal University, Faculty of Engineering, Department of Environmental Engineering, Bolu, Türkiye.

*Corresponding author e-mail: dogdu.gamze@gmail.com

Abstract

In terms of catalytic activity, MnO and ZnO Nps exhibit the desired physicochemical characteristics. Hereby, in this study, manganese oxide and zinc oxide nanocatalyzes were green synthesised with kale *(Brassica oleracea*) and einkorn wheat (*Triticum monococcum),* and catalyzes' advanced oxidation process (AOPs) performances such as sonolysis, photolysis, and hybrid photocatalysis (UV+catalyst), sonocatalysis (US+catalyst), and sonophotocatalysis (UV+US+catalyst/H_2O_2) were reported for the removal of Rhodamine B (RhB) dye from aqueous media. The operation conditions of the study were: 4.80 for pH value at 22.5 °C, 10 ppm for the initial dye concentration, and 0.5 mL of the hydrogen peroxide (H_2O_2) concentration, respectively. The results were obtained that with MnO synthesized from *T. monococcum*, 99% of dyestuff removal was achieved in 15 minutes in the hybrid sonophotocatalysis process, and 98% of RhB in 30 minutes in the hybrid sonophotocatalysis process with ZnO. While, MnO synthesized with *B. oleracea was performed* 96% of RhB removal in the 15th min in the sonophotocatalysis process, 99% of the RhB dye was removed in the 15th min in the hybrid sonphotocatalysis process with ZnO. In summary, the green synthesized MnO and ZnO nanoparticles from *B. oleracea* and *T. monococcum* have huge potential candidate as photocatalysts and they are considered to be a promising alternative in AOPs considering its low cost and high efficiency for the degradation of selective contaminants in water/wastewater systems.

Keywords: Green chemistry, hybrid advanced oxidation, *Brassica oleracea*, *Triticum monococcum*, dye removal, water treatment

DRAFT

ORAL PRESENTATION

Determination of the suitable cell count in flow-cytometrical analyzes of frozen bull semen

Kemal Tuna Olğaç (*ORCID: https://orcid.org/0000-0001-9216-7059)*

Ankara University, Faculty of Veterinary Medicine, Department of Reproduction and Artificial Insemination, Ankara, Türkiye.

*Corresponding author e-mail: ktolgac@ankara.edu.tr

Abstract

The aim of this study is to determine the most suitable number of spermatozoa to be used in the post-thawed flow-cytometrical analyzes of frozen bull semen. In the study, a total of 20 frozen semen from 5 different bulls, 4 from each bull, were used. After thawing the straws in a water bath at 37 °C for 30 seconds, they were analyzed in a flow-cytometer device (Beckman Coulter, Fullerton, CA, USA) in terms of Acrosomal Integrity, High Mitochondrial Membrane Potential and Viability parameters. Analyzes were performed by counting 1000, 5000, 10000, 15000, 20000 spermatozoa for each straw. In the study, analysis of variance was used to determine the difference between groups in the measured post-thawed spermatological characteristics. All statistical analyzes were performed by IBM SPSS 23 statistical package program and evaluated considering the criterion of $p<0.05$. As a result of the study, it was observed that the different spermatozoon counted in the flow-cytometrical analyzes of the Acrosomal Integrity ($p=0.149$), High Mitochondrial Membrane Potential ($p=0.548$) and Vitality ($p=0.347$) parameters resulted in statistically similar values and had no effect on the results obtained. According to these results, it was concluded that sperm counts between 1000-20000 were appropriate to use in post-thawed flow-cytometrical analysis of frozen bull semen.

Keywords: Flow-cytometer, frozen bull semen, spermatozoon count

Acknowledgements: This research was supported by The Scientific and Technological Research Council of Turkey (TÜBİTAK; 123O702)

DRAFT

ORAL PRESENTATION

In silico Analysis of Elucidating MicroRNAs in Secondary Metabolite Production in Medicinally Important Plants

Perihan Yigit[1*] (*https://orcid.org/0000-0002-0649-1483*) and Sevgi Marakli[2] (*https://orcid.org/0000-0001-5796-7819*)

[1]Yildiz Technical University, Graduate School of Science and Engineering, Department of Molecular Biology and Genetics, Istanbul, Turkey.
[2]Yildiz Technical University, Faculty of Arts and Sciences, Department of Molecular Biology and Genetics, Istanbul, Turkey.

*Corresponding author e-mail: yigitperihann@gmail.com

Abstract

Epigenetics is heritable or non-heritable changes in gene expression without any alterations in DNA. Micro RNAs (miRNAs) are small ncRNAs with 22-24 nt in length, regulating gene expression especially, at the post-transcriptional level. Secondary metabolites such as flavonoids, terpenoids and alkaloids in plants have been commonly used in cosmetics, medicine and other industrial areas. This study aimed to determine the regulatory roles of miRNAs in nine important enzymes involved in the metabolism of flavonoid, phenolic acid, alkaloids, terpenoids, coumarin and stilbene metabolites via *in silico* analyses. For this purpose, reference miRNAs in mountain strawberry (*Fragaria vesca* L.), papaya (*Carica papaya* L.), asparagus (*Asparagus officinalis* L.) and soybean (*Glycine max* L.) were retrieved from miRBase. The number of identified miRNAs was given as strawberry, soybean, papaya, and asparagus, respectively. We determined 10, 40, 5 and 14 miRNAs which target cinnamate-4-hydroxylase while 5, 59, 4 and 9 miRNAs were obtained for chalcone isomerase. Moreover, chalcone synthase enzyme was targeted by 16, 49, 6 and 9 miRNAs whereas 8, 40, 5 and 12 miRNAs were detected for caffeoyl CoA O-methyltransferase. Caffeic acid O-methyltransferase was estimated to have 7, 40, 8, and 16 miRNAs, whereas flavonol synthase was found to have 7, 26, 6, and 12 miRNAs. It was determined that 7, 40, 8 and 16 miRNAs were targeted by caffeic acid O-methyltransferase, while flavonol synthase was targeted by 7, 26, 6 and 12 miRNAs. Another three enzymes showed similar results. 17 miRNAs in strawberry, 41 in soybean, 11 in papaya and 16 in asparagus targeted phenylalanine ammonia lyase enzyme, 6 in strawberry, 50 in soybean, 9 in papaya and 11 in asparagus targeted aromatic-L-amino-acid decarboxylase enzyme, and even 8 in strawberry, 32 in soybean, 6 in papaya and 10 in asparagus targeted tropinone reductase I. Obtaining results of this study are expected to provide possible new insight for further studies related to the optimisation of plant secondary metabolite and also production of novel metabolites with therapeutic benefits.

Keywords: Epigenetics, non-coding RNAs, gene expression regulation, medicinal plants

DRAFT

ORAL PRESENTATION

Explant Culture of Cartilage Tissue and Multi Differentiation of Derived Cells

Özlem Özden-Akkaya[1*] (*ORCID: https://orcid.org/0000-0001-6372-915)*

[*1]Afyon Kocatepe University, Faculty of Veterinary Medicine, Department of Histology and Embryology, Afyonkarahisar, Türkiye

*Corresponding author e-mail: ozlemozden@aku.edu.tr

Abstract

The injured or damaged articular cartilage is very difficult to regenerate, and the resulting lesion may spread locally. Cell therapy applications based on autologous mesenchymal stem cells (MSCs) with high proliferation and differentiation potential, is recognized as a striking option for treating cartilage injuries. In addition, it is known that progenitor cells in the cartilage tissue has multipotent properties, similar to MSCs. While chondrogenic progenitor cells express some mesenchymal markers, they also express some chondrogenic markers unlike MSCs. In this study, it was aimed to obtain progenitor/stem cells derived from bovine cartilage tissue by explant culture method, and to characterize the isolated cells. The use of slaughterhouse material isn't only eliminating ethical problems but also provides an advantage in terms of the amount of tissue to be collected from the cattle. After washing the hind legs of the cattle, the metatarsophalangeal joint area was disinfected, the joint was opened, and the parts of the articular cartilage was taken from the surface with the help of a scalpel blade. Cartilage tissue pieces were thoroughly washed in PBS with antibiotics, and placed into well plates as 6-7 small pieces in each well, containing culture medium. Cartilage tissue pieces were incubated at 37 °C in an environment with 5% CO_2. After incubation, the cells were left to proliferate. Progenitor/Stem cells derivation took approximately three weeks. The obtained cells had heterogeneous morphology. In the first passage, the cells showed fibroblast-like morphology. Strong chondrogenic differentiation was noted as a result of differentiation studies. The osteogenic differentiation was evident, but adipogenic differentiation was weak. In future studies, it is planned to determine the gene and protein expression profiles of the isolated progenitor/stem cells. This study was conducted following approval from the experimental Animals Ethical Committee of Afyon Kocatepe University, Türkiye (AKÜHADYEK-50-23; 20.06.2023).

Keywords: Cartilage Tissue, Explant Culture, Stem Cells, Progenitor Cells.

DRAFT

ORAL PRESENTATION

Çoklu Aromatik Organik Bileşiklerin Sentezi ve Biyofilm Engelleyici Etkilerinin İncelenmesi

Kübra Yeşilkaya[1*] *(ORCID: 0009-0001-2559-0815)*, Gökçe Merey[2] *(ORCID: 0000-0003-0759-1299)*

[*1]Marmara Üniversitesi, Sağlık Bilimleri Enstitüsü, Biyogüvenlik ve Biyoemniyet A.B.D., İstanbul, Türkiye
[2]Marmara Üniversite, Sağlık Bilimleri Fakültesi, Temel Sağlık Bilimleri Bölümü, İstanbul, Türkiye

*Sorumlu yazar e-mail: dyt.kubrayesilkaya@gmail.com

Özet

Bakterilerin, kendilerini korumak ve üremelerini kolaylaştırmak için oluşturdukları polisakkarit yapılı biyofilm tabakası dezenfeksiyonu zorlaştıran önemli bir faktördür. Bununla birlikte, özellikle patojen bakterilerin oluşturduğu biyofilm tabakaları, hastane enfeksiyonlarının yaygınlaşmasına neden olmakta ve gıda endüstrisinde ürünlerin işlenmesinde problem oluşturmaktadır. Biyofilm tabakaları nedeniyle üretim bantları, borular, zemin ve çeşitli yüzeylede bakteri birikmekte ve bunlar, patojenlerin çevreye yayılması açısından risk oluşturmaktadır.

Özellikle flagella fonksiyonu olan ve sıklıkla karşılaşılan *Pseudomonas aeruginosa*, *Escherichia coli*, *Salmonella enterica* bakterileri, biyofilm oluşturma potansiyeli yüksek olan bakterilerdir. Bununla birlikte dünya çapında yaygın ve gıda kaynaklı bir patojen olan *Campylobacter jejuni* de biyofilm oluşturma potansiyeli olan bir bakteri olup gıda ve ilaç endüstrilerinde kalıcı bir kirletici haline gelebilir. *C. jejuni*, saf kültüre göre *E. coli* veya *P. aeruginosa* ile birlikte olduğunda daha fazla biyofilm oluşturabilir.

Bakterilerde biyofilm oluşumunu kontrol eden mekanizmada sorumlu molekülün dimerik formda halkalı guanozin monofosfat (c-di-GMP) olduğu anlaşılmıştır. Bakterilere özgü bir nükleotid olan c-di-GMP, biyofilm oluşumunun yanısıra bakteriler arası etkileşim, virulans, flagella fonksiyonları ve motilite gibi bakterinin hayati fonksiyonlarını da kontrol etmektedir. Dolayısıyla c-di-GMP aktivitesinin engellenmesi, bakterilerin hayati fonksiyonlarını sonlandırıbilecek, biyofilm oluşumunu engelleyebilecek bir etki yaratır. Molekül yapısı incelendiğinde c-di-GMP, yapısında bulunan dimerik formdaki guanozin grupları nedeniyle belirli şartlarda cis formda dört katlı ve sekiz katlı yapılar oluşturabilmektedir. Düşük konsantrasyonda K^+ tuzu varlığında bu yapıların oluştuğu gözlenmiştir. Bununla birlikte yüksek konjugasyona sahip çoklu aromatik bileşikler de c-di-GMP'nin katlı yapılar oluşturmasını sağlamaktadır. Katlı yapılar oluşturan c-di-GMP çökerek aktivitesini kaybeder.

Yapılan çalışmada, yüksek konjugasyona sahip çoklu aromatik bileşikler sentezlenerek bunların biyofilm engelleme kabiliyetleri incelenmiştir. Aromatik gruplar içeren aldehit ve amin bileşiklerinin basit bir imin oluşum reaksiyonu ile yüksek konjugasyona sahip çoklu aromatik bileşikler elde edilmiş ve özellikle sağlık ve gıda alanında biyogüvenlik açısından tehdit oluşturan bakterilerden *E. coli* ve *C. jejuni* üzerinde denenmiştir. Deneylerde kullanılan çoklu aromatik bileşiklerin c-di-GMP ile etkileştiği UV spektrskopisi ile izlenerek anlaşılmıştır.

Anahtar Kelimeler: Biyofilm, c-di-GMP, *C. jejuni*, *E. coli*

DRAFT

ORAL PRESENTATION

Investigation of synthesis, characterization and *in vitro* drug release of *Ferula assafoetida* fabricated Fe_3O_4@chitosan magnetic nanoparticles

Münteha Özsoy[1,2*](ORCID: https://orcid.org/0000-0002-5853-5150), Vesen Atiroğlu[2,3] (ORCID: https://orcid.org/0000-0001-7551-133X), Atheer Atiroğlu[2,3] (ORCID: https://orcid.org/0000-0003-2409-7030), Mahmut Özacar[1,2*](ORCID: https://orcid.org/0000-0002-1783-7275).

[1*]Sakarya University, Faculty of Science, Department of Chemistry, 54187 Sakarya, Turkey
[2]Sakarya University, Biomaterials, Energy, Photocatalysis, Enzyme Technology, Nano and Advanced Materials, Additive Manufacturing, Environmental Applications and Sustainability Research and Development Group (BIOENAMS R&D Group), 54187 Sakarya, Turkey
[3]Sakarya University, Biomedical, Magnetic and Semiconductor Materials Application and Research Center (BIMAS-RC), 54187 Sakarya, Turkey

*muntehaozacar@gmail.com

Abstract

Fe_3O_4 nanoparticles have emerged as promising candidates for drug delivery applications due to their inherent benefits, including small particle sizes, thermal stability, biocompatibility, and large surface areas. This study focuses on the synthesis of Fe_3O_4 nanoparticles as a drug delivery system using the co-precipitation method and their subsequent modification with chitosan as a binder. Through the co-precipitation method, Fe_3O_4 nanoparticles were successfully synthesized and subsequently functionalized with chitosan, creating a stable and efficient drug carrier system. Asafoetida, a natural medicinal substance known for its therapeutic properties, was chosen as the drug active ingredient and successfully loaded onto the nanoparticles. Characterization studies were conducted to validate the successful synthesis, functionalization, and drug loading process. The drug release behavior from the Fe_3O_4-chitosan-*F. assafoetida* nanoparticles as a potential controlled drug release system was systematically investigated. The findings demonstrated the successful synthesis and modification of Fe_3O_4 nanoparticles with chitosan as a drug delivery system, effectively encapsulating the therapeutic Asafoetida extract. This study lays the foundation for utilizing these novel nanoparticles in targeted drug delivery applications, offering a promising avenue for enhancing therapeutic efficacy and minimizing side effects through controlled drug release mechanisms.

Key words: Fe_3O_4 MNPs, Chitosan, *Ferula assafoetida*, Drug delivery systems, Drug release.

DRAFT

ORAL PRESENTATION

Etanol İntoksikasyonu Vakalarında Kandaki Alkol Konsantrasyonu ile Etil Glukronid ve Etil Sülfat arasındaki ilişkinin araştırılması

Yeter Erol Öztürk[1] (*ORCID:* 0000-0001-9503-7057)

[*1]Adli Tıp Kurumu, Ankara Grup Başkanlığı, Kimya İhtisas Dairesi, Ankara, 06300, Türkiye

*Sorumlu yazar e-mail: yetererol@hotmail.com

Özet

Postmortem örneklerde ölüm nedeni tayininde alkolün varlığı ve ölüm nedeni üzerindeki etkisini tespit etmek oldukça zordur. Alkol antemortem alkol alımı, postmortem alkol sentezi ya da mikroorganizmaların varlığına bağlı olarak vücütta oluşabilir. Ölüm koşulları, ortam sıcaklığı, örnek alma yöntemleri, örneklerin taşınması, ölüm ile alkol analizi arasından geçen süre gibi parametreler yüksek miktarda alkol oluşmasına neden olabilir. Oluşan alkolün endojen olup olmadığının tespiti için farklı teknikler kullanılmaktadır. Bu yöntemlerden en güvenilir olanı Etanol Faz II metabolitleri olan etil glukronid (EtG) ve etil sülfatın (EtS) dedeksiyonu ve miktarlı analizidir. Bu konjuge metabolitler ölümden önce etanol alımını kanıtlamaktadır. EtG ve EtS, Etanol alımını takiben oluşur ve belirli bir süre sonra maksimum konsantrasyona ulaşır. (EtG) ve etil sülfatın (EtS) yokluğu kanda tespit edilen etanolün endojen olduğunun kanıtıdır ayrıca bu metabolitlerin kanda tespiti ölüm öncesi alkol alındığının kanıtıdır. Bu çalışmada kan etanol konsantrasyonu 10 mg/dL üzerindeki örnekler çalışmaya kabul edilmiştir. Kan örnekleri protein çökürme yöntemi ile hazırlanarak LC-MS/MS yöntemi ile EtG ve EtS analizi yapılmış ve vakalardaki alkolün endojen olup olmadığı tespit edilmiştir. Kanda EtG ve EtS tespit edilmiş örneklerle kandaki etanol konsantrasyonu ile EtG ve EtS konsantrasyonları arasında bir ilişki olup olmadığı araştırılmıştır.

Anahtar Kelimeler: Etil Glukronid, Etil Sülfat, ETG, ETS, LC-MS/MS, postmortem

DRAFT

ORAL PRESENTATION

Monoamine oxidase inhibition and toxicity evaluation of phenylurea-bearing hydrazones

Cem Yamalı[1](*ORCID: https://orcid.org/0000-0002-4833-7900)*, Göksun Demirel[2*] (*ORCID: https://orcid.org/ Orcid ID: 0000-0002-2994-5505*)

[1]Cukurova University, Faculty of Pharmacy, Department of Basic Pharmaceutical Sciences, Adana, Turkey.
[*2]Cukurova University, Faculty of Pharmacy, Department of Pharmaceutical Toxicology, Adana, Turkey.

*Corresponding author e-mail: demirelgoksun3@gmail.com

Abstract
Parkinson's disease (PD) is an increasingly prevalent neurodegenerative disorder that impacts a rising global population (1). The pharmacological treatments for PD include a range of medications, including levodopa, dopamine agonists such as ropinirole, MAO-B inhibitors including selegiline, COMT inhibitors such as entacapone, and medicines with cholinergic activity, exhibited by benzatropine (2).
In the context of therapeutic applications, inhibitors targeting MAO-B have predominantly been employed for the management of Parkinson's and Alzheimer's diseases (3). Also, it is essential to advance the development of medication candidates that exhibit a reduced prevalence of adverse effects. Hence, the objective of this study was to conduct an initial assessment of the efficacy and specificity of phenylurea-bearing hydrazones as monoamine oxidase inhibitors.
Phenylurea-bearing hydrazones were synthesized, and their chemical structures were confirmed via 1H NMR, 13C NMR, and HRMS methods. The synthesized compounds were investigated for their hMAO-A and hMAO-B inhibitory activity by an in vitro fluorometric method. MTT test was employed to identify the cytotoxic properties of the lead compound against SH-SY5Y cell line which has been used in various areas of neuroscience (4).
Hydrazones represent a category of compounds that bear similar to hydrazine and exhibit advantageous biological characteristics, notably their significant capacity to inhibit monoamine oxidase. Among the compounds tested in this study, 1-(2-(2-(4-chlorobenzylidene) hydrazine-1-carbonyl) phenyl)-3-phenylurea showed selective and potent MAO-B inhibition. The compound's inhibitor concentration 50 (IC_{50}) value was 0.042±0.001 μM against MAO-B similar to reference selegiline. MTT assay was carried out for the compound to clarify whether the compound is toxic or non-toxic towards SH-SY5Y cell line. MTT results showed that the IC_{50} value of the compound was 166 μM. Consequently, the compound tested was found to be non-cytotoxic at its effective concentration against MAO-B. Neurotoxicity-induced SH-SY5Y model (5) is planned to see whether the compound has also neuroprotective effect for further studies.

Keywords: Parkinson's disease, phenylurea-bearing hydrazones, cytotoxicity test.

Acknowledgments: We thank to Assoc. Prof. Dr. B. Nurpelin Saglik for enzyme assay study.

References

Ntetsika, T.; Papathoma, P.E.; Markaki, I. Novel Targeted Therapies for Parkinson's Disease. Mol. Med. 2021, 27, 17.
Armstrong, M.J.; Okun, M.S. Diagnosis and Treatment of Parkinson Disease: A Review. JAMA-J. Am. Med. Assoc. 2020, 323, 548–560.
Tan YY, Jenner P, Chen SD. Monoamine Oxidase-B Inhibitors for the Treatment of Parkinson's Disease: Past, Present, and Future. J Parkinsons Dis. 2022;12(2):477-493. doi: 10.3233/JPD-212976. PMID: 34957948; PMCID: PMC8925102.
Yamali, C., Gul, H.I., Sakarya, M.T., Saglik, B.N., Ece, A., Demirel, G., Nenni, M., Levent, S. and Oner, A.C., 2022. Quinazolinone-based benzenesulfonamides with low toxicity and high affinity as monoamine oxidase-A inhibitors: Synthesis, biological evaluation and induced-fit docking studies. Bioorganic Chemistry, 124, p.105822.
Xicoy H, Wieringa B, Martens GJ. The SH-SY5Y cell line in Parkinson's disease research: a systematic review. Mol Neurodegener. 2017, 12(1):10.

DRAFT

ORAL PRESENTATION

Setup a Sensor Network for Multiple Measurement by the Fiber Loop Ringdown Spectroscopy (FLRDS) Technique

Burak Malik KAYA* (*ORCID: https://orcid.org/my-orcid?orcid=0000-0002-1251-6915*)

* Eskisehir Osmangazi University, Vocational School of Health Service, 26480, Eskisehir, Türkiye.

*Corresponding author e-mail: malikkaya@ogu.edu.tr

Abstract

The Fiber Loop Ringdown Spectroscopy (FLRDS) technique is an ultra-high sensitive measurement technique that merges the features of fiber optic sensing and the Cavity Ringdown Spectroscopy (CRDS) technique. The FRLDS sensor system consists of a light source, a photodiode, single mode fiber (SMF) and electronic equipment can be employed either individually or as a network platform for sensing several quantities such as strain, pressure, refractive index, temperature, biological cells, chemical species etc. In the FLRDS system, optical loss of a light pulse in the fiber loop is measured by the light decay time constant. To detect a quantity in FLRDS system, time is measured, hence the FLRDS is called as a time-domain sensing technique. FLRDS sensor system has high sensitivity, fast response, real-time monitoring, portability, and low cost. In this study, a strain sensor and a pressure sensor were connected in parallel and setup a sensor network to monitor strain and pressure change simultaneously. Two different fiber loops were utilized with a same continuous laser light source at 1550 nm central wavelength. The strain sensor of 20 cm length was stretched up to 5 cm, resulting 0.12 ε strain sensitivity and the pressure was applied up to 15.68 x 10^5 Pa. Changes in parameters were recorded separately and simultaneously monitored. Results proved that the FLRDS sensors can be utilized for the early detection in structural health monitoring applications as well as biomedical, physical, chemical applications.

Keywords: FLRDS technique, sensor network, fiber optic sensor, fiber loop, ringdown, single mode fiber.

DRAFT

ORAL PRESENTATION

Ciprofloxacin in combination with quercetin and curcumin act strong synergistically to inhibit proliferation of colon carcinoma cells

Selma Ustürk[1*] (0000-0003-2507-2951), Ergül Mutlu Altundağ[2] (0000-0001-5355-4654)

[1]Department of Chemistry, Faculty of Arts and Sciences, Eastern Mediterranean University, Famagusta, North Cyprus, 99628, via Mersin 10, Turkey
[2]Department of Medical Biochemistry, Faculty of Medicine, Eastern Mediterranean University, Famagusta, North Cyprus, 99628, via Mersin 10, Turkey

Correspondence email: selma.usturk@emu.edu.tr

Abstract

The importance of synergistic combinations, as well as the usage of medicines alone, is growing in today's cancer treatment. In this study, the pharmacological effect of the synergistic combination of Ciprofloxacin, Quercetin and Curcumin on colon carcinoma cells (HCT-116), as well as its antiproliferative activity, were studied.

Various concentrations of Ciprofloxacin (Cpx) (10, 20, 40, 80, and 100 M) and various combinations of ciprofloxacin (Cpx), curcumin (Cur), and quercetin (Que) were treated with HCT-116 cells in a dose- and time-dependent manner (24, 48 hours). Using the CompuSyn software, after 48 hours of treatment, the IC50 values for Cpx were discovered to be 56.49 µM. The IC_{50} values of quercetin and curcumin were determined before and reported as 454 µM and 12.0 µM respectively [1].

Cell viability analysis was repeated using varying concentrations of the combinations of ciprofloxacin (Cpx), curcumin (Cur), and quercetin (Que), and the data were entered into the Compusyn software to identify the doses at which the three chemicals were synergistic. The CI values calculated for various combinations of Cpx, Cur, and Que. Results suggest that every combination that was examined has high or extremely strong synergistic effects.

When ciprofloxacin (Cpx), curcumin (Cur), and quercetin (Que) were used together, the ciprofloxacindoses used alone were lowered by approximately 10 times while achieving the same 50% killing on cancer cells. The efficacy of this combination in cancer treatment should be investigated through mechanisms as well as evaluated *in vivo*.

Keywords: Ciprofloxacin, strong synergistic, Cell Proliferation, HCT-116 cells

References

[1] Altundağ, E. M., Özbilenler, C., Ustürk, S., Kerküklü, N. R., Afshani, M., & Yilmaz, E. (2021). Metal-based curcumin and quercetin complexes: cell viability, ROS production and antioxidant activity. *Journal of Molecular Structure*, *1245*, 131107.

ORAL PRESENTATION

Koyunlarda reprodüktif organlarda Doppler ultrasonografinin kullanım alanları

Çağla Nur Küçükbekir[1*] (*ORCID: https://orcid.org/0000-0002-9687-1702)*

[*1]İstanbul Üniversitesi-Cerrahpaşa Lisansüstü Eğitim Enstitüsü, İstanbul, Türkiye

*Sorumlu yazar e-mail: caglakucukbekir@hotmail.com

Özet

Ultrasonografi, iç organ ve dokuların gerçek zamanlı, non-invaziv, zararsız görüntülenmesine olanak sağladığından, tanı ve araştırmalarda sıkça kullanılan bir tekniktir. Ultrasonografinin bir tekniği olan Doppler, kan akışının belirlenmesini sağlar. Doppler ultrasonografi; gebelik muayenesi, fetal-plasental bağın izlenmesi, uterustaki fizyolojik veya patolojik değişiklikler, ovaryum muayenesi, üreme potansiyelinin değerlendirilmesi gibi olguların incelenmesine olanak sağlar. Kedi, köpek, inek, at gibi türlerle birlikte koyun ve keçilerde de tanı amaçlı tercih edilmektedir. Koyunlarda Doppler ultrasonografi, fetal gelişimin araştırmalarında kullanılmaya başlanmıştır. Power Doppler, küçük çaplı damarlardaki kan akışını ölçmede hassastır. Bu nedenle ovaryumlardaki kan akışını değerlendirmede oldukça yararlıdır. Renkli Doppler ile yalnızca graaf foliküldeki kan akışı saptandığından, buradaki değişiklikleri belirlemek, ovulatör olan ve olmayan foliküllerin vaskülaritelerini ayırt etmede yararlıdır. Ayrıca, üreme dönemlerinde genital organların kan akışındaki değişikliklerin belirlenmesinde uterus ve ovaryumların vasküler perfüzyonunun belirlenmesinde kullanılmaktadır. Gebe hayvanlarda fetal dokudaki artan veya azalan vaskülarite ve fetomateryal kan akışının değerlendirilmesinde Doppler ultarsonografiden yararlanılır. Koyun fetüslerinde sık incelenen damarlar, umbilical arter, aorta ve umbilikal vendir. En sık değerlendriilen maternal damarlar ise uterin arter ve uteroplasental arterdir. Umbilikal arter, sıklıkla fetomaternal kan dolaşımının değerlendirilmesinde incelenir. Fetoplasental kılcal yatak hakkında bilgi vererek, direncin artması halinde fetal riskin erken belirlenmesinde faydalıdır. Kan akımının incelenmesi, fetüs büyümesinin kısıtlandığı durumlarda plasental yetmezliği saptamada önemlidir. Embriyo transferlerinde, donörlerde kullanılan luteal fonksiyonun değerlendirmesinde de Doppler ultrasonografiden yararlanılmaktadır. Luteal vaskülarizasyon ile korpus luteumdan progesteronunun üretilmesi arasında bir bağlantı olduğu, fonksiyonel luteolizin yapısal luteolizden önce olduğu düşünülecek olursa, fonksiyonel korpus luteum ile gerilemiş korpus luteum arasında bir ayrım yapılması sağlanır. Bundan dolayı, koyunlarda intraovulator aralığın belirli aşamalarında ve erken gebelikte korpus luteumun sağlığı hakkında kullanılabilmektedir. Koyunlarda yukarıda belirtilen reprodüktif organların patofizyolojik hallerinin irdelenmesinde Doppler' in güncel kullanımı anlatılmıştır.

Anahtar Kelimeler: Koyun, Doppler, reprodüksiyon

DRAFT

ORAL PRESENTATION

Impact of chiral dopant concentration on frequency dependent dielectric properties of nematic liquid crystal

Gülsüm Kocakülah[1*] (*ORCID: https://orcid.org/0000-0001-5423-6957*)

[*1]Düzce University, Faculty of Arts and Sciences, Department of Physics, Düzce, Turkey.

*Corresponding author e-mail: gulsum.kocakulah@gmail.com

Abstract

Nematic liquid crystals have been extensively used in optoelectronic devices, especially in displays. Furthermore, many dopant materials are preferred to improve and develop the properties of nematic liquid crystals and new composites are formed. This study reports the impact of 4'-[(S)-2-Methylbutyl]biphenyl-4-carbonitrile (CB15) chiral dopant concentration on frequency dependent dielectric properties of 4-pentyl-4'-cyanobiphenyl (5CB) nematic liquid crystal. Dielectric parameters (dielectric constants, dielectric anisotropy, relaxation frequency and relaxation time) were determined with dielectric spectroscopy method. Enhancement in the dielectric constants was observed with increasing CB15 concentration in 5CB. Moreover, significant changes were seen in relaxation frequency and relaxation time with CB15 contribution. From this perspective, outcomes of the current study are remarkable and will provide insights for composites to be prepared with 5CB and CB15.

Keywords: 5CB nematic liquid crystal, CB15 chiral dopant, dielectric properties.

DRAFT

ORAL PRESENTATION

Enzymatic Lactate Detection by Nanorod Modified Electrodes

Ezgi Aynı[1] (*ORCID*: https://orcid.org/0000-0002-9168-4895) , Hamza Dünya[2] (ORCID: https://orcid.org/0000-0002-4336-0271) , Nimet Yıldırım Tirgil [3] (ORCID: https://orcid.org/0000-0002-5973-8830)

[1]Ankara Yildirim Beyazit University, Materials Engineering Department, Ankara, Turkey
[2]Bartın University, Biotechnology Department, Bartın, Turkey
[3]Ankara Yildirim Beyazit University, Biomedical Engineering Department, Ankara, Turkey

Corresponding author e-mail: ayniezgi@gmail.com

Abstract

Lactate, in particular, has emerged as a salient biomarker with applications in health surveillance and the evaluation of athletic performance. This research is centered on the creation of an electrochemical biosensor system tailored for the analysis of lactate, facilitated by the incorporation of nanorod-modified electrodes. Nitrogen-doped carbon nanorods were synthesized through a hydrothermal approach and subsequently applied to the electrode surface in various configurations (drop casting and electrodeposition) and concentrations (1 mg/mL, 1.5 mg/mL, and 2 mg/mL). The biosensor's design entails the incorporation of lactate oxidase (LOx) enzymes onto the surface of nanorod-modified electrodes, which present a heightened surface area. These nanorods enhance enzyme immobilization and foster efficient electron transfer, thus optimizing the identification of the analyte.
The biosensor's efficacy was evaluated in terms of its sensitivity, selectivity, and stability. The nanorod-modified electrode demonstrated remarkable sensitivity and selectivity in detecting lactate, attributed to its extensive surface area and augmented enzyme loading. Consequently, precise quantification of low lactate concentrations was rendered feasible. Moreover, the biosensor's practical utility was gauged by introducing lactate into serum samples. The outcomes gleaned from these samples corroborated the biosensor's dependability and its capacity for real-time lactate monitoring. The biosensor's stability and consistency were also subjected to comprehensive examination to ensure sustained operational integrity and practical viability. This research highlights the significant progress made in the field of electrochemical biosensors, specifically with regards to the detection of analytes.

Keywords: Biosensor, Enzymatic biosensor, Electrochemistry, Lactate detection, Nanomaterials, Nanorod

DRAFT

ORAL PRESENTATION

Rizosferik *Trichoderma* İzolatlarının Domateste Fusarium Kök ve Kök boğazı Çürüklüğü Hastalık Etmeninin Miseliyal Gelişimine Etkisi

Süleyman ÇAĞLAR[1*] (*ORCID: 0009-0005-7873-3647*), H. Handan ALTINOK[1] (*ORCID: 0000-0002-4267-1107*.

[*1]Erciyes Üniversite, Ziraat Fakültesi, Bitki Koruma Bölümü, Kayseri, TÜRKİYE

*suleyman.caglarr1@gmail.com:

Özet

Solanaceae familyasından domates (*Solanum lycopersicum* L.) ülkemizde başta Akdeniz, Ege, Marmara, Karadeniz ve Güneydoğu Anadolu Bölgeleri'nde açıkta ve örtü altında geniş alanlarda yetiştiriciliği yapılan önemli kültür bitkilerimizden biridir. Domateste Fusarium solgunluğu; *Fusarium oxysporum* f. sp. *lycopersici* (Sacc.) W. C. Snyder & H. N. Hans (FOL) ile kök ve kök boğazı çürüklüğü hastalığı; *F. oxysporum* f. sp. *radicis-lycopersici* Jarvis & Shoemaker (FORL) ekonomik kayıplara neden olan önemli hastalıklardır. Bu çalışmada, yerel rizosferik *Trichoderma* izolatlarının domateste Fusarium kök ve kök boğazı çürüklüğü hastalığına karşı (FORL) etkinlikleri in vitro'da ikili kültür çalışması ile değerlendirilmiştir. *Trichoderma* izolatlarının hiçbiri 37°C'de gelişme göstermediğinden insanda patojenik olmadıkları değerlendirilmiştir. Kültür koleksiyonundan temin edilen ve antibiyosis etkinliği bilinen iki adet *T. harzianum* izolatı da (T16 ve T23) denemelere dahil edilmiştir. İkili kültür testinde toplam 36 adet izolattan sadece bir izolat Kny/T1-2 (Konya) patojene karşı hiperparazitik etki gösterirken diğerleri antibiyosis etkisi göstermiştir. Antibiyosis etkisi gösteren iki izolat T23 ve Dyb/T10-6 (Diyarbakır) sırasıyla %72,56 ve %58,75 inhibisyon oranı ile patojenin miseliyal gelişimine karşı diğer izolatlardan daha başarılı bulunmuştur. *Trichoderma* izolatları FORL miseliyal gelişimini %35,34'den %72,56'ya kadar değişen inhibisyon oranlarında sınırlandırmıştır. Analizler sonucunda 14 adet *Trichoderma* izolatının ikili kültür testinde kontrole göre başarılı oldukları saptanmıştır. Bu izolatların tohum çimlendirme denemelerinde de kolonizasyon yetenekleri açısından benzer başarıyı gösterdikleri belirlenmiştir.

Anahtar Kelimeler: Domates, FORL, *Trichoderma*, ikili kültür

Dipnot: Bu çalışma Erciyes Üniversitesi Bilimsel Araştırma Projeleri BAP-YL-12545 nolu proje kapsamında desteklenen yüksek lisans tez çalışmasının bir kısmını içermektedir.

DRAFT

ORAL PRESENTATION

Synthesis and İnvestigation of Aggregation Properties of Water-Soluble Octa-Substituted Manganese(III) Phthalocyanines Bearing Pyridine Groups.

Hüseyin Baş[1] (https://orcid.org/0000-0002-8722-3359),
Zekeriya Biyiklioglu[1] (https://orcid.org/0000-0001-5138-214X),

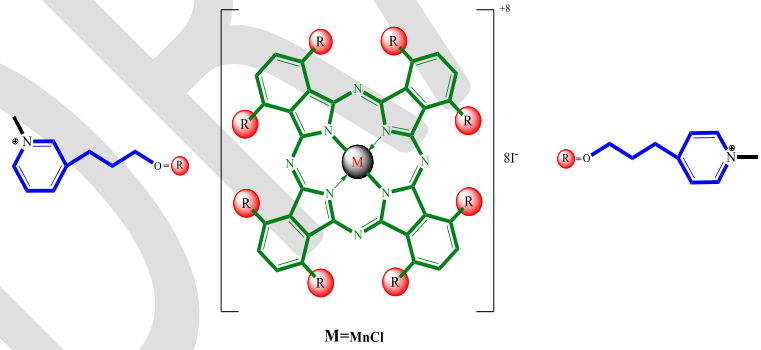
[a]Karadeniz Teknik Üniversitesi, Fen Fakültesi Kimya Bölümü, 61080 TRABZON

hsyn__bs@hotmail.com

Abstract

Pyridine-containing compounds constitute a class of significantly N-heterocycles. The compounds frequently occur in bioactive natural products, and are widely utilized as versatile building blocks for the preparation of pharmaceuticals, organic materials and agrochemicals. Therefore, the development of synthetic materials bearing pyridine groups has attracted considerable interest in organic, inorganic and medicinal chemistry [1]. As a most known class of macrocyclic aromatic compounds, phthalocyanines (Pc) are compounds that have demonstrated several interesting properties such as high chemical and thermal stability, rich redox chemistry, delocalized 18 p electron system suitable for electrontransfer processes. Besides, Pcs are compatible with chemical modifications with changing of central metal and/or changing of number and position of substituted groups to improve their required features [2]. Because of these properties, phthalocyanines are preferred as functional materials in different technologies, such as electrochemical sensors solar cells, gas sensors, nonlinear optics, semiconductors, liquid crystals, photovoltaics, catalysts, electrochromics, and photosensitizers in photodynamic therapy (PDT) [3]. However, low solubility of phthalocyanines in water and organic solvents and strong aggregation are the biggest disadvantages of these molecules. In this study, manganese(III) phthalocyanines containing water-soluble non-perieral octa-pyridine groups were synthesized and the agrgasyon özellikleri were investigated.

This work was supported by KTU-BAP (Project No: 10412).



Keywords: Photocatalysis, Phthalocyanine, Synthesis, Solubility in water,

References

[1] Zhang, X. Y., Liu, K., Li, S., Li, Y., Wu, Y., Li, X., Ran, X., Liu, A., Yuan, J., A metal-free strategy for the synthesis of tetrasubstituted pyridines: dimethyl sulfoxide as carbon source, Tetrahedron Letters 126 (2023) 154623. https://doi.org/10.1016/j.tetlet.2023.154623 0040-4039

[2] Demir, S., Yuksel, F., Novel highly water soluble zinc(ıı) phthalocyanines: synthesis, photochemistry and DNA binding behaviours. Inorganica Chimica Acta 548 (2023) 121373. https://doi.org/10.1016/j.ica.2022.121373

[3] Baş, H., Kahriman, N., Bıyıklıoğlu, Z., Synthesis and electrochemical properties of copper(II), manganese(III) phthalocyanines bearing chalcone groups at peripheral or nonperipheral positions, Turkish Journal of Chemistry, (2020) 44: 1549-1555. doi:10.3906/kim-2006-21.

DRAFT

ORAL PRESENTATION

Compatibilizer synthesis and characterization to be used in the preparation of polyal® and polyolefin masterbatches obtained from composite packaging wastes

Songül Şahin DUMANKAYA[1] (0000-0001-9224-8393), Osman ŞERBETÇİ[1] (0000-0003-3001-6954), Gökhan CEYHAN[1,2](0000-0002-9127-2348), Ahmet TUTUŞ[1,3](0000-0003-2922-4916)

[1*]Kahramanmaras Sutcu Imam University, Department Of Materials Science And Engineering, Kahramanmaras Turkey
[2]Kahramanmaras Sutcu Imam University, Technical Sciences MYO, Food Processing Division, Kahramanmaraş, Turkey
[3]Kahramanmaras Sutcu Imam University, Forestry Industrial Engineering, Kahramanmaras, Turkey

* sonqulsahin@hotmail.com

Abstract

Tetra Pak®, known as Composite Packaging, was patented by Ruben Rausing for liquid foods and came to life as Erik Wallenberg's idea. [1]. This original packaging was originally made of wax paper.Today, multi-layer Tetra Pak® packaging consists of laminated rigid paper, LDPE and aluminum foil.In general, technologies applied to PCBCs are divided into those that reprocess them as a blend and those that involve a previous step to remove cellulosic fibers. In the first case, PCBCs are subjected to thermal treatments (incineration, pyrolysis, gasification) together with other products for energy recovery. In the second case, the removal of paper is carried out by hydropulping. This process; in the presence of water, cellulosic fibers are separated from the plastic and aluminum layers by centrifugal forces. [2]. Waste of paper recovery, outer LDPE layer and Al-PE laminate, usually contains up to 4-5% paper fibers. Due to its high heat treatments it is mostly used for energy recovery in heat treatments. Al2O3 obtained as a result of these processes is generally used in the cement industry because it is beneficial for cement production. [3]. Alcoa Aluminio also uses gasification applications, such as the Klabin and TSL Ambiental plant, which use plasma gasification to convert plastic on the other hand to paraffin and recover aluminum in its pure form. [4]. In general, there are methods of recovery of PolyAl® from Tetra Pak® wastes in the literature, but the difficulties of using this material with polyolefins are expressed.
In this study, the use and synthesis of a compatibilizing agent was carried out to turn PolyAl® granules obtained from Tetra Pak® wastes into a masterbatch with polymers such as polyethylene and polypropylene. For this purpose, acidic modification of sepiolite with maleic anhydride was carried out. Analytical and spectroscopic characterization of the obtained powder material was carried out. Then, PE and PP extrusion was carried out with the additive obtained at the rate of 5%. PPRC pipe was produced from the obtained granules. Strength and rheological tests for PPRC pipe show that the strength of the PE-compatibility-PolyAl® (A2) masterbatch used in the PPRC pipe interlayer has improved by 64% compared to the PE-PolyAl® (A1) masterbatch. In the optical microscope images, it is seen that the microcracks have disappeared.

Keywords: Polyolefins, Tetra Pak®, Masterbatch, Recycling
We would like to thank Kahramanmaraş Paper Factory and TÜBİTAK (2244, Project No: 118C060) for this study.

DRAFT

ORAL PRESENTATION

Cyanogenic Glycosides as Potential Anticancer Agents: Molecular Docking, and ADME Study

Seda ŞİRİN[1,*]

[1] Gazi University, Faculty of Science, Department of Biology, Ankara, Türkiye

*E-mail: sdasirin@hotmail.com

Abstract

Cancer is a fatal disease with a growing mortality rate each year, and it is one of the most serious threats to human health worldwide. Drug resistance, recurrence, and metastasis are all risks of conventional therapy. As a result, there is an urgent need to discover new medications that are both effective and have few adverse effects in order to prevent and treat various forms of cancer. Natural products are at the forefront of anti-cancer chemotherapeutic approach because they have shown considerable promise in preventing and treating cancer. Cyanogenic glycosides are a significant and common class of plant natural products. The objective of this research is to examine at the ADME features and molecular docking of cyanogenic glycosides (amygladin, dhurrin, linamarin, lotaustralin, prunasin, and taxiphyllin) with apoptotic proteins (BAX, BCL2, CASP3, CASP8, CASP9, CYCS, and P53). The ADME study was carried out using the SwissADME program, which was used to compute physicochemical descriptors as well as forecast ADME parameters, pharmacokinetic features, druglike nature, and medicinal chemistry friendliness of one or more small compounds to aid in drug development. The molecular docking analysis were performed by using an CB-Dock2 program to find the binding mode of the ligands to target proteins. The SwissADME demonstrated that results for cyanogenic glycosides (especially amygladin, dhurrin, prunasin, and taxiphyllin) are mostly in the acceptable range. The molecular docking demonstrated a high affinity interaction between the cyanogenic glycosides and the apoptotic proteins. The results showed the significant potential of *cyanogenic glycosides* to be used as anticancer agents.

Keywords: ADME study, Alzheimer disease's, anticancer agent, cyanogenic glycosides, molecular docking.

DRAFT

ORAL PRESENTATION

Eco-friendly synthesis of graphene/ZnO nanocomposites with visible light-driven photocatalytic activity for water remediation: A preliminary study

Can ERGUN[1*] (*ORCID: https://orcid.org/0000-0003-2423-3274*), Hakan ESKIZENGIN[1] (*ORCID: https://orcid.org/0000-0003-1709-7497*), Saniye Cevher OZEREN[1] (*ORCID: https://orcid.org/0000-0002-8509-0548*)

[1]Ankara University, Faculty of Science, Department of Biology, 06560, Ankara, Turkey.

*ergunc@ankara.edu.tr

Abstract

A facile and environmentally friendly strategy for the synthesis of graphene/ZnO nano-photocatalysts for the removal of methylene blue under natural sunlight irradiation is described. The graphene/ZnO nanocomposites were successfully synthesized by a green synthesis method. Zinc acetate in an aqueous solution was reduced with corn starch and the graphene was synthesized by a modified Hummer's method from graphite waste derived from pencil rods. The morphology, structure and photocatalytic activity of the nanocomposites were characterized by scanning electron microscopy, transmission electron microscopy, X-ray diffraction spectroscopy and UV-Vis absorption spectroscopy. The ZnO nanoparticles were incorporated between the monolayers of graphene nanosheets by sonication. Based on the electronic conductivity and band gap property, the photocatalytic activity of the graphene/ZnO nanocomposites showed >95% degradation of methylene blue under natural sunlight. Photocatalytic degradation of methylene blue showed no significant decrease after the 5th cycle under natural sunlight. Although ZnO NP-only showed significant removal efficiency under sunlight irradiation, graphene/ZnO nanocomposites facilitate decontamination faster. The obtained results show better degradation rate, waste recovery, environmentally friendly simple synthesis, cost-effective strategy, and promise the development of large-scale adaptation.

Keywords: Green synthesis, Nanotechnology, Water treatment, Photocatalysis

DRAFT

ORAL PRESENTATION

The effect of boron solution application on mycotoxin amount

Aysegul Karapinar Arif [1*], Doruk Nil [2], Elif Miray Dogan [3], Mehtap Kara [4], Gonca Erkose Genç [5], Bural Pural [6]

[*1] TED Bodrum College, High School, Science, Mugla, Turkey.
[2] Ozel Modern Bilimler Akademisi, Faculty, Department, City, Country.
[4] Istanbul Universtiy, Faculty of Pharmacy, Department of Pharmaceutical Toxicology, Istanbul, Turkey.
[5, 6] Istanbul Universtiy, Faculty of Pharmacy, Department of Pharmaceutical Microbiology, Istanbul, Turkey

*Corresponding author e-mail: karapinar.aysegul@gmail.com

Abstract

Molds, which are very common in nature and threaten animal health and human health; It is accepted as one of the contaminants that cause hazards in processed, unprocessed or semiprocessed food products along with agricultural products. Mitotoxins are considered as metabolites of molds that cause health problems. Mycotoxicosis is the whole of the health problems experienced as a result of the consumption of these toxins as food by animals and humans. Mycotoxins have many effects such as estrogenic, teratogenic, carcinogenic, mutagenic, dermatoxic, hepatotoxic, nephrotoxic effects. In this project, food products that are frequently used in daily life among the public were purchased in two forms, packaged and open. All food products were kept for a certain period of time to mold after washing with tap water and boron. The amount of mold formed on the moldy food products was measured in accordance with the Eliza kit and test kit protocols. There was no difference in the amount of T-2 toxin in food products washed with boron solution and tap water. However, it was determined that aflatoxin increased more in products washed with tap water.

Keywords: food,food supply security,public health mycotoxin,boron

DRAFT

ORAL PRESENTATION

Production of antioxidant cream from vegetable and fruit peel residues within the scope of protecting nature and sustainability

Aysegul Karapinar Arif [*1] , Demircan Isik [2] , Ada Dinneden [3] , Mehtap Kara [4] , Fatma Nur Yilmaz [5] , Gozde Hasbal Celikok [6]

[*1] TED Bodrum College, High School, Science, Mugla, Turkey.
[2 ,3] BJK Kabatas Foundation Schools, High School, Science, Istanbul, Turkey.
[4] Istanbul Universtiy, Faculty of Pharmacy, Department of Pharmaceutical Toxicology, Istanbul, Turkey.
[5] Istanbul Universtiy, Faculty of Pharmacy, Department of Pharmaceutical Microbiology, Istanbul, Turkey.
[6] Istanbul Universtiy, Faculty of Pharmacy, Department of Pharmaceutical Biochemistry, Istanbul, Turkey.

*Corresponding author e-mail: karapinar.aysegul@gmail.com

Abstract

The aim of this project was to prevent the waste of vegetables and fruits in the cafeterias of schools and in daily life from going to waste and to utilize them for human health and life. Examples of common vegetables and plants used in daily life (parsley (Petrosilinum crispum), leek (Allium ampeloprasum), green apple, red apple (Malus domestica), orange (Citrus sinensis), grapefruit (Citrus paradisi), lemon (Citrus limonum), kiwi (Actinidia deliciosa) were selected. It was observed that not only in school cafeterias, but also in places such as street markets and markets where these vegetables and plants are sold, wastes of vegetables and fruits remain and go to the garbage. As a result of these observations, these wastes of vegetables and fruits were collected. Then, samples were taken from each of them by weighing them to the same extent and mixed. This cell extract was first obtained from the mixture of these vegetable and fruit wastes. This cell extract was then tested for antibacterial, antifungal and antioxidant properties. As a result of these tests, it was found to be antibacterial, antifungal and antioxidant. In addition, it was determined that the same extract was not toxic as a result of the cytotoxicity test. This extract was used in the production of cream. Thus, a useful product for human health and life was obtained from the useful parts of vegetable and fruit wastes used in daily life.

Keywords: Antioxidant, antibacterial, antifungal, food waste, cosmetics

DRAFT

ORAL PRESENTATION

Anti-apoptotic and anti-inflammatory effects of dexpanthenol against nicotine-induced liver injury in rats

Nuray Üremiş[1*] (ORCID: https://orcid.org/0000-0002-3958-4352), Meral Aslan[1] (ORCID: https://orcid.org/0009-0008-2271-8220), Elif Gürel[1] (ORCID: https://orcid.org/0000-0002-4070-7014), Elif Taşlıdere[2] (ORCID: https://orcid.org/0000-0003-1723-2556)

[*1] Inonu University, Medical Faculty, Department of Medical Biochemistry, Malatya, Türkiye.
[2] Inonu University, Medical Faculty, Department of Histology and Embryology, Malatya, Türkiye.

*Corresponding author e-mail: nurayuremis@gmail.com

Abstract

The aim of this research was to explore the correlation between the mechanisms at the molecular level responsible for liver damage caused by long-term tobacco usage, the caspase cascade, and the Akt/NF-kB signaling pathway. Additionally, the study investigated the protective effects of dexpanthenol (DEX) in this context. Male rats were administered intraperitoneal injections of nicotine at a concentration of 0.5 mg/kg/day and/or DEX at a concentration of 500 mg/kg/day for 8 weeks. After administration, liver function tests were performed on serum samples, and tissue samples were examined for Akt, NF-κB, Bax, Bcl-xL, Caspase-3, and Caspase-9 protein levels, as well as histopathological changes. Additionally, analyses of oxidative stress markers and proinflammatory cytokines were performed. The administration of nicotine resulted in elevated levels of IL-6, IL-1β, MDA, TOS, and OSI, while TAS levels exhibited a reduction. In addition, nicotine administration led to a decrease in the p-Akt/Akt ratio, an increase in NF-κB, Bax, Caspase-3, and Caspase-9 protein levels, and a decline in the antiapoptotic protein Bcl-xL. Notably, DEX intervention effectively mitigated these indicators, restoring them to levels akin to those seen in the control group. The liver damage initiated by nicotine prompted oxidative stress, inflammation, and apoptosis via the modulation of Bax/Bcl-xL, Caspase-3, Caspase-9, and Akt/NF-kB pathways. In contrast, DEX intervenes by mitigating nicotine-induced liver injury through the regulation of apoptosis involving NF-κB, Caspase-3, Caspase-9, inhibition of Bax, and activation of Bcl-xL.

Keywords: Nicotine, dexpanthenol, liver, apoptosis, inflammation, oxidative stress

DRAFT

ORAL PRESENTATION

The Role of Aurora Kinases in Human Colorectal Cancer

Elif Rumeysa Özsoy[1*] (*ORCID:https://orcid.org/0009-0008-6040-9875*),
Sibel Sarı[2] (*ORCID: https://orcid.org/0000-0002-2505-5804*)

[*1,2] Abdullah Gul University, Faculty of Life and Natural Sciences, Molecular Biology and Genetics, Kayseri, Turkey.

*Corresponding author e-mail: elifrumeysa.ozsoy@agu.edu.tr

Abstract

Colorectal cancer (CRC) was the third most common cancer worldwide in 2020 and the second leading cause of cancer deaths. Although the common treatment strategy for CRC is chemotherapy; toxicity, acquired resistance and low tumor-specific selectivity are still the main impediments to current clinical success. In recent years, it has been stated that changes in the cellular expression levels of specific proteins with roles in checkpoint pathways through carcinogenesis such as cell cycle regulators contribute to malignant phenotypes, making them as possible therapeutic targets for cancer therapy. Among these proteins, one group of cell cycle regulators is Aurora Kinases (AURKs) which contains three members: Aurora Kinase A (AURKA), Aurora Kinase B (AURKB), and Aurora Kinase C (AURKC). While AURKA and AURKB are expressed in most somatic cells, AURKC is limited to meiotic cells. AURKs are belonging to the family of serine/threonine kinases that serve as mitotic regulators with an important role in various stages of the cell cycle such as centrosome separation and maturation, kinetochore-microtubule interaction, regulation of cytoskeletons, spindle-assembly checkpoint, completion of cytokinesis and chromosome compaction. The overexpression of AURKs is evident in numerous different human cancers or solid tumors, including colorectal cancer, which is associated with its clinicopathological parameters and overall survival time. Therefore, Aurora Kinase inhibition is a potent anticancer strategy and there are many selective and pan Aurora Kinase Inhibitors (AKIs) on different phases of clinical development. In preclinical studies, AKIs have exhibited promising results by showing antiproliferative activity and their ongoing Phase-I and Phase-II clinical trials as anticancer molecules have also yielded in encouraging results for future studies.

Keywords: Colorectal Cancer, Cell Cycle Regulators, Aurora Kinases, Aurora Kinase Inhibitors

DRAFT

ORAL PRESENTATION

Exploring the Impact of Pan-AURKs and Selective AURKB Inhibitors on Non-Small Cell Lung Cancer

Elif Rumeysa Özsoy[1*] (*ORCID:https://orcid.org/0009-0008-6040-9875*),
Sibel Sarı[2] (*ORCID: https://orcid.org/0000-0002-2505-5804*)

[*1,2] Abdullah Gul University, Faculty of Life and Natural Sciences, Molecular Biology and Genetics, Kayseri, Turkey.

*Corresponding author e-mail: elifrumeysa.ozsoy@agu.edu.tr

Abstract

Aurora Kinase B (AURKB) is a mitotic serine/threonine protein kinase belonging to the highly conserved Aurora Kinase family of mitotic kinases, along with Aurora Kinase A and Aurora Kinase C. AURKB is a kinase module of the Chromosomal Passenger Complex, which plays critical roles in the cell cycle, including chromosome compaction and segregation, the spindle-assembly checkpoint and cytokinesis. While AURKB is ubiquitously expressed in healthy cells, it is overexpressed in various human cancers, including lung cancer. Lung cancer is the leading cause of global cancer mortality and incidence, with approximately 2.2 million diagnoses and 1.8 million deaths. Non-small cell lung cancer (NSCLC) is the most common type of lung cancer, accounting for 80-85% of all cases. In NSCLC, AURKB overexpression has been found to be associated with a negative prognosis, poor therapeutic response, and decreased overall survival rate, suggesting that AURKB may be a potential prognostic biomarker. In the past decades, pan-AURKs and selective AURKB inhibitors have been tested in preclinical studies, and some of them have progressed to different stages of clinical trials. Based on preclinical studies conducted on NSCLC cell lines, selective AURKB inhibitors lead cancer cells to apoptosis by causing the formation of cytokinesis defects, polyploidy and impaired chromosome alignments. Pan-AURKB inhibitors that target all AURKs for inactivation exhibit antitumor activity, and some of them tested in patients with NSCLC have shown satisfactory tolerance and a manageable safety profile.
Since AURKB has different roles directly or indirectly in NSCLC progression, both pan-AURK and selective AURKB inhibitors have shown anticancer properties based on preclinical studies. Some of their Phase-I and Phase-II clinical trials have yielded desired results, encouraging future studies in this direction.

Keywords: Non-Small Cell Lung Cancer, Aurora Kinases, Aurora Kinase B, Aurora Kinase B Inhibitors

DRAFT

ORAL PRESENTATION

Electroactive Hydrogel Preparation, Characterization and Investigation of Drug Release Potential by Electrical Stimulation

Zeynep Sude Çifcibaşı[1] (*ORCID:*https://orcid.org/0009-0005-9432-259X),
Melike Fırlak Demirkan[1] *(ORCID: https://orcid.org/0000-0003-1674-9086)*

[1]Department of Chemistry, Gebze Technical University, Kocaeli, Turkiye

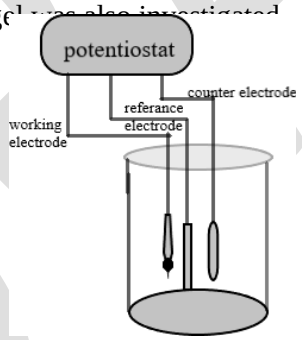
z.cifcibasi2018@gtu.edu.tr

Abstract

Controlled drug delivery systems are platforms that enable the drug to reach its effect in a certain speed-time, while minimizing the side effects of the drug in the body and providing the drug in a personalized and correct dose. Its main task is to transfer the active ingredient of the drug directly to the therapeutic area and to transfer the concentration of the right dose to that pathological area at the right time. Considering the disadvantages of the traditional drug release (i.e. repeated dosing, cost, causing toxicity, poor bioavailability, poor adsorption from target side) from capsules, injections, creams, it is possible to understand the reason for the increased interest in controlled drug release. [1,2]

Pemetrexed disodium 2,5 hydrate (PEM) compound is a drug active ingredient used in the treatment of pleural mesothelioma and non-small cell lung cancer. This active substance is sent to the organism by traditional methods. However, by traditional ways, this active substance has access to many areas outside of the pathological area. As a result, many side effects are seen in patients who receive the treatment. Realization of PEM release with electrical field sensitive hydrogels is a promising way to solve such problems. [3-4]

In this study, a novel drug delivery system consists of polypyrrole (PPy)/Polyethylene glycol diacrylate (PEGDA)/2-Hydroxyethyl acrylate (HEA)/ Ethylene glycol dimethacrylate (EGDMA) hydrogel was prepared for electrically triggered release of PEM and characterized by electrochemical and spectroscopical techniques. The electrically controlled in vitro drug release profile of the prepared electroactive PPy/PEGDA/HEA/EGDMA hydroge[illegible]



Scheme 1 : Schematic representation of the 3-electrode system to be used in PEM loading and release

Keywords: PEM release, controlled drug delivery system, electrically triggered drug release, electroactive hydrogel

DRAFT

ORAL PRESENTATION

Exfoliation of Graphite in PVA/PVP Aqueous Solution and Production of Silver Nanoparticle/Multilayer Graphene/PVA/PVP Nanocomposites

Yasemin Çelik[1*] (*ORCID: https://orcid.org/0000-0003-3993-6095*),
Özge Yıldız[1] (*ORCID: https://orcid.org/0009-0002-9965-8947*)

[*1]Eskişehir Technical University, Engineering Faculty, Department of Materials Science and Engineering, Eskişehir, Turkey

*ybozkaya@eskisehir.edu.tr

Abstract

Graphite is a three-dimensional form of carbon, which is composed of stacked graphene layers, which are bound together by van der Waals forces that can be overcome by applying mechanical forces within appropriate organic solvents. However, the most widely available, non-toxic, environmentally friendly and low cost solvent, water, is considered to be a poor solvent, since graphite is hydrophobic. To be able to exfoliate graphite in water, graphite can be oxidized using strong oxidizers to convert it into the hydrophilic form, so-called graphite oxide. However, this is a time consuming and risky process with a relatively high cost, which make this method non-practical. The objectives of this study were to directly exfoliate graphite in water using polyvinyl alcohol (PVA) and polyvinyl pyrrolidone (PVP) to obtain few or multilayer graphene, and then to fabricate silver nanoparticle/multilayer graphene-based nanocomposites by synthesis of silver nanoparticles within the dispersion of the exfoliated material. The reason for using PVA and PVP as dispersing agents is that their ability to act as stabilizers by coating silver nanoparticles, providing shape and size controlled silver nanoparticle synthesis, as well as their help for the direct exfoliation of graphite in water. At this scope, PVA and PVP aqueous dispersions were prepared at specific concentrations and mixed. The required amount of natural graphite (at different initial concentrations) was added into the PVA-PVP aqueous solution and bath sonicated to exfoliate natural graphite. After resting the prepared dispersions for a certain time, supernatant was collected. Silver nanoparticles were synthesized within the prepared dispersion via chemical reduction method. The exfoliated material and the synthesized nanocomposites were characterized by UV-vis spectrometer, Raman spectrometer, X-ray diffractometer, scanning electron microscope and transmission electron microscope. It was revealed that natural graphite was exfoliated into multilayer graphene sheets, and silver nanoparticles/multilayer graphene/PVA/PVP nanocomposites were successfully produced.

Keywords: Natural graphite, Exfoliation, PVA/PVP aqueous solution, Multilayer graphene, Silver nanoparticle, Nanocomposite

DRAFT

ORAL PRESENTATION

Utilizing Active and Passive Fungal Cell Immobilization to Create an Eco-Friendly Biocomposite for Acidic Dye Biodecolorization

Halime ÇINAR[1*] (*ORCID: https://orcid.org/0009-0009-3774-7814*),
Sema ÇELİK[2] (*ORCID: https://orcid.org/0000-0003-4284-823X*)

[*1] Eskisehir Osmangazi University, Graduate School of Natural and Applied Science, Department of Chemistry, 26480, Eskisehir, Turkey.
[2] Eskisehir Osmangazi University, Faculty of Science, Department of Chemistry, 26480, Eskisehir, Turkey.

*Corresponding author e-mail: halime172126_26@outlook.com

Abstract

In recent years, the growth of industries has led to a surge in environmental pollution, particularly concerning the rapid increase in various forms of pollution. One prominent issue is the contamination of aquatic environments due to industrial effluents containing synthetic dyes. Even at very low concentrations, these dyes become noticeable pollutants, contributing to visible color pollution. Furthermore, their presence in water sources poses risks to ecosystems due to potential toxicity. Despite attempts using conventional physico-chemical methods, these persistent pollutants remain largely untreated in aquatic systems due to their non-biodegradable nature. To address this, an effective alternative method is biosorption, which has shown promise in tackling water pollution caused by diverse pollutants including synthetic dyes and metals. Notably, fungal biomass has proven to be an efficient biosorbent, often utilized in immobilized forms due to certain limitations within industrial processes.

In this study, a novel biocomposite sorbent obtained that amalgamates both active and passive immobilization techniques. This inventive approach was effectively utilized for the biosorption of Acid Red 1 (AR1) contamination. Initially, the cells of *Thamnidium elegans* (*T. elegans*) were subject to passive immobilization onto an industrial waste product derived from olive pomace. Subsequently, this *T. elegans*/*olive pomace* biomass combination underwent active immobilization via chitosan entrapment.

The research investigated the influence of various parameters on the batch decolorization process, encompassing initial pH values ranging from 3.0 to 9.0, sorption times ranging from 5 to 30 minutes, , and sorbent dosage within the range of 0.001 to 0.012 g. The peak biosorption efficiency reached an impressive 78.09% at a pH of 3.0, utilizing 0.01 g. Equilibrium in biosorption was achieved within 10 minutes. The study's findings shed new light on the advancement of biosorption techniques using natural materials, which could contribute significantly to pollution control strategies.

Keywords: Acid red 1, biocomposite, biodecolorization, biosorption, immobilization,

DRAFT

ORAL PRESENTATION

Assessing the Impact of Shoc2 on Hair Growth Cycle

Sibel Sari[*1] (*ORCID: https://orcid.org/0000-0002-2505-5804)* and Pablo Rodriguez-Viciana[2] (*ORCID: https://orcid.org/0000-0002-8846-543X)*

[*1]Abdullah Gul University, Faculty of Life and Natural Sciences, Molecular Biology and Genetics, Kayseri, Turkey.
[2]University College London (UCL) Cancer Institute, London, UK

*Corresponding author e-mail: sibel.sari@agu.edu.tr

Abstract

Hair, which is one of the distinguishing features of mammals, serves many essential biological functions, including thermoregulation, physical protection, sensory activity, and the distribution of sweat gland products, such as pheromones. Hair shafts are produced by thousands of hair follicles in mammalian skin, and these follicles continuously regenerate throughout life. The hair follicle represents a unique, highly regenerative system that physiologically undergoes cycles of growth (anagen), apoptosis-mediated regression (catagen), and rest (telogen) numerous times in life.
The EGFR/RAS/RAF/MEK/ERK pathway has a well-described role in skin and hair follicle development. Its function in the skin and its appendages, including hair follicles, is necessary for proper development and tissue homeostasis, and its deregulation rapidly results in defects in cellular proliferation and differentiation. The SHOC2 phosphatase complex, which comprises SHOC2, MRAS, and PP1, is a key regulatory node required for efficient RAS-RAF-MEK-ERK pathway activation. Here, we have used a conditional SHOC2 knockout mouse model to examine the effect of SHOC2 ablation on hair growth in adult mice. Our earlier findings demonstrate that when SHOC2 is systemically deactivated in adult mice, it leads to skin dermatitis and various histopathological changes, including an increased number of anagen-phase hair follicles within the hypodermis. Therefore, to investigate whether aberrant anagen-phase hair follicles affect hair growth, we depilated age-matched SHOC2 KO and WT male mice and monitored them for 28 days.
The results indicate that there were no significant differences in hair regeneration between SHOC2 KO and WT mice. This suggests that the increased number of hair follicles in the anagen phase in SHOC2 KO mice does not necessarily correlate with increased hair growth.

Keywords: SHOC2, Transgenic Mouse Models, Hair Growth Cycle

DRAFT

ORAL PRESENTATION

İstatistik destekli Yalın Altı Sigma ile uygulanan teknik tarım desteğinin hayvancılık işletmelerinde verime etkisi

İrfan ÖZTÜRK* (https://orcid.org/my-orcid?orcid=0000-0002-6421-5604)
Yılmaz GÖKSU** (https://orcid.org/my-orcid?orcid=0000-0002-6421-560)

* Harran Üniversitesi Organize Sanayi Bölgesi –MYO/Şanlıurfa -TÜRKİYE
** Tarım Kredi Yem şirketi -OSB/ Şanlıurfa - TÜRKİYE

*Sorumlu yazar e-mail: ozirfan23@yahoo.com

Özet

Günümüzde birçok küçük ve orta büyüklükteki tarım işletmeleri geleneksel yöntemlerle çiftçilik yapmaktadır. Bu durum hem ülke ekonomisine hem de işletmeye büyük ekonomik kayıplara neden olmaktadır. İşletmelerde artan giderler, çiftçilerin yeterli bilgi ve teknik donanıma sahip olmamaları sonucunda birçok insan çiftçiliği bırakmak durumunda kalmaktadır. Çiftçilerin söz konusu üretimi sürdürülebilir hale getirebilmeleri için ise; devlet kurumları tarafında sürekli çeşitli desteklemelerle (gübre desteği, mazot, litre başına verilen süt desteği gibi…vs.) desteklenmek zorunda bırakılmaktadır. Ancak söz konusu yapılan nakdi desteklemeler işletme sahiplerinin bilgi ve tecrübesine katkı sağlamamaktadır. Yapılan nakdi desteklemelerin yerine, verimi artırmaya yönelik teknik bilgi desteğinin uzman eşliğinde sağlanması daha verimli sonuçlar doğuracaktır. Teknik bilgi desteği çiftçilerin üretimdeki verimini artırdığı gibi, çiftçilerin bu süreç zarfında uygulanan yeni teknik bilgileri de öğrenmelerini sağlayacaktır. Kısacası bu süreç zarfında çiftçilerimiz uygulamalı olarak eğitilmiş olacaktır. Teknik bilgi desteğinin sağlayacağı katkıyı ortaya koymak için, yaptığımız iki farklı çalışmada hem süt veriminde hem de canlı ağırlık artışında anlamlı verim artışları elde edilebileceği ortaya konulmuştur. Bu çalışmada, süt veriminin arttırılmasında Adıyaman Gerger ilçesinde bulunan Bakırcıoğlu süt işletme çiftliğinde haziran, temmuz, ağustos ve eylül aylarında holstein ırkı 25 adet süt sığırı üzerinde Yalın Altı Sigma metodolojisinin prosesi uygulanmıştır. Bu çalışmanın sonucunda süt veriminde yaklaşık %67 oranında verim artışı sağlandı. Haziran ayında altı sigma metodolojisi ile işletmenin mevcut durumu izlenip verim kayıplarına sebebiyet veren etkenler tanımlanarak, 2.laktasyonda bulunan süt sığırlarının süt ortalaması 15.3 litre/gün'den, Eylül ayı sonunda 22.53 litre/gün'e çıkarılmıştır. Diğer bir çalışma ise Erzincan Refahiye'de bulunan Zeybekler hayvancılık besi işletmesinde canlı ağırlık artışı konusunda yapılmıştır. Bu çalışmada da işletmenin verimini artırma sürecinde istatistiksel destekli Yalın Altı Sigma prosesi uygulanmıştır. Zeybekler besi işletmesinde rastgele seçilen 50 tane Holstein sığırın kulak numaraları kayıt altına alınmış ve besinin 3. ayında bir ay süre ile sığırlar (İso 9001 sertifikalı) ticari yem ile beslenerek canlı ağırlık artışları kaydedilmiştir. Besinin 4. ayında ise sığırlar Yalın Altı Sigma prosesleri sonunda hazırlanan ve Altı Sigma rasyonu olarak tanımladığımız yeni rasyonla beslenmiş ve günlük canlı ağırlık artışları 0,79 kg/gün'dan 1,250 kg/gün'a çıkarılmıştır. Sonuç olarak besi işletmesinde günlük canlı ağırlık artışında ortalama % 62 oranında artış sağlanmıştır ($p<0.01$). Çiftçilerimize nakdi yardım yerine teknik tarım desteğinin sağlanması daha etkili olmuş ve bu uygulamanın hem ülke ekonomisine hem de çiftçinin gelirinin artışında daha fazla katkı sağlayacağı ortaya çıkmıştır.

Anahtar Kelimeler: Yalın Altı sigma metodolojisi, süt verimi, canlı ağırlık artışı, verim kaybı, rasyon

DRAFT

ORAL PRESENTATION

Yemli uzaktan sualtı video kayıt sistemleri (BRUVs) ile balık topluluk yapılarının incelenmesi

Adnan Çağlar Oruç[1*] (*ORCID: https://orcid.org/0000-0003-4675-1654*)

[*1] İzmir Kâtip Çelebi Üniversitesi, Su Ürünleri Fakültesi, Temel Bilimler Bölümü, İzmir, Türkiye

*Sorumlu yazar e-mail: acaglaroruc@gmail.com

Özet

Sucul ekosistemler günümüzde kirlilik, aşırı avcılık, yapılaşma, istilacı türler ve iklim değişikliği gibi baskıların giderek artan olumsuz etkilerine maruz kalmaktadır. Balıklar ise sucul ekosistemlerin sağlığı ve işleyişindeki kilit rolleri ile elzem canlılardır. Balık biyoçeşitliliğinin tespiti ve periyodik takipleri sucul ekosistemlerin mevcut sağlık durumunun belirlenmesi, ilerleyen süreçteki değişimleri hakkında yorum yapılmasına olanak sağlamaktadır.

Balık topluluklarını etkili bir şekilde izlemek ve yönetmek için gerekli verilerin toplanmasında kullanılan yöntemler geçmişten günümüze değişime uğramış bilim insanları artık trol ağları, dinamitler gibi tahrip edici yöntemlerden ziyade sualtı görsel sayım tekniği, dalıcı kumandalı video tekniği, uzaktan kumandalı araçlar ve yemli uzaktan sualtı video kayıt sistemleri gibi canlıyı ortamından ayırmayan, gözleme dayalı yöntemler kullanmaya başlamıştır.

Yemli uzaktan sualtı video kayıt sistemleri (BRUVs) gelişen teknoloji ile birlikte son yirmi yıldır balık topluluklarının incelenmesinde kullanılmaya başlanmış, görsel sayım tekniğinde yaşanan belli derinlik, zaman ve dalışa elverişlilik gibi kısıtlamaları olmaması, farklı koşullar ve habitatlarda zamansal-mekânsal değişimlerin tespiti için uygun ölçümler sağlaması, düşük maliyetli ve istatistiksel olarak güçlü bir yöntem oluşuyla öne çıkmıştır.

Dünya genelinde büyük çoğunluğu Avustralya ve Yeni Zelanda'da gerçekleştirilmiş olan üç yüze yakın BRUVs çalışması mevcut olmasına karşın Akdeniz basenindeki araştırmalar toplamın ancak %3' lük kısmını oluşturmaktadır. Ülkemiz özelinde ise BRUVs tekniği ile gerçekleştirilmiş bir araştırma bulunmamakla birlikte sucul ekosistemlerimizin, sualtı zenginliklerimizin korunması ve izlenmesine yönelik çalışmalarda bu sistemlerden verimli şekilde faydalanılmalıdır.

Anahtar Kelimeler: BRUVs, balık, sucul ekosistem

DRAFT

ORAL PRESENTATION

Optimization of Alkaline Pectinase Production By *Bacillus* Sp. VGA7 Isolate

Kudret BULUT*[1] (ORCID ID: 0009-0009-4958-1161), Ayşe AVCI[1] (ORCID ID: 0000-0001-7102-397X), Muhammed HAMK[2] (ORCID ID: 0000-0003-0748-1743)

[1]Sakarya University, Faculty of Engineering, Department of Food Engineering, Sakarya, Türkiye
[2]Food Science and Quality Control Department, Halabja Technical College of Applied Sciences, Sulaimani Polytechnic University, Iraq

kudretbulut1@gmail.com

Abstract

Alkaline pectinases are one of the most important enzymes and are widely used in many industrial areas such as wastewater treatment, textile and paper industry, tea and coffee fermentation. Fungi (*Aspergillus, Penicillium*), yeast (*Saccharomyces, Kluyveromyces*) and bacteria (*Bacillus*) are used in microbial pectinase production. Bacterial production is more advantageous due to ease of process and shorter fermentation time. In this study, 25 Bacillus strains that were previously isolated from Lake Van and deposited in the culture collection of Sakarya University Food Engineering Department Food Biotechnology Laboratory were screened for pectinase production. The enzyme production was optimized by choosing the best producer isolate (*Bacillus* sp. VGA7). The bacterium was activated by incubating at 33 °C in a shaking incubator (120 rpm) for 24 hours on nutrient agar and then in nutrient broth. A medium consisting of pectin (10g/L), yeast extract (10g/L), NaCI (1.5g/L), K_2HPO_4 (1g/L), $MgSO_4.7H_2O$ (0.1g/L) was used for enzyme production. The effects of carbon sources (gum, lactose, fructose, sucrose, glucose, starch), nitrogen sources (ammonium sulfate, casein, peptone, soy flour, lentil meal, skimmed milk powder), pectin concentration (0.2-2), pH (7-12), and temperature (25-40 °C) on production of enzyme were determined. Enzyme activity was determined by DNS method. As a result of the study, the maximum enzyme production (3.33 U/mL) was achieved in the presence of pectin and yeast extract, at pH 10 and 33 °C.

Keywords: pectinase, *Bacillus*, DNS, enzyme production

DRAFT

ORAL PRESENTATION

Hasat Sonrası Melatonin ve Modifiye Atmosfer Paketleme Uygulamalarının Kiraz MeyvelerininMuhafazası Üzerine Etkileri

Neslihan Ekinci[1*] (*ORCID: https://orcid.org/0000-0001-7022-5289.*),
Safigül EROĞLU[2] (*ORCID: https://orcid.org/0009-0001-7205-2163.*)

[*1]Çanakkale Onsekiz Mart Üniversitesi, Ziraat Fakültesi, Bahçe Bitkileri Bölümü, Çanakkale, Türkiye
[2]Çanakkale Onsekiz Mart Üniversitesi, Lisansüstü Eğitim Enstitüsü, Bölüm, , Çanakkale, Türkiye

*nekinci@comu.edu.tr

Özet

Önemli ticari değere sahip olan kiraz (*Prunus avium* L.), uzun süre depolamaya elverişli olmayan bir meyve türü olup, aynı zamanda muhafazası sırasında da bir çok kalite kayıpları görülmektedir. Kiraz meyvesinde kalite özelliklerinin korunması amacıyla öncelikli olarak ürünün soğutulması ve soğuk zincirin ürün hasadından başlayarak tüketiciye ulaşıncaya kadar her aşamada korunması gerekmektedir. Yapılan bu çalışmada üretici bahçesinde yetiştirilmiş olan regina kiraz çeşidi meyvelerine hasat sonrası melatonin uygulamasının ve Modifiye Atmosferde paketlemenin (MAP), kiraz meyvelerinin soğukta muhafazası üzerine etkilerini belirlemek amaçlanmaktadır. Çalışma kapsamında regina çeşidi kiraz meyvelerine hasat sonrası değişik dozlarda melatonin uygulamaları 0 (Kontrol), 50,100 µmol L^{-1} ile, Modifiye Atmosfer uygulamaları yapılmış ve 21 gün süre ile depolanmıştır. Depolama süresince hafta da bir kalite parametrelerini (ağırlık kaybı, meyve kabuk ve et rengi,meyve eti sertliği,suda çözünebilir kuru madde miktarı, pH, titre edilebilir asitlik ve tadım testi) ve bazı biyokimyasal özellikleri (C vitamini) belirlemek amacıyla yapılan ölçüm ve analizlerle, uygulamaların kiraz meyvelerinin muhafazasına olan etkileri incelenmiştir. Melatonin, Kontrol MAP, 50µmol +MAP, 100µmol+MAP üzere 6 gruba ayrılmış ve MT uygulamaları, 5 dk. süre ile daldırma yöntemi ile uygulanmıştır. . Kontrol grubu meyveler ise aynı süre saf su içerisinde bekletilmiştir. Ön soğutma sonrası MAP uygulamaları da yapıldıktan sonra tümgruplardaki meyveler, 0°C sıcaklıkta ve %90-95 oransal nem koşullarında meyvelerin dayanma süresine göre 21 gün (3 hafta) süresince depolanmıştır. Çalışma sonucunda başlangıç ve son hafta kalite ve bazı biyokimyasal özellikleri belirlemek amacıyla yapılan analizlerde uygulamalar arasında farklılıklar tespit edilmiştir. 100 µmol+MAP uygulamasının kontrol ve diğer uygulamalara göre kalite ve bazı biyokimyasal özellikleri bakımından daha iyi sonuçlar verdiği belirlenmiştir.

Anahtar Kelimeler: Melatonin, Kalite, Muhafaza, Modifiye Atmosfer Paketleme

DRAFT

ORAL PRESENTATION

Contrasting and Assessing Genome-Wide Association Studies and Marker-Assisted Selection Applications in Animal Breeding

Nursen ŞENTÜRK[1] (0000-0003-1975-0103),
Sena ARDIÇLI[1*] (0000-0003-2758-5945)

[*1]Bursa Uludag University, Faculty of Veterinary Medicine, Department of Genetics, Bursa, Turkiye

*Corresponding author e-mail: sardicli@uludag.edu.tr

Abstract

The application of genomic studies in livestock species aims to comprehend the genetic underpinnings of specific traits, fortify disease resistance, and elevate product quality and production efficiency. The outcomes are integrated into animal breeding initiatives, which adopt a methodology aimed at enhancing economic attributes within livestock through heightened selection efficacy. Given the understanding that yield-related characteristics result from the additive influence of multiple genes, the identification of Quantitative Trait Loci (QTL) is employed to delineate chromosomal regions governing these traits. The shared objective of QTL identification techniques, along with genomic assessment methodologies, is to juxtapose markers and phenotypic values related to the specific trait. The intricacy inherent in the desired attributes has spurred the development of genomic selection methodology, which is rooted in predictive approaches rather than empirical testing. The utilization of selection strategies, coupled with genotyping for production-related traits and marker identification, has accelerated the pace of genetic advancement. Given that numerous genes contribute to the manifestation of most economic traits, the mere identification of a limited subset of these genes through DNA markers will merely account for a fraction of the overall genetic variance. Furthermore, individual genes are inclined to exert modest effects, necessitating a substantial volume of data to estimate their impact accurately. This challenge becomes more pronounced when utilizing a haplotype of markers to track QTL, as the need to estimate numerous haplotype effects arises. While the markers employed for Marker-Assisted Selection (MAS) may be associated with QTL, they might also exist in linkage equilibrium rather than linkage disequilibrium (LD) with the QTL. In instances of LD, it is plausible that either the QTL or the marker itself could indeed function as the QTL. In this work, our aim was to compare these methodologies, evaluating their efficacy in the context of pivotal selection practices integral to livestock breeding.

Keywords: Genetic marker, Genomic selection, GWAS, MAS, QTL

DRAFT

ORAL PRESENTATION

Electrochemical sensor based on Ag-doped ZnO nanocomposite modified pencil graphite electrode for uric acid determination

Mukerrem Findik (*ORCID: https://orcid.org/0000-0002-9441-0814*)

Neighbourhood of Meliksah, Ulusoz Street, Number: 8/17, 42090, Konya, Turkey.

*Corresponding author e-mail: mmukerremm@gmail.com

Abstract

Uric acid is one of the most important diagnostic metabolites resulting from the breakdown of purines in the liver and used in disease diagnosis. Changes in the level of uric acid in human serum and urine are symptoms of various diseases such as leukemia, pneumonia, hyperuricemia, and gout. Determination of uric acid opens the possibility of early intervention in cases of hyperuricemia and prevents deterioration of kidney function. Therefore, a simple detection method for uric acid is very important for clinical and biological fields. In recent years, several methods have been used to detection uric acid. Electrochemical sensors for uric acid detection are preferred due to their many advantages such as low cost, simplicity, high sensitivity, easy handling, and miniaturization. Metal oxides have an important place in the development of biosensors. Especially ZnO has attracted great interest in biosensors due to its desirable properties such as large specific surface area, non-toxicity, good biocompatibility, easy formation of stable nanostructures and high electron communication. The rapid and highly selective detection of ZnO-based electrochemical biosensors for the detection of various diseases has been documented. To improve their sensing performance, Ag can be loaded on the metal oxide surface as promoter, thereby enhancing catalytic activity and sensing performance. In the present study, a nanosensor based on Ag-doped ZnO nanocomposite modified pencil graphite electrode (PGE) was developed for the electrochemical determination of uric acid.

Keywords: Uric acid, Ag-doped ZnO, pencil graphite electrode, electrochemical nanosensor

DRAFT

ORAL PRESENTATION

ctDNA' nın Kanser Tanısında Kullanımında Yeni Yaklaşımlar ve Teknolojiler

Günsel Bingöl

Ankara Yıldırım Beyazıt Üniversitesi, Mühendislik ve Doğa Bilimleri Fakültesi, Biyomedikal Mühendisliği Bölümü, Ankara, Türkiye

Özet

Sirküler Tümör DNA'sı (ctDNA), kanser tanısında giderek daha fazla kullanılan yenilikçi bir yaklaşımdır. Bu yöntem, kanserli hücrelerin vücutta bıraktığı DNA fragmanlarının analiz ederek hastalığın varlığının ve özelliğinin belirlenmesini sağlamaktadır. Kanser, erken evrelerde teşhis ve etkili bir tedavi yönetimi gerektiren ciddi bir sağlık sorunudur. Kanser tanısında geleneksel olarak kullanılan doku biyopsisi ve görüntüleme gibi yöntemlerin invazivlik, zaman, hassasiyet açısından sınırlılıkları bulunmaktadır. Bunun aksine, ctDNA, minimal invaziv bir yaklaşım ile daha hızlı ve hassas bir şekilde kanserli hücrelerin genetik materyalini tespit etme potansiyeline sahiptir. ctDNA analizi, doku örneği almak yerine, kan örneğiyle yapılmaktadır. Yüksek duyarlılıkla çalışan yeni nesil DNA sekanslama teknolojileri (NGS), dijital PCR, tek molekül sekanslama gibi yöntemler sayesinde ctDNA'nın çok düşük konsantrasyonlarda bile tespit edilmesi mümkün olmaktadır. Bu teknolojiler sayesinde kanser türüne ve evresine özgü genetik değişiklikler daha etkili bir şekilde belirlenebilmektedir. Özellikle çoklu kanser taraması, minimal rezidüel hastalığın izlenmesi ve tedaviye yanıtın değerlendirilmesi gibi konularda ctDNA analizi etkili bir araç olarak kullanılabilir. Bunun yanında, yapay zeka ve derin öğrenme gibi yöntemler de ctDNA analizini hızlandırmak ve sonuçları daha doğru bir şekilde yorumlamak için kullanılabilmektedir. Sonuç olarak, ctDNA, kanser tanısı için umut vaat eden bir yaklaşımdır. Erken evre kanserlerin tespiti, tedaviye cevap izleme ve hastalığın yayılmasını takip etme gibi alanlarda önemli katkılar sağlayabilir. Diğer taraftan, bu yaklaşımın klinik uygulamada daha fazla benimsenmesi için standartlaştırılması ve güvenirliğinin sağlanması gerekmektedir. ctDNA analizinin geniş ölçekte kullanılabilmesi için maliyetin düşürülmesi ve altyapının geliştirilmesi de diğer üzerinde düşünülmesi gereken konular arasındadır.

Keywords: ctDNA, kanser, tümör

DRAFT

ORAL PRESENTATION

Sensing behavior of the Al-doped graphene structure toward ethylene and 1-methylcylopropen (1-MCP) molecules: A DFT study

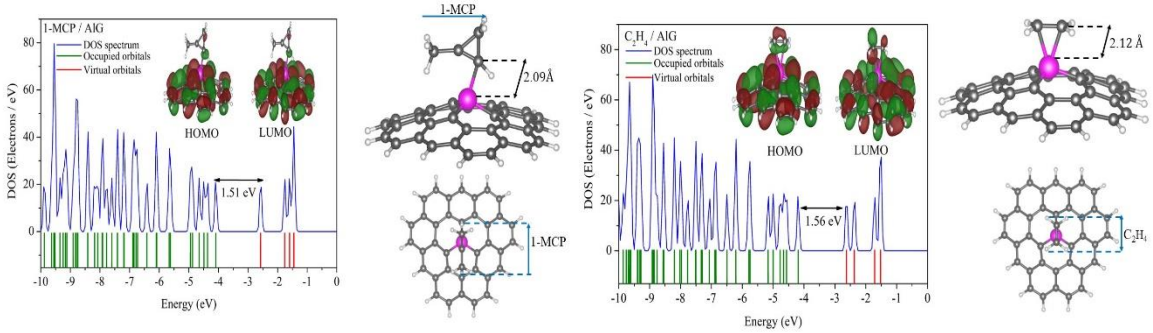
Fatma AYDIN[1] *(https://orcid.org/0000-0002-7219-6407)**, Kıvanç SEL[2] *(https://orcid.org/0000-0002-4623-5206)*

*[1]Çanakkale Onsekiz Mart Üniversitesi, Fen Fakültesi, Kimya Bölümü, Çanakkale, 17100, TÜRKİYE
[2]Çanakkale Onsekiz Mart Üniversitesi, Fen Fakültesi, Fizik Bölümü, Çanakkale, 17100, TÜRKİYE
*faydin@comu.edu.tr

Abstract

Ethylene (C2H4) is naturally produced in plants via biochemical reactions and widely known as the phytohormone responsible for the ripening of fruits, vegetables and some flowers [1]. 1-Methylcyclopropene (1-MCP) is a cycloalkene compound, exists as a gas at room temperature and commercially developed and used as a synthetic plant regulator. Since 1-MCP inhibits the ethylene receptor, it is used to delay the ripening and softening process of the plants by reducing the respiration rate for extending the postharvest shelf life of the plants.

To control the ripening and extent the shelf life of the plants, the adsorption and activity of ethylene and 1-MCP molecules on Al-doped graphene structure (AlG) were characterized by the DFT calculations, through structural geometry optimization, HOMO and LUMO, adsorption energy and ESP calculations [2-5]. Global reactivity analyzes confirmed that ethylene and 1-MCP molecules showed electrophilic properties, while AlG showed nucleophilic properties on the Al atom. The chemical adsorption of these gas molecules by AlG was effectively confirmed by different analyses such as ESP and DOS. According to these results, AlG structure is capable to be a new gas sensor for the adsorption of both ethylene and 1-MCP molecules.



Key words: Ethylene, 1-MCP, Al-doped graphene, DFT calculations

The numerical calculations reported in this paper were fully performed at TUBITAK ULAKBIM, High Performance and Grid Computing Center (TRUBA resources).

References

[1] S.F. Yang, N.E. Hoffman, Ethylene Biosynthesis and its Regulation in Higher Plants, Annual Review of Plant Physiology. 35 (1984) 155–189. https://doi.org/10.1146/annurev.pp.35.060184.001103.
[2] P. Giannozzi, *et al*, J.Phys.: Condens.Matter 21, 395502 (2009).
[3] P. Giannozzi, O. Baseggio, P. Bonfà, D. Brunato, R. Car, I. Carnimeo, C. Cavazzoni, S. de Gironcoli, P. Delugas, F. Ferrari Ruffino, A. Ferretti, N. Marzari, I. Timrov, A. Urru, S. Baroni, J. Chem. Phys. 152, 154105 (2020).
[4] K. Momma, F. Izumi, VESTA 3 for three-dimensional visualization of crystal, volumetric and morphology data, Journal of Applied Crystallography. 44 (2011) 1272–1276.
[5] A. Kokalj, Computer graphics and graphical user interfaces as tools in simulations of matter at the atomic scale, Computational Materials Science. 28 (2003) 155–168.

ORAL PRESENTATION

Taşıyıcı Sistem Olarak Kullanılan Cr-MOF-Aljinat Hidrojellerinden Sefaleksinin in vitro İlaç Salım Özelliklerinin Araştırılması

Şevval Beyza Bozkurt[1*] *(ORCID: https://orcid.org/0009-0008-8552-2428)*, Lalehan Akyüz[2*] *(ORCID: https://orcid.org/0000-0001-8548-3037)*

[*1]Aksaray Üniversite, Fen Bilimleri Enstitüsü, Biyoteknoloji ve Moleküler Biyoloji Ana Bilim Dalı, Aksaray, Türkiye
[*2] Aksaray Üniversite, Fen Edebiyat Fakültesi, Moleküler Biyoloji ve Genetik Bölümü, Aksaray, Türkiye

*[1]E-mail: sevvalbeyzabozkurt1310@gmail.com
*[2]E-mail: lale_akyuz@hotmail.com

Özet

Metal organik çerçeveler (MOF) metalle organik ligand arasında kovalent bağ oluşturan, yüksek yüzey alanı, gözeneklilik ve aktif bölge içermesinden dolayı ilaç taşıyıcı sistemlerde kullanılmaktadır. Biyopolimerler ise düşük maliyetli ve biyouyumludur. Sodyum aljinat gibi biyopolimerler hidrojel özellik gösterdiğinden ilaç taşıyıcı sistemlerde kullanılmaktadır. Bu nedenle MOF'lar sodyum aljinat mikroküreler üzerinde sentezlenerek ilaç taşıyıcı sistemlerin in vitro salım özelliklerinin iyileştirilmesi hedeflenmektedir. Sefaleksin (SF), solunum ve idrar yolu, cilt enfeksiyonlarında kullanılan birinci nesil sefalosporin antibiyotiğidir. SF'nin dezavantajı ise yarı ömrünün kısa olmasıdır. Günde 3-4 doz reçete edilmektedir. Bu çalışmada, model ilaç olan SF, ilaç taşıyıcı sistem olan Cr-MOF-Aljinat'lara yüklenerek ilacın in vitro salım özellikleri araştırılmıştır. MOF tabanlı hidrojelleri sentezlemek için yerinde sentez ve doğrudan sentez yöntemleri kullanılmıştır. Metal olarak krom klorür ($CrCl_3$) MOF ligandı olarak 2-metilimidazol (2-MI) kullanılmıştır. Yerinde sentez yönteminde $CrCl_3$ sulu çözeltisine, aljinatın sulu çözeltisi damla damla eklenerek hidrojeller oluşturulmuştur. 2-MI çözeltisine hidrojeller aktarılarak teflon otoklavda reaksiyon gerçekleştirilmiştir. Doğrudan sentez yönteminde Cr-aljinat hidrojelleri 2-MI çözeltisine eklenmiştir. Cr-aljinat hidrojeller Cr-Alg, doğrudan sentez ve yerinde sentez yöntemi ile elde edilen MOF yapıları ise sırasıyla 2-MI-DS ve 2-MI-YS olarak adlandırılmıştır. Sentezlenen Cr-MOF-Aljinatların karakterizasyonları SEM, FT-IR, TGA ve XRD analizleri kullanılarak yapılmıştır. İlaç yükleme kapasitesi 2-MI-DS, 2-MI-YS ve Cr-Alg için sırasıyla %3,431, 3,97 ve 3,867 olarak bulunmuştur. İn vitro ilaç salım çalışmaları pH 7.4'te fosfat tamponunda (PBS) ile 24 saat gerçekleştirilmiştir. Kontrol olarak saf sefaleksin kullanılmıştır. Ortama salınan ilacın konsantrasyonu UV-Vis spektrofotometre kullanılarak 262 nm'de absorbans ölçülerek belirlenmiştir. Sonuç olarak, kontrol grubunda SF'nin yüzeye salımı ilk dört saatte hızlı gerçekleşmiş sonra yavaşlamıştır. Elde edilen verimler, kontrol grubunda 5.saatte %83,20, Cr-Alg'de 8.saate %18,14, 2-MI-DS'de 7.saate %55,83 ve 2-MI-YS'de 24.saate %13,81 olarak gerçekleşmiştir. 2-MI-YS salım uzun süreli gerçekleşmesi, MOF yapısının Cr-Alg hidrojel porlarının içine yerleşmesi olabilir. Özetle Cr-MOF'ların aljinat üzerinden sentezlenebildiği, SF için Cr-MOF-Aljinatların taşıyıcı sistem olarak kullanılabileceği ve yüksek dozda ilaç kullanımını azaltabileceği öngörülmüştür.

Kelimeler: MOF-aljinat, hidrojel, sefaleksin, ilaç salım.

DRAFT

ORAL PRESENTATION

A research on the determination of some bioactive properties of *Helichrysum arenarium* (L.) oil obtained by maseration method

Kıvılcım YILDIZ[1] (ORCID:0000-0001-6807-6472), Buse TEZCAN[1*](ORCID: 0009-0008-5176-8454), Elif Beyza LERMİOĞLU[1](ORCID: 0009-0007-0694-6488), Ebru İŞİTMEZOĞLU[1](ORCID: 0009-0008-0784-8799), Pelin GÜNÇ ERGÖNÜL[1] (ORCID: 0000-0003-4993-7219)

[1]Manisa Celal Bayar University, Engineering Faculty, Department of Food Engineering, 45140, Manisa, Türkiye

Abstract

Helichrysum arenarium (L.) Moench, a perennial herbaceous plant, is a member of the Asteraceae family and originates from Europe, Central Asia, and China. The flowers of *Helichrysum arenarium* are rich in phenolic compounds, including flavonoids, chalcones, phenolic acids, coumarins, and pyrones. Additionally, these flowers exhibit various biological activities, including antiviral, anti-inflammatory, antiproliferative, antimicrobial, antiallergic, antioxidant, antiradical, cholinergic, hepatoprotective, and detoxification properties. These qualities support a wide range of applications, including those in pharmacy, alternative medicine, and natural treatment. The aim of this study is to obtain oil from the health-beneficial *Helichrysum arenarium* plant by utilizing the traditional maceration method and allowing it to steep in olive oil for one month, and to determine some bioactive properties of this oil. In this context, analyses of total carotenoid, total phenolic, total flavonoid contents and DPPH-radical scavenging activity were performed on both *Helichrysum arenarium (L.)* plant and its oil. To facilitate comparative assessment of the analysis outcomes, identical analyses have also been carried out in olive oil. When comparing the results between the two oils, *Helichrysum arenarium* oil statistically demonstrates higher values compared to olive oil. The richness of bioactive compounds in the *Helichrysum arenarium* plant has facilitated the achievement of quality oil.

Keywords: antioxidant, *Helichrysum arenarium*, maceration, olive oil, phenolic

DRAFT

ORAL PRESENTATION

Aspir (*Carthamus tinctorius* L.) bitkisi kullanılarak yeşil sentez yoluyla TiO_2 ve SiO_2 nanoparçacıklarının sentezi ve karakterizasyonu

Nesrin Korkmaz[1*] (https://orcid.org/0000-0002-7896-1042), Fatma Nur Kaçan[2] (https://orcid.org/ 0000-0003-0657-6433),Battal Doğan[3] (https://orcid.org/ 0000-0001-5542-4853), Murat Kadir Yeşilyurt [3](https://orcid.org/0000-0003-0870-7564,)Hayri Yaman[3] (https://orcid.org/ 0000-0002-9663-7027)

[*1]Yozgat Bozok Üniversitesi, Kenevir Araştırmaları Enstitüsü, Temel Bilimler ve Sağlık, Yozgat, Türkiye
[2]Yozgat Bozok Üniversitesi, Lisansüstü Eğitim Enstitüsü, İleri Malzemeler ve Nanoteknoloji, Yozgat, Türkiye
[3]Ankara Gazi Üniversitesi, Teknoloji Fakültesi, Enerji Sistemleri Mühendisliği, Ankara, Türkiye
[3]Yozgat Bozok Üniversitesi, Mühendislik Mimarlık Fakültesi, Makine Mühendisliği, Yozgat, Türkiye
[3]Kırıkkale Üniversitesi, Kırıkkale Meslek Yüksekokulu, Otomotiv Teknolojisi Programı, Kırıkkale, Türkiye

*nesrin.korkmaz@bozok.edu.tr, nesrinokumus@gmail.com

Özet

Nanoteknoloji günümüzde birçok araştırma alanında kullanılmaktadır. Nanoparçacıklar da nanoteknolojinin önemli bir nanomalzemesi olarak bilinir. Son zamanlarda nanopartikül sentezine ilgi gittikçe artmıştır. Ayrıca metal nanoparçacıklar arasında titanyum oksit nanoparçacıklarının (TiO_2 NP'ler) ve silisyum oksit nanoparçacıkların (SiO_2 NP) özel bir yeri vardır. Ultra ince TiO_2 NP'lerin, SiO_2 NP'lerin ve ZnO NP'ları ile birlikte en çok üretilen üç nanomalzemeden biridir. TiO_2 değerli bir yarı iletken geçiş metali malzemesidir ve kolay kontrol, düşük maliyet, toksik olmama gibi özel özellikler gösterir; kimyasal erozyona karşı iyi direnç, daha sonra güneş pillerinde, kimyasal sensörlerde ve çevresel dönüşüm uygulamalarında kullanılmaktadır. SiO_2 insan, hayvan ve çevre sağlığı için zararlı değildir. SiO_2 NP'ler elektrot yüzeyler üzerinde ince film oluşturmada sentezlenen metallerden biridir. Metalik nanoparçacıkların hazırlanması için yerleşmiş birçok kimyasal ve fiziksel yöntemin yanı sıra Yeşil sentez yöntemi sürdürülebilir, hijyenik, basit, güvenli ve çevre dostu olayların geliştirilmesinde önemli bir yöntemdir. Biyosentez (yeşil sentez), kimyasal olarak toksik etkinin üstesinden gelmek için bir alternatif olarak ortaya çıkmıştır. Bu çalışmada Titanyum oksit nanopartiküllerin (TiO_2 NP'ler) ve silisyum oksit (SiO_2 NP'ler) sentezi için Aspir (*Carthamus Tinctorius L.*) sulu özü kullanılarak yeşil sentez yöntemiyle sentezlenmiştir. Biyosentezlenen nanopartiküller XRD, SEM ve EDX ile karakterize edildi. TiO_2 NP'lerin XRD analizinde sentezlenen nanoparçacıkların ortalama kristal boyutu 47 nm olarak hesaplandı. SEM analizinde TiO_2 NP'lerin küresel olduğunu ve 24 -35 nm arasında partiküller gözlemlendi. EDX analizinde TiO_2 NP'ler 4–5 KeV'de yoğun bir absorpsiyon spektrumu sergilemiştir. SiO_2 NP'lerin XRD analizinde SiO_2 NP'lerin partikül büyüklüğü yaklaşık olarak 41 nm olarak hesaplandı. SEM analizinde SiO_2 NP'lerin partiküllerin ortalama boyutu 22-36 nm aralığında bulundu. EDX sonucu, "Si" elementinin varlığını %21.2 olarak rapor edildi. Kullanılmış olan yeşil sentez yöntemi diğer sentez yöntemlerine göre daha ekonomik, hızlı, basit, çevre dostu,ve tehlikesiz olduğu kanıtlanmıştır.

Bu çalışma TÜBİTAK 1002- Hızlı Destek Programı kapsamında (**122M850 proje kodu**) desteklenmiştir.

Anahtar Kelimeler: Nanopartikül, Yeşil sentez yöntemi, SiO_2NP, TiO_2NP

ORAL PRESENTATION

Afinite kromotagrafisi uygulamalarına yönelik Fe^{+3} ile çapraz bağlanmış alginat mikroküreler içeren p(HEMA) esaslı kriyojel membranların üretilmesi ve karakterizasyonu

Aykut Arif Topçu* (ORCID: : 0000-0002-5434-4920)

*Aksaray Üniversitesi, Sağlık Hizmetleri Meslek Yüksekokulu, Tıbbi Hizmetler ve Teknikler Bölümü, Aksaray, Türkiye

*Sorumlu yazar e-mail: aykuttopcu@aksaray.edu.tr

Özet

Afinite kromatografisi uzun yıllardan beri biyomoleküllerin saflaştırılmasında kullanılan etkili bir yöntemdir ve yöntemde saflaştırılmak istenen hedef moleküle karşı hem uygun ligandın hem de destek malzemesinin seçimi oldukça önemlidir.

Yapılması düşünülen bu çalışmada; afinite kromatografisinde kullanılmak üzere Fe^{+3} ile çapraz bağlanmış olan alginat mikrokürelerin elde edilmesi amaçlanmaktatır. Ardından hazırlanmış olan mikroküreler, 2-hidroksietil metakrilat monomerin -14ºC'de polimerleştirilmesi sırasında ortama ilave edilerek komposit bir afinite destek malzemesi oluşturulması hedeflenmektedir.

Sentez aşamasını takiben elde edilen kompozit destek malzemesi, Fourier Dönüşümlü Kızılötesi Spektroskopisi (FTIR), taramalı elektron mikroskobu (SEM), enerji dağılım X- ışını spektrofotometresi (EDX) ve şişme testi ile karakterize edilerek hazırlanmış olan destek malzemesinin hem kimyasal yapısı hem de fiziksel özellikleri değerlendirilerek, ileri ki dönemde yeni bir destek malzemesi olarak afinite kromotagrafisi uygulamaları için uygunluğu araştırılacaktır.

Anahtar Kelimeler: Afinite kromatografisi, 2-hidroksietil metakrilat, alginat, kriyojel.

DRAFT

ORAL PRESENTATION

Yeni bir Fonksiyonlaştırılmış Manyetik Grafen Oksit Kullanarak Enzim İmmobilizasyonu ve Atık Sulardaki Boyaların Uzaklaştırılması

Hilal Soykan Erdoğan[1](https://orcid.org/0009-0006-1918-4665), Şerife Saçmacı[1,2], Mustafa Saçmacı[3], Nalan Özdemir[1]

[1]Erciyes Üniversitesi, Fen Fakültesi, Kimya Bölümü, TR-38039, Kayseri, TÜRKİYE

[2]Nanoteknoloji Araştırma Merkezi, Erciyes Üniversitesi, TR-38039, Kayseri, TÜRKİYE

[3]Yozgat Bozok Üniversitesi, Fen-Edebiyat Fakültesi, Kimya Bölümü, TR-66200, Yozgat, TÜRKİYE

Sorumlu yazar e-mail:hilalsoykan38@gmail.com

Özet

Enzim immobilizasyonu, katalitik aktifliğini koruyarak, tekrar ve sürekli kullanımını sağlamak amacıyla organik veya inorganik taşıyıcılara fiziksel veya kimyasal yöntemlerle tutulmasıdır. İmmobilizasyon işlemi, enzimin dayanıklılığını arttırdığı gibi, enzimin istenildiğinde tepkime ortamından kolayca uzaklaştırılabilmesini de sağlar [1]. Bu çalışmada, kimyasal sentez yöntemlerinden en yaygın olarak bilinen Hummers metodu [2] ile grafen oksit sentezlenerek, manyetik hale getirildi. Daha sonra manyetik grafen oksit'e (Fe_3O_4@GO) 2-metil 8-hidroksikinolin bağlanarak yeni bir fonksiyonel manyetik grafen oksit sentezlenmiştir (mGO@Fe_3O_4/2-metil 8-hidroksikinolin). Karekterizasyon için Fourier Dönüşümlü Kızılötesi Spektroskopisi (FT-IR), taramalı elektron mikroskobu (SEM), Enerji Dağılımı X-Işını spektrometresi (EDX), Raman ve UV-Vis kullanılmıştır. Yaban turpu peroksidaz (Horseradish Peroksidaz, HRP), mGO@Fe_3O_4/2-metil 8-hidroksikinolin nanokompozit üzerine immobilize edilmiştir. Yöntemin optimum ortam şartlarını elde etmek için pH, mGO@Fe_3O_4/2-metil 8-hidroksikinolin miktarı, enzim miktarı, sıcaklık gibi parametreler taranmış, enzim için şartlar optimize edilmiştir. Daha sonra optimize edilen koşullarda atık sularda bulunabilecek Kongo Kırmızısı, Malahit Yeşili, Evans Blue, Reaktif Black boyalarının giderilmesinde kullanımı araştırılmıştır.

Anahtar Kelimeler: Yaban turpu peroksidazı (HRP), enzim, immobilizasyon, fonksiyonlaştırılmış manyetik grafen oksit, atık sularda boya giderimi.

Bu çalışma, Erciyes Üniversitesi Bilimsel Araştırma Projeleri Birimi tarafından FYL-2019-9137 kodlu proje ile desteklenmiştir.

Kaynaklar

[1] Malik, V., & Pundir, C. S. 2002. Determination of total cholesterol in serum by cholesterol esterase and cholesterol oxidase immobilized and co-immobilized on to arylamine glass. Biotechnology and applied biochemistry, 35(3), 191-197.

[2] Hummers Jr, W.S., Offeman, R. E., Preparation of Graphitic Oxide, J. Am. Chem. Soc., 80, 1958, 1339-1339.

DRAFT

ORAL PRESENTATION

Preparation of probiotic-prebiotic formulations for the development of synbiotic products

Hilal DİKMEN MERAL[1*] (https://orcid.org/0000-0002-3917-4804), Fahriye Şeyma ÖZCAN[2] (https://orcid.org/0000-0002-1816-6557), Osman SAĞDIÇ[1] (https://orcid.org/0000-0002-2063-1462)

[*1]Yildiz Technical University, Chemical and Metallurgical Faculty, Food Engineering Department, İstanbul,Turkey.
[2]TÜBİTAK, Marmara Research Center Food Institute, Kocaeli, Turkey.

*****E-mail: hilaldikmen11@gmail.com

Abstract

Functional products containing probiotics and prebiotics are frequently preferred by consumers to improve body metabolism and achieve a healthier lifestyle. In recent years, a new concept called "synbiotic" has emerged to further increase the positive effects of prebiotics and probiotics that are consumed separately.
When preparing synbiotic combinations, studies involving the growth rate and fermentation profile of different probiotic strains in the presence of different oligosaccharides should be conducted to bring together the species with the best synergistic effect. For this purpose, the synbiotic effects of different prebiotic varieties on probiotic strains were investigated in the present study. The synbiotic effects of *L. rhamnosus* GG, a probiotic strain, with different prebiotic varieties (resistant starch, maltooligosaccharide, pectin, alternan, fructooligosaccharide, isomaltooligosaccharide, FOS LP85, and FOS LP95) added to the medium as a carbon source were investigated. *L. rhamnosus* GG strain was inoculated into modified media with different prebiotics, pH and viability values were measured for 48 h. At the end of the incubation period, the lowest pH and the highest viability values were found to 3.83±0.02-8.92±0.05 log CFU/mL and 3.96±0.05-8.86±0.01 log CFU, respectively, in media added FOS LP85 and FOS LP95. After fermentation, viability values were measured once a week for a month at +4°C. At the end of 28 days, the media with the highest viability of the probiotic bacteria were FOS LP95 (8.16±0.02 log CFU/mL) and isomaltooligosaccharide (8.02±0.03 log CFU/mL) added media, respectively.
These findings showed that the viability and stability of probiotic bacteria varied depending on the rate of prebiotic sugar usage. Furthermore, this study will enable the development of products that keep viable probiotic cells at the appropriate level for a longer time with appropriate prebiotic-probiotic combinations.

Keywords: Probiotics, Prebiotics, Synbiotic combinations

DRAFT

ORAL PRESENTATION

Meldrum's Acid-based Chain Extenders for PET Recycling

Yonca Alkan Goksu[1*] (*ORCID: https://orcid.org/0000-0002-6699-8761*)

[*1] Istanbul Technical University, Faculty of Chemical and Metallurgical Engineering, Department of Metallurgical and Materials Engineering, Istanbul, Turkiye.

*Corresponding author e-mail: alkanyo@itu.edu.tr

Abstract

The global demand for single-use plastics is gradually rising. The amount of global plastic waste is expected to grow from 260 million tons/year in 2016 to 460 million tons/year by 2030 Only 16% of polymer waste were subjected to recycling process while 40% were sent to landfill and 25% of them were incinerated in 2016. Poly(ethylene terephthalate) (PET) composes the majority among those single-use plastics, especially in packaging applications of beverage bottles. Hence, accumulation of its waste in both soil and marine environments is enormous and it is imperative to provide efficient recycling routes immediately to reduce this accretion. The aim of this project is to design a versatile chemical approach to recycle PET waste and provide its reuse in many applications instead of virgin PET.
During melt recycling process of PET, due to high temperatures and applied shear forces, PET's molecular weight significantly decreases. To prevent this, reactive chain extenders such as pyromellitic dianhydride (PMDA), isocyanates or epoxide functionalized oligomers are added to regain the molecular weight by reconnecting the broken chain ends of PET. In this study, novel Meldrum's acid bearing oligomers with tailorable compositions were synthesized to be used in PET recycling. It is known that ketenes are highly reactive intermediates that could be thermally formed from Meldrum's acid derivatives in situ and rapidly react with nucleophiles such as alcohols, carboxylic acids and amines. Our results have revealed that those ketene-based chain extenders resulted in crystallinity increase in recycled PET that could resul better processability during manufacturing processes like extrusion, injection molding, and blow molding.

Keywords: PET, recycling, chemical recycling, ketenes, Meldrum's acid

DRAFT

ORAL PRESENTATION

Isolation, Characterization, and Three-Dimensional Culture of Gemcitabine-Resistant CD326+CD133+CD44+ Lung Cancer Stem Cells Derived from A549 Cancer Cell Line

Bahar DEMİR CEVİZLİDERE [1,2*](ORCID: 0000-0002-2444-8833.), Onur UYSAL[1,2,3](ORCID: 0000-0001-6800- 5607), Hüseyin AVCİ[1,2,4,5] (ORCID: 0000-0003-2079-18 9756), Sibel GUNES BAGİS [1,2,3] (ORCID: 0000-0003- 0846-1170), Tugba SEMERCİ SEVİMLİ [1,2] (ORCID: 0000-0003-19 4856-2304), Murat DİNCER[6] (ORCID: 0000-0002-9146-813X), Aynaz GHORBANİ [1,2] (ORCID: 0000-0001-5516-027X), Gülay BÜYÜKKÖROĞLU [7] (ORCID: 0000-0002-5089-6007, Ayla EKER SARİBOYACİ [1,2,3] (ORCID: 0000-0003-4536-9859)

[1]Cellular Therapy and Stem Cell Production Application and Research Centre, ESTEM, Eskisehir Osmangazi University, Eskisehir, Turkey.
[2]Department of Stem Cell, Institute of Health Sciences, Eskisehir Osmangazi University, Eskisehir, Turkey.
[3]Department of Medical Laboratory Techniques, Vocational School of Health Services, Eskisehir, Turkey.
[4]Department of Metallurgical and Materials Engineering, Engineering and Architecture Faculty, Eskisehir Osmangazi University, Eskisehir, Turkey.
[5]Translational Medicine Research and Clinical Center, Eskisehir Osmangazi University, Eskisehir, Turkey.
[6]Faculty of Medicine, Department of Medicinal Oncology, Eskisehir Osmangazi University, Eskisehir, Turkey.
[7]Department of Pharmaceutical Biotechnology, Faculty of Pharmacy, Anadolu University, Eskisehir, Turkey.

*Corresponding E-mail: bdemir92@gmail.com

DRAFT

Abstract

Cancer remains a global health challenge, with increasing incidence and mortality rates worldwide. The disease's
complexity arises from diverse cell populations within tumors, where specific subsets, known as cancer stem cells (CSCs), drive tumor growth, resistance to treatment, invasion, and metastasis. Conventional therapies like surgery, radiotherapy, and chemotherapy often struggle to target CSCs effectively. Consequently, researchers are actively exploring new drug models to combat various cancer types, driven by CSCs' inherent resistance to standard treatments. Developing innovative strategies to tackle these resilient cancer stem cells is crucial to improve treatment outcomes and patient survival.The primary aim of this study was to establish a Gemcitabine-resistant cancer stem cell line derived from the non-small lung cancer cell line (A549) and to culture it using a Three-Dimensional Culture (3D) approach, enabling the investigation of diverse therapeutic approaches. Initially, in this study a gemcitabine- resistant lung cancer cell line was established through the gradual exposure of A549 cells to gemcitabine. Subsequently, a population of cancer stem cells (CSCs) characterized by a cluster of differentiation (CD)326+CD133+CD44+ phenotypes was isolated through immunoselection using filtration column method, employing gemcitabine-resistant A549 cells purified via colony forming technology. The resistance mechanisms of the obtained gemcitabine-resistant CSCs were elucidated by investigating P-glycoprotein (P-gp, MDR, ABCB1), which has a prominent impact on resistance, utilizing flow cytometry device. Moreover, the positivity of cancer stem cell markers in the gemcitabine-resistant (CD)326+CD133+CD44+ CSCs was assessed using flow cytometry and immunofluorescence staining methods, confirming their CSC characteristics. In addition to investigate their tumorigenic capabilities, the obtained gemcitabine-resistant (CD)326+CD133+CD44+ CSCs were subjected to three- dimensional (3D) in vitro culture. This study establishes gemcitabine-resistant lung cancer cell lines and lung cancer stem cells (CSCs) for the first time. These models offer valuable insights into gemcitabine-resistant CSCs and hold great potential for further research.
This study was supported by grant (TUBİTAK-1002/221S319) from THE SCIENTIFIC AND TECHNOLOGICAL
RESEARCH COUNCIL OF TURKIYE (TUBİTAK)

Keywords: Gemsitabine resistance, Cancer stem cell, Tumorsphere culture

ORAL PRESENTATION

Endotel hücre fonksiyonu devamlılığı için karvakrol ve/veya naringin kullanılabilir mi?

Nadire Kıyak[1,3*](*ORCID:https://orcid.org/0000-0002-7126-2775)*, Eda Becer[2,3](*ORCID: https://orcid.org/0000-0002-2378-128X)*, Hafize Seda Vatansever[3,4](*ORCID: https://orcid.org/0000-0002-7415-9618)*, Aysel Kükner [1] *(ORCID: https://orcid.org/0000-0001-5387-2628)*

[1*]Yakın Doğu Üniversitesi, Tıp Fakültesi, Histoloji ve Embriyoloji Anabilim Dalı, Lefkoşa, Mersin 10 Türkiye.
[2]Doğu Akdeniz Üniversitesi, Eczacılık Fakültesi, Biyokimya Anabilim Dalı, Gazimağusa, Mersin 10 Türkiye.
[3]Yakın Doğu Üniversitesi, DESAM Enstitüsü, Lefkoşa, Mersin 10 Türkiye.
[4]Manisa Celal Bayar Üniversitesi, Tıp Fakültesi, Histoloji ve Embriyoloji Anabilim Dalı, Manisa, Türkiye.

*Sorumlu yazar e-mail: nadire.kiyak@neu.edu.tr

Özet

Bitkisel kaynaklı doğal bileşikler ile yapılan çalışmalar, doğal bileşiklerin çeşitli hastalıklarla ilgili koruycu etkiye sahip olduğunu desteklemektedir. Karvakrol ve naringin'in immun sistem modülatörü olması yanısıra antimikrobiyal, anti-inflamatuar, anti-kanserojen, anti-oksidan ve antifungal etkilere sahip olduğu birçok çalışma ile gösterilmiştir. Karvakrol (2-metil-5-(1-metiletil)-fenol) başlıca kekik ve çok sayıda aromatik bitki tarafından üretilen monoterpenik bir fenoldür. Kekik bitkisinin ana bileşeni olan karvakrol, kekiğin tıbbi bitki olarak kullanılmasında önemli bir etkendir. Naringin ise ağırlıklı olarak portakal ve greyfurt gibi turunçgillerde bulunan biyolojik aktivite bakımından zengin flavonoid grubu üyesidir. Bu çalışmada karvakrol ve naringin'in endotel hücrelerin canlılığı ve proliferasyonu üzerindeki etkilerinin araştırılması amaçlanmıştır. Çalışmada ECV304 endotelyal hücre hattı, 15mM HEPES ve %10 FBS içeren, M199 büyüme medium kullanılarak 37 °C'de ve %5 $C0_2$ içeren nemli bir ortamda kültüre edildi. Farklı karvakrol (10, 20, 50, 100 ve 150µg/ml) ve naringin (10, 25, 50, 100, 150 µM) konsantrasyonları ECV304 hücrelerine 48, 72 ve 96 saatlik sürelerde inkübe edildi. İnkübasyon sonrasında karvakrol ve naringin'in hücre canlılığı ve sitotoksisite etkileri MTT (3- (4,5-dimetiltiazol-2il)-2,5-difenil tetrazolyum bromür) testi ile analiz edildi. MTT analizi sonrasında ECV304 hücrelerinde 10 µg/ml doz ve 72 saat uygulanan karvakrolün hücre sayısını arttırdığı, naringin'in ise 50 µM dozda ve 72 saat inkübasyon süresinde koruyucu olduğu saptandı. Sonuç olarak, bitkisel kaynaklı karvakrol ve naringin'in ECV304 endotel hücresinde hücre canlılığını koruyucu yönde etkisi olabileceği düşünülmüştür. Sonuçlarımız, endotel hasarı oluşturabilen hastalıklarda doğal yollardan alınabilen kalvakrol ve naringin'in endotel hücre fonksiyonuna destekleyici etkilerinin incelenebileceği in vitro ve in vivo deneysel modeller için bir ön veri niteliğindedir.

Anahtar Kelimeler: Karvakrol, Naringin, ECV304, MTT

DRAFT

ORAL PRESENTATION

Assessment of measurement uncertainty of blood lipid parameters according to ISO/TS 20914 guidance

Bagnu Orhan*[1] (ORCID: https://orcid.org/0000-0003-1779-7784), Büşra Üresin Yazlak[1] (ORCID: https://orcid.org/0000-0002-5089-4167), Merve İnce[1] (ORCID: https://orcid.org/0000-0002-7655-2851), Berrin Berçik İnal[1] (ORCID: https://orcid.org/0000-0002-9098-4140)

[1] University of Health Sciences, Istanbul Training and Research Hospital, Department of Medical Biochemistry, Istanbul, Turkey

*Corresponding author e-mail: bagnuorhan@gmail.com

Abstract

Measurement uncertainty (MU), as a value indicating the distribution of measurement results, is a worthful tool for the assessment of analytical performance and interpretation of results in medical laboratories. The recommendation for calculating MU is outlined in the ISO/TS 20914:2019 guideline. In this study, it's aimed to calculate the MU values for the parameters of total cholesterol, high- density lipoprotein (HDL) cholesterol, low- density lipoprotein (LDL) cholesterol, and triglyceride according to the calculation model in the ISO/TS 20914:2019 guideline, and to compare these with the maximum allowable measurement uncertainty (MAU) values. The MU values of total cholesterol, HDL cholesterol, LDL cholesterol, and triglyceride parameters were calculated using the long-term imprecision (u_{Rw}) acquired from 6-month internal quality control data and the calibrator uncertainty (u_{cal}). Bias (%) calculated from 12-month external quality assessment data was compared with the desirable bias (%) values obtained from Westgard's biodatabase. In accordance with the recommendation of the ISO/TS 20914 guideline, for all parameters, the obtained biases (%) were disregarded in MU calculations as they were within the desirable bias (%) limits. The combined standart uncertainty, and expanded uncertainty were calculated for these blood lipid parameters analyzed using the Roche Cobas 8000 c 702 autoanalyzer (Roche Diagnostics, Mannheim, Germany), and compared with the European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) 2022 MAU values. The MU values of total cholesterol, HDL cholesterol, LDL cholesterol, and triglyceride analytes were evaluated as within MAU limits. The MU value is an important indicator of quality in the assessment of analytical performance.

Keywords: Measurement uncertainty, quality assurance, medical laboratory, blood lipid parameters

DRAFT

ORAL PRESENTATION

Derin ötektik çözücülerle gıda atıklarından biyoaktif bileşen eldesi

Ayşenur Erdem[1*] (*ORCID: https://orcid.org/0009-0003-4202-6866.*), Senem Kamiloğlu Beştepe[1,2] (*ORCID: https://orcid.org/0000-0003-3902-4360*)

[*1]Bursa Uludağ Üniversitesi, Ziraat Fakültesi, Gıda Mühendisliği Bölümü, Bursa, Türkiye
[*2]Bursa Uludağ Üniversitesi, Bilim ve Teknoloji Uygulama ve Araştırma Merkezi (BİTUAM), Bursa, Türkiye

*Sorumlu yazar e-mail: aysenurrerdemm@hotmail.com

Özet

Gıda israfı dünya genelinde artarak büyük bir problem haline gelmeye başlamıştır. Gıda israfı yalnızca gıdaların çöpe gitmesi demek değildir. Aynı zamanda doğal kaynakların da hızla tükendiği anlamına gelmektedir. Bu durum doğal kaynakları, ekonomiyi, tarımı, çevre ve doğal dengeyi ciddi bir şekilde etkileyen önemli bir konudur. Bu sebeple gıda israfının önüne geçilmeli ve gıda atıkları değerlendirilmelidir. Ayrıca gıda yan ürünlerinin biyoaktif bileşik kaynağı olarak kullanılması, günden güne artan organik atıkların sonucunda ortaya çıkan çevre sorununu en aza indirebilmek için iyi bir fırsattır. Ancak gıda atıkları değerlendirilirken çevreyi ve doğayı unutmamak gerekmektedir. Gıda yan ürünlerinden biyoaktif bileşik ekstraksiyonu için yüksek miktarda organik çözücü kullanılmaktadır. Günümüzde ekstraksiyon işlemi için yaygın olarak kullanılan geleneksel organik çözücüler, uçucu olmaları ve ekstraktlarda kalıntı bırakarak toksik etkiye sebep oldukları için oldukça tehlikelidir, çevreye zarar vermektedir. Yeşil ve sürdürülebilir kimya çerçevesinde derin ötektik çözücüler dikkat çekmektedir. Derin ötektik çözücüler son zamanlarda biyoaktif bileşiklerin ektraksiyonu için organik çözücülere alternatif olarak kullanılmaktadır. İki veya daha fazla bileşiğin karışması sırasında, bu karışımı oluşturan bileşiklerin erime noktalarından daha düşük değere sahip olan karışım derin ötektik çözücü olarak adlandırılmaktadır. Derin ötektik çözücüler bir hidrojen bağ alıcısı ve bir hidrojen bağ donörünün uygun bir sıcaklıkta karıştırılmasıyla hazırlanmaktadır. Son dönemdeki çalışmalar, derin ötektik çözücüler kullanılarak yapılan ekstraksiyon işleminde, kuersetin, antosiyaninler, antioksidanlar ve kateşinler gibi bazı biyoaktif bileşiklerin yüksek verim ile elde edilebildiğini göstermektedir.

Anahtar Kelimeler: Biyoaktif bileşen, derin ötektik çözücüler, gıda atıkları

DRAFT

ORAL PRESENTATION

Kurak koşullarda yapılan bağcılıkta yağmur suyu hasadının etkinliği

İbrahim Samet GÖKÇEN (https://orcid.org/0000-0002-1857-7911)

Kilis 7 Aralık Üniversitesi, Ziraat Fakültesi, Bahçe Bitkileri Bölümü, Kilis, Türkiye

ibrahimsgokcen@kilis.edu.tr

Özet

Su, canlı yaşamı için en önemli doğal kaynaktır. Bunun yanında yeryüzündeki tatlı su miktarı, dünyanın toplam su varlığının %1'inden bile azdır. Bu kapsamda Türkiye düşünülenin aksine, su zengini bir ülke olmayıp, yılda kişi başına düşen su miktarı 1566 m^3 ile "su sıkıntısı çeken" bir ülkedir. Ülkemiz iklim yapısı olarak yarı kurak bir iklime sahip olup, son 50 yıllık yağış miktarına bakıldığında ortalama 618.9 mm yağış aldığı görülmektedir. Kuraklığın artışı sonucu yağış miktarının azalmasıyla tarımsal üretimde çeşitli sorunlar ortaya çıkmaktadır. Ülkemizin en önemli tarım ürünlerinden biri olan üzüm, yetiştiriciliği yapılan bazı bölgelerde su yetersizliğinden dolayı gelişim ve ürün verimi açısından yetersiz kalmaktadır. Ayrıca iyi beslenemeyen omcalarda, hastalık ve zararlılara karşı direnç daha düşük kalmaktadır. Tarımsal üretimde başarılı olabilmek için yağmur sularını idareli kullanmak ve arazinin uygun şekilde yönetilmesi gerekmektedir. Kurak ve yarı kurak alanlara düşen, az ve verimsiz yağmur sularının yerinde biriktirilmesi veya yüzey akışa geçirilerek depolanması işlemi yağmur su hasadı olarak tanımlanır. Amaç, bağlarda arazinin yukarı bölümlerinin yüzeyini geçirimsiz hale getirerek, yağmur suyunu yüzey akışı ile havzanın alt bölümünde bulunan omcaların etkili kök bölgesine yönlendirmektir. Metrekareye 40 mm yağışın düştüğü bir günde, $4m^2$'lik yağmur su hasadı uygulanmış bir omcaya, 160 mm (16 kg) su sağlanabilmektedir. Sonuç olarak yağmur suyu hasadı uygulaması sürdürülebilir ve çevre dostu bir uygulamadır. Bağcılıkta verimi arttırıp, ilaç kullanımını azaltmaya yönelik üreticiye ekonomik anlamda yarar sağlayacaktır. Yağmur suyu hasadı ülkemizde çok yeni bilinen ve üreticilerimize tanıtılması gereken, ekonomik ve faydalı bir uygulamadır.

Anahtar kelimeler: Bağcılık, kuraklık, su hasadı, yağmur.

DRAFT

ORAL PRESENTATION

Toprak havuzlarda deniz balıkları üreten işletmelerde bir üretim periyodunda sedimentte meydana gelen element miktarı değişimi

Rifat Tezel[1*] (*ORCID: https://orcid.org/0000-0002-0870-7049*),
Kenan Güllü[2] (*ORCID: https://orcid.org/ 0000-0002-8604-8798*)

[*1]Muğla Sıtkı Koçman Üniversitesi, Su Ürünleri Fakültesi, Su Ürünleri Yetiştiriciliği Bölümü, Muğla, Türkiye

*Sorumlu yazar e-mail: rifattezel@mu.edu.tr

Özet

Toprak havuzlarda deniz balıkları üretimi; çoğunluğu küçük aile işletmelerinden oluşan su ürünleri işletmelerinde, tabana yayılmış bir ekonomik faaliyet olarak yürütülmektedir. Büyük hacimli toprak havuzlarda üretim yapıldığından ve günlük su değişim miktarı, diğer su ürünleri işletmelerine göre düşük olduğundan, bir üretim periyodu süresince havuz tabanında organik madde birikimi meydana gelmektedir. Bu çalışmada, toprak havuz işletmelerinde bir üretim periyodu süresince, havuz tabanındaki element madde miktarının değişimi araştırılmıştır. Milas Savran'da bulunan bir toprak havuz işletmesinde 3 adet toprak havuz, bu çalışma kapsamında kullanılmıştır. Kullanılan havuzlar kontrol (K), taban tahliyeli (T) ve de taban tahliyeli ve havalandırmalı (TH) olarak kodlanmıştır. Kontrol havuzunda geleneksel tahliye sistemi, diğer havuzlarda ise havuz tabanındaki organik madde birikimini azaltmak amacıyla geliştirilen havuz taban tahliye sistemi kullanılmıştır. Havuzların tabanlarında element miktarı değişimlerini izlemek için 6 ayda bir sediment örnekleri alınarak analiz edilmiştir. Örneklerde toplam organik karbon, azot, fosfor, potasyum, kalsiyum, magnezyum, kükürt, sodyum, demir, mangan, çinko, bakır, bor, molibden miktarları belirlenmiştir. K, T ve TH havuzlarında sedimentte toplam organik karbon miktarı çalışma başlangıcında sırasıyla 50,16±2,9; 51,3±5,2; 49,2±1,1 g/kg olarak ve çalışma sonunda ise 59,6±0,7; 45,8±3,4; 51,4±5,1 g/kg olarak belirlenmiştir. Toplam azot miktarı çalışma başlangıcında K, T ve TH havuzları için sırasıyla 1457±133, 1620±776, 1430±459 mg/kg olarak ve çalışma sonunda ise 2847±827, 2797±1116, 2257±165 mg/kg olarak belirlenmiştir. Fosfor miktarı ise çalışma başlangıcında sırasıyla 602±124, 546±49, 558±446 mg/kg olarak ve çalışma sonunda ise 1332±96, 1107±83, 1156±230 mg/kg olarak bulunmuştur. Yapılan çalışma sonucunda, bir üretim dönemi sonrasında, K havuzunun taban çamurunda toplam organik karbon miktarının arttığı T havuzunda azaldığı ve TH havuzunda sabit kaldığı görülmüştür. Tüm havuzlarda sedimentte azot ve fosfor miktarlarının arttığı görülmüş ancak başlangıç ve bitiş değerleri arasında istatistiksel fark bulunamamıştır.

Anahtar Kelimeler: Su ürünleri yetiştiriciliği, Balık üretimi, Su kalitesi, Tarla balıkçılığı

Teşekkür: Bu çalışma, Muğla Sıtkı Koçman Üniversitesi Bilimsel Araştırma Projeleri Koordinasyon Birimi tarafından 18/050 proje numarası ile Doktora Tez Projesi olarak desteklenmiştir. Desteklerinden ötürü Bilimsel Araştırma Projeleri Koordinasyon Birimi'ne teşekkürlerimizi sunarız.

DRAFT

ORAL PRESENTATION

Synthesis, spectral characterization and molecular docking studies on new metal complexes of a schiff base derived from 3,5-dibromo-4-methoxysalicylaldehyde

Kadir Merkit[1*] *(ORCID: https://orcid.org/0000-0001-6116-1121),*
Sümeyra Tuna Yıldırım[2] *(ORCID: https://orcid.org/0000-0001-5564-9630)*

[1]Erzincan Binali Yıldırım University, Health Sciences Institute, Department of Pharmaceutical Sciences, Erzincan, Türkiye
[2]Erzincan Binali Yıldırım University, Faculty of Pharmacy, Department of Analytical Chemistry, Erzincan, Türkiye

*Corresponding author e-mail: eczmerkit@gmail.com

Abstract

Salicylaldehyde, is a common chemical raw material, has frequently been used to synthesis aromatic schiff base ligands. Its hydroxyl and aldehyde group are in the adjacent position, so it is easy to take place substitution reaction at two positions. Novel Schiff base ligands can be prepared by the reaction of salicylaldehyde with different amines at 1:1, 1:2 and 2:1 ratio. The chelating ligands containing O and N donor atoms show widely biological activity and have special interest on bonding to metal ions [1] and substituted salicylaldehydes are potent antioxidant, anticancer, antimicrobial, antibacterial and antifungal agents and may have the potential of chemotherapy [2]. In particular, the effective application of cisplatin as an antitumor treatment in the clinic has led to the widespread interest in inorganic metal drugs [3]. Today designing new molecules that can interact with nucleic acids and trigger apoptosis, the discovery of new DNA-targeted anticancer drugs for chemotherapy is one of the most promising strategies. In this study, a Schiff base ligand was synthesized and then eight complexes prepared from this ligand. Elemental analysis and some spectroscopic techniques such as FT-IR, ^{1}H-NMR, ^{13}C-NMR, XRD, SEM and UV-Vis were used at structural characterization [4, 5]. To elucidate the mechanism of action of the synthesized compounds, some descriptive molecular properties were calculated using the Schrödinger Suite computer-aided molecular modeling program, the active region where the coupling would take place with the Receptor Grid Generation module was determined, and the interactions of the possible drug molecule candidates synthesized with the Structure-Based Drug Design (SBDD) method with the target receptor region were calculated.

Keywords: Ligand, Molecular Modelling, Salicylaldehyde, Schiff Base, Metal Complex

References

[1] Böhme, U., & Günther, B. (2007). Five and six-coordinate silicon complexes with an O, N, O′-chelating ligand derived from o-hydroxyacetophenone–N-(2-hydroxyethyl) imine. *Inorganic Chemistry Communications*, *10*(4), 482-484.
[2] Shekhar, S., Khan, A. M., Sharma, S., Sharma, B., & Sarkar, A. (2022). Schiff base metallodrugs in antimicrobial and anticancer chemotherapy applications: a comprehensive review. *Emergent Materials*, *5*(2), 279-293.
[3] Wang, X., Wang, X., Jin, S., Muhammad, N., & Guo, Z. (2018). Stimuli-responsive therapeutic metallodrugs. *Chemical reviews*, *119*(2), 1138-1192.
[4] Sharma, A., & Shah, M. (2013). Synthesis and characterization of some transition metal complexes derived from bidentate Schiff base ligand. *Journal of Applied Chemistry*, *3*(5), 62-66.
[5] Rajimon, K. J., Elangovan, N., Khairbek, A. A., & Thomas, R. (2023). Schiff bases from chlorine substituted anilines and salicylaldehyde: Synthesis, characterization, fluorescence, thermal features, biological studies and electronic structure investigations. *Journal of Molecular Liquids*, *370*, 121055.

ORAL PRESENTATION

Synthesis and spectroscopic studies of a new schiff base and its metal complexes

Sümeyra Tuna Yıldırım[1] *(ORCID: https://orcid.org/0000-0001-5564-9630)*,
Kadir Merkit[2*] *(ORCID: https://orcid.org/0000-0001-6116-1121)*

[1]Erzincan Binali Yıldırım University, Faculty of Pharmacy, Department of Analytical Chemistry, Erzincan, Türkiye
[2]Erzincan Binali Yıldırım University, Health Sciences Institute, Department of Pharmaceutical Sciences, Erzincan, Türkiye

*Corresponding author e-mail: eczmerkit@gmail.com

Abstract

Schiff bases or azomethines are among the most important groups of biomolecules. These compounds have both remarkable biological activities and a variety of practical applications. There is increasing interest in the discovery of new Schiff base ligands because of their proven usefulness as attractive precursors, sometimes with a different mechanism of action than clinically approved anticancer agents, in the design of new cytotoxic and cytostatic agents [1]. Metal complexes prepared by the coordination of Schiff bases with metal ions play an important role in pharmaceutical sciences due to their multiple and important activities. Schiff bases are versatile pharmacophores capable of forming chelating complexes with several metals in different oxidation states. Although the biological and pharmacological effects of these ligand and metal complexes are known, their use is increasing, especially in cancer treatment [2]. With the determination of the chemotherapeutic effect of cisplatin, there have been developments in other metal complexes with anticancer activity. Cisplatin-related compounds such as oxaliplatin/carboplatin are known to be used worldwide for the treatment of cancer. However, various side effects such as toxicity and drug resistance are observed that limit the clinical use of these drugs. These limitations need to be overcome by discovering new Schiff base ligands for cancer therapy. In this research, a Schiff base ligand and its metal complexes were successfully synthesized [3]. Elemental analysis and some spectroscopic techniques such as FT-IR, ^{1}H-NMR, ^{13}C-NMR, XRD, SEM and UV-Vis were used at structural characterization [4]. It was determined that the metal/ligand ratio was 1:2 in all complexes and that Schiff base ligand was bonded to the metal atom with the nitrogen atom in the imine and the phenolic oxygen atom.

Keywords: Azomethine, Ligand, Metal Complex, Schiff Base, Structural Characterization

References

[1] Tadele, K. T., & Tsega, T. W. (2019). Schiff Bases and their metal complexes as potential anticancer candidates: A review of recent works. *Anti-Cancer Agents in Medicinal*, *19*(15), 1786-1795.
[2] Wang, X., Wang, X., Jin, S., Muhammad, N., & Guo, Z. (2018). Stimuli-responsive therapeutic metallodrugs. *Chemical reviews*, *119*(2), 1138-1192.
[3] Sharma, A., & Shah, M. (2013). Synthesis and characterization of some transition metal complexes derived from bidentate Schiff base ligand. *Journal of Applied Chemistry*, *3*(5), 62-66.
[4] Rajimon, K. J., Elangovan, N., Khairbek, A. A., & Thomas, R. (2023). Schiff bases from chlorine substituted anilines and salicylaldehyde: Synthesis, characterization, fluorescence, thermal features, biological studies and electronic structure investigations. *Journal of Molecular Liquids*, *370*, 121055.

DRAFT

ORAL PRESENTATION

PARP-1 Levels in Breast Cancer Patients

Merve Alpay* (*ORCID: 0000-0002-8782-9561)*

Duzce University, Faculty of Medicine, Department of Medical Biochemistry, Düzce, TR

*E-mail: mervealpay@duzce.edu.tr

Abstract

PARP-1 (Poly ADP-ribose polymerase 1) is an enzyme with crucial functions in cells, including DNA repair and preservation of genetic integrity. Drugs known as PARP inhibitors are used in the treatment of certain cancer types, especially breast cancer. The relationship between PARP-1 and breast cancer involves the formation of BRCA mutations, which impact the DNA repair mechanism and lead to genetic damage. In this context, PARP-1 detects DNA damage and works to repair it. Both the PARP-1 enzyme and PARP inhibitors are utilized as effective treatment strategies in breast cancer patients, particularly those carrying BRCA mutations, by targeting the disruption of DNA repair mechanisms. The research was conducted on a group of 28 patients diagnosed with breast cancer within the age range of 18-65 years, who sought treatment at the Oncology Clinic of Düzce University Hospital, along with 19 healthy individuals. Within the patient group, samples were isolated from individuals who had not commenced active treatment protocols and whose malignant proliferation had not been suppressed, and their ADP-r levels were examined. Routine parameters related to breast cancer diagnosis and prognosis were analyzed using an autoanalyzer, while ADP-r levels were measured using a spectrophotometric method. According to the findings, the ADP-r level in the untreated breast cancer group increased by 34.76% compared to healthy individuals, and a 20.66% increase was observed in the Stage 2 breast cancer patients who had undergone the same treatment protocol. PARP enzymes play a significant role in ADP ribosylation processes. Evidence suggests that dysregulation of ADP ribosylation exists in some cancer types. These abnormalities can render cancer cells vulnerable to DNA damage. Additionally, ADP ribosylation has been observed to increase excessively or abnormally. In the study, a significant increase in ADP-r was observed in breast cancer patients diagnosed with Stage 2, compared to healthy individuals. In the group of patients receiving chemotherapy treatment, this increase was attributed to the direct impact of PARP-1 inhibition on lowering ADP ribosylation and affecting DNA repair mechanisms. For targeted treatment strategies in breast cancer patients, considering the relationship between the ADP ribosylation mechanism, PARP-1 levels, and the patient's individual condition, a personalized approach must be planned.

Keywords: PARP-1, Breast cancer, ADP ribosylation

DRAFT

ORAL PRESENTATION

Synthesis, Characterization of Gallic acid Nanoflowers and Anticancer Activity on MCF7, A549 cell line

Burcu Yılmaz Somtürk[1*] (ORCID:https://orcid.org/0000-0002-4375-8669)

[1]Drug Application and Research Center, Erciyes University, 38280 Kayseri, Turkey

*burcu_smtrk@hotmail.com

Abstract

Gallic acid has been reported to possess several biological activities such as antioxidant, antimicrobial, anticancer and anti-inflammatory. Nanoparticles synthesized using organic molecule have many advantages such as easy availability and low cost. Nanoparticles are widely used in pharmaceuticals, drug delivery, drug release and diagnostic applications. Nanoflowers are a kind of nanoparticle with a high surface area due to their flower-like structure. This high surface area makes the molecule more open to biological interactions. Metallic nanoflowers synthesized using plant extracts have attracted great interest in the scientific community due to their enhanced biological activities and unique physicochemical properties. In this study, nanoparticle synthesis was carried out for the first time by using extracts of gallic acid as organic part and metal ions (Cu^{2+}, Co^{2+} and Zn^{2+}) as inorganic part. Characterization of nanoflowers was performed by SEM, EDX, XRD, FTIR and elemental mapping. Then, the anticancer activities of the synthesized nanoflowers were evaluated. A549 and MCF7 cell lines were used for anticancer activity. In the end, the anticancer effect of the nanoflowers were higher than that of the gallic acid alone. It is planned to investigate the nanoflower showing anticancer activity with further in vitro and in vivo methods.

Keywords: Gallic acid, Anti cancer, nanoflower

References

[1] Somturk, B., Dayan, S., Ozdemir, N., Kalaycıoğlu Özpozan, N. Catalytic performance improvement with metal ion changes for efficient, stable, and reusable superoxide dismutase–metalphosphates hybrid nanoflowers, Chemical papers, 2022, 76 (7), 4245-4260 https://doi.org/10.1007/s11696-022-02179-z

[2] Alhayali, N.I., Kalaycıoğlu Öpozan, N., Dayan, S., Özdemir, N., Somtürk Yılmaz, B., (2021), Catalase/Fe_3O_4@Cu^{2+} hybrid biocatalytic nanoflowers fabrication and efficiency in the reduction of organic pollutants Polyhedron 194, 114888

DRAFT

ORAL PRESENTATION

Effects of medical ozone therapy against fluoxetine induced DNA damage

Dilek Aşcı Çelik[1*] *(ORCID: https://orcid.org/0000-0002-2914-4695)*,
Vehbi Atahan Toğay[1] *(ORCID: https://orcid.org/0000-0003-4722-3845)*

[*1]Süleyman Demirel University, Faculty of Medicine, Department of Medical Biology, Isparta, Türkiye

*Corresponding author e-mail: dilekasci@sdu.edu.tr

Abstract

Fluoxetine is an antidepressant which is selective serotonin reuptake inhibitor. High doses may have serious side effects. It has been reported to cause an increase in oxidative stress which is one of the most important causes of DNA damage. There have been studies on the therapeutic effect of ozone (O_3), including antioxidant activity in recent years. Therefore, it was aimed to determine the effect of acute high-dose fluoxetine treatment and ozone therapy on DNA damage. 32 wistar rats were divided into 4 groups and exposed to 24 mg/kg oral fluoxetine, 1 mg/kg intraperitoneal ozone (95% O_2 + 5% O_3) or their combination for 7 days, and a negative control group was formed. At the end of the experiment, blood was taken, and alkaline comet assay was applied, 100 cells per rat were photographed under a fluorescent microscope. The photographs were analyzed with the OpenComet, and the results were compared using a one-way anova. The "%TailDNA" parameter was determined as mean ± standard error, and DNA damage was found to be 5.67 ± 0.58, 6.63 ± 0.64, 8.16 ± 0.70, and 6.90 ± 0.76 for control, ozone, fluoxetine, and fluoxetine + ozone groups, respectively. The fluoxetine group caused a statistically significant increase in DNA damage compared to the control group ($p<0.05$). Interestingly, there was also an increase in DNA damage in the ozone-treated group, but this increase was not statistically significant ($p>0.05$). Fluoxetine + ozone treatment reduced fluoxetine-induced damage, but it was not statistically significant ($p>0.05$). The DNA damage of the fluoxetine + ozone group was still higher than the control group, but there was also no statistically significant difference when compared to the control group ($p>0.05$). Accordingly, acute high-dose fluoxetine treatment increased DNA damage in rat blood tissue, although the damage was reduced by ozone therapy, it was not therapeutic.

Keywords: Fluoxetine, Intraperitoneal ozone, Comet Assay

DRAFT

ORAL PRESENTATION

Kırgızistan'ın Kırmızı Kitap'ında yer alan tıbbi bitkilerin Türkiye'deki yakın türler ile karşılaştırılması

Zhoomart Moldaliev[1*] (*ORCID: https://orcid.org/*0000-0001-5525-7629),
Hasan Özdemir[2] (*ORCID: https://orcid.org/0009-0009-0218-7554*),
İsmail Kaşoğlu[3] (*ORCID: https://orcid.org/0000-0003-2948-913X*)

[*1]Oş Devlet Üniversitesi, Tıp Fakültesi, Genel Klinik Biyokimya ve Patofizyoloji, Oş, Kırgızistan
[2]Oş Devlet Üniversitesi, Doğa Bilimleri, Turizm ve Ziraat Teknolojileri Fakültesi, Biyoloji, Oş, Kırgızistan
[3]Sanko Üniversitesi, Lisansüstü Eğitim Fakültesi, Moleküler Tıp, Gaziantep, Türkiye

*Sorumlu yazar e-posta: joomart77@oshsu.kg

Özet

Modern tıbbın evrimi sürecinde bitkilerin insan vücudundaki gösterdikleri etkileri yani -farmakodinamik özellikleri- sayesinde başlıca kullanılmışlardır. İlk ilkel farmakopeler belli standartları olmayan şekilleriyle günümüzde bazı aktarlarda bulunabilmektedir. Bununla birlikte geleneksel tıbbi bitkilerin modern tıpta kullanımı ve içerdikleri biyokimyasal bileşiklerine bağlı olarak yeni potansiyel tedavi potansiyelleri hala araştırılmakta ve bu araştırmalara dayanarak ana tedavi hammaddesi veya ana tedavi yanında -bazen suistimal de edilerek- takviye gıda olarak sunulmaktadır. Modern ilaçların üretiminde yeni moleküllerin keşfi ve suistimallerin engellenmesi açısından bitkilerin tıbbi özelliklerinin araştırılıp kayıt altına alınması önemlidir.
Daha önce tıpta kullanılan bitkilerle ilgili bilgilerin kaybolması, dahası florada yok olan türler nedeniyle bu potansiyelin de yitirilmesi bir çeşit değer kaybıdır. Bu elde edilmiş bilgilerden en önemli kaynaklar Biruni ve İbni Sina tarafından kazandırılmıştır. Bu iki bilim insanı aynı dönemde Türkistan ve güneyinde önemli çalışmalar yapmış ve diğer bilimlerin yanında eczacılık ve tıp dünyasına önemli katkılar vermişlerdir. Türkistan'daki bu tıp tarihinde önemli rol oynayan iki bilim insanının bulunduğu yerlerdeki tıbbi bitkilerin özelliklerinin bilinmesi önemlidir.
Çalışmamızda Kırgızistan'da türü tükenme tehlikesine girmesi nedeniyle ülkede "Kırmızı Kitap" (Kırgızca: Кызыл китеп) adında envanter kitabında koruma listesinde yer alan endemik bitkilerinden daha önce çeşitli çalışmalarla ortaya konmuş olan 15 bitkinin tıbbi özellikleri derlenmiştir. Ayrıca bu türlere yakın ülkemizdeki türler ile tıbbi özellikleri karşılaştırılmıştır. Böylece bu karşılaştırmada bitkilerin biyokimyasal kompozisyon karşılaştırması sayesinde ülkemizdeki veya Kırgızistan'daki bu yakın türlerin diğer potansiyel tıbbi özellikleri konusunda bilgi edinilmesi hedeflenmiştir.

Anahtar kelimeler: Tıbbi bitkiler, Kırgızistan Kırmızı Kitap, Kırgızistan Endemik Florası, Geleneksel Tıp

DRAFT

ORAL PRESENTATION

18-25 yaş arası yurtta kalan kız öğrencilerin uyku kalitesinin beslenme davranışlarına etkisi

Aleyna Sancaklı[1]* (ORCID: https://orcid.org/0009-0006-2554-8867), Emine Soyupek[1] (ORCID: https://orcid.org/0009-0002-7970-7334), Nisanur Tuna[1] (ORCID: https://orcid.org/0009-0009-6499-5143), Ayşe Hümeyra İslamoğlu[1] (ORCID: https://orcid.org/0000-0002-2138-5996)

Marmara Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik, İstanbul, Türkiye

*aleynasancakli.9@gmail.com

Özet

Bu çalışmada İstanbul'daki yurtlarda kalan 18-25 yaş arası kız öğrencilerin uyku kalitesinin beslenme davranışları üzerine etkisinin incelenmesi amaçlanmıştır. Çalışma İstanbul ili Kadıköy, Maltepe ve Ataşehir ilçelerindeki kız öğrenci yurtlarında Mart 2023-Mayıs 2023 tarihleri arasında yapılmıştır. Çalışma çevrim içi anket yoluyla, yurtta kalan gönüllü 163 kız öğrenciyle yürütülmüştür. Veriler; Pittsburgh Uyku Kalite İndeksi (PUKİ) ve Sağlıklı Beslenmeye İlişkin Tutum Ölçeği (SBİTÖ) aracılığıyla toplanmıştır. İstatiksel analiz SPSS 21.0 paket programı ile yapılmıştır. Çalışmaya katılan bireylerin yaş ortalaması 21.75±1.43 yıldır. Bireylerin PUKİ skoru ortalaması 7,75±2,74'tür. Çalışmaya katılan bireylerin PUKİ ölçek puanı sınıflamasına göre %23,9'unun (n=39) sağlıklı uyku, %76,1'inin (n=124) kötü uyku sınıfında olduğu bulunmuştur. Katılımcıların toplam SBİTÖ puan ortalaması 71,34±10,87 olarak elde edilmiştir. Sağlıklı Beslenmeye İlişkin Tutum Ölçeği'nin alt boyutlarından "Olumlu Beslenme"nin puan ortalaması 15,83±4,57; "Kötü Beslenme"nin puan ortalaması 18,25±4,39 olarak saptanmıştır. Toplam SBİTÖ puanı ile PUKİ skoru arasında negatif yönlü istatistiksel olarak anlamlı bir ilişki bulunmuştur (p=0,020; r=-0,183).

Anahtar Kelimeler: Beslenme durumu, uyku kalitesi, üniversite öğrencisi, sağlıklı beslenme tutumu

DRAFT

ORAL PRESENTATION

An Indispensable Method from Separation to Determination: Adsorption

Kazım Köse* (*ORCID: https://orcid.org/0000-0003-1467-2898*)

*Hitit University, Alaca Avni Çelik Vocational School, Medical Laboratory Techniques, Çorum, Turkey

*Corresponding author e-mail: kazimkose@hitit.edu.tr

Abstract

Adsorption, which is based on the principle of attachment of molecules to a solid support as a result of the interaction of molecules with ligands, is a phenomenon that exists in the functioning of nature itself, considering that flower dust adhering to the feet of bees helps plants reproduce. Adsorption and its techniques, which are as old as living things, have come a long way scientifically in the last hundred years and many adsorbents/ligands have been developed. Finding a suitable ligand for the target molecule, modifying the polymeric structure and performing separation, removal and determination after interaction without any problems is a very challenging and knowledge-intensive task. However, although it is very advantageous in terms of easy applicability and cost, considering the environmental and health effects of the chemicals used, there is a need for more 'green' adsorbent designs. Although adsorbents using cellulose materials have been developed recently, it is urgent to introduce ‘greener’ modifying agents into this field, considering the future of the method, since organic chemicals cannot be completely removed. In this presentation, besides adsorbent development methods, examples of adsorption will be given and information will be shared especially on the development of adsorbents based on cellulose-based materials.

Keywords: Adsorption, Separation, Removal, Ligand, Modification, Detection.

DRAFT

ORAL PRESENTATION

Trend Analysis of Some Economic Fishes in the Mediterranean Sea

Ferhat BÜYÜKDEVECİ[1*] (*ORCID: https://orcid.org/ 0000-0002-8531-525X*)

[*1] Adana Directorate of Provincial Agriculture and Forestry, Adana, Türkiye

*Corresponding author e-mail: ferhat.bykdeveci@gmail.com

Abstract

The Turkish Ministry of Agriculture and Forestry and the Turkish Statistical Institute (TUİK) regularly gather statistical data regarding commercial fish species captured in the Turkish coast of the Mediterranean. This collected statistical data is periodically shared with international platforms, especially FAO. Although the accuracy of the catch from fisheries in Turkey has been debated by many studies, it has been proven by different studies that the trends of time series catch data are reliable. Fisheries statistics are therefore a valuable source of data for stock assessment and fisheries management. In this study, the changes in the catches of some economically important species on the Mediterranean coast of Turkey were evaluated by trend analysis. These species include red mullet (*M. barbatus*), gilthead seabream (*S. aurata*), true lizardfish (*S. undosquamis*), grey mullet (*M. cephalus*), European seabass (*D. labrax*), bluefish (*P. saltatrix*), common pandora (*P. erythrinus)*, white grouper (*E. aeneus*) and common sole (*S. solea*). Twenty-year data sets derived from TUIK databases covering the years 2000–2020 were used in this context. When the time series catches are examined generally, it is observed that the catches peaked at the end of 2010 and subsequently had a massive decline. *M. barbatus* (24.46%) was captured the most frequently among these nine species, whereas *D. labrax* (2.1%) was the least frequently caught, according to an analysis of the catch amounts provided by TUIK databases in terms of species. According to the trend analysis results, red mullet ($p>0.05$), true lizardfish ($p<0.001$), common pandora ($p<0.001$) showed positive trend while gilthead seabream ($p<0.001$), grey mullet ($p<0.001$), european seabass ($p<0.001$), bluefish ($p<0.001$), white grouper ($p>0.05$) and common sole showed negative trend.

Keywords: Mediterranean Sea, Trend Analysis, TUİK, Economic Fishes

DRAFT

ORAL PRESENTATION

The Phe19, Leu22, and Trp23 residues of p53 play a role in efficient Ser15 phosphorylation by DNA-PK

Merve Ateş[1] (*ORCID: 0000-0002-9665-9963*), Münire Sinim[1] (*ORCID: 0000-0003-2512-1768*), Ümit Uzun[2] (*ORCID: 0000-0002-0585-2865*), Ali Osman Kılıç[3] (*ORCID: 0000-0002-5506-0866*), Osman Birol Özgümüş[4] (*ORCID: 0000-0002-3665-6584), Serap Pektaş[1*] (ORCID:0000-0003-0497-6257)*

[*1]Recep Tayyip Erdogan University, Faculty of Arts and Sciences, Department of Chemistry, Rize, Turkey.
[2]Karadeniz Technical University, Faculty of Medicine, Department of Medical Biology, Trabzon, Turkey.
[3]Karadeniz Technical University, Faculty of Medicine, Department of Medical Microbiology, Trabzon, Turkey.
[4]Recep Tayyip Erdogan University, Faculty of Medicine, Department of Medical Microbiology, Rize, Turkey.

*Corresponding author e-mail: serap.pektas@erdogan.edu.tr

Abstract

The p53 cellular level is negatively regulated by oncoprotein Mouse Double Minute 2 Homolog (MDM2). The transactivation domain (TAD) of p53 interacts with MDM2, which directs the p53 to proteasomal degradation. Mutagenesis studies showed that Phe19, Leu22, and Trp23 of TAD are the key residues involved in the p53-MDM2 interaction. In several studies, the alteration of these residues through mutations has been reported to dysregulate the transactivation function of p53. Since these residues are located near the Ser15 phosphorylation site of TAD, the effect of Phe19Ala and Leu22Gln-Trp23Ser mutations on the Ser15 phosphorylation by DNA-PK was investigated in this study. The Phe19Ala and Leu22Gln-Trp23Ser mutations were generated by site-directed mutagenesis, and Ser15 phosphorylation was monitored by western blot using phospho-p53 (Ser15) antibody. Our results showed that Ser15 phosphorylation was approximately 57% slower in the Phe19Ala and about 28% slower in the Leu22Gln-Trp23Ser mutant variant compared to the wild type. Obtained results indicate that Phe19, Leu22, and Trp23 play a crucial role in efficient Ser15 phosphorylation by DNA-PK, in addition to their role in MDM2 interaction.

Keywords: p53, DNA-PK, Site-Directed Mutagenesis, Ser Phosphorylation.

DRAFT

ORAL PRESENTATION

Balık yağından çoklu emülsiyon üretimi ve bazı özelliklerinin araştırılması

Gülistan Okutan[1*] (*ORCID: https://orcid.org/0000-0002-1936-7633*),
Gökhan Boran[2] (*ORCID: https://orcid.org/0000-0002-8871-8433*)

[*1]Siirt Üniversitesi, Teknik Bilimler Meslek Yüksekokulu, Gıda İşleme Bölümü, Siirt, Türkiye
[2]Van Yüzüncü Yıl Üniversitesi, Mühendislik Fakültesi, Gıda Mühendisliği Bölümü, Van, Türkiye

*Sorumlu yazar e-mail: gulistan.okutan@siirt.edu.tr

Özet

Çoklu emülsiyonun tekli emülsiyona göre bazı avantajları bulunmaktadır ve bu nedenle çoklu emülsiyon çalışmaları son zamanlarda popüler bir konu haline gelmiştir. Balık yağının içermiş olduğu çoklu doymamış yağ asitlerinin oksidasyona karşı hassas olması balık yağının çoklu emülsiyona işlenmesine yönelik çalışmaları teşvik etmektedir. Bu çalışmada, saf ringa balığı yağı kullanılarak S/Y/S çoklu emülsiyonu üretilmiş ve elde edilen emülsiyonun bazı özellikleri araştırılmıştır. Tek adımlı emülsiyon yöntemi kullanılarak üretilen çoklu emülsiyonun damlacık boyutu 135.7±2.6 nm ve zeta potansiyeli -48.56±1.06 mV olarak tespit edilmiştir. Balık yağı çoklu emülsiyonunun balık yağına göre daha yüksek iletkenlik değerine sahip olduğu, balık yağının iletkenlik değeri 0.33±0.02 µS/cm iken balık yağı çoklu emülsiyonunun iletkenlik değeri 9.82±0.11 mS/cm olarak bulunmuştur. Balık yağı emülsiyonunun asidik olduğu (pH=2.89±0.01), balık yağının asitlik değerinin 3.06±0.29, çoklu emülsiyonun asitlik değerinin ise 1.60±0.12 mg KOH/g yağ olduğu tespit edilmiştir. Emülsiyon stabilitesini belirlemek amacıyla kremalaşma indeksi analizi yapılmış ve kremalaşma indeksi %13.58±0.54 olarak bulunmuştur. Emülsiyonun renk değerleri söz konusu olduğunda; L* değeri 96.09±0.82, a* değeri -7.77±0.23, b* değeri 8.44±0.73 olarak bulunmuştur. Renk değerleri kullanılarak hesaplanan beyazlık indeksi çoklu emülsiyonda 87.87±0.87 iken balık yağında 46.19±0.18 olarak bulunmuştur. Elde edilen sonuçlar, balık yağının çoklu emülsiyona başarılı bir şekilde işlenebileceğini ancak elde edilen emülsiyonun balık yağından oldukça farklı özellikler gösterebileceğini ortaya koymuştur. Çalışmanın sonraki aşamalarında balık yağının çoklu emülsiyona işlenerek raf ömrü ve depolama stabilitesi üzerine çalışmaların yapılması planlanmaktadır.

Anahtar Kelimeler: Balık yağı, çoklu emülsiyon, damlacık boyutu, S/Y/S emülsiyonu.

DRAFT

ORAL PRESENTATION

Biogenic amines indexes used to determine the quality of seafood

İsmail Yüksel Genç[1*] (*ORCID: https://orcid.org/0000-0002-4816-806X*)

[*1]Isparta University of Applied Sciences, Eğirdir Fisheries Faculty, Fishing and Processing Technology Department, Isparta, Türkiye.

*Corresponding author e-mail: ismailgenc@isparta.edu.tr

Abstract

Biogenic amines in seafood are generally nitrogenous compounds formed by decarboxylation of amino acids as a result of microbial activity. Biogenic amines can be found in canned and marinated seafood as well as unprocessed, raw seafood. Studies show that the most abundant BAs in seafood are histamine, putricin, cadaverine, tryptamine, and tyramine. Furthermore, it has been reported that the microorganisms responsible for biogenic amine formation are mostly *Hafnia alvei*, *Morganella morganii*, and *Klebsiella pneumoniae*. The amount of biogenic amines in seafood may pose a risk to food safety. However, biogenic amine indices developed using the total amounts of BAs provide a perspective on the quality and safety of seafood products and also allow quantitative evaluation of the palatability of seafood products. In this study, studies on BAs in different seafood products and quality assessment of seafood products according to biogenic amine index were examined. Moreover, it was reached the conclusion that the determination of the presence and quantities of BAs in seafood is an important factor in terms of public health and that biogenic amine indices are also an important parameter about the quality and safety of seafood products.

Keywords: Biogenic amines, quality indexes, seafood, shelf life.

DRAFT

ORAL PRESENTATION

Synthesis of Ag(I)-DpNap and its antiproliferative effect on glioblastoma cells

Asuman Uçar[1*] *(ORCID: https://orcid.org/0000-0003-2674-3120)*,
Suray Pehlivanoglu[2] *(ORCID: https://orcid.org/0000-0001-7422-2974)*

[*1] Department of Science Education, Education Faculty, Agri Ibrahim Cecen University, Agri, Turkey.
[2] Department of Molecular Biology and Genetics, Faculty of Science, Necmettin Erbakan University, Konya, Turkey.

* Corresponding author e-mail: asucar340@gmail.com

Abstract

Silver complexes of Schiff bases are biologically active agents and have a wide range of biological applications [1]. Glioblastoma multiforme cells (GBM) is the most common and aggressive primary malignant tumor of the central nervous system. Currently, standard clinical treatment stategies such as surgical resection, radiotherapy and chemotherapy have mostly limited therapeutic efficacy in association with poor prognosis [2]. Herein, we aimed to evaluate the potential anticancer abilities of the newly synthesized Ag(I)- DpNap as a complex (Dp: 1,3-Diaminopropane; Nap: 2-Hydroxy-1-naphthaldehyde) and DpNap as a schiff base on GBM cells (U87-MG) *in vitro*. The cytotoxic effects of the compounds were evaluated by MTT assay for 72h. The compounds showed a significant dose-dependent cytotoxicity at increasing concentrations (0-100µg/mL) on the cells. Ag(I)- DpNap had higher cytotoxic properties than DpNap. In detail, Ag(I)- DpNap affects cells 2.5-fold more than DpNap at 100µg/mL concentration with approximately 81% cell death. According to the results, the silver complex and its ligand can be debate due to their anticancer potential against drug-resistant glioblastoma cells.

Keywords: Schiff base, Mtt assay, Glioblastoma multiforme.

[1].Khan S. Alhumaydhi FA. Ibrahim MM. Alqahtani A. Alshamrani M. Alruwaili AS. Hassanian AA. and Khan S. Recent Advances and Therapeutic Journey of Schiff Base Complexes with Selected Metals (Pt, Pd, Ag, Au) as Potent Anticancer Agents: A Review, Anti-*Cancer Agents in Medicinal Chemistry*, (2022) 22, 3086-96.
[2]. Davis ME. Epidemiology and Overview of Gliomas. *Semin Oncol Nurs* (2018) 34,420–9.

DRAFT

ORAL PRESENTATION

Determination of TGF-β and CXCL12 signaling pathway changes after mesenchymal stem cell-cancer cell co-culture

Şefika Özcan[1] (ORCID: *https://orcid.org/0009-0000-2542-4684*), Handan Sevim Akan[1] (ORCID: *https://orcid.org/0000-0002-8511-5258)*, Mehmet Ali Onur[1] (ORCID: *https://orcid.org/0000-0002-3630-7982)*

[*1]Hacettepe University, Faculty of Science, Department of Biology, Ankara, Turkey

*Corresponding author e-mail: sefikaozzcan@gmail.com

Abstract

The tumor microenvironment and signaling pathways play an important role in carcinogenesis. Our study aims to examine the changes in transforming growth factor (TGF-β) and chemokine-dependent ligand 12 (CXCL12) signaling pathways and cell viability after co-culture of cancer cells with MSCs, which is one of the important tumor microenvironment elements. The human bone marrow-derived MSCs-cancer cells (T98G; Glioblastoma and MDA-MB-231; Breast cancer) were co-cultured using transwell systems. Cell viability was assessed with the MTT method. Changes in the TGF-β (pTGFβRI and pSMAD3) and CXCL12 (pCXCR4, pERK1/2) pathways were measured using ELISA, immunohistochemistry, and immunofluorescence techniques. Results showed that MSCs co-cultured with cancer cells suppressed the viability of cancer cells for both cancer types. Cell viability was also decreased in MSCs after co-culture. TGF-β pathway was examined, and expression of pTGFβRI in cancer cells and MSCs showed similarity with control groups. The pSMAD3 molecule was decreased in cancer cells and MSCs after co-culture compared to control groups. CXCL12 pathway, pCXCR4 expression was mild to moderate increased in both groups in cancer cells-MSCs co-culture. While the increase in pCXCR4 in cancer cells increased the activity of pERK1/2, the increase in MSCs did not affect the expression of pERK1/2. In the TGF-β signaling pathway, the pTGFβRI receptor activity increases, while the decrease in pSMAD3 indicates that MSCs suppress the signal. Increased CXCL12 signaling pathway activity in MSCs and cancer cells after co-culture does not prevent cancer cell viability from being suppressed by MSCs. On the contrary, it is thought that CXCL signaling pathway activity contributes to the retention of MSCs in the cancerous region and suppression of cancer cell viability through bioactive molecules secreted from MSCs.

Keywords: Mesenchymal stem cell, cancer cells, tumor microenvironment, TGF–β, CXCL12.

DRAFT

ORAL PRESENTATION

Evaluation of *in vitro* Genotoxicity of Midazolam

Bahar Köklü[1*] (*ORCID: https://orcid.org/0009-0007-0316-6769*),
Berrin Ayaz Tüylü[2] (*ORCID: https://orcid.org/0000-0002-3156-2853*)

[*1]Eskisehir Technical University, Institute of Graduate Programs, Programme in Molecular Biology, Eskisehir, Turkey.
[2]Eskisehir Technical University, Faculty of Science, Department of Biology, Eskisehir, Turkey.

*baharkoklu@ogr.eskisehir.edu.tr

Abstract

One of the most widely used options for sedation in patients is midazolam, as it presents a short acting benzodiazepine that can be administrated by different routes. The aim of this study was to reveal the previously unknown genotoxic effects of midazolam on healthy human lymphocytes. To explore this, the researchers conducted a comet assay, a straightforward, rapid, and sensitive method, for genotoxic damage arising from single-strand breaks in DNA. The investigation revealed that midazolam, when administered at concentrations ranging from 0.1 to 0.4 mg/mL, triggers harmful effects in healthy human lymphocytes. Notably, the genotoxic impact of midazolam demonstrated a correlation with the dosage, with the 0.4 mg/mL dose exhibiting genotoxicity closely resembling that of the positive control group.
Furthermore, the study found that midazolam, across concentration levels of 0.1, 0.2, 0.3, and 0.4 mg/mL, led to DNA damage characterized by strand breaks. This was evident through substantial increases in the comet evaluation parameters; DNA tail length and DNA tail intensity when compared to negative and positive control values, indicating a significant impact.
In summary, the research suggests that midazolam could exert cytotoxic, cytostatic, and genotoxic effects on healthy human lymphocytes *in vitro*, with the intensity of these effects contingent on the concentration of exposure. As the investigation progresses, it is expected that these results will receive reinforcement from subsequent studies.

Keywords: Midazolam, DNA damage, Comet assay, Genotoxicity, Human lymphocytes.

DRAFT

ORAL PRESENTATION

Artemisinin and Cancer: A Molecular Docking Analysis Perspective

Özlem Kaplan[1*] (*ORCID: https://orcid.org/*0000-0002-3052-4556)

[*1]Alanya Alaaddin Keykubat University, Rafet Kayış Faculty of Engineering, Department of Genetics and Bioengineering, Antalya, Turkey.

*Corresponding author e-mail: ozlem.kaplan@alanya.edu.tr

Abstract

Artemisinin is a natural compound traditionally used to treat malaria and has been the focus of attention in recent years due to its potential effects on cancer treatment. However, the anti-tumor mechanism of artemisinin is very complex; a review of the action mechanism is still in its infancy, and more detailed and in-depth research is needed. In this research, molecular docking studies were performed to reveal the possible effects of artemisinin in cancer treatment. ER-α, HSP90, HSP70, HSP27, Bcl-2, mTOR, PR, EGFR, and HIF1-α protein targets known to be involved in cancer, and artemisinin were visualized using Chimera 1.16. Preparation for docking was performed using the Chimera 1.16 DocPrep tool. Active sites in target proteins were determined by CASTp analysis. The docking procedure was performed using AutoDock Vina. Then, Biovia Discovery Studio was used to visualize the complex interaction between the ligand and residues located in the active site of the targeted protein. The docking results demonstrated the significant binding affinities of artemisinin revealed interactions, especially with mTOR (-9.2 kcal/mol), HSP27 (-8.8 kcal/mol), and ER-α (-7.7 kcal/mol). The extensive information from this study provides a basis for a better understanding of artemisinin's potential in cancer therapy and the development of innovative, targeted therapeutic approaches.

Keywords: Artemisinin, molecular docking, cancer therapy, mTOR, HSP27, ER-α

DRAFT

ORAL PRESENTATION

Investigating the potential of baicalein: an *in silico* assessment for breast cancer

Nazan Gökşen Tosun[1*] (*ORCID: https://orcid.org/0000-0001-5269-1067*)

[*1]Tokat Gaziosmanpaşa University, Tokat Vocational School of Health Services, Department of Medical Services and Techniques, Tokat, Turkey.

*Corresponding author e-mail: nazan.goksen@gop.edu.tr

Abstract

Breast cancer continues to pose a significant health problem worldwide. As conventional treatments show limitations, interest in new potential therapeutic agents has increased. Baicalein, a flavonoid compound found in various plant sources, is noted for its natural anticancer properties. In this in silico study, it was aimed to investigate the potential therapeutic mechanisms of baicalein as a promising agent in the treatment of breast cancer. The binding affinities of baicalein with crucial proteins such as ER-α, HSP90, HSP70, HSP27, Bcl-2, mTOR, PR, EGFR, and HIF1-α, which play an essential role in breast cancer, were evaluated. Both proteins and baicalein were visualized using Chimera 1.16 and prepared for docking via the Chimera 1.16 DocPrep tool. Active sites of target proteins were determined using CASTp analysis. AutoDock Vina was used to identify potential interaction sites between ligand and target proteins. Biovia Discovery Studio was used to visualize the interactions between baicalein and target proteins. Baicalein exhibited a significant affinity for multiple target proteins, particularly with strong binding with HSP70 (-9.7 kcal/mol), HSP27 (-9.4 kcal/mol), and mTOR (-8.4 kcal/mol). The results demonstrated the potential of baicalein to interact favorably with proteins involved in critical pathways within breast cancer cells. This research provides a strong rationale for future laboratory-based and clinical trials and will potentially enable the development of new targeted therapies.

Keywords: Breast cancer, Baicalein, Molecular docking, Targeted therapies

DRAFT

ORAL PRESENTATION

Synthesis and Characterisation of Egg white/Polyacrylamide Hydrogels via UV Curing

Fatma Nur PARIN* (*ORCID: https://orcid.org/0000-0003-2048-2951)*

*1 Bursa Technical University, Faculty of Engineering and Naturel Sciences, Polymer Materials Engineering Department, Bursa, Turkey.

*Corresponding author e-mail: nur.parin@btu.edu.tr

Abstract

Essential oils are volatile components show bioactivity and produced from aromatic plants [1]. They are widely used in agriculture, cosmetics, pharmacology, and biomedical industry. Poor solubility, volatility, and sensitivity to environmental conditions of the essential oil limited to usage as wound healing material [2]. In this study, lavandula essential oil-loaded emulsion hydrogels are rapidly and effectively produced by free radical photopolymerization of egg white/acrylamide in the continuous phase of oil-in-water Pickering emulsion templates. Lavandula essential oil / β-cyclodextrin (LAV/β-CD) complexes were prepared and added into egg white/acrylamide blends. Egg white/acrylamide blends were used as oil phase whereas LAV/β-CD complexes were used as water phase. β-CD, a type of carbonhydrate, acts as an efficient emulsifier in the preparation of LAV-in-water Pickering emulsions. The results show that the LAV can be efficiently the loaded polymer matrix. The chemical groups in the hydrogels were verified using Attenuated Total Reflectance-Fourier Transform Infrared Spectroscopy (ATR-FT-IR), and cross-sectional images of hydrogels and emulsions were examined using optical microscopy. Moreover, the swelling capacity of hydrogels was measured. Antibacterial activity of them against Gram (-) *(E.coli)* and Gram (+) *(S. aureus)*. According to FT-IR analysis, LAV was successfully loaded in hydrogels. As the amount of essential oil increases, the antibacterial activity increases against *Escherichia coli, Staphylococcus aureus* bacteria, while the swelling capacity of hydrogels decreases. In line with the findings, the obtained hydrogels have the potential to be used as wound dressings.

Keywords: Egg white, hydrogel synthesis, lavandula oil, polyacrylamide, UV curing.

[1] Fasihi, H., Noshirvani, N., Hashemi, M., Fazilati, M., Salavati, H., & Coma, V. (2019). Antioxidant and antimicrobial properties of carbohydrate-based films enriched with cinnamon essential oil by Pickering emulsion method. *Food Packaging and Shelf Life*, *19*, 147-154.

[2] Cimino, C., Maurel, O. M., Musumeci, T., Bonaccorso, A., Drago, F., Souto, E. M. B., ... & Carbone, C. (2021). Essential oils: Pharmaceutical applications and encapsulation strategies into lipid-based delivery systems. *Pharmaceutics*, *13*(3), 327.

DRAFT

ORAL PRESENTATION

Arka Çapraz Bağ Kesen Total Diz Artroplastisinde Orta Dönem Sonuçlarımız

Beytullah Unat[1*] (*ORCID: https://orcid.org/0009-0006-1036-6710),* Günhan Karakurum[1] *(0000-0003-3004-9326)*

[*1]Gaziantep Üniversitesi, Tıp Fakültesi, Ortopedi ve Tramvatoloji Bölümü, Gaziantep, Türkiye

*Sorumlu yazar e-mail: beytullahunat@hotmail.com

Özet

Bu çalışmada, arka çapraz bağ (AÇB) kesen (PCL- substituting) total diz artroplastisi uygulanan olgular retrospektif olarak incelenerek orta dönem sonuçlarının ortaya konulması amaçlanmıştır. Kliniğimizde Temmuz 2006 - Haziran 2012 tarihleri arasında AÇB kesen TDP tatbik edilen ve yeterli takibi olan 61 hastanın 80 dizi bu çalışmaya dahil edilmiştir. Hastalarımızın 52'si kadın, 9'u erkekti. Yaş ortalaması 65,4 (42-85), ortalama takip süresi 32,6 ay (12-72) olarak saptandı. Hastalar Amerikan Diz Cemiyeti kriterlerine göre; diz skoru ve diz fonksiyonel skoru bakımından değerlendirildi. Radyolojik değerlendirme ise, TDP Radyolojik Değerlendirme kriterlerine göre yapılmıştır. Diz skoru ameliyat öncesi 38-71 (ortalama 43,7) iken, ameliyat sonrası 78-100 (ortalama 95) olarak saptanmıştır. Diz skoru bakımından dizlerin 69'unda (%86,25) mükemmel, 11'inde (%13,75) iyi, Fonksiyonel skoru ameliyat öncesi 10-60 (ortalama 35,08) iken, ameliyat sonrası 60-100 (ortalama 82,2) olarak saptanmıştır. Diz fonksiyonel skoru baz alınarak yapılan değerlendirmede dizlerin 45'inde (%56,25) mükemmel, 29'unda (%36,25) iyi, 6'sında (%7,5) orta sonuç elde edilmiştir. Hastalarda ameliyat öncesi ortalama 9,7° varus (8°valgus-20°varus) tespit edilmiş olup, ameliyat sonrası ortalama 4,75° valgus (2°varus-10°valgus) dizilimi sağlanmıştır. 1 hastada geç derin enfeksiyon nedeniyle revizyon protezi tatbik edilmiştir. Bir hastada travma nedeniyle insert kırılması sonucu insert değişimi yapılmıştır. AÇB kesen total diz artroplasti uygulamalarımız sonucunda %86,25 mükemmel diz skoru, %56,25 mükemmel fonksiyonel skor elde edilmiştir. Bu değerler pek çok çalışmadaki sonuçlara yakın bulunmuştur. Uygun hasta seçimi, yeterli ameliyat öncesi hazırlık ve dikkatli cerrahi teknik kullanıldığında, AÇB kesen TDP yüz güldürücü bir ortopedik cerrahi girişimdir.

Anahtar kelimeler: AÇB, TDP, gonartroz, hareket açıklığı, fonksiyonel sonuçlar.

DRAFT

ORAL PRESENTATION

Chloroplast genome targeted phylogenetic analysis in *Cucurbitaceae* family

Mısranur Yıldırım[1] (0009-0004-1577-2027), Emre Yörük[1*] (0000-0003-2770-0157)

[1]Istanbul Yeni Yuzyil University, Faculty of Arts and Sciences, Department of Molecular Biology and Genetics, Istanbul, Turkey.

*Corresponding author e-mail: emreyoruk@outlook.com

Abstract

The distribution of *Cucurbitaceae* family members is known for tropical and warmer regions. Interspecific variation of the most common members is still represented by limited information. In this study, chloroplast (cp) genome based phylogenetic analysis were carried out in 10 predominating species (*Cucumis melo subsp. melo*, *Lagenaria siceraria*, *Luffa acutangula*, *Luffa aegyptiaca*, *Momordica charantia*, *Cucumis sativus*, *Cucurbita argyrosperma*, *Cucurbita moschata*, *Cucurbita pepo*, and *Cucurbita maxima*) of the family *Cucurbitaceae*. Nucleotide sequences and characteristic data of cp genomes were obtained from GenBank. Correlation matrix and principal component analysis (PCA) for numeric and characteristic data were obtained via GraphPad Prism software. Sequence similarity and UPGMA trees were constructed by using MAFFT software. Bayesian topology analysis were carried out using MrBayes software via TRUBA terminal. R Studio was used to obtain PCA graphics for cp genomes. Correlation matrix showed that there was positive correlation between mRNA number and GC% for each species. In PCA analysis, *C. melo* showed distinction from remaining species by geographic origin, soil characteristics, and the taste. UPGMA tree showed that intraspecific distribution was clear with 3 distinct sub-divisions. The minimum and maximum genetic variation values were recorded as 3.33% and 0.88%, respectively. Bayesian topology analysis and PCA analysis showed that at least three dimensions were present for cp genomes, and PC1 and PC2 were as 51.2% and 28.9%, respectively. In phylogenetic analyses, it was observed that cp genomes can strongly discriminate between genus as in common polymorphism studies; thus, marker genes can be identified in future studies for species distinction in the *Cucurbitaceae* family.

Keywords: cpDNA, *Cucurbitaceae*, PCA, sequence similarity

DRAFT

ORAL PRESENTATION

Mini-prep protocol for simplified RNase A extraction

Sergen Ünal[1] (0009-00058227-0649), Aleyna Eren Samur[1] (0009-0009-4920-9300), Ezgi Savaş[1] (0009-0006-3490-2602), Fatma Seda Çatuk[1] (0009-0009-5196-8752), Emre Yörük[1*] (0000-0003-2770-0157)

[1]Istanbul Yeni Yuzyil University, Faculty of Arts and Sciences, Department of Molecular Biology and Genetics, Istanbul, Turkey.

*Corresponding author e-mail: emreyoruk@outlook.com

Abstract

The enzymes produced by biotechnological procedures present variation in terms of the need for them in laboratory. Enzymes like DNA-directed DNA polymerase could be accepted as highly needed and frequently used enzyme in molecular biology laboratories. It is easy to find out DNA-directed DNA polymerase with the range of different cost and quality. However, there is limited options available for some other enzymes like RNase. In this study, it was aimed to extract RNase A enzyme by ethanol precipitation method by short time cost-effectively. First, expression cassette of the pOpenTaq plasmid was amplified by inverse PCR. Then, *RNase A* gene from pOpen-RNAseA was amplified by conventional PCR. Expression cassette and *RNase A* gene were ligated as a circulated vector, and then transformed into *Escherichia coli* JM107 strain. Confirmed colonies were used in RNase A extraction. Transformants grown in ampicillin (100µg/mL) added medium overnight, then they were induced with IPTG (1 mM) overnight. After extraction step, purification was carried out by 70% ethanol precipitation. In SDS-PAGE assays, 16 kDa protein was checked, and concentration was calculated by BCA protocol. The extracted RNase A (1:10, V:V) was used in SDS-based DNA extraction from 7-day-old mycelium culture of *Fusarium culmorum*. No RNA was detected in gDNA samples of RNase A-treated ones. The findings showed that the ethanol precipitation method could be easily used in any molecular biology laboratories involving common equipments to obtain RNase A enzyme within in short time.

Keywords: Enzyme extraction, molecular biology, PCR, RNase A

DRAFT

ORAL PRESENTATION

A Novel Composite Structure for Electrochemical Uric Acid Sensor with SWCNT and Pyrene-Substituted-Anthracene π-π Interaction

Vildan Şanko* (*ORCID: https://orcid.org/0000-0003-0331-5967*)

Gebze Technical University, Department of Chemistry, Gebze, Kocaeli 41400, Turkey.

*vildan_sanko@hotmail.com

Abstract

Uric acid, known as the end product of purine metabolism, is excreted from the body through the intestines and kidneys. The presence of more than normal levels in body fluids can be considered an indicator of kidney disease. It is also stated that changes in plasma levels may be an indicator of pneumonia and leukemia [1]. For this reason, the interest in sensitive and selective sensors that can detect uric acid continues. Carbon-based nanoparticles allow the design of sensitive sensors that can be used in many areas [2]. Carbon nanotubes, grouped as single (SWCNT) or multi-walled (MWCNT) according to the number of layers, have great advantages in the production of biosensors and chemosensors, thanks to their modifiable surfaces and very large surface areas, and extraordinary conductivity. In addition to these features, their chemical and mechanical resistance also brings this group to the forefront for many applications [3]. Although surface modification of carbon nanotubes is a very interesting subject, non-covalent interactions also provide useful results in functionalization. For such functionalizations, it is usually π-π interactions between the aromatic molecules and the nanotube surface. Some aromatic compounds such as anthracene, pyrene, ferrocene, and phthalocyanine derivatives can be used for this purpose and contribute to sensor performance [4]. In this study, the SWCNT structure modified with pyrene-substituted-anthracene was synthesized for the first time and used in the electrochemical uric acid biosensor platform. Cyclic voltammetry (CV) and differential pulse voltammetry (DPV) methods were used for electrochemical measurements and elucidation of the redox mechanism. With the inclusion of a pyrene-substituted-anthracene structure in the system, a sharper peak of uric acid and a higher peak current value were determined.

Keywords: Uric acid detection, Electrochemical sensor, Carbon nanotube, Non-covalent functionalization.

References

[1] Aafria, S., et al., *Electrochemical biosensing of uric acid: A review.* Microchemical Journal, 2022: p. 107945.
[2] Xie, F., et al., *Carbon-based nanomaterials–a promising electrochemical sensor toward persistent toxic substance.* TrAC Trends in Analytical Chemistry, 2019. **119**: p. 115624.
[3] Karimi-Maleh, H., et al., *Recent advances in carbon nanomaterials-based electrochemical sensors for food azo dyes detection.* Food and Chemical Toxicology, 2022: p. 112961.
[4] Zhou, Y., Y. Fang, and R.P. Ramasamy, *Non-covalent functionalization of carbon nanotubes for electrochemical biosensor development.* Sensors, 2019. **19**(2): p. 392.

DRAFT

ORAL PRESENTATION

Karyology of monkey goby

Sevgi Unal-Karakus[1*] (*ORCID: https://orcid.org/0000-0002-6409-7783*), Muradiye Karasu-Ayata[2] (*ORCID: https://orcid.org/0000-0001-8890-8547*), Muhammet Gaffaroğlu[3] (*ORCID: https://orcid.org/0000-0001-7436-5828*)

[*1]Bartın University, Faculty of Science, Department of Molecular Biology and Genetics, 74100, Bartın, Türkiye.
[2]Kırsehir Ahi Evran University, Faculty of Health, Department of Nutrition and Dietetics, 40100, Kırsehir, Türkiye.
[3]Kırsehir Ahi Evran University, Faculty of Science and Arts, Department of Molecular Biology and Genetics, 40100, Kırsehir, Türkiye.

*Corresponding author e-mail: sunal@bartin.edu.tr

Abstract

Karyological properties of *Neogobius fluviatilis* (Pallas, 1814) from the population inhabiting Sakarya Creek, Bilecik, Türkiye were studied by examining conventional metaphase chromosome spreads obtained from the kidney, spleen, and gill tissues. Specimens were captured by electrofishing method and carried as alive to genetic laboratory in well-aerated carrying aquariums. Cell suspansion was obtained hypotonisation, fixation and staining steps and chromosomes were harvested using air-drying technique. Observing and analysing of obtained chromosomes were carried out via Leica DM 3000 research microscope. The investigation of 40 metaphase spread from four (three females and one male) individuals evidenced that the chromosome numbers of this species was determined 2n = 46 and the arm number was found as NF = 46. The prepared karyotype of the invasive monkey goby was consisted of 23 pairs subtelo-acrocentric (st-a) chromosomes that gradually decreased in size, and they can be assessed as uni-armed chromosomes. The chromosome morphology can be noticed as 2n = 46 (st-a). No morphologically differentiated sex chromosomes were found in the monkey gobies studied. This study was revealed chromosome number and karyotype of *N. fluviatilis* for the first time from Turkey. The process was approved by the Local Animal Ethics Committee of Türkiye (Protocol Number: 68429034/09).

Keywords: Chromosomes, karyology, karyotype, *Neogobius fluviatilis*.

DRAFT

ORAL PRESENTATION

Biosynthesis of high value-added carotenoids by engineered microorganisms

Ozkan Fidan[1*] (*ORCID:* https://orcid.org/0000-0001-5312-4742),
Nuriye Arslansoy[1] (*ORCID:https://orcid.org/0009-0008-5514-8711*)

[*1]Abdullah Gül University, Faculty of Natural and Life Sciences, Department of Bioengineering, Kayseri, Türkiye.

*Corresponding author e-mail: ozkan.fidan@agu.edu.tr

Abstract

Carotenoids are pigment molecules that play an important role in coloring plants, algae, and other organisms. These molecules exhibit various biological activities such as anticancer, antiviral and antioxidant activities. They have a huge market size and are mainly used in the food, feed, and cosmetic industries. The current supply chain for carotenoids is mostly relied on the extraction from plants and/or chemical synthesis for certain carotenoids. However, these strategies have various bottlenecks and disadvantages such as being affected by climate change, more difficult and costly extraction processes, and environmental issues. These can be overcome with microbial biosynthesis, which not only addresses the previous problems but also provides advantages of producing in a short time and scale-up for industrial production. In this research, we aimed to biosynthesize the high value-added carotenoids by engineered microorganisms. The genome of a native producer of zeaxanthin diglucoside, identified as endophytic *Pseudomonas* sp. 102515, was edited by CRISPR-Cas9 to knock out zeaxanthin glucosyltransferase (*CrtX*), lycopene β-cyclase (*CrtY*) and beta-carotene hydroxylase (*CrtZ*). This led to Δ*CrtX*, Δ*CrtY* and Δ*CrtZ* mutant strains of *Pseudomonas* sp. 102515. Gene knock-outs were confirmed by colony PCR. HPLC analysis of extracts from mutant strains revealed that all mutants produced the corresponding carotenoids such as zeaxanthin, β-carotene, and lycopene. Thus, this study paved the way for the biosynthesis of valuable carotenoids in the engineered endophytic bacteria.

Keywords: Carotenoids, CRISPR-Cas9, Biosynthesis, *Pseudomonas*

DRAFT

ORAL PRESENTATION

Investigation of Wastewater Remediation Performance of Cost Effective Graphene Supported Photo Catalyst

Hüseyin GUMUS (https://orcid.org/0000-0002-2029-7978)

Bilecik Şeyh Edebali University, Osmaneli Vocational School, Bilecik, TURKEY

Cossepongind author e-mail: huseyin.gumus@bilecik.edu.tr

Abstract

Photocatalysts are effective structures that convert water polutants to unharmful components. In this study, the MO removal efficiency of graphene supported photocatalyst in the presence of ultraviolet light was examined. 5 grams of graphite powder (Gr) was soaked in 0.1 mol/L iron (III) chloride ($FeCl_3$). The mixture was heated at 250 ºC for 1 hour with nitrogen flow (3 ml/min). The precipitate was cooled, washed and dried at 50 °C for 24 h and then grinded to 150 µm, Gr-Fe. 1 gram of Gr-Fe was mixed with 10 mL of 75 ppm MO until adsorption equilibrium. Samples was analyzed by UV-Vis spectrometry at 466 nm. MO adsorption of Gr and Gr-Fe were calculated as 13% and 39%, respectively. Solution was placed horizontally in front of a 30W ultraviolet light source. 1 ml of 0.01 mol/L H_2O_2 was added to the solution and UV radiation was applied for 20 min. The same experiments were repeated in the presence of 1 g Gr. The characteristic XRD graphite peak was obtained at 27º and it shifted to 25.5º. Wide 10º peak belonging to the GO peak was observed in Gr-Fe. Gr-Fe structure is partially delaminated with amorphous structure. The interlayer spaces of the graphite are significantly opened and Fe particles are placed. The surface area of Gr was 5.6 m^2/g, while it was only 6.4 m^2/g. Fe penetrated into the pores, causing the surface shrink. The MO removal rates of Gr and Gr-Fe after 20 min UV irradiation in the presence of peroxide were 24% and 91%, respectively due to HO• radicals are formed. These radicals interacted with MO and breakdown of the dye according to the fenton process.

Keywords: Graphite, Photocatalysis, Impurities Removal, Methyl Oranj.

DRAFT

ORAL PRESENTATION

Investigation of Photocatalytic Dye Removal Performance of Activated Carbon-Polymeric Composites Modified by Phosphoric acid[1]

Bülent Büyükkıdan (https://orcid.org/0000-0001-9619-3246)

Kütahya Dumlupınar University, Faculty of Arts and Sciences, Department of Chemistry, Kütahya, TURKEY

* Corresponding author e-mail: bulent.buyukkidan@dpu.edu.tr

Abstract

Activated carbon that is formed by burning plants, trees, and synthetic materials is an effective adsorbent. In this study the dye removal performance of polymeric composites (H3-CTW-P), which are prepared according to previous publications, was studied under 50W UV radiation[1]. Textile wastes pretreated with H_3PO_4 were pyrolyzed at 350˚ and grounded (H3-CTW). 10%wt H3-CTW was added to a 14%wt polyvinyl fluoride-dimethylformamide (PVDF-DMF) solution and the mixture was phase separated. According to SEM and BET analysis results, the surface area of the %10-H3-CTW-P increased by 182%. The dye removal tests were conducted in a batch system at 298 K. 100 mL of 75 ppm MO aqueous solution and 0.5 g of composite adsorbent (1x1 cm in size) were placed 10 cm away from a 50W ultraviolet lamp. The solution was stirred at 150 rpm for 90 minutes until adsorption equilibrium. Then ultraviolet radiation was applied for 20 minutes. The MO was analyzed by UV-Vis spectrometry at 466 nm. The removal for H3-CTW-P was 36% at 90 min. and this value was calculated as 8.9% for raw PVDF. After 20 minutes of UV interaction it was recorded as 77% while it was approximately 90% for powder H3-CTW. The IR bands at 998 and 1700 cm^{-1} of H3-CTW showed that functional groups on the activated carbon was formed. Although the photocatalytic efficiency of H3-CTW powder was higher, it provided ease of use to the composite activated carbon. H3-CTW-P composites can be used in filtration systems.

Keywords: Adsorption, Active carbon, Polymeric composite, PVDF, Recycling waste.

[1] This study was carried out with the financial support of Kütahya DPÜ BAP unit (Project No: 2020-08)"."Bu çalışma Kütahya DPÜ BAP biriminin finansal desteği ile yürütülmüştür (Proje No:2020-08)"

[1] A Simple and Green Preparation Route of Waste Textile Based Photocatalytic Biochars for Pollution Removal, Chemistry Africa https://doi.org/10.1007/s42250-023-00625-3

DRAFT

ORAL PRESENTATION

Controlled electrode potential as a way towards more rapid and homogeneous deposition of self-assembled monolayers

Ebru Ercan ER, Ahmet UÇAR* (*ORCID: https://orcid.org/0000-0001-7696-7726*)

Ankara Yıldırım Beyazıt University, Faculty of Engineering and Natural Sciences,
Department of Energy Systems Engineering, 06010, Ankara, Turkey

*Corresponding author e-mail: ahmet.ucar@aybu.edu.tr

Abstract

Self-assembled monolayer (SAM) coatings are formed by the self-assembly of molecules as a single layer by adsorption on a substrate. Following the surface attachment as the initial step, the molecules often reorganize to form an ordered and uniform monolayer structure. SAM-based technologies have been used in many different areas from biosensing to smart and functional surfaces and energy storage, due to being a cheap and easy-to-apply surface modification technique. SAM formation mechanism and kinetics have always been an interesting research area. In addition to controlling the SAM adsorption kinetics and mechanism, reducing the long deposition times is a significant step to increase their applicability. Metallic surfaces such as gold, silver and platinum, where SAM-based molecules are deposited, typically have uncontrolled potential and thus variable surface charges and energies. Deposition solutions of SAM molecules might also have varying redox compositions and potentials. Therefore, the use of an active, potentiostatic (potential-controlled) method for selective SAM adsorption may offer many advantages. This study presents the feasibility of an active, potential-controlled technique in producing faster and more uniform coatings on gold screen-printed electrode surfaces. To achieve this, thiol-containing molecules with different functional groups and chain lengths have been used and results have been discussed in comparison to coatings prepared using passive (potential-uncontrolled) 24-hour incubation. The optimal potential range and solution concentration for the deposition has also been studied. Complementary characterization techniques have been performed for the detailed analysis; (i) differences in surface electroactivity before and after deposition have been examined by electrochemical techniques (CV, SWV, EIS), (ii) chemical changes for the deposited surfaces have been assessed by X-ray Photoelectron Spectroscopy (XPS) and (iii) the effects of SAM molecules, selected to have different functional groups, on the surface energy and thus on the surface wettability have been checked by water contact angle measurements.

Keywords: self-assembled monolayers, electrochemical deposition, electrode modification, screen-printed electrodes, surface characterization

DRAFT

ORAL PRESENTATION

Steroidal Konjugatların Biyolojik Aktiviteleri: Antimikrobiyal ve Antiproliferatif Özelliklerin İncelenmesi ve Moleküler Bağlanma Çalışmaları

Sevinç İlkar Erdağı[1*] (ORCID: https://orcid.org/0000-0001-5811-2302)

[*1]Kocaeli Üniversitesi, Fen Edebiyat Fakültesi, Kimya Bölümü, Kocaeli, Türkiye

* sevinc.ilkar@kocaeli.edu.tr

Özet

Steroidler, doğal bileşikler arasında önemli bir gruptur ve farklı organizmalarda önemli roller oynarlar. Önceki çalışmalar, steroid konjugatlarının kanser hücrelerini hedefleme ve kanser tedavisinde kullanma potansiyelini göstermektedir. Bu çalışmada, farklı steroidler, biyolojik olarak aktif bileşik ile konjuge edilerek yeni konjugatlar geliştirildi ve bu konjugatların seçici farmakolojik özellikleri incelendi. Antikanser aktiviteleri çeşitli kanser hücre hatları üzerinde *in vitro* olarak değerlendirildi. Seçicilik indeksinin belirlenmesi amacıyla sağlıklı hücre hattına karşı sitotoksiteleri *in vitro* olarak test edildi. Konjugatların Gram-pozitif ve Gram-negatif bakteriler ile mantarlar üzerindeki antimikrobiyal aktiviteleri de araştırıldı. Steroidal konjugatların farmakokinetik özellikleri, *in silico* ADME çalışmaları ile açıklandı ve değerlendirildi. Ayrıca, bu çalışmada ilk olarak, etkin konjugatlar üzerinde protein-ligand etkileşimleri, bağlanma ilgilerini belirlemek amacıyla *in siliko* moleküler bağlanma çalışmaları yapıldı. Bu çalışmanın sonuçları, bu konjugatların çeşitli hastalıkların tedavisine çok yönlü bir yaklaşım sunma potansiyeline sahip olduğunu göstermektedir.

Anahtar Kelimeler: Antimikrobiyal aktivite, Antiproliferatif aktivite, Biyokonjugat, Moleküler bağlanma, Hidroksi kumarin, Steroid.

DRAFT

ORAL PRESENTATION

Kemik doku mühendisliği uygulamaları için kalsiyum fosfat esaslı biyoseramiklerden doku iskelesi üretimi ve ürün performanslarının karşılaştırılması

Ayşe Seda ERGÜN[1] (*https://orcid.org/0009-0002-5144-3779*), Abdullah TORAMAN[1](*https://orcid.org/0000-0002-7315-1235*), Afife Binnaz HAZAR YORUÇ[1](*https://orcid.org/0000-0001-7281-2305*)

[**1]Yıldız Teknik Üniversitesi, Kimya-Metalurji Fakültesi, Metalurji ve Malzeme Mühendisliği Bölümü, İstanbul/ Türkiye

ergunseda0@gmail.com

Özet

Kemik doku mühendisliği uygulamaları; kemik defektleri, doku kayıpları ve kırıkların tedavisinde büyük rol oynamaktadır. Günümüzde, çeşitli nedenlerle ortaya çıkan kemik hasarlarının tedavi yöntemleri konusundaki araştırmaların sayısı hızla artmaktadır. Her yıl yaklaşık 2 milyon adet kemik grefti kullanılmaktadır. Bazı durumlarda kemikler belirli bir boyuta kadar kendiliğinden iyileşebilse de, hastaların yaklaşık %10'u, dokuda oluşan tam kaynamama, yavaş iyileşme ve yüksek tedavi maliyetleri gibi ciddi kemik hasarı sorunlarıyla karşılaşmaktadır. Özellikle kemik defekti ve kırıklarında kullanılan bir tedavi olarak kemik greftleri için olası sorunların hepsine yanıt verebilecek tedavi yöntemi mevcut olmadığından, altın standart olarak otogreftler son yıllarda sıklıkla kullanılmaktadır. Otogreftler, immünolojik açıdan allogreftlere göre daha başarılı olmasına rağmen yüksek morbidite, limitli donör sahası gibi sebeplerden dolayı dezavantajları bulunmaktadır. Bu tez çalışması kapsamında, kemik doku iskeleleri geleneksel (sponge) ve modern (3D baskı) yöntemler kullanılarak üretilmiş ve sonuçlar karşılaştırılmıştır. Sponge yöntemi; uygulama kolaylığı, düşük maliyetli üretim gibi avantajlar sunmaktadır. Bu teknik temel olarak; seramik çamurunun hazırlanması, süngerlere emdirilmesi ve kurutma işlemlerini içermektedir. Üç boyutlu baskı yöntemi ise sınırlı donör alanı ve morbidite gibi sorunları aşarak daha hızlı ve daha doğal bir yapı elde etmeye olanak tanımıştır. Ayrıca, üç boyutlu baskı teknolojisi; istenilen gözenek deseni, katman kalınlığı ve kürleme süresine göre ayarlanabilen kontrol edilebilir ürünlerin eldesi açısından geleneksel metoda göre daha avantajlıdır. Doku iskeleleri, reçine ve biyoseramik tozlarının birleştirilip üç boyutlu basım yöntemi ile oluşturulmuştur. Son aşamada ise üretilen doku iskeletlerinin dokularla arasındaki uyumunun arttırılmasını sağlamak ve implantasyon sonrasında vücuttaki olası enfeksiyonları antibakteriyel özelliği ile engellemek için kemik doku iskeletleri biyomalzeme ile kaplanmıştır. Doku iskeletlerinin kimyasal (FTIR, XRD), morfolojik (SEM, porozite) ve mekanik (basma testi) özellikleri analiz edilmiştir. Yapılan test sonuçlarına göre mekanik özellikleri sponge yöntemiyle yapılan greftlerde 2-3 MPa, 3D ile üretilen greftlerde 3-5 MPa olarak belirlenmiştir. Uygulama kolaylığı ve kontrol edilebilir yapısı nedeniyle 3D basılmış kemik doku iskelelerinin, kemik defektlerinde kullanımında umut verici olduğu öngörülmektedir.

Anahtar Kelimeler: Kemik doku mühendisliği, doku iskelesi, kemik defekti, sponge yöntemi, 3B baskı

DRAFT

ORAL PRESENTATION

Turbidity Removal by Adsorption of Coal-Based Powder Activated Carbon (CBPAC) from Regular Landfill Leachate and Evaluation of Experimental Parameters

Aysenur OGEDEY[1,*](ORCID-ID: 0000-0003-2662-106X), Ensar OGUZ[2] (ORCID-ID: 0000-0001-7720-4739)

[1] Munzur University, Faculty of Engineering, Department of Civil Engineering, Tunceli, Turkey,
[2] Atatürk University, Faculty of Engineering, Department of Environmental Engineering, Erzurum, Turkey,

aysenurcumurcu@munzur.edu.tr

Abstract

Solid waste problem constitutes an important part of environmental pollution in our country. Various factors such as various physical, chemical, biochemical interactions, high moisture content in the waste, rainwater infiltration and contact of water with solid wastes are effective in the formation of leachate. Physico-chemical methods are frequently preferred methods for the practical removal of contaminants from leachate. Among these methods, the adsorption process is an economical and convenient method in terms of applicability in removing pollutants from leachate. In this study, the removal of COD from the leachate formed in the sanitary landfill by the adsorption process was investigated. The effects of adsorbent dose, pH, temperature and mixing speed on the operating conditions were investigated. In addition, suitable operating conditions were determined with the data obtained by using adsorption kinetics and isotherms. According to the results obtained, the alternativeness of the adsorption process as a purification method was evaluated. The highest turbidity removal efficiency obtained with the CBPAC adsorption process was 51.8% at pH 5, 5 g/L adsorbent dose, 20°C temperature and 300 rpm stirring speed. It was determined that CBPAC adsorption from leachate and turbidity removal fit the pseudo-first-order model due to the high correlation coefficient (R^2 0.995). Langmuir isotherm (R^2 0.990) was determined as the most suitable isotherm for turbidity removal from leachate with CBPAC. The ΔH° (11,319 kj/mol) value of the thermodynamic parameters of turbidity removal from leachate with CBPAC was positive, indicating that the adsorption process was endothermic. The positive ΔS° (0.0370 kj/mol.K) value showed that the irregularity of the particles increased after the adhesion on the surface of the CBPAC particles. The positive ΔG° (0.478 kj/mol) value indicated that CBPAC adsorption could not occur spontaneously. In the light of the data obtained, it was determined that the CBPAC adsorption process is a suitable process for turbidity removal from leachate.

Keywords: Leachate, adsorption, turbidity removal.

NOT: Bu çalışmadaki veriler, “FARKLI ARITIM YÖNTEMLERİ KULLANARAK BİNGÖL KATI ATIK DEPOLAMA SAHASI SIZINTI SUYUNUN ARITILABİLİRLİĞİNİN ARAŞTIRILMASI VE KİNETIK ÇALIŞMALAR” adlı doktora tezinden alınmıştır.

DRAFT

ORAL PRESENTATION

Potential AHAS enzyme inhibitor containing sulfone and tetrazole groups: Synthesis, Characterization, Molecular Docking and ADME Investigations

Nüveyre Canbolat[1*] (*ORCID: https://orcid.org/0000-0003-4466-4602*), Ümmühan Özdemir Özmen[1] (*ORCID: https://orcid.org/0000-0001-9161-9367*), Doğukan Doyduk[1](*ORCID: https://orcid.org/0000-0002-1260-8879*), Yılmaz Yıldırır[1] (*ORCID: https://orcid.org/0000-0001-9091-5324)*

[*1]Gazi University, Science Faculty, Chemistry Department, Ankara, Turkey.

*Corresponding author e-mail: nuveyrecanbolat@gazi.edu.tr

Abstract

The research on the synthesis, characterization, experimental, and in silico analyses of ligands containing sulfon groups and their transition metal complexes is an important subject in fields such as drug discovery and development, biochemistry, and bioinorganic chemistry. The presence of pyrimidine, pyrrolidino, pyridyl, and tetrazole groups in conjunction with the sulfone moiety is known to enhance activities like herbicidal and antifungal effects. The tetrazole ring is known to exhibit favourable interactions with biological targets, making it a valuable scaffold for creating compounds with enhanced binding affinity and bioactivity. Herbicides, used to control unwanted plants, face the challenge of resistance, with acetohydroxyacid synthase (AHAS) enzyme inhibitors being a prominent concern due to their widespread use and importance in various organisms, i.e., AHAS, a critical enzyme in amino acid biosynthesis, serves as a target for herbicides through oxidative inactivation, making it a focal point for addressing resistance. One-pot reactions have become widely used in chemical synthesis due to their advantages including simplified procedures, efficient resource utilization, and versatility. These reactions find applications in drug design, combinatorial chemistry, and natural product synthesis, providing environmentally friendly and streamlined approaches to compound synthesis. In these studies, the synthesis of new ligands containing sulfon groups, their characterization, and the determination of their effects on enzyme inhibition are performed with experimental results through in silico analyses and molecular docking calculations. In this study, we have synthesized new sulfonyl hydrazone containing tetrazole group (4-ethyl-tetrazole-ethane sulfonyl hydrazone, 4etzesh) by one-pot reaction process. The structure of the compound was characterized using spectroscopic methods (FT-IR, NMR). In addition, enzyme inhibition studies were examined through molecular docking calculations (Schrödinger Suite 2020-3) with Glide XP OPLS2005 force field docking to elucidate non-covalent interactions. ADME (absorption, distribution, metabolism, and excretion) calculation also performed QikProp in Schrödinger Suite.

Keywords: Herbicidal resistance, AHAS enzyme, Sulfon group, One-Pot reaction, Molecular docking, ADME.

DRAFT

ORAL PRESENTATION

Immunofluorescence investigation on follicular dendritic cells resident in the pyloric tonsil of Turkey (*Meleagris gallopavo*)

Nuh YILDIRIM[1*] (*ORCID:https://orcid.org/0000-0002-5315-923X*),
Hikmet ALTUNAY[1] *(ORCID:https://orcid.org/0000-0002-2983-8582*)

*[1]Ankara University, Veterinary Medicine, Histology-Embryology, Ankara, Türkiye.

*Corresponding author e-mail: nuhyldrm1988@gmail.com

Abstract

The immune system, which is of mesenchymal origin, is not in direct contact with the antigens in the body. However, microorganisms can invade organs developed from the ectoderm or endoderm layer embryologically. Immunological cells indirectly encountering intruders are primary cells responsible for the extermination of invaders. In humans and animals, the mucosal layer of the digestive system exposed to antigenic stimulation has a vastitude surface area. The gut-associated lymphoid tissue (GALT), responsible for the extermination of food-borne pathogens, comprises the tonsils, Peyer's patches, appendix, colonic and cecal patches, and isolated lymphoid follicles in avian species. A specific area called the pyloric tonsil abounding with lymphoid follicles has recently been reported as a novel component of GALT in chickens. $CD45^-$ Follicular dendritic cells (FDCs) are a unique population of cells essential for efficient germinal center (GC) formation and maintenance in lymphoid follicles because they have a unique feature to retain intact antigens for extended periods. In this study, we aimed to detect residential dendritic cells in the pyloric lymphoid follicles using the immunofluorescence method. For this purpose, ten adult American bronze turkeys [male: five adults (10-12 kg); female: five adults (8-9 kg)] were taken from farms engaged in turkey breeding and slaughtering in Ankara province. The tissues were fixated with a 10% neutral buffered formalin (NBF) solution, and then tissue samples underwent the tissue preparation protocol. Anti-vimentin antibody (1:50) was dropped on serial sections (5 µm) to demonstrate FDCs, professional antigen-presenting cells that inform the fight against invasive pathogens. Positive reactions within lymphoid follicles revealed that FDCs harboring in the germinal center expressed the intermediate filament vimentin. In addition, it was realized the presence of cellular spiny protrusions. These findings implicate that the pyloric region in turkeys may have lymphoid follicles rather than lymphocyte infiltrations.

Keywords: Follicular dendritic cell, GALT, Germinal center, Lymphoid follicle, Pyloric tonsil, Vimentin.

DRAFT

ORAL PRESENTATION

Importance of cellular mitophagy (mitochondrial autophagy) and the role of mitochondrial dysfunction in the occurrence of several diseases

Nuh YILDIRIM[1*] (*ORCID:https://orcid.org/0000-0002-5315-923X*)

*[1]Ankara University, Veterinary Medicine, Histology-Embryology, Ankara, Türkiye.

*Corresponding author e-mail: nuhyldrm1988@gmail.com

Abstract

Autophagy is a catabolic process of degradation of cytoplasmic components and imperfect organelles by membrane vesicles. Mitophagy, a selective form of autophagy, involves the isolation of defective or superfluous mitochondria in autophagosomes. Being a mechanism that controls cellular degeneration, mitophagy plays a crucial role in cell fate. Several mitochondrial parameters, such as membranal potential, structure, and ROS, can ignite the activation of mitophagic machinery. These parameters regulate not only mitophagy but also the mitochondrial Ca^{2+} uptake. Calcium handling is fundamental in regulating ATP production by mitochondria and mitochondrial quality control processes. Under these stressful circumstances, damaged mitochondria in mammalian cells have some difficulties (membrane potential loss, isolation, fusion, or fission). Mitochondrial fragmentation due to imbalanced fission and fusion of mitochondria is a prerequisite for mitophagy. On molecular levels, PINK1, a mitochondrial serine/threonine-protein kinase, activates ubiquitin through phosphorylation, and ubiquitin binds to a variety of substrates which are found on the outer mitochondrial membrane (OMM) in a process called substrate ubiquitination. Parkin, an E3-ubiquitin ligase responsible for specific recognition and ubiquitination of target proteins, is recruited from the cytosol. PINK1 phosphorylates Parkin to activate Parkin-enhancing E3 ligase activity. Subsequently, many autophagy receptors migrate from the cytoplasm towards damaged mitochondria and interact with ubiquitinated proteins in OMM. The phagophore engulfs the organelle to form an autophagosome, and then a bi-layered membranous vesicle fuses with the lysosome to degrade the organelle. Mitophagy has recently received increasing attention related to the pathogenesis of clinical diseases, such as cancer, cardiovascular diseases, metabolic diseases, neurodegenerative diseases, renal interstitial fibrosis (RIF), retinal diseases (glaucoma, diabetic retinopathy, retinitis pigmentosa, and age-related macular degeneration). Mitophagy research, a novel and cutting-edge field, is attracting growing interest from the scientific community. As our perspective on mitophagy deepens, we will harvest promising potential therapies against diseases related to dysfunctional mitophagy.

Keywords: Cancer, Cardiovascular diseases, Mitophagy, Neurodegenerative diseases, Parkin, PINK1.

DRAFT

ORAL PRESENTATION

Synthesis, Characterization and *In Silico* ADME Screening of Some New 1,3,4-Thiadizole Derivatives

Beyzanur AKDUMAN[1] (*ORCID: https://orcid.org/000-0003-1542-9099*),
Sevil ŞENKARDEŞ[2*] (*ORCID: https://orcid.org/ 0000-0002-0523-459X*)

[1]Marmara University, Faculty of Pharmacy, Istanbul, Turkey.
[*2]Marmara University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, Istanbul, Turkey.

*Corresponding author e-mail: sevil.aydin@marmara.edu.tr

Abstract

1,3,4-Thiadiazole is a heterocyclic compound that holds significant prominence in various scientific and industrial domains due to its distinctive structural and functional characteristics. Comprising a five-membered ring with two carbon atoms, two nitrogen atoms, and one sulfur atom, this versatile moiety has captured the attention of researchers across fields such as chemistry, pharmacology, and materials science. Building on this information, we synthesized thiosemicarbazones from thiosemicarbazide and aldehydes. The resulting thiosemicarbazone intermediates were converted into 1,3,4-thiadiazoles by a reaction with acetic anhydride used as the solvent. We assessed the synthesized compounds for purity through HPLC and elemental analysis. Their structures were elucidated via IR, ^{1}H-NMR and LC-MS analyses. Furthermore, we conducted *in silico* ADME studies for our compounds. These studies involved utilizing SwissADME and Osiris software to calculate various properties such as physicochemical attributes, pharmacokinetics, lipophilicity and water solubility. The outcomes of the ADME analysis indicate that the compounds adhere to Lipinski's rule of 5, suggesting favorable oral bioavailability. Additionally, the compounds generally exhibited the potential to interact with kinase enzymes, which could position them as promising macromolecular targets. The potential biological activities of the obtained new compounds are still under investigation.

Keywords: 1,3,4-Thiadiazole, synthesis, characterization, ADME screening

DRAFT

ORAL PRESENTATION

Investigation of glucose-related changes of connection channels between pancreatic beta cells and their role in oscillation recovery

Mesut Akyüz[1,2*(0000-0001-8161-2479)], Özel Çapik[1,2(0000-0003-2827-2537)], Murat An[3(0000-0003-1363-980X)], Vehpi Yildirim[4(0000-0003-3837-4756)], Elanur A. Karataş[1,2(0000-0001-8992-6931)], Omer F. Karatas[1,2(0000-0002-0379-2088)]

[1]Erzurum Technical University, Faculty of Science, Molecular Biology and Genetics, Erzurum, Türkiye
[2]Erzurum Technical University, High Technology Application and Research Center, Erzurum, Türkiye
[3]Erzurum Technical University, Faculty of Science, Department of Basic Sciences, Erzurum, Türkiye
[4]Erzurum Technical University, Faculty of Science, Department of Mathematics, Erzurum, Türkiye

*e-mail:mesut.akyuz@erzurum.edu.tr

Abstract

Increased plasma glucose levels are associated with decreased insulin secretion and increased insulin resistance during the development of diabetes. Changes in the expression levels of the gap-junctional connection channels between the beta cells has been the focus of research related to diabetes in recent years. Long-term high plasma glucose levels lead to reduction in the expression of genes encoding the Gap-Junctions between beta cells and thus alteration in the cell network dynamics. Decreased intercellular connection strength due to the increase in the amount of glucose in the diabetes model significantly affects the electrical activity of the cells. Based on their electrical excitability, beta cells in an islet can be categorized as silent, active and overactive. Increased number of silent cells in the population decreases the activity of the cell population and decreases insulin secretion. The unhealthy cells that increase in number with the development of diabetes reduce the insulin-secreting potential of the entire cell population. Cells that are electrically less excitable have a higher K_{ATP} channel activity on their plasma membrane. The maximum conductivity between two adjacent cells in a pancreatic islet is expressed as the gap-junctional conductance (g_c). In a cell population with a high level of unhealthy cells, when g_c is high, all beta cells remain silent. At moderate g_c levels, active healthy cells disengage from the unhealthy cells, act independently and oscillate in a synchronized manner, which results in increase in Ca^{+2} levels. However, when g_c is reduced to zero, each active cell oscillates independently, while the unhealthy cells remain silent and thus the synchronization is completely lost due to low levels of Ca^{+2}. In our study, we investigated the dose- and time-dependent effects of glucose concentration on the expressions of Kir6.1, a gene encoding K_{ATP} channel, and Connexin36, which is a part of intercellular Gap-Junction channels, in an *in vitro* mouse diabetes model. In addition, we showed that reducing the expression of Connexin36 rescued the Ca^{+2} oscillation, which is accepted as the precursor of insulin release from pancreatic beta cells in our *in vitro* diabetes model.

Keywords: Beta cell, Connexin36, Diabetes, Calcium, Potassium ATP channel

DRAFT

ORAL PRESENTATION

Multi-Faceted Analysis of a Novel Sulfonylurea for Potent Herbicidal Activity: Synthesis, Characterization, ADME, Conformational Analysis, Molecular Docking, and Induced Fit Docking

Nüveyre Canbolat[1*] (*ORCID: https://orcid.org/0000-0003-4466-4602*), Ümmühan Özdemir Özmen[1] (*ORCID: https://orcid.org/0000-0001-9161-9367*), Mehmet Yakan[2](ORCID: *https://orcid.org/0000-0003-4954-2073*), İrfan Koca[2](*ORCID: https://orcid.org/0000-0001-7873-159X*), Nurcan Karacan[1](*ORCID: https://orcid.org/0000-0002-9273-7987*)

[*1]Gazi University, Science Faculty, Chemistry Department, Ankara, Turkey.
[2]Yozgat Bozok University, Science Faculty, Chemistry Department, Yozgat, Turkey.

*Corresponding author e-mail: nuveyrecanbolat@gazi.edu.tr

Abstract

Herbicides are substances used to control undesired plants, also known as weeds. Herbicide resistance is a significant problem in agriculture as unwanted plants evolve to withstand herbicides like sulfonylureas, which target the acetohydroxyacid synthase (AHAS) enzyme. These inhibitors are widely used due to their significance across various organisms. The AHAS enzyme is pivotal in amino acid biosynthesis, making it a crucial target for herbicides through oxidative inactivation. As a result, AHAS inhibition disrupts amino acid synthesis in plants, leading to the demise of targeted weeds while posing relatively lower risks to animals and the environment. Sulfonylurea resistance is a significant problem in agriculture where weeds have developed the ability to withstand the effects of sulfonylurea herbicides due to genetic mutations that affect the target enzyme's binding site, reducing the herbicide's efficacy, and necessitating the use of alternative weed management strategies. For that reason, it is needed to synthesize new types of sulfonylureas with additional sub-chemical groups becomes imperative. Among the potential candidates are pyrimidinones, a class of chemicals that have shown promise in inhibiting plant growth processes, particularly photosynthesis, thereby enabling effective weed control. In this study, Combining the attributes of sulfonylurea and pyrimidinone, we envisioned a hybrid compound (Cl-Et) that exhibits potent herbicidal activity. The structure of Cl-Et compound (2-chloro-N-((1-ethyl-2-oxo-4-phenyl-1,2-dihydro pyrimidine-5-yl))benzenesulfonylurea) was characterized using FT-IR, NMR, and MS spectroscopic methods. In addition, enzyme inhibition studies were examined through molecular docking calculations (Schrödinger Suite 2020-3) with Glide XP OPLS2005 force field docking and Induced Fit Docking (IFD) to elucidate non-covalent interactions. ADME (absorption, distribution, metabolism, and excretion) calculation also performed QikProp in Schrödinger Suite. Our work includes conformational studies that help identify the most stable and relevant ligand conformations for effective binding to the target protein.

Keywords: Herbicidal resistance, AHAS enzyme, Sulfonylureas, Pyrimidinone, Molecular docking, IFD.

DRAFT

ORAL PRESENTATION

Pet bird diseases admitted to Ankara University Small Animal Hospital: A retrospective evaluation of 276 visits

Nevra KESKİN YILMAZ[1*] (*ORCID: https://orcid.org/ 0000-0002-6287-1157*)

[*1]Ankara University, Veterinary Faculty, Department of Internal Medicine, Ankara, Türkiye.

*Corresponding author e-mail: keskinnevra@gmail.com

Abstract

As exotic animals including pet birds become more common around the world, exotic animal medicine is also becoming increasingly popular as a broad field of study and research for veterinarians. From this point of view, within the scope of this study, data on pet birds brought to Ankara University Veterinary Faculty Small Animal Hospital Internal Medicine Clinic between September 2019 and September 2022 were examined and evaluated, retrospectively. It is determined that, a total of 216 budgerigars (78.2%), 54 parrots (19.5%) and 6 canaries (2.17%) were presented between the relevant dates. When control visits were excluded from these, 103 budgerigars, 28 parrots and 3 canaries were examined. The mean age (±std) was 4.62±3.17, 4.46±3.26, and 7.66±4.12 of the budgerigars, parrots, and canaries, respectively. The most common diagnosis was categorized as gastrointestinal (54.36%), respiratory (15.53%), and dermatologic diseases (11.65%) in budgerigars while it is gastrointestinal (28.57), dermatologic (25%,) and respiratory diseases (25%) in parrots and gastrointestinal diseases in canaries. Most avian illnesses can affect most of the bird breeds. However, as it is not always possible to list and eliminate all the diseases in the clinical practice, knowing breed-specific diseases, predispositions and some common health conditions contributes significantly to diagnosis and treatment processes. For this reason, it would be beneficial for veterinarians to consider the above disease rates, especially on a regional basis, when examining pet birds.

Keywords: avian, exotic animal medicine, pet birds

DRAFT

ORAL PRESENTATION

İyonik sıvı ve bakır nanopartiküller ile modifiye edilmiş kalem grafit elektrotların elektrokimyasal karakterizasyonu

Betül Bozdoğan[1*] (*ORCID: https://orcid.org/ 0000-0003-1546-3895*)

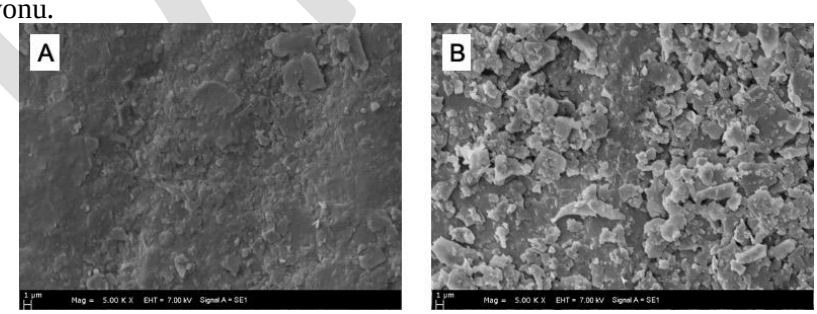
[*1]Aksaray Üniversitesi, Fen-Edebiyat Fakültesi, Kimya Bölümü, Ankara, Türkiye

*Sorumlu yazar e-mail: betulbozdogan@aksaray.edu.tr

Özet

İyonik sıvılar (İS), erime noktası 100°C'nin altında olan sıvı organik tuzlardır. Organik katyonlar ve organik veya inorganik anyonların sıvı bir formda bir arada bulunduğu yapılardır. Tamamen iyonlardan oluşan ve oda sıcaklığında sıvı halde olan organik tuzlardır [1]. İyonik sıvılar geniş elektrokimyasal pencere, mükemmel iyonik iletkenlik, yüksek viskozite, ihmal edilebilir buhar basıncı, güçlü termal ve kimyasal stabilite gibi sahip oldukları eşsiz fizikokimyasal özelliklerden dolayı elektrokimyasal sensör uygulamalarında modifikasyon ajanı olarak sıklıkla kullanılmaktadır. İyonik sıvı elektrot yüzeyi ile etkileşime girerek reolojik ve iletkenlik gibi elektrotun bazı kritik özelliklerini değiştirmektedir. İS ile modifikasyonun elektrot iletkenliğini ve elektron transferini artırarak sensör cevabını iyileştirdiği bilinmektedir [2]. İS ve metalik nanopartiküllerin bir arada yer aldığı kompozit ve hibrit modifikasyonlar ise bu iki bileşenin sinerjik etkisiyle sensöre daha üstün elektrokimyasal özellikler sağlamaktadır [3-5].
Sunulan çalışmanın ilk kısmında, hidrofobik ve aprotik bir iyonik sıvı olan 1-butil-3-metilimidazolyum hekzaflorofosfat ($BMIMPF_6$) ile modifiye edilmiş kalem grafit elektrotların (KGE) morfolojik ve elektrokimyasal karakteristikleri incelenmiştir (Şekil 1). Adsorpsiyon, fiziksel tutuklanma, elektrokaplama ve tabaka-tabaka düzenlenme gibi çeşitli modifikasyon yöntemlerinin ve parametrelerinin İS ile modifiye edilmiş KGE'nin elektrokimyasal cevabı üzerine etkileri dönüşümlü voltametri tekniği ile değerlendirilmiştir. Çalışmanın ikinci kısmında ise, KGE'nin bakır oksit nanopartiküller ve $BMIMPF_6$ ile hibrit modifikasyonunun sinerjik etkisi fizikokimyasal ve elektrokimyasal olarak incelenmiştir. Bu kapsamda, bakır oksit nanopartiküller KGE yüzeyinde elektrokimyasal çöktürme yöntemiyle sentezlenmiştir. Çöktürme esnasında (in situ deposition) ve çöktürme sonrasında (post deposition) İS ile modifikasyonun sensör performansı üzerine etkisi dönüşümlü voltametri tekniği ile araştırılmıştır. Deneysel çalışmaların sonucunda, KGE'nin yalnızca İS ile modifikasyonu ve bakır oksit nanopartiküllerle İS'nin hibrit modifikasyonu karşılıklı sistematik olarak değerlendirilmiştir. Elde edilen sonuçlar doğrultusunda, ileriki çalışmalarda sensör olarak kullanılmak üzere KGE için en uygun modifikasyon yöntemi ve modifikasyon parametreleri belirlenmiştir.

Anahtar Kelimeler: iyonik sıvı, elektrokimyasal sensör, kalem grafit elektrot, bakır oksit nanopartikül, elektrot modifikasyonu.



Şekil 1: A) Modifiye edilmemiş ve B) İS ile modifiye edilmiş kalem grafit elektrotların SEM görüntüsü.

Kaynaklar

[1]Yavir K, Marcinkowski Ł, Marcinkowska R, Namieśnik J and Kloskowski A 2019 *Analytica Chimica Acta* **1054** 1-16
[2]Cetinkaya A, Kaya SI, Yence M, Budak F and Ozkan SA 2023 *Trends in Environmental Analytical Chemistry* **37** e00188
[3]He Z and Alexandridis P 2017 *Advances in Colloid and Interface Science* **244** 54-70
[4]Khorsandi D, et al. 2022 *Clinical and Translational Discovery* **2-3** e127
[5]Moreira F, Santana ER and Spinelli A 2020 *Scientific Reports* **10-1** 1955

ORAL PRESENTATION

Metal complexes based on substituted salicylidene: synthesis and characterization

Ahmet Oral Sarıoğlu (*ORCID: https://orcid.org/0000-0001-7787-7968*)

Gaziantep University, Department of Chemistry and Chemical Processing Technologies, Naci Topçuoğlu Vocational School, Gaziantep, Turkey

*Corresponding author e-mail: aoralsarioglu@gmail.com

Abstract

Schiff base derivatives have attracted attention for a long time because they are widely used in a variety of fields, including industrial organic chemistry, biochemistry, and medicinal chemistry. Schiff base molecules may be formed by the reflux reaction of aldehydes or ketones with primary amines in organic solvents like ethanol or methanol. Shiff bases (-C=N-) obtained from carbonyl compounds of amines form complexes with metals. These complex compounds are used in many different fields today, and their usage areas are increasing day by day. Some of these areas are: dyestuffs, color pigments, stabilizers in polymers, anti-inflammatory, anti-cancer, anti-HIV, anti-tuberculosis, antipyretic agent molecules, and Schiff bases and their indispensable metal complexes in the field of pharmacology. In this study, Schiff base ligand was synthesized from 2-hydroxy-5-nitrobenzaldehyde and 4-bromoaniline compounds. Complex compounds were synthesized from some transition metal salts with the obtained Schiff base ligand. The structures of the synthesized complex compounds were elucidated by spectroscopic methods such as UV-Vis and FT-IR, and by measurements such as elemental analysis, magnetic susceptibility, and electrolytic conductivity.

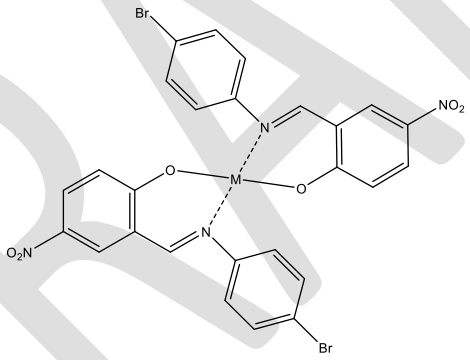


Figure. Structure of synthesized complexes

Keywords: Schiff base, Metal complexes, Characterization, Synthesis

References

Dyk, H. v., Jacobs, F. J. F., Kroon, R. E., Makhafola, T. J., Brink, A. Characterisation, structural investigations and biological activity of substituted salicylidene-based compounds, J. Mol. Struct., 2023, 1276, 134737, https://doi.org/10.1016/j.molstruc.2022.134737

Uddin, M. N., Ahmed, S. S., Alam, S. M. R. REVIEW: Biomedical applications of Schiff basemetal complexes, J. Coord. Chem., 2020, 73(23), 3109–3149. https://doi.org/10.1080/00958972.2020.1854745

A.O. Sarıoğlu et al. Synthesis of a new ONNO donor tetradentate schiff base ligand and binuclear Cu(II) complex: Quantum chemical, spectroscopic and photoluminescence investigations. J. Lumin. 2016, 176, 193–201. https://doi.org/10.1016/j.jlumin.2016.03.021

DRAFT

ORAL PRESENTATION

Tıbbi cihazların ve ürünlerinin değerlendirilmesinde mikrobiyolojik testlerin önemi

Pınar Mursaloğlu Kaynar[1*] (*ORCID: https://orcid.org/0000-0002-8451-1777*)

[*1]Ankara Medipol Üniversitesi, Sağlık Hizmetleri Meslek Yüksekokulu,
Tıbbi Hizmetler ve Teknikler Programı, Ankara, Türkiye

*Sorumlu yazar e-mail: pinar.mursaloglu@ankaramedipol.edu.tr

Özet

Hastalıkların tanı ve tedavisinde kullanılan tıbbi cihazlar ve ürünleri sürekli artmaktadır. Tıbbi cihazların/ürünlerinin geliştirilmesi sürecinden başlayarak güvenlikleri ve etkinlikleri değerlendirilmelidir. Tıbbi cihazların/ürünlerinin üretimi, nakliyesi, depolaması veya kullanımı gibi çeşitli aşamalarında mikroorganizmalar bulunabilmektedir. Bu mikroorganizmalar da enfeksiyonlara, advers reaksiyonlara veya cihaz arızalanmaları gibi hasta güvenliğini ve sağlık sonuçlarını tehlikeye atabilecek olumsuzluklara neden olabilmektedir. Bu nedenle tıbbi cihazlarına/ürünlerine yönelik mikrobiyolojik testlerin yapılması önemlidir. Mikrobiyolojik testler, tıbbi cihazlarında/ürünlerinde mikrobiyal kontaminasyona karşı biyolojik tehlikeleri belirlemek ve potansiyel risklerini değerlendirmek için ihtiyaç duyulmaktadır. Ayrıca, olası kontaminasyon kaynaklarını saptayarak kontaminasyon risklerini azaltmak için en uygun üretim süreçleri, sterilizasyon yöntemleri, uygun kullanımları ve bakım prosedürleri belirlenmektir. Üretim sonrası da tıbbi cihazın/ürünün kalitesinin sürdürülmesini ve mevzuatta uygunluğunun kontrol edilmesini mikrobiyolojik testlerle sağlanmaktadır. Mikrobiyolojik testleri biyoyük, sterilite, bakteriyel endotoksin, çevresel izleme, mikrobiyal tanımlama ve mikrobiyal limitleri belirleme testleri şeklinde sınıflandırılmaktadır. Biyoyük testi ile bir tıbbi cihazdaki/üründeki canlı mikroorganizmaların sayısı ölçülmektedir. Test sonuçları, biyolojik yükü kabul edilebilir bir seviyeye indirmek için gereken uygun sterilizasyon yöntemini ve dozunu belirlemeye yardımcı olmaktadır. Sterilite testi, tıbbi cihazın/ürünün canlı mikroorganizmalardan arındırılmış olduğundan emin olmak için yapılmaktadır. Özellikle cerrahi aletler veya implant edilebilir cihazlar gibi steril ortamlarda kullanılması amaçlanan tıbbi cihazlar için önemlidir. Bakteriyel endotoksin (LAL) testi, Gram negatif bakterilerin hücre duvarlarının bir parçası olan endotoksinin varlığını ve düzeylerini belirlemek amacıyla uygulanmaktadır. Çevresel izleme testiyle tıbbi cihazların/ürünlerin üretim ve depolama süreçlerinde olası mikrobiyal kontaminasyonu önlemek için alanlarındaki hava, su ve yüzeylerin mikroorganizmalardan arındırılmış olduklarından emin olmak için yapılmaktadır. Mikrobiyal tanımlama testi, tıbbi cihazla/ürünle ilişkili mikroorganizmaların türlerini anlamak için önemlidir. Test sonuçları ile uygun sterilizasyon yöntemini belirlemek ve temizleme prosedürlerinin etkinliğini değerlendirmek için de kullanılmaktadır. Mikrobiyal limit testiyle bir tıbbi cihazın/ürünün kalite standartlarını karşıladığından emin olmak için belirli mikroorganizmaların/patojenlerin varlığı araştırılmaktadır. Sonuç olarak, tıbbi cihazların/ürünlerinin geliştirilmesi ve değerlendirilmesinde uygun mikrobiyolojik testler seçilmelidir. Mikrobiyolojik test sonuçlarının uygunluklarının hasta güvenliğinin sağlanmasında ve sağlık hizmetlerinin iyileştirilmesinde önemli olduğu görülmektedir.

Anahtar Kelimeler: Mikrobiyolojik test, Tıbbi cihaz, Medikal Ürün, Güvenlik.

DRAFT

ORAL PRESENTATION

Investigation of the Effect of Oxidative Stress on Autophagic Gene Expressions in Gastric Cancer

M. Burcu Irmak Yazıcıoğlu[1*] (*ORCID: https://orcid.org/0000-0002-1651-305X*),
Erna Çopuroğlu[2] (*ORCID: https://orcid.org/0009-0008-6783-5965*)

[*1]Atlas University, Faculty of Engineering and Natural Sciences, Department of Molecular Biology and Genetics, Prof. Dr., Istanbul, Turkey
[2]Haliç University, Institute of Graduate Studies, Department of Molecular Biology and Genetics, Istanbul, Turkey

* Corresponding author e-mail: burcu.yazicioglu@atlas.edu.tr

Abstract

Gastric cancer is a malignant tumor that is seen in the fifth place worldwide and in the third place in cancer-related deaths. Oxidative stress is a state of imbalance between oxidants and antioxidants in the cell. Increased reactive oxygen derivatives (ROS) in the cell contribute to the development of cancer by causing cellular damage. It is extremely important to suppress oxidative stress in the cell by antioxidants in order to maintain cellular homeostasis. Caffeic acid phenethyl ester (CAPE), the main component of propolis, is a flavonoid compound with antioxidant and anti-cancer effects. Autophagy; it is a self-destruct mechanism activated as a result of stress factors such as nutrient deficiency, hypoxia and oxidative stress. Beclin-1 gene is involved in processes such as autophagy and apoptosis, and it is known that gene expression increases under stress conditions. In this study, it was aimed to investigate the changes in autophagy-related Beclin-1 gene expressions in AGS and MKN-45 gastric cancer cells under oxidative stress conditions. AGS and MKN-45 gastric cancer cell lines were cultured in media containing 89% Dulbecco's Modified Eagle Medium (DMEM), 10% Fetal Bovine Serum (FBS) and 1% Penicillin-Streptomycin at 37 °C and 5% CO_2. After the cells were cultured, hydrogen peroxide (H_2O_2) and H_2O_2+CAPE were applied to AGS and MKN-45 cells. After cell culture, total RNA isolation and cDNA synthesis were performed from control group, H_2O_2 and H_2O_2+CAPE treated cells. Changes in Beclin-1 gene expression levels in oxidative stress conditions and antioxidant-treated gastric cancer cells were investigated by quantitative real-time-polymerase chain reaction (qRT-PCR). The results were statistically analyzed by the $2^{-\Delta\Delta CT}$ method. Our findings showed that Beclin-1 gene expressions increased in H_2O_2-treated AGS and MKN-45 cells compared to the control group, while Beclin-1 expression decreased in H_2O_2+CAPE-treated AGS and MKN-45 cells compared to the control group.

Keywords: Gastric cancer, Oxidative stress, CAPE, Beclin-1

This study was carried out at Haliç University, Department of Molecular Biology and Genetics, Cancer Genetics Research Laboratory.

DRAFT

ORAL PRESENTATION

Effective Removal of Trypan Blue From Aqueous Solutions by Using Cu-Vanadate

Sedef ŞİŞMANOĞLU[1*] (*ORCID: https://orcid.org/0000-0002-1379-4423*)

[*1]Karabuk University, Science Faculty, Department of Chemistry, Karabuk, Turkey

*Corresponding author e-mail: sedefsismanoglu@karabuk.edu.tr

Abstract

The azo acid dye Trypan blue used in the textile industry is carcinogenic, and its wastes pose a threat to the environment, human and animal health. In this study, the adsorption of Trypan blue (TRB) dye at 5-10-25-50ppm concentrations with Cu-Vanadate adsorbent at 25-35 and 45 °C was investigated. In the study performed with UV-Vis spectrometry at 590 nm, the time for adsorption to reach equilibrium was found to be 60 minutes and V(L)/m(g) = 1. As adsorption isotherms; Freundlich, Langmuir, Tempkin, Frumkin or Fowler-Guggenheim (FFG) Dubinin and Radushkevich (D-R), Flory-Huggins (F-H) isotherms were used. In addition, thermodynamic and kinetic studies were carried out. After 60 minutes of Langmuir adsorption isotherm, it is seen that the maximum adsorbing capacity (q_{max}) of the adsorbent at equilibrium changes as 103.1mg/g-37mg/g-33mg/g at 25 ˚C-35 ˚C -45 ˚C temperature values, respectively, and the highest adsorption capacity is at room temperature (25 ˚C). The characterization of the adsorbent was performed before and after adsorption by FTIR/ATR, SEM and TGA.

Keywords: Cu-Vanadate, Trypan blue, Adsorption

Acknowledgement
I would like to thank Prof. Dr. Gülin Selda POZAN SOYLU for her help at every stage of this study.

DRAFT

ORAL PRESENTATION

Immobilization of La-ZnFe LDH on polydimethylsiloxane sponge for photocatalytic degradation of antibiotic

Sultan Akdag Turkay[1*] (*ORCID: https://orcid.org/0000-0003-2168-3341*), Alireza Khataee[1,2*] (*ORCID: https://orcid.org/0000-0002-4673-0223*), Nevin Atalay Gengeç[3] (*ORCID: https://orcid.org/0000-0003-0993-4398*), Tannaz Sadeghi Rad[1] (ORCID: *https://orcid.org/0000-0002-5609-8751*)

[*1] Gebze Technical University, Faculty of Engineering, Department of Environmental Engineering, Gebze, Turkey.
[2] University of Tabriz, Faculty of Chemistry, Department of Applied Chemistry, Tabriz, Iran.
[3] Kocaeli University, Department of Environmental Protection, Kocaeli, Turkey.

*Corresponding authors: akhataee@gtu.edu.tr; sakdag@gtu.edu.tr

Abstract

The overuse of antibiotics and their subsequent release into the environment pose a potential threat to the ecosystem. Advanced oxidation processes like photocatalysis, activated persulfate, sonocatalysis, and so on are among the most effective methods for removing highly stable antibiotics resistant to biodegradation. Photocatalysis is preferred for its simple operation, cost-effectiveness, absence of secondary pollution, and high efficiency in pollutant degradation. This process relies on using semiconductor materials to degrade pollutants through redox reactions under visible or ultraviolet light. Numerous semiconductor materials, such as layered double hydroxides (LDHs), TiO_2, and ZnO can be utilized as photocatalysts. However, the low stability and durability of some photocatalysts restrict their long-term applicability. To solve this problem, catalysts can be immobilized onto supporting substrates such as glass, polymers, and carbon nanotubes, which increases their stability. In the present study, La-ZnFe LDH was immobilized on polydimethylsiloxane (PDMS) sponge to increase the catalyst's stability and reusability. The photocatalytic activity of the La-ZnFe LDH-embedded PDMS sponges was evaluated for the degradation of rifampicin as an antibiotic. By optimizing the operating factors, 92.8% of rifampicin occurs within 60 min with 15 mg/L rifampicin, 118 pieces of LaZnFe LDH@PDMS sponge, 0.2 mmol/L of peroxymonosulfate, and 100 W of light intensity and at natural pH of 8. After seven cycles, the degradation efficiency of rifampicin only decreased by 1.5%, indicating that the immobilized catalyst exhibits remarkable reusability and stability. The results confirmed that immobilizing LDH onto a supporting substrate such as PDMS sponge is a promising strategy for improving the catalyst's reusability.

Keywords: Antibiotic degradation, Immobilize catalyst, Polydimethylsiloxane, Photocatalysis, Peroxymonosulfate activation.

DRAFT

ORAL PRESENTATION

Investigation of the effects of acute caffeine intake on oxidative stress in the brain and depression-like behaviors in ovariectomized mice

[1]Dilek Ozbeyli[*] (*ORCID: https://orcid.org/0000-0002-4141-6913)*

[*1]Marmara University, Vocational School of Health Services, Department of Medical Services and Techniques, Istanbul, Türkiye.

*Corresponding author e-mail: dilekozbeyli@marmara.edu.tr

Abstract

It is noteworthy that the risk of depression increases in women during the transition to menopause. In ovariectomized rodents, some studies have been reported that oxidative stress increases in areas of the brain related to anxiety and depression-like behaviors due to hormone imbalance. However, there is controversial data on the effects of moderate doses of caffeine on depression, anxiety-like behaviors, and brain oxidative status in the literature. Our hypothesis is that acute caffeine administration alleviates the possible oxidative state in the brain and reduces depression-like behaviors in ovariectomized Balb-C mice. After ethical approval (06.2023.mar), the groups were designed as Sham + Vehicle (Sham+V), Sham + Caffeine (Sham+C), Ovariectomy + Vehicle (OVX+V), and Ovariectomy + Caffeine (OVX+C) (n = 6 per group). Four weeks after ovariectomy, on the day of the experiment, single dose 20 mg/kg caffeine (moderate dose) to the caffeine groups and an equivalent volume of serum physiologic to the vehicle groups were given. Following 4 hours, depression-like behaviors were evaluated by a tail suspension test, and after euthanasia, the brains were removed. Brain oxidative status was assessed by measuring malondialdehyde and glutathione levels. Statistical analysis of the parameters was performed with a one-way analysis of variance (ANOVA) supported by Tukey's multiple comparison tests; variances were considered significant if $p<0.05$ and mean ± S.D. The findings showed no significant changes in MDA or GSH levels between groups. The immobility time of the Sham + C and OVX+C groups significantly decreased compared to the Sham+V and OVX+V groups, respectively ($p<0.05$; $p<0.001$). In conclusion, a 4-week estrogen hormone deficiency did not cause significant oxidative damage to the brain, and acute caffeine administered did not affect oxidative parameters. On the other hand, acute caffeine treatment significantly reduced the increased immobilization time, which is considered a marker of depression-like behavior due to ovariectomy.

Keywords: Caffeine, depression-like behavior, tail suspension, ovariectomy, oxidative stress, mice.

DRAFT

ORAL PRESENTATION

Habitat Preferences of Anatolian Water Frog Lineages (Genus: *Pelophylax*)

Çiğdem Akın Pekşen [1,2*] *(ORCID: https://orcid.org/0000-0001-5736-3062)*,
Banu Kaya Özdemirel[1] (*ORCID: https://orcid.org/0000-0003-2039-4601*)

[*1]Country Başkent University, Faculty of Science and Letters, Department of Molecular Biology and Genetics, Ankara, Türkiye.
[2] Başkent University, Institute of Transplantation and Gene Sciences, Ankara, Türkiye.

*Corresponding author e-mail: cerigensis@gmail.com

Abstract

Obtaining ecological knowledge about the terrestrial demands of frogs such as habitat preference is critical to compose effective conservation strategies for changing environmental conditions due to global climate change (Toledo and Jared 1993; Wikström, 2018). There are limited studies on water frog's habitat preferences. For example, in Central Europe P. lessonae avoids running waters and can tolerate ponds with hypoxic water; P. ridibundus prefers habitats under strong river influence with well-oxygenated water; their hybrid species P. esculentus could perform well in both types of environments with variable extent (Lada et al., 2995; Plenet et al., 2000). Anatolian water frog complex (Genus: *Pelophylax*) consists of narrow (*Caralitanus, Cerigensis, Cilician*) and widely distributed lineages (*Cf.bedriagae, Euprates*). Although these frog lineages occupy a wide variety of habitat types like lake, river, stream, brook, pond, artificial pond or pool, marsh, brook, and water puddle within their distribution range, it is not known whether there is a significant difference in habitat preferences among frog lineages. Therefore, in this study, we aimed to understand whether there is a difference in habitat preferences of frog lineages distributed throughout Anatolia. In this context, the Kruskal Wallis and post-hoc Dunn tests were respectively applied in accordance with the unpaired nonparametric conditions, and thus habitat preferences across different altitude gradients were investigated and tested both for all water frog lineages and pairs of water frog lineages. Accordingly, the results showed that there is a statistically significant difference between habitat preferences of Anatolian water frog lineages. This means that the distributions of these lineages do not overlap with each other, and they have different habitat preferences. In addition, *Cf. bedriagae, Caralitanus*, and *Euphrates* lineages exhibit a generalist behaviour by occupying a wide variety of habitats in a wide altitude range, while *Cerigensis* lineage occupies brook, water puddle habitat types and so exhibit a more selective behaviour. Thus, these findings are very important to developing effective conservation strategies especially for narrowly distributed *Cerigensis* lineage with a more selective behaviour.

Keywords: Habitat preference, water frog, *Pelophylax*, lineage, Anatolia.

DRAFT

ORAL PRESENTATION

Comparison of Phenolic Content and Antioxidant Activity of Two Different White Honey Samples

Tuğba Nigar BOZKUŞ[1*] *(ORCID: https://orcid.org/0000-0001-9613-6911)*

[*1]Artvin Coruh University, Artvin Vocational School, Department of Chemistry and Chemical Processing Technologies, Artvin, Türkiye

*Corresponding author e-mail: tugbancakiroglu@artvin.edu.tr

Abstract

White honey is a type of honey produced by Caucasian bees in very limited areas around the world. White honey, which has a crystal structure in the mountainous regions of the Caucasus with the effect of altitude and flora, is produced in Turkey, especially in the high parts of Artvin and Ardahan regions. This study aimed to determine the polyphenol and flavonoid content amounts and antioxidant capacity of the aqueous extracts of two different white honey samples produced in Yusufeli district of Artvin province and to compare the phenolic content and antioxidant activity of the white honey samples from the same region. Total polyphenol content and total flavonoid content of 600 mg/mL aqueous extracts of white honey samples was determined by modified Folin-Ciocalteu method, and Aluminum chloride colorimetric method, respectively. Antioxidant activities of the extracts were determined by ferric (Fe^{3+}) reducing antioxidant power. As a result of the analyses; total polyphenol content of the aqueous white honey extract of Bakırtepe and Kılıçkaya villages of Yusufeli district in Artvin province was respectively, 16.37 ± 0.78 mg Gallic acid/100 g honey and 15.27 ± 0.91 mg Gallic acid/100 g honey, total flavonoid content was respectively, 1.54 ± 0.25 mg Quercetin/100 g honey and 4.28 ± 0.65 mg Quercetin/100 g honey, and ferric (Fe^{3+}) reducing antioxidant power was respectively 95.43 ± 1.35 mg Trolox/100 g honey and 97.55 ± 0.73 mg Trolox/100 g honey. While it was determined that the aqueous extracts of two different white honey samples were close to each other in terms of polyphenol content and antioxidant activity, the amount of flavonoid content was found to be much higher in the Kılıçkaya sample. In this respect, it was concluded that different white honey samples from the same region showed similar phenolic content and antioxidant activity, but the difference in altitude and vegetation showed variation in flavonoid content.

Keywords: White honey, Polyphenol, Flavonoid, Antioxidant activity, Artvin

DRAFT

ORAL PRESENTATION

Serotonin ve Melatonin Uygulanmış Meme Kanseri Hücrelerinde MT-1 ve MT-2 Gen Ekspresyon Seviyelerinin İncelenmesi

M. Burcu Irmak Yazıcıoğlu[1*] (ORCID: https://orcid.org/0000-0002-1651-305X)
Berna Türkmen[2] (*ORCID: https://orcid.org/0009-0006-8515-3840)*
Erna Çopuroğlu[3] (ORCID: https://orcid.org/0009-0008-6783-5965)

[*1]Atlas Üniversitesi, Mühendislik ve Doğa Bilimleri Fakültesi, Moleküler Biyoloji ve Genetik Bölümü, Prof. Dr., İstanbul, Türkiye
[2]Haliç Üniversitesi, Fen Edebiyat Fakültesi, Moleküler Biyoloji ve Genetik, İstanbul, Türkiye
[3]Haliç Üniversitesi, Lisansüstü Eğitim Enstitüsü, Moleküler Biyoloji ve Genetik Anabilim Dalı, İstanbul, Türkiye

*Sorumlu yazar e-mail: burcu.yazicioglu@atlas.edu.tr

Özet

Meme kanserinin tüm dünyada ki görülme sıklığı (özellikle kadın popülasyonunda) akciğer kanserinden sonra 2. sırada gelmektedir. Meme kanseri, tipik olarak hormon-bağımlı kanser türüne örnektir ve literatürde hastaların büyük çoğunluğunun hormon bağımlı meme kanseri olduğu bildirilmiştir. Melatonin, sirkadiyen ritmin düzenlenmesinde büyük öneme sahip olan bir hormondur. Hormon-bağımlı bir kanser olan meme kanserinde, melatoninin onkostatik etki gösterdiği bilinmektedir. G proteinine bağlı melatonin reseptörü olan melatonerjik tip 1 reseptörü (MT-1) (yüksek afiniteli) bir melatonin reseptörüdür. Melatoninin bir diğer reseptörü olan Melatonerjik tip 2 reseptörü (MT-2) (düşük afiniteli) ile olan ilişkisi incelendiğinde MT-2 reseptörünün immun fonksiyonda önemli rol oynadığı bilinmektedir. Yumurtalık kanseri ve kanser kök hücreleri üzerinde yapılan bir çalışmada, melatoninin MT-1 ve MT-2 ifadelerini yukarı regüle ettiği gösterilmiştir. Triptofan aminoasidi hem melatonin hem de serotonin hormonunda bulunmaktadır ve meme bezi gelişiminde büyük öneme sahiptir. Serotonin, insanda mutluluk hissi verdiği bilinen ve çeşitli hücre tiplerinde mitojen olarak aktivite gösteren bir nörotransmitterdir. Literatürde, serotonin hormonun MT-1, MT-2 gen ifadeleri üzerindeki etkisi bilinmemektedir ve bu projede melatonin ve serotoninin MT-1 ve MT-2 genlerinin üzerindeki etkisinin araştırılması amaçlanmıştır. Bu çalışmada MCF-7 meme kanseri hücreleri %89 Dulbecco's Modified Eagle Medium (DMEM), %10 Fetal Bovine Serum (FBS) ve %1 Penisilin-Streptomisin besiyeri ortamında, 37 °C ve %5 CO_2 koşullarında kültüre edilmiş ve sonrasında MCF-7 hücrelerine 24 saat süre ile 50 nM serotonin ve melatonin uygulaması yapılmıştır. Uygulama sonrasını takiben total RNA izolasyonu, cDNA sentezi ve gerçek zamanlı-polimeraz zincir reaksiyonu (qRT-PCR) yapılmış ve veriler istatistiksel olarak analiz edilmiştir. Bulgularımız, melatonin uygulanan meme kanseri hücrelerinde MT-1 ve MT-2 gen ekspresyonunun kontrol grubuna göre arttığını gösterirken, serotonin uygulanmış hücrelerde MT-1 ve MT-2 gen ekspresyon seviyelerinin kontrol grubuna göre azaldığını göstermiştir.

Anahtar Kelimeler: Meme kanseri, Serotonin, Melotonin, MT-1, MT-2

Bu çalışma, 1919B012203901 proje numaralı TÜBİTAK 2209-A, Üniversite Öğrencileri Araştırma Projeleri Desteği Programı tarafından desteklenmiştir ve Haliç Üniversitesi, Moleküler Biyoloji ve Genetik Bölümü, Kanser Genetiği Araştırma Laboratuvarında gerçekleştirilmiştir.

DRAFT

ORAL PRESENTATION

Does different concentration of high fructose corn syrup impact some hematological parameters in broilers?

Taha Altuğ[1*] *(ORCID: https://orcid.org/0000-0002-7181-4074)*, Gökhan Şen[2] *(ORCID: https://orcid.org/0000-0001-6222-6986)*, Ruhi Kabakçı[1] *(ORCID: https://orcid.org/0000-0001-9131-0933*)
[1]Kirikkale University, Faculty of Veterinary Medicine, Department of Physiology, Kirikkale, Turkey.
[2]Kirikkale University, Faculty of Veterinary Medicine, Department of Animal Nutrition, Kirikkale, Turkey.

*Corresponding author e-mail: tahaaltug@kku.edu.tr

Abstract

Fructose is widely used as a sweetening substitute for glucose or sucrose in food processing of modern human diet. Besides fruit by-products, fructose is used on poultry farms and, its' acute effect on the chicks' metabolism is still unclear. We therefore aimed to investigate the effects of high fructose corn syrup (HFCS) consumption on hematological parameters in broilers. Total 36 chicken broilers were divided into three groups with equal number of animals, and fed *ad libitum* with isonitrogenic-isocaloric feeds and water for 42 days. Animals in the first group served as control and were given no other treatment, while the diets of broilers in the second and third groups contained 50 mg/kg (low) or 100 mg/kg (high) HFCS, respectively. At the last day of experiment, animals were killed by cervical dislocation and blood samples (5 mL) were collected into test tubes with anticoagulant (K_3-EDTA) for hematological analysis. Our findings showed that the RBC of broilers treated low or high HFCS were not statistically different from those of control animals and HGB value was not affected by both concentrations of HFCS treatments ($P<0.05$). HFCS treatment significantly ($P<0.01$) increased PCV and MCV in the high-dose group but not in the low-dose group compared with the control. On the other hand, MCH increased ($P<0.01$) by low-HFCS and MCHC decreased ($P<0.05$) by high-HFCS compared to untreated control group. WBC and monocyte values were not affected from any HFCS treatment ($P>0.05$). However, the treatment of low HFCS increased basophil ($P<0.05$), heterophil and heterophil/lymphocyte ratio, and decreased lymphocyte ($P<0.001$) while the treatment of high HFCS decreased eosinophil ($P<0.05$) and heterophil, and increased lymphocyte ($P<0.001$). In conclusion, this study revealed that dietary fructose intake may change some hematological parameters which can be fore symptoms of further systemic or metabolic diseases.

Keywords: high fructose, broilers, blood parameters, heterophil to lymphocyte ratio

DRAFT

ORAL PRESENTATION

Mide Kanseri Hücrelerinde Oksidatif Stresin VEGF Gen Ekspresyonu Üzerindeki Etkisi

M. Burcu Irmak Yazıcıoğlu[1*] (*ORCID: https://orcid.org/0000-0002-1651-305X*),
Merve Yılmaz[2] (*ORCID: https://orcid.org/0000-0001-8223-8933*)
Erna Çopuroğlu[3] (*ORCID: https://orcid.org/0009-0008-6783-5965*)

[*1]Atlas Üniversitesi, Mühendislik ve Doğa Bilimleri Fakültesi, Moleküler Biyoloji ve Genetik Bölümü, Prof. Dr., İstanbul, Türkiye
[2]Haliç Üniversitesi, Fen Edebiyat Fakültesi, Moleküler Biyoloji ve Genetik, İstanbul, Türkiye
[3]Haliç Üniversitesi, Lisansüstü Eğitim Enstitüsü, Moleküler Biyoloji ve Genetik Anabilim Dalı, İstanbul, Türkiye

[*]Sorumlu yazar e-mail: burcu.yazicioglu@atlas.edu.tr

Özet

Mide kanseri, dünya çapında sık görülen malign bir tümördür ve kansere bağlı ölümler arasında üçüncü sırada yer alan küresel bir sağlık sorunudur. Oksidatif stres, pro-oksidanlar ve antioksidanlar arasındaki denegesizlik durumu olarak tanımlanır. Hücre içerisinde süperkoksit, hidroksil ve hidrojen peroksit gibi serbest radikallere bağlı olarak reaktif oksijen türleri (ROS) seviyeleri artar. Hidrojen peroksitin (H_2O_2) tümör hücre büyümesi, proliferasyon, anjiyogenez, metastaz ve apoptoz gibi süreçlerde yer aldığı ve ROS seviyelerini arttırdığı bilinmektedir. Artan ROS seviyeleri oksidatif stres ile sonuçlanır ve kanser gelişimine katkıda bulunur. Vasküler endotelyal büyüme faktörü (VEGF), anjiyogenezin ana aracısı olmakla birlikte tümör metastazı ve büyümesinde de oldukça önemlidir. Endojen ROS kaynaklarının çeşitli hücrelerde VEGF'i indüklediği, hücre proliferasyonunu ve göçünü desteklediği bilinmektedir. Literatürdeki mevcut kanıtlar, mide kanserinde tümörün ilerlemesi ile birlikte VEGF gen ifadelerinin arttığını yapılan çalışmalar ile ortaya koymuştur. Bu çalışma kapsamında, AGS ve MKN-45 mide kanseri hücreleri AGS ve MKN-45 hücreleri %89 Dulbecco's Modified Eagle Medium (DMEM), %10 Fetal Bovine Serum (FBS) ve %1 Penisilin-Streptomisin besiyeri ortamında, 37 °C ve %5 CO2 koşullarında kültüre edilmiştir. 100 mm petri kaplarına 10^6 hücre olacak şekilde ekilen AGS ve MKN-45 mide kanseri hücrelerine kontrol grubu hücreleri hariç tutularak 24 saat süre ile 100 µM ve 200 µM H_2O_2 uygulaması yapılmış ve uygun koşullarda inkübe edilmiştir. Kontrol grubu ve oksidatif stres koşulları oluşturulan mide kanseri hücrelerinden total RNA izolasyonu ve cDNA sentezi yapılmıştır. Sentezlenen cDNA örnekleri kullanılarak kantitatif gerçek zamanlı-polimeraz zincir reaksiyonu (qRT-PCR) gerçekleştirilmiş ve H_2O_2 aracılı oksidatif stres koşulları oluşturulmuş AGS ve MKN-45 mide kanseri hücre hatlarında anjiyogenez ile ilişkili VEGF gen ekspresyonu seviyeleri incelenmiştir. qRT-PCR'den elde edilen sonuçlar $2^{-\Delta\Delta Ct}$ yöntemi ile analiz edilmiştir. Bulgularımız, H_2O_2 aracılı oksidatif stres koşullarında VEGF gen ekspresyonunun hem AGS hem de MKN-45 hücrelerinde kontrol grubuna göre arttığını göstermiştir.

Anahtar Kelimeler: Mide kanseri, Oksidatif stres, VEGF

Bu çalışma, 1919B012203119 proje numaralı TÜBİTAK 2209-A, Üniversite Öğrencileri Araştırma Projeleri Desteği Programı tarafından desteklenmiştir ve Haliç Üniversitesi, Moleküler Biyoloji ve Genetik Bölümü, Kanser Genetiği Araştırma Laboratuvarında gerçekleştirilmiştir.

DRAFT

ORAL PRESENTATION

Pharmaceutical Preparation and Characterizations of Anti-Hypertensive Nanocrystals Using Anti-Solvent Re-crystallization Methods

Abdulmohsin Al Airoa[1] (*ORCID: https://orcid.org/0009-0008-9777-9423*),
İbrahim İnanç[2] (*ORCID: https://orcid.org/0000-0003-1988-1197*)

[1]Ondokuz Mayıs University, Institute of Graduate Education, Nanoscience and Nanotechnology Department.
[2]Ondokuz Mayıs University, Faculty of Engineering, Metallurgical and Materials Engineering Department.

20281219@stu.omu.edu.tr

Abstract

Heart disease and high blood pressure are among the most dangerous, significant, and widespread diseases in the present time, which need rapid treatment to control them. Nanotechnology has contributed to a major development in the pharmaceutical industry, improving its positive effect on controlling and improving the performance of traditional medicines in treating diseases. Nanomedicines can target cells that conventional drugs cannot reach at a high rate. Anti-hypertensive nanocrystals and colloidal particles at various concentrations with a suitable stabilizer as an anti-agglomeration agent were produced using the Antisolvent re-crystallization technique. The raw Anti-hypertensive active ingredients nanocrystals and obtained colloidal particles and re-crystallized powders were characterized by using scanning electron microscopy (SEM), X-ray diffraction (XRD), Fourier transform infrared spectroscopy (FTIR), and thermogravimetry analysis (TGA).

Keywords: Anti-solvent re-crystallization, anti-hypertensive**,** nanodrug, characterization

DRAFT

ORAL PRESENTATION

Synthesis of Di-cationic Surfactant Containing Pyridinium Ion as Positively Charged Nitrogen Atom and Inhibition Efficiency Against Metal Oxidation in 1.0 M HCl

Gülşen AKGÜL* (*ORCID: https://orcid.org/0000-0003-1231-8385*), Serkan ÖZTÜRK (*ORCID: https://orcid.org/0000-0002-9396-1403*)

Bursa Uludag University, Faculty of Arts and Sciences, Department of Chemistry, Bursa, Turkey

*081730051@ogr.uludag.edu.tr

Abstract

The metal industry is a sector that has to fight corrosion. Corrosion, known as the oxidation of metal atoms, leads to the deterioration of the metal surface and the reduction of its metallic qualities. It is therefore appropriate to use inhibitors to protect the metal surface from corrosion. Among the effective inhibitors used to prevent metal corrosion, which occurs rapidly in acidic media, are cationic surfactants containing pyridinium ions. Based on this idea, two di-cationic surfactants containing one amide group, one ester group, two pyridinium ions and two positively charged nitrogen atoms thanks to pyridinium ions were synthesized to prevent metal corrosion in 1.0 M HCl solution. Its structure was confirmed by FT-IR spectroscopy. The ability of the compound to inhibit corrosion in acidic corrosive media was evaluated by weight loss technique. For this purpose, different concentrations of inhibitor solutions containing 1.0 M HCl were used and the effect of inhibitor concentration on corrosion was investigated by immersing metal samples in these solutions for 24 hours at room temperature. After the synthesis of di-cationic surfactants, it was revealed that they increased the corrosion resistance of low carbon steel in acidic environment as a result of tests by weight loss method. In these tests, corrosion inhibition efficiencies in the range of 74.61 - 87.95% were determined at different inhibitor concentrations and it was observed that the inhibition efficiency generally increased as the inhibitor concentration increased. Preventing the oxidation of iron atoms in mild steel depends on the adsorption of the inhibitor on the metal surface. As a result of this adsorption, the metal surface gains hydrophobic (water disliking) properties. As the best way to determine this hydrophobic property, the contact angle between the surface and the water dripped onto the metal surface was measured. In addition, surface images were taken with an optical microscope camera with 1600-fold magnification in order to compare the oxide layer images that were visible in the inhibitor-free environment, but not on the surfaces of the metal coupons immersed in the inhibitor (50 ppm) environment, and surface examinations were made by making the surface appearance differences more apparent.

Keywords: Synthesis, Metal corrosion, Corrosion inhibitor, Acidic environment, Contact angle

DRAFT

ORAL PRESENTATION

Kemik doku mühendisliğine yönelik farklı ajanlarla modifiye edilmiş montmorillonit katkılı nanokompozit filmlerin geliştirilmesi

Esma Aleyna Yeşilyurt[1] (*ORCID: https://orcid.org/0009-0007-5708-5630*), Sema Çörtoğlu[1] (*ORCID: https://orcid.org/0009-0002-6338-5713*), Şükran Melda Eskitoros Toğay[1*] (*ORCID:https://orcid.org/0000-0002-7473-8417)*

[*1]Sağlık Bilimleri Üniversitesi, Gülhane Sağlık Meslek Yüksekokulu, Eczane Hizmetleri Programı, Ankara, Türkiye

*Sorumlu yazar e-mail: meskitoros@gmail.com

Özet

Biyomalzemelerin kemik doku hasarlarının tedavisinde kullanımı son yıllarda hızlı bir şekilde artmıştır. Özellikle bu tedavilerde, uygun yapıya sahip biyolojik olarak parçalanabilen malzemelerin seçilmesi en kritik adımdır. Ayrıca biyofonksiyonel şablon oluşturmak ve doğal yapıyı taklit etmek için polimerler ile inorganik dolgu maddelerinin kombinasyonundan oluşan nanokompozitler, tek başına kullanılan polimer matrislerinden daha başarılı sonuç vermektedir. İnorganik dolgular, yüksek mukavemetleri ve geniş spesifik yüzey alanları nedeniyle matristen aktarılan strese dayanabildikleri için biyomalzemelerin mekanik özelliklerini güçlendirmek amacıyla yaygın olarak kullanılmaktadır. Bu çalışmada, dolgu maddesi olarak kullanılan montmorillonit (MMT), iki farklı silan ajanı ((3- aminopropil)trietoksisilan (APTES) ve (3-glisidoksipropil)trimetoksisilan (GPTMS)) ile modifiye edilmiş, organik faz ile ara yüzey etkileşiminin arttırılması ve polimer matrisi içerisinde homojen bir şekilde dağılması sağlanmıştır. Polimer matris olarak ise biyobozunur ve biyouyumlu özelliklere sahip sentetik polimerlerden polikaprolakton (PCL) ve biyouyumlu ve hidrofilik özelliğe sahip polivinilpirolidon (PVP) birlikte kullanılmış ve çözelti döküm yöntemiyle biyomalzemeler üretilmiştir. Kemik doku mühendisliğine yönelik doku iskelesi olarak kullanılacak biyomalzemelerin morfolojik, fizikokimyasal ve mekanik özellikleri, atomik kuvvet mikroskobu (AFM), Fourier transform kızılötesi spektroskopisi (FTIR), temas açısı analizi ve su absorplama kapasitesi analizi ile karakterize edilmiştir. AFM sonuçları, PCL/PVP polimer matrisine modifiye edilmemiş ve edilmiş MMT eklenmesiyle pürüzlülüğün düştüğünü göstermiştir. Pürüzlülük oranı en yüksek GPTMS ile modifiye edilmiş MMT katkılı biyomalzemelerde 0,530 µm olarak bulunmuştur. Temas açı değerleri ve su absorplama kapasiteleri karşılaştırıldığında modifiye edilmiş MMT katkılı biyomalzemelerin daha hidrofilik olduğu ve su absorplama kapasitesinin MMT katılmamış polimer matristen ve modifiye edilmemiş biyomalzemeden daha yüksek olduğu görülmüştür. Sonuç olarak, APTES ile modifiye edilmiş MMT katkılı PCL/PVP biyomalzemeleri, kemik doku hasarlarının tedavisinde doku iskelesi olarak kullanılabilir.

Anahtar Kelimeler: Montmorillonit, Silan, Polikaprolakton, Polivinilpirolidon, Doku iskelesi

Teşekkür
Bu çalışma, TÜBİTAK 2209-A Üniversite Öğrencileri Araştırma Projeleri Destekleme Programı tarafından 1919B012214355 numaralı proje kapsamında desteklenmiştir.

DRAFT

ORAL PRESENTATION

Novel Asymmetric Bisthiocarbohydrazone and Its Mixed Ligand Ni(II) Complex: Synthesis, Characterization and Antioxidant Property

Yeliz Kaya[1*] (*ORCID: https://orcid.org/0000-0001-5606-8088*)

[*1]Istanbul University-Cerrahpaşa, Faculty of Engineering, Department of Chemistry, Istanbul, Turkey.

*Corresponding author e-mail: yeliz.kaya@iuc.edu.tr

Abstract

Thiocarbohydrazide can react with aldehydes or ketones to form mono or bisthiocarbohydrazones. Thiocarbohydrazones have the ability to form chelates with various metal ions through their donor groups. Thiocarbohydrazones and their metal complexes have some biological activities such as antitumor, antioxidant, antimicrobial, antiviral, antitubercular and enzyme inhibition [1,2]. Pyridoxal is an aldehyde in the form of vitamin B6 with a heterocyclic pyridine ring. Vitamin B6 deficiency is associated with depression, dermatitis, microcytic anemia, confusion, celiac disease and weakened immune function in humans [3]. Isatin, a flexible heterocyclic chemical, also known as 1H-indole-2,3-dione, can be used to synthesize a wide range of heterocyclic compounds. Isatin derivatives have also shown excellent interest as they express anticancer, antibacterial, antiviral, antiinflammatory, antioxidant, antimalarial, antifungal, antitubercular, anticonvulsant, analgesic and antidiabetic activity [4]. In this study, a new bioactive bisthiocarbohydrazone ligand (L) was obtained from biological molecules such as isatin and pyridoxal. Along with their remarkable efficiency in transition metal-catalyzed processes, phosphine-based compounds also possess important bioactivities such as antitumor, antiviral, antifungal, antibacterial and antioxidant [5,6]. A new mixed ligand nickel(II) complex with the general formula [Ni(L)(PPh_3)] (PPh_3: triphenylphosphine) was synthesized from the reaction of L with $NiCl_2.6H_2O$ in the presence of triphenylphosphine (Figure 1). The structures of the ligand and complex were characterized by elemental analysis, IR, ^{1}H NMR and UV-VIS spectroscopic techniques. In addition, all compounds were found to show good antioxidant capacity according to the antioxidant activity assay results.

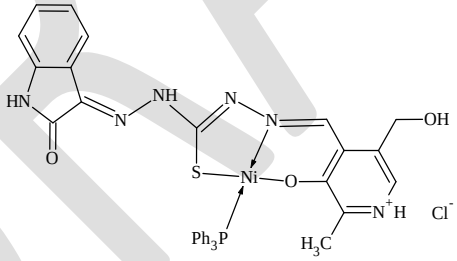


Figure 1. Structure of Ni(II) complex.

Keywords: Bisthiocarbohydrazone, Nickel(II) complex, Isatin, Pyridoxal, Antioxidant activity.

References

[1] C. Bonaccorso, T. Marzo, D. La Mendola, *Pharmaceuticals*, 2020, 13, 1-19.
[2] Y. Kaya, A. Erçağ, Y. Zorlu, Y. Demir, İ. Gülçin, *Journal of Biological Inorganic Chemistry*, 2022, 27, 271-281.
[3] Q. Poladian, O. Şahin, T. Karakurt, B. İlhan-Ceylan, Y. Kurt, *Polyhedron*, 2021, 201, 115164.
[4] A.K. Yadav, S. Srivastava, B. Yogi, S.K. Gupta, *Chemistry Research Journal*, 2022, 7, 33-42.
[5] Y. Ma, S. Li, S. Yang, *Accounts of Chemical Research*, 2017, 50, 1480-1492.
[6] Y. Kaya, A. Erçağ, A. Koca, *Journal of Molecular Structure*, 2020, 1206, 127653.

DRAFT

ORAL PRESENTATION

Dondurma üretiminde üvez pulpu kullanılmasının dondurmanın ilk damla süresine etkisi

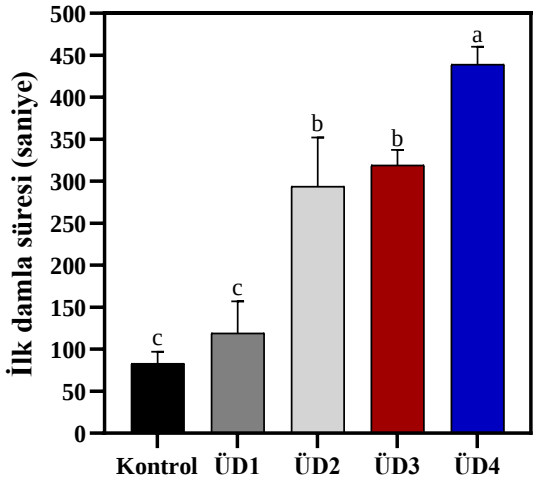
Hamza GÖKTAŞ[1*] (https://orcid.org/0000-0001-9802-9378)

[*1] İstinye Üniversitesi, Meslek Yüksekokulu, Gıda Teknolojisi Programı, İstanbul, Türkiye

*Sorumlu yazar e-mail: hamzagoktas@yandex.com

Özet

Dondurma, süt, şeker, emülgatör ve stabilizatör gibi bileşenlerden oluşan besinsel özellikleri ve serinletici etkisi nedeniyle dünya genelinde hemen her yaş grubu insan tarafından severek tüketilen bir süt ürünüdür. Fonksiyonel gıdalar insanların sağlık durumlarını olumlu yönde etkileyebilecek bileşenler içeren gıdalardır ve son yıllarda fonksiyonel gıdalara olan talep hızla artmaktadır. Bunun yanında dondurma antioksidan bileşikler, probiyotikler ve prebiyotikler gibi fonksiyonel bileşenler açısından oldukça fakirdir ve tüketici talebinin yüksek olması nedeniyle özellikle fenolik bileşikler açısından zengin meyvelerle fonksiyonelleştirilebilir. Üvez meyvesi klorojenik asitler, flavonoller, hidroksisinamik, p-kumarik asit, ferulik asit ve kafeik asit gibi fonksiyonel bileşikler açısından zengindir ve geleneksel olarak reçel, şurup ve likör gibi şekerleme ürünlerinin üretiminde kullanılmaktadır. Ancak üvez meyvesinin sahip olduğu buruk tat nedeniyle bu meyvenin fonksiyonel gıda ürünlerinin üretiminde kullanımına yönelik çalışma mevcut değildir. Bu çalışmada üvez pulpu farklı oranlarda (%2,5 (ÜD1), %5 (ÜD2), %10 (ÜD3) ve %20 (ÜD4)) dondurmaya ilave edilerek fonksiyonel dondurma üretimi hedeflenmiştir ve üvez pulpunun dondurmanın erime süresine (first dripping time-ilk damla süresi) etkisi belirlenmiştir (Şekil 1.). Üvez pulpu içermeyen kontrol dondurma grubunun ilk damla süresi 83,00±14,14 s olarak belirlenmiştir. ÜD1, ÜD2, ÜD3 ve ÜD4 örneklerinin ilk damla süreleri ise sırasıyla 119,00±38,18, 294,00±57,98, 319,00±18,38 ve 439,00±21,21 s olarak tespit edilmiştir. Dondurma üretiminde üvez pulpunun kullanılması dondurma örneklerinin ilk damla süresini arttırmıştır ve artan üvez pulpu konsantrasyonu daha uzun ilk damla süresiyle dolayısıyla daha geç erimeyle sonuçlanmıştır. Bunun nedeni şeker içeriği ve meyve konsantrasyonun artması absorbe edilen su miktarını arttırmıştır ve böylece viskozite arttığı için daha uzun ilk damla süresiyle sonuçlanmıştır.



(Kontrol: Üvez pulpu içermeyen dondurma, ÜD1: %2,5 üvez pulpu içeren dondurma, ÜD2: %5 üvez pulpu içeren dondurma, ÜD3: %10 üvez pulpu içeren dondurma, ÜD4: %20 üvez pulpu içeren dondurma) (Farklı harfler istatistiksel farklılığı göstermektedir (P≤0,05).

Şekil 1. Dondurma örneklerinin ilk damla süreleri

Anahtar Kelimeler: Dondurma, Üvez pulpu, İlk damla süresi

ORAL PRESENTATION

The effect of co-administration of Rosmarinic Acid and Carvacrol on wound tissue nitric oxide and protein carbonyl levels in diabetic rats

Sevim Kömür[1*] (*ORCID: https://orcid.org/* 0000-0002-2835-5351), Kanuni Barbaros Balabanlı[1] (*ORCID: https://orcid.org/* 0000-0002-6670-8904), Fatma Nur Tuğcu Demiröz[2] (*ORCID: https://orcid.org/* 0000-0002-9468-3329), Şule Coşkun Cevher[1] (*ORCID: https://orcid.org/* 0000-0001-6204-2845)

[*1]Gazi University, Science Faculty, Biology Department, Ankara, Turkey
[2]Gazi University, Pharmacy Faculty, Pharmaceutical Technology Department, Ankara, Turkey

*Corresponding author e-mail: sevim.komur@gazi.edu.tr

Abstract

Oxidative stress is closely associated with the development of diabetic wounds. Increased oxidative stress and decreased antioxidant activity cause diabetic wounds to fail to heal. Abnormal production of Nitric oxide (NOx) is closely associated with impaired wound healing and the development of chronic wounds. During wound healing, not only lipids but also proteins are affected by increased oxidative stress. This study aimed to determine the effect of the combination of Rosmarinic Acid (RA) and Carvacrol (CAR), which are known to have natural antioxidant properties, on wound tissue Nitric oxide (NOx) and Protein carbonyls(Pc) levels during diabetic wound healing. 72 male Wistar-albino rats with streptozotocin-induced (STZ) diabetes were divided into five groups: control, untreated group, vehicle-treated group, topical treated group with a combination of RA (10 mg/kg) and CAR (10 mg/kg) and ıntraperitoneal (ı.p.) treated group with a combination of RA (10 mg/kg) and CAR (10 mg/kg). NOx and Pc levels in the wound tissues obtained after the experiment were determined by spectrophotometric methods. When the untreated groups were compared with the treatment groups, a significant decrease was determined in NOx levels ($p<0.05$). When the untreated groups (3 and 7 days) were compared with the treated groups (3 and 7 days), a statistically significant decrease in Pc levels was determined in the treated groups ($p<0.05$). Combination of RA and CAR successfully contributed to wound healing by reducing the NOx and Pc levels in the healing process of diabetic wounds. It has been determined that the strong synergistic effect of these two antioxidants can be a powerful therapeutic agent in reducing or eliminating oxidative stress in diabetic wound healing.

Acknowledgement: This research was supported by Gazi University Scientific Research Projects (BAP), Ankara, Turkey (Project Number: FYL-2022-7703).

Keywords: Diabetic wound healing, Rosmarinic Acid, Carvacrol, Nitric oxide, Protein Carbonyls, Oxidative stress

DRAFT

ORAL PRESENTATION

Investigation of biosorption properties of Pyracanthacoccinea L. for color removal

Nurhan TURAN[1*] (*ORCID: https://orcid.org/0000-0003-2597-914X*),
Sevgi GÜNEŞ DURAK[1] (*ORCID: https://orcid.org/0000-0003-4273-7417)*

[*1]Nevsehir Haci Bektas Veli University, Institute of Science, Environmental Engineering, Nevsehir, Turkey.

*Corresponding author e-mail: turannurhan15@gmail.com

Abstract

One of the factors causing water pollution is color formation in water. Especially in textile wastewater, color is formed due to dyestuffs and when discharged to the receiving environment without proper disposal, it negatively affects the water quality and accordingly the living life in the environment. The use of natural substances in the removal of pollutants in water is quite common. Adsorption mechanism is generally used for removal. Biosorption is the uptake of metal ions from aqueous media by biomass. In this study, pure and dimethylglyoxime modified Pyracanthacoccinea L. plants growing in Nevşehir province were used as adsorbents. The decolorization potential of the modified and unmodified biomass of synthetic dyestuff-containing solution was investigated and different biomass amounts (0.1 g, 0.2 g, 0.4 g, 0.6 g, 0.8 g, 1.0 g) and at different temperature values (5 ^{o}C, 15 ^{o}C, 30 ^{o}C and 45 ^{o}C). It was found that better removal was achieved at lower temperatures and under conditions where the solution was basic. However, the effect of the amount of adsorbent used varied according to the analysis temperature.

Keywords: Adsorption, biosorption, color removal, *Pyracanthacoccinea* L., water pollution.

DRAFT

ORAL PRESENTATION

L-Fenilalanin Esterinde Türeyen Tiyosemikarbazit Türevlerinin Sentezi ve Yapı Aydınlatılması

Kudret AKŞİT* (*ORCID: 0009-0004-8545-901X*), Emine Elçin ORUÇ-EMRE (*ORCID: 0000-0001-6840-9660*), Ayşegül KARAKÜÇÜK İYİDOĞAN (*ORCID: 0000-0002-8088- 6010*)

*Gaziantep Üniversitesi, Fen Edebiyat Fakültesi, Kimya Bölümü, Gaziantep, Türkiye

*Sorumlu yazar e-mail: kudretmertaksit@gmail.com

Özet

Tiyosemikarbazit (NH_2-NH-$CSNH_2$), tiyokarbamik asidin en basit hidrazin türevi olarak bilinmektedir. Çok sayıda biyoaktif heterosiklik bileşiklerin sentezinde çok yönlü bir anahtar ürün olması ve içerdikleri substitüentlere bağlı olarak farklı farmakolojik aktivitelerinden dolayı araştırmacılar arasında geniş bir ilgi odağı olmuştur. Daha önce gerçekleştirilen birçok çalışmada tiyosemikarbazitlerin sentezine yer verilmiş ve kayda değer sonuçlar elde edilmiştir. Bu amaçla başlangıç maddesi olarak kullanılan *L*-fenilalanin etil esterinden hareketle; öncelikle 4-metoksibenzen sülfonil klorür ile reaksiyon sonucunda başlangıç maddesi olan benzensülfonamit elde edildi. Sonra bu bileşiğin ester kısmı uygun çözücü içerisinde % 98'lik hidrazin monohidrat kullanılarak hidrazit türevine dönüştürüldü. Asit hidrazit türevi, asetonitrilli ortamda ticari olarak bulunan sübstitüe izotiyosiyanatlar ile tiyosemikarbazit türevleri sentezlendi. Sentezlenen bileşiklerin saflıkları İTK ve HPLC ile kontrol edildikten sonra yapıları elementel analiz (C, H, N, S), IR, ^{1}H-NMR ve ^{13}C-NMR spektroskopik yöntemler kullanılarak aydınlatılmıştır.

Anahtar Kelimeler: *L*-fenilalanin, Sülfonamit, Tiyosemikarbazit, Sentez

DRAFT

ORAL PRESENTATION

Protective roles of fullerene C_{60} nanoparticle against chromium-induced oxidative damage in *Saccharomyces cerevisiae*

Seda Beyaz[1]* (ORCID: https://orcid.org/ 0000-0003-0436-8112)

[1*]Firat University, Faculty of Science, Department of Biology-Molecular Biology and Genetics Program, Elazig, Turkiye

*Corresponding author: beyazseda23@gmail.com

Abstract

Fullerene C_{60} is used in applications in a variety of fields, including nanotechnology, materials science and medicine. Due to its propensity to encapsulate pharmaceuticals and other compounds, it is also preferred in nanomaterial research and as a potential drug delivery mechanism. In this study, the protective roles of fullerene C_{60} against chromium-induced oxidative damage in *Saccharomyces cerevisiae* (*S. cerevisiae*) were evaluated. The experimental groups are as follows: (i) Control Group; (ii) Chromium (30 mM); (iii) Fullerene C_{60} (30 mM); (iv) Fullerene C_{60} (30 mM) + Chromium (30 mM). *S. cerevisiae* cultures were grown at 30 °C for 1, 3, 5 and 24 hours.
Malondialdehyde analyzes, catalase activities, glutathione levels, total protein and cell growth were determined by spectrophotometer. According to the results, it was determined that total protein synthesis, cell growth, catalase activities and glutathione levels increased, malondialdehyde levels decreased in the Fullerene C_{60} + Chromium applied groups compared to the chromium applied groups. These results showed that fullerene C_{60}, which exhibits many biological activities, reduces oxidative damage by providing protection in *S. cerevisiae* culture.

Keywords: Chromium, Fullerene C_{60}, Nanoparticle.

DRAFT

ORAL PRESENTATION

Presence of aquatic insects in freshwater areas of the Mediterranean region (Antalya and Mersin provinces) at the family level and their distribution on the map

Mehmet Bektaş[1*] (ORCID: https://orcid.org/0000-0002-7420-6883),
Yahya Tepe[2] (ORCID: https://orcid.org/0000-0002-4589-9860)

[*1]Ataturk University, Hınıs Vocational Collage, Erzurum, Turkey.
[2] Ataturk University, Science Faculty, Department of Biology, Erzurum, Turkey.

*Corresponding author e-mail: mehmet.bektas@atauni.edu.tr

Abstract

In recent years, faunistic research have been carried out on aquatic insect families. However, it is necessary to detect and monitor the animal diversity of these aquatic areas, which are gradually shrinking as a result of global warming, the expansion of agricultural areas and the effects of pesticides used in these agricultural regions. In this framework, aquatic Coleoptera and aquatic Hemiptera species collected in the Mediterranean coastal regions from the freshwater areas of Antalya and Mersin provinces in October, May and July between 2022 and 2023 were evaluated. 2784 insects were collected in the study area: These are; 99 individuals belong to the order Hemiptera (Corixiidae, Hydrometridae, Pleidae, Gerridae and Notonectidae) and 2685 individuals belong to the order Coleoptera (Dryopidae, Dytiscidae, Helophoridae, Heteroceridae, Hydranidae, Hydrophilidae, Hydrochidae, Haliplidae, Noteridae and Spercheidae). Those collected have yet only been determined at the family level. Hydrophiliade (Coleoptera) and Corixiidae (Hemiptera) families were caught in large numbers individually as ordo, but Spercheidae (Coleoptera) and Pleidae (Hemiptera) families were caught in almost non-existent numbers compared to previous localities. Additionally, insect distributions are shown on maps; greenhouse farming activities, which are known to involve the use of pesticides, and rivers, streams, streams, etc. It has been noticed that the number of insect samples collected near the coast and in the lower regions of greenhouse gardens is very low as a result of the use of water resources such as water resources for agricultural purposes. With this study, aquatic insects were monitored on a map for the first time in the research area. In conclusion, Turkey's aquatic insects in order to fully detect its existence, ecological and faunistic studies are increasing to cover all provinces. Moreover, it must continue.

Keywords: Aquatic insects, Mediterranean region, Pesticides, Turkey.

Acknowledgements
This study was supported by Ataturk University Scientific Research Unit (Project No: FCD-2022-8297, BAP) with financial support. We would like to thank Management of Atatürk University and BAP unit.

DRAFT

ORAL PRESENTATION

Evaluation of in vitro anticancer activity of Silver Nanoparticles Biosynthesized from *Cistus salviifolius* L. and *Ferula communis* L. in MCF-7 and HT-29 cells

Funda ULUSU[*1] (*ORCID: 0000-0002-0321-2602.*), Yakup ULUSU[2] (*ORCID: 0000-0002-8755-2822*)

[*1]Karamanoglu Mehmetbey University, Vocational School of Technical Sciences, Department of Crop and Animal Production, Karaman, Turkey.
[2]Karamanoglu Mehmetbey University, Faculty of Engineering, Department of Bioengineering, Karaman, Turkey.
[2]University, Faculty, Department, City, Country.

*fulusu@kmu.edu.tr

Abstract

In this investigation, we employed a microwave-assisted green synthesis methodology utilizing extracts derived from *Cistus salviifolius* L. and *Ferula communis* L. to synthesize silver nanoparticles (AgNPs). Subsequently, we conducted a comprehensive characterization of the biosynthesized AgNPs, denoted as *Cs*-AgNPs (obtained from *C. salviifolius*) and *Fc*-AgNPs (obtained from *F. communis*), employing various analytical techniques, including UV-Vis spectroscopy, Fourier-transform infrared (FT-IR) spectroscopy, X-ray diffraction (XRD) analysis, scanning electron microscopy (SEM), and transmission electron microscopy (TEM). The AgNPs exhibited a spherical morphology, with an average particle size measuring below 11 nm. Two different cell lines, breast adenocarcinoma (MCF-7) and colon carcinoma (HT-29), were used to evaluate their anticancer potential. *Cs*-AgNPs demonstrated elevated cytotoxic efficacy against the MCF-7 and HT-29 cell lines in contrast to *Fc*-AgNPs. Particularly noteworthy, *Cs*-AgNPs displayed the most pronounced cellular inhibition on the HT-29 cell line, with an observed IC_{50} value of 57.10 µg/mL. Our findings conclusively demonstrate that biosynthesized *Cs*-AgNPs and *Fc*-AgNPs have significant anticancer activities.

Keywords: *Cistus salviifolius*, *Ferula communis*, Silver Nanoparticles, Anticancer activity.

DRAFT

ORAL PRESENTATION

Investigation of the usability of *T. molitor* chitins for the removal of textile industry dyestuffs

Ebru Ceren FİDAN[1*] (*https://orcid.org/0000-0002-6121-4585*), Belma NURAL YAMAN[2] (*ORCID: https://orcid.org/0000-0003-2576-1300)*

[*1]Eskişehir Osmangazi University, Faculty of Science, Department of Biology, Eskişehir, Turkiye.
[2] Eskişehir Osmangazi University, Faculty of Engineering and Architecture, Department of Biomedical Engineering, Eskişehir, Turkiye.

*Corresponding author e-mail:ebruceren@ogu.edu.tr

Abstract

T. molitor L. is one of the most researched insects with its potential as a model organism, as well as being cultivated and used as animal feed and edible. Due to its rapid reproduction compared to other insect groups, industrial production is carried out in many parts of the world. A considerable amount of chitin residue is obtained during the molting process between larval stages. The aim of the study is highlighted the importance of the *Tenebrio molitor* chitin usage on the environmental biotechnology. In the scope of this study, four textile dyes (RR3:1, RO13, RS8, RB13) were treated by insect chitins. The experimental setup was designed in 50 ml working volume with pH 2 dye solution. 50 mg (1 g/L) biomass were used, and incubation was completed at 25 °C by 150 rpm for 30 min. The experiments were practiced in duplicate. Biosorption % and biosorption capacity was calculated. According to biosorption yield, *T. molitor* chitin was found to be effective for removal of RB8 and was used for the following optimization experiments. As a result of the optimization study carried out using *Tenebrio molitor* chitin, the chitin concentration was found to be as 1g/L, the dye concentration was found to be 50 ppm and the incubation time was 30 minutes. To sum up, *T. molitor* chitin can be used to treatment of textile dye wastewater including especially Reactive Black 8.

Keywords: biosorption, chitin, dye, *Tenebrio molitor*, wastewater

DRAFT

ORAL PRESENTATION

Impacts of zinc fertilization on *Ocimum basilicum* L. under water stress: Alterations in essential oil composition

Funda ULUSU* (*ORCID: 0000-0002-0321-2602*)

*Karamanoglu Mehmetbey University, Vocational School of Technical Sciences, Department of Crop and Animal Production, Karaman, Turkey.

*fulusu@kmu.edu.tr

Abstract

Abiotic water stress is a hindrance to plant growth and productivity, with a significant impact on both yield and quality traits. In the face of ongoing global climate changes, the imperative to reduce the detrimental effects of water deficit on plant systems has increased exponentially. This research focuses on increasing the resilience of *Ocimum basilicum* L., a species particularly sensitive to the challenges of water stress. For this purpose, we studied the effects of zinc fertilizer supplementation applied at varying rates (2.5, 5 and 10 mg/kg) on *O. basilicum* grown in silty sandy soil and exposed to water stress conditions. Changes in sweet basil were investigated by assessments including mineral uptake dynamics as well as essential oil composition. The findings underline the negative consequences of water stress on the profile of the studied mineral uptake secondary metabolites. In the spectrum of essential oil constituents, estragole emerged as the dominant compound and recorded the highest concentration (69.37%) in the treatment group receiving 10 mg/kg zinc fertilizer. In contrast, the lowest estragole concentration (66.14%) appeared in the group under water stress without fertilizer supplementation. The application of zinc fertilizer, particularly at concentrations of 5 and 10 mg/kg, demonstrated significant ameliorative effects on the detrimental impacts induced by water stress. Furthermore, zinc exhibited multifaceted mechanisms of action, influencing the uptake dynamics of other essential nutrients from the soil.

Keywords: *Ocimum basilicum*, Water stress, Zinc fertilization, Essential oil.

DRAFT

ORAL PRESENTATION

Normal vücut ağırlığındaki kadınlarda şeker tüketim sıklığı ile yaşam kalitesi arasındaki ilişkinin değerlendirilmesi

Özlem Baran[1*] (*ORCID: https:// 0000-0003-3868-4850.org*), Muhammed Enes Kartal[1] (*ORCID: https:// 0000-0001-9015-1491.org*), Birgül Dağ[1] (*ORCID: https:// 0000-0003-4631-3910.org*), Cemaliye Süt Kurt[1] (*ORCID: https:// 0000-0001-9016-2959.org*), Ayhan Dağ[1] (*ORCID: https:// 0000-0002-8291-3414.org*), Sevinç Yücecan[1] (*ORCID: https:// 0000-0003-4751-0924.org*)

[*1]Lokman Hekim Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Ankara, Türkiye

*Sorumlu yazar e-mail: ozlem.baran@lokmanhekim.edu.tr

Özet

Beslenme bireyin büyüme ve gelişmesi, yaşam boyu sağlığın korunması ve yaşam kalitesinin arttırılması için ihtiyaç duyulan besin ögelerinin alınıp vücutta kullanılmasıdır. Bireyin yaşamını sürdürebilmesi için yeterli ve dengeli beslenme önemlidir. Vücudun ihtiyacı olan besin ögelerinin ve yeterli enerjinin alınması sağlık açısından gereklidir. Şeker ve şeker içeren yiyecekler enerji yoğunluğu yüksek besinlerdir. Tüketim sıklık ve miktarlarının yüksek olması ideal vücut ağırlığının sağlanması ve korunmasında olumsuz etkiler ortaya çıkarır. Obezitenin varlığı yaşam kalitesini olumsuz etkileyebilmektedir. Bu çalışmanın amacı normal ağırlıktaki kadınlarda şeker tüketim sıklığı ile yaşam kalitesi arasındaki ilişkiyi değerlendirmektir. Bu çalışma Haziran – Temmuz 2022 tarihleri arasında, 18-65 yaş arası, 489 normal vücut ağırlığına sahip kadınlar üzerinde yürütülmüştür. Katılımcılara yüz yüze anket uygulaması yapılmıştır. Anket 3 bölümden oluşmaktadır. Birinci bölümde demografik bilgiler, ikinci bölümde beslenme alışkanlıkları ile ilgili bilgiler ve üçüncü bölümde ise SF-12 Yaşam Kalitesi Ölçeği yer almaktadır. Katılımcıların antropometrik ölçümleri araştırmacılar tarafından yapılmıştır. Çalışmaya katılan toplam 489 kadının yaş ortalaması 32.75±9.76'dır. Kadınların %66.2'si (n=324) evli ve yine %66.2'si (n=324) çocuk sahibidir. Kadınların %40.3'ü (n=197) üniversite mezunudur ve %49.1'i (n=240) ev hanımıdır. Sigara içenlerin oranı ise %23.1 (n=113) olarak bulunmuştur. Şeker tüketim sıklıklarına bakıldığında hiç tüketmeyenler, her gün, haftada 1-3 kez, haftada 4-6 kez ve ayda 1-3 kez tüketenlerin oranı sırasıyla %9.0, %49.9, %27.8, %4.5 ve %8.8'dir. Yaşam kalitesi mental ve fiziksel özet puanları en yüksek hiç şeker tüketmeyenlerde görülmüştür (sırasıyla 41.9±12.2 ve 43.9±0.26). Şeker tüketim sıklığı ile yaşam kalitesi arasında anlamlı bir ilişki tespit edilmemiştir ($p<0.05$). Normal vücut ağırlı kadınlarda şeker tüketim sıklığı ile yaşam kalitesi arasında anlamlı bir ilişki tespit edilmemiştir. Yaşam kalitesinin arttırılması için ideal vücut ağırlığının korunması önemlidir. Bu noktada şeker tüketim sıklığı ve miktarına gösterilen dikkat önemlidir.

Anahtar Kelimeler: Sağlık beslenme, şeker tüketimi, yaşam kalitesi

DRAFT

ORAL PRESENTATION

Yetişkin kadınlarda covid-19 geçirme durumunun beslenme davranışı ile ilişkisinin değerlendirilmesi

Muhammed Enes Kartal[1*] (*ORCID: https:// 0000-0001-9015-1491.org*), Özlem Baran[1] (*ORCID: https:// 0000-0003-3868-4850.org*), Birgül Dağ[1] (*ORCID: https:// 0000-0003-4631-3910.org*), Cemaliye Süt Kurt[1] (*ORCID: https:// 0000-0001-9016-2959.org*), Ayhan Dağ[1] (*ORCID: https:// 0000-0002-8291-3414.org*), Sevinç Yücecan[1] (*ORCID: https:// 0000-0003-4751-0924.org*)

[*1]Lokman Hekim Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Ankara, Türkiye

*Sorumlu yazar e-mail: muhammed.kartal@lokmanhekim.edu.tr

Özet

Beslenme, açlık-tokluktan etkilendiği gibi psikolojik etmenlerden, sosyal çevreden, kültürel alışkanlıklardan veya hastalık durumlarından da etkilenebilmektedir. Dünya Sağlık Örgütü'nün pandemi olarak kabul ettiği Covid-19, beslenme davranışlarını etkileyebilen önemli bir viral hastalıktır. Covid-19 sebebiyle evden çalışma durumu, hastalığın ortaya çıkardığı kaygı, değişen uyku düzeni gibi sebepler beslenme davranışlarını etkileyebilmektedir. Yeterli ve dengeli beslenme bağışıklığı etkileyebilen ve sistemik inflamasyonu sınırlayabilen önemli bir faktördür. Dolayısıyla yeterli ve dengeli beslenmeyi sağlayabilecek beslenme davranışlarının benimsenmesi önemlidir. Bu çalışmanın amacı Covid-19 geçirme durumu ile beslenme davranışı arasındaki ilişkinin değerlendirilmesidir. Bu çalışma Ankara'nın Mamak İlçesi'nde Haziran – Temmuz 2022 tarihleri arasında, 18-67 yaşları arasında olan 1205 kadın üzerinde yürütülmüştür. Katılımcıların, yüz yüze anket uygulaması ile, demografik özellikleri, beslenme alışkanlıkları ve Covid-19 geçirme durumları sorgulanmıştır. Ayrıca katılımcıların beslenme davranışları ile ilgili bilgi edinebilmek adına üç faktörlü yeme ölçeği uygulanmıştır. Çalışmaya katılan toplam 1205 kadının yaş ortalaması 38.0±11.4 olup, %77.5'i (n=934) evli ve %36.1'i (n=435) lise mezunudur. Katılımcıların %64.3'ü (n=775) ev hanımıdır ve %54.3'ünün (n=654) geliri giderine eşittir. Sigara içme durumlarına bakıldığında %78.1'inin (n=941) sigara içmediği belirlenmiştir. Katılımcıların %49.8'i (n=600) Covid-19 geçirmiştir. Kontrolsüz yeme, bilişsel kısıtlama ve duygusal yeme puanları Covid-19 geçirenlerde sırasıyla 37.3±25.5, 47.1±31.8, 36.1±38.7 olarak bulunmuştur. Covid-19 geçirenlerde, Covid-19 geçirmeyenlere göre kontrolsüz ve duygusal yeme puanları daha yüksek tespit edilmiştir ve aradaki ilişki istatistiksel açıdan anlamlıdır ($p<0.05$). Covid-19 geçirme durumu ile kontrolsüz ve duygusal yeme puanları arasında anlamlı bir ilişki tespit edilmiştir. Ciddi bir pandemiye sebep olan Covid-19 pek çok yaşam şekli alışkanlığını etkilediği gibi beslenme davranışlarını da etkilemiştir. Değişen beslenme alışkanlıklarının sağlığı koruyucu ve iyileştirici yönde olmasını sağlamak önem taşımaktadır.

Anahtar Kelimeler: Beslenme davranışı, Covid-19, sağlıklı beslenme

DRAFT

ORAL PRESENTATION

Development of waste biomass-based activated carbon-coated carbon fibers for potential applications in multifunctional structural supercapacitors

Şevval Nur Karademir[1] (*ORCID: https://orcid.org/ 0009-0008-7958-3654*), İ. Işıl Gürten İnal[1*] (*ORCID: https://orcid.org/0000-0001-5144-2800*)

[1]Ankara University, Faculty of Engineering, Department of Chemical Engineering, Ankara, Turkey.

*Corresponding author e-mail: igurten@eng.ankara.edu.tr

Abstract

In this study, the conventional carbon fiber fabrics (CFF) were modified by coating them with a waste biomass-derived porous carbon material. The primary objective of this modification was to enhance the poor energy storage performance of the CCFs to create potential structural supercapacitor electrode materials. The process involved first; the preparation of activated carbon from red pepper industrial wastes which were collected from a red pepper paste factory. The waste biomass was activated with K_2CO_3 in 1:1 mass ratio, followed by heat treatment at 800ºC under an inert atmosphere. The resultant solid was washed until the neutral pH was reached, and the activated carbon (AC) was obtained. The surface characterization of AC was conducted utilizing N_2 adsorption-desorption, FTIR, and SEM techniques. The AC sample was determined to have a highly microporous structure with a BET 1120.96 m²/g surface area. The electrochemical performance of AC was tested as symmetrical coin-cell type supercapacitor electrodes by cyclic voltammetry (CV), galvanostatic charge/discharge (GCD), and electrochemical impedance spectroscopy (EIS) analysis methods using 6 M KOH electrolyte. To investigate the energy storage performance of the modified CFFs, the CFFs in 1x1.5 cm were coated with AC by spray coating at different AC mass ratios of 2%, 5%, 10%, and 15%. Finally, the supercapacitor cells were assembled using the coated CFF electrodes and tested under the same conditions as the previous electrochemical measurements. As a result, the original CFFs, which exhibited poor energy storage performance, have shown a significant increase in energy storage capability by being coated with a highly porous low-cost activated carbon produced from waste biomass. These modified CFFs are promising candidates as reinforcement/electrodes for multifunctional structural energy storage devices.

Keywords: Carbon fiber fabric, Activated carbon, Structural supercapacitors

DRAFT

ORAL PRESENTATION

Synthesis and Antimicrobial Activities of Some Phenyl-Substituted Azaheterocyclic Compounds

Huseyin Istanbullu[1*] (*https://orcid.org/0000-0002-0102-4181),*
Ismail Ozturk[2] (*https://orcid.org/0000-0002-2669-3090*)

[*1] Izmir Katip Celebi University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, Izmir, Türkiye

[2] Izmir Katip Celebi University, Faculty of Pharmacy, Department of Microbiology, Izmir, Türkiye

*Corresponding author e-mail: huseyin.istanbullu@ikc.edu.tr

Abstract

Antimicrobial resistance (AMR) is defined by the WHO as a threat to the effective prevention and treatment of an increasing number of infections caused by bacteria, parasites, viruses and fungi[1]. AMR is a competition between natural selection and human intelligence, requiring not only the use of fewer antimicrobials but also the rapid development of new and effective antimicrobials. A number of azaheterocyclic compounds exhibit a wide range of biological activities, including the various antimicrobial agents that are available in literature[2]. Considering this, we designed novel o-chlorophenyl substituted fused triazoles as new antimicrobials. Antimicrobial activity of the compounds was tested against standard strains of Gram-positive (S. aureus), Gram-negative (E. coli) and fungal (C. albicans) bacteria. Inhibition zone diameters and minimum inhibitory concentration (MIC) values were determined by disc diffusion and microdilution methods[3,4]. Changes in biofilm production levels were determined by the spectrophotometric microplate method using crystal violet staining[5]. All experiments were performed in triplicate and statistical analyses (t-test) were performed. According to the results, the MIC values of the compounds were found to be 32-2048 µg/mL against S. aureus, E. coli and C. albicans. Biofilm production levels of S. aureus, E. coli and C. albicans were inhibited in the presence of the compounds in the range of 8.8% - 68.5%. The results obtained showed that the compounds have promising activities. The pharmacokinetic properties of the compounds were evaluated using SwissADME. Targeting virulence factors, including biofilm structures, may play an important role in the control of infections. In addition to antimicrobial activity, microscopic scanning and gene expression experiments are planned to investigate the effects of the compounds against different strains and clinical isolates.

Keywords: Antimicrobial resistance, anti-biofilm activity, fused triazoles, chlorophenyl, ADME

References:

[1]https://www.who.int/health-topics/antimicrobial-resistance
[2]Abu-Hashem et al., (2017), Med Chem Res, 26, 120-130. Doi: 10.1007/s00044-016-1733-5
[3]Kotmakçı, M., Öztürk, İ., Kantarcı, G., Ermertcan, Ş. (2015), Lat Am J Pharm. 34(3), 529–536.
[4]Başpınar, Y., Kotmakçı, M., Öztürk, İ. (2018), Celal Bayar University Journal of Science. 14(2), 223-228, doi: 10.18466/cbayarfbe.403152
[5]Öztürk İ, Tekintaş Y, Temel A, Ermertcan Ş, Kurutepe S, Hoşgör Limoncu M. (2020), J Res Pharm. 24(6), 821-832. https://doi.org/10.35333/jrp.2020.241

DRAFT

ORAL PRESENTATION

Assessing pesticide biodegradability through predictive tools and databases

Gökçin Gül[1*] (https://orcid.org/0000-0001-9886-9676),
Ferhan Çeçen[1] (https://orcid.org/0000-0002-5353-1441)

[*1]Boğaziçi University, Institute of Environmental Sciences, İstanbul, Turkey.

*Corresponding author e-mail: gokcin.gul@boun.edu.tr

Abstract

This study presents a comprehensive analysis of the biodegradation properties of five selected pesticides, namely carbofuran, chlorpyrifos, cyprodinil, propanil, and malathion, using various predictive tools and databases. The aim of the study is to identify potential biodegradation pathways and metabolites for these pesticides, thereby providing valuable insights into their environmental fate and potential impact. The results reveal significant variations in biodegradability and persistence among the selected pesticides, with some exhibiting rapid biodegradation, while others are predicted to exhibit minimal biodegradation. The study provides a reliable methodology for predicting the biodegradation potential of pesticides, which can be useful for environmental risk assessments and management.

Keywords: Biodegradation, EPI Suite, EAWAG-BBD PPS, MetaCyc, Pesticides,

DRAFT

ORAL PRESENTATION

Kökeni bilinen belirli bir çeşit buğdaydan elde edilen kepeğin mikrobiyal biyoproses yoluyla modifikasyonu ve modifiye kepek kullanılarak ekmek üretimi

Esra Şık[1]* (*ORCID: https://orcid.org/0000-0002-6771-146X*),
Görkem Özülkü[2] (*ORCID: https://orcid.org/0000-0003-0495-5667*)

[*1]Haliç Üniversitesi, Meslek Yüksekokulu, Otel Lokanta ve İkram Hizmetleri Bölümü, İstanbul, Türkiye
[2]Yıldız Teknik Üniversitesi, Kimya Metalürji Fakültesi, Gıda Mühendisliği Bölümü, İstanbul, Türkiye

*Sorumlu yazar e-mail: esrask@halic.edu.tr

Özet

Çalışmada YTÜ Gıda Mühendisliği Bölümü koleksiyonunda bulunan laktik asit bakterileri (*Lactobacillus plantarum* LABE29, *Lactobacillus brevis* LABE32) ve maya (*Saccharomyces cerevisiae* TGM55) kullanılarak buğday kepeğine (kökeni bilinen tek bir çeşit, Tekirdağ) mikrobiyal biyoproses ile modifikasyon işlemi uygulanmıştır. Modifiye kepeklerde toplam diyet lifi (TDF) ve toplam fenolik içerik (TPC) – antioksidan aktivite (AA) belirlenmiş; viskozite, hidratasyon özellikleri, fitik asit (PA) gibi teknofiziksel özellikler tespit edilmiştir. Modifiye kepekler kullanılarak baz un ile paçal edilen unlarda (ağırlıkça %15 kepek) farinograf özellikleri incelenmiş, ekmek üretim parametrelerine karar verilmiştir. Modifiye kepeklerden ekmek üretimi gerçekleştirilmiş; ekmeklere tekstür, spesifik hacim, nem, renk, TDF, TPC, AA ve PA analizleri uygulanmıştır. Modifikasyon iki farklı uygulama şeklinde gerçekleştirilmiştir: *S. cerevisiae* ve *L. plantarum* kombinasyonu M29, *S. cerevisiae* ve *L. brevis* kombinasyonu M32 olarak adlandırılmıştır. M32, M29'a göre TDF'de işlem görmemiş kepeğe göre ağırlıkça %3,4 artış sağlamıştır. M29 ve M32 serbest fenolik içeriği sırasıyla %25,2 ve %26,6 oranında artırmıştır. Bağlı fenolik içerik ise M29 ile %33,4 oranında artarken, M32'de artış %41,3 olmuştur. M29, örneklerin AA değerini işlem görmemiş kepeğe göre %181 oranında artırmış, bu artış M32'de %203 şeklinde ortaya çıkmıştır. Buğday kepeği ekstraktlarının viskozite değeri üzerinde M32 daha etkili bir artış yaratmıştır. Toplam su tutma kapasitesi üzerinde hem M29 hem M32 literatürü destekler şekilde artış sağlamıştır. M29, PA üzerinde %80.7 azalma sağlamış, bu azalma M32'de %72.5 olarak ortaya çıkmıştır. Modifiye kepeklerden elde edilen ekmeklerin spesifik hacim değerleri kontrol örneğine göre düşük tayin edilmiştir. Ekmekler renk özellikleri açısından farklı bulunmamıştır. Her iki modifikasyon uygulamasında da ekmeklerde çözünür diyet lif içeriğinin bir miktar arttığı, suda çözünmeyen diyet lif miktarının ise azaldığı tespit edilmiştir. Buğday kepeği analiz sonuçları ile paralel olarak ekmeklerin modifikasyonla toplam TPC değeri de artmış, M32 kontrol ekmeğine kıyasla TPC değerini %19.3 oranında artırmıştır. Modifikasyon uygulamaları ekmek tekstürel özelliklerinde anlamlı bir farklılık yaratmamıştır ancak nihai ürünün PA içeriğinde önemli bir azalmaya katkı sunmuştur.

Anahtar Kelimeler: Buğday kepeği, mikrobiyal modifikasyon, besinsel lif, toplam fenolik madde, ekmek

DRAFT

ORAL PRESENTATION

Removal of Crystal Violet using sawdust adsorbent prepared from wood wastes: isotherm, kinetic and thermodynamic studies

Ali Rıza Kul[1] (*ORCID: https://orcid.org/0000-0001-9331-775X),*
Adnan Aldemir[2] (*ORCID: https://orcid.org/0000-0001-9884-0961)*

[1]Van Yüzüncü Yıl University, Faculty of Science, Chemistry Department, Van, 65080, Turkey.
[2]Van Yüzüncü Yıl University, Faculty of Engineering, Mechanical Engineering Department, Van, 65080, Turkey.

*Corresponding author e-mail: adnanaldemir@yyu.edu.tr

Abstract

In this study, sawdust adsorbent prepared from fig wood which is grow Adana/Turkey was used for removal of crystal violet (CV) dye from aqueous solutions by batch adsorption process. Effects of initial dye concentration, solution temperature and contact time on adsorption were studied. It was seen that the removal of crystal violet (CV) increased with increasing initial dye concentration (from 10 to 40 mg/L), solution temperatures (298 K, 308 K, 318 K) and contact time (start to 200 min). The initial dye concentration rises from 10 to 40 mg/L, removal percentage on wood sawdust adsorbent increases from 63.30 to 68.80 (%), 73.40 to 78.60 (%) and 82.50 to 83.50 (%) for 298 K, 308 K and 318 K, respectively. Adsorption isotherms were analyzed by Langmuir, Freundlich and Temkin models at different temperatures, and Freundlich model shows more suitable results for removal of crystal violet. Adsorption kinetics were analyzed by pseudo first order (PFO), pseudo second order (PSO) and intraparticle diffusion (IPD) models for different temperatures and initial dye concentrations. Kinetic model results were best fit the PSO model which kinetic constants were higher than PFO and IPD models. Moreover, the thermodynamics of adsorption process were also studied. It was found that the values of standard free energy (ΔG°) were negative and standard enthalpy (ΔH°) and entropy (ΔS°) values were found to be positive for this removal process. Thermodynamic results were shown that removal process is physisorption and these values suggest feasibility and spontaneous nature of adsorption. The results of this study showed that prepared sawdust adsorbent from wood waste could be employed as effective and low-cost materials for the removal of dyes from aqueous solution.

Keywords: Adsorption, crystal violet, wood sawdust adsorbent, isotherm, kinetic, thermodynamic

DRAFT

ORAL PRESENTATION

Prediction of current and future distributions of *Chalcophora detrita* (Coleoptera: Buprestidae) under climate change scenarios

Arif Duyar[1*] (https://orcid.org/*0009-0002-3931-4448*), Muhammed Arif Demir[1] (ORCID: https://orcid.org/0000-0001-7141-0814, Mahmut Kabalak[2,3] (ORCID: https://orcid.org/0000-0001-6073-2551)

[1] Hacettepe University, Graduate School of Science and Engineering, Ankara, Türkiye.
[2]Hacettepe University, Faculty of Science, Biology Department, Ankara, Türkiye.
[2]Hacettepe University Biosphere Advanced Biodiversity Research and Application Center, Beytepe, Ankara, Türkiye

*Corresponding author e-mail: arifduyar@hacettepe.edu.tr

Abstract

The consequences of climate change resulting from anthropogenic activities cause different effects in different ecosystems, and the severity of these effects is predicted to increase in the near future. The number of studies for the responses of forest ecosystems to these changes is increasing day by day. However, studies on how saproxylic organisms will experience the consequences of climate change, which is one of the crucial factors for the healthy functioning of these sensitive ecosystems, are still insufficient. In our study, we carried out ecological niche modeling of *Chalcophora detrita* (Klug, 1829), both as a saproxylic species and as an organism of economic importance, under climate change scenarios. Our findings indicate that in two different scenarios (SSP2-4.5, relatively optimistic and SSP5-8.5, most pessimistic) suitable habitats for this species will be decreased gradually in future climate change scenarios. This study provides a theoretical framework of reference for habitat conservation and improvement of management plans on species belonging to the *Chalcophora* genus and the other saproxylic beetles.

Keywords: *Chalcophora detrita*, Buprestidae, Saproxylic beetles, Climate change, Ecological niche model

DRAFT

ORAL PRESENTATION

The role of apocynin against the effects of chloroquine-enhanced radiotherapy on healthy tissues

Ayca Sezen Us [1*] (ORCID: https://orcid.org/0000-0001-7102-848X), Eda Dagsuyu [2] (ORCID: ORCID: https://orcid.org/0000-0003-0395-1058), Huseyin Us [1] (ORCID: https://orcid.org/0000-0002-7421-3821), Melis Coremen [3] (ORCID: https://orcid.org/0000-0002-5054-2604), Omur Karabulut Bulan [3] (ORCID: https://orcid.org/0000-0002-6591-7317), Refiye Yanardag [2] (ORCID: https://orcid.org/0000-0003-4185-4363)

[*1]Istanbul University, Institute of Science, Biology Section, Istanbul, Turkey.
[2]Istanbul University-Cerrahpaşa, Faculty of Engineering, Department of Chemistry, Istanbul, Turkey.
[3]Istanbul University, Faculty of Science, Department of Biology, Istanbul, Turkey.

*Corresponding author e-mail: sezenayca@gmail.com

Abstract

One of the reasons that reduces the effectiveness of radiotherapy is that radiation can increase the survival rate of cancer cells by stimulating autophagy in various types of cancer. Chloroquine (CQ), which has been used in the treatment of malaria for many years, has been shown to inhibit autophagy in recent years. Apocynin (APO) is a substance known as an effective NADPH oxidase inhibitor with antioxidant and antiapoptotic properties. In our study, it was aimed to determine how the use of CQ together with radiotherapy will affect healthy tissues and to reveal whether APO shows protective effects with this combination. Randomly selected animals were divided into eight groups: Control, radiation (RAD), CQ, APO, RAD+CQ, RAD+APO, RAD+CQ+APO and CQ+APO, respectively. The first and second groups were injected saline, the third and fifth groups were injected 50 mg/kg CQ, the fourth and sixth groups were injected 20 mg/kg APO, the seventh and eighth groups were injected with CQ and APO. All injections were administered intraperitoneally (i.p.) five times every 24 hours for 5 days. One hour after the fifth injection, 8 Gray (Gy) whole body irradiation was applied to the radiation groups. Small intestinal tissues were obtained from dissected rats one day after radiation administration. As a result of our study, radiation causes significant histological damage, causes a significant increase in oxidative damage parameters, weakens the antioxidant system, has antiproliferative, inflammatory and apoptotic effects, and also stimulates autophagy are determined. CQ given together with RAD inhibits autophagy, increases oxidative stress, weakens antioxidant capacity, has antiproliferative and anti-inflammatory effects, and does not cause a significant change in apoptosis alone are observed. The fact that APO significantly reverses these changes caused by RAD and CQ shows its protective effect. We anticipate that our results may shed light on clinical applications.

Keywords: Gamma Radiation, Chloroquine, Apocynin, Autophagy, Intestine.

DRAFT

ORAL PRESENTATION

Niche Partitioning between Two Sympatric Lizards in Mediterranean Biodiversity Hotspot

Mehmet Kürşat Şahin[1*] (*ORCID: https://orcid.org/0000-0003-0834-5081*),
Muammer Kurnaz[2] (*ORCID: https://orcid.org/0000-0002-0498-0208*)

[*1]Hacettepe University, Faculty of Science, Department of Biology, Ankara, Türkiye.
[2] Gümüşhane University, Kelkit Sema Doğan Vocational School of Health Services, Department of Medical Services and Techniques, Gümüşhane, Türkiye.

*Corresponding author e-mail: yasambilimci.kursat@gmail.com

Abstract

Competition for resources between sympatric species can result in diminished fitness. Resource partitioning promotes the minimizing of competitive pressures, thereby supporting the coexistence of a diversity of organisms. In this study, we aimed to examine the idea that Danford's Lizard (*Anatololacerta danfordi*) and Golden Grass Mabuya (*Heremites auratus*), which coexist in the northern slopes of Mediterranean Biodiversity Hotspot, Anatolia exhibit discernible ecological niches as a means of reducing interspecific competition. In order to evaluate the hypothesis of resource partitioning, we conducted a comparative analysis of the activity times, perch microhabitat parameters, and thermal dynamics under investigation. Despite the absence of discernible disparities in the timing of morning activity between the two species, distinct variations were seen in their utilization of structural perch microhabitats and thermal preferences. Golden Grass Mabuya exhibited a preference for elevated and narrower perches that offered a greater degree of canopy coverage compared to Danford's Lizard. Additionally, it was observed that male Golden Grass Mabuya tended to occupy higher perches in comparison to their female counterparts. Moreover, the body temperature of *A. danfordi* was found to be higher than that of *H. auratus*, and solar radiation was identified as a key parameter affecting the thermal requirements of both species. The utilization of distinct microhabitats by perch and thermal differentiation species may potentially mitigate interspecific competition, hence facilitating coexistence between these sympatric species.

Keywords: competition, microhabitat, thermal biology, activity, reptile, Anatolia

DRAFT

ORAL PRESENTATION

Detection and characterization of thermophilic bacilli associated with pasteurized milk in markets and determination of their biofilm production properties

Basar Karaca[1*] (*ORCID: https://orcid.org/0000-0001-6943-8965*)

[*1]Ankara University, Faculty of Science, Biology, Ankara, Turkey.

*Corresponding author e-mail: karaca@ankara.edu.tr

Abstract

Thermophilic bacilli are an important group that pose a problem as contaminants in the dairy industry. In this study, thermophilic bacilli were isolated from pasteurized milk sold in markets. The isolates were first genotyped, then their biofilm production characteristics were defined and the risks they may cause in the dairy industry were assessed. In this context, the biofilm morphotypes of thermophilic isolates identified in Congo red media and their pellicle formations were studied to determine their ability to form biofilms at the liquid-air interface. Subsequently, the biofilm production of the isolates on polystyrene and stainless steel surfaces was determined. In addition, the ability of the isolates to produce biofilm on abiotic surfaces commonly used as surfaces for product processing in the dairy industry was tested. While one of the isolates was identified as *Aeribacillus pallidus*, all other isolates were identified as *Bacillus licheniformis*. Based on their biofilm morphotypes and pellicle formations, it could be seen that all isolates were strong biofilm producers. All isolates were able to produce biofilms on both polystyrene and stainless steel surfaces in a wide temperature range (45-50-55-60°C). The isolates *A. pallidus* A1 and *B. licheniformis* C1, which were selected as the strongest biofilm producers, produced biofilms mainly on polystyrene and Teflon surfaces in the milk-containing environment. According to these results, *B. licheniformis* can be detected as a common contaminant in pasteurized milk sold in markets, and it is predicted that the detected isolates could be a problem in the dairy industry due to their ability to produce biofilm.

Keywords: *Aeribacillus pallidus*, *Bacillus licheniformis*, biofilm, dairy industry, thermophilic bacilli

DRAFT

ORAL PRESENTATION

The effects of meteorological factors and atmospheric pollutants on *Acer* pollen loads in urban area

Aydan Acar Şahin[1*] (*ORCID: https://orcid.org/0000-0002-5350-5534*)

[*1] Ankara University, Faculty of Science, Department of Biology, Ankara, Türkiye.

*Corresponding author e-mail: aydanacar24@gmail.com

Abstract

Pollen allergies can have a significant impact on public health, leading to increased healthcare costs and reduced quality of life for allergy sufferers. Choosing non-allergenic plants can contribute to a healthier urban population. However, *Acer* trees (maple) are commonly found as an ornamental plant in various urban landscapes, parks, gardens and along roadsides. These trees produce pollen that can trigger allergies in susceptible individuals during the spring months when they typically release their pollen. In this study, I investigated the effects of meteorological factors and air pollutants on airborne *Acer* pollen levels. Airborne pollen sampling was performed using a 7-day Hirst type volumetric spore trap for 2023 in Ankara province. The length of the whole season was found to be only 32 days. Spearman's correlation test was performed to determine the relations between the pollen concentrations and meteorological factors. As a result, the annual pollen integral values (APIn) were recorded as 802 pollen*day/m^3 and the highest pollen concentration was observed on March 27. Daily minimum temperature and carbon monoxide (CO) were the most relevant meteorological factor and air pollutant influencing the daily airborne pollen concentrations. Nevertheless, the pollen season is short, significant levels of allergenic *Acer* pollen are released into the air in the spring. It is very important that plants with allergenic pollen, such as maple, are not recommended to be included in urban landscape planning. Ultimately, collaboration among landscape planners, healthcare professionals, and environmental experts can help strike the right balance in urban landscaping decisions.

Key words: Acer, pollen, temperature, CO, urban

DRAFT

ORAL PRESENTATION

Microbial Inactivation of Foodborne Pathogens by Ultrasound Technology

Fatma AL (*ORCID: https://orcid.org/0000-0002-4124-2677*),
Güliz HASKARACA* (*ORCID: https://orcid.org/0000-0002-4641-866X*)

Sakarya University, Faculty of Engineering, Department of Food Engineering, Sakarya, Turkey

*Corresponding author e-mail: gyaldirak@sakarya.edu.tr

Abstract

From the past to date, conventional microbial inactivation methods have been used in the food industry to ensure food safety and security and prolong the shelf life. However, these conventional methods lead to losses in quality parameters, such as losses in nutritional components and the sensory quality of food products. As a novel inactivation method, ultrasound technology (UST) attracts attention with its advantages compared to conventional methods. Minimized flavor loss, increased sphere of influence homogeneity, significant energy savings, and being green technology are reported as important advantages of UST.
UST is an alternative non-thermal technology for inactivating microorganisms in food products. During ultrasound treatment, with the effect of ultrasounds pressure and sudden temperature changes occur, leading to the formation of cavitation. Cavitation helps to thin the microbial cell membrane, and with increased heat's synergistic effect, microbial inactivation occurs. Furthermore, during ultrasound treatment, various parameters such as frequency, amplitude, exposure time, process temperature, type of microorganisms, volume of the treated food, and food composition have an impact on microbial inactivation. Thus, it is important to evaluate the effects of UST on each microorganism under different conditions and UST parameters.
Although UST has remarkable microbial inactivation in both liquid and solid foods, many researchers have revealed an increased antimicrobial effect of ultrasound when combined with other methods such as chemical treatment, mild heat application, pressure, steam, or organic acids applications. This study aims to review the effects of UST on the inactivation of main foodborne pathogens such as Listeria monocytogenes, Escherichia coli, Salmonella spp. either alone or combined inactivation methods.

Keywords: Ultrasonication, microbial inactivation, combined treatment, food safety

DRAFT

ORAL PRESENTATION

A Novel Approach in Poultry Carcass Decontamination: Ultrasound Technology

Fatma AL (*ORCID: https://orcid.org/0000-0002-4124-2677*),
Guliz HASKARACA* (*ORCID: https://orcid.org/0000-0002-4641-866X*)

Sakarya University, Faculty of Engineering, Department of Food Engineering, Sakarya, Turkey

*Corresponding author e-mail: gyaldirak@sakarya.edu.tr

Abstract

Due to factors like contaminated water and surfaces in poultry processing facilities, the risk of microbial contamination increases in processes. Although all necessary precautions are tried to be taken during the processing of poultry products, ensuring complete product quality and microbial safety remains a challenge. Thus, poultry meats may contain pathogenic and spoilage microorganisms in the final product. In order to protect products from microorganisms, several physical, chemical, and biological sanitation methods have been used alone or in combination from the past to date. Among these sanitation methods, as well as suitability for use in the product, efficiency in decontamination, cost-efficiency, and harmlessness for the environment and health are important matters. At this point, ultrasound technology (US) emerges as a technology that can be useful in decontamination. The US is one of the non-thermal technologies accepted as green technology and reported as an effective decontamination method. Also, it is reported that US technology improves some of the processing features of meats. This study focuses on the application possibilities of the US in poultry carcass decontamination and provides a short literature review.

Keywords: Poultry meat, microbial decontamination, food safety, non-thermal technologies

DRAFT

ORAL PRESENTATION

Synthesis and application of thiourea-modified cellulose as an organocatalyst in asymmetric Mannich reaction

Ayşe Halıç Poslu[1*] (*ORCID: https://orcid.org/0000-0001-8705-6522*),
Gamze Koz[2] (*ORCID: https://orcid.org/0000-0003-3276-1413*)

[*1]Bursa Technical University, Faculty of Engineering and Natural Science, Chemistry, Bursa, Turkey
[2] Bursa Technical University, Faculty of Engineering and Natural Science, Chemistry, Bursa, Turkey

*Corresponding author e-mail: aysehalic@btu.edu.tr

Abstract

Asymmetric synthesis plays an important role in the field of organic chemistry, targeting the selective production of optical isomers. Organocatalysis has emerged as a powerful tool in the field of asymmetric synthesis, enabling the efficient production of chiral compounds with high selectivity. Cellulose, an abundant and renewable biopolymer, offers several advantages such as biocompatibility, low cost, and environmental sustainability. Cellulose derivatives have been successfully used in a wide variety of asymmetric reactions, including Michael addition, Aldol, and Mannich reactions. These derivatives exhibit the ability to form non-covalent interactions such as hydrogen bonding, van der Waals forces, and π-π interactions. The use of thiourea-modified cellulose derivatives as organocatalysts in asymmetric Mannich reactions represents an exciting area in organic chemistry that could replace conventional sustainable and biocompatible organocatalysts. Future research in this area could enable the discovery of new thiourea-derived cellulose structures with improved catalytic properties, expanding efficient and environmentally friendly asymmetric synthesis methodologies. Herein, microcrystalline cellulose was modified with 3,5-bis(trifluoromethyl)phenyl isocyanate, and new organocatalysts were synthesized. The synthesized catalysts were systematically characterized and their properties were tested in asymmetric Mannich reactions (Fig. 1).

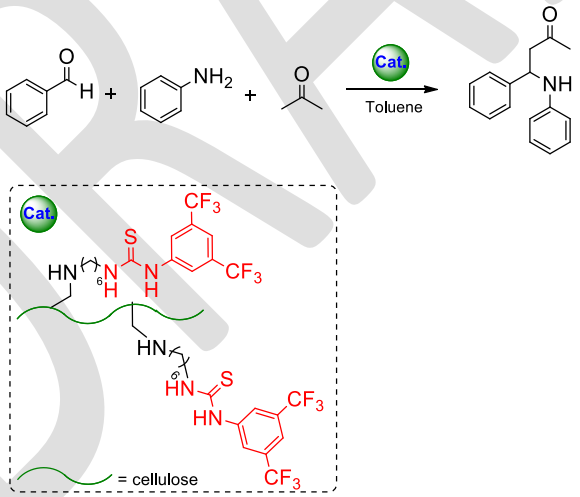


Fig.1. Thiourea-modified cellulose catalyst and the standard Mannich reaction.

Keywords: Asymmetric synthesis, Mannich reaction, Thiourea, Cellulose.

DRAFT

ORAL PRESENTATION

Ag_2O Np Katkılı PVA Yapılı İlaç Salım Sistemlerinin Hazırlanması ve Karakterizasyonu

Ezgi Altınay[1] (*ORCID:* https://orcid.org/009-006-1748-076X),
Kadriye Kızılbey[2*] (*ORCID: https://orcid.org/0000-0002-0297-0057*)

*[1] İstanbul Yeni Yüzyıl Üniversitesi, Fen Bilimleri Enstitüsü, Biyomedikal Mühendisliği, İstanbul, Türkiye
*[2]Acıbadem Üniversitesi, Mühendislik ve Doğa Bilimleri Fakültesi, Temel Bilimler, İstanbul, Türkiye

* ezgiibaran@gmail.com

Özet

İlaç, hastalığın tedavisinde veya önlenmesinde kullanılması amaçlanan bir madde olarak bilinmektedir. İlaç dağıtımı, vücudun bazı bölgelerindeki ilaç konsantrasyonunu diğerlerine kıyasla özellikle artıracak şekilde ilacın hastaya verilmesi yöntemidir. Her doz miktarının formu, ilaç/aktif farmasötik bileşenler ile katkı maddeleri adı verilen ilaç dışı bileşenlerin bir kombinasyonudur. İlaç etken maddeleri hastalıkları tedavi etmek için kullanılan asıl kimyasal bileşenlerdir. Konvansiyonel sistemlere göre kontrollü ilaç sistemleri daha farklı bir sistematiğe dayanmaktadır. Kontrollü ilaç salım sistemleri ilaç etken madde miktarını en aza indirerek canlı sistemde kalış süresini uzatır ve ilaç miktarına bağlı olarak hastada oluşabilecek zararlı yan etkileri azaltmayı hedefler. Bu çalışmada, biyouyumlu bir polimer olan polivinil alkol (PVA) ve antimikrobiyal özellikli Gümüş oksit nanoparçacıklar (Ag_2O Np) kullanılarak hidrojel yapılı filmler hazırlanmış ve karakterize edilmiştir. Oluşturulan filmlerin ilaç salım kapasitelerini incelemek üzere hidrojellere ibuprofen etken maddesi yüklenmiştir. Gümüş bazlı ürünlerin antimikrobiyal özelliğinden yararlanılmak üzere, yeşil sentez ile Equisetum telmateia bitkisinden gümüş nanoparçacık sentezi yapılmıştır. PVA hidrojelleri donma-çözme tekniği ile hazırlanmıştır. Hazırlanan filmlerin yapısal karakterizasyonu için FTIR spektrofotometre ve morfolojik özelliklerinin anlaşılması için SEM analizleri yapılmıştır. Ayrıca şişme testleri yapılmış olan filmlerin mekanik dayanımlarının belirlenmesi için çekme testi gerçekleştirilmiştir. İlaç yüklenen hidrojel yapılı filmlerin ilaç salım profilleri incelendikten sonra sitotoksisite özellikleri ve antibakteriyel etkileri belirlenmiştir. Yeşil sentez ile elde edilen gümüş oksit nanopartiküllerin parçacık boyutları Zeta Sizer cihazı ile yapılan analiz edilmiştir. İlaç etken maddesi salım deneylerinde Ag_2O Np katkılı %10'luk (w/v) (E1) ve %5'lik (w/v)'lik (B1) PVA filmleri kullanılmıştır. Salım deneyleri sonucunda; kullanım amacına ve kullanılan süreye göre daha hızlı bir salım isteniyor ise E1, daha yavaş bir salım hedefleniyor ise B1 hidrojeli tercih edilebileceği düşünülmektedir. Konvansiyonel yöntemlerden farklı olarak düşük maliyetli ve çevre dostu olarak hazırlanan nanopartikül katkılı PVA hidrojel yapılı sistemleri içeren bu çalışmanın sonucuna göre, belirlenen zaman aralığında ve ilacın istenilen salım miktarına göre hidrojel yapılı film tasarımının gerçekleştirilebileceği ve düşünülmektedir.

Anahtar Kelimeler: İlaç salımı, Polivinil alkol, Gümüş oksit nanopartikül, Hidrojel

DRAFT

ORAL PRESENTATION

3 Boyutlu Biyoyazıcı Tasarımı

Volkan Baki[1*] (*ORCID: https://orcid.org/0009-0008-3293-7857*),
Kadriye Kızılbey[2] (*ORCID: https://orcid.org/0000-0002-0297-0057*)

[*1]İstanbul Yeni Yüzyıl Üniversitesi, Fen Bilimleri Enstitüsü, Biyomedikal Mühendisliği Anabilim Dalı, İstanbul, Türkiye
[2]Acıbadem Üniversitesi, Mühendislik ve Doğa Bilimleri Fakültesi, Temel Bilimler, İstanbul, Türkiye

* volkanbaki34@gmail.com

Özet

Üç boyutlu biyoyazıcılar, özellikle Rejeneratif Tıp ve Doku Mühendisliği alanında kullanılan ve pek çok sorun için umut vaat edici çözümler üreten ileri teknolojik biyomedikal cihazlardır. Biyoyazıcılar geleneksel doku mühendisliği tekniklerindeki kısıtlamaların üstesinden gelebilmek için, 3B karmaşık dokuların/organların, baskılama veya 3B fabrikasyon teknolojilerinin kullanılarak üretildiği yeni geliştirilen bir yöntemdir. Ayrıca bu yöntem biyomalzemeler içerisinde hücrelerin kontrollü bir şekilde yerleşimine izin verir. Fabrikasyon tekniği, bilgisayar destekli tasarım (CAD) çizimlerinin sanal olarak tasarlanan verilerini okuyarak 3 boyutlu nesnelerin tabaka tabaka inşa edilmesiyle oluşturulan hızlı prototipleme prensibine dayanmaktadır. Bu çalışmada bilgisayar destekli bir çizim programı olan SolidWorks kullanılarak tek başlıklı bir üç boyutlu biyoyazıcı tasarımı ve prototip imalatı yapılmıştır. Biyoyazıcının metal ve plastik parçaların çoğunluğu CNC ve Torna makineleri ile işlenmeye uygun şekilde çizilerek üretilmiştir. Üretilmiş olan yazıcının ekstruder kısmı bir şırınga pompası şeklinde tasarlanmış olup dişliler sayesinde motorun dönme hareketini şırıngaya aktarmaktadır. Elektronik devrenin kontrolü için Duet3d firmasının "Duet 2" elektronik kartı ve ekran olarak ise aynı firmanın "PanelDue" ürünü tercih edilmiştir. Biyoyazıcıda X,Y,Z eksenlerinde hareketi sağlayan birer tane ve şırıngada baskıyı aktif hale getirmek için de 1 adet olmak üzere toplam 4 adet motor bulunmaktadır. Baskı işlemlerinde tabladan dökülen, saçılan sıvıların kızaklara zarar verebileceği ihtimaline karşın X eksen hareketi kayış-kasnak sistemi ile sağlandı. Eksen hareketleri 3 adet optik limit switch ile sınırlandırılmıştır. Şırıngaya eklenen diş macunu ile bazı denemeler yapılarak sistemin verimli çalışıp çalışmadığı kontrol edilmiştir. Yapılan denemeler sonucunda üretilmiş olan bu biyoyazıcının istenilen kalitede baskılar alınabilecek bir 3 boyutlu biyoyazıcı olduğu görülmüştür. Bu çalışmada hala gelişmekte olan 3B biyoyazıcı teknolojisine uygun olarak tasarlanan bu ürün ile; ülkemize, geleceğin öncelikli tıbbi araştırma alanları içerisinde bulunan, Rejeneratif Tıp ve Doku Mühendisliği alanında araştırma imkânı sağlayacak, yerli imkânlarla üretilmiş bir 3B biyoyazıcı kazandırılmıştır.

Anahtar Kelimeler: 3B yazıcı teknolojileri, 3 boyutlu biyoyazıcı, biyobaskı, doku mühendisliği, biyomedikal mühendisliği

DRAFT

ORAL PRESENTATION

Fermented marine algae source in aquaculture

Şehira Kabalatimardinli [1*] (*https://orcid.org/0000-0002-9813-2647*.)

[*1]Yalova University, ArmutluVocational School, Aquaculture, Yalova, Turkey.

*Corresponding author e-mail: shahykabalaty@gmail.com

Abstract

Aquatic enterprises in our time are no longer limited to fishing and fish and bivalve farming but have also begun to focus on culturing other products. One of these examples is the products obtained from aquatic algae. Marine algae have been used in various industries, from food production to biofuel, medical and pharmaceutical products, fish feed, and water purification. Marine algae contain biomolecules that can produce biomaterial by treatments like fermentation. Enzyme production by algae fermentation is one of the recent trends to benefit from algae. Enzymes used in the animal feed industry are generally obtained from animal or plant sources. Still, the production cost is high, and their stability is lower than bacterial enzymes. At the same time, algae can produce proteins and enzymes either through the process of photosynthesis or through the process of fermentation using different microorganisms on substrates to produce specific enzymes. Algae and its enzymes have critical industrial applications, but we focus on their uses as feed additives in aquafeed. The fermented algae and enzymes could be potential sources of protein and additives in fish feed to enhance digestion and immunity in fish, which achieves economic benefits for aquaculture projects and environmental sustainability. This study will present information about obtaining new products from algae through fermentation and their effects on fish.

Keywords: enzymes, ulva, bromealin, yeast, marine algae, fermentation, aquafeed.

DRAFT

ORAL PRESENTATION

üzerine inhibisyon etkilerinin incelenmesi ve moleküler docking yöntemiyle enzim-inhibitör etkileşimlerinin araştırılması

Fikret Türkan[1*] (*ORCID: https://orcid.org/0000-0002-0538-3157*),
Meryem Ayan Üstün[2] (ORCID:*https://orcid.org/0009-0007-4761-8122*)

[*1]Iğdır Üniversitesi, Diş Hekimliği Fakültesi, Temel Bilimler Bölümü, Iğdır, Türkiye
[2]Iğdır Üniversitesi, Lisansüstü Eğitim Enstitüsü, Biyokimya Bölümü, Iğdır, Türkiye

*Sorumlu yazar e-mail:fikret.turkan@gmail.com

Özet

Bu çalışmasında primidin türevlerinden olan 4,6-Dikloro-2-(metiltio) pirimidin-5-karbonil klorür ve 4,6-Dikloro-2-(metiltio) pirimidin-5-karboksiamid maddelerinin, hücre içi detoksifikasyonu sağlayan Glutatyon ile konjuge çalışmasıyla bilinen GST enzimi üzerine olan inhibisyon etkileri incelendi. Elde edilen veriler GST inhibitörü olarak kabul edilen etakrinik asit değerleri ile kıyaslandı. Molekül a, molekül b ve etakrinik asit için IC_{50} değerleri sırasıyla 38,5 nM, 46,2 nM, 5,82 nM olarak hesaplandı. İnhibisyon çalışmasının ikinci basamağında ise IC_{50} grafiklerinden hareketle Ki değerlerine ulaşıldı. Ki değerleri yine sırasıyla 57,61 nM, 43,75 nM, 4,43nM olarak bulunurken; a ve b moleküllerinin yarışmasız, etakrinik asitin ise yarışmalı inhibisyon tipine sahip olduğu görüldü. Çalışmanın ikinci bölümünde ise ilgili moleküllerin antioksidan kapasitelerini belirlemek amacıyla DPPH (2,2-difenil-1-pikrilhidrazil) yöntemi kullanıldı. a ve b molekülleri DPPH yönteminde standart olarak kabul edilen askorbik asit (vitamin C) ile karşılaştırıldığında standarda oldukça yakın değerler verdiği görüldü. Ayrıca bu tez çalışmamızda GST enzimi ile ilgili pirimidin moleküllerinin bağlanma mekanizmasını gösteren doking çalışması yapılmış olup molekül a için -5,6 kcal/mol, molekül b için -6,2 kcal/mol olarak bulunmuştur.

Anahtar Kelimeler: Glutatyon S-transferaz, Pirimidin, Doking

DRAFT

ORAL PRESENTATION

Phytochemical Study on *Gypsophila venusta*

Fırat TAŞLIYÜK[1] (https://orcid.org/0000-0002-2382-9570), Rabia Nur ÜN[1] (https://orcid.org/0000-0001-7903-9167), Özgen ALANKUŞ[1] (https://orcid.org/0000-0003-2017-3382), Tamer KARAYILDIRIM[1,*] (https://orcid.org/0000-0001-7451-0810)

[*1] Ege University, Faculty of Science, Chemistry Department, Izmir, Turkey.

*tamer.karayildirim@ege.edu.tr

Abstract

The Caryophyllaceae family consists of approximately 90 genera, traditionally utilized across various ethnic communities for treating numerous ailments. In particularly, Caryophyllaceae family has great significance in Chinese traditional medicine as a remedy for a wide range of health conditions [1]. Members of the Caryophyllaceae family are rich in natural compounds, such as triterpenes, phenolics, and alkaloids. These compounds are known to have medicinal properties [2]. Among the most enormously studied metabolites of the Caryophyllaceae circle of relatives are flavonoids, that are classified as polyphenolic compounds and display an extensive type of organic and pharmacological activities along with antioxidant, anti-oedemic, anti-inflammatory, antimicrobial, and immunomodulatory outcomes [3].

Gypsophila is the third largest genus in the Caryophyllaceae of Turkey. *Gypsophila* species are annual, biennial, or perennial herbaceous plants. This genus is mainly distributed in the Mediterranean areas of Turkey, Iran and Turan. There are 56 species in 10 sections of *Gypsophila*, 33 species in Turkey [1, 4]. In literature, there are several studies on *Gypsophila* species [5, 6]. Mainly saponins and phenolic compounds have been isolated from *Gypsophila*. However, there is no phytochemical study on *Gypsophila venusta*. This is the first study that describes the isolation and identification of secondary metabolites from *Gypsophila venusta*.

Saponins and flavonoids are specialized metabolites that remain the most extensively studied constituents of the *Gypsophila* species. Recently, the flavonoids variety in the Caryophyllaceae was reviewed by Jakimiuk et al. [7]. Cheikh-Ali et al. (2019) highlighted structural organization of saponins in the family [8].

Gypsophila venusta was collected from Malatya, Turkey. Air-dried and powdered plant material was extracted with *n*-Hexane, DCM and MeOH. The DCM phase was chromatographed over column chromatography. The fractions repeatedly subjected to normal phase silica gel CC and reverse phase silica gel column chromatography.

In this study, one saponin and one phenolic compound were isolated from the DCM extract. Their structures were elucidated by extensive spectroscopic methods including ^{1}H and ^{13}C-NMR techniques.

Keywords: Caryophyllaceae, *Gypsophila*, Secondary metabolities

References

[1] Chandra S., Rawat D.S. 2015. Medicinal plants of the family Caryophyllaceae; a review of ethno-medicinal uses and pharmacological properties, Integrative Medicine Research; 4, 123-131.
[2] Wink M. 2010. Biochemistry of plant secondary metabolism, 2nd ed., Wiley-Blackwell, Chichester, 464.
[3] Jakimiuk K., Wink M., Tomczyk M. 2022. Flavonoids of the Caryophyllaceae, Phytochemistry Reviews; 21, 179-218.
[4] Davis P.H., Mill R.R, Tan K., Edmondson. J.R. 1982. Flora of Turkey and the East Aegean Islands. Edinburgh, Scotland: University Press.
[5] Gevrenova R., Zengin G., Balabanova V., Voynikov Y., Zheleva-Dimitrova D. 2021. C, O-flavonoid glycosides and oleanane-type bidesmosides from Gypsophila perfoliata L. "tekirae" (Caryophyllaceae): Chemophenetic implications, Biochemical Systematics and Ecology, 99, 104353.
[6] Gevrenova R. 2022. Bidesmosidic Triterpenoid Saponins from Wild and Cultivated Gypsophila Paniculata L. (Caryophyllaceae), Proceedings of the Bulgarian Academy of Sciences, 75, 367-378.
[7] Jakimiuk K., Wink M., Tomczyk M. 2021. Flavonoids of the Caryophyllaceae, Phytochemistry Reviews, 21, 179-218.
[8] Cheikh-Ali, S., Farman, M., Lacaille-Dubois, M.A., Semmar, N. 2019. Structural organization of saponins in Caryophyllaceae, Phytochemistry Reviews, 18, 405-441.

ORAL PRESENTATION

SDS Sedimantasyon analizinde SDS çözelti miktarı ve analiz süresinin değişiminin sonuçlar üzerine etkisinin değerlendirilmesi.

Alaettin KEÇELİ* (https://orcid.org/0000-0003-1263-8952)

*Pamukkale Üniversitesi, Uygulamalı Bilimler Fakültesi, Organik Tarım İşletmeciliği Bölümü Çivril/DENİZLİ

*Sorumlu yazar e-mail:alaettink@pau.edu.tr

Özet

Buğday ıslahında genellikle erken kademe materyallerinin miktarının azlığı sebebiyle protein kalitesinin tahmini için SDS (Sodyum Dodesil Sülfat) Sedimantasyon analizi yoğun olarak kullanılmakta ve hızlı sonuç alınmaktadır. Bunun yan sıra buğday tanesinde emgi zararı olup olmadığını test etmek amacıyla Beklemeli SDS Sedimantasyon analizi yapılır. Ekmeklik buğdayda zeleny sedimantasyon analizine karşılık kırma ve makarnalık buğdayda SDS sedimantasyon analizi kullanılmaktadır. Bu çalışmada; %100 Tosunbey ekmeklik buğday unu, %100 siyez buğday unu, Tosunbey ve siyez tam buğday unlarının farklı oranlardaki karışımlarına ait örneklerde 15., 20 ve 25. dakikada yapılan SDS sedimantasyon ve beklemeli sedimantasyon analizleri yapılmıştır. Sonuçlar toplu olarak değerlendirildiğinde; buğday paçallarında un ile tam buğday karışımım miktarı arttıkça SDS sedimantasyon değerleri düşüş göstermektedir. Ayrıca Normal koşularda 15. dakika da ölçülen SDS sedimantasyon sonuçları ile karşılaştırıldığında 20. ve 25. dakikada yapılan ölçümlerden elde edilen sonuçlarda önemli ölçüde düşüşler meydana gelmektedir. Sonuçlar açık olarak göstermektedir ki; un analizi sonuçları kırma veya tam buğday örneklerine göre daha yüksek sonuçlar vermektedir. Ayrıca standart metot tarafından bildirilen süre olan 15. dakika sonrasında yapılan 20. ve 25. dakika ölçümlerin normal ölçümlere göre daha düşük olduğu belirlenmiştir. Sonuç olarak SDS sedimantasyon ve beklemeli SDS sedimantasyon analizi için belirtilen 15. dakika ölçümleri daha yüksek olarak ölçülmüş ve kalite değerlendirilmesinde kullanılmasının uygun olduğu ve bekleme süresinde meydana gelecek artışların değerler üzerinde olumsuz etkiler meydana getireceği sonucuna varılmıştır.

Anahtar Kelimeler: SDS sedimantasyon, beklemeli SDS, siyez, ekmeklik buğday, SDS ölçüm süresi,

DRAFT

ORAL PRESENTATION

Removal of methyl orange (MO) and reactive blue 4 (RB4) from aqueous solution by CTAB-modified coffee waste-based activated carbon

Eda Gökırmak Söğüt* (https://orcid.org/0000-0002-7707-3924)

* Van Yuzuncu Yil University, Van Security Vocational School, Van, Turkey

*Corresponding author e-mail: edagokirmak@yyu.edu.tr

Abstract

Coffee is the second most-traded commodity in the world, after oil and one of the world's most popular beverages [1]. Coffee waste is used for various purposes such as biofuels [2], polymeric films [3], porous materials [4], activated carbon [5] and adsorbents [6–8]. In this context, activated carbon (CAC) samples obtained from waste coffee were modified with the cationic surfactant cetyltrimethylammonium bromide (CTAB) (CCAC), and the obtained samples were characterized by scanning electron microscopy (SEM) and X-ray diffraction (XRD). The adsorption behaviour of methyl orange (MO) and reactive blue 4 (RB4) onto CAC and CCAC from aqueous solution and the adsorption mechanism were investigated. Scanning electron microscopy (SEM) revealed irregular pores on the CAC surface characterized by deep voids and long, irregular hollow protrusions. This increased porosity is the reason why carbon-containing adsorbents are mostly preferred for adsorption processes. The CCAC surface exhibits an irregular and dense morphology with enhanced and clearly visible macro- and micropores as well as high pore density and heterogeneity. The XRD pattern showed that the crystal structure of CCAC was not affected. Both diffraction patterns show characteristic peaks around 2 $\theta = 28°$, indicating the presence of an amorphous structure [9]. The results showed that equilibrium was reached within 45 minutes and the optimal pH for MO and RB4 was 2 and 5, respectively. It was observed that the percentage of MO removal from the aqueous solution was increased from 83% to 99% and RB4 from 67% to 82% by the effect of CTAB modification. The results were consistent with the pseudo-second-order model and showed that the adsorption mechanism of MO and RB4 on CAC and CCAC mainly involved electrostatic attraction and surface adsorption [10]. In conclusion, CCAC is a promising material for dye wastewater adsorption as well as an environmentally friendly and effective material for water treatment applications.

Keywords: Active carbon, adsorption, CTAB, dye, surfactant, waste coffee.

Reference

[1] R. Campos-Vega, G. Loarca-Piña, H.A. Vergara-Castañeda, B. Dave Oomah, Spent coffee grounds: A review on current research and future prospects, Trends Food Sci. Technol. 45 (2015) 24–36. https://doi.org/10.1016/J.TIFS.2015.04.012.

[2] X.J. Lee, H.C. Ong, W. Gao, Y.S. Ok, W.H. Chen, B.H.H. Goh, C.T. Chong, Solid biofuel production from spent coffee ground wastes: Process optimisation, characterisation and kinetic studies, Fuel. 292 (2021) 120309. https://doi.org/10.1016/J.FUEL.2021.120309.

[3] M.J.P.A. Batista, M.B.F. Marques, A.S. Franca, L.S. Oliveira, Development of Films from Spent Coffee Grounds' Polysaccharides Crosslinked with Calcium Ions and 1,4-Phenylenediboronic Acid: A Comparative Analysis of Film Properties and Biodegradability, Foods. 12 (2023) 2520. https://doi.org/10.3390/FOODS12132520/S1.

[4] H. Akasaka, T. Takahata, I. Toda, H. Ono, S. Ohshio, S. Himeno, T. Kokubu, H. Saitoh, Hydrogen storage ability of porous carbon material fabricated from coffee bean wastes, Int. J. Hydrogen Energy. 36 (2011) 580–585. https://doi.org/10.1016/J.IJHYDENE.2010.09.102.

[5] H. Laksaci, B. Belhamdi, O. Khelifi, A. Khelifi, M. Trari, Elimination of amoxicillin by adsorption on coffee waste based activated carbon, J. Mol. Struct. 1274 (2023) 134500. https://doi.org/10.1016/J.MOLSTRUC.2022.134500.

[6] P. Deivasigamani, P. Senthil Kumar, S. Sundaraman, M.R. Soosai, A.A. Renita, K. M, N. Bektenov, O. Baigenzhenov, V. D, A. Kumar J, Deep insights into kinetics, optimization and thermodynamic estimates of methylene blue adsorption from aqueous solution onto coffee husk (Coffee arabica) activated carbon, Environ. Res. 236 (2023) 116735. https://doi.org/10.1016/J.ENVRES.2023.116735.

[7] W. Cherdchoo, S. Nithettham, J. Charoenpanich, Removal of Cr(VI) from synthetic wastewater by adsorption onto coffee ground and mixed waste tea, Chemosphere. 221 (2019) 758–767. https://doi.org/10.1016/J.CHEMOSPHERE.2019.01.100.

[8] J. Shin, M. Choi, C.Y. Go, S. Bae, K.C. Kim, K. Chon, NaOH-assisted H2O2 post-modification as a novel approach to enhance adsorption capacity of residual coffee waste biochars toward radioactive strontium: Experimental and theoretical studies, J. Hazard. Mater. 435 (2022) 129081. https://doi.org/10.1016/J.JHAZMAT.2022.129081.

[9] C.F. Ramirez-Gutierrez, R. Arias-Niquepa, J.J. Prías-Barragán, M.E. Rodriguez-Garcia, Study and identification of contaminant phases in commercial activated carbons, J. Environ. Chem. Eng. 8 (2020) 103636. https://doi.org/10.1016/J.JECE.2019.103636.

[10] A.S. Franca, L.S. Oliveira, M.E. Ferreira, Kinetics and equilibrium studies of methylene blue adsorption by spent coffee grounds, Desalination. 249 (2009) 267–272. https://doi.org/10.1016/J.DESAL.2008.11.017.

ORAL PRESENTATION

HPLC-DAD phenolic content investigation, antioxidant, and antimicrobial activities of *Polygonum cognatum* Meissn

Reyhan AKPINAR[1*] (*ORCID: 0000-0001-5910- 4160*),
Gizem YILDIRIM BAŞTEMUR[2] (*ORCID: 0000-0002-4634-4525)*,
Nazmiye Ozlem SANLI [3] (*ORCID: 0000-0001-8969-5125*),
Sabriye PERÇİN ÖZKORUCUKLU[2] (*ORCID: 0000-0001-9778-2034*)

[*1]Istanbul University, Institute of Science, Programme of Molecular Biotechnology and Genetics, Istanbul, Turkey.
[2] Istanbul University, Faculty of Science, Department of Molecular Biology and Genetics, Istanbul, Turkey.
[3] Istanbul University, Faculty of Science, Department of Biology, Istanbul, Turkey.

*Corresponding author e-mail: reyhanakpnr1@gmail.com

Abstract

Polygonum cognatum Meissn., belonging to the *Polygonaceae* family, is a perennial plant that grows spontaneously in our country. This plant is rich in phenolic content. Phenolic compounds constitute the most important secondary metabolite group of plants and are known to have antioxidant and antimicrobial activities. In this study, a new HPLC-DAD method has been developed and validated for the simultaneous qualitative and quantitative determination of 10 phenolic compounds (protocatechuic acid, catechin, gentisic acid, caffeic acid, p-coumaric acid, polydatin, coumarin, resveratrol, ellagic acid, quercetin) in the plants which were harvested from different regions at different times. Chromatographic determination was performed with the gradient program on the SUPELCOSIL LC-18 (25cmx4.6mm,5µm) column. Evaluating of the validation parameters, the method proved to have remarkable linearity, sensitivity, and precision. LOD and LOQ values were calculated in the range of 0.051-0.188 µg/mL and 0.169-0.628 µg/mL, respectively. The plant extracts which include phenolic compounds were analyzed as simultaneous utilizing the chromatographic method that developed and validated. The antioxidant, and antimicrobial activities of the plants which are collected from Sivas (Plant A) and Ordu (Plant B) and extracted by soxhlet method were investigated. Antioxidant activities of the plants were determined by DPPH and CUPRAC method. IC_{50} (IC_{50}=0.781 mg/mL for Plant A, IC_{50}=0.657 mg/mL for Plant B) and EC_{50} values (EC_{50}=368.0 µg/mL for Plant A, EC_{50}=364.3 µg/mL for Plant B) were calculated by DPPH and CUPRAC methods, respectively. Antimicrobial activities of *Polygonum cognatum* methanolic extracts were investigated by diffusion and dilution method against both Gram-negative/positive bacteria and fungi. *Candida albicans* was exhibited to be greater sensitive 24.60 ± 0.55 mm inhibition zone and detected MIC values of 4.75 mg/mL and MBC values of ≥ 4.75 mg/mL.

Keywords: *Polygonum cognatum* Meissn., Phenolic Compounds, HPLC, Antioxidant Activity, Antimicrobial Activity.

DRAFT

ORAL PRESENTATION

Microbial conversion of dehydroepiandrosterone and pregnenolone by two *Penicillium* species

Mena Albayaty* (*ORCID: https://orcid.org/0009-0009-6630-2902*), Atheer Al-Samarrai (*ORCID: https://orcid.org/0000-0001-5294-9224*), Halil İbrahim Yilmaz (*ORCID: https://orcid.org/0009-0001-5546-7228*), Ali Kuru (*ORCID: https://orcid.org/*0000-0002-8300-1572) and Kudret Yildirim (*ORCID: https://orcid.org/*0000-0002-7889-5903).

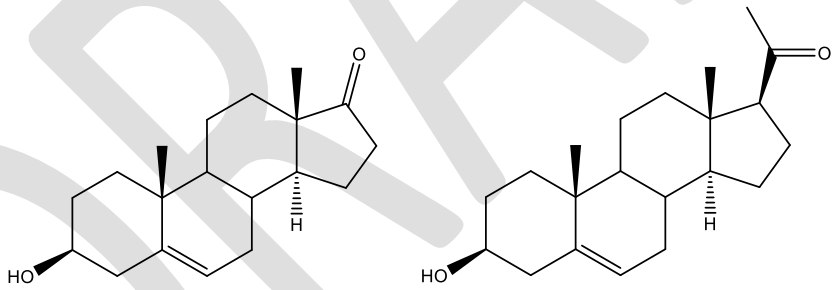
Sakarya University, Faculty of Sciences, Chemistry Department, Sakarya, Türkiye.

*mena.albayaty@ogr.sakarya.edu.tr

Abstract

Microbial steroid biotransformations have found worldwide application for preparing more valuable and functionalized compounds due to their high chemo-, regio- and stereoselectivities. There are still enormous efforts to increase microbial biotransformations' efficiency and find new useful microorganisms and reactions[1]. Different *Penicillium* species have been used for the biotransformations of some steroids. Literature has shown that Penicillium species can mainly perform bioconversion of steroidal substrates via 5α-reduction, hydroxylation or Baeyer-Villiger oxidation[2].

In this work, dehydroepiandrosterone **1** and pregnenolone **2** were incubated with *Penicillium olsonii* MRC 500780 and *Penicillium hirsutum* MRC 500372 for 5 days. Incubation of **1** with *P. olsonii* MRC 500780 gave androstenedione, 15α-hydroxyandrostenedione, 7β-hydroxydehyroepiandrosterone, 7α- hydroxydehyroepiandrosterone and 15α-hydroxytestosterone. Incubation of **2** with *P. olsonii* MRC 500780 gave progesterone and 15α-hydroxyprogesterone. Incubation of **1** with *P. hirsutum* MRC 500372 only gave androstenedione, whilst incubation of **2** with *P. hirsutum* MRC 500372 only gave progesterone.



All steroids were separated by column chromatography. Structures of the steroids were determined by comparison of their melting points, ^{1}H NMR, ^{13}C NMR and IR spectra with those of the starting materials.

Keywords: Biotransformation, *Penicillium olsonii, Penicillium hirsutum,* Dehydroepiandrosterone and Pregnenolone.

References:

1. Nassiri-Koopaei N, Faramarzi MA. 2015. Recent developments in the fungal transformation of steroids. Biocatal Biotransform. 33:1–28

2. Kolek T, Szpineter A, Swizdor A. N. 2008. Baeyer-Villiger oxidation of DHEA, pregnenolone, and androstenedione by Penicillium lilacinum AM111. Steroids, 73: 1441-1445.

DRAFT

ORAL PRESENTATION

Some important fungal diseases in rice (*Oryza sativa* L.) fields in the Thrace region in Turkey

Arzu COŞKUNTUNA* (ORCID ID: https://orcid.org/0000-0003-2232-7592)

* Tekirdağ Namık Kemal University, Agricultural Faculty Department of Plant Protection Tekirdağ, Türkiye.

*acoskuntuna@nku.edu.tr

Abstract

The diagnosis, symptoms, biology of some important fungal diseases that are problems in rice growing areas in the Thrace region of Turkey, and how to control the these diseases are reported in this review. The emergence of fungal diseases in rice, together with changing climate parameters, leads to significant crop losses day by day. The most common disease in rice is rice blast, and the disease agent is the fungus *Magnaporthe grisea* (teleomorph, TT Hebert ME Barr, anamorph *Pyricularia oryzae* Cav.). In terms of prevalence, this disease agent is followed by *Cochliobolus miyabeanus* (teleomorph, synonym: *Helminthosporium oryzae*, anamorph *Bipolaris oryzae*), which causes brown spot disease. The leading seed-borne disease in plants is *Gibberella fujikuroi* (bakanae-disease) (anamorph, *Fusarium fujikuroi*), also known as *Fusarium verticillioides* (anamorph, Syn [*F. moniliforme*]) in recent years.

Keywords: Rice (*Oryza sativa*), *Magnaporthe grisea*, *Cochliobolus miyabeanus*, *Gibberella fujikuroi*

DRAFT

ORAL PRESENTATION

Oligodendrosit hasarı ve bağırsak mikrobiyom çeşitliliği arasındaki ilişkinin araştırılması

Mehmet Bostancıklıoğlu[1*] (ORCID: https://orcid.org/0000-0001-7169-719X), Davut Sinan Kaplan[1*] (ORCID: https://orcid.org/0000-0003-4663-209X) Ebru Temiz[2*](ORCID: https://orcid.org/0000-0001-8911-7763), Elif Yiğit[1*](ORCID: https://orcid.org/0000-0002-9224-9924)

[1*]Gaziantep Üniversitesi, Tıp Fakültesi, Fizyoloji Anabilim Dalı, Gaziantep, Türkiye
[2*]Harran Üniversitesi, Sağlık Hizmetleri Meslek Yüksekokulu, Tıbbi Tanıtım ve Pazarlama Programı, Şanlıurfa, Türkiye

*elifyigit19961@hotmail.com

Özet

Bu çalışma, kafa içi lizolesitin enjeksiyonu ile indüklenen miyelin hasarının bağırsak florasına olan etkisini incelemeyi amaçlamıştır. Bu bağlamda, 10-12 haftalık erkek Wistar Albino sıçanlarında antibiyotik kokteyl tedavisiyle bağırsak flora hasarı ve hipokampal lizolesitin enjeksiyonuyla lokal miyelin hasarı modelleri oluşturulmuştur. Bağırsak florasındaki değişiklikler metagenomik sekanslama yöntemi kullanılarak belirlenmiştir. Ayrıca, hipokampal miyelin hasarı ile bağırsak mikrobiyom çeşitliliği arasındaki ilişkiyi incelemek amacıyla Miyelin Temel Proteini (MBP), Oligodendrosit Spesifik Proteini (OSP), Alfa-Sinüklein (aSyn), Interlökin-3 (IL-3), Tümör Nekroz Faktör-α (TNF-α) ve Klaudin-5 (CLDN5) protein ve gen ifade seviyeleri ELISA ve qPCR yöntemleriyle analiz edilmiştir. Elde edilen sonuçlara göre, kafa içi lizolesitin enjeksiyonu ve oral antibiyotik uygulamasının bakteri florasında benzer değişikliklere yol açtığı ve bakteri besleyici oral prebiyotik ile miyelin üretimini artıran klemastin tedavisinin bakteri florası üzerinde benzer yatıştırıcı etki gösterdiği görülmüştür. Ayrıca, lizolesitin uygulanan grupta hipokampüsteki TNF-α ve IL-3 gen ifadelerinin anlamlı bir şekilde artmıştır ($p<0.05$). Klemastin tedavisi, hipokampal MBP ve CLDN5 seviyelerini anlamlı bir şekilde artırırken, TNF-α seviyesini azaltmıştır. Kolon ve vagus örneklerindeki IL-3 seviyeleri de klemastin tedavisiyle anlamlı bir şekilde azalmıştır ($p<0.05$). Davranış deneyi sonuçlarına göre, lizolesitin uygulanan grupta anksiyete benzeri davranışların arttığı, ancak klemastin tedavisiyle birlikte bu davranışlarda azalma gözlemlenmiştir. Uyumlu bir şekilde, Morris Su Labirenti davranış testi sonuçlarına göre lizolesitin uygulanan grup ile klemastin uygulanan grup arasında S3 çeyrekliğinde geçirilen süre bakımından anlamlı bir farklılık bulunmuştur. Sonuç olarak, hipokampusta yapay olarak oluşturulan miyelin hasarının bağırsak mikrobiyom çeşitliliğini değiştirdiği ve davranış üzerinde etkili olduğu önerilmektedir. Ancak, bu çalışmanın sonuçlarına dayanarak kesin neden-sonuç ilişkisi önermek için transgenik model kullanılan daha detaylı araştırma gerekmektedir ve "sinaptik plastisite" alanındaki görüş ayrılıklarının dikkate alınması önerilmiştir.

Anahtar Kelimeler: Bağırsak disbiyozu, Lokal miyelin hasarı, Kolon, Miyelin-mikrobiyom aksı.

DRAFT

ORAL PRESENTATION

Evaluation of the anticancer effects of maleic anhydride/N-vinyl caprolactam copolymer and copolymer/organo-MMT nanocomposites

Buse Bekar[1*] (*ORCID: 0009-0001-1308-4915)*, Burcu Akar[2] *(ORCID: 0000-0003-4511-7440)*, Handan Sevim Akan[1] *(ORCID: 0000-0002-8511-5258)*, Hatice Kaplan Can[2] *(ORCID: 0000-0002- 2886-0788)*

[*1]Hacettepe University, Faculty of Science, Department of Biology, Ankara, Turkey
[2]Hacettepe University, Faculty of Science, Division of Polymer Chemistry, Ankara, Turkey

[*]Corresponding author e-mail: bekarbuse@gmail.com

Abstract

Organo-clays, such as organo-montmorillonite (Org-MMT/O-MMT), are the most widely used silicates in polymer nanotechnology (1). By using modified clays, it is possible to eliminate the weak mechanical structure and low thermal stability of the polymers, thus resulting in more durable nanocomposites (2). Our study aims to assess the anticancer effects of polymer-Org-MMT nanocomposites synthesized at different time intervals and containing varying concentrations of clay material. The anticancer effects of O-MMT, poly(MA-alt-VCL), and poly(MA-alt-VCL)/O-MMT polymer clay materials, were assessed using HeLa (cervical carcinoma) and A549 (lung cancer) cell lines, and HDF (human dermal fibroblast) cells as healthy control. To evaluate the cytotoxic effects of the materials, cells treated with materials in concentrations ranging from 0.0002 mg/ml to 10 mg/ml and an MTT analysis performed after 24 hours treatment. Furthermore, we conducted a colony formation assay with a concentration of 0.2 mg/ml to investigates the impact of the materials on cancer cell colony formation. The results showed that a concentration of 2 mg/ml gave an IC50 value in cancer cells, especially the substances were more effective in the A549 cell line, whereas this value was 10 mg/ml in healthy cells. Also O-MMT, poly(MA-alt-VCL), and poly(MA-alt-VCL)/O-MMT polymer clay materials suppressed colony formation in cancer cells. The data indicated an inverse correlation between clay content in the copolymer complex and cell viability. The copolymer complex without clay had no negative impact on the cells. According to these results, the nanocomposites containing organo-clays appear to hold promise for use in anticancer drug studies.

Keywords: Organo-clay, Montmorillonite, Copolymer, Anticancer, Drug delivery

References

Kaplan Can, H. (2016) Charge transfer complex formation in in-situ maleic anhydride and N-vinyl caprolactam copolymer and copolymer/organo-montmorillonite nanoarchitectures, *Journal of Macromolecular Science, Part A, 53:1*, 26-33. DOI: 10.1080/10601325.2016.1110454

Uysaler, B. (2019) *Preparation of functional anhydride copolymer-clay nanocomposites of organically modified montmorillonite (orgmmt) and silver determination of biological activities* (Yüksek Lisans Tezi). Hacettepe Üniversitesi, Ankara, Turkey.

DRAFT

ORAL PRESENTATION

Isolation and Structural Determination of Secondary Metabolites from *Moringa oleifera* Plant

Ozlem Buse Keskin[1], Ahmet Aykac[2] and Nazli Sarikahya[1]

[1]Department of Chemistry, Faculty of Science, Ege University, 35100, Bornova-Izmir/Turkiye
[2]Department of Bioengineering, Faculty of Engineering and Architecture, İzmir Katip Celebi University, 35620 Cigli-Izmir/Turkiye

ozlembusekeskin@hotmail.com

Abstract

There is only the *Moringa* genus in the Moringaceae family, and the most important species among the 13 species in this genus is *Moringa oleifera*. Although *Moringa oleifera* was first grown in the North Indian Regions in the foothills of the Himalayas[1], it is now widespread in many parts of the world such as Indonesia, Mexico, South America and the Middle East[2]. It is a species that can grow in semi-arid, tropical and subtropical climates. The root, seed, fruit and leaf parts of the plant are known to contain important vitamins and minerals such as vitamins A and C, calcium, protein and potassium, as well as secondary metabolites such as flavonoids, phenolic acids, glucosinolates, terpenes and alkaloids[3-5]. The biological activity studies on the plant have shown that it has anti-inflammatory, antidiabetic, antimicrobial, antioxidant, anticancer activities and sexual function regulating, wound healing properties[6-9].

The main aim of this study is isolation, purification and structural characterization of biologically active compounds from *Moringa oleifera* plant. After the seeds of the tree were dried and grounded under suitable conditions, MeOH and EtOH extracts were prepared. Afterwards, isolation procedures were continued with various chromatographic applications such as CC, RP-CC, RP-VLC, MPLC, TLC methods. So far, 2 compounds have been obtained in pure form and their chemical structures have been determined by advanced spectroscopic methods (1D, 2D-NMR and HR-Mass techniques). The isolation studies on the other fractions and structural determinations are still ongoing.

References

National Research Council. 27 Ekim 2006. "*Moringa*, Lost Crops of Africa: Volume II: Vegetables, Lost Crops of Africa", 2. National Academies Press, ISBN 978-0-309-10333-6.

Leone, A., Spada, A., Battezzati, A., Schiraldi, A., Aristil, J., Bertoli, S. 2015. "Cultivation, Genetic, Ethnopharmacology, Phytochemistry and Pharmacology of *Moringa oleifera* Leaves: An Overview", International Journal of Molecular Sciences, 16(6), 12791-12835.

Abd, R. N. Z., Husain, K., Kumolosasi, E. 2018. "*Moringa* Genus: A Review of Phytochemistry and Pharmacology", Frontiers in Pharmacology, 9, 108.

Saa, R. W., Fombang, E. N., Ndjantou, E. B., Njintang, N. Y. 2019. "Treatments and uses of *Moringa oleifera* seeds in human nutrition: A review", Food science & nutrition, 7(6), 1911-1919.

Leone, A., Spada, A., Battezzati, A., Schiraldi, A., Aristil, J., Bertoli, S. 2016. "*Moringa oleifera* Seeds and Oil: Characteristics and Uses for Human Health", International Journal of Molecular Sciences, 17(12).

Kou, X., Li B., Olayanju, J. B., Drake, J. M., Chen, N. 2018. "Nutraceutical or Pharmacological Potential of *Moringa oleifera* Lam.", Nutrients, 10(3).

Koike, M. K., Kochi, A. K., Pinto, D. Y. G. 2020. "Use of *Moringa Oleifera* Seeds in Water Treatment", Arquivos Brasileiros de Cardiologia, 114(6), 1038-1039.

Suphachai, C. 2014. "Antioxidant and anticancer activities of *Moringa oleifera* leaves", Journal of Medicinal Plants Research, 8(7), 318-325.

Wang, F., Bao, Y., Zhang, C., Zhan L., Khan, W., Siddiqua, S., Xiao, J. 2021. "Bioactive components and anti-diabetic properties of *Moringa oleifera* Lam.", Critical Reviews in Food Science and Nutrition, 1-25.

DRAFT

ORAL PRESENTATION

Molecular investigation of the role of papillomaviruses in the etiology of transmissible venereal tumors (CTVT) in dogs

İlker Şahinkesen[1*] (https://orcid.org/0000-0002-1915-6908), Seçkin Salar[2] (https://orcid.org/0000-0001-9303-6253),
Fırat Doğan[3] (https://orcid.org/0000-0001-8656-3645), Ayhan Baştan[2](https://orcid.org/0000-0001-8291-1147), Tarık Fedai[1](https://orcid.org/0009-0002-3166-2640), Ayşe Merve Köse[4](https://orcid.org/0000-0003-1863-5955), Ece Koldaş Ürer[4](https://orcid.org/0000-0002-9631-8501), Seval Bilge-Dağalp[5](https://orcid.org/0000-0002-1166-721X)

[1]The Graduate School of Health Sciences, Department of Virology, Ankara University, Ankara,Turkey.
[2]Ankara University, Faculty of Veterinary Medicine, Department of Obstetrics and Gynecology, Ankara,Turkey.
[3]Hatay Mustafa Kemal University, Faculty of Veterinary Medicine, Department of Virology, Hatay,Turkey.
[4]Hatay Mustafa Kemal University, Faculty of Veterinary Medicine, Department of Obstetrics and Gynecology, Hatay,Turkey.
[5]Ankara University, Faculty of Veterinary Medicine, Department of Virology, Ankara,Turkey.

*Corresponding author e-mail: ilkersahinkesen68@gmail.com

Abstract

Transmissible venereal tumors (TVT) are malignant (cancerous) round cell tumors. Different from other cancers, TVTs are transferrable sexually between dogs through direct skin-to-skin contact with the tumor. Although the exact source of TVT cells remains unclear, the presence of inclusion bodies in tumor cell cytoplasm indicates the involvement of a viral agent. However, this hypothesis has not been fully revealed. Considering that papillomaviruses, which can cause cancer in many animal species, may play a role as an etiological agent in TVT cases, in this study, the presence of papillomaviruses in samples taken from dogs with TVT was investigated by PCR using different primer pairs. All samples were analyzed with FAP59/64, MY09/11, PVF/FAP64 primer sets targeting the L1 gene of PVs, and CP4/5 primer sets targeting the E1 gene of CPV. PV DNA was detected in 33 (45.20%) of 73 dogs with TVT. After carrying out PCR, positive samples were used for typing using sequence analysis. The MY09/11 primers amplified a 450 bp band in 27 out of 33 positive dogs (81.81%) as single. The MY09/11-CP4/5, PVF/FAP64, and PVF/FAP64-FAP59/64-CP4/5 primer sets showed positivity in 4, 1, and 1 of the animals, respectively. After molecular studies, only the oral biopsy sample of one dog showed a 98-99% nucleotide sequence similarity with the L1 and E2 genes of canine PV type 1 for three primer sets but not for MY09/11. The other sequenced samples showed 70-73% similarity to Leptoncychotes weddellii papillomavirus 6 and 7. This therefore represents a novel PV, indicating that papillomaviruses belonging to different genera may cause TVT cases. These results suggest PV infection may be involved in TVT pathogenesis in sampled dogs with TVT. Comprehensive studies on this subject can both clarify the etiology of these case and also indicate effective practices for controlling the disease, such as treatment and vaccination.

Keywords: Etiology, Dog, Papillomaviruses, Transmissible Venereal Tumors.

DRAFT

ORAL PRESENTATION

Ahşap koruyucu kimyasallar ile yapılan emprenye işleminin uygunluğuna ilişkin yapılan testler ve yorumlanması

Mesut YALÇIN[1*] (*ORCID: https://orcid.org/0000-0002-5181-9484*)

[*1]Düzce Üniversitesi, Orman Fakültesi, Orman Endüstri Mühendisliği Bölümü, Düzce, Türkiye

*Sorumlu yazar e-mail: mesutyalcin@duzce.edu.tr

Özet

Çok çeşitli kullanım yerinde değerlendirilen özellikle doğal dayanıklığı düşük ahşap malzemeler, kullanım yerlerinde karşılaştıkları birçok biyolojik faktöre karşı çeşitli koruyucu kimyasallar ile koruma altına alınarak daha uzun bir servis ömrüne sahip olması sağlanabilmektedir. Koruyucu kimyasalların ahşaba uygulanması çeşitli emprenye yöntemleri ile gerçekleştirilmektedir. Ancak ahşap malzemenin türü ve anatomik yapısı, kullanılan kimyasalın özellikleri, uygulama yöntemi gibi faktörler emprenye işleminin başarısını etkileyen unsurlardır. Ahşap malzemenin emprenye işleminin yeterli olup olmadığına ilişkin değerlendirmeler çeşitli standartlarda belirtilen huşulara göre yapılabilmekte ve özellikle ahşabın kullanım yeri sınıfına ve kendisinden beklenilen servis ömrüne göre minimum retensiyon ve penetrasyon değerleri ile değerlendirilmektedir. Bu değerler tespit edilirken; ağaç türü, dayanıklılık sınıfı ve diri/öz odun kısmı, ahşabın kullanım yeri sınıfı, ağaç malzeme kesit şekli (yuvarlak, kereste) ve minimum servis ömrü beklentisi ve sonuç olarak müşterilerin belirlediği kabul edilebilir kalite seviyesi dikkate alınmalıdır. Retensiyon miktarının tespiti emprenye işleminden önce ve hemen sonra yapılacak ölçümler ile tespit edilmektedir. Penetresyon tespiti ise emprenye işleminden kısa bir süre sonra montaj öncesi veya montaj sonrası kullanım halinde iken tespit edilmesi mümkündür.

Anahtar Kelimeler: Ahşap koruyucu kimyasal, emprenye işlemi, retensiyon testi, penetrasyon testi

DRAFT

ORAL PRESENTATION

Yüksek fruktoz diyetinin neden olduğu metabolik ve hepatik değişikliklerde Mirsetin'in potansiyel etkilerinin araştırılması

Sümeyra Uslu-Bulut[1*] (https://orcid.org/0009-0008-5759-4977),
Nur Banu Bal[1] (https://orcid.org/0000-0001-7865-0917)

[1]Gazi Üniversitesi, Eczacılık Fakültesi, Farmakoloji Anabilim Dalı, Ankara, Türkiye

[*]s_uslu03@hotmail.com

Özet

Kronik fruktoz tüketimi başta karaciğer olmak üzere birçok dokuda yapısal ve işlevsel bozukluklara neden olmaktadır. Mitokondri ile ilişkili hücresel stres cevaplarının fruktoz diyetinin neden olduğu hepatik değişikliklerde rolü olabileceği düşünülmektedir. Son yıllarda flavonoid yapıdaki bileşiklerin metabolik hastalıkların önlenmesinde ve/veya tedavisinde yararlı etkileri olduğu gösterilmiştir. Bu çalışmada güçlü antioksidan aktiviteye sahip olan Mirsetin'in yüksek fruktoz tüketiminin neden olduğu metabolik ve hepatik değişikliklerde potansiyel etkileri araştırılmıştır. Fruktoz, erkek Wistar sıçanlara içme suyu içerisinde (%20, a/h) 15 hafta boyunca verilmiştir. Mirsetin, (50mg/kg/gün) çalışmanın son 6 haftasında oral gavaj yoluyla uygulanmıştır. Sıçanların vücut ağırlıkları, sıvı ve yem tüketimleri haftalık olarak takip edilmiştir. Deney sonunda anestezi altındaki hayvanlardan alınan kan örneklerinden plazma elde edilmiş ve glukoz, trigliserid, LDL ve HDL düzeyleri ölçülmüştür. Karaciğer dokusu çıkarılarak tartılmış ve Western Blot yöntemi ile Dynamin ilişkili protein-1 (DRP1), Mitofusin-2 (Mfn2) ve PTEN ile indüklenen kinaz-1 (PINK1) protein ekspresyonları belirlenmiştir. Fruktozla beslenen sıçanların vücut ağırlığı, karaciğer ağırlığı, karaciğer ağırlığının vücut ağırlığına oranı, plazma glukoz ve trigliserid düzeylerinde anlamlı artış görülürken, Mirsetin uygulaması bu parametreleri etkilememiştir. Plazma HDL kolesterol seviyesi fruktoz tüketen hayvanlarda azalırken, Mirsetin tedavisi bu azalmayı tersine çevirmiştir. LDL kolesterol seviyesi ise tüm gruplarda benzerdir. Ayrıca içme suyu içerisinde fruktoz alan sıçanların yem ve sıvı tüketimleri azalmış, Mirsetin uygulaması ortalama sıvı tüketimini artırmıştır. Mitokondriyal fizyon belirteci DRP1'in ekspresyonu fruktozla beslenen sıçanların karaciğer dokusunda artarken, Mirsetin uygulaması bu artışı önlemiştir. Mitokondriyal füzyon belirteci Mfn2'nin protein ekspresyonu tüm gruplarda benzerdir. Fruktoz tüketen sıçanların karaciğer dokusunda mitofaji belirteci PINK1'in ekspresyonu önemli ölçüde artarken, Mirsetin ile tedavi edilen grupta bu artış gözlenmemiştir. Bulgularımız, yüksek fruktoz diyetiyle beslenen sıçanlarda Mirsetin uygulamasının plazma HDL kolesterol seviyesindeki düşüşü önlediğini ve karaciğerde mitokondri ile ilişkili hücresel stres moleküleri DRP1 ve PINK1'in ekspresyonunu düzenlediğini ortaya koymuştur. Sonuç olarak Mirsetin, fruktoz tüketimine bağlı metabolik bozukluklarda terapötik bir potansiyel olarak dikkate alınabilir.

Anahtar Kelimeler: Fruktoz, Mirsetin, Karaciğer, Mitokondriyal füzyon, Mitokondriyal fizyon, Mitofaji.

DRAFT

ORAL PRESENTATION

Catalytic performance of lipase-inorganic hybrid nanoflowers (L@ihNFs) in the presence of commercial oils/waste oils and detergents/surfactants

Onur Atakisi[1] (*ORCID: https://orcid.org/*0000-0003-1183-6076), Kezban Yildiz Dalginli[2*] (*ORCID: https://orcid.org/*0000-0002-1483-348X.) Melek Ozturkler[2] (*ORCID: https://orcid.org/* 0000-0002-2917-6371)

[1]Kafkas University, Faculte, Faculty Science and Letter, Department of Chemistry, Kars, Turkey
[2] Kafkas University, Kars Vocational High School Kars, Department of Chemistry and Chemical Processing Technologies, Kars, Turkey

*kezbandalginli@gmail.com

Abstract

The objective of this study is to assess the performance of lipase-inorganic hybrid nanoflowers (L@ihNFs) across various parameters, including their activity on different types of commercial liquid/solid oils and waste oils, as well as their compatibility with liquid/solid laundry detergents, surfactants, organic solvents, and variations in pH and temperature. For this, lipase-inorganic hybrid nanoflowers (L@ihNFs) were synthesized using a mixture of lipase enzymes and Cu^{2+} ions. L@ihNFs were characterized by their morphology and chemical point of view by using techniques including SEM and IR. Then, the enzyme activity and stability of L@ihNFs were compared with free enzyme spectrophotometrically at different pH and temperatures. The optimum pHs of free enzyme and L@ihNFs were pH 8 and 9, respectively. The optimum temperatures of the free lipase and L@ihNFs were 40 °C and 50 °C, respectively. Even in the presence of detergents/surfactants, where free lipase could not fully protect its activity, L@ihNFs were able to maintain their activity. L@ihNFs were able to retain 50.38% of their activity in 10 reuses. When their activities in commercial solid /liquid oils and waste oils were evaluated, L@ihNFs was determined to be higher compared to the free enzyme. In addition, when the storage stability was evaluated, it was determined that L@ihNFs retained 50% activity compared to the free enzyme at the end of the 20th day.
Our data reveal that enzyme activities and stability of L@ihNFs were significantly higher compared to the free enzyme and it was determined that L@ihNFs have several novel properties that are important for industrial value. In conclusion, can be a good choice L@ihNFs which can be used for the treatment of waste oil degradation, detergent and biocatalysts applications in the future.

Keywords: Lipase-inorganic hybrid nanoflowers, Biocatalyst, Oils/waste oils, Detergents/surfactants

DRAFT

ORAL PRESENTATION

Detection of Heparanase Enzyme Activity with the Colorimetric Assays

Samet Kocabay[1*] (*https://orcid.org/0000-0002-0120-2910*),
Birnur AKKAYA [2] (*https://orcid.org/0000-0001-9139-1884)*

[1] İnonu University, Science and Art Faculty, Department of Molecular Biology and Genetics, Malatya, Türkiye.
[2] Sivas Cumhuriyet University, Science Faculty, Department of Molecular Biology and Genetics, Sivas, Türkiye.

*Corresponding author e-mail:samet.kocabay@inonu.edu.tr

Abstract

Cancer is a disease in which human deaths are widespread worldwide and is continuing to increase day by day. Cancer development is associated with uncontrollable proliferation of abnormal cells. Uncontrolled growth that took place in epithelial cells can lead to formation of malignant tumors, these are called carcinomas. Some growth factors that are produced from these tumor cells enable heparanase and other different enzymes to be activated and this leads to the degradation of heparan-sulphate proteoglycans on the cell surfaces. The breakdown of heparan-sulfate chains causes growth factors such as FGF and VEGF to be produced, these growth factors increase tumor growth and angiogenesis. To reduce tumor growth and metastasis, various drug designs can be made by modifying chitosan and its derivatives. In this study, the activity of heparanase enzyme was studied by DNS method and WST-1 method. The activity was analyzed by degradation of fondaparunix molecules, a synthetic substrate, in a pH 5 40 Mm acetate buffer at 37°C for 24 hours. A previously synthesized heparin mimic molecule was used as an enzyme inhibitor. The released enzyme products were read in the spectrophotometer with WST-1 and DNS reagents. The results have been saved. Two reagents detected the enzyme products. However WST-1 is better than DNS method due to its sensitivity to ultra-low products. Novelty inhibitor agent showed the inhibition effect on the heparanase at 0,01 mg/ml concentration of inhibitor. According to the results obtained, it was concluded that it can be applied as an alternative to the WST method, since the reducing end released as a result of enzyme activity reacts with the DNS reagent.

Keywords: Heparanase, Colorimetric, DNS, WST-1

*****Thanks to Scientific and Technological Research Council of Turkey (TÜBİTAK) (ID: 222Z122)

DRAFT

ORAL PRESENTATION

Colletotrichum fiorinae'nin neden olduğu antraknoz meyve çürüklüğüne karşı türkiye'deki bazı zeytin çeşitlerinin duyarlılık gruplarının belirlenmesi

Canan Vardar Kor[1*](*ORCID: https://orcid.org/*0000-0003-0535-2040),
Latife Erten Caran[2](*ORCID: https://orcid.org/*0000-0001-5374-401X)

[1-2]Zeytincilik Araştırma Estititüsü, Bitki Sağlığı Bölümü, İzmir, Türkiye

[*] cananvardarkor@gmail.com

Özet

Zeytin, ülkemiz ekonomisi için oldukça önemi bir tarımsal üründür. Son yıllarda pek çok hastalık etmeninden dolayı zeytinde önemli ürün kayıpları yaşanmaktadır, Zeytinde meyve çürüklüğüne neden olan pek çok hastalık etmeninin varlığı bildirilmiştir. Bu hastalık etmenlerinin en önemlilerinden birisi Glomerellaceae familyasına bağlı *Colletotrichum fiorinae*'dir. Bu çalışmada, ülkemizin Akdeniz Bölgesi zeytin çeşitlerinin farklı olgunluk derecelerinde antraknoz meyve çürüklüğüne karşı çeşit dayanıklılığı belirlenmiştir. Denemelerde*, İzmir Kemalpaşa'da Zeytincilik Araştırma Enstitüsüne ait koleksiyon bahçesinde bulunan* Büyük Topakulak, Sarı Ulak, Küçük Topakulak, Çelebi (Silifke), Halhalı (Hatay), Sarı Habeşi, Saurani, Sayfi, Karamani, Elmacık, Yağlık Sarı Zeytin, Maraş No:7 zeytin çeşitlerinin meyveleri kullanılmıştır. *Çalışmada 12 çeşitten alınan, yeşil (1), sarı-yeşil (3) ve siyah (5) olarak 3 farklı olgunluk derecesinde zeytin meyveleri, iklim odası koşullarında Colletotrichum fiorinae'ye karşı hastalık şiddeti belirlenmiştir. Değerlendirmeler,* meyve inokulasyonun 3. gününde lezyonlar gözlemlendiği için 7.günden sonra lezyon gelişimi duyarlılık grubunu değiştirebilecek bir artış olmadığından sonlandırılmıştır. Halhalı (Hatay) %0, Sayfi %2,4, Yağlık Sarı Zeytin %3,2, Saurani %5,2 Maraş No:7 %13,6, Sarı Habeşi %16,4 çeşitlerinin *1.olgunluk derecesindeki meyveleri inokulasyondan 3 gün sonra yüksek dayanıklı ve dayanıklı gruptadır. Diğer çeşitler ise orta duyarı-duyarlı gruptadır. Ancak denemenin 5.ve 7. gün sonunda hastalık şiddetinin sırasıyla Sarı Habeşi'nin %31,8-%83,6 'ya ve Maraş No:7'nin %56,0-%90,4 ile yüksek duyarlı gruptadır. 3. olgunluk dönemindeki meyvelerin, inokulasyonun 3. gününde* Halhalı (Hatay) %0, Saurani%6,4, Büyük Topakulak %8,4 hastalık şiddeti ile yüksek dayanıklı, Maraş No:7 %16,4,Yağlık Sarı Zeytin %17,6 ve Sayfi %22,8 çeşitleri dayanıklıdır. Denemenin 7. gün sonunda ise Halhalı (Hatay) %0'ın yüksek dayanıklı, Saurani'nin %24,4 ile dayanıklı, diğer çeşitlerin duyarlı ve yüksek duyarlı olduğu belirlenmiştir. Çeşitlerin 5. olgunluk derecelerindeki hastalık şiddetleri 3. olgunluk dereceleri ile benzer değerlerdedir. Bu çalışmada, ülkemizin Akdeniz Bölgesi zeytin çeşitlerinin farklı olgunluk derecelerinde antraknoz meyve çürüklüğüne karşı en dayanıklı çeşit Halhalı (Hatay) iken en duyarlı çeşit Sarı Habeşi'dir. Araştırma sonuçlarından elde edilen verilerin bundan sonraki dayanıklılık çalışmalarına ışık tutacağı düşünülmektedir.

Anahtar Kelimeler: Zeytin, antraknoz, hastalık şiddeti, meyve çürüklüğü

DRAFT

ORAL PRESENTATION

Zeytinde yaprak testleri ile *colletotrichum fioriniae'ye* karşı duyarlı ve dayanıklı çeşitlerin belirlenmesi

Latife Erten Caran[1*] *(ORCID: https://orcid.org/*0000-0001-5374-401X),
Canan Vardar Kor[2] (*ORCID: https://orcid.org/*0000-0003-0535-2040)

[1-2]Zeytincilik Araştırma Estititüsü, Bitki Sağlığı Bölümü, İzmir, Türkiye

[*] latife.erten@tarimorman.gov.tr

Özet

Antraknoz (*Colletotrichum spp.*), Glomerellaceae familyasına bağlı dünya çapında zeytin (*Olea europaea* L.) bitkisinin en önemli hastalıklarından biridir. Hastalık etmeni, zeytin yetiştiriciliğini olumsuz yönde etkilemesinin yanısıra, zeytinyağı sektöründe de ciddi sorunların ortaya çıkmasına neden olmaktadır. Bu çalışmada, ülkemizin önemli zeytin çeşitlerinden Trabzon Yağlık, Marantelli, Samsun Tuzlamalık, Halhalı (Hatay), Samsun Yağlık, Taş Arası, Hurmakara, Çakır, Tavşan Yüreği, Çekişte, Girit, Yağ Zeytini, Dilmit, Saurani, Butko, Erkence, Memeli, Eşek (Tekirdağ) ve antraknoza duyarlı olduğu bilinen İspanyol çeşitlerinden Manzallina' nın duyarlılık grupları belirlenmiştir. Denemelerde*, İzmir Kemalpaşa'da Zeytincilik Araştırma Enstitüsüne ait koleksiyon bahçesinde bulunan zeytin çeşitlerinin* genç (≤ 3 aylık yaprak) ve yaşlı (≤1 yaşında yaprak) yaprakları kullanılmıştır. *Yapraklar* 22-24 °C'de 12 saat ışık, 12 saat karanlık fotoperiyotta % 90-95 nem oranında iklim odasında inkübasyona bırakılmıştır. Deneme kurulduktan sonraki 5, 10, 15 ve 20. gün değerlendirme sonuçları skala değerlerine göre yapılıp hastalık şiddeti Towsend-Heuberger formülü ile hesaplanmıştır. Bu sonuçlara göre çeşitlerin *Colletotrichum fioriniae'*ye karşı yapraktaki duyarlılık grupları belirlenmiştir. Zeytin çeşitlerinin genç yapraklarına yapılan inokulasyonun 5. ve 10. gün değerlendirmesinde Marantelli %66, Samsun Tuzlamalık %52,0 ,Memeli % 63 duyarlı, Butko %32,0, Erkence %45,0, Eşek (Tekirdağ) orta duyarlı olup, diğer tüm çeşitler patojene karşı dayanıklıdır. Denemenin 15. gününde ise Trabzon Yağlık %10, Samsun Yağlık %2.0, Hurmakara %7,0 ve Yağ Zeytini %6,0 ile yüksek dayanıklıdır. Yaşlı yapraklar *inokulasyondan 5 gün sonra Colletotrichum fiorinae 'ye karşı Manzallina dışındaki tüm çeşitler yüksek dayanıklı, 10 gün sonra ise dayanıklıdır. Denemenin 20. günü sonunda Trabzon Yağlık-Yağ Zeytini %10 ve Hurmakara'nın %7 ile yüksek dayanıklı, Marantelli %61, Erkence %64 ve Manzalina'nın %95 duyarlı-yüksek duyarlı olduğu belirlenmiştir.* Kimyasal mücadeleye alternatif, yöntemlerin başında hastalıklara karşı dayanıklı çeşitlerin kullanımı gelmektedir. Bu çalışma Türkiye'de, zeytinde *Colletotrichum fioriniae'*ye karşı yapılan ilk yaprakta dayanıklı çeşit çalışmasıdır.

Anahtar Kelimeler: Zeytin, antraknoz, dayanıklılık, yaprak

DRAFT

ORAL PRESENTATION

Determination of optimum modification conditions of glassy carbon electrode with Pt/CNT for HCOOH dehydrogenation

Berdan Ulaş[1,2*] (*ORCID: https://orcid.org/0000-0003-0650-0316*)

[*1]Van Yuzuncu Yil University, Engineering Faculty, Mining Engineering Department, Van, Turkey.
[2]Van Yuzuncu Yil University, Engineering Faculty, Chemical Engineering Department, Van, Turkey.

*berdanulas@yyu.edu.tr

Abstract

Herein, a glassy carbon electrode was modified with carbon nanotube-supported Pt catalysts (Pt/CNT), and its performance for HCOOH dehydrogenation reaction was investigated. Pt/CNT catalysts were polished by chemical reduction and characterized by scanning electron microscopy with X-ray energy dispersive (SEM-EDX), X-ray diffraction (XRD), and inductively coupled plasma mass spectrometry (ICP-MS) methods. Electrochemical measurements were carried out by cyclic voltammetry (CV) and chronoamperometry (CA). Independent parameters affecting specific activity of Pt/CNT for HCOOH dehydrogenation were optimized with response surface methodology (RSM). For the modification conditions of GCE with Pt/CNT, the amount of catalyst sludge transferred to the electrode surface, ultrasonification time, and drying time of the catalyst sludge transferred to the GCE surface were selected as independent parameters, and the optimum values were determined as 9 µl, 50 min, and 9.07 min, respectively. The specific activity for HCOOH dehydrogenation under optimum conditions was obtained as 12.8 mA cm^{-2}.

Keywords: Formic acid, Dehydrogenation, Response surface method, Platinum.

DRAFT

ORAL PRESENTATION

Evaluation of the land use/land cover (LULC) change effects on land surface temperature (LST): A case study of Kağıthane Watershed

Betül Uygur Erdoğan[1*] (*ORCID: https://orcid.org/0000-0002-8179-4564*), Reyhan Sağlam[1] (*ORCID: https://orcid.org/0000-0002-8772-9704)*, Vildan Rabia Yar[3] (*ORCID: https://orcid.org/0000-0002-6514-1387)*

[*1] Istanbul University-Cerrahpasa, Faculty of Forestry, Department of Watershed Management, Istanbul, Turkey.
[2] Istanbul Medipol University, General Coordination of Foreign Languages, Istanbul, Turkey.

*Corresponding author e-mail:uygurb@iuc.edu.tr

Abstract

Along with the increasing population, mis-land use, natural resource destruction and loss have been experienced. In this process, the land use/land cover (LULC) changes in urban areas and the increase in impervious surfaces play an important role. In this study, it has been aimed to reveal how the land surface temperature (LST) changed according to the land uses in the Kağıthane watershed between 2002-2021, which is located in the province of Istanbul. In this context, the satellite images of Landsat 5 for 2002 and Landsat 8 for 2021 were used and analyses were carried out with remote sensing (RS). When the LULC results of the study were examined, the major increase was observed in urban areas with 1014.7 ha and the major decrease was in forest areas with 933.3 ha. In addition, the highest LST values related to LULC were observed in urban and open areas while the lowest values were observed in forest areas and water bodies. Besides, the lowest increase in LST was 0.6 °C in forest areas, whereas the highest increase was detected in urban areas with 2.6 °C. These results can be considered as a clear indication of the cooling effect of the forests in the northern part of the watershed. Therefore, this study has shown the importance of protecting the forest areas in the watershed from fragmentation and how necessary it is to plan forests or green areas in the urban areas in order to increase the cooling effect in the urban climate.

Keywords: Land use/land cover (LULC), Land surface temperature (LST), Forest areas, Urban watersheds, Cooling effect

DRAFT

ORAL PRESENTATION

Aromatik sülfonik asit katalizörler varlığında levülinik asit üretimine biyokütle içeriğinin etkisi

Doğan Emre Yüksel[1*] (*ORCID: https://orcid.org/0000-0002-8913-4501*),
Levent Ballice[2] (*ORCID: https://orcid.org/0000-0002-3137-1352*)

[*1]Ege Üniversitesi, Aliağa Meslek Yüksekokulu, Kimya Teknolojisi, İzmir, Türkiye
[2] Ege Üniversitesi, Mühendislik Fakültesi, Kimya Mühendisliği, İzmir, Türkiye

*Sorumlu yazar e-mail: dogan.emre.yuksel@ege.edu.tr

Özet

Dünyada yılda oluşan lignoselülozik biyokütlenin %97'ye yakın kısmı değerlendirilmemektedir. Bu büyük potansiyelden kimyasal ve biyokimyasal yöntemlerle sıvı ve gaz yakıtlar ile çeşitli kimyasalları elde etmek için laboratuvar ve pilot ölçekte yoğun çalışmalar yapılmaktadır. Lignoselülozik biyokütleden üretilebilecek 12 önde gelen kimyasaldan (Building Blocks) oluşan bir liste 2004 yılında yayınlanmıştır. Bu kimyasallar çok yakın bir gelecekte petrokimya rafinerilerinin yerini alması planlanan biyorafinerilerin temelini oluşturacaktır. Bu 12 kimyasaldan biri de LEVÜLİNİK ASİT (LA)'dir. Bu çalışmada, ülkemiz için önemli olan biyokütlelerin atıklarından, aromatik sülfonik asit katalizörleri varlığında otoklav reaktör sistemi kullanılarak LA oluşum koşulları araştırılmıştır. Belirlenen parametreler için optimum koşulların belirlenmesi amacıyla, selüloz-hemiselüloz içeriği yüksek olan aspir sapı ile çalışmalara başlanması uygun görülmüştür. Reaksiyon sıcaklığı (120, 140, 160, 180, 200 ve 220°C), pH (0,5, 1,0 ve 1,5), solvent / biyokütle oranı (ağırlıkça 10, 15 ve 20) ve katalizör tipinin (BSA, PTSA ve LABSA) maksimum LA verimi üzerindeki etkileri incelenmiştir. Minitab-19 Yazılımı ile oluşturulan L18 Taguchi deney tasarımına göre yapılan denemelerde maksimum LA verimi için optimum koşullar, 200°C sıcaklıkta 0.3 M konsantrasyonda para-toluensülfonik asit (PTSA) ve çözücü/biyokütle oranı 20 olarak bulunmuştur. Elde edilen bu sonuçlara göre ayçiçeği sapı, mısır koçanı, fındık kabuğu ve palamut denemeleri optimum noktalarda gerçekleştirilmiştir. Mısır koçanı ile yapılan denemelerde maksimum LA verimi 120. dakikada 94 g/kg olarak bulunmuştur. Bu değer optimum koşullarda aspir sapında bulunan değerden (88 g/kg) daha yüksektir. Ayçiçeği sapı denemesinde 120. dakikada maksimum LA verimi 49.38 g/kg olarak bulunmuştur. Bu değerin aspir sapı ile optimum koşullarda bulunan değerden (88 g/kg) oldukça düşük olması dikkat çekmektedir. Fındık kabuğu ve palamut ile yapılan deneylerde maksimum LA verimi sırasıyla 70 g/kg (20. dakikada) ve 61 g/kg (20. dakikada) olarak bulunmuştur. Bu değerler, optimum koşullarda aspir sapında bulunan değerden (88 g/kg) daha düşüktür.

Anahtar Kelimeler: Hidrotermal dönüşüm, levulinik asit, lignoselülozik biyokütle, Taguchi yaklaşımı, aromatik sülfonik asitler.

DRAFT

ORAL PRESENTATION

Synthesis and characterization of poly (ε-caprolactone-*b*-4-vinyl benzyl-*g*-ethylene glycol) block-graft copolymers using reversible addition/fragmentation chain transfer polymerization (RAFT), ring opening polymerization (ROP) and "click" chemistry methods

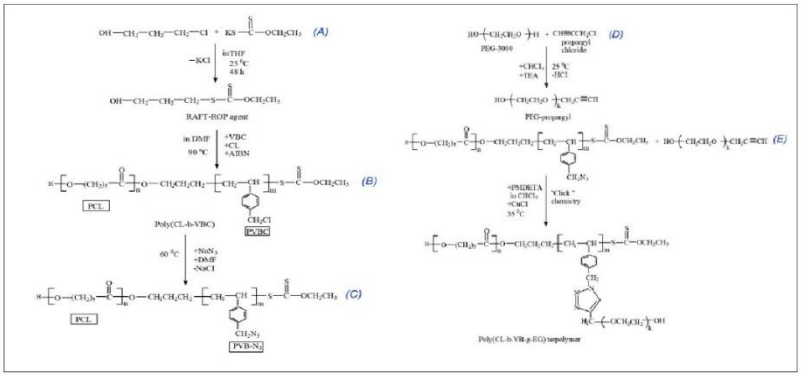
Cengiz Aykaç*[1], Melahat Göktaş[2], Temel Öztürk[3], Bedrettin Savaş[4,]

[1]Van Yüzüncü Yıl University, Institute of Science, Van, 65080 Turkey
[2]Van Yüzüncü Yıl University, Faculty of Education, Department of Chemistry, Van, 65080 Turkey
[3]Giresun University, Department of Chemistry, Giresun, 28200 Turkey
[4]Kars Vocational School, Kafkas University, 36100 Kars, Turkey

*****Corresponding author: E-mail: cengzaykac@gmail.com

Abstract

Reversible addition/fragmentation chain transfer (RAFT) polymerization is a unique method for the synthesis of polymers of desired molecular weights with low polydispersity **[1]**. Ring opening (ROP) polymerization is widely used in the polymerization of lactones **[2]**. The "Click" chemistry method introduced by Sharpless et al. is a unique technique for copolymer synthesis and has attracted great attention from many researchers **[3]**. In this study, well-known poly(ε-caprolactone-*b*-4-vinylbenzyl-*g*-ethyleneglycol) [P(ε-CL-*b*-VB-*g*-EG)] block-graft terpolymer were synthesized by recycling addition/fragmentation chain transfer (RAFT) polymerization, ring-opening polymerization (ROP) and click chemistry method. For this purpose; in the first step, a novel RAFT-ROP agent was synthesized by the reaction 3-chloro-1-propanol and potassium ethyl xanthogenate salt. In the second step, poly(ε-caprolactone-*b*-4-vinyl benzyl chloride) P(ε-CL-*b*-VB) block copolymer was synthesized by RAFT polymerization of 4-vinyl benzyl chloride and ROP of ε-caprolactone using synthesized a novel RAFT-ROP initiator in the one-step. In the third step, poly(ε-caprolactone-*b*-4-vinyl benzyl chloride)-N_3 was obtained by reaction synthesized poly(ε-caprolactone-*b*-4-vinyl benzyl chloride) block copolymer and NaN_3. On the other hand, PEG-propargyl (alkyne-terminated-PEG) was obtained by the reaction polyethylene glycol (PEG-3000) with propargyl chloride. In the last step, [P(ε-CL-*b*-VB-*g*-EG)] block-graft terpolymer were obtained by the reaction PEG-propargil and poly(ε-caprolactone-*b*-4-vinyl benzyl chloride)-N_3 via the "click" chemistry method. The synthesized products were characterized by using multi-instruments such as GPC, ^{1}H-NMR, FT-IR, DSC ve SEM.



Scheme 1: The chemical synthesis reaction of RAFT-ROP initiator ***(A)***, synthesis mechanism of poly(CL-*b*-VBC) block copolymers ***(B)***, chemical synthesis reaction of poly (CL-*b*-VB)-N3 ***(C)***, synthesis of PEG-propargyl ***(D),*** synthesis mechanism of poly(CL-*b*-VB-*g*-EG) block-graft terpolymers ***(E).***

Keywords: "click" chemistry, Reversible addition/fragmentation chain transfer (RAFT), block-graft terpolymer

References: **1.** Chiefari, J.; Chong, YK.; Ercole, F.; Krstina, J.; Jeffery, J.; Le, TPT.; Mayadunne, RTA.; Meijs, GF.; Moad, CL.; Moad, E.; Rizzardo, E.; Thang, SH. (1998) Macromolecules 16:5559-5562.
2. Göktaş M, Aykaç C, Öztürk T (2022) J Chem Sci 134:73.
3. Kolb, HC.; Finn, MG.; Sharpless, KB. (2001) Angew Chem Int Ed. 40:2004-2021.

ORAL PRESENTATION

Removal of heavy metals from wastewater using activated carbon synthesized from coffee silverskin

Aizada Myrzageldi[1*], Nurcan Tuğrul[2]

[*1] Yildiz Technical University, Faculty of Chemical and Metallurgical Engineering, Department of Chemical Engineering, Istanbul, Turkey.
[*2] Yildiz Technical University, Faculty of Chemical and Metallurgical Engineering, Department of Chemical Engineering, Istanbul, Turkey.

*Corresponding author e-mail: aizada.myrzageldy@gmail.com

Abstract

The aim of this study was to produce a low-cost adsorbent using coffee wastes. Activated carbon prepared from coffee silverskin will be used for heavy metal adsorption from wastewater. The present study describes a synthesis of the activated carbon by the combination of chemical activation and microwave treatment in the oven without radiation leakage, which is a safe, simple, and fast method compared to the hydrothermal pyrolysis. The combination of microwave pyrolysis with chemical activation and the use of lignocellulosic waste (coffee silverskin) was investigated as an alternative method to the hydrothermal pyrolysis. The zinc chloride and phosphoric acid were used as an activation agent followed by microwave treatment at 600W for 5 min with 30 sec time intervals. When the zinc chloride was used at 1:4, 1:5 molar rations, maximum metal adsorption capacity of the activated carbon was 76,95 mg/g, 115,1 mg/g for 2 hours. When phosphoric acid was used in similar molar rations the maximum adsorption capacity of activated carbon was registered as 115,2 mg/g, 100 mg/g for 2 hours respectively. The removal of heavy metal ions was studied by UV-spectroscopy and had shown 96% and 82,3%. Additionally, the effect of the temperature, time and concentration on the adsorption were investigated. These results illustrate that coffee silverskin can be used as a low-cost adsorbent for the removal of heavy metal ions from wastewater.

Keywords: Activated carbon, Microwave treatment, Wastewater, Heavy metals.

DRAFT

ORAL PRESENTATION

Should chitosan films be referred to as natural or semi-synthetic?

Behlül Koç Bilican[1*] (*ORCID: https://orcid.org/0000-0001-9943-771X*)

[*1]Aksaray University, Faculty of Science and Letters, Department of Molecular Biology and Genetics, Aksaray, Turkey.

*Corresponding author e-mail: behlulkoc.bk@gmail.com

Abstract

In recent years, there has been a growing trend toward the production of biopolymer-based films due to consumer demand. This shift is driven by consumers' increasing awareness of the harmful effects of non-biobased films on human health and the detrimental impact of non-biodegradable materials on the environment. In the literature, natural films made from biopolymers such as gelatin, alginate, cellulose, and chitosan have emerged as alternatives to petroleum-based plastics. Among these, chitosan-based films stand out prominently. Chitosan, derived from the deacetylation of chitin, is a biodegradable, non-toxic, antimicrobial, antioxidant, and biocompatible polysaccharide. Its unique properties have led to a growing interest in chitosan. Films made from chitosan find applications in various fields, including food, medicine, and agriculture, among others. However, it is worth noting that in the existing literature, all chitosan films are typically prepared by dissolving chitosan in a weak acid and forming it into a gel. Therefore, these films should be classified as synthetic or semi-synthetic chitosan films.

Keywords: Chitosan film, Natural films, Semi-synthetic films

DRAFT

ORAL PRESENTATION

Relationship of Diabetes Mellitus and Cancer

Filiz YILDIRIM[1], Hasan KARAGEÇİLİ[2]

[1]Department of Internal Medicine, Polatlı Duatepe State Hospital, Ankara, Turkey, drfyildirim@yahoo.com;

[2]Siirt University, Faculty of Healthy Sciences, Siirt, Turkey

Abstract

Cancer is the most dangerous disease of our contemporary day, and it ranks high on the list of causes of human mortality. While cancer is the leading cause of death in wealthy societies, it is steadily growing in emerging countries. Diabetes mellitus affects 250 million individuals globally. This figure is expected to reach 380 million during the next 20 years. Cancer rates in diabetes people are greater than in non-diabetic patients, according to studies. Furthermore, diabetes medicines, food, and metabolism control level smay all play a part as defining variables in the link with cancer. Diabetes people are more likely to develop malignancies of the liver, pancreas, endometrial, colon and rectum, breast, bladder, prostate, and esophageallining. Cancer in type 2 diabetes patients is caused by three factors: (1) direct effects such as hyperglycemia, hyperinsulinemia, and insulin resistance, inflammation (IL-6 secreted from adipocytes, TNF-α, PAI-1, adiponectin, and leptin), (2) indirect effects such as advanced age, obesity, sedentary lifestyle, malnutrition, and (3) incorrect therapies. The examination of the relationship between cancer and diabetes revealed that diabetic people had a greater risk of many forms of cancer than healthy ones. Diabetics are about twice as likely as healthy people to develop liver, pancreatic, and uterine malignancies. Given that the control group in this research includes people who have never been diagnosed with diabetes, it is reasonable to assume that the risk is higher. In the case of prostate cancer, the scenario is just the contrary. Diabetic persons were shown to have a decreased chance of acquiring prostate cancer than healthy ones. The hormone testosterone is thought to play a role in the development of prostate cancer. The comparatively low amount of testosterone hormone in diabetic people lowers the chance of acquiring this malignancy. Diabetic medications also have a role in this. Some diabetic medicines have been demonstrated to lessen the chance of acquiring cancer. Metformin, the first-line medication for diabetes, inhibits proliferation and prevents cancer. In recent research, the prevalence of cancer in insulin users has raised concerns regarding the use of insulin in the treatment of diabetes. In conclusion, the incidence of cancer in diabetes individuals is higher than in the general population. Routine cancer tests should be done more carefully in the first 5 years, especially in the event of new diabetes appearing at an elderly age.

Keywords: Diabetes mellitus, Cancer, Metformin

DRAFT

ORAL PRESENTATION

Simple and green colorimetric method for Iron (III) determination using *Helichrysum arenarium* Infusion

Batuhan Yardımcı[1*] (*ORCID: https://orcid.org/0000-0002-4041-8183)*

[*1]Zonguldak Bülent Ecevit University, Science and Technology, Application and Research Center (ARTMER), Zonguldak, Turkey.

*Corresponding author e-mail: batuhan.yardimci@beun.edu.tr

Abstract

This study presents a novel and environmentally friendly approach for the determination of Iron (III) utilizing *Helichrysum arenarium* infusion. The increasing demand for green analytical methods has led to the exploration of natural sources for reagents. In this context, *Helichrysum arenarium*, known for its rich chemical composition [1-3], has been harnessed for its potential as a colorimetric agent in iron quantification. According to the principles of green chemistry, this method is simple, eco-friendly, and simple to implement. A colorimetric reaction occurs between Iron (III) and the infusion, resulting in a measurable color change that corresponds to the concentration of the analyte. This reaction was optimized under various conditions to achieve the best analytical performance. The developed method demonstrates a linear range for Iron (III) quantification, with a good correlation coefficient. Moreover, the method showcases exceptional sensitivity, as indicated by the low limit of detection and quantification values.

As a result, an efficient and environmentally friendly colorimetric method has been developed for determining iron (III) using Helichrysum arenarium infusion as a reagent. The method's efficiency, accuracy, and compatibility with green analytical principles make it a promising alternative for iron quantification in various applications. Incorporating natural reagents like *Helichrysum arenarium* infusion underscores the potential of sustainable practices in analytical chemistry.

Keywords: Green analytical chemistry, Iron(III)-polyphenol complex, colorimetry, spectrophotometry.

References :

[1]. Grinev, V. S., Shirokov, A. A., Navolokin, N. A., Polukonova, N. V., Kurchatova, M. N., Durnova, N. A., ... & Maslyakova, G. N. (2016). Polyphenolic compounds of a new biologically active extract from immortelle sandy flowers (Helichrysum arenarium (L.) Moench.). *Russian Journal of Bioorganic Chemistry*, *42*, 770-776.

[2]. Gradinaru, A. C., Silion, M., Trifan, A., Miron, A., & Aprotosoaie, A. C. (2014). Helichrysum arenarium subsp. arenarium: phenolic composition and antibacterial activity against lower respiratory tract pathogens. *Natural product research*, *28*(22), 2076-2080.

[3]. Czinner, E., Hagymasi, K., Blazovics, A., Kery, A., Szőke, É., & Lemberkovics, E. (2000). In vitro antioxidant properties of Helichrysum arenarium (L.) Moench. *Journal of ethnopharmacology*, *73*(3), 437-443.

DRAFT

ORAL PRESENTATION

Antioxidant activity and total phenolic and flavonoid content of *Pistacia khinjuk* Stocks ethanolic extract

Neşe Eray Vuran[1*] (*ORCID: https://orcid.org/.0000-0001-6387-1493*)

[*1]Van Yüzüncü Yıl University, Faculty of Science, Department of Molecular Biology and Genetic, Van, Turkey.

*Corresponding author e-mail: neseeray@yyu.edu.tr

Abstract

Pistacia khinjuk Stocks is a species of plant in the Anacardiaceae family, which is contained commercially important species. *Pistacia* genus has been used in traditional medicine to treat many diseases duo to their therapeutic properties. The aim of the study is to reveal the total phenol (TPC) and flavonoid content (TFC) and antioxidant activity of ethanolic extract of *Pistacia khinjuk* Stocks. The plant was collected from Siirt/Turkey and diagnosed by Assoc. Prof. Dr. Süleyman Mesut Pınar. A voucher specimen was stored at Van Yüzüncü Yıl University's herbarium (VANF) (Herbarium number: 164257).The ethanolic extract of the plant was prepared. Total phenolic (TPC) and flavonoid (TFC) contents were measured spectrophotometrically. Antioxidant potentials of the extracts were evaluated by DPPH (1, 1-diphenyl-2-picrylhydrazyl) and ABTS [2,2′-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)] radical scavenging capacity tests. The extraction yield was 5, 5 % (w/w) for the ethanolic extracts of *Pistacia khinjuk*. TPC was calculated as 329± 9, 68 µg GAE/mg. TFC of the ethanolic extracts were determined as 416,6±9, 99 QE/ mg dry extracts and 105± 8,85 µg CE/ mg dry extracts according to Quercetin (QE) and Catechin (CE) standards, respectively. IC50 values were determined as 0, 56 and 0, 46 mg/ml for DPPH and ABTS+ assay, respectively. As a result, the extraction yield was low due to the hard nature of the seed. A different extraction method should be used to increase the extraction yield in the plant. Thanks to its high TPC and TFC, the ethanolic extract of *Pistacia khinjuk* Stocks has strong antioxidant properties. This situation must also have resulted from the polarity of the extraction solvent which affects the contents of the plant extracts. Duo to the high antioxidant activity, *Pistacia khinjuk* Stocks may have considered as an experimental material to protect against the oxidative stress-related diseases.

Keywords: Antioxidant activity, Flavonoid, Phenolic, *Pistacia khinjuk*

DRAFT

ORAL PRESENTATION

Mikrodizi Verilerinde Eksik Veri Atama ve Normalizasyon Sıralamasının Sınıflama Performansına Etkisi

Asena Ayça Özdemir [1,*](*ORCID: https://orcid.org/ 0000-0002-0108-1880*),
Damla Hazal Sucu[2](*ORCID:https://orcid.org/ 0000-0002-8823-3727*)

[*1]Mersin Üniversitesi, Tıp Fakültesi, Tıp Eğitimi Anabilim Dalı, Mersin, Türkiye
[2]Mersin Üniversitesi, Tıp Fakültesi, Biyoistatistik ve Tıbbi Bilişim Anabilim Dalı, Mersin, Türkiye

*Sorumlu yazar e-mail: a.aycaozdemir@hotmail.com

Özet

Mirodizi, genlerin ifade seviyelerini aynı anda ölçmeye yarayan bir teknolojidir. Binlerce genin aynı anda ölçümü yapılabilen bu teknolojide cihaz ya da çevre kaynaklı eksik gözlemler oluşabilmektedir. Bu eksik gözlemlerin oranı ise değişkenlik göstermektedir. Literatürde genellikle ilk olarak eksik gözlemlere atama işlemi yapılmakta ve sonrasında diğer analizlere devam edilmektedir. Ayrıca mikrodizi verilerine herhangi bir analiz yapılmadan önce normalizasyon uygulanması gerekmektedir. Biz de araştırmamızda mikrodizi verilerinde eksik veri atama ve normalizasyon uygulaması sıralamasında sınıflama performanslarında gözlenen değişkenliği değerlendirmeyi amaçladık. Bu amaçla R programı Madsim paketinde gen sayısı 100, 500 ve 1000, örnek genişlikleri ise 10, 20, 40,
100 ve 200 olacak şekilde sentetik mikrodizi verisi üretilmiştir. Oluşturulan veri setlerinde %1, %5, %10, %20 ve
%50 Tamamen Rastgele Kayıp yapısına uygun olarak eksik veriler oluşturulmuştur. Her durum 1000 kez tekrar olacak şekilde simüle edilmiştir. Eksik veri oluşturulan veri setlerine ilk olarak KNN ve CART ile atamalar yapılmış sonrasında ise Kantil normalizasyon uygulanmıştır, ikinci durumda ise önce normalleştirme sonra eksik veri atama adımı gerçekleştirilmiş ve sınıflama performansları Tek Katmanlı İleri Beslemeli Sinir Ağı ile değerlendirilmiştir. Gen sayısı 100 ve 500 iken önce normalizasyon sonra atama yönteminin kullanılması durumunda daha yüksek doğruluklar elde edilirken, gen sayısı 1000 olduğunda önce atama sonra normalizasyonun uygulanması daha yüksek performans göstermiştir. Örnek genişliklerine ve gen sayısına göre doğruluk oranları oldukça değişiklik göstermekte olup, genel olarak CART algoritmasının KNN'e göre daha yüksek performansa sahip olduğu sonucuna ulaşılmıştır. Bu sebepler doğrultusunda, mikrodizi verileri gibi örnek genişliği küçük, değişken sayısı fazla olan veri setlerinde klasik bilinen eksik veri atama ve normalizasyon sıralamasının aksine önce normalizasyon sonrasında eksik veri ataması uygulanarak sonuçların karşılaştırılması, verinin yapısına göre değerlendirme yapılarak analiz performanslarının oldukça önemli ölçülerde değişebileceği düşünülmektedir.

Anahtar Kelimeler: Mikrodizi, eksik veri, normalizasyon

DRAFT

ORAL PRESENTATION

Green synthesis of silver, zinc, and calcium nanoparticles from a psychrophilic bacterium

Simge Emlik[1]* *(https://orcid.org/0000-0002-2299-6158)*,
Sevgi Marakli[1] *(https://orcid.org/0000-0001-5796-7819)*

[1]Yildiz Technical University, Faculty of Arts and Sciences, Department of Molecular Biology and Genetics, 34220, Istanbul, Türkiye

simge.emlik@std.yildiz.edu.tr

Abstract

Nanotechnology deals with nano-sized particles ranging from 1 to 100 nm. Nanoparticles (NPs) are used in a wide variety of fields such as biology, medicine, agriculture, chemistry, and engineering. There are various synthesis methods and green synthesis is a fairly new and rapidly developing technique among them. When compared to other procedures, green synthesis avoids different problems such as reaction complications of chemical methods, high cost, environmental pollution, toxic effects, and safety. Bioactive agents such as plants, bacteria, fungi, etc. are utilized for green synthesis. Psychrophiles, as a subgroup of extremophiles, thrive in extremely cold habitats within the temperature range of -20°C to -15°C and are also used for NP preparation. We aim to synthesize silver (Ag), zinc (Zn), and calcium (Ca) NPs from *Psychrobacter* sp. *TaeBurcu001* which was identified by our group in Antarctica. The formations of NPs were observed by the color change in the solution. In addition, NPs were also analyzed with SEM and Zetasizer. Sizes of Ag NPs in SEM were in the range of 10-50 nm, ZnO NPs were in the range of 80-150 nm and CaO NPs in the range of 100-250 nm. Zeta potential measurements also showed negative charge in the averages as −15.8 mV, −22.8 mV, and −12.8 mV for Ag NP, ZnO NP, and CaO NP, respectively. We know that NPs can be different shapes, sizes, and stability when obtained from different species. In addition, these different characteristics have been used for different purposes in numerous areas. Our analyses continue to determine the potential use of obtained NPs, especially in agriculture.

Keywords: Nanoparticle, Green synthesis, Psychrophilic bacteria, Antarctica

DRAFT

ORAL PRESENTATION

Bilecik İli Küre ve Çaltı Köyleri Florası İçin Ön Çalışmalar

Mertcan Zengin[1*] (*0009-0003-3980-3847*), Ebru Ataşlar[2*] (*0000-0001-5755-4256*)

[*1]Eskişehir Osmangazi Üniversitesi, Fen Bilimleri Enstitüsü, Biyoloji Anabilim Dalı, 26040, Eskişehir, Türkiye

[*2]Eskişehir Osmangazi Üniversitesi, Fen Fakültesi, Biyoloji Bölümü, 26040, Eskişehir, Türkiye

*zenginmertcan23@gmail.com

Özet

Ülkemizin geçit kuşağı bölgesi üzerinde bulunan Bilecik ili, bitki çeşitliliği açısından önemli bir yere sahiptir. Bu sayede birçok bitki türüne ev sahipliği yapmaktadır. Bölgenin, Sakarya Nehri'nin kıyısında konumlanmış ve birçok medeniyete ev sahipliği yapmış olması biyoçeşitlilik açısından önemlidir. Çalışmada, Bilecik ilinin Söğüt ilçesinin floristik yapısını incelenmek için ilçenin biyoçeşitlilik açısından önemli bir kısmını oluşturan Küre ve Çaltı Köyleri ile çevresindeki bölge seçilmiştir. Bölgede, 2023 yılının Nisan ve Ağustos ayları arasında on beş günlük periyotlar halinde arazi çalışmaları gerçekleştirilmiş olup bitkilerin yaprak, çiçek ve meyve/tohum durumlarından örnekler toplanılmış ve tür tayinleri yapılmıştır. Yapılan arazi çalışmaları sonucunda bölgede floristik yapıya ait tür sayısı açısından zengin bazı familyalar şu şekilde sıralanmıştır: Asteraceae, Brassicaccae, Fabaceae, Moraceae, Oleaceae, Rosaceae.

Anahtar Kelimeler: Flora, Biyoçeşitlilik, Bilecik, Söğüt

DRAFT

ORAL PRESENTATION

Biyoaktif bileşiklerin glikasyon ürünleri üzerine etkisi

Esranur Seyyar[1*] (*ORCID: https://orcid.org/0000-0002-6773-5541*),
Merve Tomas[1] (*ORCID: https://orcid.org/0000-0003-1057-7914*)

[*1]İstanbul Sabahattin Zaim Üniversitesi, Müh. ve Doğa Bil. Fakültesi., Gıda Mühendisliği Bölümü, İstanbul, Türkiye

*esranur.seyyar@std.izu.edu.tr

Özet

Beslenme alışkanlıklarındaki değişimler ve günümüzdeki diyetlerin büyük ölçüde ısıyla işlenmesi beraberinde yüksek seviyelerde glikasyon ürünlerine maruziyetini de arttırmaktadır. Glikasyon ürünleri, indirgeyici şekerler ile proteinlerin, lipidlerin veya nükleik asitlerin serbest amino grupları arasındaki enzimatik olmayan bir reaksiyon yoluyla oluşur. Bu reaksiyon aynı zamanda Maillard veya esmerleşme reaksiyonu olarak da bilinmektedir. Maillard reaksiyonuna dayanan glikasyon ürünleri üç aşamada gerçekleşmektedir. Birinci aşama glukoz enzimatik olmayan bir yolla protein, lipit veya DNA'nın serbest bir amino asidine bağlanarak Schiff bazı oluşturur. İkinci aşamada ise Shiff bazı, Amadori ürünlerine (daha stabil ve geri dönüşümlü olan, erken glikasyon ürünleri olarak da bilinir) dönüşür. Üçüncü aşamada ise Amadroi ürünleri bileşiklerin dehidrayonu ve oksidasyonu ile haftalar veya aylar süren bir zamanda geri dönüşümü olmayan glikasyon ürünleri oluşur. Mevcut araştırmalar, farklı gıda bileşenlerinin (şeker, protein, yağ, su vb.), saklama yöntemlerinin (depolama sıcaklığı, süresi, nem, gaz vb.) ve işleme yöntemlerinin (buharda pişirme, kaynatma, kızartma, fırınlama, kavurma vb.) gıdalardaki glikasyon ürünlerinin oluşumunu etkilediğini bildirmektedir. Gıda yoluyla alınan glikasyon ürünlerinin bir kısmı insan vücudunda birikerek bir dizi kronik hastalığa neden olmaktadır. İnsan vücudunda ve işlenmiş gıdalarda glikasyon ürünlerin oluşumunu engelleyebilen biyoaktif bileşikler, birçok bitki de bulunmaktadır. Son yıllarda bazı bitki ekstraktlarının glikasyon ürünleri oluşumu üzerindeki etkileri değerlendirilmektedir. Bu bitki ekstraktlarının çoğunun glikasyon ürünlerinin oluşumu üzerindeki önleyici etkilerinin esas olarak içerdikleri büyük miktardaki fenolik antioksidanlardan kaynaklandığı belirtilmektedir ve bunların glikasyon sürecinde serbest radikal oluşumunu engellemeleri ve ardından proteinlerin modifikasyonunun engellenmesi, anti glikasyon aktivitelerine aracılık eden ana mekanizmalar olarak kabul edilmiştir. Bu çalışmada amaç biyoaktif bileşiklerin, gıdalarda bulunan glikasyon ürünleri üzerine etkilerini incelemektir.

Anahtar Kelimeler: Biyoaktif bileşikler, glikasyon ürünleri, anti glikasyon aktivite

DRAFT

ORAL PRESENTATION

The effect of caffeic acid phenethyl ester (CAPE) on the viability, apoptosis, and vascular endothelial growth factor (VEGF)-A and VEGF-B gene expression in human umbilical vein endothelial cells

Ecem Kaya-Sezginer[1*] (*ORCID: https://orcid.org/0000-0002-8490-6293*), Omer Faruk Kırlangıc[2] (*ORCID: https://orcid.org/0000-0003-0219-3312*), Taner Ozgurtas[3] (*ORCID: https://orcid.org/0000-0003-1110-6671*)

[*1]Ankara University, Faculty of Pharmacy, Department of Biochemistry, Ankara, Turkey.
[2]Ankara University, Vocational School of Health, Ankara, Turkey.
[3]University of Health Sciences, Gulhane Faculty of Medicine, Department of Medical Biochemistry, Ankara, Turkey.

*Corresponding author e-mail: ecemkaya@ankara.edu.tr

Abstract

A phenolic compound caffeic acid phenethyl ester (CAPE) is a main substance of propolis with its antiinflammatory, antiviral, and anti-tumor properties. Human umbilical vein endothelial cell (HUVEC) is used model for research on human endothelium. This study was designed to evaluate the effect of different concentrations of CAPE on HUVECs. The effects of CAPE on cell viability and apoptosis were determined by MTT and annexin V/7-aminoactinomycin D (7-AAD) assay. The gene expression levels of vascular endothelial growth factor (VEGF)-A and VEGF-B were measured using RT-PCR. Our results demonstrated that CAPE treatment at 7.5 µM and higher concentrations for 24 h significantly suppressed the viability of HUVECs with IC_{50} value of 25.3 µM. 48 h treatment of HUVECs with CAPE reduced cell proliferation between 5-40 µM with IC_{50} value of 19.8 µM. The percentage of necrotic cells treated with 19.8 µM CAPE for 48 h was remarkably higher than control group ($p<0.05$). No significant difference was found in the percentage of early and late apoptotic cells after treatment with IC50 value of CAPE (19.8 µM) for 48 h. The gene expressions of VEGF-A and VEGF-B were partially decreased after 48 h treatment with CAPE at 19.8 µM concentration ($p=0.061$ and $p=0.117$, respectively). Treatment with CAPE revealed a significant cytotoxicity and induced necrosis in HUVECs possibly via a slight downregulation of VEGF-A and VEGF-B gene expression. CAPE could demonstrate dose-dependent effects on endothelial cells.

Keywords: caffeic acid phenethyl ester, endothelial cells, vascular endothelial growth factor, cell proliferation

DRAFT

ORAL PRESENTATION

Exploring the Function of a Candidate Calcium-Binding Protein in Cilio-biogenesis in C. elegans

Sebiha Çevik-Kaplan*

*Rare Disease Laboratory, Department of Molecular Biology and Genetics, School of Life and Natural Sciences, Abdullah Gul University, Kayseri, Turkey

Abstract

Microtubule-structured cilia, ubiquitously present in diverse organisms, including Chlamydomonas reinhardtii, nematode C.elegans and humans ect., serve pivotal roles in essential cellular processes encompassing both motility and sensory functions. The significance of cilia in these critical cellular events is underscored by the emergence of ciliopathies in humans, which stem from structural and functional aberrations within these cellular organelles. These ciliopathies can be attributed to genetic mutations affecting ciliary genes or genes modulating ciliary function, as well as disruptions in protein transport mechanisms vital for cilia functionality. Symptoms include retinal degeneration, polycystic kidney disease, obesity, and polydactyly. This phenotypic heterogeneity complicates the comprehension of ciliopathy pathogenesis and, consequently, the development of effective therapeutic interventions. Consequently, the investigation of mutations in genes associated with ciliopathies assumes paramount importance, providing valuable insights into these conditions and necessitating the exploration and characterization of novel ciliary genes. The primary objective of this research project is the comprehensive characterization of a candidate gene, which represents the ortholog of the human gene. The candidate gene posseses calcium biding domain suggesting a role in the positive regulation of protein localization to the ciliary membrane and exhibit Calcium ion binding activity. To investigate role of a candidate calcium binding gene in ciliobiogenesis we apply C.elegans as a model organism. Our initial data shown that mutation of the candidate calcium binding gene shown significantly shorter body length of animal. Since our candidate gene has calcium binding domain, external and internal calcium intake may essential for body length. Collectively, this endeavor is poised to contribute significantly to the elucidation of the functional role of the newly characterized ciliary gene. The observation of distinct phenotypes promises to illuminate our understanding of cilia and ciliopathy mechanisms, paving the way for future research endeavors in this domain.

Keywords: A Candidate Calcium-Binding Protein, cilia, ciliopathy, C.elegans

ORAL PRESENTATION

In vitro biological activity studies of *Bilacunaria scabra* (Fenzl) Pimenov et V.N.Tikhom.

Tugsen Buyukyildirim[1*] (*ORCID: https://orcid.org/0000-0003-0101-9742*), Nuraniye Eruygur[2] (*ORCID: https://orcid.org/0000-0002-4674-7009*), Yavuz Bağcı[3], *(ORCID: https://orcid.org/0000-0002-2343-3672)*

[*1]Selcuk University, Faculty of Pharmacy, Department of Pharmacognosy, Konya, Türkiye.
[2]Selcuk University, Faculty of Pharmacy, Department of Pharmacognosy, Konya, Türkiye.
[3]Selcuk University, Faculty of Pharmacy, Department of Pharmaceutical Botany, Konya, Türkiye.

*Corresponding author e-mail: tugsen095@gmail.com

Abstract

Bilacunaria scabra (Apiaceae) (syn: *Ferula scabra* Fenzl, *Hippomarathrum scabrum* (Fenzl) Boiss., *Cachrys scabra* (Fenzl) Meikle.) is a perennial herb that grows in Southeastern Anatolia, Cyprus, and Syria. In this study, enzyme inhibitory activities of acetyl/butryl cholinesterase (AChE/BChE) associated with Alzheimer's disease and tyrosinase (TYR) associated with skin lightening effect and antioxidant activities (DPPH, ABTS) of ethanol (BSE) and hexane (BSH) extracts prepared from *B. scabra* aerial parts were evaluated with using ELISA microplate reader. In addition, the total phenol and flavonoid contents (TPC/TFC) of the extracts were also examined. According to the results, BSH extract (43.29%) has remarkable AChE and high BChE (94.20%) inhibitory activity. BSH (16.13%) and BSE (23.81%) showed moderate inhibitory activity against TYR. BSE has higher TPC (84.85 gallic acid mg/g extract) content than BSH (69.95 gallic acid mg/g extract). The reason why BSE (66.83%; 87.64%, respectively) had higher antioxidant activity than BSH (34.77%; 48.00%, respectively) in both DPPH and ABTS radical scavenging activity is probably due to the richness of phenolic compounds. *B. scabra* may also be a possible treatment option with the contribution of antioxidant activity in neurodegenerative diseases.

Keywords: *Bilacunaria scabra*, Apiaceae, cholinesterase, tyrosinase, antioxidant.

DRAFT

ORAL PRESENTATION

A novel fluorometric chemosensor for the selective, qualitative and quantitative detection of cyanide ions in water samples

Melek Güçoğlu[1*] (*ORCID:https://orcid.org/0000-0001-9426-9278*),
Nuray Şatıroğlu[2] (*ORCID:https://orcid.org/0000-0001-8201-9896*)

[*1, 2] Hacettepe University, Faculty of Science, Department of Chemistry, Ankara, TURKEY

* melekalp@hacettepe.edu.tr

Abstract

Cyanide ions are one of the most toxic anions and are unfortunately extensively utilized in a wide range of chemical and industrial activities, including the manufacture of plastics, the resin industry, organic synthesis, metallurgy, and gold mining. Thus, cyanide is present around us in various forms. It can be absorbed through the skin, lungs, and digestive system, which can result in vomiting, convulsions, unconsciousness, and even death. Therefore, it is imperative to design fluorometric sensors that are sensitive, selective, affordable, and simple for the detection of cyanide. Among various approaches for detecting cyanide, fluorescence spectroscopy is crucial in molecular sensing because of its high sensitivity, quick response time, and relatively low cost. In this study, a novel fluorometric chemosensor **(L)** was used to determine cyanide in water samples. The sensing properties of the newly synthesized sensor **L** were investigated by fluorescence spectroscopy. The sensor **L** showed a notable fluorescence after interacting with cyanide ions. The detection limit and association constant were examined by fluorimetric titration data and the Benesi-Hildebrand equation. The detection limit of the sensor for cyanide ions is 0.23 μM, which is lower than the maximum permissible level of cyanide in drinking water set by WHO. The sensor **L** has been successfully applied to determine cyanide ions in natural water samples and has showed selectivity towards cyanide over other common anions in the mixed solution.

Keywords: Chemosensor, cyanide, fluorescence spectroscopy, water samples

DRAFT

ORAL PRESENTATION

Synthesis route of smart halloysite nanocontainers and their use in anti-corrosion coatings

Müge AKBIYIK DEMİREL[1*] (*ORCID:0009-0009-2958-615X),*
Hatice KAPLAN CAN [1,2] *(ORCID:0000-0002- 2886-0788)*

[*1] Hacettepe University, Institute of Science and Technology, Polymer Science and Technology Division, Beytepe, 06800 Ankara, Turkey

[2] Hacettepe University, Faculty of Science, Deparment of Chemistry, Division of Polymer Chemistry, Beytepe, 06800 Ankara, Turkey

[*] Corresponding author e-mail: akbiyikmuge@gmail.com

Abstract

Corrosion is defined as the chemical or electrochemical wear of materials by interacting with their environment and the formation of physical and chemical changes in their structures. Many sectors directly and indirectly affected by corrosion, such as aviation, energy, chemistry etc. Today, the aviation sector is the leading sector when considering safety and speed factors. There are two main approaches to applying smart coatings with self-healing properties used for anti-corrosion applications. The first method involves localized repair of the damaged coating by the controlled release of polymerizable agents, while the second method requires electrochemical protection of the exposed metallic substrate using corrosion inhibitors stored in nanocontainers [1,2]. Nanoscale reservoirs provide uniform and long-term release of corrosion inhibitors [3]. Within the scope of this study; a self-healing nanostructure developed with the use of smart nano-containers that provide protection against corrosion, and the synthesis of pH-sensitive poly(maleic anhydride-*alt*-acrylic acid) copolymers to be used in the end stop system has been carried out in the layer-by-layer bonding (LBL) method. Within the scope of the study, aluminium 7075 was used as the main material.
The morphology and properties of nanocontainers were characterized by scanning electron microscopy (SEM) and Fourier transform infrared spectroscopy (FTIR). The loaded amount of benzotriazole (BTA) the halloysite lumen was determined by thermogravimetric analysis (TGA). Dynamic Mechanics Analysis (DMA) was used to follow dynamical mechanical properties of nanocontainers. The control of the healing of the defective area subjected to corrosion was illuminated with emission scanning electron microscopy.

Keywords: Self-repair, halloysite, LBL method, BTA, poly (MA-*alt*-AA)

References

[1] Nazari M. H., Shi X., ‘‘Polymer-based nanocomposite coatings for anticorrosion applications’’, *Industrial Applications for Intelligent Polymers and Coatings,* pp. 373-399, 2016, New York. doi: 10.1007/978-3-319-26893-4_18
[2] Stankiewicz A., Szczygieł I., Szczygieł B., ‘‘Self-healing coatings in anti-corrosion applications’’, *J Mater Sci,* vol.48, pp. 8041-8051, 2013. DOI 10.1007/s10853-013-7616-y
[3] Kartsonakis I., Balaskas A. C., Koumoulos E. P., Charitidis C. A., Kordas G., ‘‘Evaluation of corrosion resistance of mahnesium alloy ZK10 coated with hybrid organic – inorganic film including containers, *Corrosion Science,* vol.65 , 2012. DOI:10.1016/j.corsci.2012.08.052

DRAFT

ORAL PRESENTATION

Pollen diversity in the *Iberis simplex* species complex

Edibe Özmen-Baysal[1,2*] (*ORCID: https://orcid.org/ 0000-0002-6991-376X*),
Emre Çilden[1,2] (*ORCID: https://orcid.org/ 0000-0002-4950-0327*)

[1]Hacettepe University, Faculty of Science, Biology Department, Botany Section, Ankara, Turkey
[2] Hacettepe University, Biological Diversity Research and Application Center, Ankara, Turkey

*edibeozmen@gmail.com

Abstract

In this research, *Iberis simplex* species complex was studied in the terms of pollen morphology. Pollen samples were obtained from 31 population of *I. attica, I. carica, I. gypsicola, I. halophila, I. simplex* and *I. spruneri*. The palynological characteristics were measured under light microscope (LM) on 30 pollen grains for each sample. We evaluated different characteristics including polar axis, equatorial axis, colpus length and width, Amb diameter, apocolpium, exine structure and sculpture. Besides, the detailed exine sculpture of each sample examined by scanning electron microscopy (SEM). Principal component analysis (PCA) and cluster analysis were performed to reveal similarities between studied populations in terms of pollen morphology. As a result, it was determined that the pollen grains of the taxa included in *Iberis simplex* species complex had a wide variation at the intraspecific and interspecific levels. The diversity in pollen grains was compatible with anatomical, morphological and phylogenetic studies. However, pollen data did not elucidate the taxonomic confusion in the *Iberis simplex* species complex.

Keywords: Brassicaceae, intraspecific diversity, interspecific diversity, PCA, pollen morphology.

DRAFT

ORAL PRESENTATION

Echinodermata Filumunun Türkiye'deki Gelişimi (2003-2023), Bibliyometrik Bir Analiz

Sera Övgü KABADAYI YILDIRIM (*ORCID: https://orcid.org/0000-0002-0060-2445*)

[1]Isparta Uygulamalı Bilimler Üniversitesi, Eğirdir Su Ürünleri Fakültesi, Temel Bilimler ABD, Isparta, Türkiye

serakabadayi@isparta.edu.tr

Özet

Echinodermata phylumu, deniz ekosistemlerinin önemli bir bileşeni olan ve biyolojik çeşitliliği ve ekosistem işlevini etkileyen; deniz kestaneleri, deniz yıldızları, deniz hıyarları gibi önemli türleri içerir ve bilimsel araştırmalar için büyük bir potansiyele sahiptir. Derisi dikenliler, okyanusların ve denizlerin çoğunda yaygın olarak bulunur ve çeşitli ekosistemlerin anahtar türleridir. Ancak, ticari nedenlerle avlanmaları ve toplanmaları aynı zamanda balıkçılık av araçlarının baskısı bazı türlerin popülasyonlarının tehlikede olmasına yol açmaktadır. Özellikle deniz kestaneleri ve deniz hıyarları, ticari avcılıkla karşı karşıya olan türlerdir. Bu türlerin popülasyon biyolojisi hakkında eksik bilgi, artan avcılık baskısı ve sıkı balıkçılık yönetimi eksikliği, dünya çapında popülasyonlarının azalmasına katkıda bulunmaktadır. Echinodermata filumuna dair yapılan bilimsel araştırmaların sayısı giderek artmaktadır. Fakat bu araştırmaların büyük bir bölümü farklı bilimsel kaynaklarda yayınlanmakta ve bu da bu alandaki veri bütünlüğünü ve erişilebilirliği zorlaştırmaktadır. Bir veri tabanı oluşturularak, bu dağınık verilerin bir araya getirilmesi ve daha iyi bir araştırma altyapısının oluşturulması mümkün olacaktır. Bibliyometrik bir çalışma, Echinodermata phylumuyla ilgili yapılan araştırmaların analizini ve sentezini sağlayarak, bu organizmaların ekolojik, biyolojik ve ekonomik önemini daha iyi anlamamıza yardımcı olacaktır. Bu analizler, gelecekteki araştırmalara yön verme açısından da büyük önem taşır. Bibliyometrik çalışmalar, bu alandaki bilimsel verilerin toplanması, analizi ve sentezi için önemli bir araç olabilir. Ayrıca, kendi veritabanını oluşturarak bu çalışmanın daha da etkili hale getirilmesi mümkündür. Bu çalışma, Echinodermata phylumunu daha iyi anlamamıza ve korumamıza yardımcı olabilir ve literatürdeki boşlukları analiz ederek gelecekteki araştırmalara yol gösterebilir. Çalışmamız ile Türkiye ve Akdeniz'de son 20 yılda Echinodermata filumu ile ilgili yapılan çalışmalar analiz edilmiş ve literatürdeki boşluklar belirlenmiştir.

Anahtar Kelimeler: Bibliyometrik analiz, Echindermata, Benthos, Mediterranean

DRAFT

ORAL PRESENTATION

Effect of modification with surfactant on organic dye adsorption capacity of char based adsorbents

İsmail Cem Kantarlı[1*] (*ORCID: https://orcid.org/0000-0002-5911-3152)*

[*1] Ege University, Ataturk Medical Technology Vocational Training School, İzmir, 35100, Türkiye

*Corresponding author e-mail: ismail.cem.kantarli@ege.edu.tr

Abstract

In this study, the effect of modification with surfactant on organic dye adsorption capacity of char based adsorbents, hydrochar and its activated carbon, was investigated. Adsorbents were modified by sodium dodecyl sulphate, an anionic surfactant, with the aim of increasing the negative charge on the adsorbent surface and hence its adsorption capacity of methylene blue, a basic and cationic organic dye present in waste streams. Equilibrium adsorption study of the adsorbents was carried out with 100 mL methylene solutions at concentrations between 50 and 350 mg. L^{-1} and at adsorbent concentration of 1 $g.L^{-1}$. The residual methylene blue in the filtrated solutions were determined by the UV-visible spectrophotometer (UV-160A, Shimadzu) at the wavelength of 665 nm. The isothermal equilibrium data obtained from tests were interpreted by applying Langmuir and Freundlich isotherm equations. Adsorption of methylene blue on all adsorbents displayed better fit to the Langmuir model in comparison with Freundlich model as implied by the R^2 values. The Langmuir capacity, S_M, of hydrochar was calculated as 34.2 $mg.g^{-1}$, while that of activated carbon was much higher (294.1 $mg.g^{-1}$) due to its improved porosity and surface area as a result of $ZnCl_2$ activation. The methylene adsorption capacity calculated for adsorbents modified with sodium dodecyl sulphate were found to be lower than that of unmodified adsorbents which was attributed to the dominance of pore blocking effect of sodium dodecyl sulphate over the positive effect of its anionic functional groups during methylene blue adsorption.

Keywords: Modification, Hydrochar, Activated Carbon, Adsorption, Methylene Blue

DRAFT

ORAL PRESENTATION

Novel 2D Cu (II) coordination polymer based on the flexible 3, 3'-dimethylglutaric acid and 4,4'-bipyridine: Hydrothermal synthesis and crystal structure

Figen Arslan Biçer* (*ORCID: https://orcid.org/0000-0002-4024-8233)*

*Karabük University, Faculty of Science, Department of Chemistry, 78050 Karabük, Türkiye

farslan@karabuk.edu.tr

Abstract

The design and synthesis of coordination polymers have received intense interest not only their intriguing structural topologies but also have their potential applications such as chemical sensors, magnetism, adsorption, catalysis and so on. [1-3]. One of the most effective methods for design and synthesize of targeted CPs is mixed ligand strategy [4]. Polycarboxylates because of their diverse coordination modes and bridging abilities, and N-donor ligands with different conformations, and flexibility, play a vital role in these process [5]. This study has aimed to hydrothermal synthesis of $\{[Cu(\mu\text{-dmglu})(\mu\text{-bipy})]\cdot H_2O]\}_n$ (**1**) (H_2dmglu: 3,3'-dimethyl glutaric acid, bipy: 4,4'-bipyridine) and to analyse its structural characteristics using elemental analysis and single crystal X-ray diffraction techniques. X-ray structural analysis of the complex **1** reveal that **1** crystallize in the monoclinic systems with the $P2_1/n$ space group. The asymmetric unit of **1** contain one Cu(II) ion, one dmglu ligand and one lattice water molecule. Each Cu(II) ion is coordinated by three carboxylate oxygen atoms from two different dmglu ligands and one nitrogen atom from bipy ligand to give a distorted square pyramidal geometry (τ = 0236) [6] (Fig. 1.).

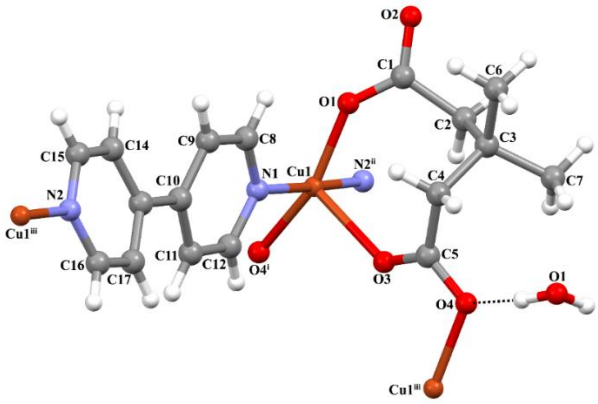


Figure 1. View of the molecular structure for **1** showing the atom numbering scheme

Keywords: 3,3'-Dimethyl glutaric acid, bipy: 4,4'-bipyridine, Cu(II) coordination polymer.

References:

1. S. Qiu and G. Zhu. *Coord. Chem. Rev.*, **2009**, *253*(23/24), 2891.
2. W. P. Lustig, S. Mukherjee, N. D. Rudd, A. V. Desai, J. Li, and S. K. Ghosh. *Chem. Soc. Rev.*, **2017**, *46*(11), 3242.
3. M. Du, R. Banerjee, and G. K. H. Shimizu. *CrystEngComm*, **2013**, *15*(45), 9237.
4. M. Du, , C. P. Li, , C. S. Liu, and S. M. Fang. *Coordination Chemistry Reviews*, **2013**. *257*(7-8), 1282.
5. P. Du, Y. Yang, J. Yang, Y.-Y. Liu, W.-Q. Kan, and J.-F. Ma. *CrystEngComm*, **2013**, *15*(35), 6986.
6. A.W. Addison, T.N. Rao, J. Reedijk, J. Vanrijn, G.C. Verschoor. J Chem Soc Dalton, **1984** 1349.

ORAL PRESENTATION

Exitation energy transfer between bodipy dyes on the same cyclotriphosphazene platform

Semiha Yıldırım Sarıkaya* (*ORCID: https://orcid.org/0000-0002-5993-3660*)

*Karabük University, Faculty of Science, Department of Chemistry, Karabük, Türkiye

semihayildirim@karabuk.edu.tr

Abstract

Effective harvesting of solar radiation needs absorbers that have been wisely designed. Thanks to a well-designed multichromophoric system, it is possible to work in a wide spectral region. In a multichromophoric system, if one of the chromophores is excited by light with an appropriate wavelength, energy transfer can occur when the donor returns to its ground state and the acceptor is simultaneously brought to its excited state [1]. In this work, two different BODIPY dyes were brought together by a cyclotriphosphazene ring (Figure 1). The structural integrity of the molecule was demonstrated by mass and ^{1}H-NMR studies. Intermolecular interactions in a newly synthesized BODIPY-cyclotriphosphazene-distyryl-BODIPY triad have been established from spectral studies. As predicted from the literature [1,2], occurrence of energy transfer from the donor BODIPY units to the distyryl-BODIPY acceptor within the triad was witnessed. This construction ensured strong absorption in a large part of the visible spectrum, but emission occurred highly from the distyryl BODIPY units by energy transfer processes.

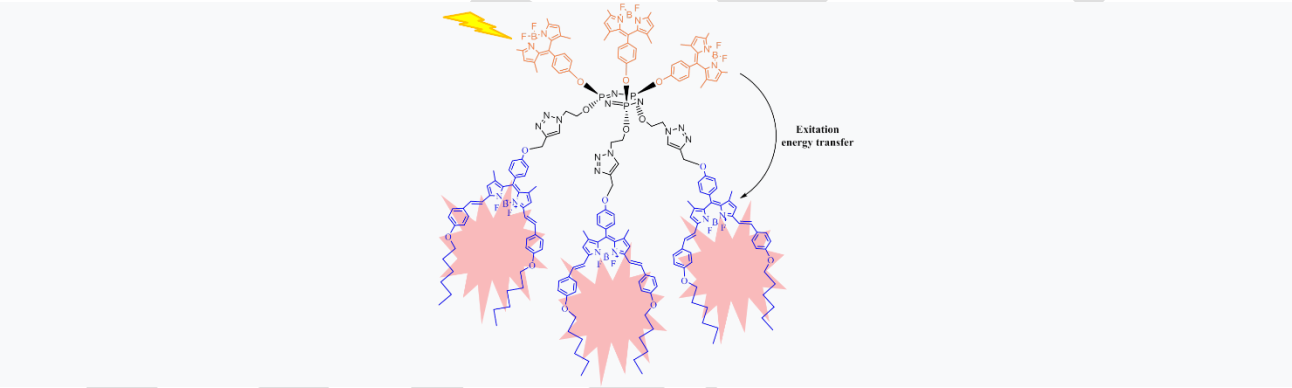


Figure 1. Illustiration of energy transfer processes in the BODIPY-cyclotriphosphazene-distyryl-BODIPY triad

Keywords: BODIPY, cyclophosphazene, energy transfer

References:

[1] Barin, G., Yilmaz, M. D., Akkaya, E. U. "Boradiazaindacene (Bodipy)-based building blocks for the construction of energy transfer cassettes", Tetrahedron Letters, 2009, 50, 1738-1740.
[2] Kostereli, Z., Ozdemir, T., Buyukcakir, O., Akkaya, E. U. "Tetrastyryl-BODIPY-Based Dendritic Light Harvester and Estimation of Energy Transfer Efficiency", Organic Letters , 2012, 14, 3636-3639.

ORAL PRESENTATION

Capnodis tenebricosa (Olivier, 1790) (Coleoptera: Buprestidae)'da dişi üreme sisteminin morfolojisi

Hanife Gözüpek[1*] (*ORCID: https://orcid.org/0000-0003-3620-3056*), Nurcan Özyurt Koçakoğlu[2] (*ORCID: https://orcid.org/0000-0001-7137-8631*), Selami Candan[3] (*ORCID: https://orcid.org/0000-0002-7402-1360*)

[*1, 2,3]Gazi Üniversitesi, Fen Fakültesi, Biyoloji Bölümü, Ankara, Türkiye

*Sorumlu yazar e-mail: hanifegozupekk@gmail.com

Özet

Coleoptera takımı dünya üzerinde 350.000'in üzerinde tanımı yapılmış tür sayısı bakımından en zengin böcek guruplarından birisidir. Bu böcek takımı içerisinde yer alan Buprestidae familyası da tanımlanmış 16.000 tür ile temsil edilmektedir. Bu familyanın üyelerinin birçoğu ekonomik zarara sebep olmaktadır. *Capnodis tenebricosa* (Olivier, 1790) (Coleoptera: Buprestidae) meyve ağaçlarında önemli zarara sebep olmaktadır. Bu çalışmada zararlı bir tür olan *C. tenebricosa* 'nın dişi üreme sisteminin anatomisi ve histolojisi stereo, ışık ve taramalı elektron mikroskobu teknikleri kullanılarak incelenmiştir. Yapılan çalışmalar sonucunda *C. tenebricosa* 'nın dişi üreme sisteminde çok sayıda lineer dizilmiş ovaryoller ayırt edilmiştir. Ayrıca dişi üreme sistemi 1 spermateka, 1 bursa copulatrix, bir çift lateral kanal, 1 ortak kanal ihtiva etmektedir. Her bir ovaryolde terminal filament, germaryum, vitellaryum, pedisel kısımları bulunmaktadır. Germaryumda trofositlere, vitellaryumda farklı gelişim aşamalarında oositlere (previtellogenez, vitellogenez ve koryogenez aşamasındaki oositler) rastlanmıştır. Bu oositlerde gelişim ovaryolün distalinden proksimaline doğru artmakta olup oositi çevreleyen epitel hücrelerinin şeklinde ve oosit lümen içeriğinde farklılıklar ayırt edilmiştir. Pedisel ve lateral kanal yüzeyinde boyuna kaslar dikkat çekmektedir. Bursa copulatrix yüzeyinde enine kaslara rastlanmıştır. Bursa copulatrix lümeni salgı materyaliyle doludur. Spermateka bursa copulatrix'ten oldukça küçük olup kas yapısı daha zayıftır. Spermatekanın lümeninin spermle dolu olduğu görülmüştür. Sonuç olarak, bu çalışma, önemli bir zararlı tür olan *C. tenebricosa* 'nın dişi üreme yapısı hakkında temel bilgiler sağlamaktadır. Bu bulgular, Buprestidae dahil Coleoptera'daki üreme sistemi morfolojisinin çeşitliliği üzerine yapılacak çalışmalarda ve bu türün zararı ile mücadele konusunda yapılacak çalışmalarda temel oluşturması açısından önem taşımaktadır.

Anahtar Kelimeler: ovaryum, oosit, spermateka, bursa copulatrix, ışık mikroskobu, elektron mikroskobu

DRAFT

ORAL PRESENTATION

In vitro inhibition effects of Acai berry extracts on human AChE

Bülent Şengül (*ORCID: https://orcid.org/0000-0002-9998-6564*)

Department of Health Care Services, Vocational School of Health Services, Bayburt University, Bayburt, Türkiye.

bulentsengul@bayburt.edu.tr

Abstract

Plant species are frequently used in the pharmaceutical, agriculture, and food industries because they are rich in antioxidants and bioactive components. Many plant extracts have been studied for their effects on enzyme inhibition, and many herbs have been utilized to treat cognitive impairment and memory loss. In fact, the first clinical application of Acetylcholine esterase (AChE) inhibitors in Alzheimer's Disease (AD) began with the use of physostigmine isolated from plants. As a result, studies aimed at lowering AD symptoms have focused on herbal therapies with little adverse effects. In this study, the *in vitro* inhibition effect of aqueous, ethanol, and methanol extracts of Acai berry on the AChE enzyme isolated from human erythrocytes by ion exchange chromatography was investigated. For this purpose, dried and powdered Acai Berry was mixed with water, ethanol, and methanol in a ratio of 1:5 by a magnetic stirrer at room temperature for 24 hours. The effects of the obtained extracts on enzyme activity were studied *in vitro*. As a result of the experimental studies, it was found that aqueous, ethanol, and methanol extracts of Acai berry inhibited hAChE with IC_{50} values of 0.15 µg/ml, 0.49 µg/ml, and 0.62 µg/ml respectively.

Keywords: Acetylcholine esterase, Acai berry, inhibition

s

DRAFT

ORAL PRESENTATION

Capnodis tenebricosa (Olivier, 1790) (Coleoptera: Buprestidae)'da arka bağırsağın ultrastüktürel yapısı

Gizem Dal[1*] (*ORCID: https://orcid.org/0000-0002-7845-5430*), Selami Candan[2] (*ORCID: https://orcid.org/0000-0002-7402-1360*), Nurcan Özyurt Koçakoğlu[3] (*ORCID: https://orcid.org/0000-0001-7137-8631*),

[*1, 2,3]Gazi Üniversitesi, Fen Fakültesi, Biyoloji Bölümü, Ankara, Türkiye

*Sorumlu yazar e-mail: gizemdal2007@gmail.com

Özet

Capnodis tenebricosa (Olivier, 1790) (Coleoptera: Buprestidae) larvaları özellikle sert çekirdekli meyve ağaçlarının köklerinde beslenerek zararlı olmaktadır. Ülkemizde kiraz, kayısı ve zerdali gibi meyve ağaçlarında ve kuzukulağında zararlıdır. Bu zararlı ile mücadele için sindirim yapısının bilinmesi oldukça önem taşımaktadır. Bu çalışmada tarımsal zararlı bir tür olan ve daha önce çalışılmamış olan *C. tenebricosa*'nın sindirim sisteminin bir kısmı olan arka bağırsak yapısı detaylı olarak tanımlanmıştır. Bu çalışmada kullanılan ergin *C. tenebricosa* örnekleri Haziran 2019'da Sivas'tan toplanmıştır. Stereomikroskop altında disekte edilen örnekler ışık mikroskobu ve elektron mikroskobunda incelenmiştir. Elde edilen verilere göre *C. tenebricosa*'nın sindirim sisteminin ön, orta ve arka bağırsak kısımlarından oluştuğu görülmüştür. Arka bağırsak dışkı ve idrardan suyun, tuzların ve diğer yararlı maddelerin emilmesini sağlamaktadır. Sindirimi sisteminin üçüncü kısmı olan arka bağırsak; ileum, kolon ve rektum kısımlarından oluşmaktadır. Arka bağırsağın ilk kısmı olan ileum yıldız şeklinde bir lümene sahip olup ürik asit kristallerine rastlanmıştır. İleum duvarı intima, tek tabakalı silindirik epitelle çevrelenmiştir. İkinci kısım olan kolon duvarı Malpighi tübülleriyle bağlantılıdır. Kolon duvarı lümenden itibaren intima, tek tabakalı kübik epitel ve kas tabakasıyla çevrelenmiştir. Sindirimin son kısmı olan rektum yüzeyi oldukça kalın enine kas tabakasıyla çevrelenmiştir. Histolojik incelemelerde rektumu çevreleyen intima tabakası, tek tabakalı kübik epitel ve kas tabakası görülmektedir. Bu çalışmada sunulan verilerin gelecekte bu konularla ilgili yapılacak çalışmalara ilaveten taksonomik, sistematik ve tarım zararlısı olan türlerle mücadelede yapılacak çalışmalara temel oluşturarak katkı sağlayacağı düşünülmektedir.

Anahtar Kelimeler: İleum, kolon, rektum, ışık mikroskobu, elektron mikroskobu

DRAFT

ORAL PRESENTATION

Türkiye akar faunası için yeni bir tür kaydı; *Erythraeus* (*E.*) *opilionoides* (C. L. Koch, 1837) (Acari: Erythraeidae)

İbrahim Karakurt[1*] (*https://orcid.org/0000-0003-4056-0822*)

[1]Erzincan Binali Yıldırım Üniversitesi, Sağlık Hizmetleri MYO, Sağlık Bakım Hizmetleri, Erzincan, Türkiye

*e-mail:ikarakurt24@gmail.com

Özet

Oldukça zengin tür çeşitliliğine sahip ve dünya genelinde yayılış gösteren Erythraeidae Robineau-Desvoidy, 1828 ailesi, Erythraeoidea üst ailesini oluşturan iki aileden en büyük olanıdır ve günümüzde 900'ün üzerinde tür sayısı ile temsil edilmektedir. Heteromorfik larva yapıları sayesinde bazı türler yalnızca larva veya yalnızca postlarva (larva sonrası) evrelerinden tanımlanmışken; bir kısmı ise artan deneysel çalışmalarla birlikte her iki evreden de tanımlanabilmiştir. *Erythraeus* (*Erythraeus*) Latreille, 1806 alt cinsi bu ailenin en fazla tür çeşitliliğine sahip olan *Erythraeus* Latreille, 1806 cinsine dahildir. Ülkemizden bu alt cinse ait şu ana kadar yalnızca 9 türün kaydı yapılmıştır. 2023 yılı Mayıs ayında, Erzincan ili, Kemaliye ilçesinden (39º36′K 39º09′D 1162 m) alınan çam ağacı altı yosunlu döküntü örnekleri, araştırma laboratuvarına getirilmiş ve Berlese sistemine yerleştirilerek, akar örnekleri ayıklanmıştır. Elde edilen örnekler % 70'lik etilalkol ile muhafaza edilmiş ve Hoyer ortamında preparatları hazırlanmıştır. Preparatlar Olympus BX63 mikroskobunda incelenerek, tür teşhisleri yapılmıştır. Yapılan inceleme sonucunda; *Erythraeus* (*E.*) *opilionoides* türünün nimf evresi teşhis edilmiş ve Türkiye akar faunası için yeni bir kayıt olarak listelenmiştir. Böylece ülkemizden bu alt cinse ait kaydedilen tür sayısı 10'a ulaşmıştır.

Anahtar Kelimeler: Karasal Parasitengona, *Erythraeus*, Erythraeinae, Erzincan

DRAFT

ORAL PRESENTATION

Evaluating microplastic administration methods for honeybees in controlled cage experiments

Aygün Schiesser[*1] (*ORCID: 0000-0002-1480-892X*),
Seçil Karahisar Turan[1] (*ORCID: https://orcid.org/0000-0002-9735-968X)*

[1] Hacettepe University, Faculty of Science, Department of Biology, Ankara, Turkey.

*Corresponding author e-mail: aygun@hacettepe.edu.tr

Abstract

Microplastics are plastic particles generated by human-induced environmental pollution. These particles pose a growing threat to ecosystems worldwide. These tiny plastic fragments, often less than five millimeters in size, arise from deliberate industrial and cosmetic production or the gradual disintegration of larger plastic items. Research on microplastics has primarily focused on marine environments and aquatic invertebrates, with limited attention given to the interaction between microplastics and terrestrial ecosystems, particularly pollinators like honeybees. This study aims to evaluate different methods of applying microplastic particles to honeybees using controlled cage experiments. Cage experiments are recognized for their capacity to provide comprehensive data, especially regarding nutrition regimens and lifespan. To conduct our research, emerging adult worker honeybees were carefully collected from an apiary. These bees were then placed in a test cabinet, maintaining a temperature of 34 ± 1°C and a relative humidity of 55 ± 5% RH. Subsequently, groups of 50 bees were put within specialized cages designed for experimentation. Microplastic pieces smaller than 130 microns in diameter were obtained from a local manufacturer and measured using a microscope. Microplastic particles were presented to bees using 4 different methods: in water, in sugar syrup, in pollen cake and in pollen gel. The experiment was continued for 14 days with the types of food that the bees accepted to consume. Feeds were renewed every day and daily consumption amounts were determined by weighing. The results were evaluated statistically. Our findings demonstrate the effectiveness of various microplastic administration methods in controlled cage experiments. These methods allowed us to quantify the consumption rates of microplastics by honeybees and assess their potential effects on the bees. This study highlights the importance of selecting appropriate administration methods to accurately evaluate the impact of microplastics on honeybees and emphasizes the need for further research in this evolving field.

Keywords: Honey bee, microplastics, nutrition, cage experiments

DRAFT

ORAL PRESENTATION

A new Sulfonylamide compound containing 4-methoxy cinnamaldehyde with potential bioactivity: Synthesis, characterization, and detailed theoretical calculations

Nurana Ibrahimova[1*] (*ORCID: https://orcid.org/0000-0003-2378-9174*),Ümmühan Özdemir Özmen[1] ORCID :*https://orcid.org/* 0000-0001-9161-9367 , Servet Çete[1] (*ORCID: https://orcid.org/0000-0002-9316-2812*), Deniz Akın Anakök[1] (ORCID: https://orcid.org/0000-0002-7984-9159), and Ayla Balaban Gündüzalp[1] ORCID :*https://orcid.org/* 0000-0002-5740-3674

[1]Gazi University, Science Faculty, Department of Chemistry.Ankara, TURKEY

*****Corresponding author e-mail: nurana.ibrahimova@gazi.edu.tr

Abstract

Sulfanilamide is an organic compound belonging to the sulfonamide class of antibacterial drugs and is a sulfa drug that mimics para-aminobenzoic acid, an important component of bacterial cell wall synthesis. Sulfanilamide has been widely used to prevent microbial infections, particularly it continues to be used topically to treat vulvovaginitis caused by *Candida albicans.* Many effective antibacterial drugs derived from sulfa drugs have been used and discovered in medicine. Examples of some common sulfa drugs are sulfanilamide (SA), sulfaquinoxaline, silver sulfadiazine, sulfasalazine, and sulfamethoxazole. 4-Methoxycinnamaldehyde (4MeOCA) is a natural compound of Cinnamomum cassia. Cinnamaldehyde; It contains essential oil components and shows antibacterial activity. Essential oils have been widely used for bactericidal, virucidal, fungicidal, antiparasitic, insecticidal, medical, and cosmetic purposes since the Middle Ages. This study synthesized a new compound SA-4MeOCA from sulfanilamide (4-aminobenzenesulfonamide) and 4-methoxy cinnamaldehyde, which may have potential drug properties. The synthesized compound was analyzed using various spectral tools such as FTIR, 1H-NMR, and 13C-NMR and compared with highly correlated simulated results. Since it is a new compound, frontier orbitals, HOMO and LUMO energies, chemical reactivity descriptors, and molecular electrostatic potential (MEP) map were calculated with detailed theoretical calculations. That's why we use the structure of the newly synthesized SA-4MeOCA molecule, which is a sulfa drug derivative, optimized with the DFT/B3LYP method and 6-31+G (d,p) basis set. All calculations were performed using the Gaussian 09 package program.

K**eywords**: Sulfa drug, Sulfanilamide, 4-methoxy cinnamaldehyde, Theoretical Calculations

DRAFT

ORAL PRESENTATION

The Entomofauna Visiting *Salvia cryptantha* Flowers

Çiğdem Özenirler* *ORCID: https://orcid.org/0000-0003-0390-2416*

* Hacettepe University, Faculty of Science, Department of Biology, Ankara, Türkiye.

*Corresponding author e-mail: cozenir@hacettepe.edu.tr

Abstract

Salvia cryptantha Montbret & Aucher ex Bentham, is an endemic species for Türkiye. Its high nectar and pollen content makes it an atractive foraging plant for insects. *Orobanche anatolica* is a parasitic plant that thrives on *Salvia cryptantha.* The research was conducted at Hacettepe University's Beytepe Campus, located in Ankara, Türkiye. The study aimed to ascertain whether the parasitic relationship between these plants influences the diversity of insects that feed on them. Over a 10-day observation period during the flowering season, insects that visited Salvia cryptantha plants, both infected and uninfected with *Orobanche anatolica*, were monitored. Sampling was carried out using insect nets for a total of 6 days. These insects were identified by their species and individual numbers. Additionally, a comparison was made at the order level using percentage values. In the case of *Salvia cryptantha* plants not infected by *Orobanche anatolica*, 297 individuals from 97 species were observed. The order Hymenoptera accounted for 66.33%, Coleoptera for 12.12%, Homoptera for 9.76%, Diptera for 4.72%, Lepidoptera for 2.02%, and Hemiptera for 1.68% of the visitors. On the other hand, Salvia cryptantha plants infected with *Orobanche anatolica* attracted 161 individuals from 75 species, with the following percentages: Hymenoptera 57.14%, Homoptera 15.13%, Coleoptera 11.8%, Diptera 7.46%, Orthoptera 5.59%, Lepidoptera 1.86%, and Hemiptera 0.62%. In conclusion, Salvia cryptantha plants infected with Orobanche anatolica were found to be visited by relatively fewer insects. As a result, *Salvia cryptantha* individuals infected with *Orobanche anatolica* were found to be visited by relatively few insects.

Keywords: insects, *Salvia cryptantha*, parasitism, biodiversity

DRAFT

ORAL PRESENTATION

Üç fazlı üç boyutlu elektro oksidasyon sisteminde degradasyon

Canan Samdan[1*] (*ORCID*: http://orcid.org/0000 0001 8755 0790)

[*1]Eskişehir Osmangazi Üniversitesi, Mühendislik-Mimarlık Fakültesi, Kimya Mühendisliği Bölümü, Eskişehir, Türkiye

*Sorumlu yazar e-mail: : caydin@ogu.edu.tr

Özet

Pestisit dünyasında Bentazone, geniş yapraklı yabani otları hedef alan seçici bir herbisittir. Bu çalışma, Bentazon'un üç boyutlu bir elektro-oksidasyon işlemi kullanılarak sulardan uzaklaştırılmasını konu almıştır. Pozitif yada negatif yüzey yüküne sahip tanecikler elektrik alan altında yüklü partikül elektrotlara dönüşürler. Anot ve katot arasında yerleştirilen yüklü partiküller elektrot yüzeyini arttırarak üç fazlı üç boyutlu elektro oksidasyon reaktörünü oluşturur. Bu çalışmada organik yapılı kirleticinin degredasyonunda üç boyutlu reaktöre uygulanan potansiyel farkın ve akım yoğunluğunun etkisi araştırılmıştır. Partikül elektrotlar olarak TEPA (Tetraethylenepentamine) yüklü aktif karbon pa kullanılmıştır. Anot ve katot olarak karbon fiber kullanılmıştır. Destek elektrolit olarak NaCl'ün kullanıldığı çalışmalarda 4V ila 7V arasında değişen potansiyel farklar, ve 2,4 mA/cm2 ila 9,6 mA/cm2 arasında değişen akım yoğunlukları kullanılarak degredasyon verimi araştırılmıştır. Potansiyel farkın 4V dan 7 V a arttırılması degredasyon vermini olumlu yönde etkilemiştir. Bentazonun degredsasyon verimi %76.80'den %81.66'ya yükselmiştir. Akım yoğunluğunun 2.4'den 9.6 mA/cm2'ye arttırılmasının degradasyon verimi üzerinde oldukça önemli etkiye sahip olduğu belirlenmiştir. 2.4 mA/cm2 akım yoğunluğunda %61.85 degradasyon verimi elde edilirken, 9.6 mA/cm2 akım yoğunluğunda %91.53 bentazon degredasyonuna ulaşılmıştır. Elde edilen sonuçlar kinetik açıdan değerlendirildiğinde degradasyonun sözde birinci mertebeden hız ifadesine uyduğu belirlenmiştir. Yapılan çalışmada TEPA-yüklü aktif karbon partiküllerinin bentazonun degredasyonunu desteklediği belirlenmiştir. 3 fazlı üç boyutlu elektro oksidasyon sisteminin organik yapılı moleküllerin degredsayonunda oldukça etkili olabileceği ortaya konulmuştur.

Anahtar Kelimeler: TEPA-modifiye aktif karbon, üç fazlı üç boyutlu elektro oksidasyon reaktörü, bentazon

DRAFT

ORAL PRESENTATION

Nanolif yapılı çok katmanlı yara örtüsü sentezi, karakterizasyonu ve antibakteriyel etkisi

Selvinaz Erdoğan[1] (*ORCID: https://orcid.org/0000-0001-6557-0125*), Şükran Melda Eskitoros Toğay[2] (*ORCID: https://orcid.org/0000-0002-7473-8417*), Nursel Dilsiz[1*] *(ORCID: https://orcid.org/0000-0002-6496-0487)*

[*1]Gazi Üniversitesi, Mühendislik Fakültesi, Kimya Mühendisliği Bölüm, Ankara, Türkiye
[2]Sağlık Bilimleri Üniversitesi, Gülhane Sağlık Meslek Yüksekokulu, Eczane Hizmetleri Programı, Ankara, Türkiye

*Sorumlu yazar e-mail: ndilsiz@gazi.edu.tr

Özet

Vücudun dış örtüsü ve örtü sisteminin en büyük organı deridir. Derideki yaralar veya yaralanmalar; yanık, kazara travma veya kronik ülserasyonlardan kaynaklanabilmektedir. Derideki bu yaralar kontaminasyona müsait olup enfeksiyon riski taşımaktadır. Günümüzde yara bakımı, enfeksiyon riskini en aza indirerek hızlı ve etkili bir iyileşme sağlanması açısından büyük bir öneme sahiptir. Bu kapsamda antibakteriyel yara örtüleri bu alandaki önemli gelişmelerden biridir. İdeal bir pansuman, yarada hemostaz ve nemli ortam sağlayarak enfeksiyondan korumalıdır. Ayrıca su ve hava geçirgenliğine izin vermeli ve epitelizasyonu desteklemelidir. Tıp alanında yeni pansuman türlerinin keşfedilmesi ve geliştirilmesi büyük önem taşımaktadır. Bu çalışmanın amacı, polikaprolakton (PCL) ve kitosan (CS) polimerlerine tigesiklin yükleyerek elektrospinleme yöntemiyle kontrollü salım hedefli, antibakteriyel nanolif yapılı çok katmanlı bir membranının sentezlemesi ve yara örtüsü olarak uygulanabilirliğinin gösterilmesidir. Tigesiklin, geniş antimikrobiyal etkiye sahip bir minosiklin çeşidi olup ABD Gıda ve İlaç Dairesi (FDA) tarafından onaylanan ilk glisilsiklin antibiyotiktir. Bu antibiyotik çok sayıda Gram pozitif ve Gram negatif mikroorganizmaya karşı *in vitro* etkinlik göstermektedir. Yara örtüsü olarak kullanılacak ilk polimer olan PCL, yüksek mekanik stabiliteye sahip olup zayıf hücre tutunma özelliği gösteren, biyolojik olarak parçalanabilen, biyolojik olarak uyumlu ve hidrofobik bir sentetik polimerdir. Diğer polimer olan CS ise, doğada bulunan bir polisakkarit olup kitinden türetilen biyouyumlu ve biyobozunur yapıya sahiptir. İyi hücre yapışması ve çoğalması özellikleri göstermektedir. Bu polimerlerin avantajlarından yararlanarak, yüksek hücre tutunma ve hücre proliferasyonuna sahip aynı zamanda mekanik stabilitesi yüksek çok katmanlı PCL-CS-Tigesiklin kompozit malzemesi elektrospinleme yöntemiyle üretilmiştir. *In vitro* salım çalışmalarında çok katmanlı yara örtüsünden tigesiklin salımı, tek ve iki katmanlı membranlara oranla %65 oranında kontrollü salım ile gerçekleşmiştir. *S. aureus* ve *E. coli* bakterileri üzerinde yapılan antibakteriyel analiz sonuçları ise, iki ve çok katmanlı memranların tigesiklin yüklenmemiş yara örtüsüne göre belirgin oranda antibakteriyel etkiye sahip olduğunu göstermiştir. Sonuç olarak, bu çalışma çok katmanlı biyouyumlu ve biyobozunur polimerik matris yapısıyla yara örtüsü alanındaki araştırmaların ilerlemesine katkı sağlamıştır.

Anahtar Kelimeler: Yara örtüsü, antibakteriyel, çok katmanlı, polikaprolakton, kitosan, tigesiklin.

DRAFT

ORAL PRESENTATION

Türkiye'deki Akdeniz midyesi (*Mytilus galloprovincialis* Lamarck, 1819) yetiştiricilik sektörünün mevcut durumu

Nihat Sami KUYUMCU[1*] *(0009-0007-2058-5439)*, Sefa ACARLI[2] *(0000-0002-5891-5938)*

[1*]Çanakkale Onsekiz Mart Üniversitesi, Lisansüstü Eğitim Enstitüsü, Su Ürünleri Yetiştiriciliği Anabilim Dalı.,
Çanakkale, Türkiye
[2] Çanakkale Onsekiz Mart Üniversitesi, Deniz Bilimleri ve Teknolojisi Fakültesi, Su Ürünleri Yetiştiriciliği Bölümü Çanakkale, Türkiye

*nskuyumcu@gmail.com

Özet

Yüksek miktarda protein ve düşük miktarda yağ içeriğine sahip olmalarının yanı sıra Omega-3 yağ asitleri ve vitamin-mineral bakımından zengin besin içerikleri sebebiyle çift kabuklu yumuşakça türlerine olan ilgi Türkiye'de ve dünyada her geçen gün artış göstermektedir. Çift kabuklu türlerinden birisi olan Akdeniz midyesi (*Mytilus galloprovincialis*) ekonomik değeri ve tüketim popülaritesinden dolayı Türkiye'de son yıllarda avcılık ve yetiştiricilik üretimi artan tek tür olarak istatistiklerde yerini almaktadır. Akdeniz midyesinin gerek iç gerekse dış piyasadaki talebini karşılamak üzere üretim çiftliklerinin kurulumuna ilişkin başvurular son yıllarda artış göstermiştir. Bu bağlamda Akdeniz midyesi üretimi yapan tesislerin durumunu ortaya koymak ve geleceğe yönelik bir planlama yapmak sektörün doğru bir şekilde gelişimi açısından oldukça önemlidir. Bu çalışmada, 2023 yılı Ocak-Nisan dönemini kapsamak üzere, Türkiye'de yetiştiricilik belgesine sahip toplam 47 Akdeniz midyesi üretim tesisi tespit edilmiş ve bu tesisler ile anket çalışması yapılmıştır. Anket çalışmasında tesislerin kapasiteleri, alanları, üretim yöntemleri, personel durumları, pazarlama stratejileri ve karşılaştıkları sorunlara dair konularla ilgili istatistiki veriler alınmıştır. Sonuç olarak, bu çalışma ile midye üretim sektörünün gelişime açık bir sektör olduğunu ancak, malzeme temini ile birlikte kurdaki dalgalanmalar, yetişmiş personel ve bilgi eksikliğinin yanı sıra ulaşım ve lojistikte yaşanılan altyapı eksikliklerinin en çok karşılaşılan sorunlar olduğu ortaya konulmuştur.

Anahtar Kelimeler: Akdeniz midyesi, *Mytilus galloprovincialis*, long-line, yetiştiricilik

Not: Bu makale ÇOMÜ Lisansüstü Eğitim Enstitüsü Su Ürünleri Yetiştiricilik Anabilim Dalı Öğrencisi Nihat Sami KUYUMCU'ya ait "Türkiye'deki Akdeniz midyesi (*Mytilus galloprovincialis*) yetiştiriciliğinin mevcut durumu, sorunları ve çözüm önerileri" isimli Yüksek Lisans tez çalışmasından türetilmiştir.

DRAFT

ORAL PRESENTATION

Synthesis of a New Quinaldinium-Based Sensor and Investigation of Its Fluorescence Properties Towards Specific Metals

Arwa Alsaleh[1] (*ORCID*: *https://orcid.org/0000-0002-0587-3228)*, İbrahim Uyanık[2] (*ORCID: https://orcid.org/0000-0002-1028-8293*), Arzu Uyanık[1,*] *(ORCID: https://orcid.org/0000-0001-5545-7970*), and Mustafa Yılmaz[1] (*ORCID: https://orcid.org/0000-0003-2904-160X*)

[*1]Selçuk University, Faculty of Science, Chemistry Department, Konya, Türkiye.
[2]Selçuk University, Technology Faculty, Metallurgical and Materials Engineering Department, Konya, Türkiye.

*Corresponding author e-mail: a.uyanik80@gmail.com

Abstract

In the field of molecular fluorescence sensors, the design and synthesis of novel, highly selective, and sensitive probes for the detection of metal ions is considered one of the most important methods of detecting low concentrations of metal ions, comprising environmental monitoring and biological systems. Here the synthesized sensor contains a quinaldinium group bound to an imidazole moiety via Knoevenagel reaction, forming a new fluorescence sensor. The ion binding affinity of quinaldinium-imidazole sensor towards specific metal ions leads to increase selectivity and sensitivity in the detection process. Among various metal ions, the sensor displays a remarkable fluorescence response towards Cd^{2+}, Zn^{2+}, Cu^{2+}, and Fe^{3+} ions in acetonitrile, presenting a distinct emission spectrum for each metal ion. Moreover, the sensor's detection limits fall well within the environmentally relevant concentration ranges, making it highly suitable for trace metal ion analysis.

Keywords: Quinaldinium derivative, Fluorescence sensor, Metal-ion detection, Knoevenagel reaction.

DRAFT

ORAL PRESENTATION

Effect of temperature and salt applications on kojic acid production in *Aspergillus flavus*

Neslihan Bayrak[1*] (ORCID: https://orcid.org/0000-0002-4143-3027),
Gülsüm Ebru Özer Uyar [2] (ORCID: https://orcid.org/ 0000-0001-7164-0222)

[1]Kocaeli University, Faculty of Arts and Sciences, Department of Biology, Kocaeli, Türkiye.
[2]Kocaeli University, Faculty of Agriculture, Department of Plant Protection, Kocaeli, Türkiye.

*Corresponding author e-mail: bayrakneslihan024@gmail.com

Abstract

Kojic acid (KA) is a secondary metabolite of economic value produced by a limited number of microorganisms, including *Aspergillus oryzae, Aspergillus flavus, Aspergillus tamari*, and some other species, during the stationary phase of growth. Secondary metabolites are produced by many organisms, including molds. Secondary metabolites; they play very important roles in mold ecology by providing survival functions to the mold that produces it. One of the main roles of KA in molds is its involvement in iron uptake. The ability to bind iron is important for molds because iron is an essential nutrient for the growth and metabolism of molds. KA may also exhibit antimicrobial properties, inhibiting the growth of other microorganisms and potentially providing a competitive advantage to molds that thrive in the same environment. This antimicrobial activity helps mold protect its territory and resources. KA is also an important industrial secondary metabolite for the food and cosmetic industries. While it is used as a color regulator and preservative in the food industry, it is used as a skin lightener and antioxidant in cosmetic products. In this study, the effects of different temperatures (35°C, 40°C, and 45°C) and different salts (NaCl and KCl) at different concentrations (0.1M, 0.5M, 1M) on KA production in *A. flavus* were examined. According to the study results, it was seen that temperature had a significant effect on KA production. The highest KA yield was observed at 35°C, while the lowest yield was recorded at 45°C. Additionally, salt applications were found to negatively affect KA production. This study guides in understanding the factors affecting KA production by *A. flavus* and identifying potential strategies to increase the production of this important secondary metabolite. Additionally, the results obtained will contribute to optimizing the industrial production and use of KA.

Keywords: *Aspergillus flavus*, Kojic acid, temperature, salt

DRAFT

ORAL PRESENTATION

Metilen Mavisi Boyar Maddesinin Hibrit Nanokompozit ile Atık Sulardan Gideriminin Yanıt Yüzey Yöntemi ile Modellenmesi

Gözde Kılmen Öztürk[1*] (*https://orcid.org/0009-0007-4365-955X*),
Elçin Demirhan Yılmaz[2] (*https://orcid.org/0000-0002-4885-2902*)

[*1]Yıldız Teknik Üniversitesi, Kimya Metalurji Fakültesi, Kimya Mühendisliği, İstanbul, Türkiye.
[2] Yıldız Teknik Üniversitesi, Kimya Metalurji Fakültesi, Kimya Mühendisliği, İstanbul, Türkiye.

*gozdekilmen@gmail.com

Abstract

Teknolojinin gelişmesi ve endüstriyel faaliyetlerin artması sonucunda açığa çıkan çeşitli atıkların sulara salınması; su, toprak ve atmosfer gibi farklı ekolojik sistemlere zarar vermektedir. Özellikle boya, tekstil, kâğıt endüstrisi gibi çeşitli sektörlerin işletmelerinde uygulanan kimyasal prosesler sonucu oluşan atık sular yüksek miktarda boyar madde içermektedir. Ürünlerin üretimi sonucunda çevreye salınan atıkların minimum seviyede olması, arıtımı yapılacak su içerisindeki boyar maddelerin yok denecek kadar aza indirgenmesi gibi kriterler göz önünde bulundurulduğunda sulardan boyar madde giderimi oldukça önem taşımaktadır. Günümüzde boyar maddelerin giderimi için farklı yöntemler uygulanabilmektedir. Literatürde bu uygulama için en çok adsorpsiyon yöntemi kullanılmakta ve farklı adsorbent maddeler araştırılmaktadır. Kullanılan adsorbent maddenin kolay ulaşılabilir olması, ucuz ve giderilecek madde ile uyumlu olması önemli kriterlerdir. Bu çalışmada, özellikle endüstriyel faaliyetler sonucunda yaygın bir şekilde ortaya çıkan atık sulardan boyar madde bertarafını sağlamak için yeni bir nanoadsorban malzeme sentezlenmiştir. Nanoadsorban madde, atık olarak değerlendirilen kestane kabuğu ve TiO_2 malzemesinin sol-gel yöntemiyle bir yapı oluşturması sonucu elde edilmiştir. Bu malzeme sentezinin amacı atık malzemeyi, farklı bir atığı gidermek için kullanarak değerlendirmektir. Kestane kabuğu ve TiO_2 den elde edilen adsorbent madde ile metilen mavisi boyar maddesinin giderimi üzerindeki etkisi incelenmiştir. Deneysel çalışmalar, Box-Behnken deneysel tasarımı ile bağımsız değişken olan sıcaklık, pH ve adsorbent miktarı parametreleri için bağımlı değişken %giderim değerleri olacak şekilde planlanmıştır. Farklı sıcaklık, pH ve adsorbent miktarı için deneysel çalışmalar yapılarak Metilen Mavisi için % giderim değerleri belirlenmiştir. Bu değerler kullanılarak, Design Expert programı ile çok değişkenli analiz gerçekleştirilmiştir. Bu deneysel tasarım planı için elde edilen boyar madde giderim verimliliği modellenerek, bu verimliliği en iyi şekilde temsil eden model denklemi belirlenmiş ve en iyi deney koşulları için optimizasyon yapılmıştır.

Keywords: Kestane kabuğu, TiO_2, Sol-jel yöntemi, Boyar madde giderimi, Nanoadsorban Madde, Box-Behnken

Teşekkür: Bu çalışma Yıldız Teknik Üniversitesi Bilimsel Araştırma Projeleri Koordinasyon Birimi tarafından FYL-2022-5157 nolu proje kapsamında desteklenmiştir

DRAFT

ORAL PRESENTATION

Luteolin alleviates genotoxicity by attenuating micronucleus formation and cellular damage in acrylamide-exposed embryonic fibroblast cells

Burcu Keskin[1] (ORCID: *https://orcid.org/0000-0003-1856-4798)*, Banu ORTA YILMAZ[2] (*ORCID: https://orcid.org/0000-0002-8006-1107*), Yasemin AYDIN[2*] (*ORCID: https://orcid.org/0000-0002-4338-4687)*

[1] Istanbul University, Institute of Graduate Studies in Science and Engineering, Department of Biology, Istanbul, Turkey
[2] Istanbul University, Faculty of Science, Department of Biology, Istanbul, Turkey

*Corresponding author e-mail: yastun@istanbul.edu.tr

Abstract

Acrylamide is a white, crystalline, odorless compound that is formed during processes such as frying and roasting used in the preparation of foods. Acrylamide has been identified in cigarette smoke and sewage, in addition to being used as polyacrylamide in industry. Acrylamide intake is inevitable due to its existence in foods that are often consumed in daily life, such as bread, coffee, and french fries. Researchers have been attempting to find ways to mitigate the cytotoxic and genotoxic effects of chemicals, including acrylamide, that are always present in our bodies. Knowledge of the harmful effects of acrylamide has led researchers to undertake studies to minimize these effects. Flavonoids, plant secondary metabolites, have been studied for their antioxidant properties, which protect against cellular damage and preserve vitamin and enzyme deposits in fruits and vegetables. Compared to other flavonoids, luteolin is a potent antioxidant with strong DNA-protective activity. In this study, the cytotoxicity of acrylamide in 3T3 embryonic fibroblast cells was determined by the cell viability and lactate dehydrogenase tests. The DNA damage caused by acrylamide in 3T3 embryonic fibroblast cells and the protective effect of luteolin against this damage were evaluated by cytokinesis-blocked micronucleus analysis. As a result, the concentrations of acrylamide exposed to embryonic fibroblast cells have been found to significantly decrease cell viability while increasing the levels of lactate dehydrogenase enzymes and the incidence of micronuclei. On the other hand, luteolin treated in combination with acrylamide has been shown to prevent genotoxicity as well as its therapeutic effects on embryonic fibroblast cells.

Keywords: Acrylamide, Cytotoxicity, Embryonic fibroblast cells, Genotoxicity, Luteolin.

DRAFT

ORAL PRESENTATION

DFT Studies of A Salicylaldehyde Based Hydrazone Ligand

Ayşin Zülfikaroğlu[1*] (*ORCID: https://orcid.org/0000-0002-2871-844X*)

[*1]Amasya University, Faculty of Arts and Sciences, Department of Chemistry, Amasya, Türkiye.

*Corresponding author e-mail: aysin.zulfikaroglu@amasya.edu.tr

Abstract

In this study, some quantum chemical calculations were carried out using density functional theory (DFT) on N'-(3-ethoxy-2-hydroxybenzylidene)acetohydrazide ligand. The moleculer structure of this compound was optimized at the B3LYP/6-311G+(d,p) level. The Natural Population Analysis (NPA) charges of the ligand in gas, aqueous and ethanol medium were obtained from Natural Bond Orbital (NBO) analysis, and its coordination mode was estimated. To predict the chemical reactivity of this compound, its molecular electrostatic potential (MEP) surface map was investigated, and some of its global reactivity descriptors (chemical potential μ, electronegativity χ, hardness η and electrophilicity index ω) were calculated using DFT. The interaction of the hydrazone ligand with some heavy metals such as Sn, Hg, Cd, and Zn was investigated by the Charge Transfer (ΔN) and Energy Lowering (ΔE) parameters.

Keywords: Hydrazone, DFT, Metal-Ligand interaction.

DRAFT

ORAL PRESENTATION

Investigation of the effects of the phenolic extracts obtained from the different industrial food wastes on the gelatin modification

Huseyin Demircan[1*, 2] (*ORCID: https://orcid.org/0000-0003-3375-8777*), Omer Said Toker[1] (*ORCID: https://orcid.org/0000-0002-7304-2071*), Rasim Alper Oral[2] (*ORCID: https://orcid.org/0000-0001-7530-7478*)

[*1]Yıldız Technical University, Faculty of Chemistry and Metallurgy, Food Engineering Department, İstanbul, Turkey.
[2]Bursa Technical University, Faculty of Engineering and Natural Sciences, Food Engineering Department, Bursa, Turkey.

*Corresponding author e-mail: huseyin.demircan@btu.edu.tr

Abstract

From production to consumption, the rheological and textural properties of gelatin play a crucial role in determining the quality of gelatin based products. In this study, covalent modification of bovine skin gelatin was conducted under alkaline conditions using oxidised phenolic extracts from four different industrial food wastes, namely pomegranate peel (PP), grape pomace and seed waste (GPS), black tea waste (BT), grean tea waste (GT), at varying concentrations (1%, 3%, and 5% w/w, based on gelatin). Dynamic oscillatory measurements were performed by using stress and strain-controlled rheometer (Anton Paar, MCR-302, Austria) equipped with a parallel plate (PP25). To obtain bloom, rupture strength, brittleness and adhesiveness values, a TA-HD Plus Texture Analyzer (Stable Micro Systems, Godalming, UK) equipped with a AOAC plunger (P/0.5) was used. The effect of the modification on the color (L, a, b), clarity, total phenolic content, ABTS and DPPH antioxidant activity, free amino content (FAC), microstructure (scanning electron microscopy (SEM)), Fourier transform infrared (FTIR) spectroscopy, differential scanning calorimetry (DSC), circular dichroism (CD) spectroscopy, zeta potential, swelling, water holding capacity, emulsion activity and sability, foam capacity and stability properties of the gelatin was also investigated. Extraction yields of the PP, GPS, BT, and GT were 40.74%, 14.41%, 6.71%, and 4.22% on a dry weight basis, respectively. Bloom value was increased for all concentration of GPS and 5% of BT, while decreased for rest of the extract and concentration combinations. Gelling and melting temperatures increased with increasing extract concentration, with PP having the highest effect, followed by BT, GPS, and GT, respectively. Our results showed that gelatin properties can be modified using covalent interaction with various type of oxidised industrial food waste phenolic extracts at different concentration levels.

Keywords: Gelatin, Covalent modification, Industrial food waste, Phenolic extract, Rheology, Texture.

DRAFT

ORAL PRESENTATION

Unrevealing the EECC Water Reduction Mechanism of Cobalt based Planar Catalyst: Insights from Ab-initio Molecular Dynamics and Density Functional Theory

Ipek Guclu[1] (*ORCID: https://orcid.org/0000-0003-0375-9111),*
Yeliz Gurdal[1*] (*ORCID: https://orcid.org/0000-0002-6245-891X)*

[1]Adana Alparslan Türkeş Science and Technology University, Faculty of Engineering, Department of Bioengineering, Adana, Turkiye.

*ygurdal@atu.edu.tr

Abstract

Finding a sustainable replacement for fossil fuels is one of today's challenges, and hydrogen appears to be a promising option. One sustainable method for molecular hydrogen (H_2) production is photocatalytic water reduction, catalyzed by molecules that have transition metals as reaction centers. Studies have demonstrated that Cobalt-based catalysts exhibit significant activity in photocatalytic water reduction reactions, providing an economical alternative to costly transition metals such as ruthenium and platinum. In this study, we investigate the EECC water reduction reaction mechanism of a planar Co-based poly-pyridyl water reduction catalyst in explicit solvent environment using Ab-initio Molecular Dynamics (AIMD), Density Functional Theory (DFT), and Free Energy Perturbation Theory. We model two reduction and two protonation steps through AIMD simulations, analyze allowable spin states, and examine solvent responses following electron injections. The electronic structures of selected AIMD snapshots are further characterized using the hybrid density functional within the DFT framework. Finally, we calculate the first and second reduction free energies utilizing Marcus theory of electron transfer.

Keywords: H_2 production, Cobalt based poly-pyridyl catalyst, reduction free energy

This study has received funding from the TUBITAK under the 3501 Career Development Program (grant agreement No: 120Z240).

DRAFT

ORAL PRESENTATION

Farklı pişirici sistemlerle çapraz bağlanan floroelastomerlerin (FKM) gerilim-gevşeme özellikleri

Derya Kaya[1,2] (0000-0001-6217-0775), Tuba Ünügül[1,3] (0000-0003-0509-8425), Bağdagül Karaağaç[1*] (0000-0001-8747-8004)

[1]Kocaeli Üniversitesi, Mühendislik Fakültesi, Kimya Mühendisliği Bölümü, Kocaeli, Türkiye.
[2]Ünver Group, 16315, Nilüfer, Bursa, Türkiye.
[3]Özka Lastik, 41140, Başiskele, Kocaeli, Türkiye.

*Sorumlu yazar e-mail: bkaraagac@kocaeli.edu.tr

Özet

Floroelastomerler (FKM), ana zincirinde ve/veya karbon atomlarına bağlı yan zincirlerde flor atomları içeren sentetik kauçuklardır. Koruyucu etkisi ve yapısında bulunan C-F bağının yüksek bağ enerjisi sayesinde, diğer kauçukların karşılayamadığı yüksek sıcaklıkta sürekli performans gösterebilme (225 °C'ye kadar), değişken atmosferik koşullara dayanım, aşındırıcı kimyasallara ve petrol türevli hidrokarbonlara (yağ, yakıt gibi) karşı mükemmel direnç ve düşük gaz geçirgenliği gibi özellikleri ile bilinirler. Bu özellikleri sayesinde savunma sanayi uygulamaları başta olmak üzere, reaktör kaplamaları ve turbo şarj hortumları gibi özel uygulamalarda tercih edilmektedirler. Çok sayıda üstün özelliği birlikte taşıyan FKM kauçuk, yapısındaki güçlü flor bağları nedeniyle klasik vulkanizasyon yöntemleri ile çapraz bağlanamayan, kullanım alanındaki gereksinimlere göre çapraz bağlayıcı sistem tasarımı gerektiren bir hammaddedir. FKM vulkanizasyonunda diamin vulkanizasyonu (iyonik reaksiyon), bisfenol AF vulkanizasyonu (iyonik reaksiyon) ve peroksit vulkanizasyonu (serbest radikal reaksiyonu) olmak üzere üç farklı mekanizma uygulanmaktadır. Diamin vulkanizasyonu kauçuk-metal yapışmasının istendiği ve dinamik uygulamalarda tercih edilirken, bisfenol AF vulkanizasyonu kalıcı deformasyonun azaltılması ve ısıl dayanımın geliştirilmesi amacıyla, peroksit vulkanizasyonu ise hızlı ve görece düşük maliyetli uygulamalarda tercih edilir. Literatürde, FKM için adı geçen vulkanizasyon sistemlerinin çaprazbağ yoğunluklarının ve vulkanizat mekanik özelliklerinin değerlendirildiği çalışmalara rastlanmaktadır. Bununla birlikte, genellikle savunma sanayi gibi yüksek performans gerektiren uygulamalarda sıklıkla karşılaşılan gerilme etkisinde ısınma sırasındaki malzeme davranışlarının incelendiği ve vulkanizasyon sistemine bağlı olarak oluşan çaprazbağ tiplerinin malzeme özellikleri üzerindeki etkilerinin değerlendirildiği bir çalışmaya rastlanmamıştır. Bu çalışmada, farklı vulkanizasyon sistemleriyle hazırlanan FKM esaslı hamurların çaprazbağ yapıları ve çaprazbağ yoğunlukları kauçuk proses analizörü (RPA) kullanılarak Lee-Pawlowski-Coran yaklaşımına göre belirlenmiş, ısınma sırasındaki gerilim durulması davranışları ve karakteristik sıcaklıkları sıcaklık taramalı gerilim-durulması cihazı (TSSR-metre) yardımıyla araştırılmıştır. Diamin, bisfenol-AF ve peroksit vulkanizasyonu sistemleri birbirleriyle ve kendi içlerinde pişirici sistem bileşenlerinin seviyeleri açısından karşılaştırmalı olarak değerlendirilmiştir.

Teşekkür: Bu çalışma, Kocaeli Üniversitesi BAP Birimi tarafından FYL-2023-3374 no'lu proje kapsamında desteklenmiştir.

Anahtar Kelimeler: Çapraz bağlanma, Floroelastomer (FKM), Sıcaklık taramalı gerilme-durulma (TSSR) analizi

DRAFT

ORAL PRESENTATION

Synthesis of novel carvacrol-triazole conjugates as anticancer agents

Selma CRNISANIN[1*] (*ORCID: https://orcid.org/0009-0009-5902-3139*), Necla KULABAŞ[2] (*ORCID: https://orcid.org/0000-0003-2273-5094*), Merve GÜRBOĞA[3] (*ORCID: https://orcid.org/0000-0003-4614-7094*), Özlem BİNGÖL ÖZAKPINAR[3] (*ORCID: https://orcid.org/0000-0003-0287-5639*), Esra AYDEMİR[4] (*ORCID: https://orcid.org/0000-0002-6965-2838*), Fikrettin ŞAHİN[5] (*ORCID: https://orcid.org/0000-0003-1503-5567*), Sinem HELVACIOĞLU[6] (ORCID: *https://orcid.org/ 0000-0002-8463-3571*), Muhammed HAMİTOĞLU[7] (*ORCID: https://orcid.org/0000-0002-4545-0756*), İlkay KÜÇÜKGÜZEL[8] (*ORCID: https://orcid.org/0000-0002-7188-1859*)

[*1] Institute of Health Sciences, Marmara University, İstanbul, Türkiye
[2] Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Marmara University, İstanbul, Türkiye
[3] Department of Biochemistry, Faculty of Pharmacy, Marmara University, İstanbul, Türkiye
[4] Department of Biomedical Engineering, Faculty of Engineering and Natural Sciences, Biruni University, Zeytinburnu, 34010, İstanbul, Turkey
[5] Department of Genetics and Bioengineering, Faculty of Engineering, Yeditepe University, İstanbul, Türkiye
[6] Department of Pharmaceutical Toxicology, Faculty of Pharmacy, İstinye University, İstanbul, Türkiye
[7] Department of Pharmaceutical Toxicology, Faculty of Pharmacy, Yeditepe University, İstanbul, Türkiye
[8] Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Fenerbahçe University, İstanbul, Türkiye

* e-mail: selmacrnisanin@marun.edu.tr

Abstract

Some novel triazole-bearing acetamide derivatives **7**-**20** were synthesized starting from carvacrol. Structures of all synthesized compounds were characterized by spectral techniques. *In vitro* cytotoxic activities of these molecules against five cancer lines (human breast cancer MCF-7, human lung cancer A549, human prostate cancer PC-3, human chronic myelogenous leukemia K562, human neuroblastoma SH-SY5Y cell lines) were evaluated by MTT assay. Compounds were also tested on mouse embryonic fibroblast cells (NIH/3T3) to determine their selectivity. As a result of MTT assays, it was determined that compound **9** had antiproliferative activity on MCF-7 cells with an IC_{50} value of 3.41 µM. Since the compound **10** showed over 80% antiproliferative activity on both PC-3 and MCF-7 cells, studies were carried out to determine the IC_{50} value on both cell lines. While the IC_{50} value of compound **10** for PC-3 cells was determined as 3.24 µM; IC_{50} value of this compound was >100 µM in MCF-7 cells. Apoptosis, invasion and migration studies showed that compounds **9** and **10** might be important candidates as anti-cancer agents for the treatment of breast cancer and prostate cancer, respectively. Angiogenesis studies of compounds **9** and **10** did not show toxicity on HaCaT cells at 48 hours in viability analysis. At the same time, these compounds have been shown to significantly inhibit tube formation compared to untreated cells. It was determined that compounds **9** and **10** were not genotoxic according to the OECD 487-*In vitro* Mammalian Cell Micronucleus Test, one of the genotoxicity tests. Finally, it was also determined that these compounds were not mutagenic with TA98 and TA102 strains in the presence and absence of S9.

Acknowledgments

This work was supported by The Scientific and Technological Research Council of Turkey (TÜBİTAK - Grant no. 218S549).

Keywords: 1,2,4-Triazole; carvacrol; cancer; apoptosis; angiogenesis; mutagenicity.

DRAFT

ORAL PRESENTATION

Bioadsorption of Lead (II) on Teff Seeds

Yasemin İşlek Coşkun* *(ORCID: https://orcid.org/0000-0003-3207-4381)*

*Ege University, Faculty of Science, Department of Chemistry, İzmir, Türkiye.

*Corresponding author e-mail: yasemin.islek@ege.edu.tr

Abstract

The removal of heavy metal ions, such as lead (II) from wastewater is of utmost importance due to their propensity for bioaccumulation, toxicity, persistence, and resistance to biodegradation in the environment [1]. The accumulation of lead in the human body can lead to reproductive issues, damage to the central nervous system, developmental delays in physical and mental activities, especially in childhood. Moreover, it poses potential carcinogenic and neurological risks and can even be fatal. Hence, there has been a growing interest in conducting effective studies on the removal of Pb (II) ions [1,2]. In wastewater, Pb (II) primarily originates from metal plating and finishing processes, battery manufacturing, soldering materials, printing and pigments, glass industries, ceramics, ammunition, as well as steel and iron manufacturing facilities [3]. Conventional approaches for eliminating heavy metal ions, including membrane processes, chemical precipitation, and ion exchange, have long been employed to purify water from these contaminants. Nonetheless, the use of these chemical techniques is frequently constrained by technical or economic challenges. As a result, the utilization of biomaterial-based adsorption has emerged as a viable and cost-effective alternative for industrial wastewater treatment, offering both affordability and environmental safety [1,2,4]. In the present study, the effectiveness of an agricultural material- Teff seeds (Eragrostis tef)- for the removal of Pb (II) from an aqueous solution was investigated for the first time. The effects of pH, adsorbent dose, contact time, initial Pb (II) concentration and temperature were analysed. The equilibrium isotherm, kinetic and thermodynamic models were studied. The adsorbents were also applied to real wastewater samples. Optimal experimental conditions were found to be pH 5, 0.25 g/L of adsorbent dose with 1440 min of contact time. The adsorption isotherm was described by the Langmuir model. The thermodynamic studies revealed that the adsorption was spontaneous and exothermic.

Keywords: Lead, biosorption, Teff seeds, flame atomic absorption spectrometry.

References:

[1] Biosorbent from tomato waste and apple juice residue for lead removal, E. Heraldy et al., Journal of Environmental Chemical Engineering 6 (2018) 1201–1208.
[2] Kinetics and equilibrium studies on biosorption of Pb(II) from aqueous solution by a novel biosorbent: Cyclosorus interruptus, K. Zhou et al., Journal of Environmental Chemical Engineering 3 (2015) 2219–2228.
[3] Removal of lead from aqueous solutions by agricultural waste maize bran, K.K. Singh et al., Bioresource Technology 97 (2006) 2124–2130.
[4] Adsorption of Lead on Cucumber Peel, M. Basu et al., Journal of Cleaner Production 151 (2017) 603-615.

DRAFT

ORAL PRESENTATION

Quantification of Sulforaphane from Raphanus sativus species by Reverse Phase HPLC

Esra Maltas Cagil[1] (ORCİD: 0000-0001-7574-6016), Kubra Yilmaz[2*] (ORCİD: 0000-0002-3841-3156)

[*1,2] Selcuk University, Faculty of pharmacy, Konya, Turkey.

*Corresponding author e-mail: yilmazkubra19@gmail.com

Abstract

Raphanus sativus is a plant from Brassicaceae family. There are several species such as Raphanus sativus var. niger, Raphanus sativus longipinnatus and Raphanus sativus var. Radicula. Their root and seed have been reported to be used for the prevention and treatment of gallstones, jaundice, bloating, indigestion, and various stomach ailments in traditional medicine. The sulforaphane molecule is one of their natural compounds which is known to be an anticancer molecule. In this study, sulforaphane was extracted from two types of radishes, Raphanus sativus var. Niger and Raphanus sativus Longipinnatus by a simple solid-phase extraction (SPE) method. The amount of the compound was measured by HPLC analysis. The optimal conditions in HPLC methods were developed by using a silica SPE C18 cartridge and carbon tetrachloride and dichloromethane as washing and eluting solvents, which could eliminate interferences originating from the radish. Firstly, acidic water at pH 6 was used to dissolve, and then we let it warm at 45 degrees for 2 and a half hours. After the sample was warmed, carbon tetrachloride was added to the sample to separate the two phases, and the upper phase was taken to HPLC column C18. After impurities were removed from the extracts, they were directly injected into high-performance liquid chromatography (HPLC) for further chromatographic analysis. Good linearity was obtained from 10 to 166 µg/mL ($r = 0.9999$) for sulforaphane with the relative standard deviations. The SPE method provides a higher yield of sulforaphane from crude extracts compared to conventional liquid-liquid extraction. The mean recoveries of sulforaphane from broccoli were more than 90.8%, and the detection limit was 2.70 µg/mL.

Keywords: radish, sulforaphane; solid-phase extraction; HPLC analysis

DRAFT

ORAL PRESENTATION

Chymotrypsin, trypsin, ureaz inhibitory anti antioxidant activities of *Centaurea helenioides* Boiss. & Hausskn.

Elif Dilmaç[1], Melike Sucu[2], Tuğba Günbatan[3]

[1]Department of Pharmacognosy, Faculty of Pharmacy, Lokman Hekim University, 06510, Ankara, Turkey
[2]Department of Pharmacognosy, Faculty of Pharmacy, Başkent University, 067900, Ankara, Turkey
[3]Department of Pharmacognosy, Faculty of Pharmacy, Gazi University, 06330, Ankara, Turkey

Abstract

Chymotrypsin and trypsin are key players in the digestive process, where it acts to break down dietary proteins. However, maintaining the balance of proteolytic processes is crucial for overall health and the proper functioning of biological systems. Chymotrypsin and trypsin inhibitors can be used as therapeutic agents to treat conditions characterized by excessive protease activity, such as inflammatory diseases. Inhibitors of chymotrypsin-like proteases are under investigation for their potential to modulate proteolytic activity in diseases like cancer and neurodegenerative disorders. Urease allows *Helicobacter pylori* to survive in the acidic environment of the stomach by neutralizing the acid through the production of ammonia. Therefore, urease inhibitors have become one of the mechanisms investigated for the treatment of *H. pylori* positive ulcers. In these study, it was aimed to investigate the in vitro trypsin, chymotrypsin and urease inhibitory activities of *Centaurea helenioides* Boiss. & Hausskn. In addition, its antioxidant activity were determined by three different methods (ABTS, DPPH and CUPRAC). For these purpose 80% ethanol extract and liquid-liquid fractions were prepared from flowers and analysed by in vitro methods. According to the activity test results, *n*-hexane fraction showed important chymotrypsin inhibitory activity (IC_{50}: 64.36 ± 1.36 µg/mL); while the remaining water fraction exhibited significant urease inhibition (IC_{50}: 46.59 ± 1.35 µg/mL). The prepared extracts and fractions generally showed strong antioxidant activity in all studied antioxidant methods. Based on these results, it is planned to carry out further studies to determine the compounds responsible for the activity in *Centaurea helenioides*.

Key Words: Chymotrypsin, trypsin, urease, *Centaurea helenioides*, antioxidant

Acknowledgements
We would like to thankto Prof. Dr. İlhan Gürbüz for his guidance in this research.

DRAFT

ORAL PRESENTATION

Karbazolil ve benzildiamino pendant kolları içeren (N/N)dispirosiklotrifosfazen türevlerinin sentezi: spektral, kristallografik, fotofiziksel, stereojenik özelliklerinin ve biyolojik aktivitelerinin incelenmesi

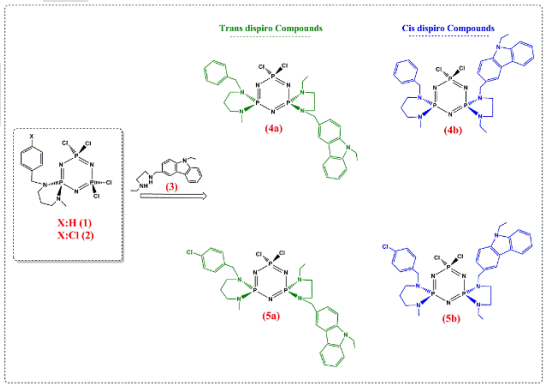
Reşit Cemaloğlu[1*] (0000-0002-2802-0754), Nuran Asmafiliz[1] (0000-0002-9335-4101), Zeynel Kılıç[1] (0000-0003-1061-8122), Bünyemin Çoşut[2] (0000-0001-6530-0205), Büşra Nur Sabah[3] (0000-0003-1678-843X), Leyla Açık[3] (0000-0002-3672-8429), Tuncer Hökelek[4] (0000-0002-8602-4382)

[1]Ankara Üniversitesi, Fen Fakültesi, Kimya Bölümü, 06560, Ankara, Türkiye
[2]Gebze Teknik Üniversitesi, Kimya Bölümü, 41400, Kocaeli, Türkiye
[3]Gazi Üniversitesi, Fen Fakültesi, Biyoloji Bölümü, 06560, Ankara, Türkiye
[4]Hacettepe Üniversitesi, Mühendislik Fakültesi, Fizik Mühendisliği Bölümü, 06800, Ankara, Türkiye

* E-mail: rcemaloglu@ankara.edu.tr

Abstract

Hekzaklorosiklotrifosfazen bileşiği (trimer, HCCP, $P_3N_3Cl_6$) fosfor ve azot atomlarının birbirine ardışık olarak bağlanması ile oluşan inorganik heterohalkalı bir bileşiktir. Termal kararlılık, biyouyumluluk ve çok yönlü reaktivite gibi önemli özelliklere sahip olan trimerik fosfazen bileşiğinin yapısında bulunan klor atomları, inorganik ve organik gruplar ile yer değiştirebilmektedir [1-4]. Bu nükleofilik sübstitüsyon reaksiyonları neticesinde, tercih edilen sübstitüentlere bağlı olarak farklı kimyasal, biyolojik özelliklere sahip halkalı fosfazen bileşikleri sentezlenebilmektedir. Ayrıca, bu nükleofilik sübstitüsyon reaksiyonları, kiral bileşiklerin sentezlenmesine de imkân tanımaktadır [5-8]. Çalışmaların ilk aşamasında, ligant olarak kullanılmak üzere benzildiamin bileşikleri sentezlenmiştir. Ardından bu diamin bileşiklerinin, trimerik fosfazen ile regioselektif olarak etkileşmesinden monospirofosfazenler (**1**, **2**) ve oluşan bu bileşiklerin karbazolildiamin (**3**) ile tepkimelerinden cis/trans dispirofosfazenler (**4a**/**4b**, **5a**/**5b**) hazırlanmıştır. Bu bileşikler, spiro halkadaki metil ve/veya etil gruplarının uzaysal oryantasyonu uyarınca cis ve trans geometrik izomerler halinde elde edilmiştir. Bu orijinal bileşiklerin; **spektroskopik**, **stereojenik özellikleri**, **kristal yapıları**, **fotofiziksel özellikleri** ve **biyolojik aktiviteleri** araştırılmıştır. Yapısal karakterizasyon çalışmaları, tek boyutlu NMR teknikleri ve X-ışını kırınımmetre yöntemi kullanılarak gerçekleştirilmiştir. Bileşiklerin kiral özellikleri, X-ışını kristallografi yöntemi ve kiral çözücü (chiral solvating agent; CSA) ilaveli ^{31}P NMR spektroskopisi yöntemi ile detaylı bir şekilde incelenmiştir. Biyolojik aktivite çalışmaları kapsamında, bileşiklerin antibakteriyel ve antifungal aktiviteleri belirlenen veriler yardımı ile karşılaştırmalı olarak değerlendirilmiştir. Ayrıca, bileşiklerin fotofiziksel özellikleri UV-Vis ve Floresans spektroskopisi yöntemleri ile incelenmiştir. Bununla birlikte, bileşiklerin pBR322 plazmid DNA ile etkileşimleri araştırılmıştır.



Anahtar kelimeler: Dispirofosfazenler, trimer, spektroskopi, kristallografi, biyolojik aktivite, fotofiziksel özellikler.

Bu çalışma "122Z180" numaralı Tubitak Projesi ile desteklenmiştir.

Referanslar

1. Zibarov, A., Oukhrib, A., Aujard Catot, J., Turrin, C. O. and Caminade, A. M. 2021. "AB_5 Derivatives of Cyclotriphosphazene for the Synthesis of Dendrons and Their Applications", Molecules, 26(13), 4017.
2. Şenkuytu, E., Öztürk, E., Aydınoğlu, F., Eçik, E. T., and Okutan, E. 2020. "Cyclotriphosphazene cored naphthalimide-BODIPY dendrimeric systems: Synthesis, photophysical and antimicrobial properties", Inorganica Chimica Acta, 502, 119386.
3. Wang, Y., Wang, Z., Wang, S., Chen, Z., Chen, J., Chen, Y. and Fu, J. 2019. "Magnetic poly(cyclotriphosphazene-co-4,4′-sulfonyldiphenol) nanotubes T modified with glacial acetic acid for removing methylene blue: Adsorption performance and mechanism", European Polymer Journal, 120, 109198.
4. Jamain, Z., Khairuddean, M., and Saidin, S. A. 2019. "Synthesis and characterization of 1,4-phenylenediamine derivatives containing hydroxyl and cyclotriphosphazene as terminal group", Journal of Molecular Structure, 1186, 293–302.
5. Usri, S. N. K., Jamain, Z., and Makmud, M. Z. H. 2021. "A Review on Synthesis, Structural, Flame Retardancy and Dielectric Properties of Hexasubstituted Cyclotriphosphazene", Polymers, 13(17), 2916.
6. Beytur, A., Tekin, Ç., Çalışkan, E., Tekin, S., Koran, K. Görgülü, A. O. and Sandal, S. 2022. "Hexa-substituted cyclotriphosphazene derivatives containing hetero-ring chalcones: Synthesis, *in vitro* cytotoxic activity and their DNA damage determination", Bioorganic Chemistry, 127, 105997.
7. Asmafiliz, N., Civan, M., Özben, A., Kılıç, Z., Ramazanoğlu, N. Açık, L. and Hökelek, T. 2018. "Phosphorus-nitrogen compounds. Part 39. Syntheses and Langmuir-Blodgett thin films and antimicrobial activities of N/N and N/O spirocyclotriphosphazenes with monoferrocenyl pendant arm", Applied Organometallic Chemistry, 32(4), e4223.
8. Cemaloğlu, R., Asmafiliz, N., Çoşut, B., Kılıç, Z., Sabah, B. N., Açık, L., Mergen, H., Hökelek, T. 2023. "Phosphorus-nitrogen compounds: Part 69—Unsymmetrical dispiro(*N*/*N*)cyclotriphosphazenes containing different pendant arms: syntheses, characterization, stereogenism, photophysical and bioactivity studies", Research on Chemical Intermediates, 49, 2071-2098.

DRAFT

ORAL PRESENTATION

Valorization of de-oiled *Botryococcus braunii* biomass for bioethanol production

Benan İnan[1*] (ORCID: http://orcid.org/0000-0002-2315-3099)

[*1]Yildiz Technical University, Faculty of Chemical and Metallurgical Engineering, Bioengineering Department, Istanbul, Turkey

*Corresponding author e-mail: benan.inan@yildiz.edu.tr

Abstract

In the last two decades, microalgae have been evaluated for biodiesel production due to their high lipid content, but microalgal biomass is also a raw material for bioethanol production, because of its cellulosic structure with- out the need for other applications. *Botryococcus b*raunii is a single-celled, slow-growing photosynthetic microalgae of the green algae, that produces large amounts of biomass, lipid, carbohydrate and other bioactive molecules. It is one of the most evaluated microalgae species for biodiesel production due to its high lipid content. Although it is a prominent feedstock for biodiesel production, the biomass remained after lipid extraction should be also utilized in the aspects of zero waste management. In this context, investigating the effect of microalgal bioethanol production parameters on bioethanol yield is of great importance. In this study, de-oiled *B. braunii* microalgae was valorized for bioethanol production by evaluating the process parameters. Highest bioethanol yield was obtained as 20.42% under condition of 1 N H_2SO_4, pre-treatment temperature of 100°C, and 45 minutes of pre-treatment time, respectively.

Keywords: Bioethanol, microalgae, fermentation, biofuel, biomass

DRAFT

ORAL PRESENTATION

Expression, Purification, and Characterization of Intracellular Xylanase from *Anoxybacillus caldiproteolyticus*

Hakan KARAOĞLU[1*] (ORCID: 0000-0003-4615-1157)

[*1] Recep Tayyip Erdogan University, Faculty of Fisheries and Aquatic Sciences, Department of Basic Sciences Rize, Turkey.

*Corresponding author e-mail: hakan.karaoglu@erdogan.edu.tr

Abstract

Xylanases are the enzymes that can hydrolyse xylan, the major hemicellulose, which is found in the structure of the plant cell wall in nature. Nowadays, xylanases have been intensively used in many industrial areas such as food, textile, pulp and paper industries, ethanol production, and waste treatment. Due to its industrial importance, the discovery of more effective xylanases still attracts the attention of the scientific world. In this study, the 329 amino acid gene encoding *Anoxybacillus caldiproteolyticus* xylanase was cloned into the pET-28a(+) expression vector and the gene product was expressed in *E.coli BL21* for the first time. The produced enzyme was purified by column chromatography methods and evaluated in terms of biochemical and kinetic parameters. As a result of the study, the optimum conditions in which the enzyme showed the best activity were determined as pH 6.50 and 60 °C. The enzyme showed over 80% activity in a wide pH range such as pH 5.00-9.00. The enzyme did not lose activity between pH 6.00-9.00 at +4 °C for 75 days. K_m, V_{max}, k_{cat} and k_{cat}/K_m values of *Acal*XYN329 were calculated as 3.5929 ± 0.166 µg/µL, 58.767 ± 0.02 µmoL/min/mg protein, 75.64 1/sec, and 21.05 1/mMs $^{-1}$, respectively. In conclusion, the xylanase of *A. caldiproteolyticus* has excellent potential to be utilized in many industrial processes.

Keywords: *Anoxybacillus caldiproteolyticus*, xylanase, xylan, expression.

DRAFT

ORAL PRESENTATION

Analysis of Blood Chemistry Changes after Systemic SHOC2 Inactivation in Adult Mice

Sibel Sari[*1] (*ORCID: https://orcid.org/0000-0002-2505-5804),*
Pablo Rodriguez-Viciana[2] (*ORCID: https://orcid.org/0000-0002-8846-543X)*

[*1]Abdullah Gul University, Faculty of Life and Natural Sciences, Molecular Biology and Genetics, Kayseri, Turkey.
[2]University College London (UCL) Cancer Institute, London, UK

*Corresponding author e-mail: sibel.sari@agu.edu.tr

Abstract

The RAS-RAF-ERK pathway orchestrates a cascade of intracellular events that regulate cell growth, proliferation, and differentiation and its aberrant activation is a major driver of human cancer. The MRAS-SHOC2-PP1 (SHOC2 phosphatase) heterotrimeric complex plays an essential role in RAF-ERK pathway activation by dephosphorylating a specific phosphoserine site on RAF kinases. To investigate the function of SHOC2 in tissue homeostasis, we have generated Tamoxifen-inducible Cre mice that are homozygous for floxed SHOC2. To this end, we crossed $SHOC2^{fl/fl}$ conditional knockout (KO) mice with Rosa26-$CreER^{T2}$ mice to generate $SHOC2^{fl/fl}$ KO Rosa26-$CreER^{T2}$ mice, which carry an inducible ubiquitously expressed $CreER^{T2}$ recombinase. The administration of tamoxifen results in highly effective recombination of more than 80% across all investigated tissues, excluding the brain. Serum biochemical assessment helps predict pathological processes in vital internal organs of the body, such as the liver, muscle, heart, pancreas, and kidney. To elucidate alterations in metabolism and organ functions resulting from systemic SHOC2 deletion, blood was collected from wildtype and SHOC2 knockout mice via cardiac puncture at morbidity, and serum biochemical parameters were analysed. The blood test for the biochemistry profile included a series of biomarkers for liver and kidney functions, lipid levels, electrolytes, as well as additional metabolic markers such as glucose, fructose, total protein, and albumin. Most of the serum chemistry parameters were unaffected suggesting SHOC2 inactivation does not cause major and/or clear alterations in liver or kidney function. However, albumin, total cholesterol, high-density lipoprotein, glucose, fructose and alkaline phosphatase levels were significantly decreased in SHOC2 KO mice compared to wild type controls. While the significance of these changes is unclear, they are consistent with the weight loss that was observed in the SHOC2 KO mice at the time of analysis and thus may be the reflection of weight loss like consequence rather than the cause.

Keywords: SHOC2, Mouse Models, Blood Biochemical Markers

DRAFT

ORAL PRESENTATION

Determination of Biological Activities of DMSO Extracts Obtained from Sesame and Black Cumin Seed Oils

Aleyna OGRETEN[1] (*ORCID: https://orcid.org/0009-0001-9975-8916*),
Erdal EROGLU[*1] (*ORCID: https://orcid.org/0000-0001-7281-3906)*

[*1]Manisa Celal Bayar University, Faculty of Engineering, Bioengineering, Manisa, TURKIYE.

*Corresponding author e-mail: erdal.eroglu@cbu.edu.tr

Abstract

Cancer is one of the most common disease threads lives of billions worldwide. The increasing number of cancer-related deaths, the lack of a definitive cure, and the serious side effects of treatment for patients have accelerated the discovery of natural based cancer preventives or therapeutics. Essential oils extracted from different plants such as sesame and black cumin have been studied for a long time as natural anticancer agents. However, insolubility of oily agents in aqueous solutions limits the accuracy of the in vitro assays such as cell culture, antioxidant, phenolic content assays etc. In this study, we report, for the first time, content analysis and biological activities of DMSO extracts of sesame and black cumin oils in vitro. To determine the phenolic content, antioxidant capacity and flavonoid content of DMSO extracts, Folin-Ciocalteu, CUPRAC and aluminum chloride calorimetric assays were performed, respectively. In biological tests, MTT cell proliferation assays against human bladder cancer (T-24) for anticancer activity and disk diffusion assay against S. *aureus* and E. *coli* for antibacterial activity were conducted. According to content analysis results, DMSO extracts were found to be rich in phenol (4391 mg gallic acid equivalent/ml extract for sesame; 6200 mg gallic acid equivalent/ml extract for black cumin) and flavonoid (546 mg catechin equivalent/ml extract for sesame; 1100 mg catechin equivalent/ml extract for black cumin) contents. In antibacterial tests of DMSO extracts, which were also found to have high antioxidant capacity, showed that zone formation was found in DMSO extract obtained from black cumin. According to in vitro MTT cell proliferation assay, the IC_{50} value of the obtained DMSO extracts from sesame and black cumin were found 1.47% and 0.37% against T-24 cells, respectively. A novel method, presenting an alternative extraction technique for herbal products characterized by complex phytochemical components, was introduced for the first time in this study.

Keywords: sesame, black cumin, natural agents, anticancer

DRAFT

ORAL PRESENTATION

Determination of total antioxidant capacity and GC-MS content of *Malus floribunda coccinella* fruit

Ebru Coteli* (*ORCID: https://orcid.org/0000-0002-9473-0914*)

*Ahi Evran University, Vocational School of Health Services, Kirsehir, Turkey

*Corresponding author e-mail: e.coteli@ahievran.edu.tr

Abstract

Malus floribunda coccinella is a plant used for landscaping purposes because it has small red fruits and pinkish flowers. The aim of the study is to determine whether the fruits of the plant have different usage areas by performing GC-MS analyses of their total antioxidant capacity. To determine the total antioxidant capacity of methanol extracts of plant fruits, total phenolic and flavonoid analyses, and DPPH· radical scavenging activities were examined. The content of methanol extracts from plant fruits was determined by the GC-MS (gas chromatography/mass spectroscopy) method. A spectrophotometer device was used in total antioxidant capacity analyses. The total phenolic amount (46.55±2.15 mg GAE/g extract), total flavonoid amount (61.37±2.49 mg QAE/g extract) of the methanol extract of the fruits were determined. Solutions of fruit methanol extract at 250, 500, and 1000 µg/mL concentrations were prepared, and their inhibition was calculated. Additionally, the IC_{50} values of samples with known inhibition percentages were also calculated. The IC_{50} value of the methanol extract of the plant fruit was determined as (876.91 µg/mL). In addition, fifteen different substances were identified as a result of the GC-MS analysis of the fruit methanol extract. Especially 1,6-Anhydro-beta-D-glucofuranose and beta-D-Glucopyranose, 1,6-anhydro- substances were found in high amounts. As a result, the fruits of this plant, whose phytochemical content was determined, were found to have antioxidant properties. It was concluded that the substances identified as a result of the GC-MS analysis were important and used in different fields. It is thought that this study will contribute to the plant literature.

Keywords: *Malus floribunda coccinella*, Fruit, Total antioxidant capacity, GC-MS

DRAFT

ORAL PRESENTATION

Comparison of the Effects of Lead Toxicity on Chlorophyll Content in Some Wheat Varieties with Classical and Modern Methods

Betülnur ÖZEL[1] , Elif ÖZTETİK[2] (*ORCID: https://orcid.org/0000-0002-9663-6884*)

Eskişehir Technical University, Science Faculty, Biology Department, Eskişehir, Türkiye.

*Corresponding author e-mail: betulnurozel@ogr.eskisehir.edu.tr, eoztetik@eskisehir.edu.tr

Abstract

Events that disrupt or stop the development and physiological processes in plants are called stress. In this sense, heavy metals (HMs) are one of the important stress factors that threaten the organism by accumulating in ecosystems. HMs affect many cellular activities negatively, such as protein synthesis, respiration, photosynthesis, mineral nutrient intake and membrane structure. Of those, Lead (Pb), is used for a wide variety of purposes and produced over several million tons annually. Photosynthesis, as a physiological processes, is one of the most sensitive processes to Pb toxicity and chlorophyll is the key pigment of this process for all living things demanding energy. Chlorophyll converts light energy into chemical energy. Therefore, chlorophyll content is an important indicator in determining the characteristics of a plant such as development, photosynthetic capacity, stress and yield. The level of stress in the plant can be determined by measuring the amount of chlorophyll. Extraction of leaf tissue and subsequent spectrophotometric measurements is a common method used in chlorophyll measurement and determines the amount of chlorophyll per gram in leaf tissue. As an alternative to this method, portable chlorophyll meters have been developed that save time. Chlorophyll meters give a result expressed as SPAD, indicating only the relative chlorophyll content. So, it is important to create reference curves for the SPAD value and the total amounts of chlorophyll obtained by the extraction method to decipher the level of relationship between the two methods. In this study, the effect of different concentrations of lead chloride ($PbCl_2$) exposure on chlorophyll contents in two different wheat varieties *(Triticum aestivum cv.* Bezostaya and *cv*. Yunus) were investigated. Measurements were made by two different methods, portable chlorophyll meter and extraction method. When the of both methods were compared, it was determined that the results of these two varieties were consistent with each other.

Keywords: Heavy metal (Pb), Wheat, Chlorophyll Content

DRAFT

ORAL PRESENTATION

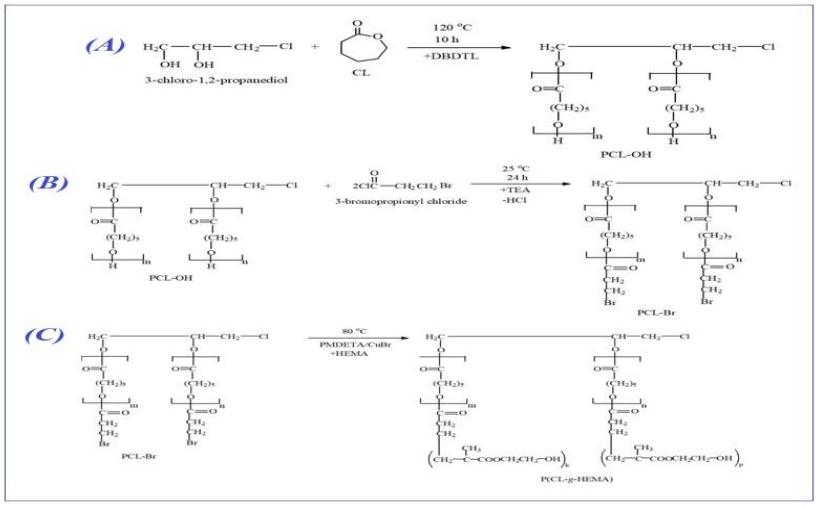
Graft copolymer hydrogel synthesis and characterization via controlled/living radical polymerization techniques and investigation of hydrogel properties

Melahat Göktaş*[1] and Ümran Aslan[2]

[1]Van Yüzüncü Yıl University, Faculty of Education, Department of Chemistry, Van, 65080 Turkey
[2]Van Yüzüncü Yıl University, Institute of Science, Van, 65080 Turkey
*Corresponding author: E-mail: melahat_36@hotmail.com

Abstract

Graft copolymers continue to be of increasing importance, with numerous studies from synthesis to application reported each year. With the use of controlled radical polymerization (CRP) techniques, it is possible to create perfectly controlled complex macromolecular structures **[1,2].** While a graft copolymer obtained from a single polymerization method enables a facile synthesis, the combination of two different polymerization techniques can be an advantageous way to incorporate classes of monomers that would not be compatible, such as lactones and vinyl monomers **[3]**. In this contribution, poly(ε-caprolactone-g-2-hydroxy ethyl methacrylate) [P(ε-CL-g-HEMA)] graft copolymer hydrogels were synthesized in two polymerization steps by atom transfer radical polymerization (ATRP) and ring-opening polymerization (ROP) methods. For these syntheses, hydroxyl-terminated poly ε-caprolactone (PCL-OH) was obtained by the ring-opening polymerization method of ε-caprolactone by using 3-chloro-1,2-propanediol initiator, which is suitable for the ring-opening polymerization method, in the first step. From the reaction of this synthesized PCL-OH and 3-bromine propionyl chloride, a new bromine-terminated poly ε-caprolactone (PCL-Br) was synthesized to be used as an ATRP initiator. In the second step, poly(ε-caprolactone-g-2-hydroxy ethyl methacrylate) [P(ε-CL-g-HEMA)] graft copolymer hydrogels were synthesized by ATRP polymerization of 2-hydroxy ethyl methacrylate (HEMA) in the presence of the new synthesized ATRP initiator, and hydrogel properties were examined. The routes of these syntheses are shown in Scheme 1. The obtained initiator and graft copolymer hydrogels were characterized by spectroscopic methods such as 1H-NMR, FT-IR, TGA, DSC, GPC and SEM.



Scheme 1. Synthesis mechanism of PCL-OH ***(A),*** Chemical synthesis reaction of PCL-Br ***(B),*** Synthesis mechanism of PCL-*g*-PHEMA graft copolymer hydrogels (***C)***.

Keywords: Atom transfer radical polymerization (ATRP); graft copolymer; hydrogels; Ring-opening polymerization (ROP); bromine-terminated poly(ε-caprolactone) (PCL-Br).

References: 1. Chiefari, J.; Chong, YK.; Ercole, F.; Krstina, J.; Jeffery, J.; Le, TPT.; Mayadunne, RTA.; Meijs, GF.; Moad, CL.; Moad, E.; Rizzardo, E.; Thang, SH. (1998) Macromolecules 16:5559-5562.
2. Wang, J. S., Matyjaszewski, K. (1995) Macromolecules 28: 7901-7910.
3. Göktaş M, Aykaç C, Öztürk T (2022) J Chem Sci 134:73.

ORAL PRESENTATION

Vincristin ile indüklenen bağırsak toksisitesinde quercetin'in koruyucu etkilerinin incelenmesi

Mustafa İleritürk[1*] (*ORCID: https://orcid.org/0000-0002-4581-4492*),
Özge Kandemir[2] (*ORCID: https://orcid.org/0000-0001-8884-4168*)

[*1] Ataturk University, Horasan Vocational College, Department of Animal Science, Erzurum, Türkiye
[2] Aksaray University, Aksaray Technical Sciences Vocational School, Aksaray, Türkiye

*Sorumlu yazar e-mail: m.ileriturk@atauni.edu.tr

Özet

Bu çalışmada, çeşitli kanser türlerinin tedavisinde etkin olarak kullanılan vinkristin'in neden olduğu bağırsak hasarında quercetin'in koruyucu etkileri araştırılmıştır. Çalışmada, erkek Sprague Dawley sıçanlarına 12 gün boyunca oral olarak 25 ve 50 mg/kg dozlarda quercetin verildi ve 30 dakika sonra intraperitoneal olarak 0,1 mg/kg dozda vinkristin (kümülatif doz 1,2 mg/kg) uygulandı. Oksidatif stres düzeyi biyokimyasal metotlarla tespit edildi. Nuclear Factor kappa B (NF-κB), Interleukin-1 beta (IL-1β), Bcl-2 ile ilişkili X proteini (Bax), B hücreli lenfoma-2 (Bcl-2) ve caspase-3'ün mRNA transkript seviyeleri RT-PCR ile belirlendi. Bağırsak dokusunda glutatyon (GSH) düzeyleri ile süperoksit dismutaz (SOD), katalaz (KAT), glutatyon peroksidaz (GPx) aktivitelerinin azalması, malondialdehit (MDA) düzeylerinin artması, vinkristin kaynaklı NF-κB, IL-1β, Bax, Bcl-2 ve caspase-3 ekspresyonlarındaki artışın sırasıyla oksidatif strese, inflamasyona ve apoptoza neden olduğu, ancak quercetin'in vinkristin kaynaklı oksidatif stresi, inflamasyonu ve apoptozu giderdiği tespit edildi. Bu sonuçlar sıçanlarda quercetin'in vinkristin kaynaklı bağırsak toksisitesine karşı koruyucu etki gösterebileceğini göstermektedir.

Anahtar Kelimeler: Apoptoz, Bağırsak toksisitesi, İnflamasyon, Oksidatif stres, Quercetin, Vincristin

DRAFT

ORAL PRESENTATION

Synthesis and characterization of some new coumarin-perimidine hybrid compounds

Nesrin Ünal Karaali[1*] (*ORCID: https://orcid.org/0000-0003-0459-6975*)

[*1]Recep Tayyip Erdogan University, Art and Science Faculty, Department of Chemistry, Rize, Turkey

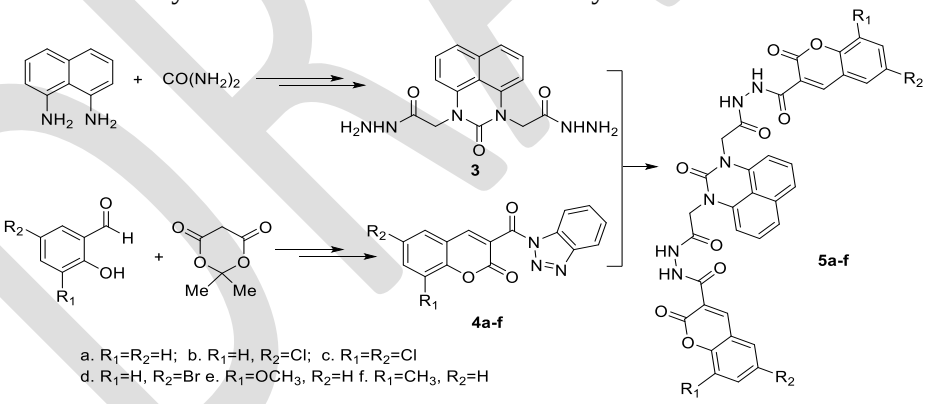
*Corresponding author e-mail: nesrin.karaali@erdogan.edu.tr

Abstract

Coumarin and its derivatives are important chemicals used in various fields such as fluorescent probes, cosmetic industries, and coumarin dyes. Also, coumarin derivatives have shown a wide spectrum of pharmacological activities such as antibacterial, anticoagulant, antifungal, antimycobacterial, and anti-inflammatory. In the present times, great importance is attached to the research of perimidines and their derivatives. This interest is not only due to its various biological activities such as anti-cancer, anti-inflammatory, antioxidant, antibacterial, but also due to various applications such as dyes, photochemical sensor, DNA binding properties. Molecular hybridization has become a powerful method for the synthesis of new and more activated molecules by bringing together active compounds in pharmaceutical or other fields. Considering the literature activities of coumarin and perimidine structures, the use of these heterocycles in hybrid form may lead to the formation of more active structures. With this in mind, in this study, the hydrazide derivative perimidine compound was synthesized, starting from the reaction of 1,8-naphthalidiamine and urea. And also the synthesis of coumarin derivatives was carried out in several steps, starting with the reaction of different 2-hydroxybenzaldehyde compounds and 2,2-dimethyl-1,3-dioxane-4,6-dione compound. In the final step, coumarin-perimidine hybrid compounds were obtained as a result of the reaction of the obtained hydrazide derivative perimidine and coumarin compounds.

Keywords: Coumarin, Perimidine, Hybrid Molecule.

Synthesis of Coumarin-Perimidine Hybrid Molecule



DRAFT

ORAL PRESENTATION

Fabrication and characterization of hydrogels from bovine achilles tendon collagen and human hair keratin

Yasemin Pazarçeviren[1] (*0000-0001-5966-4715*), Özge Erdemli[1*] (*0000-0001-8606-8863*)

[*1]Başkent University, Faculty of Science and Letters, Molecular Biology and Genetics, Ankara, Turkey

*Corresponding author e-mail: oerdemli@baskent.edu.tr

Abstract

In recent years, there has been a growing interest in the development of hydrogels that utilize natural and sustainable sources instead of relying solely on synthetic materials. In this study, the aim was to fabricate sustainable hydrogels made up of collagen/keratin/alginate (C/K/A) and characterize their physical, chemical and biological properties. In this regard, keratin was extracted from waste human hair by reduction method, and collagen was obtained from bovine achilles tendon via acid solubilization and enzymatic hydrolysis. Hydrogels, cross-linked with calcium chloride and EDC/NHS, were produced by using different weight ratios of 1:1:1 and 2:2:1 C/K/A. The FTIR spectra obtained from isolated keratin and collagen demonstrated the existence of characteristic absorption peaks. In SDS-PAGE analysis, α-keratin and β-keratin bands and the γ-collagen, β-collagen and α1(III), α1(I), α2(I) collagen bands were observed to confirm the keratin and collagen structures. Physicochemical properties of hydrogels such as water uptake, *in vitro* hydrolytic degradation, porosity, chemical and microstructural properties were examined. Porosity was higher for the 1:1:1 C/K/A (80%) than 2:2:1 C/K/A (47%), yet the amount of weight loss was observed to be higher for the 2:2:1 C/K/A group at the end of 21 days. The swelling ratio of 2:2:1 C/K/A (1337%) is found to be lower than 1:1:1 C/K/A (2063%). Lastly, L929 mouse fibroblast cells interacted with the hydrogels, and it was observed that 1:1:1 C/K/A group supported the highest cell proliferation rate.

Keywords: Keratin, collagen, alginate, hydrogel

DRAFT

ORAL PRESENTATION

Enjeksiyonluk bir ürün için liyofilizasyon prosesi geliştirilmesi ve ölçek büyütmesi

Göksu Yağ Çilingiroğlu[1]* (*https://orcid.org/0000-0002-3344-7920*), İrem Karasu[1](https://orcid.org/0009-0005-3209-1669), Sakine Ulusoy[1](*https://orcid.org/0000-0001-9044-9079*), Adem Şahin[1,2] (*https://orcid.org/0000-0002-3996-2931*)

[1] Department of R&D, Centurion Pharma, Ankara, Turkey

[2] Department of Pharmacy Service, Vocational School of Health Services, Bilecik Seyh Edebali University, Bilecik, Turkey

*Sorumlu yazar e-mail: goksucilingiroglu@centurion.com.tr

Özet

Günümüzde çözücüsünde veya dispersiyon ortamında stabil kalmayan, ısıya hassas enjeksiyonluk ürünlerin stabil bir şekilde saklanması için en sık tercih edilen kurutma metodu liyofilizasyondur. Dondurarak kurutma olarak bilinen liyofilizasyon, sulu çözelti/dispersiyon halinde bulunan katıların kurutma yöntemlerinden biri olup, buzun süblimleşmesiyle suyun uzaklaştırılması esasına dayanmaktadır. Liyofilizasyon işlemi dondurma, birincil kurutma ve ikincil kurutma olarak adlandırılan üç basamaktan oluşur. Bu üç basamağı içeren proses, liyofilize ürünlerin üretiminde en çok zaman alan ve enerji harcanan üretim basamağıdır. Diğer yandan doğru yürütülmemiş bir liyofilizasyon prosesi ürünün kalitesini doğrudan etkileyerek bitmiş ürünün hedeflenen özellikleri sağlayamamasına neden olabilmektedir. Tüm bu nedenlerle çalışmamız kapsamında üretmeyi hedeflediğimiz kapreomisin içeren liyofilize enjeksiyonluk toz için liyofilizasyon prosesi geliştirme süreçleri yürütülmüştür. Bu kapsamda öncelikle liyofilizasyon prosesinde etkili olabilecek kritik proses parametreleri belirlenmiş ve bu parametrelerdeki değişikliklerin ürünün hedeflenen kalite özelliklerine nasıl etki ettiği incelenmiştir. Çalışma sonunda ürünün kuruma süresi için en önemli faktörlerin doğru primer kurutma ve sekonder kurutma sıcaklığı olduğu belirlenmiştir. Doğru olmayan sıcaklıklarda yürütülen süreçlerin liyofilizasyon süresini uzattığı ve tamamen kuruyamayan ürünlerin geri erimeye neden olduğu gösterilmiştir. Belirlenen bu reçete üretim boyutuna aktarılmış ve geliştirilen süreç valide edilmiştir.

Anahtar Kelimeler: Liyofilizasyon, Kapreomisin, Ölçek Büyütme

DRAFT

ORAL PRESENTATION

Serotonin Treatment Decreases miR21 Expression And Cell Proliferation Of SA0S-2 Human Osteogenic Sarcoma Cells

Murat SEVİMLİ[1] (ORCID: 0000-0001-8463-6943), Zarifa AHMADOVA[2] (ORCID: 0000-0003-0883-2752), Tuğba SEMERCİ SEVİMLİ[2] (ORCID: 0000-0003-4856-2304)

[1]Suleyman Demirel University, Faculty of Medicine, Department of Histology and Embryology.
[2]Eskisehir Osmangazi University, Department of Stem Cell, Cellular Therapy and Stem Cell Production Application and Research Center (ESTEM).

zarifa.ahmadova@outlook.com

Abstract

Osteosarcoma (OS) is a primary and highly aggressive malignant bone tumor frequently seen in children. Chemotherapy is the current therapeutic strategy used to treat OS. However, due to its severe side effects, there is a need to develop a new agent with increased effectiveness and reduced toxicity in treatment. Serotonin (5-HT) is a central nervous system neurotransmitter. In addition to its known functions in neuroendocrine mechanisms, Serotonin has also been shown to be associated with cancer. miRNAs have essential biological functions in various processes and carcinogenesis. miR21 is one of the best-known and most essential oncomiRs. In this study, we aimed to investigate the effect of Serotonin on the proliferation of human osteogenic sarcoma cells (Saos-2) by modulating miR21 expression. The effect of Serotonin on the proliferation of Saos-2 cells was performed by MTT analysis at 24, 48, and 72 hours. The expression level of miR21 was analyzed by qPCR. The apoptotic effect was determined by Annexin V/PI analysis. In the MTT analysis, it was observed that the proliferation of Saos-2 cells decreased in response to Serotonin. miR21 expression was down-regulated in Serotonin-treated Saos-2 cells. Serotonin has a potential therapeutic strategy for OS.

Keywords: Osteosarcoma, Serotonin, miR21, Cytotoxicity, Apoptosis

DRAFT

ORAL PRESENTATION

Preparation and Characterization of Biomaterial-based Antibacterial Wound Dressing

Ozlem Ayse Tosyalı[1] (*https://orcid.org/0000-0002-8878-5863*),
Oksan Karal-Yilmaz[2*] *(https://orcid.org/0000-0001-6780-9814*)

[*1]İstanbul Beykent University, Faculty of Engineering&Architecture, Department of Biomedical Engineering, Istanbul, Turkey
[*2]İstanbul Beykent University, Faculty of Engineering&Architecture, Department of Chemical Engineering, Istanbul, Turkey

Corresponding author *e-mail: oksanyilmaz@beykent.edu.tr; oksankaralyilmaz@gmail.com

Abstract

Wound repair is a biological process that varies with regard to the period of time to attain healing. In recent years, biomaterials-based wound dressings have received much attention due to their potentials and many studies have been performed based on them. After development of hydrogel appearance which facilitated the production of wound dressings from the passive material to active and functionalized ones. The chitosan (CS) based hydrogel wound dressing encapsulated with drugs serves better in wound healing and exudate management. CS is the common biopolymer used in wound and burn dressings, due to their antimicrobial activity, resistance against environmental conditions, adhesive nature, antifungal, and excellent oxygen permeability. Polyvinyl alcohol (PVA), is one of the most frequently synthetic polymer have been employed as wound dressings and drug delivery systems.It is a biological friendly polymer due to its biocompatibility, lack of toxicity and appropriate mechanical properties. In this study, we prepared PVA/CS hydrogel films containing a model drug (amphotericin B) to develop a suitable material with potential application as high performance wound dressing with controlled drug release. For that, the physicochemical, thermal, morphological and biological characterizations of the biocomposite hydrogel films were evaluated, as well as the release assessment of the drug. The interactions between the constituents of the hydrogels were analyzed by FTIR spectroscopy. This biocomposite films in all compositions showed desirable swelling behavior at pH 7,4. To develop the potential application of chitosan based biocomposite film dressings, the antibacterial potentials, as well as the wound healing activities, were investigated *in vitro*. The studies also showed a sustained release of drug from the hydrogel films. Overall, the results signaled the potential application of amphotericin containing PVA/CS based hydrogel films as wound dressings.

Keywords: chitosan, hydrogel , swelling, drug release, wound healing

DRAFT

ORAL PRESENTATION

Kaspofungin liyofilize toz için elementel impürite metodu geliştirilmesi ve validasyonu

Sena Özlem Gündoğdu[1](*https://orcid.org/0000-0001-8509-7371*), Seda Turhan[1*] (*https://orcid.org/0009-0004-9473-5514*), Tuğçe Çatak[1](*https://orcid.org/0000-0002-2648-2066*), Adem Şahin[1,2] (*ORCID: https://orcid.org/0000-0002-3996-2931)*

[*1] Department of R&D, Centurion Pharma, Ankara, Turkey

[2]Department of Pharmacy Service, Vocational School of Health Services, Bilecik Seyh Edebali University, Bilecik, Turkey

*Sorumlu yazar e-mail: sedaturhan@centurion.com.tr

Özet

Elementel impüriteler kullanıma hazır ilaçların içerisinde bulunabilen ve hasta haslığını riske atan impüriteler olarak dikkat çekmektedir. Bu impüriteler ilacın içerisine sentez sırasında bulaşmış olabileceği gibi ilacın hazırlanması sırasında da bitmiş ürün içerisine geçebilmektedir. Üretim sırasında kullanılan ekipman, filtreler, su, yardımcı maddeler, bitmiş ürün ambalajları, kapakları gibi faktörler tüm süreçte risk yaratabilen unsurlardır. Bitmiş ürün dozaj formları içerisinde elementel impüritenin doğrudan kana karışmasına neden olabilecek paranteral kullanım yolu en fazla dikkat edilmesi gereken dozaj formlarından birini oluşturmaktadır.[1,2] Endüktif Eşleşmiş Plazma Kütle Spektrometresi (ICP–MS) geniş spektrumda elementel impüriteyi düşük konsantrasyonlara kadar belirleyebilmesi ile avantaj sağlayan önemli bir analiz tekniğidir. ICP kaynağı elementlerin atomlarını pozitif yüklü iyonlara çevirir ve bu iyonlar da kütle spektrofotometresi tarafından kütle yük oranına göre ayrılır ve dedektöre iletilir. Avantajlarına rağmen cihazın pahalı olması, kullanımında ve metot geliştirilmesinde uzman personel gerektirmesi tekniğin olumsuz yönleri olarak dikkat çekmektedir.[1] Bu çalışma kapsamında ürettiğimiz Kaspofungin Liyofilize Toz ürününde elementel safsızlıkların takibini sağlamak için ICP-MS analiz metodunun geliştirilmesi ve validasyonunun gerçekleştirilmesi amaçlanmıştır. Bu amaçla ICH Q3D'de belirtilen 24 elementin hedeflenen limitlerde analiz edilmesini sağlayan analiz metodu geliştirilerek valide edilmiştir. Sonuç olarak tüm bu elementler için hedeflenen konsantrasyon aralığında doğrusallık gösterilmiş, LOD ve LOQ değerleri hesaplanmış, tekrarlanabilirlik, geri kazanım gibi tüm validasyon parametreleri başarı ile sağlanmıştır.

Anahtar Kelimeler: Elementel İmpürite, ICP-MS, Kaspofungin

1- F. Thomas, "Approaching Elemental Impurity Analysis," Pharmaceutical Technology 45 (2) 2021.
2- Balaram, V., Recent advances in the determination of elemental impurities in pharmaceuticals – Status, challenges and moving frontiers. TrAC Trends in Analytical Chemistry, 2016. 80: p. 83-95.

DRAFT

ORAL PRESENTATION

Colorimetric determination of organic and inorganic pollutants in different environmental and biological samples

Hakan Serbest[1*] (*ORCID: https://orcid.org/0000-0003-2303-0408*)

[*1]İstanbul Health and Technology University, Faculty of Engineering and Natural Sciences, Department of Chemical Engineering, İstanbul, Türkiye

*Corresponding author e-mail: hakan.serbest@istun.edu.tr

Abstract

Organic and inorganic pollutants are known to occur worldwide as a result of human activities such as agriculture, industry, and spillage as well as natural resources. Since low levels of these pollutants pose a threat to human health and the environment, it is vital to perform qualitative and quantitative determinations with high accuracy and precision. Numerous techniques have been reported in the literature for organic and inorganic pollutants. Among these techniques, colorimetric detection has become very interesting in recent years as an alternative to complex instrumentation with advantages such as rapid response, simple experimental setup, easy operation, low cost, and high sensitivity. Thanks to the colorimetric sensors developed within the scope of the method, many organic / inorganic analytes such as heavy metals and metal salts, pesticides and drug residues can be determined even using a smartphone digital camera with the high accuracy. In this presentation, the limit of detection and limit of quantitation of the developed colorimetric systems for target analytes and the percent recovery results to investigate the feasibility in real samples are evaluated.

Keywords: Colorimetric detection, pollutants, sensor

DRAFT

ORAL PRESENTATION

Harnessing the potential of quince seed extracts to enhance *Chlamydomonas reinhartii* cultivation

Gülay Karagülle[1*] (*ORCID: https://orcid.org/0000-0002-7516-6625)*,
Murat Telli[2] (*ORCID:https://orcid.org/0000-0001-5546-0379)*

[*1]Bolu Abant İzzet Baysal University, Faculty of Science, Biology, Bolu, Turkey
[2] Bolu Abant İzzet Baysal University, Faculty of Science, Biology, Bolu, Turkey

*gulaykalmaoglu@gmail.com

Abstract

Hydrocolloids derived from various seed sources are known for their unique functional properties, characterized by polysaccharide polymers. While the utilization of hydrocolloids has been studied, that remains an untapped potential in unexplored sources. Quince seed mucilage (QSM) has received attention due to its antioxidative, anti-inflammatory, and antimicrobial attributes. Its biocompatibility, non-toxicity, and biodegradability have found applications in diverse industries, including food, cosmetics, and surfactants. Remarkably, its potential in biotechnology, concerning its antimicrobial properties, has yet to be investigated. The aim of the present study is to test potential contribution of quince seed on growth rate of green algae specifically on *Chlamydomonas reinhardtii*, as a growth medium additive. The content of quince seed has been extracted by distilled water by mixing magnetic stirrer. Then in a proportion of 1:2 (50ml QSM/100ml TAP), 1:1(75ml QSM/75ml TAP), and 2:1(100ml QSM/50ml TAP) combination of quince seed mucilage and TAP growth medium respectively were used as growth medium for *C. reinhardtii*. Cultures were maintained in 25 µmol $s^{-1}m^2$ light intensity and aerated with 5% CO_2. The results revealed a remarkable outcome, with algae cultivated in the presence of quince mucilage exhibiting more than a twofold increase (OD 672nm value was 1,51) in growth compared to the control group (OD672nm was 0,73) in the combination of 1:1. Moreover, a synchronized statistically significant enhancement in chlorophyll a (16,54µg/mL) , chlorophyll b (8,915µg/mL), total chlorophyll (25,46µg/mL) and carotenoid content (4,34µg/mL)) coincided with the growth curve in treatment of 1:1 than control groups (7,07µg/mL, 3,65µg/mL, 10,73µg/mL and 1,76µg/mL respectively). These findings suggest a promising role for quince mucilage as an inducer and a sustainability agent, serving to mitigate contamination, in the cultivation of algae. This study underscores the potential of quince seed mucilage as a valuable resource in biotechnological applications, opening new avenues for sustainable and efficient algae production.

Keywords: Quince extract, *Chlamydomonas reinhardtii*, microalgae, chlorophyll, and carotenoids

DRAFT

ORAL PRESENTATION

Establishment of single barcode-harbouring chemotherapy-resistant HCT116 cell lines via limiting dilution approach

Ahmet Acar[*1] (*ORCID: https://orcid.org/ 0000-0002-2478-8029*)

[*1]ODTÜ Biyolojik Bilimler Bölümü, Üniversiteler Mahallesi, 06800, Çankaya, Ankara, Türkiye

*Corresponding author e-mail: acara@metu.edu.tr

Abstract

The selection of drug-resistant clones can inevitably result in treatment failure due to resistance to cancer treatments. With the development of single-cell barcoding, it is now possible to quantify medication resistance by calculating the frequency of chosen clones that contain distinctive cellular barcodes. To characterise barcode frequencies, in this study, it was aimed to generate a single barcode containing chemotherapy-resistant HCT116 cells. Before the development of their chemotherapy-resistant derivatives, the initial HCT116 cell line was integrated with cellular barcodes using a lentiviral barcode library. Amplicon-based NGS method was used to identify the mechanism of resistance, whether pre-existing or de novo drug resistance was present. To create a new cell line from a single barcode, drug-resistant barcoded HCT116 cells have been utilised through a single cell dilution experiment. A single barcode's characterisation and confirmation were carried out using newly developed drug-resistant HCT116 cell lines. The outcomes show the effectiveness of using single-cell dilution and cellular barcoding technology to create single-cell derived colonies in HCT116 cells under the influence of chemotherapeutic selection pressure. The barcoding method displays the frequency of barcode enrichment in drug-resistant HCT116 cell variants. The usefulness of this experimental model system to examine drug resistance at the single-cell level was also demonstrated by unique-barcode containing drug-resistant HCT116 single cell-derived cell lines. With the introduction of recently discovered cellular barcoding technology, it is now possible to monitor treatment resistance at the single cell level, giving the capacity to take advantage of the tumour's susceptibility.

Keywords: treatment resistance, cellular barcoding, cancer

DRAFT

ORAL PRESENTATION

Immunohistochemical Expression of Matrix Metalloproteinase-9 (MMP-9) and Tissue Inhibitors of Matrix Metalloproteinases-1 (*TIMP-1)* in Natural Canine Parvoviral Enteritis Infection

Mehmet Önder KARAYİĞİT[1*] 0000-0002-7958-180X

[*1]University of Cukurova, Faculty of Veterinary Medicine, Department of Pathology, ADANA, Turkey.

*Corresponding author e-mail: karayigit09@hotmail.com

Abstract

Canine parvoviral enteritis is highly contagious infection in intestines caused high morbidity and mortality in untreated dogs younger than 6 months. The aim of this study was to examine the expression of Matrix Metalloproteinase-9 (MMP-9) and Tissue Inhibitors of Matrix Metalloproteinases-1 (*TIMP-1)* in Natural Canine Parvoviral Enteritis Infection of 25 dogs diagnosed with parvoviral enteritis by clinical tests and histopathology. On histopathology, shedding and blunting of the villi epithelium, severe mononuclear inflammation in the lamina propria and locally enlarged crypts with lymphocytolysis in peyer's patches were noted in the ileum. Immunohistochemically, mean strong (+++) expression for MMP-9 and mean moderate (++) expression for TIMP-1 were observed in the crypt epithelium and inflammatory cells in the small intestines of infected animals. As a result, overexpression of MMP-9 and weaker expression of TIMP-1 as its inhibitor in canine parvoviral enteritis may determine the development of the disease.

Keywords: MMP-9, TIMP-1, Parvoviral Enteritis, Immunohistochemistry

DRAFT

ORAL PRESENTATION

Design and evaluation of methionine-bearing 1,2,4-triazole-3-thione derivatives as potential SARS-CoV-2 RdRp inhibitors

Mazın HAMDAN[1*] (*ORCID:* https://orcid.org/0000-0001-7794-5043), Necla KULABAŞ[2] (*ORCID: https://orcid.org/0000-0003-2273-5094*), İlkay KÜÇÜKGÜZEL[3] (*ORCID: https://orcid.org/0000-0002-7188-1859*)

[*1] Institute of Health Sciences, Marmara University, İstanbul, Türkiye
[2] Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Marmara University, İstanbul, Türkiye
[3] Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Fenerbahçe University, İstanbul, Türkiye

* e-mail: mazen.hamdan.10@gmail.com

Abstract

The SARS-CoV-2 has been declared by the WHO as the most devastating pandemic affecting the world's population and economy. Although many vaccines and drugs are available against SARS-CoV-2 infection, the need for promising treatments and drugs remain. RdRp is an attractive and promising therapeutic target for the treatment of SARS-CoV-2 disease due to it is one of the key enzymes that control viral replication (Bekheit et al., 2023; Brindani et al., 2023; Rehman et al., 2023). It is also known that the biologically active 1,2,4-triazol-3-thione ring has antiviral activity. In this study, novel *N*-[3-(methylsulfanyl)-1-(4-aryl/alkyl-5-sulfanyliden-4,5-dihydro-1*H*-1,2,4-triazol-3-yl)propyl]benzamide derivatives were designed and their potential antiviral effect were evaluated against SARS-CoV-2 RdRp enzyme using molecular modeling studies. Autodock Vina, an open licensed software developed by Scripps Research Institute, was used in molecular modeling studies. SARS-CoV-2 RdRp (pdb code: 6M71) (Gao et al., 2020) structure were obtained from the protein database and prepared for use in docking studies using The AutoDock Tools program. The conformations of the designed compounds were scanned with the semi-experimental PM3 method using the Spartan4 quantum chemistry program (SPARTAN 04, Wavefunction, Inc., Irvine, USA). The most stable conformation for each ligand was selected and made ready for the molecular modeling process. Enzyme-ligand interactions were examined with open license imaging software such as Discovery Studio Visuliazer to determine the best binding conformation for each ligand, and the binding mechanisms to the active site of the enzyme were elucidated.

Keywords: 1,2,4-triazoles, SARS-CoV-2 RdRp, molecular modeling, antiviral.

Reference

Bekheit MS, Panda SS, Girgis AS 2023. Potential RNA-dependent RNA polymerase (RdRp) inhibitors as prospective drug candidates for SARS-CoV-2. European Journal of Medicinal Chemistry, 252: 115292.

Brindani N, Munafò F, Menichetti A, Donati E, Nigro M, Ottonello G, Armirotti A, De Vivo M 2023. Design, synthesis, docking, and biochemical characterization of non-nucleoside SARS-CoV-2 RdRp inhibitors. Bioorganic Medicinal Chemistry, 80: 117179.

Gao Y, Yan L, Huang Y, Liu F, Zhao Y, Cao L, Wang T, Sun Q, Ming Z, Zhang L, Ge J, Zheng L, Zhang Y, Wang H, Zhu Y, Zhu C, Hu T, Hua T, Zhang B, Yang X, Li J, Yang H, Liu Z, Xu W, Guddat L, Wang Q, Lou Z, Rao Z 2020. Structure of RNA-dependent RNA polymerase from 2019-nCoV, a major antiviral drug target. Science, 368(6492): 779–782.

Rehman HM, Sajjad M, Ali MA, Gul R, Naveed M, Aslam MS, Shinwari K, Bhinder MA, Ghani MU, Saleem M, Rather MA, Ahmad I, Amin A 2023. Identification of RdRp inhibitors against SARS-CoV-2 through E-pharmacophore-based virtual screening, molecular docking and MD simulations approaches. International Journal of Biological Macromolecules, 237: 124169.

DRAFT

ORAL PRESENTATION

The antibacterial property of silver nanoparticles embedded cryogel

Ilgım Göktürk* (*ORCID: https://orcid.org/0000-0001-7292-7241*)

Hacettepe University, Science Faculty, Department of Chemistry, Ankara, Turkey.

*Corresponding author e-mail: ilgim@hacettepe.edu.tr

Abstract

Membrane fabrication using silver nanoparticles (AgNPs) and polymers has gained popularity in recent years due to its superior optical, conductivity, and antibacterial properties. AgNPs are employed in water treatment because of their exceptional antibacterial capabilities even at low concentrations due to interaction with microorganisms. Macroporous hydrogels, called cryogels, are a type of hydrogel with a macroporous structure and durability that are intrinsically interconnected. Cryogels are formed by cryogelation at sub-zero temperatures by freezing the initial polymer solution containing a crosslinker. In this study, Poly(2-hydroxyethyl methacrylate-vinylimidazole) cryogels were synthesized as solid support. Particle size analysis was used to determine the size of AgNPs. Also, swelling test and scanning electron microscopy (SEM) were performed for the characterization of the cryogel. The antibacterial test was carried out with AgNPs embedded cryogels at three different concentrations.

Keywords: Antibacterial, silver nanoparticles, cryogel.

DRAFT

ORAL PRESENTATION

The Cytotoxic Effect of Royal Jelly Produced in Ardahan and Ordu Regions on Lung and Colon Cancer Cell Lines

Cansu Korkmaz[1*] (*ORCID: https://orcid.org/0000-0003-3027-7687*),
Mehmet Emin Duru[2] (*ORCID: http://orcid.org/0000-0001-7252-4880*)

[*1] * Department of Biology, Faculty of Science, Muğla Sıtkı
Koçman University, 48000 Muğla, Türkiye.
[2] Department of Chemistry, Faculty of Sciences, Muğla Sıtkı Koçman University, 48000 Muğla, Türkiye.

*Corresponding author e-mail: cansukorkmaz08@gmail.com

Abstract

According to GLOBOCAN 2020 statistics, the rates of lung and colon cancer are 11.4% and 18.0%, respectively. Natural products like royal jelly have drawn the attention of many researchers due to their potential anti-cancer properties. One of the important bee genetic resources in the Anatolian region is the Caucasian bee. This bee species, which is widespread in our country and extends to the provinces of Kars, Ardahan, and Artvin in Northeast Anatolia, is known to be an excellent nectar collector with high honey yields. This study aims to investigate the cytotoxic effect of lyophilized royal jelly produced in the Ardahan and Ordu regions on lung (A 549) and colon (HT 29) cancer cell lines, as well as on healthy human (CCd18Co) cell lines, at concentrations ranging from 0 to 200 µg/mL over a 24-hour period.The significant cytotoxic activity of royal jelly produced in the Ardahan region on lung (A 549) and colon (HT-29) cell lines (IC50=53.59 ±1.16, 109.54±2.65 µg/mL) can be attributed to the presence of the Caucasian bee (Apis mellifera caucasica) in the Ardahan region, where it is widely distributed in Turkey. Its non-toxic effect on healthy cells (CCD18Co) (IC_{50}>200 µg/mL) suggests a supportive role in cancer treatment and raises hopes for pre-clinical research against the side effects of chemotherapeutic products. In evaluating the effectiveness of royal jelly on lung and colon cancer, which are widespread worldwide, it is believed that there is a need for further evaluation of additional mechanisms of action to reduce the side effects of chemotherapy. Additionally, the assessment should include histopathological and additional biomarkers for cancer treatment, along with the parameters and genes/proteins tested in vitro.

Keywords: Royal jelly, cytotoxic effect, lung and colon cancer

DRAFT

ORAL PRESENTATION

Comparison of antimicrobial effects of ethanol extracts of some medicinal aromatic plants

Sibel Kerem[1], Özlem Özbek[2]* (*ORCID: https://orcid.org/0000-0002-7683-4197*)

[1]Hitit University, Graduate Education Institute, Department of Molecular Biology and Genetics, Çorum-Turkiye
[2]Hitit University, Faculty of Arts and Sciences, Department of Molecular Biology and Genetics, Çorum-Turkiye

*Corresponding author e-mail: ozbekozlem@gmail.com

Abstract

In this study, plant samples of *Mentha piperita*, *Salvia officinalis*, *Melissa officinalis*, *Achillea filipendulina*, *Echinacea purpurea* from Çorum province, and *Lavandula angustifolia* from Eskişehir, Çorum and Kütahya provinces were collected. The aim of the study is to compare the antimicrobial effects of the extracts obtained from these plants on *Escherichia coli* ATCC-25922, *Pseudomonas auriginosa* ATCC-27853, *Staphylococcus aureus* ATCC-29213 and *Enterococcus faecalis* ATCC-29212. The plant extracts obtained using ethanol in the Soxhlet device and the EUCAST disc diffusion method used. Bacteria were suspended to a density standard of 0.5 McFarland (108 CFU/ml). Three replicates at 3 different doses (15, 20 and 25µL) were used. Ethanol and standard Ciprofloxacin antibiotic disc (5mcg) were used as negative and positive control respectively. Then the petries were incubated at 37 ºC for 24 hours in the incubator. When all plant extracts applied in three volumes, no considerable antimicrobial effect was observed on *E.coli* and *P.auriginosa*, and the zone diameters ranging from 4.67-14.33mm and 21.67-23.67mm were detected on *S.aureus* and *E.faecalis* respectively. The antimicrobial effect of the positive control was higher than that plant extracts. However, it was observed that the *L.angustifolia* from Eskişehir and Kütahya produced the zone diameters close to the positive control on *S.aureus* and *E.faecalis*. The negative control displayed no antimicrobial effect on any bacteria in all volumes. According to the results of this study, *E.coli* and *P.auriginosa* were determined to be the most resistant and *S.aureus* and *E.faecalis* were the most sensitive microorganisms against plant extracts. *L.angustifolia*-Eskişehir, *M.piperita* and *S.officinalis* attract attention with their high antimicrobial effects. Conclusively, using different solvents and extraction methods more effective secondary metabolites could be obtained from medicinal aromatic plants and used more widely as natural products in alternative medication against pathogenic diseases, in food packaging, and in areas such as the pharmaceutical and cosmetic industries.

Keywords: *Mentha piperita*, *Salvia officinalis*, *Lavandula angustifolia*, Medicinal aromatic plant, Antimicrobial, Disc Diffusion,

DRAFT

ORAL PRESENTATION

Cloning and sequencing of *Bacillus mojavensis* TH309 phytase gene

Mevlüt ARSLAN[1*] (*ORCID: https://orcid.org/0000-0003-4883-4736*),
Ali Osman ADIGÜZEL[2] (*ORCID: https://orcid.org/0000-0002-5602-5886*)

[*1]Van Yüzüncü Yıl University, Faculty of Veterinary Medicine, Department of Genetics, Van, Turkey
[2]Ondokuz Mayıs University, Faculty of Science, Department of Molecular Biology and Genetics, Samsun, Turkey

*Corresponding author e-mail: mevlutarslan@yyu.edu.tr

Abstract

Phytate is the main storage form of phosphorous in inositol ring in the plants. Phytase hydrolyses phytate and releases phosphorus from the structure. Monogastric animals including poultries, pigs, and fishes lack phytate-degrading enzymes in their gastrointestinal systems. Therefore, the animals cannot benefit from the phosphorus found in phytate in feed. Furthermore, phytate in the gastrointestinal systems of these animals cause chelation of metallic cations such as calcium, ferric, zincs, minerals, amino acids and proteins; therefore, it has anti-nutritional effects in feeding the monogastric animals. To eliminate its anti-nutritional effects and utilize phosphorus from phytate, phytase is added to feeds, and it has been used in the feeding of monogastric animals. Phytases from fungal and bacterial sources can be used for this purpose. Phytases obtained from the Bacillus species have several advantages such as high thermal stability, requirement of low calcium level for activation, substrate specity, pH profile, and resistance to proteolysis. Even though phytase from several *Bacillus* species have been cloned and purified, phytase from *Bacillus mojavensis* has not studied, yet. Here, phytase gene from *B. mojavensis* TH309 strain was cloned into pet20b(+) expression vector for extracellular synthesis of phytase in *E. coli* BL21. Using special primers, phytase gene from *B. mojavensis* TH309 was amplified by PCR. The vector and PCR product were digested by *BamH*I and *EcoR*I restriction enzymes and ligated by T4 DNA ligase. After obtaining recombinant plasmid called pet20b(+)-phytase, transformation and colony PCR were carried out to select clone. After isolation of the plasmid, DNA sequencing was carried out bidirectionally. BLAST analysis indicated that *B. mojavensis* TH309 phytase gene contained several mutations compared to *B. mojavensis* reference sequence. Three of the mutations are missense, R290G, T301K and N366D, which indicates the unique structure of the enzyme. Expression, purification, and characterization studies of the enzyme are in progress.

Keywords: Cloning, Sequencing, Phytase, *Bacillus mojavensis*

DRAFT

ORAL PRESENTATION

Recombinant Production, Purification and Activity of Microbial Hyaluronidase Enzyme

Mehmet AYTAR[1*] (*ORCID: https://orcid.org/0000-0002-8083-7358*), Gamze BAŞBÜLBÜL[2] (*ORCID:* https://orcid.org/0000-0001-8151-6321) Bülent BOZDOĞAN[3] (*ORCID: https://orcid.org/0000-0003-2469-9728*)

[*1] Aydın Adnan Menderes University, Institute of Natural and Applied Sciences, Department of Biology, Aydın, Turkey
[2] Aydın Adnan Menderes University, Faculty of Science, Department of Biology, Aydın, Turkey
[3] Aydın Adnan Menderes University, REDPROM Research Center, Aydın, Turkey

* Corresponding author e-mail: maytar90@gmail.com

Abstract

Hyaluronic acid injection has increased rapidly in cosmetic applications. Although it seems safe, it causes complications such as nodules or papules, Tyndall effect (bluish discoloration of the skin), hypersensitivity, tissue necrosis, infections, microbial biofilms and granulomas. The use of hyaluronidase to treat is increasing day by day. Hyaluronidase enzyme is an endoglucosidase that breaks its substrate, hyaluronic acid, from its glucosidic bonds and separates it into monosaccharides. In our study, the hyaluronidase enzyme gene from *S. aureus* was amplifiex, cloned in pET-30a(+) expression vector and transformed into *E. coli* BL-21 (DE3) competent cell. Recombinant hyaluronidase enzyme production was induced with 5g/L lactose for 24 hours. Purification was performed by his-tag nickel affinity using the IMAC colulactos HPLC. It was determined by the agar-well diffusion method that there was a high level of enzyme activity in the fractions at the elution stage. Today, in addition to the complications listed above, hyaluronidase is used in the treatment of lumps, bumps and over corrections as a filling dissolving agent in incorrectly performed filling procedures. It is considered an effective tool in preventing serious complications. The recombinant microbial hyaluronidase (HysA-9) enzyme may have potential for use in cosmetics and medicine.

Keywords: Hyaluronidase, Hyaluronic acid, Recombinant, HPLC

DRAFT

ORAL PRESENTATION

Determination of adhesion properties of lactic acid bacteria isolated from traditional food products.

Mehmet Burak YİĞİT[1] (*ORCID: https://orcid.org/0000-0002-6777-6773*),
Aysun CEBECİ AYDIN[2*] (*ORCID: https://orcid.org/0000-0002-6158-8798*)

[1]Abdullah Gül University, Faculty of Life and Natural Sciences, Molecular Biology and Genetics Department, Kayseri, Türkiye.
[*2]Abdullah Gül University, Faculty of Engineering, Nanotechnology Engineering Department, Kayseri, Türkiye.

*Corresponding author e-mail: aysun.cebeciaydin@agu.edu.tr

Abstract

Probiotics are microorganisms that live in our bodies and positively affect health when consumed regularly. One of the ways to have a healthy body is to have a healthy microbiota. Because of that, the importance given to the consumption of probiotic foods among the public is increasing. Since probiotics are especially abundant in fermented and traditional foods, consuming these foods is vital to have a healthy microflora. Among the specific properties of probiotics, adhesion is the one of the most significant features. Since the adhesion is the requirement for growth and colonization of probiotic bacteria and support the colonization resistance, adhesion abilities of probiotic bacteria should be evaluated. In this study, adhesion of bacteria isolated from tarhana, einkorn sourdough, Turkish and Bulgarian-type boza and pickled beetroot foods were investigated. According to the results, 7 strains showed over 35%, 4 strains showed over 50% adhesion and especially, two of the strains showed over 85% adhesion to Caco-2 cells. Afterwards, 16S rRNA sequencing was conducted for identification of bacteria at species level and identifying them under the *Lactobacillus* and *Enterococcus* genera.

Keywords: Probiotics, Lactic acid bacteria, traditional foods, adhesion

DRAFT

ORAL PRESENTATION

Determination of pH Value Using Conventional pH Strips by Image Processing

Farah Eid[1*], (ORCID:https://orcid.org/0009-0006-6247-9637), Mert Akin Insel[1] (ORCID: https://orcid.org/0000- 0002-4347-1190), Hasan Sadikoglu[1] (ORCID: https://orcid.org/0000-0002-0234-8428)

[*1]Yildiz Technical University, Faculty of Chemical and Metallurgical Engineering, Chemical Engineering, Istanbul, Turkiye.

*Corresponding author e-mail: farahsyr91@gmail.com

Abstract

Determination of pH value is considered to be a critical measurement utilized in a large number of applications. It is mainly used to evaluate the acidity of aqueous solutions in order to classify them as acidic or alkaline. There are several pH measurement methods. The most conventional and widely utilized method is the use of pH strips. While they have a lower level of precision than pH meters, they are known to be an economical and affordable measurement method to be used by non-experts users. The major drawback of this method is the large possibility of having scientific errors due to the limited ability of human eyes in precisely reading the strip's value, especially when highly accurate results are required. However, the availability of advanced technology and mobile phones nowadays provides a great advantage to overcome this challenge. For this purpose, a literature review was conducted about utilization of image processing in determining the accurate pH value from reading pH strips; by taking a picture of it using a mobile phone camera and analysing the color differences. Many researchers reveal that it is possible to determine the pH value by comparing the color of the strip with the reference solutions associated with the pH levels. Each researcher used this method for determining pH value for a specific substance. Our goal is to investigate the most recent applications that determine the pH value of solutions from the pH strips, providing essential information for researchers that aim to develop a mobile application regarding the determination of the pH value.

Keywords: Conventional pH strips, Image Processing, Mobile Phone, Artificial Intelligence

DRAFT

ORAL PRESENTATION

Investigation of the effect of metformin on epithelial-mesenchymal transition in MCF-7 and MDA-MB-231 breast cancer cells with increased expression of miR-506

Özge Rencüzoğulları (*ORCID: 0000-0002-2157-1289*)

*Istanbul Kultur University, Faculty of Science and Letter, Department of Molecular Biology and Genetics, Istanbul, Turkey.

e-mail: ozgeberrak@gmail.com

Abstract

Breast cancer is the second most common cancer when its incidence is evaluated. Increases in breast cancer aggressiveness due to AMPK deregulation have triggered the development of AMPK-targeted therapy models. Metformin is a drug used in the treatment of type 2 diabetes mellitus, which reduces blood glucose levels by inhibiting gluconeogenesis, decreasing hepatic glucose production and improving insulin sensitivity. A 2014 meta-analysis showed that individuals using metformin had a lower incidence of breast cancer. Metformin suppresses anti-proliferative mammalian target of rapamycin and inhibits cell growth due to activation of AMP-activated protein kinase (AMPK). miR-506 has been shown to be anti-oncogenic and suppress tumor growth in vitro and in vivo studies. At the same time, miR-506 has been examined to have a role in regulating epithelial-mesenchymal transition (EMT) in breast cancer cell lines. It has been shown that high expression of miR-506 inhibits the proliferation and metastasis of breast cancer cells. The aim of this study is to examine the effect of metformin on the EMT signaling mechanism in MDA-MB-231 and MCF-7 breast cancer cells with increased miR-506 expression. According to the results obtained, the inhibitory effect of metformin on cell proliferation, colony formation, and migration in breast cancer cells was significantly increased when miR-506 expression was increased. As a result, miR-506 expression acts as a tumor suppressor in breast cancer cells and its enhancing effect of metformin has provided preliminary data for future in vivo studies.

Keywords: Breast cancer, AMPK, EMT, miR-506

DRAFT

ORAL PRESENTATION

Antiulcerative effect of Apilarnil on ethanol-induced gastric injury model in rats

İhsan Karaboğa[1]* *(ORCID: 0000-0001-6708-1229)*, Mehmet Akif OVALI[2] *(ORCID: 0000-0002-3672-871X)*

*[1]Kirklareli University, Faculty of Medicine, Department of Histology and Embryology, Kirklareli, Türkiye
[2]Çanakkale Onsekiz Mart University, Faculty of Medicine, Department of Physiology, Canakkale, Türkiye

*Sorumlu yazar e-mail: ihsankaraboga@gmail.com

Abstract

The therapeutic properties of apilarnil, an apitherapy product, in a gastric ulcer model were examined in our study. Male Wistar albino rats (32) were used in the study. Rats were divided into 4 groups; Group I (Control, n=8), Group II (Ethanol, n=8), Group III (Apilarnil +Ethanol, n=8), Group IV (Lansoprazole + Ethanol, n=8). Ulcer was induced by oral administration of 1 ml absolute ethanol to rats in groups II, III and IV following the 24-hour fasting process. Apilarnil (0.8 g/kg) was administered to Group III rats 1 hour before ulcer induction, and Lansoprazole (30 mg/kg) was administered to Group IV rats. Ulcer index was calculated in the gastric mucosa. Histopathological changes in the gastric mucosa were revealed by Hematoxylin-Eosin (H&E) staining. Immunohistochemical inducible nitric oxide (iNOS) expression level was also determined in gastric tissue. Cellular death in the mucosa was shown by the TUNEL method. Superoxide Dismutase (SOD), Malondialdehyde (MDA) and Myeloperoxidase (MPO) levels that are the oxidative status markers in the stomach tissue were studied spectrophotometrically. Moreover, Interleukin 1-beta (IL-1β) and Interleukin-10 (IL-10) levels in stomach tissues were determined by ELISA method. While apilarnil administration significantly improved the gastric ulcer index, gastric iNOS expression and mucosal apoptosis are significantly reduced ($p<0.05$). Apilarnil application caused a significant increase in SOD level compared to the ulcer model group ($p<0.05$), on the oether hand, it caused a significant decrease in MDA and MPO levels ($p<0.05$). In addition, Apilarnil administration provided a significant decrease in the level of IL-1β compared to Group II, and a significant increase in the level of the anti-inflammatory cytokine IL-10. The outcomes obtained in our study reveal the antioxidative and anti-inflammatory effects of Apilarnil and its protective properties on the gastric mucosa in the ethanol-induced gastric ulcer model.

Key words: Apilarnil, gastric ulcer, infalmmation, oxidative stress

DRAFT

ORAL PRESENTATION

Immunohistochemical Investigation of the Relationship Between the Disease and Apoptosis in Dogs Naturally Infected with Parvoviral Enteritis.

Mehmet Önder KARAYİĞİT[1*] (0000-0002-7958-180X)

[*1]University of Cukurova, Faculty of Veterinary Medicine, Department of Pathology, ADANA, Turkey.

*Corresponding author e-mail: karayigit09@hotmail.com

Abstract

Parvoviral enteritisis a very common disease in dogs in Turkey. The treatment of the disease is difficult and often results in puppy deaths. There are many studies on the pathogenesis of parvoviral enteritis, but the role of apoptosis little known. The aim of this study is to investigate the relationship between disease and apoptosis in dogs naturally infected with parvovirus. Intestinal tissues of 18 dogs diagnosed with parvoviral enteritis were used as material. TUNEL method was used to determine apoptosis. Histopathological examination revealed shedding of intestinal villus epithelium, villus mass, dilatation of crypts, and desguamation with necrosis of crypt epithelium. TUNEL staining found few tunnel positive epithelial and connective tissue cells in control tissues. A statistically significant number of TUNEL positive cells were found in infective tissues compared to control tissues. These results suggested to us that there was a positive correlation between Parvoviral enteritis and apoptosis. Apoptosis studies with different markers will contribute to the mechanism of the disease.

Keywords: TUNEL, Parvoviral Enteritis, Immunohistochemistry

DRAFT

ORAL PRESENTATION

Differential Effects of Natural p-Menthan-3,8-diol Synthesized by Green Chemistry Method and N, N-Diethyl-meta-toluamide on Developing Zebrafish Embryos

Zülal Mızrak[1*] *(ORCID: 0009-0004-7647-7267*), Semanur Işıkoğlu [1] *(ORCID: 0009-0003-9779-2867)*, Merih Beler[1] *(ORCID: 0000-0002-3828-4630)*, Gizem Eğilmezer [1] *(ORCID: 0000-0002-1231-5232)*, İsmail Ünal[1] (*ORCID: 0000-0002-8664-3298*), Derya Cansız[2] *(ORCID:0000-0002-6274-801X*), Gökhan Özokan[3] (*ORCID:0000-0003-1140-1996*), Ebru Emekli-Alturfan[4] (*ORCID:0000-0003-2419-8587*)

[1] Marmara University, Institute of Health Sciences,Department of Biochemistry, , Turkey
[2] Istanbul Medipol University, Faculty of Medicine, Department of Biochemistry, Turkey
[3] Yıldız Technical University, BioArge Laboratories, Turkey
Marmara University, Faculty of Dentistry, Department of Basic Medical Sciences, Turkey

* zulalmizrak26@gmail.com

Abstract

DEET, also known by its chemical name N,N-diethyl-meta-toluamide, is the main component in many repellents. It is frequently used to keep off biting insects like ticks and mosquitoes. As natural and regarded as safer, p-menthan-3,8-diol (PMD) is a well-known repellent active product against mosquitoes exhibiting an exceptional repelling action. *Corymbia citriodora*, formerly known as *Eucalyptus citriodora*, is a plant with leaves that yield an essential oil that contains trace amounts of PMD. Aim of this study was to evaluate the effects of the synthetic n,n-diethyl-meta-toluamide (DEET) and PMD (Bioarge) produced by green chemistry method on the development and oxidant antioxidant parameters of zebrafish embryos. Zebrafish embryos were exposed to DEE) and PMD (Bioarge) for up to 72 hours post fertilization (hpf). Developmental parameters, mortality and hatching rates were monitored and documented daily during embryonic development. Oxidant-antioxidant status of the embryos were evaluated through lipid peroxidation, glutathione S-transferase and superoxide dismutase activities determined at 72 hpf. Significant alterations were observed in the oxidant-antioxidant parameters of the DEET and PMD exposed embryos. Delayed hatching, developmental defects and increased mortality rates were observed in the zebrafish embryos exposed to DEET when compared with PMD. When all these results are evaluated, naturally sourced PMD produced by the green chemistry method was more biocompatible and showed less toxic effects on zebrafish embryos than synthetic DEET in terms of mortality and hatching rates. These results have emphasized the importance of green chemistry method in producing insect repellents.

Keywords: Insect repellent, N,N-diethyl-meta-toluamide, p-menthan-3,8-diol, green chemistry, zebrafish embryos

DRAFT

ORAL PRESENTATION

Investigation of apoptotic effects of organosulfur compounds as multitarget directed ligands

Sümeyra Çetinkaya[1*] *(https://orcid.org/0000-0002-5811-8832)*

[*1] Sümeyra Çetinkaya[*1], Biotechnology Research Center, Field Crops Central Research Institute, Ankara, Türkiye

*e-mail: cetinkayasumeyra0@gmail.com

Abstract

Apoptosis, also known as programmed cell death, is a critical process for all organisms and serves as an essential component of the cell cycle. It plays a vital role in the elimination of damaged or dysfunctional cells. Certain plants, such as garlic, onion, and broccoli, are rich in organosulfur compounds. Compounds like allicin, diallyl trisulfide, and S-allyl cysteine have the potential to inhibit the growth of cancer cells and promote apoptosis. In this study, molecular docking analysis was used to understand how the mentioned organosulfur compounds (S-allyl cysteine, dially trisulfide, allicin, allyl mercaptan, captopril, glucosinolate) interact with genes crucial to the apoptosis process (CASP3, CASP7, CASP8, CASP9, BAX, BCL-2, p53 ve TNFα). Additionally, the SWISS ADME tool was utilized to assess the physicochemical properties, bioavailability, and potential toxicities of these compounds. The results indicate a positive correlation between organosulfur compounds found in garlic and critical proteins in the apoptosis process. SWISS ADME and molecular docking assessments have shown that glucosinolate and captopril ligands exhibit strong binding to proteins associated with the apoptotic pathway. The interaction between glucosinolate and CASP9 protein (PDB ID: 2ar9) has the highest binding affinity with a value of -7.3 kcal/mol. The low free energy value and identified interactions indicate that glucosinolate and captopril ligands may serve as effective potential activators for protein targets associated with the apoptotic pathway. SMILES data reveal the potential of allicin, allyl mercaptan, diallyl trisulfide, and captopril as drug candidates. These findings pave the way for more extensive studies to deeply explore the potential of organosulfur compounds in modulating apoptotic pathways and their possible treatment in cancer therapy.

Keywords: ADME analysis, apoptosis, molecular docking, organosulfur compounds

DRAFT

ORAL PRESENTATION

Yenilenebilir kaynaklardan doymamış poliüretanların sentezi ve karakterizasyonu

Ayşe Ebra TEMUR[1] (*ORCID: https://orcid.org/0000-0001-9624-1586.*), Gökhan ÇAYLI[1*] (*ORCID: https://orcid.org/0000-0002-3395-5642*)

[*1]Istanbul University-Cerrahpaşa, Faculty of Engineering, Department of Engineering Sciences, Istanbul, Türkiye.

*Sorumlu yazar e-mail: gokhan.cayli@iuc.edu.tr

Özet

Poliüretanlar sağlıktan inşaat sektörüne kadar oldukça farklı alanlarda yaygın kullanım alanına sahip polimerik malzemelerdir. Poliüretanlar diizosiyanatların ve diollerin reaksiyonuyla elde edilmektedirler. Bu reaksiyon sonunda elde edilen polimerler lineer olup çözünürdürler. Ancak ikiden fazla izosiyanat ve hidroksil grubuna sahip reaktiflerle sentezlendiğinde termoset malzemeler elde edilmektedir. Günümüzdeki en büyük problemlerden bir tanesi polimerlerin çevre ile olan etkileşimidir. Biyo esaslı poliüretanların yapısına bakıldığına genellikle diol (poliol) komponentinin biyo esaslı olduğu görülmektedir. Bu çalışmamızda biyo esaslı bir diol olan mono olein daha çevreci bir metot olan borik asit esteri asidolizi reaksiyonu ve atık zeytinyağından elde edilen oleik asit ile sentezlenmiştir. Daha sonra elde edilen mono olein MDI ve H-MDI ile polimerize edilerek lineer doymamış poliüretanlar sentezlenmiştir. Bu doymamış poliüretanlar sahip olduğu çift bağ sebebiyle vulkanizasyondan halojenlendirme reaksiyonuna kadar birçok reaksiyonu verebilmektedir. Bu polimerlerin daha sonradan modifikasyonunu yapmak mümkündür. Elde edilen tüm ara maddelerin ve polimerlerin spektral karakterizasyonları bu çalışmada yapılmıştır.

Anahtar Kelimeler: Yenilenebilir kaynaklar, Bitkisel trigliseritler, monoolein, doymamış poliüretanlar, borik asit esteri

DRAFT

ORAL PRESENTATION

Kuru ceviz yapraklarından biyochar üretimi ve piroliz kinetiği parametrelerinin belirlenmesi

Anıl Tevfik Koçer[1*] (*ORCID: https://orcid.org/0000-0003-1519-1711*)

[*1]Yıldız Teknik Üniversitesi, Kimya-Metalurji Fakültesi, Biyomühendislik Bölümü, İstanbul, Türkiye

*Sorumlu yazar e-mail: anilkocer66@gmail.com

Özet

Tarım atıklarının katma değeri yüksek, değerli ürünlere dönüştürülmesi hem ekonomik hem de çevresel olarak oldukça önemli bir konudur. Özellikle ceviz gibi bol bulunan ve kullanımı geniş olan ağaçların atıkları bu bağlamda büyük potansiyele sahiptir. Bu atıkların yapısı itibariyle en uygun değerlendirme yöntemlerinden birisi biyochar üretmektir. Biyochar, piroliz adı verilen bir termokimyasal reaksiyon sonucu açığa çıkan ve biyoyakıt, adsorbent ve toprak iyileştirici gibi önemli alanlarda kullanılabilen değerli bir katı üründür. Bu çalışmanın amacı ceviz ağaçlarından kuruyup düşerek çevresel kirliliğe sebep olan yaprakların biyochar üretiminde kullanılma potansiyellerinin belirlenmesi ve piroliz reaksiyonu mekanizmasının termogravimetrik yöntemle incelenmesidir. Bu kapsamda kuru ceviz yapraklarından farklı sıcaklıklarda (400, 500 ve 600 °C) biyochar üretilmiş ve üretilen biyocharların karakterizasyonları yapılmıştır. Ayrıca piroliz kinetiği parametreleri Coats-Redfern yöntemi kullanılarak hesaplanmış ve en uygun kinetik modeller ve piroliz kinetiği parametreleri belirlenmiştir. Elde edilen sonuçlara göre en yüksek verimde biyochar 400 °C sıcaklıkta üretilmiş olup, sıcaklık arttıkça biyochar verimi düştüğü görülmüştür. Kinetik hesaplamalarına göre ceviz yaprağının pirolizi için en uygun model ikinci-derece (F2) modeldir ve bu modelde hesaplanan piroliz aktivasyon enerjisi ve regresyon katsayısı değerleri sırasıyla 66.90 kJ/mol ve 0.966'dır. Elde edilen biyochar ve kinetik sonuçlarına göre kuru ceviz yaprağı atıklarının biyochar üretimi için oldukça uygun bir kaynak olduğu ve üretilen biyocharların katı biyoyakıt olarak kömüre alternatif ya da kömürle karıştırılarak yakma sistemlerinde kolaylıkla kullanılabileceği anlaşılmıştır.

Anahtar Kelimeler: Ceviz yaprağı, Biyochar, Piroliz, Kinetik, Termogravimetrik analiz, Coats-Redfern

DRAFT

ORAL PRESENTATION

Dopamin, ürik asit ve askorbik asit eşzamanlı deteksiyonu için güncel elektrokimyasal sensör çalışmaları: Kısa inceleme

Mehmet Selçuk Erdoğan[1*] (0000-0002-0423-6972)

[*1]Kütahya Dumlupınar Üniversitesi, Altıntaş Meslek Yüksekokulu, Kimya ve Kimyasal İşleme Teknolojileri, Kütahya, Türkiye

*mselcuk.erdogan@dpu.edu.tr

Özet

Bilindiği gibi dopamin (DA), ürik asit (UA) ve askorbik asit (AA) insan serumundaki önemli bileşiklerdir ve bu bileşiklerin elektrokimyasal olarak deteksiyonu alanında dünya çapında birçok sensör çalışmaları yapılmaktadır. Geliştirilen elektrokimyasal sensörlerde performans, maliyet, dayanıklılık, doğruluk, kesinlik, kullanışlılık gibi değerler açısından istenilen değerler elde edilmeye çalışılmaktadır. Bu çalışmada öncelikle genel sensör türlerinden ve deteksiyonu yapılan biyomoleküllerden ve yapılarından teorik olarak bahsedilmiştir. Daha sonra da dopamin (DA), ürik asit (UA) ve askorbik asit (AA) tayini yapabilen bazı güncel elektrokimyasal sensör çalışmaları incelenmiş ve kısaca bu çalışmalar açıklanmıştır. Üretilen elektrokimyasal sensörlerin kimyasal bileşimleri, algılama sınırları (LOD) ve performansları da tablo halinde sunulmuştur. Ayrıca ele alınan sensörlerin kimyasal olarak üretim aşamaları da gösterilmiştir. İncelenen sensörler birbirleriyle malzeme, algılama sınırı, maliyet vb. değerler açısından kıyaslanarak bir değerlendirme yapılmıştır.

Anahtar Kelimeler: Elektrokimyasal sensör, dopamin, ürik asit, askorbik asit

DRAFT

ORAL PRESENTATION

Comparative Effects of Natural Cosmetic Preservative Synthesized by Green Chemistry Method and Synthetic Cosmetic Preservative on Developing Zebrafish Embryos

Semanur Işıkoğlu [1*] *(ORCID: 0009-0003-9779-2867)*, Zülal Mızrak[1] *(ORCID: 0009-0004-7647-7267*), Merih Beler[1] *(ORCID: 0000-0002-3828-4630)*, Gizem Eğilmezer [1] *(ORCID: 0000-0002-1231-5232)*, İsmail Ünal[1] (*ORCID: 0000-0002-8664-3298*) , Derya Cansız[2] *(ORCID:0000-0002-6274-801X*), Gökhan Özokan[3] (*ORCID:0000-0003-1140-1996*), Ebru Emekli-Alturfan[4] (*ORCID:0000-0003-2419-8587*)

[1] Marmara University, Institute of Health Sciences,Department of Biochemistry, , Turkey
[2] Istanbul Medipol University, Faculty of Medicine, Department of Biochemistry, Turkey
[3] Yıldız Technical University, BioArge Laboratories, Turkey
Marmara University, Faculty of Dentistry, Department of Basic Medical Sciences, Turkey

* yusem.2912@gmail.com

Abstract

In order to preserve the safety and quality of cosmetic products during their shelf life, preservatives are included in their compositions to give protection against microbiological growth. Preservative selection and quantification are crucial in the design and development of cosmetic formulations. According to the regulations, benzoic acid and benzyl alcohol are among the preservatives that are allowed to be utilized in limited quantities in cosmetic formulations. Aim of this study was to evaluate the effects of the liquid natural preservative (Preallnat 65, Bioarge) on the development and oxidant antioxidant parameters of zebrafish embryos and compare with the synthetic preservative. The natural preservative contained natural benzoic acid, benzyl alcohol, phenylpropanol from *Cinnamomum Cassia*, and natural pentylene glycol from corn and sugar cane whereas, the synthetic preservative consisted of synthetic benzoic acid and synthetic benzyl alcohol. Zebrafish embryos were exposed to the natural and synthetic preservative for up to 72 hours post fertilization (hpf). Developmental parameters, mortality and hatching rates were monitored and documented daily during embryonic development. Oxidant-antioxidant status of the embryos were evaluated through lipid peroxidation, glutathione S-transferase and superoxide dismutase activities determined at 72 hpf. Significant alterations were observed in the oxidant-antioxidant parameters of the natural and synthetic preservative exposed embryos. Zebrafish embryos exposed to the synthetic preservative hatched later, delayed in development, and had a higher mortality rate than those exposed to natural preservative. According to our findings, natural preservative was found to be more biocompatible in zebrafish embryos than synthetic preservative in terms of mortality rates, embryo development, hatching rates and malformations. Our results have shown the importance of green chemistry method in producing preservatives used in cosmetic products.

Key words: Preservatives, cosmetic products, green chemistry, zebrafish embryos

DRAFT

ORAL PRESENTATION

Efficacy of the spectrotaxonomic method in the classification of female individuals of the species belonging to the genus Laccophilus (Coleoptera, Dytiscidae)

Ergün ERGENEKON[1] (*ORCID: https://orcid.org/0000-0002-8303-4585*), Nihal ŞİMŞEK ÖZEK[2*] (*ORCID:* https://orcid.org/0000-0002-7326-5670), Ömer Köksal ERMAN[1] (*ORCID:* https://orcid.org/0000-0001-8901-2750)

[1]Atatürk University, Faculty of Science, Department of Biology, Erzurum, Türkiye
[2]Atatürk University, Department of Biology and East Anatolia High Technology Application and Research Center (DAYTAM), Erzurum, Türkiye

*Corresponding author e-mail: nihal.ozek@atauni.edu.tr

Abstract

Insects are found almost everywhere, including deserts and mountainous regions. Identification and classification of these creatures, which have a wide variety and distribution, are generally based on morphological parameters, and this is not easy and requires a long time. Furthermore, the experience of the researcher is significant in their diagnosis. Barcoding based on gas and liquid chromatography-mass spectrometry analytical methods and DNA-based techniques have been developed to overcome these classification obstacles. Although these techniques are susceptible, they often require many individuals, sophisticated, specialized equipment and expensive reagents. Therefore, as an alternative to morphology-based methods, there is a need for an efficient, user-independent, fast, inexpensive and non-destructive method without requiring sample preparation. One of the techniques with these advantages is Fourier transform infrared (FTIR) spectroscopy. Based on reflectance measurement, this technique's attenuated total reflectance (ATR) module enables direct spectrum acquisition from samples and is, therefore, widely used in biological spectroscopy studies. Recent studies have shown that FTIR spectroscopy effectively determines the biochemical content (lipids, proteins, cellular processes) of plants and insects. Information about differences in biochemical components can be obtained from FTIR spectra, which can be used to discriminate between species based on their spectral characteristics. Because the cuticle of each species has a unique biochemical composition, FTIR spectrum is a method that reveals this composition as a fingerprint. This study developed a method for identifying female individuals of 3 different species of the genus Laccophilus based on FTIR spectra using multivariate analysis techniques. Chemometric findings based on the spectral data obtained show that all three species are distinguished with 100% sensitivity and specificity.

Keywords: Insect, Dytiscidae, Laccophilus, Classification, FTIR.

DRAFT

ORAL PRESENTATION

Çet koyunlarında DGAT1/AluI gen polimorfizminin belirlenmesi

Mervan Bayraktar

Çukurova Üniversitesi, Ziraat Fakültesi, Zootekni Bölümü

mervan.bayraktar@gmail.com

Özet

Trigliserid sentezinin ana düzenleyicisi olan Diacylglycerol acyltransferase 1 (DGAT1), anahtar bir enzim olarak kabul edilmektedir. Koyunlarda, DGAT1 geni süt, karkas ve büyüme özelliklerini etkileyen güçlü bir fonksiyonel aday gendir. Bu çalışmada, genetik materyal olarak 45 Çet koyunu kullanılmıştır. Genotipleme, PCR-RFLP yöntemi ile gerçekleştirilmiştir. 309 bp'lik PCR ürünü, AluI kesim enzimi ile muamele edilerek genotipik farklılıklar ortaya konulmuştur. Elde edilen üç farklı genotip; TT, TC ve CC'dir. Saptanan genotip ve allel frekansları sırasıyla şu şekildedir: 0.60 (TT), 0.33 (TC), 0.07 (CC); 0.77 (T) ve 0.23 (C). Çet koyunlarında tespit edilen DGAT1/AluI gen polimorfizmi, bu hayvanların genetik çeşitliliğinin ve ekonomik özellikler üzerindeki potansiyel etkisinin bir göstergesidir. Baskın olan T allelinin olumlu özelliklerle ilişkilendirilmesi, hayvancılıkta daha verimli üretim stratejileri oluşturulması için bu genotipin seçilmesi gerekliliğini ortaya koymaktadır. Bu bulgular, Çet koyunlarında genetik ıslah çalışmalarının yönünü belirlemek için kritik öneme sahiptir.

Anahtar kelimeler: Çek koyunu, DGAT1, Polimorfizm, PCR-RFLP, Allel frekansı

DRAFT

ORAL PRESENTATION

Atık su arıtımında biyoteknoloji: *Tetrahymena thermophila* giderim performansı ve gelecek beklentileri"

Erdal Başkut[1], Handan Açelya Kapkaç[1*] *(https://orcid.org/0000-0002-2812-1686),* Çağdaş Saz[2] *(https://orcid.org/0000-0001-6230-5908),* Cengiz Türe[1] *(https://orcid.org/0000-0001-6137-1866)*

[*1]Eskişehir Teknik Üniversitesi, Fen Fakültesi, Biyoloji Bölümü, Eskişehir, Türkiye
[2]Benli Recycling Technologies, Eskişehir, Türkiye

*haakdamar@eskisehir.edu.tr

Özet

Gelişmekte olan ülkelerdeki kentleşme ve nüfus artışı, tatlı su talebini ve atık su üretimini artırmaktadır. Kontrolsüz atık su deşarjı, çevresel kirliliğe neden olmakta, tatlı su kaynaklarını, ekosistemleri ve insan sağlığını tehdit etmektedir. Bu nedenle, atık suyun arıtılması küresel bir zorunluluktur. Atık su arıtımının maliyetleri ve aşırı kimyasal kullanımı çeşitli sorunlar yaratmaktadır. Bu bağlamda, bilim insanları, yeşil ve sürdürülebilir teknolojilere odaklanmıştır. Mevcut literatürde, ökaryotik bir organizma *olan Tetrahymena thermophila*'nın atık su arıtımı üzerine yapılmış bir çalışma bulunmamaktadır. Bu çalışma, literatüre yeni bir bakış açısı getirmek amacıyla yürütülmüş ve elde edilen ön veriler ışığında, *Tetrahymena thermophila*'nın atık sulardaki organik madde giderimi için kullanılabilirliğine yönelik potansiyel sunması ve gelecekteki çalışmalar için bir temel oluşturması hedeflenmiştir. Çalışma kapsamında, *Tetrahymena thermophila*'nın sentetik atık sulardaki KOI (Kimyasal Oksijen İhtiyacı) üzerindeki etkisini değerlendirebilmek için öncelikle atık suyun toksisitesinin belirlenebilmesi için MTT testi yapılmış ve LD50 değeri (350 mg/L) belirlenerek kinetik (10 dakikadan 24 saate kadar), pH (4-9 arası) ve sıcaklık (20-40 °C arası) parametreleri analiz edilmiştir. Kinetik çalışmanın sonuçları, 20. dakikadan sonra ortamdaki kirliliğin arttığını göstermiş ve en yüksek giderim verimliliğinin %27,97 ile 10. dakikada olduğunu belirlemiştir. Ayrıca, pH 7'de %25 ve 30 °C'de %23 arıtım performansı gözlemlenmiştir. Bu sonuçlar, *Tetrahymena thermophila*'nın atık su gideriminde potansiyel bir uygulama için umut vermektedir. Bu nedenle, protokolün optimize edilmesi için çeşitli çalışmaların yapılması planlanmaktadır.

Anahtar Kelimeler: Mikrobiyal Biyoteknoloji, Biyoremediasyon, Atık su giderimi, *Tetrahymena thermophila.*

DRAFT

ORAL PRESENTATION

Synthesis of aerogel materials from biowaste: comparison of supercritical carbon dioxide drying with atmospheric drying

M. Serhat Ekinci [1] (*ORCID: https://orcid.org/0000-0001-7240-9380),*
Emine Ekinci [2*] *(ORCID: https://orcid.org/0000-0002-0958-1513)*

[*1] Republic of Türkiye, Ministry of Labour and Social Security, 06520, Ankara, Türkiye.
[2] Gazi University, Engineering Faculty, Department of Chemical Engineering, 06570, Ankara, Türkiye.

*Corresponding author e-mail: eminekaya@gazi.edu.tr

Abstract

Aerogel materials have emerged as a highly versatile class of materials due to their exceptional properties, including low density, high porosity, and excellent thermal insulation. These unique characteristics make aerogels suitable for a wide range of applications, such as energy storage, catalysis, and environmental remediation. Biowaste, comprising organic residues from agricultural, forestry, and food industries, represents a promising and sustainable feedstock for the synthesis of aerogel materials. Utilizing biowaste not only addresses the issue of waste management but also contributes to the development of eco-friendly materials. The drying process employed during the synthesis significantly influences the final properties and performance of the aerogels. Various drying methods, including atmospheric, freeze, and supercritical drying, can be employed to eliminate the solvent from the gel skeleton. The drying process plays a crucial role in the fabrication of aerogels, as it involves the removal of liquid solvents from the wet gels while preserving their nanoporous structure to prevent shrinkage and cracking during the drying phase. Therefore, it is imperative to explore and compare different drying methods to optimize the synthesis of aerogel materials from biowaste. Atmospheric drying relies on the capillary force generated by the surface tension of the solvent. However, this method can cause the wet gel to collapse due to its low mechanical properties. Freeze-drying, on the other hand, is suitable for gels prepared using water-soluble polyamic acid as a precursor. Supercritical fluid drying, utilizing supercritical carbon dioxide, has emerged as a reliable method for producing high-performance aerogels, both inorganic and organic. Supercritical drying is a single-phase process that avoids the surface tension at the phase boundary, thus preserving the nano-structure of the aerogel. In this study, two drying methods; supercritical carbon dioxide drying and atmospheric drying, were employed to synthesize aerogel materials from biowaste. The results revealed that the aerogels dried using the supercritical carbon dioxide method exhibited a more uniform pore size distribution and satisfactory surface area with an amorphous structure.

Keywords: Aerogel, Biowaste, Supercritical drying

DRAFT

ORAL PRESENTATION

Rheological characterization of the tiger nut (*Cyperus esculentus* L.) oil-beeswax oleogels prepared with some flavors

Eda KESKİN USLU[1*] (*ORCID: https://orcid.org/0000-0002-8266-7137*)

[*1]Çanakkale Onsekiz Mart University, Ezine Vocational College, Department of Food Processing, Ezine, Çanakkale, Turkey

*Corresponding author e-mail: eda.keskinuslu@comu.edu.tr

Abstract

Within the FHD-2023-4261 project, flavored spreadable oleogels were prepared and rheological characterized using cold-pressed Tiger nut oil (*Cyperus esculentus L.*) and beeswax. For this purpose, unflavored (TBO), butter (TBO-BU) and honey (TBO-HM) flavored oleogels were prepared with beeswax and tiger nut oil. First of all, the oscillatory strain (%), *G´* and *G´´* changes of the samples were detected in the linear viscoelastic region (LVR), which is a range where gel structures can be kept under a certain strain without deterioration. The LVR strain values TBO, TBO-BU and TBO-HM were found to be % 0.0641, 0.0254 and 0.0163, respectively. Frequency sweep tests were performed to identify time-dependent behaviour in the LVR. It was detected that all of the oleogels met the *G´>G´´* condition, did not show free flow but exhibited solid-like behaviour and had a gel structure with the results of the frequency sweep test. The thixotropic behaviour of oleogels was examined by the time sweep test. Different strain values were applied to the oleogels at constant temperature and constant frequency at three different time intervals (in the first region; (strains determined in LVR), in the second region; (LVR strain≤Strain), in the third region; (LVR strain≥Strain)). The oleogels had thixotropic behaviour. Temperature ramp tests at constant amplitude and constant frequency were performed to examine their flow behaviour at different temperatures. It was observed that the flow behaviour of all oleogels was greatly affected by temperature and *G´* and *G´´* values gradually decreased with the increase in temperature. Rheological characterization of the produced oleogels is extremely important in determining the application areas of the food product.

Key words: wax oleogels, tiger nut oil, rheology, stability, flavor

DRAFT

ORAL PRESENTATION

Platinum(II) complexes with 2-/3-/4-(benzimidazol-2-yl)phenol: Synthesis, characterization and *in vitro* cytotoxic activities

Mahmut Gozelle[1*] (*ORCID:https://orcid.org/0000-0003-0234-6577*),
Yaprak Dilber Şimay Demir[2] (*ORCID:https://orcid.org/0000-0002-8248-0268*)

[*1]Gazi University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, Ankara, Türkiye.
[2]Gazi University, Faculty of Pharmacy, Department of Pharmacology, Ankara, Türkiye.

*Corresponding author e-mail: mgozelle@gazi.edu.tr

Abstract

Cisplatin and its derivatives continue to play a significant role in tumor therapy today, although drug resistance and cell toxicity limit their usage. Platinum(II) complexes containing benzimidazole ligands (Fig 1.) have been reported as cytotoxic against various cancer cell types. The objective of this investigation was to construct platinum(II) complexes [Pt(L)(DMSO)Cl_2] containing 2-/3-/4-(benzimidazol-2-yl)phenol ligands and evaluate their *in vitro* cytotoxic effects on HeLa human cervix and A549 human lung cancer cell lines. The benzimidazole ligands were synthesized through the reaction between 1,2-phenylenediamine and 2-/3-/4-hydroxybenzaldehyde. The formation of [Pt(L)(DMSO)Cl2] complexes was achieved through the reaction between benzimidazole ligands and cis-Pt(DMSO)2Cl2. The synthesized complexes were characterized, and their *in vitro* cytotoxic activity was examined using the MTT test. As a result, the **Pt2** and **Pt3** complexes have demonstrated significant potential for further investigation owing to their moderate in vitro cytotoxic activity, which is comparable to carboplatin.

Keywords: Platinum complexes, Cytotoxicity, A549, HeLa

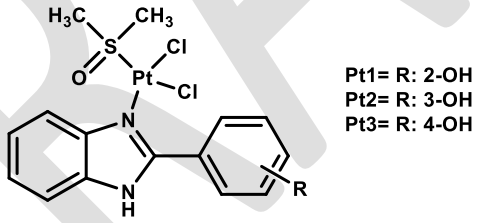


Figure 1. The structure of synthesized platinum(II) complexes

DRAFT

ORAL PRESENTATION

Sudaki boron kirliliği'ne karşı alternatif bir biyoremediasyon yöntemi

Şahin Erkaycan Yılmaz[1], Handan Açelya Kapkaç[1*] *(https://orcid.org/0000-0002-2812-1686)*, Çağdaş Saz[2] *(https://orcid.org/0000-0001-6230-5908)*, Cengiz Türe[1] *(https://orcid.org/0000-0001-6137-1866)*

[*1]Eskişehir Teknik Üniversitesi, Fen Fakültesi, Biyoloji Bölümü, Eskişehir, Türkiye
[2]Benli Recycling Technologies, Eskişehir, Türkiye

*haakdamar@eskisehir.edu.tr

Özet

Günümüzde, sanayileşme, nüfus artışı ve globalleşme gibi etkenler, tatlı su kaynaklarının kirlenme ve tükenme sorunlarını ciddi bir şekilde artırmaktadır. Bu etkenler hem ekosistemler hem de insan sağlığı üzerinde olumsuz etkiler oluşturmaktadır. Özellikle sulama amaçlı kullanılan tatlı su kaynaklarının sürdürülebilir bir şekilde korunması, gıda güvenliği açısından son derece kritik bir öneme sahiptir. Ancak, sulama sularına karışan bor (B) gibi kirleticiler, yalnızca çevresel sorunlara yol açmakla kalmayıp aynı zamanda ürünlerde ve insanlarda toksik etkilere neden olmaktadır. Su kaynaklarından borun (B) etkili bir şekilde uzaklaştırılması ise geleneksel yöntemlerle oldukça karmaşık ve zor bir süreç içermektedir Silli protozoanlar arasında Tetrahymena türleri, çevre araştırmaları ve toksikoloji çalışmalarında kullanılan en popüler model organizmalardandır. Literatürde, *Tetrahymena thermophila* üzerinde biyoremediasyon çalışmaları oldukça sınırlı olup Bor giderimi açısından bir çalışmaya rastlanmamıştır. Önerilen bu proje ile *Tetrahymena thermophila* protistinin Boron biyoremediasyonu potansiyeli araştırılmıştır. Öncelikle organizma üzerinde Bor'un farklı konsantrasyonlardaki toksisitesi analiz edilip, LD50 dozu MTT testi ile belirlenmiş ve 20 ppm olarak bulunmuştur. Çalışma kapsamında, farklı temas sürelerinde (15 dakikadan 24 saate kadar), pH değerleri (4-9) ve sıcaklık aralıkları (20-40°C) kullanılarak gerçekleştirilen kinetik, pH ve sıcaklık belirleme çalışmaları yapılmıştır. Sonuçlar, 2 saatlik temas süresi, pH 8 ve 30 °C'de %24,17 ile %24,67 arasında değişen maksimum B giderim verimliliği elde edildiğini göstermektedir. Bulgular, ökaryot *Tetrahymena thermophila* organizmasının biyoteknolojik araştırmalarda kullanılabilirliğinin ötesinde ekoteknolojik uygulamalara da umut verdiğini ve bu organizmanın sulama sularındaki B'un toksisitesini azaltma amacıyla kullanım potansiyelini yansıtmaktadır. Bu sonuçlar ışığında çalışmanın ileri düzeyde geliştirilmesi ve optimize edilmesi ile B biyoremediasyonu için alternatif bir yöntem geliştirilebilecektir.

Anahtar Kelimeler: Çevre Biyoteknolojisi, Biyoremediasyon, Boron giderimi, *Tetrahymena thermophila.*

DRAFT

ORAL PRESENTATION

Anti-apoptotik Bcl-2 proteinlerinin yaşlanma üzerindeki koruyucu etkilerinin maya modelinde araştırılması

Ayşenur Güler[1*] (*ORCID: 0000-0001-7276-0189*), Berna Kavakcıoğlu Yardımcı[2] (*ORCID: 0000-0003-0719-9094*)

[*1] Pamukkale Üniversitesi, Fen Bilimleri Enstitüsü, Kimya Bölümü, Denizli, Türkiye
[2] Pamukkale Üniversitesi, Fen Fakültesi, Kimya Bölümü, Biyokimya Anabilim Dalı, Denizli, Türkiye

*Sorumlu yazar e-mail: byardimci@pau.edu.tr

Özet

Genetik olarak düzenlenen ve organizmayı çevresel faktörlerin de etkisiyle meydana gelen yapısal/işlevsel değişmelerle ölüme sürükleyen olaylar bütünü olarak tanımlanan yaşlanma, hücrenin karmaşık ve merak uyandıran fenomenlerinden biridir. Yüksek ökaryotlardaki yaşlanma ilişkili yolakların mayalar gibi basit ökaryotlarda da iyi korunduğunun bilinmesi bu organizmayı yaşlanma süreci hakkında yeni bilgilerin elde edilmesi çalışmalarında populer bir model organizma haline getirmiştir. Bu çalışmada, insan Bcl-2, Bcl-xL ve Mcl-1 proteinlerinin hücre yaşlanması üzerindeki olası koruyucu etkileri ilgili proteinleri ektopik olarak ifade eden *Saccharomyces cerevisiae* BY4741 suşu kullanılarak incelenmiştir. Çalışmada öncelikle yaşlı geç stasyoner faza ulaştırılan hücrelerin FE-SEM görüntüleri alınarak morfolojik değerlendirmeleri yapılmış ve genç eksponansiyel faz hücrelerine göre çarpıcı değişimler gösterdikleri kaydedilmiştir. Boş vektörü taşıyan ve kontrol grubunu oluşturan geç stasyoner faz hücrelerine kıyasla plazmid DNA taşıyan yaşlı hücrelerdeki canlılık koloni oluşturan birim (CFU) yöntemi ile tespit edilmiş ve özellikle Bcl-2 proteinini ifade eden transforme suşun daha fazla prolifere olduğu görülmüştür. Söz konusu proteinlerin geç stasyoner faza giren yaşlı hücrelerdeki canlılık-ölüm yüzdeleri üzerindeki etkilerini aynı zamanda flow sitometrik Annexin V-PI testi ile de irdelenmiştir. CFU yöntemi ile uyumlu olarak kontrol grubuna kıyasla özellikle Bcl-2 proteinini ifade eden transforme suşun hücre canlılığında anlamlı bir artış belirlenmiştir. Ayrıca, ilgili proteinlerin ifadelenmesine bağlı olarak hücrenin biyokimyasal kompozisyonunda meydana gelen değişimler Fourier Dönüşümlü Kızılötesi Spektroskopisi (FTIR) tanımlanmıştır. Elde edilen verilere göre kontrol grubuna kıyasla plazmid DNA'larını taşıyan hücrelerde özellikle lipid ve mannan konsantrasyonlarının değiştiği görülmüştür. Elde edilen sonuçlar, yaşlanma ilişkili oksidatif stres karşısında başta Bcl-2 proteini olmak üzere bu proteinlerin önemli koruyucu roller oynadığına işaret etmektedir.

Anahtar Kelimeler: *Saccharomyces cerevisiae*, Yaşlanma, Oksidatif Stres, Bcl-2, Bcl-xL, Mcl-1

DRAFT

ORAL PRESENTATION

Antimikrobiyal maddeler için lipozomal formülasyon

Münevver Müge Çağal* (ORCID ID: 0000-0002-1786-1216)

Bursa Teknik Üniversitesi, Mühendislik ve Doğa Bilimleri Fakültesi, Biyomühendislik Bölümü, Bursa, Türkiye

*munevver.cagal@btu.edu.tr

Özet

Lipozomlar sulu bir merkezi çevreleyen bir ya da daha fazla fosfolipit çift tabasından oluşan veziküler yapılardır. Lipozomlar yapısal özellikleri, kompozisyonları, çeşitleri ve hazırlama yöntemleri ile birçok avantaja sahip olmaktadır. Günümüzde gıda, kozmetik, tıp, eczacılık, tekstil gibi birçok farklı alanda kullanılabilmektedir. Lipozomlar bu alanlarda ilaç taşıyıcı sistemler olarak işlev görmektedir. Lipozomların ilaç taşıyıcı sistemler olarak işlev görmesi; test edilecek maddenin lipozom içine enkapsüle olmasını ve hedef lokasyonda salınmasını sağlamaktadır. Lipozomların bir enkapsülasyon aracı olarak kullanılması antimikrobiyal açıdan da önemlidir. Günümüzde sıklıkla antimikrobiyal açıdan test edilmek istenen ajanlar; peptitler, antibiyotikler, biyoaktif bileşikler, ekstraktlar, esansiyel yağlar ve metal iyonları lipozomlarda enkapsüle edilmektedir. Lipozomal enkapsülasyon, biyoaktif bileşiklerin hidrofilik, hidrofobik özellikte olmasına göre lipozomların membranındaki fosfolipit kısımlarına ya da sulu merkezine tutunmasıdır. Lipozomal formülasyon bir ilaç taşıyıcı sistem olarak kullanılıyorsa enkapsülasyon canlı organizmadaki hedefe ulaşılana kadar devam etmektedir. Enkapsülasyon farklı parametrelere bağlı olsa da (ilaç/lipit oranı, ilacın alıkonması, ilaç ve lipozomun stabilitesi vb.) temelde 2 farklı teknik ile biyoaktif bileşikler lipozomlar içerisine enkapsüle olabilmektedir. Bu teknikler; ilaç enkapsülasyonunun vezikül oluşumu sürecinde gerçekleştiği pasif enkapsülasyon ve oluşumu tamamlanmış veziküllere biyoaktif bileşiklerin yüklendiği aktif enkapsülasyondur. Genel olarak lipozomların en önemli avantajları ise enkapsüle edilen maddenin toksisitesini azaltması ile daha az miktarda etken maddeden daha çok biyoyararlanım elde edilmesidir.

Anahtar Kelimeler: Lipozom, Antimikrobiyal Maddeler, Formülasyon

DRAFT

ORAL PRESENTATION

Üleksit'in insan periferal lenfositlerinde genotoksik ve hidrojen peroksit (H_2O_2)'e karşı antigenotoksik potansiyelinin komet testi ile değerlendirilmesi

Açelya Seyrek[1*] (*ORCID:* 0000-0003-0334-5154), Fatma Ünal[2] (*ORCID:* 0000-0002-7468-6186), Deniz Yüzbaşıoğlu[2] (*ORCID:* 0000-0003-2756-7712)

[*1]Gazi Üniversitesi, Fen Bilimleri Enstitüsü, Biyoloji Anabilim Dalı, Ankara, Türkiye.
[2]Gazi Üniversitesi, Fen Fakültesi, Biyoloji Bölümü, Ankara, Türkiye.

*Sorumlu yazar e-mail: acelya.seyrek@gazi.edu.tr

Özet

Üleksit (UX), bor bileşiklerinden biridir. Epoksi ve polipropilen kompozitlerde, otomotiv fren balatalarında ve sürtünme malzemelerinde, vitrifiye karo üretiminde, radyasyon koruyucu betonlarda, katalitik kağıt üretiminde ve dekoratif cam kaplama gibi çok çeşitli endüstriyel alanlarda kullanılmaktadır. Diğer yandan UX, çeşitli organizmalarda, antioksidan savunma sistemini teşvik etmesi, manyetik nanopartiküllerin neden olduğu oksidatif stresi azaltarak, ağır metallere karşı koruma sağlaması kısaca koruyucu yönde etkili olması nedeniyle önemle incelenmeye başlanan bir bileşiktir. Bu nedenle, bu çalışmada, UX'in *in vitro* genotoksik ve antigenotoksik potansiyeli, insan periferal lenfositlerinde komet testi ile araştırılmıştır. İki sağlıklı genç donörden elde edilen izole lenfositler, UX'in tek başına dört farklı konsantrasyonu (2,5; 5, 10 ve 20 µg/mL) ve aynı dozlar ile eş zamanlı şekilde H_2O_2 (100 µM) ile muamele edilmiştir. Her muamele için bir negatif (saf su) ve bir pozitif (100 µM H_2O_2) kontrol kullanılmıştır. Komet kuyruk uzunluğu (µm), kuyruk yoğunluğu (%DNA) ve kuyruk momenti değerlendirilmiştir. Floresan mikroskobuna bağlı "Comet Assay IV" programı kullanılarak (Perceptive Instruments Ltd., İngiltere), her konsantrasyon için toplam 200 (her donör için 100) nükleoid değerlendirilmiştir. Elde edilen sonuçlara göre, negatif kontrole kıyasla, UX'in tek başına uygulamasında, kuyruk uzunluğunda 2,5 ve 5 µg/mL'lik konsantrasyonlarda istatistiksel olarak anlamlı azalmalar meydana gelirken, 10 ve 20 µg/mL'lik konsantrasyonlarda ise anlamlı artış olmuştur. UX+H_2O_2 eş zamanlı uygulamasında, incelenen tüm parametrelerde (kuyruk uzunluğu, kuyruk yoğunluğu ve kuyruk momenti), pozitif kontrole kıyasla anlamlı düşüşler gözlenmiştir. Tüm veriler birlikte değerlendirildiğinde, UX'in DNA'da genelde anlamlı bir DNA hasarına neden olmadığı, daha ziyade, H_2O_2'nin sebep olduğu DNA hasarına karşı antigenotoksik bir potansiyele sahip olduğu görülmektedir. Ancak UX'in olası genotoksik ve antigenotoksik potansiyeli için daha farklı testler kullanılarak hem *in vitro* hem de *in vivo* detaylı araştırmalar yapılması gerekmektedir.

Teşekkür: Bu çalışma, Gazi Üniversitesi Bilimsel Araştırma Projeleri Koordinasyon Birimi tarafından (Proje kodu: FYL-2023-8246) desteklenmiştir.

Anahtar Kelimeler: Üleksit, Hidrojen peroksit, Genotoksisite, Antigenotoksisite, Komet testi

DRAFT

ORAL PRESENTATION

Borik asitin hidrojen peroksite ve bakır oksit nanopartikülüne karşı antigenotoksik potansiyelinin komet testi ile incelenmesi

Aleyna Halıcı[1*] (*ORCID:* 0000-0002-0423-8441), Fatma Ünal[2] (*ORCID:* 0000-0002-7468-6186), Deniz Yüzbaşıoğlu[2] (*ORCID:* 0000-0003-2756-7712)

[*1]Gazi Üniversitesi, Fen Bilimleri Enstitüsü, Biyoloji Anabilim Dalı, Ankara, Türkiye.
[2]Gazi Üniversitesi, Fen Fakültesi, Biyoloji Bölümü, Ankara, Türkiye.

*Sorumlu yazar e-mail: aleyna.halici@gazi.edu.tr

Özet

Boratların en yaygın şekli olan borik asit (BA), günümüzde sağlık, kozmetik, cam, pestisit, yangın geciktirici, gıda ve biyositler gibi pek çok alanda yaygın bir şekilde kullanılmaktadır. BA, bitki, insan ve hayvanlarda hücre büyümesini, çoğalmasını ve gelişimini etkilemektedir. BA, kanserojen, mutajen ve genotoksinlerin neden olduğu DNA hasarına karşı antigenotoksik özellik göstermektedir. Bu çalışmada, BA'in hem tek başına olası genotoksik ve hem de hidrojen peroksit (H_2O_2-100 µM) ve bakır oksit nanopartikülüne (CuO NP-100 µg/mL) karşı olası antigenotoksik potansiyeli, *in vitro* insan lenfositlerinde komet testi kullanılarak araştırılmıştır. İzole lenfositler, BA'in beş farklı konsantrasyonu (0,5; 0,25; 0,125; 0,0625 ve 0,0313 µg/mL) ile hem tek başına ve hem de hidrojen peroksit (BA+H_2O_2) ve CuO NP (BA+CuO NP) ile eş zamanlı olarak 1 saat muamele edilmiştir. Ayrıca, bir negatif kontrol (distile su) ve 2 farklı pozitif kontrol (100 µM H_2O_2 ve 100 µg/mL CuO NP) grubu kullanılmıştır. Her konsantrasyon için 200 nükleoid incelenmiş ve DNA hasarı, komet kuyruk uzunluğu (µm), kuyruk yoğunluğu (%DNA) ve kuyruk momenti açısından değerlendirilmiştir. Yapılan analizlere göre, BA'in tek başına muamelesi, negatif kontrole kıyasla, DNA hasarını hiç bir parametrede artırmamıştır. Tam tersine, BA+H_2O_2 eş zamanlı muamelesi, kuyruk uzunluğu ve kuyruk yoğunluğunda, pozitif kontrole (H_2O_2) kıyasla anlamlı bir azalma oluşturmuştur. BA+CuO NP eş zamanlı uygulaması, pozitif kontrole (CuO NP) kıyasla, kuyruk uzunluğunu anlamlı olmayan şekilde azaltmıştır (0,5 µg/mL hariç). Kuyruk yoğunluğu ve momentinde ise anlamlı olmayan değişiklikler görülmüştür (0,5 µg/mL hariç). Bu sonuçlar, BA'in tek başına DNA hasarı oluşturmadığını, tam tersine, H_2O_2 ve CuO NP'nin oluşturduğu DNA hasarlarını iyileştirici/antigenotoksik potansiyele sahip olduğunu göstermektedir. Ancak daha güvenilir sonuçlara ulaşabilmek için, diğer *in vitro* ve *in vivo* genotoksisite testleri ile daha detaylı araştırmalar yapılması gerekmektedir.

Anahtar Kelimeler: Borik asit, hidrojen peroksit, bakır oksit nanopartikülü, genotoksisite, antigenotoksisite, komet testi.

DRAFT

ORAL PRESENTATION

Innovative Strategies in KRAS Inhibitor Development: Integrating Pharmacophore-Based Virtual Screening, Molecular Docking, and AI-Powered ADMET Insights

Mazlum TÜRK[1] *(ORCID: https://orcid.org/0000-0003-1683-9284)*, Harun NALÇAKAN[2] *(ORCID: https://orcid.org/0000-0003-3821-8681)*, Gülbin KURTAY[1*] (*ORCID: https://orcid.org/0000-0003-0920-8409*)

[*1]Hacettepe University, Faculty of Science, Department of Chemistry, Ankara, Türkiye.
[2]Ankara University, Faculty of Science, Department of Chemistry, Ankara Türkiye.

*Corresponding author e-mail: gulbinkurtay@hacettepe.edu.tr

Abstract

The RAS proteins, specifically KRAS, HRAS, and NRAS, serve as binary molecular switches, undergoing dynamic transitions between their active (GTP-bound) and inactive (GDP-bound) states. These proteins hold a pivotal role in the regulation of critical cellular signalling pathways governing fundamental processes such as cellular differentiation, proliferation, and viability. Particularly, KRAS stands out as the most frequently mutated oncogene in human malignancies, manifesting in approximately 30% of cases. Historically, targeting KRAS has posed a formidable challenge due to its robust affinity for GDP and GTP molecules within intracellular environments. However, recent scholarly literature has advanced several pharmaceuticals as potent inhibitors of KRAS, thus offering promising prospects in the realm of anticancer therapeutics.

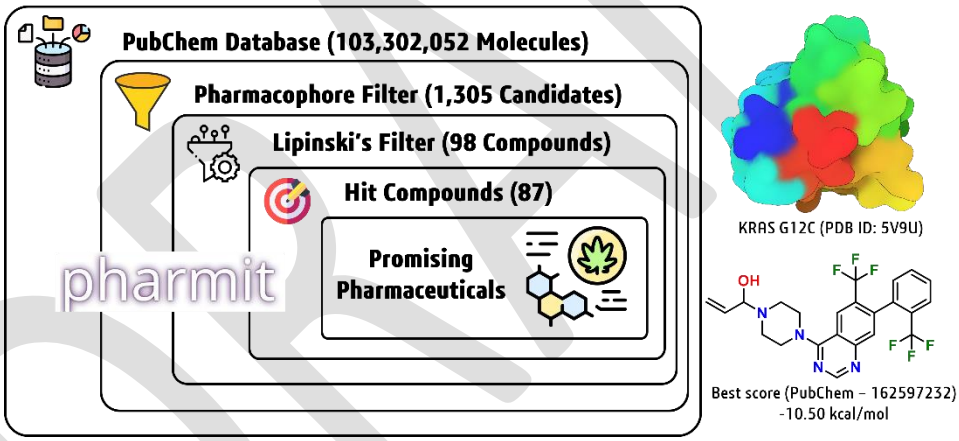


Figure 1. (*left*) Pharmacophore-based virtual screening flowchart (*right*) screened ligand with the highest docking score along with KRAS protein

In this scope, we have applied various computational techniques, including pharmacophore-based virtual screening through Pharmit, to uncover potent KRAS inhibitors. Specifically, PubChem database, which contains 103,302,052 compounds, was employed using a pharmacophore model based on the 91S/KRAS co-crystallized receptor structure (PDB ID: 5V9U). After implementing pharmacophore and Lipinski's filters, we identified 87 promising compounds for subsequent molecular docking simulations. To this purpose, target molecules were constructed using GaussView 5.0.8, and further refined their geometry through optimization studies using Gaussian 09 with the DFT/B3LYP/6-31G(d,p) method. To assess the compounds' efficacy and their compatibility with drug-like characteristics, we conducted ADMET evaluations utilizing SwissADME, OSIRIS, Molinspiration, Toxtree and an AI-driven platform, Syntelly. Simultaneously, we also performed molecular docking simulations (via SAMSON platform/2022-R2 and OneAngstrom/AutoDock Vina Extension) to unveil the binding potential of these screened candidates as potent KRAS inhibitors.

Keywords: KRAS, pharmit, virtual screening, molecular docking, ADMET.

ORAL PRESENTATION

Farklı habitatlardan izole edilen bazı aktinobakterilerin antimikrobiyal aktivitelerinin taranması

Çiğdem YÜKSEL DEMİRCİ* (*ORCID: https://orcid.org/0000-0001-8506-9899*),
Mustafa OSKAY (*ORCID: https://orcid.org/0000-0001-8693-5621*)

Manisa Celal Bayar Üniversitesi, Fen-Edebiyat Fakültesi, Biyoloji Bölümü, Temel ve Endüstriyel Mikrobiyoloji Anabilim Dalı, Manisa, Türkiye

*Sorumlu yazar e-mail:cigdem.yuksel@cbu.edu.tr

Özet

Aktinobakteriler, tedavide kullanılan antibiyotikler başta olmak üzere, bitki büyümesini teşvik edici maddeler, antimikrobiyal, antitümöral ve antiinflamatuar ajanlar olarak da bilinen önemli kimyasalları ve metabolitleri üretme potansiyeline sahiptirler. Bu metabolitlerin çoğunluğu, endüstriyel olarak tekstil, fırıncılık, tıp, biyoteknoloji ve tarım sektörü gibi farklı alanlarda uygulama olanağı bulmaktadır. Birçok mikrobiyal patojenin mevcut tedavide kullanılan antibiyotiklere karşı direncinin ortaya çıkması ve fungal enfeksiyonlarının sayısındaki artış, biyolojik olarak aktif bileşiklerin yeni rezervlerinin bulunmasına olan ilginin artmasına neden olmuştur. Bu nedenle, araştırmacılar son zamanlarda daha etkili yeni biyolojik aktif bileşiklerin keşfi için yoğun çalışmalar gerçekleştirmektedir. Bu araştırmada, farklı habitatlardan alınan toprak örneklerinden aktinobakteri izolasyonları gerçekleştirilmiş ve antimikrobiyal aktiviteleri belirlenmiştir. Aktinobakteri izolasyonu için toprak örneklerinin her birinden 10 g alınarak steril petrilere aktarılmış ve üzerine 0.1 g $CaCO_3$ ilave edilerek 2 gün ön işlemden geçirilmiştir. Sonrasında farklı dilüsyonları hazırlanarak uygun seyreltmelerden 1'er ml steril petri kabına aktarılarak üzerine 45-50 ^oC'ye kadar soğutulmuş International Streptomyces Project (ISP) 2 ve Potato Dekstroz Agar (PDA) besiyerlerinden eklenmiştir. Homojen karışım sağlandıktan sonra petriler 30 ^oC'de 7-21 gün inkübasyona bırakılmıştır. İnkübasyonun 2. gününden itibaren ekimi yapılan besiyerleri çıplak gözle ve ışık mikroskobu altında (10x) tipik aktinobakteri varlığı açısından taranmıştır. Aktinobakteri olduğu tespit edilen izolatlar numaralandırılarak belirlenmiştir. Çalışma sonunda toplamda 80 aktinobakteri izole edilmiştir. İzole edilen aktinobakterilerin antimikrobiyal aktiviteleri nokta ekim yöntemi ile kültür koleksiyonundan temin edilen *Staplylococcus aureus*, *Escherichia coli*, *Bacillus cereus* ve *Candida albicans'a* karşı tespit edilmiştir. Taranan aktinobakterilerden UZ-74, UZ-78, TA-13, TA-14, TA-15, TA-16 ve TA-25 kodlu izolatlar tüm test organizmalarına karşı farklı oranlarda (4-30 mm) inhibisyon zonu oluşturarak antimikrobiyal aktivite göstermiştir. Antimikrobiyal aktivitesi yüksek çıkan izolatların tanımlanması ve biyoaktif metabolitin üretimi, saflaştırılması ve tipinin belirlenmesi çalışmaları halen devam etmektedir. Seçilen antimikrobiyal aktivitesi yüksek izolatlardan elde edilebilecek metabolitlerin yeni ve farklı alanlarda kullanılabilme ihtimali bulunmaktadır.

Anahtar Kelimeler: Aktinobakteriler, antibiyotik, antimikrobiyal, metabolit tarama, izolasyon

DRAFT

ORAL PRESENTATION

Evaluation of black elderberry tea samples in the market and grown with organic farming in terms of European Pharmacopoeia and analysis of their phenolic contents by RP- HPLC techique

Merve Türker (*ORCID: https://orcid.org/0009-0000-8245-9349*), Hatice Nur Negiz *(ORCID: https://orcid.org/0009-0009-7384-4881*), Burçin Özüpek (*ORCID: https://orcid.org/0000-0003-2159-9860*), Didem Deliorman Orhan (*ORCID: https://orcid.org/0000-0003-3916-4048*)

Gazi University, Faculty of Pharmacy, Department of Pharmacognosy, Ankara, Turkey

*Corresponding author e-mail: didemdeliorman@gmail.com

Abstract

In this study, quality control studies were carried out on the flowers of *Sambucus nigra* L. (Viburnaceae), known as black elderberry in Anatolia. Among the people, the flowers of the plant are used in the form of tea for diuretic, diaphoretic, and laxative purposes, and all samples in this study were purchased from herbalists. Only one sample was obtained from a farm producing organically in the Konya region. The samples were analyzed in terms of the criteria (microscopic analysis, foreign matter, loss of drying, determination of total ash content, analysis of thin layer chromatography) specified in the "Sambuci flos" monograph in the European Pharmacopoeia 10.0, and the rutin, caffeic acid, chlorogenic acid, and hyperoside contents of the samples were determined using the High Performance Liquid Chromatography technique. Except for the ash amounts of all samples, the results were incompatible with the Pharmacopoeia. By HPLC analysis, it was determined that the phenolic compounds analyzed in all samples were in different amounts. While caffeic acid could not be detected in flowers produced by organic farming, coarse pedicel fragments were also found in high amounts. It was predicted that these differences were related to the collection time, collection, and drying methods of the samples. In terms of public health, these drugs should be standardized in terms of pharmacopoeial criteria.

Keywords: Elderberry, Sambucus nigra, European Pharmacopoeia, RP-HPLC

DRAFT

ORAL PRESENTATION

The investigation of the anticancer effects of royal jelly on A549 lung cancer cell line and BEAS-2B normal human bronchial epithelial cells

Abdullah Taşkın[1*] (0000-0001-8642-1567), Hasan Ulusal[2] (0000-0003-3890-2088)

[*1]Harran University, Faculty of Health Sciences, Department of Nutrition and Dietetics, Şanlıurfa, Türkiye
[2]Gaziantep University, Faculty of Medicine, Department of Biochemistry, Gaziantep, Türkiye

*abdullahtaskin52@harran.edu.tr

Abstract

Royal jelly is a product made by honey bees that contains bioactive peptides. These peptides provide unique therapeutic effects, such as anticancer, antioxidant, antimicrobial, anti-aging, and anti-inflammatory properties. In addition to these unique properties, royal jelly has also been reported to have minimal toxicity compared to other bee products. One major challenge in cancer treatment is minimizing harm to healthy cells during chemotherapy. This study aimed to investigate the anticancer effects of royal jelly on the A549 lung cancer cells and BEAS-2B human normal bronchial epithelial cells. Concentrations of royal jelly at 0.001, 0.01, 0.1, 1, 10, and 100 mg/ml were prepared. These concentrations were incubated with A549 and BEAS-2B cells for 24, 48, and 72 hours. The cytotoxic effect of royal jelly concentrations was analyzed by the MTT method. IC50 values were calculated with the GraphPad Prism 8 program. According to MTT assay, it was found that 0.001-100 mg/mL royal jelly concentrations had a cytotoxic effect on A549 and BEAS-2B cells in a concentration-related manner ($p<0.001$). At the end of 24, 48, and 72 hours of incubation periods, the IC50 values of A549 cells were calculated as 0.862, 2.872, and 0.697, respectively, and the IC50 values of BEAS-2B cells were calculated as 7.420, 3.275 and 6.328, respectively. After three incubation periods, it was found that royal jelly had more cytotoxic effects on cancer cells than on normal cells. Our results show that royal jelly reduces cell proliferation more in the A549 cells than in BEAS-2B cells, depending on dose and time, at 24, 48, and 72 hours of incubation periods. This difference between cancer and normal cells shows that royal jelly may be a potential cancer agent candidate. Supporting these results with further molecular analysis may be an alternative option in the treatment of lung cancer and other cancers.

Keywords: Lung cancer, Royal Jelly, Cytotoxicity

DRAFT

ORAL PRESENTATION

Alzheimer's Alchemy: Unveiling the Dual-Inhibitory Potential of Tacrine-Donepezil Hybrids

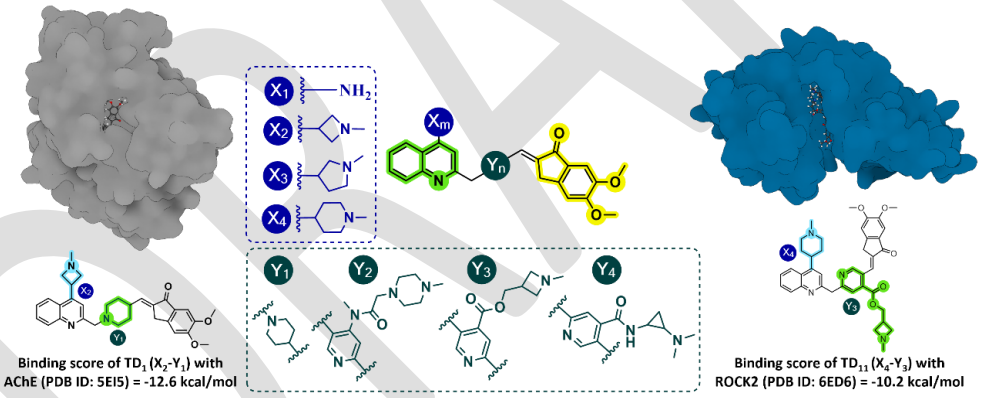
Harun Nalçakan[1] *(ORCID: https://orcid.org/0000-0003-3821-8681)*, Mazlum Türk[2] *(ORCID: https://orcid.org/0000-0003-1683-9284)*, Gülbin Kurtay[2*] (*ORCID: https://orcid.org/0000-0003-0920-8409*)

[1]Ankara University, Faculty of Science, Department of Chemistry, Ankara Türkiye.
[2]Hacettepe University, Faculty of Science, Department of Chemistry, Ankara, Türkiye.

*Corresponding author e-mail: gulbinkurtay@hacettepe.edu.tr

Abstract

Alzheimer's disease (AD) is a multifaceted neurodegenerative disorder characterized by progressive memory decline, cognitive dysfunction, and pronounced behavioral anomalies. Accumulating evidence suggests pivotal roles of acetylcholinesterase (AChE) and Rho-associated coiled-coil kinase 2 (ROCK2) proteins in the pathogenesis of AD. Contemporary therapeutic strategies for AD predominantly focus on enhancing cholinergic neurotransmission, specifically by elevating cerebral acetylcholine concentrations. This modality offers symptomatic relief, ameliorating the clinical manifestations of AD. Concurrently, the ROCK2 protein, implicated in AD pathophysiology, modulates neural processes and synaptic retraction, influencing neural information processing and retention. Recent advancements spotlight hybrid molecules with dual-inhibitory capacities. A salient hybrid derivative, amalgamating FDA-sanctioned tacrine and donepezil, has demonstrated potential in attenuating the activities of these pivotal proteins, heralding a promising avenue for AD therapeutics.



Within this framework, our study embarked on an exhaustive computational exploration, entailing structural modifications of the piperidine and amine moieties of the hybrid molecule (TA8Amino), culminating in designing 71 avant-garde therapeutic candidates. Employing Gaussian 09 (DFT/B3LYP/6-31G(d,p)), corresponding geometry optimization studies were performed, while GaussView 5.0.8 rendered visual representations of the target entities. These novel compounds' pharmacokinetic profiles and drug-likeness attributes were assessed using the AI-powered Syntelly platform, with corroborative analyses conducted via SwissADME, OSIRIS, and Molinspiration. Toxicological assessments were executed using Toxtree software. Furthermore, molecular docking simulations conducted via the SAMSON platform (2022-R2) and OneAngstrom/AutoDock Vina Extension delineated the prospective inhibitory efficacies of the designed entities (TD1-71) against AChE (PDB ID: 5EI5) and ROCK2 (PDB ID: 6ED6).

Keywords: AChE, ROCK2, pharmacophore modeling, virtual screening, molecular docking, ADMET

DRAFT

ORAL PRESENTATION

Green syntesis of rGO-NiO-CuO nanocomposite for photocatalytic aplications

Salih Açıl[1*](*ORCID: https://orcid.org/0000-0001-5948-9216)*, Furkan Soysal[2] (*ORCID: https://orcid.org/0000-0002-2558-2014*), Zafer Çıplak[3] (*https://orcid.org/*0000-0003-2449-5274), Nuray Yıldız[1] (*https://orcid.org/*0000-0003-2428-3474)

[*1]Ankara University, Faculty of Engineering, Department of Chemical Engineering, Ankara,Turkey
[2]Ankara Yildirim Beyazit University, Department of Chemical Engineering, Faculty of Engineering and Natural Sciences, Ankara, Turkey
[3] Sivas Cumhuriyet University, Faculty of Engineering, Department of Chemical Engineering, Sivas, Turkey

sacil@ankara.edu.tr

Abstract

In this study, rGO-NiO-CuO three-component nanocomposite was produced with a simple, green and one-step production method using *Cetraria Islandica L. Ach* lichen extract which undertook the roles of both a green reducing agent and an effective stabilizing agent. The obtained samples were characterized by Fourier conversion fluorescence spectrophotometry (FTIR), UV-visible spectrophotometry (UV), Scanning electron microscopy (SEM) and X-ray diffraction (XRD). As a result of the strong reducing power of *Cetraria Islandica L. Ach* the chemical reduction of GO and the production of NiO and CuO nanostructures on the surface of rGO nanosheets were successfully obtained simultaneously with this one-step synthesis approach. Moreover, as an effective stabilizing agent the lichen extract provide uniform coating of rGO nanosheets with NiO and CuO nanoparticles and prevent aglomeration of the nanostructures. The rGO-NiO-CuO ternary nanocomposite was used in the photocatalytic removal of methylene blue (MB). In the study, it was determined that rGO-NiO-CuO nanocomposite removed 72% of MB in visible region irradiation in 180 minutes. It has been observed that the rGO-NiO-CuO nanocomposite, produced in a single step with an environmentally friendly method, has great potential for photocatalytic applications.

Keywords: rGO,NiO,CuO, photocatalytic, greeen synthesis

DRAFT

ORAL PRESENTATION

AI-Guided ADMET Analysis and Molecular Docking in the Discovery of Novel LIM Kinase 1 Inhibitors: A Pharmacophore-Based Virtual Screening Approach

Gülbin KURTAY (*ORCID: https://orcid.org/0000-0003-0920-8409)*

Hacettepe University, Faculty of Science, Department of Chemistry, 06800, Ankara, Turkey

Abstract

LIM kinases (LIMKs) are a group of protein kinases that play a crucial role in regulating actin dynamics. This group consists of two members, known as LIMK1 and LIMK2. LIMK1 plays an essential function in regulating the morphology and synaptic strength of dendritic spines in a well-functioning brain. On the other hand, in patients suffering from dementia, an increased enzymatic activity of LIMK1 leads to the deterioration of the structural integrity of these dendritic spines. Consequently, the regulation of LIMK1 in the context of dementia yields enhancements in both the structural characteristics and abundance of dendritic spines, so mitigating the impact of Alzheimer's Disease (AD). In pursuit of this objective, a number of small compounds have emerged as a promising therapeutic target for Alzheimer's disease (AD).

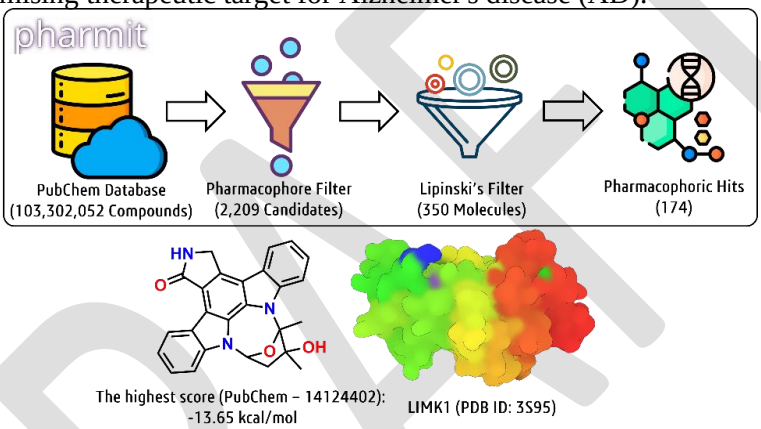


Figure 1. (*top*) Virtual screening flowchart in Pharmit server (*bottom*) screened candidate with the highest binding pose along with LIMK1 receptor

In order to identify potent inhibitors of LIMK1, a diverse range of computational methodologies have been employed, including the utilization of Pharmit for pharmacophore-based virtual screening. In this study, a pharmacophore model was constructed using the co-crystallized receptor structure of STAUROSPORINE/LIMK1 (PDB ID: 3S95). The model was applied to the PubChem database, which has a vast collection of 103,302,052 compounds. After the integration of the pharmacophore and Lipinski's filter, a total of 87 compounds were identified as promising candidates for subsequent molecular docking simulations. In the subsequent analysis, the Gaussian 09 software package was employed to do geometry optimization investigations using the DFT/B3LYP/6-31G(d,p) method. Additionally, the software GaussView 5.0.8 was applied to visually represent the molecular structures. The evaluation of the pharmacological properties of the ligands was conducted using ADME analysis, employing several tools such as SwissADME, OSIRIS, and Molinspiration. In addition, the assessment of toxicity characteristics for these ligands was conducted utilizing the Syntelly platform, which is an AI-guided tool, as well as the Toxtree program. The process of molecular docking simulation was conducted utilizing the SAMSON platform/2022-R2 and OneAngstrom/AutoDock Vina Extension. This was done in order to do a detailed examination of the binding interactions and affinities of the identified candidates against LIMK1.

Keywords: Staurosporine, LIMK1, virtual screening, DFT, ADMET, molecular docking

DRAFT

ORAL PRESENTATION

Lipid-rich Microalgae for Sustainable and Renewable Bio-fuel Production

Ecem AKSU*, Mesude İSAR, Gamze TURAN

Ege University, Fisheries Faculty, Aquaculture Department, 35100, Bornova, İzmir, TÜRKİYE

*Corresponding author e-mail: gamze.turan@ege.edu.tr

Abstract

Due to continued use of fossil fuels is not sustainable as they are a finite resource and their combustion lead to environmental problems, the recent investigations started to focus on more renewable energy resources. As an alternative energy resource Biodiesel is an environmentally friendly and renewable fuel source obtained from vegetable oils and used in diesel motors. Since some of terrestrial plants, such as soybean, canola, corn, coconut and palm tree oils used in food purposes and they require huge areas to grow, in recent years studies on microalgae as renewable fuel resources gained more attention due to their surprising ability to grow in unused areas. Like plants, microalgae use sunlight to produce oils however their production is more efficient than crop plants. Oil productivity of many microalgae is higher than the best oil producing crops (Ahmad et al., 2011). Microalgae have been suggested as very good candidates for biodiesel production, because they have some advantages such as higher photosynthetic efficiency, higher biomass production and faster growth compared to other energy crops (Miao and Wu, 2006). Additionally, it provides advantage for usage of unfertile lands, inefficient for agriculture, for biodiesel production instead of using productive lands for food production. Aim of this study was to summarized biomass yield and lipid production of various microalgal species of cultured in phototrophic systems.

Keywords: Microalgae, energy, biofuel, biodiesel, global warming.

DRAFT

ORAL PRESENTATION

Vücut sıcaklığının düzenlenmesinde UCP1'in fonksiyonu

Hülya Yıldız[1*] (0000-0001-6862-8011)

[*1] Burdur Mehmet Akif Ersoy Üniversitesi, Fen Bilimleri Enstitüsü, Biyoloji Anabilim Dalı, Türkiye

*yildizhulya1687@gmail.com

Özet

Mitokondriyal anyon taşıyıcı ayrılma proteinleri (uncoupling proteinler; UCP) mitokondrinin kristasında sentezlenir. Bu ayrılma proteinlerin farklı dokularda etkili ve farklı fonksiyona sahip 1-5 aralığında üyesi bulunmaktadır. UCP1 olarak adlandırılan ilk üyesi kahverengi yağ dokuya spesifiktir. UCP1, termogenez olarak bilinen vücut sıcaklığının korunmasında ve aynı zamanda adipoz dokuda depolanan enerjinin kullanılmasında işlev görür.

Kahverengi yağ dokusunun asıl görevi yağı depolamak değil, yağı yakma yoluyla vücuda ısı sağlamak yani termogenezistir. Kahverengi yağ dokusu termogenezinde β-adrenerjik aracılı lipoliz aktivasyonu ve yağ asitlerinin tekrarlayan yıkımı olmaktadır. Bu işlevlerini termogenin olarak da adlandırılan mitokondriyal anyon taşıyıcı ayrılma proteini (UCP1) ile yapar. Aynı zamanda kahverengi yağdaki kahverengi adipositler gibi beyaz yağ dokuda bulunan kahverengi adipositlere benzeyen bej adipositlerinde UCP1 ifadesi ile karakterize edilen termojenik özelliklere sahip olduğu bilinmektedir. Yağ asidi varlığında UCP1 üzerindeki kapanma (Shutting) dediğimiz bölgeyle ve oluşan proton gradyanına bağlı olarak ısı oluşmaktadır. UCP1 bunu, mitokondri iç membranında protonların ATP sentaz kompleksinden geçmeden mitokondri matriksine yeniden girmesine izin veren bir kanal oluşturarak yapar. Böylece solunumda ATP sentezi kısmen engellenirken, adipositlerdeki yağ asitlerinin sürekli oksidasyonu ile enerjinin ısı olarak dağılımı gerçekleşir. Soğuğa maruz kalma ve beslenme kahverengi yağ dokusunun aktivitesini ve Santral Sinir Sistemi'nden salınan norepinefrin UCP1 ekspresyonunu arttırır. β-adrenerjik antogonistleri, troid hormonu, insülin ve cAMP analogları da UCP1 ekspresyonunu arttırır. Kahverengi yağ dokuda UCP1 aktivasyonunun artması ile lipolizisin uyarılması hem de UCP1 gen transkripsiyonu da gerçekleşmektedir. Ayrıca mitokondriyal biyogenezis, kahverengi yağ dokunun hiperplazisi, beyaz yağ dokudan kahverengi ve bej adipozitlerin oluşması gerçekleşmektedir. Bu sebeple UCP1 sistemi, tüm vücut enerji dengesinin, yağlanmanın ve obezitenin kontrolü için bir hedef olmaktadır.

Anahtar Kelimeler: termogenez, ayrılma proteinleri, UCP1.

DRAFT

ORAL PRESENTATION

Producing Cellulose from a Green Seaweed, *Ulva lactuca,* for Paper and Bioplastic Production

Kürşat BAĞCI * , Mesude İSAR, and Gamze TURAN

Ege University, Fisheries Faculty, Aquaculture Department, 35100, Bornova, İzmir, TÜRKİYE

*Corresponding author e-mail: gamze.turan@ege.edu.tr

Abstract

Cellulose is an indispensable raw material and also the world demand for cellulose has been increasing day by day. Apart from the basic usage areas of cellulose in paper, and cardboard, in the petrochemical and construction sector, in the paint and coating industry. it is an essential raw material used in food, medicine, cosmetics, the textile industry, and many industrial categories.
Algae, especially green algae, are good sources of cellulose and developments in the paper pulp industry have made Macroalgae practical alternatives to the use of trees (De Poli et al., 1994; You, 2008).
In this research, the usage of *Ulva lactuca* in cellulose production was determined. The study results showed that the content of raw cellulose was 22.21 grams per 100 gr dry weight of *Ulva lactuca*. And, the amounts of α (Alpha) and β (Beta) cellulose fractions were 6.58 and 23.35 grams per 100 gr dry weight of *Ulva lactuca*. From the study result, it was concluded that it is effective and favorable to use high α(Alpha) cellulose containing *Ulva lactuca* in paper production and high β (Beta) cellulose contenting *U. lactuca* in bioplastic production.

Keywords: *Ulva*, raw cellulose, α (Alpha) cellulose, β (Beta) cellulose, paper production, bioplastic production.

DRAFT

ORAL PRESENTATION

Production of Natural and Functional β-carotene Pigment from A Green Microalga, *Dunaliella viridis* (Chlorophycea), Cultivated in Laboratory Conditions

Mesude İSAR*,Kürşat BAĞCI, and Gamze TURAN

Ege University, Fisheries Faculty, Aquaculture Department, 35100, Bornova, İzmir, TÜRKİYE

*Corresponding author e-mail: gamze.turan@ege.edu.tr

Abstract

β-carotene is a pigment that has a significant position in clearing free radicals, protecting the cell against destructive rays such as UV, regulating immunity, and preventing cancer as a potential antioxidant. In the food industry, β-carotene can be utilized to color foods and preserve the color of fruit juices. It can also be mixed with feeds to color salmon, trout, shellfish, and poultry meat (Ben-Amotz, 1999; Garcia-Gonzalez et al., 2000; Santin-Montanya, 2007; Takagi et al., 2005). A green microalga, *Dunaliella* sp. (Chlorophyceae) has achieved a critical point in algal biotechnology with the β-carotene production. Under salt and/or light stress conditions, *Dunaliella* spp. can accumulate β-carotene between 2-5% of its dry weight, depending on the system utilized. *Dunaliella* spp. naturally spread in saltwork fields sach as in Ayvalık Tuzlası, Çamaltı Tuzlası, etc. in Türkiye. However, β-carotene production of natural species of *Dunaliella* in Türkiye is very limited. For this reason, this study was contacted to produce *Dunaliella viridis* biomass and β-carotene production and to explore inexpensive natural β-carotene sources assembled in our country that can be used in coloring fish, in cosmetic products such as blush, lipstick, and UV-protected sunscreens, and as vitamin A pills. Study results showed that the cultivation of the vegetative cells and β-carotene production of the green alga *Dunaliella viridis* is possible under the laboratory conditions. During the experients, the vegetative growth of *D. viridis* was achieved at 67 µmol $m^{-2}s^{-1}$ ligh density, while at 8000 µmol m-2s-1 light intensity, Cystic *Dunaliella* cell formation started in the 5th week of the experiment followed by β-carotene production in the system and β-carotene amount accumulated in *D. viridis* was measured as 2,542.12 ± 781.35 µg.g^{-1} of dry weight (dw).

Keywords: *Dunaliella viridis*, Chlorophyceae, Natural and Functional Pigments, Chlorophyll, β-carotene

DRAFT

ORAL PRESENTATION

Seaweed Biorefinery

José Stênio[1], Carlos Prentice[1], Gamze Turan[1,2*]

[1)] Federal University of Rio Grande-Marine Aquaculture Station- Rua do Hotel, nº 02, 96210-000, Rio Grande-RS, Brazil.
[2)]Ege University Fisheries Faculty, Aquaculture Department 35100, Bornova, İzmir, Türkiye.

*Corresponding Author: gamze.turan@ege.edu.tr

Abstract

Seaweeds are a lot of bio-compounds and others chemical sources that can be used in industry, and in the last years came gained attention, mainly in areas of food, drug, agriculture, aquaculture, bio-energy, and degradable film and others products. They can be found in the oceans and fresh water as well that have light access. Macro-algae are eukaryotic, macroscopic multicellular algae commonly know by seaweeds. They need to be fixed on solids subtracts to live, can be rock, ship hulls, immersed woods, even pipes because they benthonic habits. They don't have a complexes structure at all, owing thallus, lamina, kelp, holdfast and frond sorus, thus morphologically differentiating it from the typical terrestrial plant, consisting of complex tissue and organ organization. (Milledge et al. 2014). Seaweeds are divided in three groups based on their most important pigments, are they: Chlorophyceae (green algae, with chlorophylls a and b), Phaeophyceae (brown algae, with chlorophylls a and c) and Rhodophyceae (red algae, with chlorophylls a and d). Green algae can absorb a huge amount of light energy, while red and brown algae cannot as they live in deeper waters where there is insufficient sunlight. Brown algae account for approximately 59% of the total macroalgae cultivated in the world, followed by red algae at 40% and green algae at less than 1% (Christaki et al., 2012). The purpose of this review article is to show some products extracted from macroalgae, such as fertilizers, drugs, drugs, pigments and biofuels.

Keywords: Seaweeds, Bio-fuel, Bio-fertilizer, Pigments, Pigments, Drugs

ORAL PRESENTATION

Investigation of antibiotic resistance and transferable resistance gene profiles of *Stenotrophomonas maltophilia* strains isolated from ready-to-eat foods

Tolga UYANIK[1*] (*ORCID: https://orcid.org/0000-0002-3181-3878*)

[1]Ondokuz Mayis University, Faculty of Veterinary Medicine, Department of Food Hygiene and Technology, Samsun, Turkey.

*Corresponding author e-mail: tolga.uyanik@omu.edu.tr

Abstract

Stenotrophomonas maltophilia is an important opportunistic pathogen that poses a considerable threat to susceptible patients. This microorganism is reported be intrinsically resistant to most of the antibiotics and can be found in multiple sources, including water, plant root environments, animals, and various food products. This study was carried out to determine the resistance status of *Stenotrophomonas maltophilia* strains against various antibiotics and to investigate transferable plasmid resistance genes. Four *S. maltophilia* strains isolated from ready-to-eat sandwiches were used as material. In accordance with CLSI instructions, in order to determine the antibiotic resistance profiles of the isolates, disc diffusion method was used to investigate trimethoprim-sulfamethoxazole and levofloxacin resistance, and E-test method was used to investigate ceftazidime and chloramphenicol resistance. Presence of transferable antibiotic resistance genes, including $bla_{\text{CTX-M}}$, bla_{TEM}, bla_{SHV}, bla_{KPC}, $bla_{\text{OXA-48}}$, bla_{NDM}, bla_{VIM} and bla_{IMP} were investigated by polymerase chain reaction. According to the results all isolates were found resistant to ceftazidime. All isolates were susceptible to trimethoprim-sulfamethoxazole, levofloxacin and chloramphenicol. Based on PCR results, only one of the four analyzed isolates was found $bla_{\text{CTX-M}}$ positive. All remaining isolates were found as negative for the analyzed resistance genes. In the future stages of the study, it is aimed to investigate cefiderocol and minocycline resistance and to determine the presence of chromosomal genes responsible for beta lactam resistance.

Keywords: *Stenotrophomonas maltophilia*, Antibiotic resistance, Ready to eat, Food, PCR

DRAFT

ORAL PRESENTATION

Production of urease enzyme by *Proteus app.* immobilized cryogel

Kıvılcım Çaktü Güler* (https://orcid.org/0000-0002-3096-1246)

Hacettepe University, Science Faculty, Department of Chemistry, Ankara, Turkey.

*Corresponding author e-mail: caktukivilcim@gmail.com

Abstract

Cryogels are gel matrices formed by freezing polymeric solutions. Cryogels prepared by the freeze-thaw method contain a network of interconnected pores ranging from 5-100 μm. They also have high flexibility and spongy morphology. Urease enzyme is used as a model enzyme for the detection of heavy metal ions in food samples, industrial wastewater, groundwater and environmental heavy metal ions, as well as to prevent the formation of ethyl carbamate by removing urea from fruit juices, food and wine. In this study, *Proteus app.* immobilized Poly(2-hydroxyethyl methacrylate) cryogel was synthesized as solid support. The characterization of cryogel was confirmed by swelling test and scanning electron microscopy (SEM). Besides, urease production of Proteus was determined by quantitative analysis based on color change in the medium.

Keywords: *Proteus app.*, urease, cryogel.

DRAFT

ORAL PRESENTATION

In silico identification of potential miRNAs that target *Avr* genes in *Fusarium oxysporum* f. sp. *lycopersici*

Aydin Atakan (*https://orcid.org*/0000-0001-9794-4427)

Department of Plant and Animal Production, Araban Vocational School, Gaziantep University, Turkey

*Corresponding author e-mail: aydinatakan@gantep.edu.tr

Abstract

Tomato (*Solanum lycopersicum* L.) is one of the most cultivated and consumed plants in the world. It is exposed to many biotic stress factors, especially fungi. Tomato wilt caused by *Fusarium oxysporum* f. sp. *lycopersici* (FOL) is one the most important diseases in tomato cultivation and causes crucial yield and quality losses. miRNAs are small non-coding molecules and play also important roles in plant immunity against plant pathogens. miRNAs mediate the regulation of the expression of defense response genes by mRNA cleavage and translational inhibition. In this study, previously identified mature miRNA sequences and related *Avr* genes of FOL were retrieved from miRBase and NCBI, respectively. RNAhybrid and psRNATarget algorithms were used to predict sly-miRNAs and *Avr* targets relationships. As a result of target identification, 13 miRNAs (miR477-5p, 9471b-3p, 9469-3p, 6022, 169e-3p, 9471a-5p, 164a-5p, 164b-5p, 5303, 319a, 395a, 395b, 482c) that target *Avr1* and *Avr3* genes of FOL were determined. Their inhibition types were translation and cleavage or both. Furthermore, a phylogenetic tree was constructed by using MEGA X. The neighbour-joining (NJ) method was operated with p-distance model. It was observed that all miRNAs were divided into two main groups. Only translation, only cleavage and even both of them showed very similar homology. Because of their importance in plant-pathogen interactions, miRNAs offer new opinions on the cross-kingdom immune regulation in plant-pathogen interactions.

Keywords: non-coding RNA, disease control, plant immunity

DRAFT

ORAL PRESENTATION

Are nanoparticle-hydrogel combinations the next generation of control strategies against plant diseases?

Mehmet Mensur Karpuz[1] (*https://orcid.org/0000-0003-4853-7950*), Aydin Atakan[2*] (*https://orcid.org/*0000-0001-9794-4427), Sevgi Marakli[1] (*https://orcid.org/*0000-0001-5796-7819)

[1]Department of Molecular Biology and Genetics, Faculty of Arts and Sciences, Yildiz Technical University, Turkey

[2]Department of Plant and Animal Production, Araban Vocational School, Gaziantep University, Turkey

*Corresponding author e-mail: mensur.karpuz@std.yildiz.edu.tr

Abstract

Hydrogels are defined as hydrophilic and water-insoluble polymeric macromolecules used in agriculture for various purposes such as reducing stress and increasing crop yield [1]. Nanoparticles (NPs) are bio-permanently small particles in the nanoscale (1-100 nm in size) and they have gained interest as alternatives in agricultural practices due to their characteristics [2, 3]. Plant pathogens cause crucial yield and quality losses in economically important crop plants. Therefore, they are a constant threat to agricultural industry [4]. To prevent negative effects of pathogens, pesticides have been widely used but excessive utilisation of them also cause negative environmental impact on animal, human and even all ecosystem. For this reason, eco-friendly control methods have gained importance in control of plant diseases. The combination of NPs and hydrogel can offer a promising alternative tool with their antibacterial and antifungal effects [5]. For this purpose, Shang et al. [5] studied the antifungal efficacy of copper oxide (CuO) embedded hydrogels against *Fusarium* wilt of lettuce. Results showed that a combination of NP and CuO NP-embedded hydrogels increased P, Mn, Zn and Mg uptake and organic acid levels compared to control. Abdelaziz et al. [6] investigated the positive effects of an eco-friendly superabsorbent hydrogel based on zinc oxide nanoparticles (ZnO-NPs) to protect the pepper plants against *Fusarium* wilt. Moreover, irrigation water usage for these pepper plants decreased by one-third after application. In addition, these materials can be successfully used as biotechnological tools in sustainable agriculture. de Oliveira et al. [7] synthesised hydrogel-based repellent systems containing botanical compounds emulsified or encapsulated in zein (Zein is a type of prolamin protein) nanoparticles. The synthesised hydrogels were reported to exhibit high repellent activity against whiteflies and spider mites, which are major agricultural pests [9]. As a result, hydrogels show promise not only for plant diseases but also in agricultural technology with their effects such as water retention capacities, water release control, soil aeration and plant development.

Keywords: plant disease, eco-friendly control method, biotechnological tool, sustainable agriculture

[1] Sahmat, S. S., Rafii, M. Y., Oladosu, Y., Jusoh, M., Hakiman, M., & Mohidin, H. (2022). A systematic review of the potential of a dynamic hydrogel as a substrate for sustainable agriculture. Horticulturae, 8(11), 1026.
[2] Kurczyńska, E., Godel-Jędrychowska, K., Sala, K., & Milewska-Hendel, A. (2021). Nanoparticles—plant interaction: what we know, where we are?. Applied Sciences, 11(12), 5473.
[3] Farooq, M. A., Hannan, F., Islam, F., Ayyaz, A., Zhang, N., Chen, W., ... & Zhou, W. (2022). The potential of nanomaterials for sustainable modern agriculture: present findings and future perspectives. Environmental Science: Nano, 9(6), 1926-1951.
[4] Venbrux, M., Crauwels, S., & Rediers, H. (2023). Current and emerging trends in techniques for plant pathogen detection. Frontiers in Plant Science, 14, 1120968.
[5] Shang, H., Ma, C., Li, C., Zhao, J., Elmer, W., White, J. C., & Xing, B. (2021). Copper oxide nanoparticle-embedded hydrogels enhance nutrient supply and growth of lettuce (*Lactuca sativa*) infected with *Fusarium oxysporum* f. sp. *lactucae*. Environmental science & technology, 55(20), 13432-13442.
[6] Abdelaziz, A. M., Dacrory, S., Hashem, A. H., Attia, M. S., Hasanin, M., Fouda, H. M., ... & ElSaied, H. (2021). Protective role of zinc oxide nanoparticles based hydrogel against wilt disease of pepper plant. Biocatalysis and Agricultural Biotechnology, 35, 102083.
[7] de Oliveira, J. L., Campos, E. V., Camara, M. C., Della Vechia, J. F., de Matos, S. T. S., de Andrade, D. J., ... & Fraceto, L. F. (2019). Hydrogels containing botanical repellents encapsulated in zein nanoparticles for crop protection. ACS Applied Nano Materials, 3(1), 207-217.

DRAFT

ORAL PRESENTATION

Removal of Nitrate (NO_3^-) from Aqueous Solution by Shallow Shell resin

Elif Çendik (https://orcid.org/0009-0004-2848-8973), Mügenur Saygı (https://orcid.org/0009-0002-3585-7429),
Özgür Arar (https://orcid.org/0000-0002-3687-9534) *

*Ege University, Faculty of Science, Chemistry Department, Izmir, Türkiye

*Corresponding author e-mail: ozgur.arar@ege.edu.tr.

Abstract

Nitrate, the conjugate base of nitric acid, is inherently neutral in aqueous solution. However, nitrate pollution is a pervasive issue in both surface and groundwater across our country. Its primary sources can be attributed to (1) uncontrolled fertilization of agricultural lands, (2) the oxidation of ammonia resulting from the decomposition of proteins in plant and animal waste, and (3) the discharge of untreated domestic and industrial wastewater. The proliferation of these compounds in our water bodies contributes to significant environmental problems, including eutrophication, algal blooms, and adverse health effects [1,2].
This study investigated the removal of NO_3^- from aqueous solutions using a strongly basic anion exchange resin of Shallow Shell Technology (SST). The influence of resin dosage and solution pH on removing NO_3^- by the strong basic anion exchange resin was examined. Furthermore, modeling sorption isotherms and conducting kinetic studies determined the NO_3^- removal mechanism and the rate-determining step. Sorption experiments were conducted at various temperatures, ranging from 15°C to 30°C, and thermodynamic analysis was performed. The NO_3^- analysis was carried out using a UV-visible spectrometer.

Keywords: Anion exchange resin, Separation Techniques, Ion exchange, Water treatment

Acknowledgments
This study was supported by the Scientific and Technological Research Council of Turkey (TUBITAK program code: 2209 A).

References
[1] Yücel, A., Uçar, D., & Demir, Ö. Yeraltı sularından biyolojik nitrat giderimde kullanılan reaktör tipleri. Dicle Üniversitesi Fen Bilimleri Enstitüsü Dergisi, 8(3), 77-83.
[2] Kaçmazoğlu, E. (2021). Kalsiyum Aljinatta Tutuklanmış Paracoccus denitrificans ile Sabit Yatak Biyoreaktörde İçme Sularından Nitrat Giderimi. Journal of the Institute of Science and Technology, 11(4), 2632-2644.

DRAFT

ORAL PRESENTATION

Evaluation of Eber Lake (Afyonkarahisar) Macrophytes in Terms of Restoration Ecology

Mustafa Aljumaili (*ORCID: 0000-0003-1167-1582*), Ahmet Serteser * (*ORCID: 0000-0003-2792-7239*

Afyon kocatepe Üniversitesi, Fen Edebyat Fakültesi, Moleküler Biyoloji ve Genetik, Afyonkarahisar, Türkiye.

*Corresponding author e-mail: aserteser@aku.edu.tr

Abstract

In this research, some physical and chemical parameters of the soil and water properties of Lake Eber and the *Phragmites australis* species in Lake Eber were evaluated in terms of restoration ecology. In this context, physical and chemical analyzes of soil samples taken from 5 different locations on the shore of Eber Lake were made. The average pH value of the soil samples was determined as 7.54, the salt (%) value was 0.53, the lime value was 23.94%, the organic matter % value was 16.21, and the phosphorus value was 28.72, and the potassium value was 314.6. According to this, it was determined that the soil structure of Eber Lake is heavy clay, salty, and calcareous, with a very high value in terms of organic matter, phosphorus, and potassium. Average pH value of water samples taken from 4 different locations of the lake is 8.63, sodium value is 33 me/l, potassium value is 2.5 me/l, boron value is 0.6815 me/l, Ca+ Mg value is 5.1 me/l, carbonate value is 2.34m e/l, bicarbonate value is 8.57 me/l, chlorine value is 14.17 me/l, sulfate value is 15.55 me/l, RSC value is 5.77 me/l, SAR value is 20.8 me/l. Eber Lake water is alkaline and has good electrical conductivity, sulfate, chlorine, and RSC value sare very high within the scope of analysis. On the other hand, SAR, boron, sodium, potassium, calcium, and magnesium parameters were recorded within Standard criteria. With the current data obtained, contributions have been made to the identification and protection of the bioecological features of Lake Eber.

Keywords: Eber Lake, Ecological Restoration, Macrophyte, *Phragmites australis*.

DRAFT

ORAL PRESENTATION

Degradation and mineralization of methyl orange by advanced oxidation processes

Baran Özyurt* (ORCID: https://orcid.org/0000-0003-0011-8351)

*[1]Ankara University, Faculty of Engineering, Department of Chemical Engineering, Ankara, Turkey.

*Corresponding author e-mail: bozyurt@ankara.edu.tr

Abstract

Methyl orange (MO) is a kind of synthetic azo dye which is not only widely utilized in the printing and dyeing process of textile, but also used as colorant in plastics, rubber and paint, etc. Due to low reactivity and biodegradation, this type of compound is resistant to degradation in wastewater treatment. Under certain special conditions, azo dyes are able of decomposing to produce more than 20 kinds of carcinogenic aromatic amines, which will change the DNA structures of the human body through the activation inducing lesions and cancer. Advanced oxidation processes (AOPs) for the removal of these pollutants from water became important technologies today. AOPs are carried out on the basis of forming hydroxyl radicals ($^{\bullet}OH$), which can oxidize many pollutants in water, and the process efficiency is evaluated depending on the formation of $^{\bullet}OH$. Various AOP mechanisms are classified depending on whether $^{\bullet}OH$ formation occurs chemically, photochemically, sonochemically or electrochemically. In the present work degradation and mineralization of synthetic dye MO by means of various AOPs including, homogeneous Fenton (F), homogeneous photo-Fenton (PF), homogeneous electro-Fenton (EF), homogeneous photoelectro-Fenton (PEF), heterogeneous photoelectro-Fenton (hPEF), UV photodegradation (PD), TiO_2/UV photocatalytic degradation (PC), TiO_2/UV/H_2O_2 photoelectrocatalytic process (PEC) were investigated. Fe_{2+} was used as catalyst in F, PF, EF and PEF processes, whereas Fe_3O_4 magnetic nanoparticles synthesized by co-precipitation method was used in hPEF process. Anatase phase of TiO_2 used in PC and PEC processes was synthesized by means of a hydrothermal process, where hydrolysis of titanium butoxide was took place. Platinum anode and a carbon felt (CF) cathode was used in EF, PEF, hPEF and PEC processes. Complete degradation of MO was achieved at 5, 7, 20, 24, 30, 36 mins by PF, F, PEF, EF, hPEF and PEC processes, respectively under pH=3, 6W UVA light and 100 mA current intensity conditions.

Keywords: advanced oxidation processes, environmental remediation, catalysis

DRAFT

ORAL PRESENTATION

Effect of the cathode material on the efficiency of the electro-Fenton process for the electrocatalytic destruction of azo dye methyl orange

Şule Camcıoğlu[1*] (*ORCID: https://orcid.org/0000-0002-8983-3193*)

[*1] Ankara University, Faculty of Engineering, Department of Chemical Engineering, Ankara, Turkey.

*Corresponding author e-mail: camcioglu@eng.ankara.edu.tr

Abstract

A significant amount of micropollutants from various chemical industries are continuously released into the natural water bodies, where they accumulate due to their resistance to biodegradation. Nowadays, they are detected in natural water streams, even frequently in drinking water sources. These pollutants are toxic to aquatic life as well as to humans. Conventional water treatment plants remain generally inefficient in treatment of water polluted with persistent and/or toxic organic pollutants. On the other hand, advanced oxidation processes (AOPs) which are effective water treatment technologies, were developed. Based on in-situ generation of strong oxidants such as hydroxyl radicals (•OH) with very high oxidation power and reactivity, AOPs are able to oxidize organic micropollutants until mineralization. Among them, electrochemical AOPs (EAOPs) were revealed as versatile, clean and efficient processes for degradation/mineralization of organic pollutants. The electro-Fenton process is one of the most popular EAOPs that generates H_2O_2 in situ from two-electron reduction of dissolved O_2 on a suitable cathode. •OH produced through electrocatalytically generated Fenton's reagent (H_2O_2+Fe^{2+}) in the bulk react quickly with organic pollutants. Therefore, cathode material plays a crucial role for the efficiency of electro-Fenton process since it determines the yield of H_2O_2 which controls the production rate of •OH. For this purpose, this study focuses on the comparative efficiency of cathode materials (i.e., stainless steel, carbon felt and graphite) based on the oxidative degradation and mineralization of azo dye methyl orange as the target pollutant. Performance was compared based on the following parameters: H_2O_2 generation ability, capacity of oxidation and mineralization, mineralization current efficiency and energy consumption as a function of applied current. According to the results, carbon felt showed better oxidation and mineralization efficiency, followed by graphite. Stainless steel cathode was an inappropriate material for electro-Fenton process due to its poor H_2O_2 generation ability.

Keywords: Electrochemical advanced oxidation processes, electro-Fenton, mineralization, decay kinetics, methyl orange

DRAFT

ORAL PRESENTATION

Akdeniz'de Nesli Tükenme Tehlikesiyle Karşı Karşıya Olan *Pinna nobilis*'in Korunmasına ve Rehabilitasyonuna Yönelik Yapılan Uygulamalar

Sefa ACARLI[1*] (0000-0002-5891-5938), Harun YILDIZ[1] (0000-0002-8619-7814),
Deniz ACARLI[2] (0000-0001-8537-9969)

[*1]Çanakkale Onsekiz Mart Üniversitesi, Deniz Bilimleri ve Teknolojisi Fakültesi, Çanakkale, Türkiye
[2]Çanakkale Onsekiz Mart Üniversitesi, Deniz Teknolojileri Meslek Yüksekokulu, Çanakkale, Türkiye

* e-mail: sefaacarli@comu.edu.tr

Özet

Akdeniz'e özgü bir tür olan *Pinna nobilis* (Linnaeus, 1758) deniz suyunu süzme kapasitesi ve bir çok epibiyotik türe sert kabuk yapısından dolayı ev sahipliği yaptığı için önemli bir ekolojik role sahiptir. Ancak insan aktivitelerinin sonucunda uzun yıllardan beri *P. nobilis* popülasyonları zarar görmüş ve sonuç olarak 1992 yılında itibaren Avrupa Konseyi kararları ile kırmızı listeye alınarak koruma altına alınmıştır. 2016 yılından bu zaman kadar ise, Batı Akdeniz den başlayarak *P. nobilis* popülasyonlarında %100 ölüm rapor edilmiştir. Bu kitlesel ölümlere patojenik parazit olan *Haplosporidium pinnae*'nin ve diğer potansiyel patojenlerin birlikte etkili olduğu söylenilmektedir. IUCN, *P. nobilis* türünün statütüsünü karşılaşılan kitlesel ölümlerden dolayı kırmızı listede "*kritik seviye*" ye yükseltti. 2020 yılından itibaren ise Ege Denizi, Çanakkale Boğazı ve Marmara Denizi'nde farklı noktalarda toplu ölümlerin rapor edilmiştir. Aynı zamanda sınırlıda olsa Akdeniz'in farklı noktalarında ve Çanakkale Boğazı ile birlikte Marmara Denizinde sağlıklı popülasyonların bildirilmesi oldukça umut vericidir. Bu bağlamda hastalığa maruz kalmasına rağmen hayatta kalan dirençli bireylerin ve sağlıklı olan popülasyonların belirlenmesi, sürekli izleme çalışmalarının yapılması ve özel koruma alanlarının oluşturulması, hasarlı *P. nobilis* yataklarının rehabilitasyonuna katkı sağlayacaktır. Tüm bu yapılan çalışmalara ilaveten yapılacak olan yetiştiricilik çalışmaları yani larva üretimi, doğadan toplanan spatın büyütmeye alınması ve transplantasyon çalışmaları da oldukça önemlidir. Sonuç olarak bu çalışmada Akdeniz'de türün sürdürülebilir üretimine ve korunmasına yönelik yapılan uygulamaların genel bir değerlendirilmesi yapılmıştır.

Anahtar Kelimeler: *Pinna nobilis*, yetiştiricilik, transplantasyon, hayatta kalma

DRAFT

ORAL PRESENTATION

Adölesanların hedonik açlık durumunun uyku kalitesi, diyet kalitesi ve beslenme durumu ile ilişkisinin değerlendirilmesi

Gamze Yurtdaş Depboylu[1*] (*ORCID: https://orcid.org/0000-0001-5410-7231*), Başak Şimşek[1] (*ORCID: https://orcid.org/0009-0002-0452-2378*)

[*1]İzmir Katip Çelebi University, Faculty, Department of Nutrition and Dietetics, İzmir, Turkiye.

*Corresponding author e-mail: gmzyurtdas@hotmail.com

Abstract

Hedonik açlık fizyolojik açlığın yokluğunda yemeğe karşı aşırı duyarlılık, yemekten zevk alma ve yeme dürtüsü ile karakterize olan durumdur. Bu çalışmada adolesanlarda hedonik açlık durumunun diyet kalitesi, uyku kalitesi ve beslenme durumu ile ilişkisini değerlendirmek amaçlanmıştır. Bu çalışmada İzmir merkezde bulunan 3 ortaokulda gerçekleştirilmiştir. Araştırma verileri anket kullanılarak sınıf ortamında yüz yüze görüşme yöntemiyle elde edilmiştir. Adolesanların hedonik açlık durumu Besin Gücü Ölçeği (BGÖ) ile, uyku kalitesi ise Pittsburgh Uyku Kalitesi İndeksi (PUKİ) ile değerlendirilmiştir. Diyet kalitesi akdeniz Diyeti Kalite İndeksi (KIDMED) kullanılarak değerlendirilmiştir. Adölesanların hedonik açlık ile çeşitli besinleri tüketmeye olan arzularını değerlendirmek amacıyla görsel analog skalası kullanılmıştır. Beslenme durumunu değerlendirmek için antropometrik ölçümler (boy uzunluğu, bel çevresi, kalça çevresi, üst orta kol çevresi, boyun çevresi) araştırmacılar tarafından uygun yöntemlerle alınmıştır. Besin alımını değerlendirmek için bir günlük besin tüketim kaydı alınmıştır. Çalışmaya katılan çocukların yaş ortalamaları 12,2∓1,19 idi. Kız ve erkek öğrencilerin BGÖ ortalama toplam puanları sırasıyla 3,16±0,80 ve 2,83±0,78 puan idi (p=0,258). PUKİ kesim noktasına göre değerlendirildiğinde adölesanların %58,9'unun uyku kalitesi iyi, %71,1'inin uyku kalitesinin kötü olduğu belirlendi. Hedonik açlığı olan adolesanların çikolata, kek, cips, gazlı içecekler, fast food, patates kızartması, makarna, dondurma, hamur işleri, kremalı pasta ve pastane ürünleri tüketme isteklerinin hedonik açlığı olmayanlara göre yüksek olduğu belirlendi (p<0,05). Adolesanların hedonik açlık durumları göre antropometrik ölçümleri arasında anlamlı fark bulunmadı (p>0,05). BGÖ skoru PUKİ skoru (r=0,135, p<0,001) ile pozitif yönlü, KIDMED skoru ile (r=-0,224 p=0,001) negatif yönlü korele idi. Bu sonuçlar adolesanlarda hedonik açlık durumunun diyet ve uyku kalitesi ile ilişkili olduğunu göstermiştir. Bu bağlamda adolesanların hedonik açlık hakkında eğitilmesi önemlidir.

Keywords: hedonik açlık, adolesan, uyku kalitesi, diyet kalitesi, beslenme durumu

DRAFT

ORAL PRESENTATION

Investigation of individual variation in *Macrovipera lebetinus* venom by Fourier transform infrared (FTIR) spectroscopy

Naşit İĞCİ[1*] (*ORCID: https://orcid.org/0000-0001-6151-808X*)

[*1] Nevşehir Hacı Bektaş Veli University, Faculty of Science and Arts, Department of Molecular Biology and Genetics, Nevşehir, Türkiye.

*Corresponding author e-mail: igcinasit@yahoo.com.tr

Abstract

Snake venom is a molecular cocktail that has diverse biological activities. Its main constituent is polypeptides (eg. proteins, peptides). These proteins and peptides are responsible for the toxicity of venom in the prey. Snake venom is also an important source for drug discovery. Although venoms of different species share common protein families, their abundancy in venoms show variation. Moreover, different species may have distinct proteoforms. Venom variation is important topic since this phenomenon affects the pathology of snakebite and antivenom production, as well. Fourier transform infrared spectroscopy (FTIR) is a useful analytical technique that provides qualitative and quantitative information about the bioorganic molecules in the sample. Studies regarding the venom variation of *Macrovipera lebetinus*, a medicinally important viper species, are limited in the literature. In the present study, venom variation in *M. lebetinus* was investigated using FTIR. Crude venom samples from different individuals were directly measured using attenuated total reflectance method in the mid-infrared region. Obtained absorbance spectra were analysed in the fingerprint region between 800-1800 cm-1 and peaks were assigned to proteins (mainly), carbohydrates and nucleic acids. Protein secondary structures were also compared using the sub-bands of Amide I band. Qualitative and quantitative differences were observed both in the absorbance spectra and Amide I sub-bands obtained by derivatization. The results revealed that *M. lebetinus* venom shows individual variation at molecular level and FTIR is a useful technique for venom research.

Keywords: FTIR, snake venom, variation.

DRAFT

ORAL PRESENTATION

Akuakültür Yoluyla Akdeniz Midyesi (Mytilus galloprovincalis Lamarck, 1819) Üretiminin Sağlık Açısından Önemi

Harun YILDIZ[1*] (0000-0002-8619-7814), Sefa ACARLI[1] (0000-0002-5891-5938),
Pervin VURAL[2] (0000-0002-1810-1112)

[*1]Çanakkale Onsekiz Mart Üniversitesi, Deniz Bilimleri ve Teknolojisi Fakültesi, Çanakkale, Türkiye

[2]Çanakkale Onsekiz Mart Üniversitesi, Bayramiç Meslek Yüksekokulu, Çanakkale, Türkiye

* e-mail: harunyildiz@comu.edu.tr

Özet

Akdeniz Midyeleri (*Mytilus galloprovincialis*, Lamarck 1819); yüksek protein değerleri, düşük yağ içerikleri, kolay sindirilebilme ve dietetik özellikleriyle değerli besin maddeleri olarak karşımıza çıkmaktadır. Fakat; sulardaki primer prodüktiviteyi süzerek beslenme (filter-feeding) özellikleri, denizlerdeki kirleticilerle kontamine olma risklerini de beraberinde getirmektedir. Dolayısıyla; toksikolojik, biyotoksikolojik ve mikrobiyolojik açıdan takip edilmeyen bölgelerden toplanan Akdeniz midyeleri, birçok sağlık sorununun da sebebi olabilmektedir. Örneğin, evsel atıkların boşaltıldığı bölgelerden toplanan midyeler tüketildiğinde; bağırsak enfesiyonlarına sebep olan *Escherichia coli*, campylobacter ve salmonella gibi bakteriler ile rotavirüs ve norovirüs gibi virüsler insanlara bulaşabilir. Özellikle endüstriyel atıkların kirlettiği bölgelerden elde edilen midyelerin tüketimi neticesinde manganez, nikel kurşun, kalay ve alüminyum gibi ağır metaller vücuda alındığında; alzheimer, kısırlık, böbrek ve karaciğer yetmezliği gibi önemli hastalıklar görülebilir. Yine DSP (Diarrhetic shellfish poisoning), ASP (Amnesic shellfish poisoning) ve PSP (Paralytic shellfish poisoning) gibi biyotoksinleri akümüle eden midyelerin tüketilmesi; bulantı, kusma, baş dönmesi, hafıza kaybı, solunum yetmezliği, midye tümörleri ve daha birçok enfeksiyona sebep olabilmektedir. Akdeniz midyelerin, bu parametreler açısından insan sağlığını tehdit etmeyecek ve tolere edilebilir limitlere sahip olması gerekmektedir. Bütün veriler bir arada değerlendirildiğinde, tüketim için en uygun ürünler kültür yoluyla elde edilen midyedir. Burada ki en önemli unsur, Akdeniz midyesi yetiştiricilik tesislerinin kurulumundan itibaren ilgili kurumlar tarafından sürekli denetim altında olmasıdır. Bu çalışmanın amacı, yetiştiricilik yoluyla elde edilen Akdeniz midyesinin üretimi ve ürün kalitesini etkileyen unsurlar değerlendirilerek insan sağlığı açısından önemini ortaya koymaktır.

Anahtar Kelimeler: *Mytilus galloprovincialis,* Yetiştiricilik, filter-feeding, tüketim

DRAFT

ORAL PRESENTATION

Synthesis, Characterization and Antioxidant Activities of Ni(II) and Cu(II) Complexes of Salicylaldehyde-N(4)-phenyl Thiosemicarbazone

Elif AVCU ALTIPARMAK[1] (*https://orcid.org/0000-0002-5491-8445*), Namık ÖZDEMİR[2] (https://orcid.org/0000-0003-3371-9874), Tülay BAL DEMİRCİ[1*](https://orcid.org/0000-0003-4663-2209)

[*1]Istanbul University-Cerrahpasa, Faculty of Engineering, Department of Chemistry, Istanbul, Turkey.
[2] Ondokuz Mayıs University, Faculty of Education, Department of Mathematics and Science Education, Samsun, Turkey.

*Corresponding author e-mail: tulaybal@iuc.edu.tr

Abstract

Thiosemicarbazones are compounds obtained by the reaction of a thiosemicarbazide with carbonyl group compounds and can coordinate to metal atoms via more than one donor atom. Thiosemicarbazones and their metal complexes exhibit a wide range of biological activities such as anticancer [1], antiviral [2], antibacterial [3], antidiabetic [4], antioxidant [5], antimalerial [6], anti-HIV [7] and have capabilities of inhibition of enzyme [8] and binding to DNA [9].
In this work, mixed-ligand Ni(II) **(I)** and Cu(II) **(II)** complexes of salicylaldehyde-N(4)-phenyl thiosemicarbazone ligand were synthesized by using 3,5-lutidine as a secondary ligand. The structures of ligand and complexes were characterized by elemental analyses, IR, UV-Vis, ^{1}H-NMR, ESI-MS spectroscopies, and also the complexes by single crystal X-ray diffraction. The antioxidant activities of the compounds were investigated by CUPRAC and DPPH assays. Both the ligand and the complexes were found to have antioxidant capacity.

Keywords: Thiosemicarbazone, Transition Metal Complexes, Antioxidant Activity, CUPRAC, DPPH.

References:
[1] Hu, W., Zhou, W., Xia, C., Wen, X., 2006, Synthesis and anticancer activity of thiosemicarbazones, Bioorganic & Medicinal Chemistry Letters, 16, 8, 2213-2218.
[2] West, D., El-Sawaf, A., Bain, G., 1998, Metal complexes of N(4)-substituted analogues of the antiviral drug methisazone {1-methylisatin thiosemicarbazone}, Transition Metal Chemistry. 23 (1), 1–6.
[3] Khan, S.A., Yusuf, M., 2009, Synthesis, spectral studies and in vitro antibacterial activity of steroidal thiosemicarbazone and their palladium (Pd (II)) complexes, European Journal of Medicinal Chemistry, 44(5), 2270-2274.
[4] Yanardag, R., Bal Demirci, T., Ülküseven, B., Bolkent, S., Tunali, S., Bolkent, Ş., 2009, Synthesis, characterization and antidiabetic properties of N1-2, 4-dihydroxybenzylidene-N4-2-hydroxybenzylidene-S- methyl-thiosemicarbazidato-oxovanadium (IV), European Journal of Medicinal Chemistry, 44, 2, 818-826.
[5] Bal-Demirci, T., Güveli, Ş., Yeşilyurt, S., Özdemir, N., Ülküseven, B., 2020, Thiosemicarbazone ligand, nickel(II) and ruthenium(II) complexes based on vitamin B6 vitamer: The synthesis, different coordination behaviors and antioxidant activities, Inorganica Chimica Acta, 502, 119335.
[6] Scovill, J.P., Klayman, D.L., Franchino C.F., 1982, 2-Acetylpyridine Thiosemicarbazones. 4. Complexes with Transition Metals as Antimalarial and Antileukemic Agents, Journal of Medicinal Chemistry, 25 (10), 1261-1264.
[7] Bal, T.R., Anand, B., Yogeeswari, P., Sriram, D., 2005, Synthesis and evaluation of anti-HIV activity of isatin β-thiosemicarbazone derivatives, Bioorganic & Medicinal Chemistry Letters, 15 (20), 4451-4455.
[8] Pervez, H., Chohan, Z. H., Ramzan, M., Nasım, F. H. ve Khan, K. M., 2009, Synthesis and biological evaluation of some new N4-substituted isatin-3-thiosemicarbazones, Journal of Enzyme Inhibition and Medicinal Chemistry, 24 (2), 437-446.
[9] Bal-Demirci, T., Congur, G., Erdem, A., Erdem-Kuruca, S., Özdemir, N., Akgün-Dar, K., Varol, B., Ülküseven, B., 2015, Iron(III) and nickel(II) complexes as potential anticancer agents: synthesis, physicochemical and structural properties, cytotoxic activity and DNA interactions, New Journal of Chemistry, 39, 5643-5653.

ORAL PRESENTATION

Prebiotics in Aquatic Animals

Semra KÜÇÜK (*ORCID:* https://orcid.org/0000-0002-9299-5986)

Adnan Menderes University, Faculty of Agriculture, Department of Aquacultural Engineering and Fisheries, Aydın, Turkey,

E-mail:skucuk@adu.edu.tr

Abstract

The global aquaculture industry has been growing fast. Almost 1 billion people have been working in this sector. Accordingly FAO 2020, aquaculture production has formed 46% of the total world fish production. The largest aquatic animal producer is the China. And current aquaculture practices have been carrying on sustainable and environment friendly and produced safe products for the end user.
Prebiotics are long chain complex carbohydrates that give energy to beneficial microorganisms (probiotics) to increase an animal health. Prebiotics are mostly derived from plant based products (mushroom) and only a few gaining from animal dairy products. Prebiotics are actually found in vegetables, fruits, seaweed, microalgae and milk in natural. Prebiotics can protect epithelial cells against to harmful microorganisms and decrease harmful microorganisms from intestinal cells as a receptor or modulator in the host immune system. They can improve growth, enhance immune response and disease resistance and decrease stressor effects in aquaculture. There have been some studies about using prebiotics in aquatic animals. In this study, it is going to explain the definition, mechanisms and function of prebiotics and indicate the previous studies having been done on the prebiotics in aquatic animals.

Keywords: Probiotics, prebiotics, aquatic animals

DRAFT

ORAL PRESENTATION

Herbal Anaesthetics in Aquaculture

Semra KÜÇÜK (*ORCID:* https://orcid.org/0000-0002-9299-5986)

Adnan Menderes University, Faculty of Agriculture, Department of Aquacultural Engineering and Fisheries, Aydın, Turkey.

E-mail:skucuk@adu.edu.tr

Abstract

For prevention of physical injuries and easy working during applications in fish farming or for research purposes
Anesthetics substances are used for handling, artificial spawning, vaccination,sorting of specimens, taking biopsy, labeling, transportation blood collection, surgery, and for euthanasia. Some essential oils have been used for sedative and anaesthetic substances such as clove oil, lavanter oil, thyme oil. The most commonly used plants for anesthesia in aquaculture are basil, thyme, mint, rosemary, lavender, citronella, verbena and camphor.They have different active substances such as eugenol, menthol, myrcene, 1,8-cineole, linalool, limonene, citronellal, thymol, carvacrol, spathulenol, α- and β-pinene, 4-allylphenyl acetate and globulol in plants. They cause some effects on the modulation of fish behavior, physiology and biochemistry. The natural anesthetics could have potential to be more environmentally friendly, cost-effective and safer products than synthetic drugs (2-phenoxyethanol, tricaine methane-sulfonate-MS-222, benzocaine, metomidate, etomidate, quinaldine sulfate). In this study, it is going to give some information about herbal anaesthetics and explain some previous studies have been done about them in aquaculture.

Keywords: Herbal anaesthetics, synthetic anaesthetics, aquaculture

DRAFT

ORAL PRESENTATION

Biostimulant effect of brown algae extracts on soil grown broccoli

Yağmur ARIKAN[1,2*] (*ORCID: https://orcid.org/0000-0002-1252-2979*), Hande MUTLU DURAK[1] (*ORCID: https://orcid.org/0000-0003-4351-5795*), Bahar YILDIZ KUTMAN[1] (*ORCID: https://orcid.org/0000-0002-7891-7527*), Ümit Barış KUTMAN[1] (*ORCID: https://orcid.org/0000-0002-9158-0332)*

[*1] Gebze Technical University, Institute of Biotechnology, Kocaeli, 41400, Turkey
[2] GÜBRETAŞ R&D Center, Kocaeli, 41400, Turkey

*yarikan2018@gtu.edu.tr

Abstract

Macroalgae are the main class of biostimulants because of the benefits of their numerous bioactive components that encourage plant development without compromising crop quality, even with small amounts. Although the brown seaweed *Cystoseira barbata*, which is common in the seas of Turkey, has a significant potential for stimulating plant development and growth, it has never been applied as a biostimulant. The high-value crop broccoli contains an array of phytochemicals and mineral minerals that support human health, so it is important to drive production toward increased organoleptic quality. The purpose of the study is to determine whether the *C. barbata* extracts, which were made as three distinct extracts and applied by irrigation at two different concentrations, may improve the growth characteristics of broccoli (*Brassica oleracea* L. cv. Maraton) grown in soil. The plants were harvested after 100 days, and their growth features were evaluated. Chlorophyll, flavonoids, and anthocyanin levels in the plants' leaves were measured during their intermediate growth stage. To further understand the possible beneficial effects of biostimulants, experiments for the biochemical characterization of seaweed extracts were also conducted. The group treated with low concentrations of acid extraction showed increases of up to 55% in the dry weight of broccoli heads. It was also noted that the application of a high concentration of hot water extraction caused the plants' total dry weight to rise by nearly 43%. This research reveals the positive biostimulating effects of different *C. barbata* extracts on broccoli growing in a soil system. It is possible to say that various amounts of potentially bioactive components can affect biostimulants in multiple ways. This study will advance sustainable agricultural methods, support global food security, and reduce broccoli production-related economic losses. This study is supported by TÜBİTAK 2244 (119C030) collaborated with GUBRETAS.

Keywords: Broccoli, *Cystoseira barbata*, Plant Biostimulant, Seaweed Extract, Sustainable Agriculture

DRAFT

ORAL PRESENTATION

Membrane Cholesterol Affects P2X7-Mediated Permeability Pathways Differently

Serife CANKURTARAN-SAYAR[1*] (*ORCID: https://orcid.org/0000-0002-3757-8709*), Kemal SAYAR [2]

[*1]Yuksek Ihtisas University, Faculty of Medicine, Department of Biophysics, Ankara, TÜRKİYE
[2]Ankara University, Faculty of Medicine, Department of Medical Pharmacology, Ankara, TÜRKİYE

*Corresponding author e-mail: serifecankurtaransayar@gmail.com

Abstract

In this study, we planned to examine the effect of membrane cholesterol on anionic and cationic permeabilities mediated by the P2X7 receptor, which is a purinergic receptor. In this study, RAW 264.7 cells, a mouse-derived macrophage cell line, were used. These cells endogenously express the P2X7 receptor. We grew the cells in flasks or glass coverslips and tested them after 24 hours. Flowcytometer and Fluorescent Microscopy: To monitor membrane permeability, we stimulated the cells with the receptor agonist ATP (1 mM) in the medium containing the cationic YO-PRO1 or the anionic Lucifer Yellow fluorophores. After 20 min, we calculated the ratio of the number of cells uptaking fluorophore to the total number of cells. We considered this ratio as a measure of P2X7-mediated permeability. To test the role of membrane cholesterol, we incubated cells with the membrane cholesterol inhibitor methyl-β-cyclodextrin (MβCD, 5 mM) for 20 min and repeated the experiments. In this study, we observed that methyl-β-cyclodextrin, a membrane cholesterol inhibitor, did not affect the permeability rate of cationic YO-PRO-1 (MβCD: 98.5 and MβCD+: 2), but increased the permeability rate of anionic Lucifer Yellow (MβCD -: 41,8 and MβCD+ : 57.9). We also observed that the Lucifer Yellow fluorescence intensity increased. We observed that methyl-β-cyclodextrin did not affect P2X7-mediated cytoskeleton reorganization. Although membrane cholesterol inhibition does not affect the permeability of cations, it increases the permeability of anions. This finding is very important as the first in the literature. Investigating the roles of membrane cholesterol-rich lipid rafts in P2X7-mediated functional responses will help explain the signal transduction mechanism of the P2X7 receptor. Considering that membrane cholesterol changes membrane fluidity and that membrane fluidity is of vital importance for the cell, the importance of our findings will be better understood.

Keywords: Macrophage, P2X7, Fluorescence Microscopy, membrane cholesterol

DRAFT

ORAL PRESENTATION

EC Effect on Growth, Quality, and Nutrient Uptake of Hydroponic Lettuce

Ayşenur BAYRAK[*1] (*ORCID: https://orcid.org/0000-0002-2680-29040*),
Ümit Barış KUTMAN[1](*ORCID: https://orcid.org/0000-0002-9158-0332*)

Gebze Technical University, Institute of Biotechnology, Kocaeli, Turkey.

*abayrak2018@gtu.edu.tr

Abstract

Hydroponic systems are frequently preferred for commercial lettuce cultivation. In closed hydroponic production, ionic balance in the nutrient solution can be disturbed over time, which can cause yield loss, deficiency, or toxicity symptoms. The optimal EC (an indicator of the strength of the solution) can vary depending on environmental factors. In this study, the plants were grown in a greenhouse and a growth chamber to explore the impact of the growing environment on an EC characteristic. The treatments' ECs, which we designated EC1X, EC2X, and EC3X, were 0.8, 1.6, and 2.4 dS/m. Greenhouse plants exhibited higher fresh and dry weights than growth chamber plants. The highest fresh weight was found in EC3X greenhouse plants, while the lowest fresh weight was found in EC3X growth chamber plants. With rising EC levels, the total phenolic content increased and was higher in growth chamber plants. Statistically, EC3X-growth chamber plants had higher vitamin C content than all other applications. In all treatments, tissue nitrates were at a nutritionally safe level. The macroelements of EC1X solution were nearly depleted in a week while the EC2X and EC3X solutions still had enough nutrients to allow for a biweekly solution change. There was a correlation between these pH dynamics and ammonium consumption in the solutions. The rate of ammonium depletion differed in two different locations. The solution pH of EC3X-growth chamber plants was low enough to negatively impact plant root development. Results indicate that EC2X is suitable for lettuce cultivation in hydroponic conditions for both locations in terms of the providing optimum pH of the solution, the advantage it creates in the frequency of changing the nutrient solution, and the nutritional quality.

Keywords: EC, hydroponic, lettuce, nutritional quality, nutrient uptake

DRAFT

ORAL PRESENTATION

Kurna Köyü (Burdur) ve Çevresindeki Bazı Tıbbi Bitkiler

Veysi ERGÜL* (*https://orcid.org/0009-0002-8888-1270),* Neslihan BALPINAR (*https://orcid.org/0000-0002-4469-8629*), Ümit KEBAPÇI

Burdur Mehmet Akif Ersoy Üniversitesi, Fen Edebiyat Fakültesi, Biyoloji Bölümü, Burdur, Türkiye

*ergulveysi@gmail.com

Özet

Bitkiler, geçmişte olduğu gibi günümüzde de geleneksel tedavide ilaç olarak yararlanılan doğal kaynakların başında gelmektedir. Sentetik ilaçlarla kıyaslandığında birçok avantaja sahip olması nedeniyle, dünyada olduğu gibi ülkemizde de tıbbi bitkilere olan ilgi hızla artmaktadır. Bu çalışma Kurna Köyü'nde doğal olarak yetişen bazı tıbbi bitkileri belirlemek amacıyla yapılmıştır. Bitkilerin çiçeklenme dönemlerinde yapılan arazi çalışmaları sonucunda 21 familyaya ait 39 cins ve bu cinslere ait 40 tıbbi ve bitki taksonu tespit edilmiştir. Araştırma alanında tanımlanan familyalardan sırasıyla *Asteraceae* familyası 7 takson, *Lamiaceae* familyası 5 takson, *Brassicaceae* familyası 3 takson, *Boraginaceae* familyası 3 takson ve *Fabaceae* familyası 3 takson ile en çok takson sayısına sahip ilk beş familyayı oluşturmaktadır. Tıbbi bitkilerin bilimsel adları, yöresel adları, kullanılan kısımları ve kullanım alanları sunulmuştur. Ayrıca bu taksonların korunması, geliştirilmesi ve yaygınlaştırılarak insanlığın hizmetine sunulabilmesi konularında önerilerde bulunulmuştur. Böylece doğal kaynaklarımızın belirlenmesi, korunması ve sürdürülebilirliğine katkı sağlanacağı kanaatindeyiz.

Anahtar Kelimeler: Burdur, Kurna Köyü, Tıbbi Bitki

DRAFT

ORAL PRESENTATION

The New Trend in Nutrition: Edible Flowers

Ugur BAYRAM[1*] (*ORCID: https://orcid.org/0000-0003-2167-2933)*,
Ilkay KOCA[2] (*ORCID*: *https://orcid.org/0000-0001-6089-8586)*

[*1] Giresun University, Dereli Vocational School, Department of Hotel, Restaurant and Catering Services, Cooking Pr., Giresun, Turkey.
[2] Ondokuz Mayıs University, Faculty of Engineering, Department of Food Engineering, Samsun, Turkey.

*ugur.bayram@giresun.edu.tr

Abstract

Edible flowers have had an important place in human nutrition since ancient times. Nowadays, globalization and consumer awareness and increasing interest in healthy natural foods have made edible flowers popular again. Many different types of flowers have been used as both food and medicine in alternative medicine in different cultures. Nowadays, it is used to serve buffet meals or to decorate salads, desserts, fruits, ice creams and drinks. In addition to contributing to the appearance of food, they add flavor and smell. Edible flowers can be consumed raw or cooked, hot or cold. In our country, they are often used to produce herbal tea, jam and syrup, or as colorants. Edible flowers are an excellent natural food source. They contain mineral substances, phenolic compounds and various bioactive compounds. Some of the edible flowers are important sources of pigments such as anthocyanins and carotenoids. Studies have revealed that some edible flowers have antioxidant, anti-inflammatory, antidiabetic and anticarcinogenic properties. In this review, edible flowers commonly used in our country, their uses and biological properties are discussed.

Keywords: Edible flowers, alternative food, functional food.

DRAFT

ORAL PRESENTATION

Mangiferin: Değerli bir Antioksidan

Engin AYDIN[1*] (*ORCID: https://orcid.org/*0000-0003-4288-5911 *),*
İlkay KOCA[2] (*ORCID*: *https://orcid.org/0000-0001-6089-8586)*

[*1]Giresun Üniversitesi, Dereli Meslek Yüksekokulu, Otel, Lokanta ve İkram Hizmetleri Bölümü, Aşçılık Programı, Giresun, Türkiye.
[2]Ondokuz Mayıs Üniversitesi, Mühendislik Fakültesi, Gıda Mühendisliği Bölümü, Samsun, Türkiye.

*engin.aydin@giresun.edu.tr

Özet

Mangiferin, mango meyvesinin kabuk, sap, yaprak, kabuk ve çekirdeklerinde bulunan bir ksantondur. Geleneksel tıpta farklı kültürler, kardiyovasküler hastalık, diyabet, çeşitli enfeksiyonlar ve kanser dahil olmak üzere bir dizi hastalığın tedavisinde mangiferin açısından zengin bitkilerinden yararlanmaktadırlar. Çin, Küba ve Doğu Asya'da magniferin içermesi nedeniyle mango anti-inflamatuar, anti-viral, anti-diyabetik ve anti-kanserojen ajan olarak kullanılmaktadır. Mangiferinin bir diğer önemli kaynağı Güney Afrika'da popüler olan ve *Cyclopia sp*.'den elde edilen Honeybush çayıdır. Honeybush çay yapraklarında yaklaşık %4 mangiferin bulunduğu bildirilmiştir. Suda iyi çözünen mangiferinin ısıya dayanıklı olması, bu maddeyi değerli kılmaktadır. Son çalışmalar, önemli biyolojik aktiviteye sahip olan magniferinin farmasötik ve kozmetik alanlar için potansiyel bir terapötik ajan olacağına işaret etmektedir. Bu derlemede, magniferinin özellikleri ve sağlık üzerine pozitif etkileri üzerinde durulmaktadır.

Anahtar kelimeler: Mangiferin, sağlık, polifenol

DRAFT

ORAL PRESENTATION

Phytic acid immobilized cryogels: Studying the effect on the L929 cell proliferation for biomedical applications

Koray ŞARKAYA[1*] (*https://orcid.org/0000-0003-0177-5134*),
Doğukan Mutlu[2] (*https://orcid.org/0000-0003-3259-5822*)

[*1]Pamukkale University, Faculty of Science, Department of Chemistry, Denizli, Turkey
[21]Pamukkale University, Faculty of Science, Department of Biology, Denizli, Turkey

*Corresponding author e-mail: ksarkaya@pau.edu.tr

Abstract

Hydrogels have been employed as support materials for the growth and proliferation of mammalian cells because they closely mirror the gel-like characteristics and, in some instances, the chemical characteristics of the extracellular matrix (ECM), which naturally surrounds the cells of any biological tissue [1]. The fields of tissue engineering and in vitro cell culture have recently shown a great deal of interest in macro-porous hydrogels, also referred to as "cryogels," that are set below the freezing point of the solvent due to their inherent interconnected macro-porous structure and ease of formation in comparison to other macro-pore forming techniques [2]. In addition, a hexaphosphate-substituted inositol ring molecule known as phytic acid is a renewable resource. It is a common plant product that is easily accessible and water soluble. Also, the primary phosphorus storage form in plant seeds and bran is phytic acid (inositol hexakisphosphate), which supports several cellular processes [3]. Mammalian cells also have abundant it, with concentrations ranging from 10 to 100 mol/L [4]. In this study, poly(2-Hydroxyethyl-methacrylate) based phytic acid containing cryogels were prepared and analyzed by various characterization methods including swelling, FT-IR, SEM. Then, the cytotoxicity of phytic acid immobilized cryogel was demonstrated on the L929 mouse fibroblasts cell line. For this purpose, cryogels were extracted (0.05 g mL−1) and diluted in a DMEM complete medium to obtain a series of dilutions with 100, 75, 50, 25, and 12.5% concentrations. Cells were plated in a 96-well plate (2.5x103 cell/well) for 24 h, and the culture medium was replaced with cryogel extracts. After 24 h incubation, the cytotoxic effect was evaluated by MTT assay.
Cell viability was similar for control and treatment groups, and extracts caused no cytotoxicity to L929 cells. In conclusion, phytic acid immobilized cryogels have appropriate biological properties and are, therefore, suitable for application in biomedicine.

Keywords: Cryogels, Phytic acis, cytotoxicty, MTT, L929

DRAFT

ORAL PRESENTATION

Meme Kanseri Hücre Hatlarında WISP-1 Gen Ekspresyonu

Beyza Akın Mutlu*[1,2](*ORCID: https://orcid.org/0000 0004 1759 9216),* Demet Akdeniz Ödemiş*[1](*ORCID: https://orcid.org/0000-0002-2271-8481)*, Hülya Yazıcı*[1,3](*ORCID: https://orcid.org/0000-0002-8919-0482)*

*[1]İstanbul Üniversitesi, Onkoloji Enstitüsü ve [2]Sağlık Bilimleri Enstitüsü Temel Onkoloji Kanser Genetiği Bölümü, 34093 Çapa-Fatih, İstanbul, Türkiye.

[3]İstanbul Arel Üniversitesi, Tıp Fakültesi, Tıbbi Biyoloji ve Genetik Anabilim Dalı, 34010 Cevizlibağ-Zeytinburnu, İstanbul, Türkiye.

Sorumlu yazar e-mail:beyzakin07@gmail.com

Özet

Meme kanseri kadınlarda en sık görülen malign hastalıktır. Çeşitli genomik farklılıkların, özellikle de gen ekspresyon düzeylerinin, meme kanserinde kötü prognoz ile ilişkili olduğu bilinmektedir. Meme tümörogenezinde önemli bir role sahip olan *BRCA1* ve ilişkili *WISP1(CCN4)* mutasyonu aktivasyonu *TP53* geni aracılı apoptozu inhibe eder. Ayrıca *WISP1*'i negatif olarak düzenleyen *TGFβ1*'in meme kanserinde terapötik bir hedef olabileceğini düşündürmektedir. *WISP1* geni kanserde ilerleme, uzak metastaz ve kötü prognoz ile ilişkilidir. Ancak bu gen bazı doku tiplerinde onkogen gibi davranmaktadır. Farklı meme kanseri hücre hatlarında, MDA-MB-231, MDA-MB-453, MCF-7 ve MDA-MB-436'da *WISP-1* gen ekspresyon düzeylerinin çeşitli gen mutasyonları ile değişimi TGFβ1 tedavisi ile ilişkili olarak değerlendirilmiştir. TGFβ1 uygulamasına bağlı olarak değişen *WISP-1* gen ekspresyonu düzeyi üç tekrarlı olarak gerçek zamanlı kantitatif PZR ile değerlendirildi. MDA-MB-231, MDA-MB-453 ve MCF-7 hücre hatlarının *WISP1'*de görülen ekspresyon artışı nedeniyle kanser oluşumunda onkogen görevi gördüğü gösterilmiştir. MDA-MB-436 hücre hattında *WISP1* ekspresyonu neredeyse yok denecek kadar az olduğu görülmüştür yani bu da onkogenik özelliklere sahip olmadığı anlamına geliyor. Yapılan çalışmada ER-pozitif MCF-7 hücre hattında TGFβ1 uygulaması ile *WISP1* ekspresyonunun 6 kat arttığı gösterildi. TGFβ1'in MCF-7 hücre hattında hücre proliferasyonunu arttırdığı gözlenmiştir. MDA-MB-453, *CDH1* mutant bir hücre hattıdır. Bu hücre hattında *TP53* yolağının düzenli çalışmasına bağlı olarak TGF-β1 uygulamasının *WISP-1* ekspresyonunu azalttığı düşünülmektedir. *BRCA1* ve *RB1* mutant olan MDA-MB-436 hücre hattında ise *WISP-1* gen ekspresyonu yok denecek kadar az olduğu görülmüştür. *WISP-1* geni, meme kanseri hücre hatlarında gen mutasyonlarına bağlı olarak biyolojik belirteç niteliği taşıyabilirken, TGFβ1'de terapötik bir hedef olabileceği gösterilmiştir, ancak daha detaylı ve klinik düzeyde çalışmalar yapılmalıdır.

Anahtar Kelimeler: Meme kanseri, WISP1, TGFβ1,biyobelirteç, ekspresyon.
Bu çalışma İstanbul Üniversitesi Araştırma Fonu tarafından desteklenmiştir. Proje numarası: TYL-2019-34703; ID: 34703

DRAFT

ORAL PRESENTATION

Aromataz inhibitölerinin hayvanlarda reprodüktif amaçlı kullanımı

Mehmet Buğra KIVRAK[1*] (*ORCID: https://orcid.org/0000-0002-4772-874X)*

[*1]Sivas Cumhuriyet Üniversitesi, Veteriner Fakültesi, Doğum, Sivas, Türkiye.

*Corresponding author e-mail: mbkivrak@cumhuriyet.edu.tr

Abstract

Anti-östrojenik maddeler uzun yıllardır insanlarda meme tümörü tedavisinde post-operatif olarak kullanılmaktadır. Non-steroidal anöstrojenik ilaçlardan tamoksifen ilk olarak 1967 yılında geliştirilmiş ve uzun yıllar meme tümörü tedavisinde post operatif olarak kullanılmıştır. Ancak bu tür antiöstrojenik maddeler aynı zamanda uzun vadede östrojenik aktivite de göstermektedir. Bu nedenle potansiyel yan etkileri daha az olan aromataz inhibitörleri bu amaçla kullanılmaya başlanmıştır. Aromataz CYP 450 enzim ailesinin bir üyesidir ve ovaryum, yağ doku, beyin, adrenal bez, meme, karaciğer ve plasenta gibi dokularda bulunur. Aromataz, adrenal bezden üretilen C19 androjenlerden C18 östrojenik steroidlerin oluşumunda önemli rol oynar ve bu nedenle östrojen üretiminde hız kısıtlayıcı basamak olarak kabul edilir. Aromataz maddeler östrojenik feedback mekanizmasını değiştirerek gonadotropin salınımını artırdığından insanlarda ovaryum stimulasyonu amacıyla güvenle kullanılmaktadır. Östrojenin hidroksil metabolitleri östrojen reseptör yolaklarını indükleyerek epidermal büyüme faktörünün aktivasyonuna neden olur ve bu da meme kanserinin başlaması ve ilerlemesinde önemlidir. Bu nedenle postmenopozal kadınlarda meme kanseri tedavisinde östrojen üretimini engellemek için aromataz inhibitörleri kullanılır. Bunun yanında azospermik ve oligozoospermik erkekler de dahil olmak üzere infertil hastalarda aromataz inhibitörlerinin olumlu etkilerini bildiren birçok çalışma bulunmaktadır. Yapılan bu çalışmaların ışığında aromataz inhibötlerinin hayvanlar üzerindeki etkileri de son yıllarda ilgi gören bir konu haline gelmiştir. Özellikle potansiyel toksisiteleri sebebiyle steroidlerin insan tüketimine sunulan hayvanlarda kullanımı Avrupa Birliği tarafından yasaklanması nedeniyle aromataz inhibtörleri çiftlik hayvanlarda non-steroidal senkronizasyon methodları geliştirme amacıyla kullanılmıştır. Sığırlar üzerinde yapılan çalışmalarda aromtaz inhibötlerinin LH salınım ritmini değiştirerek daha büyük foliküllerin ovule olmasını ve daha büyük bir corpus luteum şekillenmesini sağlamıştır. Anöstrustaki koyunlarda kullanıldığında FSH salınımında belirgin bir artışa sebep olmuştur. İnfertil erkek keçi ve köpeklerde uzun süreli kullanımı sonucunda spermatalojik parametrelerde belirgin artışlar görülmüştür. Bu nedenle yapılan çalışmalar incelendiğinde anti-östrojenik maddelerin hayvanlarda reprodüktif amaçlı kullanımının yaygınlaşacağı değerlendirilmektedir.

Keywords: Antiöstrojen, aromataz inhibitörü, Üreme, Sığır, Koyun

DRAFT

ORAL PRESENTATION

Reproductive Evaluation of Two Different Progesterone Assisted Sexual Stimulation Methods in Acyclic Simmental Breed Heifers

Abdurrahman TAKCI[1]

[1]Department of Obstetrics and Gynecology, Faculty of Veterinary Medicine, University of Sivas Cumhuriyet, 58140, Sivas, Turkey

*Corresponding Author's E-Mail: abdurrahmantakci@cumhuriyet.edu.tr

Abstract

In order for cows to reach maximum productivity throughout their lives, their first calving must be around 24 months. To meet this reproductive parameter, heifers must be ensured to become pregnant at approximately 15 months of age. Before getting pregnant, They are required to go through several estrous cycles, provided that they are regular. However, in most farm this reproductive process does not occur as desired. In heifers, estrus either does not occur at all or is so weak that it cannot be detected. The material of this study consisted of 72 Simmental breed heifers whose ages were between 18-21 months, but whose oestrus was not determined. These heifers were first subjected to ultrasonographic examination twice consecutively, with an interval of 8 days (day -8 and day 0). Animals that did not have a corpus luteum on the ovary in both examinations were considered acyclic (not cycling). Thus, 60 heifers considered to be acyclic were included in the study. The diameters of the largest candidate follicles found on the ovaries of these 60 heifers were measured. Two groups were created, each group containing 30 heifers. In the second ultrasonographic examination (day 0), follicle diameters were measured and an intravaginal device containing progesterone was placed in the heifers in both groups. While the animals in the first group were injected with GnRH while the intravaginal device was inserted, this procedure was not performed in the animals in the second group. After 5 days (day 5), the intravaginal materials were removed from the animals in both groups and Prostaglandin f2 alpha (PGF2α) injection was made. One day later, the second PGF2α injection was given to the animals in group 1, while the second PGF2α injection was not given to the animals in group 2. GnRH injection was applied to the heifers in both groups 72 hours after the intravaginal device was removed (day 8) and fixed-time insemination was performed. Pregnancy examination was performed using ultrasonographic method 35 days after the insemination date. It was determined that 14 animals in Group 1 and 16 animals in Group 2 were pregnant, and the difference was found to be statistically insignificant. In the light of this information, it was concluded that it would be more appropriate to apply the protocol applied to group 2, as it involves less hormone injection to acyclic heifers.

Keywords: , Breed, Estrus, Heifer, Progesterone, Simmental

DRAFT

ORAL PRESENTATION

Biyoinformatik araçlar kullanılarak TAOK1 genindeki tek nükleotid varyasyonların (SNV'lerin) değerlendirilmesi

Tuğba KAMAN (ORCID ID : 0000-0002-5885-0193)

Üsküdar Üniversitesi, Sağlık Hizmetleri Meslek Yüksek Okulu, Tıbbi ve Aromatik Bitkiler, Üsküdar, İstanbul, Türkiye

Sorumlu yazar e-mail: tugba.kamanuskudar.edu.tr

Özet

Serin-treonin kinaz kodlayan bir gen olan TAOK1 geni, hem nörogelişimsel bozukluk hem de otizm spektrum bozukluğu (ASD) ile ilişkilendirilmektedir. TAOK1'in beyin içindeki nöronlarda yüksek düzeyde eksprese edildiği ve fosfoinositidlerle doğrudan ilişki yoluyla plazma zarının yeniden şekillenmesinde işlevsel bir role sahip olduğunu gösterilmiştir. Bu çalışmada, TAOK1 geninde yer alan tek nükleotid varyasyonlardan (SNV) zararlı veya hastalıkla ilişkili olanların tespiti, ortak zarar verici etkiye sahip olan varyasyonların filtrelenerek protein stabilizasyonu üstüne etkileri ve üç boyutlu modellemelerinin biyoinformatik araçlar kullanılarak belirlenmesi amaçlanmıştır. Ayrıca gen-gen etkileşimleri ve protein-protein etkileşimleri değerlendirilmiştir. TAOK1 genindeki SNV'ler NCBI dbSNP veritabanından 2023 yılı Mart ayında elde edilmiştir. Aminoasit değişimine sebep olduğu bilinenlerin fonksiyonel etkileri SIFT (Sorting Intolerant From Tolerant), PolyPhen-2, SNP&GO, SNAP2, PANTHER ve Mutation Assessor yazılım araçları kullanılarak tespit edilmiştir. Bu SNV'lerden ortak zarar verici etkiye sahip olanlar filtrelenerek protein stabilizasyonuna olan etkilerinin belirlenebilmesi için I-Mutant 2.0 ve MuPRO, üç boyutlu modellemelerin oluşturulmasında ise Project HOPE yazılım araçları kullanılmıştır. Gen-gen etkileşimleri için GeneMANIA, protein-protein etkileşimleri için STRING yazılım aracından veri sağlanmıştır. TAOK1 geninde 462 missense SNV, SIFT, PolyPhen-2, SNP&GO, SNAP2, PANTHER, MutationAssessor yazılım araçları ile taranmış olup 9 SNV'nin ortak zarar verici etkiye sahip olduğu tespit edilmiştir. I-Mutant 2.0 sonuçları, 9 SNV'den 6 tanesinin protein stabilizasyonunu azalttığı 3 tanesinin ise artırdığını, MuPRO sonuçları ise tüm SNV'lerin protein stabilizasyonunu azalttığını göstermiştir. Project HOPE yazılım aracı ile yabanıl ve mutant rezidülerin yük, boyut, hidrofobiklik ve yapısal özelliklerine ait bilgilerine ulaşılmıştır. Biyoinformatik araçlar kullanılarak yapılan taramalar sonucunda TAOK1 geninde 9 SNV'nin protein yapısı ve stabilizasyonu üzerine zarar verici etkisi olabileceğini göstermiştir. Elde edilen sonuçların ileri yapılacak deneysel çalışmalara veri sağlayacağı öngörülmektedir.

Anahtar Kelimeler: TAOK1 geni, nörogelişimsel bozukluk, tek nükleotid varyasyon (SNV), biyoinformatik

ORAL PRESENTATION

Boron-complexed polylipoic acid as metal-free self-healing polymer

Efdal Teknikel[1*] (*ORCID: https://orcid.org/0000-0003-3218-9437*),

[*1] HacettepeUniversity, Faculty of Science, Department of Chemistry, Ankara, Türkiye.

*Corresponding author e-mail: efdalt@hacettepe.edu.tr

Abstract

Restoring the polymer to its original state after mechanical changes such as cracks, ruptures, punctures or cuts in a polymer is important for the development of long-term usable materials. A structure that has been frequently studied recently for the development of such self-healing polymers has been poly-lipoic acid. Different working groups have shown that self-healing polymers can be developed by adding different metal cations (Fe3+, Zn, Cu2+) to this polymer. In our study, it was shown that boron-chelated lipoic acid chains also have self-healing properties. No crosslinker was used in the synthesis of this polymer. Characterization of the polymer was made by mass spectroscopy and infra-red spectroscopy and it was determined that it polymerized in a cyclic form. As a result of the tests, the material was observed to repair itself very efficiently and quickly. The carboxyl acid groups in this polymer are of course involved in this self-healing process, but the crystalline feature of the non-boron-chelated polymer reveals the effect of the boron atom in the self-healing process.

Keywords: Boron, Lipoic acid, Polymer, Self-healing

DRAFT

ORAL PRESENTATION

In terms of biological activity: Carob

Falah Saleh Mohammed[1*] (*ORCID: https://orcid.org/0000-0001-9083-1876*), Mustafa Sevindik[2] (*ORCID: https://orcid.org/0000-0001-7223-2220*), Muhittin Doğan[3] (*ORCID: https://orcid.org/0000-0001-5400-8065*), İmran Uysal[4] (*ORCID: https://orcid.org/0000-0003-0942-9658*), Ali Erdem Şabik[5] (*ORCID: https://orcid.org/0000-0001-6182-3834*)

[1] Zakho University, Science and Literature Faculty, Department of Biology, Duhok, Iraq
Email: falah.sindy@uoz.edu.krd
Cep: +964 750 450 2150
[2] Osmaniye Korkut Ata University, Science and Literature Faculty, Department of Biology, Osmaniye, Turkey
Email: sevindik27@gmail.com
Cep: +905327484228
[3] Gaziantep University, Science and Literature Faculty, Department of Biology, Gaziantep, Turkey
Email: doganm@gantep.edu.tr
Cep: +905061250842
[4] Bahçe Vocational School of Higher Education, Department of Food Processing, Bahçe-Osmaniye, Turkey
Email: imranuysal@osmaniye.edu.tr
Cep: +905068759306
[5] Osmaniye Korkut Ata University, Bahçe Vocational School of Higher Education, Department of Chemistry and Chemical Processing Technologies, Bahçe-Osmaniye, Turkey
Email: erdemsabik@osmaniye.edu.tr
Cep: +90 506 860 16 88

Abstract

Plants have been preferred for many different purposes from past to present. Plants that have been widely used throughout human history attract attention with their nutritional properties. In addition to its nutritional properties, many plant species are also very important medicinally thanks to the bioactive compounds they produce. In this study, the biological activities of *Ceratonia siliqua* L. reported in the literature were compiled. *C. siliqua* is known as "Carob". Carob, which belongs to the Fabaceae family, is widely distributed in the Mediterranean region. The pod-shaped fruits of carob are edible. Carob is in the form of evergreen shrubs and trees. As a result of literature research, *C. siliqua* has been shown to have activities such as antioxidant, antimicrobial, cytotoxic, anti-inflammatory and antiproliferative. As a result, it is thought that the carob plant can be used in different studies in the field of pharmacology.

Keywords: Biological activities, Carob, *Ceratonia siliqua*, medicinal plants

DRAFT

ORAL PRESENTATION

Biological Activities of Edible Girolle Mushrooms

Vadim Tagirovich Khassanov[1] (*ORCID: https://orcid.org/0000-0002-9054-5551*), Celal Bal[2] (*ORCID: https://orcid.org/0000-0001-6856-3254*), Mustafa Sevindik[3*] (*ORCID: https://orcid.org/0000-0001-7223-2220*)

[1] Saken Seifullin Kazakh Agrotechnical University, Astana, Kazakhstan
Email: vadim_kazgatu@mail.ru
Cep: +7 701 421 2134
[2]Gaziantep University, Science and Literature Faculty, Department of Biology, Gaziantep, Turkey
Email: bal@gantep.edu.tr
Cep: +905308767611
[3]Osmaniye Korkut Ata University, Science and Literature Faculty, Department of Biology, Osmaniye, Turkey
Email: sevindik27@gmail.com
Cep: +905327484228

Abstract

In addition to their nutritional importance, mushrooms are also used in the field of health to treat different ailments and problems. In this study, the biological activities of *Cantharellus cibarius* Fr. reported in the literature were compiled. *C. cibarius* is known as "girolle". Girolle is in the Cantharellaceae family. It is widespread from Europe across the Mediterranean to Scandinavia, especially in deciduous and pine forests. As a result of literature research, *C. cibarius* has been shown to have biological activities such as antioxidant, antimicrobial, anti-inflammatory, antihyperglycemic, cytotoxic, anticancer and pesticidal. As a result, it is thought that *C. cibarius* can be preferred as a natural source in the field of pharmacology.

Keywords: Antimicrobial, antioxidant, girolle, medicinal mushroom, *Cantharellus*

DRAFT

ORAL PRESENTATION

A compilation on the biological activities of gum tree

Falah Saleh Mohammed[1*] (*ORCID: https://orcid.org/0000-0001-9083-1876*), Mustafa Sevindik[2] (*ORCID: https://orcid.org/0000-0001-7223-2220*), Muhittin Doğan[3] (*ORCID: https://orcid.org/0000-0001-5400-8065*), İmran Uysal[4] (*ORCID: https://orcid.org/0000-0003-0942-9658*), Ali Erdem Şabik[5] (*ORCID: https://orcid.org/0000-0001-6182-3834*)

[1] Zakho University, Science and Literature Faculty, Department of Biology, Duhok, Iraq
Email: falah.sindy@uoz.edu.krd
Cep: +964 750 450 2150
[2] Osmaniye Korkut Ata University, Science and Literature Faculty, Department of Biology, Osmaniye, Turkey
Email: sevindik27@gmail.com
Cep: +905327484228
[3] Gaziantep University, Science and Literature Faculty, Department of Biology, Gaziantep, Turkey
Email: doganm@gantep.edu.tr
Cep: +905061250842
[4] Bahçe Vocational School of Higher Education, Department of Food Processing, Bahçe-Osmaniye, Turkey
Email: imranuysal@osmaniye.edu.tr
Cep: +905068759306
[5] Osmaniye Korkut Ata University, Bahçe Vocational School of Higher Education, Department of Chemistry and Chemical Processing Technologies, Bahçe-Osmaniye, Turkey
Email: erdemsabik@osmaniye.edu.tr
Cep: +90 506 860 16 88

Abstract

Plants are indispensable elements of diet lists that stand out with their nutritional properties. Many plant species have been used by humans for many purposes such as shelter, heating, equipment, food and medicine. It is known that different plant species are used as tools in the fight against different diseases. In this study, the biological activities of *Pistacia lentiscus* L. reported in the literature were compiled. *P. lentiscus* is known as the "gum tree". It is generally distributed in the Mediterranean region, does not shed its leaves in winter and has a shrub form. The fruits are round, small and reddish black in colour. As a result of literature research, *P. lentiscus* has been shown to have activities such as antioxidant, antimicrobial, anticancer, cytoprotective, antiparasitic, anti-inflammatory, hepatoprotective and antidiabetic. As a result, it is thought that the use of the gum tree plant can be expanded in many areas.

Keywords: Biological activities, Gum tree, medicinal plants, *Pistacia lentiscus*

DRAFT

ORAL PRESENTATION

Biological activities of Hawthorn, an important fruit

İmran Uysal[1*] (*ORCID: https://orcid.org/0000-0003-0942-9658*), Ali Erdem Şabik[2] (*ORCID: https://orcid.org/0000-0001-6182-3834*), Falah Saleh Mohammed[3] (*ORCID: https://orcid.org/0000-0001-9083-1876*), Mustafa Sevindik[4] (*ORCID: https://orcid.org/0000-0001-7223-2220*), Şevket Sümer[5] (*ORCID: https://orcid.org/0000-0001-5495-1299*)

[1] Bahçe Vocational School of Higher Education, Department of Food Processing, Bahçe-Osmaniye, Turkey
Email: imranuysal@osmaniye.edu.tr
Cep: +905068759306
[2] Osmaniye Korkut Ata University, Bahçe Vocational School of Higher Education, Department of Chemistry and Chemical Processing Technologies, Bahçe-Osmaniye, Turkey
Email: erdemsabik@osmaniye.edu.tr
Cep: +90 506 860 16 88
[3] Zakho University, Science and Literature Faculty, Department of Biology, Duhok, Iraq
Email: falah.sindy@uoz.edu.krd
Cep: +964 750 450 2150
[4] Osmaniye Korkut Ata University, Science and Literature Faculty, Department of Biology, Osmaniye, Turkey
Email: sevindik27@gmail.com
Cep: +905327484228
[5] Department of Pharmacy Services, Vocational School of Health Services, University of Osmaniye Korkut Ata, Osmaniye, 80000, Turkey
Email: sevketsumer@osmaniye.edu.tr
Cep: +90 5545927106

Abstract

Natural products are preferred in traditional medicine, and plants are the most important of these natural products. Plants are the most commonly used natural products by humans. In this study, the biological activities of *Crataegus monogyna* Jacq. reported in the literature were compiled. *C. monogyna* is known as " Hawthorn". It is in the Rosaceae family. Hawthorn usually grows in forest areas. Its flowers, which bloom in spring, are white and pink. The fruits are red or dark red, 6-10 mm in diameter. Jam, marmalade and paste are made from its fruits. As a result of literature research, it has been observed that the *C. monogyna* species has activities such as antioxidant, antimicrobial, antidiabetic, antihyperglycemic, anti-inflammatory and antiproliferative. As a result, it is thought that the common hawthorn plant can be used as a natural product in the field of health.

Keywords: Biological activities, *Crataegus monogyna*, Hawthorn, medicinal plants

DRAFT

ORAL PRESENTATION

A review on the biological activities of Raspberry

Ali Erdem Şabik[1*] (*ORCID: https://orcid.org/0000-0001-6182-3834*), Falah Saleh Mohammed[2] (*ORCID: https://orcid.org/0000-0001-9083-1876*), İmran Uysal[3] (*ORCID: https://orcid.org/0000-0003-0942-9658*), Muhittin Doğan[4] (*ORCID: https://orcid.org/0000-0001-5400-8065*), Mustafa Sevindik[5] (*ORCID: https://orcid.org/0000-0001-7223-2220*)

[1] Osmaniye Korkut Ata University, Bahçe Vocational School of Higher Education, Department of Chemistry and Chemical Processing Technologies, Bahçe-Osmaniye, Turkey
Email: erdemsabik@osmaniye.edu.tr
Cep: +90 506 860 16 88
[2] Zakho University, Science and Literature Faculty, Department of Biology, Duhok, Iraq
Email: falah.sindy@uoz.edu.krd
Cep: +964 750 450 2150
[3] Bahçe Vocational School of Higher Education, Department of Food Processing, Bahçe-Osmaniye, Turkey
Email: imranuysal@osmaniye.edu.tr
Cep: +905068759306
[4] Osmaniye Korkut Ata University, Science and Literature Faculty, Department of Biology, Osmaniye, Turkey
Email: sevindik27@gmail.com
Cep: +905327484228
[5] Gaziantep University, Science and Literature Faculty, Department of Biology, Gaziantep, Turkey
Email: doganm@gantep.edu.tr
Cep: +905061250842

Abstract

Plants are important natural products responsible for many biological activities. Many studies have shown that plants have different biological activities. In our study, the biological activities of *Rubus idaeus* L. reported in the literature were compiled. *R. idaeus* (Rosaceae) is known as raspberry or raspberry. It is a plant species that produces red and sweet fruits in summer and autumn. They develop in places where forests are open or in areas previously cleared by fire or logging. They grow at altitudes of 900-2200 m, under forests, on the edges of meadows and on stony slopes. According to literature data, *R. idaeus* has been reported to have many biological activities such as antioxidant, antimicrobial, anticancer, cytotoxic, antiproliferative and anti-inflammatory. Important findings have been found in the literature, especially in terms of antioxidant and antimicrobial activity. It is thought that this plant species, which stands out with its nutritional properties, may also be an important natural material from a medical perspective.

Keywords: Raspberry, biological activities, medicinal plants, *Rubus idaeus*

DRAFT

ORAL PRESENTATION

Biological activity of Silverberry

İmran Uysal[1*] (*ORCID: https://orcid.org/0000-0003-0942-9658*), Ali Erdem Şabik[2] (*ORCID: https://orcid.org/0000-0001-6182-3834*), Falah Saleh Mohammed[3] (*ORCID: https://orcid.org/0000-0001-9083-1876*), Mustafa Sevindik[4] (*ORCID: https://orcid.org/0000-0001-7223-2220*), Şevket Sümer[5] (*ORCID: https://orcid.org/0000-0001-5495-1299*)

[1] Bahçe Vocational School of Higher Education, Department of Food Processing, Bahçe-Osmaniye, Turkey
Email: imranuysal@osmaniye.edu.tr
Cep: +905068759306
[2] Osmaniye Korkut Ata University, Bahçe Vocational School of Higher Education, Department of Chemistry and Chemical Processing Technologies, Bahçe-Osmaniye, Turkey
Email: erdemsabik@osmaniye.edu.tr
Cep: +90 506 860 16 88
[3] Zakho University, Science and Literature Faculty, Department of Biology, Duhok, Iraq
Email: falah.sindy@uoz.edu.krd
Cep: +964 750 450 2150
[4] Osmaniye Korkut Ata University, Science and Literature Faculty, Department of Biology, Osmaniye, Turkey
Email: sevindik27@gmail.com
Cep: +905327484228
[5] Department of Pharmacy Services, Vocational School of Health Services, University of Osmaniye Korkut Ata, Osmaniye, 80000, Turkey
Email: sevketsumer@osmaniye.edu.tr
Cep: +90 5545927106

Abstract

Plants; It has been used in different areas such as shelter, nutrition, dressing and alternative medicine. These features put the plants at the top of functionally used natural products. Many studies have shown that plants have important biological activities. In this study, the biological activities of *Elaeagnus angustifolia* L. reported in the literature were compiled. *E. angustifolia* is known as "bird oleaster". It is in the Elaeagnaceae family. Buckthorn grows as a native species in Western and Central Asia, including Turkey and Iran. Its fruit, whose leaves resemble willow leaves, is generally orange yellow in color and reaches up to 7-8 meters in length. As a result of literature research, *E. angustifolia* has been shown to have activities such as antioxidant, antimicrobial, antimutagenic, antitumor, insecticidal, anticancer and anti-inflammatory. As a result, it is thought that the bird oleaster plant can be used in different subjects by expanding its biological activity areas.

Keywords: Biological activities, *Elaeagnus angustifolia*, medicinal plants, silverberry

DRAFT

ORAL PRESENTATION

Strict-branch Coral Mushrooms and Biological Activities

Vadim Tagirovich Khassanov[1] (*ORCID: https://orcid.org/0000-0002-9054-5551*), Celal Bal[2] (*ORCID: https://orcid.org/0000-0001-6856-3254*), Mustafa Sevindik[3*] (*ORCID: https://orcid.org/0000-0001-7223-2220*)

[1] Saken Seifullin Kazakh Agrotechnical University, Astana, Kazakhstan
Email: vadim_kazgatu@mail.ru
Cep: +7 701 421 2134
[2]Gaziantep University, Science and Literature Faculty, Department of Biology, Gaziantep, Turkey
Email: bal@gantep.edu.tr
Cep: +905308767611
[3]Osmaniye Korkut Ata University, Science and Literature Faculty, Department of Biology, Osmaniye, Turkey
Email: sevindik27@gmail.com
Cep: +905327484228

Abstract

Mushrooms, like plants, are evaluated nutritionally by humans. Apart from its nutritional aspects, its use in the field of pharmacology has recently become widespread. In this study, the antioxidant and antimicrobial activities of *Ramaria stricta* (Pers.) Quél. species reported in the literature were compiled. *R. stricta*, commonly known as tight branch coral, is a coral fungus of the genus *Ramaria*. Tight branch coral is in the Gomphaceae family. It has a cosmopolitan distribution and grows on dead wood, logs, trunks and branches of both deciduous and coniferous trees. As a result of literature research, *R. stricta* was found to have antioxidant and antimicrobial activities. As a result, it is thought that *R. stricta* mushroom can be preferred as a natural source of antioxidant and antimicrobial properties in pharmacological designs.

Keywords: Antimicrobial, antioxidant, coral, medicinal mushroom, *Ramaria*

DRAFT

ORAL PRESENTATION

Wolf grapes in terms of antioxidant, antimicrobial and other activities

Ali Erdem Şabik[1*] (*ORCID: https://orcid.org/0000-0001-6182-3834*), Falah Saleh Mohammed[2] (*ORCID: https://orcid.org/0000-0001-9083-1876*), İmran Uysal[3] (*ORCID: https://orcid.org/0000-0003-0942-9658*), Muhittin Doğan[4] (*ORCID: https://orcid.org/0000-0001-5400-8065*), Mustafa Sevindik[5] (*ORCID: https://orcid.org/0000-0001-7223-2220*)

[1] Osmaniye Korkut Ata University, Bahçe Vocational School of Higher Education, Department of Chemistry and Chemical Processing Technologies, Bahçe-Osmaniye, Turkey
Email: erdemsabik@osmaniye.edu.tr
Cep: +90 506 860 16 88
[2] Zakho University, Science and Literature Faculty, Department of Biology, Duhok, Iraq
Email: falah.sindy@uoz.edu.krd
Cep: +964 750 450 2150
[3] Bahçe Vocational School of Higher Education, Department of Food Processing, Bahçe-Osmaniye, Turkey
Email: imranuysal@osmaniye.edu.tr
Cep: +905068759306
[4] Osmaniye Korkut Ata University, Science and Literature Faculty, Department of Biology, Osmaniye, Turkey
Email: sevindik27@gmail.com
Cep: +905327484228
[5] Gaziantep University, Science and Literature Faculty, Department of Biology, Gaziantep, Turkey
Email: doganm@gantep.edu.tr
Cep: +905061250842

Abstract

Plants have been used for different purposes, from the most primitive societies to the most developed societies. The most important of these different purposes is the treatment of diseases. In this study, the biological activities of *Lycium barbarum* L. reported in the literature were compiled. *L. barbarum* plant is known as Chinese wolfberry, Goji berry, barbary matrimony vine, red medlar or matrimony vine. It is in the Solanaceae family. The homeland of Goji berry is known as China. It is a deciduous woody shrub that usually grows to a height of 1-3 meters. Its fruits are bright orange-red. As a result of literature research, *L. barbarum* has been shown to have activities such as antioxidant, antimicrobial, anti-inflammatory, anti-tumor and anti-diabetic. As a result, it is thought that the wolfberry plant can be used for different purposes in the field of health apart from its nutritional properties.

Keywords: Biological activities, Goji berry, medicinal plants, wolfberry

DRAFT

ORAL PRESENTATION

Silsesquioxane-based nanoadsorbent for the wastewater treatment

Asuman Celik-Kucuk* (*ORCID: 0000-0002-3509-1497)*

[*] Marmara University, Metalurgical and Materials Engineering 34722 Istanbul, Turkey

*Corresponding author e-mail: asumancelikkucuk@gmail.com

Abstract

In this study, polyhedral oligomeric silsesquioxanes (POSS) and chitosan based nanocomposite materials (PSCS-(1,2,3)) as nanoadsorbents were prepared and investigated for the first time. POSS with eight arms of di(ethylene glycol) oxides units (POSS-DEG) has been synthesized by direct hydrosilylation reaction of POSS-SiH with di(ethylene glycol)vinyl ether.[1] POSS-DEG has been characterized by FT-IR, 1H and 13C NMR. POSS -DEG and chitosan based nanocomposite materials (PSCS-(1,2,3)) were prepared by varying the cross-linker ratio while keeping POSS and chitosan quantities constant. The sol-gel method was used to develop nanocomposites (PSCS-(1,2,3)) by using POSS-DEG and chitosan as precursors. TEOS has been preferred to utilize as a cross-linking agent due to the having potential to keep the active amine groups as in pristine chitosan. The prepared nanocomposite materials (PSCS-(1,2,3)) have been characterized (Figure 1a) by FT-IR spectroscopy, TGA and SEM (Figure 1b). The effectiveness of PSCS-(1,2,3) nanocomposites in wastewater treatment has been investigated and compared to the chitosan beads. It is well known that chitosan beads cross-linked by TEOS showed efficient cationic Cd (II) adsorption. Among the prepared nanocomposites, PSCS-2 was found to be the most effective nanoadsorbent for the treatment of wastewater containing cadmium (Cd (II)) because of having a balance between the availability of functional sites and the stability of the nanocomposites (Figure 1c). The influence of pH, saturation time, initial metal concentration was determined under the optimum conditions. The adsorption property of the nanocomposite material was investigated by the Langmuir and Freundlich adsorption isotherms. Characterization and analytical results demonstrated that Cd (II) ions were adsorbed by functional groups available in nanocomposite (PSCS;1,2,3). PSCS-2 showed higher adsorption capacity of 98.6% compared to chitosan (70%), PSCS-1 and PSCS-3 due to the good balance between the availability of functional sites and the stability of the nanocomposites.[2] The contribution of nanohybrid POSS on the adsorption of Cd (II) is probably due to the having extra OH groups and increasing surface area of nanoadsorbant. Although the availability of functional sites of OH decrease with increasing the amount of TEOS, the stability of nanocomposite composite increases. Therefore, PSCS-2 has showed higher recovery than PSCS-1 due to the stability of PSCS-2. The lower capacity of PSCS-3 compared to PSCS-2 might be because of lowering functional sites and nonhomogeneous film formation. From this result, it can be emphasized that the balance between the stability and availability of the functional sites have significant role on the adsorption. Moreover, competitive adsorption test showed that PSCS-2 exhibited high selectivity for Cd (II), Cu (II), and Ni (II) ions. Good fitting with the Langmuir model was attributed to succeeding a smooth surface morphology thanks to the good solubility and dispersibility of POSS molecules in organic environments. Therefore, a high adsorption capacity value (98.6%) with incorporation of POSS (for PSCS-2) compare to that of chitosan (70%) was succeeded. It can be concluded that the use of POSS reinforced chitosan nanocomposite to treat heavy metal pollutants in wastewater is promising and deserves further investigation.

Keywords: Polyhedral oligomeric silsesquioxanes (POSS), POSS/chitosan based nanocomposite, Sol-gel method, AAS, Cadmium (II) Adsorption

DRAFT

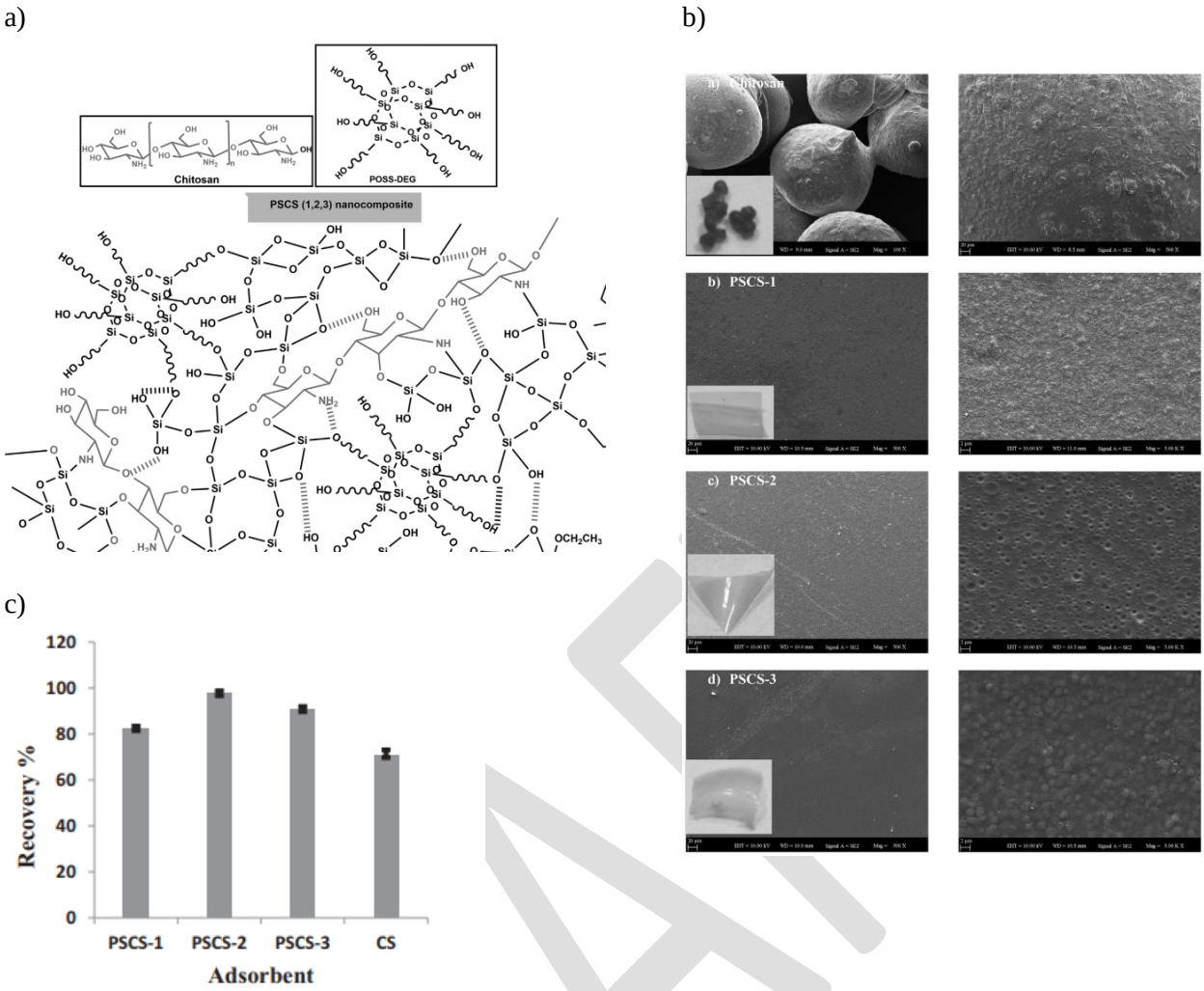


Figure 1 (a) Proposed interactions for PSCS-(1,2,3) nanocomposites. POSS-DEG and chitosan have been expected to bind to each other directly and also through TEOS.Expected hydrogen bonds are marked by dotted line**. (b)** SEM images for chitosan and the nanocomposites (PSCS-(1,2,3)) obtained after the sol-gel by using POSS-DEG and chitosan as precursors. **(c)** Cd (II) adsorption capacity of PSCS-1, PSCS-2, PSCS-3 and chitosan at pH 7 and room temperature.

References

A.C. Kucuk, J. Matsui, T. Miyashita, Langmuir-Blodgett films composed of amphiphilic double-decker shaped polyhedral oligomeric silsesquioxanes, J. Colloid. Interf. Sci. 355 (1) (2011) 106–114, https://doi.org/10.1016/j.jcis.2010.12.033

G.Z. Li, L.C. Wang, H.L. Ni, C.U. Pittman, Polyhedral oligomeric silsesquioxane (POSS) polymers and copolymers: a review, J. Inorg. Organomet. 11 (3) (2001) 123–154, https://doi.org/10.1023/A:1015287910.

ORAL PRESENTATION

Assessing Generation-Specific Chemical Profiles of Calliphora Adult Flies

Canan KULA

Hacettepe University, Graduate School of Science and Engineering, Department of Forensic Science, Ankara, Turkey

Abstract

The discipline of forensic entomology plays a pivotal role in the estimation of the post-mortem interval (PMI) through the examination of colonising insect species. Calliphoridae (Diptera) occupies an interest among insects due to their well-defined life cycle and close association with decaying organic matter. Species identification is a fundamental aspect of forensic entomology, as it provides valuable information about the location and circumstances surrounding a death. Estimating the age of adult flies is another critical element in determining the PMI. Traditional methods rely on morphological changes. Nevertheless, the chemical composition of the cuticle is species-specific, like a fingerprint, and undergoes systematic changes over a period of time, which can be used as an indicator of species and their age. The cuticle is coated with a waxy layer consisting of hydrocarbons, fatty acids, and alcohols.

This research aims to examine variations in Calliphora adult flies throughout different generations. This will be achieved via the use of gas chromatography-mass spectrometry (GC-MS) and principal component analysis (PCA) to analyse cuticular hydrocarbons. PCA, a statistical methodology, is used to examine and interpret intricate patterns seen in the cuticular hydrocarbon profiles and facilitates the identification of notable distinctions between several generations of adult Calliphora flies by lowering the dimensionality of the data. This methodology facilitates a more complete comprehension of changes peculiar to different generations.

The aforementioned results provide significant contributions to the field of forensic entomology. However, they also underscore the need for more study to better understand the complex correlation between generation-specific chemical profiles and their possible influence on PMI prediction. The acquisition of these findings has the potential to significantly improve the accuracy and reliability of the use of cuticular hydrocarbon analysis in criminal investigations, hence providing valuable assistance to law enforcement organisations in the resolution of complex criminal cases.

Keywords: Calliphoridae, cuticular hydrocarbon analysis, generation differences

DRAFT

ORAL PRESENTATION

Protected fatty acid feeding influences production parameters and rumen digestibility in mid and late lactation dairy cows

Halil İbrahim Tosun[1*] (*ORCID: https://orcid.org/0000-0001-5117-0390*),
David P. Casper[2] (*ORCID: https://orcid.org/0000-0002-0666-8808*)

[*1]Rumico Livestock Management and Nutrition, Aydın, Turkiye
[2] North Carolina A&T State University, Department of Animal Sciences, Greensboro, NC, USA and Colorado State University, Department of Animal Sciences, Fort Collins, CO, USA

*Corresponding author e-mail: halil.tosun@rumico.com.tr

Abstract

Feeding fatty acids to lactating dairy cows improve milk yield, milk fat, energy balance, reproduction and other biological functions. Protecting fats from ruminal bio-hydrogenation provides concentrated energy without reducing total track digestibility. This study will discuss a recent experiment feeding calcium salts of palm fatty acids (CSPF) on nutrient digestibility and milk production. Forty multiparous dairy cows (Jersey × Holstein) producing 20 ± 1.69 kg of milk/d and 20 ± 5.0 d in milk were assigned to 1 of 4 treatments using a randomized complete block design. Feeding increasing CSPF amounts linearly decreased dry matter intake (1.20 kg/d), while linearly increasing neutral detergent fiber digestibility (3.90%), and quadratically increasing total fat digestibility (6.30% at 0.4 kg/d CSPF). Increasing CSPF feeding rate increased energy-corrected milk (ECM; 3.30 kg/d), milk fat (0.11 kg/d), and feed efficiency (ECM/dry matter intake, 0.34 kg/kg) compared with cows fed no CSPF. Palm fatty acid, which is a 16-carbon saturated fatty acid is known to increase milk fat concentration and production. In controlled research studies, the increase in milk production has been greater than 1 kg per cow per day. Another important fatty acid is oleic acid, an 18-carbon fatty acid having one double bond. Previously, feeding oleic acid increases NDF digestibility, probably through stimulating fiber digesting bacteria. Oleic acid is easily absorbed as a micelle in the small intestine and the first fatty acid attached to glycerol when synthesizing a triglyceride for export as milk fat by the mammary gland. Oleic acid is thought to influence metabolism while increasing the sensitivity to insulin. Reducing oleic acid concentrations in the rations during Mid- to Late-lactation could be advantageous by minimizing the increase in body condition scores. Fatty acids are more than just an energy source for milk fat, they also impact immunity, metabolism, and body condition score.

Keywords: Fatty acid, Milk, Milk fat, Dairy cow, Energy, Yield.

DRAFT

ORAL PRESENTATION

Sağlık profesyonellerinin genetik ve genetik uygulamalara yönelik tutum ve anlayışlarının belirlenmesi

Abdullah Karabey[1*] (*ORCID: https://orcid.org/0000-0002-3507-7599*),
Nevin Karakuş[2](*ORCID: https://orcid.org/0000-0002-1916-7471*)

[*1]Tokat Gaziosmanpaşa Üniversite Hastanesi Tokat, Türkiye
[2]Tokat Gaziosmanpaşa Üniversitesi, Tıp Fakültesi, Tıbbi Biyoloji Anabilim Dalı, Tokat, Türkiye

*Sorumlu yazar e-mail: abdullahkarabey58@hotmail.com

Özet

Bu araştırma sağlık profesyonellerinin genetik ve genetik uygulamalara yönelik tutum ve anlayışlarının belirlenmesi amacı ile yapılmıştır.
Araştırma tanımlayıcı ve kesitsel olarak yürütülmüştür. Bu araştırma Türkiye'de bulunan bir üniversite hastanesinde görev yapan 350 sağlık profesyoneli ile yapılmıştır. Araştırmada veriler, tanıtıcı özellikler formu ve Genetik ve Genetik Uygulamalarına Yönelik Tutum ve Anlayış Ölçeği kullanılarak toplanmıştır. Verilerin değerlendirilmesinde Student t testi ve Anova testi kullanılmıştır.
Bu araştırmada katılımcıların %54.90'ının kadın olduğu, %79.70'inin 18-30 yaş aralığında olduğu, %51.43'ünün bekar olduğu, %17.70'inin idari birimde görev yapığı saptanmıştır. Yine katılımcıların %33.10'unun mesleğinin ebe/hemşire olduğu, %49.40'ının eğitiminin ön lisans/lisans düzeyinde olduğu, %50.00'sinin 5-10 yıl süre ile çalıştığı ve %61.70'inin genetik ve genetik uygulamalarına yönelik eğitim almadığı bulunmuştur. Sağlık profesyonellerinin Genetik ve Genetik Uygulamalarına Yönelik Tutum ve Anlayış Ölçeği puan ortalamaları ile cinsiyet, yaş, medeni durum, eğitim durumu, görev yapılan birim, meslek ve genetik konusunda eğitim alma durumu arasında istatistiksel olarak anlamlı fark olduğu belirlenmiştir ($p<0.05$).
Bu araştırmanın sonuçlarına göre sağlık profesyonellerinin genetik uygulamalarına yönelik tutum ve anlayış düzeylerinin orta düzeyde olduğu ve cinsiyet, yaş, medeni durum, eğitim durumu, görev yapılan birim, meslek ve genetik konusunda eğitim alma durumu ile istatistiksel olarak anlamlı bir ilişki olduğu bulunmuştur.

Anahtar Kelimeler: sağlık, genetik, tutum, anlayış, sağlık profesyonelleri

DRAFT

ORAL PRESENTATION

In silico comparison of the binding energies of some antiviral plant compounds to non-structural proteins of SARS-COV-2 virus

Fadime Taşdemir[1*] (*https://orcid.org/0009-0004-1953-0789)* , Esma Eryılmaz Doğan[2] (*https://orcid.org/0000-0001-6809-7513*)

[*1] University of Selçuk, Faculty of Technology, Department of Biomedical Engineering, Konya, Turkey.
[2] University of Selçuk, Faculty of Technology, Department of Biomedical Engineering, Konya, Turkey.

*Corresponding author e-mail: eeryilmaz@selcuk.edu.tr

Abstract

With the Coronovirus disease-2019 (Covid-19) showing its effect all over the world, a pandemic was declared by the World Health Organization in March 2020. The lack of a specific treatment method for the disease has led all researchers to seek new drug molecules that can be curative or inhibit the activity of the virus. With the urgent need for a specific antiviral drug, Computer Aided Drug Design (CADD) technology gains more attention due to its time and cost effective characteristic. Demonstrating the structure of the virus with experimental methods accelerated CADD processes even more. Researchers performed molecular docking with different herbal compounds using the AUTODOCK program to inhibit the main non-structural protein Mpro of the virus, compared the docking results with the existing antiviral drugs, and made inferences on the effectiveness of plant compounds on the virus function. After the SARS-CoV-2 virus enters the human body, it can transforms and reproduce itself. During this replication process, the virus expresses itself with different non-structural proteins and those are used as important targets in process of therapy. The most important of these are 3CLpro, PLpro, RNA polymerase RdRp, endoribonuclease, 2′-O-methyltransferase, and helicase.
In the current study, some herbal compounds known to have antiviral effects will be computationally investigated using all non-structural proteins of the SARS-CoV2 listed above. The important binding parameters like energy, and bonds will be examined for recommendation of further experimental investigation of covid-19 therepatics.

Keywords: CADD, covid-19, docking, non-structural protein, binding enegy

DRAFT

ORAL PRESENTATION

Removal of Copper (II) from Mining Waste Water by Adsorption onto Activated Carbons Produced from Hazelnut Shell

Pınar BOZBEYOĞLU (Orcid ID: 0000-0002-3704-2701)

Gümüşhane University, Gümüşhane Vocational School, Electronics and Automation Department, Gümüşhane, Turkey

pinarbozbeyoglu@gumushane.edu.tr

Abstract

Mining wastewater is one of the most important environmental problems today with the heavy metal ions they contain. Waste waters containing heavy metal pollution are generally acidic waters with low BOI (Biochemical oxygen demand) value.Various processes such as mineral processing waste disposal, illegal mining, domestic waste disposal and others result in the release of heavy metals into waters.
One of the heavy metals that cause problems in terms of environmental pollution is copper. Copper (Cu) metal is commonly found in metal and metal plating industry wastewater. Although metal industry wastewater is low in quantity, they are toxic wastes. For this reason, industrial wastewater must be treated before being discharged into the receiving environment.
In this study, the amount of Cu in wastewater of the mining operation in Gümüşhane was determined. Heavy metal pollution was eliminated with an adsorbent produced from agricultural biowaste. It has been tried to determine the amount of adsorbent, contact time and treatment efficiency of wastewater at different pHs of activated carbon produced by chemical activation.
As a result of the study, an average of 88.7% efficiency was obtained with activated carbon activated with KOH, while 22,4% copper removal efficiencies were obtained with raw material (hazelnut shell) used without activation. It was determined that the appropriate working pH was 5.5,the amount of adsorbent was 0.05 g.Due to the high ionexchange capacity of activated carbon, the process result on the recovery of Copper bound on activatedcarbon works are in progress. After recovery, processed the reusability of activated carbon will be examined.

Keywords: Activated carbon, Chemical activation, adsorption, wastewater, hazelnut Shell

DRAFT

ORAL PRESENTATION

Eye-on-chip technology and potential use in ophthalmology

Gözde NARİN[1,* *] (*ORCID: 0000-0003-0807-2298*), Onur İNAM[2,3] (*ORCID: 0000-0002-4726-1190*), Mehmet YÜKSEKKAYA[4] (*ORCID: 0000-0002-2665-5799*)

[1]Gazi University, Institute of Health Sciences, Department of Biophysics
[2]Department of Ophthalmology, Edward S. Harkness Eye Institute, Vagelos College of Physicians and Surgeons, Columbia University Irving Medical Center, New York, New York, USA
[3]Gazi University, Faculty of Medicine, Department of Biophysics
[4]Ankara University, Faculty of Engineering, Department of Biomedical Engineering

* Corresponding Author e-mail: gozdeondernarin@gmail.com

Abstract

The eye is one of the most important organs of the body. Therefore, especially laboratory and drug research for ophthalmology is gaining importance. Studies conducted in recent years have shown that a more functional working environment is required in this field. For this reason, there are approaches to apply organ-on-chip technology for ophthalmological studies. This study aimed to compile and examine eye-on-chip applications. In this study, 'Eye-on-chip, microfluidic chip, neuroscience, ophthalmology, biophysics' are utilizedin Web of Science, Scopus, Pubmed and Google Scholar databases in order to reach the relevant articles about nutrition and biophysical effects on eye health.
The organ-on-chip should be manufactured using materials that do useful affect the cellular microenvironment components and provide a regular fluid connection such as polydimethylsiloxane (PDMS), Polymethylmethacrylate (PMMA). In addition, it is very popular to use 3D software to be able to use a chip together the scaffold. Pressure generators and different pumps such as a compressor, a pressure regulator and a manometer, to measure the current pressure value are used. Eye-on-chip models designed for eye studies such as various neurodegenerative diseases and retina-blood barrier have been planned considering all these methods. Studies conducted in the literature includes, designs on cornea, retina, retinal stem cells and organoids, blood-retina barrier, neurodegenerative diseases and they were seemed to have modelled within the scope of eye-on-chip.
Retina-on-a-chip designs promise to reduce reliance on animal models in retinal drug testing. Retina-on-a-chip systems can be used to study the effects on human retinas before starting clinical trials of new drugs. In the future, retina-on-chip model could have a huge impact on drug discovery and safety testing.

Keywords: Eye-on-chip, microfluidic chip, neuroscience, ophthalmology, biophysics.

ORAL PRESENTATION

Relationship between metabolizable protein supply with production, performance and health parameters in early-lactation dairy cows

Halil İbrahim Tosun[1*] (*ORCID: https://orcid.org/0000-0001-5117-0390*), David P. Casper[2] (*ORCID: https://orcid.org/0000-0002-0666-8808*)

[*1]Rumico Livestock Management and Nutrition, Aydın, Turkiye
[2] North Carolina A&T State University, Department of Animal Sciences, Greensboro, NC, USA and Colorado State University, Department of Animal Sciences, Fort Collins, CO, USA

*Corresponding author e-mail: halil.tosun@rumico.com.tr

Abstract

The dairy cow's energy requirements during the close-up period are increased due to fetal development and colostrum production. After calving, the mammary glands demand for glucose increases 3-fold, amino acids 2-fold and fatty acids 3-fold compared to the late lactation period. High protein rations can be fed to fresh cows to maximise milk and milk protein yields, however over feeding protein to meet energy requirements increases ration costs and reduces N efficiency. Most studies have focused on fresh cows evaluating dietary crude protein (CP) whereas this study discusses a recent experiment on metabolizable protein (MP) amounts fed during early lactation on milk yield, blood parameters, and health. Three rations containing 17.0, 16.2, 15.3% CP to supply 114, 107, and 101 g MP/kg were fed in different combinations to 100 multiparous Holstein cows during weeks 1-3 or 4-13 of lactation. All rations were considered deficient in MP. Milk yield, dry matter intake, and feed efficiency were similar for lactating dairy cows fed all treatments except for cows fed 101MP producing lower energy corrected milk and feed efficiency during week 1-3 compared with cows fed 114MP. Milk composition and yields were similar except for cows fed 101MP being lower during week1-3 compared cows fed 114MP. Body weight, body condition score, and disease incidences were similar for cows fed all treatment combinations. Serum concentrations of total fatty acids, albumin, and aspartate aminotransferase were similar for cows fed all ration combinations, however, serum β-hydroxybutyrate concentrations were lower in cows fed 101MP during the first 3 weeks of lactation indicating reduce ketosis incidences. Feeding lower MP ration combinations sustained lactational performance and improved milk N efficiency without negatively affecting the frequency of diseases in dairy cows during early lactation. Reducing ration MP fed to early lactation dairy cows improves N efficiency, reduce ration costs and ammonia excretion.

Keywords: Crude protein, Metabolizable protein, Milk yield, Early lactation, Milk components.

DRAFT

ORAL PRESENTATION

Green synthesis and biomedical applications of selenium nanoparticles via *Rosa damascene* Herrm. leaf extract

Nabeel AHMAD[1]* (*ORCID: https://orcid.org/0000-0001-7525-0950*), Alevcan KAPLAN[2*] (*ORCID: https://orcid.org/ 0000-0001-6738-7527*), Muhammad Nauman KHAN[3] (*ORCID: https://orcid.org/0000-0003-3790-0902*)

[1*]School of Allied Sciences Dev Bhoomi Uttarakhand University Dehradun-248007 Uttarakhand, India

[2*]Batman University, Sason Vocational School, Department of Crop and Animal Production, Batman, Türkiye

[3]Department of Botany, Islamia College Peshawar, 25120 Peshawar, Pakistan

[*]Corresponding author's e-mail: nabeel.biotech@gmail.com; kaplanalevcan@gmail.com

Abstract

In this study, we reported the green synthesis of SeNP*s* (selenium nanoparticles) by simply mixing *Rosa damascena* Herrm. leaf extract and sodium selenite (Na_2SeO_3) salt. This mixture was stirred to give a dispersion of SeNP*s* conjugate. The use of plant extracts in green synthesis has become very popular because no toxic solvents or high temperatures are required. The synthesized PdNP*s* were characterized by UV-Vis spectrophotometer, Fourier transform infrared spectroscopy (FTIR), transmission electron microscope (TEM), and scanning electron microscope (SEM). The synthesis of SeNPs determined by the UV-vis spectrum showed an SPR peak at 315 nm. The prepared nanoparticles had a spherical and polydisperse shape and a size of 40-60 nm. These green synthesized SeNP*s* showed significant activity against both antibacterial and antifungal species. The present results highlight the advantages of the green method for the synthesis of SeNP*s* with potential activities.

Keywords: Antibacterial and antifungal activity; green synthesis; *Rosa damascena*; selenium nanoparticles

DRAFT

ORAL PRESENTATION

Sustainable cultivation of *Anacamptis coriophora* plant in Turkey: Antioxidant properties

Ines Harzli[1*] (*ORCID: https://orcid.org/0000- 0003-4009-2993*), Erdi Can Aytar[1] (*ORCID:* https://orcid.org/0000-0001-6045-0183) and Yasemin Özdener Kömpe[1] (*ORCID:* https://orcid.org/0000-0003-1649-4298)

[*1]University of Ondokuz Mayis, Faculty of Science, Department of Biology, Samsun, Turkey.

*Corresponding author e-mail: inesharzli9@gmail.com

Abstract

Plants are among the most significant sources of natural products. Moreover, plants with medicinal uses play a vital role in numerous indigenous and rural communities worldwide, particularly in areas where government-provided medical services are limited or non-existent, and where herbal remedies are preferred over allopathic medicine. Such practices are especially prevalent in traditional Chinese and Indian medicine. These uses have historical roots, and plants like orchids have been employed for medicinal purposes in nearly every geographic region. In Turkey, for instance, orchid tubers have been used for centuries in the production of salep, a traditional beverage. However, the overharvesting of these tubers poses a threat to the survival of these plant species. This study aimed to determine the antioxidant activity of methanol extracts from the leaves and flowers of *Anacamptis coriophora*, a species grown sustainably from seeds, and to measure the total content of flavonoids, flavanols, tannins, and proanthocyanins. The results indicate that both the leaves and flowers of *A. coriophora* contain a high level of bioactive compounds and exhibit a strong DPPH free radical scavenging effect (IC50 values of 44.77 and 47.79 mg/ml, respectively). This research makes a significant contribution to the understanding of the preservation and sustainability of natural resources, as well as the exploration of their medicinal potential.

Keywords: Biological activity, Orchidaceae, Sustainable agriculture

DRAFT

ORAL PRESENTATION

Au/Fe_2O_3 Catalyst for propargylamines synthesis under Irradiation

Hamiani Zohra [1,2] (https://orcid.org/0000-0001-9014-0661), Berrichi Amina [1,2] (https://orcid.org/ 0000-0003-2586-6428), Bailiche Zohra [1,2] (https://orcid.org/0000-0003-4348-9523), Bachir Redouane [1] (https://orcid.org/0000-0001-6260-2179)

[1] University of Tlemcen, BP 119, 13000, Laboratory of Catalysis and Synthesis in Organic Chemistry, Tlemcen, Algeria.

[2] University of Ain Temouchent, BP 284, 46000, Faculty of Sciences and technologies, Departement of Chemisty, Ain Temouchent, Algeria.

*Corresponding author e-mail: Berrichi.amina@yahoo.fr

Abstract

In organic synthesis, the irradiation by UV light has attracted many researchers, so several reactions were performed using the photocatalytic route as the conversion of alcohol to aldehyde [1]. The UV visible light is considered as an energy source that can reduce the reaction temperature and modify the metal capacity by the excitation of its electrons leading to strong interaction between molecules and metal [2]. Propargylamines are interesting intermediates to synthesize built and heterocycled molecules such as pyrrole, oxazoles, pyrazoles [3]. Furthermore, a number of propargylamines derivatives were used to treat Parkinson's and Alzheimer's disease such as rasagyline [4]. Their preparation method has drawn the attention of a significant number of researchers. In view of the synthetic procedure, the three coupling component of alkyne, aldehyde and amine (A3) was used in the presence of catalysts[5].

In the present study, two types of mesoporous materials were synthesized: the mesoporous Fe_2O_3 was prepared with the hard template using SBA-15, while the mesoporous Au/Fe_2O_3 was prepared by DP-NaOH, and DPU. With the last one, different content of gold were used. Then, the effect of preparation method and nanoparticles size on the catalyst activity was demonstrated. The catalyst which gives the best yield was used also to synthesize propargylamines via AHA coupling under ordinary condition and UV light irradiation.

Keywords:
AHA coupling, Gold catalyst, Mesoporous materials, Nanoparticles, Propargylamine synthesis, UV light irradiation.

Reference

(1) R. Zhang, Y. Liu, Z. Wang, P. Wang, Z. Zheng, X. Qin, X. Zhang, Y. Dai, M.-H. Whangbo and B. Huang, *Appl. Catal. B* (**2019)**, 463-470 *254*.
(2) Z. Jiao, Z. Zhai, X. Guo and X.-Y. Guo, *J. Phys. Chem. C* (**2015)**, 3238-3243 *119*.
(3) K. Lauder, A. Toscani, N. Scalacci and D. Castagnolo, *Chem. Rev.* (**2017)**, 14091-14200 *117*.
(4) M. Sharma, J. Mangas-Sanchez, N. J. Turner and G. Grogan, *Adv. Synth. Catal.* (**2017)**, 2011-2025 *359*.
(5) S. Sadjadi, M. M. Heravi and M. Ebrahimizadeh, *J. Porous Mater.* (**2018)**, 779-788 *25*.

DRAFT

ORAL PRESENTATION

Characterization of bio-char samples obtained from biomass lignite co-gasification

Turgay KAR[1]* (ORCID: https://orcid.org/0000-0002-5543-3394)

*[1]Karadeniz Technical University, Faculty of Science, Chemistry, Trabzon, Türkiye

*Corresponding author e-mail: karturgay1984@gmail.com

Abstract

It is crucial to transform fossil-based energy resources into energy using renewable and clean energy technologies since fossil fuel reserves are depleting and environmental degradation is a real threat. Utilizing coal and lignite effectively through gasification technologies can result in energy efficiency of 40% or more when compared to burning. The gasification of lignite coal using waste biomass (tea bush hazelnut shells), which was also used in this study, is one of the most significant of these technologies. The structure of the bio-char samples was attempted to be clarified through the application of brief analyses, calorific values, SEM, BET, and XRD analyses of bio-char samples produced from co-gasification studies carried out at 700 °C. In the experiments, the value found for bio-char with the highest calorific value was found to be 7141 cal/g. The porous structure and holes on the surface of bio-char created during pyrolysis with volatile matter production will provide more adsorption areas for ions. Both open and closed vesicle formations were observed in the produced bio-char samples due to the gradual release of different volatile compounds as the temperature increased during evaporation at a high heating rate. When the BET analysis results of biochar samples are examined, the adsorption capacity can be improved by increasing the surface areas and pore diameter through activation processes, which can contribute to the production of graphene-type conductive activated carbon. In the XRD analysis spectra of the bio-char samples, peaks belonging to crystalline regular stacking could not be seen and it was determined that the structure was generally amorphous.

Keywords: Gasification, biomass, lignite, bio-char, characterization

DRAFT

ORAL PRESENTATION

Analysis of Monosodium Glutamate in Flavors, Spices and Seasonings by RP-HPLC Method

Aleyna Aydemir[1*] (*ORCID: 0009-0009-7139-7889*),
Yusuf Katrancı[1] (*ORCID: 0009-0001-4211-583X*),
Beray Kızılkaya[2] (*ORCID: 0009-0005-6148-4090*),
Gizem Yıldırım Baştemur[1] (*ORCID: 0000-0002-4634-4525*),
Sabriye Perçin Özkorucuklu[1] (*ORCID: 0000-0001-9778-2034*)

[1]Istanbul University, Faculty of Science, Department of Molecular Biology and Genetics, Istanbul, Turkey.
[2]Istanbul University, Institute of Graduate Studies in Sciences, Programme of Molecular Biotechnology and Genetics, Istanbul, Turkey.

*Corresponding author e-mail: aleyna.aydemir.1183@gmail.com

Abstract

Monosodium glutamate (MSG) is a flavour-enhancing compound that elevates the flavour profiles of various foods. However, concerns have been raised about potential health effects from excessive MSG consumption, including cardiovascular diseases, obesity, diabetes, kidney damage, hypertension, anxiety, and memory impairment. To address these concerns, the Turkish Food Codex has established regulations to protect public health by limiting the amount of MSG in foods to a maximum of 10 g/kg. This study aims to develop a method for analysing MSG content in various food products, such as seasonings, spices, and flavourings sold in Turkish local markets. Chromatographic determination was performed with the gradient program on the Mediterranea Sea 18 (15x0.46 cm, 5µm) column. The optimal method conditions were determined by analysing different formic acid percentages, mobile phase flow rates, column temperatures, and MSG:OPA ratios. The validation parameters of the developed method were examined and it was found that the developed method corresponds to the recommended values. LOD and LOQ values were calculated as 0.075 µg/mL and 0.251 µg/mL, respectively. Recovery % was determined 100.96% in intra-day and 132.22% in inter-day analyses for the precision of the method. The developed and validated method showed that it is accurate and rapid for determining MSG in both qualitative and quantitative analysis. The results compared to the values specified in the Turkish Food Codex Food Additives Regulation and MSG was detected in samples that claim not to contain MSG on their labels.

Keywords: HPLC-DAD, Method validation, Food additives, Monosodium glutamate

DRAFT

ORAL PRESENTATION

Phosphorous acid application on chestnut saplings against chestnut canker caused by *Cryphonectria parasitica*

Deniz Çakar [1*] (*ORCID: https://orcid.org/0000-0002-6269-404X*), Seçil Akıllı Şimşek [2] (*ORCID: https://orcid.org/0000-0002-5055-1391*), Salih Maden [3] (*ORCID: https://orcid.org/0000-0002-7291-4712*)

[*1] Çankırı Karatekin University, Central Research Laboratory Application and Research Center, Çankırı, Turkey
[2] Çankırı Karatekin University, Faculty of Sciences, Department of Biology, Çankırı, Turkey
[3] Ankara University, Faculty of Agriculture, Department of Plant Protection, Ankara, Turkey

*Corresponding author e-mail: denizzcakarr86@gmail.com

Abstract

Sweet chestnut (*Castanea sativa* Mill.) is an essential species in Turkey. Chestnut blight or canker caused by *Cryphonectria parasitica* (Murrill) Barr is one of the most significant diseases affecting this tree. In this study, the efficacy of applying phosphorous acid (PA) to the foliage was examined in the control of chestnut canker caused by *C. parasitica* on chestnut saplings under field conditions. An isolate of *C. parasitica* for artificial inoculations were selected. Foliage treatment of PA which is the only practical option for saplings was employed onto ten healthy chestnut saplings for each treatment group. Effect of PA was determined in two ways. The initial treatment for the first group involved spraying the chemical (4 mL/L of Agrifos) onto the saplings, followed by inoculation with the pathogen after one weeks. For the second group, the pathogen was inoculated first, followed by the application of the chemical one week later. Additionally, positive control was conducted by placing a culture disk of *C. parasitica* without any treatment. After 28 and 35 days from inoculations, evaluation of the treatment was assessed by measuring the lengths of the cankers. Foliage application of PA done on two ways did not provide any protection against chestnut canker caused by *C. parasitica*.

Keywords: chestnut blight, plant protection, foliage treatment

DRAFT

ORAL PRESENTATION

Enhancing Environmental Sustainability Using MIL-88 MOFs Photocatalysts for RO16 Dye Degredation

Elif Özcan (*ORCID:* https://orcid.org/0000-0002-4375-2320)

[1]Department of Chemistry, Gebze Technical University, Gebze, Kocaeli 41400, Türkiye

Abstract

Extensive research into cutting-edge materials and technology has been prompted by rising worries about environmental contamination and the urgent need for sustainable wastewater treatment techniques. Photocatalysis, a versatile and environmentally friendly approach, has emerged as a promising solution for the removal of persistent organic pollutants from aqueous environments. In particular, metal-organic frameworks (MOFs) have garnered attention for their unique properties, including high surface areas, tunable structures, and remarkable photocatalytic potential [1]. One such class of MOFs, MIL-88, has shown significant promise as efficient photocatalysts in various environmental applications. MIL-88 MOFs are made of metal clusters that are linked together by organic linkers. They provide a platform for designing and fine-tuning photocatalytic properties to fit specific needs. Their structural diversity, combined with the ability to incorporate different metal ions and functional groups, presents an intriguing opportunity to address the persistent challenge of organic dye removal from wastewater [2]. Among the numerous organic dyes commonly found in industrial effluents, RO16 (Reactive Orange 16) stands out due to its widespread use in the textile, leather, and dyeing industries. RO16 dye is known for its persistence in aquatic ecosystems and its adverse effects on human health, making its efficient removal a priority in environmental remediation efforts [3]. This study explores the potential of MIL-88 MOFs as advanced photocatalysts for the degradation and removal of RO16 dye from aqueous solutions. The aim of this study contribute to the development of efficient and eco-friendly dye cleanup processes by harnessing solar energy and utilizing the unique properties of MIL-88 MOFs. In this investigation, MIL-88 type MOFs as photocatalysts, namely MIL-88-A, MIL-88-B, and NH_2-MIL-88B MOFs were evaluated for photocatalytic dye degradation. The findings presented herein offer insights into the practical applications of MIL-88 MOFs in wastewater treatment, emphasizing their role in enhancing environmental sustainability and addressing the pressing challenges of organic pollutant removal.

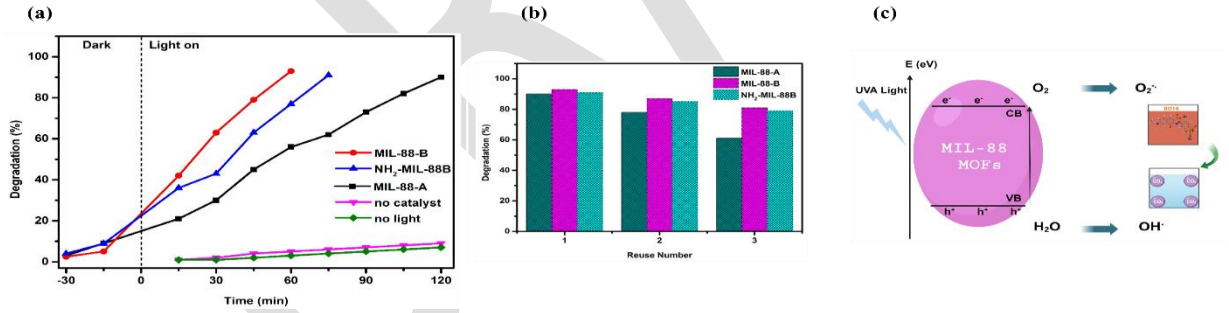


Figure 1. (a) Photocatalytic degradation rate of RO16. (b) Reuse cycle for RO16. (c) Photocatalytic mechanism of dye degradation process.

Keywords: MIL-88 MOFs, photocatalysis, RO16 dye, wastewater treatment, environmental sustainability, pollutant removal.

References

[1] Borges, M. E., de Paz Carmona, H., Gutiérrez, M., Esparza, P. 2023. Photocatalytic Removal of Water Emerging Pollutants in an Optimized Packed Bed Photoreactor Using Solar Light. *Catalysts*, *13*(6), 1023.
[2] Xu, W. T., Ma, L., Ke, F., Peng, F. M., Xu, G. S., Shen, Y. H., Yuan, Y. P. 2014. Metal–organic frameworks MIL-88A hexagonal microrods as a new photocatalyst for efficient decolorization of methylene blue dye. *Dalton transactions*, *43*(9), 3792-3798.
[3] Rawat, D., Mishra, V., Sharma, R. S. 2016a. Detoxification of azo dyes in the context of environmental processes. *Chemosphere*, *155*, 591-605.

ORAL PRESENTATION

In vitro effects of newly synthesized propane sulfonyl hydrazone derivatives on rat myometrial contractions

Esra Sumlu[1*] (*ORCID: https://orcid.org/0000-0002-5004-5958*), Ummuhan Ozdemir Ozmen[2] (*ORCID: https://orcid.org/0000-0001-9161-9367*), Kaniye Zeynep Çalışkan Sak[3] (*ORCID: https://orcid.org/ 0000-0003-0847-1168*), Ayça Yavuz[3] (*ORCID: https://orcid.org/ 0000-0002-7136-2506*), Esra Bilen Ayan[2] (ORCID: https://orcid.org/0000-0001-9194-5461, Selim Kutlu[4] (*ORCID: https://orcid.org/ 0000-0001-9257-4797*)

[1]KTO Karatay University, Faculty of Medicine, Department of Medical Pharmacology, Konya, Turkey.
[2]Gazi University, Faculty of Science, Department of Chemistry, Ankara, Turkey.
[3]KTO Karatay University, Faculty of Medicine, Department of Physiology, Konya, Turkey.
[4]Necmettin Erbakan University, Faculty of Medicine, Department of Physiology, Konya, Turkey.

*Corresponding author e-mail: esra.sumlu@karatay.edu.tr

Abstract

This study aimed to investigate the potential of newly synthesized sulfonyl hydrazone (SH) derivatives to modulate contractions in isolated rat myometrium. For this purpose, the structures of SH compounds were characterized by elemental analyses and spectroscopic methods. Virgin female rats were obtained from KONUDAM Experimental Animal Research Centre (Konya, Turkey). Longitudinal myometrial strips, rapidly isolated post-decapitation, were rapidly inserted into a tissue bath containing Krebs' solution to monitor spontaneous contractions. Separate strips from each rat were treated with two different compounds. Control contractions were initially recorded, followed by the assessment of SH effects at different concentrations (0.5-2 mg/ml) over 10-minute intervals. Despite the tendency of all SH derivatives to decrease contraction, SH 2.2 (4-diethylaminobenzaldehydepropanesulfonylhydrazone) at 2 mg/ml significantly reduced both contraction frequency and amplitude ($p<0.01$) compared to control. No statistically significant differences were found with SH 1.2 (4-nitrobenzaldehydepropanesulfonylhydrazone) and SH 3.2 (2-hydroxy-5-chloro-benzaldehydepropanesulfonylhydrazone) on spontaneous contractions. Following the determination that SH 2.2 was efficacious among the compounds, the effect of SH 2.2 on oxytocin-induced contractions was examined. SH 2.2 decreased the frequency of oxytocin-induced contractions ($p<0.05$) dose-dependent but did not significantly change the amplitude. Taken together, these findings indicate that SH 2.2 may effectively regulate uterine contraction activity. This could potentially be a promising therapeutic target for abnormal uterine contractions. Further research is needed to understand the effects of sulfonyl hydrazones on uterine contractions.

Keywords: Sulfonyl hydrazone derivates, Myometrium contractions, Tissue bath

DRAFT

ORAL PRESENTATION

Scale-up expression of *E. coli* for the production of recombinant super folder green fluorescent protein (sfGFP) using inducible system

Hülya Kuduğ Ceylan*1 (*ORCID: https://orcid.org/0000-0003-0365-2760*)

*1 Tokat Gaziosmanpaşa University, Faculty of Pharmacy, Department of Basic Pharmaceutical Sciences, Tokat, Turkey.

*Corresponding author e-mail: hulya.kudug@gop.edu.tr

Abstract

A superior Green Fluorescent Protein (GFP) mutant, known as superfolder GFP (sfGFP), is more soluble, faster folding, and is the brightest of the known GFP mutants. GFP mutants have been widely used as visual marker proteins in developmental and cell biology studies. A superior Green Fluorescent Protein (GFP) mutant, known as superfolder GFP (sfGFP) (GenBank accession no. HI069813.1) was developed, which has superior features among GFP mutants, such as higher solubility, brighter fluorescence, faster folding, and higher resistance to denaturants such as urea and formamide. Therefore, an efficient overexpression method of sfGFP to obtain sufficient amount of protein is required for further investigations. In this regard, the current study focused upon production of recombinant sfGFP in a large scale in inducible pBAD *E. coli* expression system using bioreactor. *E. coli* pBAD expression vector that inserted sfGFP gene transformed to *E. coli* BL21-AI competent cells for expression of target protein by heat shock. Transformed cells cultured in bioreactor includes 3L LB triple medium. Protein expression induced with arabinose. Purification of recombinant sfGFP was performed with affinity chromatography. The protein is engineered with 6xHis-tag on the N-terminus, which can be used for purification/removal by using Ni++ beads easily. Recombinant protein analyzed SDS-PAGE and UV spectroscopy. mCherry was produced recombinantly in bioreactor in high yield (12,8 g protein/L culture) by the induction of arabinose concentration (0.04 %) for 4 hours induction result observed high levels of fluorescent protein expression. Therefore, this method provides a quick, high-yield production route for any soluble fluorescent protein that is needed for further research.

Keywords: superfolder Green Fluorescent Protein, *E. coli*, arabinose induction, pBAD expression

DRAFT

ORAL PRESENTATION

Designing novel inhibitors targeting Nsp16 protein for SARS-CoV-2

Onur ÖZER[1] (*ORCID: https://orcid.org 0000-0002-8329-2525*),
Sefer BADAY*[2,3] (*ORCID: https://orcid.org/0000-0002-2913-6687)*

[1]Istanbul Technical University, Faculty of Science and Letters, Molecular Biology and Genetics Department, Istanbul Türkiye
[2] Istanbul Technical University, Informatics Institute, Applied Informatics Department, Istanbul Türkiye
[3] Istanbul Technical University, Faculty of Computer Informatics and Engineering, Artificial Intelligence and Data Engineering Department, Istanbul 34469, Türkiye

*badays@itu.edu.tr

Abstract

Non-structural protein 16 (Nsp16) is an important target for SARS-CoV-2. It faciliates capping of viral mRNA to disable the recoginition of the viral mRNA by host immune system. Nsp16 transfers a methyl group from S-adenosylmethionine (SAM) to 2' hydroxyl of ribose sugar in the viral mRNA. Thus, inhibiting methyl transferase activity could be a way for the treatment of SARS-CoV-2. Therefore, we screened 400.000 molecules drug-like molecule obtained from ZINC database using AutoDock Vina program. Top-ranked 5000 molecules were redocked using Schrödinger glide program. Top 200 molecules having highest docking score were selected and then clustering was applied to prevent working on very similar molecules. From 200 molecules we obtained 165 clusters and selected cluster centroids for further operation. After that ADME/T analysis was performed on these selected molecules. 100 ns long MD simulations of 10 molecules predicted to have good human oral absorption properties were performed. We selected 5 molecules exhibiting the highest stability in the MD simulation trajectories. RMSD fluctuation of some molecules were less than ~1.0 Å. Based on the analysis of the trajectories, residues Leu 6898, Cys 6913, Met 6929, Tyr 6930, Pro 6932 and Phe 6947 form hydrophobic interactions with ligands. Also, residues Asp 6897, Asp 6928 and Lys 6968 make hydrogen bonds with molecules of the interests.

Keywords: Virtual screening, MD simulations, Nsp16, SARS-CoV-2, Docking

DRAFT

ORAL PRESENTATION

Protected viticulture for sustainable grape production to cope with the adverse effects of climate change

Bushra Hussain* (*https://orcid.org/*0009*-0000-6273-135X*), Ali Sabir (*https://orcid.org/0000-0003-1596-9327)*

*University of Selcuk, Faculty of Agriculture, Department of Horticulture, Konya, Türkiye

*Corresponding author e-mail: bushrahussain159@gmail.com

Abstract

Grapes (*Vitis vinifera* L.) are among the most commonly cultivated horticultural crops on the earth covering an area of 7.5 million hectares with around 70 million tons production. Grapes being highly nutritious are majorly utilized in making beverages or used as fresh and raisin. In today's world, the most concerned topic in agriculture is continuous change in climatic conditions leading to several negative social, ecological, economical and biological problems. Grapes are delicate fruits, their vines are sensitive towards extremes in temperature, water, heat, duration and intensity of light exposure as well as carbon dioxide and humidity alterations. Hence, one can shift to drought and heat tolerant varieties and certain training and pruning methods suitable to the prevailing environmental conditions. Vineyard management practices, including farm mechanization can be amended for improving the gains, while reducing the input. Control over temperature and proper water supply are needed to help the vines against stress. Wind machines in areas facing speedy, chilling winds mix the cold and warm air, reducing the freezing injuries to plants. An old method is to cover the young plant with soil mound to conserve the heat but this can expose the plant to microbes, causing vine rots and damaging their fragile roots. In areas where summer heat is insufficient for conversion of vegetative buds into reproductive ones, it is needed to conserve the heat by covering plants with some insulator like polyethylene sheets. Mulch is also used to conserve soil moisture in drylands. Also, certain organic amendments can protect the plant from under-nutrition. Climate change renders the plants more sensitive for certain disorders so we use some pesticides or growth promotors to counteract those abnormalities. The best approach is to combine all the environment-friendly sustainable strategies and apply a joint integrated crop management (ICM) approach for successful viticulture.

Keywords: Viticulture, climate change, protected agriculture, environmental stress factors

DRAFT

ORAL PRESENTATION

Investigation of biotransformation of a chalcone derivative by *Aspergillus glaucus*

Semra Yılmazer Keskin[1*], Ayça Meral[1], Yavuz Derin[1], Ridanur Öztürk[1], Ahmet Tutar[1]

[*1]Sakarya University, Faculty of Science, Chemistry Department, Sakarya, Turkey

*syilmazer@sakarya.edu.tr

Abstract

1,3-diaryl-2-propene-1-ones and chalcones have more than one different biological activity. These compounds are formed by natural means or are easily synthesized [1]. The flavonoid-type phenolic phytochemicals called biosynthesized and open-chain flavonoids are chalcones [2]. They are considered biosynthetic precursors of flavonoids. Chemically, chalcones are usually α, β-unsaturated ketones consisting of two aromatic rings (A and B rings) bonded by a three-carbon alkenone unit, but they also contain some saturated ketones, mostly known as dihydrochalcones. instead of a three-carbon alkenone unit, there is a three-carbon alkanon unit. Decoction of one or more phenolic hydroxyl functions is present at every point between naturally occurring chalcones, and prenyl and geranyl substituents on aromatic rings are also commonly observed. There are thousands of naturally occurring chalcones in the current literature [3]. In this study, biotransformation of chalcone derivative which was synthesized with acetophenone and aldehyde using *Aspergillus glaucus* 200914 was investigated. The chalcone derivative compound was incubated with *A. glaucus* 200914 for five days in a shaker. Metabolite obtained after the biotransformation experiments was examined by thin layer chromatography. The structure of the resulting metabolite was determined by the spectra of ^{1}H NMR, and ^{13}C NMR. Accordingly, it was determined that the chalcone derivative was hydrogenated.

Keywords: Chalcone, biotransformation, *Aspergillus glaucus*

References:

[1] Dhar, D.N. “The Chemistry of Chalcones and Related Compounds”, Wiley-İnterscience: New York, (1981).
[2] Nahar, L., Sarker, S.D., “Chemistry for Pharmacy Students: General, Organic and Natural Product Chemistry”, 2nd ed.; Wiley and Sons: Chichester, UK, (2019).
[3] Zhuang, C., Zhang, W., Sheng, C., Zhang, W., Xing, C., “Chalcone:A privileged structure in medicinal chemistry”, Chem. Rev. (2017), 117, 7762–7810. [CrossRef]

DRAFT

ORAL PRESENTATION

Biotransformation of (E)-1-phenyl-3-(p-tolyl)prop-2-en-1-one by *Aspergillus glaucus*

Semra Yılmazer Keskin[1*], Ayça Meral[1], Ayşin Tuğçenur Altundal[1], Yavuz Derin[1], Eda Nur Aydın[1], Ahmet Tutar[1]

[*1]Sakarya University, Faculty of Science, Chemistry Department, Sakarya, Turkey

* syilmazer@sakarya.edu.tr

Abstract

The chemistry of chalcones has provided intensive scientific studies around the world. The name "Chalcones" was given by Kostanecki and Tambor [1]. Another well-known name for chalcones is benzylidene acetophenone or benzyl acetophenone. In chalcones, two aromatic rings and an aliphatic are connected to each other by three carbon chains. Chalcones (trans-1, 3-diaryl-2-propene-1-ones) are α, β-unslaked ketones comprise of two aromatic rings (ring A and B) with diverse substituents. The rings are connected to each other by a highly electrophonic three-carbon α, β-unslaked carbonyl system, which estimates a linear or almost planar structure [2, 3]. They include the ketomethylene group (-CO-CH =CH-). Chalcones have conjugated double bonds and there is a π-electron system completely embedded in both benzene rings. In chalcones, the purpose of use of intermediates is for decoction of compounds with therapeutic value. [4, 5]. Chalcones have been described as different compounds associated with various biological activities. In this study, biotransformation of (E)-1-phenyl-3-(p-tolyl)prop-2-en-1-one with *Aspergillus glaucus* 200914 fungus culture was performed. The chalcone compound was incubated for five days in a shaker. Metabolite was examined by thin layer chromatography. The structure of the resulting metabolite was determined by the spectra of ^{1}H NMR, ^{13}C NMR. Accordingly, the hydrogenation of the chalcone was detected.

Keywords: Chalcone, biotransformation, *Aspergillus glaucus*

References:

[1] Yerragunta, V., T. Kumaraswamy, D. Suman, V. Anusha, Patil, P., Samhitha, T., "A review on Chalcones and its importance", Pharmatutor Pharmacy Infopedia, Review Artical, ISSN:2347– 7881, (2013),1 (2), 54-59.
[2] Awasthi, S.K., Mishra, N., Kumar, B., Sharma M, Bhattacharya A, Mishra LC, Bhasin VK, "Potent antimalarial activity of newly synthesized substituted chalcone analogs in vitro", Medicinal Chemistry Research., (2009), 18, 407-420.
[3] Lim, S.S, Kim, H.S, Lee, D.U., "In-vitro antimalarial activity of flavonoids and chalcones", Bulletin of the Korean Chemical Society, (2007), 28:2495-2497.
[4] Straub, T.S., "Epoxidation of α, β-unsaturated ketones with sodium perborate", Tetrahedron Lett., (1995), 36 (5), 663-664.
[5] Bergman, E.D., Ginsibm, L., Pappo, R., "The Michael Reaction", Org. React. (2011), 10, 179.

DRAFT

ORAL PRESENTATION

Production and characterization composite films composed of chitosan/silk sericin/silver nanoparticles for potential antibacterial applications

Zeynep Niyaz[1] (*https://orcid.org/0009-0009-5761-7646*),
Zehra Gün Gök[1*] (*ORCID: https://orcid.org/0000-0001-6426-0395*)

[1] Kırıkkale University, Faculty of Engineering and Natural Sciences, Department of Bioengineering, Kırıkkale, Turkey.

*Corresponding author e-mail: zehragungok@kku.edu.tr

Abstract

The aim of this study is to obtain composite polymeric film surfaces consisting of chitosan (CHT), silk sericin (SS) and silver nanoparticles (AgNPs). In the study, firstly, composite polymeric film surfaces consisting of CHT/SS polymers containing different amounts of AgNPs were obtained by solvent casting method. For the synthesis of CHT/SS membranes, CHT and SS solutions were prepared separately. To prepare SS solution, 2 grams of SS was added to 100 mL of hot water and the pH of the mixture was increased above 7 with NaOH to increase the solubility of SS. To prepare CHT solution, 2 grams of CTH were taken and dissolved in 100 mL of 1% acetic acid by stirring for 24 hours. First of all, for the synthesis of AgNPs, 10 mM silver nitrate and SS solution were mixed in equal volumes and the mixture was heated at 100 ºC for 2 hours. At this stage, a color change was observed and the color of the silver nitrate-SS mixture turned dark brown. With this color change, it was confirmed that AgNPs were synthesized. Afterwards, SS-AgNPs solution and CHT solution were mixed in different proportions and 10 mL of this mixture was taken and transferred to petri dishes and the petri dishes were incubated at 37 ºC for 24 hours to obtain CHT/SS/AgNPs composite film surfaces by solvent casting method. The chemical structure of the film surfaces was examined by fourier transformed infrared spectroscopy (FTIR), it was confirmed that the membranes consisted of CHT and SS. The morphological structure of the membranes was examined by field emission scanning electron microscopy (FESEM) analyses, and the presence of AgNPs in the structure of films containing AgNPs was proven by energy dispersive spectroscopy (EDS) analyzes taken on FESEM images. The thermal properties of membranes were investigated by thermogravimetric analysis (TGA) and it was confirmed that AgNPs addition increased the thermal stability of the membranes. The antibacterial and cytotoxic properties of membranes will also be examined in our ongoing studies. The resulting film surfaces will have the capacity to be used in medical applications such as wound dressing or antibacterial packaging materials.

Keywords: Chitosan, Silk sericin, Silver Nanoparticles, Membrane, Wound Dressing, Antibacterial materials

DRAFT

ORAL PRESENTATION

Darbepoetin alpha has a potent anxiolytic effect on the neuroinflammation-induced rat model

Hasan Çalışkan[1] (ORCID: https://orcid.org/0000-0002-3729-1863), Deniz Önal[1] (ORCID: https://orcid.org/0000-0002-9604-4539), Erhan Nalçacı[2](ORCID: https://orcid.org/0000-0003-4850-4745)

[1]Balıkesir University, Medicine Faculty, Department of Physiology, Balıkesir, Turkey
[2]Ankara University, Medicine Faculty, Department of Physiology, Ankara, Turkey

Abstract

Erythropoietin is a hormone which has protective activity. The aim of this study was to investigate the possible anxiolytic effect of Darbepoetin alpha (DEPO), a derivative of erythropoietin, a neuroinflammation model induced by lipopolysaccharide (LPS). Adult male 40 Wistar albino rats were divided into four groups (n=10), (Control; LPS ; DEPO; DEPO+LPS). Rats were treated with 5 mcg/kg DEPO once a week for four weeks, followed by neuroinflammation with 2 mg/kg LPS. Anxiety-like behaviors were assessed by elevated plus maze, open field, and light dark box test. TNF-α, IL-1β, brain-derived growth factor (BDNF), and serotonin levels were analyzed by ELISA in the prefrontal cortex and striatum. Cortisol and total oxidant/antioxidant (TAS/TOS) levels were analyzed in serum and brain regions. Total/free thiol levels were analyzed in serum. Increased harderian gland secretion in stress was analyzed observationally according to an existing scale. Anova and Tukey tests were performed statistically.
Anxiety-like behavior decreased in the both naive rats group and the LPS group. In the neuroinflammation group, serotonin and BDNF decreased, whereas cortisol, TNF-α, and IL-1 β increased significantly compared to the control group ($p<0.001$). Serotonin and BDNF levels increased in the neuroinflammation group treated with prophylactic DEPO compared to the neuroinflammation group ($p < 0.01$), while cortisol, IL-1β, and TNF-α decreased ($p<0.0001$). While there was no significant difference between all groups in terms of TAS, TOS increased in prefrontal cortices and serum after DEPO administration compared to the control group. Free thiol increased in serum after DEPO administration compared to the control group. Harderian gland secretion is increased in the inflammation group. Furthermore, 20 percent mortality occurred in the neuroinflammation group. There were no deaths in the prophylactic DEPO group. DEPO as an erythropoietin derivative has been found to have anxiolytic effects in terms of many physiological mechanisms in the neuroinflammation model.

Keywords: Anxiety-like behaviors, BDNF, Darbepoetin alfa, Neuroinflammation, Serotonin

DRAFT

ORAL PRESENTATION

High-intensity exercise effect on self care-related grooming behaviors in rat splash test

Hasan Çalışkan[1] (ORCID: https://orcid.org/0000-0002-3729-1863) , Dilara Karakaya[2] (ORCID: https://orcid.org/0009-0002-6868-1909) , Seda Koçak[3] (ORCID: https://orcid.org/0000-0003-1183-4847), Göktuğ Ömercioğlu[2] (ORCID: https://orcid.org/0000-0002-4387-4598), Metin Baştuğ[2] (ORCID: https://orcid.org/0000-0002-8008-6515)

[1]Balıkesir University Medicine Faculty, Department of Physiology, Balıkesir, Turkey
[2]Ankara University Medicine Faculty, Department of Physiology, Ankara, Turkey
[3]Kırşehir Ahi Evran University Medicine Faculty, Department of Physiology, Kırşehir, Turkey

Abstract

The aim of this study is to examine the self-care and self-care-related grooming behavior patterns of high-intensity interval exercise. 8-week-old Wistar Albino 16 male rats were divided into Control (n=8), and Exercise (n=8). The animals in the Exercise group was exercised for 38 minutes a day, five days a week, for 8 weeks. The animals were then subjected to open field test and splash test, and the behaviors were video recorded. Splash test was conducted by spraying a 10% sucrose solution on the dorsal body surface of the rats for 5 minutes. Total distance traveled in the open field test, grooming time/frequency, and latency were analyzed in the splash test. Student t-test and Shapiro Wilk test were used as statistical tests. In the exercise group, splash-induced grooming behavior increased significantly in terms of duration and frequency ($p<0.001$). High-intensity intermittent exercise increased self-care behaviors. No significant difference was observed in the latency of grooming ($p>0.05$). In the open field test, the total distance traveled, which is a locomotor activity parameter, did not change between the groups ($p>0.05$). Intermittent high-intensity exercise increased self-care behaviors. This Exercise type can be a new option to improve depression-associated behavior. More studies are needed.

Keywords: Exercise, grooming behavior, self-care, splash test

DRAFT

ORAL PRESENTATION

Pd-modified waste based activated carbon materials as a catalyst in hydrodeoxygenation reaction

Yusuf Osman DONAR (*https://orcid.org/0000-0003-1672-4540)*

Ankara University, Faculty of Science, Department of Chemistry, Ankara, TURKEY

yodonar@ankara.edu.tr

Abstract

Bio-oil is an important product, obtained from thermal conversion (pyrolysis or liquefaction) of biomass. One of the obstacle on the usage of bio-oil as a potential transportation fuel is its higher oxygenated compund content. Hydrodeoxygenation of bio-oil is the very attractive method for the removal of oxygenated compounds on the last years. HDO catalyst is mainly consist of metal that serves as the active site and support material. In this two-component system, the metallic region activates molecular hydrogen and catalyzes the hydrogenation reaction. In addition the support material catalyzes dehydration, hydrogenation, isomerization, cracking reactions as well as provides substrate adsorption. For this purpose, there are studies conducted with many different metals immobilized on a support material. However, in order achieve high activity, noble metals must be well dispersed on the support material. Otherwise, agglomeration or post-reaction sintering cause the activity decreasing. Due to the large surface area and adjustable pore size activated carbons serve as a perfect support material for HDO reactions. Waste based carbon materials can be prepared via different techniques such as pyrolysis or hydrothermal/ionothermal carbonization. The surface areas of carbon materials obtained with these techniques can be increased to over 1000 m^2/g with a further activation methods. In this study carbon materials obtained from corn leaf by hydrothermal carbonization activated in the presence of KOH as an activation agent. Obtained activated carbons decorated with Pd particles via wet impregnation method. The activity of prepared catalyst was tested in hydrodeoxygenation reaction of carboxylic acids using batch type high pressure and high temperature autoclave.

Keywords: waste utilization, carbon, hydrodeoxygenation, catalyst

DRAFT

ORAL PRESENTATION

Accelerated solvent extraction of rosehip seeds and determination of antioxidant capacity, total phenolic and flavonoid content

Rabia Nur Bozkurt[1,2*] (*ORCID: https://orcid.org/0000-0002-2034-0737* .), Selin Şahin Sevgili[2](*ORCID: https://orcid.org 0000-0002-9989-9823)*

[*1] Istanbul Health and Technology University, Faculty of Engineering and Natural Sciences, Department of Chemical Engineering, Istanbul, Türkiye.

[2]Istanbul University-Cerrahpasa, Faculty of Engineering, Department of Chemical Engineering, Istanbul, Türkiye.

*Corresponding author e-mail: rabia.bozkurt@istun.edu.tr

Abstract

Phenolic compounds encompass a wide variety of plant substances having an aromatic ring bearing one or more hydroxyl groups. Since all phenolic compounds are aromatic, these compounds are known to have absorption in the UV region. The presence of phenolic compounds in all plant organs makes them a vital component of the human diet [1][2]. An antioxidant is a substance that delays or prevents the oxidation of a substrate [3]. Phenolic compounds show antioxidant activity and prevent oxidative damage to biomolecules through free radical-mediated reactions [4]. Extracts of rosehip (Rosa canina) species, one of the members of the Rosaceae family, are potential phytochemicals for the reduction reaction due to their hydroxyl groups [5]. Solvent-assisted extraction of bioactive compounds from solid plant materials is of great interest in the production of phytochemically rich products. Since conventional extraction methods have long processing times and low extraction yields, environmentally friendly extraction techniques that provide automation, reduce organic solvent consumption, and shorten extraction times have attracted interest in recent years [6]. Since the seeds of this fruit become waste after being used in the food industry, this study focuses on the evaluation of rosehip seeds. Rosehip fruit seeds will be extracted by accelerated solvent extraction, which is one of the advanced extraction techniques. Antioxidant capacity, total phenolic and flavonoid content of the obtained extract will be determined spectrophotometrically.

Keywords: Phenolic compounds, antioxidant, rosehip, accelerated solvent extraction

References:

[1] Harborne, J. B. (1973). Phenolic compounds. In *Phytochemical methods* (pp. 33-88). Springer, Dordrecht.

[2] Alara, O. R., Abdurahman, N. H., & Ukaegbu, C. I. (2021). Extraction of phenolic compounds: A review. *Current Research in Food Science*, *4*, 200-214.

[3] Santos-Sánchez, N. F., Salas-Coronado, R., Villanueva-Cañongo, C., & Hernández-Carlos, B. (2019). Antioxidant compounds and their antioxidant mechanism. *Antioxidants*, *10*, 1-29.

[4] Vuolo, M. M., Lima, V. S., & Junior, M. R. M. (2019). Phenolic compounds: Structure, classification, and antioxidant power. In *Bioactive compounds* (pp. 33-50). Woodhead Publishing.

[5] Jafarirad, S., Mehrabi, M., Divband, B., & Kosari-Nasab, M. (2016). Biofabrication of zinc oxide nanoparticles using fruit extract of Rosa canina and their toxic potential against bacteria: A mechanistic approach. *Materials Science and Engineering: C*, *59*, 296-302.

[6] Maran, J. P., Manikandan, S., Nivetha, C. V., & Dinesh, R. (2017). Ultrasound assisted extraction of bioactive compounds from Nephelium lappaceum L. fruit peel using central composite face centered response surface design. *Arabian journal of chemistry*, *10*, S1145-S1157.

DRAFT

ORAL PRESENTATION

Aurelia aurita türü denizanası venomu yüklü ZrO_2 kaplı $MnFe_2O_4$/aljinat kompozitler: Venom salım kinetiklerinin belirlenmesi

Sümeray Ezgi Ayan[1*] (ORCID: https://orcid.org/0009-0000-7651-6263), Nurçin Killi[2] (ORCID: https://orcid.org/0000-0002-6095-2674), Yasemin İspirli Doğaç[3] (ORCID: https://orcid.org/0000-0001-8616-0280)

[1]Muğla Sıtkı Koçman Üniversitesi, Fen Bilimleri Enstitüsü, Moleküler Biyoloji ve Genetik Anabilim Dalı, Muğla, Türkiye
[2]Muğla Sıtkı Koçman Üniversitesi, Su Ürünleri Fakültesi, Su Ürünleri Temel Bilimleri Bölümü, Muğla, Türkiye
[3]Muğla Sıtkı Koçman Üniversitesi, Muğla Meslek Yüksekokulu, Kimya ve Kimyasal İşleme Teknolojileri Bölümü, Muğla, Türkiye

* sumerayayan@gmail.com

Özet

Kanser tedavisi ile ilgili bilimsel çalışmalar devam ederken, tedavi süreci kemoterapi, radyoterapi ve cerrahi yöntemlerle sürdürülmektedir. Kanser hastalıklarının tedavisine yönelik klasik ilaçlar, kanser hücrelerinin DNA replikasyonunu önleyerek etki gösterirler. Fakat bilimsel verilere göre, ilaçların kanser hücrelerine seçici bir etki yarattığını söylemek güçtür. Çoğunlukla sağlıklı hücrelere de aynı etkiyi gösterdikleri için, alternatif yöntem arayışları, yüzyıllar boyunca geleneksel olarak tedavi amaçlı kullanılan ve doğal kaynaklardan elde edilen biyotoksinlerle antikanser ilaç geliştirilmesi çalışmalarına yönlenmiştir. Bu çalışmalar kapsamında, çeşitli hayvan türlerinin (yılan, akrep, kurbağa, arı ve deniz anemonu gibi) venom proteinleri kansere karşı terapötik potansiyel göstermiştir. Bu bakış açısından yola çıkarak önerilen bu çalışmanın ilk aşamasında, Muğla kıyılarında literatürde dağılım gösterdiği saptanan denizanası türlerinden biri olan *Aurelia aurita* türünün örneklenmesi hedeflendi. Denizanası örneklerinin ekstraksiyonu ile ham venom elde edildi. Bu aşamayla eş zamanlı olarak gerçekleştirilecek diğer aşamada, ZrO_2 kaplı $MnFe_2O_4$/aljinat kompozitler üretildi. Manyetik kompozitlere ham venom enkapsülasyonu gerçekleştirilip, salım deneyleri, farklı metabolik pH'larda ayrı ayrı yürütüldü ve salımlarının kinetik modeli belirlendi. Venom kinetikleri, Peppas, Higuchi, sıfırıncı ve birinci mertebe modellerine ait grafikler çizilerek belirlendi. Hazırlanan venom salım sistemlerinin tüm salım modellerine uyduğu yani polimerik yapılardan oluşan taşıyıcıların salımı geciktirmediği, ani salıma neden olmadığı, salınmayan etken madde miktarının zamanla logaritmik olarak azaldığı, gözenekliliğinin yani yüzey alanlarının yüksek olduğu, salınan etken madde miktarını zamana bağlı olarak sabit tutmayı başardığı görüldü ve venom ile hazırlanan sistemin kontrollü bir salım sistemi sonucuna ulaşıldı. Bu çalışma sırasında sentezlenen ZrO_2 kaplı $MnFe_2O_4$/aljinat manyetik kompozitlerle ilgili literatürde, ilaç veya venom salımıyla ilgili bir çalışmaya rastlanmamıştır. Ayrıca, denizanası venomuyla yapılan herhangi bir salım çalışması da mevcut değildir. Bu yönleriyle bu çalışmada elde edilen verilerin, özgünlüğü yönüyle literatüre önemli katkılar yaptığı düşünülmektedir.

Anahtar kelimeler: Kanser, venom, manyetik kompozit, salım sistemi.

Teşekkür: Bu çalışma MSKÜ-BAP 22/134/07/1/4 no'lu proje ile desteklenmiştir.

DRAFT

ORAL PRESENTATION

Bisfenol-A giderimi için lakkaz yüklenmiş manyetik $CuFe_2O_4$/aljinat boncukların sentezi ve karakterizasyonu

Elif Koçaslan[1*] (ORCID: 0009-0001-7016-2932), Gökmen Arabacı[2*](ORCID: 0000-0002-5848-6117),Yasemin İspirli Doğaç[3] (ORCID: 0000-0001-8616-0280)

[*1] Muğla Sıtkı Koçman Üniversitesi, Fen Fakültesi, Moleküler Biyoloji ve Genetik Bölümü, Muğla, Türkiye
[2]Muğla Sıtkı Koçman Üniversitesi, Fen Bilimleri Enstitüsü, Moleküler Biyoloji ve Genetik Anabilim Dalı, Muğla, Türkiye
[3]Muğla Sıtkı Koçman Üniversitesi, Muğla Meslek Yüksekokulu, Kimya Ve Kimyasal İşleme Teknolojileri Bölümü, Muğla, Türkiye

* gokmen_arabaci@hotmail.com

Özet

Bisfenol-A (BPA), endokrin bozucu bileşiklerden biridir. Biyolojik uzaklaştırma işlemlerinde enzimlerin kullanılması, toksik olmama, yüksek katalitik verimlilik, yüksek substrat spesifikliği ve kısa reaksiyon süreleri gibi avantajlar sunar. Ancak bu enzimlerin çözünür formlarında kullanılması pratik uygulamalarını sınırlandırmakta ve tekrar kullanılamama, denatüre edici ajanlara karşı hassasiyet ve düşük stabilite gibi bazı dezavantajlara sahiptir. Enzim immobilizasyonu bu sınırlayıcı özelliklerin üstesinden gelmek için önemli bir yöntemdir. Çalışmada BPA'nın uzaklaştırılması enzimatik bir işlemle gerçekleştirildi. Lakkaz yüklenmiş manyetik $CuFe_2O_4$/aljinat boncukların sentezi ilk kez gerçekleştirildi ve sentez optimize edilerek, taramalı elektron mikroskobu ile termal analiz yapılarak karakterize edildi. Optimum koşullar altında sentezlenen lakkaz yüklenmiş boncukların sıcaklık, pH, tekrar kullanım ve kinetik özellikleri gibi enzimatik özellikleri serbest lakkazla kıyaslanarak araştırıldı. Lakkazın katalitik performansı immobilizasyondan sonra artarak, aktivite gösterdiği sıcaklık profili genişledi ve alkali bölgede daha iyi aktivite gösterdi. Düşük sıcaklıklarda (30ºC ve altı) serbest lakkaz aktivite göstermezken, immobilize lakkaz % 40 aktivitesini sabit tutmayı başardı. 60ºC'de ise serbest lakkazın aktivitesi %45'e düşerken İmmobilize lakkaz %87 oranında aktive göstermiştir. pH kararlılığı çalışmasında pH 7.5'dan itibaren serbest lakkaz aktivite göstermezken, immobilize lakkaz aktivitesini %76 oranında korudu. Ayrıca, 7 kullanımdan sonra immobilize lakkaz aktivitesi %64 oranında korundu ve BPA giderimi için 120 dakika sonra %50 BPA giderimi sağlandı.

Anahtar Kelimeler: BPA, lakkaz, immobilizasyon, enkapsülasyon

Teşekkür: Bu çalışma TÜBİTAK 2209/A kapsamında desteklenmiştir.

DRAFT

ORAL PRESENTATION

Invertebrate iridescent virus 6 (IIV6)'ya ait helikaz geninin kodon kullanım eğilimi analizi

Yeşim AKTÜRK DİZMAN[1*] (*ORCID: 0000-0003-0185-502X*)

[*1]Recep Tayyip Erdoğan Üniversitesi, Fen Edebiyat Fakültesi, Biyoloji Bölümü, Rize, Türkiye

*Sorumlu yazar e-mail: yesim.akturk@erdogan.edu.tr

Özet

Invertebrate iridescent virüs 6 (IIV6), *Iridoviridae* familyasının *Betairidovirinae* alt familyasında yer alan *Iridovirus* cinsinin tip türüdür. IIV6, 215 açık okuma zinciri (ORF) kodlayan, yaklaşık 212 kbp büyüklüğünde lineer çift zincirli DNA genomuna sahiptir. IIV6 genomu, *Saccharomyces cerevisiae*'nin PIF1 helikaz proteinine homolog bir protein kodlayan açık okuma zinciri (ORF 030L, IIV6 helikaz geni) içerir. IIV6 helikaz geni sadece viral DNA replikasyonunda rol oynamakla kalmaz, aynı zamanda viral konak seçiciliğinde de rol oynar. Bu çalışmada, IIV6 ve 9 referans iridovirüsün helikaz genindeki kodon kullanım eğilimi (CUB), nükleotid içeriklerini, göreli sinonim kodon kullanımını (RSCU), kodonların etkili sayısını (ENC), kodon adaptasyon indeksini (CAI), dinükleotid içeriklerini ve diğer indeksleri karşılaştırarak analiz edilmiştir. Hem nükleotid içeriği hem de RCSU analizi, IIV6'nın helikaz geninde çoğunlukla A ve T ile biten kodonların tercih edildiğini göstermiştir. ENC değeri 36.20 olan IIV6 helikaz geninde düşük bir kodon kullanım eğilimi (CUB) olduğu belirlenmiştir. ENC grafiği, neutrality grafiği ve parity rule 2 grafiği, hem mutasyon baskısının hem de doğal seçilimin IIV6 ve referans iridovirüslerin helikaz genlerinin kodon kullanım eğilimi üzerinde etkisi olduğunu ortaya koymuştur. Kodon adaptasyon indeksi analizi, IIV6 DNA ligaz geninin konakçısına güçlü bir şekilde adapte olduğunu göstermiştir. Ayrıca, correspondence analizi (COA) ve korelasyon analizleri, nükleotid bileşimlerinin, dinükleotid içeriğinin, mutasyon baskısının ve doğal seçilimin helikaz genlerindeki kodon kullanım eğilimini şekillendirdiğini ortaya koymuştur. Bu bulgular, IIV6 helikaz geninin ve referans genlerinin kodon kullanım eğilimini anlamamıza katkıda bulunacak ve bu genlerin temel evrimsel analizi için gerekli bilgileri sağlayabilecektir.

Anahtar Kelimeler: Invertebrate iridescent virus 6, ORF 030L, Helikaz, Kodon kullanım eğilimi

DRAFT

ORAL PRESENTATION

Tıp fakültesi öğrencilerinin genetik okuryazarlık konularına yönelik tutumlarının incelenmesi

Derya Batmaz https://orcid.org/0009-0008-1795-7307, Nevin Karakuş

Gaziosmanpaşa Üniversitesi, Lisansüstü Eğitim Enstitüsü, Tıbbi Biyoloji Anabilim Dalı, Tokat, Türkiye

Patoloji2173@gmail.com

Özet

Genetik okuryazarlık: genetik kavramını anlama, algılama ve bunu yaşamla ilişkilendirilme şeklinde ifade edilir (Acra, 2006). Hızla gelişmekte olan gen teknolojisindeki gelişmelerin sebep olduğu sorunları çözen, sonuç verme kısmına dahil olan ve bunu bir sonraki nesillere aktaracak genetik okuryazar bireylerin yetiştirilmesi savunulmaktadır (Tsui & Treagust, 2010).

Genetik ve genetik uygulamalar kapsamında alınan görüşlere bakıldığında, ilgili literatür kapsamında genetik okuryazarlığın, genelde genetik uygulamalara yönelik bilgi ve tutum ölçmek adına yapıldığı görülmüştür (Bowling , vd., 2008; Cebesoy ve Öztekin, 2018; Henneman, Timmermans , & Wal, 2006; Shaw III & Bassi; Smith, Wood, & Knight, 2008). Sınırlı sayıdaki araştırmalar dahilinde yapılan bu çalışma tıp fakültesi öğrencilerinin genetik okuryazarlık düzeyinin belirlenmesi amacı ile yazılmaktadır.

Bütün bu tanımlar, genetik okuryazarlığa sahip öğrencilerin yetiştirilmesinin önemini vurgulamaktadır. Özellikle lisans düzeyinde verilecek eğitim oldukça önem taşımaktadır (Erdoğan , Özsevgeç, & Özsevgeç, 2014).Eğitim, öğrencilerin toplumu yakından ilgilendiren konularda etkin karar verme ve genetik bilimi ile ilgili kavramlar hakkında yeterli bilgi düzeyine ulaşması açısından da oldukça önem arz etmektedir (Acra, 2006).

Anahtar Kelimeler: Genetik okuryazarlık, Tıp fakültesi öğrencileri, Anlayış

DRAFT

ORAL PRESENTATION

Determination of oxidative stress levels (malondialdehyde), and some antioxidant activities (superoxide dismutase, glutathione peroxidase and reduced glutathione) in unoperated breast cancer patients

Safa Ali Ahmed AMAR , Halit DEMİR, Canan DEMIR , Mehmet Naci ALDEMİR , Kasım TURAN

* Van Yüzüncü Yıl University Institute of Science and Technology

asafa1278@gmail.com

Abstract

In this study, it was aimed to determine malondialdehyde (MDA) level and some antioxidant activities such as superoxide dismutase (SOD), reduced glutathione (GSH) and glutathione peroxidase (GPx) in patients with non-operated breast cancer. SOD, GSH and GPx activities were found to be significantly lower in the unoperated breast cancer patient group compared to the healthy control group ($p<0.05$). The MDA levels of the unoperated breast cancer patient group were found to be significantly higher than the healthy control group ($p<0.05$). In conclusion, increased oxidative stress may cause cellular damage in non-operated breast cancer patients. As a result, lipid peroxidation level (MDA) increased, while antioxidant activity decreased.

Keywords: Antioxidant enzymes, Breast cancer, Malondialdehyde, Oxidative stress, ROS.

DRAFT

ORAL PRESENTATION

HMGA2 Proteini inhibitörlerinin ilaç yeniden konumlandırma sistemi ile araştırılması

Turgut ŞEKERLER[1*] (*ORCID: https://orcid.org/0000-0002-3120-2911*)

[*1] Marmara Üniversitesi, Eczacılık Fakültesi, Biyokimya Anabilim Dalı, İstanbul, Türkiye
[1]Üniversite, Fakülte, Bölüm, Şehir, Ülke

*Sorumlu yazar e-mail:turgut.sekerler@marmara.edu.tr

Özet

HMGA2 (The mammalian high-mobility-group protein AT-hook 2)1970'li yıllarda izole edilen ve üzerinde bir çok çalışma yapılan HMGA2 geninin ürünü olan bir proteindir.Bu protein DNA üzerindeki ceplere yerleşerek konformasyonel değişikliğe sebep olur. Bu değişimlerin sonucunda ise;embryonik kök hücrelerinin regülasyonunun sağlanması ve preadiposit hücrelerden adipositlerin sentezlenmesinde rol oynadığı ileri sürülmektedir. Son yıllarda yapılan çalışmalarda ise nadir hastalık olarak kabul edilen ailesel multiple lipomatozis ile de ilgili olabileceği ileri sürülmüştür (1-2). Yeni çalışmalar, bu proteine özgü ilaç geliştirmeyi önemli hale getirmiştir. Bununla ilgili çalışmalar mevcut olsa da daha çok araştırma yapılmaya ihtiyaç vardır. Bu çalışmada; proteinin yapısı veritabanlarından alınarak incelenmiş ve mevcut piyasadaki ilaçlarla olan etkileşimi in siliko olarak araştırılmıştır. Çalışmanın sonucunda mevcut ilaçlarla, protein arasındaki etkileşimin zayıf olduğu görülmüştür. En iyi etkileşime sahip ajanın (Masoprocol) kenetlenme enerjisi -5.8kcal/mol olarak tespit edilmiştir.Bununla beraber doğal ürün kökenli moleküller ile yapılan in siliko çalışmada ise rottlerin isimli molekülün -7.0kcal/mol değerinde bir bağlanma enerjisi verdiği tespit edilmiştir.Bu çalışmanın sonucunda rottlerin isimli kimyasal ajanın bu protein üzerine etkili olabileceği düşünülmektedir. Tüm bu hesaplamalı biyolojik çalışmalara göre HMGA2 proteini önemli biyokimyasal reaksiyonlarda görev almakta olup, üzerine daha çok çalışmanın yapılmasına ihtiyaç duyulmaktadır.

Anahtar Kelimeler: HMGA2, İlaç yeniden konumlandırma, Moleküler docking

Kaynaklar:

1- Su, L., Deng, Z., & Leng, F. (2020). The mammalian high mobility group protein AT-Hook 2 (HMGA2): biochemical and biophysical properties, and its association with adipogenesis. *International journal of molecular sciences*, *21*(10), 3710.
2-Mejia Granados, D. M., de Baptista, M. B., Bonadia, L. C., Bertuzzo, C. S., & Steiner, C. E. (2020). Clinical and molecular investigation of familial multiple lipomatosis: variants in the HMGA2 gene. *Clinical, Cosmetic and Investigational Dermatology*, 1-10.

DRAFT

ORAL PRESENTATION

Surface characterization of the newly developed ion–selective electrodes

Onur Cem Altunoluk[1*] (*ORCID: https://orcid.org/0000-0003-3558-3499*), Oğuz Özbek[2] (*ORCID: https://orcid.org/ 0000-0001-5185-9681*)

[*1]Tokat Gaziosmanpaşa University, Faculty of Science and Arts, Department of Chemistry, Tokat, Turkey
[2]Zonguldak Bülent Ecevit University, Science and Technology, Application and Research Center, Zonguldak, Turkey

*Corresponding author e-mail: altunolukonurcem@gmail.com

Abstract

In this study, new ion–selective electrodes using 1-(3-carboxyphenyl)-2-thiourea as an ionophore were prepared and their potentiometric properties were tested. As a result of the potentiometric studies, it was determined that the ionophore exhibited a highly selective behaviour towards copper(II) ions. The developed copper(II)–selective electrode had a wide working range, lower detection limit, fast response time, good stability and repeatability. Surface images of the prepared electrodes were examined with a scanning electron microscope (SEM). Additionally, energy dispersive X–ray (EDX) and mapping studies of the electrodes were performed. According to the results obtained, it was determined that copper(II) ions adhered to the porous areas on the electrode surface, and in mapping studies, copper(II) ions showed a wide distribution on the electrode surface. This result shows that the ionophore can interact with copper(II) ions. Finally, the newly developed electrode detected copper(II) ions in various water samples with very high recoveries.

Keywords: Scanning electron microscopy, ion–selective electrodes, potentiometry, thioure.

DRAFT

ORAL PRESENTATION

Fishery Cooperatives in Çanakkale

Mustafa Zabun[1*] (*ORCID: https://orcid.org/0009-0006-8509-5281*), Semih Kale[2] (*ORCID: https://orcid.org/0000-0001-5705-6935*)

[*1]Çanakkale Onsekiz Mart University, School of Graduate Studies, Department of Marine Sciences and Limnology, 17020, Çanakkale, Türkiye.
[2]Çanakkale Onsekiz Mart University, Faculty of Marine Sciences and Technology, Department of Fishing and Fish Processing Technology, 17020, Çanakkale, Türkiye.

*Corresponding author e-mail: mustafazabun3317@gmail.com

Abstract

Fishery cooperative is defined as a form of organization focused on the development of the fisheries industry and the improvement of the welfare of fishermen, with objectives such as increasing income, increasing the standard of living and increasing production, and bringing together those involved in fishing, processing, aquaculture or marketing activities in the sector. Fisheries cooperatives exist in almost every country where fishing is carried out. These cooperatives are generally active in inland fisheries and marine fisheries, with a focus on capture fisheries, but they also play an active role in aquaculture. Çanakkale has Çanakkale has coastlines to the Çanakkale Strait, the Aegean Sea and the Marmara Sea. There are a total of 26 fisheries cooperatives established in Çanakkale and affiliated to the Çanakkale Region Fisheries Cooperatives Union. The total number of members of these cooperatives is 1207. The total number of members of the fishery cooperatives in Çanakkale varies between 7 and 328. While the number of cooperatives whose members mostly fish in the Çanakkale Strait is 9, there are 14 cooperatives in the Aegean Sea and 3 cooperatives in the Marmara Sea. Fisheries cooperatives play an important role in the social, cultural and economic survival of fisheries. Fisheries can be more efficient and sustainable if fisheries cooperatives play a more active role.

Keywords: Fisheries, Fishery cooperation, Fisheries management.

This study is a part of MSc thesis of the first author at Çanakkale Onsekiz Mart University, School of Graduate Studies, Department of Marine Sciences and Limnology.

DRAFT

ORAL PRESENTATION

The Atherton-Todd reaction for functionalization of DOPO-derivatives

Nese Cakir Yigit[1*] (*ORCID: https://orcid.org/0000-0002-4714-4488*)

[*1] Yalova University, Faculty of Engineering, Department of Polymer Materials Engineering, Yalova, Turkey.

nese.cakir@yalova.edu.tr

Abstract

Atherton–Todd reaction is a versatile tool, as defined by Atherton, Openshawand, and Todd in 1945, as a new way of synthesizing phosphoramidates from dialkylphosphite with a primary amine in the presence of a base in carbon tetrachloride in mild reaction conditions. These reaction conditions are appropriate for nucleophiles such as amines, alcohols, and phenols. Each reaction demonstrates stereospecific behavior, leading to the inversion of configurations at the phosphorus centers. Given the importance and versatility of phosphorus compounds in various applications, innovative synthetic methods interest academic and industrial communities across various fields. For instance, phosphorus-based flame retardants have been preferred over halogenated ones for an extended period due to their ability to generate a protective char during combustion. In particular, 9,10-dihydro-9,10-oxa-10-phosphaphenanthrene-10-oxide (DOPO) derivatives are significantly interested in developing halogen-free flame retardant additives. Studies in the literature reported the modification of DOPO to react with the P-H bond (phosphinate) to introduce nitrogen or oxygen-containing groups. Despite the mechanism of the Atherton–Todd reaction is not fully understood, especially the alteration in the oxidation state of the P-atom, the Atherton–Todd reaction of DOPO is highly efficient and straightforward, yielding a high quantity of products.
Herein, we reported the thiol-based nucleophiles for the functionalization of DOPO by adapting the Atherton–Todd reaction conditions. The optimum reaction conditions were determined by changing reaction time, solvent, and reactant amounts. The success of the reaction was confirmed by both ^{1}H NMR and ^{31}P NMR spectrums. In addition, to confirm the purity of the obtained thiol-functional DOPO compounds further coupling reactions and characterizations were performed.

Keywords: Atherton–Todd reaction, phosphorous groups, DOPO, coupling reactions.

DRAFT

ORAL PRESENTATION

Antalya doğal florasından toplanan kocayemiş (*Arbutus unedo* L.) ve sandal (*Arbutus andrachne* L.)'ın, bazı fiziksel ve kimyasal özelliklerinin belirlenmesi

Arzu BAYIR YEĞİN[*] (*ORCID: https://orcid.org/ 0000-0002-2194-6730)*, Ahu ÇINAR[1] (*ORCID: https://orcid.org/ 0000-0001-7095-1192)*, Demet YILDIZ TURGUT[1] (*ORCID: https://orcid.org/0000-0002-7486-3701)*, Tuba SEÇMEN[1] (*ORCID: https://orcid.org/ 0000-0001-9730-7057)*

[*1]Batı Akdeniz Tarımsal Araştırma Enstitüsü Müdürlüğü, Gıda teknolojisi ve Tıbbi Aromatik Bitkiler Bölümü, Antalya, Türkiye
*Sorumlu yazar e-mail: arzu.bayir@tarimorman.gov.tr

Özet

Ericaecea ailesinin bir üyesi olan Arbutus cinsi içinde yer alan Arbutus unedo L. (kocayemiş) ve Arbutus andrachne L. (sandal) ülkemizin doğal florasında bulunan ve ekonomik öneme sahip olan iki önemli türdür.

Çalışmada, Antalya florasında yetişen kocayemiş ve sandal bitkisinin farklı kısımlarının bazı fiziksel ve kimyasal özelliklerinin belirlenmesi, üstün özelliklere sahip genotiplerin tespit edilmesi amaçlanmıştır.

Bu amaçla 60 adet sandal ve 9 adet kocayemiş genotipinin yaprakları, meyveleri ve çiçekleri kullanılmıştır. İncelenen özellikler bakımından genotipler arasında önemli farklılıklar olduğu ortaya konulmuştur. Her iki türde de en düşük fenolik ve flavonoid madde miktarı meyvelerde görülmüş, bunu çiçekler ve yapraklar takip etmiştir. Antioksidan aktivite DPPH ve FRAP yöntemleri ile değerlendirilmiş, her iki yöntemde de en fazla aktivite yapraklarda gözlenmiştir. Sandal türünün meyvelerinin ortalama 103.16 mg/100 g, kocayemiş meyvelerinin ise ortalama 108.12 mg/100 g C vitamini, sandal yapraklarının ortalama 0.87 g/100 g, kocayemiş türünün yapraklarının ise ortalama 0.79 g/100 g arbutin içerdiği belirlenmiştir.

Anahtar Kelimeler: Arbutus L., fenolik madde, flavonoid madde, antioksidan aktivite, arbutin

DRAFT

ORAL PRESENTATION

Binding of breast milk-derived exosomes to the amniotic membrane

Ferda Işık[1*] (https://orcid.org/0009-0002-2798-0988), Tuğba Tunalı Akbay[2] (https://orcid.org/ 0000-0002-2091-9298)

*[1]Marmara University, Institute of Health Sciences, Faculty of Pharmacy, Department of Biochemistry, Istanbul, Turkey

[2]Marmara University, Faculty of Dentistry, Department of Biochemistry, Istanbul, Turkey

* Corresponding author e-mail: ferdaaaisik@gmail.com

Abstract

Exosomes are the smallest known group of extracellular vesicles secreted by all cells in the body. They are 40-100 nm in size and surrounded by a double phospholipid layer. Exosomes are also found in the body fluids, such as blood, urine, amniotic fluid, breast milk, serum, and plasma. They can carry proteins, lipids, messenger RNA, micro RNA, long non-coding RNA, and DNA fragments. Through these biomolecules, exosomes regulate gene expression in recipient cells and play a role in many biological functions, such as immune regulation, cell differentiation, intercellular communication, and cell migration. Human amniotic membrane (HAM) is the innermost placental membrane. It is used as a biomaterial that supports wound healing. It has anti-inflammatory, antiangiogenic and antimicrobial properties and can promote epithelialization and inhibit fibrosis. This study aimed to increase the therapeutical effects of HAM by ensuring the attachment of breast milk-derived exosomes to HAM. In the first stage of this study, the fat layer was removed from the breast milk by centrifugation. Then, exosomes were isolated by ultracentrifugation. The total protein level was determined. The exosomes were then analyzed using flow cytometry for the presence of the most frequent exosomal membrane proteins: CD9 and CD63. Particle counting was also performed. Commercially produced lyophilized HAMs (22 × 40 mm) were placed in petri dishes and incubated with different concentrations of exosomes. The presence of exosomes on the membrane was visualized using scanning electron microscopy. In conclusion, exosomes derived from breast milk will further contribute to the use of amniotic membranes for therapeutic purposes, as they successfully bind to HAM.

Keywords: Exosomes, breast milk, amniotic membrane

DRAFT

ORAL PRESENTATION

The effect of myrtle extract added to drinking water on performance, hematological and biochemical parameters in rats

*Ümit Özçınar[1] (ORCID: https://orcid.org/0000-0002-1143-1215), İsmail Bayram[1](ORCID: https://orcid.org/0000-0002-9993-7092), Ali Çalık[2] (ORCID: https://orcid.org/0000-0003-4550-9321),Mehmet Fatih Bozkurt[3] (ORCID: https://orcid.org/0000-0002-1669-0988), Syed Rizwan Ali Shah[1] (ORCID: https://orcid.org/0000-0002-2752-4854), Mustafa Midilli[4] (ORCID: https://orcid.org/0000-0003-3575-3304), İsmail Hakkı Özsandık[1] (ORCID: https://orcid.org/0000-0003-4943-1760), Eyüp Eren Gültepe[1] (ORCID: https://orcid.org/0000-0002-2404-1232), Mudassar Zafar[3] (ORCID: https://orcid.org/0009-0009-4356-7449), Barış Denk [5] (ORCID: https://orcid.org/0000-0002-7586-0895), İbrahim Sadi Çetingül[1] (ORCID: https://orcid.org/ 0000-0002-7608-6176)

[1] University of Afyon Kocatepe, Faculty of Veterinary Medicine, Department of Animal Nutrition, ANS Campus, Afyonkarahisar-TURKEY
[2] University of Ankara, Faculty of Veterinary Medicine, Department of Animal Nutrition and Nutritional Diseases, Ankara-TURKEY
[3] University of Afyon Kocatepe, Faculty of Veterinary Medicine, Department of Pathology, ANS Campus, Afyonkarahisar-TURKEY
[4] Abant İzzet Baysal University, Faculty of Agriculture and Nature Science, Department of Poultry Science, Bolu- Turkey.
[5] University of Afyon Kocatepe, Faculty of Veterinary Medicine, Department of Biochemistry, ANS Campus, Afyonkarahisar-TURKEY

*umitozcinar@gmail.com

Abstract

This study was conducted to determine the effects of myrtle extract added to drinking water on performance, blood physiology, biochemical and small intestine histomorphological parameters in rats. 80 Wistar albino rats were used and divided into 5 groups and each group had 8 subgroups. Myrtle plant extract was supplemented at 0% (Control group), 2.5%, 5%, 7.5%, 10% levels. The research was continued for 35 days. Weekly body weights and feed consumption were measured to calculate Feed conversion ratios (FCR). To determine the water consumption, the water given to the rats and the refusal were measured daily. At the end of the experiment, blood, liver and small intestine samples were taken to perform analyses. Myrtle extract at different levels did not influence performance parameters. The glucose level was the lowest in the 2.5% group ($p=0.005$) and there was a cubic effect among the groups .The blood bilirubin level was determined at the highest level in the group given 2.5% ($p=0.008$). There was a statistical significance in serum urea ($p=0.017$) and BUN ($p=0.022$) values; In both parameters, quadratic effects of mrytus are also observed. There were significant differences, between the treatment groups and control group for Basophil ($p=0.001$), Mean Corpuscular Hemoglobin (MCH) ($p=0.009$), Mean Corpuscular Hemoglobin Concentration (MCHC) ($p=0.017$) and platelete ($p<0.001$) also P-LCR ($p=0.045$) values among treatment groups. Liver cytokine and heat shock protein mRNA abundance were not influenced. Myrtus extract had a statistically significant effect on histomorphological traits of small intestines both between control and treatment groups and among treatment groups. As a result, it was concluded that the addition of myrtle extract has a cubic effect on the blood serum glucose also intestinal morphology was affected positively. Some of hemogram parameters were influenced in dose dependent pattern also water consumption increased without adversely affecting performance parameters.

Key words: Rat, Myrtle, performance, metabolism, cytokine

DRAFT

ORAL PRESENTATION

A voltammetric study on the interaction of (E)-1-((2-chloro-4-nitrophenylimino)methyl)naphthalen-2-ol with Human Serum Albumin at physiological pH

Ender Biçer[1,*] (*ORCID: https://orcid.org/0000-0001-5501-5160*), Neslihan Özdemir[2] (*ORCID: https://orcid.org/0000-0003-4558-8955*), Mustafa Macit[1] (*ORCID: https://orcid.org/0000-0001-6593-4291*)

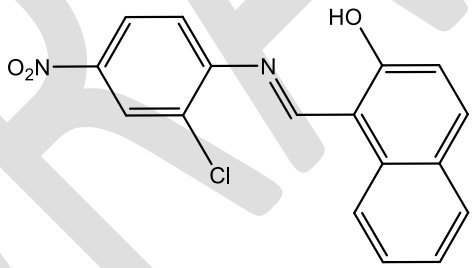
[1]Ondokuz Mayıs University, Faculty of Science, Department of Chemistry, Samsun, Turkey.
[2]Amasya University, Merzifon Vocational School, Department of Machinery and Metal Technologies, Amasya, Turkey.

*Corresponding author e-mail: ebicer@omu.edu.tr

Abstract

In this study, the interaction of (*E*)-1-((2-chloro-4-nitrophenylimino)methyl)naphthalen-2-ol (see Scheme, abbreviated as CNPIMN) with human serum albumin (HSA) has been investigated at pH 7.40 by means of square-wave voltammetry technique. Under the applied experimental conditions, CNPIMN has given two cathodic peaks, attributed to the reduction processes of nitro and imine moieties, respectively. On the other hand, it has been observed that the currents of these peaks have decreased in the presence of HSA. It has been also found that the decreases in peak current depend on the concentration of HSA. These peak current decreases have been sourced from the intermolecular interactions between CNPIMN and HSA. By using of the peak current values of CNPIMN in the absence and presence of HSA, the stoichiometry and formation constant values of CNPIMN-HSA molecular complex have been determined.

Keywords: Schiff base, HSA, interaction, voltammetry.



Scheme. Molecular structure of CNPIMN

DRAFT

ORAL PRESENTATION

Sıçanlarda gentamisin kaynaklı nefrotoksisite üzerinde D vitamininin etkileri

Elif Aksöz[1] (*ORCID:* https://orcid.org/0000-0002-4827-804X), Fazilet Şen[1*] (*ORCID:* https://orcid.org/0000-0002-8433-1194), Murat Çelebi[2] (*ORCID:* https://orcid.org/0000-0002-1769-2512), Nurullah Güçlü[1] (*ORCID:* https://orcid.org/0000-0002-7985-1910), Oğuzhan Korkut[1] (*ORCID:* https://orcid.org/0000-0002-2057-4197)

[*1]Balıkesir Universitesi, Tıp Fakültesi, Tıbbi Farmakoloji AD, Balıkesir, Türkiye.
[2] Balıkesir Universitesi, Savaştepe Meslek Yüksek Okulu, Veterinerlik Bölümü, Balıkesir, Türkiye.

*Corresponding author e-mail: aksoz@balikesir.edu.tr

Özet

Nefrotoksisite, aminoglikozidlerin, özellikle de gentamisinin en önemli yan etkilerinden biridir. Aminoglikozid kullanımı ile %25'e varan oranda nefrotoksisite gelişme riski bulunmaktadır. Literatürde D vitamininin gentamisin nefrotoksisitesindeki etkileri ile ilgili az sayıda çalışma bulunmaktadır. Bu çalışmalarda D vitamini farklı dozda ve farklı sürelerde uygulanmıştır.Çalışmaların sonuçları çelişkilidir. Bu çalışmada, gentamisin ile birlikte veya öncesinde verilen D vitamininin gentamisinin neden olduğu nefrotoksisite üzerine etkilerinin karşılaştırılması ve D vitamininin olası nefroprotektif etkisinin araştırılması amaçlandı. Gentamisin ile nefrotoksisite oluşturulan sıçanlara, gentamisinle aynı anda ve 1 hafta öncesinden başlanarak D vitamini uygulandı. Gentamisin dozu 100 mg/kg/gün, D3 vitamini dozu 1000 IU/kg/gün olarak belirlendi. Wistar Albino 28 adet sıçan rastgele 4 gruba ayrıldı: 1. Kontrol; 2. Gentamisin (1 hafta boyunca) uygulandı; 3. D vitamini + Gentamisin (1 hafta boyunca eşzamanlı başlanarak) uygulandı; 4. D vitamini (gentamisinden 1 hafta önce başlanarak 2 hafta boyunca) + Gentamisin (2. hafta eklenerek 1 hafta boyunca) beraber uygulandı. İlaç uygulamalarının bitiminde tüm sıçanlar sakrifiye edildi; biyokimyasal analizler için kan ve doku örnekleri alındı. D vitamini toksisitesini ekarte etmek için değerlendirdiğimiz kan kalsiyum düzeylerinde gruplar arasında fark görülmedi. Lipid peroksidasyonunun göstergesi olan MDA'da gentamisine bağlı artış, D vitamini uygulaması ile her iki grupta da kontrol değerlerine geri döndürülürken; oksidatif strese karşı savunmanın ilk basamağı olan SOD'da gentamisine bağlı oluşan azalma sadece gentamisinden önce başlanarak iki hafta D vitamini uygulanan grupta kontrol değerlerine döndü. TNF-α ve IL-6'da gentamisine bağlı artış D vitamini uygulaması ile her iki grupta da anlamlı olarak geri çevrildi. Kreatinin ve üre seviyelerinde gentamisine bağlı yükselme D vitamini uygulaması ile azalmakla birlikte kontrol değerlerine getirilemedi. D Vitamini, gentamisin nefrotoksisitesini önleme potansiyeli göstermekle birlikte, bu vitaminin uygun doz ve uygulama şemasının belirlenmesi için daha fazla çalışmaya ihtiyaç bulunmaktadır.

Anahtar Kelimeler: Gentamisin, D vitamini, nefrotoksisite

DRAFT

ORAL PRESENTATION

In silico identification of newly designed potent class I and class IIb histone deacetylase enzyme inhibitors

İsmail Akçok[1*] (*ORCID: https://orcid.org/0000-0002-5444-3929*)

[*1] Abdullah Gül University, Faculty of Life and Natural Sciences, Bioengineering Department, Kayseri, Türkiye.

*Corresponding author e-mail: ismail.akcok@agu.edu.tr

Abstract

There are a lot of studies that show dysregulation of histone deacetylases (HDACs) is related to cancer development and progression. HDAC enzymes catalyze the removal of the acetyl group from lysine residues in the N-terminal tails of histones and non-histone proteins. HDACs are fascinating therapeutic targets because they are overexpressed in cancer cells, and their inhibition offers hope for the treatment of cancer and other diseases related to them. Because of this relation between HDACs and development of cancer, scientists have been quite interested in HDACs as a target to battle against cancer. Four HDAC inhibitors, vorinostat, romidepsin, belinostat, and panobinostat have been approved by the Food and Drug Administration (FDA) so far. In this study, a series of novel compounds that contains hydroxamic acid Zn^{2+} binding group, N-acyl hydrazone linker, and pyrrolopyrimidine or purine cap region were designed for targeting class I and class IIb HDAC enzymes (Figure 1). Through the use of molecular docking studies and molecular dynamics (MD) simulation analysis, the potential interactions between ligands and enzymes were revealed. Molecular Mechanics-Poisson-Boltzmann Surface Area (MM-PBSA) scores were calculated to predict the binding affinity.

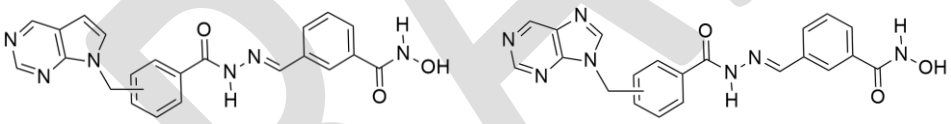


Figure 1: Structures of novel potent class I and class IIb HDAC inhibitors

Keywords: Cancer, HDACs, Molcular Docking, Molecular Dynamics (MD), MM-PBSA

DRAFT

ORAL PRESENTATION

Development of Electromagnetic Field Exposure System in Zebrafish Embryos

Derya CANSIZ (*ORCID:0000-0002-6274-801X*)

Istanbul Medipol University, Istanbul, Turkey

cansizderya@yahoo.com

Abstract

Electronic devices such as mobile phones, computers and tablets cause disruption of many systems in living organisms with the radiofrequency-electromagnetic field (RF-EMF) they emit. Nowadays, with the advancement of technology and the inclusion of electronic devices in every aspect of our lives, the RF-EMF created by these devices has also increased. RF-EMF plays a role in the formation of various diseases (such as, headache, cancer, heart disease, obesity) by penetrating into the tissues, causing the movement of charged particles such as electrons and ions within the cell and the formation of radicals, preventing the synthesis of various proteins by disrupting enzyme activity, and causing apoptosis and cell differentiation. Zebrafish embryo is accepted as a suitable model organism in modeling human diseases because zebrafish genes have 70% genetic homology with humans, they can be monitored under a microscope during the development period, and experiments can be completed in a short time. We established a special RF-EMF exposure system to examine the effects of RF-EMF exposure on zebrafish embryos. With this system developmental, behavioral, and molecular changes that occur as a result of RF-EMF exposure can be demonstrated in zebrafish embryos. Although many studies have been conducted on RF-EMF exposure, studies on the effect of RF-EMF exposure on fetal development are limited. The first 120 hours of zebrafish after fertilization refer to the embryonic period. Accordingly the RF-EMF exposure system we established allows the examination of the effects of RF-EMF exposure on the embryonic development of zebrafish embryos.

Key words: Radiofrequency-Electromagnetic field, embryonic development, Zebrafish embryos

DRAFT

ORAL PRESENTATION

Molecular Substrates of COVID-19 and Pulmonary Arterial Hypertension Identified Through Signalling Network Construction

Defne Cig[1] (*ORCID: https://orcid.org/0000-0002-2761-5246*), Ceyda Kasavi[2*] (*ORCID: https://orcid.org/0000-0003-2970-387X*)

[1]Marmara University, Faculty of Engineering, Bioengineering Department, Istanbul, Turkey.

*Corresponding author e-mail: ceyda.kasavi@marmara.edu.tr

Abstract

Pulmonary arterial hypertension (PAH) is a serious condition that occurs when high blood pressure builds up in pulmonary arteries, affecting blood circulation from the heart to the lungs, which can ultimately lead to heart failure and death. COVID-19 pandemic exhibits genuine challenges for PAH patients. Therefore, novel strategies for the management of PAH patients with COVID-19 are needed. However, molecular mechanisms underlying the crosstalk between two diseases have not been fully understood. In the current study, we elucidated similarities and differences in the re-organization of transcriptional response in the presence of PAH and COVID-19 and identify molecular substrates of both diseases by constructing signalling networks. For this purpose, gene expression data obtained from lung tissues of PAH and COVID-19 patients were comparatively analysed and differentially expressed genes (DEGs) were identified. Then, reporter molecules (i.e., metabolites, transcription factors (TFs) and microRNAs (miRNAs)) were discovered by integrating transcriptome data with human metabolic and regulatory networks. Consequently, signalling networks were constructed by merging the interactions between regulatory molecules and DEGs. COVID-19 network contained 2546 interactions between 558 DEGs, 89 metabolites, 6 TFs and 128 miRNAs, whereas PAH network contained 29266 interactions between 3955 DEGs, 112 metabolites, 22 TFs and 356 miRNAs. A total of 207 reporter molecule-DEG interactions were found to be common in both diseases. Functional enrichment analysis of these reporter molecules was performed via interacting DEGs, and revealed significant alterations in cardiac muscle contraction, oxidative phosphorylation, non-alcoholic fatty liver disease, diabetic cardiomyopathy, thermogenesis, and several cardiovascular, infectious, and neurodegenerative disease pathways. Moreover, gene-based drug repositioning analysis revealed candidate drugs including cardiac glycosides, insulin sensitizers, and drugs with antifibrotic, anti-inflammatory, and antiproliferative effects, for consideration in future clinical drug development.

Keywords: Pulmonary arterial hypertension, COVID-19, Transcriptomics, Signalling network, Drug repositioning

DRAFT

ORAL PRESENTATION

Investigation of the corrosion performance of aluminum 7075 alloy used as submarine material in the Sea of Marmara environments

Tuba Ünsal Özgüvenç[1*] (*ORCID: 0000-0003-0057-1260*)

[*1]Istanbul University, Institute of Marine Science and Management, Fatih, Istanbul, Türkiye

*Corresponding author e-mail: tunsal@istanbul.edu.tr

Abstract

Aluminum (Al) has a wide range of uses in many industries with its low cost, high conductivity and easy processing properties. AA7075 is the most commonly used Al alloy in submarines, aircraft and rail vehicles with its high corrosion resistance, fracture toughness, ductility and low density. These alloys are used as an excellent material to produce marine vehicles, especially in the maritime industry. In this study, the corrosion performance of AA7075 alloy used as submarine material under seawater environment was examined. For this purpose, seawater samples were taken from MD24 and MD26 stations in the Sea of Marmara and the corrosion performance of AA7075 alloy exposed to these environments was determined using corrosion potential, potentiodynamic polarization and electrochemical impedance spectroscopy (EIS) methods. The coupons were abraded from 600 up to 1200 grit using abrasive paper then cleaned with isopropanol and dried under UV light before testing. Only the top 1 cm^2 surface was exposed to the solutions. All experiments were carried out in corrosion cell. In this work, AA7075 alloy exposed to seawater taken from MD24 and MD26 stations has different corrosion behavior. The corrosion potentials of AA7075 alloy were found as -433 mV/s and -746 mV/s in the MD24 and MD26 stations, respectively. The corrosion rate of AA7075 alloy was found lower in MD26 than in MD24. AA7075 alloy showed high impedance resistance in both environments. All the results indicated that AA7075 alloy is an excellent material against corrosion in the Sea of Marmara environments.

Keywords: AA7075 alloy, The Sea of Marmara, Corrosion, Electrochemical methods.

DRAFT

ORAL PRESENTATION

Suda ve Yağda Eriyen Vitaminlerin Nörona Farklılaştırılmış SH-SY5Y Hücrelerinde Apoptoz Koşullarına Karşı Koruyucu Etkilerinin İncelenmesi

Gül ÖNCÜ[1*] (*ORCID: https://orcid.org/0000-0003-0376-0112*),
Hale SAYBAŞILI[2] (*https://orcid.org/0000-0002-6016-4651*)

[*1,2]Boğaziçi Üniversitesi, Biyomedikal Mühendisliği Enstitüsü, İstanbul, Türkiye.

*gul.oncu@boun.edu.tr; saybasil@boun.edu.tr,

Özet

Oksidatif stres, beynin enerjiye duyduğu yüksek ihtiyaç ve oksijenli solunum nedeniyle nöronların özellikle açık olduğu bir tehlikedir. Nörodejeneratif hastalıklar dışında, oksidatif stresin kanser oluşumuyla, yaşlanmayla ve koroner kalp hastalığı ile ilişkili olduğu tespit edilmiştir. Antioksidanlar, "Bir maddenin oksidatif hasar görmesini geciktiren, engelleyen ya da oksidatif hasarı ortadan kaldıran tüm maddeler" olarak tanımlanmıştır. Hücrelerin, serbest radikallerin etkisini azaltmak için kullanabileceği iki antioksidan sistem vardır: antioksidan enzimler ve küçük moleküllü antioksidan maddeler. Enzimatik olmayan antioksidan maddeleri vücut kendisi üretebildiği gibi (metabolik antioksidanlar), beslenme yoluyla dışarıdan alınan antioksidanlar da bulunmaktadır (besinsel antioksidanlar). Önerilen çalışma, sinir hücresi benzeri hücrelere çeşitli antioksidan maddeler uygulayıp, sonrasında bu hücreleri çeşitli oksidatif strese bağlı apoptoz modellerine maruz bırakarak uygulanmış olan antioksidan maddelerin oksidatif strese karşı hücreleri koruyup korumadığını incelemeyi hedeflemektedir. Antioksidan uygulaması yapılmamış hücrelerde de apoptoz modellenerek bu maddelerin hücreleri kontrole göre ne oranda koruduğu anlaşılabilecektir. Antioksidan maddeler olarak literatürdeki çalışmalara dayanarak askorbik asit, α-tokoferol ve bu vitaminlerin beraber kullanılması seçilmiştir. Oksidatif stress yaratması amacıyla da C2-seramit maddesi seçilmiştir. C2 seramidin yaratacağı oksidatif stresi ölçebilmek adına ise literatürde negative kontrolü olarak bilinen dihidroseramid maddesi kullanılmıştır. Öncelikli olarak seramidin yaratacağı oksidatif stresin dihidroseramide kıyasla yarattığı etki canlılık testi ve elektrofizyolojik metodla ölçülmüş. Sonrasında yapılan çalışmada ise seçilen vitaminlerin koruyucu etkisi yine yapılan canlılık testleriyle (alamar blue assay) ve elektrofizyoloji deneyleriyle kanıtlanmıştır. Bu çalışma kapsamında, askorbik asit ve α-tokoferol'ün retinoik asit ve beyin kaynaklı nörotrofik faktör kullanılarak farklılaştırılmış insan nöroblastoma hücrelerinde indüklenmiş apoptoza karşı koruyucu etkisi olup olmadığı ilk kez incelenmiştir.

Anahtar Kelimeler: Oksidatif stres, Seramit, A-tokoferol, Askorbik asit, SH-SY5Y, Antioksidan

DRAFT

ORAL PRESENTATION

Synthesis, Characterization, and Enzyme Activity of a New Series Schiff Base Metal Complex

Sümeyra Tuna Yıldırım [1] (ORCID: https://orcid.org/0000-0001-5564-9630), Cüneyt Türkeş [2] (ORCID: https://orcid.org/0000-0002-2932-2789), Meryem Topal [3] (ORCID: https://orcid.org/0000-0002-2107-8603), Fevzi Topal [4] (ORCID: https://orcid.org/ 0000-0002-2443-2372), Oktay Eroğlu [5,*] (ORCID: https://orcid.org/0000-0001-6045-7051), Şükrü Beydemir [6,7] (ORCID: https://orcid.org/ 0000-0003-3667-6902)

[1] Erzincan Binali Yıldırım University, Faculty of Pharmacy, Department of Analytical Chemistry, Erzincan, Türkiye
[2] Erzincan Binali Yıldırım University, Faculty of Pharmacy, Department of Biochemistry, Erzincan, Turkey
[3] Gümüşhane University, Gümüşhane Vocational School, Department of Health Services, Gümüşhane, Turkey
[4] Gümüşhane University, Gümüşhane Vocational School, Department of Chemical and Chemical Processing Technologies, Gümüşhane, Turkey
[5] Erzincan Binali Yıldırım University, Health Sciences Institute, Department of Pharmaceutical Sciences, Erzincan, Türkiye
[6] Anadolu University, Faculty of Pharmacy, Department of Biochemistry, Eskişehir, Turkey
[7] Bilecik Şeyh Edebali University, Bilecik, Turkey

*Corresponding author e-mail: oktayeroglu6124@gmail.com

Abstract

Schiff base ligands constitute an important class of synthetic phenolic compounds and are known for their biological and pharmacological activities [1]. Additionally, the presence of hydroxyl groups attached to the aromatic ring makes them effective agents for some diseases caused by free radical damage [2]. In this research, 2-[(2-hydroxybenzilidene)amino)]phenol and its some metal complexes were designed and synthesized. The structures of these compounds were elucidated using various spectroscopic techniques [3] and investigated as therapeutic agents against Alzheimer's disease [4,5]. After screening the acetylcholinesterase and butyrylcholinesterase inhibitory activities, Mn(II)- and Co(II)-complexes (IC_{50}s of 36.24 ± 1.63 nM and 90.60 ± 6.86 nM, respectively) were determined as potent inhibitors of cholinesterases.

Keywords: Enzimatic Activity, Ligand, Schiff Base, Metal Complex

Acknowledgements

This study was supported by a grant of The Research Fund of Erzincan Binali Yıldırım University grant number TSA-2021-756.

References

Kostova, I., & Saso, L. (2013). Advances in research of Schiff-base metal complexes as potent antioxidants. *Current Medicinal Chemistry*, 20(36), 4609-4632.

Petrović, Z. D., Đorović, J., Simijonović, D., Petrović, V. P., & Marković, Z. (2015). Experimental and theoretical study of antioxidative properties of some salicylaldehyde and vanillic Schiff bases. *RSC Advances*, 5(31), 24094-24100.

Wang, X., Wang, X., Jin, S., Muhammad, N., & Guo, Z. (2018). Stimuli-responsive therapeutic metallodrugs. *Chemical Reviews*, *119*(2), 1138-1192.

Türkeş, C., Akocak, S., Işık, M., Lolak, N., Taslimi, P., Durgun, M., ... & Beydemir, Ş. (2022). Novel inhibitors with sulfamethazine backbone: synthesis and biological study of multi-target cholinesterases and α-glucosidase inhibitors. *Journal of Biomolecular Structure and Dynamics*, *40*(19), 8752-8764.

Özler, E., Topal, F., Topal, M., & Öztürk Sarıkaya, S. B. (2023). LC-HRMS Profiling and Phenolic Content, Cholinesterase, and Antioxidant Activities of Terminalia citrina. *Chemistry & Biodiversity*, e202201250.

DRAFT

ORAL PRESENTATION

Penetration Possibility of K^+ Channel Blocker Antiarrhytmia Drugs of CNS and Their Potential Results

Gül ÖNCÜ[1]* (*ORCID: https://orcid.org/0000-0003-0376-0112*),
Hale SAYBAŞILI[2] (*https://orcid.org/0000-0002-6016-4651*)

*[1,2]Boğaziçi Üniversitesi, Biyomedikal Mühendisliği Enstitüsü, İstanbul, Türkiye.

*gul.oncu@boun.edu.tr; saybasil@boun.edu.tr,

Abstract

Arrhythmia is a general term for the heartbeat to be abnormal in any other than normal sinus rhythm. The sinus rhythm is obtained via depolarization with the opening of Na+ channels and Na+ influx, repolarization with the opening of K+ channels, and efflux of K+ and Ca2+ ions as the electrical signal generated in the brain. Like neuronal action potential generation, serum Na+, K+, and Ca2+ levels also affect a cardiac action potential. Even though ion channels have the same purpose and mechanism, they have different subgroups according to their protein chemistry. Many voltage-gated ion channel subgroups are in the brain and heart. This phenomenon affects the central nervous system (CNS) from drugs that target cardiac voltage-gated ion channels and vice versa. This effect is observed with some of the antiarrhythmic drugs. Patients who took the drugs show side effects related to CNS, such as tremors, sleep disorders, disorientation, confusion, abnormal taste and smell, chronic anxiety, and even psychosis. However, each drug has a different side effect and frequency due to its ability to penetrate BBB. This research studies the BBB penetrative ability of class III antiarrhythmic channel blockers to shed light on their potential effects on the central nervous system. Their popularity is increasing, and they are preferred over the other classes due to their safety profile.

Keywords: BBB, cardiac drugs, arrhythmia, K+ channel blockers, lipophilicity, CNS

DRAFT

ORAL PRESENTATION

Expression Analysis of Salinity-associated miRNAs in Gamma-induced Salinity Tolerant Rice (*Oryza sativa*) Mutants

Büşra İmece[1*] (https://orcid.org/0000-0001-7318-5155), Tamer GÜMÜŞ[1] (*ORCID: https://orcid.org/0000-0002-5674-4987*), Sinan MERİÇ[1] (*https://orcid.org/0000-0003-0336-0004*), Alp AYAN[1] (*https://orcid.org/0000-0003-3749-0472*), Çimen ATAK[1] (*https://orcid.org/0000-0001-6045-562X*)

[*1]Istanbul Külür University, Faculty of Science and Letter, Department of Molecular Biology and Genetics, Istanbul, Türkiye.

*Corresponding author e-mail: s.meric@iku.edu.tr

Abstract

Rice (Oryza sativa L.) is one of the most important staple foods along with wheat, barley, and maize in the world. Rice is the main daily nutrition for approximately three billion people. It also has essential minerals and vitamins as nutritional supplements and contains the highest digestible proteins. The present study emphasized the profiles of miR156a, miR159a, miR169b, miR393a, miR398a and miR820a under salinity stress in rice. Experiments were conducted on 90 mM NaCl tolerant rice mutant which was previously obtained from Osmancık-97 variety induced by using gamma radiation. Also, target gene expressions of SPL16, MYB, HAPC2, AFB2, TIR1, CSD2 and DRM2 were evaluated in response to selected miRNAs. The original variety and its resistant mutant were grown in salt-free conditions for 14 days. Perlite filled plastic pods were watered with Yoshida nutrient solution. 14 days old seedlings were subjected to 150 mM NaCl stress for 7 days by adding corresponding amount of NaCl to Yoshida solution. miRNA isolations and cDNA conversions were performed from the fresh leaves of plants after the end of the stress period. Probe-based real-time PCR analysis was performed for miRNA evaluation. All selected miRNA groups presented alteration in response to the salinity stress. There was a decrease in gene regions associated with methylation targeted by miRNA groups, transcription factor HAPC2, root development and flowering. However, an increase was found in the gene region associated with SOD pathways. Since the mutant material was derived through forward genetics approach, we associated salinity tolerant phenotype to specific miRNA expression alterations compared to the original rice variety.

Keywords: miRNA, *Oryza sativa*, salinity, mutation breeding

DRAFT

ORAL PRESENTATION

Effect of alumina source on the final properties of cordierite ceramics

Merve Şeyma Sürel[1*] (ORCID: https://orcid.org/0009-0002-4314-2319), Gülsüm Topateş[2] (ORCID: https://orcid.org/0000-0003-4453-8219

[1] Ankara Yıldırım Beyazıt University, Graduate School of Natural and Applied Sciences, Materials Engineering, Ankara, Turkiye.
[2] Ankara Yıldırım Beyazıt University, Faculty of Engineering and Natural Sciences, Department of Metallurgical and Materials Engineering, Ankara, Turkiye

*merveseymasurel@gmail.com

Abstract

Cordierite ($2MgO.2Al_2O_3.5SiO_2$) is a promising ceramic material for microwave-millimeter wave dielectric and wide-band radome material due to its outstanding mechanical, thermal and electrical properties. These properties directly related with raw materials used during the production of cordierite ceramics. In this study, three different alumina sources were used to obtain cordierite ceramics. A cordierite composition (13.70 MgO - 34.90 Al_2O_3 - 51.40 SiO_2 in wt.%) were designed and pure oxides were selected as raw materials. Al_2O_3 were supplied by using three different sources. The first two foreign sources have high purity and fine particle size, the last local source consisting of coarser particles with lower purity. The compositions were designated as A1, A2 and A3. Prepared ceramics from each composition were sintered at 1350°C for 3 h. The density values of all three compositions were directly affected by alumina source. The bulk density and open porosity values of A1 and A2 were measured as 2.45 g/cm^3 - %0.13 and 2.40 g/cm^3 - %0.22, respectively. Lower bulk density and higher open porosities with the values of 1.85 g/cm^3 and %25.54 were obtained for A3. The dielectric constant and loss values of all three compositions were measured as 3.74 and 0.0082 for A1, 3.86 and 0.0027 for A2, and lastly 3.0041 and 0.0064 for A3, correspondingly. According to X-ray diffraction (XRD) analysis, finer Al_2O_3 particles accelerates cordierite formation. Sample A1 and A2 composed of single cordierite in indialite form. For A3, secondary phases such as mullite and corundum were detected besides indialite. The formation of pure cordierite grains also proved by microstructures from scanning electron microscopy (SEM) images. Results showed that the source of oxides directly affected cordierite synthesis. The finer and purer oxide eases formation of cordierite during sintering. By using proper type of oxide, the properties of cordierite can be improved.

Keywords: cordierite ceramics, composition, alumina, dielectric

DRAFT

ORAL PRESENTATION

Sinop İli Ayancık ve Erfelek ormanlık alanlarında tuzak kamera kayıtları ile büyük memeli türlerinin belirlenmesi

Pınar Çam İcik *ORCID: https://orcid.org/0000-0002-0714-3536*

Sinop Üniversitesi, Fen Edebiyat Fakültesi, Biyoloji Bölümü, Sinop, Türkiye

*Sorumlu yazar e-mail: pinar82mail@gmail.com

Özet

Yaban hayatının önemli bileşenlerinden olan memeliler, besin zincirinde hem av hem de avcı rolündedirler ve farklı iklim koşullarında ve oldukça farklı habitatlarda yaşamlarını sürdürebilirler. Büyük memeliler, birçok ekosistemdeki temel unsurlardır. Büyük etoburlar sıklıkla avlarının sayısını, dağılımını ve davranışını şekillendirirler. Büyük otçullar ise bitki örtüsünün yapısı ve tür bileşimini etkileyen ekolojik mühendisler olarak işlev görürler. Yaban hayvanların geniş alanlarda izlenmesi, periyodik kayıtlarının alınması, günlük aktivitelerinin izlenmesi oldukça zordur. Bu sebeple, memelilerin belirli bir alanda tespiti, aktivitelerinin ve bazı davranış özelliklerinin belirlenebilmesi, yayılış sınırlarının belirlenebilmesi için harekete ya da ısıya duyarlı tuzak kameralar (fotokapanlar) kullanılmaktadır. Fotokapanlar, özellikle mevcut arazi ve çevre koşullarının doğrudan gözlem yöntemine imkân vermemesi ya da dolaylı gözlemin daha etkili ve güvenli olması durumlarında kullanılmaktadır. Özellikle sarp arazilerde, yoğun vejetasyonla kaplı alanlarda ya da hedef türün geceleri daha aktif olması durumlarında fotokapanlar yaban hayatı araştırmalarında başarılı bir şekilde kullanılmaktadır.

Bu çalışmada, Sinop İli Erfelek ve Ayancık ilçeleri ormanlık alanlarında ağaç gövdelerine sabitlenen tuzak kameralar (fotokapan kameralar) (JUO JH-770 ve JUO JH-780) kullanılmıştır. 15 farklı kapan özellikle ormanlık bölgelerin geçit noktalarına yerleştirilmiş ve haftada bir veri kaydı alınmıştır. Çalışmalar 2022-2023 yılları arasında yürütülmüştür. Fotokapanlar, 20 farklı noktaya kurularak, toplam 304 gün arazide bırakılmıştır. Çalışma süresi içerisinde memeli hayvanlara ait değerlendirilebilir 117 adet video- resim elde edilmiştir. Fotokapan çalışmaları neticesinde Ayancık ve Erfelek ormanlık alanlarında, Carnivora, Cetartiodactyla ve Lagomorpha takımlarına dahil 8 büyük memeli yaban hayvanı tespit edilmiştir. Kaydedilen bu türler, Çakal (*Canis aureus*), Karaca (*Capreolus capreolus*), Tilki (*Vulpes vulpes*), Porsuk (*Meles meles*), Sansar (*Martes foina*), Tavşan (*Lepus europaeus*), Yaban domuzu (*Sus scrofa*) ve Yaban kedisi (*Felis silvestris*)'dir. Alanda en çok kaydedilen türler, *Vulpes vulpes* (tilki) ve *Martes foina* (sansar) iken en az kayıt *Felis silvestris* (yaban kedisi)'e aittir. Bu çalışma, alandaki büyük memeli çeşitliliğin tespitini sağlayarak, tür koruma çalışmalarına altlık sağlayacaktır.

Anahtar Kelimeler: Sinop, Erfelek, Ayancık, Mammals, Camera trap

DRAFT

ORAL PRESENTATION

Effect of reboxetine treatment on the cognitive parameters and the hippocampal BDNF levels of the scopolamine-induced amnesic rats

Nazlı Turan Yücel[1*] (*ORCID: https://orcid.org/0000-0002-0371-2703*)

[*1]Anadolu University, Faculty of Pharmacy, Department of Pharmacology, Eskişehir, Türkiye

*Corresponding author e-mail: nazlituran@anadolu.edu.tr

Abstract

Dementia is a degenerative disease which leads to a decrease in cognitive and intellectual functions. The number of medications approved for the treatment of dementia is fairly limited; thus, there is a prominent need for discovering and developing new agents beneficial for its pharmacotherapy. Reboxetine is a noradrenaline reuptake inhibitory drug prescribed for the treatment of major depression. In this study, it was aimed to investigate the anti-amnesic effect of this drug on rats and also the changes induced by reboxetine on the brain-derived neurotrophic factor (BDNF) levels in the hippocampal subfields. Male Sprague-Dawley rats (adult, weighted 250-300 g) were used in the experiments. The animals received reboxetine treatment at doses of 5 and 10 mg/kg/day (*i.p.*) for 14 days. Scopolamine (0,5 mg/kg, *i.p.*) was used to induce experimental amnesia model. Following the treatment procedure, spatial learning and memory parameters of the rats were evaluated by the Morris water maze test, while locomotor activity was assessed by activity cage test. Subsequently, alterations of the BDNF levels in the $CA_{1\text{-}2}$, CA_3, and dentate gyrus subfields of the hippocampus were investigated by immunohistochemical analyses. In the water maze test, escape latency values of the scopolamine-treated group were higher than the control group. On the other hand, reboxetine treatment reduced these values of the amnesic animals at both doses. Moreover, it was observed that reboxetine administrations significantly prolonged the time spent in the target quadrant when compared to the amnesic rats. Data from the immunohistochemical evaluations indicated that reboxetine also increased the reduced BDNF levels induced by scopolamine administration in the hippocampal formations of the amnesic rats. Obtained results pointed out that reboxetine treatment ameliorated the scopolamine-induced impairments and may have a potential to improve cognitive disorders if confirmed with validated clinical studies.

Keywords: Reboxetine, Amnesia, Morris water maze, BDNF, Hippocampus, Immunohistochemistry

DRAFT

ORAL PRESENTATION

Synthesis and antifungal activity studies of azole derivative compounds

Merve Saylam[1*] (*ORCID: https://orcid.org/0000-0002-7602-4565*), Aybala Temel[2] (*ORCID: https://orcid.org/0000-0003-1549-7219*), Halil Mert Karasu[1] (*ORCID: https://orcid.org/0009-0004-9861-7187*), Hilal Özdemir[1] (*ORCID: https://orcid.org/ 0009-0002-3941-9717*)

[*1]Izmir Katip Celebi University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, Izmir, Turkey.
[2]Izmir Katip Celebi University, Faculty of Pharmacy, Department of Pharmaceutical Microbiology, Izmir, Turkey.

*Corresponding author e-mail: merve.saylam@ikc.edu.tr

Abstract

Invasive fungal infections (IFI) pose a serious threat to immunocompromised patients and are associated with high mortality rates. The eukaryotic cell structure of fungal species makes it difficult for antifungal agents to provide selective toxic effects and many antifungal drugs with a broad spectrum cause serious side effects. In addition to the side effect profile like antibiotics, fungal species may develop natural or acquired resistance against antifungal drugs. Lack of knowledge about invasive fungal infections, uncertainties about the treatment strategies, and formulation deficiencies, as well as the drug resistance and associated side effects lead to the discovery of new antifungal agents in the treatment. Azoles (itraconazole, fluconazole etc.), which are the most widely used antifungal drugs in clinical practice today, show their effects by inhibiting ergosterol synthesis, a component of the fungal cell membrane. Within the scope of this study, synthesis, purification, and structure elucidation studies of potentially antifungal compounds bearing azole moieties. Additionally, minimum inhibitor concentrations (MIC) values have been determined against *Candida albicans* and *Candida parapsilosis.*

Keywords: invasive fungal infection, azole, drug discovery, antifungal

DRAFT

ORAL PRESENTATION

Covid-19'DA Fasl (844 T/C) Gen Polimorfizminin Rolü

Gülseren YILMAZ[1] (ORCID:0009-0005-5289-7061), Nihan BOZKURT[1] (ORCID: 0000-0002-2283-0828), Kübra ŞAHİN[1] (ORCID: 0000-0001-9870-0176), Sümeyya Deniz AYBEK[2*] (ORCID:0000-0001-9957-5485), Figen Güzelgül[3] (ORCID: 0000-0002-2796-9511), Sadegül ŞAVKIN[1] (ORCID: 0000-0002-9284-9102), Saime SEZER SONDAŞ[4] (ORCID:0000-0002-7849-3445)

[1]Tokat Gazi Osmanpaşa Üniversitesi, Tıp Fakültesi, Tıbbi Biyoloji AD, Tokat, Türkiye
[2]Ordu Üniversitesi, Tıp Fakültesi, Tıbbi Biyoloji AD, Ordu, Türkiye
[3]Tokat Gazi Osmanpaşa Üniversitesi, Tıp Fakültesi, Tıbbi Biyokimya AD, Tokat, Türkiye
[4]Malatya Turgut Özal Üniversitesi, Tıp Fakültesi, Tıbbi Biyoloji AD, Malatya, Türkiye

Corresponding autor e-mail: sumeyyadenizaybek@odu.edu.tr

Özet

Şiddetli akut solunum sendromu koronavirüs 2 (SARS-CoV-2) enfeksiyonu, Covid-19 küresel pandemisinden sorumludur. Dünya çapında yayılmasına rağmen, patofizyolojisi halen netlik kazanmayan Covid-19, asemptomatik olgulardan çoklu organ yetmezliğine ve ölüme sebep olan şiddetli solunum yetmezliğine kadar değişen, şok ve trombotik olaylarla karakterize karmaşık bir hastalıktır. SARS-CoV-2, epitel hücrelerini hedef alıp lokal inflamasyona yol açarak anti-koagülan ve protrombotik faktörler arasında bir dengesizliğe yol açmaktadır. Şiddetli SARS-CoV-2 enfeksiyonları lenfopeni ile karakterizedir, ancak ilgili mekanizmalar hala belirsizliğini korumaktadır. SARS-CoV-2 enfeksiyonunun aracılık ettiği lenfopeninin T hücresi apoptozu ile de ilişkili olabileceği düşünülmektedir. Yapılan bu çalışmada yoğun bakım ünitesindeki donör ile Covid-19 tanısı almamış sağlıklı donörlerle karşılaştırarak, Covid-19 hastalık seyrinde FasL 844 T/C polimorfizminin olası rolünün araştırılması amaçlanmıştır. Bu amaçla çalışmaya Tokat Devlet Hastanesine başvurup Covid-19 PCR sonucu pozitif olan, yoğun bakım ihtiyacı olan 25 kişi ve Covid-19 PCR test sonucu negatif olan 25 kişi dahil edildi. Çalışma gruplarından alınan kan örneklerinden DNA izolasyon kiti kullanılarak DNA izolasyonu yapıldı. FasL 844 T/C polimorfizmi için BsrDI restriksiyon enzimi kullanılarak restriksiyon fragmenti uzunluk polimorfizmi (RFLP-PZR) tekniği ile allel tipleri belirlendi. Verilerin istatistiksel analizi için *p* değeri 0.05'in altında bulunan değerler anlamlı kabul edildi. Çalışma sonuçlarımıza göre FasL 844 T/C gen polimorfizminde genotip dağılımı bakımından kontrol grubuna kıyasla hasta grubunda istatistiksel olarak anlamlı bir fark bulunmuştur (p= 0.02). Kontrol grubuna kıyasla hasta grubunda FasL 844 T/C gen polimorfizminin genotip dağılımında istatistiksel olarak anlamlı bir fark bulunması Covid-19'a yatkınlıkta bu polimorfizmin etkili olabileceğini düşündürmektedir.

Anahtar Kelimeler: Covid-19, Fas L, Polimorfizm, PCR-RFLP

DRAFT

ORAL PRESENTATION

Investigation of photocatalytic degradation and disinfection activity characteristics of one-dimensional TiO_2 nanorods decorated potato peel derived carbon dot (P-CD/TiO_2 NRs)

Melek Koç Keşir[1*] (*https://orcid.org/0000-0001-8905-2664*),
Mahmut Deniz Yılmaz[2] (*https://orcid.org/0000-0001-5793-0805*)

[*1]Karadeniz Technical University, Faculty of Science, Department of Chemistry, Trabzon, Turkey
[2]Necmettin Erbakan University, Faculty of Engineering-Architecture, Department of Basic Sciences, Konya, Turkey

[*]kocmelek612013@gmail.com

Abstract

In the recent years, carbon dot (CD) materials serve as a mediator to improve photocatalytic efficiency of TiO_2 nanomaterial derivatives by the way of Z-scheme heterojunction mechanism. The new type fluorescence CD samples possess photoluminescence and photoexcitation properties, photostability, excellent biocompatibility/environmental sustainability, low toxicity, and unexpensive of production of CD solution. As a starting carbon source, potato peel samples were utilized to synthesize the potato carbon dots (P-CD), since tons of potato peels wastes are being generated owing to increasing the consumed of potatoes and expansion of the chip industry. A novel potato peel carbon dot (P-CD) and CD impregnated TiO_2 nanorod photocatalyst (P-CD/TiO_2 NRs), which are used to yield good photocatalytic for degradation of Methyl Blue (MB), Hexavalent Chromium (Cr(VI)), Escherichia-Coli (e-coli) and Candida Albicans (C. albicans) in wastewater, were synthesized with fixed-bed pyrolysis and hydrothermal process without adding any kind of extra chemical reagent. The photocatalytic activity and photokilling effect of P-CD decorated TiO_2 nanorods were investigated under visible light. The structure, optical and electrochemical properties of as-prepared samples were characterized by means of TG-DTA, FTIR, UV-vis and fluorescence properties, XPS, TEM, SEM-EDS, and XRD analyses. The results showed that the photocatalytic performance of the P-CD/TiO_2 composite is slightly increased compared to that of pristine TiO_2 NRs after optimum conditions have been established.

Keywords: Biomass derived carbon dot, TiO_2 nanorod, MB degradation, Cr (VI) reduction, photokilling of e-coli and C. Albican

DRAFT

ORAL PRESENTATION

Cisplatin and Ethacrynic Acid Combination Treatment Inhibits Cell Proliferation of Cholangiocarcinoma Cells

Münevver Yenigül Karaca[1] (*ORCID: 0000-0003-0468-721X),*
Emel Başak Gencer Akçok[2*] (*ORCID: 0000-0002-6559-9144*)

[*1] Abdullah Gul University, Graduate School of Engineering and Science, Bioengineering Department, Kayseri, Turkey.
[2] Abdullah Gul University, Faculty of Life and Natural Sciences, Molecular Biology and Genetics Department, Kayseri, Turkey.

*Corresponding author e-mail: emelbasak.gencerakcok@agu.edu.tr

Abstract

Cholangiocarcinoma(CCA) is a heterogeneous group of malignancies formed by the differentiation of epithelial cells in the biliary tract and is the second most common primary liver tumor. The heterogeneity of CCA and its resistance to chemotherapy is one of the main reasons for treatment failure. Chemotherapy drugs used in cancer treatment either directly bind to DNA or target proteins necessary for cell division, causing cell death. Cisplatin is the most widely used chemotherapy drug among them. However, it has cytotoxic properties and the response of platinum in its structure is high, the disease will result in relapse. Glutathione-S-transferase(GST) is overexpressed in tumor cells, and its activity enhances chemotherapeutic drug detoxification. Ethacrynic acid(EA), a diuretic agent, is a GST enzyme inhibitor. By inhibiting GST in hematological malignancies and solid tumors, EA makes cells more sensitive. In this study, the effects of cisplatin combination with EA on CCA cell lines are examined for the first time. In order to investigate the cytotoxic effects, the CCA cell lines, TFK-1 and EGI-1, were treated with increasing concentrations of cisplatin and EA for 48h by MTT assay. Treating TFK-1 and EGI-1 cells with 2-100µM cisplatin reduced the growth of both cell lines in a dose-dependent manner by up to 64% and 85%, respectively. Besides, 10-150µM EA treatment decreased the proliferation of both cell lines in dose-dependent manner by up to 92% and 89%, respectively. The cells were treated with the IC50 of cisplatin (9,2µM for EGI-1 and 32µM for TFK-1) and increasing concentrations of EA (10µM-150µM). A significant decrease was observed in a concentration-dependent manner in response to increasing concentrations of EA in both cell lines compared to the control cells and only cisplatin. This study suggests a novel combination therapy approach for CCA after further studies are performed to reveal the mechanism of action.

Keywords: cholangiocarcinoma, cisplatin, ethacrynic acid, combination treatment

DRAFT

ORAL PRESENTATION

Türkiye'den iki *Asplenium* L. (Aspleniaceae) türünün spor morfolojisi

Züleyha ASLAN ERGENEKON[1*] (*ORCID: https://orcid.org/0000-0002-5078-2317*), Tülay EZER[2] (*ORCID: https://orcid.org/0000-0002-6485-5505*)

[*1]Niğde Ömer Halisdemir Üniversitesi, Fen Edebiyat Fakültesi, Biyoloji Bölümü, Niğde, Türkiye
[2]Niğde Ömer Halisdemir Üniversitesi, Mimarlık Fakültesi, Peyzaj Mimarlığı Bölümü, Niğde, Türkiye

*Sorumlu yazar e-mail: zuleyhaaslan@ohu.edu.tr

Özet

Asplenium L. cinsi Dünya'da 700'den fazla eğrelti türü ile temsil edilmektedir. Bu çalışmada Türkiye'de 17 türü doğal olarak yayılış gösteren *Asplenium* cinsine ait iki türün (*Asplenium ceterach* L. ve *Asplenium trichomanes* L.) spor morfolojileri incelenerek taksonomilerine katkı sağlanması amaçlanmıştır. Erdtman metodu ile hazırlanan spor preparatları, ışık mikroskobu (LM) ile incelenerek sporların morfolojik özellikleri belirlenmiştir. Yapılan incelemeler sonucunda her iki türün spor şeklinin oblat olduğu gözlemlenmiştir. *A. ceterach* monolet ve trilet sporlara sahip olup heteromorftur. Monolet sporlarda apertür uzunluğu 33-45 µm arasında değişkenlik göstermektedir. Boyut olarak orta büyüklükte sınıflandırılan sporların polar eksenleri ortalama 35 µm, ekvatoral eksenleri ise ortalama 48 µm'dir. *A. ceterach*'ın sporları, *A.trichomanes*'in sporlarından nispeten daha büyüktür. *A. trichomanes* ise yalnızca monolet sporlara sahiptir. Apertür uzunluğu 25-35 µm arasında değişkenlik göstermektedir. Sporların polar eksenleri ortalama 29 µm, ekvatoral eksenleri ise 42 µm'dir. Her iki türün sporlarının ekzin tabaka kalınlıkları birbirine yakın olup 1-2 µm aralığında değişiklik göstermektedir. Yapılan çalışmalar sonucunda elde edilen bulgular ilgili türlerin taksonomik sınıflandırılmasında spor morfolojilerinin de ayırt edici karakter olarak kullanılabileceğini göstermektedir.

Anahtar Kelimeler: *Asplenium, Asplenium trichomanes, Asplenium ceterach* Eğrelti, Spor morfolojisi.

DRAFT

ORAL PRESENTATION

Gümüş nanoparçacık (nanoAg) katkılı içi boş antibakteriyel nanolif yapıların üretimi ve karakterizasyonu

Hatice Bilge İŞGEN[1*](*ORCID: 0000-0002-6201-5836),* Sema Samatya YILMAZ[2] (*ORCID: 0000-0002-2682-2892),* Ayşe AYTAÇ[1,2] (*ORCID: 0000-0002-9566-7881)*

[1] Kocaeli Üniversitesi, Fen Bilimleri Enstitüsü, Polimer Bilimi ve Teknolojisi Anabilim Dalı, Kocaeli, Türkiye
[2] Kocaeli Üniversitesi, Mühendislik Fakültesi, Kimya Mühendisliği Bölümü, Kocaeli, Türkiye

*Sorumlu yazar e-mail: bilgecebislitr@gmail.com

Özet

Gümüş nanoparçacıklar (nanoAg'ler) optik, ısıl ve elektrik özelliklerinin yanı sıra antimikrobiyal etkileri sayesinde çok ilgi görmektedir. Gümüşün yüzyıllardır bilinen antimikrobiyal özelliğinin, nano boyutlarda görülen geniş yüzey alanı ve küçülen parçacık boyutu ile arttığı kanıtlanmıştır. Özellikle antibakteriyel lif olarak yara örtüsü, doku mühendisliği, gıda ambalajı, hava temizleme ve su arıtma sistemleri gibi alanlarda [1], ayrıca cerrahi, anesteziyoloji alanlarında kullanılan tıbbi alet ve malzemelerin kaplanması, tanı, yanık tedavisi, ilaç dağıtımı, kimyasal ve biyolojik sensörler, biyo-ilaçlar, biyo-işaretler ve medikal cihazlar dahil olmak üzere birçok tıbbi uygulamada, elektronik devreler ve kozmetikte de nanoAg'ler kullanılmaktadır. Elektroçekim yöntemi ise nanoAg katkılı nanoliflerin üretimini için kullanılan ve çeşitli alanlarda yüksek performans sağlayan bir uygulamadır. Bu çalışmada, eş-zamanlı elektroçekim yöntemi ile nanoAg katkılı ve katkısız içi boş Polibütilen Süksinat (PBS)/Termoplastik Poliüretan (TPU) nanoliflerinin üretimi ve karakterizasyonu yapılmıştır. Elde edilen iki-bileşenli nanoliflerin içyapısında saf polivinilpirolidon (PVP), dış yapısında ise nanoAg eklenmiş PBS/TPU polimer karışımı bulunmaktadır. PBS/TPU (60/40 a/a) çözelti karışımlarına kütlece farklı yüzde oranlarında (2; 4; 6) nanoAg eklenerek çeşitli parametrelerde çözünme işlemi gerçekleştirilmiş, ardından bu çözeltiler elektroçekim sürecinde kullanılmıştır. NanoAg katkılı nanolifler üretildikten sonra, saf etanol ile içyapıdaki PVP'nin çözünmesi sağlanarak, yapıdan uzaklaştırılmıştır. Dolayısıyla, içi boş polimer nanolifler üretilmiştir. Üretilen nanoliflerin yapısal özelliklerini ve antimikrobiyal aktivitelerini belirlemek için Fourier Dönüşümlü Kızılötesi Spektroskopisi (FTIR), Termo Gravimetrik Analiz (TGA) ve Antimikrobiyal Aktivite Analizi (AAA) testleri uygulanmıştır. FTIR ve TGA sonuçlarından, nanoliflerin çekirdeğindeki PVP'nin uzaklaştırıldığı ortaya koyulmuştur. Nanoliflerin antibakteriyel etkinliği *Escherichia coli* ve *Staphylococcus aureus* bakterilerine agar plak koloni sayma metodu uygulanarak belirlenmiştir. AAA sonuçlarına göre, karışımlarda nanoAg miktarının artmasına bağlı olarak antibakteriyel etkinlik artış göstermiştir. Katma değerli, nanoteknoloji ürünü PBS/TPU/nanoAg nanolifi biyomedikal uygulamalar için gelecek vaat etmektedir.

Bu çalışma Türkiye Bilimsel ve Teknolojik Araştırma Kurumu (TÜBİTAK) tarafından desteklenmiştir (Proje No: 122M880).

Anahtar Kelimeler: Nanolif, polimer, elektroçekim, gümüş nanoparçacık.

[1] Li, H.; Chen, X.; Lu, W.; Wang, J.; Xu, Y.; Guo, Y. Application of Electrospinning in Antibacterial Field. Nanomaterials **2021**, 11, 1822. https://doi.org/10.3390/nano11071822.

DRAFT

ORAL PRESENTATION

Investigation of the Effect of Cyanidine-3-Glucoside and Metformin in Pancreatic Cancer Cells

Seda Nur Özer[1*] (*ORCID: https://orcid.org/0009-0005-6502-6679*), Eray Kalabalıkoğlu[2] (*ORCID: https://orcid.org/0009-0008-6644-3513*), Burcu Ayhan Şahin (*ORCID: https://orcid.org/0000-0001-8321-0860*)

[*1]Istanbul Kultur University, Faculty of Science and Letter, Department of Molecular Biology and Genetics, Istanbul, Turkiye.

*Corresponding author e-mail: burcu.sahin@iku.edu.tr

Abstract

Pancreatic cancer is a type of aggressive cancer which progresses rapidly and has a significantly low survival rate. It was observed that, in patients, Type II Diabetes also develops before or after pancreatic cancer pathogeny. Metformin, a drug that is frequently used for Type II Diabetes, has been proved for anticarcinogenic effects besides its antidiabetic effects. It is also stated in literature that, elevated glucose level is related with epithelial-mesenchymal transition in tumor cells. Anthocyanins are edible plant secondary metabolites and found in red-purple fruits and vegetables in abundance. Cyanidine-3-glucoside (C3G) is an anthocyanin which has many significant roles as antioxidant, hepatoprotective, anti-inflammatory, antidiabetic and antiobesity. Its anticarcinogenic effect was also observed but the underlying mechanism has not been enlightened. In this study, metastatic (MIA PaCa-2) and non-metastatic (Capan-2) pancreas cancer cells were grown in low and high glucose media in order to *in vitro* mimic the glucose level elevation due to obesity or Type II Diabetes. Cell viability was assessed by MTT assay in response to C3G and metformin treatment. Cells were stained with H_2DCFDA and observed in order to see the intracellular reactive oxygen species (ROS) formation after treatment. Lipid droplets were observed by Bodipy staining. As a result, it was confirmed that, high glucose levels increased cell viability and cell survival in MIA PaCa-2 and Capan-2 cells in different manner. C3G and Metformin impedes not only cellular proliferation, but also 2D and 3D colony formation. When the cells were treated with both drugs, they showed synergistical effect. C3G and Metformin treatment also induced intracellular ROS generation. To conclude with, C3G and metformin co-treatment repress cellular proliferation and colony formation whether triggering ROS formation.

Keywords: Pancreas cancer, cyanidine-3-glucosite, metformin, type II diabetes, reactive oxygen species (Use commas to separate words.).

DRAFT

ORAL PRESENTATION

Synthesis and Characterization of P- and S-Containing Polymers in the Main Chain

Ufuk Saim Gunay [1,*](*ORCID: https://orcid.org/ 0000-0002-3085-3763*)

[1,*]Istanbul Technical University, Faculty of Science and Letters, Department of Chemistry, Istanbul, Turkey.

*Corresponding author e-mail: gunayuf@itu.edu.tr

Abstract

Phosphorus-containing polymers have long been regarded as highly advantageous in the field of polymer research, primarily due to their extensive utilization across different fields, notably in applications related to biology. Therefore, it is of vital importance for synthetic and industrial polymers to have phosphorus units. This not only expands their potential applications in the field of biology but also enhances their ease of processing and simplifies their operation for a wide range of applications. In this study, polymers containing phosphorus units in the main chain were synthesized and characterized. As a result of the reaction of the phenylphosphonic dichloride compound with various dithiol compounds, polymers containing S-P-S bonds in the main chain were obtained, respectively. First, 1,6-hexanedithiol was selected as a representative dithiol compound and reacted with the compound phenylphosphonic dichloride in the presence of triethylamine. The progress of the reaction was monitored, and polymerization was terminated after 6 hours. The obtained polymer had both P and S atoms in the main chain. Then, other dithiol compounds were used to obtain various polymer chains. All polymers were characterized with NMR and GPC techniques.

Keywords: phosphorus, dithiols, phenylphosphonic dichloride

DRAFT

ORAL PRESENTATION

Meat quality characteristics of Central Anatolian Merino and Ramlic lambs

Zekeriya Kiyma[1*] (*ORCID: https://orcid.org/ 0000-0002-1760-6442*),
Ousmane Barry[1] (*ORCID: https://orcid.org/ 0000-0002-9044-534X*)

[*1]Eskişehir Osmangazi University, Faculty of Agriculture, Department of Animal Science, Eskişehir, Türkiye.

*Corresponding author e-mail:zkiyma@gmail.com

Abstract

This study was conducted to reveal the physicochemical properties of different muscle types from Central Anatolian Merino (CAM) and Ramlic (RAM) lambs that were raised under producer conditions. For this purpose 7 CAM and 7 RAM lambs were slaughtered at 42kg live weight and meat samples were taken from the semimembranosus (SM), longissimus dorsi (LD), and semitendinosus (ST) muscles after the carcasses had been chilled at 4°C for 24 hours. Both muscle type and breed did not affect meat color parameters, however, interaction between studied breeds, muscle types, and time has been observed for L* and H° color parameters. Additionally, interactions between breed x muscle types and between time x muscle types were determined for meat a*, b*, and C values. It was determined that pH at 24 h. (pH24) were lower in CAM lambs compared to RAM lambs ($p<0.05$). Breed had no effect on water holding capacity (WHC), as well as thawing loss (TL). However, LD muscle had a higher value in both traits when compared to other muscles. CAM lamb had a higher cooking loss value, whereas SM muscle expressed higher CL ($p<0.05$). Meat hardness, springiness, and cohesiveness were not affected by breed or muscle type, chewiness was higher only in ST muscle. Drip loss (DL) was not affected by breed, however, both LD and ST muscles expressed higher DL values on day 7. Chemical analysis revealed that, despite the absence of substantial differences in certain characteristics, in general meat from RAM lambs showed better quality measures. Examining and comparing the meat quality of these breeds at different slaughter ages/weights will make it possible to reveal the meat quality of the breeds more clearly and to reveal the correct slaughter weights of the breeds for higher quality meat production.

Keywords: Muscle type, Meat quality, Ramlıç, Central Anatolian Merino.

DRAFT

ORAL PRESENTATION

Evaluation of lomefloxacin as a cholinesterase enzyme inhibitor: An *in vitro* and *in silico* analysis

Busra Dincer[1*] (*ORCID: https://orcid.org/0000-0002-3365-7741)*

[*1]Ondokuz Mayıs University, Faculty of Pharmacy, Department of Pharmacology, Samsun, Türkiye.

*Corresponding author e-mail: busra.dincer@omu.edu.tr

Abstract

Alzheimer's disease (AD) is a progressive neurodegenerative disorder marked by the gradual deterioration of cognitive function, including memory loss and difficulty with language, reasoning, and perception. It stands as the predominant etiological factor behind dementia, accounting for approximately 60-70% of the cases. Although its etiology remains incompletely elucidated, it is recognized that AD has a complex interplay of genetic, environmental, and lifestyle factors. AD's current treatments are primarily symptomatic and include the inhibition of acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) enzymes, which are involved in the breakdown of acetylcholine. However, there is a need to develop more efficacious treatments capable of delaying or halting the progression of the disease. This study aims to investigate the potential of lomefloxacin, a fluoroquinolone antibiotic commonly used to treat bacterial infections in AD. Results indicate that lomefloxacin exhibits low inhibition as a noncompetitive inhibitor of AChE and BChE at micromolar concentrations, with K_Is of 3.19 ± 0.21 µM (R^2: 0.9845) and 4.21 ± 0.20 µM (R^2: 0.9890), respectively, compared to the reference drug tacrine. Additionally, molecular docking studies have been conducted to elucidate lomefloxacin's binding interactions and affinities at the active site of AChE and BChE, offering detailed insights into its potential effects on AD.

Keywords: Alzheimer's disease, Acetylcholinesterase, Butyrylcholinesterase, Lomefloxacin.

DRAFT

ORAL PRESENTATION

Oğuzeli ve Karkamış (Gaziantep) ilçelerinde yayılış gösteren Asteraceae ve Lamiaceae taksonlarının etnobotanik özellikleri

Hülya ÇELİK (*ORCID: https://orcid.org/0009-0005-3936-8114*),
Mustafa PEHLİVAN *(ORCID:https://orcid.org/0000-0002-8277-6085*)

Gaziantep Üniversitesi, Fen-Edebiyat Fakültesi, Biyoloji Bölümü, Gaziantep, Türkiye

Sorumlu yazar e-posta: ihcelik@msn.com

Özet

Bu çalışmada Gaziantep ili Oğuzeli ve Karkamış ilçelerine ait 10'ar köy tespit edilerek 147 katılımcı ile yapılan görüşmeler sonucunda bu bölgede yayılış gösteren Asteraceae ve Lamiaceae familyalarına ait taksonların yerel adları ve kullanım amaçları ve kullanım şekilleri belirlenmiştir. Çalışma alanında Asteraceae familyasından doğal yayılış gösteren 20, yetiştirilen kültür bitkisi olarak 4 olmak üzere 24 taksonun, Lamiaceae familyasından ise doğal yayılış gösteren 10, yetiştirilen kültür bitkisi olarak 3 olmak üzere 13 taksonun etnobotanik açıdan kullanım alanına sahip olduğu belirlenmiştir. Asteraceae familyasındaki taksonların tıbbi (15), gıda (10), eşya (5) ve yem (2) amaçlı kullanıldığı tespit edilmiştir. En yaygın kullanımın *Gundelia tournefortii L. var. tournefortii L., Silybum marianum (L.)GAERTNER, Anthemis hyalina* DC., *Centaurea solstitialis* L. subsp. *solstitialis* L., *Tragopogon pterocarpus* DC. taksonlarına ait olduğu görülmüştür. Lamiaceae familyasındaki taksonların ise tıbbi (11), gıda (6) ve yem (1) amaçlı kullanıldığı tespit edilmiştir. *Thymbra spicata* L. var. *spicata* L., *Sideritis syriaca* L. subsp. *nusairiensis* (POST) HUB.-MOR., *Mentha longifolia* (L.) HUDSON subsp. *typhoides* (BRIQ.) HARLEY var. *typhoides* (L.) HUDSON, *Teucrium polium* L. taksonlarının en yaygın kullanıma sahip oldukları belirlenmiştir.

Anahtar Kelimeler: Etnobotanik, Asteraceae, Lamiaceae, Gaziantep, Oğuzeli, Karkamış

DRAFT

ORAL PRESENTATION

Theranostics: past, present and future

Ergin YALÇIN (*ORCID: https://orcid.org/0000-0003-3710-7242*)

Iskenderun Technical University, Department of Engineering Basic Sciences, Hatay, Türkiye

*Corresponding author e-mail: ergin.yalcin@iste.edu.tr

Abstract

Cancer remains a significant global health challenge, with traditional treatments facing limitations such as postoperative recurrence, harm to normal tissues, and the emergence of resistance. However, advancements in genomics, proteomics, and bioinformatics have paved the way for the development of new anticancer agents. These innovations aim to minimize drug misuse, improve the safety and precision of treatments, and offer more efficient and targeted therapies for cancer patients. Theranostics, which integrates therapy and diagnosis, presents an attractive approach for chemotherapy in medicine. It offers improved biodistribution, selective cancer targeting, reduced toxicity, masked drug efficacy, and minimal side effects. In this approach, diagnostic tools play a crucial role by collecting information about the diseased state before and after specific treatment, enabling effective cancer region targeting. The overexpression of reactive oxygen species (ROS), glutathione (GSH), thiols, acidic pH, and various enzymes within the tumor microenvironment provides valuable insights for refining the chemical structure of theranostics. This refinement enhances therapeutic efficacy and diagnostic tools, such as bioimaging, in comparison to healthy cells.

Keywords: Theranostic, cancer cell, overexpression, small molecules.

DRAFT

ORAL PRESENTATION

Synthesis, characterization and theoretical calculations of Schiff base derived from 3-amino-1,2,4-triazole-5-thiol as potent antimicrobial agent

Musa ALKAN[1*] (*ORCID:* https://orcid.org/0009-0004-5580-4839), Ayla BALABAN GÜNDÜZALP[1] (*ORCID*: *https://orcid.org/*0000-0002-5740-3674)

[1]Gazi University, Science Faculty, Department of Chemistry, Ankara, TURKEY

musaalkan1@gmail.com

Abstract

Schiff bases are important chemical compounds in various fields such as inorganic, analytical and medicinal chemistry due to their versatilities. Schiff bases are formed by the condensation of aldehydes or ketones with primer amines. Their active group called azomethine (CH=N) or imine (C=N) makes them ideal candidates for developing new drugs. They can form numerous stable complexes when they are coordinated with different transition metal ions. Thus, metal complexes of Schiff bases have been studied extensively due to their various applications and chemical activities. And also, the compounds containing triazole derivatives possess chemotherapeutic effects including antibacterial activities against drug-sensitive as well as drug-resistant pathogens. 1,2,4-triazoles are a very important class of compounds that have attracted the attention of many scientists in medical and pharmaceutical fields due to their various biological activities such as anticancer, antimicrobial, anticonvulsant.

In this study, we synthesized Schiff base derived from 3-amino-1,2,4-triazole-5-thiol (ATT) and 5-chlorosalicylaldehyde (5ClSA), and characterized using spectroscopic method. In theoretical studies, the molecular structure of Schiff base was optimized using the density functional theory (DFT) with B3LYP/6-31+G (d,p) level of theory. In addition, the frontier molecular orbitals (FMOs:HOMO and LUMO) and the molecular electrostatic potential (MEP) map were performed by the same basis set using Gaussian 09 software. The global reactivity descriptors such as electronegativity (χ), hardness (η), softness (S), chemical potential (μ) and electrophilicity (ω) were also calculated by using HOMO and LUMO energies. The optimized moleculer structure and electronic properties of schiff base was determined by computational studies using Gaussian 09 package program.

K**eywords**: Schiff bases, triazoles, DFT calculations, HOMO and LUMO energies

DRAFT

ORAL PRESENTATION

4-methoxybenzaldehydesulfisoxazole derived from Sulfa drug: Synthesis, characterization, theoretical calculations and antimicrobial studies

Selingün ÇAKIR[1*] (*ORCID: https://orcid.org/0000-0001-5226-3599*), Ayla BALABAN GÜNDÜZALP[1] (*ORCID: https://orcid.org/0000-0002-5740-3674*), Ümmühan ÖZDEMİR ÖZMEN[1] (*ORCID: https://orcid.org/0000-0001-9161-9367*), Ebru AKTAN[1] (*ORCID: https://orcid.org/0000-0001-9412-9160*), Ali Öztürk[2] (*ORCID: https://orcid.org/0000-0003-2428-1831*)

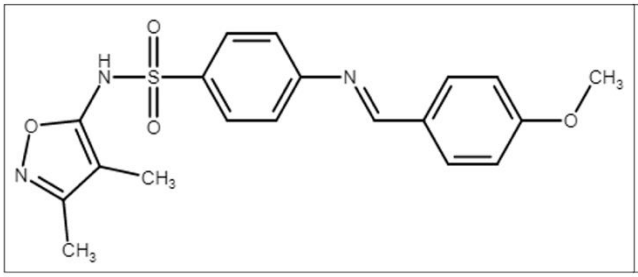
[1]Gazi University, Faculty of Science, Department of Chemistry, Teknikokullar, Ankara, Turkey
[2]Niğde Ömer Halisdemir University, Faculty of Medicine, Department of Medical Microbiology, Niğde, Turkey

*selinguncakir@gmail.com

Abstract

Sulfonamides are ones of the oldest synthetic antimicrobial agents and used in the treatment of many bacterial, protozoal and fungal infections. Although the use of penicillin and other antibiotics is increased, sulfonamides are still used actively in ophthalmic, urinary and gastrointestinal infections. Due to significant usage of sulfonamides/sulfonylhydrazines/sulfonylhydrazones in pharmacological applications, they have gained importance in medicinal chemistry as sulfa drugs. Sulfisoxazole is well known sulfonamide derivative as sulfa drug that are most commonly used in combination with trimethoprim as is the drug Bactrim. Schiff bases are known as imines or azomethines containing C=N moiety which is present in various natural and nonnatural compounds and responsible for their biological activities including antifungal, antibacterial, antiproliferative, anti-inflammatory, antiviral and antipyretic.

In this study, Schiff base; 4-methoxybenzaldehydesulfisoxazole was synthesized by the reaction of sulfisoxazole with 4-methoxybenzaldehyde and characterized by spectroscopic methods (^{1}H-NMR, ^{13}C-APT and FT-IR). In theoretical studies, the ground state geometry of Schiff base was obtained by Density Functional Theory (DFT) with B3LYP/6-31+G(d,p) basis set at Gaussian 09 package program. Frontier molecular orbitals (HOMO and LUMO), global reactivity descriptors and the molecular electrostatic potential surface (MEP) map were determined by this set. Also, antimicrobial activities of Schiff base were evaluated against reference bacterial and yeast strains (two Gram-negative bacteria; *Escherichia coli* ATCC 8739 and *Pseudomonas aeruginosa* ATCC 27853, two Gram-positive bacteria; *Staphylococcus aureus* ATCC 29213, and *S. epidermidis* ATCC 35984), and one fungus; *Candida albicans* ATCC 10231). Antibacterial activities of S2M-4-MetBA compound were found higher than the reference drugs. But, it was observed that the antifungal activity of the compound against *C. albicans* yeast is lower than the reference drug Fluconazole (Figure 1.).



	mg/mL
E.coli	0,005
P.aeruginosa	0,156
S.aureus	0,005
S.epidermidis	0,005
C.albicans	0,156

Figure 1. Antimicrobial activity results of compound

Keywords: Schiff base, Sulfisoxazole, Sulfa drugs, DFT calculations, Antimicrobial activities

DRAFT

DRAFT

1.2. FULL-TEXT PAPERS

DRAFT

DRAFT

DRAFT

ORAL PRESENTATION

Phosphate adsorption study on activated Albanian montmorillonite

Sonila Vito[1*], Ilirjana Boci[1], Maria Hoxhaj, Sidorela Vishkulli

[1]Department of Industrial Chemistry, Faculty of Natural Sciences, Tirana University, Albania

[*]sonila.vito@fshn.edu.al

Abstract

This paper presents the experimental results of the removal of aqueous phosphate ions through adsorption on activated clay. The selected clay is montmorillonite from the area of Prrenjas, Albania, while synthetic phosphate containing water is used. The clay is subjected to chemical and thermal activation to increase the adsorption capacity. Therefore, the impact of the activation method, as well as treatment parameters, on the adsorption capacity is studied. The influence of temperature and acid concentration is taken into consideration. First the kinetics and then the adsorption model is evaluated. The experimental results show that, among the considered models, the pseudo second order model describes better the process suggesting that the process is controlled by chemical adsorption. Both thermal and chemical treatment have a positive effect on increasing the adsorption capacity of phosphates from clay. The chemically treated clay exhibits better adsorption capacity, compared to the thermally treated one. In addition, the increase in the treatment temperature and acid concentration lead to improvement of phosphate removal efficiency by the clay. At all cases, a favourable multilayer adsorption on heterogeneous sites is noticed.

Keywords: montmorillonite, activation, adsorption, phosphate.

INTRODUCTION

In recent years, due to eutrophication concerns, treatment of liquid discharges containing phosphate compounds has taken on most of the scientific and technological interest. In this regard, various methods have been investigated and developed. They are based on the processes of sedimentation, coagulation/flocculation, filtration, biodegradation, adsorption, chemical precipitation, etc. Practiced alone or in combination with each other, to increase the cleaning efficiency, they have resulted efficient in removing not only phosphate concentrations but also a myriad of other pollutions. Among these technologies, adsorption is a simple, sustainable, cost-effective, and environmental-friendly technique (Rashid et al., 2021). It involves the use of a variety of adsorbents, with distinctive properties, from the conventional to novel materials, such as activated carbon, natural-sorbents (clay, zeolite), nano-sorbents, bio-sorbents, etc. (Chai et al., 2021).

Among the sorbents being studied for water treatment clays appear to be highly promising. It is found that they have distinctive ability to adsorb various water pollutants, being attributed to the high surface area, porosity, surface electrical charge, cation exchange capacity, acidity, and different types of active sites (Barakan and Aghazadeh, 2021). Many studies have documented this extraordinary ability of theirs for antibiotics (Chauhan et al., 2020), biocidal compounds, dyes (Teğin and Saka, 2021), other organic pollutants (Fairos et al., 2020), oils, heavy metals (Obayomi and Auta, 2019), nutrients (phosphates) (Huo et al., 2021), and so on. There are various types of clays used for this purpose: natural clays, activated clays (Komadel and Madejová, 2006), organo-clays, nano-clays (Annan et al., 2018), and each of them has been investigated for treatment of synthetic and real wastewater purposes. Among the many kinds of natural clay minerals, bentonite or montmorillonite has been most often used in adsorption due to large surface area, high CEC, and strong adsorption capacity (Barakan and Aghazadeh, 2021). They can be very effective in the natural state but may also need pre-treatment. There are different treatment techniques, but thermal and chemical (acid/base) activation have been reported the most effective ones, mostly acid than alkaline or thermal treatment. During the treatment procedure occurs the enlargement of the pore volume, specific surface area, and the number of surface acid sites able to sorb pollutants (Teğin and Saka, 2021).

Albania, although with a relatively small area, is rich in clays. They are widespread and found among sedimentary rocks, mostly terrigenous being part of molasses deposits. The largest clay deposits are found in the Near Adriatic Lowlands (UPA) and in inner lowlands. The most common clay formation eras are those of

DRAFT

the Neogene, Pliocene and Quaternary. The most widespread are the clays in the Lower and Middle Miocene to which are associated the largest sources of Tirana (Vora), Elbasan (Bradashesh), Alarup, Bilisht, etc. While in terms of quality, the best clays are the Piacesian clay deposits in Koplik, as well as the clays in Currila, Mamel, Polovina, in the valley of Shkumbin (Peqin), Prrenjas, Vlora, Shushica, etc. Despite their optimal characteristics, the use of clay in our country has been mostly for construction practices (to produce cement, brick or tile). Although the environmental benefits of using clays are universally recognized in principle, in Albania industrial exploitation for these purposes has not yet started. Even the research studies in this direction have only developed in recent years (Xhaxhiu et al., 2013), (Mele et al., 2018), (Prifti and Xhaxhiu, 2021). From the literature review these studies are based on the structure characterization of Prrenjasi, Brari, Currila, Dardha clays and evaluation of the adsorptive capacity for some metals (Pb dhe Cd) and pesticides (dimethionate and methomyl). The effect of their activation by thermal (Keri et al., 2017) and acidic methods (Mele et al., 2018) (sulphuric acid) has been investigated. From the batch experiments for synthetic water samples the adsorption isotherm of the cited pollutants and the clay adsorption capacity were observed. It is worth noting that the clays behaved very well towards acidic activation. The authors concluded that their use for surface or groundwater treatment to remove the studied pollutants would be of great environmental interest.

Based on these scientific study circumstances, the contribution in this direction is considered interesting. Thus, further on, it is presented the experimental work, which aims at the determination of the ability of Albanian montmorillonite to adsorb phosphate ions present in synthetic wastewaters. Furthermore, in order to understand the mechanisms according to which the process occurs, the kinetics and adsorption isotherms are also studied. This information would be necessary for the conventional wastewater treatment systems design (Ho and McKay, 1999).

MATERIALS AND METHODS

Clay sampling and pre-treatment: The clay used for this study is montmorillonite of the region of Prrenjas in Albania. It is sampled according to ISO 22475-1:2021 standard method, at 20cm depth. Firstly, it is subjected to pre-treatment for the sand fraction removal by water rinsing and sedimentation technique. For that, clay and distilled water are mechanically mixed at 100g/l ratio for 2 hours. After mixing, the suspension is left to rest for 10 min for sand particles sedimentation. Upon separation of the sediment from the supernatant, the later one is subjected to the same procedure for other 3 minutes of sedimentation. The suspension is then left to settle for 24 hours. The decanted material (the clay) is filtered and dried at 90°C until constant weight.

Thermal and chemical treatment of Clay: The thermal treatment of clay is performed at temperatures 500°C and 600°C for 2 hours. Whilst the chemical treatment was achieved through sulphuric acid contact for 3 hours and in the presence of heat. The process is carried out in a flask equipped with a refrigerant, to capture the acid vapours. After treatment the flask is cooled and the clay is separated from the liquid by decantation. It is then rinsed with distilled water until the natural pH is reached. Further, it is dried until constant weight, ground to less than 100μm particle size diameter and used for the evaluation of adsorption capacity. The acid concentrations used in the study are 15, 27 and 40% (evaluated as mass of acid/mass of clay).

Batch experiments for the study of phosphate adsorption on clay: All clay-synthetic wastewater treatment experiments are realized after the same protocol. In a series of chemical flasks, 50ml of phosphate solutions (synthetic wastewater), with certain concentration, are treated with 1g clay, under mechanical stirring. After a certain contact time, they are subjected to centrifugation and the supernatant is analysed for phosphate content.

The synthetic wastewater samples, of different phosphate concentration, are prepared by dilution of the 200mg/l KH_2PO_4 stock solution. The latter one is prepared by dissolving 0.2865g KH_2PO_4 in 1l distilled water.

The kinetics study batch experiments are performed for same initial aqueous phosphate concentration and different contact times. Whilst, the adsorption isotherm experiments are performed for different initial phosphate concentration and same contact time as that evaluated as the equilibrium time by the kinetics studies.

Aqueous phosphate spectrophotometric determination method (APHA, 1998): The reagents used are: ammonium molibdate solution (9.5 g of $(NH_4)_6Mo_7O_{24}\cdot 4H_2O$ dissolved in 100ml water); 4.5M sulphuric acid; ascorbic acid solution (7g $C_6H_8O_6$ diluted in 100ml water); potassium antimonile tartrate solution (3,25g of $K(SbO)C_4H_4O_6\cdot 1/2H_2O$ diluted in 100ml water); mixed reagent (45ml of ammonium molibdate solution is added in 200mL sulphuric acid and 5mL of potassium antimonile tartrate solution).

In 20ml of water sample is added 0.5ml of ascorbic acid, 1ml of combined reactive and water till 25ml volume. After 15 minutes the absorption value in the wave length 880nm is determined. Measurement are performed using UV 1200 spectrophotometer.

Data analysis for adsorption kinetic model evaluation: Three adsorption kinetic models have been taken in consideration: pseudo first-order model; pseudo second-order model; Elovich model; and intraparticle diffusion model. Nonlinear regression is applied for all cases, as strongly recommended by literature (Kostoglou and Karapantsios, 2022), and the analyses have been made based on the regression coefficient (R^2).

The pseudo first-order model: $q_t = q_e(1 - e^{K_1 t})$ (1)

The pseudo second-order model: $q_t = \frac{{q_e}^2 * K_2 * t}{1 + K_2 * q_e * t}$ (2)

Elovich model: $q_t = \frac{1}{\beta}\ln(\alpha\beta t)$ (3)

Intraparticle diffusion model: $q_t = c + t^{0.5} K_{diff}$ (4)

Where: t is the time of adsorption (hour); q_t is the amount of phosphate adsorbed at time t (mg/g clay); q_e is the amount of phosphate adsorbed until equilibrium time is reached (mg/g clay); K_1 is the pseudo first-order model constant (min^{-1}); K_2 is the pseudo second-order model constant (min^{-1}); α is the initial adsorption rate (mg/g.min) and β is adsorption constant; c and K_{diff} are the intraparticle diffusion model constants.

Evaluation of q_t and q_e is done according to equations:

$q_t = \frac{(C_0 - C)V}{M}$; $q_e = \frac{(C_0 - C_e)V}{M}$ (5)

Two adsorption isotherms has been considered for this purpose: Langmuir isotherm and Freundlich isotherm; as two of the most used ones in the literature. Linear regression has been considered for both of them.

Linear Langmuir isotherm: $\frac{C}{q_e} = \frac{1}{ab} + \frac{1}{a}C$ (6)

Linear Freundlich isotherm: $\ln(q_e) = \ln(K_F) + \frac{1}{n}\log(C)$ (7)

Where: C is the concentration of phosphate in solution at time t (mg/l) of adsorption; q_e is the amount of phosphate adsorbed until equilibrium time is reached (mg/g clay); a and b are the Langmuir constants; K_f and n are the Freundlich constants.

RESULTS AND DISCUSSION

Adsorption kinetics study. Evaluation of the adsorption equilibrium time

The study of sorption kinetics in wastewater treatment is important to understand the detailed mechanism and characteristics of the pollutant removal process. Since it provides information about the solute uptake rate, it determines very important process parameters such as the time required for reaching the adsorption equilibrium (adsorbate-adsorbent residence time). Furthermore, it enables the mathematical modelling of the process, facilitating the prediction of the behaviour of the system in different conditions (Ho and McKay, 1999).

For that, 50ml of synthetic phosphate wastewater, with initial concentration 3ppm, is treated for varying contact time, in the range of 0.5-24hours, with 1g of 500°C thermally activated clay. The experimental results of the phosphate removal over time are presented in figure 1.

DRAFT

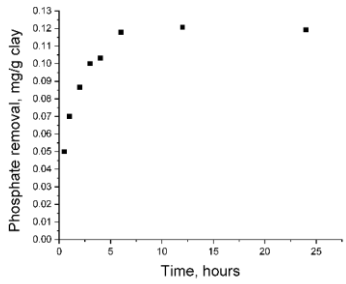


Figure 1. The phosphate removal, mgPO_4^{3-}/g clay, over time.

It is observed that with the increase of the contact time, the amount of phosphate ions adsorbed by the clay increase, due to the presence of the abundance free adsorptive sites in the clay surface, to asymptotically approach the maximum value of 0.12mgPO_4^{3-}/g clay at times greater then 6hours. After this time the adsorption capacity remains constant, indicating that all the adsorptive sites have been fully occupied. Thus, the minimum time that clay needs to show maximum adsorption capacity (adsorption equilibrium time) is 6 hours. All the following experiments are carried out at this contact time.

Further, to understand the mechanism according to which the process occurs, as well as to evaluate the mathematical model that best describes it, four kinetic models are taken in consideration: Pseudo first-order model, pseudo second-order model, Elovich model, and intraparticle diffusion model. Nonlinear regression methodology is used. The experimental results are presented in figure 2.

It can be seen that the adsorption of phosphate ions in thermally treated clay corresponds better to the pseudo second-order model, as similarly by other studies (Edet and Ifelebuegu, 2020), (Cheng et al., 2018), with a correlation coefficient, R^2, of 0.9815. This indicates that the removal of phosphate from the solution to the clay is due to physicochemical interactions between the two phases. Thus, the process is controlled by chemical adsorption and the adsorption rate is dependent on adsorption capacity of the clay (not on concentration of phosphate) (Ho and McKay, 1999). The pseudo second order rate constant, k_2, resulted 9.69hour^{-1}. Furthermore, the experimental results also fit the Elovich model, with a correlation coefficient, R^2, 0.8764. The initial adsorption rate and the adsorption constant, α and β, resulted respectively 0.82mg/g*hour and 52.27.

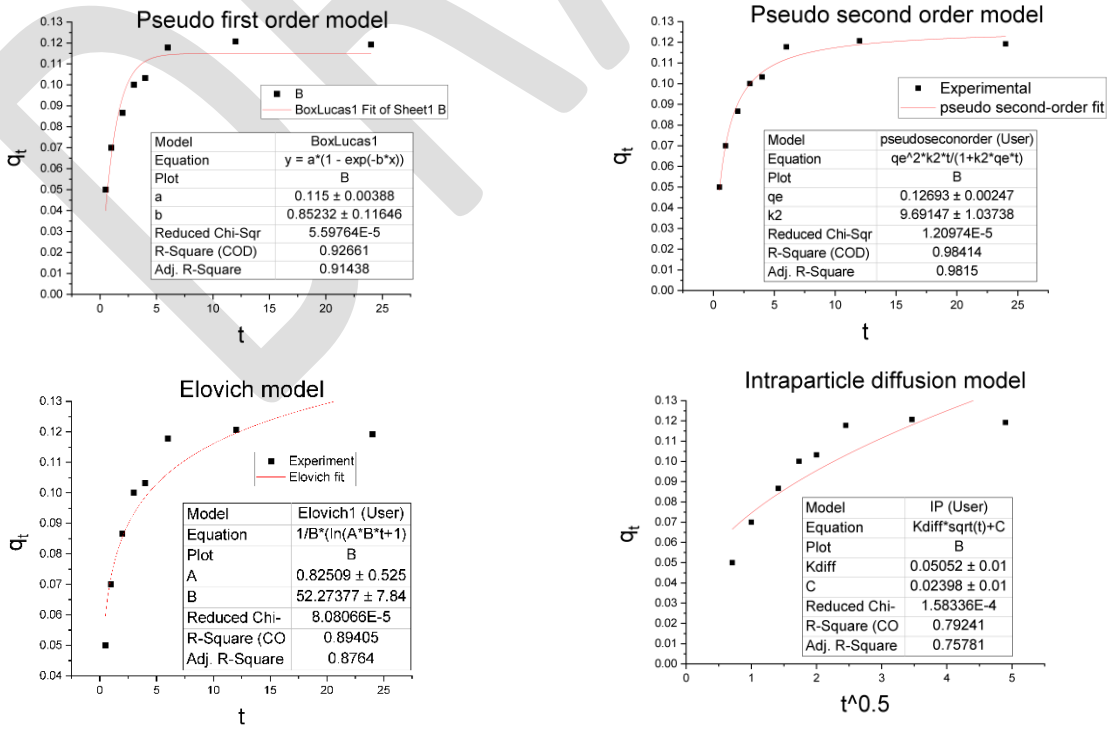


Figure 2. The kinetic models for the adsorption of phosphate ions in montmorillonite.

Evaluation of adsorption isotherms

The aim of these experiments is to evaluate the adsorption capacity of the clay samples taken in consideration, as well as the mathematical approach that represents the process. Thus, the influence of the initial concentration of the phosphate aqueous solution on the adsorption capacity of clays has been studied. The batch experiment series consist on the treatment of 50ml of phosphate solution, with an initial concentration of 2-6mg/l, with 1g of clay, for a contact time of 6 hours. After this time, the solution is centrifuged and the supernatant is analysed for phosphate content. The experimental results of the dependence of phosphate adsorbed, in mg/g clay and %, versus the initial concentration are presented in figure 3 and 4. The thermal activation is realized in two temperatures 400 and 500°C. While chemical activation is achieved by treatment of the clay with sulphuric acid at concentrations, in mass percentage with respect to the mass of clay, 15%, 27% and 40%.

As can be seen, for all cases, the increase in the solution initial concentration (for the considered concentration range) results in an increase of the adsorbed phosphate amount. It seems that the initial concentration of phosphate ions in the solution is so low that the number of active positions on the surface of the clay is sufficient to adsorb them. Therefore, the effect of ion competition is not relevant. It is also established that both thermal and chemical treatment have a positive effect on increasing the adsorption capacity of phosphates from clay. More specifically, the chemically treated clay exhibits better adsorption capacity, compared to the thermally treated one. In addition, both the increase in the treatment temperature and acid concentration lead to increase of the adsorption capacity of the clay. Thus, clay activated with 40% sulphuric acid exhibits better adsorption capabilities, with 0.28mg/g of clay maximum adsorption and 99.5% phosphate removal rate, compared to the 500°C clay, with 0.21mg/g clay maximum adsorption and 81.78% removal rate, and natural clay, with 0.15mg/g clay maximum adsorption and 57% removal rate. However, it should be noted that, although the increase in acid concentration leads to an increase in phosphate removal capacity, the rate of increase decreases with increasing concentration. The curves for Clay 27 and Clay 40% are close to each other. Apparently, as stated by literature (Teğin and Saka, 2021), calcination has caused dihydroxylation of the structure. It has changed the structure of the clay from a uniform thickness distribution and thick layer firmly stacked on the surface to a more heterogeneous and rough one, thus increasing the porosity and therefore the adsorption sites density.

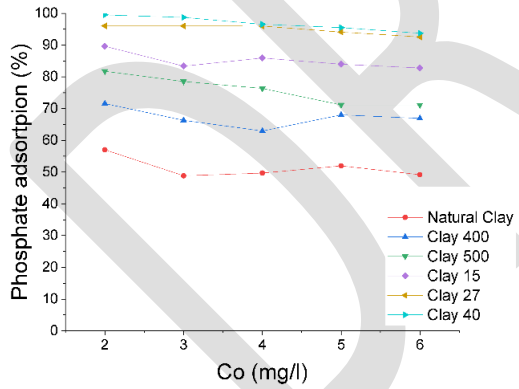


Figure 3. Experimental results of phosphate adsorption efficiency vs initial concentration.

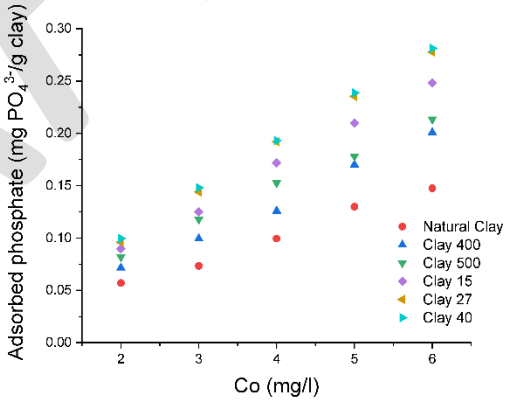


Figure 4. Experimental results of phosphate adsorption capacity in mg/g clay vs initial concentration.

While the explanation for the behaviour of chemically activated clay can be explained by the fact that because of the acid treatment of the clay surface, the reaction of the surface cations (like Al^{3+}, Fe^{3+} and $Mg^{2+}$from the octahedral and tetrahedral sites as well as Na^{+}, K^{+}, and Ca^{2+}) with the acid has occurred to give sulphates, easily leached from the structure, and living behind active sites readily to adsorb. The reduction in phosphate removal from clays activated at higher acid concentrations may be a result of the excessive leaching of OH^{-} groups and central Al cations from the tetrahedral layers due to the deeper penetration of the acid, providing a solid surface that contains unchanged layers and amorphous silica (Teğin and Saka, 2021).

To evaluate the adsorption isotherm, the dependence of the adsorbed phosphate amount against the equilibrium concentration of the solution is graphed, figure 5. Langmuir and Freundlich isotherms are taken into

consideration, figure 6, 7, 8, 9. Linear regression has been applied and analysis are been made upon R^2 correlation coefficient. Table 2 summarizes the data of the isotherm coefficients and the respective correlation coefficients. It is noticed that, in most cases, adsorption takes place after Freundlich isotherm, with R^2 greater than 0.93. For clay chemically activated with sulphuric acid of concentration 27 and 40%, it happens that the adsorption also matches the Langmuir isotherm with R^2 slightly higher than that of Freundlich, respectively 0.97 and 0.95 for clay 27%.

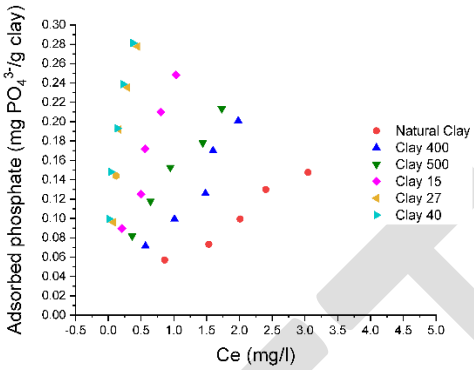


Figure 5. Experimental results of adsorption isotherm of phosphate ions in clays.

However, it can be generalized that in all cases, a multilayer adsorption on heterogeneous sites occur. In addition, it could be concluded that phosphate adsorption on these clays is advantageous, as the Freundlich constant values, 1/n, are in the range 0-1, implying for a monolayer chemical adsorption process.

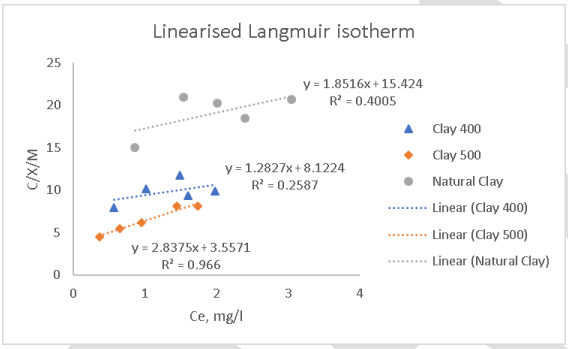


Figure 6. Linearized Langmuir isotherms for the adsorption of phosphate on thermal activated clays.

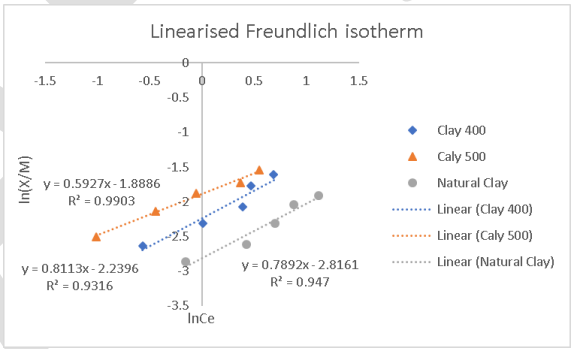


Figure 7. Linearized Freundlich isotherms for the adsorption of phosphate on thermal activated clays.

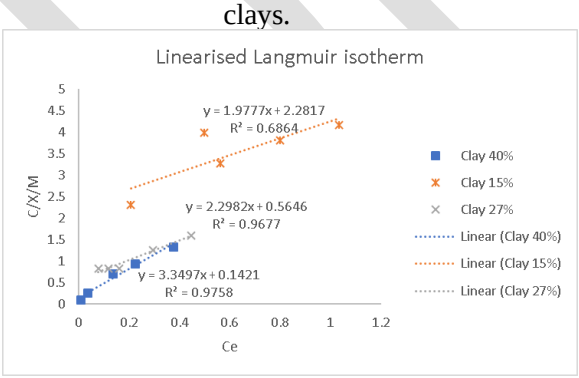


Figure 8. Linearized Langmuir isotherms for the adsorption of phosphate on chemical activated clays.

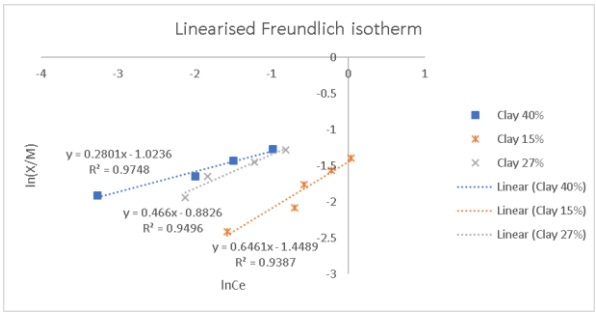


Figure 9. Linearized Freundlich isotherms for the adsorption of phosphate on chemical activated clays.

Table 2. The adsorption isotherm constants and regression coefficients.

Clay	Langmuir Isotherm			Freundlich Isotherm		
	a	b	R^2	K_F	1/n	R^2
Natural Clay	0.54	0.12	0.40	0.06	0.79	0.947
400	0.78	0.16	0.26	0.11	0.81	0.932
500	0.35	0.80	0.97	0.15	0.59	0.99
15%	0.30	23.57	0.69	0.36	0.28	0.94
27%	0.51	0.87	0.97	0.23	0.65	0.95
40%	0.44	4.07	0.98	0.41	0.47	0.98

CONCLUSIONS

The experimental results of synthetic phosphate wastewater treatment by Albanian activated montmorillonite concluded that this clay is very effective in water depollution from phosphates. This characteristic is reinforced even more through thermal and chemical treatment of the clay. The chemical treatment (with sulphuric acid) resulted much more effective than the thermal one. Furthermore, the adsorption efficiency is a factor of the temperature and the acid concentration used for activation. The dependence of the change in their value (within the studied limits) is proportional to the change in adsorption effectiveness. Meanwhile, the adsorption process of the thermally treated clay at 500°C, corresponds to the pseudo second-order model, indicating that the removal of phosphate from the solution to the clay is due to physicochemical interactions between the two phases. Thus, the process is controlled by chemical adsorption and the adsorption rate is dependent on adsorption capacity of the clay. In addition, it is noticed that, adsorption takes place after Freundlich isotherm indicating for a multilayer adsorption on heterogeneous sites and it is advantageous.

REFERENCES

Annan E, Agyei-Tuffour B, Bensah YD, Konadu DS, Yaya A, Onwona-Agyeman B, Nyankson E 2018. Application of clay ceramics and nanotechnology in water treatment: A review. Cogent Engineering, 5(1), 1–35. https://doi.org/10.1080/23311916.2018.1476017

Barakan S, Aghazadeh V 2021. The advantages of clay mineral modification methods for enhancing adsorption efficiency in wastewater treatment: a review. In Environmental Science and Pollution Research, Vol. 28, Issue 3. https://doi.org/10.1007/s11356-020-10985-9

Chai WS, Cheun JY, Kumar PS, Mubashir M, Majeed Z, Banat F, Ho SH, Show PL 2021. A review on conventional and novel materials towards heavy metal adsorption in wastewater treatment application. Journal of Cleaner Production, 296, 126589. https://doi.org/10.1016/J.JCLEPRO.2021.126589

Chauhan M, Saini VK, Suthar S 2020. Ti-pillared montmorillonite clay for adsorptive removal of amoxicillin, imipramine, diclofenac-sodium, and paracetamol from water. Journal of Hazardous Materials, 399, 122832. https://doi.org/10.1016/J.JHAZMAT.2020.122832

Cheng P, Chen D, Liu H, Zou X, Zhang Y, Xie J, Qing C, Chen T 2018. Enhanced adsorption capacity for phosphate in wastewater from thermally activated flue gas desulfurization gypsum. Journal of Chemical Technology and Biotechnology, 93(6), pp. 1733–1741. https://doi.org/10.1002/jctb.5546

Edet UA, Ifelebuegu AO 2020. Kinetics, Isotherms, and Thermodynamic Modeling of the Adsorption of Phosphates from Model Wastewater Using Recycled Brick Waste. Processes, 8(6), 665. https://doi.org/10.3390/pr8060665

Fairos S, Shattar A, Zakaria A, Keng & Foo Y 2020. One step acid activation of bentonite derived adsorbent for the effective remediation of the new generation of industrial pesticides. https://doi.org/10.1038/s41598-020-76723-w

Ho YS, McKay G 1999. Pseudo-second order model for sorption processes. Process Biochemistry, 34(5), pp. 451–465. https://doi.org/10.1016/S0032-9592(98)00112-5

Huo J, Min X, Wang Y 2021. Zirconium-modified natural clays for phosphate removal: Effect of clay minerals. Environmental Research, 194, 110685. https://doi.org/10.1016/J.ENVRES.2020.110685

Keri J, Mele A, Fetallari M, Ylli F 2017. Physical – Chemical changes during thermal treatment of Prrenjas clay. The Austrian Journal of Technical and Natural Sciences 7-8, Premier Publishing, Vienna –Prague, pp. 51 – 58.

Komadel P, Madejová J 2006. Chapter 7.1 Acid Activation of Clay Minerals. Developments in Clay Science, 1(C), pp. 263–287. https://doi.org/10.1016/S1572-4352(05)01008-1

Kostoglou M, Karapantsios TD 2022. Why Is the Linearized Form of Pseudo-Second Order Adsorption Kinetic Model So Successful in Fitting Batch Adsorption Experimental Data? Colloids and Interfaces, 6(4). https://doi.org/10.3390/colloids6040055

Mele A, Taraj K, Korpa A 2018. Acid Activation of Prrenjas Clay Mineral. Journal of Engineering & Processing Management, 4(7), pp. 37–44. https://doi.org/10.7251/jepmen1507037m

Obayomi KS, Auta M 2019. Development of microporous activated Aloji clay for adsorption of lead (II) ions from aqueous solution. Heliyon, 5(11), e02799. https://doi.org/10.1016/J.HELIYON.2019.E02799

Prifti E, Isak N, Xhaxhiu K 2021. Comparison of the Adsorptive Properties of Some Albanian Clays Toward Dimethoate and Methomyl. International Journal of Ecosystems and Ecology Science (IJEES), 11(2), pp. 277–292. https://doi.org/10.31407/ijees11.212

Rashid R, Shafiq I, Akhter P, Iqbal MJ, Hussain M 2021. A state-of-the-art review on wastewater treatment techniques: the effectiveness of adsorption method. Environmental Science and Pollution Research, 28(8), 9050–9066. https://doi.org/10.1007/s11356-021-12395-x

Teğin İ, Saka C 2021. Chemical and thermal activation of clay sample for improvement adsorption capacity of methylene blue. International Journal of Environmental Analytical Chemistry. https://doi.org/10.1080/03067319.2021.1928105

Xhaxhiu K, Sula L, Shllaku J, Xhelaj A, Kota T 2013. Comparison of the Adsorption Capacities of Two Albanian Clays for the Removal of Metribuzine from Contminated Waters. 12(3–4), pp. 949–965.

DRAFT

ORAL PRESENTATION

Determining the Change of hTERT and PTEN Protein, Total mRNA and Transcript Variant Levels in YKG-1 and HUVEC Upon Capsaicin Treatment

Sabahattin Cömertpay[1,2] (0000-0003-4850-6927), Feyza Nur Zabun[1,*] (000-0002-9595-061X)

1: Kahramanmaraş Sütçü İmam University, Department of Biomedical Engineering and Sciences, Kahramanmaraş, Turkey.

2: Kahramanmaraş Sütçü İmam University, Faculty of Agriculture, Department of Agricultural Biotechnology, Kahramanmaraş, Turkey.

* Corresponding Author: feyzanurzabun@gmail.com

Abstract

The term 'transcript variants' refers to different mature mRNAs generated from the same precursor mRNA during post-transcriptional modifications. It is an established fact that these variants can acquire important roles in cancer pathways and can have opposing effects despite being produced from the same gene. In this study, the effects of capsaicin on protein, total mRNA and transcript variant levels of human telomerase (hTERT) and tensin homolog (PTEN) were examined in a cancerous [glioblastoma cell line (YKG-1)] and a healthy [(human umbilical vein endothelial cells (HUVEC)] human cell population. Firstly, the cells were treated with capsaicin for 24 hours, and IC_{50} values for YKG-1 and HUVEC were determined as 351.1 µM and 365.5 µM, respectively, through MTS assay. According to the statistical analysis, the healthy HUVEC cells were found to be significantly resistant to capsaicin than YKG-1 ($p<0.05$). Subsequently, Western Blotting was performed to observe the changes in human telomerase and PTEN protein levels. It was found that capsaicin treatment caused a slight decrease in PTEN levels while triggering an increase of approximately 20% in hTERT protein levels in both cell types. When the relative levels of total mRNAs were examined by qRT-PCR, a statistically significant increase was observed in PTEN and hTERT for both cell types ($p<0.05$), except for the PTEN in YKG-1, which turned out to increase by ethanol, the solvent used to solve capsaicin. Subsequently, an attempt was made to determine the changes in the variants expressed from the PTEN and hTERT gene regions, respectively consisting of 3 and 4 variants. As a result, it was found that capsaicin treatment led to an increase in variant expressions in all cases, and this increase was statistically significantly higher than both the control group and the ethanol-treated group ($p<0.05$).

Keywords: Post-transcriptional Modifications, Cancer, human, Alternative Splicing, pepper.

Kapsaisin Muamelesi Sonrası YKG-1 ve HUVEC Hücrelerinde hTERT ve PTEN Protein, Toplam mRNA ve Transkript Varyant Seviyelerinin Belirlenmesi

Özet

Transkript varyantları, DNA'da kodlanmış tek bir genden üretilen öncül mRNA'nın post-transkripsyonel değişimler sırasında, bazı ekzon kısımlarının çıkarılması ile oluşturulan farklı olgun mRNA'ların her birine verilen isimdir. Bu varyantların, kanser yolaklarında önemli görevler edinebildiği ve aynı genden üretilmelerine rağmen farklı varyantların birbirlerine zıt etkilere neden olabildiği bilinmektedir. Bu çalışmada, kapsaisinin bir glioblastoma hücre hattı (YKG-1) ile sağlıklı hücreleri temsil ettiği düşünülen HUVEC hücrelerinde insan telomeraz (hTERT) ve tensin homolog (PTEN) genlerinden üretilen protein, toplam mRNA ve transkript varyantlarına nasıl etki ettiği gösterilmeye çalışılmıştır. Bunun için öncelikle hücreler kapsaisinle 24 saat muamele edilerek YKG-1 ve HUVEC hücreleri için IC_{50} değerleri MTS testi aracılığıyla sırasıyla 351,1 ve 365,5 µM olarak tespit edilmiştir. Yapılan istatistiksel analizde kapsaisinin hücreler üzerindeki etkisinin istatistiksel olarak anlamlı derecede farklı olduğu ($p<0.05$) ortaya konulmuştur. Sonrasında, IC_{50} değerinde kapsaisinle muamele edilen hücrelerde insan telomeraz ve PTEN protein seviyeleri Western Blotlama ile incelenmiş; çalışılan iki hücre tipinde de kontrol grubuna kıyasla, PTEN protein seviyesinde küçük bir azalma, hTERT protein seviyesinde %20 dolaylarında bir artış gözlenmiştir. hTERT ve PTEN gen bölgelerinden ifade edilen toplam mRNA'ların bağıl seviyelerine qRT-PCR yöntemiyle bakıldığında, yine her iki hücre tipi için hem PTEN hem de hTERT mRNA seviyelerinde kontrol grubuna kıyasla istatistiksel olarak anlamlı bir artış gözlenmiştir ($p<0.05$). Ancak, PTEN'in YKG-1 hücrelerindeki artışı dikkatle incelendiğinde, gözlenen bu değişimin, kapsaisinin çözücüsü olarak kullanılan etanolden kaynaklandığı ortaya çıkarılmıştır. Ek olarak,

PTEN ve hTERT gen bölgelerinden ifade edilen transkript varyantlarının kapsaisin uygulamasıyla nasıl değiştiği saptanmaya çalışıldığında; hücre tipine ve çalışılan varyanta bağlı olarak şiddetlerinde farklılıklar görünse de tüm durumlarda kapsaisin uygulamasının varyant ifadelerinde artışa neden olduğu ve bu artışın hem kontrol grubundan hem de etanol ile muamele edilmiş gruptan istatistiksel olarak anlamlı ölçüde yüksek olduğu tespit edilmiştir ($p<0.05$).

Anahtar Kelimeler: Post Transkripsiyonel Modifikasyonlar, Kanser, insan, AS, biber

INTRODUCTION

Messenger RNA (mRNA) is produced through a process called transcription, which converts the genetic information of a gene into an RNA molecule. In eukaryotic cells, following transcription, post-transcriptional modifications are carried out to prepare the precursor mRNA molecules for translation and to regulate their movement within the cell. One of these modifications involves the removal of introns present in the precursor mRNA chain. During the process of intron removal, occasionally exons can also be removed, resulting in the production of different mature mRNA molecules from the same precursor mRNA. This process, which can be summarized as alternative splicing (AS), leads to different forms of mature mRNAs being generated. Each of these mRNAs is named as a transcript variant. Transcript variants can have different functions and activities, and they can vary depending on the cell's role and environmental conditions. These variants are also important for regulating gene expression levels and tissue specificity such that their misregulation can lead to diseases (Keren et al., 2010).

Cancer is a group of diseases that share certain common characteristics, such as uncontrolled cell division, evasion of apoptosis, production of their own growth signals, insensitivity to growth inhibitory signals, or the ability to create their own blood vessels (Hanahan and Weinberg, 2011). Despite significant advancements in treatment methods in recent years, cancer remains one of the leading causes of death worldwide (World Health Organization, 2023).

The uncontrolled and unlimited capacity for cell division in cancer cells, also known as immortality, has been attributed to the high activity of an enzyme called telomerase. Telomeres are non-coding "TTAGGG" repeat DNA sequences located at the terminal ends of all vertebrate chromosomes (Montpetit et al., 2014). Telomere length decreases by 50-150 base pairs (bp) per cell division in human somatic cells in culture. While telomerase activity is very low in somatic cells due to tight regulation of hTERT (Cifuentes and Shippen, 2012), the majority of malignant tumors exhibit significantly elevated levels of telomerase (Kim et al., 1994; Koziel et al., 2011; Shay and Wright, 1996). The hTERT gene in humans produces four different transcript variants, and the roles of these variants are not yet fully understood (Khosravi-Maharlooei et al., 2015).

PTEN is another signaling molecule known to play an important role in cancer development. Its role in limiting the cell proliferation in healthy cells makes it an important tumor suppressor (Guo et al., 2018). In cancer cells, the PTEN gene is either completely lost or it its function is vastly reduced (Dillon and Miller, 2014). Three transcript variants expressed from the PTEN gene have been identified, and ongoing discussions revolve around the roles of these variants in cancer cells (Hasle et al., 2019).

Capsaicin is a sharp-smelling, pungent phenolic (8-methyl-N-vanillyl-6-nonenamide) compound synthesized in plants of the Capsicum genus (Cao et al., 2015). Numerous studies conducted thus far have demonstrated that capsaicin has diverse biological and physiological functions, such as pain relief (Maihofner and Heskamp, 2013), anti-obesity (Baboota et al., 2014) and anti-inflammatory properties (Toyoda et al., 2016). In addition, the anti-cancer properties of capsaicin have also drawn the attention of researchers in recent years (Cao et al., 2015; Helvaci and Comertpay, 2018; Comertpay and Ozcelik Demirbanka, 2020). In this study, YKG-1 glioblastoma cancer cell line and HUVEC (Human Umbilical Vein Endothelial Cells), which could represent human healthy tissue, were treated with capsaicin. Telomerase (hTERT) and PTEN proteins in the cells were detected using Western blotting, and the relative quantities of total RNA were measured using qRT-PCR. In the same cells, the changes in the expression of four hTERT (hTERT1, hTERT2, hTERT3, and hTERT4) and three PTEN (PTEN1, PTEN2, and PTEN3) transcript variants upon capsaicin treatment were also determined using qRT-PCR technique.As far as our knowledge is concerned, this study is the first attempt to explore the effects of capsaicin application on transcript variants in glioblastoma and HUVEC cells.

DRAFT

MATERIALS AND METHODS

Cell Culture

YKG-1 (ATCC, USA) and HUVEC (ATCC, USA) cells available in our laboratory were cultured in DMEM (Dubelco's Modification of Eagle's Medium; Hyclone, USA) supplemented with 10% FBS (Fetal Bovine Serum; Hyclone, USA) and 1% Penicillin-Streptomycin (invitrogen, USA). The cells were grown in an incubator (Panasonic, Japan) at 37°C with 5% CO_2.

Determination of IC_{50} Values

After cell counting, the wells of a 96-well plate were seeded with 5,000 cells per well. To achieve the desired concentration of capsaicin, different concentrations including 0 were used. Each concentration was seeded in a column with 4 replicates. The cells were seeded with 100 µL of culture medium. In each concentration, two wells without cells were reserved to measure the absorbance of the culture medium. The concentrations for YKG-1 cells were 0-100-150-200-225-250-275-300-325-350-400 µM, and for HUVEC cells, the concentrations were 0-100-150-200-250-300-350-400-450-500-550-600 µM. The continuation of the process was carried out following the method described by Helvaci and Comertpay (2018) to obtain the IC_{50} values.

Treatment of Cells with Capsaicin

The calculated IC_{50} value for each cell type was used for the treatment. The treatment was carried out as described by Turan (2019). In summary, 1,173,000 cells were seeded in three T75 flasks. After overnight incubation at 37°C and 5% CO_2, the medium in the flasks was replaced with capsaicin-containing medium at the IC_{50} concentration (M2028; Sigma-Aldrich, Germany) (C), medium containing an equivalent amount of ethanol (E), or plain medium (K). After 48 hours, live cells were counted and re-seeded into capsaicin-containing (or ethanol-containing) medium. This process was repeated after 96 hours, and live cells were harvested at 120 hours.

Western Blotting (WB)

The procedure was carried out following the method described by Comertpay and Ozcelik Demirbanka (2020). Briefly, protein was isolated from the cells collected after capsaicin treatment, and 50 µg portions of these proteins, along with a protein marker (abcam, USA), were loaded onto an SDS-PAGE gel, with each in a separate well. Subsequently, the proteins were electrophoretically separated based on their sizes, transferred onto a PVDF membrane, and the membrane was incubated with antibodies against telomerase (abcam-ab32020, USA), PTEN (abcam-ab133532, USA), mTOR (abcam-25880), and GAPDH (ThermoFisher, PA1-9046, USA) as a control. Following incubation, specific secondary antibodies were applied to each primary antibody, and chemiluminescence was generated using an ECL Substrate (Huang et al., 2009). The membranes were then photographed using a UVP device, and the obtained images were analyzed using Image J software. The intensities of the observed bands were expressed numerically. Band values obtained for the studied proteins (Telomerase, PTEN) were normalized by dividing them by the values of GAPDH obtained from the same membrane, and then they were further normalized by dividing them by the value obtained for the control group.

Measurement of Relative mRNA Levels by qRT-PCR

This step was performed following the method described by Comertpay et al. (2022). After total RNA isolation from the cells in the C, E, and K groups, cDNAs were synthesized. Specific primers were designed to determine the total amount of human telomerase reverse transcriptase (hTERT) and PTEN mRNAs, obtained from the National Center For Biotechnology Information (NCBI) (2023a, 2023b). These primers can be seen in Table 1. The beta-actin (β-Actin) primer, generated using the same method, was used as an internal control. After performing qRT-PCR, the Ct values obtained from triplicate measurements for each sample were compared, and the 2^-ΔΔCt formula was used to calculate the relative mRNA levels (Schmittgen and Livak, 2008).

Table 1. Sequences of primers used in RT-PCR analysis and the expected product length (bp)

Primer Name	Sequence	Expected Product Length (bp)
Beta-Actin (Forward)	CCCTGGACTTCGAGCAAGAG	323
Beta-Actin (Reverse)	GATCTTCATTGTGCTGGGTGC	
PTEN (Forward)	CACACGACGGGAAGACAAGTTC	161
PTEN (Reverse)	CCTCTGGTCCTGGTATGAAGAATG	
Telomerase (Forward)	CCACATAGGAATAGTCCATCC	220
Telomerase (Reverse)	TTTACTCCCACAGCACCTC	

Variant-Specific Analysis

Variant-specific analysis was conducted as described above, after determining the relative quantities of total RNA expressed from each gene region. qRT-PCR was performed using the primers listed in Table 2 to determine how the levels of each transcript variant produced from the PTEN and hTERT gene regions, according to the NCBI database, changed. It was assumed that the designed primers would enable the discrimination of variants.

Table 2. a. Primers designed for the separation of 4 variants transcribed from the hTERT gene region and the resulting variant after the relevant amplification.

	PRIMER			Recognized Variants	Amplified Variants
hTE.RT	1	Forward	TCTTCCTACGCTTCATGTG	1, 2	1
		Reverse	AGTAGTCGCTCTGCACCT	1	
	2	Forward	TCTTCCTACGCTTCATGTG	1, 2	1, 2
		Reverse	GCAAGACCCCAAAGAGTT	1, 2, 3, 4	
	3	Forward	ACGGGCGCGTACGACACCAT	1, 2, 3	1, 2, 3
		Reverse	AAACAGCTTGTTCTCCATGT	1, 2, 3, 4	
	4	Forward	TGAAGGCACTGTTCAGCGT	1, 2, 3, 4	1, 2, 3, 4
		Reverse	AAACAGCTTGTTCTCCATGT	1, 2, 3, 4	

DRAFT

Primer designed for the separation of the variant transcribed from the PTEN gene region and the resulting variant after the relevant amplification.

	PRIMER			Recognized Variants	Amplified Variants
PTEN	1	Forward	GCTGGCACATCCAGGGA	1	1
		Reverse	GATTGTATATCTTGTAATGG	1, 2	
	2	Forward	GCGGCACATCCAGGGA	2	2
		Reverse	GATTGTATATCTTGTAATGG	1, 2	
	3	Forward	GATGTAGTAAGTTGTGCTG	3	3
		Reverse	TTAGCCTTGGCCTCTACA	3	

Statistical Analysis

Graphs representing the IC_{50} values of the cells were generated based on the MTS results, and they were compared using two-way ANOVA with GraphPad Prism software (CA, USA). The p-value given under the "interaction" heading in the results table was taken into consideration. Numerical results obtained from the qRT-PCR experiments were compared between groups ("C," "E," and "K") or treatments using the Student's t-test, also with GraphPad Prism software. In all comparisons where the p-value was equal to or less than 0.05, the difference between the compared groups was considered statistically significant, and this data was indicated with an asterisk (*) on the generated graphs. If the p-value was less than 0.01, 0.001, or 0.0001, the corresponding symbols (**, ***, ****) were placed on the graph at the respective points.

RESULTS

Calculation of IC_{50} values for cell sensitivity to capsaicin

Previous studies have determined IC_{50} values for cancer cell lines, which formed the basis of our study. However, considering that the passage of time may have led to differences in cytotoxicity, and in order to achieve simultaneous results with the healthy cell line HUVEC, IC_{50} values for capsaicin were determined for all cells. The results are shown in Figure 1.

DRAFT

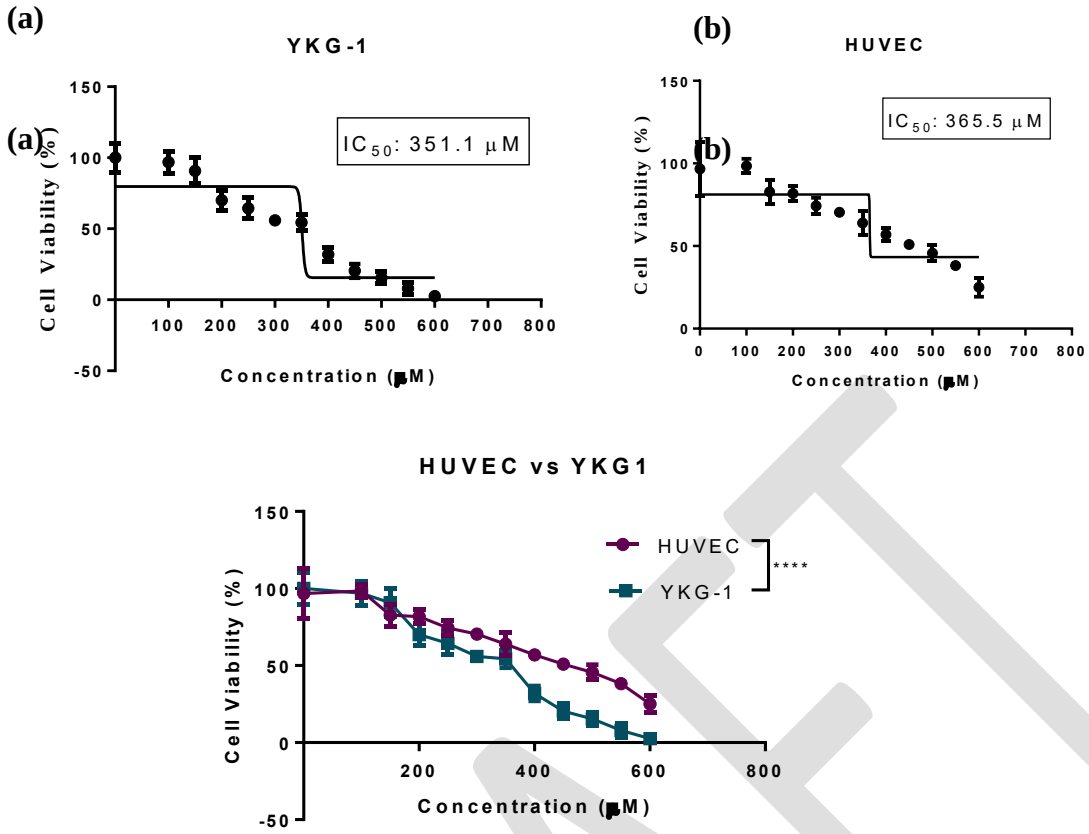


Figure 1. Measurement of cytotoxicity of capsaicin on (a) YKG-1 and (b) HUVEC cell lines using MTS and the corresponding IC_{50} values determined. (c) Comparison of differences in cell viability vs. concentration graphs between the cells using two-way ANOVA.

PTEN Protein Levels

Cells treated with capsaicin (C), cells treated with ethanol (E), and experimental control cells (K) were used to isolate proteins based on the calculated IC_{50} value for each cell line. PTEN protein levels were evaluated using Western Blot technique. The results are presented separately for each cell line (Figure 2).

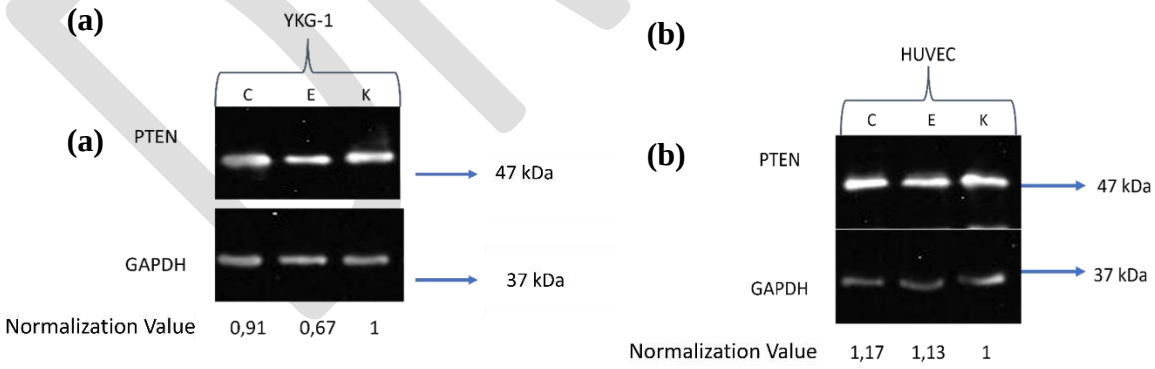


Figure 2. Relative levels of PTEN protein in (a) YKG-1 and (b) HUVEC cells treated with capsaicin. GAPDH was used as a loading control, and the calculated value in the Control Group was set as 1.00, representing the levels in other groups. Normalization values reflect the relative level of the protein.

DRAFT

Relative PTEN mRNA Levels

The relative evaluation of the total PTEN levels in the mRNA obtained from YKG-1 and HUVEC cells in groups (C), (E), and (K) is shown in Figure 3. The relative evaluations of the quantities corresponding to each variant are presented below.

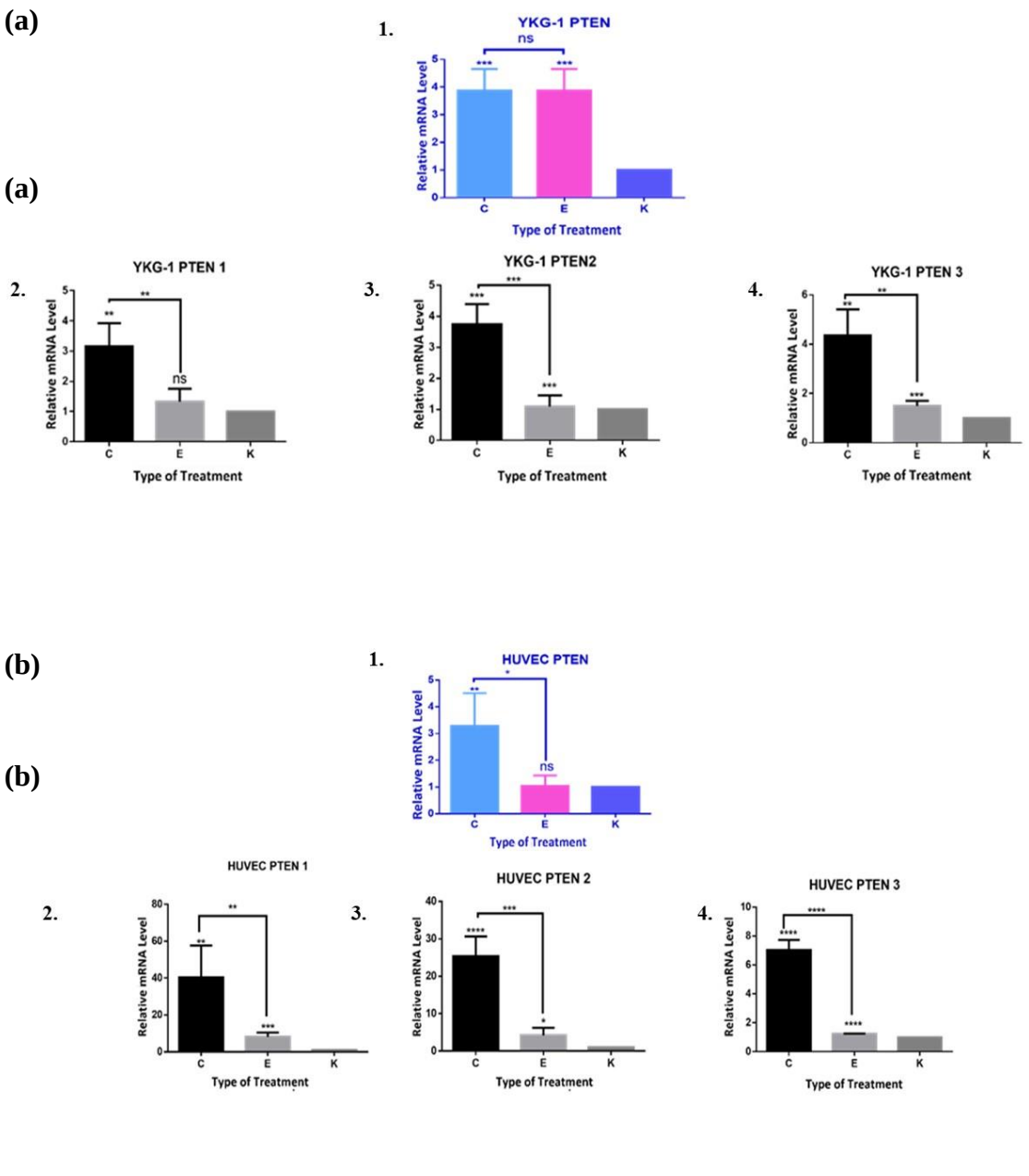


Figure 3. (a) Relative mRNA levels of (1) total PTEN, (2) PTEN1, (3) PTEN2, (4) PTEN3 in YKG-1 cells after capsaicin treatment. (b) Relative mRNA levels of (1) total PTEN, (2) PTEN1, (3) PTEN2, (4) PTEN3 in HUVEC cells after capsaicin treatment. C: Cells treated with capsaicin, E: Cells treated with ethanol, K: Control cells grown in regular medium. Experiments were repeated twice. C and E groups were compared to K, and the indicator bar representing the p-value was placed on the bar. The comparison between the C and E groups is represented by a line that appears to merge the two bars. *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001, ns: not statistically significant. β-Actin was used as an internal control.

hTERT Protein Levels

Proteins were isolated from cells treated with capsaicin, cells treated with ethanol, and experimental control cells based on the calculated IC_{50} value for each cell line. hTERT protein levels were evaluated using Western Blot technique. The results are presented separately for each cell line (Figure 4).

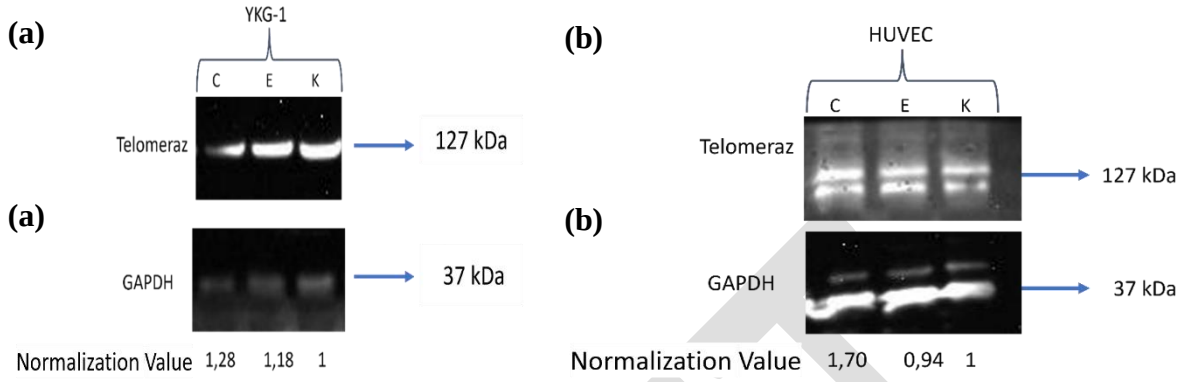


Figure 4. Relative levels of hTERT protein in YKG-1 and HUVEC cells treated with capsaicin.

C: Cells treated with capsaicin at the IC50 value for 120 hours. E: Cells treated with ethanol at the same volume as capsaicin. K: Control cells grown in regular medium without any additions. GAPDH was used as a loading control, and the value calculated in the Control Group was accepted as 1.00, representing the levels in other groups. Blue arrows indicate the size of observed protein bands in kilodaltons. Normalization values reflect the relative levels of the protein.

Relative mRNA Levels of hTERT

Following the protein measurements, the relative levels of both total hTERT and each hTERT variant were measured in the RNA isolated from cells subjected to the same treatments (Capsaicin, Ethanol, Control) (Figure 5.). Since hTERT has 4 different variants according to the NCBI database (Materials and Methods; 3.6. Variant-Specific Analysis), the results are presented based on these variants.

DISCUSSION

Our study is the first to investigate the effect of capsaicin at the IC_{50} level in HUVEC and YKG-1 cells in terms of transcript variants expressed from the hTERT and PTEN coding gene regions in response to capsaicin treatment in the cell lines used.

The first phase of our study involved determining the cytotoxic effects of capsaicin. Commercially obtained capsaicin was dissolved in ethanol and applied to our cell lines at varying concentrations. Subsequently, the MTS assay was performed to determine the IC_{50} value for each cell line. This approach has been demonstrated previously for the cell lines used in our study. When the IC_{50} values were evaluated together, a significant difference was found among them ($p<0.05$). Specifically, glioblastoma cells were highly sensitive to capsaicin, while HUVEC cells were less affected in terms of sensitivity to capsaicin.

Finding evidence for the use of YKG-1 cells in capsaicin studies is difficult in the literature. In a master's thesis completed by Alsayed (2019), the IC_{50} value for this cell line was determined to be 280 µM. In the present study, this value was measured as 351.1 µM (Figure 1). This difference can be understood with the explanation provided by Ben-David et al. (2018). According to their explanation, cells maintained in culture can behave differently towards the same agent due to genetic and transcriptional changes they undergo.

Regarding HUVEC cells, the cytotoxicity of capsaicin has been evaluated in different studies. For example, Chularojmontri et al. (2010) reported that 25 µM capsaicin did not induce cytotoxic effects in HUVEC cells. Although the direct determination of capsaicin IC_{50} was not reported in their study, Chularojmontri's report aligns with our observations. Indeed, the IC_{50} value for HUVEC was found to be high, at 365.5 µM.

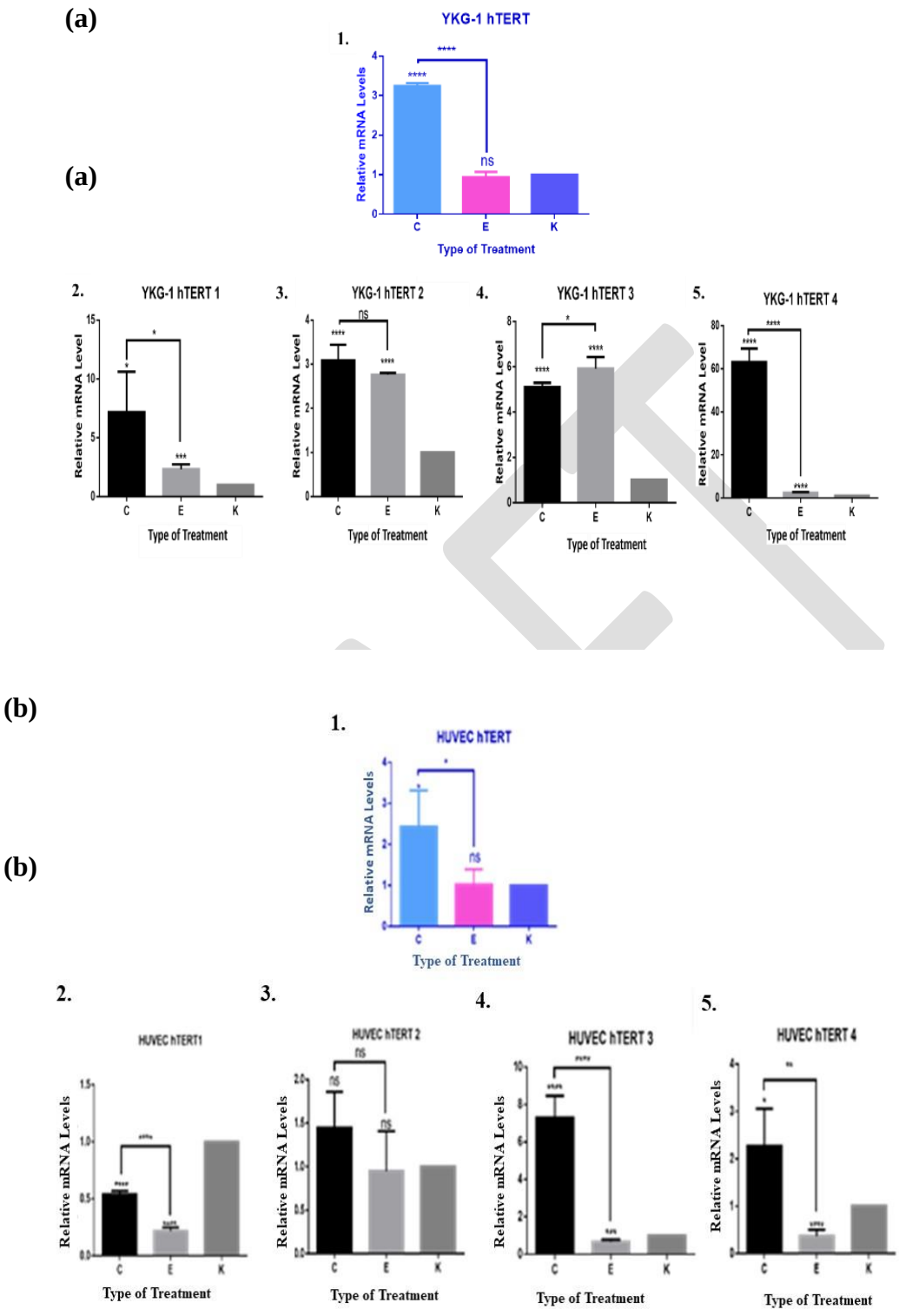


Figure 5. (A.) Relative mRNA levels of total hTERT and hTERT1, hTERT2, hTERT3, and hTERT4 variants in YKG-1 cells after capsaicin treatment. (B.) Relative mRNA levels of total hTERT and hTERT1, hTERT2, hTERT3, and hTERT4 variants in HUVEC cells after capsaicin treatment. C: Cells exposed to capsaicin; E: Cells exposed to ethanol treatment; K: Control cells grown in regular medium. Experiments were repeated twice. C and E groups were compared with K group, and the significance level (p-value) was indicated on the error bars. The comparison between C and E groups was represented by a line that appears to merge the two bars. *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001, ns: not statistically significant. β-Actin was used as an internal control.

In the next phase, capsaicin was applied to the cells at the determined IC_{50} value, and mRNA and protein were isolated from the cells along with those treated with ethanol, and untreated control group. No significant changes were observed in PTEN protein levels following capsaicin treatment. Our findings contradict the literature information indicating the suppression of the PTEN pathway in capsaicin-treated cancer cells (Sánchez et al., 2019), suggesting that PTEN suppression may not exist, at least in the studied cells. However, it is also possible that the decrease in PTEN protein levels observed by Sánchez et al. can be explained by a substantial halt in protein synthesis or the degradation of certain unstable proteins. Another possibility is that the differences between our results and the literature can be attributed to the cell type studied and the behavioral differences developed by cells depending on their environmental conditions, as mentioned by Ben-David et al. (2018).

On the other hand, the observed PTEN levels in HUVEC will be evaluated solely based on our results since there is no study specifically examining PTEN levels in response to capsaicin in this cell type. Our observation of a roughly 17% increase in PTEN aligns with the role of this molecule in cell survival (Kuo et al., 2017), suggesting that, although slight, the increased PTEN levels in cells that survived capsaicin stress resulted from a population selection in which cells with higher PTEN levels resisted the treatment and were subsequently subjected to protein and RNA isolation.

In all cells studied, an increase in the relative value of total PTEN mRNA was observed using a common primer recognizing all variants. This observation is consistent with our previous findings in YKG-1 cells (Alsayed, 2019). For HUVEC cells, since no such study has been conducted before, it is challenging to compare our results. However, the fact that both the protein and mRNA levels point the same direction suggests that HUVEC cells might have activated suitable molecular pathways for cell survival.
The protein level of telomerase in YKG-1 cells exhibited an increase of approximately 28%. Considering the role of telomerase in cell survival (Cifuentes et al., 2012), this increase suggests that the upregulation of the relevant enzyme in cells that survived capsaicin treatment may enhance cell viability. A similar observation was made in HUVEC cells, where the difference was calculated to be around 70%.

As for the limiting factors of our study, it was not possible to design separate hTERT primers for each variant due to the high similarity between the variants (NCBI, 2023a). The analysis of hTERT variants was generally weaker compared to the analysis of PTEN variants. This can be attributed to the difficulty in distinguishing hTERT variants solely based on primer design and the susceptibility of Nested PCR to errors. Therefore, it is crucial to have the results related to hTERT confirmed through repeated studies in the conclusion section of the article.

CONCLUSION

i. The IC_{50} value of capsaicin was determined as 351.1 µM for YKG-1 cells and 365.5 µM for HUVEC cells.

ii. Capsaicin treatment does not seem to significantly affect PTEN and but increase the telomerase protein levels in cells.

iii. Capsaicin treatment increased the relative levels of total hTERT and PTEN mRNA in both cell types. However, this affect might be the result of ethanol for PTEN in YKG-1.

iv. Capsaicin treatment led to an increase in all PTEN variants in HUVEC and YKG-1 cells.

v. In YKG-1 cells, capsaicin treatment increased the levels of all hTERT transcript variants compared to control. However, ethanol also increased the hTERT2 and hTERT3 levels.

vi. Capsaicin treatment resulted in a decrease in hTERT1 variant and an increase in hTERT3 and hTERT4 variants in these cells while hTERT2 remained unaffected.

ACKNOWLEDGEMENT

This study was supported by the Scientific and Technological Research Council of Turkey (TÜBİTAK) under Project No. 120 Z 775.

DRAFT

REFERENCES

Alsayed MM, 2019. "In vitro Examination of the Effect of Capsaicin on YKG-1 Glioblastoma Cell Line" *In Turkish* - Master's Thesis, Institute of Science, Kahramanmaraş Sütçü İmam University, Faculty of Science.

Baboota RK, Murtaza N, Jagtap S, Singh DP, Karmase A, Kaur J, Bhutani KK, Boparai RK, Premkumar LS, Kondepudi KK, Bishnoi M 2014. Capsaicin-induced transcriptional changes in hypothalamus and alterations in gut microbial count in high fat diet fed mice, The Journal of Nutritional Biochemistry, 25(9), 893-902.

Ben-David U, Siranosian B, Ha G, Tang H, Oren Y, Hinohara K, Strathdee CA, Dempster J, Lyons NJ, Burns R, Nag A, Kugener G, Cimini B, Tsvetkov P, Maruvka YE, O'Rourke R, Garrity A, Tubelli AA, Bandopadhayay P, Tsherniak A, Vazquez F, Wong B, Birger C, Ghandi M, Thorner AR, Bittker JA, Meyerson M, Getz G, Beroukhim R, Golub TR 2018. Genetic and transcriptional evolution alters cancer cell line drug response. Nature. 560 (7718):325-330.

Cao S, Chen H, Xiang Sh, Hong J, Weng J, Zhu H, Liu Q 2015. "Anti-Cancer Effects and Mechanisms of Capsaicin in Chili Peppers", American Journal of Plant Sciences, 6, 3075-3081.

Chularojmontri L, Suwatronnakorn M, Wattanapitayakul S 2010. Influence of capsicum extract and capsaicin on endothelial health. Journal of the Medical Association of Thailand, Suppl 2. S92-101.

Cifuentes RC, Shippen DE 2012. Telomerase regulation. Mutation Research, 730(1-2), 20–7.

Cömertpay S, Gül A, Delibaş M, Tekin Turhan MS 2022. Investigating the Efficacy of Zingerone on Mesothelioma and the Role of TRPV1 in This Effect. Nutr Cancer.;74(6):2174-2183.

Cömertpay S, Özçelik Demirbanka FG 2020. Lowered Cyclin E levels increase the efficiency and the specificity of capsaicin against cancerous cells of mesothelium. Cellular and Molecular Biology (Noisy-le-grand), 30, 66(6), 98-104.

Dillon LM, Miller TW 2014. Therapeutic targeting of cancers with loss of PTEN function, Current Drug Targets, 15(1), 65–79.

Garrity A, Tubelli AA, Bandopadhayay P, Tsherniak A, Vazquez F, Wong B, Birger C, Ghandi M, Thorner AR, Bittker JA, Meyerson M, Getz G, Beroukhim R, Golub TR 2018. Genetic and transcriptional evolution alters cancer cell line drug response, Nature, 560(7718), 325-330.

Guo Y, Wang LP, Li C, Xiong YX, Yan YT, Zhao LQ, Xian CJ 2018. Effects of Ginsenoside Rb1 on Expressions of Phosphorylation Akt/Phosphorylation mTOR/Phosphorylation PTEN in Artificial Abnormal Hippocampal Microenvironment in Rats, Neurochemical Research, 43(10), 1927–1937.

Hanahan D, Weinberg RA 2011. Hallmarks of cancer: the next generation, Cell, 144, 646-674.

Hasle N, Matreyek KA, Fowler DM. 2019. The Impact of Genetic Variants on PTEN Molecular Functions and Cellular Phenotypes. Cold Spring Harb Perspect Med. 1;9(11):a036228.

Helvaci N, Comertpay S 2018. In vitro evaluation of the effects of capsaicin on normal and cancerous cells of human cartilage, Turkish Journal of Biology, 42, 422-434.

Keren H, Lev-Maor G, Ast G. 2010. Alternative splicing and evolution: diversification, exon definition and function, Nature Reviews Genetics, 11, 345–355.

Khosravi-Maharlooei M, Jaberipour M, Hosseini Tashnizi A, Attar A, Amirmoezi F, Habibagahi M. 2015. Expression Pattern of Alternative Splicing Variants of Human Telomerase Reverse Transcriptase (hTERT) in Cancer Cell Lines Was not Associated with the Origin of the Cells. International journal of molecular and cellular medicine, 4(2), 109–119.

Kim NW, Piatyszek MA, Prowse KR 1994. Specific association of human telomerase activity with immortal cells and cancer, Science, 266(5193), 2011–5.

Kuo YH, Chiang EI, Chao CY, Rodriguez RL, Chou PY, Tsai SY, Pai MH, Tang FY 2017. Dual inhibition of key proliferation signaling pathways in triple-negative breast cancer cells by a novel derivative of Taiwanin A, Molecular Cancer Therapy, 16(3), 480–493.

DRAFT

Koziel JE, Fox MJ, Steding CE, Sprouse AA, Herbert BS 2011. Medical genetics and epigenetics of telomerase, Journal of Cellular and Molecular Medicine , 15(3), 57–67.

Maihofner C, Heskamp ML 2013. Prospective, Non-Interventional Study on The Tolerability and Analgesic Effectiveness over 12 Weeks After A Single Application of Capsaicin 8% Cutaneous Patch in 1044 Patients with Peripheral Neuropathic Pain:First Results of The QUEPP Study Current Medical Research Opinion, 29 (6), 673-683.

Montpetit JA, Alhareeri AA, Montpetit M, Starkweather, RA, Elmore WL, Filler K, Mohanraj L, Burton WC, Menzies SV, Lyon ED, Collins BJ, Teefey MJ, Jackson-Cook KC 2014. Telomere Length: A Review of Methods for Measurement, Nursing Research and Practice, 63(4), 289–299.

National Center for Biotechnology Information. 2023a. TERT telomerase reverse transcriptase [Homo sapiens (human)], https://www.ncbi.nlm.nih.gov/gene/7015 Last Access Date: 27.09.2023

National Center for Biotechnology Information. 2023b. PTEN phosphatase and tensin homolog [Homo sapiens (human)], https://www.ncbi.nlm.nih.gov/gene/5728 Last Access Date: 27.09.2023

Sánchez BG, Bort A, Mateos-Gómez PA, Rodríguez-Henche N, Díaz-Laviada I. 2019. Combination of the natural product capsaicin and docetaxel synergistically kills human prostate cancer cells through the metabolic regulator AMP-activated kinase, Cancer Cell International, 8, 19:54.

Schmittgen TD, Livak KJ 2008. Analyzing Real-Time PCR Data by the Comparative CT Method, Nature Protocols, 3(6), 1101-1108.

Shay JW, Wright WE 1996. The reactivation of telomerase activity in cancer progression, Trends in Genetics , 12(4), 129–31.

Toyoda T, Shi L, Takasu S, Cho YM, Kiriyama Y, Nishikawa A, Ogawa K, Tatematsu M, Tsukamoto, T. 2016. Anti-Inflammatory Effects of Capsaicin and Piperine on Helicobacter pylori-Induced Chronic Gastritis in Mongolian Gerbils, Helicobacter, 21 (2), 131-142.

Turan E 2019. Analysis of the Effects of Capsaicin on Telomere Length and Telomerase Enzyme Levels in H2452 Mesothelioma Cell Line. *In Turkish* - Master's Thesis, Institute of Science, Kahramanmaraş Sütçü İmam University, Faculty of Science.

World Health Organization. 2023. “Cancer”, https://www.who.int/news-room/fact-sheets/detail/cancer Last Access Date: 27.09.2023

DRAFT

ORAL PRESENTATION

Pharmacological effect of juice and extract fruits of Algerian *Ecballium elaterium* (L) on Phenylhydrazine induced hyperbilirubinemia in rats

Farah Ramdane [1,2], Sara Djemal [1] , Salima Lellah [1]

[1] Faculty of Nature sciences and Life, El Oued University. PO Box 789. 39000 . Algeria
[2]Biogeochemistry Laboratory in Desert Environments. Kasdi Merbah University.PO Box 511, 30000. Ouargla. Algeria

*Corresponding author e-mail: farahramdane@yahoo.fr

Abstract

This study aims to evaluate for the first-time bilirubin lowering activity of an Algerian *Ecballium elaterium*. The juice was pressed from the mature fruits. The decoction method was used to prepare aqueous powder fruits extract. the current study examined the bilirubin-lowering activity of powder fruit extract (300mg/kg) and fruits juice at 0.2 mL/kg and 0.7 mL/kg for three days in jaundiced Wistar rats administrated phenylhydrazine 0,05 ml/kg (I.P). The biochemical parameters included albumin, bilirubin, alkaline phosphatase, were restored to a normal level and the liver damage was improved in rats treated with Ecballium elaterium. Therefore, both juice and fruits aqueous extract can be used for their hepatoprotective

Key words: *Ecballium elaterium* (L), Rats, Jaundice, Hepatoprotective activity, Biochemical parameters

INTRODUCTION

Jaundice is also known as icterus (Muhammad et al.,2016) is one of the most common medical condition affecting infants, children, and adults (Janghel et al., 2019). It results from a liver disorders or hyperbilirubinaemia consequently lead to a yellow pigmentation in the skin, sclera and mucous membranes (Arthur et al.,2012; Ayeni et al., 2017) the eyes, tongue and urine (Bipin Kumar et al.,2016). Which is developed in relation to several diseases or abnormal condition such as hepatitis, liver cancer, hemolytic anaemia…etc (Abbasi et al.,2009). Medicinal plants and their natural products are being used long ago by our ancestors for the treatment of such hepatic disorders (Tewari et al.,2017; Janghel et al., 2019). Ecballium elaterium (E. Elaterium) or squirting cucumber (Attard and Scicluna-Spiteri 2001;Toker et al.,2003; Uslu et al., 2006; Bourebaba et al.,2018) commonly known as Fegous el hemir is a species of herbaceous plant (Bizid et al.,2015) with sub-succulent, hairy leaves and stems belonging to the Cucurbitaceae family (Greige-Gerges et al., 2007;Christodoulakis et al.,2011; Bizid et al.,2015) mainly distributs in the Mediterranean regions (Uslu et al., 2006; Greige-Gerges et al., 2007). It is reported that squeeze fruit gives a juice with an excellent therapeutic remedy effect known as elaterium. It also contains proteins, lipids, cucurbitacins (B, D, E, I and L,) and cucurbitacin derivatives such as glycosylcucurbitacins and triterpenoids glycosides (Touhiri et al.,2019). Many studies have been carried out to explore the therapeutic effect of this plant, particularly its anti-inflammatory, rhinosinusitis, analgesic, antipyretic and antiphlogistic (El Naggar et al.,2015; Bourebaba et al.,2018) hepathitis, cancer, rheumatism, hemorrhoids, liver disorders, earache, and uroclepsia, antimalarial, antimicrobial, antijaundice and anti-proliferative properties and anti-cancer effects (Toker et al.,2003; Touihri-Barakati et al.,2016: Bourebaba et al.,2018). Based on the traditional use of E. Elaterium fruits in Algerian folk medicine to treat jaundice. The present study is the first has been carried out to investigate its antioxidant and antijaundice activities by using in vitro and in vivo studies.

MATERIALS AND METHODS

Juice and aqueous extract preparation

The green fruits of E. elaterium was collected from Jijel (Northeast Algeria) in February 2020 and identified by Dr. Halice.Y at (CRSTRA. Touggourt. Algeria), then air-dried at room temperature in the shade. Fruits were ground to a fine powder using domestic blunder and used for the preparation of aqueous extract. The juice was pressed from the green fresh fruits and then filtered in till yielded a clear crude fruit juice that was conserved in sterile tubes at 4 °C. 10g of E. elaterium powder fruits was boiled in 100 ml distilled water for 20 minutes according to traditional practitioners (data not shown here). After cooling the extract was filtered and concentrated by rotary evaporator (Büchi Rotavapor R- 200), stored away the light and humidity for further analysis.

DRAFT

Blood biochemical and histopathological evaluation

All animals were sacrificed on day 8 after an overnight fast. At sacrifice, blood was obtained from the heart and liver was taken and recorded after rinsing in normal saline before remaing it in formol 10% for histopatholigical study. The serum total bilirubin, glutamic pyruvate transaminase (SGPT), glutamic oxaloacetate transaminase (SGOT), and alkaline phosphatase (ALP) levels were estimated using commercial kits.

Statistical analysis

Statistical analysis was done in triplicate and represented as the mean ± SEM using MINITAB17. ANOVA process was adopted for the analysis of variance. Statistical comparisons between means were determined by Student test. P values < 0.05 were considered as significant.

RESULTS

Blood biochemical parameters evaluation

The biochemical parameters are illustrated in Figure1. Total bilirubin concentration in group treated with phenylhydrazine (PC) was highly significantly increased when compared with the normal group (NC). However, The level of total bilirubin of the *E. elaterium* treated groups (EJ1,Ej2.EE) were significantly lower than PC. The enzymatic activity of SGPT, SGTO and alkaline phosphatase (ALP) were significantly hihger only in this later in the (PC) when compared with (NC) and has been decreased significantaly in the groups administred the juice or aqueous extract compared with jaundiced group (PC) nevertheless this activity was significantly lower in EGJ2,EE for SGTP and SGTO respectively when compared to group (PC) .

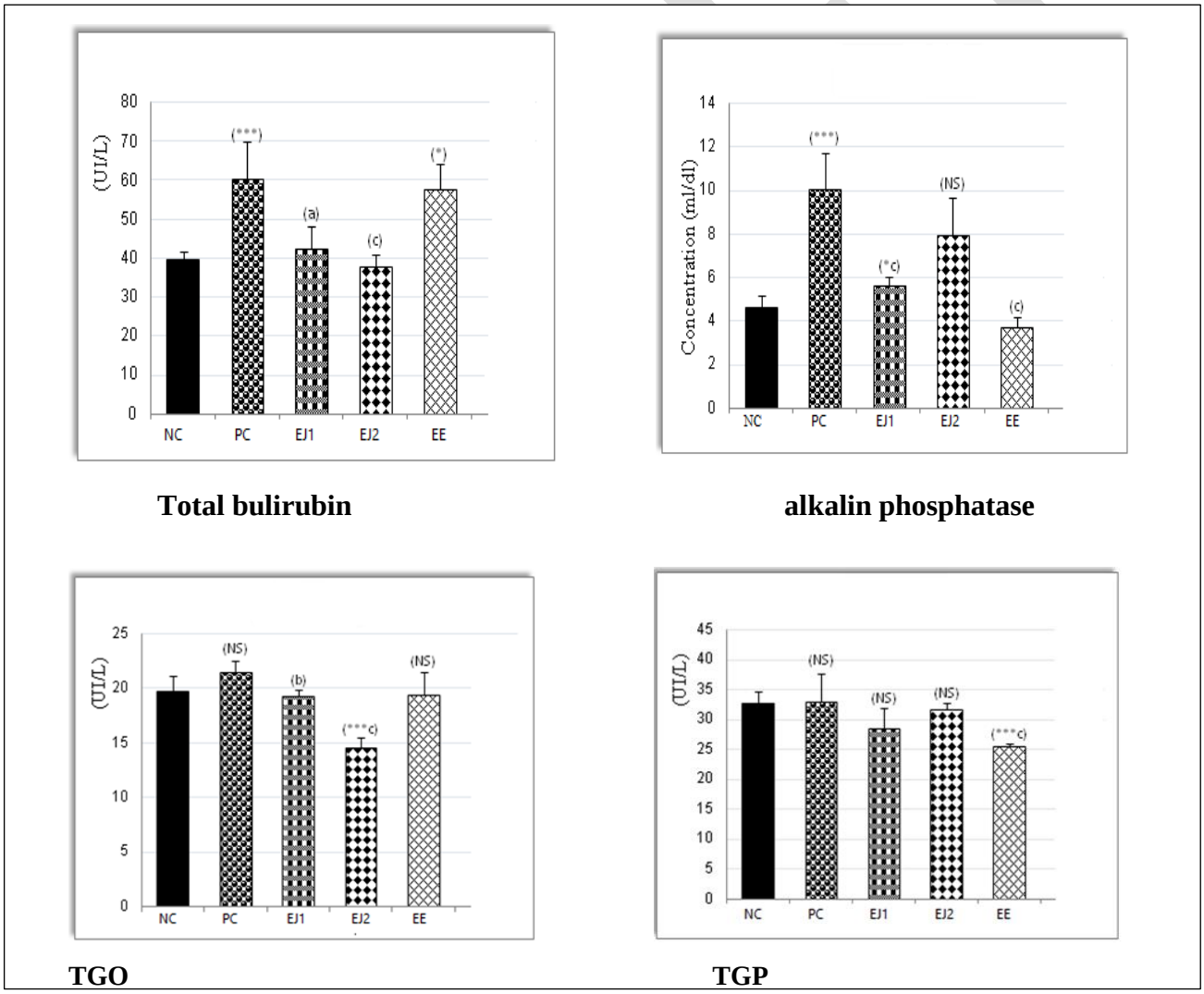


Figure 1: Liver enzymes and biochemical parameters variation in all groups
Values represent the means ±SEM of three measures. Student test.
(*.**.***) P < 0.05 compared with negative control.
(a,b,c) P < 0.05 compared with positive control

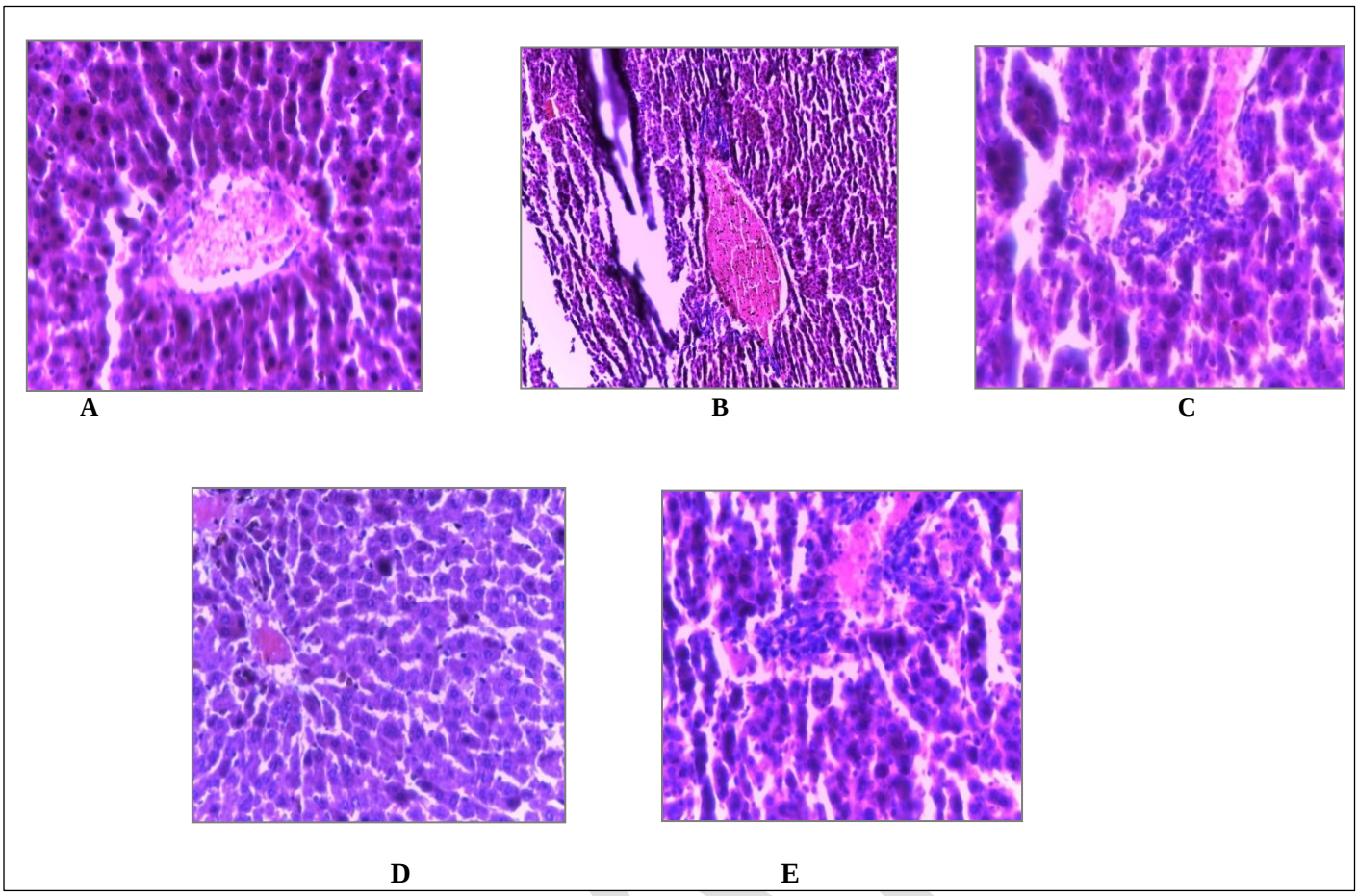


Figure2. Histopathological results for all groups

Section of hepatocyte was stained with haematoxylin and eosin and observed under optical microscope (×400)
A: Negative control: central veins, normal hepatocytes B: Positive control, dilated veins, bile pigment, vascular congestion
C: EJ1, bile pigment, cholangitis.
D: EJ2, bile pigment
E: EE, bile pigment, cholangitis.

DISCUSSION

The present research summarized the antijaundice activity of *E. elaterium* juice and aqueous extract, this plant belong to the cucurbitateae family. This latter contains about 965 species and 120 genera (Omokhua-Uyi and Van Staden, 2020). Phenylhydrazine induces hyperbilirubinaemia by increasing enzymatic transformation of haemoglobinhaem to bilirubin (Arthur et al., 2012; Maity et al., 2013). However, the current study revealed the decrease of the bilirubin and enzymatic activity of ALP, SGPT and SGTO after administration of phenylhydrazine on rats using *E elaterium* juice (EJ1, EJ2) and aqueous extract with diffenrence effectiveness on these biochemical parameters. These latter were the basis of hepatocyte functions (Maity et al., 2013) . Results are in agreement with those of (Greige-Gerges et al.,2007), the juice contains proteins, lipids and several active compounds known as cucurbitacins (Touihri et al.,2019) responsible for this activity mainly cucurbitacin B by modification of the complex albumin-bulirubin binding (El Naggar et al.,2015) .
Histopathological analysis of hepatic sections of all groups was presented in (Fig.2). In the jaundice group some changes were noted such as dilation of central veins, congestion, and the presence of bile pigments compared to the control. *E. Elaterium* juice and extract restored the structure damage of the liver.

CONCLUSION

The effect of *E. elaterium* aqueous extract juice fruits on hyperbilirubinemia has been studies. Results demonstrate that *E. elaterium* juice and aqueous extract are effectively able to decrease the levels of bilirubin in human plasma. Therefore, this is the first study of the Algerian *E. elaterium* may serve to the development of new potent drugs in the treatment of jaundice.

ACKNOWLEDGEMENTS

Athors thank Dr Halice.Y (CRSTRA. Touggourt. Algeria) for plant identification

REFERENCES

Abbasi, A,M., Khan, M,A., Ahmad, M., Zafar, M., Khan, H., Muhammad, N., Sultana, S. 2009. Medicinal plants used for the treatment of jaundice and hepatitis based on socio-economic documentation. African Journal of Biotechnology, 8 (8), 1643-1650. DOI:http://www.academicjournals.org/AJB

Attard, E.G., & Scicluna-Spiteri, A. 2001. Ecballium elaterium: an in vitro source of cucurbitacins. Fitoterapia,72,46-53. DOI: https://doi.org/10.1016/S0367-326X(00)00256-2

Arthur, F,K,N., Woode, E., Terlabi, E.O., Larbie, C. 2012. Bilirubin Lowering Potential of Annona muricata (Linn.) in Temporary Jaundiced Rats. American Journal of Pharmacology and Toxicology, 7 (2): 33- 40. DOI: https://doi.org/10.3844/ajptsp.2012.33.40

Ayeni, M, I., Akanmu, M. A., Bolaji, O, O., Osasan, S, A., Olayiwola, G., Afolabi, M. O., Morohunfola, A., M. 2017. Bilirubin lowering potential of aqueous Carica papaya extract in induced jaundice in rats. Journal of Pharmacy and Pharmacology,7: 457- 466. DOI:10.17265/2328-2150/2017.07.011

Bizid, S., Sabbaha, M., Msakni, I., Ben Slimene, B., Mohamed, G., Bouali, R.,... Abdelli., N.2015. Cholestatic hepatitis due to Ecballium elaterium ingestion. Clinic and Research in Hepatology and Gastroenterology, 39, e61-e63. DOI: https://doi.org/10.1016/j.clinre.2014.11.004

Bipin, K. S., Ramashanker Sushanta, G., Lovely, R., Nandita, N., David, L. K. 2016. Plant based folk treatments from North East India for jaundice. (Anoverview). Journal of Medicinal Plants Studies. 4(5), 234 -247.

Bourebaba, L., Gilbert-López, B., Oukil, N., Bedjou, F. 2020. Phytochemical composition of Ecballium elaterium extracts with antioxidant and anti-inflammatory. Arabian Journal of Chemistry,13, 3286 -3300. DOI: https://doi.org/10.1016/j.arabjc.2018.11.004

Christodoulakis, N, S., Kollia, K., Fasseas, C. 2011. Leaf structure and histochemistry of Ecballium elaterium (L.) A. Rich (squirting cucumber). Flora Morphology Distribution Functional Ecology of Plants , 206 (3), 191-197. DOI:10.1016/j.flora.2010.03.004.

El Naggar, E,B., Chalupová, M., Prazanovà, G., Parák, T., Śvajdlenka, E., Zemlićka, M., Suchý, P.2015. Hepatoprotective and proapoptotic effect of Ecballium elaterium on CCl4-induced hepatotoxicity in rats. Asian Pacific Journal of Tropical Medicine, 8(7), 526 -531. DOI: https://doi.org/10.1016/j.apjtm.2015.06.012

Greige-Gerges, H., Abou Khalil, R., Abou Mansour, E., Magdalou, J., Chahine, R., Ouaini, N. 2007. Cucurbitacins from Ecballium elaterium juice increase the binding of bilirubin and ibuprofen to albumin in human plasma. Chemico- Biological Interaction,169, 53-62.DOI: https://doi.org/10.1016/j.cbi.2007.05.003.

Janghel, V., Patel, P., Chandel, S,S. 2019. Plants used for the treatment of icterus (jaundice) in Central India: A review. Annals of Hepatology.18, 658- 672. DOI: https://doi.org/10.1016/j.aohep.2019.05.003

Maity, S., Nag, N., Chatterjee, S., Adhikari, S., Mazumder, S.2013. Bilirubin clearance and antioxidant activities of ethanol extract of Phyllanthus amarus root in phenylhydrazine-induced neonatal jaundice in mice. Journal of Physiology and Biochemistry. DOI: 10.1007/s13105-013-0234-y.

Muhammad, W,A., Shamshad, T., Muhammad, A,A., Rukhsar, J.2016. Jaundice: a basic review. International Journal of Research in Medicinal Sciences, 4,1313-9. DOI: https://doi.org/10.18203/2320-6012.ijrms20161196.

Omokhua-Uyi, AG., & Van Staden, J. 2020. Phytomedicinal relevance of South African Cucurbitaceae species and their safety assessment: A review. Journal of Ethnopharmacology, 259, 112967. DOI: https://doi.org/10.1016/j.jep.2020.112967.

Tewari, D., Mocan, A., Parvanov, E,D., Sah, A,N., Nabavi, S,M., Huminiecki, L., Ma, Z,F., ……Atanasov, A,G. 2017. Ethnopharmacological Approaches for Therapy of Jaundice: Part II.

Highly used plant species from Acanthaceae, Euphorbiaceae, Asteraceae,Combretaceae, and Fabaceae Families. Frontiers in Pharmacology, 8,519. DOI: doi: 10.3389/fphar.2017.00519.

Tokera, G., Memişoğlua, M., Tokerb, M,C., Yeşiladaa, E.2003. Callus formation and cucurbitacin Baccumulation in Ecballium elaterium callus cultures. Fitoterapia. 74, 618- 623.DOI: https://doi.org/10.1016/S0367-326X(03)00165-5.

Touihri-Barakati,I., Kallech-Ziria, O., Boulila, A., Khwaldia, K., Marrakchib, N., Hanchid, B., …….Luise, J. 2016. Targetting avb3 and a5b1 integrins with *Ecballium elaterium* (L.) A. Rich. seed oil. Biomedecine and Pharmacotherapy. 84,1223-1232. DOI; https://doi.org/10.1016/j.biopha.2016.10.035

Touihri, I., Kallech-Ziri, O., Boulila, A., Fatnassi, S., Marrakchi, N., Luis, J., Hanchi, B. 2019. Ecballium elaterium (L.) A. Rich. seed oil: Chemical composition and antiproliferative effect on human colonic adenocarcinoma and fibrosarcoma cancer cell lines. Arabian Journal of Chemistry,12,2347-2355.DOI: https://doi.org/10.1016/j.arabjc.2015.02.023

Uslu, C., Karasen, R,M., Sahin, F,Taysi, S., Akcay, F. 2006. Effect of aqueous extracts of Ecballium elaterium rich, in the rabbit model of rhinosinusitis. International Journal of Pediatric Otorhinolaryngology,70,515-518. DOI : https://doi.org/10.1016/j.ijporl.2005.07.020.

DRAFT

ORAL PRESENTATION

Recent advances in wound healing using silk fibroin-based microparticles

Chidi Wilson Nwekwo (*ORCID: https://orcid.org/0000-0001-7621-3019*)

Near East University, Faculty of Engineering, Department of Biomedical Engineering, Nicosia, TRNC.

*Corresponding author e-mail: chidiwilson.nwekwo@neu.edu.tr

Abstract

Silk fibroin, a biopolymer obtained from the silkworm cocoon, has been extensively studied for its potential applications in medical devices, drug delivery systems, and tissue engineering scaffolds. Its biodegradable, non-toxic, and biocompatible properties make it a promising candidate for wound healing. Silk fibroin offers significant advantages by allowing drug delivery to the site of injury, controlling the release rate of drugs, and minimizing toxicity. Additionally, silk fibroin microparticles show promise in targeted delivery of anticancer agents and gene therapies. This review explores the potential of silk fibroin-based microparticles loaded with antibiotics to enhance wound healing outcomes.

Keywords: silk fibroin, microparticles, wound healing

INTRODUCTION

Wound healing plays a crucial role in restoring tissue integrity and preventing infections (Niederstätter et al., 2021). A retrospective analysis of Medicare claims from 2014-2019 revealed that wound prevalence increased by 13% from 14.5% to 16.4%, especially among individuals under 65 who are disabled in the United States (Carter et al., 2023). Traditional methods for wound healing often face challenges such as delays in recovery time, scarring issues, and susceptibility to infection (Kim et al., 2019). Therefore, there is a need for novel approaches that can enhance the effectiveness and efficiency of healing treatments.

Extensive research has been conducted on the use of silk fibroin, which is biocompatible and biodegradable to expedite wound healing and promote skin regeneration (Ceccarini et al., 2023; He et al., 2019; J. Liu et al., 2021; Tariq et al., 2021; Vidya & Rajagopal, 2021; W. Zhang et al., 2017)

Studies have demonstrated that silk fibroin films can effectively improve wound healing and facilitate skin regeneration (J. Liu et al., 2021). This is because these films possess properties such as biocompatibility and minimal inflammatory response, making them an excellent choice for various wound healing applications. However, their limited permeability has posed challenges in some scenarios (Stoica et al., 2020). To address this limitation, researchers have devised techniques to incorporate micro/nano-sized pores into silk fibroin films, enhancing their permeability and effectiveness in promoting wound healing (J. Liu et al., 2021).

In addition to films, extensive research has been conducted on using silk fibroin hydrogels for wound healing. Animal studies have discovered that hydrogels from silk fibroin loaded with growth factor 1 (FGF1) promote or facilitate wound healing. Furthermore, these hydrogels create an environment that supports cell adhesion, proliferation, and migration, thus aiding skin tissue regeneration (He et al., 2019; Mazurek et al., 2022).

Moreover, researchers have explored the potential of silk fibroin for 3D printing materials used in wound healing. They have successfully 3D printed grid piezoresistors using inks composed of functionalized silk fibers and peptide patterns. In vitro tests have shown that these materials contribute to skin healing and improved cell adhesion (Ceccarini et al., 2023).

A Brief Overview of Silk Fibroin Microparticles

Silk fibroin microparticles are biocompatible and biodegradable nanoparticles derived from silk protein sources such as silkworm cocoons or spider webs (Bobrova et al., 2022). These particles possess mechanical strength, adjustable degradation rates, and excellent drug-carrying capacity (Lehmann et al., 2022). In recent

years, silk fibroin microparticles have garnered attention for their application in various biomedical fields, including wound healing drug delivery systems (Sabarees et al., 2023).

Silk Fibroin Microparticles and Their Potential in Wound Healing and Drug Delivery

Hemostasis, inflammation, proliferation, and remodeling are the four interconnected stages of wound healing (Tottoli et al., 2020). Hemostasis aims to stop bleeding by forming blood clots at the injury site (Pourshahrestani et al., 2020). Inflammation follows as immune cells infiltrate the area to clear debris and prevent infection (Koh & DiPietro, 2011). During the proliferation phase, cells grow to restore tissue structure and functionality (Landén et al., 2016). Lastly, collagen fibers undergo reorganization during remodeling to strengthen the formed tissue (Frantz et al., 2009).

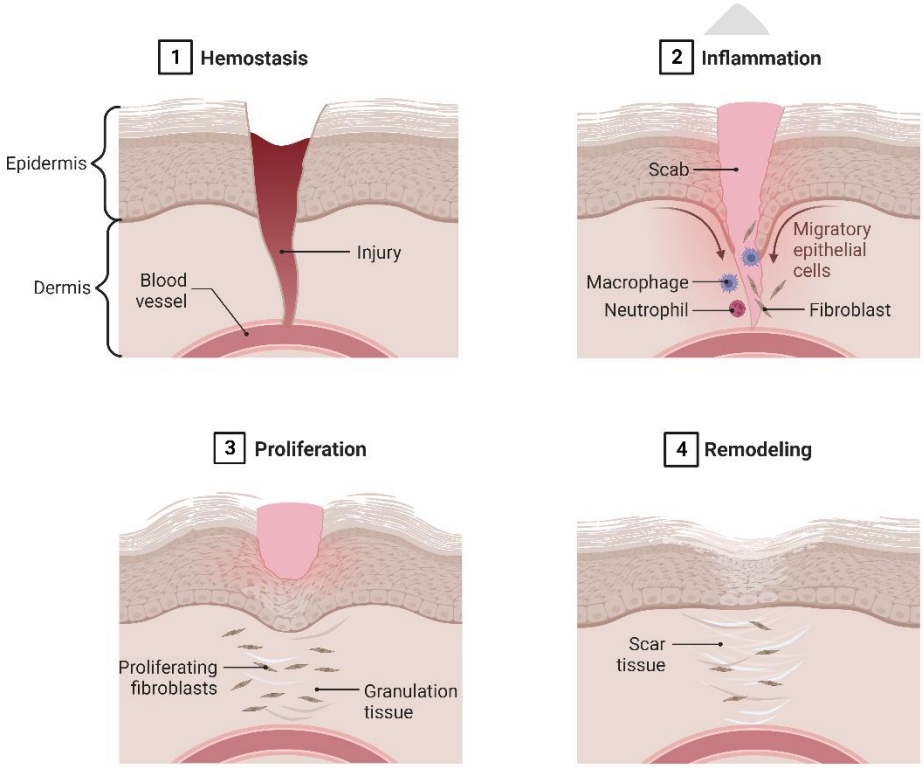


Figure 1. *The 4 stages of the wound healing process*

Due to the intricacies involved in the process, conventional wound healing methods have their limitations. These include delayed healing, risk of infection, and insufficient regeneration due to scar formation (Kim et al., 2019; Xu et al., 2020). The increased risk of infection at the injured site further complicates the healing process (Raziyeva et al., 2021).

Silk fibroin microparticles loaded with antibiotics improve wound healing outcomes by enabling sustained drug delivery at the injury site. This allows control over drug release rates and minimizes systemic toxicity risks (Pacheco et al., 2020). These antibiotic-loaded silk fibroin microparticles can be directly injected into the wound site, where they gradually disintegrate over days or weeks while releasing the medication (Wani et al., 2022). Additionally, these microparticles possess outstanding mechanical strength, allowing their integration into wound dressings or scaffolds without compromising their overall structure (Bobrova et al., 2022; Vidya & Rajagopal, 2021).

These microparticles offer advantages such as targeted delivery of anticancer agents and gene therapies because they can carry a range of molecules, including proteins, peptides, oligonucleotides, and small molecules (Tomeh et al., 2019). Moreover, silk fibroin microparticles are biodegradable and non-toxic, making optimizing their properties easy (Florczak et al., 2020). Another benefit is their capability to accommodate

drugs within a delivery system, which can be valuable for combination therapies (C. Li et al., 2019). When transformed into nanoparticles, silk fibroin microparticles hold the potential for nanomedicine by enabling interaction with specific body parts and delivering therapeutic molecules effectively (Lehmann et al., 2022). This opens up possibilities for delivering drugs to targeted locations within the body. In summary, silk fibroin microparticles offer a solution for drug delivery due to their biocompatibility and versatility in tailoring properties (Wani et al., 2022).

Hence, further exploration into the use of silk fibroin microparticles to deliver drugs could be beneficial in developing safe treatments for various diseases.

Antibiotics in Wound Healing

Antibiotics play a role in preventing infections during the process of wound healing. They can hinder growth and eliminate bacteria, thus reducing the risk of complications (Lehmann et al., 2022). Commonly used antibiotics include penicillin, erythromycin, and tetracycline. Each antibiotic has its mechanism of action, such as inhibiting cell wall synthesis or protein synthesis, which effectively combats infections (Nguyen et al., 2019).

Benefits of Silk Fibroin Microparticles Loaded with Antibiotics

Silk fibroin microparticles loaded with antibiotics offer advantages over other antibiotic drug delivery systems. Silk fibroin offers an advantage in delivering antibiotics to wounds by providing sustained release, ensuring a long-lasting therapeutic effect (Zheng et al., 2022). This sustained release feature allows for dosing and better patient compliance. Moreover, silk fibroin is highly biocompatible with the body (W. Zhang et al., 2017).

Techniques for Fabricating Silk Fibroin Microparticles Loaded with Antibiotics

Scientists have developed techniques to fabricate silk fibroin microparticles loaded with antibiotics. The methods include solvent evaporation/dispersion and emulsion-based approaches (Bobrova et al., 2022). In the evaporation/dispersion method, silk fibroin and antibiotics are dissolved in an organic solvent and precipitated through rapid desolvation or coacervation techniques (Tudora et al., 2013). On the other hand, emulsion-based approaches involve dispersing a solution containing silk fibroin and antibiotics within an organic phase before solidifying it (Amirifar et al., 2022).

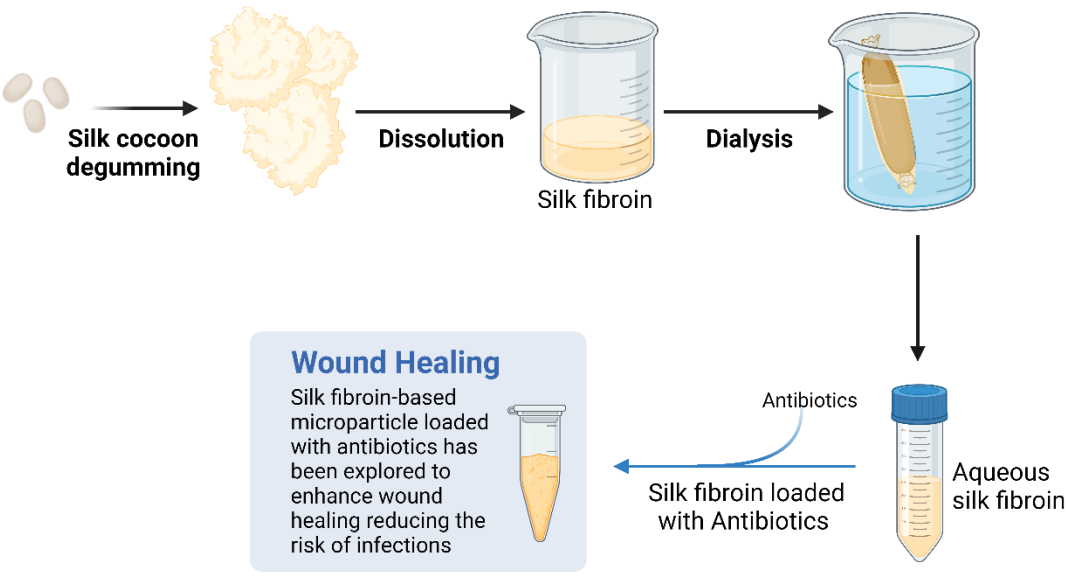


Figure 2. *Silk fibroin-based microparticles loaded with antibiotics: fabrication and application*

In Vitro Studies on Silk Fibroin Microparticles Loaded with Antibiotics

Laboratory studies have examined the effectiveness and compatibility of silk fibroin microparticles loaded with antibiotics in controlled environments. These studies often use laboratory models with cells to evaluate factors like the ability to kill bacteria, the health of the cells, and the response to inflammation. The findings from these studies show that silk fibroin microparticles are effective in delivering antibiotics over periods while maintaining their ability to fight against bacteria (S. Li et al., 2022; C. Liu et al., 2019).

In Vivo Studies on Silk Fibroin Microparticles Loaded with Antibiotics

Researchers have also conducted experiments on animals to assess how well silk fibroin microparticles loaded with antibiotics can work for wound healing. These experiments involved applying the microparticles onto wounds in animal models and evaluating aspects such as how fast wounds close, analyzing tissue samples under a microscope, and observing any inflammatory response. Encouraging results demonstrated wound healing, reduced infection rates, and improved tissue regeneration when using silk fibroin microparticles loaded with antibiotics (Sang et al., 2023; P. Zhang et al., 2023).

CONCLUSION

Using silk fibroin microparticles loaded with antibiotics shows promise in improving wound healing outcomes in biomedical engineering. These microparticles can enhance treatment effects while reducing the need for doses and addressing compliance concerns due to their unique properties like sustained release of drugs, compatibility with living tissues, and mechanical strength. Future research should focus on refining manufacturing techniques, conducting trials, and exploring opportunities for clinical use to fully realize the potential of using silk fibroin microparticle based antibiotic delivery systems in wound healing applications.

ACKNOWLEDGEMENTS

Figures 1 and 2 were created with BioRender.com. Figure 1 is adapted from “Wound Healing” and Figure 2 from “Silk scaffold: Fabrication and Application” by BioRender.com (2023). Retrieved from https://app.biorender.com/biorender-templates.

REFERENCES

Amirifar, L., Besanjideh, M., Nasiri, R., Shamloo, A., Nasrollahi, F., De Barros, N. R., Davoodi, E., Erdem, A., Mahmoodi, M., Hosseini, V., Montazerian, H., Jahangiry, J., Darabi, M. A., Haghniaz, R., Dokmeci, M. R., Annabi, N., Ahadian, S., & Khademhosseini, A. (2022). Droplet-based microfluidics in biomedical applications. In *Biofabrication* (Vol. 14, Issue 2). https://doi.org/10.1088/1758-5090/ac39a9

Bobrova, M., Safonova, L., Efimov, A., Lyundup, A., Mozheiko, N., Agapova, O., & Agapov, I. (2022). Scaffolds Based on Silk Fibroin with Decellularized Rat Liver Microparticles: Investigation of the Structure, Biological Properties and Regenerative Potential for Skin Wound Healing. *Pharmaceutics*. https://doi.org/10.3390/pharmaceutics14112313

Carter, M. J., DaVanzo, J., Haught, R., Nusgart, M., Cartwright, D., & Fife, C. E. (2023). Chronic wound prevalence and the associated cost of treatment in Medicare beneficiaries: changes between 2014 and 2019. *Journal of Medical Economics*, *26*(1). https://doi.org/10.1080/13696998.2023.2232256

Ceccarini, M. R., Palazzi, V., Salvati, R., Chiesa, I., De Maria, C., Bonafoni, S., Mezzanotte, P., Codini, M., Pacini, L., Errante, F., Rovero, P., Morabito, A., Beccari, T., Roselli, L., & Valentini, L. (2023). Biomaterial Inks from Peptide-Functionalized Silk Fibers for 3D Printing of Futuristic Wound-Healing and Sensing Materials. *International Journal of Molecular Sciences*, *24*(2). https://doi.org/10.3390/ijms24020947

Florczak, A., Grzechowiak, I., Deptuch, T., Kucharczyk, K., Kaminska, A., & Dams-Kozlowska, H. (2020). Silk particles as carriers of therapeutic molecules for cancer treatment. In *Materials* (Vol. 13, Issue 21). https://doi.org/10.3390/ma13214946

DRAFT

Frantz, S., Bauersachs, J., & Ertl, G. (2009). Post-infarct remodelling: Contribution of wound healing and inflammation. In *Cardiovascular Research* (Vol. 81, Issue 3). https://doi.org/10.1093/cvr/cvn292

He, S., Shi, D., Han, Z., Dong, Z., Xie, Y., Zhang, F., Zeng, W., & Yi, Q. (2019). Heparinized silk fibroin hydrogels loading FGF1 promote the wound healing in rats with full-thickness skin excision. *BioMedical Engineering Online*, *18*(1). https://doi.org/10.1186/s12938-019-0716-4

Kim, H. S., Sun, X., Lee, J. H., Kim, H. W., Fu, X., & Leong, K. W. (2019a). Advanced drug delivery systems and artificial skin grafts for skin wound healing. In *Advanced Drug Delivery Reviews*. https://doi.org/10.1016/j.addr.2018.12.014

Kim, H. S., Sun, X., Lee, J. H., Kim, H. W., Fu, X., & Leong, K. W. (2019b). Advanced drug delivery systems and artificial skin grafts for skin wound healing. In *Advanced Drug Delivery Reviews* (Vol. 146). https://doi.org/10.1016/j.addr.2018.12.014

Koh, T. J., & DiPietro, L. A. (2011). Inflammation and wound healing: the role of the macrophage. In *Expert reviews in molecular medicine* (Vol. 13). https://doi.org/10.1017/S1462399411001943

Landén, N. X., Li, D., & Ståhle, M. (2016). Transition from inflammation to proliferation: a critical step during wound healing. In *Cellular and Molecular Life Sciences* (Vol. 73, Issue 20). https://doi.org/10.1007/s00018-016-2268-0

Lehmann, T., Vaughn, A. E., Seal, S., Liechty, K. W., & Zgheib, C. (2022). Silk Fibroin-Based Therapeutics for Impaired Wound Healing. In *Pharmaceutics*. https://doi.org/10.3390/pharmaceutics14030651

Li, C., Wang, J., Wang, Y., Gao, H., Wei, G., Huang, Y., Yu, H., Gan, Y., Wang, Y., Mei, L., Chen, H., Hu, H., Zhang, Z., & Jin, Y. (2019). Recent progress in drug delivery. In *Acta Pharmaceutica Sinica B* (Vol. 9, Issue 6). https://doi.org/10.1016/j.apsb.2019.08.003

Li, S., Shi, X., Xu, B., Wang, J., Li, P., Wang, X., Lou, J., Li, Z., Yang, C., Li, S., & Zhen, P. (2022). In vitro drug release and antibacterial activity evaluation of silk fibroin coated vancomycin hydrochloride loaded poly (lactic-co-glycolic acid) (PLGA) sustained release microspheres. *Journal of Biomaterials Applications*, *36*(9). https://doi.org/10.1177/08853282211064098

Liu, C., Lin, L., Huang, Z., Wu, Q., Jiang, J., Lv, L., Yu, X., Quan, G., Li, G., & Wu, C. (2019). Novel Inhalable Ciprofloxacin Dry Powders for Bronchiectasis Therapy: Mannitol–Silk Fibroin Binary Microparticles with High-Payload and Improved Aerosolized Properties. *AAPS PharmSciTech*, *20*(2). https://doi.org/10.1208/s12249-019-1291-5

Liu, J., Huang, R., Li, G., Kaplan, D. L., Zheng, Z., & Wang, X. (2021). Generation of Nano-pores in Silk Fibroin Films Using Silk Nanoparticles for Full-Thickness Wound Healing. *Biomacromolecules*, *22*(2). https://doi.org/10.1021/acs.biomac.0c01411

Mazurek, Ł., Szudzik, M., Rybka, M., & Konop, M. (2022). Silk Fibroin Biomaterials and Their Beneficial Role in Skin Wound Healing. In *Biomolecules*. https://doi.org/10.3390/biom12121852

Nguyen, T. P., Nguyen, Q. V., Nguyen, V. H., Le, T. H., Huynh, V. Q. N., Vo, D. V. N., Trinh, Q. T., Kim, S. Y., & Van Le, Q. (2019). Silk fibroin-based biomaterials for biomedical applications: A review. In *Polymers* (Vol. 11, Issue 12). https://doi.org/10.3390/polym11121933

Niederstätter, I. M., Schiefer, J. L., & Fuchs, P. C. (2021). Surgical Strategies to Promote Cutaneous Healing. In *Medical sciences (Basel, Switzerland)* (Vol. 9, Issue 2). https://doi.org/10.3390/medsci9020045

Pacheco, M. S., Kano, G. E., Paulo, L. de A., Lopes, P. S., & de Moraes, M. A. (2020). Silk fibroin/chitosan/alginate multilayer membranes as a system for controlled drug release in wound healing. *International Journal of Biological Macromolecules*. https://doi.org/10.1016/j.ijbiomac.2020.02.140

Pourshahrestani, S., Zeimaran, E., Kadri, N. A., Mutlu, N., & Boccaccini, A. R. (2020). Polymeric Hydrogel Systems as Emerging Biomaterial Platforms to Enable Hemostasis and Wound Healing. In *Advanced Healthcare Materials* (Vol. 9, Issue 20). https://doi.org/10.1002/adhm.202000905

Raziyeva, K., Kim, Y., Zharkinbekov, Z., Kassymbek, K., Jimi, S., & Saparov, A. (2021). Immunology of acute and chronic wound healing. In *Biomolecules* (Vol. 11, Issue 5). https://doi.org/10.3390/biom11050700

Sabarees, G., Tamilarasi, G. P., Velmurugan, V., Alagarsamy, V., Sibuh, B. Z., Sikarwar, M., Taneja, P., Kumar, A., & Gupta, P. K. (2023). Emerging trends in silk fibroin based nanofibers for impaired wound healing. In *Journal of Drug Delivery Science and Technology* (Vol. 79). https://doi.org/10.1016/j.jddst.2022.103994

Sang, S., Wang, S., Wu, J., & Zhang, X. (2023). Sprayable Berberine-Silk Fibroin Microspheres with Extracellular Matrix Anchoring Function Accelerate Infected Wound Healing through Antibacterial and Anti-inflammatory Effects. *ACS Biomaterials Science and Engineering*, *9*(6). https://doi.org/10.1021/acsbiomaterials.3c00030

Stoica, A. E., Chircov, C., & Grumezescu, A. M. (2020). Nanomaterials for wound dressings: An Up-to-Date overview. In *Molecules* (Vol. 25, Issue 11). https://doi.org/10.3390/molecules25112699

Tariq, M., Tahir, H. M., Butt, S. A., Ali, S., Ahmad, A. B., Raza, C., Summer, M., Hassan, A., & Nadeem, J. (2021). Silk derived formulations for accelerated wound healing in diabetic mice. *PeerJ*, *9*. https://doi.org/10.7717/peerj.10232

Tomeh, M. A., Hadianamrei, R., & Zhao, X. (2019). Silk fibroin as a functional biomaterial for drug and gene delivery. In *Pharmaceutics* (Vol. 11, Issue 10). https://doi.org/10.3390/pharmaceutics11100494

Tottoli, E. M., Dorati, R., Genta, I., Chiesa, E., Pisani, S., & Conti, B. (2020). Skin wound healing process and new emerging technologies for skin wound care and regeneration. In *Pharmaceutics* (Vol. 12, Issue 8). https://doi.org/10.3390/pharmaceutics12080735

Tudora, M. R., Zaharia, C., Stancu, I. C., Vasile, E., Truşcă, R., & Cincu, C. (2013). Natural silk fibroin micro- and nanoparticles with potential uses in drug delivery systems. *UPB Scientific Bulletin, Series B: Chemistry and Materials Science*, *75*(1).

Vidya, M., & Rajagopal, S. (2021). Silk Fibroin: A Promising Tool for Wound Healing and Skin Regeneration. In *International Journal of Polymer Science* (Vol. 2021). https://doi.org/10.1155/2021/9069924

Wani, S. U. D., Zargar, M. I., Masoodi, M. H., Alshehri, S., Alam, P., Ghoneim, M. M., Alshlowi, A., Shivakumar, H. G., Ali, M., & Shakeel, F. (2022). Silk Fibroin as an Efficient Biomaterial for Drug Delivery, Gene Therapy, and Wound Healing. In *International Journal of Molecular Sciences* (Vol. 23, Issue 22). https://doi.org/10.3390/ijms232214421

Xu, Z., Han, S., Gu, Z., & Wu, J. (2020). Advances and Impact of Antioxidant Hydrogel in Chronic Wound Healing. In *Advanced Healthcare Materials* (Vol. 9, Issue 5). https://doi.org/10.1002/adhm.201901502

Zhang, P., Sun, Y., Yang, H., Liu, D., Zhang, F., Zhang, Y., Zhong, W., Zuo, B., & Zhou, Z. (2023). Vancomycin-loaded silk fibroin microspheres in an injectable hydrogel for chronic osteomyelitis therapy. *Frontiers in Bioengineering and Biotechnology*, *11*. https://doi.org/10.3389/fbioe.2023.1163933

Zhang, W., Chen, L., Chen, J., Wang, L., Gui, X., Ran, J., Xu, G., Zhao, H., Zeng, M., Ji, J., Qian, L., Zhou, J., Ouyang, H., & Zou, X. (2017). Silk Fibroin Biomaterial Shows Safe and Effective Wound Healing in Animal Models and a Randomized Controlled Clinical Trial. *Advanced Healthcare Materials*. https://doi.org/10.1002/adhm.201700121

Zheng, H., Huang, Z., Chen, T., Sun, Y., Chen, S., Bu, G., & Guan, H. (2022). Gallium ions incorporated silk fibroin hydrogel with antibacterial efficacy for promoting healing of Pseudomonas aeruginosa-infected wound. *Frontiers in Chemistry*. https://doi.org/10.3389/fchem.2022.1017548

DRAFT

ORAL PRESENTATION

The innovation of the student's portfolio in the competency-based curriculum, an instrument measuring the level of students' knowledge, as well as their self-esteem in the subject of biology

Gëzim Bara[1], Brilanda Berdica [2], Enkelejda Bara[3]

[1]University of Tirana,Faculty of Natural Sciences,Department of Biology,Tirana,Albania
[2]University of Shkodra 'Luigj Gurakuqi', Faculty of Natural Sciences,Shkoder,Albania
[3]Curriculum specialist at the University of Tirana,Tirana ,Albania

*Corresponding author e-mail: gezim_bara@live.com

Abstract

Teaching and learning involve the teacher, the learner and the curriculum. All three of these elements work in unison inseparable from each other. This is due to the fact that if one of these elements is missing, then the chain is disconnected and no longer functions as a process. Assessment as an important component from which the level of students' knowledge is derived is of several forms. In addition to the continuous evaluation, the evaluation by means of tests has been implemented today and the evaluation called "Student's Portfolio". The student's portfolio serves as a bridge between the process of teaching, learning and the reflection of the teacher and the students. In order to carry out an accurate study of this problem, we managed to do a case study in the "Partizani" gymnasium in Tirana and in the "Ibrahim Rrugova" gymnasium in Kamëz. The purpose of the study is to bring out the importance of the "Student's Portfolio" in the evidence of its level, at the same time how it is valued and recognized by the teacher as one of the important alternatives in the evaluation. In the realization of this study, we relied on research studies where we browsed the relevant literature as well as on the qualitative method where we studied biology teachers and students of class X-XI at the "Partizani" gymnasium in Tirana and at the gymnasium " Ibrahim Rrugova" in Kamez. From the study as well as the analysis of the case study, it results that the teachers had adequate knowledge about this assessment alternative, argued its importance, knew what the student's portfolio contains, but what emerges from this study is that there is negligence in the importance of this evaluation by means of the portfolio. The student's portfolio, as one of the student assessment techniques, occupies an important place in the teaching and learning process, because through it the quality of curriculum implementation is achieved, teaching and learning are documented, and student achievements can be monitored.

Keywords: Portfolio, competence, instrument, criteria, teaching, learning.

INTRODUCTION

In the study that is used to evaluate the students, there is also the "Student's Portfolio". A portfolio is a student-organized body of work that represents the student's learning outcomes.

The correctly designed and well-organized portfolio proves the students' abilities to participate in the entire learning process and expresses the level of students' activity in independent work.

From the interviews conducted in addition to the evaluation given to this evaluation, such opinions appear that the portfolio is considered a waste of time, because according to them the information is readily available on the Internet.

Chang states, "The portfolio in its entirety is seen as a stimulus for students' multiple intelligences, self-reflection, critical thinking, responsibility for learning, as well as a stimulus for the creation of skills and knowledge in a certain field (2003). The content of the portfolio should to meet all the evaluation criteria and standards. The benefits of the "Student's Portfolio" are for the student, the teacher, the parents.

For the student, the "Portfolio" is a panorama of his work, where from the evaluation and analysis of the students' works, the achievements, the knowledge they have acquired, and the reflection on the future are highlighted.

For the teacher, the "Portfolio" in its entirety shows the level of knowledge of the students, where this knowledge serves as an indicator to discover the abilities, tendencies, weaknesses, deficiencies as well as the needs of the students.

For parents, the "Portfolio" is a documented record of their children's level, observing how their child is progressing, relying on the record that the teacher makes with his notes.

DRAFT

According to Diane Hart, "Portfolio" is like a "container" that holds evidence of students' knowledge, ideas, interests and achievements.
According to Nunan, in "Portofol students tell about their progress and progress"
"The use of the portfolio as an assessment tool is related to teaching, as long as portfolios are seen as one of the most widely used alternatives in assessment". (Broën.H.D.2004).
The portfolio as an instrument supports the achievement of student results, encourages teachers and supports the reflection of parents, increases the range of elements that are evaluated and manages to evaluate their abilities. The quality of the portfolio is very important, where an important role in its quality the teacher plays, but also other factors such as: How much it refers to the curriculum, the type of methodology used for its design, the criteria that are set for evaluation, transparency in evaluation and communication of results. In order to monitor the quality of the "Portfolio", we must rely on several criteria, such as:
The content which must be precisely defined, to which field it belongs, which are the materials to be presented, and do these materials represent the progress of the students.
The collection of materials has been made accurate in order to reflect the evaluation criteria and materials.
Reflection and self-evaluation. All materials refer to the function, content, as well as the degree of reflection by students, teachers, parents. Today, in the framework of curriculum reform, (Student's Portfolio) occupies an important place in the evaluation of students' knowledge, where this is supported by instruction No. 34 of the Ministry of Education and Sports dated 11.09.2015, which states that: "Student portfolio" is a tool for evaluating students, in basic education, in the curriculum based on competences in our country. Finally, "Portfolio of students" is a document that provides necessary information to students, teachers, and parents.

MATERIALS AND METHODS

The "Student's portfolio" which accounts for 20% of the student's evaluation in the entire volume that is realized is a very important tool because it proves the progress of the students and their abilities, giving the teacher and parents information about the level of the students theirs. Recently, portfolio assessment is taking an important place in the teaching process. In daily practice, three portfolios are used to carry out a quality assessment of students.
The portfolio of work which contains completed or unfinished work but which is systematized daily by the students. With the help of this portfolio, the students' performance is monitored every day.
The document portfolio contains the finished works that are used to prepare the products. Its purpose is to evaluate products.
The exhibition portfolio contains the best quality works of the student, with the help of which the best quality students are identified.
The importance of this study is to obtain information on how this assessment is known and applied by teachers, what is the performance of students who demonstrate this assessment. With the help of the conducted research as well as the case study with the interviews of teachers and students, we managed to get a detailed information as well as identify some problems in terms of evaluation with the "Portfolio". The study was conducted within ethical rules and norms, both in the way interviews were conducted, and in data collection and analysis. During the conduct of the study, the teachers were willing to give their explanations and arguments for this form of assessment, and from the students' interview conducted with this questionnaire, we were able to identify some problems and finally give recommendations for this problem.
Population and sample: The two gymnasiums "Partizani" in Tirana and the "Ibrahim Rrugova" gymnasium in Kamëz were studied as the population. Samples were taken in the study through interviews, biology teachers of two high schools as well as students of classes X - XI, selecting the best classes.
Instrument and data collection: Pre-prepared interviews with teachers and students served as an instrument for this study. An oral interview was conducted with the teachers and a written interview with the students. At the same time, the control and evidence of this assessment by the teachers during the school year served as an instrument, as well as the study of some of the best quality portfolios of the students. The collection and conclusion of the data was carried out after the analysis of the responses of the interviews witnessed by the teachers and students. Based on the data analysis, discussions and recommendations have been made.

RESULTS AND DISCUSSION

The results and discussions of this paper are derived from the analysis of the case study, where two gymnasiums, more specifically, "Partizani" Tirana and "Ibrahim Rrugova" Kamëz gymnasiums, where with a predetermined theme, the biology teachers were interviewed as and students of grades 10 and 11, selecting the best classes. The topic presented in this interview was about some essential issues for the "Student's Portfolio", where from the answers of teachers and students, we were able to create a panorama clear for this study, where in the end we concluded with concrete conclusions and recommendations. During the research I studied some

of the best quality students' portfolios, where from their analysis I identified the level of the portfolios, the types of works, are the works in accordance with the realization competences. The results are concentrated from the evidence of the answers of the interviews of biology teachers and students of these high schools of grades 10 and 11, the best quality classes were selected. In general, from the answers of teachers and students, some problems are evident, in addition to the achievements that are visible. We are presenting a summary of the questions and answers of teachers and students.

Interview with high school biology teachers:

-What place does the "Student's Portfolio" take in the evaluation of his skills? Which elements of the "Portfolio" are valued the most?

In general, teachers give this answer; In general, they show interest in this type of evaluation, where they emphasize that "Portfolio" is an effective form of evaluation, but they also show opinions that there are cases that they don't evaluate it so much, since the volume of work it contains creates a little subjectivity.

For the second part of the question, they say that they value more the works related to projects, experiments and multimedia presentations.

-In giving the task where are you based? Why? What are the rubrics that the portfolio is built on? How much time do you spend checking a portfolio?

As you can see, the question had to do with the orientation that should be given to the students, the construction of the portfolio, as well as the importance of time devoted to its correction.

The summary answer of the teachers was: In determining the tasks of the teacher, as an orientation we took the curriculum, the key competencies, as well as the teaching topics, in order for the student to be more clear and dedicated in the work that will be presented. The main headings to focus on in the design of the portfolio are: Projects, written works, experiments, videos, etc. While the time spent on evaluating the portfolio depends on its volume, as well as the quality of the tasks. But it varies from 30 min to an hour.

-Should key competencies be included in the construction of the portfolio? During the evaluation of the student, which of the qualities do you see as primary, its content or creativity?

In the answer to the first part of the question, all teachers agree that the inclusion of key competencies in the portfolio curriculum is a curricular obligation, since the motto of the new curriculum is learning based on competencies. But they also accept the fact that there are times when a such request. As for the second part of the question, their answer is different, where 60% of them prefer creative demonstrations, while 40% prefer educational content.

-During the evaluation, how do you manage to evaluate each element of the portfolio? What about correspondence and scoring? During the evaluation, are you objective, focusing on the level of the portfolio, or do you consider its performance during the year?

Regarding the first part of the question, they all give the same answer, where the placement of points is based on two criteria. First, the amount of total points that the portfolio should have, and secondly, setting the amount of points for each element of the portfolio is conditioned by the weight of each element in the weight of the portfolio. As for the grading, this is determined by a table where from the total points of the portfolio it is possible to determine the points for each grade based on a correctly calculated table. As for the third part of the question, their answer with 2 exceptions was that they thought that the performance of the students should be taken into consideration as it could be a corruption or underestimation of the moment. The others say that the portfolio is an individual work and should not be taken considering other assessments or student performance.

So evaluation only according to the real condition of the work.

- In the works listed, which of the following works will you give priority in the evaluation of the portfolio:

Essays, projects, presentations, experiments, certificates, tests, videos, written assignments, creative work, multimedia presentations.

In their answers, they give different opinions, in the selection of works, specifically:

40% prefer more: Projects, collections, written assignments.

30% prefer more: Presentations, essays, experiments.

30% prefer more: Created videos, multimedia videos, tests, creative work.

Student survey

- What is (Portfolio) for you? Are you interested in this learning innovation? For each 3-month period, how many assignments should you submit?

In the first part of the question, 100% of the students in their answer show that they had the necessary knowledge about this problem and admit that the portfolio is a job where we demonstrate our knowledge and skills. For the second part of the question, 80% say that they accept it as an evaluation technique, where through

it they manage to express the learning information in the way they like. While for the 3rd part of the question, they say that for every 3 months they have to submit 4 papers determined by the subject teacher.
-Are you given clear instructions by the teacher about the portfolio? Is the level of the task given easy, difficult, or beyond your ability?
To the first part of the question, 80% of the students answer that the teachers give clear and sufficient instructions to complete the portfolio. While 20% say that they are not very clear in the instructions given by the teacher. For the second part of to the question we get this answer: 80% of the students state that their level is within their abilities, 20% state that there are also cases that the tasks are difficult. beyond their abilities, there are also cases that the tasks are difficult, beyond their abilities, but this does not stop them from engaging, on the contrary, according to them, it helps them to increase their skills.
-In the realization of the "Portfolio" work, where do you get the source of information? Give your opinion on this assessment?
For the first part of the question, they give this answer: 50% from the Internet, 40% from the teacher, 10% from other sources. As for the second part of the question, they answer: 70% evaluate it as a valid and important assessment, while 30% not very valid.
- In the jobs you want to plan for "Portfolio", who guides you?
We get this answer: 60% agree that they are guided by the teacher, 20% together with the teacher, while 20% agree that they determine it themselves.
-Are you satisfied with the teacher's assessment of the portfolio? How informed are you by this assessment?
For the first part of the question, they say: 70% are satisfied and aware of the evaluation, while 30% express dissatisfaction. As for the information, they say: 80% admit that they are regularly informed about the evaluation, while 20% express dissatisfaction.
Give your opinion on the usefulness of the wallet? What can you specify from this assessment?
Regarding the answer to this question, they have different approaches. 80% of them say that the portfolio is important in assessing their skills, while 20% say that it is a waste of time, because it is simply a printout of some materials from the Internet. While in terms of the specification of this assessment, in general they use the "Portfolio", an important form of assessment, which affects their creative and research work, makes them more active in digital work, as well as affects the increase in the quality of the final grade.

CONCLUSIONS

From an analysis and expertise of this study, we reached some conclusions, relying on the issues arising from this study where the analysis of the problem was at the center, relying on the qualitative methods where it was focused on the survey of teachers and students.
From the interviews of teachers and students, we come to the following conclusions:
In many cases, the "Portfolio" turned into a routine process, turning into tedious and unproductive work.
There were sporadic cases where the purpose and content were not made clear from the start
A part of the students saw it as a waste of time, seeing it as a form of assessment only to increase the average, and not as their creative work.
They also showed objections in the way of evaluation as well as in giving results.
In the student's "Portfolio" the key competences achieved must be documented.
The "portfolio" should be an incentive for students' awareness of their level and achievements.
The teacher uses portfolio data to promote learning, record and demonstrate their progress, and document student performance.
The design of the portfolio should be guided by the function it has, take into account the age and abilities of the students, as well as document the learning process.

RECOMMENDATIONS

Teachers should give due importance to the "Student's Portfolio", especially biology teachers, where in its design they should cooperate with students, parents, to be clear about the purpose, content and organization of the work for its design.
In the work that should be done for the portfolio, the students should be oriented to the biological teaching topics, such as cells, reproduction, organisms and the environment, DNA,
To focus more on creative, proactive and experimental work, where they concretize the topic with concrete means.
The students should have the necessary spaces in the preparation of the materials, both for the volume of the tasks and for the degree of difficulty.

During the evaluation of the "Portfolio", students' abilities in the way of using information resources should be taken into account.
To encourage students' self-esteem
During the evaluation, each element of the portfolio should be evaluated, giving importance to the evaluation according to the weight of the element in the portfolio. At the same time, the assessment is made known to students and parents.
Training of teachers on how to design and evaluate the "Portfolio"
The portfolio should contain important elements of the curriculum, where the learner compares, reflects and analyzes the strengths and weaknesses of his level

REFERENCES

Angela, Simmons, Jill Lumnsden, Florida State University: Preparing a portfolio, 2016,.
Dona, M. Zucker, Massachusetts University; Hoë to do a case study research, 2009,.
European pre-accession institute: Teaching and learning, 2013,.
Marsh, J. Colin: Basic concepts for understanding the curriculum, New York, 2009, p.85-88
Mita, Nikoleta: Assessment in education, Tirana, 2009, p.57-62.
Musaj, Bardhyl: Teaching methodology, 1994
Mustafa, Kadi; Methods for evaluation, 2011,.
Vesna, Hostanovic."Action research in educational educational practice, 2015,.
WW.izha.edu.al/.../Approved instruction-34-evaluation for curriculum-with-competencies.

DRAFT

ORAL PRESENTATION

Physico-chemical quality of some honeys produced in the region of Beni Mellal-Khenifra in Morocco

Mourad Chikhaoui[1*] (*ORCID: https://orcid.org/0009-0001-1782-8205*), Chaimaa Elagdi[1] (*ORCID: https://orcid.org/0009-0007-4161-7567*) , Wadi Badri[1] (*ORCID:* https://orcid.org/0000-0003-1529-9474), Jamal Mouslim[1] (*ORCID:https://orcid.org/0009-0004-7149-0067*), Houda El Hajjouji[1](*ORCID:*https://orcid.org/*0000-0002-2347-3703*)

[1] Hassan II University, Faculty of Sciences Ben M'sik, Laboratory of Ecology and Environment, Casablanca, Morocco.

*Corresponding author: Mourad Chikhaoui ; E-mail : chimoura7@gmail.com

Abstract

Due to its essential floristic, faunistic, and landscape diversity, Morocco has a significant and unique beekeeping potential, making it one of the most fascinating biological and biogeographical plans. Moroccan culture reserve an important place to the benefits of honey, the product is widely consumed by the population, and its quality control is important before it is put on the market to protect consumers. In order to study their physico-chemical quality, four types of honey samples were collected from Beni Mellal-Khenifra region in Morocco: agrume honey, euphorbe honey, anis honey and carob honey. Nine physico-chemical parameters were analysed: Moisture content, electrical conductivity, ash content, pH, free acidity, hydroxymethyl furfural, diastasis activity and sugar content. The results showed differences from one honey sample to another. However, the mean values of the studied physio-chemical properties of the honey samples were within the ranges of international standards.

Keywords : Honey, Beni Mellal-Khenifra region, Physico-chemical characteristics.

INTRODUCTION

Due to its essential floristic, faunistic, and landscape diversity, Morocco has a significant and unique beekeeping potential, making it one of the most fascinating biological and biogeographical plans (Moujanni et al., 2017). Thus, Moroccan beekeeping is supported actively by the Government through the Green Morocco Plan (GMP), with the objective of reaching 16,000 tons of production annually (Belahsen et al., 2016). Moreover, Moroccan culture reserve an important place to the benefits of honey, and the consumption of the product by the population (0,520 à 0,750) kg/ person/year) is relatively high compared to the countries neighbors including Algeria with an average of 0.200 kg/person/year and Tunisia with a range of (0.170 at 0.200) kg/person/year (Haderbache and Mohammedi, 2015 ; Hussein, 2000 ; Moujanni et al., 2017). According to the High Commission for Planning (HCP), Beni Melal-khenifra region ranks third nationally in terms of honey production place with an annual production of 274 tons (HCP, 2017). Nevertheless, 50% of this production is marketed through traditional informal channels without quality control (Moujanni, 2017). In this context, the present study aims to determine the physico-chemical characteristics of four honey samples that were collected from the Beni Mellal-Khenifra region in Morocco, to assess their quality and verify their compliance with international standards.

MATERIAL AND METHODS

Samples collection

Four pure crude honey samples were obtained directly from beekeepers from different localities of Beni Mellal-Khenifra region during March to October 2021 (Tbles 1). All honey samples were filtred with sterile gauze to remove debris, and stored in sterile glass containers at 4 ° C in the dark until analysis (Bogdanov and Blumer 2001).

DRAFT

Table 1 : Honey samples according to their botanical origin, localities and harvest date

Honey samples	Botanical origin	Localities	Harvest date
Citrus honey	*Citrus*	Tadla	24/3/2021
Spurge honey	*Euphorbia*	Tagzirt	14/07/2021
Anise honey	*Pimpinella anisum*	Souk sebt	06/09/2021
Carob honey	*Ceratonia siliqua*	Tagzirt	05/10/2021

Physico-chemical analysis

In order to study the quality of honey samples, nine physicochemical parameters were analyzed (Water content, electrical conductivity, ash content, pH, free acidity, hydroxymethylfurfural (HMF) content, diastasis activity and sugar content) because they were most frequently used as indicators of quality and stability of honey and having a great influence on its organoleptic properties. The parameters: Water content, electrical conductivity, ash content, pH and free acidity were carried out in the Ecology and Environment laboratory (Faculty of Sciences Ben M'sik, Hassan II University, Casablanca). However, HMF content, diastasis activity and sugar content were realised by the National Center for Scientific and Technical Research (NCSTR) in Rabat.

Physicochemical endpoints were analyzed using official analytical methods from the Association of Official Analytical Chemists (AOAC, 1990) and harmonised methods from the European Honey Commission (Bogdanov et al., 2002). To ensure uniform conditions and comparability, the samples were examined in triplicate and over a period of time.

Water content (g/kg of honey)

The water content is determined by measuring the index of refraction at 20°C using an Atago type refractometer.

Electrical conductivity

To determine the electric conductivity of all honey samples, the resistance of 20 g of honey was measured in 100 ml of distilled water at 20°C. we use a portable conductivity meter (HI99300).

pH and free acidity

The pH is measured using a Digital pH meter (Sension+ MM 374 GLP). calibrated by standard solutions on a 10% honey solution.

Free acidity was determined by titration of 10 g of honey dissolved in 75 ml of distilled water, using phenolphthalein as indicator and 0.1 N NaOH as titrator. The results are presented as milliequivalents (meq) of acid per kg of honey as 10 times the volume of NaOH used in titration.

Ash content

Ash content is based on incineration of honey in an oven. 5 g of honey are added with a few drops of olive oil and the whole was put in a crucible and heated to 350- 400°C for 1 hour. These measurements were expressed as a percentage (%).

Diastase activity

Spectrophotometry was utilized to determine the diastatic activity by incubating a buffered solution of soluble starch and honey in a thermostatic bath at 40°C. The results were expressed in Gothe degrees as the amount (ml) of 1% starch hydrolysed by the enzyme in 1 g of honey in 1 h (de Almeida-Muradian *et al*. 2013).

HMF content

To determine the HMF content, HPLC-UV was utilized, with absorption maximum at 285 nm. The GraceSmart RP-C18 column (250 × 4.6 mm, 5 µm) was used; water/methanol mobile phase (90 : 10, v/v) with flow rate of 1 ml/min and total analysis time of 7 minutes. Injection volume was 10 µl, and column temperature 30°C.

DRAFT

Sugar content

The chromatographic separation is carried out using a Shodex Asahipak NH2P-504E column (C18, 250 nm x 4.6 nm) and acetonitrile:water (78:22, v/v) as mobile phase (1 ml/min). The injection volume was 10 µl, the temperature is set at 35°C and the analysis time was 30 min.

RESULTS AND DISCUSSION

The physico-chemical characteristics parameters of the honey samples studied are summarised in table 2.

Table 2 : Physico-chemical parameters of the honey samples studied

Physico-chemical parameters	Honey samples				*Codex Alimentarius*
	Citrus honey	Spurge honey	Anise honey	Carob honey	
Moisture content	17.4 ± 0.125	18.92 ± 0.196	19.55 ± 0.113	20.8 ± 1.041	17,5<H<21,0 g/100g
conductivity (µs/cm)	121,2 ± 0.55	74,4 ± 1.15	93 ± 1.15	117,5 ± 1.5	< 800
% Ash	0,50 ± 0.036	0,34 ± 0.015	0,38 ± 0.036	0,38 ± 0.032	<0,6 g/100g
pH	3,49 ± 0.055	3,78 ± 0.215	3,58 ± 0.055	3,66 ± 0.005	3,5 < pH <4,5
Free Acidity (meq/kg)	12,0 ± 0.5	18.5 ± 1.1	19,7 ± 0.5	19,4 ± 0.3	<50 meq/kg
HMF (mg/kg)	20,96 ± 0.11	25,23 ± 0.755	19,95 ± 0.216	13,39 ± 0.15	<60 mg/kg
DA	12,2 ± 0.2	8 ± 0.1	10,0 ± 0.5	12,0 ± 0.2	≥ 8
Glucose %	38,4 ± 1.2	38,3 ± 1.5	39,2 ± 0.5	36,2 ± 1.2	Glucose 26 à 36 %
Fructose %	41,57 ± 0.1	40,1 ± 0.6	32,7 ± 0.2	29,1 ± 0.2	Fructose 32 à 42 %
Saccharose %	3,3 ± 0.2	5,0 ± 0.1	2.8 ± 0.2	2,2 ± 0.4	Saccharose 0 à 5 %

DA : Diastase activity
HMF : Hydroxyméthylfurfural

The water content is a parameter linked to the degree of maturity. It is responsible for the stability of honey during storage (Bagdanov et al., 1999). Honey with high humidity is more likely to ferment, making storage more difficult (Rostislav et al., 2016). In our study, the values obtained are between 17.4% and 20.2%. These values were less than 21%, the maximum recommended by the European standards (Thrasyvoulou et al., 2018). These results were indicative of a good storage of the studied honeys.

The electrical conductivity expresses the ability of the aqueous solution to conduct an electric current. it is positively correlated with the concentration of mineral salts, organic acids and proteins (Bettar et al., 2019). This parameter varies greatly depending on the floral origin, it is regarded as one of the parameters used to differentiate between floral honeys and honeydew honeys. In this study, the electrical conductivity showed values between 74,4 and 121,2 µs/cm, and they were within the bounds of the European legislation (under 800 µs/cm) (Thrasyvoulou et al., 2018).

The ash content is a parameter of quality that depends on the botanical origin of honey. The mineral matter in the analyzed honey samples ranged between 0,34% and 0,50%. These values were in line with the European legislation (less than 0.6%) (Thrasyvoulou et al., 2018).

Honey pH is impacted by conditions processing and storage. moreover, an extreme pH reveals a biochemical degradation due to poor conditions harvesting or conservation. The pH values of our studied honey samples tend towards acidity, they were between 3.49 and 3.78.These values were in accordance with the recommendations of the *Codex Alimentarius (*3,5 < pH <4,5) (Thrasyvoulou et al., 2018.

The free acidity of honey deprnds on the presence of organic acids, or internal esters, and certain inorganic ions, such as as phosphate (Gomes et al., 2010). High acidity can be indicative of fermentation of sugars into organic acids (Rostislav et al., 2016). The values of free acidity of the investigated samples ranged between 12.0 and 19.7 meq/kg, and they were within the limits of the European legislation (lesse than 50 meq/kg) (Thrasyvoulou et al., 2018), indicating no undesirable fermentation.

The HMF content is largely recognized as a freshness parameter for honey samples, it is absent from the fresh honey immediately stored by bees and tends to increase by aging of honey (Gomes et al. 2010). In this study, spectrometric analysis of honey samples revealed HMF contents which were between 13,39 and 20,96 mg/kg.

These values comply with the standards set by the *Codex Alimentarius*, which is less than 60 mg/kg (Thrasyvoulou et al., 2018).

Diastase is an enzyme in honey, it is an index of freshness and overheating the honey. Spectrophotometry showed that the DA values of honey samples studied vary from 8 to 12.2. These values were within the normal range set by the *Codex Alimentarius*, which is ≥ 8 (Thrasyvoulou et al., 2018).

Sugars were the main components of honey types. Fructose and glucose were considered as a major constituents of honey (Küçük et al., 2007). The glucose and fructoses values of the samples studied vary successively from 36,2 to 39.2% and from 29,1 to 41.57%. These values meet the standards required by the *Codex Alimentarius*, which is 26 to 36% for glucose and 32 to 42% for fructose (Thrasyvoulou et al., 2018).

Briefly, The results of this study showed that honey samples analyzed were good quality and according to international standards.. Neverless, there were differences from one honey sample to another in studied physiochemical properties. These differences can be explained by different environmental conditions such as the climate, the botanical origin of the honey samples, (Nandaa et al., 2003; Bogdanov et al., 2004), the conditions storage (Ozcan et al., 2006), and the harvest season (Pe'rez-Arquillue et *al.*, 1995).

CONCLUSION

It appears from this study that the honey samples analyzed exhibited variable physiochemical properties. They comply with international standards. Future studies will be necessary to analyze other honey types produced at the regional or national level. In addition, It will be crucial that the quality control of the product be systematic before it is put on the market in order to protect consumers.

ACKNOWLEDGEMENTS

We are thankful to the National Center for Scientific and Technical Research (NCSTR) for the contribution to the realization of this study.

REFERENCES

AOAC 1990. Official methods of analysis. 15th Ed. Virginia, Association of Official Analytical Chimists, 770-771.

Belahsen S, Khellaf A, Belahsen M 2016. Evaluation de la stratégie agricole du Maroc (Plan Maroc Vert) : Une analyse en équilibre général dynamique. Dossiers de Recherches en Economie et Gestion, 413(3756), 1-22.

Bettar I, González-Miret ML, Hernanz D, Marconi A, Heredia FJ, Terrab A 2019. Characterisation of Moroccan Spurge (Euphorbia) honeys by their physicochemical characteristics, mineral contents and colour. Arabian Journal of Chemistry, 12(8) : 2052-2060.

Bogdanov S, Blumer P 2001. Propriétés antibiotiques naturelles du miel. Revue suisse d'agriculture, (5), 219-222.

Bogdanov S, Martin P, Lullmann C 2002. Harmonised methods of the international honey commission. Swiss Bee Research Centre, FAM, Liebefeld, 5 : 1-62.

Bogdanov S, Ruoff K, Persano L 2004. Physico-chemical methods for characterization of unifloralhoneys: a review. Apidologie, 35: 4-17.

Bogdanov S, Lüllmann C, Martin P., Von Der Ohe W, Russmann H, Vorwohl G, Vit P 1999. Honey quality and international regulatory standards : review by the International Honey Commission. Bee world, 80(2) : 61-69.

Codex Alimentarius 2001. Revised Codex Standard for Honey. Codex Stan. 12-1981, Rev. 1 (1987). World Health Organization, Rome. Rev, 2.

Gomes S, Dias L, Moreira LG, Rodrigues P, Estevinho LM 2010. Physicochemical, microbiological and antimicrobial properties of commercial honeys from Portugal. Food and Chemical Toxicology, 48: 544–548.

Haderbache, L, Mohammedi, A 2015. Etude sur le comportement de consommation du miel en Algérie : attentes et préférences.

Haut-Commissariat au Plan 2017. Monographie régional Béni Mellal-Khénifra.

Küçük M, Kolayh. KaraogIu S, Ulusoy E, Baltaci C, Candan F 2007. Biological activities and chemical composition of three honeys of different types from Anatolia. Food Chem. 100: 526-530.

Hussein MH 2000. A review of beekeeping in Arab countries. Bee World, *81*(2) : 56-71.

Moujanni A, Essamadi AK, Terrab A 2017. L'apiculture au Maroc : focus sur la production de miel/Beekeeping in Morocco : focus on honey production. International Journal of Innovation and Applied Studies, 20(1) : 52.

Nandaa V, Sarkara BC, Sharmaa HK, Bawa ASJ 2003. Determination of Some major and minor elements in the east of Morocco honeys through inductively coupled plasma optical emission spectrometry. Food Comp. Anal, 16:613-619.

Ozcan MD, Arslam DA 2006. Phenolic profiles and antioxidant capacities of Chinese unifloral honeys from different botanical and geographical sources. Food Chem, 99:24-27

Pérez-Arquillue C, Conchello P, Ariňo A, Juan T, Herrera A 1995. Physico-chemical attributes and pollen spectrum of some unifloral Spanish honeys. Food Chem, 54:167–172.

Rostislav H, Petr T, Ćavar ZS 2016. Characterisation of phenolics and other quality parameters of different types of honey. Czech Journal of Food Sciences, 34(3) : 244-253.

Thrasyvoulou A, Tananaki C, Goras G, Karazafiris E, Dimou M, Liolios V, Gounari S 2018. Legislation of honey criteria and standards. Journal of Apicultural Research, 57(1) : 88-96.

DRAFT

ORAL PRESENTATION

Yem börülcesi (*Vigna unguiculata* (L.) Walp.) genotiplerinin bazı morfolojik ve tarımsal özelliklerinin karşılaştırılması

Serkan Bayazit[1] (*ORCID: https://orcid.org/0000-0002-5105-9489*), Ertan Ateş[2*] (*ORCID: https://orcid.org/0000-0002-3048-497X*)

[1]Tekirdağ Namık Kemal Üniversitesi, Fen Bilimleri Enstitüsü, Tekirdağ, Türkiye
[*2]Tekirdağ Namık Kemal Üniversitesi, Ziraat Fakültesi, Tarla Bitkileri Bölümü, Tekirdağ, Türkiye

*Sorumlu yazar e-mail: ertan_ates@hotmail.com

Özet

Bu araştırma Kırklareli ili, Babaeski ilçesi, Taşköprü köyü üretici arazisinde, USDA' dan temin edilen ve yöreden toplanan yem börülcesi genotiplerinin bazı morfolojik ve tarımsal özelliklerinin belirlenmesi amacıyla sulu koşullarda yürütülmüştür. Araştırma ilk yılında (2021) temin edilen az miktardaki tohumun çoğaltılması amacıyla 120 cm sıra arası ve 66 cm sıra üzeri mesafede 4,2 m uzunluğundaki sıralara elle ocak usulü ekim yapılarak, ikinci yılında (2022) ise tesadüf blokları deneme desenine göre 3 tekrarlamalı olarak, sıra arası 25 cm ve 5 m uzunlukta 8 sıradan oluşan parsellere her parselde 400 bitki olacak şekilde ekim yapılmıştır. Araştırmada yem börülcesi genotiplerinin bitki boyu (cm), dal sayısı (adet), ana sap kalınlığı (mm), yeşil ot verimi (kg/da), kuru ot verimi (kg/da) ile bazı kalite özelikleri (ham protein oranı, NDF, ADF, Ca, Mg, P, K oranları) belirlenmiştir. En yüksek bitki boyu İ.H.K. genotipinde (100.30 cm) ölçülmüştür. Dal sayısı en fazla 6.3 adet ile İ.H.B. genotipinde bulunurken, ana sap kalınlığı en fazla 40 genotipinde (8.87 mm) ölçülmüştür. En yüksek yeşil ot verimi 40 genotipinden (4845.00 kg/da), en yüksek kuru ot verimi ise 1 genotipinden (1038.00kg/da) elde edilmiştir. Araştırma sonuçları incelendiğinde, 40 ve 1 genotipleri genetik ıslah materyali olarak değerlendirilerek silaj ve kaba yem üretimine fayda sağlayacağı sonucu ortaya çıkmaktadır.

Anahtar Kelimeler: Yem Börülcesi, Genotip, Ot Verimi, Kalite Özellikleri

Comparison of some morphological and agronomic traits of cowpea (*Vigna unguiculata* (L.) Walp.) genotypes

Abstract

This research was conducted in the village of Taşköprü, Babaeski district, Kırklareli province, Turkey, with the objective of assessing the morphological and agronomic traits of forage cowpea genotypes obtained from the USDA and collected from the local region. The study involved two consecutive years: the first year (2021) focused on seed multiplication, while the second year (2022) involved a randomized complete block design with three replications. The planting method in the first year included manual sowing in rows measuring 4.2 m in length, with a spacing of 120 cm between rows and 66 cm between plants. In the second year, the experiment was conducted under irrigated conditions, with each plot consisting of 8 rows, 5 m in length, and a spacing of 25 cm between rows. A total of 400 plants were sown in each plot. The investigated traits of the forage cowpea genotypes included plant height (cm), number of branches (pcs), main stem diameter (mm), fresh forage yield (kg/ha), dry forage yield (kg/ha), some quality parameters (crude protein ratio, NDF, ADF, Ca, Mg, P, K). Among the genotypes studied, the highest plant height of 100.30 cm was observed in the İ.H.K. genotype. The İ.H.B. genotype exhibited the highest number of branches with 6.3 branches, while the genotype 40 had the greatest main stem diameter of 8.87 mm. The highest green forage yield of 48450.0 kg/ha was obtained from genotype 40, while the highest dry forage yield of 10380.0 kg/ha was recorded in genotype 1. Based on the research findings, genotypes 40 and 1 can be considered promising genetic breeding materials for the production of silage and forage due to their superior performance in terms of yield and other agronomic traits. These genotypes have the potential to contribute positively to forage production systems in the region.

Keywords: Forage Cowpea, Genotype, Forage Yield, Quality Parameters

DRAFT

GİRİŞ

Dünyada tarım ve hayvancılık faaliyetleri ülkelerin gelişmeleri ve kalkınmaları açısından önemli bir yere sahiptir. Ülkelerin ekonomik bağımsızlığının güçlendirilmesi, yoksulluğun azaltılması, gelir ve refah düzeyinin artırılması için tarım ve hayvancılık sektöründe sürdürülebilirliğin sağlanması gerekmektedir. Bu sektörün en temel işlevi insanların besin ihtiyacını karşılamaktır. İnsan beslenmesinde kullanılan hayvansal ürünlerin üretilebilmesi için yem bitkileri yetiştiriciliği ve çayır meraların etkin kullanımı önem arz etmektedir. Ancak ülkemiz meraları üzerinde uzun yıllardır devam eden ağır ve kontrolsüz otlatma sonucu hayvanlar tarafından tercih edilen yem bitkileri ya tamamen kaybolmuş ya da hayvanların otlayamadıkları yerlerde az miktarda kalmış durumdadır. Meralar üzerinde yıllardan beri devam eden aşırı ve erken otlatma, bu alanların verim kapasitesinin büyük oranda azalmasına neden olmuştur. Yurdumuz hayvancılığının ana sorunlarından birisi de yem bitkileri üretimidir. Hayvanların ihtiyacı olan kaba yemlerin çok büyük bir bölümünün çayır-mera ve anız otlatılmasıyla veya besleme değeri çok düşük tahıl sap ve samanından karşılanması, hayvansal ürünlerdeki verim düşüklüğünün başlıca nedenleri arasında yer almaktadır (Açıkgöz, 2001). Ülkemizde yetiştirilen hayvanlar yeterli ve kaliteli kaba yemlerle beslenemedikleri için et ve süt verim ortalamaları düşük olduğu gibi hayvansal ürünlerin kalitesi de düşük düzeydedir (Yücel ve ark., 2004). Bu sebeple ülkemizde var olan kaliteli kaba yem eksikliğini gidermek için yem bitkileri ekim alanları artırılmalıdır.

Yem bitkileri üretimi tarımsal faaliyetleri hayvansal üretim açısından çok önemli bir yere sahiptir. Tarım arazilerinde üretilen otlar öncelikle hayvanlar tarafından kullanılmakta, et, süt vb. ürünlere dönüştürülerek bu ürünlerden de insanlar faydalanmaktadır (Soya ve ark., 2004). Hayvancılık işletmelerinin kaba yemlerini kendilerinin üretmeleri işletmenin karlılığı açısından önemlidir. Tarım ve Orman Bakanlığı tarafından verilen desteklerle yem bitkileri ekim alanlarının arttırılması hedeflenmektedir. Hayvancılık işletmeleri ihtiyaçları olan kaba yemi karşılayabilmek için yonca, korunga, yem bezelyesi, hayvan pancarı, burçak, fiğ türleri ve silajlık mısır dışında farklı yem bitkileri arayışı içerisindedirler.

Börülce tek yıllık bir sıcak iklim baklagil bitkisidir. Genel karakteri gereği sıcaktan hoşlanır. Sıcaklığın 20-30 °C olduğu bölgelerde börülce bitkisinin çok rahat yetiştirildiği, yaz mevsiminde 1,5-2 ay gibi bir sürede gelişmesini tamamlayarak yeşil ot oluşturduğu, toprak yüzeyinde su buharlaşmasını azaltarak, köklerini çok derine gönderip susuzluğa karşı dirençli olduğu bilinmektedir (Günay, 1992). Küresel iklim değişikliği nedeniyle sıcaklığın arttığı ve su kaynaklarının azaldığı günümüz ve gelecekte börülce yem bitkileri arasında daha çok talep edilen bir bitki olacaktır. Çünkü börülce baklagiller içerisinde sıcağa ve kurağa dayanıklılığı en yüksek olan bitkilerden biridir. İklim istekleri mısıra benzerdir. Fakat kurak koşullara dayanıklılığı ve sıcaklık isteği mısıra göre daha fazladır (Şehirali, 1988).

Hayvancılık sektöründe hayvanların verimliliğinin yıl boyunca sağlanabilmesi için silajlık yemler kullanılmaktadır. Mısır silajı, üretim maliyetinin düşük olması, depolamanın kolay olması, içerdiği yüksek enerji, yüksek verim kapasitesi ve hayvanlar tarafından sevilerek tüketilmesi nedeniyle en çok tercih edilen silo yemi olmuştur. Mısır, fermantasyon ve yem değeri açısından hamur olum ya da süt olum döneminde hasat edilerek silaj yapılır. Kolay fermente olabildikleri için, yalın olarak silolanma yetenekleri az olan, protein yönünden zengin baklagil yem bitkileri ile birlikte silolanarak katkı maddesi olarak kullanılabilmektedir. Mısır silajının ham protein oranı %7.65, börülce silajında ham protein oranı %17.36, mısır+börülce karışımı silajının ham protein oranı ise %9.57' dir (Alaca ve Özaslan Parlak, 2017). Baklagil yem bitkileriyle mısır karışımından elde edilen silajın ham protein oranı artarak silaj kalitesini olumlu yönde etkilemektedir. Börülce yüksek protein içeriğiyle mısır ile karışık yetiştirilebilecek önemli bir baklagil yem bitkisi olarak dikkat çekmektedir.

İstatistikler incelendiğinde, 1962 yılında ülke nüfusumuz 28.23 milyon iken bu nüfusa karşılık gelen büyük baş hayvan birimi (BBHB) 13.47 milyon adettir (Düzgüneş ve ark., 1965). Türkiye İstatistik Kurumu 2022 yılı verilerine göre ise büyükbaş hayvan varlığı 17 024 000 adet, küçükbaş hayvan varlığı 56 266 000 adet olsa da geçen 60 yıla göre ülkemizin nüfus artışına paralel olarak hayvan sayımız artmadığı gibi hayvancılık yapan çiftçi sayısı azalmış ve azalmaya devam etmektedir. Ayrıca, günümüzde tarım ile uğraşan çiftçilerin büyük çoğunluğu hayvancılık yapmadığı gibi hayvancılık yapan çiftçi ve işletmelerinde kaba ot ihtiyacını yeteri kadar üretecek arazilerinin olmaması, hayvancılık ile buna bağlı olan hayvansal üretimi sınırlandıran ve azaltan faktörlerin başında gelmektedir. Tarımsal desteklemeler planlanırken, tarım ve hayvancılığı bir arada yürüten ve toprakların organik madde miktarını artıracak şekilde ekim nöbeti uygulayan çiftçilerimize kat sayılı bir destek sistemi getirilerek, hayvancılık sorunu ile birlikte toprak koruma ve ıslahı, yetiştiriciliğinden kaçınılan bitkilerin üretimi konularıyla, bu faaliyetlerle bağlantılı birçok sorun da çözüme kavuşturulabilir. Bu şekilde, geniş arazi varlığına rağmen sadece bitkisel üretim yapan ve üretimden kalan hasat artıklarını (saman vb.) kendi hayvanlarına yedirmeden satan çiftçilerin hayvan bakmaları da teşvik edilmiş olacaktır (Baş ve Ateş, 2022). Ülkemizde var olan kaliteli kaba yem açığını kapatmak için ekolojik koşulları uygun olan bölgelerdeki

tarım alanlarında özellikle 2. ürün olarak yem bitkisi üretimi yapılmalıdır. Trakya'da 1. ürün olarak yetiştirilen serin iklim tahıllarından ve yeşil/kuru ot üretimi amacıyla yetiştirilen fiğ+buğdaygil, yem bezelyesi+buğdaygil karışımlarının hasadından sonra sulama imkânı olan tarım arazilerinde yem börülcesi yeşil/kuru ot üretimi için yalın olarak veya yüksek protein içeren kaliteli silaj elde etmek için mısır ile karışık olarak yetiştirilebilir.

Bu araştırma, farklı yem börülcesi genotiplerinin bazı morfolojik ve tarımsal özellikleri ile kimi kalite parametrelerinin belirlenmesi amacıyla yürütülmüştür.

MATERYAL VE METOT

Araştırma, Kırklareli ili Babaeski ilçesi Taşköprü köyü üretici arazisinde 2021-2022 yılları arasında iki yıl süreyle tesadüf blokları deneme desenine göre 3 tekrarlamalı olarak sulu koşullarda yürütülmüştür. Materyal olarak USDA'dan temin edilen ve yöreden toplanan börülce genotipleri kullanılmıştır. Her parsel 25 cm sıra arası açıklıkta, 5 m uzunluğunda, 8 sıradan oluşmuştur. Parsellerde 400 bitki olacak şekilde elle ekim yapılmıştır. Toprak hazırlığı yapılırken tabana 5 kg/da saf azot ve 5 kg/da saf fosfor olacak şekilde 20-20-0 kompoze gübre ile gübreleme yapılmıştır. Yabancı ot mücadelesi kültürel yöntemlerle gerçekleştirilmiştir. İlk yıl tohum çoğaltmak amacıyla 12.05.2021 tarihinde, ikinci yıl ise 15.05.2022 tarihinde ekim yapılmıştır. Çalışmada toprak nemi ve bitkinin su ihtiyacına göre yağmurlama sulama yöntemiyle sulama uygulanmıştır.

Yeşil ot veriminin (kg/da) belirlenebilmesi için hasatlar ilk baklanın olgunlaştığı dönemde (Ayan ve ark., 2012), Gazioğlu ve İ.H.K. genotiplerinde 24.07.2022, İ.H.B. genotipinde 29.07.2022, 30 numaralı genotipte 07.08.2022, 5 numaralı genotipte 09.08.2022, 11 numaralı genotipte 23.08.2022, 40 numaralı genotipte 12.09.2022, 1 numaralı genotipte 18.09.2022 tarihlerinde parsellerin kenar tesirleri dışında kalan 2 m²'lik kısmı 7,5 cm yukarıdan orak yardımı ile elle biçilerek yapılmıştır. Aynı dönemde her parselden rastgele seçilen 10 bitkide bitki boyu (cm), ana sap kalınlığı (mm) ve bitkide dal sayısı (adet/bitki, ana sap dışındaki dallar sayılmıştır) saptanmıştır. Ana sap kalınlığı dallanmanın başladığı noktada ölçülmüştür. Parsellerden biçilen yeşil otlar gölgede kurutulduktan sonra tartılıp dekara kuru ot verimi (kg/da) hesaplanmıştır.

Kuru örnekler 0.5 mm elek açıklığında öğütülmüş ve kimyasal analizler iki paralel olarak (ham protein, asit deterjanda çözünmeyen lif (ADF), nötr deterjanda çözünmeyen lif (NDF), kalsiyum (Ca), potasyum (K), magnezyum (Mg) ve fosfor (P) oranları) yapılmıştır. Ham protein (%) oranı (%) AOAC (2019)'a göre, NDF ve ADF oranları (%) ise Van Soest ve ark. (1991)'nın belirttikleri yöntem kullanılarak tespit edilmiştir. Ca, K, Mg oranları (%) fleymfotometrede, P oranı (%) ise vanadamolibdofosforik sarı renk yöntemine göre spektrofotometrede okunmuştur (AOAC, 2019). Elde edilen veriler TARİST (Açıkgöz ve ark., 1993) istatistik programında değerlendirilmiştir.

BULGULAR ve TARTIŞMA

En uzun bitki boyu 100.30 cm ile İ.H.K. yem börülcesi genotipinde ölçülmüştür ($P<0.05$, Tablo 1). Bitkideki dal sayısı en fazla İ.H.B. (6.3 adet) ve 1 (6.1 adet) genotiplerinde belirlenirken, ana sap kalınlığı 8.87 mm ile yem börülcesi genotipi 40'ta ölçülmüştür (Tablo 1). En yüksek yeşil (4382.0-4845.0 kg/da) ve kuru ot (966.0-1038.0 kg/da) verimleri 40 ve 1 nolu genotiplerde saptanmıştır ($P<0.01$, Tablo 1).

Tablo 1. Farklı yem börülcesi genotiplerinin bazı morfolojik özellikleri ile ot verimleri

Genotipler	Bitki Boyu (cm)	Dal Sayısı (adet/bitki)	Ana Sap Kalınlığı (mm)	Yeşil Ot Verimi (kg/da)	Kuru Ot Verimi (kg/da)
Gazioğlu	32.30g	3.4c	5.17f	1845.0e	416.0e
İ.H.K.	100.30a	1.9d	5.56e	2002.0e	433.0e
İ.H.B.	68.80d	6.3a	7.23c	2579.0d	546.0c
30	50.26f	5,0b	7.47b	2633.0d	455.0de
5	66.13e	4.1bc	6.61d	3081.0c	589.0b
11	95.93c	2.0d	6.91d	2881.0c	644.0b
40	98.10b	3.2c	8.87a	4845.0a	966.0a
1	94.46c	6.1a	7.53b	4382.0a	1038.0a
EKÖF[&]	1.489**	1.087**	0.223**	463.011*	73.233*

[&]: En Küçük Önemli Fark, *: $P<0.05$, **: $P<0.01$

Ayan ve ark. (2017) yem börülcesinde bitki boyunun 70.60-203.40 cm arasında değiştiğini belirlerlerken, Katlan (2018) iki yıl süreyle yaptığı çalışmada ilk yıl yem börülcesinde bitki boyunu 46.10-54.60 cm, ikinci yıl 28.20-51.10 cm arasında değiştiğini saptamıştır. Araştırma sonuçları Katlan (2018)'ın belirlediği değerlerden yüksek, Ayan ve ark. (2017)'nın bulgularından ise düşüktür. Başaran ve ark. (2011) ana sap kalınlığını 8.60-9.80 mm arasında, Sallam ve İbrahim (2016) 5.20-9.20 mm arasında olduğunu belirlemişlerdir. Ana sap kalınlığın ilişkin sonuçlar Başaran ve ark. (2011)'in bulgularından düşük, Sallam ve İbrahim (2016)'in sonuçlarıyla benzerdir. Beycioğlu (2016) yem börülcesinde yeşil ot verimini 1971.38-5840.75 kg/da, kuru ot verimini de 451.40-1338.00 kg/da olarak tespit etmiştir. Ot verimlerine ilişkin bulgularımız araştırıcının elde ettiği verimler ile benzerdir.

Ham protein, ADF, K, Ca, Mg ve P oranları bakımından yem börülcesi genotipleri arasında fark belirlenmezken ($P>0.05$); en düşük NDF oranı 11 nolu (% 34.83) genotipte tespit edilmiştir ($P<0.01$, Tablo 2).

Tablo 2. Farklı yem börülcesi genotiplerinin bazı kalite özellikleri

Genotipler	Ham Protein Oranı (%)	NDF (%)	ADF (%)	K (%)	Ca (%)	Mg (%)	P (%)
Gazioğlu	20.45	36.94a	24.48	2.13	1.60	0.30	0.33
İ.H.K.	21.42	36.25a	24.58	2.14	1.57	0.36	0.35
İ.H.B.	20.80	37.02a	25.08	2.04	1.68	0.31	0.32
30	20.68	35.71b	24.60	2.10	1.59	0.35	0.32
5	20.83	35.82b	24.91	2.08	1.64	0.33	0.31
11	20.95	34.83c	24.57	2.09	1.64	0.35	0.32
40	20.71	35.23b	25.01	2.05	1.59	0.34	0.32
1	20.80	36.17a	24.72	2.08	1.60	0.35	0.34
EKÖF[&]	ÖD	0.877**	ÖD	ÖD	ÖD	ÖD	ÖD

[&]: En Küçük Önemli Fark, ÖD: Önemli Değil, **: $P<0.01$

Otçul hayvanlar ihtiyaçları olan proteinin büyük bölümünü doğadaki bitkilerden karşılarken, yetiştiriciliği yapılan hayvanlar bu ihtiyaçlarını kaba yemler ile hazırlanan kesif yemlerden karşılamaktadır (Tenikecier, 2019). Ruminantların beslenmesinde rasyonların protein düzeyi önemli bir kriterdir. Hayvanların kaliteli ve yüksek enerjili yem rasyonlarıyla beslenebilmeleri için protein değeri yüksek olan yem bitkilerinin üretilmesi gerekmektedir. Bununla birlikte, yem bitkilerinden elde edilen kuru otların NDF içerikleri yemin kalitesini ve sindirile bilirliğini ortaya koyan bir değerdir. NDF oranı %41-46 arasında olan kuru otlar çok iyi, % 47-53 arasında olanlar iyi, %54-60 arasında olanlar orta, %61-65 arasında olanlar ise kötü kalite olarak sınıflandırılmaktadır (AFGC, 2009). Yem börülcesinde NDF oranını Beycioğlu (2016) % 24.51-42.55, Ayan ve ark. (2017) % 29.42-37.25 arasında tespit etmişlerdir. Çalışmada tespit edilen NDF oranı Beycioğlu (2016) ile Ayan ve ark. (2017)'nın sonuçlarıyla benzerdir. Hayvan bünyesinde bulunan fosfor iskelet büyümesinde, diş, kıl-tüy oluşumunda, kanın kimyasal yapısında, bazı organların işlevlerinde, besinlerin enerjiye dönüştürülmesinde, vitamin ve enzimlerin aktivitesinde önemli rol oynamaktadır. Hayvan bünyesinde beraber işlev gösteren Ca/P oranının 2:1 veya 1:1 olması arzu edilmektedir (Ates ve Tekeli, 2005). Hayvanların bünyesinde bulunan toplam mineral maddenin yaklaşık olarak %4-5' ini oluşturan potasyumun büyük bölümü kas sisteminde bulunmaktadır. İşkembeli hayvanlar işkembe mikroorganizmalarının faaliyetleri için işkembesiz hayvanlara göre daha fazla potasyuma ihtiyaç duyarlar. Hayvan vücudundaki enzimlerin aktivasyonunda rol oynayan, kas ve iskelet sistemi işlevleri açısından önemli bir elementte magnezyumdur. Sinir ve kas fonksiyonlarının düzenlenmesi ve kemiğin mineral yapısının oluşumunda görev almaktadır. Bu nedenle, hayvanların beslenmesinde kullanılan otlardaki K ve Mg oranları ile birlikte diğer besin elementi içeriklerinin de bilinmesi oldukça önemlidir (Ateş, 2009). Ayan ve ark. (2012) yem börülcesinde Mg, Ca, K ve P oranlarını sırasıyla %0.46-0.51, %1.04-1.32, %1.30-1.65 ve %0.34-0.37 aralığında belirlemiştir. Saptanan makro element içerikleri bu araştırıcıların sonuçları ile benzerlik göstermektedir.

DRAFT

SONUÇ

Elde edilen sonuçlar ışığında; araştırmada kullanılan 40 ve 1 nolu yem börülcesi genotipleri ıslah materyali olarak değerlendirilerek, kaba yem üretimine katkı sağlayacak çeşitlerin geliştirilmesinde kullanılabileceklerdir.

TEŞEKKÜR

Bu makale Tekirdağ Namık Kemal Üniversitesi Fen Bilimleri Enstitüsü tarafından 2023 yılında kabul edilmiş olan Serkan BAYAZİT'a ait Yüksek Lisans Tezinin bir bölümü olup sağladığı destek ve katkılardan dolayı adı geçen enstitüye teşekkür ederiz.

KAYNAKLAR

Açıkgöz E 2001. Yem Bitkileri (Yenilenmiş 3. Baskı). Uludağ Üniversitesi Güçlendirme Vakfı Yayınları, Bursa.

Açıkgöz N, Akkaş E, Moghaddam A, Özcan K 1993. TARİST, Pc'ler İçin Türkçe İstatistik Paketi. Ulusal Ekonometri ve İstatistik Sempozyumu, İzmir.

AFGC 2009. Relative Feed Value. American Forage and Grassland Council. http://www.buckevenutrition .com /eQuinetechnical/EB22%20RELATIVE%20 FEED%20V ALUE.pdf (Erişim tarihi: 18.04.2023).

Alaca B, Özaslan Parlak A (2017). Mısır, sorgum Sudanotu melezi ile soya, börülce ve guarın karışık ekimlerinin silaj verimi ve kalitesine etkileri. ÇOMÜ Ziraat Fakültesi Dergisi, 5: 99-104.

AOAC 2019. Association of Official Analytical Chemists. The 21st Ed., Maryland, USA.

Ates E, Tekeli AS 2005. Forage quality and tetany potential of orchardgrass (*Dactylis glomerata* L.) and white clover (*Trifolium repens* L.) mixtures. Cuban Journal of Agricultural Science, 39: 97-102.

Ateş E 2009. Bakı ve yüksekliği farklı mera vejetasyonlarında değişik üçgül türleri (*Trifolium* sp.)'nin kimi morfolojik ve yem niteliği özellikleri (Doktora Tezi), Namık Kemal Üniversitesi Fen Bilimleri Enstitüsü, Tekirdağ.

Ayan I, Mut H, Basaran U, Acar Z, Ascı OO 2012. Forage potential of cowpea (*Vigna unguiculata* L. Walp). Turkish Journal of Field Crops, 17(2): 135-138.

Ayan I, Mut H, Acar Z, Can M 2017. Impacts of row spacing and sowing date on forage cowpea (*Vigna unguiculata* (L.) Walp.) hay yield and quality. Book of Proceedings, VIII. International Scientific Agriculture Symposium ''AGROSYM 2017'', October 05-08, Jahorina, Bosnia and Herzegovina, p. 1959-1964.

Baş M, Ateş E 2022. Farklı bezelye (*Pisum sativum* L.) genotiplerinin yem verimleri ve bazı kalite özellikleri. 5th International Eurasian Conference on Biological and Chemical Sciences (EurasianBioChem 2022), November 23-25, Ankara, Türkiye, s. 498-505.

Başaran U, Ayan I, Acar Z, Mut H, Aşçı O 2011. Seed yield and agronomic parameters of cowpea (*Vigna unguiculata* L.) genotypes grown in the Black Sea Region of Turkey. African Journal of Biotechnology, 10: 13461-13464.

Beycioğlu T 2016. Kahramanmaraş koşullarında börülce (*Vigna unguiculata* (L.) Walp) bitkisine uygulanan farklı sıra arası ve sıra üzeri mesafelerin verim unsurlarına etkisi (Yüksek Lisans Tezi), Kahramanmaraş Sütçü İmam Üniversitesi Fen Bilimleri Enstitüsü, Kahramanmaraş.

Düzgüneş O, Bakır Ö, Aksoy S, Akyüz Ö, Alınoğlu N 1965. Mer'alarımızla ilgili problemler ve çözüm önerileri. T.M.M.O.B. Ziraat Mühendisleri Odası Yayınları, Sıra No: 21, s.11.

Günay A 1992. Özel Sebze Yetiştiriciliği. Çağ Matbaası, Ankara.

Katlan E 2018. Farklı kükürt dozlarının yemlik börülce (*Vigna unguiculata* (L.) Walp)' nin ot verimi ve ot kalitesine etkisi (Yüksek Lisans Tezi), Bozok Üniversitesi Fen Bilimleri Enstitüsü, Yozgat.

Sallam MA, İbrahim IM 2016. Morphological, physiogical and chemical traits of some forage cowpea genotypes. American-Eurasian J. Agric.& Environ Sci.,16(2): 302-311.

Soya H, Avcıoğlu R, Geren H 2004. Yem Bitkileri. Hasad Yayıncılık, s.223.

Şehirali S. 1988. Yemeklik Dane Baklagiller. Ankara Üniv., Ziraat Fak. Yay. No: 1089, Ders Kitabı No: 314, Ankara, s. 435.

Tenikecier HS 2019. Bazı koca fiğ (*Vicia narbonensis* L.) çeşitlerinde farklı ekim zamanlarının morfolojik ve fizyolojik karakterler ile verim ve verim unsurlarına etkisinin saptanması üzerine araştırmalar (Doktora Tezi). Namık Kemal Üniversitesi Fen Bilimleri Enstitüsü, Tekirdağ.

DRAFT

Van Soest PJ, Robertson JB, Lewis BA 1991. Methods for dietary fibre, neutral detergent fibre, and nonstarch polysaccharides in relation to animal nutrition. Journal of Dairy Science, 74: 3583-3597.

Yücel C, Avcı M, Yücel H, Çınar S 2004. Çukurova taban koşullarında adi fiğ (*Vicia sativa* L.) hat ve çeşitlerinin ot verimi ve kalitesi ile ilişkili özelliklerinin saptanması. Tarla Bitkileri Merkez Araştırma Enstitüsü Dergisi, 13: 47-57.

DRAFT

ORAL PRESENTATION

Yapıştırıcı Kimyasal Üretim Atık Sularının Ardışık Koagülasyon–Elektrokimyasal/ Fotokimyasal Proseslerle Arıtımının Araştırılması

Berfin Boyraz[1*] (*ORCID: https://orcid.org/0009-0009-9284-0835*), Ezgi Ünal Yılmaz[1] (*ORCID: https://orcid.org/0000-0003-4178-0990*), Senem Yazıcı Güvenç[1] (*ORCID: https://orcid.org/0000-0002-2877-0977),* Gamze Varank[1] (*ORCID: https://orcid.org/0000-0003-3437-4505),* Emine Can Güven[1] (*ORCID: https://orcid.org/0000-0002-3540-3235),*Ahmet Demir[1] (*ORCID: https://orcid.org/0000-0001-6902-5626)*

[1]Yıldız Teknik Üniversitesi, İnşaat Fakültesi, Çevre Mühendisliği Bölümü, 34220, Esenler, İstanbul-Türkiye

*Sorumlu yazar e-mail: berfin.boyraz@std.yildiz.edu.tr

Özet

Makine, kimya, elektronik ve daha pek çok endüstriyel alanda kullanılan polimer bazlı yapıştırıcı kimyasal maddelerin üretim prosesleri su tüketiminin yüksek miktarlarda olduğu sistemlerdir. Üretim ve işleme proseslerinde kullanılan yüksek su miktarı bu endüstrilerden yüksek oranda atık su oluşumunu tetiklemektedir. Yüksek oranda organik kirlilik yüküne sahip bu atık suların arıtılmadan yüzeysel su kaynaklarına deşarj edilmesi alıcı ortamda çözünmüş oksijen oranını azaltmakta, ötrofikasyon meydana gelmekte, suyun rengi ve kokusu bozulmakta ve sucul sistem yoğun olarak etkilenmektedir. Yapıştırıcı kimyasal üretim atık suları çeşitli yöntemler ile arıtılsa bile uygulamada bazı sınırları bulunmaktadır. Biyolojik ayrışabilirliği düşük olan bu atık suları tekil veya hibrit ileri arıtma yöntemleri arıtılması gerekmektedir. Bu çalışmada, askıda ve kolloidal partikül gideriminde etkin koagülasyon prosesi ile onu takiben ileri arıtma teknikleri ile yapıştırıcı kimyasal üretim atık sularının arıtılabilirliği araştırılmıştır. Koagülasyon prosesi için poli alüminyum klorür (PAC) koagülant olarak kullanılmış ve KOİ, AKM, UV_{254} ve renk giderim verimleri sırasıyla %73.8, % 75, %87.3 ve %88.4 olmuştur. PAC ile koagülasyonun ardından elektrokimyasal proses olarak elektrooksidasyon (EO) ve persülfat ile iyileştirilmiş elektrooksidasyon (EO/PS) uygulanmıştır. PAC ile koagülasyonu takiben yapılan elektrokimyasal prosesler ile KOİ giderimi EO ile %36.5 ve EO/PS ile % 40'ı bulmuştur. PAC ile koagülasyonun ardından fotokimyasal proses olarak UV bazlı persülfat (UV/PS) ve UV bazlı perkarbonat (UV/PC) prosesleri uygulanmış ve degradasyon potansiyelini arttırmak üzere Fe^{+2} katalizör olarak ilave edilmiştir. PAC ardından fotokimyasal prosesler ile KOİ giderimi UV/PC ile %41 ve UV/PS ile %34.2 iken Fe^{+2}'nin katalizör olarak kullanılması sonucu KOİ giderimi UV/PC/Fe ile % 46.5 ve UV/PS/Fe ile %44'e yükselmiştir. Her ne kadar kanala deşarj kriterleri sağlanmış olsa da proseslerin detaylı optimizasyonu ile KOİ konsantrasyonunun alıcı ortam deşarj kriterlerine indirgenmesi mümkün olabilir.

Anahtar Kelimeler: Endüstriyel Atık Su, Hibrit Sistemler, Elektrokimyasal Prosesler, Fotooksidasyon

Investigation of Treatment of Adhesive Chemical Production Wastewater by Sequential Coagulation-Electrochemical/Photochemical Processes

Abstract

The production processes of polymer-based adhesive chemicals used in machinery, chemistry, electronics and many other industrial fields are systems with high water consumption. The high amount of water used in the production and processing processes triggers the formation of wastewater at high rates from these industries. The discharge of these wastewaters with high organic pollution load to surface water resources without treatment reduces the dissolved oxygen content in the receiving environment, eutrophication occurs, the color and odor of the water deteriorates and the aquatic system is intensely affected. Even if adhesive chemical production wastewater is treated with various methods, there are some limits in practice. These wastewaters with low biodegradability should be treated by single or hybrid advanced treatment methods. In this study, effective coagulation process for the removal of suspended and colloidal particles followed by advanced treatment techniques was investigated for the treatment of adhesive chemical production wastewater. Poly aluminium chloride (PAC) was used as coagulant for the coagulation process and COD, UV_{254} and color removal efficiencies were 83.8%, 97.3% and 98.4%, respectively. Coagulation with PAC was followed by electrochemical processes of electrooxidation (EO) and persulfate enhanced electrooxidation (EO/PS). COD removal by electrochemical processes following coagulation with PAC reached 36.5% and 40%. After

coagulation with PAC, UV-based persulfate (UV/PS) and UV-based percarbonate (UV/PC) processes were applied as photochemical processes and Fe^{+2} was added as a catalyst to increase the degradation potential. While COD removal by photochemical processes after PAC was 41% with UV/PC and 34.2% with UV/PS, COD removal increased to 46.5% with UV/PC/Fe and 44% with UV/PS/Fe when Fe^{+2} was used as a catalyst. Although the discharge criteria to the canal were met, with detailed optimization of the processes, it may be possible to reduce the COD concentration to the receiving environment discharge criteria.
Keywords: Industrial Waste Water, Hybrid Systems, Electrochemical Processes, Photooxidation

GİRİŞ

Çeşitli malzemeleri yapıştırma amacıyla; mobilya, denizcilik, otomotiv ve havacılık gibi pek çok alanda kullanılmakta olan polimer bazlı kimyasal maddeler yapıştırıcı olarak adlandırılmaktadır. Endüstriyel uygulamalar için yapıştırıcı üretimi, her formülasyon için temiz karışım kapları gerektirmektedir. Kapların yıkanma işlemi sonrasında, oldukça kirli bir atık su akışı elde edilmekte dolayısıyla yapıştırıcı maddelerin üretim prosesleri sonrasında çok büyük miktarda ve kirlilik yüküne sahip yıkama atık suyu açığa çıkmaktadır. Yapıştırıcı kimyasal üretim atık suyu esas olarak polimerler ve inorganik dolgu maddeleri, çözücüler ve deterjanlar gibi yapıştırıcıların seyreltilmiş bileşenlerini ve yüksek oranda metakrilat, ksilen, sodyum lauril sülfat, stiren ve reçine gibi organik bileşenleri içermektedir. Bu organik maddeler atık suda yüksek KOİ değerine ve amonyak azotu konsantrasyonuna sebep olmaktadır. KOİ 60.000 ile 150000 mg/L arasında değerlere ulaşırken $BOİ_5$ 15000 mg/L'yi aşmamaktadır (Kleine, Peinemann, & vd., 2002). Toplam kütle akışının yaklaşık %3-10'u arasında yüksek katı madde fraksiyonuna sahiptir. Bu nedenle yapıştırıcı kimyasal üretim atık suyunun, uygun arıtma yöntemleri vasıtasıyla arıtılmadan bir alıcı ortama deşarj edilmesi son derece tehlike arz etmektedir. Yapılan deşarj sudaki çözünmüş oksijenin azalmasına dolayısıyla su kütlesinin ötrofikasyonuna yol açarak siyah ve kötü kokulu su kütlesine ve balıkların ölümüne neden olmaktadır.
Yapılan çalışmalarda yapışkan malzeme üretim prosesinden kaynaklanan atık suların arıtımı için pek çok arıtma yöntemi denenmiştir. Bunlar; fotokataliz (Kleine, Peinemann, & ark., 2002), biyolojik arıtma (Posey, Cyr, & ark., 2014), kimyasal flokülasyon, fenton prosesi, aktif karbon adsorpsiyonu ve membran filtrasyonudur. Fakat bu teknolojilerin uygulama aşamasında pek çok sorunla karşılaşılmaktadır. Örneğin membran filtrasyonu ya da adsorpsiyon uygulamaları bu atık suların yüksek katı madde içeriği nedeniyle kullanışlı değildir. Buna ek olarak atık suyun düşük biyolojik parçalanabilirliği ve değişken karakterizasyonu nedeniyle biyolojik arıtma yönteminin de uygulanması tercih edilmemektedir (Lu, Zhuo, & ark., 2022). Arıtma veriminin diğer yöntemlere kıyasla daha yüksek olduğu kimyasal koagülasyon ve fenton prosesleri ise alüminyum tuzu, demir tuzu ve H_2O_2 gibi çok sayıda madde gerektirmektedir. Nakliye maliyetlerinin artması, depolama alanı ihtiyacı ve kullanım sırasında yaşanmakta olan güvenlik riskleri nedeniyle bu teknolojiler su arıtımında sınırlı kalmaktadır.

Elektrokimyasal arıtma yöntemleri; kolay işletilebilirliği, yüksek verimliliği ve ilave oksidatif madde ihtiyacı duymaması nedeniyle son yıllarda hızla gelişmiştir. Elektrokoagülasyon (EC), elektrooksidasyon (EO), elektro-fenton (EF) gibi prosesler atık su arıtımında kullanılmaktadır. Elektrokoagülasyon teknolojisi geniş uygulama yelpazesi ve düşük maliyeti nedeniyle en yaygın kullanılan elektrokimyasal teknolojilerden biridir (Lu, Zhuo, & ark., 2022).

EC prosesinin temel prensibi elektrolizdir. Elektroliz; elektrik akımının kullanılmasıyla bileşiklerin parçalanması işlemidir (Boinpally, Kolla, & ark., 2023). EC prosesinin kirleticileri uzaklaştırmadaki prensibi alüminyum ve demir gibi metal anotların anodik çözünmeye uğraması ve hidrolizi ile çok az çözünen $Al(OH)_3$, $Fe(OH)_2$ ve $Fe(OH)_3$ gibi metal hidroksitlerin oluşması şeklindedir (Özyonar & Karagözoğlu, 2012). Son olarak ortaya çıkan bu metal hidroksitlerin metal-polimer kompleksleri oluşturmasıyla birlikte kirleticiler adsorplanarak koagüle olmaktadır.

EC prosesinin kirleticilerin atık su içerisinden uzaklaştırılmasında çok etkili olduğu kanıtlanmıştır ve çamur üretiminin azalması, kimyasal kullanımına gerek olmaması, kullanım kolaylığı, basit ekipman, düşük maliyet ve daha az reaksiyon süresi gibi çeşitli avantajlara sahiptir. Bununla birlikte, tekniğin 'kurban elektrotların' atık suda çözünmesi ve düzenli olarak değiştirilmesi gerekmesi ve elektrik kullanımı nedeniyle bazı dezavantajları da vardır (Negash, Tibebe, & ark., 2023). EC prosesinde genellikle Al ya da Fe elektrotlar kullanılmaktadır. Al elektrot kullanıldığı takdirde meydana gelecek reaksiyonlar şu şekildedir (Yakamercan, Bhatt, & ark., 2023):

DRAFT

Anot için alüminyum elektrot: $Al \rightarrow Al^{+3} + 3e^{-}$ (1)

Alkali koşullar: $Al + 3OH^{-} \rightarrow Al(OH)_3$ (2)

Asidik koşullar: $Al^{+3} + 3H_2O \rightarrow Al(OH)_3 + 3H^{+}$ (3)

Katottaki reaksiyonlar: $2H_2O + 2e^{-} \rightarrow H_2 + 2OH^{-}$ (4)

Elektrooksidasyon, çeşitli endüstriyel sektörlerden açığa çıkan atık suların arıtılması amacıyla yaygın olarak kullanılan elektrokimyasal yöntemlerden biridir. EO prosesi ile amaçlanan atık su içerisindeki kirletici maddelerin daha az toksik ve biyolojik parçalanabilirliği daha yüksek olan CO_2 ve H_2O gibi nihai ürünlere dönüştürülmesidir. EO doğrudan veya dolaylı oksidasyon yoluyla gerçekleşmektedir (Titchou, Zazou, & vd., 2021). Dolaylı anodik oksidasyon sırasında aşağıdaki reaksiyonlar meydana gelmektedir (Saeed, Hameed, & ark., 2023):

Anot: $2Cl^{-} \rightarrow Cl_2 + 2e^{-}$ (5)

Toplu reaksiyonlar:$Cl_2 + H_2O \rightarrow HOCl + H^{+} + Cl^{-}$ (6)

$Cl_2 + OH^{-} \rightarrow HOCl + Cl^{-}$ (7)

$HOCl \rightarrow H^{+} + OCl^{-}$ (8)

$R + OCl^{-} \rightarrow 2CO^{2} + Cl^{-} + H_2O$ (9)

Bu yöntemin en büyük dezavantajı yüksek oranda askıda katı madde içeren atık suların arıtılmasında uygulanamaması ve uzun çalışma süresidir. Bu nedenle EO prosesini kullanmak için atık sudaki askıda katı maddelerin ilk önce farklı teknikler kullanılarak giderilmesi gerekmektedir (Asfaha, Tekile, & ark., 2021). Önerilen en yaygın teknik EC'dir.

Hibrit EC-EO prosesinde atık sudaki askıda katı maddeler EC ile giderilmekte, ardından EO ile kalan kirleticiler okside edilmektedir. EC ve EO'nun hibrit prosesi sadece EC veya sadece EO kullanılarak etkili bir şekilde elde edilemeyen KOİ, TOK, NH_4^{+}-N, nitratlar ve fenol giderimini artırmaktadır (Asfaha, Tekile, & ark., 2021). Bu nedenle, hibrit yöntem hem askıdaki ve kolloidal hem de kararlı kalıcı organik bileşikleri pratik bir sürede izin verilen sınıra kadar giderebilir. Böylece elektrotların ömrünü arttırırken enerji tüketimini azaltabilir (Almukdad, Hafiz, & ark., 2021). Yapılan bazı çalışmalarda ilaç, deri, tekstil, boya üretimi, yapıştırıcı üretimi, otomotiv, kimya, metal terbiye ve gıda işleme gibi çeşitli endüstri atık sularının, hibrit elektrokoagülasyon-elektrooksidasyon prosesi ile yüksek verimlilikle arıtıldığı gösterilmiştir (Asfaha, Tekile, & ark., 2021), (Saeed, Hameed, & ark., 2023), (Hernández, Díaz, & ark., 2010), (Lu, Zhuo, & ark., 2022).

Parçalayıcı prosesler olarak tanımlanan ileri oksidasyon prosesleri hidroksil ve sülfat radikalleri gibi yüksek reaktif oksijen türleri üretimi esasına dayanmaktadır ve üretilen reaktif serbest radikaller organik moleküllerin büyük kısmını parçalamaktadır (Ghanbari, Khatebasreh, & ark., Evaluation of peroxymonosulfate/O3/UV process on a real polluted water with landfill leachate: Feasibility and comparative study, 2021). Reaktif oksijen türlerinin üretimi için hidrojen peroksit, peroksidisulfat (PS), peroksimonosülfat (PMS) ve perkarbonat (PC) gibi farklı oksidantlar kullanılmaktadır (Eslami, Mehdipour, & ark., 2020) (Hassani, Eghbali, & ark., 2020). Peroksitlerin bağları ultraviyole (UV), ultrases (US), plazma, mikrodalga gibi yöntemler ile ayrılarak reaktif serbest radikaller meydana getirmektedir (Zhang, Zhang, & ark., 2015), (Ghanbari, Moradi, & ark., 2014). UV, kuvvetli oksidantların varlığında organik kirleticilerin gideriminde iyi performans göstermektedir (Lin, Qin, & ark., 2020). Oksidantların UV ile etkili bir şekilde aktive olması sonucu $OH^{\cdot}$, $O_2^{\cdot -}$, $SO_4^{\cdot -}$ reaktif türleri oluşmakta ve organik bileşikler toksik olmayan düşük moleküler ağırlıklı ürünlere ayrışmaktadır. UV bazlı ileri oksidasyon prosesleri çamur oluşumunun olmaması, ikincil bir kirletici oluşmaması, faz transferi problemi olmaması, kolay işletim, düşük yatırım maliyeti gibi avantajlara sahiptir (Gorito, Pesqueira, & ark., 2021).

$H_2O_2 + UV \rightarrow HO\cdot + HO\cdot$ (10)

$S_2O_2^{2-} + UV \rightarrow SO_4^{\cdot -} + SO_4^{\cdot -}$ (11)

$HSO_5^{-} + UV \rightarrow HO\cdot + SO_4^{\cdot -}$ (12)

Son zamanlarda PC hidrojen peroksite alternatif olarak kullanılmaktadır. Sodyum perkarbonat perkarbonatın en yaygın olarak kullanılan kaynağıdır.

$$Na_2CO_3.1.5H_2O_2 \rightarrow Na_2CO_3 + 1.5H_2O_2 \quad (13)$$

Buna ilave olarak aktivatör olarak UV uygulandığında hidrojen peroksit bikarbonat ile reaksiyona girerek peroksikarbonat oluşur (14) ve UV etkisi ile peroksikarbonattan hidroksil ve karbonat radikalleri üretilir (15).

$$HCO_3^- + H_2O_2 \rightarrow HCO_4^- + H_2O \quad (14)$$

$$HCO_4^- + UV \rightarrow HO^{\cdot} + CO_3^{\cdot -} \quad (15)$$

Sodyum perkarbonat iyi derecede termal stabilite ve suda çözünürlük özelliklerine sahip katı fazlı oksidanttır. Katı fazlı olduğu için sıvı peroksite göre depolanması, taşınması ve işlenmesi daha güvenilir ve daha kolaydır (Fu & Chen, 2015). Geniş pH aralığında etkin olup aynı zamanda organik kirleticileri ayrışması esnasında oluşan ara ürünlerin sebep olduğu pH düşüşlerine karşı tampon görevi yapmaktadır (Zhang, Zhang, & ark., 2017). Ayrıca sodyum perkarbonat toksik bir bileşik değildir (Zhang, Ren, & ark., 2017). Diğer peroksitlere oranla maliyeti de düşüktür (Gao, Song, & ark., 2020). Sodyum perkarbonat kirleticilerle doğrudan da reaksiyona girmekte ancak aktive olduğunda performansı artmaktadır. PC aktivasyonu sonucu hidroksil, süperoksit ve karbonat radikalleri oluşmaktadır (Zhan, Kuang, & ark., 2021).

Son yıllarda ultraviyole (UV) bazlı ileri oksidasyon prosesleri de atık su arıtımında kullanılmakta olan en etkili teknolojilerdendir. Fiziksel bir dezenfeksiyon işlemi olan UV dezenfeksiyon, kimyasal girdi olmaması, zararlı dezenfeksiyon yan ürünlerinin düşük oluşumu, düşük maliyet ve küçük ayak izi gibi çeşitli avantajlara sahiptir (Sun, Li, & ark., 2022). Bunlardan UV/hidrojen peroksit (UV/H_2O_2) yaygın olarak uygulanmaktadır. Ancak sıvı H_2O_2, nakliye ve depolama sırasında hızlı ayrışma ve patlama nedeniyle güvenlik tehlikesi oluşturmaktadır (Wu, Chi, & ark., 2010). Bu nedenle sıvı H_2O_2'nin sodyum perkarbonat veya persülfat ile değiştirilmesi önerilmiştir (Gao, Song, & ark., Comparative toxicity reduction potential of UV/sodium percarbonate and UV/hydrogen peroxide treatments for bisphenol A in water: An integrated analysis using chemical, computational, biological, and metabolomic approaches, 2021).

Bu çalışmada yüksek askıda katı madde içeriğine sahip yapışkan kimyasal üretim atık suları ilk olarak koagülasyon prosesine tabi tutulup, ardından dirençli kirleticilerin giderimi için elektrokimyasal ve fotokimyasal yöntemler ile arıtılabilirliği incelenmiştir. İlk arıtma kademesi olan koagülasyon prosesi PAC koagülantı kullanılarak gerçekleştirilmiştir. Böylece askıda katı madde konsantrasyonu düşürülen atık suyun hibrit ileri arıtma yöntemleri ile organik kirlilik yüklerinin indirgenmesi araştırılmıştır.

MATERYAL VE METOT

Atık Su Karakterizasyonu

Yapıştırıcı kimyasal üretim atık suyu, İstanbul Haramidere'de işletilen bir kimyasal üretim fabrikasının kazan yıkama suyundan alınmıştır. Tek seferde alınan atık su numunesi laboratuvara getirilerek, deneysel çalışmalar boyunca biyolojik reaksiyonları engellemek için +4 °C'de muhafaza edilmiştir. Atık su karakterizasyonu Tablo 1'de verilmiştir.

Tablo 1. Tutkal Atık Suyu Karakterizasyonu

Parametre	**Birim**	**Değer**
pH	-	7,1
İletkenlik	µS/cm	1900
KOİ	mg/L	11209
UV_{254}	ABS	75
Renk	mg/L	65400
AKM	mg/L	2193

Kullanılan Kimyasallar

Deneysel çalışmalarda kullanılan tüm kimyasallar analitik saflıktadır. Atık suyun pH seviyesini ayarlamak için 1 N sülfürik asit (H_2SO_4) ve sodyum hidroksit (NaOH) kullanılmıştır. Koagülasyon prosesinde poli alüminyum klorür (PAC), elektrooksidasyon ve fotokimyasal oksidasyon proseslerinde potasyum peroksidisülfat ($K_2S_2O_8$, PS) ve sodyum perkarbonat ($Na_2CO_3 \cdot 1.5H_2O_2$, PC) kullanılmıştır.

Deneysel Çalışma

Koagülasyon Prosesi

Yapıştırıcı kimyasal üretim atık suyu PAC ile koagülasyon prosesine tabi tutulmuştur. Koagülasyon prosesi 100 mL atık su hacmi alınarak, ham pH değerinde 1 dk 200 rpm hızla hızlı karıştırılıp, 20 dk boyunca 30 rpm hızla yavaş karıştırılarak gerçekleştirilmiştir. Ardından, 30 dk sedimantasyona tabi tutulup üst fazdan alınan atık su numunesinde analizler yapılmıştır.

Elektrooksidasyon Prosesi

Deneysel çalışmalar laboratuvar ölçekli ve 200 cm^3 hacme sahip pleksiglass malzemeden yapılmış dikdörtgen prizma reaktörde yürütülmüştür. Elektrooksidasyon çalışmasında 100 mL atık su içerisine, monopolar bağlı biri anot (Pt/Ti) ve biri katot (Grafit) elektrotlar, aralarında 2 cm mesafe olacak şekilde yerleştirilmiştir. PS eklenen proseste 0.5 g/L oranında PS eklenmiş ve diğer tüm şartlar tek başına EO prosesi ile aynı tutulmuştur. Reaksiyon başlamadan önce atık su pH değeri 5 seviyesine ayarlanmıştır. Atık suya 90 dk reaksiyon süresi boyunca 0.3 A elektrik akımı verilmiştir. Reaksiyon süresi sonunda elektrik bağlantıları kesilerek alınan atık su numunelerinde analizler yapılmıştır.

Fotokimyasal Oksidasyon Prosesi

Deneysel çalışmalar laboratuvar ölçekli ve 8 cm çapında 16 cm yükseklikte silindirik quartz reaktörde gerçekleştirilmiştir. 100 mL atık suyun pH değeri 5'e ayarlanarak 90 dk boyunca fotoliz yürütülmüştür. Oksidan olarak prosese PS ve PC 0.5 g/L eklenmiştir. Katalizör olarak 0.1 g/L Fe^{+2} ilavesi yapılmıştır. Manyetik karıştırıcı ile atık suyun reaksiyon boyunca homojen bir şekilde dağılımı sağlanmıştır. Her bir deney seti sonunda Al(OH)x ve Fe(OH)x'lerin çökebilirliğini arttırmak üzere pH seviyesi 7-8 aralığına çıkarılmış ve numuneler 4000 rpm'de 5 dakika santrifüjlenmiştir. Santrifüj sonrası üst fazdan numune alınarak muhafaza edilmiş ve analizler yapılmıştır.

Analitik Yöntemler

Koagülasyon, elektrooksidasyon ve fotokimyasal oksidasyon prosesleri ile KOİ, UV_{254} ve renk giderim verimleri (%) aşağıda verilen eşitlik kullanılarak hesaplanmıştır:

$$R\ giderim\ verimi\ (\%) = \left(\frac{(R)_i - R_t}{R_i}\right).100 \quad (16)$$

Burada $(R)_t$ t anındaki kirletici konsantrasyonunu, $(R)_i$, başlangıç anındaki kirletici konsantrasyonunu ve t reaksiyon süresini (dk) ifade etmektedir.

Optimum koşullarda elektrokoagülasyon prosesi maliyetini hesaplamak için aşağıda yer alan formüller kullanılmıştır (17-18).

$$SEC\ (kWh/m^3) = \frac{U \times i \times t}{10^3 \times V} \quad (17)$$

$$SEC\ (kWh/kg\ KOİ) = \frac{U \times i \times t}{10^3 \times V \times (KOİ_0 - KOİ_t)} \quad (18)$$

Burada $KOİ_0$ ve $KOİ_t$ sırasıyla t_0 ve t_t zamanlarında KOİ konsantrasyonlarını (kg/L), U uygulanan voltaj miktarını, i akım değerini (A), t arıtma süresini (saat) ve V atık su hacmini temsil etmektedir.

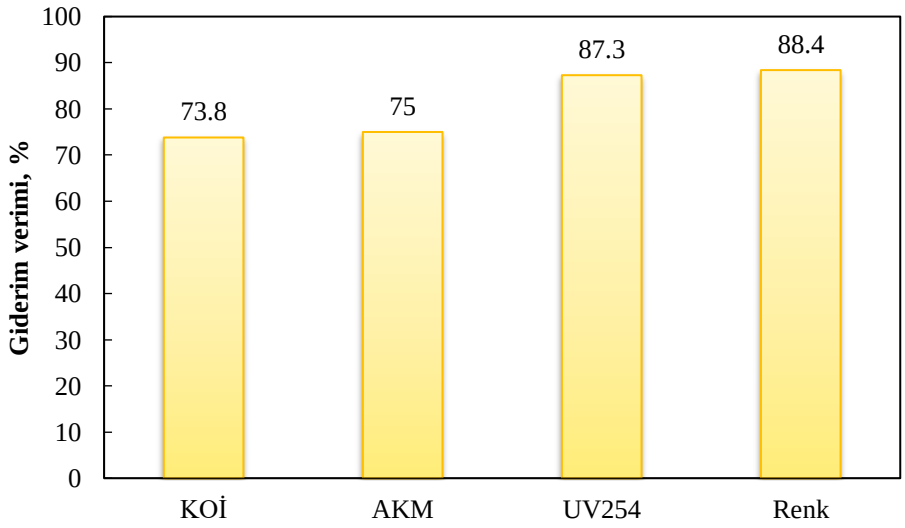
BULGULAR ve TARTIŞMA

Koagülasyon Prosesi

Atık su arıtımında koagülasyon-flokülasyon işlemi oldukça pratik ve kontrol edilebilir bir teknik olup, özellikle ön arıtmada yaygın olarak kullanılmaktadır. Toplam askıda katı madde miktarının, organik içeriğin ve rengin azaltılması için gerekli bir adım olduğu gibi, pıhtılaşma işleminden sonraki ikinci işlemin verimliliğinin arttırılması açısından da önemlidir (Amor, Torres-Socías, & ark., 2015). Koagülasyon çalışmasının temeli partikül destabilizasyonunu içermektedir. Bu çalışmada parçacık kararsızlaştırması için koagülant olarak PAC ilave edilerek gerçekleştirilen deneysel çalışmanın sonuçları Şekil 1'de verilmiştir. Ham atık su pH değerinde (pH=7.1) ve 1 g/L PAC dozlanarak gerçekleştirilen koagülasyon prosesi sonucu % 75

AKM, % 73.8 KOİ, % 87.3 UV_{254} ve % 88.4 renk giderimi sağlanmıştır. Koagülasyon prosesi yüksek oranda pH'a bağlıdır. Atık suyun pH değeri, atık suya metal bazlı pıhtılaştırıcılar ekleyerek çözünebilir ve çözünmeyen polimerik metal türlerinin doğasını doğrudan etkiler. Koagülasyon işlemindeki pH dengesi, birbiriyle yarışan iki kuvvetin dengesine dayanır. Bu kuvvetlerden biri H^+ iyonları ile metal hidroliz ürünleri arasında organik ligandlarla etkileşime giren kuvvet, diğeri ise hidroksit iyonları ile organik anyonlar arasında metal hidroliz ürünleriyle etkileşime giren kuvvettir (Maleki & Zazouli, 2009). Düşük pH değerlerinde H^+ iyonları, organik ligandlar için metal hidroliz ürünleriyle rekabet eder. Bu nedenle oluşan organik asitlerin bir kısmı çökelmez ve giderim verimleri düşük kalır. Bununla birlikte, yüksek pH değerlerinde, hidroksit iyonları, metal adsorpsiyon bölgeleri için organik bileşiklerle rekabet eder ve metal hidroksitlerin çökeltilmesi, temel çökeltme işlemi yerine ağırlıklı olarak yarı çökeltme işlemiyle gerçekleşir. Bu nedenlerden dolayı ham atık su pH değerleri olan nötr seviyede çalışılmıştır.

Koagülant dozu artışı ile oluşan çamur hacminin de artacağı düşünüldüğünden ve uygulanacak hibrit sistemin ilk adımı için elde edilen kirletici giderimi yeterli görüldüğünden optimum doz çalışmaları yürütülmemiştir. Ayrıca, kolloidal parçacıklar negatif yüklüdür. Uygun dozda koagülant ilavesi ile katyonlar optimum oranda verilir ve negatif yüklü kolloidler nötralize edilir. Ancak optimum değerin üzerinde bir koagülant eklendiğinde ortamda daha fazla pozitif yüklü iyon olacak ve buna bağlı olarak yük itmesi sonucu kolloidler tekrar stabil hale gelecektir. Bunlara bağlı olarak seçilen koagülant PAC için 1 g/L dozlama ile arıtma işlemi yapılarak, 2. kademe arıtma işlemi için koagüle edilmiş atık su biriktirilmiştir.



Şekil 1. PAC kullanılan koagülasyon prosesi ile kirletici giderimi

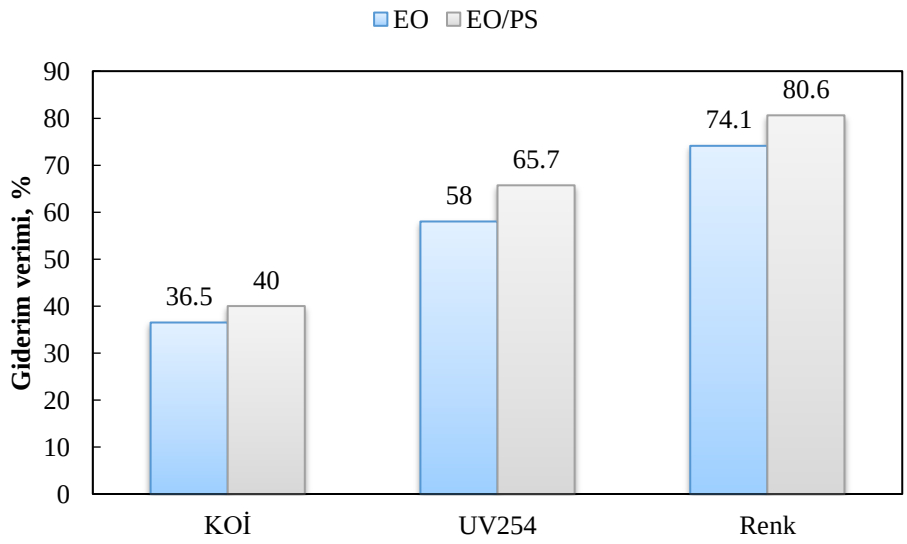
Hibrit Koagülasyon – Elektrooksidasyon Prosesi

Yapıştırıcı kimyasal atık suyu 2. kademe olarak elektrooksidasyon prosesine tabi tutulmuştur. Elektrooksidasyon ile organik madde ve renk giderim verimleri incelenmiştir. Elde edilen verimleri iyileştirebilmek için persülfat gibi çevre dostu oksidan ilavesi yapılmıştır. Hem tek başına EO hem de persülfat ilaveli EO prosesi sonuçları Şekil 2'de verilmiştir.

Elektrooksidasyon prosesinde atık su pH değeri, doğrudan ve dolaylı oksidasyonun iki mekanizması üzerinde meydana gelen etkili bir parametredir. Asidik pH değerlerinde hidroksil radikalleri üzerinde temizleyici etki yapan karbonat ve bikarbonat iyon konsantrasyonu azaltılarak istenmeyen oksijen oluşum reaksiyonları azaltılır (Anglada, Urtiaga, & ark., 2010). Buna bağlı olarak reaksiyon hızı artar. Atık sudaki klorür iyonunun şekli doğrudan pH ile ilişkilidir (Anglada, Urtiaga, & ark., 2010), (Mandal, Dubey, & ark., 2017). Sulu klor, 3,3'ün altındaki pH seviyelerinde elektroliz sırasında oluşan aktif klor türleri arasında baskın olan türdür. pH 3,3'ün altında elektroliz sonucunda klorlu organik bileşiklerin oluşumu artacaktır ki bu da istenmeyen bir durumdur. Bu nedenle EO prosesinde pH' ın 3,3'ün altında olmaması gerekir. Ham atık su pH değerinin nötr seviyelerde olması ve kimyasal ilavesini minimize etmek için bu çalışmada atık su pH değeri 5'de sabit tutulmuştur. Şekil 2'den de anlaşıldığı gibi EO prosesi ile %36.5 KOİ, %58 UV_{254} ve %74.1 renk giderimi sağlanmıştır.

Bu çalışmada elektrolitik reaktöre oksidan olarak PS ilave edilmiş ve PS ilavesinin DSA anotları kullanılarak EO prosesi ile atık sudan KOİ, UV_{254} ve renk giderilmesine etkisi araştırılmıştır. Dirençli organik maddelerin giderim potansiyelini arttırmak için yüksek reaktif türlerin oluşmasını sağlayan persülfat ilavesi gerçekleştirilmiştir. Persülfat ilaveli EO prosesinde 40 KOİ, %65 UV_{254} ve %80.6 renk giderimi elde edilmiştir. Tek başına EO prosesine kıyasla EO/PS prosesi ile her bir kirletici parametrenin giderim verimi en az yaklaşık %5 oranında artış göstermiştir. Burada, sistem içerisinde gerçekten dolaylı oksidasyonun (sülfat radikallerinin oluşumu) pozitif etkisi görünmektedir.

Hibrit koagülasyon – EO/PS prosesi ile toplam kirletici giderim verimleri %84.3 KOİ, %95.6 UV_{254} ve % 97.7 renk olarak hesaplanmıştır.



Şekil 2. PAC ile koagülasyon sonrası elektrooksidasyon prosesi ile kirletici giderimi

Hibrit Koagülasyon - Fotokimyasal Oksidasyon Prosesi

Bir diğer 2. kademe arıtma prosesi olarak fotokimyasal oksidasyon prosesi çalışılmıştır. Fotokimyasal oksidasyon prosesi için oksidan olarak PS ve PC iki tür seçilmiştir. PS ve PC oksidanlarının organik maddeyi oksitlemesi için daha uygun pH değerlerinin asidik koşullar olduğu pek çok çalışmada kanıtlanmıştır (Li, Xiao, & ark., 2019), (Can-Güven, Daniser, & ark., 2022), (Türk, Adalar, & ark., 2023). Bu sebeple fotokimyasal oksidasyon deneyleri pH 5 değerinde sabit tutularak yürütülmüştür. UV/PS ve UV/PC proseslerinde meydana gelen mekanizma yüksek oksidasyon potansiyeline sahip serbest radikallerin oluşmasıdır. UV/PS prosesi radikal türleri olarak hidroksit ve sülfat radikalleri oluşurken, UV/PC prosesinde hidroksit ve karbonat radikalleri oluşmaktadır. Farklı radikal türlerin etkin olduğu bu prosesler ile kirletici giderim verimleri de farklılık göstermektedir. Şekil 3 fotokimyasal oksidasyon prosesi sonuçlarını göstermektedir. Şekil 3'ten de anlaşıldığı gibi UV/PC prosesi KOİ giderim verimi %41, UV/PS prosesi ile KOİ giderim verimi %34.2'dir. Karbonat radikallerinin daha yüksek organik madde parçalama etkisi gösterdiği anlaşılmaktadır. Ayrıca, UV ışınlarının yanı sıra ortamda Fe^{+2} iyonlarının bulunması durumunda ikincil bir aktivatör etkisi göstererek serbest radikallerin oluşumunu desteklemektedir (Rivas, Gimeno, & ark., 2010), (Hassani , Scaria, & ark., Sulfate radicals-based advanced oxidation processes for the degradation of pharmaceuticals and personal care products: A review on relevant activation mechanisms, performance, and perspectives, 2023).

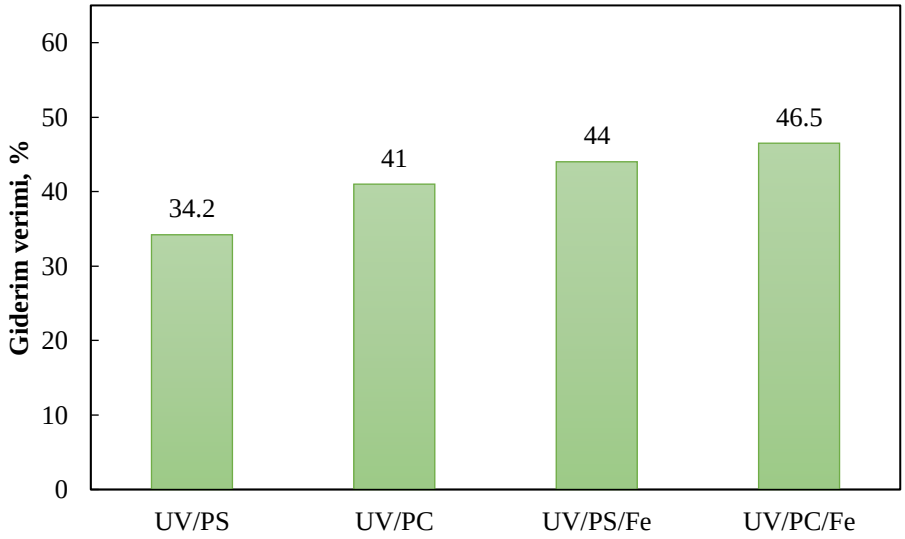
$$Fe^{2+} + S_2O_8^{2-} \rightarrow Fe^{3+} + SO_4^{\cdot-} + SO_4^{2-} \quad (19)$$

$$2Na_2CO_3.3H_2O_2 \rightarrow 2Na_2CO_3 + 3H_2O_2 \quad (20)$$

$$H_2O_2 + Fe^{2+} \rightarrow {}^{\bullet}OH^{+} + OH^{-} + Fe^{3+} \quad (21)$$

Bu çerçevede Şekil 3'e bakılarak Fe^{+2} ' nin katalizör olarak kullanıldığı her iki fotokimyasal oksidasyon proseslerinde KOİ giderim verimlerinde artışlar gözlenmiştir. UV/PS/Fe prosesi ile KOİ giderim verimi %44'e, UV/PC/Fe prosesi ile KOİ giderim verimi %46.5'e yükselmiştir.

Hibrit koagülasyon – UV/PC/Fe prosesi toplam KOİ giderim verimi %85.9 olmuştur. Ancak diğer kirletici parametrelerin ölçümleri spektrofotometrik olarak yapıldığından ve Fe kullanıldığında suya kendisi de renk verdiğinden negatif girişim meydana getirmektedir. Bu sebeple fotokimyasal oksidasyon prosesleri sadece KOİ giderim verimi üzerinden değerlendirilmiştir.



Şekil 3. KOİ giderim verimi üzerine fotokimyasal proseslerin etkisi

SONUÇ

Bu çalışmada yapıştırıcı kimyasal üretim atık sularının alternatif arıtma yöntemleri kullanılarak kirletici parametrelerinin indirgenmesi amaçlanmıştır. Yüksek askıda katı madde ve organik madde konsantrasyonunu sahip bu atık suların öncelikle koagülasyon prosesi ve ardından elektrooksidasyon/fotokimyasal oksidasyon proseslerini içeren hibrit arıtma sistemleri ile arıtılabilirlikleri araştırılmıştır. Koagülasyon prosesi ile AKM, KOİ, UV_{254} ve renk giderim verimleri sırasıyla %75, %73.8, %87.3 ve %88.4 olmuştur. Ardından sırasıyla EO ve EO/PS prosesleri ile %36.5 ve %40 KOİ giderimleri elde edilirken, sırasıyla UV/PS/Fe ve UV/PC/Fe ile prosesleri ile %44 ve %46.5 KOİ giderimleri elde edilmiştir. Hibrit koagülasyon – EO/PS prosesi ile toplam KOİ giderimi %84.3 ve hibrit koagülasyon – UV/PC/Fe prosesi ile toplam KOİ giderimi %85.9 olarak hesaplanmıştır. Her iki hibrit sistem de her ne kadar yüksek oranlar organik madde giderimi sağlamış olsalar da organik madde konsantrasyonu için alıcı ortam deşarj kriterlerine indirilememiştir. Proseslerin hem kirletici konsantrasyonu bakımından hem de maliyet analizleri bakımından işletme parametrelerinin optimizasyonu gerekmektedir.

TEŞEKKÜR

Bu araştırma Yıldız Teknik Üniversitesi-Bilimsel Araştırma Projeleri Koordinatörlüğü tarafından FBA-2023-5541 araştırma projesi numarasıyla desteklenmiştir.

KAYNAKLAR

Almukdad, A., Hafiz, M., & ark. (2021). Unlocking the application potential of electrocoagulation process through hybrid processes. *Journal of Water Process Engineering*.

Amor, C., Torres-Socías, E. D., & ark. (2015). Mature landfill leachate treatment by coagulation/flocculation combined with Fenton and solar photo-Fenton processes. *Journal of Hazardous Materials*, 261-268.

Anglada, Á., Urtiaga, A. M., & ark. (2010). Laboratory and pilot plant scale study on the electrochemical oxidation of landfill leachate. *Journal of Hazardous Materials*, 729-735.

Asfaha, Y. G., Tekile, K. A., & ark. (2021). Hybrid process of electrocoagulation and electrooxidation system for wastewater treatment: A review. *Cleaner Engineering and Technology*.

Boinpally, S., Kolla, A., & ark. (2023). A state-of-the-art review of the electrocoagulation technology for wastewater treatment. *Water Cycle*, 26-36.

Can-Güven, E., Daniser, Y., & ark. (2022). Effective removal of furfural by ultraviolet activated persulfate, peroxide, and percarbonate oxidation: Focus on influencing factors, kinetics, and water matrix effect. *Journal of Photochemistry and Photobiology A: Chemistry*.

Eslami, A., Mehdipour, F., & ark. (2020). Sono-photo activation of percarbonate for the degradation of organic dye: the effect of water matrix and identification of by-products. *Journal of Water Process Engineering*.

Fu, Y.-H., & Chen, C.-Y. (2015). Tuning of hydrogen peroxide-responsive polymeric micelles of biodegradable triblock polycarbonates as a potential drug delivery platform with ratiometric fluorescence signaling. *Polymer Chemistry*.

Gao, J., Song, J., & ark. (2020). Comparative toxicity reduction potential of UV/sodium percarbonate and UV/hydrogen peroxide treatments for bisphenol A in water: An integrated analysis using chemical, computational, biological, and metabolomic approaches. *Water Research*.

Gao, J., Song, J., & ark. (2021). Comparative toxicity reduction potential of UV/sodium percarbonate and UV/hydrogen peroxide treatments for bisphenol A in water: An integrated analysis using chemical, computational, biological, and metabolomic approaches. *Water Research*.

Ghanbari, F., Khatebasreh, M., & ark. (2021). Evaluation of peroxymonosulfate/O3/UV process on a real polluted water with landfill leachate: Feasibility and comparative study. *Korean Journal of Chemical Engineering*, 1416-1424.

Ghanbari, F., Moradi, M., & ark. (2014). Textile wastewater decolorization by zero valent iron activated peroxymonosulfate: compared with zero valent copper. *Journal of Environmental Chemical Engineering*, 1846-1851.

Gorito, A. M., Pesqueira, J. F., & ark. (2021). Ozone-based water treatment (O3, O3/UV, O3/H2O2) for removal of organic micropollutants, bacteria inactivation and regrowth prevention. *Journal of Environmental Chemical Engineering*.

Hassani , A., Scaria, J., & ark. (2023). Sulfate radicals-based advanced oxidation processes for the degradation of pharmaceuticals and personal care products: A review on relevant activation mechanisms, performance, and perspectives. *Environmental Research*.

Hassani, A., Eghbali, P., & ark. (2020). Acetaminophen removal from aqueous solutions through peroxymonosulfate activation by CoFe2O4/mpg-C3N4 nanocomposite: insight into the performance and degradation kinetics. *Environmental Technology & Innovation*.

Hernández, I. L., Díaz, C. B., & ark. (2010). A combined electrocoagulation–electrooxidation treatment for industrial wastewater. *Journal of Hazardous Materials*, 688-694.

Kleine, J., Peinemann, K.-V., & ark. (2002). Multifunctional system for treatment of wastewaters from adhesive-producing industries: separation of solids and oxidation of dissolved pollutants using doted microfiltration membranes. *Chemical Engineering Science*, 1664-1666.

Li, D., Xiao, Y., & ark. (2019). A metal-free protonated g-C3N4 as an effective sodium percarbonate activator at ambient pH conditions: Efficiency, stability and mechanism. *Materials Chemistry and Physics*, 225-232.

Lin, Z., Qin, W., & ark. (2020). Kinetics and mechanism of sulfate radical-and hydroxyl radical-induced degradation of Bisphenol A in VUV/UV/peroxymonosulfate system. *Journal of Water Process Engineering*.

Lu, J., Zhuo, Q., & ark. (2022). Treatment of wastewater from adhesive-producing industries by electrocoagulation and electrochemical oxidation. *Process Safety and Environmental Protection*, 527-536.

Maleki, A., & Zazouli, M. A. (2009). Composting Plant Leachate Treatmentby Coagulation-Flocculation Process. *American-Eurasian J. Agric. & Environ. Sci.*, 638-643.

Mandal, P., Dubey, B. k., & ark. (2017). Review on landfill leachate treatment by electrochemical oxidation: Drawbacks, challenges and future scope. *Waste Management*, 250-273.

Negash, A., Tibebe, D., & ark. (2023). A study of basic and reactive dyes removal from synthetic and industrial wastewater by electrocoagulation process. *South African Journal of Chemical Engineering*, 122-131.

Özyonar, F., & Karagözoğlu, B. (2012). Elektrokoagülasyon Prosesi ile Tekstil Sanayi Atıksuyunun Arıtımı. *Erciyes Üniversitesi Fen Bilimleri Enstitüsü Dergisi*, 29-37.

Posey, J., Cyr, P., & ark. (2014). Unconventional Bench And Pilot Approaches For Chemical Adhesive Wastewater Treatment. *WEFTEC.* New Orleans: Water Environment Federation.

DRAFT

Rivas, J., Gimeno, O., & ark. (2010). Influence of oxygen and free radicals promoters on the UV-254 nm photolysis of diclofenac. *Chemical Engineering Journal*, 35-40.

Saeed, O. F., Hameed, K. W., & ark. (2023). Treatment of vegetable oil refinery wastewater by sequential electrocoagulation-electrooxidation process. *Journal of Environmental Management*.

Sun, Z., Li, M., & ark. (2022). A review of the fluence determination methods for UV reactors: Ensuring the reliability of UV disinfection. *Chemosphere*.

Titchou, E. F., Zazou, H., & vd. (2021). An overview on the elimination of organic contaminants from aqueous systems using electrochemical advanced oxidation processes. *Journal of Water Process Engineering*.

Türk, O. K., Adalar, G., & ark. (2023). Photodegradation of oxytetracycline by UV-assisted persulfate and percarbonate processes: kinetics, influencing factors, anion effect, and radical species. *Environmental Science and Pollution Research*, 869-883.

Wu, S.-H., Chi, J.-H., & ark. (2010). Thermal hazard analyses and incompatible reaction evaluation of hydrogen peroxide by DSC. *Journal of Thermal Analysis and Calorimetry*, 563-568.

Yakamercan, E., Bhatt, P., & ark. (2023). Comprehensive understanding of electrochemical treatment systems combined with biological processes for wastewater remediation. *Environmental Pollution*.

Zhan, B.-T., Kuang, L., & ark. (2021). Application of percarbonate and peroxymonocarbonate in decontamination technologies. *Journal of Environmental Sciences*, 100-115.

Zhang, B.-T., Zhang, Y., & ark. (2015). Sulfate radical and its application in decontamination technologies. *Critical Reviews in Environmental Science and Technology*, 1756-1800.

Zhang, Q., Zhang, F., & ark. (2017). Structure–property correlations of reactive oxygen species-responsive and hydrogen peroxide-eliminating materials with anti-oxidant and anti-inflammatory activities. *Chemistry of Materials*, 8221-8238.

Zhang, X., Ren, B., & ark. (2017). Is sodium percarbonate a good choice in situ remediation of deltamethrin pollution?. *Frontiers of Environmental Science & Engineering*, 1-8.

DRAFT

ORAL PRESENTATION

Kardiyovasküler hastalıklarda yeni biyobelirteçler

Yahya ALTINKAYNAK[1*] (ORCID ID: 0000-0003-2060-4576), Buket AKCAN[2] (ORCID ID: 000000-0002-4516-6528)

1 Ardahan Üniversitesi, Sağlık Hizmetleri Meslek Yüksekokulu, Tıbbi Hizmetler ve Teknikler Bölümü, Ardahan, TÜRKİYE

2 Ardahan Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Ardahan, TÜRKİYE

*Sorumlu yazar e-posta: yahyaaltinkaynak@ardahan.edu.tr

Özet

Biyobelirteç; normal biyolojik süreçler, patolojik süreçler veya terapötik bir işleme karşı farmakolojik cevabın göstergesi olabilen, objektif olarak ölçülüp değerlendirilmesi yapılabilen özelliktir. Temel olarak biyobelirteçler hastalığın tanı ve takibinde kullanılır. Bu nedenle biyobelirteçler ucuz, kolay ulaşılabilir, tekrar edilebilir ve objektif olmalıdır.

Kardiyovasküler Hastalıklar dünya genelinde mortalite ve morbiditenin önde gelen nedenidir. Tanı tedavi ve prognozda önemli ilerlemeler kaydedilmiş olmakla birlikte özellikle teşhis için yani biyobelirteçlere ihtiyaç duyulmakta, yeni terapötik yöntemler için çalışmalar yapılmaktadır.

Kardiyovasküler Hastalık prevelansı yaşla birlikte artmaktadır. Bunun yanında sigara ve alkol kullanımı, fiziksel inaktivite, dengesiz beslenme ve obezite gibi faktörler de kardiyavasküler hastalık riskini artırmaktadır. Kardiyovasküler Hastalıklar için standardize ve valide edilmiş kardiyovasküler risk skorları bulunmaktadır. Bunlar temel olarak, yaş, cinsiyet, hipertansiyon ve diabetis mellitus varlığı ile total kolesterol, düşük ve yüksek yoğunluklu kolesterol (LDL-K, HDL-K) değerleridir.

Kardiyovasküler Hastalıklar için yeni biyobelirteçler arasında; myokard hasarı için Kardiyak Troponinler, Kalp-tipi yağ asidi bağlayıcı protein ve Kardiyak myosin bağlayıcı protein-C, myokard stresi için A ve B tip Natriüretik peptidler, Orta bölgesel pro-adrenomedullin ve Kopeptin, inflamasyon için C-reaktif protein, İnterlökin-6, Büyüme farklılaşma faktörü 15, Tümörijenisitenin çözünebilir baskılanması 2 ve Galektin-3, platelet aktivasyonu için çözünebilir CD40 ligandı ve P-selektin, plak değişkenliği için Lipoprotein ilişkili fosfolipaz A2 ve Matriks Metaloproteinaz-9, sistemik stres için Katekolaminler ve Granin proteinleri kalsiyum homestazı için Sekretonörin sayılabilir. Bunun yanında son çalışmalarda kardiyavasküler hastalıklar için teşhis ve tedavide kullanılmak üzere potansiyel biyobelirteç olarak mikro RNA'ların da kullanılabileceği ortaya konmuştur.

Anahtar Kelimeler: Kardiyovasküler Hastalıklar, Biyobelirteç, Teşhis, Tedavi, Prognoz

Novel biomarkers in cardiovascular diseases

Abstract

A biomarker is a property that can be objectively measured and evaluated, which can be an indicator of normal biological processes, pathological processes, or a pharmacological response to a therapeutic procedure. Basically, biomarkers are used in the diagnosis and follow-up of the disease. Therefore, biomarkers should be inexpensive, easily accessible, reproducible and objective.

Cardiovascular Diseases are the leading cause of mortality and morbidity worldwide. Although significant progress has been made in diagnosis, treatment and prognosis, biomarkers are needed especially for diagnosis, and studies are carried out for new therapeutic methods.

The prevalence of cardiovascular disease increases with age. In addition, factors such as smoking and alcohol use, physical inactivity, unbalanced diet and obesity increase the risk of cardiovascular disease. There are standardized and validated cardiovascular risk scores for Cardiovascular Diseases. These are basically age, gender, presence of hypertension and diabetes mellitus, and total cholesterol, low- and high-density cholesterol (LDL-C, HDL-C) values.

Among the new biomarkers for Cardiovascular Diseases; Cardiac Troponins, Heart-type fatty acid-binding protein and Cardiac myosin-binding protein-C for myocardial injury, A and B-type Natriuretic peptides for myocardial stress, Mid-regional pro-adrenomedullin and Copeptin, C-reactive protein for inflammation, Interleukin-6, Growth differentiation factor 15, Soluble suppression of tumorigenicity 2 and Galectin-3, soluble CD40 ligand and P-selectin for platelet

activation, Lipoprotein-associated phospholipase A2 and Matrix Metalloproteinase-9 for plaque instability, Catecholamines for systemic stress, and Granin proteins Secretoneurin for calcium homestasis. In addition, recent studies have shown that microRNAs can be used as potential biomarkers for use in the diagnosis and treatment of cardiovascular diseases.

Key words: Cardiovascular Diseases, Biomarkers, Diagnosis, Treatment, Prognosis

Giriş

Biyobelirteç; normal biyolojik süreçler, patolojik süreçler veya terapötik bir işleme karşı farmakolojik cevabın göstergesi olabilen, objektif olarak ölçülüp değerlendirilmesi yapılabilen özelliktir. Temel olarak biyobelirteçler hastalığın tanı ve takibinde kullanılır. Bu nedenle biyobelirteçler ucuz, kolay ulaşılabilir, tekrar edilebilir ve objektif olmalıdır (Aronson ve Ferner 2017; Lyngbakken ve ark. 2019)

Kardiyovasküler hastalıklar (KVH) dünya genelinde mortalite ve morbiditenin önde gelen nedenidir (Zhou ve ark. 2018). KVH'lar kalp ve damar bozuklukları ile karakterize olan ve kanın kalbe, beyine ve vücudun periferal bölgelerine akışını etkileyen bir grup hastalıktır. KVH' lar içerisinde koroner kalp hastalığı, serebrovasküler hastalık, romatizmal ve doğumsal kalp hastalıkları yer almaktadır (Hijová 2023).

Kardiyovasküler Hastalıklar

KVH'lar dünya genelinde prematüre ölüm nedenlerinin başında gelmektedir (Van Camp 2014). KVH prevelansı yaş ve bunun yanında tütün kullanımı, alkol tüketimi, obezite ve fiziksel inaktivite gibi modifiye edilebilir faktörler ile artmaktadır (Lyngbakken ve ark. 2019).

KVH subklinik ateroskleroz gelişimi ile karakterize bir yaşam boyu hastalıktır (Santhakumar, Battino, ve Alvarez-Suarez 2018) İnflamasyon aterosklerozun başlangıcı ve gelişiminde son derece önemli bir rol oynar. Aterosklerotik kardiyovasküler hastalıklar kronik inflamatuar hastalıklar arasında yer almaktadır (Irwandi ve ark. 2022).

KVH'lara sebep olan faktörler önlenebilir ve önlenemez olmak üzere iki gruba ayrılır. Yaş, cinsiyet ve genetik yatkınlık gibi faktörler değiştirilemeyen etkenler olduğu için önlenebilir değildir. Bunun yanında değiştirilebilen etkenler olan yaşam tarzı, beslenme alışkanlıkları, fiziksel aktivite, kan glukoz ve lipid düzeyleri ise önlenebilir faktörlerdir (Hijová 2023). Dengeli beslenmenin KVH riskinin önlenmesinde önemli rol oynadığı yapılan çalışmalar ile ortaya konulmuştur (Anon 2013; Anon n.d.; Llorente-Cortés ve ark. 2010; De Lorgeril ve ark. 1999; Widmer ve ark. 2015)

Kardiyovasküler Hastalıklarda Kullanılan Biyobelirteçler

Yapılan çalışmalara göre birçok biyobelirteç artmış KVH riski ile ilişkilendirilmiştir. Bu biyobelirteçler arasında C Reaktif protein (CRP), B tip natriüretik peptid (BNP), Lipoprotein (a), Homosistein ve fibrinojen bulunmaktadır (Mahmood ve ark. 2014)

Kardiyovasküler biyobelirteçler yaygın olarak patofizyolojik süreçlere göre sınıflandırılır. Buna göre kardiyovasküler patoloji grupları; miyokard hasarı, miyokardiyal stres, inflamasyon, trombosit aktivasyonu, plak instabilitesi, sistemik stres ve kalsiyum homeostazisi olarak sıralanabilir. Bu gruplara göre kullanılan biyobelirteçler ise; miyokard hasarı için Kardiyak Troponinler, Kalp-tipi yağ asidi bağlayıcı protein ve Kardiyak miyosin bağlayıcı protein-C, miyokard stresi için A ve B tip Natriüretik peptidler, Orta bölgesel pro-adrenomedullin ve Kopeptin, inflamasyon için C-reaktif protein, İnterlökin-6, Büyüme farklılaşma faktörü 15, Tümörijenisitenin çözünebilir baskılanması 2 ve Galektin-3, platelet aktivasyonu için çözünebilir CD40 ligandı ve P-selektin, plak değişkenliği için Lipoprotein ilişkili fosfolipaz A2 ve Matriks Metaloproteinaz-9, sistemik stres için Katekolaminler ve Granin proteinleri kalsiyum homestazı için Sekretonörin şeklindedir (Lyngbakken ve ark. 2019).

Bunların dışında son yıllarda yapılan çalışmalarda kardiyavasküler hastalıklar için teşhis ve tedavide kullanılmak üzere potansiyel biyobelirteç olarak mikro RNA'ların da kullanılabileceği ortaya konmuştur (Ultimo ve ark. 2018; Zhou ve ark. 2018).

Aterosklerotik kardiyovasküler hastalık gelişiminin başlangıç basmağı olan endotel disfonksiyonu için de bazı biyobelirteçler bulunmaktadır (Zhang 2022). Bu biyobelirteçler özellikle kardiyovasküler hastalık gelişim riski ve yeni tedavi hedefleri açısından önem arz etmektedir (Szmitko ve ark. 2003). Kronik inflamatuar bir süreç olan aterosklerozun başlangıç basmağı ile ilişkili olan bu biyobelirteçler Tablo 1 de sunulmuştur.

Tablo 1. Endotel disfonksiyonu için biyobelirteçler (Szmitko ve ark. 2003).

Biyobelirteç	Alternatif isim	Aile-Süper aile	Major Hücresel Kaynak	Potansiyel Kullanım Alanı
E-Selektin	Endotelyal lökosit adezyon molekülü-1, CD62E	Selektin	Aktive endotel	Endotel aktivasyonu için spesifik ve sentitif belirteç
P-Selektin	CD62P, GMP140, PADKEM	Selektin	Aktive endotel	Endotel aktivasyonu için sensitif belirteç
ICAM-1	Hücreiçi adezyon molekülü-1, CD54	Ig G gen super ailesi	Aktive endotel	Endotel aktivasyonu için sensitif ve güvenilir belirteç
VCAM-1	Vasküler hücre adezyon molekülü-1, CD106	Ig G gen super ailesi	Aktive endotel	Endotel aktivasyonu için sensitif ve güvenilir belirteç
IL-6	İnterlökin-6	Sitokin ailesi	Aktive endotel	Spesifik olmayan, Endotel aktivasyonu için sensitif ve güvenilir belirteç
CRP	C reaktif protein	Akut faz proteini	Aktive endotel	Spesifik olmayan, Endotel aktivasyonu için sensitif ve güvenilir belirteç
LOX-1	Lektin benzeri okside LDL reseptörü-1	Okside LDL için A reseptörü	Aktive endotel	Spesifik olmayan, endotel aktivasyonu için gelecek vaad eden yeni belirteç
CD40L	CD Ligand, CD154, TNF ilişkili aktivasyon proteini	TNF süperailesi	Aktive endotel	Spesifik olmayan, endotel aktivasyonu için gelecek vaad eden yeni belirteç
ADMA	Asimetrik dimetil arjinin	Endojen NOS ailesi	Aktive endotel	Spesifik olmayan, endotel aktivasyonu için gelecek vaad eden yeni belirteç
Endocan	Endotelyal hücre spesifik molekül-1	Proteoglikan	Aktive endotel	Endotel aktivasyonu için gelecek vaad eden yeni belirteç
EMPler	$CD62E^{+}$ Endotelyal Mikropartiküller		Aktive endotel	Endotel aktivasyonu için gelecek vaad eden yeni belirteç

Kardiyak Troponinler

Troponin bir kas protein kompleksi olup çizgili kaslarda bulunur. Troponin T, Troponin I ve Troponin C olmak üzere üç alt tipi vardır (Lyngbakken ve ark. 2019). Troponinlerin tüm izoformları hem kalp kası hem de iskelet kasında bulunmakla beraber kardiyak Troponin (cTnt) ve kardiyak Troponin I (cTnt I) miyokarda spesifiktir (Mohammed ve Januzzi 2010).

DRAFT

Kalp Tipi Yağ Asiti Bağlayıcı Protein

Yağ asiti bağlayıcı proteinler yağ asitlerinin membrane boyunca taşınmasından sorumlu olan protein ailesidir. Karaciğer, ince bağırsak, adipositler, çevresel sinirler, beyin ve kalp olmak üzere pek çok doku izoformu bulunmaktadır (Lyngbakken ve ark. 2019). Kalp tipi yağ asiti bağlayıcı protein aynı zamanda FABP 3 olarak da bilinmekte olup iskelet kası ile kalbin baskın izoformudur (Otaki, Watanabe, ve Kubota 2017). Kalp tipi yağ asiti bağlayıcı protein miyokard hasarı göstergesi olarak akut miyokard iskemi sonrası dolaşımda görülmektedir (Lyngbakken ve ark. 2019; Otaki ve ark. 2017).

A ve B Tip Natriüretik Peptidler

Natriüretik Peptidler; Atriyal ya da A tip, Beyin ya da B tip ve C tip olmak üzere üç çeşittir. Natriüretik peptidler ağırlıklı olarak atriyum ve ventriküllerde yapılır ve böbrekten sodyum atılımından sorumludurlar. Natriüretik peptidlerin konsantrasyonları yaşla, böbrek fonksiyonlarının azalmasıyla ve atriyal fibrilasyonun varlığıyla birlikte artar. A tip natriüretik peptid konsantrasyonları ile akut kalp yetmezliği arasında güçlü bir ilişkili bulunmuştur (Lyngbakken ve ark. 2019).

Mikro RNA ve Kardiyovasküler Hastalıklar

MikroRNA' lar, (miRNA) kısa (yaklaşık 22 nükleotid), kodlanmayan tek sarmallı RNA molekülleridir (Zhou ve ark. 2018). MiRNA'lar çeşitli fizyolojik süreçlerin düzenleyicileri olup aynı zamanda da Kardiyovasküler Hastalıklar, Obezite, Tip 2 Diyabet ve Kanser gibi patolojik durumlar ile de ilişkilidirler.

Yapılan çalışmalarda miRNA'lar ile hipertansiyon, kalp yetmezliği, aortik anevrizma, aritmiler, akut miyakord infarktı, konjenital kalp hastalıkları, vasküler hastalıklar ve aterosklerozun ilişkili olduğu gösterilmiştir (Çakmak ve Demir 2020). MiRNA' lar farklı sinyal yollarını kullanarak birçok geni etkileyen yeni terapotik ajanlar ve biyobelirteçler olarak sınıflandırılmaktadır.

Sonuç

Dünya genelinde ölüm nedenlerinin başında gelen kardiyovasküler hastalıklar için yeni teşhis ve tedavi yöntemlerinin bulunması son derece önemlidir. Klasik risklerden bağımsız olarak kardiyovasküler olaylarla ilişkilendirilebilen yeni biyobelirteçler kardiyovasküler risk belirleme ve tedavi geliştirme açısından yararlı olabilecektir.

Kaynaklar

Anon. 2013. "Primary Prevention of Cardiovascular Disease with a Mediterranean Diet." *Zeitschrift Fur Gefassmedizin* 10(2):28.

Anon. n.d. *Mediterranean Alpha-Linolenic Acid-Rich Diet in Secondary Prevention of Coronary Heart Disease Summary*.

Aronson, Jeffrey K., and Robin E. Ferner. 2017. "Biomarkers—a General Review." *Current Protocols in Pharmacology* 2017:9.23.1-9.23.17.

Çakmak, Hüseyin Altuğ, and Mehmet Demir. 2020. "Microrna and Cardiovascular Diseases." *Balkan Medical Journal* 37(2):60–71. doi: 10.4274/balkanmedj.galenos.2020.2020.1.94.

Van Camp, G. 2014. "Cardiovascular Disease Prevention." *Acta Clinica Belgica: International Journal of Clinical and Laboratory Medicine* 69(6):407–11.

Hijová, Emília. 2023. "Benefits of Biotics for Cardiovascular Diseases." *International Journal of Molecular Sciences* 24(7).

Irwandi, Rizky A., Scott T. Chiesa, George Hajishengallis, Venizelos Papayannopoulos, John E. Deanfield, and Francesco D'Aiuto. 2022. "The Roles of Neutrophils Linking Periodontitis and Atherosclerotic Cardiovascular Diseases." *Frontiers in Immunology* 13.

Llorente-Cortés, Vicenta, Ramón Estruch, Mari Pau Mena, Emilio Ros, Miguel Angel Martínez González, Montserrat Fitó, Rosa María Lamuela-Raventós, and Lina Badimon. 2010. "Effect of Mediterranean Diet on the Expression of Pro-Atherogenic Genes in a Population at High Cardiovascular Risk." *Atherosclerosis* 208(2):442–50. doi: 10.1016/j.atherosclerosis.2009.08.004.

De Lorgeril, Michel, Patricia Salen, Jean-Louis Martin, Isabelle Monjaud, Jacques Delaye, and Nicole Mamelle. 1999. *Mediterranean Diet, Traditional Risk Factors, and the Rate of Cardiovascular Complications After Myocardial Infarction Final Report of the Lyon Diet Heart Study*.

Lyngbakken, Magnus Nakrem, Peder Langeland Myhre, Helge Røsjø, and Torbjørn Omland. 2019. "Novel Biomarkers of Cardiovascular Disease: Applications in Clinical Practice." *Critical Reviews in Clinical Laboratory Sciences* 56(1):33–60.

Mahmood, Syed S., Daniel Levy, Ramachandran S. Vasan, and Thomas J. Wang. 2014. "The Framingham Heart Study and the Epidemiology of Cardiovascular Disease: A Historical Perspective." *The Lancet* 383(9921):999–1008.

Mohammed, Asim A., and James L. Januzzi. 2010. "Clinical Applications of Highly Sensitive Troponin Assays." *Cardiology in Review* 18(1):12–19.

Otaki, Yoichiro, Tetsu Watanabe, and Isao Kubota. 2017. "Heart-Type Fatty Acid-Binding Protein in Cardiovascular Disease: A Systemic Review." *Clinica Chimica Acta* 474:44–53.

Santhakumar, Abishek B., Maurizio Battino, and José M. Alvarez-Suarez. 2018. "Dietary Polyphenols: Structures, Bioavailability and Protective Effects against Atherosclerosis." *Food and Chemical Toxicology* 113:49–65.

Szmitko, Paul E., Chao Hung Wang, Richard D. Weisel, John R. De Almeida, Todd J. Anderson, and Subodh Verma. 2003. "New Markers of Inflammation and Endothelial Cell Activation Part I." *Circulation* 108(16):1917–23.

Ultimo, Simona, Giorgio Zauli, Alberto M. Martelli, Marco Vitale, James A. Mccubrey, Silvano Capitani, and Luca M. Neri. 2018. *Cardiovascular Disease-Related MiRNAs Expression: Potential Role as Biomarkers and Effects of Training Exercise*. Vol. 9.

Widmer, R. Jay, Andreas J. Flammer, Lilach O. Lerman, and Amir Lerman. 2015. "The Mediterranean Diet, Its Components, and Cardiovascular Disease." *American Journal of Medicine* 128(3):229–38.

Zhang, Jun. 2022. "Biomarkers of Endothelial Activation and Dysfunction in Cardiovascular Diseases." *Reviews in Cardiovascular Medicine* 23(2).

Zhou, Shan Shan, Jing Peng Jin, Ji Qun Wang, Zhi Guo Zhang, Jonathan H. Freedman, Yang Zheng, and Lu Cai. 2018. "MiRNAS in Cardiovascular Diseases: Potential Biomarkers, Therapeutic Targets and Challenges Review-Article." *Acta Pharmacologica Sinica* 39(7):1073–84.

DRAFT

ORAL PRESENTATION

Su ve Atıksulardan Kirletici Gideriminde Uygulanan Üç Boyutlu Elektrokimyasal Proseslerde Kullanılan Parçacık/Partiküler Elektrot Türlerinin İncelenmesi

Ezgi Unal Yilmaz[1*] (*ORCID: https://orcid.org/0000-0003-4178-0990*),
Senem Yazici Guvenc[1] (*ORCID: https://orcid.org/0000-0002-2877-0977),*

*Sorumlu yazar e-mail: ezgi.yilmaz3@std.yildiz.edu.tr

Özet

Konvansiyonel atıksu arıtma tekniklerine kıyasla proses maliyetlerinin daha yüksek olması sebebiyle elektrokimyasal prosesler özellikle biyolojik olarak ayrışamayan, toksik içeriği yüksek, dirençli ve kalıcı kirleticilerin su ve atıksu ortamından uzaklaştırılmasında kullanılmaktadır. Bu prosesler uygulama kolaylığı, alan ihtiyacının düşüklüğü, ikincil kirlilik oluşumunu engellemesi, yüksek giderim verimi potansiyeli ve dirençli ve kalıcı kirleticilere cevap verebilmesi gibi avantajları ile pek çok araştırmada tercih edilmektedir. Son yıllarda kullanılan iki boyutlu elektrokimyasal sistemlere anot-katot çiftinin yanı sıra parçacık/partiküler elektrot gibi üçüncü bir elektrotun eklenmesi ile sistem üç boyutlu (3D) elektrokimyasal sisteme modifiye edilmekte ve böylece su ortamında daha yüksek iletkenlik, daha fazla reaktif alan ve düşük voltajlarda bile daha yüksek akım yoğunluğu gibi sistem performansını iyileştiren kolaylıklar sağlanbilmektedir. 3D elektrokimyasal proses performansının artması kullanılan parçacık/partiküler elektrotun tipi, cinsi, boyutu ve kimyasına göre değişim göstermektedir. 3D elektrokimyasal proseslerde kullanılan parçacıkların yüksek spesifik yüzey alanı içermesi kirleticilerin giderim mekanizması olarak adsorpsiyon prosesinin de etkin olduğunu göstermektedir. Geniş yüzey alanı ve gözenekli yapıları sayesinde adsorpsiyon kapasitesi yüksek olan granüler aktif karbon yaygın olarak kullanılan parçacık elektrotların başında gelmektedir. Buna ilaveten, yapılan çalışmalar metal oksitler, kaplanmış karbon malzemeler, arıtma çamuru ve cüruf gibi parçacıkların da bu sistemlerde kullanımının mümkün olduğunu göstermektedir. Üç boyutlu elektrokimyasal sistemler, iki boyutlu sistemlere kıyasla daha yüksek kirletici giderimi sağlarken daha düşük enerji ve proses maliyetleri gerektirmektedir. Bu çalışma ile su ve atıksu ortamından üç boyutlu elektrokimyasal sistemler kullanılarak farklı kirletici türlerin gideriminde etkin rol alan parçacık/partiküler elektrot türleri incelenmiştir. Ayrıca, bu farklı türdeki parçacık elektrotların hem fiziksel ve kimyasal özellikleri hem de kullanıldığı işlemlerin maliyetleri de belirlenmiştir.

Anahtar Kelimeler: Üç boyutlu elektrokimyasal proses, su ve atıksu arıtımı, parçacık elektrodu

Examination of Particulate Electrode Types Used in Three-Dimensional Electrochemical Processes Applied in Pollutant Removal from Water and Wastewater

Abstract

Electrochemical processes are especially used to remove non-biodegradable, high toxic, resistant and persistent pollutants from water and wastewater environment due to the higher process costs compared to conventional water and wastewater treatment techniques. These processes are preferred in many studies with their advantages such as ease of application, low space requirement, prevention of secondary pollution, high removal efficiency potential and ability to respond to resistant and persistent pollutants.
By adding a third electrode, such as a particle electrode, to the two-dimensional electrochemical systems used in recent years, the system is modified into a three-dimensional electrochemical system, and thus, facilities that improve system performance such as higher conductivity in the aquatic environment, more reactive area and higher current density even at low voltages can be provided. The increase in 3D electrochemical process performance depends on the type, size and chemistry of the particle electrode. The high specific surface area of the particles used in 3D electrochemical processes shows that the adsorption process is also effective as a mechanism for removing pollutants. Granular activated carbon, which has a high adsorption capacity thanks to its large surface area and porous structure, is one of the most commonly used particle electrodes. In addition, studies show that it is possible to use particles such as metal oxides, coated carbon materials, sewage sludge and slag in these systems. 3D electrochemical systems provide higher pollutant removal compared to two-dimensional systems and require lower energy and process costs. In this study, particle electrode types that play an active role in the removal of different types of pollutants from the water and wastewater environment

were investigated using three-dimensional electrochemical systems. In addition, both the physical and chemical properties of these different types of particle electrodes and the costs of the processes in which they are used have been determined.

Keywords: Three-dimensional electrochemical process, water and wastewater treatment, particle electrode

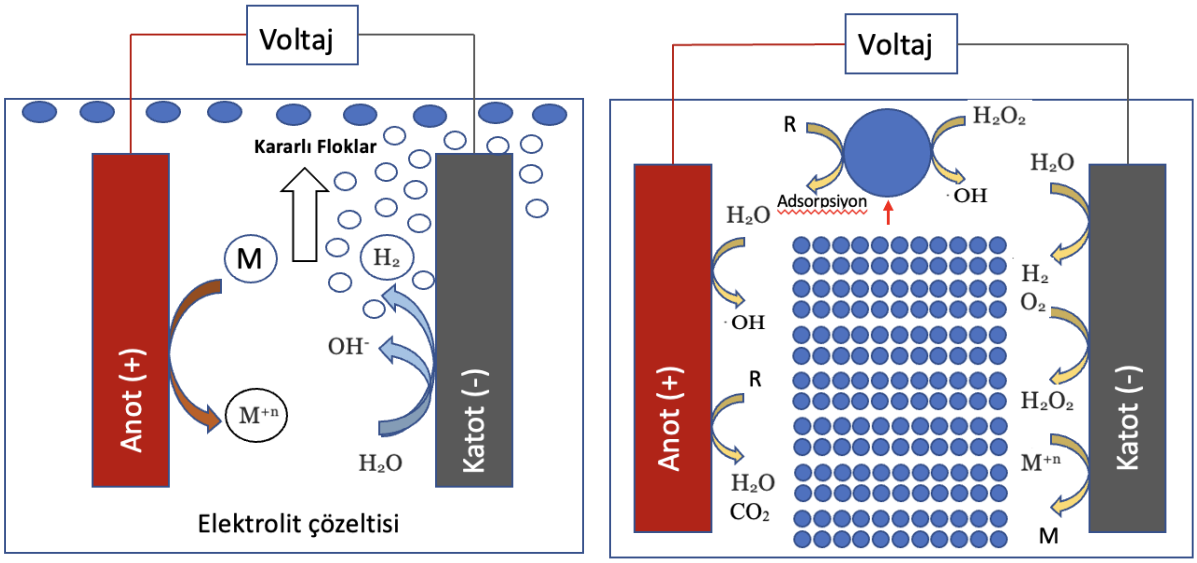
GİRİŞ

Elektrokimyasal arıtım elektrokimyasal hücre olarak adlandırılan bir yapı içerisinde gerçekleştirilir. Elektrokimyasal hücre ise iki farklı elektrotun iletken bir ortamda oluşturduğu sistemler olarak tanımlanabilir. Bu hücrelerde elektrik akımı sayesinde kimyasal reaksiyonların gerçekleşmesi sağlanır (Kaygusuz, 2013). Hücre içerisinde indirgenme ve yükseltgenme reaksiyonları gerçekleşirken, indirgenme gerçekleşen elektrota katot, yükseltgenme gerçekleşen elektrota anot denir. Anot ve katot çiftinin tek başına kullanıldığı bu sistemler iki boyutlu (2D) sistemler olarak adlandırılırlar. 2D sistemler elektrotlar ile elektrolitik çözelti arasında meydana gelen elektron transferi esasına dayanmaktadır. Elektron transferi elektrokimyasal teknoloji için uygun spesifik materyallerden yapılan anot ve katot arasında elektriksel alan uygulanarak elde edilmektedir. Uygulanan akım/potansiyel farkı, elektrokimyasal hücre içinde yer alan çözeltide, oksidasyon-redüksiyon reaksiyonlarını başlatmak için gerekli itici gücü sağlamaktadır (Fernandes ve ark., 2015). Anot ve katot olarak genellikle metal, karbon veya yarı iletken özellikte elektrotlar kullanılır. Anot ve katotta meydana gelen redoks reaksiyonları Tablo 1'de verilmiştir. Evsel atıksuların arıtımının yanı sıra tekstil atıksuları, mezbaha atıksuları, sızıntı suları, deterjan atıksuları ve kağıt sanayi atıksuları gibi sanayi atıksularının arıtımınında da elektrokimyasal prosesler kullanılmaktadır. Organik madde, ağır metal, askıda katı madde (AKM), renk, nitrat ve fenol gibi kirleticilerin gideriminde oldukça etkili yöntemlerdir (İlhan ve ark., 2007). Arıtımın genel mekanizması ise adsorpsiyon, koagülasyon, absorpsiyon, çöktürme ve flotasyondur.

Tablo 1. Elektrokimyasal proseslerde gerçekleşen anot ve katot reaksiyonları (Sivri, 2020)

Anot Reaksiyonları	**Katot Reaksiyonları**
e^- vererek yükseltgenir	e^- alarak indirgenir
Anotta çözünme $(Al \rightarrow Al^{+3} + 3e^-)$	Katotta birikme $(Cu^{+2} + 2e^- \rightarrow Cu)$
Anotta oksijen çıkışı $2H_2O\text{-}4e^- \rightarrow O_2(g)\uparrow + 4H^+$	Katotta hidrojen çıkışı $2H_2O + 2e^- \rightarrow H_2(g)\uparrow + 2OH^-$
Anotta klor olması durumu $2Cl^- + 2e^- \rightarrow Cl_2\uparrow$	Gaz indirgeme durumu $O_2 + 4H^+ + 4e^- \rightarrow 2H_2O$

2D sistemine eklenen parçacık/partiküllerin elektrik alanı içerisinde elektrot görevi görmesiyle beraber üç boyutlu (3D) sistem oluşturulmaktadır. Şekil 1'de 2D ve 3D sistemlerin genel mekanizması gösterilmiştir.



Şekil 1. 2D ve 3D sistem genel mekanizması

Reaktöre eklenen parçacıklar elektrik alanı içerisinde bulunduğundan elektrostatik indüksiyon yoluyla yüklü hale gelir. Bu yüklü parçacıklar sistemde bağımsız 3 boyutlu elektrotlar haline gelir ve bir tarafı pozitif yüklü iken diğer tarafı negatif yüklü olacaktır (Foroughi ve ark., 2018; Zhang ve ark., 2013; Zhu ve ark., 2011). Her bir yüklü parçacık "mikro elektroliz hücresi" oluşturacağından elektrokimyasal reaksiyonun gerçekleştiği alan yüksek miktarda artacaktır (Li ve ark., 2021a). 2D elektrokimyasal sistemlerde anotta gerçekleşen oksidasyona ek olarak parçacık üzerinde de doğrudan ve dolaylı oksidasyon mekanizması gerçekleşir. Sistemde hidroksi radikali gibi aktif ürünlerin oluştuğu da gözlenmiştir. Ayrıca bu parçacıkların yüksek adsorpsiyon kapasitesi sayesinde kirleticilerin gideriminde adsorpsiyon mekanizmasının da etkili olduğu söylenebilir. Oluşan elektriksel alanın artması ve adsorpsiyon kapasitesi sayesinde, 2D sisteme kıyasla daha düşük voltajlarda daha yüksek giderim veriminin elde edilir ve neticesinde daha düşük işletim maliyeti sağlanmış olur.

Yapılan literatür çalışmalarında görülmüştür ki toksik içeriği yüksek, biyolojik olarak arıtılması zor ve dirençli kirleticilerin yoğun olduğu tekstil endüstrisi (Ren ve ark., 2023), kömür atıksuyu (Sun ve ark., 2021), ilaç endüstrisi (Zhan ve ark., 2019), petrol rafinerisi atıksuyu (Yan ve ark., 2016), çelik üretimi (Hu ve ark., 2022), kağıt endüstrisi (Chi ve ark., 2018), kimya ve gıda endüstrisi (Can ve ark., 2014) gibi kirletici özelliği yüksek atıksuların arıtılması için 3D sistemler çoğunlukla tercih edilmektedirler. Bu çalışmada, 3D elektokimyasal sistemlerde kullanılan farklı parçacık/partikül elektrot türleri ve kullanılan elektrotların fiziksel ve kimyasal özellikleri ve 3D elektrokimyasal proseslerin elektrot, enerji ve işletme maliyetleri araştırılmış ve parçacık/partikül elektrot kullanımının proses maliyeti üzerindeki etkileri belirlenmiştir.

2D Sistemlerde Kullanılan Elektrot Türleri

Elektrokimyasal proseslerde arıtma verimini etkileyen parametrelerin en önemlisi seçilen elektrot tipidir. Farklı elektrot tiplerinin kullanımında farklı reaksiyonlar gerçekleşmekte dolayısıyla giderim verimleri de farklı olmaktadır. Elektrokimyasal sistemlerde anot ve katot malzemesi olarak çoğunlukla çözünerek suya geçen ve koagülant özellikleri yüksek olan alimünyum ve demir elektrotlar kullanılmaktadır. Kolay ulaşılabilir olmaları, ucuz olmaları ve etkili olmalarından kaynaklı olarak elektrokimyasal proseslerde sıklıkla kullanıldıkları bilinmektedir (Zazou ve ark., 2019).

2D sistemlerde demir ve alüminyum gibi çözünen elektrotların yanında Ti, Ru, bor katkılı elmas gibi tükenmeyen elektrotlarda kullanılmaktadır. Genellikle kullanılan elektrotlar ise bor katkılı elmas, PbO2/Ti, PbO2/SnO2, PbO2, TiO2/TiRuO2, SnO2, Ti/Pt-Ir, Ti/PdO-CO3O4, Ti/RhOx-TiO2, Ti-SnO2/Pt elektrotlardır (Andrade ve ark., 2007; Cabeza ve ark., 2007). Özellikle bor katkılı elmas yüksek gerilim potansiyeline sahiptir ve geniş aralıklı voltajlarda çalışabilirler. Bu sayede yüzeylerinde daha fazla OH radikali adsorplayabilirler. Böylelikle de kirlilik daha çabuk okside olduğundan genellikle tercih edilen elektrot tipidir. Literatürde 2D sistemlerde kullanılan elektrot türlerinin özetleri Tablo 2'de verilmiştir.

Tablo 2. 2D Elektrokimyasal proseslerde elektrot tipleri ve verimleri

Sistem	Anot	Katot	Kirletici türü	Giderim verimi (%)	İşletme Maliyeti	Kaynak
EC	Al	Al	KOİ	76	-	(Asfaha ve ark., 2022)
EO	Fe	Karbon keçe	Escherichia coli	97.9	-	(Özyonar ve Korkmaz, 2022)
EO/O2	Al	Al	Arsenit	99.2	0.883 dolar/m^3	(Goren ve ark., 2020)
EO	Ti/RuO2	Ti	Toksik amonyum nitrojen	100	$2.7x10^{-5}$ kWh/mg NH_4	(Romano ve ark., 2020)
EO	Grafit	Grafit	Bromür	99	-	(Thangamani ve ark., 2022)
EC	Ti/Pt	Çelik	KOİ	80.5	210.7 kWh/m^3	(Fil ve Günaslan, 2022)
EO	Ti/IrO2/RuO2	Ti	KOİ	96.93	462.5 kWh/m^3	(Kul ve ark., 2022)
EO	Ti/Pt	Ti	Bulanıklık	99.18	210.7 kWh/m^3	(Fil ve Günaslan, 2023)
EC	Al	Al	KOİ	100	1.58 dolar/m^3	(Arslan ve ark. 2022)
EO	Bor katkılı elmas	Pt	KOİ	100	0.61 dolar/m^3	(Arslan ve ark. 2022)

3D SİSTEMLERDE KULLANILAN PARÇACIK/PARTİKÜL ELEKTROT TÜRLERİ

Parçacık/Partikül elektrotların seçimi 3D sistemlerin verimi etkileyen en önemli parametrelerden biridir. Parçacık/partikül elektrotların yeterli adsorpsiyon kapasitesi için spesifik yüzey alanının yüksek olması ve gözenekli yapıda olması istenir. Ayrıca, elektrot iletkenliğinin yüksek olması da sistem verimi etkilediğinden aranan bir diğer özelliktir. Literatürdeki yapılan 3D elektrokimyasal çalışmalarında kullanılan parçacık/partikül elektrot türleri satın alınanlar, sentezlenenler ve atıktan üretilenler üç farklı kategoride sınıflandırılmıştır. Yapılan çalışmalarda çoğunlukla ekonomik uygunluğu, kolaylıkla temini sebebiyle aktif karbonun sıklıkla parçacık/partikül elektrot olarak kullanıldığı görülmüştür. Buna ek olarak metal parçacıklar, metal oksit kaplanmış aktif karbon, doğal madeni taşlar, atıktan üretilmiş biochar, atık çamur, cüruf gibi malzemeler de parçacık/partikül elektrot olarak kullanılmaktadır.

Satın Alınanlar

Şekil 2'de satın alınan parçacık elektrotların türleri verilmiştir. Aktif karbon ulaşılabilirliği ve adsorpsiyon kapasitesinin yüksek olması nedeniyle en çok tercih edilen elektrot olmuştur. Foroughi ve ark. (2020), tatrasiklin giderimi için granül aktif karbonun (GAC) kullanıldığı 3D sistem kullanılmıştır. 3D sistemde tetrasiklin giderimi 20 dk sonunda %90 iken 2D sistemde verim %65.16 olmuştur. Yapılan bir diğer çalışmada (Ulucan-Altuntas ve ark., 2022) ise Oksitetrasiklin (OTC) gideriminde kullanılan GAC sonucunda 3D sistem 2D sisteme göre %21 daha iyi performans göstermiştir. Doğal madenler ve volkanik kayalarda geniş yüzey alanları, yüksek iletkenlik ve düşük maliyet gibi özellikleri nedeniyle tercih edilen diğer elektrotlardır. Volkanik kaya Norfloksasin gideriminde oldukça etkili sonuçlar vermiştir (Song ve ark., 2021). Doğal bir maden olan zeolit ise hem tek başına (Li ve ark., 2021b) hem de GAC ile birlikte (Feng ve ark., 2018) kullanılmaktadır. Li vd., (2021b)'nin yaptıkları çalışmada zeolitin GAC ile benzer performans gösterdiği görülmüştür. Piroluzit doğal kaynağı yüksek adsorpsiyon kapasitesi yanında içerdiği geçiş metalleriyle katalitik özelliği yüksektir. İletkenliğinin düşük olmasına çözüm olarak grafit toz ile beraber kullanılan çalışmada (Hong ve ark., 2018) %49,56 KOİ giderimi elde edilmiştir. Aynı çalışmada partikül elektrot olarak GAC kullanılması da araştırılmış ve verim %35 civarında kalmıştır.



Şekil 2. Doğrudan satın alınarak kullanılan parçacık/partikül elektrot türleri

Yüksek iletkenlik ve mükemmel kimyasal stabiliteye sahip aerojel elektrotlar 3D sistemlerde kullanılan diğer elektrotlardır. Grafen aerojellerin kullanıldığı sistemde Bisfenol A (BAS) kirleticisinin gideriminde %98 giderim verimi elde edilirken (Zhang ve ark., 2018), karbon aerojellerin kullanıldığı sistemde %90 üzerinde fenol giderimi gözlenmiştir. 3D elektro fenton (3D-EF) prosesinde kullanılan demir köpüğü hem sistemde parçacık elektot olarak görev görür hem de sistemin ihtiyacı olan demiri karşılar (Zheng ve ark., 2019). Folik asit gideriminde kullanılan 3D-EF ile %70.4 ile yüksek giderim verimi elde edilmiştir (Zheng ve ark., 2019). Titanyum köpük ve nikel köpüğün kullanıldığı bir diğer 3D-EF sisteminde ise 30 dakika gibi kısa bir sürede Rhodamine B (RhB) kirleticisinin %99 giderimi nikel köpük kullanımı sonucunda elde edilmiştir. Titanyum köpük ise 90 dakika sonunda %53 seviyesinde kalmıştır. Nikel köpüğün iletkenliğinin daha yüksek olması verimin bu derece yüksek olmasına etki ettiği yazarlar tarafından bildirilmiştir (Liu ve ark., 2012). Sıfır değerlikli demir düşük maliyetli olması, indirgeme potansiyelinin yüksek olması ve toksik olmaması gibi avantajlarıyla 3D sistemlerde kullanılan bir diğer parçacık türüdür. Ozon ile desteklenmiş 3D sistemde kullanılan sıfır değerlikli demir parçacıklar ile 120 dk sonunda %85.2 TOK giderimi elde edilirken 20 dk sonunda %90.5 nitrobenzen giderimi elde edilmiştir (Wang ve ark., 2021).

Sentezlenenler

Sentezlenerek elde edilen parçacık elektrotların özeti Şekil 3'de verilmiştir. Elektrokatalitik etkinin elde edilmesi veya arttırılması, yüzey alanının genişletilmesi ve kütle transferinin teşfik edilmesi amacıyla parçacık elektrotlarının üzerine metal veya metal oksitler kaplanmaktadır (Li ve ark., 2021a; Zhang ve ark., 2013). Yaygın olarak GAC, alüminyum oksit (Al2O3), doğal taşlar taşıyıcı olarak kullanılırlar. Katalizör olarak ise Zn, Mn, Sn, Ti, Cu, Ni, Ce, Zr ve Co kullanılmaktadır. Katalizörlerin düşük maliyetli olması ve uzun ömürlü olması tercih edilmelerinin sebeplerindendir. 3D sistemlerde en çok tercih edilen GAC tek başına kullanıldığı için birtakım katalizörlerin üzerine sentezlenmesi sonucunda da 3D sistemlerde sıklıkla kullanılmaktadır. Tekstil atıksularından sıklıkla görülen RhB kirleticisinin giderimi için Mn-Sn kaplanmış GAC parçacık elektrot olarak kullanılmıştır (Yang ve ark., 2023). %94.3 giderim verimi elde edilen sistemde katalizörlerin kullanılmadığı GAC tek başına olduğunda verim %80.7'ye düşmüştür. Pu ve ark., (2023)'nin yaptıkları diğer bir çalışmada SnO2-Sb2O3 kaplanmış GAC parçacık elektrodunun GAC'dan daha yüksek katalitik aktiviteye sahip olduğunu (GAC için 1.0 V iken SnO2-Sb2O3/GAC için 1.6 V) ve bunun sonucu olarak GAC kullanılan sisteme kıyasla Fenol gideriminin %31.68 ve KOİ gideriminin %42.26 daha fazla olduğunu görülmüştür (Pu ve ark., 2023). Al, Si ve diğer elementlerce zengin olan kaolin mükemmel elektriksel iletkenliğe ve stabiliteye sahiptir. Cu ve Fe elementlerinin kaolin üzerine sentezlenerek elde edildiği CuFeO/Kaolin parçacığı Orange G kirleticisinin giderilmesi için 3D-EF sisteminde kullanılmıştır (Zhou ve ark., 2023). CuFeO/Kaolin parçacığının kullanıldığı durumda giderim verimi %99.48 iken tek başına kaolin kullanılan sistemde verim %52.63 olmuştur. Kaolin parçacığına sentezlenen elementler ile katalitik aktif bölge arttırılmış ve daha fazla H2O2 ile OH radikali üretimi sağlanmıştır.

DRAFT



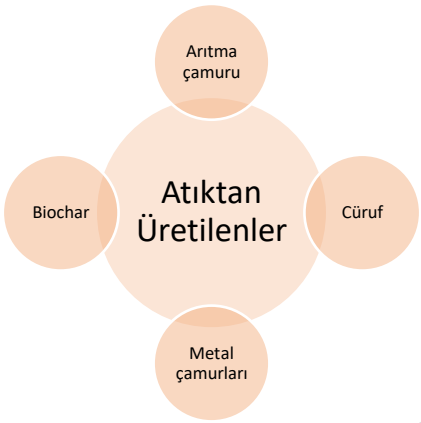
Şekil 3. Farklı malzemelerden sentezlenerek elde edilen parçacık/partikül elektrot türleri

Son yıllarda elektrokimyasal proseslerde enerji ihtiyacını azaltmak, kütle transferi teşvik etmek ve reaktif türlerin üretimini arttırmak için karbon nanotüplerin uygulaması giderek artmaktadır (Mengelizadeh ve ark., 2019). Diğer parçacık elektrotlarda da olduğu gibi nanotüpler yüksek iletkenlik, yüksek mekanik ve kimyasal dayanım ve geniş yüzey alanına sahiptirler. Fe3O4 kaplanmış bir karbon nanotüp'ün kullanıldığı 3D-EF prosesinde diklofenak giderimi 60 dk sonunda %95.6 olmuştur (Iranpour ve ark., 2018). Al2O3 ise geniş kanalları ve yüzey alanı sayesinde geçiş metallerinin sentezlenmesi için etkili bir taşıyıcıdır (Zhang ve ark., 2013). Co, Ce ve Zr elementlerinin Al2O3 üzerine sentezlenmesi sonucu elde edilen Co-Ce-Zr/γ-Al2O3 parçacığı bir çeşit antibiyotik olan Siprofloksasin giderimi için kullanılmıştır (Liu ve ark., 2021). Hazırlanan parçacıkla hedef kirleticinin giderimi boş Al2O3 parçacığa göre 2 kat daha yüksek olmuştur. TOK gideriminde ise %17.3 daha yüksek verim elde edilmiştir.

Atıktan Üretilenler

Atıktan üretilen parçcık elektrotların özet şeması Şekil 4'te verilmiştir. Son yıllarda döngüsel ekonomi kavramıyla birlikte ortaya çıkan bütünsel yaklaşım neticesinde cüruf, arıtma çamuru, biochar ve endüstriyel yan ürünlerin hammadde olarak kullanılması giderek ilgi odağı haline gelmiştir (Petala ve ark., 2022). Biochar, biyokütle kalıntılarının (arıtma çamuru, gıda atıkları, organik çöpler vb.,) oksijensiz ortamda ya da sınırlı oksijen altında düşük sıcaklıklarla yakılması sonucu elde edilir. Karbon açısından oldukça zengin bir bir malzeme olmasının yanında geniş yüzey alanına da sahiptir. Pirinç kabuğundan elde edilen biocharın kullanıldığı 3D sistemde Norfloksasin giderim verimi incelenmiştir. 120 dakika sonunda hedef kirleticide %87 giderim verimi elde edilirken TOK giderim verimi %90 olmuştur (Wang ve ark., 2023). Yapılan bir diğer çalışmada ise (Xie ve ark., 2021), şeker kamışından üretilen biochar üzerine Mn, Sn ve Sb katalizörleri ayrı ayrı kaplanmış ve 4-klorofenol giderim verimleri karşılaştırılmıştır. En iyi verim Mn kaplı parçacık ile %99,93 olurken verimlerim sıralaması Mn/AC > Sn/AC > Sb/AC şeklinde görülmüştür. Çelik, nikel, lityum ve manganez gibi metallerin cürufları da parçacık elektrot olarak kullanılan diğer türlerdir. Manganez cürufunun üzerine Cu-Fe katalizörlerinin kaplanarak kullanıldığı 3D-EF sisteminde Diklofenak giderimi araştırılmıştır (Long ve ark., 2019). İlave oksidan olarak Persülfatın da kullanıldığı sistemde %96.3 gibi yüksek bir verim elde edilebilmiştir. Boksit mineralinden alüminyum oksit üretimi için kullanılan bayer prosesinden kırmızı çamur olarak adlandırılan bir atık oluşmaktadır. Kırmızı çamur yüksek miktarda demir içeriğinin yanında silisyum ve titanyum gibi bileşikleri de içerisinde barındırır. Yüksek kimyasal stabiliteye ve yüksek yüzey morfolojisine sahip olmasının yanında elektron transferinin çok iyi sağlanamadığı düşük iletkenliğe ve yüksek elektrik direncine sahiptir. Bu nedenle katalizörler yardımıyla iletkenliği geliştirilerek kullanılması tercih edilmektedir (Lu ve ark., 2022). Lu ve ark., (2022) yaptıkları çalışmada kırmızı çamurun üzerine CuO katalizörünü kaplamış ve 3D-EF prosesinde Siprofloksasin giderimini incelemişlerdir. 80 dk sonunda 3D-EF sisteminde giderim %78.43 iken 2D sistemde verim %54.6'da kalmıştır. Evsel atıksu arıtma tesisinden elde edilen arıtma çamuru ve demir çamurunun sentezlenerek parçacık elektrot olarak kullanıldığı bir diğer çalışmada (Hou ve ark., 2017), N-heterosiklik bileşiklerden olan indol, kinolin ve piridin giderimi üzerine çalışmalar yürütülmüştür. 120 dk sonunda giderim verimleri sırasıyla %99.1, %96.7 ve %64.2 olmuştur. Arıtma çamurlarından elde edilen parçacık elektrotlarının 3D sistemler için iyi bir parçacık elektrot olduğu böylece görülmüştür.

DRAFT



Şekil 4. Çeşitli atıklardan üretilen parçacık/partikül elektrot türleri

Elektrokimyasal Proseslerde Elektrot Tipinin Enerji Verimliliği Üzerine Etkisi

3D sistemlerde anot ve katot olarak genellikle Ti, Ti/RuO2, Ti/IrO2, çelik gibi çözünmeyen elektrotlar tercih edilir. Bu nedenle arıtma işlemi sırasında sürekli bir maliyet oluşturmazlar. Parçacık elektrotlar ise yapılan çalışmalarda göstermiştir ki tekrar tekrar kullanıldıklarında bile yüksek verim elde edilmesine olanak sağlamaktadırlar (Gu ve ark., 2023; Miao ve ark., 2022; Ta ve ark., 2023). Bu özellikleri sayesinde sistemde maliyet oluşturan esas kalemlerden biri olmanın dışındadır. 3D sistemlerin asıl maliyeti ise sistemin işletme gücü olan elektrik maliyetidir. 2D sisteme kıyasla 3D sistemlerde katalitik kapasitenin ve enerji aktif bölgenin yüksek olması sonucu daha fazla serbest radikal oluşumu gözlenir. Yani daha düşük enerji ihtiyacıyla daha yüksek giderim verimi elde edilebilir. Miao ve ark., (2022) yaptıkları çalışmada 2D sistemde enerji verimliliğinin 2.02 kWh/m^3 atıksu iken nikel cürufunun kullanıldığı 3D sistemde 0.69 kWh/m^3 olduğunu bildirmişlerdir. Yapılan diğer bir çalışmada (Zhao ve ark., 2023), 3D sistemde enerji verimliliği 0.0077 kWh/gr KOİ iken 2D sistemde 0.0673 kWh/gr KOİ olduğu görülmüştür. Ulucan-Altuntaş ve ark., (2022) OTC gideriminde kullandıkları GAC sayesinde enerji verimliliğini 2.33 kWh/kg OTC'den 0.99 kWh/kg OTC'ye düşürmüşlerdir. Maliyette bu sayede 5.38 Euro/m^3 iken 2.81 Euro/m^3 olmuştur. Yapılan çalışmalar 2D sisteme kıyasla parçacık elektrotlarının kullanıldığı 3D sistemlerin enerji verimliliğinin daha yüksek olduğunu göstermiştir.

SONUÇ

Biyolojik olarak ayrışması zor, toksik içeriği yüksek ve dirençli kirleticilerin arıtılması için kullanılan elektrokimyasal proseslere olan ilginin artmasıyla sistemin dezavantajlarının azaltılması ile ilgili çalışmalar giderek artmıştır. 3D elektrokimyasal sistemlerde iyileştirme çalışmaları gün geçtikçe artmakta ve sistemin verimini pozitif yönde etkileyen yeni birçok parçacık/partikül elektrot literatüre kazandırılmaktadır. Bu çalışmaların bir sonucu olarak ortaya çıkan 3D elektrokimyasal prosesler ise 2D proseslere kıyasla daha az enerji ihtiyacı ile daha yüksek giderim verimi elde edilmesine olanak sağlamaktadır. 3D proseslerde kullanılan parçacık/partikül elektrotlarının türü ve yapısı 3D sistemleri etkileyen en önemli parametredir. Yapılan çalışmalar göstermiştir ki kullanılan partiküllerin geniş yüzey alanına sahip olması, yüksek iletkenlikte olması ve kimyasal ve fiziksel dayanımının yüksek olması 2D elektrokimyasal sistemlere kıyasla proses performansını iyileştirici etkiler ortaya koymaktadır. Yüzey alanı oldukça geniş olan aktif karbon en çok kullanılan parçacık olmakla birlikte iletkenliğinin arttırılması için üzerine bazı geçiş metallerinin sentezlenmesi sonucu daha iyi kirletici giderim verimlerinin elde edildiği görülmüştür. Elektrokimyasal sistemlerde en büyük maliyeti elektrik ihtiyacı oluşturmakla beraber, 3D sistemlerin düşük elektrik akımında bile yüksek kirletici giderim potansiyeli de bu maliyetin de giderek azaldığını göstermektedir. 2D elektrokimyasal sistemlerin bir ileri verisyonu olan 3D elektrokimyasal sistemlerin sağlamış olduğu bu avantajlar ile farklı karakterizasyonlara sahip endüstriyel atıksulardan spesifik ve dirençli kirleticilerin gideriminde uygulanabilir olduğu anlaşılmaltadır. Ayrıca, 3D elektrokimyasal sistemlerin çeşitli spesifik kirleticilerin su ve atıksulardan gideriminde etkin olduğu ve kompleks kirletici türleri içeren sızıntı suları gibi arıtımı birden fazla basamaktan oluşan ve yüksek maliyetli arıtma sistemlerinin kullanıldığı atıksu tiplerinde alternatif bir sistem olabileceği öngörülmektedir.

DRAFT

Sistem	Elektrotlar	Optimum şartlar	Atıksu	Kirleticiler	Verim (%)	Enerji Verimliliği		Kaynak
						2D sistem	3D sistem	
3D-EO	Anot: Ti/RuO2-IrO2 Katot: Çelik Sn–Mn–Ce/Aktif karbon	Elektrotlar arası mesafe 6 cm, pH: 6.6, voltaj: 6 V reaksiyon süresi: 90 dk	Sentetik	Fenol	74	58 kWh/kg KOİ	13.5 kWh/kg KOİ	(Jia ve ark., 2021)
				KOİ	75.6			
3D-EO/O3	Anot: Ti/Pt Katot: Ti/Pt Demir parçacıkları	Elektrotlar arası mesafe 3 cm, pH: 5.3, parçacık dozu: 1 gr/l, ozom konsantrasyonu: 5 mg/l, akım yoğunluğu: 11,43 mA/cm	Sentetik	Nitrobenzen	90.5	2.17 kWh/gr TOK	0.99 kWh/gr TOK	(Wang ve ark., 2021)
				TOK	85.2			
3D-EO/PS	Anot: Ti/RuO2 Katot: Çelik CuFe2O4/Al2O3	PH 4.95, parçacık dozu: 0.3 kg/l, PS/PNP dozu: 10, akım yoğunluğu: 24 mA/cm2	Sentetik	Fenolik bileşikler (PNP)	80.23	9.5 kWh/mg PNP	3.97 kWh/mg PNP	(Wu ve ark., 2021)
3D-EF	Anot: DSA Katot: Nikel köpük Demir yüklenmiş KOK kömürü	Elektrotlar arası mesafe 5 cm, voltaj: 11.15 V, pH: 2.62, parçacık dozu: 12.23 gr/l	Çelik üretimi atıksuyu	Toplam nitrojen	84.8	–	0.069 kWh/ mg KOİ	(Hu ve ark., 2022)
				KOİ	87			
3D-EO	Anot:Ti/RuO2 Katot: Ti Aktif karbon	Elektrotlar arası mesafe: 5 cm, pH: 4.3, parçacık dozu: 0,75 gr/l, akım yoğunluğu: 10 mA/cm2, reaksiyon süresi: 100 dk	Sentetik	OTC	99	2.33 kWh/kg OTC	0.99 kWh/kg OTC	(Ulucan-Altuntas ve ark., 2022)
3D-EO/PMS	Anot: Pt/Ti Katot: Pt/Ti Biochar	Elektrotlar arası mesafe 2,5 cm, pH: 7.2, parçacık dozu: 5 gr/l, PMS dozu: 10 mM, akım yoğunluğu: 18 mA/cm2, reaksiyon süresi 120 dk	Sentetik	Norfloksasin	87	90.2 kWh/gr TOK	29.7 kWh/gr TOK	(Wang ve ark., 2023)
				TOK	90			
3D-EO/PMS	Anot: Ti/Pt Katot: Ti/Pt Mn/Cu katkılı aktif karbon	pH: 7.3, PMS dozu: 10 mM, parçacık dozu: 1 gr/l, akım yoğunluğu: 25 mA/cm2, reaksiyon süresi: 60 dk	Sentetik	Norfloksasin	90.81	–	78.91 kWh/gr Norfloksasin	(Ta ve ark., 2023)
				TOK	85.12			
3D-EF	Anot: Ti/RuO2-Ir02 Katot: Ti karbon siyahı-politetrafloroetilen kompozit parçacıklar	Elektrotlar arası mesafe 2 cm, parçaık dozu: 4 gr, akım yoğunluğu: 30 mA/cm2, %10 cam küre ilavesi, reaksiyon süresi: 120 dk reaksiyon süresi, Fe+2 dozu: 0,2 mM	Sentetik	Fenol	100	–	0.080 kWh/gr KOİ	(Xiao ve ark., 2023)
				KOİ	80			
3D-EO	Anot: Ti/RuO2-IrO2 Katot: Ti plaka Mn/Sn/Aktif karbon	Elektrotlar arası mesafe: 2 cm, voltaj: 9.45 V, parçacık dozu: 32.37 gr/l, reaksiyon süresi: 60 dk	Sentetik	Rhodamine B	98.3	–	20.1 kWh/ kg KOİ	(Yang ve ark., 2023)
				KOİ	60.6			
3D-EF	Anot: Ti/IrO 2 –RuO 2 Katot: Aktif karbon fiber CuFeO/Kaolin	pH: 6.71, akım yoğunluğu: 10 mA/cm2, parçacık dozu: 4 gr/l, reaksiyon süresi: 60 dk	Sentetik	Orange G	99.48	–	0.88 kWh/gr TOC	(Zhou ve ark., 2023)
				TOK	58.13			
3D-EF	Anot: DSA Katot: Grafit keçe Nikel cürufu + laterit	Elektrotlar arası mesafe 4 cm, pH:3, parçacık elektrot dozu: 100 gr/l, voltaj: 4 V, reaksiyon süresi 30 dk	Sentetik	Metilen mavisi	98.64	2.02 kWh/m3	0.69 kWh/m3	(Miao ve ark., 2022)
				KOİ	87.84			

DRAFT

KAYNAKLAR

Andrade LS, Ruotolo LAM, Rocha-Filho RC, Bocchi N, Biaggio SR, Iniesta J, García-Garcia V, Montiel V 2007. On the performance of Fe and Fe,F doped Ti–Pt/PbO2 electrodes in the electrooxidation of the Blue Reactive 19 dye in simulated textile wastewater. Chemosphere, 66(11): 2035–2043. https://doi.org/10.1016/J.CHEMOSPHERE.2006.10.028

Arslan H, Gun M, Akarsu C, Bilici Z, Dizge N 2023. Treatment of turnip juice wastewater by electrocoagulation/electroflotation and electrooxidation with aluminum, iron, boron-doped diamond, and graphite electrodes. International Journal of Environmental Science and Technology, 20: 53–62.

Asfaha YG, Zewge F, Yohannes T, Kebede S 2022. Application of hybrid electrocoagulation and electrooxidation process for treatment of wastewater from the cotton textile industry. Chemosphere, 302: 134706. https://doi.org/10.1016/j.chemosphere.2022.134706

Cabeza A, Urtiaga A, Rivero MJ, Ortiz I 2007. Ammonium removal from landfill leachate by anodic oxidation. Journal of Hazardous Materials, 144(3): 715–719. https://doi.org/10.1016/J.JHAZMAT.2007.01.106

Can W, Yao-Kun H, Qing Z, Min J 2014. Treatment of secondary effluent using a three-dimensional electrode system: COD removal, biotoxicity assessment, and disinfection effects. Chemical Engineering Journal, 243: 1–6. https://doi.org/10.1016/j.cej.2013.12.044

Chi Z, Wang Z, Liu Y, Yang G 2018. Preparation of organosolv lignin-stabilized nano zero-valent iron and its application as granular electrode in the tertiary treatment of pulp and paper wastewater. Chemical Engineering Journal, 331: 317–325. https://doi.org/10.1016/j.cej.2017.08.121

Feng L, Li XY, Gan LH, Xu J 2018. Synergistic effects of electricity and biofilm on Rhodamine B (RhB) degradation in three-dimensional biofilm electrode reactors (3D-BERs). Electrochimica Acta, 290: 165–175. https://doi.org/10.1016/j.electacta.2018.09.068

Fernandes A, Pacheco MJ, Ciríaco L, Lopes A 2015. Review on the electrochemical processes for the treatment of sanitary landfill leachates: Present and future. Applied Catalysis B: Environmental, 176–177: 183–200. https://doi.org/10.1016/J.APCATB.2015.03.052

Fil BA, Günaslan S 2023. Electrooxidation treatment of slaughterhouse wastewater: investigation of efficiency of Ti/Pt anode. Particulate Science and Technology, 41(4): 496–505. https://doi.org/10.1080/02726351.2022.2119905

Fil BA, Günaslan S 2022. Comparison of Different Anodes and Investigation of Energy Consumption in the Treatment of Real Wastewater by Electrooxidation. International Journal of Environmental Research,16: 79.

Foroughi M, Rahmani AR, Asgari G, Nematollahi D 2018. Optimization of a three-dimensional electrochemical system for tetracycline degradation using box-behnken design. Fresenius environmental bulletin, 27(3):1914-1922 https://www.researchgate.net/publication/330652187

Foroughi M, Rahmani AR, Asgari G, Nematollahi D, Yetilmezsoy K, Samarghandi R 2020. Optimization and Modeling of Tetracycline Removal from Wastewater by Three-Dimensional Electrochemical System: Application of Response Surface Methodology and Least Squares Support Vector Machine. Environmental Modeling & Assessment, 25: 327–341.

Goren AY, Kobya M, Oncel MS 2020. Arsenite removal from groundwater by aerated electrocoagulation reactor with Al ball electrodes: Human health risk assessment. Chemosphere, 251: 126363. https://doi.org/10.1016/j.chemosphere.2020.126363

Gu J, Wei G, Zhu Y, Lu C, Zhang L, Huang Z, Su Q, Pan S 2023. Photoelectric activation of persulfate with a new type of red mud-based CuFe2O4 particle electrode for the efficient degradation of ciprofloxacin: Preparation, influencing factors and mechanism. Journal of Environmental Chemical Engineering, 11(1): 109137. https://doi.org/10.1016/j.jece.2022.109137

Hong L, Yang Q, Liying Z, Yingyan C, Bing W 2018. Investigation of a novel pyrolusite particle electrode effects in the chlorine-containing wastewater. Water Science and Technology, 78(7): 1427–1437. https://doi.org/10.2166/wst.2018.414

Hou B, Ren B, Deng R, Zhu G, Wang Z, Li Z 2017. Three-dimensional electro-Fenton oxidation of N-heterocyclic compounds with a novel catalytic particle electrode: high activity, wide pH range and catalytic mechanism. RSC Advances, 7(25): 15455–15462. https://doi.org/10.1039/c7ra00361g

Hu Y, Yu F, Bai Z, Wang Y, Zhang H, Gao X, Wang Y, Li X 2022. Preparation of Fe-loaded needle coke particle electrodes and utilisation in three-dimensional electro-Fenton oxidation of coking wastewater. Chemosphere, 308: 136544. https://doi.org/10.1016/j.chemosphere.2022.136544

Iranpour F, Pourzamani H, Mengelizadeh N, Bahrami P, Mohammadi H 2018. Application of response surface methodology for optimization of reactive black 5 removal by three dimensional electro-Fenton process. Journal of Environmental Chemical Engineering, 6(2): 3418–3435. https://doi.org/10.1016/j.jece.2018.05.023

İlhan F, Kurt U, Apaydın Ö, Arslankaya E, Gönüllü MT 2007. Elektrokimyasal Arıtım ve Uygulamaları: Katı Atık Sızıntı Suyu Çalışması. TÜRKAY AB Sürecinde Türkiye'de Katı Atık Yönetiminde Çevre Sorunları Sempozyumu, YTÜ Makine Fakültesi Endüstri Müh. Bölümü, İstanbul.

Jia Z, Zhao X, Yu C, Wan Q, Liu Y 2021. Design and properties of Sn–Mn–Ce supported activated carbon composite as particle electrode for three-dimensionally electrochemical degradation of phenol. Environmental Technology and Innovation, 23: 101554. https://doi.org/10.1016/j.eti.2021.101554

Kaygusuz T 2013. Elektrokimyasaloksidasyon yönteminin tekstil atıksularının arıtılabilirliğine etkilerinin tam ölçekli araştırılması. Namık Kemal Üniversitesi Fen Bilimleri Enstitüsü. Yüksek Lisans Tezi.

Kul S, Boncukcuoğlu R, Ekmekyapar Torun F, Reçber Z, Sözüdoğru O, Aladağ E 2022. Investigation of the Treatment of Olive Mill Wastewater by Electrooxidation. Water, Air, & Soil Pollution, 233: 421.

Li H, Yang H, Cheng J, Hu C, Yang Z, Wu C 2021a. Three-dimensional particle electrode system treatment of organic wastewater: A general review based on patents. In Journal of Cleaner Production, 308: 127324. https://doi.org/10.1016/j.jclepro.2021.127324

Li XY, Peng P, Wang WK, Wang SY, Feng L, Zhang YC, Xu J 2021b. Particle electrode materials dependent tetrabromobisphenol A degradation in three-dimensional biofilm electrode reactors. Environmental Research, 197: 111089. https://doi.org/10.1016/j.envres.2021.111089

Liu Y, Ma Y, Wan J, Wang Y, Sun J, Xue Y 2021. Electrocatalytic oxidation of ciprofloxacin by Co-Ce-Zr/γ-Al2O3 three-dimensional particle electrode. Environmental Science and Pollution Research, 28: 43815–43830.

Liu W, Ai Z, Zhang L 2012. Design of a neutral three-dimensional electro-Fenton system with foam nickel as particle electrodes for wastewater treatment. Journal of Hazardous Materials, 243: 257–264. https://doi.org/10.1016/j.jhazmat.2012.10.024

Long Y, Feng Y, Li X, Suo N, Chen H, Wang Z, Yu Y 2019. Removal of diclofenac by three-dimensional electro-Fenton-persulfate (3D electro-Fenton-PS). Chemosphere, 219: 1024–1031. https://doi.org/10.1016/j.chemosphere.2018.12.054

Lu C, Gu J, Wei G, Ba J, Zhang L, Li Z, Pei R, Li J, Wei J 2022. Three-dimensional electro-Fenton degradation of ciprofloxacin catalyzed by CuO doped red mud particle electrodes: Influencing factors, possible degradation pathways and energy consumption. Journal of Environmental Chemical Engineering, 10(3): 107737. https://doi.org/10.1016/j.jece.2022.107737

Mengelizadeh N, Pourzamani H, Saloot MK, Hajizadeh Y, Parseh I, Parastar S, Niknam N 2019. Electrochemical Degradation of Reactive Black 5 Using Three-Dimensional Electrochemical System Based on Multiwalled Carbon Nanotubes. Journal of Environmental Engineering, 145(5). https://doi.org/10.1061/(asce)ee.1943-7870.0001517

Miao L, Li J, Yi L, Qu W, Ma C, Feng X, Xu Y, He R 2022. Sustainable reuse of nickel converter slag as a heterogeneous electro-fenton catalyst for treating textile dyeing wastewater: Activity, mechanism and stability assessment. Journal of Cleaner Production, 378: 134421. https://doi.org/10.1016/j.jclepro.2022.134421

Özyonar F, Korkmaz MU 2022. Sequential use of the electrocoagulation-electrooxidation processes for domestic wastewater treatment. Chemosphere, 290: 133172. https://doi.org/10.1016/j.chemosphere.2021.133172

Petala A, Bampos G, Frontistis Z 2022. Using Sawdust Derived Biochar as a Novel 3D Particle Electrode for Micropollutants Degradation. Water (Switzerland), 14(3). https://doi.org/10.3390/w14030357

Pu Y, Zhao F, Chen Y, Lin X, Yin H, Tang X 2023. Enhanced Electrocatalytic Oxidation of Phenol by SnO2-Sb2O3/GAC Particle Electrodes in a Three-Dimensional Electrochemical Oxidation System. Water (Switzerland), 15(10). https://doi.org/10.3390/w15101844

Ren Y, Wang J, Qu G, Ren N, Lu P, Chen X, Wang Z, Yang Y, Hu Y 2023. Study on the mechanism of high effective mineralization of Rhodamine B in three dimensional electrochemical system with γ-Fe2O3@CNTs particle electrodes. Separation and Purification Technology, 314: 123616. https://doi.org/10.1016/j.seppur.2023.123616

Romano A, Urtiaga AM, Ortiz I 2020. Optimized energy consumption in electrochemical-based regeneration of RAS water. Separation and Purification Technology, 240: 116638. https://doi.org/10.1016/j.seppur.2020.116638

Sivri S 2020. Elektrooksidasyon yöntemi ile nonilfenoletoksilat-10 (NP10E) arıtımı ve yüzey yanıt metodu ile optimizasyonu. - T.C. Bursa Uludağ Üniversitesi Fen Bilimleri Enstitüsü. Yüksek lisans tezi.

Song B, Wang Z, Li J, Luo M, Cao P, Zhang C 2021. Volcanic rock: A new type of particle electrode with excellent performance, which can efficiently degrade norfloxacin. Chemical Engineering Journal, 426: 131940. https://doi.org/10.1016/j.cej.2021.131940

Sun W, Zhou S, Sun Y, Xu Y, Zheng H 2021. W-Ag-Ti@γ-Al2O3particle electrodes for enhanced electrocatalytic pretreatment of coal chemical wastewater. Journal of Environmental Chemical Engineering, 9(1): 104681. https://doi.org/10.1016/j.jece.2020.104681

Ta M, Wang T, Guo J, Wang Y, Zhang J, Zhao C, Liu S, Liu G, Yang H 2023. Enhanced norfloxacin degradation by three-dimensional (3D) electrochemical activation of peroxymonosulfate using Mn/Cu co-doped activated carbon particle electrode. Separation and Purification Technology, 310: 123067. https://doi.org/10.1016/j.seppur.2022.123067

Thangamani R, Periyaraman PM, Thanarasu A, Velayutham K, Dhanasekaran A, Subramanian S 2022. Electrooxidation of coragen-contaminated wastewater using graphite electrodes and sorbent nano-hydroxyapatite. Environmental Technology (United Kingdom), 43(11): 1603–1612. https://doi.org/10.1080/09593330.2020.1844306

Ulucan-Altuntas K, El Hadki A, Ilhan F, Zrineh A, El Hadki H, Kabbaj OK, Dahchour A, Debik E 2022. Electrocatalytic degradation of oxytetracycline using three-dimensional electrode and optimization via fuzzy logic modeling. Separation Science and Technology (Philadelphia), 57(3): 454–464. https://doi.org/10.1080/01496395.2021.1900867

Wang T, Ta M, Guo J, Liang LE, Bai C, Zhang J, Ding H 2023. Insight into the synergy between rice shell biochar particle electrodes and peroxymonosulfate in a three-dimensional electrochemical reactor for norfloxacin degradation. Separation and Purification Technology, 304: 122354. https://doi.org/10.1016/j.seppur.2022.122354

Wang T, Zhang J, Song Y, Liu Z, Ding H, Zhao C, Wang P 2021. Role of micro-size zero valence iron as particle electrodes in a three-dimensional heterogeneous electro-ozonation process for nitrobenzene degradation. Chemosphere, 276: 130264. https://doi.org/10.1016/j.chemosphere.2021.130264

Wu X, Song X, Chen H, Yu J 2021. Treatment of phenolic compound wastewater using CuFe2O4/Al2O3 particle electrodes in a three-dimensional electrochemical oxidation system. Environmental Technology (United Kingdom), 42(28): 4393–4404. https://doi.org/10.1080/09593330.2020.1760356

Xiao H, Hao Y, Wu J, Meng X, Feng F, Xu F, Luo S, Jiang B 2023. Differentiating the reaction mechanism of three-dimensionally electrocatalytic system packed with different particle electrodes: Electro-oxidation versus electro-fenton. Chemosphere, 325: 138423. https://doi.org/10.1016/j.chemosphere.2023.138423

Xie S, Li M, Liao Y, Qin Q, Sun S, Tan Y 2021. In-situ preparation of biochar-loaded particle electrode and its application in the electrochemical degradation of 4-chlorophenol in wastewater. Chemosphere, 273: 128506. https://doi.org/10.1016/j.chemosphere.2020.128506

Yan L, Wang Y, Li J, Shen H, Zhang C, Qu T 2016. Reduction of chemical oxygen demand from refinery wastewater by three-dimensional electrode-electro-fenton process. Bulletin of the Chemical Society of Japan, 89(1): 50–57. https://doi.org/10.1246/bcsj.20150250

Yang Z, Yang S, Shiqiao Y, Yuanhong D 2023. Enhanced Rhodamine B degradation by GAC/Mn–Sn particles electrodes. Water Science and Technology, 87(6): 1552–1570. https://doi.org/10.2166/wst.2023.063

Zazou H, Afanga H, Akhouairi S, Ouchtak H, Addi AA, Akbour RA, Assabbane A, Douch J, Elmchaouri A, Duplay J, Jada A, Hamdani M 2019. Treatment of textile industry wastewater by electrocoagulation coupled with electrochemical advanced oxidation process. Journal of Water Process Engineering, 28: 214–221. https://doi.org/10.1016/j.jwpe.2019.02.006

Zhan J, Li Z, Yu G, Pan X, Wang J, Zhu W, Han X, Wang Y 2019. Enhanced treatment of pharmaceutical wastewater by combining three-dimensional electrochemical process with ozonation to in situ regenerate granular activated carbon particle electrodes. Separation and Purification Technology, 208: 12–18. https://doi.org/10.1016/j.seppur.2018.06.030

Zhang C, Jiang Y, Li Y, Hu Z, Zhou L, Zhou M 2013. Three-dimensional electrochemical process for wastewater treatment: A general review. In Chemical Engineering Journal, 228:455–467. https://doi.org/10.1016/j.cej.2013.05.033

Zhang Y, Zhang D, Zhou L, Zhao Y, Chen J, Chen Z, Wang F 2018. Polypyrrole/reduced graphene oxide aerogel particle electrodes for high-efficiency electro-catalytic synergistic removal of Cr(VI) and bisphenol A. Chemical Engineering Journal, 336: 690–700. https://doi.org/10.1016/j.cej.2017.11.109

Zhao Z, Hao Y, Wu J, Feng Z, Feng F, Li Y, Yang Q, Jiang B 2023. Development of a three-dimensional electro-Fenton system packed with C-PTFE/Fe–Co–C hybrid particle electrodes for simultaneous H2O2 generation and activation into •OH. Separation and Purification Technology, 317: 123960. https://doi.org/10.1016/j.seppur.2023.123960

Zheng Y, Qiu S, Deng F, Zhu Y, Li G, Ma F 2019. Three-dimensional electro-Fenton system with iron foam as particle electrode for folic acid wastewater pretreatment. Separation and Purification Technology, 224: 463–474. https://doi.org/10.1016/j.seppur.2019.05.054

Zhou C, Peng X, Li X, Qi K, Gao L 2023. Stable CuFeO/Kaolin-based catalytic particle electrode in 3D heterogeneous electro-Fenton system for orange G removal: Synthesis, performance and mechanism. Journal of Environmental Chemical Engineering, 11(2): 109562. https://doi.org/10.1016/j.jece.2023.109562

Zhu X, Ni J, Xing X, Li H, Jiang Y 2011. Synergies between electrochemical oxidation and activated carbon adsorption in three-dimensional boron-doped diamond anode system. Electrochimica Acta, 56(3): 1270–1274. https://doi.org/10.1016/J.ELECTACTA.2010.10.073

DRAFT

ORAL PRESENTATION

Co_3O_4 ve Fe_3O_4 nanokatalizörlerin $NaBH_4$ hidroliziyle hidrojen üretimlerinin karşılaştırılması

Adil Umaz[1*] (*ORCID: https://orcid.org/0000-0003-2438-5454*)

[*1]Mardin Artuklu Üniversitesi, Sağlık Hizmetleri Meslek Yüksekokulu, Tibbi Hizmetler ve Teknikler Bölümü, Mardin, Türkiye

*Sorumlu yazar e-mail: adilumaz@gmail.com

Özet

Hidrojenin depolanması, dağıtımı ve kontrollü salınımı, hidrojene dayalı ekonomi için önemli bir endişe kaynağıdır. Sodyum borohidrit ($NaBH_4$), hidrojen depolama ve üretimi için kullanılan en çok çalışılan kimyasal hidritlerden biridir. Bununla birlikte, kontrol edilebilir hidrojen üretimi için dehidrojenasyonunu hızlandırmak üzere verimli katalizörler gerektirir. Bu çalışmada, ticari olarak satın alınan Co_3O_4 ve sentezlenen Fe_3O_4 nanokatalizörlerinin $NaBH_4$ hidroliziyle hidrojen üretimleri incelenmiştir. Ayrıca, bu iki nanokatalizörün hidrojen üretim hacimleri ve hidrojen üretim hızları (HGR) karşılaştırılmıştır. Fe_3O_4 nanokatalizörünün $NaBH_4$ hidroliziyle üretilen hidrojen hacmi 112 mL ve HGR değeri ise 306.67 mL H2/gkat·dk. olarak ölçüldü. Co_3O_4 nanokatalizörü kullanıldığında üretilen hidrojen hacmi 235 mL ve HGR değeri ise 713.33 mL H2/gkat·dk. olarak ölçüldü. Sonuç olarak, Co_3O_4 nanokatalizörü Fe_3O_4'ten $NaBH_4$ hidrolizi için daha yüksek bir katalitik aktivite sergilediğini göstermektedir. Ayrıca, bu çalışma Co_3O_4 bazlı sentezlenecek katalizörlerin $NaBH_4$ hidroliziyle daha yüksek hidrojen üretimi sağlayacaktır.

Anahtar Kelimeler: Hidrojen üretimi, $NaBH_4$, Co_3O_4, Fe_3O_4

Comparison of hydrogen production of Co_3O_4 and Fe_3O_4 nanocatalysts by $NaBH_4$ hydrolysis

Abstract

The storage, distribution and controlled release of hydrogen is a major concern for the hydrogen-based economy. Sodium borohydride ($NaBH_4$) is one of the most studied chemical hydrides used for hydrogen storage and production. However, controllable hydrogen production requires efficient catalysts to accelerate its dehydrogenation. In this study, hydrogen production of commercially purchased Co_3O_4 and synthesized Fe_3O_4 nanocatalysts by $NaBH_4$ hydrolysis was investigated. In addition, the hydrogen production volumes and hydrogen production rates (HGR) of these two nanocatalysts were compared. The hydrogen volume produced by the $NaBH_4$ hydrolysis of the Fe_3O_4 nanocatalyst is 112 mL and the HGR value is 306.67 mL H2/gkat·min was measured. When the Co_3O_4 nanocatalyst is used, the volume of hydrogen produced is 235 mL and the HGR value is 713.33 mL H2/gkat·min was measured. In conclusion, the Co_3O_4 nanocatalyst shows a higher catalytic activity for $NaBH_4$ hydrolysis from Fe_3O_4. In addition, this study will provide higher hydrogen production by $NaBH_4$ hydrolysis of catalysts to be synthesized based on Co_3O_4.

Keywords: Hydrogen production, $NaBH_4$, Co_3O_4, Fe_3O_4

GİRİŞ

Enerji ve enerji kaynakları ülkelerin ekonomik ve sosyal gelişimi açısından oldukça önemlidir. Bu enerji ihtiyacını karşılamak için birincil enerji kaynağı olarak çoğunlukla fosil yakıtlar kullanılmaktadır (Özkan ve ark., 2019). Ancak bu kaynakların yakın gelecekte tükenmesi ve sera gazlarının salınımından dolayı çevre kirliliğine sebep olması, temiz, güvenli, ekonomik ve verimliliği yüksek enerji kaynakları arayışını tetiklemiştir (Vernekar ve ark., 2012; Özkan ve ark., 2019).

Bu enerji kaynakları arasında Hidrojen (H_2), çevre dostu doğası ve yüksek enerji yoğunluğu nedeniyle gelecek için alternatif bir enerji taşıyıcısı olarak kabul edilmektedir (Özturk ve ark., 2018; Yang ve ark., 2021). Tüm avantajlarına rağmen hidrojene dayalı enerji teknolojisinin gelişmesi için üretim, depolama ve nakliye gibi

DRAFT

sorunların çözülmesi gerekmektedir. Basınçlı hidrojen, katı hal hidrojen, kriyojenik sıvı, karbon malzemelerde adsorpsiyon, metal hidritler gibi çeşitli hidrojen depolama teknolojileri mevcuttur. Bu teknolojiler arasında metal hidritler yüksek gravimetrik hidrojen depolama yoğunluklarına sahiptir (Dönmez ve Ayas, 2021). Bu metal hidritler; sodyum hidrit (NaH), sodyum alüminyum hidrit ($NaAlH_4$), sodyum borohidrit ($NaBH_4$), lityum hidrit (LiH), lityum alüminyum hidrit ($LiAlH_4$), lityum borohidrit ($LiBH_4$), kalsiyum hidrit (CaH_2), kalsiyum borohidrit ($Ca(BH_4)_2$), magnezyum hidrit (MgH_2), magnezyum borohidrit ($Mg(BH_4)_2$), potasyum hidrit (KH), potasyum borohidrit (KBH_4)'tir (Izgi ve ark., 2019). Bu hidritlerin çoğu su ile çok hızlı reaksiyona girer. Diğer hidritlerle karşılaştırıldığında $NaBH_4$, yüksek hidrojen kapasitesine (ağırlıkça %10,8) sahip olması, ekonomik olması, yanıcı olmaması, alkali çözeltilerde çok kararlı olması ve çevre dostu olması gibi avantajlara sahiptir. Ayrıca, $NaBH_4$ hidroliz işleminin yan ürünü ($NaBO_2$) toksik değildir ve geri dönüştürülebilir (Yolcular ve Karaoglu, 2020). NaBH4'ün hidroliz reaksiyonu (Reaksiyon 1) aşağıdaki gibidir.

$$NaBH_4 + 2H_2O \rightarrow 4H_2 + NaBO_2 \quad (1)$$

Bu çalışmada, sentezlenen Fe_3O_4 ve ticari olarak satın alınan Co_3O_4 nanokatalizörlerin $NaBH_4$ hidroliziyle hidrojen üretimleri incelenmiştir. Co_3O_4 nanokatalizörün hidrojen üretimi için $NaBH_4$, NaOH ve katalizör miktarları gibi parametreler optimize edilmiş ve en yüksek hidrojen miktarı veren parametreler belirlenmiştir. Ayrıca, optimum şartlarda bu iki nanokatalizörlerin hidrojen üretim miktarları karşılaştırılmıştır.

MATERYAL VE METOT

Kimyasallar

Reaksiyonlarda kullanılan tüm kimyasallar ve çözücüler analitik saflıktadır. Demir (III) klorür hekzahidrat ($FeCl_3 \cdot 6H_2O$, % 99.9, Merck), demir (II) klorür tetrahidrat ($FeCl_2 \cdot 4H_2O$,% 98, Alfa Aesar), amonyak (NH_3, % 28.0, İsolab), etanol (CH_3CH_2OH, % 99.9, İsolab), kobalt oksit (Co_3O_4, % 99.55, 48 nm) Nanografi şirketinden temin edildi.

Fe_3O_4 Manyetik Nanokatalizörün Sentezi

12 mmol $FeCI_3 \cdot 6H_2O$ ve 6 mmol $FeCI_2 \cdot 4H_2O$ bileşikleri iki boyunlu balona konularak 200 ml saf su eklendi ve karışım oda sıcaklığında 30 dakika boyunca karıştırıldı. Reaksiyon sıcaklığı 70 °C ye çıkartıldı. Karışım üzerine 50 mL 8 M NH_3 eklendi (Çözelti ilk başta turuncu renkte iken NH_3 eklenmesiyle siyah renge dönüştü). Oluşan siyah karışım 30 dakika daha karıştırıldı. Ardından karışım soğutuldu ve mıktanıs yardımıyla Fe_3O_4 manyetik nanokatalizör ortamdan izole edildi. Manyetik nanokatalizör 4 defa saf suyla ve 2 defa etanolle yıkandı ve 70 °C'de 24 saat etüvde kurutma işlemi yapıldı (Kutluay ve ark., 2021).

Karakterizasyon

Fe_3O_4 manyetik nanokatalizör Fourier dönüşümü kızılötesi spektroskopisi (FTIR) spektrumu, Bruker Vertex 70 spektrometresi kullanılarak kaydedildi.

Hidrojen Üretimi

Fe_3O_4 ve Co_3O_4 nanokatalizörlerin $NaBH_4$ hidroliziyle hidrojen üretim için 100 mL'lik yuvarlak tabanlı reaksiyon şişesine 75 mg $NaBH_4$ ve 25 mg nanokatalizörler ilave edildi. Hidroliz reaksiyonunu başlatmak için 25 °C'de bu reaksiyon şişesine 10 mL su ilave edildi ve 500 rpm'de çözelti karıştırıldı. Açığa çıkan hidrojen miktarı, tasarladığımız su dolu ters çevrilmiş bir gaz silindiri aracılığıyla Su-gaz yer değiştirme yöntemi kullanılarak zamana bağlı olarak kaydedildi. HGR, aşağıdaki formül kullanarak belirlendi (Wei ve ark., 2020).

$$HGR = \frac{mL\ H2}{gkat \times zaman} \quad (2)$$

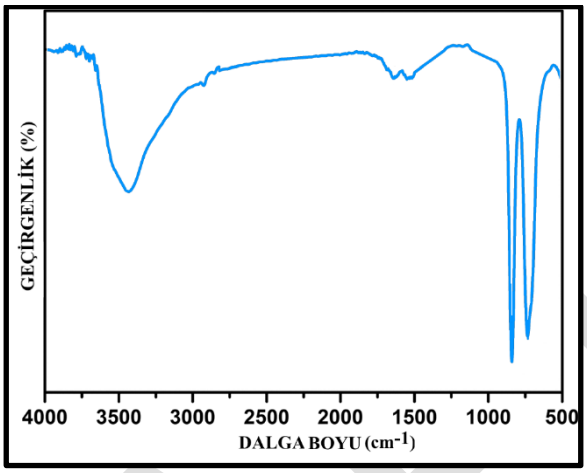
Bu formülde, mL H_2; üretilen hidrojen hacmi, gkat; kullanılan katalizör miktarı ve zaman ise reaksiyon süresiydi. Bu çalışmada, $NaBH_4$ (25, 50 ve 75 mg), NaOH (0 ve 20 mg), katalizör (10, 25 ve 50 mg) miktarları ve katalizör çeşidinin hidrojen üretimi üzerinde etkileri incelendi.

BULGULAR ve TARTIŞMA

Co_3O_4 ve Fe_3O_4 Nanokatalizörlerin Karakterizasyonu

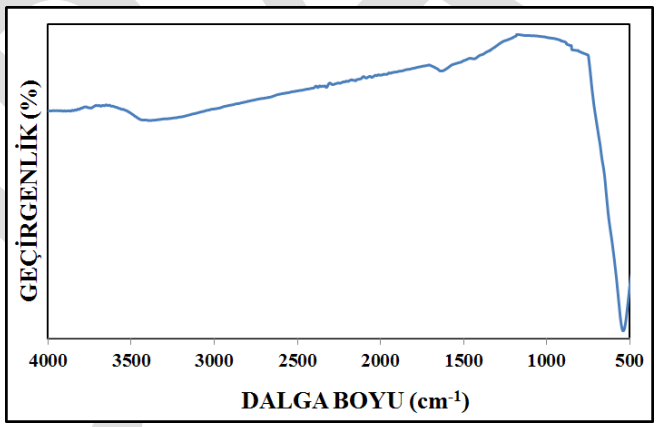
Co_3O_4 ve Fe_3O_4 nanokatalizörlerde bağ oluşumunu analiz etmek için FTIR spektrumu kullanıldı ve bu nanokatalizöre ait sonuçlar Şekil 1 ve 2'de verildi. Bu nanokatalizörlerin FTIR spektrumu, 4000 ila 500 cm^{-1} aralığında izlendi.

Co_3O_4 nanokatalizörine ait FTIR spektrumu incelendiğinde, 730 ve 830 cm^{-1}'de ortaya çıkan iki güçlü ve keskin pik, bu nanokatalizörün doğrulanması olan metal oksit sallanma titreşimine karşılık gelen Co-O'nun karakteristik bantlarına atfedilir. 3440 ve 1630 cm^{-1}'deki iki farklı titreşim bandı, adsorbe edilmiş su moleküllerinden kaynaklanan O-H gerilme ve bükülme titreşimlerine atfedilir (Şekil 1) (Packiaraj ve ark., 2019).



Şekil 1. Co_3O_4 nanokatalizörüne ait FTIR spektrumu (Packiaraj ve ark., 2019).

Fe_3O_4 nanokatalizörüne ait FTIR spektrumu incelendiğinde, 560 cm^{-1} civarındaki keskin pik, Fe-O bağıyla (gerilim titreşimi) ilişkilidir. 3400 cm^{-1} civarındaki çok geniş pik, -OH (gerilim titreşimi) gruplarının varlığının bir göstergesidir (Şekil 2) (Kutluay ve ark., 2021).



Şekil 2. Fe_3O_4 nanokatalizörüne ait FTIR spektrumu (Packiaraj et al., 2019; Kutluay et al., 2021)

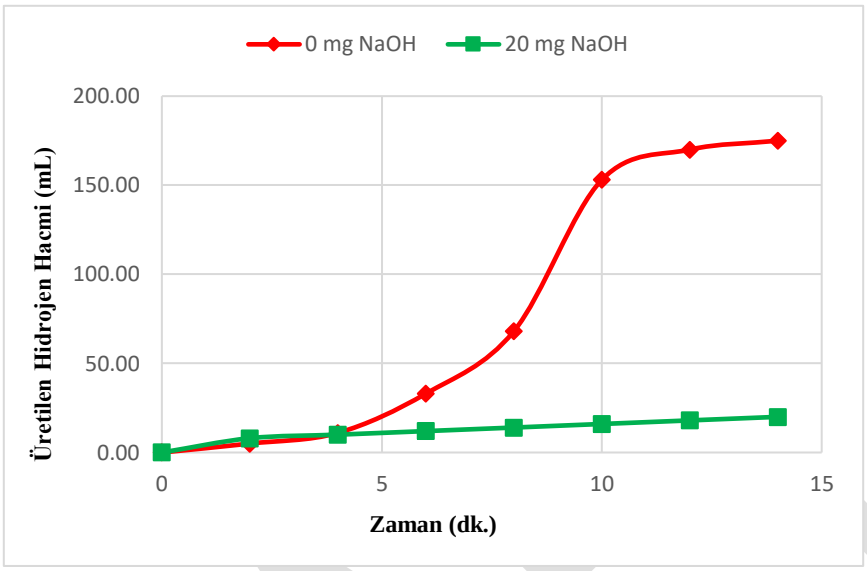
$NaBH_4$'den Hidrojen Üretimi

$NaBH_4$ hidrolizi yoluyla hidrojen üretimi, $NaBH_4$ miktarı, alkalinite için kullanılan NaOH miktarı, reaksiyon sıcaklığı ve kullanılan katalizör miktarı gibi birçok faktörden etkilenir. Bu çalışmada, bu faktörlerin detaylı incelemesi yapılmıştır. Ayrıca, katalizör olarak kullanılan Co_3O_4 ve Fe_3O_4 nanokatalizörlerin hidrojen üretim miktarları kıyaslanmıştır.

NaOH Miktarının Hidrojen Üretimine Etkisi

$NaBH_4$'ün kendi kendine hidrolizini engellemek için $NaBH_4$ çözeltisine genellikle bir dengeleyici görevi görmesi için NaOH eklenir (Li ve ark., 2021). Bu nedenle, NaOH miktarındaki bir artışın hidroliz oranını düşürmesi beklenir (Huang ve ark., 2016).

Şekil 3, 25 °C'de 25 mg nanokatalizör, 50 mg $NaBH_4$ ve farklı miktarlarda NaOH kullanılarak hidrojen üretimlerini verir. NaOH miktarı 0'dan 20 mg kadar yükseltildiğinde, üretilen hidrojen hacmi dramatik bir şekilde 175'ten 20 mL'ye kadar düştüğü belirlendi (Şekil 3). NaOH miktarındaki bir artış hidrojen hacminin azalmasına neden olmuştur. Farklı miktarlardaki NaOH'in (0 ve 20 mg) HGR değerleri sırasıyla 612 ve 64 mL H_2/gkat·dk. olarak belirlendi. HGR değerleri kıyaslandığında, Co_3O_4 nanokatalizörünün $NaBH_4$ hidroliz reaksiyonunda NaOH'in kullanılmaması en yüksek HGR değeriyle sonuçlandı.

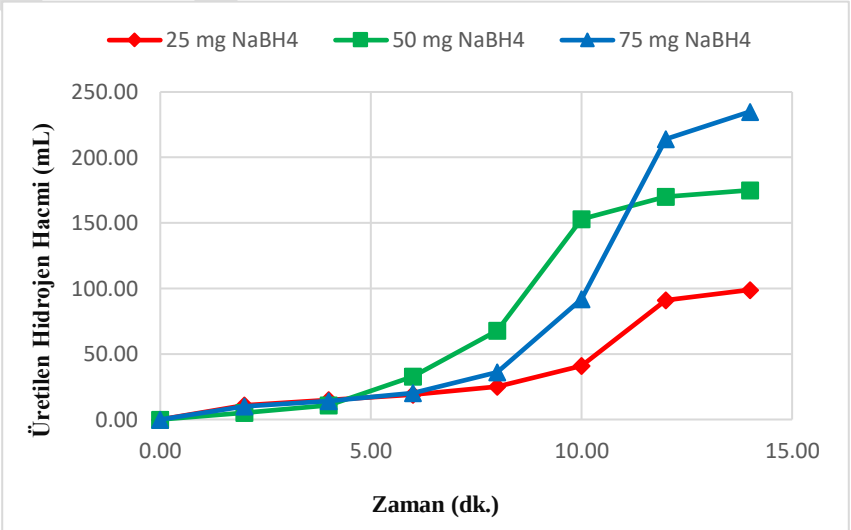


Şekil 3. Farklı miktarlardaki NaOH'in, $NaBH_4$ hidroliziyle hidrojen hacmi üzerindeki etkisi (Reaksiyon koşulları: 25 mg nanokatalizör, 50 mg $NaBH_4$, 0 ve 20 mg NaOH ve 25 °C)

$NaBH_4$ Miktarının Hidrojen Üretimine Etkisi

$NaBH_4$ miktarının Co_3O_4 nanokatalizörü kullanılarak hidrojen üretimi üzerindeki etkisi, Şekil 4'te gösterilmektedir. Hidroliz reaksiyonu için başlangıç koşulları şunlardı. 25 mg Co_3O_4 nanokatalizörü, 10 ml su ve 500 rpm karıştırma hızı ile 25°C'de 25-75 mg arasında değişen $NaBH_4$ miktarları kullanıldı.

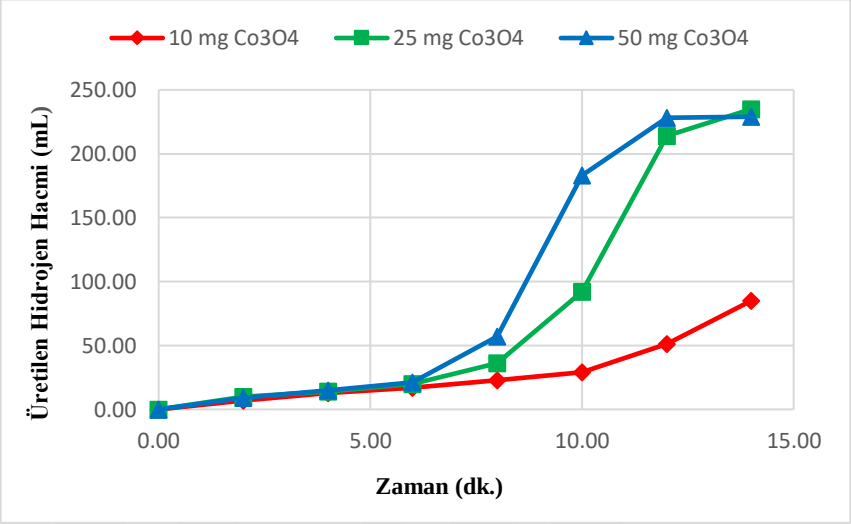
$NaBH_4$ miktarı 25'ten 75 mg kadar yükseltildiğinde, üretilen hidrojen hacmi dramatik bir şekilde 99'dan 235 mL'ye kadar yükseldi. Şekil 4 incelendiğinde, 10 dk. kadar 50 mg $NaBH_4$ daha fazla hidrojen ürettiği belirlendi. Bunun nedeni, reaksiyon ortamında daha yüksek miktarda $NaBH_4$ kullanılması sonucu bir yan ürün olan $NaBO_2$ üretebilir, bu da katalizörün aktif bölgelerini bloke edebilir ve böylece daha düşük hidrojen hacmine neden olduğu söylenebilir. Ancak 10 dk. sonra $NaBO_2$ katalizör üzerindeki etkisi azaldığı için 75 mg $NaBH_4$ daha fazla hidrojen hacmi ürettiği belirlendi. $NaBH_4$ miktarındaki artış üretilen hidrojen hacminin arttırılmasına faydalı olduğu görülmektedir.



Şekil 4. Farklı miktarlardaki $NaBH_4$'ün, Co_3O_4 katalizörü kullanılarak $NaBH_4$ hidrolizi yoluyla hidrojen hacmi üzerindeki etkisi (Reaksiyon koşulları: 25 mg nanokatalizör, 25, 50 ve 75 mg $NaBH_4$ ve 25 °C)

Katalizör Miktarının Hidrojen Üretimine Etkisi

Şekil 5, aynı koşullar altında farklı Co_3O_4 nanokatalizör miktarları (10 mg'dan 50 mg'a) için hidrojen üretimini göstermektedir. Nanokatalizör miktarı 10 mg'dan 50 mg kadar yükseltildiğinde, üretilen hidrojen hacmi dramatik bir şekilde 85'ten 235 mL'ye kadar yükseldi. Şekil 5 incelendiğinde, 5 dk. kadar farklı miktarlardaki nanokatalizörlerin üretilen hidrojen hacimleri birbirine yakın olduğu belirlendi. 5-14 dk. arasına kadar artan katalizör miktarına bağlı olarak üretilen hidrojen hacimlerinde artış gözlemlendi. Bunun nedeni muhtemelen çözeltiye eklenen nanokatalizörün tamamen $NaBH_4$'e maruz kalmış olmasıdır. 14 dk. sonra artan katalizör miktarına bağlı olarak üretilen hidrojen hacimlerinde azalış gözlemlendi. Bunun nedeni, nanokatalizör yapısının bozulmaya başladığı söylenebilir.



Şekil 5. Farklı miktarlardaki Co_3O_4 katalizörü kullanılarak $NaBH_4$ hidrolizi yoluyla hidrojen hacmi üzerindeki etkisi (Reaksiyon koşulları: 10, 25 ve 50 mg nanokatalizör, 75 mg $NaBH_4$ ve 25 °C)

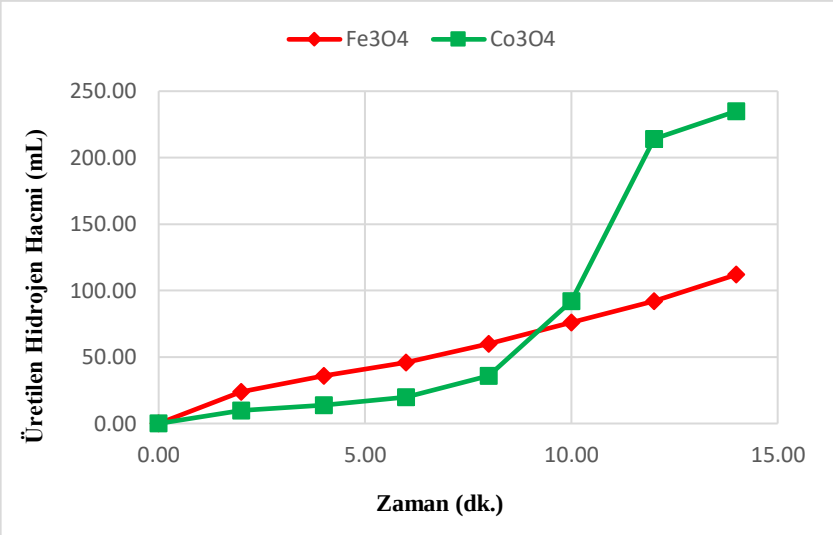
Farklı miktarlardaki nanokatalizörün (10, 25 ve 50 mg) HGR değerleri karşılaştırıldığında, 10 mg nanokatalizöre karşılık gelen HGR, 25 °C'de 425 mL H_2/gkat·dk. olarak belirlendi. Bununla birlikte, nanokatalizör miktarı 25 mg'a çıkartıldığında, hidrojen üretimi hızlandı ve 12 dakikada HGR değeri 713.33 mL H_2/gkat·dk. ulaştı. Nanokatalizör miktarı 50 mg'a çıkarıldığında, hidrojen üretimi 12 dakikada artığı, yine de 380 mL H_2/gkat·dk. gibi nispeten küçük bir HGR'ye neden olduğu belirlendi. Bu sonuçlar, daha yüksek bir nanokatalizör miktarının daha fazla H_2 üretmek üzere $NaBH_4$'ün hidrolizini kolaylaştırıyor gibi görünse de, fazla nanokatalizörün kullanımı nispeten düşük HGR'ye yol açabileceğini göstermektedir. Bunun nedeni muhtemelen eklenen nanokatalizörün tamamen $NaBH_4$'e maruz kalmamış olması ve dolayısıyla nanokatalizörün sadece bir kısmının $NaBH_4$ hidrolizinin katalizine katkıda bulunmasıdır. Bu sonuçlar ayrıca, 25 mg'lık nanokatalizör miktarının optimal miktar olduğu belirlendi.

Katalizör Çeşidinin Hidrojen Üretimine Etkisi

Co_3O_4 nanokatalizörünün hidrojen üretimi için belirlenen optimum koşullarında bir başka nanokatalizör olan Fe_3O_4'in hidrojen üretimi ve HGR değerleri belirlendi. Ayrıca, bu iki nanokatalizörün hidrojen üretimi ve HGR değerleri karşılaştırıldı. Fe_3O_4 nanokatalizörünün hidrojen üretimi, $NaBH_4$'ten çok yavaş salındığını ve hidrojen hacmi 14 dakikada 112 mL olarak belirlendi. Hidrojen üretim hızı ise 306.67 mL H_2/gkat·dk. olarak belirlendi.

Co_3O_4 nanokatalizörü kullanıldığında, hidrojen üretimi 14 dakika içinde 235 mL'ye ulaştığı ve hidrojen ürerim hızı ise 713.33 mL H_2/gkat·dk. olarak belirlendi. Co_3O_4 nanokatalizörü, Fe_3O_4'in $NaBH_4$'ün hidrolizinden daha hızlı olduğu belirlendi. Böylece, Co_3O_4 nanokatalizörü, $NaBH_4$'ün hidrolizini katalize edebildiğini ortaya çıkardı. Bu sonuçlara göre Co_3O_4 nanokatalizörü, Fe_3O_4'ten daha yüksek bir katalitik aktivite sergilediğini gösterir. Ayrıca, bu karşılaştırma aynı zamanda Co_3O_4 bazlı malzemelerin üretilmesi ve bunların kullanımı

hidrojen üretimi için $NaBH_4$ hidrolizini katalize etmek için gelecek vaat eden avantajlı bir malzeme olarak göründüğünü doğrular.



Şekil 6. Farklı nanokatalizörler kullanılarak $NaBH_4$ hidroliziyle hidrojen hacmi üzerindeki etkisi (Reaksiyon koşulları: 25 mg nanokatalizör, 75 mg $NaBH_4$,ve 25 °C)

Tablo 1 ile karşılaştırıldığında, Co_3O_4 nanokatalizörü tarafından katalize edilen $NaBH_4$'ün hidrolizi, literatürdeki ticari Co_3O_4 katalizörüne yakın bir HGR değeri gösterdiği belirlendi. Ayrıca, Co_3O_4 nanokatalizörü, diğer Co_3O_4 katalizörler ve bu katalizörlere bağlı yapılardan daha düşük bir HGR gösterdiği belirlendi. Bunun nedeni literatürdeki Co_3O_4 katalizörlerinin yapı şekli ve katalizöre bağlı olan yapılardan dolayı olduğu söylenebilir. Bu da Co_3O_4 katalizörüne bağlanacak grubun HGR artışına sebep olacağını gösterir (Tablo 1). Fe_3O_4 nanokatalizörü Tablo 1 ile karşılaştırıldığında, Fe_3O_4 bazlı katalizörlerden daha düşük bir HGR sahip olduğu belirlendi. Fe_3O_4 nanokatalizörü de Co_3O_4 katalizörü gibi bir yapı bağlandığında HGR değerinde bir atış olduğu belirlendi (Tablo 1). Böylece, Her iki Fe_3O_4 ve Co_3O_4 nanokatalizörlerine farklı yapılar bağlanarak daha yüksek HGR değerleri elde edilebilir.

Tablo 1. $NaBH_4$ hidrolizi ile hidrojen üretimi için kullanılan çeşitli katalizörlerin karşılaştırılması

Katalizör	Sıcaklık (°C)	HGR (ml H_2/ gkat·dk.)	Kaynaklar
Ticari Co_3O_4	25	860	(Krishnan ve ark., 2008)
Co_3O_4 nanoköpüğü	30	2530	(Pfeil ve ark., 2014)
Yüksek kristalli Co_3O_4	40	2500	(Patel ve ark., 2013)
Co_3O_4 tozu	30	1000	(Edla ve ark., 2016)
Co_3O_4 kaplı NPS	30	5010	(Edla ve ark., 2016)
Ni/Co_3O_4	25	1925	(Bozkurt ve ark., 2018)
Co/Co_3O_4	25	2823	(Bozkurt ve ark., 2018)
C/Co_3O_4	25	3686	(Ugale ve ark., 2022)
C/Co_3O_4	27	1120	(Ugale ve ark., 2022)
Co_3O_4 içi boş mikro küre	25	5340	(Wang ve ark., 2020)
Ticari Co_3O_4	25	713.33	Bu çalışmada
Co/Fe_3O_4@C	25	1403	(Chen ve ark., 2018)
Oksitlenmiş Fe_2O_3	25	264	(Nabid ve ark., 2019)
α-Fe_2O_3@N-C NSs	25	637	(Nabid ve ark., 2019)
Fe_3O_4@SiO_2–Pt	25	7500	(Ro ve ark., 2019)
Fe_3O_4@SiO_2–Pt/Ni	25	19000	(Ro ve ark., 2019)
Fe_3O_4@SiO_2–Pt@TiO_2	25	14000	(Ro ve ark., 2019)
Fe_3O_4	25	306.67	Bu çalışmada

SONUÇ

Bu çalışmada, ticari olarak satın alınan Co_3O_4 ve Fe_3O_4 nanokatalizörü basit, güvenilir ve ucuz bir proses ile başarılı bir şekilde hazırlanmıştır. Co_3O_4 ve Fe_3O_4 nanokatalizörleri alkali çözeltilerde $NaBH_4$ hidrolizinden

hidrojen üretimi için kullanılır. Co_3O_4 nanokatalizörünün HGR üzerindeki etkisini belirlemek için $NaBH_4$, NaOH ve katalizör miktarları incelenmiştir. Ayrıca, Co_3O_4 ve Fe_3O_4 nanokatalizörlerinin HGR değerleri karşılaştırılmıştır. Co_3O_4 nanokatalizörü için 75 mg $NaBH_4$, 25 mg katalizör, 10 ml saf su kullanılması ve reaksiyonda NaOH kullanılmamasıyla en yüksek HGR değeri elde edilmiştir. Elde edilen maksimum HGR değeri 713.33 mL H_2/gkat·dk. olarak bulunmuştur. Fe_3O_4 nanokatalizörünün HGR değeri ise 306.67 mL H_2/gkat·dk. olarak bulunmuştur. HGR değerleri literatürde bildirilen değerlerle karşılaştırılabilir. $NaBH_4$ hidrolizi yoluyla hidrojen üretimi için ekonomik ve büyük ölçekli Co_3O_4 ve Fe_3O_4 bazlı nanokatalizörlerinin üretimi, hidrojeni ana yakıt kaynağı yapmak için çok daha basit ve daha ticari bir süreç sunar.

KAYNAKLAR

Bozkurt G, Özer A, Yurtcan A.B. 2018. Hydrogen generation from sodium borohydride with Ni and Co based catalysts supported on Co_3O_4. International Journal of Hydrogen Energy, 43(49): 22205–22214.

Chen B, Chen S, Bandal H.A, Appiah-Ntiamoah R, Jadhav A.R, Kim H. 2018. Cobalt nanoparticles supported on magnetic core-shell structured carbon as a highly efficient catalyst for hydrogen generation from $NaBH_4$ hydrolysis. International Journal of Hydrogen Energy, 43(19): 9296–9306.

Dönmez F, Ayas N. 2021. Synthesis of Ni/TiO_2 catalyst by sol-gel method for hydrogen production from sodium borohydride. International Journal of Hydrogen Energy, 46(57): 29314–29322.

Edla R, Gupta S, Patel N, Bazzanella N, Fernandes R, Kothari D.C, Miotello A. 2016. Enhanced H_2 production from hydrolysis of sodium borohydride using Co_3O_4 nanoparticles assembled coatings prepared by pulsed laser deposition. Applied Catalysis A: General, 515: 1–9.

Huang Y, Wang K, Cui L, Zhu W, Asiri A.M, Sun, X. 2016. Effective hydrolysis of sodium borohydride driven by self-supported cobalt oxide nanorod array for on-demand hydrogen generation. Catalysis Communications, 87: 94–97.

Izgi M.S, Onat E, Çelik Kazici H, Şahin Ö. 2019. Hydrogen production through the cooperation of a catalyst synthesized in ethanol medium and the effect of the plasma. Energy Sources, Part A: Recovery, Utilization and Environmental Effects, 45(3): 8271–8284.

Krishnan P, Advani S.G, Prasad A.K. 2008. Cobalt oxides as Co_2B catalyst precursors for the hydrolysis of sodium borohydride solutions to generate hydrogen for PEM fuel cells. International Journal of Hydrogen Energy, 33(23): 7095–7102.

Kutluay S, Horoz S, Şahin Ö, Ekinci A, Ece M.Ş. 2021. Highly improved solar cell efficiency of Mn-doped amine groups-functionalized magnetic Fe_3O_4@SiO_2 nanomaterial. International Journal of Energy Research, 45(14): 20176–20185.

Li Z, Liu R, Liu D, Zhang Y, Si T, Li Y. 2021. Three-dimensional porous cobalt as an efficient catalyst for hydrogen production by $NaBH_4$ hydrolysis. Reaction Kinetics, Mechanisms and Catalysis, 134(2): 665–675.

Nabid M.R, Bide Y, Kamali B. 2019. Hydrogen release from sodium borohydride by Fe_2O_3@nitrogen-doped carbon core-shell nanosheets as reasonable heterogeneous catalyst. International Journal of Hydrogen Energy, 44(47): 25662–25670.

Özkan G, Akkuş M.S, Özkan G. 2019. The effects of operating conditions on hydrogen production from sodium borohydride using Box-Wilson optimization technique. International Journal of Hydrogen Energy, 44(20): 9811–9816.

Öztürk O.F, Demirci S, Sengel S.B, Sahiner N. 2018. Highly regenerable ionic liquid microgels as inherently metal-free green catalyst for H_2 generation. Polymers for Advanced Technologies, 29(5): 1426–1434. https://doi.org/10.1002/pat.4254

Packiaraj R, Devendran P, Venkatesh K.S, Asath bahadur S, Manikandan A, Nallamuthu N. 2019. Electrochemical ınvestigations of magnetic Co_3O_4 nanoparticles as an active electrode for supercapacitor applications. Journal of Superconductivity and Novel Magnetism, 32(8): 2427–2436.

Patel N, Santini A, Bello V, Mattei G, Miotello A. 2013. Cobalt/cobalt oxide nanoparticles-assembled coatings with various morphology and composition synthesized by pulsed laser deposition. Surface and Coatings Technology, 235: 784–791.

Pfeil T.L, Pourpoint T.L, Groven L.J. 2014. Effects of crystallinity and morphology of solution combustion synthesized Co_3O_4 as a catalyst precursor in hydrolysis of sodium borohydride. International Journal of Hydrogen Energy, 39(5): 2149–2159.

Ro G, Hwang D.K, Kim Y. 2019. Hydrogen generation using Pt/Ni bimetallic nanoparticles supported on Fe_3O_4@SiO_2@TiO_2 multi-shell microspheres. Journal of Industrial and Engineering Chemistry, 79:

364–369.

Ugale A.D, Ghodke N.P, Kang G.S, Nam K.B, Bhoraskar S.V, Mathe V.L, Yoo J.B. 2022. Cost-effective synthesis of carbon loaded Co_3O_4 for controlled hydrogen generation via $NaBH_4$ hydrolysis. International Journal of Hydrogen Energy, 47(1): 16–29.

Vernekar A.A, Bugde S.T, Tilve S. 2012. Sustainable hydrogen production by catalytic hydrolysis of alkaline sodium borohydride solution using recyclable Co-Co_2B and Ni-Ni_3B nanocomposites. International Journal of Hydrogen Energy, 37(1): 327–334.

Wang Q, Wei L, Ma M, Liu H. 2020. Hydrogen generation from the hydrolysis of sodium borohydride using Co_3O_4 hollow microspheres as high-efficient catalyst precursor synthesized by facile bio-template method. Energy Sources, Part A: Recovery, Utilization and Environmental Effects, 1–10.

Wei Y, Wang M, Fu W, Wei L, Zhao X, Zhou X, Ni M, Wang H. 2020. Highly active and durable catalyst for hydrogen generation by the $NaBH_4$ hydrolysis reaction: CoWB/NF nanodendrite with an acicular array structure. Journal of Alloys and Compounds, 836: 155429.

Yang L, Fan C, Zhang J, Zhang F, Li R, Yi S, Sun Y, Dong H. 2021. Poly(acrylic acid)-modified silica nanoparticles as a nonmetal catalyst for $NaBH_4$ methanolysis. International Journal of Hydrogen Energy, 46(45): 23236–23244.

Yolcular S, Karaoglu S. 2020. Hydrogen generation from sodium borohydride with cobalt boride catalysts. Alkü Fen Bilimleri Dergisi, 2(2): 84–96.

DRAFT

ORAL PRESENTATION

Hydrobiology and ecology in the context of climate change: the future of aquatic ecosystems

Abuzer Çelekli[1,2] (*ORCID: https://orcid.org/0000-0002-2448-4957*), Özgür Eren Zariç[1,2*] (*ORCID: https://orcid.org/0000-0001-5293-871X*)

[1]Gaziantep University, Environmental Research Center (GÜÇAMER), Gaziantep, Turkey.
[*2]Gaziantep University, Faculty of Art and Science, Department of Biology, Gaziantep, Turkey.

*Corresponding author e-mail: ozgurerenzaric@gmail.com

Abstract

Climate change profoundly affects aquatic ecosystems, with consequential impacts on hydrobiology and global biodiversity. This review elucidates the multifaceted relationships between climate change, aquatic ecosystem health, and human society. By synthesizing current research and case studies, it extensively examines the physical impacts of climate change, including sea level rise, temperature fluctuations, and extreme weather events on aquatic environments. The review includes the ecological responses, identifying key areas such as the effects on aquatic organisms, habitat alteration, spread of invasive species, and water quality changes. Economic and social implications are also critically evaluated, focusing on fisheries, aquaculture, water provision, and recreational and cultural values. An in-depth analysis of adaptation and mitigation strategies, technological solutions, and policy recommendations is also presented to address these challenges. Hydrobiology, as the nexus of the study, offers critical insights into the interactions and complexities within aquatic ecosystems, underscoring the vital role of water organisms in maintaining ecosystem functions. Through the lens of hydrobiology, the review emphasizes the necessity for a comprehensive approach to understanding, predicting, and managing the changes occurring in aquatic environments. This review is a foundational resource for researchers, policymakers, environmental managers, and other stakeholders, aiming to foster informed decisions and effective strategies for conserving and sustainably managing aquatic ecosystems in the face of climate change. By highlighting the interconnectedness of climate, water, biology, and human society, it underscores the urgent need for integrative and collaborative efforts to protect the vitality and resilience of our planet's aquatic life.

Keywords: Aquatic ecosystems, Climate change, Hydrobiology, Mitigation.

INTRODUCTION

Climate change, stemming from increased greenhouse gas emissions, severely impacts aquatic environments, including oceans, lakes, rivers, and wetlands (Harley et al., 2006). These systems are susceptible to temperature shifts, altered precipitation patterns, and sea-level changes. The field of hydrobiology, which examines biological phenomena within aquatic settings, is vital in understanding these effects. It aids in exploring the complex relationships shaping aquatic ecosystems and their responses to climate change. This review analyzes the multifaceted consequences of climate change on aquatic ecosystems by focusing on the interplay between hydrobiology and ecology. By synthesizing current research and case studies, the review aims to contribute to developing strategies for adapting and conserving these crucial systems and informing policy and management. It underscores the urgency in understanding and safeguarding aquatic ecosystems in a rapidly changing climate, recognizing their vital role in human sustenance and livelihood, including migration (Çelekli et al., 2023).

MATERIALS AND METHODS

This review's methodological approach offers a comprehensive view of the relationship between climate change and aquatic ecosystems. Using multiple databases like PubMed, Web of Science, and Google Scholar, and government reports and conference proceedings, a meticulous search was conducted with keywords related to hydrobiology, ecology, and aquatic ecosystems. Studies were chosen based on relevance, publication date within the last twenty-five years, and English availability. Exclusions were made for non-aligned materials, and quality assessment ensured methodological rigor. The data extraction involved gathering critical

information, leading to thematic synthesis across physical impacts, biodiversity, water quality, socio-economic impacts, and adaptation strategies. Comparative analysis was used to cross-validate findings across studies and regions, guided by systematic review protocols, providing an integrative view of current knowledge. The review synthesizes existing research and sets the stage for future inquiries, policy-making, and practical interventions, forming a solid base to understand and tackle aquatic ecosystems' challenges.

PHYSICAL IMPACTS OF CLIMATE CHANGE

Sea Level Changes

One of the most observable physical impacts of climate change on aquatic ecosystems is the alteration in sea levels. Rising sea levels, attributed to the melting of polar ice caps and the thermal expansion of seawater, have profound implications for coastal ecosystems; these changes threaten to inundate coastal habitats, erode shorelines, increase salinity in estuaries, and create more frequent and severe coastal flooding (Hallegatte et al., 2011).

Water Temperature Fluctuations

Water temperature is a critical factor in determining the health and function of aquatic ecosystems. Climate change has led to noticeable water temperature fluctuations in marine and freshwater environments. Warming waters can affect metabolic rates, breeding cycles, and migration patterns of aquatic organisms; for example, increased temperatures can cause corals to bleach, disrupt fish migration routes, and alter the distribution of phytoplankton, which is the basis of many aquatic food webs and is of economic and biotechnological importance, such as the removal of harmful dyes (Çelekli et al., 2011; Doney et al., 2012; Frederick & Major, 1997; Zariç et al., 2022).

BIODIVERSITY AND ECOSYSTEM RESPONSES

Effects on Aquatic Organisms

Climate change's physical impacts trigger biological responses in aquatic ecosystems. Warming waters and other altered environmental conditions directly affect species by challenging their physiological limits, leading to differences in reproduction, survival, and behavior (Reid et al., 2019). Fish and invertebrate species migrate to calmer waters, forming new community structures. Some species suffer from increased mortality rates due to thermal stress (Sebens, 1994). Warmer water temperatures can cause coral reef bleaching, a stress response that can lead to coral mortality and the subsequent decline of species dependent on coral habitats (Lough, 2000). Changes in water temperature and pH levels influence phytoplankton and zooplankton dynamics, affecting food availability for higher trophic levels (Strecker et al., 2004).

Habitat Loss and Alteration

The physical changes brought about by climate change often lead to significant alterations or even complete loss of critical habitats. Rising sea levels and increased storm surges can erode or inundate coastal wetlands, vital for many fish and bird species (Fujii, 2012). Melting ice in polar regions jeopardizes species relying on ice-covered areas, including specific seals, penguins, and polar bears (Ainley et al., 2003).

Spread of Invasive Species

Altered conditions favor non-native species, enabling them to outcompete natives. In freshwater ecosystems, invasive species may thrive in warmer temperatures; in marine systems, changes in currents and temperatures can spread invasives, disrupting local ecology (Dukes & Mooney, 1999). These profound and complex impacts lead to intricate biological responses that vary across ecosystems (Kernan, 2015). Preserving aquatic life's diversity and ecological functions requires collaboration across various scientific fields. Research sheds light on strategies to protect and manage these creatures. Dealing with the effects of climate change on water health requires cooperation among different disciplines, governments, industries, and communities. This collaboration aims to develop adaptive strategies to sustain water resources.

Changes in Water Quality

Extreme weather events, such as intense rainfall, can cause increased runoff from agricultural and urban areas, carrying pollutants like nutrients, pesticides, and heavy metals into water bodies (Agrawal, 1999). Floods can also increase sedimentation, which may smother sensitive habitats like coral reefs and seagrass beds; increased

nutrient loads can lead to eutrophication, where excessive algae growth depletes oxygen levels in the water, leading to "dead zones" where aquatic life cannot survive (van Beusekom, 2018).

DRAFT

Chemical Alterations

Ocean Acidification: Increased atmospheric CO_2 leads to higher concentrations of carbonic acid in oceans, lowering pH levels. This ocean acidification affects organisms that rely on calcium carbonate for their shells and skeletons, such as mollusks and corals (Guinotte & Fabry, 2008). Changes in temperature and flow regimes can also affect the chemical composition of rivers and lakes, potentially influencing the availability of essential nutrients and the concentration of harmful substances.

ECONOMIC AND SOCIAL IMPACTS

The impact of climate change on aquatic ecosystems goes far beyond just the environment; it also profoundly affects human communities and economies. The way climate, water, living organisms, and human actions are interconnected creates complicated problems but also opportunities. This means that changes in the climate can have widespread effects on everything from the water we drink to the economy we rely on, and it requires careful consideration and planning to manage these interconnected challenges.

Fisheries and Aquaculture

Changes in water temperature and currents can cause fish populations to shift, affecting local fisheries and potentially leading to conflicts over fishing rights and territories (Munday et al., 2008). Coastal communities dependent on fishing may face significant economic challenges if fish stocks decline or move to different areas. Climate-related water quality and temperature changes may affect aquaculture operations, influencing growth rates and increasing disease susceptibility.

Effects on Tourism and Recreation

Rising sea levels and increased storm activities may threaten coastal tourist destinations, impacting local economies heavily dependent on tourism (Scott et al., 2012). Using the climate change process in biological urbanization engages nature-environment communication (Çelekli et al., 2023). Changes in water quality and biodiversity loss may diminish the appeal of lakes, rivers, and coastal areas for recreational activities such as fishing, boating, and diving.

Water Security and Public Health

Water quality and availability changes affect drinking water supplies, necessitating costly treatment or transportation and impacting human health (Mishra, 2023). Altered ecosystems can influence the spread of waterborne diseases, potentially creating new public health challenges.

ADAPTATION AND MITIGATION STRATEGIES

Adaptation strategies are plans that help us get used to the new conditions created by climate change. They aim to reduce any adverse effects and take advantage of possible benefits. In other words, these strategies help us prepare for and respond to changes in the climate so that we can lessen the damage and find new ways to thrive.

Ecosystem-Based Approaches

Restoring and preserving natural barriers such as mangroves and wetlands to enhance coastal resilience.

Flexible Water Management

Adaptive water management plans for ensuring sufficient water quality and quantity under changing climatic conditions.

Community Engagement

Involving local communities in decision-making for culturally appropriate and effective local adaptation.

Mitigation Strategies

Mitigation strategies tackle the root causes of climate change, aiming to reduce emissions or enhance the sinks of greenhouse gases.

Sustainable Fisheries Management

Implementing energy-efficient fishing methods and sustainable aquaculture practices.

DRAFT

DRAFT

Renewable Energy Transition

Transitioning from fossil fuels to renewable energy sources in water management and related sectors.

Carbon Sequestration in Aquatic Ecosystems

Protecting and restoring blue carbon ecosystems for significant CO_2 sequestration.

Integrating Adaptation and Mitigation

A holistic approach integrating adaptation and mitigation can create more resilient and sustainable pathways.

Policy Integration

Incorporating climate considerations at all levels of policy-making for coordinated strategies.

Cross-Sector Collaboration

Engaging various sectors for cohesive, comprehensive solutions.

Investment in Research and Technology

Supporting research and innovation in climate science, ecology, and technology. The challenges of climate change in aquatic ecosystems require thoughtful, targeted adaptation and mitigation strategies. From ecosystem-based approaches to policy integration, these strategies offer pathways to resilience, vulnerability reduction, and sustainability. The collaborative, integrative, and forward-thinking approaches outlined here can create a future where human societies and aquatic ecosystems flourish in a changing climate.

CASE STUDIES

Mangrove Restoration in Vietnam

Coastal erosion and increased storm surges due to climate change threatened local communities.

Strategy: Restoration and protection of mangrove forests to act as natural barriers.

Outcome: Enhanced coastal resilience, protection of livelihoods, and additional benefits in biodiversity conservation (Tri et al., 1998).

Sustainable Fisheries Management in Iceland

Challenge: Overfishing and changing fish migration patterns influenced by climate change.

Strategy: Implementing quota systems and leveraging technology for real-time data collection and adaptive management.

Outcome: Sustainable fish stocks, economic stability for fishing communities, and a model for other nations (Chambers & Kokorsch, 2017).

Adaptive Water Management in Australia's Murray-Darling Basin

Challenge: Severe drought and altered precipitation patterns affecting water availability.

Strategy: Flexible water allocation policies, stakeholder engagement, and climate-informed planning.

Outcome: Improved water efficiency, support for agricultural sectors, and enhanced ecosystem health (Connell & Grafton, 2011).

Carbon Sequestration in the Blue Carbon Project, Indonesia

Challenge: Loss of carbon-sequestering ecosystems like mangroves and seagrass beds.

Strategy: Community-led conservation and restoration projects to enhance carbon sinks.

Outcome: Significant carbon sequestration, improved local livelihoods, and increased biodiversity (Sejati et al., 2020).

Climate-Resilient Tourism in the Maldives

Challenge: Rising sea levels and coral bleaching threaten tourism.

Strategy: Investments in coral reef restoration, sustainable tourism practices, and infrastructure adaptation.

Outcome: Preservation of the tourism industry, enhanced environmental stewardship, and global recognition as a leader in climate adaptation (Kagawa, 2022).

These case studies illuminate the real-world applications and successes of adaptation and mitigation strategies in addressing the challenges of climate change in aquatic ecosystems. They highlight the importance of context-specific solutions, interdisciplinary collaboration, community engagement, and innovative thinking. Moreover, they demonstrate that addressing climate change's impacts is about averting crises and seizing opportunities for sustainability, resilience, and social equity. By learning from these examples and scaling up similar efforts, we can foster a future where human communities and aquatic ecosystems thrive harmoniously with our changing climate.

PREDICTIONS AND RECOMMENDATIONS FOR THE FUTURE

As we move into the 21st century, climate change's effects on aquatic ecosystems will unfold in complex and interconnected ways. While there are uncertainties, several predictions can be made, and corresponding recommendations can guide our approach:

Increased Climate Variability

More frequent and severe weather events are likely, affecting water quality, availability, and ecosystem stability.

Continued Shifts in Biodiversity

Changes in temperature and chemistry will continue to alter species distributions and interactions within aquatic ecosystems.

Growing Socio-Economic Pressures

Coastal communities and those dependent on fisheries and aquaculture will likely face increasing economic challenges.

Technological and Policy Innovations

As awareness and understanding grow, expect increased innovation in technology and policy aimed at mitigation and adaptation.

Invest in Research and Monitoring

Continuous research and monitoring of aquatic ecosystems are crucial for understanding ongoing changes and implementing timely responses.

Promote Cross-Sector Collaboration

Encourage collaboration between governments, industries, researchers, and communities for comprehensive and integrative solutions.

Strengthen Local Community Engagement

Empower local communities through education, resources, and active participation in decision-making processes.

Implement Adaptive Management Strategies

Develop flexible, adaptive management plans that can be adjusted as new information and conditions emerge.

Prioritize Sustainable Development

Balance ecological conservation with economic development, focusing on long-term sustainability rather than short-term gains. The future of our aquatic ecosystems in the face of climate change is fraught with challenges, but it is also ripe with opportunities for innovation, collaboration, and transformation. The predictions and recommendations provided here offer a roadmap for navigating these complexities. Investing in research, fostering collaboration, engaging communities, adopting adaptive management, and prioritizing sustainability can construct a resilient and flourishing future for aquatic ecosystems and human societies. The task is substantial but within our reach with concerted effort, thoughtful planning, and a commitment to shared values and goals. The future of our aquatic ecosystems depends on our collective willingness to understand, adapt, innovate, and act with foresight, compassion, and wisdom.

RESULTS and DISCUSSION

In our exploration of climate change's multifaceted impacts on aquatic ecosystems, we synthesized a wide array of information from diverse disciplines such as hydrobiology, ecology, and economics. Our study's key findings include the physical alterations in aquatic environments, such as changes in temperature, salinity, and circulation patterns, which have significant and cascading consequences for both ecosystem health and human welfare. Climate change is leading to alterations in species distribution, abundance, and behavior, resulting in shifts in ecosystem structure and function. Our analysis identified significant alterations in water quality parameters such as nutrient loading, oxygen levels, and contaminant concentrations, potentially affecting human health and ecosystem sustainability. The economic ramifications of climate-induced changes in aquatic ecosystems have far-reaching impacts, affecting fisheries, tourism, and water supply, and disproportionately impacting marginalized communities. A range of successful case studies demonstrated effective, regionally-tailored adaptation and mitigation strategies that are not mutually exclusive but work most effectively when integrated. The complexity of climate change effects on aquatic ecosystems makes the development of a holistic and flexible strategy essential. Our findings underscore the urgency and shared responsibility to act. The success of adaptation and mitigation strategies depends on a nuanced understanding and tailored approaches to specific regional and community contexts. Solutions must align with both ecological imperatives and the social and economic realities of affected communities, recognizing the deeply interconnected nature of these systems. The case studies examined provide tangible blueprints for action, highlighting the need for innovation, investment, and cross-sector collaboration. Our research is more than an academic exercise; it represents a profound call to responsible stewardship and sustainable development.
In conclusion, the complex interconnections within our planet's aquatic ecosystems necessitate a unified and mindful approach. The findings of this study function both as a comprehensive guide and a call to action for aligning future development with the fragile beauty of these ecosystems. The decisions made today will determine the legacy left to future generations, posing both significant challenges and unique opportunities to create a sustainable and harmonious relationship with our aquatic environment.

CONCLUSION

Our study of aquatic ecosystems highlights the urgent need for a multifaceted approach, blending empathy, innovation, and stewardship. Reflecting fragility and resilience, these systems symbolize our interconnected fate with nature. Beyond scientific and economic considerations, we underscore a moral imperative to protect and sustain these vital resources. The human potential for positive change is evident in our capacity to adapt and innovate. Recommendations include the continued interdisciplinary research, community involvement, and aligned policies aimed at preserving the aquatic resources that sustain our planet. In embracing these principles, we commit to a future that honors the intricate balance of life, recognizing our shared responsibility to foster a sustainable world where human civilization and aquatic ecosystems can flourish.

REFERENCES

Agrawal, G. D. (1999). Diffuse agricultural water pollution in India. *Water Science and Technology, 39*(3), 33–47.

Ainley, D. G., Tynan, C. T., & Stirling, I. (2003). Sea ice: a critical habitat for polar marine mammals and birds. *Sea Ice: An Introduction to Its Physics, Chemistry, Biology, and Geology. Blackwell, Oxford, UK*, 240–266.

Çelekli, A., Tanriverdi, B., & Bozkurt, H. (2011). Predictive modeling of removal of Lanaset Red G on *Chara contraria*; kinetic, equilibrium, and thermodynamic studies. *Chemical Engineering Journal, 169*(1–3), 166–172.

Çelekli, A., Yaygır, S., & Zariç, Ö. E. (2023). A review of climate change-induced migration. *Acta Biologica Turcica, 36*(2), 1–3.

Çelekli, A., Yeşildağ, İ., Yaygır, S., & Zariç, Ö. E. (2023). Effects of urbanization on bioclimatic comfort conditions. *Acta Biologica Turcica, 36*(4), 2-1–9.

Chambers, C., & Kokorsch, M. (2017). The social dimension in Icelandic fisheries governance. *Coastal Management, 45*(4), 330–337.

Connell, D., & Grafton, R. Q. (2011). Water reform in the Murray-Darling Basin. *Water Resources Research,*

47(12).

Doney, S. C., Ruckelshaus, M., Emmett Duffy, J., Barry, J. P., Chan, F., English, C. A., Galindo, H. M., Grebmeier, J. M., Hollowed, A. B., & Knowlton, N. (2012). Climate change impacts on marine ecosystems. *Annual Review of Marine Science*, *4*, 11–37.

Dukes, J. S., & Mooney, H. A. (1999). Does global change increase the success of biological invaders? *Trends in Ecology & Evolution*, *14*(4), 135–139.

Frederick, K. D., & Major, D. C. (1997). Climate change and water resources. *Climatic Change*, *37*(1), 7–23.

Fujii, T. (2012). Climate change, sea-level rise and implications for coastal and estuarine shoreline management with particular reference to the ecology of intertidal benthic macrofauna in NW Europe. *Biology*, *1*(3), 597–616.

Guinotte, J. M., & Fabry, V. J. (2008). Ocean acidification and its potential effects on marine ecosystems. *Annals of the New York Academy of Sciences*, *1134*(1), 320–342.

Hallegatte, S., Ranger, N., Mestre, O., Dumas, P., Corfee-Morlot, J., Herweijer, C., & Wood, R. M. (2011). Assessing climate change impacts, sea level rise and storm surge risk in port cities: a case study on Copenhagen. *Climatic Change*, *104*, 113–137.

Harley, C. D. G., Hughes, A. R., Hultgren, K. M., Miner, B. G., Sorte, C. J. B., Thornber, C. S., Rodriguez, L. F., Tomanek, L., & Williams, S. L. (2006). Erratum: The impacts of climate change in coastal marine systems (Ecology Letters (2006) 9 (228-241)). *Ecology Letters*, *9*(4), 500.

Kagawa, F. (2022). The Heat is On! Towards a Climate Resilient Education System in India. *Kathmandu: UNICEF Regional Office for South Asia*, *164*(14), 44.

Kernan, M. (2015). Climate change and the impact of invasive species on aquatic ecosystems. *Aquatic Ecosystem Health & Management*, *18*(3), 321–333.

Lough, J. M. (2000). 1997-98: Unprecedented thermal stress to coral reefs? *Geophysical Research Letters*, *27*(23), 3901–3904.

Mishra, R. K. (2023). Fresh water availability and its global challenge. *British Journal of Multidisciplinary and Advanced Studies*, *4*(3), 1–78.

Munday, P. L., Jones, G. P., Pratchett, M. S., & Williams, A. J. (2008). Climate change and the future for coral reef fishes. *Fish and Fisheries*, *9*(3), 261–285.

Reid, G. K., Gurney-Smith, H. J., Marcogliese, D. J., Knowler, D., Benfey, T., Garber, A. F., Forster, I., Chopin, T., Brewer-Dalton, K., & Moccia, R. D. (2019). Climate change and aquaculture: considering biological response and resources. *Aquaculture Environment Interactions*, *11*, 569–602.

Scott, D., Simpson, M. C., & Sim, R. (2012). The vulnerability of Caribbean coastal tourism to scenarios of climate change related sea level rise. *Journal of Sustainable Tourism*, *20*(6), 883–898.

Sebens, K. P. (1994). Biodiversity of coral reefs: what are we losing and why? *American Zoologist*, *34*(1), 115–133.

Sejati, A. W., Buchori, I., Kurniawati, S., Brana, Y. C., & Fariha, T. I. (2020). Quantifying the impact of industrialization on blue carbon storage in the coastal area of Metropolitan Semarang, Indonesia. *Applied Geography*, *124*, 102319.

Strecker, A. L., Cobb, T. P., & Vinebrooke, R. D. (2004). Effects of experimental greenhouse warming on phytoplankton and zooplankton communities in fishless alpine ponds. *Limnology and Oceanography*, *49*(4), 1182–1190.

Tri, N. H., Adger, W. N., & Kelly, P. M. (1998). Natural resource management in mitigating climate impacts: the example of mangrove restoration in Vietnam. *Global Environmental Change*, *8*(1), 49–61.

van Beusekom, J. E. E. (2018). Eutrophication. *Handbook on Marine Environment Protection: Science, Impacts and Sustainable Management*, 429–445.

Zariç, Ö. E., Yeşildağ, İ., Yaygır, S., & Çelekli, A. (2022). Removal of Harmful Dyes Using Some Algae. *3rd*

International Congress on Plant Biology; Rize, Turkey, 1st Edition, 173.

DRAFT

ORAL PRESENTATION

Cr(VI)'nın sulu çözeltiden FIBAN AK-22-1 anyon değiştirici fiber ile uzaklaştırılması

İlker AKIN*[1] (https://orcid.org/0000-0002-8683-0210) ,
Faysal Selimoğlu[1](https://orcid.org/0000-0003-3798-9054)

*[1]Necmettin Erbakan Üniversitesi, Fen Fakültesi, Biyoteknoloji Bölümü, Konya,Türkiye

*ilker0997@gmail.com

Özet

FIBAN AK-22-1 anyon değiştirici fiberler kullanılarak sulu çözeltilerden Cr(VI)'nın değişen pH, temas süresi, başlangıç Cr(VI) konsantrasyonu, adsorban dozajı ve sıcaklıkta gibi parametreler ile kesikli adsorpsiyonu incelenmiştir. Anyon değiştirici fiber tarafından Cr(VI) adsorpsiyonu için optimum pH aralıkları 1.0-2.0 olarak bulunmuştur. Anyon değiştirici fiber için maksimum Cr(VI) adsorpsiyonu 30 dakikada meydana gelmiştir. Fiber tarafından Cr(VI) tutumu kinetiği, ikinci dereceden kinetik modeli ile gerçekleşmiştir. Adsorpsiyon izotermleri, Langmuir, Freundlich ve üç parametreli Redlich-Peterson modelleri kullanılarak analiz edilmiştir. Adsorpsiyon için termodinamik parametreler (serbest enerji değişimi ΔG^o, entropi değişimi ΔS^o ve entalpi değişimi ΔH^o) da değerlendirilmiştir. Sıcaklıktaki artış, Cr(VI)'nın anyon değiştirici fiber tarafından adsorplanması için denge sabitinin (Kc) değerinde bir artışa neden olmuştur.

Anahtar Kelimeler: Anyon değiştirici fiber; Adsorpsiyon; Krom

Removal of Cr(VI) from aqueous solution by FIBAN AK-22-1 anion exchange fiber

Abstract

By using FIBAN AK-22-1 anion exchange fibers, batch adsorption of Cr(VI) from aqueous solutions with parameters such as changing pH, contact time, initial Cr(VI) concentration, adsorbent dosage and temperature was investigated. The optimum pH ranges for Cr(VI) adsorption by the anion exchange fiber were found to be 1.0-2.0. Maximum Cr(VI) adsorption for the anion exchange fiber occurred at 30 minutes. The kinetics of Cr(VI) uptake by the fiber was realized with the second order kinetic model. Adsorption isotherms were analyzed using Langmuir, Freundlich and three parameter Redlich-Peterson models. Thermodynamic parameters (free energy change ΔG^o, entropy change ΔS^o and enthalpy change ΔH^o) for adsorption were also evaluated. The increase in temperature caused an increase in the value of the equilibrium constant (Kc) for Cr(VI) to be adsorbed by the anion exchange fiber.

Keywords: Anion exchange fiber; Adsorption; Chromium

GİRİŞ

Deri tabaklama, metalürji, galvanik kaplama vb. gibi çeşitli endüstrilerde kromun yaygın kullanımı, kromun çevreye salınmasına neden olmuştur. Bu endüstrilerden üretilen atık sulardaki Cr(VI) varlığı, sağlık ve çevre üzerindeki zararlı etkilerinden dolayı günümüzde önemli bir endişe haline gelmiştir. $HCrO_4^-$, $Cr_2O_7^{2-}$ ve CrO_4^{2-} gibi Cr(VI) türlerinin oksitleme potansiyeli onları bakteri, bitki ve hayvanlar için oldukça toksik hale getirir [ATSDR, 2000; Edebali ve Pehlivan,2010]. Cr(VI)'nın iç yüzey sularına deşarj limiti 0,1 mg/L'dir. İçme suyu için maksimum Cr(VI) kirletici seviyesi 0,05 mg/L'dir [US EPA,1990]. Bu nedenle sulardan Cr(VI) giderimi çevre sağlığı açısından önemlidir. Geri kazanım için farklı metodolojiler (yani, iyon değişimi [Göde ve Pehlivan, 2005], solvent ekstraksiyonu [Venkateswaran ve Palanivelu, 2004], dağılmayan solvent ekstraksiyonu [Alonso ve ark. 1999], membran bazlı yöntemler [Hafiane ve ark., 2000; Cengeloglu ve ark., 2003; Tor ve ark., 2004; Yılmaz ve ark., 2008; Arslan ve ark., 2009; Venkateswaran ve Palanivelu, 2005] ve adsorpsiyon [Kobya, 2004; Khezami ve Capart,2005; Arslan ve ark., 2010] kullanılmıştır. veya Cr(VI)'nın sulu çözeltilerden çıkarılması. Bununla birlikte, iyon değişimi dışındaki bu tekniklerin, eksik metal giderimi,

DRAFT

pahalı ekipman ve izleme sistemi gereksinimleri, yüksek reaktif veya enerji gereksinimleri veya bertaraf edilmesi gereken toksik çamur veya diğer atık ürünler gibi önemli dezavantajları vardır [Dragan ve ark., 2003]. Son yıllarda, bir tip adsorban olarak cleating ve iyon değiştirici fiberler, geniş özgül yüzey, yüksek şişme kapasitesi, mükemmel mekanik dayanım gibi avantajları nedeniyle su ve atık sudan anyonların uzaklaştırılması için yaygın olarak kullanılmaktadır. Örneğin, Liu ve ark. (1999) atık sudan Cu(II), Zn(II), Cd(II) ve Hg(II)'nin poli(akrilaminofosfonik)-tipi şelatlama fiber ile uzaklaştırılmasını incelediler. Park ve Na (2006), akrilik asitle aşılanmış polipropilen dokunmamış fiber ve iyon değiştirme özelliğinin aminasyonuyla bir anyon değiştirici fiberin hazırlanışını bildirmiştir. Huang ve ark. (2009), kimyasal olarak modifiye edilmiş kavak ağacı fiberleri tarafından bakırın emilmesini tanımlamıştır. Başka bir çalışmada Zhang ve ark., (2008) tarafından güçlü bir bazik anyon değiştirici hazırlanmış, stirenin poli(tetrafloroetilen) fiber üzerine radyasyona bağlı aşılanmasına göre $Cr_2O_7^{2-}$ ve MnO_4^{2-} için adsorplama özelliği incelenmiştir. Bu çalışmanın temel amacı, aminokarboksil grubu içeren FIBAN AK-22-1 iyon değiştirici fiberle sulu çözeltiden Cr(VI) giderimi için denge ve kinetik parametrelerinin araştırılmasıdır. Başlangıç Cr(VI) konsantrasyonu, temas süresi, pH, fiber miktarı ve sıcaklık gibi adsorpsiyonu etkileyen parametreler araştırılmış ve sonuçlar tartışılmıştır.

MATERYAL VE METOT

Bu çalışmada kullanılan FIBAN AK-22-1 anyon değiştirici fiber Eichrom Technologies Inc.'den (Ecofil, Minsk, Beyaz Rusya) satın alınmıştır. Fiberin temel fiziko-kimyasal özellikleri Tablo 1'de verilmiştir. Cr(VI) stok solüsyonu, K_2CrO_4'ün çift damıtılmış suda çözülmesiyle hazırlandı. Kullanılan tüm kimyasallar analitik derecedeydi ve Merck Co.'dan (Darmstadt, Almanya) temin edildi. Deneyler boyunca tüm çözeltileri hazırlamak için çift damıtılmış su kullanıldı.

Tablo 1. FIBAN AK-22-1 anyon değiştirici fiberin özellikleri ve türleri

FIBAN AK-22-1 anyon değiştirici fiber	
Tip	Zayıf baz anyon değiştirici
Matrix	Poliakrilonitrilik fiber
Fonksiyonel grup	$-NH_2$, -COOH
Sevk edilen iyonik form	H^+, Cl^-
Amino üzerinde toplam değişim kapasitesi	-3.8 meq/g
–COOH	-1.0 meq/g üzerinde
Stabilite (pH aralığında)	1-8
Nem tutma kapasitesi	%80

Kesikli adsorpsiyon deneyleri

Cr(VI) çözeltileri, stok çözelti (K_2CrO_4'ten hazırlanan 1000 mg/L Cr(VI)) istenen konsantrasyonlara seyreltilerek hazırlandı. İyonmetre (Orion EA940, ABD) kullanılarak belirlenen çözeltilerin pH'ları 0,1 M NaOH veya 0,1 M HCl çözeltileri ile istenilen düzeye getirildi. Deneyler, bilinen miktarda fiberle 20 mL Cr(VI) solüsyonunda sıcaklık kontrollü orbital çalkalayıcıda (Gallenkamp, UK) 220 rpm'de ve 25±1 ^oC'de denge süresi boyunca çalkalanmasıyla kesikli sorpsiyon tekniğine göre gerçekleştirilmiştir. Daha sonra solüsyon Whatman No.42 filtre kağıdından süzüldü. Temas süresinin (5-240 dakika), başlangıç Cr(VI) konsantrasyonunun (5-1000 mg/L) ve adsorbent miktarının (1.5-10 g/L) ve sıcaklığın (25-65 ^oC) etkileri de incelenmiştir. FIBAN AK-22-1 anyon değiştirici fiber. Adsorplanan Cr(VI) miktarı Denk. (1).

$$q = (C_o - C_e).V/m \quad (1)$$

Burada q, Cr(VI) emilir (mg/g); C_o, Cr(VI)'nın başlangıç konsantrasyonu (mg/L); C_e, dengede çözeltideki Cr(VI) konsantrasyonu (mg/L); V, çözelti hacmi (L); m, fiber dozajı (g). Cr(VI) konsantrasyonu, hava-asetilen alevinde Atomik Absorpsiyon Spektrometresi (ContrAA 300, Analytik jena) kullanılarak belirlendi. Tüm

deneyler, iki kopya veya üç kopya halinde gerçekleştirildi ve bir deney aralığı içindeki tekrar numuneler arasındaki varyasyonlar %3'ten azdı.

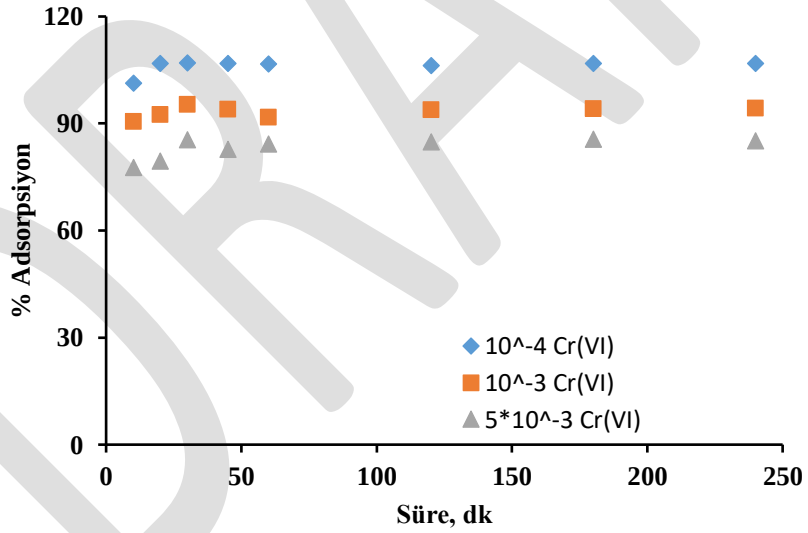
BULGULAR ve TARTIŞMA

Temas süresinin ve kinetik değerlendirmenin etkisi

Temas süresinin bir fonksiyonu olarak Cr(VI)'nın anyon değişimli fiber tarafından uzaklaştırılması Şekil 1'de gösterilmektedir. Anyon değiştirici fiber için sorpsiyon dengesine 30 dakikada ulaşılmıştır. Ayrıca denge süresinin ilk Cr(VI) konsantrasyonuna bağlı olmadığı da bulundu. Barassi ve ark., (2009) göre adsorban yüzeyindeki fonksiyonel grubun konsantrasyonu, yalancı ikinci dereceden kinetik model aracılığıyla kimyasal adsorplama hızının değerlendirilmesine izin verir. Bu nedenle, deneysel veriler Denklem. (2) [Ho ve McKay, 1999] göre değerlendirilmiştir.

$$t/qt = 1/(k_2 . qe^2) + t/ qe \quad (2)$$

qe ve qt sırasıyla dengede ve t zamanında (dk) emilen Cr(VI) miktarları (mg/g) olduğunda, k_2 yalancı ikinci dereceden kemisorpsiyonun hız sabitidir (g/(mg.dk)). İncelenen konsantrasyonlar için, hız sabiti (k_2) ve teorik denge sorpsiyon kapasiteleri, qe (hesaplanan), sözde ikinci dereceden kinetik modelin lineer çizimlerinin eğiminden ve kesişme noktasından hesaplandı ve Tablo 2'de verildi. belirleme (R^2) ve teorik ve deneysel qe değerleri, Cr(VI)'nın anyon değitirici fiber tarafından adsorplamasının ikinci dereceden tip reaksiyon kinetiğini takip ettiğini gösterdi (Tablo 2).



Şekil 1. Temas süresinin FIBAN AK-22-1 anyon değiştirici fiber tarafından Cr(VI) adsorpsiyonuna etkisi (başlangıç Cr(VI konsantrasyonu): 5, 50 ve 250 mg/L; fiber miktarı: 2,5 g /L, çözelti pH'ı: 1,0, sıcaklık: 25±1 ^oC; çalkalama hızı: 220 rpm).

Tablo 2. Yalancı ikinci dereceden kinetik model için adsorpsiyon hızı sabiti değerleri.

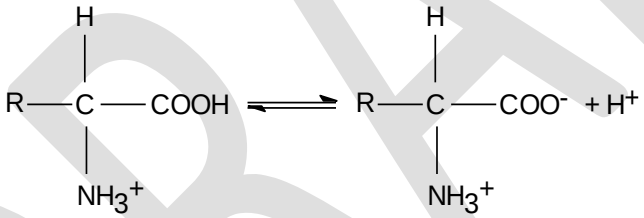
	FIBAN AK-22-1 anyon değiştirici fiber		
	5 mg/L	**50 mg/L**	**250 mg/L**
q_e (hesaplanan) (mg/g)	2.085	19.646	89.285
q_e (deneysel) (mg/g)	2.512	20.105	90.251

k_2 [g/(mg. min)]	1.44	0.116	0.010
R^2	0.999	1.000	1.000

pH'ın etkisi

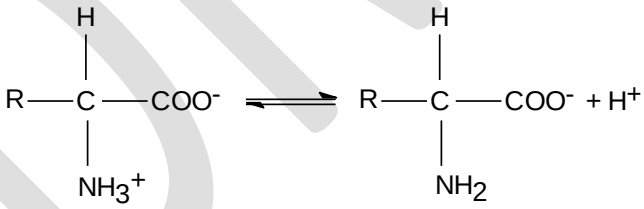
Cr(VI) iyonlarının sulu çözeltilerde pH ve konsantrasyona bağlı olarak çeşitli anyonik formlarda ($Cr_2O_7^{2-}$, $HCrO_4^-$, CrO_4^{2-} ve $HCr_2O_7^-$) bulunduğu bilinmektedir. CrO_4^{2-} anyonu bazik veya hafif asidik çözeltilerde hakimdir. Ayrıca, Cr(VI) konsantrasyonu yaklaşık 1000 mg/L'yi aştığında, $Cr_2O_7^{2-}$ asidik sulu çözeltide HCrO4- anyonuna dönüşür [Pehlivan ve Çetin, 2009]. Bu nedenle, bu çalışmada, Cr(VI)'nın çalışılan başlangıç konsantrasyonları ≤ 1000 mg/L olduğundan, Cr(VI) iyonları sulu çözeltide $HCrO_4^-$ olarak bulunacaktır. Sulu çözeltilerde anyon değiştirici ve iyonların birbirleriyle etkileşime girebilmesinin iki ana yolu vardır. En yaygın yol, iyon değişimini veya metal iyonlarının şelatlanmasını içerir. Bu etkileşimler, mevcut fonksiyonel gruplar açısından reçine yapısı ile karakterize edilir [Pehlivan ve Çetin, 2009]. Ek olarak, düşük pH değerlerinde, eşanjörün yüzeyi hidronyum iyonları ile çevrili olacaktır, bu da Cr(VI)'nın sorbentin bağlanma bölgeleri ile etkileşimini daha büyük çekici kuvvetlerle arttıracaktır. Bununla birlikte, pH arttıkça, sorbentler üzerindeki genel yüzey yükü negatif hale geldi ve iyon değişimi azaldı. Şekil 2 ayrıca, çözelti pH'ının, Cr(VI)'nın anyon değiştirici fiber tarafından adsorplanması üzerindeki etkisini göstermektedir. Anyon değiştirici reçinenin aksine, Cr(VI) için daha yüksek adsorpsiyon etkinliğinin pH 1.0-2.0 aralığında elde edildiği bulunmuştur. Ayrıca, çözelti pH'ındaki bir artış, anyon değiştirici fiberin assorpsiyon kabiliyetini azaltmıştır. pH bağımlılığından, çözelti ile fiberin amino ve karboksil grupları arasındaki etkileşimler, Denklemler izlenerek ifade edilebilir. (3) ve (4), solüsyondaki fiberin amino ve karboksil gruplarının protonasyon ve deprotonasyon reaksiyonlarını gösterir.

Karboksil grubu



pKa = 1.8 (3)

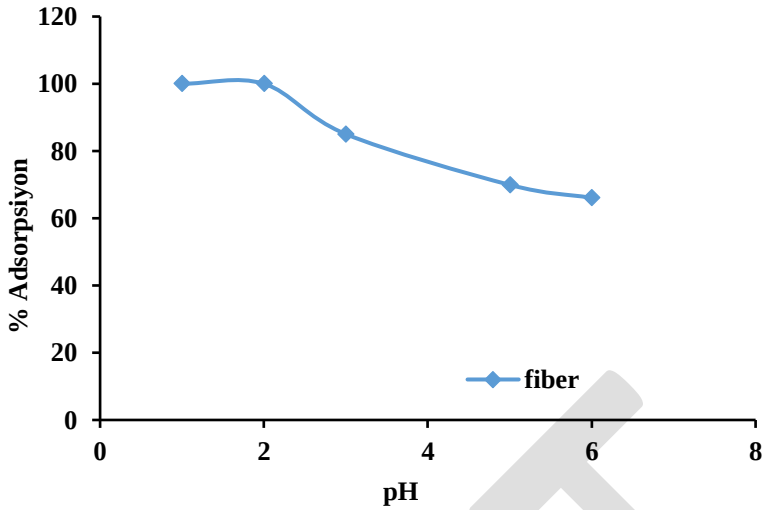
Amino grubu



pKa= 8.0 (4)

Düşen çözelti pH'ı ile, özellikle pH ≤2.0'da, Denk. (3) karboksil grubunun pKa değeri 1.8 olduğu için sola doğru ilerlemiştir [Troy, 2006]. Bu nedenle pH≤2.0'da fiberin HCrO4− iyonu ile $–NH_3^+/Cl^-$ grubu arasında iyon değişimi gerçekleşmiştir. Çözelti pH'ı 1,8 ve 8,0 arasında, fiber matrisinde protonlanmış amino ($–NH_3^+$) ve deprotanatlanmış karboksil ($-COO^-$) grupları bulunur. Bu nedenle, 2.0-8.0 pH aralığında, protonu giderilmiş karboksil grubu ile $HCrO_4^-$ arasındaki olası bir itme kuvveti sorpsiyonu azaltmıştır. Böylece FIBAN AK-22-1 anyon değiştirici fiber ile Cr(VI)'nın etkin adsorpsiyonu için optimal pH aralıkları 1.0-2.0 olmuştur.

DRAFT



Şekil 2. FIBAN AK-22-1 anyon değiştirici fiber tarafından Cr(VI) adsorpsiyonuna pH'ın etkisi (başlangıç Cr(VI konsantrasyonu): 25 mg/L; fiber miktarı: 2,5 g/ L; temas süresi: 1 saat; sıcaklık: 25±1 ˚C; çalkalama hızı: 220 rpm).

Adsorpsiyon izoterm modelleri

Adsorpsiyon izotermlerinin analizi tasarım amaçları için önemlidir. Bu nedenle deneysel veriler, Langmuir, Freundlich ve Redlich-Peterson izotermlerini içeren iyi bilinen adsorpsiyon izoterm modelleri ile analiz edilmiştir. Langmuir izotermi, adsorpsiyon yüzeylerinin tek tabakalı kaplamasını modeller ve adsorpsiyonun adsorbanın yapısal olarak homojen bir yüzeyinde gerçekleştiğini varsayar. Bu izoterm Denklem olarak verilir. (5) [Langmuir,1916].

$$q_e = Q_o.b.C_e/(1+b.C_e) \qquad (5)$$

Langmuir izoterm modelinin lineer formu Denklem. (6);

$$C_e/q_e = (1/Q_o.b)+(C_e/q_e) \qquad (6)$$

C_e, dengede Cr(VI) iyonunun konsantrasyonu (mg/L) olduğunda, Q_o adsorbanın tek tabaka kapasitesi (mg/g) ve b, Langmuir sorpsiyon sabitidir (L/mg). C_e/q_e'ye karşı C_e grafiği düz bir çizgi verir (Şekil 3 (a)) ve Q_o ve b değerleri sırasıyla grafiğin eğiminden ve kesişme noktasından hesaplanabilir.

Freundlich denklemi, çok katmanlı sorpsiyonu modellemek ve heterojen yüzeyler üzerindeki sorpsiyon için türetilmiştir. Freundlich modeli Denklem olarak formüle edilir. (7) [Freundlich,1906].

qe = k. C_e 1/n (7)

Freundlich denkleminin lineerleştirilmiş formu aşağıdaki denklemle verilir:

$$\log q_e = \log k + (1/n)\log C_e \qquad (8)$$

C_e denge konsantrasyonu (mg/L) olduğunda, k kabaca adsorpsiyon kapasitesinin [(mg/g).(mg/L)-1/n] bir göstergesidir ve n ampirik bir parametredir. Denklem(9)'a göre log qe'nin log C_e'ye karşı grafiği bir doğru verir (Şekil 3 (b)) ve k ve n değerleri bu düz çizginin kesişme noktası ve eğiminden hesaplanır, sırasıyla.

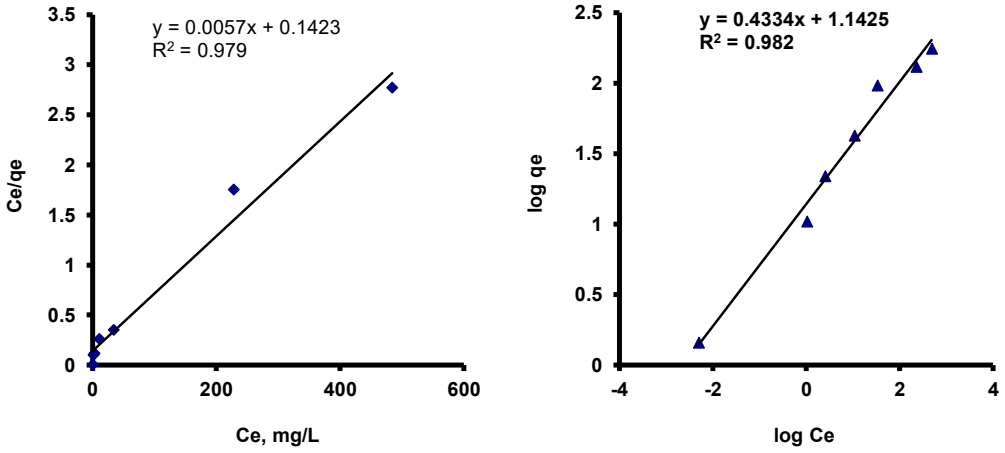
Redlich-Peterson izotermi [Redlich ve Peterson, 1959], payda konsantrasyona doğrusal bir bağımlılığa ve paydada üstel bir fonksiyona sahiptir. Freundlich modeline yüksek konsantrasyonlarda yaklaşır ve Langmuir denkleminin düşük konsantrasyon limiti ile uyumludur. Ayrıca, Redlich-Peterson denklemi ampirik bir izotermde üç parametre içerir ve bu nedenle çok yönlülüğü nedeniyle homojen veya heterojen sistemlere uygulanabilir. Redlich-Peterson denklemi:

$$q_e = K_R.C_e / \left(1 + a_R.C_e^{\ \beta}\right) \qquad (9)$$

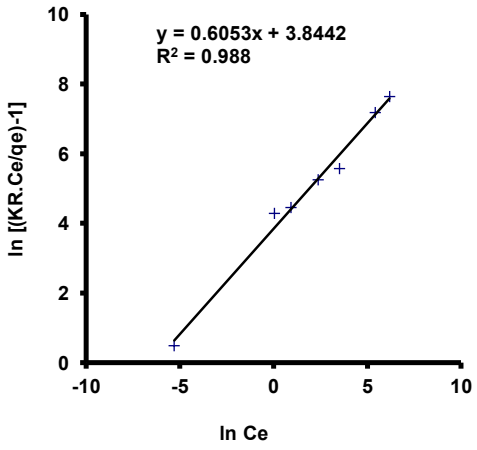
Burada KR Redlich-Peterson izoterm sabitidir (L/mg), α_R aynı zamanda bir sabittir (L/mg)β ve β sıfır ile bir arasında yer alan üsdür. β = 1 için Denk. (10) Langmuir denklemine ve β = 0 için Henry denklemine indirgenir. Denk. (10) logaritma alınarak doğrusal forma dönüştürülebilir:

$$\ln\left[\left(K_R.C_e/q_e\right)-1\right] = \ln a_R + \beta.\ln C_e \quad (10)$$

Redlich-Peterson izotermi için sabitlerin değerleri de izotermin lineer formunun (Eq. 10) çizimlerinin (Şekil 3 (c)) eğiminden ve kesişme noktalarından elde edildi. Cr(VI)'nın anyon değiştirici fiber tarafından adsorplanması, Freundlich izoterm modeli ile daha iyi tarif edilebilir. Tablo 3'teki birden (<1) küçük olan β (0.605) değeri de bu sonucu desteklemektedir [Tor, 2006].



(b)



(c)

Şekil 3. Anyon değiştirici fiber için; Langmuir izoterm grafiği (a), Freundlich izoterm grafiği (b), Redlich-Peterson izoterm grafiği (c) (fiber miktarı: sırasıyla 2.5 g/L, çözelti pH: 1.0, temas süresi: 1 saat, çalkalama hızı: 220 rpm, sıcaklık: 25±1 ˚C).

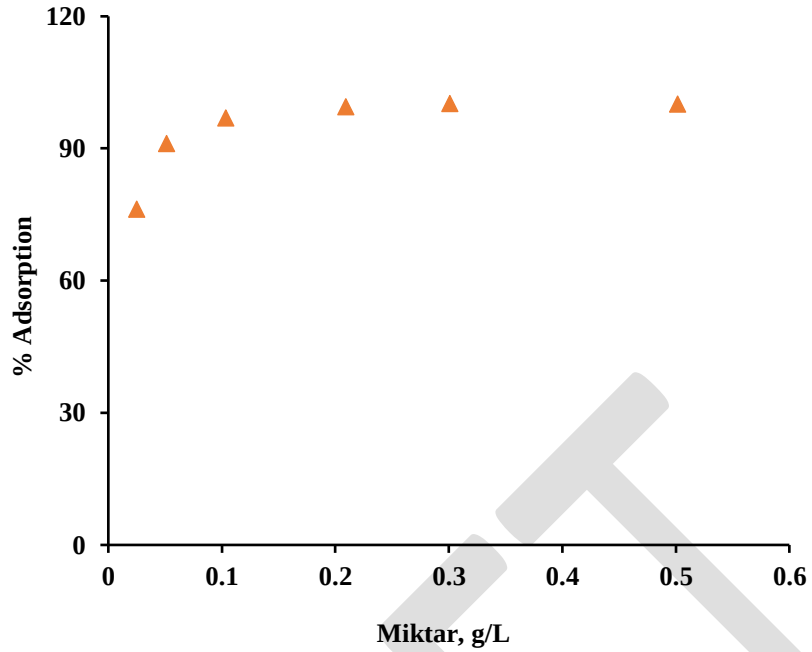
Tablo 3. Cr(VI)'nın FIBAN AK-22-1 anyon değiştirici fiber tarafından sulu çözeltiden adsorplanması için Langmuir, Freundlich ve Redlich-Peterson izoterm parametreleri.

Izoterm modeli	FIBAN AK-22-1 anyon değiştirici fiber
Langmuir modeli	
Q_o, (mg/g)	175.438
b, (L/mg)	0.040
R^2	0.979
Freundlich modeli	
k, [(mg/g).(mg/L)$^{-1/n}$]	13.883
n	2.307
R^2	0.982
Redlich-Peterson modeli	
K_R, (L/mg)	750.897
a_R, (L/mg)$^{\beta}$	47.299
β	0.605
R^2	0.988

Adsorban dozajının etkisi

Değişken miktarlarda anyon değiştirici fiber ile Cr(VI) sorpsiyonunun yüzdesi, Şekil 4'te gösterilmiştir. Genel olarak, adsorban dozundaki bir artış, adsorbatın giderilme yüzdesini artırdı. Bu, daha yüksek adsorban dozajlarının daha düşük q değerleri ile sonuçlanacağı beklentisiyle tutarlıdır. Fiber üzerindeki fonksiyonel grupların konsantrasyonu, yüzey bölgesi yoğunluğu aracılığıyla bu adsorbanların miktarı ile ilişkilidir [Sujana ve ark., 1998]. Fiber miktarının artmasıyla Cr(VI) adsorpsiyonu artmıştır, Cr(VI) adsorpsiyonu üzerinde kayda değer bir etkisi yoktur.

DRAFT



Şekil 4. Cr(VI) adsorpsiyonunun anyon değiştirici fiber miktarı ile değişimi (Cr(VI konsantrasyonu): 100 mg/L, çözelti pH'ı: 1.0, temas süresi: 1 saat, çalkalama hızı: 220 rpm, sıcaklık: 25±1 ˚C).

Sıcaklığın etkisi

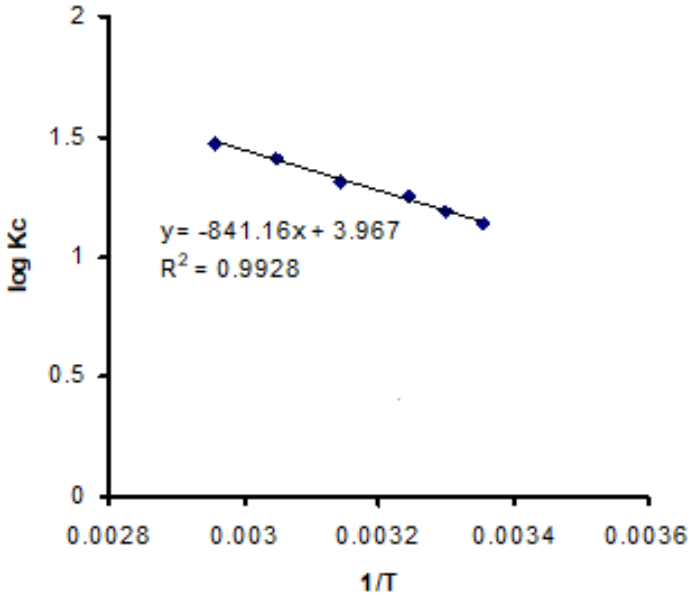
Serbest enerji değişimi (ΔG^o), entalpi değişimi (ΔH^o) ve entropi değişimi (ΔS^o) gibi termodinamik parametreler Denklemler kullanılarak hesaplanabilir. (11-13) burada Kc, metal iyonunun sırasıyla iyon değiştirici ve çözelti içindeki denge konsantrasyonlarının oranından kaynaklanan denge sabitidir [Gode ve Pehlivan, 2005]. Karışımın sıcaklığı 25 ila 65 ^oC arasında değişmiştir.

$$K_c = C_{Ae}/C_e \qquad (11)$$

$$\Delta G^o = -R.T.\ln K_c \qquad (12)$$

$$\log K_c = \left[\Delta S^o/2.303.R\right] - \left[\Delta H^o/(2.303.R.T)\right] \qquad (13)$$

Burada C_e çözeltideki denge konsantrasyonu (mg/L) ve C_{Ae} dengedeki katı faz konsantrasyonudur (mg/L). ΔG^o, ΔH^o ve ΔS^o sırasıyla serbest enerji, entalpi ve entropi değişimleridir. ΔG^o Denklem ile hesaplanabilir (12). ΔH^o ve ΔS^o, log K_c'ye karşı 1/T grafiğinden hesaplanabilir (Şekil 5 ve Denk.(13)) ve Tablo 4'te verilir. Tablo 4'te görüldüğü gibi, anyon değiştirici fiber için pozitif ΔH^o değerleri, endotermik reaksiyon meydana geldi. Pozitif ΔS^o değerleri, Cr(VI)'nın fiber üzerine adsorplaması sırasında iyon değiştirici-çözelti arayüzünde rastgeleliği önerdi. ΔG^o'nun negatif değerleri, anyon değiştirici fiber ile adsorpsiyon işlemlerinin kendiliğinden gerçekleştiğini gösterdi. Ayrıca, sıcaklığın artmasıyla birlikte ΔG^o değerlerinde meydana gelen bir düşüş, fiber tarafından adsorplanması daha yüksek sıcaklıklarda daha kendiliğinden olduğunu göstermiştir (Tablo 4).



Şekil 5. Cr(VI)'nın sıcaklığın bir fonksiyonu olarak anyon değişimli fiber üzerindeki denge sabitleri.

Tablo 4 . Cr(VI)'nın FIBAN AK-22-1 iyon değiştirici fiber üzerine adsorpsiyonu için termodinamik parametreler.

	FIBAN AK-22-1 anyon değiştirici fiber		
	ΔG° (J/mol)	**ΔH° (J/mol)**	**ΔS° (J/K.mol)**
T=298 K	-6467.72	16106.78	75.96
T=303 K	-6896.21		
T=308 K	-7395.29		
T=318 K	-8015.24		
T=328 K	-8856.34		
T=338 K	-9515.24		

SONUÇ

Mevcut çalışma, FIBAN AK-22-1 anyon değiştirici fiberin Cr(VI) iyonu için kayda değer bir adsorplama kabiliyetine sahip olduğunu göstermiştir. İyon değişimi için optimum pH aralıkları, anyon değiştirici fiber için 1.0-2.0 idi. Cr(VI) adsorpsiyonu, anyon değiştirici fiber için 30 dakikada bir dengeye ulaşmıştır. Hem reçine hem de fiber tarafından Cr(VI) adsorpsiyonunun kinetiği, ikinci dereceden kinetik modeli takip etti. Fiber üzerindeki adsorplama verileri Freundlich modeliydi, Cr(VI)'nın anyon değiştirici fiber tarafından adsorplanmasına ilişkin verileri daha iyi temsil ediyordu. Sonuç olarak, FIBAN AK-22-1 anyon değiştirici fiber, su numunelerinden Cr(VI)'nın başarıyla uzaklaştırılması için kullanılabilir bir materyal olduğu deneysel veriler ile ortaya konmuştur. Anyon değiştirici fiber için denge adsorpsiyon seviyesi, artan sıcaklıkla arttığı görülmüştür.

Kaynaklar

ATSDR, Agency for Toxic Substances and Disease Registary 2000. Division of Toxicology/Toxicology Information Branch (ATSDR), Toxicological profile for chromium, ATSDR, Atlanta, USA, pp. 1–157.

Alonso AI, Galon B, Gonzalez M, Ortiz I 1999. Experimental and theoretical analysis of a non-dispersive solvent extraction pilot plant for the removal of Cr(VI) from a galvanic process waste waters, Ind. Eng. Chem. Res. 38, 1666–1675.

Arslan G, Tor A, Muslu H, Ozmen M, Akin I, Cengeloglu Y, Ersoz M 2009. Facilitated transport of Cr(VI) through a novel activated composite membrane containing Cyanex 923 as a carrier, J. Membrane Sci. 337, 224-231.

Arslan G, Edebali S, Pehlivan E 2010. Physical and chemical factors affecting the adsorption of Cr(VI) via humic acids extracted from brown coals, Desalination 255, 117-123.

Barassi G, Valdés A, Araneda C, Basualto C, Sapag J, Tapia C, Valenzuela F 2009. Cr(VI) sorption behavior from aqueous solutions onto polymeric microcapsules containing a long-chain quaternary ammonium salt: Kinetics and thermodynamics analysis, J. Hazard. Mater. 172, 262-268.

Cengeloglu Y, Tor A, Kir E, Ersoz M 2003. Transport of hexavalent chromium through anion exchange membranes, Desalination 154, 239–246.

Dragan S, Dinu MV, Vlad CD 2003. Ion-exchange resins. II. Acrylamide crosslinked copolymers as precursors for some ion exchangers, J. Appl. Polym. Sci. 89, 2701–2707.

Edebali S, Pehlivan E 2010. Evaluation of Amberlite IRA96 and Dowex 1×8 ion-exchange resins for the removal of Cr(VI) from aqueous solution, Chem. Eng. J. 161, 161–166.

Freundlich HMF19066. Über die adsorption in losungen, Z. Phys. Chem. 57A, 385–470.

Gode F, Pehlivan E 2005. Removal of Cr(VI) from aqueous solution by two Lewatit anion exchange resins, J. Hazard. Mater. B119,175–182.

Hafiane A, Lemordant D, Dhahbi M 2000. Removal of hexavalent chromium by nanofiltration, Desalination 130, 305–312.

Ho YS, McKay G 1999. Pseudo-second order model for sorption processes, Process Biochem. 34, 451-465.

Huang L, Ou Z, Boving TB, Tyson J, Xing B 2009. Sorption of copper by chemically modified aspen wood fibers, Chemosphere 76, 1056–1061.

Kobya M 2004. Adsorption, kinetic and equilibrium studies of Cr(VI) by hazelnut shell activated carbon, Adsorpt. Sci. Technol. 22, 51–64.

Khezami L, Capart R 2005. Removal of chromium(VI) from aqueous solution by activated carbons: Kinetic and equilibrium studies, J. Hazard. Mater. B123, 223–231.

Langmuir I 1916.The constitution and fundamental properties of solids and liquids, J. Am. Chem. Soc. 38, 2221–2295.

Liu R, Tang H, Zhang B 1999. Removal of Cu(II), Zn(II), Cd(II) and Hg(II) from waste water by poly(acrylamınophosphonıc)-type chelatıng fiber, Chemosphere 38, 3169-3179.

Park HJ, Na CK 2006. Preparation of anion exchanger by amination of acrylic acid grafted polypropylene nonwoven fiber and its ion-exchange property, J. Colloid and Interface Sci. 301, 46–54.

Pehlivan E, Cetin S 2009. Sorption of Cr(VI) ions on two Lewatit-anion exchange resins and theirquantitative determination using UV–visible spectrophotometer, J. Hazard. Mater. 163 (2009) 448–453.

Redlich O, Peterson DL1959. A useful adsorption isotherm, J. Phys. Chem. 63, 1024.

Sujana MG, Thakur RS, Rao SB 1998. Removal of fluoride from aqueous solution by using alum sludge, J. Coll. Interface Sci. 206, 94–101.

Tor A 2006. Removal of fluoride from an aqueous solution by using montmorillonite, Desalination 201, 267–276.

Tor A, Buyukerkek T, Cengeloglu Y, Ersoz M 2004. Simultaneous recovery of Cr(III) and Cr(VI) from the aqueous phase with ion-exchange membranes, Desalination 171, 233–241.

Troy DB 2006. Remington: The Science and practice of pharmacy, Williams and Williams, Philadelphia.

US EPA 1990. Environmental Protection Agency, Environmental Pollution Control Alternatives, EPA/625/5-90/025, EPA/625/4-89/023, Cincinnati, US

Venkateswaran P, Palanivelu K 2004. Solvent extraction of hexavalent chromium with tetrabutyl ammonium bromide from aqueous solution, Sep. Purif. Technol. 40, 279-284.

Venkateswaran P, Palanivelu K 2005. Studies on recovery of hexavalent chromium from plating wastewater by supported liquid membrane using tri-n-butyl phosphate as carrier, Hydrometallurgy 78, 107-115.

Yilmaz A, Kaya A, Alpoguz HK, Ersoz M, Yilmaz M 2008. Kinetic analysis of chromium(VI) ions transport through a bulk liquid membrane containing p-tert-butylcalix[4]arene dioxaoctylamide derivative, Sep. Purif. Technol. 59, 1–8.

Zhang Q, Zhang S, Chen S, Li P, Qin T, Yuan S 2008. Preparation and characterization of a strong basic anion exchanger by radiation-induced grafting of styrene onto poly(tetrafluoroethylene) fiber, J. Colloid and Interface 322, 421–428.

DRAFT

DRAFT

ORAL PRESENTATION

Atık Kırmızı Çamur Katkılı Nano Gözenekli Polimerik Kompozit Membranların Hazırlanması ve Membran Performanslarının İncelenmesi

İlker AKIN* https://orcid.org/0000-0002-8683-0210

*Necmettin Erbakan Üniversitesi, Fen Fakültesi, Biyoteknoloji Bölümü, Konya,Türkiye

*ilker0997@gmail.com

Özet

Günümüzde ve yakın gelecekte en çok konuşulacak konuların su sorunu ve enerji olacağı bilinmektedir. Hem tüketimin hem de kirliliğin artması sebebiyle, bu kaynakların geri kazanımı ve çevresel kirliliklerin tespit edilip temizlenmesi gelecek yılların en önemli sorunu olarak görülmekte ve bu sorunun daha ciddi boyutlara ulaşmadan çözümünün araştırılması gerekmektedir. Membran teknolojileri günümüzde; yüksek seçicilik, verimlilik, az sarf kimyasal kullanımı, diğer ayırma ve saflaştırma yöntemleriyle birlikte kullanılabilmesi, yerinde temizlik göstermesi, düşük enerji tüketimi ve modüler esnek tasarımı kolaylığı gibi avantajları sayesinde birçok ayırma ve saflaştırma proseslerin de sürekli gelişime açık olarak tercih edilmektedir. Membran uygulamalarında en önemli problem membranın mekanik kararlılığı, akış, seçimlilik ve kirlenmenin neden olduğu membran gözenek tıkanmasıdır. Membranların bu dezavantajlarını ortadan kaldırmak için, membranlara farklı özellikte kompozit, inorganik ve/veya organik malzemeler katkılamak son yıllarda yaygın bir yöntem olmuştur. Özellikle inorganik katkı maddelerinin; killer, zeolitler, nanopartiküller vb. kompozit hibrit malzemelerin membran yapısında kullanımı membran teknolojisine yeni bir boyut kazandırmıştır. Su arıtım teknolojilerine yeni bir boyut kazandıracak, performans çalışmalarında pilot sistem kullanılarak su arıtımda birden fazla uygulama alanına hitap eden, yüksek akı, seçimlilik ve kirlenmenin neden olduğu membran gözenek tıkanmasını minimize eden ve atık bir malzemenin değerlendirilmesi açısından ayrı bir önem taşıyan kırmızı çamur katkılı polimerik kompozit membranlar elde edilmiştir. Farklı oranlarda aktif kırmızı çamur katkılı polisüfon(PSf) kompozit membran hazırlanması ve bunların gözenekli destek tabaka malzeme üzerine kaplanması ile yüksek geçirgenlik gösteren, aktif kırmızı çamur içeriğindeki metal oksitler sayesinde seçimli ve kirlenmeye karşı minimize edilen kompozit malzeme destekli membranların geliştirilmesi sağlanmış ve membranların hidrofilitesi, membranın morfolojik özellikleri SEM ve yüzey temas ölçüm teknikleri kullanarak karakterize edilip, su geçirgenliği, akı geri kazanım oranı, bovin serum albümin kullanarak kirlilik testleri ve Kütahya Emet bölgesinden temin edilen gerçek su numunesinde arsenik giderimi her bir kompozit membran için karşılaştırılarak araştırılmıştır. Elde edilen sonuçlar doğrultusunda % 90 civarı bir arsenik giderimi sağlanmıştır.

Anahtar Kelimeler: Arsenik, Kırmızı çamur, Membran

Preparation of Waste Red Mud Added Nanoporous Polymeric Composite Membranes and Investigation of Membrane Performances

Abstract

It is known that the most discussed topics today and in the near future will be the water problem and energy. Due to the increase in both consumption and pollution, the recovery of these resources and the detection and cleaning of environmental pollution are seen as the most important problem of the coming years, and the solution of this problem should be investigated before it reaches more serious dimensions. Membrane technologies today; Thanks to its advantages such as high selectivity, efficiency, low consumption of chemicals, use with other separation and purification methods, cleaning in place, low energy consumption and ease of modular flexible design, it is preferred for many separation and purification processes, open to continuous improvement. The most important problem in membrane applications is the mechanical stability of the membrane, flow, selectivity, and membrane pore clogging caused by contamination. In order to eliminate these disadvantages of membranes, it has been a common method in recent years to add composite,

inorganic and/or organic materials with different properties to membranes. Especially inorganic additives; clays, zeolites, nanoparticles etc. The use of composite hybrid materials in the membrane structure has brought a new dimension to membrane technology. Red mud added polymeric composite, which will add a new dimension to water treatment technologies, address more than one application area in water treatment by using a pilot system in performance studies, minimize membrane pore clogging caused by high flux, selectivity and pollution, and is of particular importance in terms of evaluation of a waste material. membranes were obtained. With the preparation of polysulfone (PSf) composite membrane with activated red mud in different proportions and their coating on the porous support layer material, the development of composite material supported membranes with high permeability, selective and minimized against pollution thanks to the metal oxides in the active red mud content, and the hydrophilicity of the membranes, the membrane's hydrophilicity The morphological features were characterized using SEM and surface contact measurement techniques, water permeability, flux recovery rate, pollution tests using bovine serum albumin and arsenic removal in real water sample obtained from Kütahya Emet region were compared for each composite membrane. In line with the results obtained, an arsenic removal of around 90% was achieved.

Keywords: Arsenic, Red mud, Membrane

GİRİŞ

Günümüzde ve yakın gelecekte en çok konuşulacak konuların su sorunu ve enerji olacağı bilinmektedir. Dünya nüfusunun hızlı bir şekilde artması, ülkelerin gelişim çabaları, enerji tüketimini ve doğal kaynaklarının tüketimini çok hızlı bir şekilde artırmaktadır. Enerji kaynaklarına alternatif çözümler üretilebilmektedir ancak artan nüfus ile birlikte dünyadaki temiz su kaynakları azalmakta, endüstriyelleşmenin artmasıyla da sınırlı sayıdaki kaynaklar kirlenmektedir ve suya olan talep de günden güne artmaktadır. Hem tüketimin hem de kirliliğin artması sebebiyle, bu kaynakların geri kazanımı ve çevresel kirliliklerin tespit edilip temizlenmesi gelecek yılların en önemli sorunu olarak görülmekte ve bu sorunun daha ciddi boyutlara ulaşmadan çözümünün araştırılması gerekmektedir. Bu amaçla yapılan çalışmalar; (i) temiz su kaynaklarının kullanılabilirliğinin arttırılması ve (ii) atık suların tekrar kullanıma sunulması üzerinde yoğunlaşmıştır. Bu sorunların çözümü için yapılan çalışmalarda membran teknolojisi yaygın olarak kullanılmaktadır. Membran teknolojileri günümüzde; yüksek seçicilik, verimlilik, az sarf kimyasal kullanımı, diğer ayırma ve saflaştırma yöntemleriyle birlikte kullanılabilmesi, yerinde temizlik göstermesi, düşük enerji tüketimi ve modüler esnek tasarımı kolaylığı gibi avantajları sayesinde birçok ayırma ve saflaştırma proseslerin de sürekli gelişen bir teknoloji olarak tercih edilmektedir. Membran uygulamalarında en önemli problem membranın mekanik kararlılığı, akış, seçimlilik ve kirlenmenin neden olduğu membran gözenek tıkanmasıdır. Membranların bu dezavantajlarını ortadan kaldırmak için, membran performansını ve verimliliğini artırmak amacı ile membranların geliştirilmesi gerekmektedir. Bu nedenle membranlara farklı özellikte kompozit, inorganik ve/veya organik malzemeler katkılamak son yıllarda yaygın bir yöntem olmuştur. Özellikle inorganik katkı maddelerinin; killer, zeolitler, nanopartiküller vb. kompozit hibrit malzemelerin membran yapısında kullanımı membran teknolojisine yeni bir boyut kazandırmıştır. Nanopartiküller ve kompozit yapılar; yüksek kararlılık, kimyasal ve biyolojik dayanımları, eşsiz fizikokimyasal özellikleri, geniş pH aralığında çalışma imkanı, kolay fonksiyonelleştirilebilme özelliği, bazı malzemeler için fotokatalitik, antibakteriyel, kirlilik önleme vb. özelliklerinden faydalanmak ve özellikle membranın hidrofobik polimer yapısından dolayı ortaya çıkan dezavantajı ortan kaldırmak için membran yapısına nanopartikül ve kompozit bazı malzemeler eklenmektedir. Kırmızı çamur ise alüminyum üretim tesislerinde boksitten saf alüminyum elde edilmesi sonucunda atık olarak açığa çıkan bir kompozit malzemedir. Kırmızı çamur kolloidal yapısı nedeni ile bol miktarda su içermekte ve barajlarda depolanarak rüzgarın ve çeşitli doğa olaylarının etkisiyle, çevre kirliliği ve insan sağlığı konusunda risk arz etmektedir. Ayrıca içerdiği alkali ve mikron boyutlu partiküller, kuruduğunda toz halinde havaya karışarak çevre kirliği ve insan sağlığı konusunda tehdit oluşturmaktadır. Endüstriyel faaliyetler sonucu oluşan katı atıklar, üretimin en büyük problemlerinden biridir. Bunların bertarafı için, bu katı atıkların su arıtmada düşük maliyetli absorban olarak kullanmaları iyi bir yöntem olabilmektedir. Böylelikle hem atık hacmi azaltılmış olacak hem de su arıtma işleminin maliyeti düşürülebilecektir. Kırmızı çamur, adsorban özelliği sayesinde sulardan metal iyonlarının, boyaların, fenollerin ve inorganik anyonların giderilmesinde kullanılmaktadır. Kırmızı çamurun Zn (Sahu ve diğ., 2010), Cr (Danış, 2005), Cd (Ma ve diğ., 2009), Pb (Gupta ve diğ., 2001; Luo ve diğ., 2011), Cu (Nadaroğlu ve diğ., 2017) ve As (Altundoğan ve diğ., 2002) gibi iyonları adsorplaması konusunda literatürde birçok çalışma ile karşılaşmak mümkündür. Bu çalışmaların çoğunda, yüksek yüzey alanına sahip olduğu belirtilen toz şeklindeki kırmızı çamur kullanılmıştır. Ancak

membran yapısında aktif kırmızı çamur kullanılarak membran çalışması literatürde bulunmamaktadır. Su arıtım teknolojilerine yeni bir boyut kazandıracak, performans çalışmalarında pilot sistem kullanılarak su arıtımda birden fazla uygulama alanına hitap eden, yüksek akı, seçimlilik ve kirlenmenin neden olduğu membran gözenek tıkanmasını minimize eden ve atık bir malzemenin değerlendirilmesi açısından ayrı bir önem taşıyan kırmızı çamur katkılı polimerik kompozit membranlar elde edilmiştir.

MATERYAL VE METOT

Kırmızı çamurun nötralleştirilmesi ve aktive edilmesi

Seydişehir Eti Alüminyum A.Ş. Tesislerinden (Konya) temin edilecek olan ham kırmızı çamur, saf su ile sıvı/katı oranı 2/1 (w/w) olacak şekilde sıvı kısmın pH değeri 8,0-8,5 olana kadar yıkanmış ve daha sonra 105°C'de kurutulacaktır. Kırmızı çamur nötralizasyon işlemlerinden sonra kırmızı çamurun kompozisyonu, tane boyutu dağılımı tespit edilecektir. Literatürde belirtilen kırmızı çamurun ortalama kimyasal analizi şu şekildedir: %18.71±0.59 Al_2O_3, %39.70±0.67 Fe_20_3, %14.52±0.37 $Si0_2$, %8.82±0.96 Na_2O, %4.47±0.56 CaO, %4.90±0.54 $Ti0_2$, ve diğerleri. Mineralojik açıdan bakıldığında, sodyum alüminyum silikat ve hematit bileşiklerinden oluşmaktadır. Kırmızı çamurun tane boyutu 10 µm'nin altındadır. Üretim sürecine giren boksitin yaklaşık %35-40'ı kırmızı çamur halinde atılmaktadır. Kırmızı çamur, alüminyum üreten tesislerin en önemli atık problemidir. Kırmızı çamur gerek kostik soda içermesi gerekse depolama nedeniyle önemli bir çevre sorunu oluşturmaktadır. Ayrıca, yazın kuruyan kırmızı çamurun havayı kirletmesi de ayrı bir çevresel problemdir. Aktif kırmızı çamur; ham kırmızı çamurun hem yüzey alanını genişletmek hem de yüzeyde kirlilik oluşturan metal oksitlerin uzaklaşması sağlamak koşuluyla asit ile muamelesi sonucu ortaya çıkar. Bu prosedür şu şekilde uygulanmıştır; Ham kırmızı çamur saf suyla yıkamak suretiyle nötür (pH≈7) hale getirilmiş ve 105°C de kurutulmuştur. Aktive işlemi için nötür haldeki kırmızı çamurdan 10 gr alınarak üzerine 200 ml % 20 lik HCl çözeltisi eklenerek 3 saat boyunca karıştırılmıştır. Aktivasyon işlemi için diğer asit çözeltileride duruma göre ilave edilerek (H_2SO_4 ve/veya HNO_3) karıştırma işlemi devam edilmiştir. Daha sonra aktive edilmiş kırmızı çamur daha sonra filtre kağıtından daha sonra nuçe erleninden vakum altında süzülerek nötür (pH≈7) oluncaya kadar saf suyla yıkanarak tekrar 105°C de kurutulmuştur. Böylece aktive edilmiş kırmızı çamur kullanıma hazır hale getirilmiştir.

Aktif kırmızı çamur katkılı kompozit membranların hazırlanması ve karakterizasyonu

Projenin bu basamağında yine grubumuz tarafından daha önceki membran hazırlama çalışmalarında (Arslan vd., 2009; Akın vd., 2014) başarılı bir şekilde uygulanan, su ile çöktürme tekniği kullanılarak kompozit membranlar hazırlanmıştır. Kompozit membranların hazırlanışında; ham ve aktive edilmiş atık kırmızı çamur farklı oranlarda (%w;w) polisülfon çözeltisine katkılanmasıyla, destek maddesi (hollytex 3329) üzerine döner kaplama cihazı(spin coater) yardımıyla kaplama yapılıp, su ile çöktürme(polimerleşme) tekniği kullanılarak kompozit membranlar elde edilmiştir. Kompozit membran hazırlanmasını aşamalandırırsak;

Homojen bir polisülfon çözeltisi hazırlamak için ağırlıkça %15 olacak şekilde DMF içersinde 12 saat boyunca manyetik karıştırıcıda karıştırılmıştır. Bu polimer çözeltisine farklı oranlarda (w/w)% kırmızı çamur içericek şekilde 5 mL DMF içerisinde karıştırıp homojen bir şekilde dağıtılarak polimer çözeltisine ilave edilmiştir.

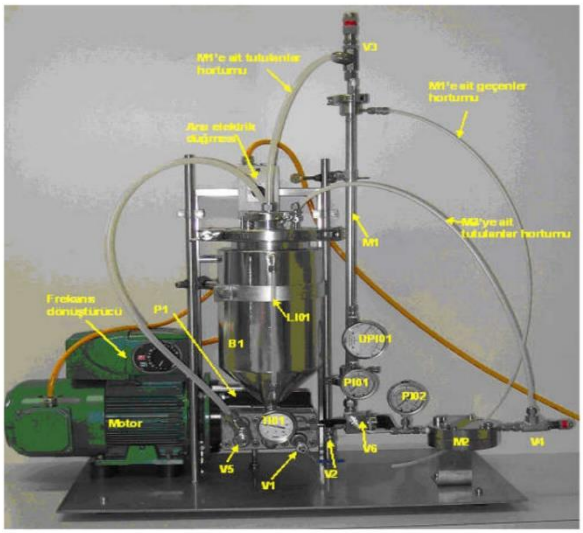
Karışım çözeltisi dokuma olmayan kumaş destek maddesi (Hollytex 3329) üzerine döner kaplama cihazı (spin coater) yardımı ile kaplama işlemi gerçekleştirilmiştir. Spin coater ile kaplama yapılırken; karışım çözeltisinin miktarı, kaplama hızı ve süresi için optimum parametreler belirlenip kaplama işlemi gerçekleştirilmiştir.

Son olarak polimerizasyonunun başlatılarak polimerin çökmesini sağlamak için oda sıcaklığında su banyosuna daldırılarak 5 dakika bekletilerek membranlar hazırlanmıştır. (Paez-Hernandez vd., 2009).

Membran Performans Çalışmaları

Hazırlanan membranların geçirgenlik çalışmalarında Şekil 1.'deki Prozesstechnik GmbH pres masaüstü membran pilot sistemi kullanılacaktır. Geçirgenlik çalışmalarında saf su akısı, tuz reddetme oranı($Mg(NO_3)_2$, $MgSO_4$ ve $MgCl_2$) ve kirlilik testleri(BSA) araştırılmıştır. Membran performans denemeleri oda sıcaklığında, besleme çözeltisinin konsantrasyonunu sabit tutmak için hem konsantre (retenteate) hem de süzüntü (permeate) besleme tankına geri döndürülmüş şekilde, işlem her membran için 1-6 saat arasında sürdürülerek, farklı konsantrasyon, farklı basınç aralıklarında(4-40 bar) gerçekleştirilmiştir. Şekil 1.'deki M2 modülü için 2,5 L uygulama hacmine sahip olan besleme tankına 2 L besleme çözeltisi kullanılacaktır. 19 Membranlar 7,5 cm çapında dairesel olarak hazırlanarak M2 modülüne yerleştirilip ve birer saat arayla hem süzüntüden hem de beslemeden numune alınarak pilot sistem çalışmaları sürdürülmüştür. Tuz reddetme oranının belirlenmesinde farklı konsantrasyonlar ve basınç parametreleri ayrı ayrı her bir membran için araştırılıp, Mg

konsantrasyonu ContrAA 300 cihazı ile, kirlilik (BSA) denemeleri ise UV spektrofotometre ile tespiti yapılmıştır. Gerçek su numunesi denemeleri; Kütahya Emet bölgesinden temin edilen gerçek su numunelerinin öncelikle kompozisyonu belirlenmiş ve sorunlu olan arseik konsantrasyonu için en iyi performans gösteren membrada orijinal numunede optimum şartlarda çalışma gerçekleştirilmiştir.



Şekil 1. Prozesstechnik GmbH pres masaüstü membran pilot sistemi

BULGULAR ve TARTIŞMA

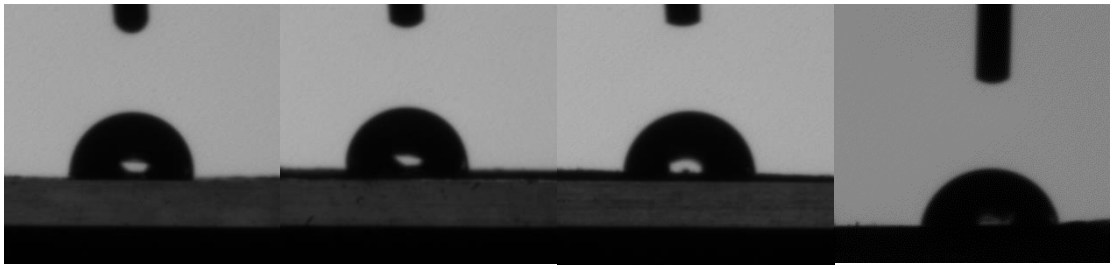
Aktif kırmızı çamur katkılı kompozit membranların karakterizasyonu

Aktive edilmiş kırmızı çamur ile hazırlanan kompozit membranların optik ve yapısal karakterizasyonları için SEM ve yüzey temas ölçüm analizleri gerçekleştirilmiştir. Katı yüzeylerde, damla şekli ve temas açısı katının serbest yüzey enerjisine bağlıdır. Hazırlamış olduğumuz membranların yüzey temas açısı ölçümlerinde KSV CAM 200 cihazı kullanılmıştır. Temas açısı ölçümleri, her bir kompozit membranın üç farklı noktasından ölçüm alınarak ölçüm değerlerinin ortalaması alınmıştır. Hazırlanan kompozit membranların temas açısını ölçmek için sabit damlası yöntemi kullanılmıştır. Temas açısı, yüzey üzerine yerleştirilen 0.10 mL'lik bir şırınga vasıtasıyla yüzeye 5 µL su damlatılarak yatay bir ışın karşılaştırıcı ile ölçülmüştür. Şekil 2'de görüldüğü üzere görüntüler dijital fotoğraf makinesi tarafından kaydedilmiştir. Bu görüntülerin temas açıları statik temas açıları hesaplama yazılımı ile hesaplanmış (Akın vd., 2014) ve sonuçlar Tablo 1'de verilmiştir.

Tablo 1. Farklı oranlardaki PSf-RM kompozit membranların yüzey temas açısı ölçüm değerleri(Ɵ)

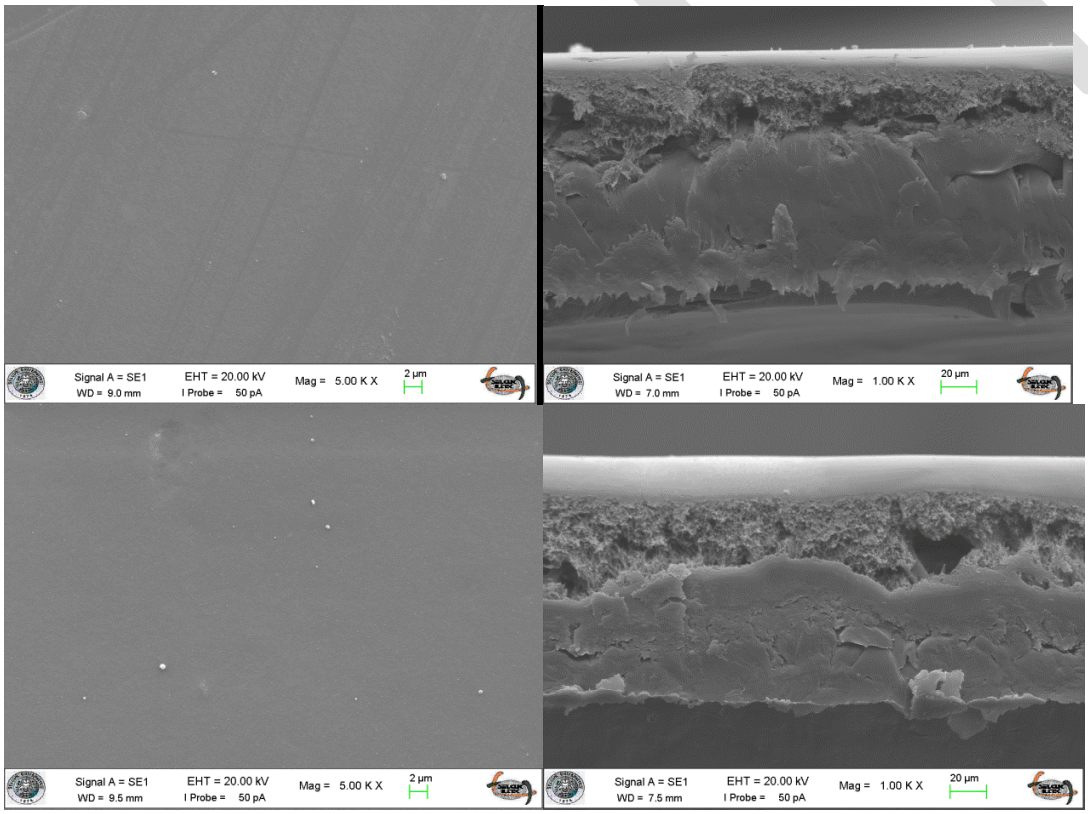
PSf-RM	Ɵ değeri
%0.1	85,23 ±0.1
%0.3	82,47 ±0,07
%0.5	73,21 ±0,12
%1	67,50 ±0,8

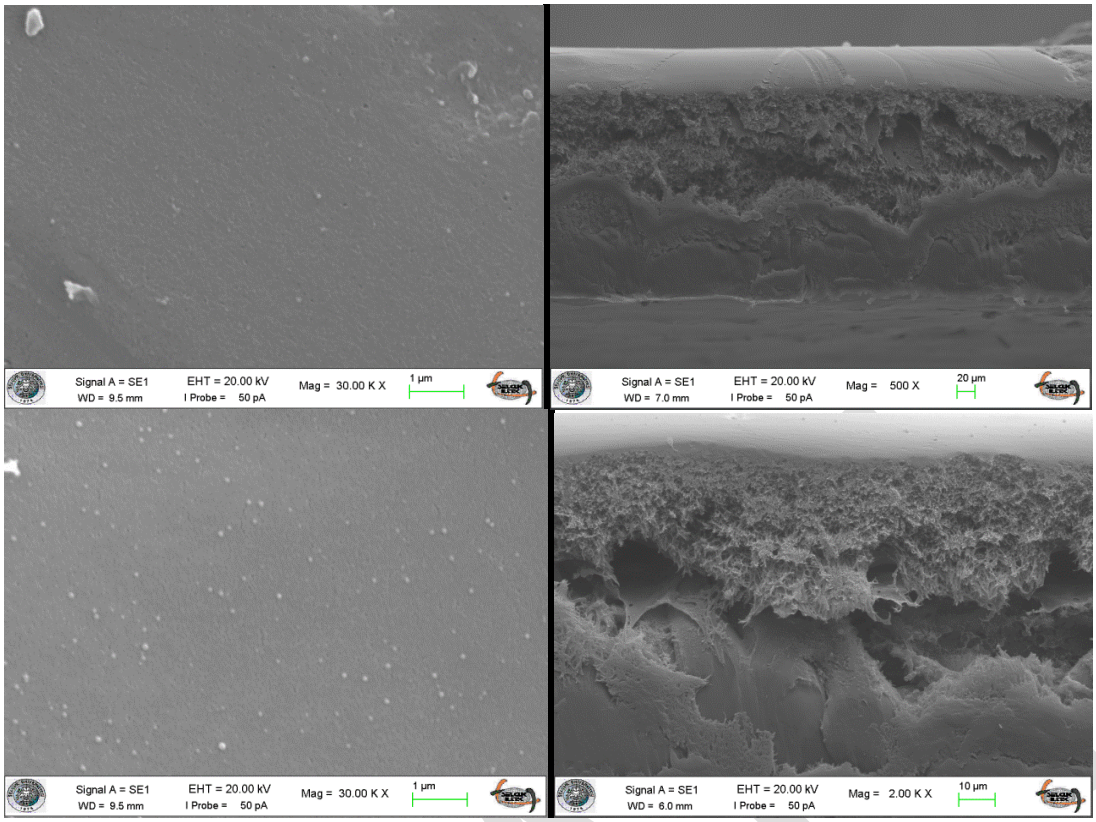
Elde edilen kompozit membranların yüzey temas açısı değerleri incelendiğinde PSf-RM kompozit membranların hidrofilik karakterinin artan madde miktarı ile değiştiği görülmüştür. Yapılarındaki hidroksil sayısının ve miktarının artması, aktive edilmiş kırmızı çamurun polimerizasyon işlemi sırasında su ve organik çözücünün hızlı yer değiştirmesi sonucunda membran içerisine çekilerek yüzeyde gözeneklilik azalması yanında ağ örgü yapıyı değiştirerek plakalar halinde boşluklar oluşturduğu (Çelik vd. 2011) yüzeydeki değişime bağlı olarakta yüzey temas açısında azalma meydane gelmesine neden olduğu düşünülmektedir.



Şekil 2. PSf-RM kompozit membranların yüzey temas açısı ölçüm görüntüleri(sırasıyla; (%w/w 0.1; 0.3;0.5;1 RM)

Farklı oranlarda katkılanmış aktive edilmiş kırmızı çamurların membran içerisindeki etkinliğini ve yapısal ve fiziksel hallerini görebilmek için farklı büyütme oranlarında SEM yüzey ve kesit görüntüleri alınarak Şekil 3.'de aşağıda verilmiştir. Elde edilen kompozit membranların SEM görüntüleri incelendiğinde katkılanan malzemeye ve orana göre yüzeyde ve kesitte farklılıklar göstermektedir. Yapısındaki metal oksitler sayesinde polimer tabaka arasında kesit görüntülerinden de görüleceği üzere ağ örgü yapı oran arttıkça daha belirgin hale gelmiştir. Membranların yüzey deformasyonu katkılanan aktive edilmiş kırmızı çamur miktarı arttıkça artmakta olduğu tespit edilmiştir. Polimerizasyon sırasında su ve organik çözücünün hızlı yer değiştirmesi esnasında oluşan boşluklara aktive edilmiş kırmızı çamur parçacıkları sürekli yer değiştirmesinden dolayı kanallar oluşturarak ağ örgü yapının oluşumunu sağlamaktadır (Vatanpour vd., 2012).





Şekil 3. Farklı oranlarda(sırasıyla; (%w/w 0.1; 0.3;0.5;1) aktive kırmızı çamur içerikli PSf kompozit membranların SEM yüzey ve kesit görüntüleri

Membran Performans Çalışmaları Sonuçları

Su tutma kapasitesi kompozit malzemelerin destek tabakasının etkinliğini tayin etmede önemli bir faktördür. Çok katmanlı aktive edilmiş kırmızı çamur katkılı kompozit membranların su tutması kapasitesi gravimetrik metot ile ölçülmüştür. 7 cm x 7 cm ebatlarında farklı oranlarda PSf-RM katkılanmış membranlar saf suda 3 saat bekletilerek, ıslak ve kuru değerleri farkından [denk. 1] kullanılarak hesaplanmıştır. Burada; membranın, sırasıyla W_w ve W_d ıslak ve kuru olarak ağılıklarını, U ise su tutma oranını göstermektedir. Elde edilen sonuçlar Tablo 2'de verilmiştir.

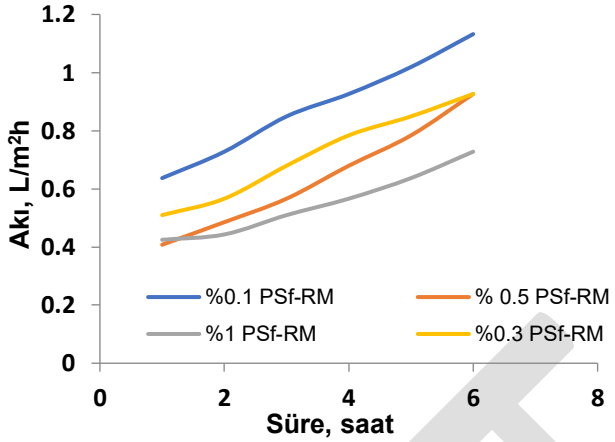
$$U = \left(\frac{W_w - W_d}{W_d} \right) x100 \quad (1)$$

Tablo 2 Farklı oranlardaki PSf-RM kompozit membranların su tutma kapasite değerleri

PSf-RM	U değerleri
%0.1	25,22
%0.3	32,48
%0.5	38,12
%1	49,14

Edilen sonuçlar gösteriyor ki PSf-RM kompozit membranların içeriğindki aktive edilmiş kırmızı çamur miktarı arttıkça su tutma kapasitesi artmaktadır. Yapısındaki metal oksitler hidroksil suyu seven gruplar olduğundan dolayı bu artışın gözlendiği düşünülmektedir. Metal nanopartiküllerin etrafında hidroksil guruplarının bulunması ve hidrfilik özellik sağlamasından dolayı su moleküllerin tutunma olasınığını arttırmaktadır. SEM kesit görüntülerinden de anlaşılacağı gibi ağ örgü yapının madde miktarı ile doğru orantılı

olarak artması sonucu su tutma kapasitesi değerlerininde fazla çıkmamasına sebep olmaktadır. Şekil 4.'de de görüldüğü gibi farklı oranlardaki saf su akı değerlerindeki değişim bu durumu doğrulamaktadır.



Şekil 4. *Farklı oranlarda (%w/w 0.1; 0.3;0.5;1) aktive kırmızı çamur içerikli PSf-RM kompozit membranların saf su akı değişimi*

Kirlilik Direnci Testleri

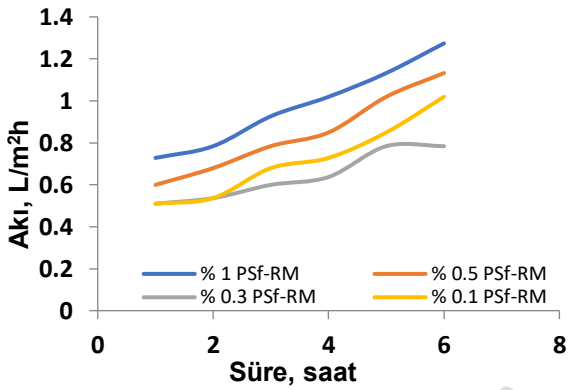
Membran performans denemeleri için tuz reddetme oranı(NaCl, Na_2SO_4, $MgSO_4$ ve $MgCl_2$) ve kirlilik testleri (BSA) gerçekleştirilmiştir. Membran performans denemeleri oda sıcaklığında, besleme çözeltisinin konsantrasyonunu sabit tutmak için hem konsantre (retenteate) hem de süzüntü (permeate) besleme tankına geri döndürülmüş şekilde, işlem her membran için 1-6 saat arasında sürdürülerek gerçekleştirilmiştir. Tuz reddetme oranının belirlenmesinde farklı konsantrasyonlar ve basınç parametreleri ayrı ayrı her bir membran için araştırılıp, Na ve Mg konsantrasyonu ContrAA 300 cihazıyla, kirlilik (Bovine Serum Albumin-BSA) denemeleri ise UV spektrofotometre ile 280 nm'de tespiti yapılmıştır. Hazırlanan kompozit membranların ayırma performansı, 25 ° C ve 1 MPa altında bir çapraz akış test sisteminde saf su akısı, BSA (moleküler ağırlık: 66 kD) reddinin ölçülmesi ile karakterize edildi. Her membran, zarın etkili alanının 44 cm^2 olduğu gözenekli bir paslanmaz çelik disk üzerindeki hücrede desteklenmiştir. Saf su, sulu bir BSA çözeltisi (100 mg/L, pH = 7) besleme çözeltileri ile test edilmiştir. Su akısı (L/m^2h^1) aşağıdaki denklemle hesaplanmıştır:

$J_{w,1}=[M/(AxΔt)]$ (2)

Burada M, nüfuz eden suyun ağırlığı (L), A membran alanı (m^2) ve süresi (h) 'dir. Permeatlar belirli bir süre boyunca toplandı ve tartıldı. Deneyler 25 °C'de gerçekleştirildi. BSA reddi, görünür ışık-ultraviyole spektrofotometre kullanılarak 280 nm'de belirlendi. Proteinin reddi (R,%) şu yollarla elde edildi;
$R(\%) = [1 - (Cp/Cf)]100$ (3)

Buradaki Cp ve Cf, sırasıyla permeat ve beslemedeki konsantrasyonlardır.

Protein çözeltisi Jp ($Lm^{-2}h^{-1}$) için akı, membranlara 6 saat boyunca 1 MPa'da nüfuz eden su miktarına göre ölçüldü. Kirlenmiş membranlar, protein çözeltisinin süzülmesinden sonra 30 dakika boyunca damıtılmış su ile yıkandı ve bundan sonra temizlenmiş membranlar Jw_2'nin (L/m^2h) su akışı tekrar ölçüldü, ayrıca Jw_1'nin (L/m^2h) saf su akı değerini göstermektedir. Filtrasyon işlemi sırasında oluşan direnç, membranın kirlenmesini gösterebilir. Kirlenme, membran yüzeyinde bir kek/jel tabakasının oluşması ve membran yüzeyine veya membran gözenekleri içine adsorpsiyon nedeniyle gerçekleşir.



Şekil 5. Farklı miktarlarda sırasıyla % 0.1, 0.3, 0.5 ve 1 oranlarında aktive edilmiş kırmızı çamur katkılı kompozit membranların süre ile BSA reddetme akı değerleri değişim

Tablo 3. Farklı miktarlarda sırasıyla % 0.1, 0.3, 0.5 ve 1 oranlarında aktive edilmiş kırmızı çamur katkılı kompozit membranların süre ile BSA reddetme değerleri(%)

	PSf-RM			
Süre(saat)	**%0.1**	**%0.3**	**%0.5**	**%1**
1	35,3	51,0	58,8	64,8
2	43,7	55,4	59,8	67,1
3	47,5	58,6	61,8	68,2
4	52,2	61,7	65,3	73,8
5	58,7	65,3	71,9	79,4
6	61,2	67,3	75,4	81,6

BSA kirlilik direnç değerleri incelendiğinde toplam kirlilik direnci değeri %0.1'lik RM içeren PSf-RM kompozit membranlarda yaklaşık %35-61 arasında sırasıyla artarak değişmektedir. %0.3'lük RM içeren PSf-RM kompozit membranlarda yaklaşık ise yaklaşık %51-67 arasında sırasıyla artarak değişmektedir. %0.5'lük RM içeren PSf-RM kompozit membranlarda yaklaşık ise yaklaşık %58-75 arasında sırasıyla artarak değişmektedir. %1'lük RM içeren PSf-RM kompozit membranlarda yaklaşık ise yaklaşık % 64-81 arasında sırasıyla artarak değişmektedir. Verilen bu sonuçlar 1 saatlik verilerin oranlarıdır. Süre ilerledikçe toplam kirlilik direncindeki değişim yapıdaki aktive edilmiş kırmızı çamur miktarı arttıkça azaldığı görülmüştür. Elde edilen BSA toplam kirlilik direnci değerleri Tablo 3.'de verilmiştir.

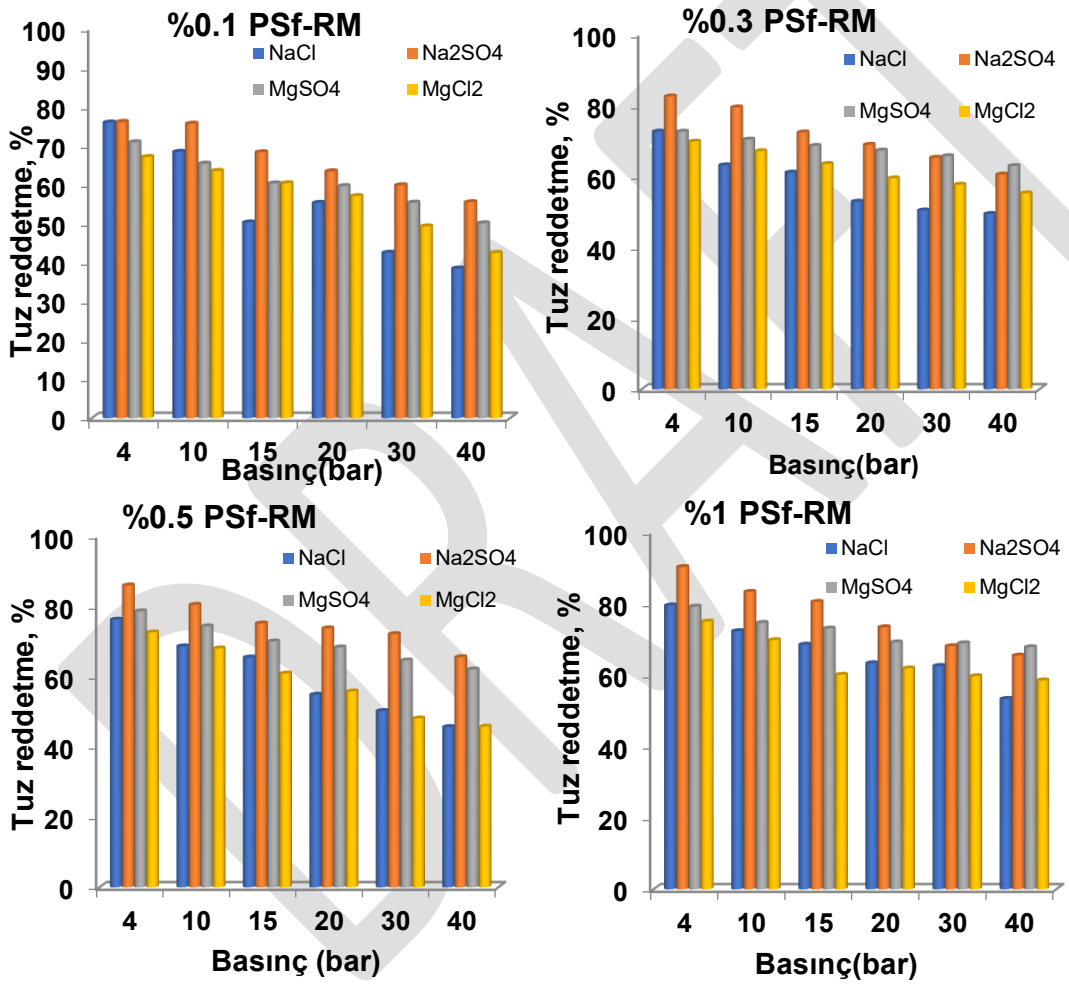
Tuz Reddetme Çalışması

Farklı oranlarda hazırlanan PSf-RM kompozit membranların performansları incelemek amacıyla, tuz reddetme (rejection) çalışmaları yapılmıştır. Tuz reddetme oranının belirlenmesinde NaCl, Na_2SO_4,$MgSO_4$ ve $MgCl_2$ tuzlarının 1000 mg/L lik çözeltileri hazırlanarak farklı basınç parametreleri ayrı ayrı her bir membran için araştırılmış ve Na, Mg konsantrasyonu ContrAA 300 cihazıyla tespit edilmiştir. Elde edilen sonuçlar Şekil 6'da verilmiştir. Elde edilen sonuçlar doğrultusunda basınç faktörünün etkili olduğu açıkça belli olmaktadır. Basıncın artmasıyla (4-40 bar) tuz reddetme oranının her bir membran için kendi içerisinde azaldığını gözlemlenmiştir. Tuz reddindeki düşüşün sebebi ise itici kuvvetin (basınç) artması membranın gözenek boyutunun değişmesine ve membranın mekanik dayanımının azalmasına sebep olduğu düşünülmektedir. Bunun sonucunda su akışını arttırırken tuz reddetme oranında düşüşler gözlenmektedir. Ganesh ve ark. (2013) yapmış oldukları PSf/GO kompozit membran ile Na_2SO_4 reddetme çalışmasında da benzer sonuçlar elde etmişlerdir. Elde edilen kompozit membranlarda en iyi tuz reddetme oranları 4 bar basınç altın elde edilmiştir. Tablo 4.'de elde edilen değerler verilmiştir. Wu ve ark. (2014) PSf çözeltisi içersine SiO_2-GO eklenmesi ile membranın hidrofilikliğini geliştirildiğini, membran ve su molekülleri arasındaki etkileşimi geliştirmek ve buna bağlı olarak sutaşıma oranını arttırdığını belirtmişlerdir. Bu durum, aynı zamanda aktive edilmiş kırmızı

çamur eklenmesi ile de literatürle uyumlu olduğu görülmüştür. Sonuç olarak hazırladığımız farklı oranlarda aktive edilmiş kırmızı çamur içeren kompozit membranların tuz reddetme performansları incelendiğinde tuz reddetme değerleri(%) Na_2SO_4> NaCl> $MgSO_4$> $MgCl_2$ şeklinde sıralandığı ve madde miktarına bağlı olarak artış gösterdiği bulunmuştur.

Tablo 4. Farklı miktarlarda sırasıyla % 0.1, 0.3, 0.5 ve 1 oranlarında aktive edilmiş kırmızı çamur içeren PSf-RM kompozit membranların 4 bar basınçta ki tuz reddetme oranı değerleri(%)

	PSf-RM			
Tuz	**%0.1**	**%0.3**	**%0.5**	**%1**
NaCl	75,86	72,56	76,24	79,50
Na_2SO_4	76.01	82,48	85,98	90,24
$MgSO_4$	70,80	72,56	78,60	79,16
$MgCl_2$	67,00	69,76	72,60	74,98

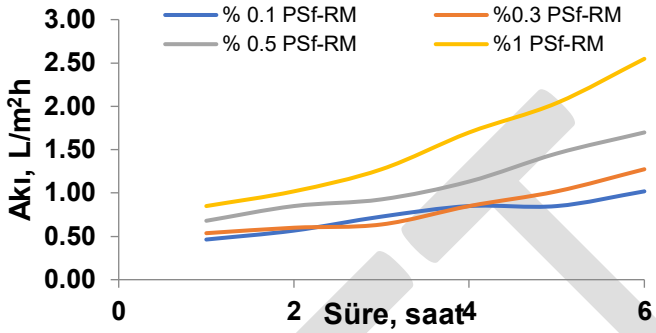


Şekil 6. Farklı oranlarda (%w/w 0.1; 0.3;0.5;1) kırmızı çamur içerikli PSf kompozit membranların tuz reddetme oranının basınç ile değişimi

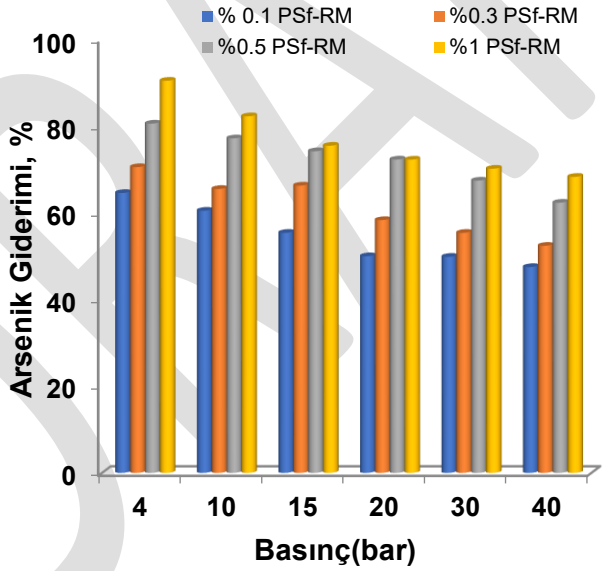
Gerçek Su Numunesi Çalışması

Prozesstechnik GmbH pres masaüstü membran pilot sistemiyle gerçek su numunesi denemeleri için Kütahya Emet ilçesinden temin edilen yeraltı suyu kullanılmıştır. Laboratuvar ortamında sentetik arsenik çözeltisi ile yapılan optimum koşullarla gerçek su numunesindeki şartlar birbirine uymayacağı düşünülerek elde ettiğimiz farklı oranlarda aktive edilmiş kırmızı çamur katkılanan PSf-RM kompozit membran ile gerçek su numunesin şartlarında(pH: 7,8; As konsantrasyonu: 21,2 ppm) pilot sistemde gerçekleştirileceği için çalışmalar basınç

faktörü göz önünde bulundurularak çalışmalar gerçekleştirilmiştir. Ayrıca gerçek su numunesi için elde ettiğimiz akı değerlerinde ki değişim sentetik numunelerdeki akı değerleriyle farklılık gösterdiği farklı orandaki PSf-RM kompozit membran denemelerinde görülmüştür.
Basınç çalışmalarında ise; 4-40 bar arasında yapılan denemeler sonucu arsenik gideriminde ve akı değerlerinde artış gözlenmiştir. Ancak basınç yükseldikçe membran yüzeyinde deformasyon ve gözenek boyutunda değişiklikler meydana gelmiştir. Basıncın artması membran ömrünü kısaltacağından optimum parametrelerin belirlenmesinde 4 bar basınç altında en yüksek verim elde edilmiştir.



Şekil 7. % 0.1-0.3-0.5 ve 1 RM içeren PSf-RM kompozit membranların gerçek su numunesindeki akı değişiminin incelenmesi



Şekil 8. % 0.1-0.3-0.5 ve 1 RM içeren PSf-RM kompozit membranlarında basınç etkisinin arsenik giderimine etkisinin incelenmesi

SONUÇ

Çalışmada su arıtım teknolojilerine yeni bir boyut kazandıracak, yüksek performanslı membran malzemeleri geliştirerek, arıtılacak su karakterine göre membran sürecinin optimizasyonunun kolaylıkla sağlanabileceği, su arıtımda birden fazla uygulama alanına hitap eden, nano gözenekli, yüksek akı, seçimlilik ve kirlenmenin neden olduğu membran gözenek tıkanmasını minimize edebilecek aktive edilmiş kırmızı çamur katkılı polimerik kompozit membranlar üretilmesi amaçlanmıştır. Bu amaç doğrultusunda yapılan çalışmalarda aşağıdaki sonuçlar elde edilmiştir.

Ham atık kırmızı çamur alınarak uzun yıkama işlemleri sonucu pH 7 civarına getirilmiştir. Daha sonra aktive etme işlemi asit karışımları ile sağlanmıştır.

DRAFT

Elde edilen aktive edilmiş kırmızı çamur polimer matriksi (PSf) içerisinde homojen bir şekilde dağıtılıp, destek malzemesi üzerine (Hollytex) kaplanmasıyla yeni bir kompozit membran oluşturularak literatüre kazandırılmıştır. Elde edilen polimerik kompozit membranların hidrofilitesi, membranların yüzey ve içyapısının morfolojik özellikleri SEM ve yüzey temas ölçüm teknikleri kullanarak başarılı bir şekilde karakterize edilmiştir.

Laboratuvar ortamında hazırlanan numuneler ile hazırlanan tüm kompozit membranların performans test çalışmaları: su tutma kapasiteleri, su geçirgenliği, tuz reddetme ve bovin serum albümin kullanarak kirlilik testleri Prozesstechnik GmbH pres masaüstü membran pilot sisteminde her bir kompozit membran için karşılaştırılarak araştırılmıştır. Membran performans denemeleri için tuz reddetme oranı(NaCl, Na_2SO_4, $MgSO_4$ ve $MgCl_2$) ve kirlilik testleri (BSA) gerçekleştirilmiştir.

Elde edilen sonuçlar doğrultusunda basınç faktörünün etkili olduğu açıkça belli olmaktadır. Basıncın artmasıyla (4-40 bar) tuz reddetme oranının her bir membran için kendi içerisinde azaldığını gözlemlenmiştir. Tuz reddinde ki düşüşün sebebi ise itici kuvvetin (basınç) artması membranın gözenek boyutunun değişmesine ve membranın mekanik dayanımının azalmasına sebep olduğu düşünülmektedir. Bunun sonucunda su akışını arttırırken tuz reddetme oranında düşüşler gözlenmektedir. Elde edilen kompozit membranlarda en iyi tuz reddetme oranları 4 bar basınç altın elde edilmiştir. Sonuç olarak hazırladığımız farklı oranlarda PSf-RM kompozit membranların tuz reddetme performansları incelendiğinde tuz türüne göre reddetme değerleri(%) Na_2SO_4> NaCl> $MgSO_4$> $MgCl_2$ şeklinde sıralandığı ve madde miktarına bağlı olarak artış gösterdiği bulunmuştur.

Prozesstechnik GmbH pres masaüstü membran pilot sistemiyle gerçek su numunesi denemeleri için Kütahya Emet ilçesinden temin edilen yeraltı suyu kullanılmıştır. Laboratuvar ortamında sentetik Arsenik çözeltisi ile yapılan optimum koşullarla gerçek su numunesindeki şartlar birbirine uymayacağı düşünülerek elde ettiğimiz farklı oranlarda aktive edilmiş kırmızı çamur katkılanan PSf-RM kompozit membran ile gerçek su numunesin şartlarında pilot sistemde gerçekleştirileceği için çalışmalar basınç faktörü göz önünde bulundurularak çalışmalar gerçekleştirilmiştir.
Basınç çalışmalarında ise; 4-40 bar arasında yapılan denemeler sonucu arsenik gideriminde ve akı değerlerinde artış gözlenmiştir. Ancak basınç yükseldikçe membran yüzeyinde deformasyon ve gözenek boyutunda değişiklikler meydana gelmiştir. Basıncın artması membran ömrünü kısaltacağından optimum parametrelerin belirlenmesinde 4 bar basınç altında denemeler gerçekleştirilmiştir. Buradan da basınç artışıyla arsenik gideriminin arttığı sonucunu çıkarabilir. Elde edilen sonuçlar doğrultusunda maksimum % 90 civarı bir As giderimi tespit edilmiştir.

Sonuç olarak; elde ettiğimiz kompozit membranlar ile yapılan denemeler her ne kadar olumlu sonuç verse de her zaman yeterli gelmemektedir. Burada da gördüğümüz gibi bazı doğal su numunelerinde tek seferde arsenik için WHO'nun belirlediği 10 µg/L limitinin altına inilememiştir. Bu sorunu ortadan kaldırmak içinde çift kademeli membran uygulaması veya diğer ayırma teknikleri ile hibrid şekli düşünülebilir veya membran sistemi ile adsorpsiyon hibridi uygulanması gerekmektedir.

Teşekkürler

Bu araştırma Necmettin Erbakan Üniversitesi Bilimsel Araştırma Projeleri Koordinatörlüğü tarafından finanse edilmiştir. (Proje numarası: 191715001)

Kaynaklar

Akin I, Zor E, Bingol H, Ersoz M 2014. Green synthesis of reduced graphene oxide/polyaniline composite and its application for salt rejection by polysulfone-based composite membranes, The journal of physical chemistry B, 118, 21,5707 – 5716

Altundoğan HS, Altundogan S, Tumen F, Bildik M 2002. Arsenic adsorption from aqueous solution by activated red mud. Waste Manage. 22 (3): 357–363

Arslan G, Tor A, Muslu H, Ozmen M, Akin I, Cengeloglu Y, Ersoz M 2009. Facilitated transport of Cr(VI) through a novel activated composite membrane containing Cyanex 923 as a carrier, Journal of Membrane Science, 337, 224–231.

Celik E, Liu L, Choi H 2011. Protein Fouling Behavior of Carbon Nanotube/Polyethersulfone Composite Membranes During Water Filtration, Water Reseach, 45, 5287–5294

Ganesh BM, Isloor AM, Ismail AF, 2013. Enhanced Hydrophilicity and Salt Rejection Study of Graphene Oxide- Polysulfone Mixed Matrix Membrane, Desalination, 313, 199−200.

Gupta Y, Hellgardt K, Wakeman RJ, 2006. Enhanced Permeability of Polyaniline Based Nano-membranes for Gas Separation, Journal of Membrane Science, 282, 60-70

Luo L, Ma C, Ma Y, Zhang S, Lv J, Cui M 2011. New insights into the sorption mechanism of cadmium on red mud. Environ Pollut, 159(5):1108-13.

Ma H, Burger C, Hsiao BS, Chu B, 2012. Nanofibrous Microfiltration Membrane Based on Cellulose Nanowhiskers. Biomacromolecules, 13, 180−186

Nadaroğlu H, Kalkan E, Çelebi N 2017. Adsorption Performance of LaccaseModified-Red Mud for Acid Fuchsin DyeRemoval fromAqueous Solutions. Annals ofChromatographyand SeparationTechniques, 3, 1027-1035

Paez-Hernandez, ME, Aguilar-Arteaga K, Valiente M, Ramirez-Silva MT, Romero-Romo M, Palmomar-Pardave M 2004. Facilitated transport of Hg(II) through novel activated composite membranes, Analytical and Bioanalytical Chemistry, 380, 690–697.

Sahu PC, Mishra MC 2018. Employment generation through environmental management in mining areas of Orissa, India: A novel approach. National Journal of Multidisciplinary Research and Development 3;1,250-255

Vatanpour V, Madaeni SS, Moradian R, Zinadini S, Astinchap B, 2012. Novel antibifouling nanofiltration polyethersulfone membrane fabricated from embedding TiO_2 coated multi walled carbon nanotubes, Separation and Purification Technology, 90, 69–82.

DRAFT

ORAL PRESENTATION

Karkamış Baraj Gölü (Gaziantep) Mavi-Yeşil Algleri

Güneş Pala[1*](*ORCID: https://orcid.org/0000-0003-0535-4177*), Özge ALİM[2] (*ORCID: https://orcid.org/0009-0003-8026-0346*)

[1*]Fırat Üniversitesi, Su Ürünleri Fakültesi, Temel Bilimler, Elazığ, Türkiye
[2]Eskişehir Teknik Üniversitesi, Fen Fakültesi, Moleküler Biyoloji, Eskişehir, Türkiye
[1*]gpala@firat.edu.tr
[2]ozgealim@ogr.eskisehir.edu.tr

Özet

Karkamış Baraj Gölü'nün (Gaziantep) mavi-yeşil alglerini belirlemek amacıyla gölden Mayıs-Ağustos 2021 tarihleri arasında örnekler alınmıştır. Çalışma süresince mavi-yeşil alglere ait toplam 23 takson tespit edilmiştir. Mavi-yeşil algler içerisinde en fazla taksonla temsil edilen cinsler *Chroococcus* sp. ve *Anabaena* sp. olmuştur. İstasyonlarda tüm aylarda kaydedilen mavi yeşil algler arasındaki Sorenson benzerlik indeksi ise en fazla ikinci ve üçüncü istasyonlar arasında çıkmıştır.

Anahtar Kelimeler: Mavi-yeşil alg, Karkamış Baraj Gölü, Gaziantep.

Abstract

In order to determine the blue-green algae of Karkamış Dam Lake (Gaziantep), samples were taken from the lake between May and August 2021. During the study, a total of 23 taxa of blue-green algae were identified. Among the blue-green algae, the genera represented by the most taxa are *Chroococcus* sp. and *Anabaena* sp. has been. The Sorenson similarity index among the blue-green algae recorded at the stations in all months was the highest between the second and third stations.

Keywords: Blue-green algae, Karkamış Dam Lake, Gaziantep.

GİRİŞ

Baraj gölleri; akarsuların üzerine inşa edilen, içme suyu temini, sulama, elektrik üretimi, balıkçılık, sel kontrolü ve rekreasyon amaçlı kullanılan yapay göllerdir. Bu göller, akış hızı yüksek, giriş suyunda askıda katı madde varlığı ve kısa su değişim süresi gibi özelliklerinden dolayı doğal göllerden farklıdır. Su toplama havzalarının daha geniş olması dolayısıyla doğal göllere göre havzadaki kirlenmeden daha fazla etkilenirler (Demir ve Atay, 1999).

Akçaalan vd., 2008; Albay vd., 2005'a göre: "*Türkiye'de baraj göllerinde yürütülen araştırmalarda daha çok baraj göllerinin fitoplankton kompozisyonu incelenmiştir. Bu araştırmalarda, Bacillariophyta ve Chlorophyta divizyolarına ait türlerin oransal olarak fazla bulunduğu, mavi-yeşil alglerin ise nadiren bulunduğu, ancak zaman zaman sayısal artışlar olduğu kaydedilmiştir*." (Fakıoğlu ve ark. 2011).
Mavi-yeşil algler (Cyanophyta), akuatik çevrede yaşam süren prokaryot organizmalardır (Saad ve Atia, 2014). Mavi-yeşil algler, rikestsiya, aktinomiset ve miykoplazma gruplarının dahil olduğu; gerçek çekirdek zarları ve membrana bağlı organelleri olmayan, fosfolipid tabakalı hücre çeperi bulunan mikroorganizmalardır. Mavi-yeşil algler çekirdek bulundurmayan hücrelerin en gelişmiş kolunu oluşturur. Hemen hemen tamamında kromozomları proteinle çevrilmiş halkasal şekilde bir DNA zinciri içerirken, mitoz bölünmezler. Her hücrede monoploid olarak bulunan tek kromozom, açılıp bir hücrenin bir ucundan, diğer bir ucuna hareket ederek kendini replike ettiğinde, hücre bölünür. Evrimsel süreçte ise, 2,5-3 milyar sene önce, kese şeklindeki ilk hücreden evrimleştiği düşünülmektedir (Paerl ve ark., 2001; URL, 1). Mavi-yeşil alglerin, diğer alg türleri gibi, farklı bir dış görünümleri vardır. Fotosentez yaparak beslenirler (Ris H., 1961). Alglerin en çok bilinenleri mavi-yeşil olanlarıdır. Sitoplazmalarında hem klorofil (yeşil renk) hem de fikosiyanin (mavi renk) renk pigmentleri bulunur (Gantt E., 1980). Çoğunlukla koloni halinde derelerde, göllerde ve denizlerde, nemli topraklarda, havuz kenarlarında, nemli kayaların üzerinlerinde ve kaplıca sularında yaşayabilirler. Bitkilerin

yeryüzünde yaptığı görevi algler sularda yaparlar. Bu algler; akuatik ortamda yaşayan canlılar için besin ve oksijen kaynağıdır (Saad A. ve Atia A., 2014; URL, 1).
Mavi yeşil algler, diğer algler gibi sucul ekosistemdeki besin zincirlerinde temel basamakta ilk sırada yerini alırlar. Yapısında barındırdığı pigmentler sayesinde, suyu ve bazı besin tuzlarını (azot, fosfor), ışık etkisiyle karbonhidratlara çevirirler. Böylelikle hem besin üretir, hem de suyun çözünmüş oksijen miktarını artırırlar. Çoğalmaları ışık, sıcaklık ve besine bağlı olarak belirlenir. Ya tek tek hücresel yapıda ya da koloniler halinde yaşarlar (URL, 2). Visser, (2015)'deki derleme makalesinde özetle: "Bu alglaer fazla çoğaldığında da sudaki oksijeni azaltarak, suda bulanıklığa sebep olurlar ve bu durum ışığın suyun derinlerine inmesine engel olur. Bu ise; derinlerde yaşam süren su canlılarının ölmesiyle sonuçlanır. Yine, bu algler; tatlı suların yüzeylerinde diğer planktonlarla birlikte "su çiçeği" denilen bir yapıyı oluşturmaktadırlar. Bu algler arasında yer alan bazı türler, salgıladıkları toksinler nedeniyle zooplanktonların, balıkların ölümüne de yol açabilmektedirler." Olarak bildirmiştir (Visser, 2015). Göllerde genişleyen insan faaliyetleri nedeniyle siyanobakteri patlamaları çok yaygın olarak görülmektedir (Christoffersen ve Kaas, 2000). Bu çalışmanın amacı, baraj göllerimizde fitoplankton üzerine yapılan araştırmaların incelenmesi ile mavi-yeşil alg artışları potansiyelini ve izlenmesinin önemini ortaya koymaktır.

MATERYAL VE METOT

Çalışma alanı olarak seçilen bölge; Güneydoğu Anadolu Projesi'nin bir kısmını oluşturan, sınır Fırat Projesi'nin ikinci ünitesi olarak bulunan, Karkamış Barajı ve HES Tesisi, Fırat Nehrinin üzerinde, Suriye Sınırı'na 4,5 km'lik uzaklıkta, beton ağırlık ve toprak dolgu tipinde ve yine Türkiye'de nehir santrali tanımlamasıyla gerçekleşen ilk uygulama alanıdır. 180 MW gücünde olan bu santralin, senede 652 GWh'lik elektrik enerjisi üretmesi amaçlanmıştır. Enerji üretmekte olan bu barajın, baraj rezervuar bölgesinde sağ-sol sahil koruma seddeleri de yapılarak, 336 m kotunda olan baraj gölünün işletme kotu 340 m'ye çıkarılarak, baraj gölü altında kalan yer ve 433 hektarlık tarım arazisinin korunması hedeflenmiştir (URL, 3). Karkamış Barajı'nın gövdesel hacmi; 2.100.000 m^3, akarsu yatağından yüksekliği ise 29,00 m normal su kotunda, gölün hacmi 157,00 hm^3, normal su kotundaki göl alanıysa 28,40 km^2'dir. Karkamış Baraj Gölü'nün mavi-yeşil alglerini belirlemek için 500 m aralıklarla üç istasyon belirlenmiştir. Üç istasyonda da kalitatif fitoplankton örnekleri plankton ağı ile toplanmıştır. Toplanan örnekler plankton ağının ucundaki kapak açılarak önceden göl suyuyla steril edilmiş cam kavanozlara alınmıştır. Fitoplankterlerin sayımları için 'Leitz' marka inverted mikroskop kullanılmıştır. Sayımlar için alınan örneklerden 1 ml alınarak sayma lamına konulmuş ve sayılmıştır. Sayımlar sırasında tek hücreli alglerde hücre sayıları baz alınırken, kolonial veya filamentli formlarda her bir koloni veya filament bir organizma olarak kabul edilmiştir. Mavi-yeşil alglerin tür bazında teşhislerinin yapılabilmesi için gliserinle geçici preparatları oluşturulmuştur. Teşhisler için ilgili kaynaklardan (Geitler, 1925; Bourelly, 1968; Bourelly,1972) yararlanılmıştır. İstasyonlardaki mavi-yeşil algler arasındaki benzerliği ortaya çıkarmak için Sorenson benzerlik indeksi uygulanmıştır.
Sorenson Benzerlik İndeksi: Q/S= 2J/A+B
A= Birinci örnekteki toplam tür sayısı
B= İkinci örnekteki toplam tür sayısı
J= Her iki örnekte ortak olan tür sayısı (Sørensen, 1948).

BULGULAR

Araştırma süresince Karkamış Baraj Gölü'nde mavi-yeşil alglere (Cyanophyta) ait toplam 23 takson kaydedilmiştir Mavi- yeşil alglerin istasyonlara göre bulunma özellikleri Tablo 1'de gösterilmiştir (Tablo 1).

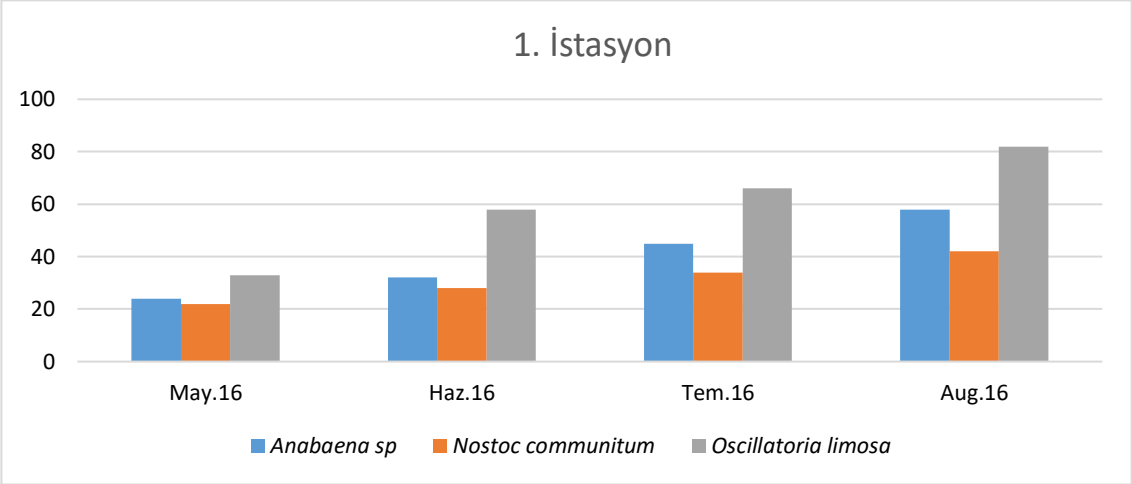
DRAFT

Tablo 1. Karkamış Baraj Gölü'nde Kaydedilen Mavi-Yeşil Alglerin İstasyonlara Göre Bulunma Özellikleri

CYANOPHYTA	I.ist.	II. İst.	III. İst.
Chroococcales			
Chroococcus minör (Kütz.) Naegeli	+	+	_
Chroococcus indicus Zeller	+	+	+
Chroococcus turgidus (Kütz.) Naegeli	+	–	–
Synechocystis endobiotica (Elenkin & Hollerbach) Elenkin	–	+	+
Dactylococcopsis rhaphidioides Hansgirg	+	+	+
Gleocapsa aeruginosa Kütz.	+	+	+
Gleocapsa rupestris Kütz.	–	+	–
Hormogonales			
Calothrix epiphytica West & West	+	+	–
Nostocales			
Anabaena augstumalis Schmidle	+	–	–
Anabaena levanderi Lemmermann	+	+	–
Anabaena verrucosa J.B. Petersen	+	+	+
Dolichospermum planctonicum (Brunnthaler) Wacklin, L.Hoffmann & Komárek	–	+	+
Nostoc comminutum Kütz.	+	+	–
Trichormus variabilis (Kützing ex Bornet & Flahault) Komárek & Anagnostidis	–	+	+
Oscillatoriales			
Geitlerinema earlei (N.L.Gardner) Anagnostidis	+	+	+
Lyngbya martensiana Meneghini ex Gomont	–	–	+
Oscillatoria limosa C.Agardh ex Gomont	+	+	–
Phormidium ambiguum Gomont	–	+	–
Phormidium diguetii (Gomont) Anagnostidis & Komárek	–	+	+
Phormidium hamelii (Frémy) Anagnostidis & Komárek	–	+	–
Planktothrix prolifica (Gomont) Anagnostidis & Komárek	–	+	+
Porphyrosiphon versicolor (Gomont) Anagnostidis & Komárek	+	+	+
Schizothrix tinctoria Gomont ex Gomont	+	+	+

Birinci İstasyonda Kaydedilen Mavi- Yeşil Algler (Cyanophyta)

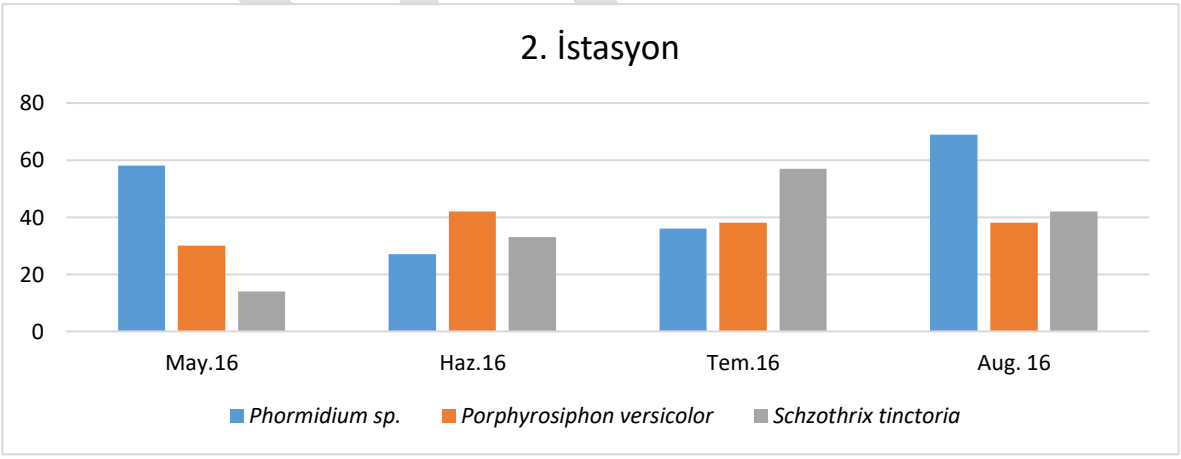
Karkamış Baraj Gölü'nden seçilen birinci istasyonda mavi- yeşil alglere ait *Chroococcus* (3 takson), *Dactylococcopsis* (1 takson), *Gleocapsa* (1 takson), *Calothrix* (1 takson), *Anabaena* (3 takson), *Nostoc* (1 takson), *Geitlerinema* (1 takson), *Oscillatoria* (1 takson), *Porphyrosiphon* (1 takson) ve *Schizothrix* (1 takson) olmak üzere toplam 14 takson kaydedilmiştir. Ortaya çıkış sıklığı ve birey sayıları bakımından en önemli cinsler, *Anabaena*, *Nostoc* ve *Oscillatoria* olmuştur. Bu alglerin birinci istasyondaki birey sayıları çalışma süresince hava sıcaklığına paralel bir şekilde düzenli olarak artmıştır. *Oscillatoria limosa'*nın Ağustos ayında kaydedilen ml'deki filament sayısı (82 fil./ml) bu istasyonun diğer mavi-yeşil algleri arasındaki en yüksek birey sayıları olmuştur (Şekil 1). Birinci istasyonda *Anabaena* cinsi, *Anabaena augstumalis*, *Anabaena levanderi* ve *Anabaena verrucosa* türleri ile; *Nostoc* cinsi yalnızca *Nostoc communitum* türü ve *Oscillatoria* cinsi ise yalnızca *Oscillatoria limosa* türü ile temsil edilmiştir. Bu istasyonda birey sayıları bakımından önemli olan diğer türler Mayıs ayında *Geitlerinema earlei* (77 fil./ml), Haziran ayında *Porphyrosiphon versicolor* (66fil./ml), Temmuz ayında *Schzothrix tinctoria* (68 fil./ml), Ağustos ayında ise *Geitlerinema earlei* (50 fil./ml) olmuştur.



Şekil 1. Birinci istasyonda tüm aylarda kaydedilen *Anabaena* sp, *Nostoc communitum* ve *Oscillatoria limosa*' nın ml'deki filament sayılarındaki aylık değişimler

İkinci İstasyonda Kaydedilen Mavi-Yeşil Algler (Cyanophyta)

Karkamış Baraj Gölü'nden seçilen ikinci istasyonda mavi-yeşil alglere ait *Chroococcus* (2 takson), *Synechocystis* (1 takson), *Dactylococcopsis* (1 takson), *Gleocapsa* (2 takson), *Calothrix* (1 takson), *Anabaena* (2 takson), *Dolichospermum* (1 takson), *Nostoc* (1 takson), *Trichormus* (1 takson), *Geitlerinema* (1 takson), *Oscillatoria* (1 takson), *Phormidium* (3 takson), *Planktothrix* (1 takson), *Porphyrosiphon* (1 takson) ve *Schizothrix* (1 takson) olmak üzere toplam 20 takson kaydedilmiştir.
İkinci istasyonda birey sayıları ve ortaya çıkış sıklığı bakımından en önemli cinsler ise *Phormidium*, *Planktothrix* ve *Schzothrix* olmuştur. *Phormidium* cinsi bu istasyonda *P. ambiguum*, *P. diguetti* ve *P. hamelii* türleri ile temsil edilirken *Porphyrosiphon* cinsi, *P. versicolor*, *Schzothrix* cinsi ise *S. tinctoria* türü ile temsil edilmiştir. Bu istasyonda dört ayda da ortaya çıkan bu cinslerin aylara göre birey sayılarında artma ve azalmalar dikkat çekici olmuştur.



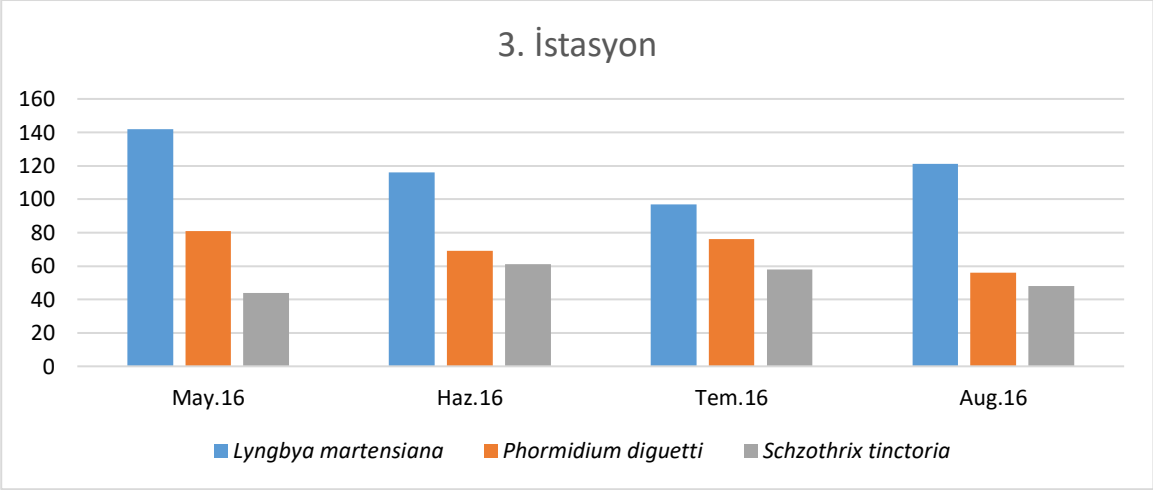
Şekil 2. İkinci istasyonda tüm aylarda kaydedilen *Phormidium* sp, *Porphyrosiphon versicolor* ve *Schzothrix tinctoria'* nın ml'deki filament sayılarındaki aylık değişimler

Karkamış Baraj Gölü'nün ikinci istasyonunda tüm aylarda kaydedilen diğer bir türler ise *Anabaena verrucosa*, *Nostoc communitum* ve *Oscillatoria limosa* olmuştur. Fakat bu türlerin ml deki filament sayıları Şekil 2'de gösterilen türlere nazaran daha düşük olmuştur. Diğer türler ise ya bir ya da iki ayın örneklerinde kaydedilmişlerdir.

Üçüncü İstasyonda Kaydedilen Mavi-Yeşil Algler (Cyanophyta)

Karkamış Baraj Gölü' nün üçüncü istasyonunda mavi-yeşil alglere ait *Chroococcus* (1 takson), *Synechocystis* (1 takson), *Dactylococcopsis* (1 takson), *Gleocapsa* (1 takson), *Anabaena* (1 takson), *Dolichospermum* (1 takson), *Trichormus* (1 takson), *Geitlerinema* (1 takson), *Lyngbya* (1 takson), *Phormidium* (1 takson), *Planktothrix* (1 takson), *Porphyrosiphon* (1 takson) ve *Schizothrix* (1 takson) olmak üzere toplam 13 takson kaydedilmiştir.

Bu istasyonda örneklemenin yapıldığı tüm aylarda ortaya çıkan taksonlar *Lyngbya martensiana*, *Planktothrix prolifica*, *Phormidium diguetti* ve *Schzothrix tinctoria* olmuştur. *Porphyrosiphon versicolor* türü ise mayıs ayı hariç haziran, temmuz ve ağustos'ta kaydedilen mavi- yeşil alg olmuştur. Diğer türlere ise bir veya iki örnekte ancak rastlanılabilmiştir. Karkamış Baraj Gölü'nün üçüncü istasyonunda tüm aylarda kaydedilen *Lyngbya matrensiana'*nın mayıs ayında kaydedilen filament sayıları (142 fil./ml) her üç istasyonda da kaydedilmiş en yüksek birey sayıları olmuştur. Bu istasyonda ortaya çıkış sıklığı fazla olan diğer bir tür *Gleocapsa aeruginosa* olmuştur. Fakat bu türün ml'deki hücre sayıları önemsiz olmuştur.



Şekil 3. Üçüncü istasyonda tüm aylarda kaydedilen *Lyngbya martensiana*, *Phormidium diguetti* ve *Schzothrix tinctoria'* nın ml'deki filament sayılarındaki aylık değişimler

Üçüncü istasyonda birey sayıları bakımından önemli olan diğer bir tür *Phormidium diguetti* ile *Schzothrix tinctoria* olmuştur. *P. diguetti*'ye ait en yüksek filament sayıları (81 fil./ml) mayıs, *S. tinctoria*'ya ait en yüksek filament sayıları (61 fil./ml) ise haziran ayında kaydedilmiştir. Sorenson Benzerlik İndeksi birinci ve ikinci istasyon arasında %70,58; ikinci ve üçüncü istasyon arasında %72,72 çıkarken, birinci ve üçüncü istasyon arasında ise %44,44 gibi bir sonuçla çok düşük çıkmıştır.

TARTIŞMA VE SONUÇ

Karkamış Baraj Gölü'nde yapılan bu çalışmada, mavi-yeşil alglere ait toplam 23 takson kaydedilmiştir. Chroococcales ordosuna ait 7 takson, Hormogonales ordosuna ait 1 takson, Nostocales ordosuna ait 6 takson ve Oscillatoriales ordosuna ait 9 takson tespit edilmiştir. Baraj Gölü'nden tespit edilen istasyonlar arasındaki Sorenson Benzerlik İndeksi en yüksek (%72,72) ikinci ve üçüncü istasyon arasında, en düşük ise (%44,44) birinci ve üçüncü istasyonlar arasında çıkmıştır (Sorenson, 1948). Birinci ve üçüncü istasyonlar arasında bu kadar düşük bir benzerlik çıkmasının nedeni ise habitat farklılığından kaynaklı olabileceği sonucunu ortaya koymaktadır. Örnek alma yeri olarak seçilen birinci istasyonda çok az makrofit bulunurken ikinci ve üçüncü istasyonlar bol makrofitli alanlardan seçilmiştir.
Sönmez ve ark. (2017)'nin Karkamış Baraj Gölü (Şanlıurfa/Türkiye)'nde "Fitoplanktonun Mekansal Temporal Dağılımı" adlı çalışmalarında mavi-yeşil (Cyanophyta) alglere ait (*Geitlerinema amphibium, Gleocapsa alpina, Lyngbya major, Microcystis aeruginosa, Oscillatoria limosa, Oscillatoria tenuis, Phormidium breve, Phormidium lucidum, Phormidium subfuscum* ve *Spirulina major*) toplamda 10 adet takson kaydedilmiştir. Aynı baraj gölü'nde yapmış olduğumuz bu çalışmanın bulguları ile Sönmez ve ark. (2017)'nın bulguları

Oscillatoria limosa hariç yalnızca cins seviyesinde benzerlik göstermiştir. Bu durum örnek alma istasyonlarının ve aylarının farklılıklarından kaynaklanmış olabileceğini ortaya koymaktadır. Fakıoğlu ve ark. (2011)'nın "Baraj Göllerinde Toksik Mavi-Yeşil Algler" adlı çalışmalarında toksik Cyanobakteria artışlarının izlenmesi ve alınması gereken önlemler araştırılmıştır. Bu çalışmada mavi-yeşil alglerden 98 adet tür teşhis edilmiş ve bu türlerden 57 tanesinin toksik olabileceği vurgulanmıştır. Bu toksik türler arasında yer alan iki tür (*Chroococcus turgidus* ve *Lyngbya martensiana*) çalışmamız bulgularında da tespit edilmiştir. Toksik mavi-yeşil alglerin içme sularında gelişiminin engellenmesi halk sağlığının korunması açısından önemlidir. Bu alglerin aşırı artması, su kirlenmesi ile orantılıdır. Ortamdaki azot ile fosfor derişiminin standart seviyelerde olmasıyla bu alglerin artışları önlenebilir. Bu bağlamda havza yönetimi önem kazanmaktadır. Çünkü dış ve iç azot ve fosfor yükünün kontrolü stratejik bir havza yönetimi ile mümkündür (Anonymous, 2010).

Açıkgöz ve Baykal (2005)'ın "Karagöl (Çubuk-Ankara) Alg Florası" isimli araştırmalarında Karagöl'ün planktonik, epipelik, epifitik ve epilitik alg florası Kasım 1999-Ekim 2000 tarihleri arasında kalitatif olarak incelenmiş ve Bacillariophyta (120), Chlorophyta (41), Cyanophytaya (24), Euglenophyta (9) ve Dinophyta (2) olmak üzere toplam 196 adet takson tespiti yapılmıştır. Cyanophyta'ya ait 24 takson içerisinde bulunan *Lyngbya, Oscillatoria, Phormidium, Anabaena* ve *Nostoc* cins bazında *Lyngbya martensiana* ise tür bazında çalışmamızın bulguları ile benzerlik göstermiştir.

Öztürk (2014)'ün "Apa Baraj Gölü Algleri Üzerine Araştırmalar" isimli doktora tezinde; gölün bentik algleri (epipelik, epifitik ve epilitik), kıyı bölgesi fitoplanktonu, mevsimsel değişimi ve bu değişime etki eden fiziksel ve kimyasal faktörler Mart 2010–Mart 2012 tarihleri arasında incelenmiştir. Fitoplanktonda *Heterokontophyta*, *Chlorophyta*, *Charophyta*, *Cyanobacteria*, *Euglenophyta* ve *Dinophyta*'ya ait toplam 116 adet takson saptanmıştır. Cyanobacteria'dan *Nostoc commune'*nin mayıs ayında bölgesel koloni oluşturduğu bunun yanısıra Ağustos-Eylül aylarında algal artış gözlendiği ve en baskın türün *Microcystis aeuroginosa* olduğu tespit edilmiştir. Bu türlere ise çalışmamız bulgularında rastlanılmamıştır.

Türkiye baraj göllerinin fiziksel, kimyasal ve biyolojik parametreleri hakkında yeterli bilgi olduğunu söylemek zordur. Ülkemizdeki bazı baraj göllerinde yürütülen çalışmaların ise sınırlı olduğu ve belirli bir amaç için yapıldığı görülmektedir. Bunun sonucu olarak, yapılan çalışmalar ancak birkaç yıllık gözlemleri içermekte, bir baraj gölü hakkında geleceğe ve çok amaçlı kullanımlarına yönelik planlama yapılması mümkün olmamaktadır. Karkamış Baraj Gölü'nde yapılan bu çalışma ile hem Türkiye alg veri tabanının oluşturulmasına hem de baraj göllerinde yapılacak çok amaçlı çalışmalara katkı sağlanması düşünülmüştür.

KAYNAKLAR

Açıkgöz, İ ve Baykal, T 2005. Karagöl (Çubuk-Ankara) Alg Florası. S.D.Ü. Eğirdir Su Ürünleri Fak. Der., Cilt I, sayı: 2, 38-55.

Anonymous 2010. Cyanobacteria California Recreational Water Badies: Provading Voluntory Guidance about Harmful Algal Blooms, Their Monitorning and Public Notification.

Bourelly, P 1968. Les Algues D' eau Douce Algues jaunes et Brunes. N. Baues, Paris.

Bourelly, P 1972. Les Algues D' eau Douce Tome: 1, Editions n. Boubee and Cie 3, Place Saınt-Andre_Des-Arts. Paris.

Christoffersen K, Kaas H 2000. Toxic cyanobacteria in water. A guide to their public health consequences, monitoring, and management. Limnology and Oceanography, 45(5): 1212–1212.

Demir N ve Atay D 1999. Kurtboğazı ve Çamlıdere Baraj Göllerinin Fitoplanktonu. X. Ulusal Su Ürünleri Sempozyumu. Adana.

Fakıoğlu Ö, Atamanalp M, Demir N 2011. Baraj göllerinde toksik mavi-yeşil algler. A.Ü. Çevre Bilimleri Der., Cilt 3, Sayı:2, 65-71.

Gantt E 1980. Structure and function of phycobilisomes: Light harvesting pigment complexes in red and blue-green algae. International Review of Cytology, 45–80.

Geitler L, Pascher A 1925. Cyanophyceae In: Pascher, A. Die Susswasser_Flora, Deutschlans, Osterreichs und der Schweiz. Heft 12, VEB Gustav Fischer Verlag., Jena, Stuttgart.

Öztürk (Yılmaz) B 2014. Apa Baraj Gölü algleri üzerine araştırmalar (Çumra/Konya). S.Ü. Fen Bil. Enst., Doktora Tezi, 216 s.

Paerl HW, Fulton RS, Moisander PH, Dyble J 2001. Harmful freshwater algal blooms, with an emphasis on Cyanobacteria. The Scientific World Journal, 1: 76–113.

Ris H 1961. Electron microscope studies on blue-green algae. The Journal of Cell Biology, 9(1): 63–80.

Saad A, Atia A 2014. Review on freshwater blue-green algae (Cyanobacteria): occurrence, classification and toxicology. Bıoscıences Bıotechnology Research Asıa, 11(3):1319-1325.

Sørensen T 1948. A method of establishing groups of equal amplitude in plant sociology based on similarity of species and its application to analyses of the vegetation on danish commons. Kongelige Danske Videnskabernes Selskab 5(4): 1–34.

Sönmez F, Kutlu B, Sesli A 2017. Spatial and temporal distribution of phytoplankton in Karkamış Dam Lake (Şanlıurfa/Turkey). Fresenius Environmental Bulletin, 26(10): 6234-6245.

Visser PM, Ibelings BW, Bormans M, Huisman J 2015. Artificial mixing to control cyanobacterial blooms: A review. Aquatic Ecology, 50(3): 423–441.

URL,1.https://www.msxlabs.org/forum/cevaplanmis/470589-cekirdekli-ve-cekirdeksiz-hucre-nedir.html [12.08.23]

URL, 2. https://yunus.hacettepe.edu.tr/~akbulut/sayfalar/alg.html [12.08.23]

URL,3.https://www.turkcebilgi.com/karkam%C4%B1%C5%9F_baraj%C4%B1_ve_hidroelektrik_santrali [12.08.23]

DRAFT

ORAL PRESENTATION

Covid-19 pandemi döneminde yeme davranış bozuklukları ile akdeniz diyeti bağlılık düzeyinin değerlendirilmesi

Ayşe Nur Elmaskaya[1] (ORCID: https://orcid.org/0000-0002-5990-5483), Fatmagül Yur[2] (*ORCID: https://orcid.org/0000-0002-5536-9169*)

[1]Muğla Muğla Sıtkı Koçman Üniversite, Sağlık Bilimleri Enstitüsü, Beslenme ve Diyetetik Ana Bilim Dalı, Muğla, Türkiye
[2]Muğla Sıtkı Koçman Üniversitesi, Fethiye Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Muğla, Türkiye

*Sorumlu yazar e-mail:aysenurelmaskaya@posta.mu.edu.tr

Özet

Bu araştırma, COVID-19 pandemi sürecindeki duygu durumlarının bireylerde yeme davranış bozuklukları ile Akdeniz diyeti bağlılıkları üzerine etkisini ve bu etmenler arasındaki ilişkiyi belirlemek amacıyla tanımlayıcı araştırma çalışması olarak planlanıp yürütülmüştür. Araştırmanın örneklemini Türkiye'de yaşayan 18-65 yaş aralığında bulunan bireyler oluşturmuştur. Bu çalışma toplam 491(190'ı erkek, 301'i kadın) birey üzerinde yürütülmüştür. Çalışma verileri bireylerin sosyodemografik bilgilerine yönelik sorular, Akdeniz Diyeti Bağlılık Ölçeği (MEDAS), COVID-19 Korku Ölçeği (FCV-19S), Üç Faktörlü Yeme Ölçeği (TFEQ-R21), beslenme alışkanlıkları ile ilgili soruların yer aldığı bir form aracılığıyla online olarak elde edilmiştir. Çalışma sonucunda elde edilen veriler Windows ortamında Statistical Package fort he Social Sciences (SPSS) 25.0 paket programı ile değerlendirilmiş ve tüm hesaplamalarda $p<0.05$ anlamlı kabul edilmiştir. Katılımcıların FCV-19S ile MEDAS ve TFEQ-R21 arasında aynı yönlü anlamlı bir ilişki bulunmuştur($p<0.05$). Katılımcıların TFEQ-R21alt boyutu bilişsel kısıtlama ile MEDAS ve FCV-19S arasında aynı yönlü anlamlı bir ilişki bulunmuştur($p<0.05$). Katılımcıların TFEQ-R21 alt boyutu duygusal yeme ile FCV-19S arasında aynı yönlü anlamlı bir ilişki bulunmuştur ($p<0.05$). Katılımcıların TFEQ-R21 alt boyutu kontrolsüz yeme ile MEDAS arasında ters yönlü anlamlı bir ilişki bulunmuştur($p<0.05$). Katılımcıların TFEQ-R21 alt boyutu kontrolsüz yeme ile FCV-19S arasında aynı yönlü anlamlı bir ilişki bulunmuştur($p<0.05$). Sonuçta yetişkin bireylerde artan COVID-19 korkusunun TFEQ-R21 toplam puanını, duygusal yeme, bilişsel kısıtlama ve kontrolsüz yeme davranış düzeylerini arttırdığı, artan kontrolsüz yeme MEDAS uyumunu düşürdüğü ve artan bilişsel kısıtlama MEDAS uyumunu arttırma eğiliminde olduğu bulunmuştur. Artan FCV-19S artan MEDAS ile ilişkili olduğu bulunmuştur. Pandemi süresince yetişin bireylerin hissettikleri korku seviyelerinin azaltılması, yeme davranışlarına pozitif anlamda etki edilerek MEDAS düzeylerinin arttırılması için disiplinler arası çalışmaların faydalı olacağı düşünülmektedir.

Anahtar Kelimeler: Covid-19, Akdeniz diyeti, Yeme davranışı

Evaluation of mediterranean diet addiction level with eating behavior disorders during the covid-19 pandemic period

Abstract

This research was planned and conducted as a descriptive research study in order to determine the effect of mood states during the COVID-19 pandemic process on eating behavior disorders and Mediterranean diet adherence in individuals and the relationship between these factors. The sample of the study consisted of individuals between the ages of 18-65 living in Turkey. This study was conducted on a total of 491(190 male, 301 female) individuals. The study data were obtained online through a form including questions about the sociodemographic information of individuals, Mediterranean Diet Adherence Scale (MEDAS), COVID-19 Fear Scale (FCV-19S), Three-Factor Eating Scale (TFEQ-R21), and nutritional habits. The data obtained as a result of the study were evaluated with the Statistical Package for the Social Sciences (SPSS) 25.0 package program in the Windows environment and $p<0.05$ was considered significant in all calculations. A statistically

DRAFT

significant correlation was found between the participants' FCV-19S and MEDAS and TFEQ-R21($p<0.05$). A significant correlation was found between the participants' TFEQ-R21 sub-dimension cognitive restriction and MEDAS and FCV-19S($p<0.05$). A significant correlation was found between the participants' TFEQ-R21 sub-dimension emotional eating and FCV-19S($p<0.05$). A significant inverse relationship was found between the participants' TFEQ-R21 sub-dimension, uncontrolled eating, and MEDAS($p<0.05$). A significant correlation was found between the participants' TFEQ-R21 sub-dimension, uncontrolled eating, and FCV-19S($p<0.05$). As a result, it was found that increased fear of COVID-19 in adults increased TFEQ-R21 total score, emotional eating, cognitive restriction and uncontrolled eating behavior levels, increased uncontrolled eating decreased MEDAS compliance, and increased cognitive restriction tended to increase MEDAS compliance. Increased FCV-19S was found to be associated with increased MEDAS. It is thought that interdisciplinary studies will be beneficial to reduce the level of fear felt by adults during the pandemic and to increase MEDAS levels by positively affecting their eating behaviors.

Keywords: Covid-19, Mediterranean diet, Eating behavior

GİRİŞ

Covid-19, tüm dünyayı etkisi altına alan, ivedilikle el atılması gerekli olan bir sağlık krizidir (Bavel vd., 2020). Türkiye'de mart ayında, ilk vakanın görülmesi ile birlikte Covid-19 virüsünün yayılmasını kontrol altına almak için diğer ülkelerde olduğu gibi sıkı tedbirler uygulanmaya başlanmıştır (T.C. Sağlık Bakanlığı, 2021). Bu tedbirler enfeksiyon oranını azaltmaya yardımcı olurken nüfusun alışkanlıklarında ani ve radikal bir değişime neden olmuştur (Hossain vd., 2020). Sosyal etkileşim kurmaya alışkın olan insanların arkadaş ve aileleri ile mesafelerini korumak zorunda kalmaları, aniden daha fazla veya tamamen yalnız kalmaları, kısıtlamalar nedeniyle günlük rutinlerinin kesintiye uğraması, medyadan sürekli olarak Covid-19 ile ilgili haberlerin duyulması veya okunması anksiyete ve stresi artırmıştır (Todisco & Donini, 2021). İnsanlar, olumsuz hislerin üstesinden gelebilmek amacıyla çeşitli yeme davranışlarına yönelebilirler (Kaner vd., 2023). Duygusal durumlar, besin alımını arttırabildiği gibi bazı bireylerde ise azaltmaktadır (Konttinen, 2020). Sağlığın en önemli yapı taşlarından biri beslenmedir. Beslenme tedavisi, birçok akut, kronik ve bulaşıcı hastalıklar olmak üzere çeşitli hastalıkların tedavisinde büyük önem taşımaktadır. Destekleyici beslenme tedavisinin Ebola virüs salgınında vaka ölüm oranını azalttığı gözlenmiştir. SARS-CoV-2 için de aynı durumun geçerli olduğu düşünülmektedir (Kartal vd., 2020). Literatürde Covid-19'dan korunmada adından en sık bahsettiren diyet çeşidi, Akdeniz diyetidir. Birçok otorite bu dönemde meyve ve sebze alımının artmasını, işlenmiş gıda alımının azaltılmasını önermektedir (Yasar & Aytekin, 2021). Yaşanılan pandemi süreci gibi toplumun yaşamını etkileyen süreçlerde, toplumun sağlıklı bir şekilde baş etmesini sağlayabilmek, sekonder hastalık riskini azaltabilmek ve uygun tedbirler alabilmek için bu süreçlerde meydana gelen duygu durum değişikliğinin yeme davranış ve diyet kaliteleri üzerine etkilerini araştırmak önemlidir. Bu çalışmada, pandemi süresince yetişkin bireylerin duygu durumlarının belirlenip yeme davranışı ve Akdeniz diyeti bağlılık düzeyi ile aralarındaki ilişkilerin araştırılması amaçlanmıştır.

MATERYAL VE METOT

Araştırmanın Türü

Bu çalışma, Covid-19 pandemi süresince yetişkinlerin duygu durumları ile yeme davranışları ve Akdeniz diyeti bağlılıklarının belirlenerek aralarındaki ilişkinin değerlendirilmesi amacıyla kesitsel ve betimleyici tipte tasarlanmıştır. Veriler 1 Ekim 2021-31 Aralık 2021 tarihleri arasında toplanmıştır.

Araştırma Evreni ve Örneklemi

Araştırmanın evrenini Türkiye'deki 18-65 yaş aralığında olan, araştırmaya katılmaya gönüllü olan, okuma yazması olan, sosyal ağ siteleri ve uygulamalarından (İnstagram, Facebook ve WhatsApp) en az birini kullanan bireyler oluşturmaktadır. G*Power 3.1.9.4 programı ile yapılan analizde 0.05 hata payı, 0.95 güç ve 0.5 etki büyüklüğü ile örneklem büyüklüğü hesaplandığında çalışmaya alınması gereken kişi sayısı minimum 210 bulunmuştur.

Veri Toplama Araçları

Genel tanıtıcı bilgiler, beslenme durumu ile ilgili sorular, Akdeniz Diyeti Bağlılık Ölçeği, Üç Faktörlü Yeme Ölçeği ve COVID-19 Korku Ölçeği'nin bulunduğu anket formu "Google forms" ile oluşturulduktan sonra bireylere araştırmacı tarafından sosyal ağ siteleri ve uygulamaları (Instagram, Facebook ve WhatsApp) ya da e-posta yoluyla ulaştırılmıştır. Genel tanıtıcı bilgiler formu, araştırmacılar tarafından oluşturulan on bir soruluk bir bölümdür. Beslenme alışkanlıkları formu, araştırmacılar tarafından geliştirilen bu form pandemi süresince ara ve ana öğün sayıları, öğün atlama durumları ile besin takviyesi kullanımının sorgulandığı dört soruluk bir bölümdür. Akdeniz Diyeti Bağlılık Ölçeği 2012 yılında Martinez-Gonzalez ve arkadaşları tarafından geliştirilmiştir (Martínez-González vd., 2012). Akdeniz Diyeti Bağlılık Ölçeği 14 sorudan oluşan bir ankettir. Tüketim miktarına göre sorulan her soru için 1 ya da 0 puan alınmakta olup, toplam puanın hesaplaması yapılmaktadır. Toplam puanın 7 ve üzerinde olması bireyin Akdeniz diyetine kabul edilebilir derece uyumunun olduğunu, 9 ve üzerinde olması ise bireyin Akdeniz diyetine sıkı uyumunun olduğunu göstermektedir. Akdeniz Diyeti Uyum Skorlaması (ADS) Akdeniz Diyeti Bağlılık Ölçeği'nin Türkçe 'ye Uyarlanması Geçerlilik ve Güvenilirliği çalışmasından alınmıştır (Özkan Pehlivanoğlu vd., 2020). Bu çalışmada, katılımcıların Covid-19 pandemi döneminde duydukları korku düzeyinin tespit edilmesi için COVID-19 Korku Ölçeği kullanılmıştır. FCV-19S, Ahorsu ve arkadaşları tarafından geliştirilen tek boyutlu yedi maddeli, beş puanlı Likert ölçeğidir (Ahorsu vd., 2020). Bu çalışmada, katılımcıların yeme davranışlarını değerlendirmek için üç faktörlü yeme ölçeği formu kullanılmıştır. Bu ölçeğin orijinali 51 madde içermekte ve iki bölümden oluşmaktadır. Baş, Bozan & Ciğerim (2008) tarafından Türkiye'de yapılan çalışmada 51 maddeden oluşan Üç Faktörlü Yeme Ölçeği kullanılmıştır (Baş vd., 2008). Buna ek olarak Karakuş, Yıldırım & Büyüköztürk (2016) tarafından bu ölçeğin 21 maddelik formu Türk kültürüne uyarlanmıştır. Bu ölçek yeme davranışını bilişsel kısıtlama, kontrolsüz yeme ve duygusal yeme adı altındaki üç alt faktör ile ölçmektedir (Karakuş vd., 2016).

BULGULAR

Araştırmaya Türkiye'de yaşayan 18-65 yaş aralığındaki 301'i kadın, 190'ı erkek olmak üzere toplam 491 yetişkin birey katılmıştır. Sosyo-demografik özelliklere ilişkin frekans dağılımı Tablo 1 'de sunulmuştur.

Tablo 1. Sosyo-demografik özelliklere ilişkin frekans dağılım tablosu

Değişkenler		N	%
Cinsiyet	Kadın	301	61.30
	Erkek	190	38.70
Medeni Durum	Evli	106	21.59
	Bekar	385	78.41
Eğitim Durumu	Ortaokul ve altı	16	3.26
	Lise	70	14.26
	Lisans	310	63.14
	Lisansüstü	95	19.35
Çalışma Durumu	Çalışıyor	206	41.96
	Çalışmıyor	285	58.04
Kronik Rahatsızlık	Evet	72	14.66
	Hayır	419	85.34
BKI Sınıflandırılması	Zayıf	30	6.11
	Normal	302	61.51
	Hafif şişman	120	24.44
	Obez	39	7.94
Covid-19 pandemi süresince vücut ağırlığınızda değişiklik olma durumu	Artış oldu	220	44.81
	Azalma oldu	77	15.68
	Değişmedi	170	34.62
	Bilmiyorum	24	4.89
Covid-19 pandemi öncesi haftada en az 150 dk egzersiz yapma durumu	Evet	182	37.07
	Hayır	309	62.93
Covid-19 pandemi sürecinde haftada en az 150 dk egzersiz yapma durumu	Evet	140	28.51
	Hayır	351	71.49

N: Sayı, %: Yüzde.

Tablo 1'de yer alan verilere göre çalışmaya katılan bireylerin %61.30'unun kadın, %38.70'inin erkek olduğu belirtilmiştir. Araştırmaya katılan yetişkinlerin %21.59'unun evli, %78.41'inin bekar medeni durumuna sahip olarak bildirilmiştir. Katılımcıların %3.26'sının eğitim düzeyinin ortaokul ve altı, %14.26'sının lise, %63.14'ünün lisans ve %19.35'inin eğitim düzeyinin lisansüstü olduğu saptanmıştır. Katılımcıların %41.96'sı çalıştığını, %58.04'ü ise çalışmadığını belirtmiştir. Çalışmaya katılanlardan %14.66'sının doktor tarafından tanısı konulmuş kronik rahatsızlığının olduğunu, geriye kalan %85.34'ünün herhangi bir kronik hastalığının olmadığı bildirilmiştir. BKİ sınıflandırılmasına göre; çalışmaya katılan bireylerin %6.11'inin zayıf, %61.51'inin normal, %24.44'ünün hafif şişman ve %7.94'ünün obez olduğu saptanmıştır. Katılımcıların %44.81'inin pandemi sürecinde vücut ağırlığında artış olduğu, %15.68'inde vücut ağırlığında azalma olduğu, %34.62'sinin vücut ağırlığında değişiklik olmadığını bildirmiştir. Çalışmaya katılanların %37.07'sinin pandemi öncesi egzersiz yaptığı, %28.51'inin pandemi sürecinde egzersiz yaptığı görülmektedir. Pandemi öncesi dönemde egzersiz yapmadığını belirtenler katılımcıların %62,93'ü iken pandemi sırasında egzersiz yapmadığını bildirenler katılımcıların %71,49'u olarak saptanmıştır.

Tablo 2. Ölçeklerin BKİ sınıflandırılmasına göre karşılaştırılması

		BKİ Kategorik		Kruskal Wallis H Testi		
		$\bar{X}$	Ss	H	P	Fark**
MEDAS	Zayıf	5.93	1.86	2.049	0.568	-
	Normal	6.33	1.83			
	Hafif şişman	6.11	1.96			
	Obez	6.13	1.89			
FCV-19S	Zayıf	15.30	7.73	4.894	0.18	-
	Normal	16.01	7.36			
	Hafif şişman	15.08	7.05			
	Obez	17.92	7.92			
TFEQ-R21	Zayıf	36.90	13.62	39.875	0.001*	1<2
	Normal	45.72	13.25			1<3
	Hafif şişman	50.12	11.83			1<4
	Obez	55.77	11.34			2<3
						2<4
Bilişsel Kısıtlama	Zayıf	10.37	4.60	23.059	0.001*	1<2
	Normal	14.05	4.96			1<3
	Hafif şişman	14.96	4.04			1<4
	Obez	14.15	4.30			2<3
						2<4
Duygusal Yeme	Zayıf	10.43	4.76	33.455	0.001*	1<2
	Normal	11.82	5.61			1<3
	Hafif şişman	13.28	6.00			1<4
	Obez	17.72	5.95			2<3
						2<4
Kontrolsüz Yeme	Zayıf	18.80	5.99	19.563	0.001*	1<2
	Normal	19.85	6.49			1<3
	Hafif şişman	21.88	6.52			1<4
	Obez	23.90	6.42			2<3
						2<4

$\bar{X}$: Aritmetik ortalama, Ss: Standart sapma, *p<0.05

Tablo 2'de bulunan verilere göre; Akdeniz diyeti bağlılık düzeyleri açısından BKİ durumları arasında istatistiksel olarak anlamlı bir farklılık bulunmamaktadır (p>0.05). Covid-19 korku düzeyleri açısından BKİ durumları arasında istatistiksel olarak anlamlı bir farklılık bulunmamaktadır (p>0.05). Üç faktörlü yeme düzeyleri açısından BKİ durumları arasında istatistiksel olarak anlamlı bir farklılık bulunmaktadır (p<0.05). Zayıf BKİ sınıflamasına sahip olan katılımcıların üç faktörlü yeme düzeyleri normal, hafif şişman ve obez olanlara göre anlamlı derecede düşük, normal kilolu olanların üç faktörlü yeme düzeyleri hafif şişman ve obez olanların düzeylerine göre anlamlı derecede düşüktür. Bilişsel kısıtlama düzeyleri açısından BKİ durumları

arasında istatistiksel olarak anlamlı bir farklılık bulunmaktadır (p<0.05). Zayıf BKİ sınıflamasına sahip olan katılımcı bireylerin bilişsel kısıtlama düzeyleri normal, hafif şişman ve obez olanlara göre anlamlı derecede düşük, normal kilolu olanların bilişsel kısıtlama hafif şişman ve obez olanların düzeylerine göre anlamlı derecede düşüktür. Duygusal yeme düzeyleri açısından BKİ durumları arasında istatistiksel olarak anlamlı bir farklılık bulunmaktadır (p<0.05). Zayıf BKİ sınıflamasına sahip olanların duygusal yeme düzeyleri normal, hafif şişman ve obez olanlara göre anlamlı derecede düşük, normal kilolu olanların duygusal yeme hafif şişman ve obez olanların düzeylerine göre anlamlı derecede düşüktür. Kontrolsüz yeme düzeyleri açısından BKİ durumları arasında istatistiksel olarak anlamlı bir farklılık bulunmaktadır (p<0.05). BKİ kategorisi zayıf olanların kontrolsüz yeme düzeyleri normal, hafif şişman ve obez olanlara göre anlamlı derecede düşük, normal kilolu olanların kontrolsüz yeme hafif şişman ve obez olanların düzeylerine göre anlamlı derecede düşüktür.

Tablo 3. Ölçek düzeyleri arasındaki ilişki

		MEDAS	FCV-19S	TFEQ-R21	Bilişsel Kısıtlama	Duygusal Yeme	Kontrolsüz Yeme
MEDAS	r	1.000					
	p						
FCV-19S	r	.116	1.000				
	p	0.010					
TFEQ-R21	r	-0.065	.336**	1.000			
	p	0.148	0.000*				
Bilişsel Kısıtlama	r	.125**	.208**	.543**	1.000		
	p	0.005*	0.000*	0.000*			
Duygusal Yeme	r	-0.084	.250**	.858**	.270**	1.000	
	p	0.062	0.000	0.000*	0.000*		
Kontrolsüz Yeme	r	-1.39**	.317**	.829**	.158**	.631**	1.000
	p	0.002*	0.000*	0.000*	0.000*	0.000*	

*p<0.05; **Korelasyon Katsayısı.

Tablo 3'te bulunan verilere göre; Covid-19 korku düzeyi ile Akdeniz diyeti bağlılık düzeyleri arasında aynı yönlü gücü zayıf dereceli istatistiksel olarak anlamlı bir ilişki bulunmaktadır (r=0.116; p<0.05). Covid-19 korku düzeyleri arttıkça Akdeniz diyet bağlılık düzeyleri de artmaktadır. Covid-19 korku düzeyi ile üç faktörlü yeme ölçek düzeyleri arasında aynı yönlü gücü zayıf dereceli istatistiksel olarak anlamlı bir ilişki bulunmaktadır (r=0.336; p<0.05). Covid-19 korku düzeyleri arttıkça üç faktörlü yeme ölçek düzeyleri de artmaktadır. Bilişsel kısıtlama düzeyleri ile Akdeniz diyeti bağlılık düzeyleri arasında aynı yönlü gücü zayıf dereceli istatistiksel olarak anlamlı bir ilişki bulunmaktadır (r=0.125; p<0.05). Bilişsel kısıtlama düzeyleri arttıkça Akdeniz diyeti bağlılık ölçek düzeyleri de artmaktadır. Bilişsel kısıtlama düzeyleri ile Covid-19 korku düzeyleri arasında aynı yönlü gücü zayıf dereceli istatistiksel olarak anlamlı bir ilişki bulunmaktadır (r=0.208; p<0.05). Bilişsel kısıtlama düzeyleri arttıkça Covid-19 korku düzeyleri de artmaktadır. Duygusal yeme düzeyleri ile Covid-19 korku düzeyleri arasında aynı yönlü gücü zayıf dereceli istatistiksel olarak anlamlı bir ilişki bulunmaktadır (r=0.250; p<0.05). Duygusal yeme düzeyleri arttıkça Covid-19 korku düzeyleri de artmaktadır. Kontrolsüz yeme düzeyleri ile Akdeniz diyeti bağlılık düzeyleri arasında ters yönlü gücü zayıf dereceli istatistiksel olarak anlamlı bir ilişki bulunmaktadır (r=-0.139; p<0.05). Kontrolsüz yeme düzeyleri arttıkça Akdeniz diyeti bağlılık ölçek düzeyleri de azalmaktadır. Kontrolsüz yeme düzeyleri ile Covid-19 korku düzeyleri arasında aynı yönlü gücü zayıf dereceli istatistiksel olarak anlamlı bir ilişki bulunmaktadır (r=0.317; p<0.05). Kontrolsüz yeme düzeyleri arttıkça Covid-19 korku düzeyleri de artmaktadır. Diğer düzeyleri arasında istatistiksel olarak anlamlı bir ilişki bulunmamaktadır (p>0.05).

TARTIŞMA

Literatürde bulunan bir araştırmada, duygusal yeme BKİ artışına sebep olduğu ve bununla birlikte duygusal yeme davranışının bireylerin ideal BKİ ulaşmalarına engel olduğu belirtilmiştir (Sainsbury vd., 2019). Başka bir çalışmada ise normal vücut ağırlığında olan katılımcıların bilişsel kısıtlama seviyeleri fazla kilolu olan katılımcılara göre anlamlı ölçüde yüksek olduğu belirtilmiştir (Şen & Kabaran, 2021). Fazla kilolu ve obez sınıflamasındaki grubun TFEQ-R21 toplam puanı anlamlı derecede daha yüksek olduğu bildirilmiştir. BKİ

seviyesi arttıkça duygusal yeme ve kontrolsüz yeme alt boyut yeme davranışları artış gösterdiği rapor edilmiştir (Valencio vd., 2022). İtalya'da genç yetişkin bireyler üzerinde yapılan bir araştırmada, katılımcıların duygusal yeme düzeyleri arttıkça BKİ seviyelerinin artış gösterdiği saptanmıştır (Guerrini-Usubini vd., 2023). Negatif hislerle baş etme durumlarında hafif şişman ve obez BKİ gruplamasına sahip olan bireyler; zayıf ve normal kiloya sahip olanlara kıyasla daha çok yeme davranışına yönelmeye yatkın oldukları bildirilmektedir (Geliebter & Aversa, 2003). Bu çalışma, BKİ sınıflamasına göre zayıf olan katılımcıların üç faktörlü yeme, bilişsel kısıtlama, kontrolsüz yeme ve duygusal yeme düzeyleri; normal, hafif şişman ve obez olan katılımcılara kıyasla anlamlı derecede düşük iken normal olanların üç faktörlü yeme düzeyleri; hafif şişman ve obez olanların düzeylerine göre anlamlı derecede düşük bulunmuştur (Tablo 2). Elde edilen bulgulara göre, literatür ile paralel olarak BKİ sınıflama derecesi arttıkça duygusal yeme, kontrolsüz yeme ve bilişsel kısıtlama seviyelerinin artış gösterdiği görülmektedir. Dolayısıyla yeme davranışlarındaki düzensizliğin vücut ağırlıklarının kontrolü üzerinde önemli bir etkisi olduğu gibi bireylerin mevcut BKİ değerlerinin yeme davranışlarını da etkilediği düşünülebilir.

Ülkemizde pandemi zamanı yapılmış olan diyet kalitesi ile Covid-19 kaynaklı kaygı durumunun aralarındaki ilişkinin araştırıldığı bir çalışmada, korku ölçek puanlaması ile Akdeniz diyeti uyumu arasında bir fark gözlemlenmediği rapor edilmiştir (Sevim vd., 2021). Yetişkinler üzerinde yapılan pandemi korku düzeyi ile Akdeniz diyeti uyum durumunun değerlendirildiği bir çalışmada, bireylerin korku düzeyine göre besin seçiminin farklılık gösterdiği belirtilmiş olup düşük korku seviyesine sahip katılımcı bireylerin Akdeniz diyetine uyum düzeyi, korku seviyesinin yüksek ve orta düzeyde olan katılımcıların Akdeniz diyeti uyum puanlarına göre daha düşük seviyede olduğunu bildirmiştir. Fakat; bireyler Covid-19 korku seviyesi ile Akdeniz diyeti bağlılıkları arasında anlamlı bir ilişki bulunmadığı belirtilmiştir (Gumus vd., 2023). Bu çalışmada, katılımcıların Covid-19 korku düzeylerinin artması ile Akdeniz diyeti bağlılık seviyelerinin de arttığı tespit edilmiştir (Tablo 3). Bireylerde pandemi döneminde duyulan virüse yakalanma korku durumunun bağışıklık sistemini güçlendirmek amacıyla daha sağlıklı ve dengeli beslenmeye yönelim konusunda etkili olduğu düşünülebilir.

Sağlık çalışanları üzerinde yapılan bir çalışmaya göre TFEQ-R18 puanlamasındaki artış stres ile korku seviyelerini azalttığı tespit edilerek; hissedilen korku seviyelerinin yeme davranışlarını olumsuz olarak etki ettiğini belirtmiştir (Üstün & Yilmaz, 2023). Covid-19 korkusunun aracılığı ile yetişkin bireyler üzerinde olumsuz hislerin duygusal yeme davranış düzeyine anlamlı derecede etki ettiği bulunmuştur (Dominte vd., 2022). Bu çalışmada, katılımcıların Covid-19 korku düzeyi artış gösterdikçe üç faktörlü yeme ölçeği toplam puanı, ölçek alt boyutu olan bilişsel kısıtlama, duygusal yeme ve kontrolsüz yeme düzeyleri de artış gösterdiği saptanmıştır (Tablo 3). Literatür araştırmasına göre, korku gibi kaygılı ve stresli dönemlerde, bireylerin duygularını beslenmeye yönelerek kontrol etmeye çalıştıklarını göstermektedir. Bu sebeple, Covid-19 korkusu yeme davranışlarında bozuklukları tetikleyeceği düşünülmektedir.

Yapılan bir çalışmada; ruh hali, beslenme davranışları ve diyet kalitesi ile ilgi çalışmada fazla kilolu ve obez metabolik sendromlu erkek bireylere Akdeniz diyeti uyumu yüksek beslenme planları 12 ay boyunca uygulanmış ve hiçbir müdahale yapılmayan grup ile çalışma sonuna değerlendirilme yapılmıştır. Müdahale edilen grubun duygusal ve kontrolsüz yeme davranışında azalma görülürken bilişsel kısıtlayıcı yeme davranışına artış olduğu bildirilmiştir. Katılımcıların müdahale sonucunda vücut ağırlıklarında herhangi bir değişim olmadığı bildirilse de Akdeniz diyeti uyumları artış gösterdiği bildirilmiştir (Paans vd., 2020). Bu çalışmada, yeme davranışları alt boyutlarından bilişsel kısıtlama arttığında Akdeniz diyeti uyum düzeyinin de yükseldiği tespit edilmiştir (Tablo 3). Ayrıca kontrolsüz yeme davranış seviyesinin artması ile Akdeniz diyeti bağlılık düzeyinin azaldığı saptanmıştır (Tablo 3). Bu bağlamda, toplumda Akdeniz diyeti bağlılığının arttırılması ile ilgili teşvik ve çalışmalarının yeme davranışlarını olumlu yönde etkileyeceğini düşündürmektedir.

SONUÇ

Covid-19 korku düzeyleri arttıkça Akdeniz diyet bağlılık ve üç faktörlü yeme davranış düzeyleri de artış göstermektedir. Bilişsel kısıtlama yeme davranış düzeyleri arttıkça Akdeniz diyeti bağlılık ve Covid-19 korku düzeyleri de artış gösterdiği bulunmuştur. Duygusal yeme düzeyleri arttıkça Covid-19 korku düzeyleri artmaktadır. Kontrolsüz yeme düzeyleri arttıkça Akdeniz diyeti bağlılık ölçek düzeyleri azalırken Covid-19 korku düzeyleri artış gösterdiği bulunmuştur. Zayıf BKİ derecesine sahip olanların üç faktörlü yeme, bilişsel

kısıtlama, kontrolsüz yeme ve duygusal yeme düzeyleri; normal, hafif şişman ve obez olanlara göre anlamlı derecede düşük, normal kilolu olanların üç faktörlü yeme düzeyleri; hafif şişman ve obez olanların düzeylerine göre anlamlı derecede düşük düzeyde bulunmuştur.

TEŞEKKÜR

Çalışmaya katılan tüm gönüllülere teşekkür ederiz.

KAYNAKLAR

Ahorsu DK, Lin CY, Imani V, Saffari M, Griffiths MD ve Pakpour AH (2020). The Fear of COVID-19 Scale: Development and Initial Validation. Int J Ment Health Addict, 1-9. DOI:10.1007/s11469-020-00270-8.

Bavel JV, Baicker K, Boggio PS, Capraro V, Cichocka A, Cikara M (2020). Using social and behavioural science to support COVID-19 pandemic response. Nature Human Behaviour, 460-471. DOI:10.1038/s41562-020-0884-z.

Bas M, Bozan N ve Cigerim N (2008). Dieting, dietary restraint, and binge eating disorder among overweight adolescents in Turkey. Adolescence, 43(171), 635. Erişim Adresi: https://www.researchgate.net/profile/Murat-Bas3/publication/23667668_Dieting_dietary_restraint_and_binge_eating_disorder_among_overweight_adolescents_in_Turkey/links/563caa7a08ae405111aa3fc7/Dieting-dietaryrestraint-and-binge-eating-disorder-among-overweight-adolescents-in-Turkey.pd

Dominte ME, Swami V ve Enea V (2022). Fear of COVID-19 mediates the relationship between negative emotional reactivity and emotional eating. Scandinavian Journal of Psychology, 63(5), 462-467. DOI:10.1111/sjop.12828.

Geliebter A ve Aversa A (2003). Emotional eating in overweight, normal weight, and underweight individuals. Eating Behaviors, 3(4), 341-347. DOI:10.1016/S1471- 0153(02)00100-9.

Guerrini-Usubini A, Cattivelli R, Scarpa A, Musetti A, Varallo G, Franceschini C ve Castelnuovo G (2023). The interplay between emotion dysregulation, psychological distress, emotional eating, and weight status: A path model. International Journal of Clinical and Health Psychology, 23(1). DOI:10.1016/j.ijchp.2022.100338.

Gumus D, Topal GG, Sevim S ve Kizil M (2023). Adherence to Mediterranean diet and dietary changes according to the fear of COVID-19 during the pandemic: a cross-sectional study. Journal of Nutritional Science, 12, e56. DOI:10.1017/jns.2023.40.

Hossain MM, Sultana A ve Purohit N (2020). Mental health outcomes of quarantine and isolation for infection prevention: a systematic umbrella review of the global evidence. Epidemiology and Health. DOI:10.4178/epih.e2020038.

Kaner G, Yurtdaş Depboylu G, Çalık G, Yalçın T ve Nalçakan T (2023). Evaluation of perceived depression, anxiety, stress levels and emotional eating behaviours and their predictors among adults during the COVID-19 pandemic. Public Health Nutrition, 26(3), 674-683. DOI:10.1017/S1368980022002579.

Karakuş SŞ, Yıldırım H ve Büyüköztürk Ş (2016). Üç faktörlü yeme ölçeğinin Türk kültürüne uyarlanması: Geçerlik ve güvenirlik çalışması. TAF Preventive Medicine Bulletin, 15(3), 229-237. DOI:10.5455/pmb.1-1446540396.

Kartal A, Ergin E ve Kanmış HD (2020). COVID-19 Pandemik Salgın Döneminde Yaşam Kalitesini Arttırmaya Yönelik Sağlıklı Beslenme ve Fiziksel Aktivite Önerileri. Avrasya Sağlık Bilimleri Dergisi, 149-155. Erişim Adres: https://dergipark.org.tr/en/pub/avrasyasbd/issue/56010/747805

Konttinen H (2020). Emotional eating and obesity in adults: the role of depression, sleep and genes. Proceedings of the Nutrition Society, 283-289. DOI:10.1017/S0029665120000166.

Martínez-González MA, García-Arellano A, Toledo E, Salas-Salvado J, Buil-Cosiales P, Corella D (2012). A 14-item Mediterranean diet assessment tool and obesity indexes among highrisk subjects: the PREDIMED trial. DOI: 10.1371/journal.pone.0043134.

Özkan Pehlivanoğlu EF, Balcıoğlu H ve Ünlüoğlu İ (2020). Akdeniz Diyeti Bağlılık Ölçeği'Nin Türkçe'ye Uyarlanması Geçerlilik ve Güvenirliği. Osmangazi Tıp Dergisi, 160-164. DOI: 10.20515/otd. 504188.

Paans NP, Bot M, Brouwer IA, Visser M, Gili M, Roca M (2020). Effects of food-related behavioral activation therapy on eating styles, diet quality and body weight change: Results from the MooDFOOD Randomized Clinical Trial. Journal of Psychosomatic Research, 137. DOI:10.1016/j.jpsychores.2020.110206.

Sainsbury K, Evans EH, Pedersen S, Marques MM, Teixeira PJ, Lähteenmäki L (2019). Attribution of weight regain to emotional reasons amongst European adults with overweight and obesity who regained weight following a weight loss attempt. Eating and Weight Disorders- Studies on Anorexia, Bulimia and Obesity, 24(2), 351-361. DOI:10.1007/s40519-018-0487-0.

Sevim S, Topal GG, Gumus D ve Kizil M (2021). Turkish population's adherence to the mediterranean diet and fear of Covid-19 during Covid-19 pandemic lockdowns. Clin Nutr ESPEN. DOI:10.1016/j.clnesp.2021.09.206.

Şen G ve Kabaran S (2021). Beslenme Durumunun Duygusal Yeme, Gece Yeme ve Uyku Kalitesi Üzerindeki Etkileri. Kocaeli Üniversitesi Sağlık Bilimleri Dergisi, 7(3), 284-295. DOI:10.30934/kusbed.952227.

TC Sağlık Bakanlığı (2021, Mayıs 6). COVID-19 Bilgilendirme Platformu. Erişim Adresi: https://covid19.saglik.gov.tr/TR-66300/covid-19-nedir-.html

Todisco P ve Donini LM (2021). Eating disorders and obesity (ED&O) in the COVID 19 storm. Eating and Weight Disorders- Studies on Anorexia, Bulimia and Obesity, 747-750. Erişim Adresi:https://link.springer.com/article/10.1007/s40519-020-00938-z

Üstün D ve Yilmaz S (2023). Determining the Effect of Stress and Anxiety on Eating Attitudes in Healthcare Providers Working on COVID-19 Wards. Ecology of Food and Nutrition. DOI:10.1080/03670244.2023.2187384.

Valencio AC, Antunes AB, Fonseca L, Araujo J, Silva MC, Costa M (2022). Associations between Overweight and Obesity and Common Mental Disorders and Eating Behaviors of Adult Women. Obesities, 2(4), 350-360. DOI:10.3390/obesities2040029.

Yasar RK ve Aytekin ÖÜ (2021). COVID-19 ve Beslenme Arasındaki İlişkiye Güncel Bir Bakış. Akademik Gıda, 108-115. Erişim Adresi: https://dergipark.org.tr/en/pub/akademikgida/issue/62016/927735

DRAFT

ORAL PRESENTATION

Mehmet Akif Ersoy Üniversitesi Öğrencilerinde Beden Memnuniyeti ve Duygusal İştahın Yeme Farkındalığı Üzerindeki Etkisi

Feraye Elvan Koçak[1](*https://orcid.org/0009-0003-7736-0493*, Sebahat Demirbilek[1] (*https://orcid.org/0000-0005-3790-2053)*, Simge Can[1](*https://orcid.org/0009-0004-1710-9325*, Mustafa ÖZGÜR[1]* (*https://orcid.org/0000-0001-9724-9571*)

[*1] Burdur Mehmet Akif Ersoy Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Burdur, Türkiye.

*Sorumlu yazar e-mail: mozgur@mehmetakif.edu.tr

Özet

Bireylerin beden algıları duygu durumlarını ve duygu durumları da beslenme davranışlarını değiştirebilmektedir. Bu çalışmanın amacı öğrencilerin yeme davranışlarının duygusal durum ve beden memnuniyetsizliği doğrultusunda farklılaşıp farklılaşmadığını belirlemektir. Kesitsel olarak planlanan araştırmanın evreni Burdur Mehmet Akif Ersoy Üniversitesi öğrencileridir. Online anket olarak planlanan çalışmaya Şubat- Mayıs 2023 tarihleri arasında toplam 141 kadın, 35 erkek olmak üzere 176 kişi katılmıştır. Katılımcılara uygulanan ankette sosyo-demografik ve antropometrik bilgiler, öğrencilerin beden memnuniyetleri, Hollanda Yeme Davranışı Ölçeği (DEBQ) ve Yeme Farkındalığı Ölçeği (YFÖ) yer almıştır. Yaş, cinsiyet ve BKİ bağımlı değişken, DEBQ ve YFÖ ise bağımsız değişken olarak değerlendirilmiştir. Çalışmaya katılan kadın öğrencilerin yaş ortalaması 22,1±2,5 yıl, erkeklerin ise 25,1±7,3 yıldır. Kadın öğrencilerin %51,8'i, erkek öğrencilerin %22,9'u daha ince bir vücuda sahip olmak istediğini belirtmiştir. Kadın öğrencilerin erkeklere kıyasla şimdiki vücut ağırlığından daha az memnun olduğu tespit edilmiştir. Kadın öğrencilerin yalnızca %12,8'i, erkek öğrencilerin yalnızca %22,9'u öğün atlamadığını belirtmiştir. Çalışmaya katılan kadın öğrencilerin DEBQ puanları erkek öğrencilere kıyasla daha yüksektir. Fakat sadece duygusal yeme puanları istatistiksel olarak anlamlı farklılık göstermektedir. Mevcut ağırlıklarından memnun olmayanların duygusal yeme ve kısıtlayıcı yeme davranış düzeyleri yüksek, dışsal yeme davranış düzeyleri ise düşüktür. Kadın öğrencilerin yeme farkındalığı ölçeği alt boyutlarından yeme kontrolü davranışı erkek öğrencilere kıyasla istatistiksel olarak anlamlı düzeyde daha yüksektir ($p<0,05$). Üniversite eğitim dönemi öğrencilerin hayatında yeni bir hayata geçiş dönemidir ve bu dönemde özellikler kadın öğrencilerde duygusal davranışlar dış faktörler ile beraber beslenme davranışlarını oldukça etkilemektedir. Bu nedenle üniversite öğrencilerine sağlıklı beslenme davranışları kazandırılmalı kadın öğrencilerde sağlıklı beden farkındalığı oluşturulmalıdır.

Anahtar kelimeler: Beslenme, duygusal yeme davranışı, beden memnuniyeti

Abstract

Individuals' perceptions of their bodies may affect their emotional states, and emotions can affect their eating habits. The aim of this study is to determine whether emotional states and body dissatisfaction have an impact on students' eating patterns. Students at Burdur Mehmet Akif Ersoy University constitute the study's population. Between February and May 2023, 176 individuals (141 women and 35 men) participated in the study, which was designed as an online survey. Students' body satisfaction, the Dutch Eating Behavior Scale (DEBQ), and the Mindful Eating Questionnare (MEQ) were among the items on the questionnaire given to the participants. Age, gender and BMI were considered as dependent variables, while DEBQ and MEQ were considered as independent variables. The mean age of the female students participating in the study was 22.1±2.5 years, and 25.1±7.3 years for males. 51.8% of female students and 22.9% of male students stated that they wanted to have a slimmer body. It has been determined that female students are less satisfied with their current body weight compared to males. Only 12.8% of students in the female and 22.9% of students in the male reported not to skip meals. Female study participants scored higher on the DEBQ than male participants.

However, the only scores that differ statistically are those related to emotional eating. High levels of emotional eating and restrictive eating behavior are present in people who are unhappy with their weight, while low levels of external eating behavior are present. Female students scored statistically substantially higher than male students on the eating control behavior sub-dimension of the eating awareness measure ($p<0.05$). University education period is the transition period to a new life in the lives of students, and in this period, emotional behaviors in female students, together with external factors, affect nutritional behaviors. For this reason, healthy eating behaviors should be taught to university students and healthy body awareness should be created in female students.

Keywords: Nutrition, emotion, emotional eating behavior, body satisfaction

Giriş

Dünya Sağlık Örgütü sağlığı "insanın fiziksel, zihinsel ve sosyal yönden tam bir iyilik halinde olması" biçiminde tanımlamaktadır (WHO, 1948). Sağlıklı beslenme; yeterli ve dengeli miktarda besin öğelerini, her bir bireye özgü gereksinim miktarınca tüketmesidir. Bunun yanı sıra besinlerden haz alarak, yeterli miktarlarda alarak yaşamın devamını sağlamak amacıyla yapılan bilinçli bir eylemdir. Sadece tüketilmek istenen besinin vücuda alınması ya da sadece açlık duygusunu bastırmak amacıyla yapılmamaktadır (Baysal ve ark., 2013). Bireylerin besin tercihi; sosyo-demografik faktörler, şişmanlık, beden algısı, iştah, içinde bulunulan duygusal durum, dini ve kültürel inanışlar, geçmiş deneyimler, hormonlar, çevre ve genetik gibi birçok etmen tarafından belirlenmektedir (Deveci ve ark., 2017). Duyguların bireylerin besin tercihlerinde ve beslenme alışkanlıklarının oluşumunda çok güçlü bir etkisi olduğu bilinmektedir (Framson ve ark., 2009). Yeme davranışı üstünde pozitif (mutluluk, sevinç vb.) ve negatif (üzüntü, korku ve öfke vb.) duyguların etkileri vardır. Örneğin öfke duyusu ile bireyler, düzensiz ve hızlı bir şekilde ne yediğini umursamaksızın dürtüsel yeme davranışı gösterebilmekte ya da mutluluk duygusunda hedonik davranışı ile yemeklerden zevk almak için yemeye yönelebilmektedir. Ayrıca öfke gibi yoğun yaşanan olumsuz duygular varlığında yemenin duygusal durumu düzenlemek ve dikkati başka yöne çekmek için yapıldığı düşünülmektedir. Bu gibi duygular bireyi yeme davranışına yönlendirmekte ve bu davranışı etkileyen birçok etmen bulunmaktadır. Bunlar arasında şişmanlık, zayıflık, beden algısı, medya, dini ve kültürel inanışlar, geçmiş deneyimler, sosyodemografik özellikler, bireyin o an ki duygusal durumu, hormonlar, çevre, genetik ve iştah sayılabilmektedir (Karakuş ve ark., 2016).

Beslenme davranışı yaşam boyunca hızla gelişmektedir (Bilici ve Özkan, 2018). Bu gelişim sürecinde yeme davranışı ve duygu durumu arasındaki ilişki bilim insanları tarafından uzun zamandan beri araştırılmaktadır. Yaşamın devamlılığı için gerekli olan beslenme davranışı kişiye verdiği hazzın yanında yetersiz veya aşırı beslenme ya da yeme bozuklukları gibi bazı sağlık sorunlarına da neden olabilmektedir (Özgen ve ark., 2012). Ergenlerde genel olarak arkadaş, aile ve soysal medyanın "ince ya da zayıf olma" konusundaki tutumlarının özellikle kızlarda beden algısını olumsuz etkilediği bilinmektedir (French ve ark., 2012). Son zamanlarda duygulardan etkilenen yeme davranışı ön plana çıkmakta olup; duygusal yeme, kontrolsüz yeme ve bilişsel kısıtlama davranışı gibi davranışlar olumsuz duygulara verilen bir tepki olarak kullanılan baş etme stratejisi olarak ele alınmaktadır (Sevinçer ve Konuk, 2013). Farkındalık temelli müdahalelerin etkinliği ve yargısal olmayan deneyim kabulü artan bir deneysel destek kazanmıştır. Yapılan bir çalışmada; farkındalık, depresyon, anksiyete, ağrı gibi birçok kronik sağlık probleminin tedavisinde manyetik rezonans görüntülemede beyin aktivasyon yönelimlerinde gözle görülür değişikliklerle etkili sonuçlar göstermiştir (Lutz ve diğerleri, 2013).

Duygusal yeme, son zamanlarda gündeme gelen ve üzerinde yoğun araştırmalar yapılan bir yeme davranışıdır. Duygusal yeme davranışı ilk olarak Kaplan ve Kaplan (1957) ve Bruch (1964) tarafından, bireylerin kaygı, depresif ruh hali, öfke ve stres gibi olumsuz duygularla baş etme yöntemi olarak yeme eğilimi olarak tanımlanmıştır. Duygusal yeme, olumsuz deneyimler sonucunda yaşanan duygulara tepki olarak aşırı yeme eğilimidir (Pinaguy ve ark., 2003). Duygusal yeme davranışı, olumsuz veya olumlu duygusal durumlara eğilim gösterir. Macht (2008), duygusal uyarıcıların iştah üzerinde %30'luk bir etkiye sahip olduğunu ve iştahı azaltmadaki etkisinin ise %48'e ulaşabildiğini rapor etmiştir.

DRAFT

Bireylerin duygu durumlarının farkına varması ve bu duygu durumları ile başa çıkabilmesi için yeme farkındalıklarının arttırılması gerekmektedir (Köse, 2017). Yeme farkındalığı kavramı; Türkçe olarak ilk defa 'Ne yenildiğinden çok, nasıl ve neden yeme davranışının oluştuğunu fark ederek, fiziksel açlık-tokluk kavramını içselleştirip duygu ve düşüncelerin etkisinin farkında olarak, çevresel etmenlerden etkilenmeden, besin seçimlerini yargılamadan o anda tüketilecek olan besine odaklanarak yeme' olarak ifade edilmiştir (Köse ve ark., 2016). Yeme davranışları üzerine yoğunlaşarak, farkındalık kavramının yeme farkındalığına uyarlanması sonucu oluşmuştur (Framson ve ark., 2009; Hulbert-Williams ve ark., 2014). Bu çalışmanın amacı Burdur Mehmet Akif Ersoy Üniversitesi'nde öğrenim gören kadın ve erkek öğrencilerin şu an ki beden memnuniyetsizliği doğrultusunda yeme davranışlarının duygusal durum ile farklılaşıp farklılaşmadığını incelemektir.

Materyal ve Metot

Araştırmanın Örneklemi ve Yapıldığı Yer ve zaman

Çalışma kesitsel bir araştırmadır ve Burdur Mehmet Akif Ersoy Üniversitesi'nde eğitim gören öğrenciler üzerinde gerçekleştirilmiştir. Araştırma verileri, Mayıs 2023-Temmuz 2023 tarihleri arasında Google formlar aracılığıyla online olarak toplanmıştır. Burdur Mehmet Akif Ersoy Üniversitesi öğrencisi olan, 18 yaş ve üzeri olan, araştırmaya gönüllü olarak katılmak isteyenler ve bilinen psikiyatrik bozukluğu olmayan bireyler çalışmaya dahil edilmiştir. Burdur Mehmet Akif Ersoy Üniversitesi öğrencisi olmayan bireyler, Araştırmaya katılmak istemeyenler, bilinen psikiyatrik bozukluğu olanlar, veri toplama araçlarını eksik dolduran bireyler çalışmadan dışlanmıştır. Buna göre çalışmaya 141 kadın, 35 erkek olmak üzere toplam 176 kişi katılmıştır.

Anket Formu

Öğrencilerin sosyodemografik özellikleri, besin tüketim alışkanlıkları ve beden memnuniyetsizliğine ilişkin veriler ile Yeme Farkındalığı Ölçeği (YFÖ-30) ve Hollanda Yeme Davranışı Ölçeğini (DEBQ) içeren anket formu oluşturulmuştur. Anket formunda bireylerin vücut ağırlığı ve boy uzunluğu bilgileri katılımcının beyanı ile alınmıştır.

Yeme Farkındalığı Ölçeği

Yeme Farkındalığı Ölçeği (YFÖ), Framson ve ark. (2009) tarafından geliştirilen ve özgün adı 'Mindful Eating Questionnare' olan ölçek ile yeme davranışı, farkındalığı ve duygusal durum arası ilişkiler incelenmektedir. Ölçeğin Türkçe geçerlilik ve güvenilirlik çalışması Köse ve ark. (2016) tarafından yapılmış ve yeni ölçekte 5'li likert skalası (1:hiç, 2:nadiren, 3:bazen, 4:sık sık, 5:her zaman) olarak belirlenmiştir. Ölçeğin alt faktörleri; disinhibisyon, duygusal yeme, yeme kontrolü, odaklanma, yeme disiplini, farkındalık ve enterferans olmak üzere 7 başlıkta incelenmiştir. Ölçeğin her bir alt boyutu için alınan yüksek puan katılımcının ilgili alt boyutun değerlendirildiği özelliği taşıdığını gösterirken, ölçek toplam yeme farkındalığı puanını da ölçmektedir, fakat kesme noktası belirtilmemiştir.

Hollanda Yeme Davranışı Ölçeği

Hollanda Yeme Davranışı Ölçeği (The Dutch Eating Behaviour Questionnaire-DEBQ) Van Strien ve ark. (1986) tarafından geliştirilmiş ve Türkçe'ye adaptasyonu Bozan ve ark. (2011) tarafından yapılmıştır. Duygusal yeme davranışlarını, dışsal yeme davranışlarını ve kısıtlı yeme davranışlarını değerlendiren 3 alt ölçekten oluşmaktadır. Ankette yer alan maddeler 5'li Likert skalası ile değerlendirilmektedir (1: hiçbir zaman, 2: nadiren, 3: bazen, 4: sık, 5: çok sık). Ölçeğin toplam skoru değerlendirilmemekte, 3 alt ölçek kendi içinde değerlendirilmektedir. Ölçeğin skorlanmasında herhangi bir kesim noktası olmazken 3 alt ölçeğin kendi içinde değerlendirilen toplam puanının yüksek olması yeme davranışı ile ilgili olumsuzluğu göstermektedir. Hollanda Yeme Davranışı anketinin Türkçe versiyonunda ilk 10 soru kısıtlı yeme, 11-23 arası duygusal yeme, 24-33 arası ise dışsal yeme tutumunu değerlendiren sorulardır.

İstatistiksel Analiz

Araştırmanın verileri IBM SPSS 22 (Statistical Package for the Social Sciences) programı kullanılarak analiz edilmiştir. Tanımlayıcı değer olarak nicel veriler için aritmetik ortalama (x) ± standart sapma (SS), nitel veriler için yüzde (%) ve frekans (n) değerleri verilmiştir. Öğrencilerin tanımlayıcı özellikleri ile YFÖ puan ortalaması ve DEBQ puan ortalaması arasında normal dağılım gösteren değişkenlerde; bağımsız örneklem t testi ve One-Way ANOVA testleri, normal dağılmayan değişkenlerde ise Mann Whitney-U ve Kruskal Wallis testleri kullanılmıştır. $p<0,05$ istatistiksel anlamlılık düzeyi olarak kabul edilmiştir.

Araştırmanın Etik Yönü

Araştırmanın yürütülmesi için Burdur Mehmet Akif Ersoy Üniversitesi Girişimsel Olmayan Klinik Araştırmalar Etik Kurulu Başkanlığından GO 2023/208 karar numaralı 05/04/2023 tarihli "Etik Kurul Onayı" alınmıştır. Öğrencilere araştırma hakkında açıklama yapılmış ve araştırmaya katılmayı kabul eden gönüllü öğrenciler ile yürütülmüştür.

Bulgular

Çalışmaya katılan 176 kişiden elde edilen veriler analiz edilmiş, çalışmaya katılan öğrencilerin sosyodemografik özellikleri Tablo 1.'de gösterilmiştir. Buna göre kadınların yaş ortalaması 22,1±2,5 yıl, erkeklerin yaş ortalaması ise 25,1±7,3 yıldır. 1. sınıfta eğitim gören kadın katılımcıların oranı %21,3 iken, erkek katılımcıların oranı %17,1'dir. 4. sınıfta eğitim gören kadın katılımcıların oranı %35,5, erkek katılımcıların ise %28,6'dir. Geliri giderinden az olan kadın öğrencilerin oranı %27,7, erkek öğrencilerin oranı ise %25,7'dir. Geliri giderine eşit olan kadın öğrencilerin oranı %61,7, erkek öğrencilerin oranı ise %48,6 olduğu saptanmıştır. Katılımcıların şimdiki vücut ağırlıklarını nasıl değerlendirdikleri incelendiğinde; daha ince olmayı isteyen kadın katılımcıların oranı %51,8 iken, erkek katılımcıların oranı %22,9'dir. Buna göre kadın öğrencilerin şimdiki vücut ağırlıklarından daha az memnun olduğu söylenebilir. Kadın katılımcıların %17,7'sinin hekim tarafından tanısı konmuş bir hastalığı varken erkek katılımcıların %14,3'ünün hekim tarafından tanısı konmuş bir hastalığı vardır. Kadın katılımcılarda en çok %16,7 oranla demir eksikliği anemisi, psikiyatrik hastalıklar ve diğer hastalıklar görülürken, erkeklerde %8,6 oranla diğer hastalıklar görülmektedir. Kadın katılımcıları %28,4 'ü sigara kullanırken erkek katılımcıları %57,1 'i sigara kullanmaktadır. Alkol kullanan kadın katılımcıların oranı %28,4 iken erkek katılımcıların oranı %51,4'tür.

Genel beslenme alışkanlıkları

Öğrencilerin bazı beslenme alışkanlıkları incelenmiş ve elde edilen sonuçlar Tablo 2.'de gösterilmiştir. Buna göre kadın öğrencilerin %42,6'sının (60 kişi) erkek öğrencilerin ise %40'ının (14 kişi) öğün atladığı saptanmıştır. Öğrencilerin tamamında en çok atlanan öğünün %43,2 ile öğle öğünü olduğu görülmüştür. Neden öğün atlıyorsunuz sorusuna kadın öğrenciler en çok uykuda olduğum için (%19,9) seçeneğini işaretlerken, erkek öğrencilerde zamanım yok (%19,3) seçeneği tercih edilmiştir. Duygusal durumunuz yeme davranışınızı etkiler mi sorusuna kadın öğrencilerin %74,5'i, erkek öğrencilerin ise %54,3'ü evet demiştir. Çay ve kahve tüketim alışkanlıkları incelendiğinde erkek öğrencilerin kadın öğrencilere kıyasla daha fazla şeker attığı (sırasıyla 10,0±7,8 ve 11,0±7,4 g), fakat farkın önemli olmadığı saptanmıştır.

DRAFT

Tablo 1. Katılımcıların sosyodemografik özellikleri

	Kadın		**Erkek**		**Toplam**	
	x±SS		**x±SS**		**x±SS**	
Yaş	22,1±2,5		25,1±7,3		22,69±4,1	
BKİ (kg/m^2)	22,0±3,4		24,2±3,1		22,4±3,4	
	n	**%**	**n**	**%**	**n**	**%**
Sınıf						
1	30	21,3	6	17,1	36	20,5
2	29	20,6	10	28,6	39	22,2
3	31	22,0	7	20,0	38	21,6
4	50	35,5	10	28,6	60	34,1
5 ve 6	1	0,7	2	5,7	3	1,6
Gelir seviyesi						
Gelirim giderimden az	39	27,7	9	25,7	48	27,3
Gelirim giderime eşit	87	61,7	17	48,6	104	59,1
Gelirim giderimden fazla	15	10,6	9	25,7	24	13,6
Şimdiki vücut ağırlığınızı nasıl değerlendirirsiniz?						
Daha ince olmayı isterdim	73	51,8	8	22,9	81	46,0
Daha kilolu olmayı isterdim	13	9,2	9	25,7	22	12,5
Kilomdan memnunum	55	39,0	18	51,4	73	71,5
Sigara kullanıyor musun?						
Evet	40	28,4	20	57,1	60	34,1
Hayır	94	66,7	12	34,3	106	60,2
İçiyordum bıraktım	7	5,0	3	8,6	10	5,7
Alkol kullanıyor musunuz?						
Evet	40	28,4	18	51,4	58	33,0
Hayır	99	70,2	16	45,7	115	65,3
İçiyordum bıraktım	2	1,4	1	2,9	3	1,7
Düzenli spor yapıyor musunuz?						
Evet	23	16,3	13	37,1	36	20,5
Hayır	118	83,7	22	62,9	140	79,5
Ne sıklıkla spor yapıyorsunuz?						
Her gün	3	2,1	3	8,6	6	3,4
Haftada 1-2 gün	71	50,4	18	51,4	89	50,6
Haftada 3-4 gün	12	8,5	6	17,1	18	10,2
Haftada 5 gün ve daha fazla	11	7,8	4	11,4	15	8,5
Hekim tarafından tanısı konulmuş bir hastalığınız var mı?						
Evet	25	17,7	5	14,3	30	17,0
Hayır	116	82,3	30	85,7	146	83,0

DRAFT

DRAFT

Tablo 2. Katılımcıların genel beslenme alışkanlıkları

	Kadın		**Erkek**		**Toplam**	
	n	**%**	**n**	**%**	**n**	**%**
Öğün atlar mısınız?						
Evet	60	42,6	14	40,0	74	42,0
Hayır	18	12,8	8	22,9	26	14,8
Bazen	63	44,7	13	37,1	76	43,2
Cevabınız evet veya bazen ise en çok hangi öğünleri atlarsınız?						
Sabah	55	39,0	14	40,0	69	39,2
Öğle	65	46,1	11	31,4	76	43,2
Akşam	3	2,1	2	5,7	5	2,8
Öğün atlama nedenlerini belirtir misiniz?						
Zamanım yok	25	17,7	9	25,7	34	19,3
Uykuda olduğum için	28	19,9	4	11,4	32	18,2
İştahım olmadığı için	13	9,2	1	2,9	14	8,0
Diyet	1	0,7	1	2,9	2	1,1
Acıkmıyorum	25	17,7	3	8,6	28	15,9
Yoğunluk	16	11,3	6	17,1	22	12,5
Diğer	15	10,6	3	8,6	18	10,2
Duygusal durumunuz yemenizi etkiler mi?						
Evet	105	74,5	19	54,3	124	70,5
Hayır	6	4,3	10	28,6	16	9,1
Bazen	30	21,3	6	17,1	36	20,5
Çay tüketir misiniz?						
Evet	134	85,0	34	87,1	168	85,5
Hayır	7	5,0	1	2,9	8	4,5
Kahve tüketir misiniz?						
Evet	137	97,2	31	88,6	168	85,5
Hayır	4	2,8	4	11,4	8	4,5
Bir bardak çay için (varsa) kullandığınız şeker miktarı (g)						
	8,9±5,2		10,0±7,8		9,1±5,7	
Bir fincan/bardak kahve için kullandığımız şeker miktarı (g)						
	7,6±3,7		11,0±7,4		8,6±5,2	
Günde kaç mL su tüketiyorsunuz?						
	344,7±143,7		371,4±169,0		350,0±148,9	

Öğrencilere ait Hollanda Yeme Davranışı Ölçeği (DEBQ) ve Yeme Farkındalığı Ölçeği (YFÖ-30) puanlarının değerlendirilmesi

Öğrencilerin yeme davranışlarını gösteren DEBQ ve YFÖ-30 ölçek skorları incelemiş, buna göre elde edilen sonuçlar Tablo 3.'te gösterilmiştir. DEBQ ölçeğine ait kısıtlayıcı yeme, duygusal yeme ve dışsal yeme skorlarının hepsinin kadın öğrencilerde daha yüksek olduğu saptanmıştır. Duygusal yeme puanlarındaki fark istatistiksel olarak anlamlıdır ($p<0,05$). YFÖ-30'un alt grupları incelendiğinde ise sadece duygusal yeme ve yeme kontrolü davranışı skorlarında cinsiyete göre anlamlı farklılık saptanmıştır ($p<0,05$). Erkeklerde duygusal yeme skorları 18,5±4,5 iken, kadınlarda bu skor 16,0±4,7'dir. Yeme kontrolü puanları incelendiğinde ise kadın ve erkek öğrencilerde sırasıyla 15,5±3,6 ve 13,5±3,1'dir.

Tablo 3. Cinsiyete göre DEBQ ve YFÖ-30 ölçek puanları

	Kadın	Erkek	Toplam	p[a]
	x±SS	x±SS	x±SS	
DEBQ Puanı				
Kısıtlayıcı yeme	24,2±8,0	22,1±1,9	23,8±8,0	0,180
Duygusal yeme	30,9±13,8	25,4±10,6	29,8±13,4	0,012
Dışsal yeme	31,0±7,1	30,5±7,0	30,9±7,0	0,708
Yeme Farkındalığı				
Disinhibisyon	17,0±4,5	17,1±3,4	17,0±4,3	0,964
Duygusal yeme	16,0±4,7	18,5±4,5	16,5±4,8	0,009
Yeme kontrolü	15,5±3,6	13,5±3,1	15,1±3,6	0,001
Odaklanma	16,1±2,6	15,6±2,9	16,0±2,7	0,131
Yeme disiplini	11,6±3,1	11,5±3,2	11,6±3,2	0,937
Farkındalık	16,0±2,1	15,4±2,3	15,9±2,3	0,325
Enterferans	7,3±1,8	7,3±2,0	7,3±1,8	0,707
Toplam	99,4±13,7	99,1±13,0	-	0,899

a: Mann Whitney U testi kullanılmıştır. *: p <0,001

Öğrencilerin şimdiki vücut ağırlığından memnun olma durumuna göre ölçek puanları Tablo 4.'te gösterilmiştir. Daha ince olmayı isteyen öğrencilerin kısıtlayıcı yeme (28,2±7,1) ve duygusal yeme skorları (34,9±14,0) istatistiksel olarak anlamlı düzeyde yüksektir (p<0,001). DEBQ ölçeği alt boyutlarından dışsal yeme skorlarında ise istatistiksel olarak anlamlı farklılık saptanmamıştır (p>0,05). Yeme farkındalığı alt boyutlarından ise sadece duygusal yeme skorlarında istatistiksel olarak anlamlı farklılık saptanmıştır (p<0,05). Bu farklılığın daha ince olmayı isterdim (15,3±4,8) seçeneğini işaretleyen öğrencilerin skorlarından kaynaklandığı görülmektedir. Öğrencilerin şimdiki vücut ağırlığından memnun olma durumları DEBQ ölçeği toplam puanında istatistiksel olarak anlamlı farklılık gösterirken (p<0,05), YFÖ-30 toplam puanları arasında istatistiksel olarak anlamlı farklılık bulunmamaktadır (p>0,05).

Tablo 4. Şimdiki vücut ağırlığından memnun olma durumuna göre DEBQ ve YFÖ-30 ölçek puanları

	Daha ince olmayı isterdim	Daha kilolu olmayı isterdim	Kilomdan Memnunum	p[a]
	x±SS	x±SS	x±SS	
DEBQ				
Kısıtlayıcı yeme	28,2±7,1	17,1±4,4	20,7±7,1	0,000*
Duygusal yeme	34,9±14,0	23,8±10,4	25,8±11,5	0,000*
Dışsal yeme	30,7±6,8	32,2±7,7	30,7±7,2	0,642
Yeme farkındalığı				
Disinhibisyon	16,5±4,6	17,2±4,3	17,5±3,9	0,294
Duygusal yeme	15,3±4,8	17,5±4,7	17,7±4,4	0,010
Yeme kontrolü	14,9±3,8	14,7±3,6	15,4±3,4	0,556
Odaklanma	15,8±2,2	15,3±1,8	16,1±2,3	0,401
Yeme disiplini	11,5±3,3	12,0±3,2	11,5±3,0	0,743
Farkındalık	16,0±2,9	15,4±2,0	16,2±2,6	0,564
Enterferans	7,4±1,7	6,9±1,9	7,4±1,9	0,552
Toplam	97,3±14,7	98,9±12,3	101,8±12,2	0,118

a: Kruskall Walls testi uygulanmıştır. *: p <0,001

Tartışma

Bu çalışmaya katılan öğrencilerin büyük bir çoğunluğu kadındır. Büyük ölçüde sağlık alanına ilişkin bölümlerde eğitim almakta olan öğrencilerle gerçekleştirilmiş olması bunun temel nedenidir. Benzer şekilde yürütülen bir çalışmada da kadın katılımcı sayısının daha yüksek olduğu görülmüştür (Köse ve ark., 2016). Öğrencilerin yaş ortalaması 22,7± 4,1 yıldır. Öğrencilerin BKİ değerleri incelendiğinde ise büyük

çoğunluğunun normal aralıkta olduğu saptanmıştır. Çalışmaya dahil olan öğrenci grubunun özellikleri nedeniyle bu durumun beklenen bir sonuç olduğu söylenebilir. Yapılan diğer bir çalışmada da öğrencilerin BKİ değerleri benzer şekilde dağılım göstermiştir (Köse, 2017).

Çalışmamıza katılan öğrencilerin büyük çoğunluğu öğün atladığını (%85,2) bildirmiştir. Yapılan bir diğer çalışmada ise benzer şekilde katılımcıların %82,8'inin öğün atladığı tespit edilmiştir. %42' si kesin olarak öğün atladığını belirtirken, %43,2'si ara sıra öğün atladığını belirtmiştir. Cinsiyetler arası öğün atlama incelendiğinde kadınların erkeklere göre daha çok öğün atladığı istatistiksel olarak anlamlı bulunmuştur (İbrahimova, 2020). Şimdiki vücut ağırlığından memnun olma durumunda da kadın öğrencilerin daha ince olmayı istediği görülmektedir. Kadın öğrencilerin daha zayıf olma istekleri nedeniyle daha fazla öğün atladıkları söylenebilir.

Hollanda yeme davranışı ölçeği incelendiğinde; kısıtlayıcı yeme, duygusal yeme ve dışsal yeme davranışı düzeylerine göre daha ince olmayı isteyen bireylerin sayısı kadınlarda daha fazladır. Anschutz, ve ark. (2008) tarafından üniversite öğrencileri üzerinde yapılan bir çalışmada, 163 kız öğrencide görülen beden memnuniyetsizliği ile kısıtlayıcı yeme ve duygusal yeme davranışları arasında pozitif bir ilişki olduğu, beden memnuniyetsizliği ile dışsal yeme davranışı arasında ise bir ilişki olmadığı saptanmıştır. Snoek ve ark.'nın (2007) adölesanlar üzerinde yaptığı bir çalışmada kız öğrencilerde erkek öğrencilere göre kısıtlayıcı yeme ve duygusal yeme skorlarının daha yüksek olduğu görülmüştür. Erkeklerde ise kızlara göre daha yüksek dışsal yeme skorları olduğu saptanmıştır.

Çalışmamızda YFÖ alt boyutlarından duygusal yeme ve yeme kontrolü ile cinsiyet arasında anlamlı fark saptanmıştır ($p<0,05$). Duygusal yemenin alt boyut kapsamları duygusal açlık, iyi hissetme ve tatmin için yeme; yeme kontrolünün alt kapsamları yeme hızını ayarlama, yeme işlevinin kontrolünü elinde tutmaktan oluşmaktadır. Erkek öğrencilerin puan ortalaması, kadın öğrencilerin puan ortalamasından daha yüksektir. Erkek öğrencilerin bu konulardaki farkındalık bilgisinin kadın öğrencilerden daha fazla olduğu söylenebilir. Fakat çalışmaya katılan erkek öğrenci sayısının az olması da bu duruma neden olmuş olabilir. Erkek öğrenci sayısının az olması çalışmanın sınırlı yönü olarak düşünülmektedir. Spor salonunda spor yapan katılımcılar ile gerçekleştirilen diğer bir çalışmada ise YFÖ-30 alt boyutlarından duygusal yeme, yeme kontrolü ve bilinçli beslenme alt boyut puanlarının cinsiyete göre istatistiksel olarak anlamlı fark gösterdiği belirlenmiştir. Spor yapan katılımcıların duygusal yeme, yeme kontrolü ve bilinçli beslenme puanları, kadınlarda erkeklere göre daha yüksektir. Spor yapan katılımcıların YFÖ-30 alt boyutlarından disinhibisyon, farkındalık, yeme disiplini, bilinçli beslenme, enterferans alt boyutları ve YFÖ-30 ölçek puanlarının cinsiyete göre anlamlı farklılık göstermediği ($p>0,05$) saptanmıştır (Üstündağ, 2020).

Sonuç

Sonuç olarak öğrenciler üzerinde gerçekleştirilen bu çalışmada özellikle kadın öğrencilerin BKİ değerleri normal aralıkta olmasına rağmen, daha ince görünmeyi istedikleri, daha fazla öğün atladıkları ve yeme davranışlarının da buna göre etkilendiği görülmektedir. Beslenme üzerine çevrenin etkileri de olduğu bilinmekte ve bu çalışmadan elde edilen sonuçların öğrencilerin beslenme alışkanlıklarının değerlendirilmesinde çevresel faktörlerden kolaylıkla etkilenebildiğini göstermektedir. Öğrencilere sağlıklı vücut ağırlığı kavramının önemini içeren eğitimler, afişler, kongre ve sempozyumların düzenlenmesi bu konuda bilinçlendirme sağlaması açısından önemlidir.

Kaynaklar

Anschutz DJ. van Strien T, Engels RC ME 2008. Exposure to slim images in mass media: Television commercials as reminders of restriction in restrained eaters. Health Psychology, 27(4): 401–408.

Baysal A, Aksoy M, Bozkurt N, Merdol TK, Pekcan G, Keçecioğlu S 2013. Diyet el kitabı. Ankara: Hatipoğlu Baskı.

Bilici S, Özkan N 2018. Yeme davranışında yeni yaklaşımlar: Sezgisel yeme farkındalığı. Gazi Üniversitesi Sağlık Bilimleri Dergisi. 3(2): 16-24.

Bozan N, Baş M, Aşçı FH 2011. Psychometric properties of Turkish version of Dutch Eating Behaviour Questionnaire (DEBQ). A preliminary results. Appetite, 56(3): 564-566.

Bruch, H. 1964. Psychological aspects of overeating and obesity, Psychosomatics, 5(5): 269-274.

Deveci B, Deveci B. Avcıkurt C 2017. Yeme davranışı: Gastronomi ve mutfak sanatları öğrencileri üzerine bir araştırma. Journal of Tourism and Gastronomy Studies, 118: 134

Framson C, Kristal AR, Schenk JM, Littman AJ, Zeliadt S, Benitez D 2009. Development and Validation of the Mindful Eating Questionnaire. Journal of the American Dietetic Association. 1439-1444.

French SA, Epstein LH, Jeffery RW, Blundell JE, Wardle J 2012. Eating behavior dimensions. Associations with energy intake and body weight. A review. Appetite. 59(2): 541-549.

Hulbert-Williams L, Nicholls W, Joy J, Nick H-W 2014. Initial Validation of the Mindful Eating Scale. Mindfulness. 719-729.

İbrahimova A. 2020. Üniversite öğrencilerinin yeme farkındalığına göre aşırı besin isteği, iştah ve beslenme durumlarının değerlendirilmesi. Yüksek lisans tezi, Başkent Üniversitesi Sağlık Bilimleri Enstitüsü, Ankara.

Kaplan HI, Kaplan HS 1957. The psychosomatic concept of obesity. Journal of Nervous and Mental Disease, 125: 181-201.

Karakuş SŞ, Yıldırım H, Büyüköztürk Ş 2016. Üç faktörlü yeme ölçeğinin Türk kültürüne uyarlanması: geçerlik ve güvenirlik çalışması. TAF PreventiveMedicine Bulletin. 15(3): 229-237.

Köse G 2017. Üniversite öğrencilerinin yeme farkındalığının üzerine bir araştırma. Doktora tezi. Başkent Üniversitesi, Sağlık Bilimleri Enstitüsü, Ankara.

Köse G, Tayfur M, Birincioğlu İ, Dönmez A 2016. Adaptation study of the Mindful Eating Questionnare (MEQ) into Turkish. Bilişsel Davranışçı Psikoterapi ve Araştırmalar. 125-134.

Lutz J, Herwig U, Opialla S, Hittmeyer A, Jäncke L, Rufer M, Brühl AB 2013. Mindfulness and emotion regulation-- an fMRI study. Social Cognitive and Affective Neuroscience. 776-785.

Macht M 2008. How emotions affect eating: A Five-Way model. Appetite, 50: 1–11.

Özgen L, Kinaci B, Arli M 2012. Ergenlerin Yeme Tutum ve Davranışları. Journal of Faculty of Educational Sciences, 45(1)

Pinaquy S, Chabrol H, Simon C, Louvet JP, Barbe P 2003. Emotional eating, alexithymia, and binge-eating disorder in obese women. Obesity research, 11(2): 195-201.

Sevinçer GM, Konuk N 2013. Emotional eating. Psychiatry and Behavioral Sciences, 3(4): 171.

Snoek HM, Engels RC, Janssens JM, van Strien T 2007. Parental behaviour and adolescents' emotional eating. Appetite, 49(1): 223-230.

Üstündağ EG 2020. Spor Salonunda Spor Yapan Bireylerde Yeme Farkındalığı ile Ortoreksiya Nervoza Belirtileri Arasındaki İlişkinin İncelenmesi. Yüksek Lisans Tezi, Çağ Üniversitesi Sosyal Bilimler Enstitüsü, Mersin.

Van Strien T, Frijters JE, Bergers GP, Defares PB 1986. The Dutch Eating Behavior Questionnaire (DEBQ) for assessment of restrained, emotional, and external eating behavior. International Journal of Eating Disorders, 5(2): 295- 315.

WHO. 1948. Preamble to the Constitution of the World Health Organization as adopted by the International Health Conference, New York, 19 June - 22 July 1946; signed on 22 July 1946 by the representatives of

61 States (Official Records of the World Health Organization, no. 2, p. 100) and entered into force on 7 April 1948.

DRAFT

ORAL PRESENTATION

Cloning of GM-CSF gene into the pVAX1 DNA vaccine vector

Ezgi Dağ Taşkesenligil[1*] *(ORCID: 0000-0002-5184-8031)*, Yağmur Ünver[2](*ORCID: 0000-0003-1497-081X*)

[*1]Atatürk University, Institute of Science, Department of Molecular Biology and Genetics, 25240, Erzurum, Turkey
[2]Atatürk University, Faculty of Science, Department of Molecular Biology and Genetics, 25240, Erzurum, Turkey
*E-mail: aezgidag@gmail.com

Abstract

Adjuvant genes used in DNA vaccines designed designed to prevent and treat cancer stimulate elements of both the innate and adaptive immune systems. Normally, GM-CSF is produced in response to cytokines, which primarily mediate immune activation and inflammation and respond to various cell types including macrophages and T cells. Because of its critical role in the development and maturation of antigen presenting cells (APCs), GM-CSF indirectly affects T cells. In addition, GM-CSF has been shown to induce the proliferation, differentiation, and activation of both APCs and T cells.These properties make GM-CSF a potent adjuvant that is often used in vaccine trials. In this study, the GM-CSF gene was cloned into the pVAX1 vector, a common DNA vaccine vector. The GM-CSF gene was synthetically obtained and amplified by PCR. After the reaction, the resulting products were analysed by agarose gel electrophoresis, and the bands were extracted from the gel. PCR products cut with *Xba*I and *Xho*I enzymes were extracted with a phenol-chloroform-isoamyl alcohol mixture. Similarly, pVAX1 was cut with the same enzymes and extracted from the gel. The resulting fragments were exposed to an appropriate ligation reaction mixture at 16°C for 16 hours. The ligation products were introduced into competent *E. coli* JM109 cells by heat shock. Transformants were plated on LB-agar medium containing 50 µg/mL kanamycin and incubated. Randomly selected colonies observed on the petri dishes were used for colony PCR. Bands specific for the GM-CSF gene were observed from the colony PCR results. Plasmid isolation was performed from one of these colonies, followed by Sanger sequencing analysis.This confirmed that the gene was correctly positioned on the pVAX1 vector. The resulting recombinant expression plasmid was named pVAX1-GM-CSF. This study was supported by Atatürk University Scientific Research Projects (BAP, FDK-2022-10925). E.D.T. is also thankful for The Council of Higher Education(CoHE,100/2000)PhD Scholarship Program,Turkey.

Keywords: GM-CSF, adjuvant, DNA vaccine, cloning, recombinant DNA

INTRODUCTION

Despite coming more than a century after infectious disease vaccines, cancer vaccines are even more promising. While infectious disease vaccines focus primarily on disease prevention, cancer vaccines aim to treat as well as prevent cancer. Most current cancer vaccine efforts target tumour-associated antigens, whether known or unknown. In addition to these antigens, adjuvants are often used in cancer vaccine research. Often used in combination with tumour-associated antigens, adjuvants enhance the specific immune response generated against the antigen, thereby increasing the efficacy of the vaccine (Madan et al., 2012; Schlom, 2012; Thomas & Prendergast, 2016).

A DNA vaccine is a recombinant DNA produced by combining a DNA sequence to be used as a vaccine with a plasmid DNA capable of expressing that sequence using recombinant DNA technology. Host cells are transfected with the plasmid DNA containing the relevant DNA sequence and carrying fragments or genes encoding immunogenic antigens. This approach effectively induces an immune response in both humoral and cellular immunity, generating an effective immune response in both aspects (Silveira et al., 2017; Yang Lee et al., 2018). Granulocyte-macrophage colony-stimulating factor (GM-CSF) is encoded by a 2.5 kb mRNA with 4 exons. It is secreted as a small glycosylated monomeric protein with a molecular weight of 23 kDa. Human

GM-CSF is derived from a precursor containing a signal peptide. GM-CSF is produced in response to various cytokines that mediate immune activation and inflammation, primarily by various cell types including macrophages, mast cells, T cells, fibroblasts and endothelial cells. Circulating GM-CSF levels are typically low or undetectable under normal conditions, but can rise to high levels in response to immune stimuli. GM-CSF is produced by activated T cells following engagement of the T cell receptor with the antigen, together with appropriate co-stimulatory signals. Many of the effects of GM-CSF on T cells are thought to be mediated indirectly through antigen presenting cells (APCs) (Farrar et al, 1988). In addition, GM-CSF has been shown to be critical for the development and maturation of antigen-presenting cells, particularly dendritic cells (DCs), within APCs. One of the most studied in vivo effects of GM-CSF in the immune system is the critical interaction between T cells and APCs in anti-tumour immunity. The outcome of this interaction determines the fate of cancer cells based on signals received by dendritic cells, referred to as either "danger" or "tolerogenic" signals (Wada et al., 1997). On the other hand, GM-CSF has been shown to induce the proliferation, differentiation and activation of macrophages, neutrophils, various antigen-presenting cells and T cells. These properties make GM-CSF a potent adjuvant and a commonly used component in vaccine trials (Mach & Dranoff, 2000). With this in mind, the GM-CSF gene was incorporated into the DNA vaccine in this study to allow expression of the GM-CSF gene as an adjuvant.

MATERIALS AND METHODS

E. coli JM109 strain, DNA ladder and restriction enzymes were purchased from Invitrogen Company (USA). The pVAX1 expression vector was purchased from ThermoFisher Scientific. The GM-CSF gene was synthesised by Invitrogen. Chemicals used in the study, including bacterial growth media, plasmid isolation kit, enzymes, DNA markers, and all other chemicals, were purchased from Invitrogen, Sigma, ThermoFisher Scientific, Intron Biotechnology, Fluka, and Merck. Initially, the following primers were used for GM-CSF gene amplification *Xho*I-GMCSF-F; 5'-ATTTCTCGAGATGTGGCTGCAGA-3' and *Xba*I-GMCSF-R; 5'-ATTCTAGATCACTCCTGGACTGG-3'. The gene was amplified using Phusion High-Fidelity DNA Polymerase (Thermo Scientific, USA) by PCR with gene-specific primers. PCR amplification included denaturation at 98 °C for 30 seconds, followed by 35 amplification cycles at 98 °C for 10 seconds, 55 °C for 30 seconds and 72 °C for 35 seconds. The final extension step was performed at 72 °C for 5 minutes. The PCR product was then digested with XhoI and XbaI enzymes. Similarly, the pVAX1 plasmid was digested with the same enzymes. The two digested products (gene and plasmid) were ligated using T4 DNA ligase to produce the recombinant plasmid. The putative recombinant plasmid was introduced into competent E. coli JM109 cells by heat shock transformation. LB agar containing 0.5% yeast extract, 1% tryptone, 0.5% NaCl, 2% agar and 50 µg/mL kanamycin was prepared to select for kanamycin-resistant transformants. To confirm positive transformants, colony PCR was performed using gene and vector specific primers. Based on the results of agarose gel electrophoresis, putative recombinant plasmids were purified from positive transformants and sequenced using the Sanger sequencing method.

RESULTS and DISCUSSION

Amplification of GM-CSF gene and obtaining recombinant vector

The GM-CSF gene was amplified by PCR using gene-specific primers (**Figure 1A**). The PCR product was then digested with specific enzymes (*Xho*I and *Xba*I). In parallel, the pVAX1 plasmid was digested with the same enzymes and analysed by agarose gel electrophoresis. The two digested products, the gene and the plasmid, were then ligated together using T4 DNA ligase to produce the recombinant plasmid. The putative recombinant vector was introduced into competent *E. coli* JM109 cells by heat shock transformation. The transformed cells were cultured on LB agar supplemented with kanamycin. After incubation, the colonies were used for colony PCR to validate the presence of positive transformants (**Figure 2**). For this purpose, colony PCR was performed using gene/vector specific primers (**Figure 1B** and **C**). The result showed successful gene amplification in almost all selected clones. Plasmid sequencing was then performed. The results of both colony PCR and nucleotide blast analysis confirmed successful insertion of the GM-CSF gene into the appropriate site of pVAX1 (**Figure 3**). The recombinant plasmid was named pVAX1-GM-CSF. The recombinant plasmid was named pVAX1-GMCSF and its map was visualized by SnapGene (SnapGene® software, www.snapgene.com) **(Figure 4**).

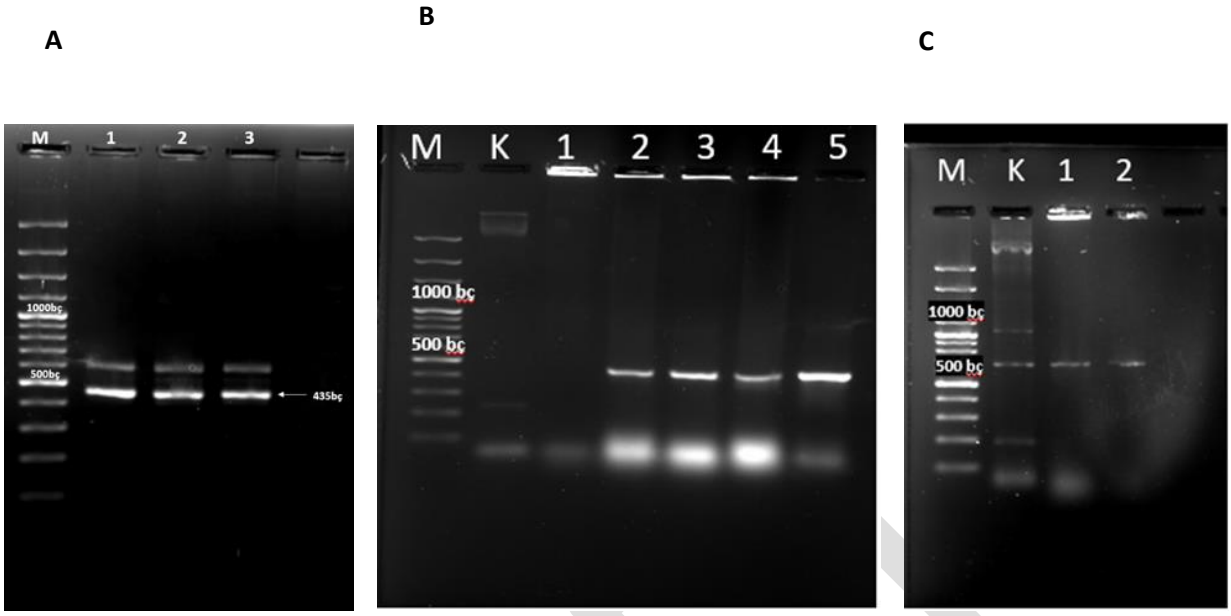


Figure 1: Agarose gel electrophoresis images, **A:** GM-CSF gene; **B:** Colony PCR results with gene spesific primers of selected colonies (435 bp); **C:** Colony PCR results with vector spesific primers of selected colonies (657bp)

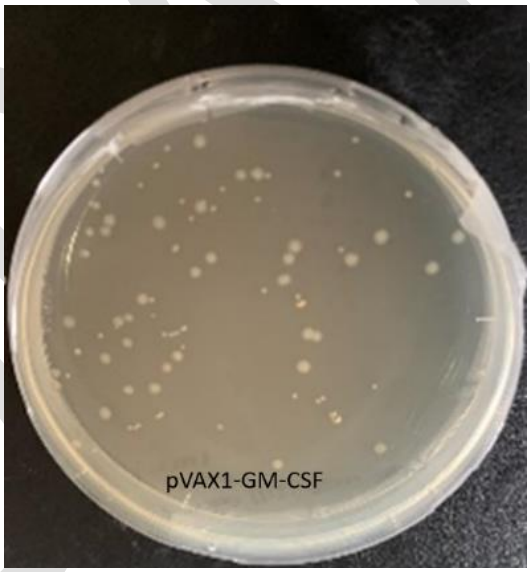


Figure 2: Bacterial colonies developed after transformation

Score	Expect	Identities	Gaps	Strand	Frame
804 bits(435)	0.0()	435/435(100%)	0/435(0%)	Plus/Plus	

```
Query  1    ATGTGGCTGCAGAGCCTGCTGCTCTTGGGCACTGTGGCCTGCAGCATCTCTGCACCCGCC  60
Sbjct  194  ATGTGGCTGCAGAGCCTGCTGCTCTTGGGCACTGTGGCCTGCAGCATCTCTGCACCCGCC  253
Query  61   CGCTCGCCCAGCCCCAGCACGCAGCCCTGGGAGCATGTGAATGCCATCCAGGAGGCCCGG  120
Sbjct  254  CGCTCGCCCAGCCCCAGCACGCAGCCCTGGGAGCATGTGAATGCCATCCAGGAGGCCCGG  313
Query  121  CGTCTCCTGAACCTGAGTAGAGACACTGCTGCTGAGATGAATGAAACAGTAGAAGTCATC  180
Sbjct  314  CGTCTCCTGAACCTGAGTAGAGACACTGCTGCTGAGATGAATGAAACAGTAGAAGTCATC  373
Query  181  TCAGAAATGTTTGACCTCCAGGAGCCGACCTGCCTACAGACCCGCCTGGAGCTGTACAAG  240
Sbjct  374  TCAGAAATGTTTGACCTCCAGGAGCCGACCTGCCTACAGACCCGCCTGGAGCTGTACAAG  433
Query  241  CAGGGCCTGCGGGGCAGCCTCACCAAGCTCAAGGGCCCCTTGACCATGATGGCCAGCCAC  300
Sbjct  434  CAGGGCCTGCGGGGCAGCCTCACCAAGCTCAAGGGCCCCTTGACCATGATGGCCAGCCAC  493
Query  301  TACAAGCAGCACTGCCCTCCAACCCCGGAAACTTCCTGTGCAACCCAGATTATCACCTTT  360
Sbjct  494  TACAAGCAGCACTGCCCTCCAACCCCGGAAACTTCCTGTGCAACCCAGATTATCACCTTT  553
Query  361  GAAAGTTTCAAAGAGAACCTGAAGGACTTTCTGCTTGTCATCCCCTTTGACTGCTGGGAG  420
Sbjct  554  GAAAGTTTCAAAGAGAACCTGAAGGACTTTCTGCTTGTCATCCCCTTTGACTGCTGGGAG  613
Query  421  CCAGTCCAGGAGTGA  435
Sbjct  614  CCAGTCCAGGAGTGA  628
```

Figure 3: Nucleotide BLAST analysis result of pVAX1-GM-CSF (Query: referenced sequence, Subject: Sequence obtained as a result of Sanger analysis)

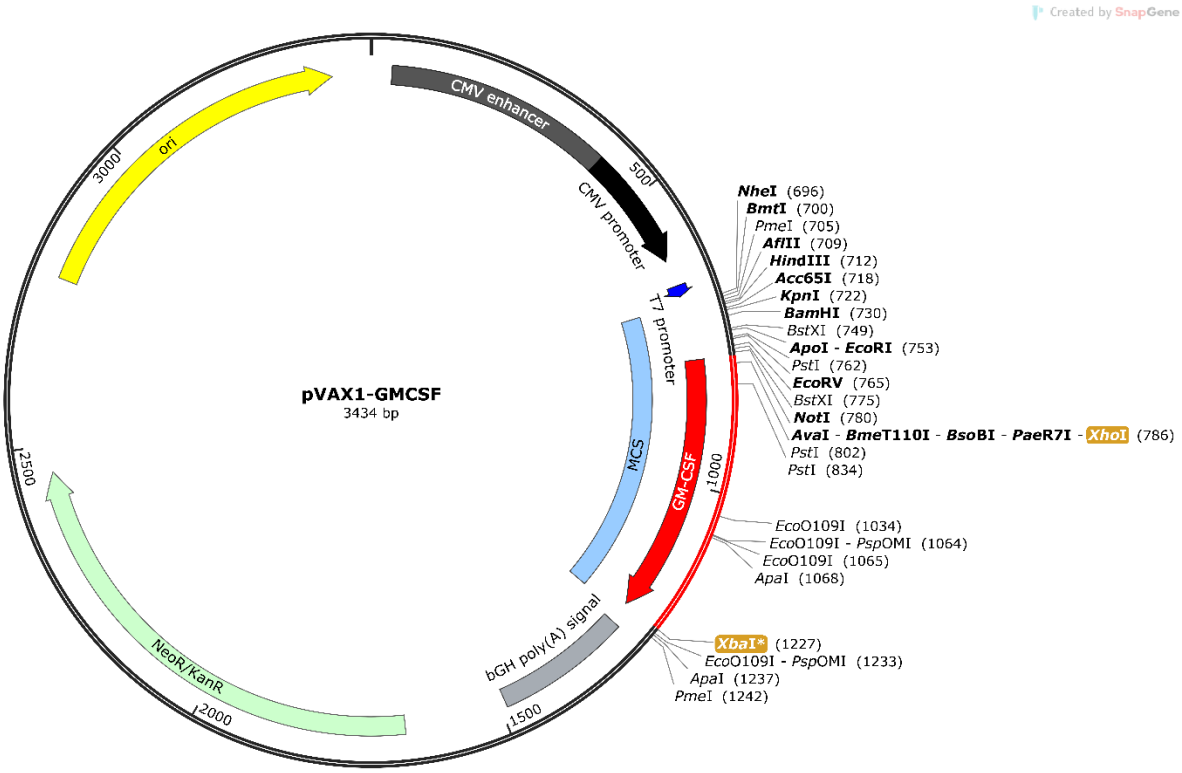


Figure 4: Construction map of pVAX1-GMCSF

CONCLUSION

In this study, we aimed to clone GM-CSF gene into pVAX1 to obtain recombinant expression vector. Colony PCR and Sanger sequencing analysis showed that the gene was inserted into the plasmid at the correct position after transformation of bacteria with the resulting putative recombinant plasmid. The DNA vaccine construct

that was successfully obtained in this study can be used as a preventive and therapeutic agent against all types of cancer.

ACKNOWLEDGEMENTS

This study was supported by Atatürk University Scientific Research Projects (BAP, FDA-2022-10925). E.D.T. is also thankful for The Council of Higher Education (CoHE,100/2000) PhD Scholarship Program,Turkey.

REFERENCES

Farrar, W. L., Harel-Bellan, A., & Ferris, D. K. (1988). Biochemical and molecular events controlled by lymphokine growth factors. In *Society of General Physiologists series* (Vol. 43, pp. 371–380).

Mach, N., & Dranoff, G. (2000). Cytokine-secreting tumor cell vaccines. In *Current Opinion in Immunology* (Vol. 12, Issue 5, pp. 571–575). https://doi.org/10.1016/S0952-7915(00)00144-8

Madan, R. A., Mohebtash, M., Arlen, P. M., Vergati, M., Rauckhorst, M., Steinberg, S. M., Tsang, K. Y., Poole, D. J., Parnes, H. L., Wright, J. J., Dahut, W. L., Schlom, J., & Gulley, J. L. (2012). Ipilimumab and a poxviral vaccine targeting prostate-specific antigen in metastatic castration-resistant prostate cancer: A phase 1 dose-escalation trial. *The Lancet Oncology*, *13*(5). https://doi.org/10.1016/S1470-2045(12)70006-2

Schlom, J. (2012). Therapeutic cancer vaccines: Current status and moving forward. *Journal of the National Cancer Institute*, *104*(8). https://doi.org/10.1093/jnci/djs033

Silveira, M. M., Oliveira, T. L., Schuch, R. A., McBride, A. J. A., Dellagostin, O. A., & Hartwig, D. D. (2017). DNA vaccines against leptospirosis: A literature review. In *Vaccine* (Vol. 35, Issue 42). https://doi.org/10.1016/j.vaccine.2017.08.067

Thomas, S., & Prendergast, G. C. (2016). *Chapter 43 Cancer Vaccines : A Brief Overview*. *1403*, 755–761. https://doi.org/10.1007/978-1-4939-3387-7

Wada, H., Noguchi, Y., Marino, M. W., Dunn, A. R., & Old, L. J. (1997). T cell functions in granulocyte/macrophage colony-stimulating factor deficient mice. *Proceedings of the National Academy of Sciences of the United States of America*, *94*(23), 12557–12561. https://doi.org/10.1073/pnas.94.23.12557

Yang Lee, L. Y., Izzard, L., & Hurt, A. C. (2018). A review of DNA vaccines against influenza. In *Frontiers in Immunology* (Vol. 9, Issue JUL). https://doi.org/10.3389/fimmu.2018.01568

DRAFT

ORAL PRESENTATION

CYP1A1 Geni m1(T>C) ve m2 (A>G) Polimorfizminin Prostat Kanserinde Olası Rolünün Araştırılması

Rojda TANRIVERDİ[1] (0000-0002-7004-790X), Şenay Balci FIDANCI[1](0000-0002-7498-604X), Lülüfer TAMER[1](0000-0002-0997-0260), Murat BOZLU[2](0000-0002-8624-0149), Melih BIYIKOĞLU[2](0000-0002-4798-7389), Didem Derici YILDIRIM[3](0000-0001-7709-6133)

[*1]Mersin Üniversitesi, Tıp Fakültesi, Tıbbi Biyokimya AD, Mersin, Türkiye
[2] Mersin Üniversitesi, Tıp Fakültesi, Üroloji AD, Mersin, Türkiye
[3] Mersin Üniversitesi, Tıp Fakültesi, Biyoistatistik ve Tıbbi Bilişim AD, Mersin, Türkiye

*rjd_tnv_73@hotmail.com

Özet

Prostat kanseri, prostat bezindeki anormal hücreler kontrolsüz bir şekilde büyüdüğünde ve kötü huylu tümörler oluşturduğunda gelişir. Prostat kanseri ile CYP450 sisteminin polimorfizmleri arasındaki ilişkiyi inceleyen çalışmalar gün geçtikçe ilgi görmektedir. Bugüne kadar yapılan çalışmalarda CYP016 enzim ailesi ve alt gruplarındaki polimorfizmlerin meme kanseri, akciğer kanseri, ağız kanseri ve kolorektal kanser gibi kanser türleri ile anlamlı şekilde ilişkili olduğu bulunmuştur. Bu bilgiler ışığında CYP-450 enzimlerinden biri olan CYP1A1 geni ile ilişkili m1(T>C; rs4646903) ve m2 (A>G; rs1048943) polimorfizmlerinin prostat kanseri ve BPH ile olası ilişkisi araştırılmıştır. (Benign Prostatik Hiperplazi) hastaları kontrol grubu olarak kullanılmış ve sağlıklı kontrol grubunun verileri ile bölgemizde CYP1A1 polimorfizmlerinin alel sıklığının belirlenmesi amaçlandı.

Çalışmaya, dahil edilme kriterlerine uyan, MEÜ Hastanesi Üroloji polikliniğine başvuran prostat kanseri tanısı almış 60 hasta, Benign prostat Hiperplazisi tanısı almış 40 hasta ve 30 sağlıklı birey dahil edildi. Hasta ve kontrol gruplarından EDTA'lı hemogram tüplerine 2 ml kan örnekleri alındı ve DNA izolasyonu, DNA izolasyon kiti kullanılarak (Roche Diagnostics, GmbHMannheim Almanya) yapıldı.Elde edilen DNA örneklerinde CYP1A1 rs4646903 ve rs1048943 polimorfizm analizi, RT-PCR cihazı kullanılarak (Roche Diagnostics LightCycler 480, GmbHMannheim, Almanya) gerçekleştirildi. İstatistiksel analizler Statistica v.13.3.0 ve MedCalc v.11.5.0. paket programı ile yapıldı ve $p<0.05$ ise sonuçlar anlamlı kabul edildi.

Çalışma verileri incelendiğinde; Hasta grubu ve kontrol grubu arasında ve kontrol grubunu alt grupları olan gruplar ile karşılaştırıldığında CYP1A1 (rs1048943) polimorfizmi AA, GG ve AG genotipleri (p=0,81) ve CYP1A1 (rs4646903) polimorfizmi TT ve TC genotipleri (p=0,007) bakımından anlamlı bir ilişki saptanmadı. Bu konuda yeterli çalışma bulunmadığından, elde ettiğimiz verilerin literatüre katkısı olacağını düşünmekteyiz.

Anahtar Kelimeler: Prostat kanseri, ksenobiyotik, Sitokrom P450

CYP1A1 Gene in Prostate Cancer OF m1(T>C) and m2 (A>G) Polymophism Investigation of the Possible role

Abtract

Prostate cancer develops when abnormal cells in the prostate gland grow uncontrollably and form malignant tumors. Studies examining the relationship between prostate cancer and polymorphisms of the CYP450 system are getting more and more attention. Studies conducted to date have found that polymorphisms in the CYP016 enzyme family and subgroups are significantly associated with cancer types such as breast cancer, lung cancer, oral cancer and colorectal cancer. In the light of this information, the possible relationship of m1(T>C; rs4646903) and m2 (A>G; rs1048943) polymorphisms associated with the CYP1A1 gene, one of the CYP-450 enzymes, with prostate cancer and BPH was investigated. (Benign Prostatic Hyperplasia) patients were used as the control group and it was aimed to determine the allele frequency of CYP1A1 polymorphisms in our region with the data of the healthy control group.

The study included 60 patients diagnosed with prostate cancer, 40 patients diagnosed with Benign Prostatic Hyperplasia, and 30 healthy individuals who applied to the Urology outpatient clinic of MEU Hospital, who met the inclusion criteria. 2 ml blood samples were taken from the patient and control groups into hemogram tubes with EDTA and DNA isolation was performed using a DNA isolation kit (Roche Diagnostics, GmbHMannheim Germany). LightCycler 480, GmbHMannheim, Germany) was performed. Statistical analyzes Statistica v.13.3.0 and MedCalc v.11.5.0. It was done with a package program and the results were considered significant if $p<0.05$.

When the study data are examined; CYP1A1 (rs1048943) polymorphism AA, GG and AG genotypes ($p=0.81$) and CYP1A1 (rs4646903) polymorphism TT and TC genotypes ($p=0.007$) were significant between the patient group and the control group and compared with the control group subgroups. no relationship was detected. Since there are not enough studies on this subject, we think that the data we obtained will contribute to the literature.

Keywords: Prostate cancer, Xenobiotic, Cytochrome P450

GİRİŞ

Dünyada erkeklerde en çok görülen kanserlerden biri prostat kanseri olup ve yılda tahmini 1.600.000 vaka ve 366.000 ölümle sonuçlanan nonkutanöz kanserdir. Bu konudaki tıbbi ilerlemelere rağmen prostat kanseri, bening tümörlerin aşırı tedavisi ve/veya metastatik prostat kanserlerindeki yetersiz tedavilerden dolayı erkekler için önemli bir tıbbi sorun olmaya devam etmektedir. Prostat kanseri, prostat bezindeki anormal hücrelerin kontrolsüz bir şekilde büyüyerek malign tümör oluşturmasıyla gelişmektedir. Prostat kanserinde, normal prostat epitelinde sekretuar farklılaşmada artış, bazal hücre tabaka yapısında bozulma meydana gelmektedir. Aynı zamanda, nükleer ve nükleolar anormalliklerde ve mikrodamar yoğunluğunda artış olmasıyla birlikte ilerleyen preneoplazi veya displazi sonucu kanser oluşmaktadır [1]. 2022'de 24.200'den fazla erkeğe prostat kanseri teşhisi konduğu tahmin edilmektedir. Dünya genelinde tüm kanser türleri içerisinde PCa %7,3 görülme sıklığı ile en çok görülen üçüncü kanser türüdür. Ülkemizde ise yapılan araştırmalarda, PCa kanseri erkeklerde en çok görülen 3. kanser türü olup kanser kaynaklı ölümlerde 4. sırada bulunmaktadır [2,3].

Prostat kanserinde, tanı anında ortalama yaş 69'dur. İleri yaşın dışında etnik köken, genetik faktörler ve aile öyküsü değiştirilemez risk faktörlerini arasındadır [4]. Hastalığın etiyolojisi tam olarak bilinmemekle birlikte, çevresel faktörler arasında bulunan beslenmenin kanser gelişiminde önemli bir rol oynadığı, sağlıksız beslenme modellerinin kanser riskini arttırdığı belirtilmiştir. Ayrıca bireylere ait yaşam stilleri ve metabolik durumlarda hastalığın risk düzeyine etki ederken, sağ kalım düzeyini de etkilemektedir[5]. Obezite, hiperglisemi, vardiyalı çalışma, inaktivite ve kimyasal veya çevresel zararlara maruziyet, inflamasyon ve enfeksiyonlar da hastalığın değiştirilebilir risk faktörleri arasında bulunmaktadır [6]. Yaşanan coğrafya ve ırk gibi değişkenlerinde PCa insidansına etkisi olduğu gösterilmiştir. PCa görülme sıklığı ile ilgili yapılan çalışmalar sonucunda Afrikalı-Amerikalı erkeklerde en yüksek, Asyalılarda ise en düşük oranda gözlendiği raporlanmıştır [6-7]. Shimizu ve arkadaşlarının yapmış olduğu bir çalışmada, Japonya'dan Amerika Birleşik Devletleri'ne yerleşen erkeklerin, yaşam stilleri ve çevresel değişiklikler sebebiyle ülkelerindeki erkeklere kıyasla prostat kanseri görülme riski daha yüksek düzeyde görülmüştür [8].

Prostat kanseri gün geçtikçe tanı bakımından sıklığı yükselen, tanı ve tedavisindeki gelişmelerle beraber lokalize olarak yakalandığında kesin olarak kür olabilecek bir hastalık türüdür. Yakın geçmişe kadar prostat kanseri yaşlı erkeklerin kaçınılmaz bir sorunu olarak görülmüş ve üzerinde çok az çalışma yapılmıştır. Son 15 yılda prostat kanseri biyolojisi ve tedavisi üzerindeki çalışmalar artmıştır. Bunun nedeni daha iyi yaşamak isteyen yaşlı popülasyonun artmasıdır. Temel araştırmalar ileri moleküler biyoloji tekniklerinin gelişimi ve erken tanı ve tedaviye verilen yanıtın değerlendirilebildiği serum prostat spesifik antijen (PSA) testinin gelişimi ile daha da bir destek bulmuştur [9]. Kansere yatkınlık ile gen polimorfizmleri arasında bir ilişki olduğuna ilişkin kanıtlar mevcuttur. Onkogen ve tümör baskılayıcı genler, DNA onarımında bozulma, genetik instabilite, anjiogenez, adhezyon, ilaç direnci ve ksenobiyotik metabolizmasından sorumlu genlerin tek ve ya kombine etkisiyle, normal prostat hücrelerinin regülasyonu bozulabilmekte ve daha agresif, metastatik ve hormona refraktör prostat hücrelerine dönüşebilmektedirler [10]. Bunlar arasında özellikle ksenobiyotik metabolizmasından sorumlu olan sitokrom P450 monooksijenaz enzim sistemlerine (CYP450) ait polimorfizmler ile prostat kanser gelişimi arasindaki bağlantı her geçen gün giderek daha da artan bir şekilde

ilgi odağı olmaktadır. CYP450 monooksijenaz enzim sistemleri, iskelet kası hücreleri ve matür eritrosit haricinde prokaryotlarda ve tüm memeli hücre türlerinde yer alan hem-protein ailesidir.
CYP450 enzimleri, pek çok çevresel karsinojen, endojen bileşiklerin, ve toksik kimyasalın ksenobiyotiklerin metabolizmasından sorumludur. CYP450 enzim sistemi üç ayrı grupta (I. Grup/ II. Grup/III. grup) incelenir. 2. grupta bulunan enzimler daha çok biyomonitorizasyonda polimorfizmleri değerlendiren enzimlerdir. Bu grupta bulunan enzimler %70-80 oranında faz I metabolizmasında rol oynar [11]. İlaç biyotransformasyonu ve metabolizmasında rol alan en 0önemli sistem olan CYP450 enzim sistemi inhibisyon veya indüksiyon gibi birçok mekanizma ile değişime uğrayabilmektedir. Bunun neticesinde, bireyler arasında oldukça değişik formları ortaya çıkabilmektedir. CYPP450 sistemi baz dizilimi benzerliklerine göre P450 40 farklı tip aile içerisinde gruplandırılır [12].
CYP450 enzim ailesinden CYP1A1, karaciğer dışı dokularda başlıca epitelyal dokularda ifade bulan ve II grupta yer alan bir enzimdir. Pek çok ksenobiyotik kimyasalın; 7-etoksikumarin, 7-etoksiresorufin, teofilin, klorzoksazon, kafein ve poliaromatik hidrokarbonların (PAH) ve Benzo(a) piren gibi polisiklik aromatik hidrokarbonların (PAH) oksidasyonunu katalizler. Karsinojen biyoaktivasyonu sonucu, akciğer kanserine yatkınlık ile CYP1A1 arasındaki ilişki olduğunu raporlayan araştırmalar bulunmaktadır. Ayrıca CYP1A1 genotiplerinin akciğer kanseri ile ilişkili olduğu ortaya çıkmış olup bunun haricinde, çok sayıda farklı tip östrojen metabolizması ilişkili kanserle bağlantılı olduğu otaya çıkmıştır. CYP1A1ve kanser bağlantısını araştıran araştırmalarda iki polimorfizm üzerinde daha fazla durulmuştur. Bunlar CYP1A1 m1(T>C) ve m2 (A>G) polimorfizmleridir [13]. Her ikisi de tek nükleotid polimorfizmi (Single nucleotide polymorphism, SNP) olup SNP numaraları sırasıyla rs4646903, rs1048943 olarak belirtilmektedir [14].
Kanserin multifaktöriyel bir hastalık olduğu bilinmekle birlikte, genetik faktörlerin etkisini tam olarak ortaya koymak adına moleküler çalışmalara ihtiyaç duyulmaktadır. Günümüze kadar yapılan araştırmalarda akciğer, meme ve kolorektal kanseri gibi kanser vakalarında, CYP016 enzim ailesi ve alt gruplarında oluşan polimorfizmlerin bu hastalıklar ile ilişkili olduğu raporlanmıştır [14-19]. Prostat kanseri etiyolojisinde de CYP016 enzim sisteminde CYP9A0, CYP8C5 gibi alt gruplarındaki meydana gelen polimorfizmlerin prostat kanseri oluşumundaki rolü incelenmiştir. Ancak CYP1A1 gen polimorfizmi ile prostat kanserinde arasındaki bağlantıyı araştıran çok az çalışma bulunmaktadır.
Bu çalışmada bu bilgilerin ışığı altında CYP-450 enzimlerinden olan CYP1A1 geni ile ilişkili m1(T>C; rs4646903) ve m2 (A>G; rs1048943) polimorfizmlerinin prostat kanseri ile olan olası ilişkisinin araştırılması amaçlandı. Ayrıca kontrol grubu olarak kullanılan BPH (Benign Prostat Hiperplazisi) hastaları ve sağlıklı kontrol grubu verileri ile de CYP1A1 polimorfizmlerinin bölgemizde allel sıklığının saptanması hedeflendi.

MATERYAL VE METOT

Çalışma Grubu

Çalışmaya Mersin Üniversitesi Hastanesi, Üroloji Anabilim Dalı polinikliğine başvuran prostat kanseri tanısı almış 18-80 yaş arasında 60 birey (Hasta grubu), benign prostat hiperplazisi tanısı almış 18-80 yaş arasında 40 birey ve Mersin Üniversitesi Hastanesi Üroloji Anabilim Dalı polinikliğine başvuran rutin muayene ve testler sonucu herhangi bir sistemik hastalığı bulunmayan sağlıklı 18-80 yaş arasında 30 birey (Kontrol grubu) olmak üzere toplam 130 birey dahil edildi.

Bu çalışma 23/03/2022 tarihli ve 2022/197 sayılı kurul kararı ile Mersin Üniversitesi Klinik Araştırmalar Etik Kurulu tarafından onaylanmış olup, çalışmaya katılan tüm bireylerden yazılı onam formu alınmıştır.

Örneklerin Toplanması ve Saklanması

Çalışmaya Mersin Üniversitesi Hastanesi Üroloji Anabilim Dalı polinikliğine başvuran prostat kanseri tanısı almış 60 birey (Hasta grubu), benign prostat hiperplazisi tanısı almış 40 birey ve Mersin Üniversitesi Hastanesi Üroloji Anabilim Dalı polinikliğine başvuran rutin muayene ve testler sonucu herhangi bir sistemik hastalığı bulunmayan sağlıklı 30 birey (Kontrol grubu) olmak üzere toplam 130 birey dahil edildi. Hastalardan EDTA'lı hemogram tüplerine alınan kan DNA izolasyonu için, biyokimya tüplerine alınan kan örnekleri kreatinin, HDL, LDL, VLDL, AST, ALT, WBC, RBC, CRP ve Üre, PSA, Ferritin analizi için kullanıldı.Çalışmaya katılmayı kabul eden hastalardan EDTA'lı tüplere alınan ve +4°C'de saklanan kan

örneklerinden DNA izolasyonu, DNA izolasyon kiti kullanılarak (Roche Diagnostics, GmbHMannheim Almanya) yapıldı. Bu işlemde uygulanan presedür;

1- Steril bir tüpe (1,5 ml'lik kapaklı) 200 µL tam kan alındı ve üzerine sırası ile 200 µL bağlayıcı tampon ve 40 µL proteinaz K eklendi. Proteinaz eklenmesini takiben tüplerin kapakları hemen karıştırıldı.

2- Tüpler 10 dakika 70 °C' de inkübe edildi.

3- İnkübasyonun ardından 100 µL izopropanol eklenir, iyice karıştırılır ve karışım filtre tüpüne aktarıldı.

4- Bir dakika 8000 g'de santrifüj edildi.

5- Toplama tüpü değiştirildi ve tüpe 500 µL Inhibitor Removal Buffer eklendi. 1 dakika 8000 g'de santrifüj edildi ve toplama tüpü tekrar değiştirildi.

6- 0Ardından tüpe 500 µL wash buffer eklendi ve 1 dakika 8000 g'de santrifüj edildi. Toplama tüpü değiştirildi.

7- Bu yıkama işlemi bir kez daha tekrar edilerek, toplama tüpü değiştirildi.

8- Tüpe herhangi bir çözelti eklenmeden 10 saniye 13.000 g'de tekrar santrifüj edildi.

9- Son olarak 1,5 ml'lik kapaklı DNA saklama tüplerine filtre tüpleri yerleştirilerek içerisine 70 °C'de bekletilen elüsyon tamponundan 200 µL eklenerek 1 dakika 8000 g'de santrifüj edildi. Bu defa DNA elüsyon tamponunun yardımı ile 1,5 ml'lik kapaklı DNA saklama tüplerine geçtiği için filtre tüpleri uzaklaştırılır ve DNA eldesi tamamlanmış olur.

CYP1A1 rs4646903 ve rs1048943 polimorfizmi analizi

Elde edilen DNA örneklerinde CYP1A1 rs4646903 ve rs1048943 polimorfizm analizi, RT-PCR cihazı kullanılarak (Roche Diagnostics LightCycler 480, GmbHMannheim, Almanya) gerçekleştirildi. Analiz sırasında üretici firmanın talimatları doğrultusunda aşağıdaki reaksiyon karışımı hazırlandı. Reaksiyon karışımının hazırlanmasında kullanılan materyaller ve miktarları Tablo 1'de verilmiştir.

Tablo 1. Reaksiyon karışımının hazırlanmasında kullanılan materyaller ve miktarlar

İçerik	Miktar
H2O	4 µl
Master Mix	10 µl
Primer-probe Assay	1 µl
Toplam karışım hacmi	15 µl
DNA	5 µl

Multiwell plate içerisine her bir örnek için 15 µL karışım ve karışımın üzerine 5 µL DNA eklendi. CYP1A1 rs4646903 ve rs1048943 polimorfizmlerinin analizi için uygulanan PCR koşulları Tablo 2'de verilmiştir.

Tablo 2. CYP1A1 rs4646903 ve rs1048943 polimorfizmlerinin analizi için PCR koşulları

PCR aşamaları		Hedefisı (ºC)	Bekleme süresi (sn)
Denatürasyon		95	600
Amplifikasyon (40 Döngü)	Denatürasyon	95	15
	Anneal/extend	60	60
Soğuma		40	30

BULGULAR VE TARTIŞMA

Çalışmaya, Mersin Üniversitesi Hastanesi Üroloji polikliniğine başvurmuş 130 birey dahil edildi.

Çalışma grupları;

Hasta grubu; Prostat kanseri tanısı konulan 60 hasta (yaş: 69),

Kontrol grubu; Benign prostat hiperplazisi tanısı konulan 40 hasta (yaş: 65,5) ve 30 sağlıklı kişi (yaş: 58,50) olmak üzere toplam 70 birey.

Çalışma gruplarına ait yaş, kilo, boy, aile öyküsü, sigara kulanımı ve alkol kullanımı gibi demografik veriler Tablo 3'te verilmiştir.

Tablo 3. Hasta ve kontrol gruplarına ait demografik veriler

			Hasta n=60	**Kontrol n=70**	**p değeri**
Yaş	medyan[min-max]		69.0[51-82]	63.0[26-85]	0,001
Kilo (kg)	medyan[min-max]		79.5[60-125]	79.0[57-178]	0,799
Boy (cm)	medyan[min-max]		1.72[1,55-1,91]	1.77[1,55-1,80]	0,001
Aile Öyküsü	n %	Var	23.0(38,3%)	13.0(18,6%)	0,12
		Yok	37.0(61,7%)	57.0(81,4%)	
Sigara	n %	Var	14.0(20%)	12.0(20%)	1,000
		Yok	56.0(80%)	48.0(80%)	
Alkol	n %	Var	10.0(16,7%)	6.0(8,6%)	,161
		Yok	50.0(83,3%)	64.0(91,4)	

DRAFT

Tablo 4. PCa, BPH ve sağlıklı bireylere ait demografik veriler

			PCa n=60	BPH n=40	Sağlıklı n=30	P değeri
Yaş	medyan[min-max]		69.0[51-82]	65.5[41-85]	58.5[26-86]	<0,001
Kilo(kg)	medyan[min-max]		79.5[60-125]	81.0[57-178]	77.0[60-90]	0,303
Boy (cm)	medyan[min-max]		1.72[1,55-1,91]	1.74[1,55-1,85]	1.56[1,65-1,80]	<0,001
Aile Öyküsü	n %	Var	23.0(38,3%)	13.0(32,5%)	0(0%)	0,001
		Yok	37.0(61,7%)	27.0(67,5%)	30.0(100%)	
Sigara	n %	Var	12.0(20%)	10.0(25%)	4.0(13,3%)	0,482
		Yok	48.0(80%)	30.0(75%)	26.0(86,7%)	
Alkol	n %	Var	10.0(16,7%)	6.0(15%)	0(0%)	,063
		Yok	50.0(83,3%)	34.0(85%)	30.0(100%)	

Erkeklerde en çok görülen kanser türü prostat kanseri'dir [20,21]. Birçok gelişmiş ülkede en fazla görülen nonkutanöz malignitedir[22,23]. Dünya çapında gittikçe artan sayıda prostat kanseri tanısı konulduğundan bu hastalık ve hastalığın önlenmesiyle ilgili bilgiler daha önemli hale gelmiştir. Prostat kanserinde önemli bir risk faktör olan yaşla birlikte hastalık insidansı ve görülme sıklığı yükselmektedir. Prostat kanseri ileri yaşlarda görülen bir hastalıktır. Esas olarak hastalık 65 yaş üstü erkeklerde sık gözlenirken nadiren 40 yaş öncesi gelişir ve yaşla birlikte artmaktadır. 70 yaş üzeri erkeklerin nerdeyse yarısında mikroskobik düzeyde prostat kanseri bulunmaktadır ve bu hasta grubunda prevelans %57,4 civarındadır[24,25]. Yaşın prostat kanseri gelişimini nasıl kolaylaştırdığını halen bilinmemekle birlikte bu yaş prostat kanseri ilişkisine androjenik etkinin aracılık ettiği ifade edilmektedir[24].

Çalışmamızda, hasta grubunun yaş ortalaması 69, kontrol grubunun yaş ortalaması 58,50 olarak bulundu. Hasta grubunun yaş ortalamalarının kontrol grubuna göre daha yüksek olduğu bulundu. Gruplar arası karşılaştırmada anlamlı farklılık bulundu($p<0,001$). Kontrol grubunun alt grupları ile hasta grubu karşılaştırıldığında da yaş bakımından anlamlı farklılık tespit edildi($p<0,001$).

Prostat kanserinde risk faktörlerinden biri de aile öyküsüdür. Birinci derece akrabaları arasında prostat kanserli bir yakını olan bireydeki risk kontrol grubuna göre iki kat, iki yakını prostat kanseri olan bir bireydeki risk beş 5 kat artmaktadır[26,27]. Prostat kanserine kalıtsal yatkınlığı olan kişi yalnızca risk altında kalmayıp erken yaş prostat kanserine de adaydır. Kalıtsal yatkınlık sadece %5-10 olarak bildirilmiş ve erken yaş prostat kanserlerinin %40'ının (55 yaş öncesi tanı konulanlar) kalıtsal bir bozukluktan kaynaklandığı ileri sürülmüştür[26]. Segregasyon analizleri, erken yaş prostat kanserlerine otozomal dominant katılımlı bir genin neden olabileceğini göstermiştir[26,28]. Henüz tanımlamanayan bu allelin popülasyonlardaki tahmini frekansı yaklaşık %0,36'dır. Bu alele sahip olan bir kişinin, 85 yaşlarında prostat kanserine yakalanma riski %88'dir. Prostat kanserli erkek kardeşi olan kişilerin, prostat kanserli babalara olanlara göre daha yüksek riske sahip olmasına dayanarak X'e bağlı resesif kalıtım gösteren bir genin prostat kanserine neden olabileceği de ileri sürülmüştür[29].

T.C. Sağlık Bakanlığı, Türkiye Halk Sağlığı Kurumu, Aile Hekimliği Uygulamasında Önerilen PSM ve Tarama Testleri Kılavuzu, ailesinde prostat kanseri öyküsü olan 40 yaşından büyük erkekler ve aile öyküsü olmayan 50 yaşından büyük erkeklerde erken tanı ve önleme amaçlı hastanın bilgilendirilmesini ve bir üroloji uzmanına yönlendirilmesini önermektedir[30].

Çalışmamızda; Hasta ve Kontrol grubu karşılaştırıldığında bakımından anlamlı bir farklılık bulunmadı (p=0,12). Kontrol grubunun alt grupları ile hasta grubu karşılaştırıldığında da sigara kullanımı bakımından anlamlı farklılık tespit edildi (p<0,001).
Prostat kanserinde risk faktörlerinden biri de sigara kullanımıdır. Sigara kullanan hastalarda görülen prostat kanseri evresinin ve hastalığın ilerlemesinin kullanmayanlara oranla daha yüksek olduğunu ve bu hastalarda prostat kanserine bağlı ölüm oranlarının anlamlı olarak artış gösteren çalışmalar bulunmaktadır [31]. Health Professionals Follow-up Study (HPFS) kohort çalışmasında, prostat kanseri tanısı alan hastaların ortalama 22 yıllık takibinin değerlendirildiği sigara içenlerde prostat kanserine bağlı ölüm ve biyokimyasal rekürrens oranlarında %60 artış göstermiştir [32]. Yapılan çalışmalarda <20 paket yılı sigara kullanımı olan, 10 yıldan uzun süredir sigarayı bırakmış olan hastalarda prostat kanseri tanı anında riskin ortadan kalktığı gözlenmiş olup; >20 paket yılı sigara öyküsü olan, 10 yıldan daha az süredir sigarayı bırakmış hastalar ile halen sigara kullanımı olan hastaların ise tanı anında eşit oranda risk altında olduğu belirtilmiştir [33,34].
Çalışmamızda; Hasta ve kontrol grubu karşılaştırıldığında sigara kullanımı bakımından anlamlı bir farklılık bulunmadı (p=1,000). Kontrol grubunun alt grupları ile hasta grubu karşılaştırıldığında da sigara kullanımı bakımından anlamlı farklılık tespit edilmedi(p=0,482).

Tablo 5. Hasta ve kontrol gruplarına ait PSA değerleri

	Hasta **n=60**	**Kontrol n=70**		**p değeri**
		BPH n=40	**Sağlıklı n=30**	
PSA ***(ng/ml)***	11,65[4,20-582]	1,050[,09-9,82]		<0,001
		1,81[,09-9,82]	,6950[10-3,29]	

Prostat kanserinin erken tanısı, evreleme ve izleminde en önemli belirleyicilerinden biri de Prostat spesifik antijen (PSA)'dır [35]. Sadece prostatik duktal epitel ve prostatik asini hücrelerinde oluşturulmakta kalmayıp malin özellikte olan hücrelerde sentezlendiği gibi normal yapılı hücrelerde de oluşturulmaktadır[36]. Prostat kanserinde serum PSA düzeyi yükselir bunun haricinde BPH, prostatit, yaşlanma, prostatik infarkt gibi benin durumlarda da artış gösterdiği görülmüştür. Prostat kanseri tanısında daha kesin sonuçlar elde etmek için toplam PSA değeri kullanılmaktadır bunun yanında yaşa bağlı PSA, PSA dansitesi, transizyonel zon dansitesi, serbest PSA oranı gibi değerlerden de faydalanılır. Prostat spesifik antijen (PSA), prostat bezindeki hücreler (hem normal hücreler hem de kanser hücreleri) tarafından üretilen bir proteindir. PSA genel olarak semende bulunurken kanda az miktarda bulunur. Kandaki PSA seviyesi, mililitre başına nanogram (ng / mL) adı verilen birimlerle ölçülür. Prostat kanseri olma riski PSA seviyesi yükseldikçe yükselir fakat bir erkeğin prostat kanseri olup olmadığını belirleyecek kesin bir yüksek değer bulunmamaktadır. Klinik olarak kararda, bazen 4 ng/mL veya daha yüksek bir PSA değeri ölçüt olarak kullanırken, bazen de 2.5 veya 3 gibi daha düşük bir seviyede daha ileri testlerin yapılmasını istenmektedir. PSA değeri prostat kanseri olmayan erkeklerin çoğunda 4 ng/mL'nin altındadır. PSA seviyesi prostat kanseri geliştiğinde, genellikle 4'ün üzerine çıkar. 4'ün altındaki bir değerde olması, kanser olmadığının bir garantisini oluşturmaz. PSA düzeyi 4 ile 10 arasında olan erkeklerin prostat kanseri olma olasılığı yaklaşık 4'te 1'dir. PSA 10'dan fazla ise, prostat kanseri olma olasılığı % 50'nin üzerinde olmaktadır [37]. Kanser dışındaki bir dizi faktörün PSA seviyelerini de etkileyebiliyor olması prostat kanseri taramasında, PSA testi ile belirli bir kesme noktası kullanmanın zor olmasının bir nedenidir [37].
Prostat spesifik antijen (PSA), Androjen reseptörü (AR5α-redüktaz tip II (SRD5A2) ve sitokrom P450'yi (CYP) kodlayanlar dahil olmak üzere bir dizi genin PCa duyarlılık genleri olduğu doğrulanmıştır [38].
Çalışmamızda; Hasta ve kontrol grubu karşılaştırıldığında PSA değeri bakımından anlamlı bir farklılık bulundu (p<0,001). Kontrol grubunun alt grupları ile hasta grubu karşılaştırıldığında da PSA bakımından anlamlı farklılık tespit edildi(p<0,001).

Hasta ve kontrol gruplarında rs1048943 polimorfizminde genotip ve allel dağılıma ait bulgular
Hasta ve kontrol gubunun rs 1048943 polimorfizmindeki genotip ve allel dağılımları Tablo 6'da verilmiştir.

DRAFT

Tablo 6. rs1048943 polimorfizminde hasta ve kontrol gruplarındaki genotip ve allel dağılımı

Genotip	Hasta n=60	Kontrol n=70	p değeri
AA	52(86,7%	61(87,1%)	0,309
GG	2(3,3%)	0(0%)	
AG	6(10%)	9(12,9)	
Allel dağılımı			
A	110(91,67%)	131(93,57%)	0,635
G	10(8,33%)	9(6,43%)	

Tablo 7. rs1048943 polimorfizminde Pca, BPH ve sağlıklı gruba ait genotip ve allel dağılımı

Genotip	Pca n=60	BPH n=40	Sağlıklı n=30	p değeri
AA	52(86,7%)	37(92,5)	24(80%)	0,353
GG	2(3,3%)	0(0%)	0(0%)	
AG	6(10%)	3(7,5%)	6(20%)	
Allel dağılımı				
A	110(91,67%)	77(96,25%)	54(90%)	0,9087
G	10(8,33%)	3(3,75%)	6(10%)	

Hasta ve kontrol gruplarında rs4646903 polimorfizminde genotip dağılıma ait bulgular

Hasta ve kontrol gubunun rs 4646903 polimorfizmindeki genotip ve allel dağılımları Tablo 8'de verilmiştir.

Tablo 8. rs4646903 polimorfizminde hasta ve kontrol gruplarındaki genotip ve allel dağılımı

Genotip	Hasta n=60	Kontrol n=70	p değeri
TT	43(71,7%)	59(84,3%)	0,81
TC	17(28,3%)	11(15,7%)	
Allel dağılımı			0,11
T	103(85,83%)	129(92,14%)	
C	17(14,17%)	11(7,86%)	

Tablo 9. rs4646903 polimorfizminde polimorfizminde Pca, BPH ve sağlıklı gruba ait genotip ve allel dağılımı

Genotip	Pca n=60	BPH n=40	Sağlıklı n=30	p değeri
TT	43(71,7%)	33(82,5%)	26(86,7%)	0,217
TC	17(28,3)	7(17,5%)	4(13,3%)	
Allel sıklığı				
T	100(85,47%)	73(91,25%)	59(93,65%)	0,3147
C	17(14,53%)	7(8,75%)	4(6,75%)	

Kanserin multifaktöriyel bir hastalık olduğu bilinmekle birlikte, moleküler çalışmaların yapılmasına gereksinim duyulmaktadır. Bugüne kadar yapılan çalışmalarda akciğer kanseri, meme kanseri, kolorektal kanseri ve ağız kanseri gibi kanser vakalarında, CYP016 enzim ailesi ve alt gruplarında meydana gelen polimorfizmlerin önemli derecede ilişkili olduğu tespit edilmiştir [14-19]. Prostat kanseri etiyolojisinde de CYP016 enzim sisteminde CYP9A0, CYP8C5 gibi alt gruplarındaki polimorfizmlerin prostat kanseri oluşumundaki rolü araştırılmıştır. Ancak CYP1A1 gen polimorfizmi ile prostat kanserinde arasındaki araştıran çok az çalışma bulunmaktadır. Karsinojen biyoaktivasyonu, CYP1A1 polimorfizmi ve akciğer kanserine yatkınlık arasında bağlantı kuran birçok çalışma bulunmaktadır. Ayrıca akciğer kanseri dışında, CYP1A1 genotiplerinin çok sayıda farklı tip östrojen metabolizması ilintili kanserle ilişkisi olduğu bulunmuştur. CYP1A1ve kanser ilişkisini araştıran çalışmalarında iki polimorfizm ön plandadır. Bunlar CYP1A1 m1(T>C; rs4646903) ve m2 (A>G; rs1048943) polimorfizmleridir [13].

CYP'ler, hayvanlarda, bitkilerde ve mikroorganizmalarda bulunan büyük bir korunmuş protein süper ailesidir. Sitokrom P450 1A1 (CYP1A1) ve 1B1 (CYP1B1), CYP1 ailesinin önemli üyeleridir. Bir faz I enzimi olarak sınıflandırılan CYP1A1, aril hidrokarbon hidroksilaz aktivitesinde yer alırken, CYP1B1 östrojenlerin hidroksilasyonunda yer alır. CYP1A1 proteini, CYP süper ailesinin bir üyesidir ve akciğer, böbrek, bağırsak, cilt, gırtlak, plasenta, lenfosit, beyin dokularında yaygın olarak dağılır [39]. Bu enzimin ana rolü, PAH'lar, heterosiklik aminler, aflatoksin B1 ve östrojen [40] gibi çevresel kanserojenleri metabolize etmek olduğundan, CYP1A1 genindeki varyasyonlar CRC oluşumunu indükleyebilir. Şu anda, CYP1A1 enzim aracılı kanserojen aktivasyonu için yaygın olarak kabul edilen bir paradigma, CYP1A1'in polisiklik aromatik hidrokarbonları reaktif epoksit ara ürünlerine metabolize etmesidir, bu da DNA'ya kovalent olarak bağlanabilir ve daha sonra tümörleri indükleyebilir [41]. Yüksek minör alel sıklığı (MAF) ve akciğer kanseri, kolorektal kanser, meme kanseri, lösemi, özofagus karsinomu ve prostat kanseri [42] dahil olmak üzere çeşitli karsinomların [43] artmış riskine olası katılımları nedeniyle, *CYP1A1* rs1048943 A > G ve rs4646903 T > C polimorfizmleri daha yaygın olarak incelenmiştir. Kanser oluşumunu indüklemenin yanı sıra, *CYP1A1'in* polimorfizmleri ülseratif kolit, kolorektal adenom, ateroskleroz, miyokard enfarktüsü ve benzeri gibi diğer hastalıklara da yol açabileceği belirtilmiştir [44,46].

He ve Feng [47], CYP1A1'in östrojen metabolizmasında rol oynadığını ve aynı zamanda prokarsinojenleri aktive ettiğini bildirmiştir. Ayrıca, CYP1A1 polimorfizminin PCa gelişimi ile ilişkili olduğu gösterilmiştir. Tokizane ve ark. [48] promotör/arttırıcı bölgenin hipometilasyonunun PCa hücrelerinde aşırı CYP1B1 ekspresyonuna yol açtığını ve CYP1B1 katalizinden gelen metabolitlerin bir hayvan modelinde PCa'yı indüklediğinin gösterildiğini bulunmuştur [49] . Androjen bağımlı bir organ olarak prostat, steroid hormonlarının metabolizmasına katılan CYP1A1 ve CYP1B1 dahil olmak üzere birkaç enzim içerir. CYP1A1 ve CYP1B1 polimorfizmleri ile PCa riski arasındaki ilişkiyi araştıran önceki meta-analizler tutarsız sonuçlar bildirmiştir.

Son yıllarda, kansere genetik duyarlılık, bilimsel toplulukta bir araştırma noktası olmuştur. Ortaya çıkan kanıtlar, özellikle CYP1A1 ve CYP1B1 dahil olmak üzere CYP1 ailesinde, gen polimorfizmleri ile kanser

riski arasındaki potansiyel ilişkiyi göstermiştir. Örneğin, CYP1A1 polimorfizmlerinin akciğer [50], mesane [51], pankreas [52] ve meme kanserleri [53] dahil olmak üzere çok çeşitli kanserlere duyarlılık ile ilişkili olduğu bilinmektedir. Benzer şekilde, CYP1B1 polimorfizmlerinin ilişkisi ve çeşitli kanser türlerinin riski araştırılmıştır [54,56]. Günümüzde, CYP1 ailesi gen polimorfizmleri ile PCa duyarlılığı arasındaki bağlantı yaygın olarak dikkat çekmektedir. Çok sayıda çalışma, CYP proteinleri ve SNP'leri arasındaki ilişkiye ve bunların PCa gelişimi üzerindeki olası etkilerine odaklanmıştır. Hem CYP1A1 hem de CYP1B1, östrojenler de dahil olmak üzere çok sayıda kanserojen ve steroidal hormonun metabolizmasında rol oynar [57,58]. Ayrıca, Cavalieri ve ark. [59], CYP1B1 katalizi tarafından üretilen kimyasal kanserojenlerin metabolitlerinin hayvan modellerinde PCa'yı indükleyebileceğini bildirmiştir. CYP1A1 * 2A (rs4646903) polimorfizmi, poliadenilasyon bölgesinin aşağı akımındaki 3801′-kodlamayan bölgenin 3 pozisyonunda bir timidin sitozin ikamesini içerir [60]. Bazı çalışmalar rs4646903 varyantının mutant enzimlerin aktivitesini önemli ölçüde arttırdığını göstermesine rağmen, sonuçlar çelişkilidir [61]. CYP1A1 * 2B (rs1048943) polimorfizmi, ekzon 1'deki kodon 1'nin 2455 pozisyonunda bir adenin-guanin geçişini içeren ikinci en yaygın CYP462A7 polimorfizmidir [62].

Wei Zhu ve arkadaşlarının yapmış oldukları bir çalışmada rs1048943 ve rs4646903 nın prostat kanseri riski arasında anlamlı bir ilişki bulmadıklarını belirtmişlerdir. Bunu PCa'nın multigenik bir hastalık olduğu ve bu nedenle tek bir polimorfizmin etkisinin sınırlı olabileceğine bağlamışlardır. Ayrıca çalışmanın rs1048943 polimorfizminin genel popülasyonda PCa duyarlılığı ile ilişkili olduğunu AA ile karşılaştırıldığında AB genotipindeki rs4646903 polimorfizminin ve AA ile karşılaştırıldığında BB genotipinin sırasıyla Asya ve Beyaz ırk popülasyonlarında PCa için risk faktörleri olarak kabul edildiğine dair kanıtlarının sağlandığına bununla birlikte, CYP1A1 rs4646903 polimorfizmi, genel olarak artan PCa riski ile ilişkili olmadığı belirtmişlerdir. Polimorfizmlerin PCa duyarlılığı üzerindeki etkisi etnik kökenden etkilendiği açıklamışlardır. Ve yaptıkları bu meta analizin bu meta-analiz, CYP1A1 rs1048943 ve rs4646903 polimorfizmlerinin, sırasıyla özellikle Asya ve Beyaz ırk popülasyonlarında PCa riski ile ilişkili olduğunu öne sürmüşlerdir [63].

Sergentanis TN ve arkadaşlarının yapmış oldukları bir çalışmada Beyaz ırk, Çin ve Afrika popülasyonları üzerindeyaptıkları bir meta analiz çalışmasında *CYP1A1* rs1048943 polimorfizminin meme kanseri ile ilişkisini incelemek için menopoz öncesi ve menopoz sonrası kadınlarda araştırıldığında sonuçlar mutant genotipin (GG) meme kanseri riskini artırdığını gösterdi ve sonuçları, bu polimorfizmin bu popülasyonlarda meme kanseri tahmini için iyi bir belirteç olacağını belirtmişlerdir [64].

Çalışmamızda; Hasta ve Kontrol grupları arasında CYP1A1 (rs1048943) polimorfizmi AA, GG ve AG genotipleri bakımından anlamlı bir farklılık bulunmadı. Kontrol grubunu alt gruplara ayırıp hasta grubuyla karşılaştırıldığında da AA, GG ve AG genotipleri bakımından anlamlı bir farklılık bulunmadı. Hasta ve kontrol grupları arasında A ve G alelleri bakımından anlamlı bir farklılık bulunmadı. Kontrol grubunu alt gruplara ayırıp hasta grubuyla karşılaştırıldığında da A ve G alelleri bakımından anlamlı bir farklılık bulunmadı.

Çalışmamızda; Hasta ve kontrol grupları arasında CYP1A1 (rs4646903) polimorfizmi TT ve TC genotipleri bakımından anlamlı bir farklılık bulunmadı. Kontrol grubunu alt gruplara ayırıp hasta grubuyla karşılaştırıldığında da TT ve TC genotipleri bakımından anlamlı bir farklılık bulunmadı. Hasta ve kontrol grupları arasında T ve C alelleri bakımından anlamlı bir farklılık bulunmadı. Kontrol grubunu alt gruplara ayırıp hasta grubuyla karşılaştırıldığında da Tve C alelleri bakımından anlamlı bir farklılık bulunmadı.

Yapılan bu çalışmanın sonucunda rs1048943 ve rs4646903 polimorfizmlerinin prostat kanseri ile ilişkisi incelendiğinde gruplar arasında anlamlı bir farklılık bulunmamıştır. Bu durumun coğrafi dağılım ve ırksal farklılıklardan kaynaklandığını düşünmektedir. Yapılan literatür taramasında bu polimorfizmlerin bazı kanser türleri ilişkili olduğu gösteren çalışmalar bulunmasına rağmen prostat kanseri ile ilişkisi hakkında yeteri sayıda çalışma bulunmamaktadır. Elde ettiğimiz verilerin literatüre katkısı olacağını düşünmekteyiz.

SONUÇ

Çalışmamızın sonucunda;

Aile öyküsü açısından gruplar incelendiğinde, PCa grubundan aile öyküsü bulunanlar %38,3 iken, kontrol grubunda %18,6 olarak tespit edildi. Gruplar arası karşılaştırmada aile öyküsü açısından anlamlı bir farklılık bulunmadı (p=0,12).

Sigara kullanımı açısından gruplar incelendiğinde her iki grupta sigara kullananlar %20 olduğu bulundu. Hasta ve kontrol grubu sigara kullanımı bakımından karşılaştırıldığında anlamlı bir farklılık bulunmadı (p=1,000).

PCa grubunda alkol kullanımı %16,7 iken, kontrol grubunda %8,6 olarak tespit edildi. Gruplar arası karşılaştırmada alkol kullanımı açısından anlamlı bir farklılık bulunmadı (p=1,61).

Gruplar arası karşılaştırmada PSA değeri açısından anlamlı bir farklılık bulundu (p<0,001).
Kontrol grubunun alt grupları ile hasta grubu karşılaştırıldığında; BPH grubunda 40 birey bulunan PSA değeri 0,9-9,82 ng/ml arasında olup medyan değeri 1,81 ng/ml olarak bulundu. Sağlıklı bireylerin olduğu grupta 30 birey bulunup PSA değeri 3,29-10 ng/ml arasında olup medyan değeri 0,6950 ng/ml olarak bulundu. Gruplar arası karşılaştırmada PSA değeri açısından anlamlı farklılık bulundu (p<0,001).
Hasta ve kontrol grupları arasında CYP1A1 (rs1048943) polimorfizmi genotipleri bakımından anlamlı bir bulunmadı (p=0,007). Hasta grubunda A aleli %91,7 G aleli %8,33 olarak, kontrol grubunda ise A aleli %93,57 G aleli %6,43 olarak bulundu. Hasta ve kontrol grubunda A ve G alelleri karşılaştırıldığında anlamlı bir farklılık bulunmadı (p=0,635).
Sağlıklı birey grubunda ise AA genotipine 24(80%) birey, GG genotipine 0(0%) birey ve AG genotipine sahip 6(20%) birey bulundu. Kontrol grubu alt gruplara ayrıldığında ve hasta grubunda AA, AG ve GG genotipleri ile karşılaştırıldığında anlamlı bir farklılık bulunmadı (p=0,353). Hasta grubunda A aleli %91,7 G aleli %8,33, BPH grubunda A aleli %96,25 G aleli %3,75 ve sağlıklı bireyler grubunda A aleli %90 G alleli %10 bulundu. Kontrol grubu alt gruplara ayrıldığında ve hasta grubunda A ve G aleli ile karşılaştırıldığında anlamlı bir farklılık bulunmadı (p=0,9087).
Hasta ve kontrol grupları arasında CYP1A1 (rs1048943) polimorfizmi genotipleri bakımından anlamlı bir bulunmadı (p=0,81). Hasta grubunda A aleli %85,83 G aleli %14,17 iken kontrol grubunda A aleli %92,14 G aleli %7,86 olarak bulundu. Hasta ve kontrol grubunda T ve C alelleri karşılaştırıldığında anlamlı bir farklılık bulunmadı (p=0,11).
Hasta grubunda A aleli %85,47 G aleli %14,53 ,BPH grubunda A aleli %91,25 G aleli %8,75 ve sağlıklı bireyler grubunda A aleli %93,65 G alleli %6,75 olarak bulundu. Kontrol grubu alt gruplara ayrıldığında ve hasta grubunda T ve C aleli ile karşılaştırıldığında anlamlı bir farklılık bulunmadı (p=0,3147).
Bu konuda yeterli çalışma bulunmadığından, elde ettiğimiz verilerin literatüre katkısı olacağını düşünmekteyiz.

KAYNAKLAR

[1] Zaorsky, N.G, Churilla, T, Egleston, B. and Fisher, S. “Causes of Death Among Cancer Patients”. Annals of Oncology, 2017; 28 (2), 400-407.

[2] Erzurum Alim, N. (2018). “Türkiye’de ve Dünyada Kanser Epidemiyolojisi”. Sağlık Bakanlığı Halk Sağlığı Genel Müdürlüğü. Erişim adresi: https://hsgm.saglik.gov.tr (Erişim tarihi: 07.04.2022).

[3] Sung, H, Ferlay, J, Siegel, R.L. and Laversanne, M. “Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries”. Cancer Journal for Clinicians, 2021; 71 (3), 209-249

[4] Xu, X, Kharazmi, E, Tian, Y. and Mukama, T. “Risk of Prostate Cancer in Relatives of Prostate Cancer Patients in Sweden: A Nationwide Cohort Study”. PLoS Medicine, 2021; 18 (6), e1003616.

[5] Pernar, C.H, Ebot, E.M, Wilson, K.M. and Mucci, L.A. “The Epidemiology of Prostate Cancer”. Cold Spring Harbor Prspectives in Medicine, 2018; 8 (12), a030361.

[6] Chung, B.H, Horie, S. and Chiong, E. “The Incidence, Mortality, and Risk Factors of Prostate Cancer in Asian Men”. Prostate International, 2019; 7 (1), 1-8.

[7] Taitt, H.E. “Global Trends And Prostate Cancer: A Review of Incidence, Detection, and Mortality as Influenced by Race, Ethnicity, and Geographic Location”. American Journal of Men's Health, 2018; 12 (6), 1807-1823.

[8] Shimizu, H, Ross, R.K, Bernstein, L. and Yatani, R. “Cancers of the Prostate and Breast Among Japanese and White Immigrants in Los Angeles County”. British Journal of Cancer, 1991; 63 (6), 963-966. https://doi: 10.1038/bjc.1991.210

[9] Taplin ME, Ho S. The endocrinology of prostate cancer. J. Clin Endoc Metab 2001; 86: 3467-3477

[10] Kallioniemi OP, Visakorpi T. Genetic basic and clonal evolution of human prostate cancer. Advances in Cancer Research 1996; 68:225-255

[11] Ingelman- Sundberg M.Human drug metabolizing cytochrome P-450 enzymes; properties and polymorphisms. Naunyn –Schmiedeberg’s Arch Pharmacol 2004; 369:89-104

[12] Journal of Turgut Özal Medical Center 1996;3(3):257-275

[13] Abbas, M.., Shrivastav, K.., Imran, M.., Banerjee, M.., Assocation of CYP1A1 gene variants rs4646903 and rs1048943 with cercival cancer in North Indian population. Eur. J. Obstet. Gynecal. Reprod. Biol. 2014; 176,68-74

[14] Habano W, Gamo T, Sugal T, Otsuka K, Wakabayaski G, Ozawa S. CYP1B1, but not CYP1A1, is downregulated by promoter methylation in colorectalcancers. Int J Oncol.2009;34(4);1085-1091

[15] Liao LH, Zhang H, Lai MP, Lau KW, Lai AK, Zhang JH et al. The association of CYP2C9 gene polymorphisms with colorectal carcinoma in Han Chinese, Clin Chim Acta, 2007;380(16):191-196

[16] Levkovich NN, Gorovenko NG, Myasoedov DV. Association of polymorphic G1934A variant (allele *4) of CYP2D6 gene with increased risk of breast cancer development in Ukrainian women. Exp Oncol. 2011; 33(3): 136–139

[17] Gan CQ, Wang XY, Cao YD, Ye WX, Liu H, Sun YY. Association of CYP2C19*3 gene polymorphism with breast cancer in Chinese women. Genet Mol Res. 2011; 10(4):3514-3519

[18] Fuster V, Badimon L, Badimon JJ, Chesebro JH. Mechanisms of disease, the pathogenesis of coronary artery disease and the acute coronary syndromes (part one). N Engl J Med. 1992; 326(1): p.242–250

[19] Nimptsch K, Rohrmann S, Nieters A, Linseisen J. Serum Undercarboxylated Osteocalcin as Biomarker of Vitamin K Intake and Risk of Prostate Cancer: A Nested Case- Control Study in the Heidelberg Cohort of the European Prospective Investigation into Cancer and Nutrition. Cancer Epidemiol Biomarkers Prev. 2009; 18(1): 49-56

[20] Epstein J.I.,Tumors of the prostate,Chapter 3 in Pathology and genetics of tumor of the urinary system and male genital organs,Ed:Eble J.N.,Souter G.,Epstein J.I.,Sesterhenn I.A.,IARC Pres Lyon,2004; 159-215 World Health Organization Classification of Tumors.

[21] Epstein J.I.:The Lower Urinary Tract and Male Genital System.Chapter:21 in Robbins and Cotran Pathologic disease,Edited by Kumar V.,Abbas A.K.,Fausto N.,7th ed,Elsevier Saunders,Philadelphia,2005; 1023-58.

[22] Young R.H., Srigley J.R.,Amin M.B.,Ulbright T.M.,Cubilla A.L.,Tumors of the prostate gland,seminal vesicles,male urethra and penis.Armed Forces Institute of Pathology.Washington,D.C,2000;1-344.

[23] Moline V.,Herve J.M.,Lugafne P.M.;Baglin A.C.:p63 and p504 S Coctail is useful in ambiguous lesions of the prostate(Letter).Histopathology 2004;44:403-4.

[24] Stamatiou KN. Elderly and prostate cancer screening. Urol J. 2011 Spring;8(2):83-7.

[25] Zeliadt SB, Hoffman RM, Etzioni R, Gore JL, Kessler LG, Lin DW. Influence of publication of US and European prostate cancer screening trials on PSA testing practices. J Natl Cancer Inst. 2011; 103: 520-3.

[26] Carter BS, Beaty TH, Steinberg GD, et al. Mendelian inheritance of familial prostate cancer. Proc Natl Acad Sci USA 1992;89: 3367-3371

[27] Steinberg GD, Carter BS, Beaty TH, et al. Family history and the risk of prostate cancer. Prostate 1990; 17: 337-347

[28] Carter BS, Bova GS, Beaty TH, et al. Hereditary prostate cancer: epidemiologic and clinical features. J Urol 1993; 150: 797-802.

[29] Narod S, Dupond A, Cusan L, et al. The impact of family history on early detection of prostate cancer. Nat Med 1995; 1: 99-101

[30] Aile Hekimliği Uygulamasında Önerilen Periyodik Sağlık Muayeneleri ve Tarama Testleri, T.C. Sağlık Bakanlığı Türkiye Halk Sağlığı Kurumu, yayın no: 991, 2015.

[31] Warren GW, Alberg AJ, Kraft AS, et al. The 2014 Surgeon General's report: "The health consequences of smoking--50 years ofprogress": a paradigm shift in cancer care. Cancer 2014;120:1914-1916.

[32] Kenfield SA, Stampfer MJ, Chan JM, et al. Smoking and prostate cancer survival and recurrence. JAMA 2011;305:2548-2555.

[33] Daniell HW. A worse prognosis for smokers with prostate cancer. J Urol 1995;154:153-157.

[34] Gong Z, Agalliu I, Lin DW, et al. Cigarette smoking and prostate cancerspecific mortality following diagnosis in middle-aged men. Cancer Causes Control 2008;19:25-31.

[35] Partin AW, Osterling JE: The clinical usefulness of prostate spesific antigen. J Urol. 1994; 152: 1358,

[36] Brawer MK, Lange PH: Prostate spesific antigen: Its role in early detection, staging and monitoring of prostatic carcinoma. J. Endourol. 1989;3: 227

[37] https://www.kanser.org/saglik/toplum/kanser-turleri-alt-kategori/prostat-kanserinde-erken-tani

[38] Ding G., Xu W., Liu H.. et al. CYP1A1 MspI polymorphism is associated with prostate cancer susceptibility: evidence from a meta-analysis. *Mol. Biol. Rep*. 2013; 40, 3483–3491 10.1007/s11033-012-2423-0 [PubMed] [CrossRef] [Google Scholar]

[39] Crewe HK, Notley LM, Wunsch RM, Lennard MS, Gillam EM. Metabolism of tamoxifen by recombinant human cytochrome P450 enzymes: formation of the 4-hydroxy, 4′-hydroxy and N-desmethyl metabolites and isomerization of trans-4-hydroxytamoxifen. Drug Metab Dispos. 2002; 30:869–874.

[40] Gonzalez J, Marchand-Geneste N, Giraudel JL, Shimada T. Docking and QSAR comparative studies of polycyclic aromatic hydrocarbons and other procarcinogen interactions with cytochromes P450 1A1 and 1B1. SAR QSAR Environ Res. 2012; 23:87–109.

[41] He X, Feng S. Role of Metabolic Enzymes P450 (CYP) on Activating Procarcinogen and their Polymorphisms on the Risk of Cancers. Curr Drug Metab. 2015; 16:850–863.

[42] Hussein AG, Pasha HF, El-Shahat HM, Gad DM, Toam MM. CYP1A1 gene polymorphisms and smoking status as modifier factors for lung cancer risk. Gene. 2014; 541:26–30.

[43] Daly AK. Polymorphic Variants of Cytochrome P450:Relevance to Cancer and Other Diseases. Adv Pharmacol. 2015; 74:85–111.

[44] Ma C, Lu XC, Fan L, Luo Y, Yang B, Gao Y, Liu XF. Irbesartan regulates inflammatory gene expressions related to atherosclerosis in EA.hy926 cells [Article in Chinese]. Nan Fang Yi Ke Da Xue Xue Bao. 2011; 31:1835–1839.

[45] Buyukgoze O, Osmanoglu N, Arslan S, Sen A. Association of the CYP1A1*2A, GSTT1 null, GSTM1 null, mEPHX*3, and XRCC1-399 genetic polymorphisms with ulcerative colitis. Int J Colorectal Dis. 2013; 28:593–595.

[46] Hamachi T, Tajima O, Uezono K, Tabata S, Abe H, Ohnaka K, Kono S. CYP1A1, GSTM1, GSTT1 and NQO1 polymorphisms and colorectal adenomas in Japanese men. World J Gastroenterol. 2013; 19:4023–4030.

[47] He X. and Feng S. Role of metabolic enzymes P450 (CYP) on activating procarcinogen and their polymorphisms on the risk of cancers. *Curr. Drug Metab.* 2015; 16, 850–863 10.2174/1389200216101512101645O1 [PubMed] [CrossRef] [Google Scholar]

[48] Tokizane T., Shiina H., Igawa M.. et al. Cytochrome P450 1B1 is overexpressed and regulated by hypomethylation in prostate cancer. *Clin. Cancer Res.* 2005; 11, 5793–5801 10.1158/1078-0432.CCR-04-2545 [PubMed] [CrossRef] [Google Scholar]

[49] Williams J., Martin F., Muir G.. et al. Metabolic activation of carcinogens and expression of various cytochromes P450 in human prostate tissue. *Carcinogenesis* 2000; 21, 1683–1689 10.1093/carcin/21.9.1683 [PubMed] [CrossRef] [Google Scholar]

[50] Nie, Q., Yang, X.-n., An, S.-j. et al. CYP1A1* 2A polymorphism as a predictor of clinical outcome in advanced lung cancer patients treated with EGFR-TKI and its combined effects with EGFR intron 1 (CA) n polymorphism. Eur. J. Cancer 2011; 47, 1962–1970,

[51] Ozturk, T., Kahraman, ¨ O.T., Toptas ¸, B. et al. The effect of CYP1A1 and GSTM1 gene polymorphisms in bladder cancer development in a Turkish population. In Vivo 2011; 25, 663–668

[52] Liu, G., Ghadirian, P., Vesprini, D. et al. Polymorphisms in GSTM1, GSTT1 and CYP1A1 and risk of pancreatic adenocarcinoma. Br. J. Cancer 2000; 82, 1646

[53] Chen, C., Huang, Y., Li, Y. et al. Cytochrome P450 1A1 (CYP1A1) T3801C and A2455G polymorphisms in breast cancer risk: a meta-analysis. J. Hum. Genet. 2007; 52, 423, https://doi.org/10.1007/s10038-007-0131-8

[54] Berber, U., Yilmaz, I., Yilmaz, O. et al. CYP1A1 (Ile 462 Val), CYP1B1 (Ala 119 Ser and Val 432 Leu), GSTM1 (null), and GSTT1 (null) polymorphisms and bladder cancer risk in a turkish population. Asian Pac. J. Cancer Prev. 2013; 14, 3925–3929, https://doi.org/10.7314/APJCP.2013.14.6.3925

[55] Ibrahim, M., Rashed, R., Hassan, N. et al. Association of cytochrome P450-1B1 gene polymorphisms with risk of breast cancer: an Egyptian study. Asian Pac. J. Cancer Prev. 2016; 17, 2861–2866

[56] Shah, I.A., Mehta, P., Lone, M.M. et al. Leu432Val polymorphism of CYP1B1 is not associated with squamous cell carcinoma of esophagus-a case-control study from Kashmir, India. Asian Pac. J. Cancer Prev. 2015; 16, 5337–5341, https://doi.org/10.7314/APJCP.2015.16.13.5337

[57] Hanna, I.H., Dawling, S., Roodi, N. et al. Cytochrome P450 1B1 (CYP1B1) pharmacogenetics: association of polymorphisms with functional differences in estrogen hydroxylation activity. Cancer Res. 2000; 60, 3440–3444

[58] Kumar, V., Singh, S., Yadav, C.S. et al. CYP1A1 and CYP3A4 polymorphic variations in Delhi population of Northern India. Environ. Toxicol. Pharmacol. 2010; 29, 126–130, https://doi.org/10.1016/j.etap.2009.12.001

[59] Cavalieri, E.L., Devanesan, P., Bosland, M.C. et al. Catechol estrogen metabolites and conjugates in different regions of the prostate of Noble rats treated with 4-hydroxyestradiol: implications for estrogen-induced initiation of prostate cancer. Carcinogenesis 2002; 23, 329–333, https://doi.org/10.1093/carcin/23.2.329

[60] Bale, A. Subchromosomal localization of the dioxin-inducible P1450 locus (CYP1) and description of two RFLPs detected with P1450 cDNA probe. Cytogenet. Cell Genet. 2002; 46, 574–575

[61] Androutsopoulos, V.P., Tsatsakis, A.M. and Spandidos, D.A. Cytochrome P450 CYP1A1: wider roles in cancer progression and prevention. BMC Cancer. 2009; 9, 187, https://doi.org/10.1186/1471-2407-9-187

[62] Hayashi, S.-i., Watanabe, J., Nakachi, K. et al. Genetic linkage of lung cancer-associated Msp I polymorphisms with amino acid replacement in the heme binding region of the human cytochrome P450IA1 gene. J. Biochem. 1991;110, 407–411, https://doi.org/10.1093/oxfordjournals.jbchem.a123594

[63] Zhu W, Liu H, Wang X, Lu J, Zhang H, Wang S, Yang W. Associations of CYP1 polymorphisms with risk of prostate cancer: an updated meta-analysis. Biosci Rep. 2019 Mar 1;39(3):BSR20181876. doi: 10.1042/BSR20181876. PMID: 30765615; PMCID: PMC6395298.).

[64] Sergentanis TN, Economopoulos KP. Four polymorphisms in cytochrome P450 1A1 (CYP1A1) gene and breast cancer risk: A meta-analysis. *Breast Cancer Res Treat.* 2010;122:459–69.

ORAL PRESENTATION

Balıkçı teknelerinde tekne ve makine sigortası

Ece Akın[1] (*https://orcid.org/0000-0002-6521-0274*), Okan Akyol[2] (*https://orcid.org/0000-0001-7738-2156*)

[1]Ege Üniversitesi, Fen Bilimleri Enstitüsü, Su Ürünleri Avlama ve İşleme Teknolojisi ABD, İzmir, Türkiye
[2]Ege Üniversitesi, Su Ürünleri Fakültesi, Avlama ve İşleme Teknolojisi Bölümü, İzmir, Türkiye

eetkesen@gmail.com

Özet

Balıkçılık çok eski uğraşlardan biri olmakla birlikte zaman içerisinde avlanma biçimleri oldukça değişmiştir. Teknolojinin gelişimiyle kullanılan av araçları ve yardımcı ekipmanların da gelişimini arttırmıştır. Bu gelişme balıkçılığın daha büyük araç ve ekipmanlar ile ticari boyutta yapılmasının önünü açmıştır. Avlanan ürün miktarının artışı ve bu avcılığı gerçekleştirmek üzere teknelerle birlikte denizde kalınan sürenin artması sonucu, denizde karşılaşılan risklerin sayısı ve maruz kalma olasılığı da buna bağlı olarak yükselmiştir. Riskin varlığı, bu riskin oluşturabileceği kayıpların bir şekilde telafi edilmesi gereğini doğurmuştur. Buradaki en büyük telafi mekanizması ve risk ile mücadele edecek araç "sigorta"dır. Balıkçı teknelerinde temel olarak üç çeşit sigorta teminatı uygulanabilir. Direkt olarak tekne ve üzerinde bulunan ekipmanın karşılaşabileceği riskleri kapsayan Tekne ve Makine Sigortası, Teknenin Üçüncü Şahıslara verebileceği zarardan dolayı Sorumluluk Sigortası (P&I : Protection and Indemnity : Koruma ve Tazmin) ve son olarak da tekne üzerinde görevli personel (kaptan, mürettebat), yolculara gelebilecek riskleri kapsam altına alan Ferdi Kaza Sigortası. Bu bildiride balıkçı teknelerine uygulanabilecek sigortalardan Tekne ve Makine sigortası hakkında pratik bilgiler verilmiştir.

Anahtar Kelimeler: Balıkçılık, Sigorta, Tekne, Makine.

Hull and machinery insurance for fishing vessels

Abstract

Fisheries are one of the oldest types of occupation, and the way of fishing has changed considerably over time. With the development of technology has increased the fishing equipment and auxiliary equipment used. This development has enriched the way for commercial fishing with larger tools and equipment. As a result of the increase in the number of products caught and the time spent at sea with the boats to carry out this fishing, the number of risks encountered on the sea has increased and the probability of exposure has also increased accordingly. The existence of the risk has necessitated the compensation of the losses that this risk may cause in some way, the prominent compensation mechanism here and the tool to handle the risk is "insurance". Basically, 3 types of insurance coverage can be applied to fishing boats. Hull and Machinery Insurance covers the risks that the boat and the equipment on it may face directly, Liability Insurance due to the damage that the boat may cause to third parties (P&I: Protection and Indemnity) and finally the personnel on the boat (captain, crew) Personal Accident Insurance, which covers risks. In this study, practical information about Hull and Machinery insurance, which can be applied to fishing boats, is given.

Keywords: Fishing, insurance, vessel, machine.

GİRİŞ

Balıkçı tekneleri avladıkları tür ve kullandıkları av araçları ve avlanma miktarına göre farklılık gösterir. Balıkçı gemileri genel olarak denizlerde ve tatlı sularda su ürünlerinin avlanmasında, üretilmesinde, korunmasında, işlenmesinde, taşınmasında kullanılan gemilerdir (Abur, 2023). Tahmini olarak tüm dünya genelinde 2020 yılı itibari ile 4,1 milyon balıkçı gemisi mevcuttur (FAO,2022) .Türkiye'de Balıkçılık ve Su Ürünleri Genel Müdürlüğü 2022 verilerine göre kayıtlı gemi sayısı içsular ve denizlerde toplam 18.444'dür.

Balıkçı tekneleri, balıkçıların hem mesleki faaliyetlerine devam edebilmelerini sağlayan bir araç hem de şahsi ya da tüzel olarak bir mal varlığıdır. Bu minvalde, kendilerine ya da teknelerine gelebilecek bir zarar ya da kayıp, gelir kaynağının sekteye uğramasına, refah seviyesinin düşmesine dolayısıyla ülke ekonomisine de negatif etkide bulunabilir. Bu sebeple, riskin transferi ve yönetimi oldukça önemlidir. Günümüzde tüm dünyada risk yönetiminin en etkili aracı 'sigortadır'.

Dünya çapında faaliyet gösteren tahmini 67.800 büyük ölçekli balıkçı gemisinin en az %90'ı (>24 m uzunluğunda) tekne ve makine sigortası kapsamındadır. Tekne ve Makine Sigortası kapsamındaki balıkçı gemilerinin sayısının ise global olarak 450.000 civarında olduğu tahmin edilmektedir. Sigortalı balıkçı gemilerinin %61'i Asya'da, %18'i Amerika'da, %14'ü Avrupa'da ve %6'sı Afrika'dadır. Yarı endüstriyel balıkçı gemilerinin (12-24 m) %50 ve %60'ı tekne ve makine sigortası kapsamındadır. Dünya çapında faaliyet gösteren tahmini 430.000 yarı endüstriyel balıkçı teknesinin on binlercesi ise herhangi bir sigortaya sahip değildir. 2,3 milyon motorlu küçük ölçekli balıkçı gemilerinin (<12 m) %95'inden fazlası sigortasızdır. Çoğunlukla küçük ölçekli balıkçıların özel ihtiyaç ve şartları sağlayabilecek yeterli sigortaya erişimleri yoktur (FAO, 2022).

Literatür taraması sırasında eksikliği görülen özellikle balıkçılık faaliyetlerinde, ihtiyaca yönelik bir sigorta uygulamasının bulunmaması, sigorta sektörü içerisinde bu mesleki faaliyetin tam olarak bilinmemesi ve risk unsurlarının karmaşık yapısı sebebi ile uygun bir teminat türünün olmayışı, balıkçılık faaliyetinde bulunan işveren ve çalışanların sigorta konusundaki bilgi ve tecrübe eksikliği riske bu denli açık bir sektörün doğru risk yönetimi yapamamasına neden olmaktadır. Bu bildiride balıkçı teknelerine uygulanabilecek sigortalardan Tekne ve Makine sigortası hakkında pratik bilgiler verilmiştir.

MATERYAL VE METOT

Türkiye'de uygulanan Tekne ve Makine sigortasının genel şartları itibari ile teminat altına aldığı riskler ve teminat harici olan riskler belirtilecektir. Üç yıldır düzenli olarak sigortalanan 19 adet teknenin bedel ve prim bilgileri grafik halinde gösterilerek yıllar içerisindeki değişimi karşılaştırılacaktır.

BULGULAR VE TARTIŞMA

Tekne ve Makine Sigortası; tekne ve teknenin içerisinde bulunan makine, vinç, jeneratör, ırgat gibi donanımları ve teknenin tüm ekipman ve teçhizatlarını sigortacı ile karşılıklı mutabakat ile teminat altına alır. Balıkçı teknelerinde bulunan ağ makarası (power block), balık saklama makinesi, buzluk, dondurucu, balık pompası, balık ağları gibi ekipmanlar da dâhil edilebilir. Bir Tekne ve Makine Poliçesinde aşağıdaki detaylar yer almaktadır;

Tekne Adı
Tekne Bayrağı
Teknenin Yapım Malzemesi
İnşa Yılı
Ana Makine
Sigorta Bedeli
Sefer Sahası
Tekne Tipi
Boy, Genişlik

Sigortacı yukarıdaki bilgiler ile birlikte sigortalıya teklif iletebilir. Teklif verilirken, sigorta şirketlerinin kullandığı, geçmiş yıllardaki bilgilerin istatistiksel verileri ile tasarlanmış özel modüller ve/veya aktüerler tarafından teknenin boyu, yaşı, sefer sahası göz önünde bulundurularak bir fiyatlandırma yapılır. Bu fiyat çarpanı sigorta bedeli olarak belirtilmiş olan tekne bedeline uygulanır. Tekne bedeli içerisinde teknenin kabuk hali birlikte, tekne üzerindeki seyir ve seyire yardımcı ekipmanlar, vinçler, ırgatlar, jeneratörler, makineler, şaft, pervane tümüyle sigorta bedeline dâhil edilir. Yukarıda belirtilen fiyat çarpanı sigorta bedeline uygulanarak net prim bilgisi elde edilir. Özellikle Tekne ve Makine sigortalarını fiyatları ya da sigortacının riski kabul edebilirliği teknenin bedeli, teknenin yaşı ve kondisyon durumuna göre değişmektedir. Özellikle 30 yaşını geçmiş tekneler çoğu sigorta şirketinin kabul kriterlerine uymamaktadır. Bunun yanı sıra sigortacının

tekneye ekspertiz talebi doğrultusunda da teknenin göz ile muayenesi ve buna uygun olarak risk kabulü veya reddi gerçekleştirilir.

Tekne ve Makine Sigortalarının Rizikoları:

1) Deniz, ırmak, göllerin veya diğer sefer yapılabilir suların tehlikeleri.
2) Yangın, patlama (infilak).
3) Geminin dışındaki kişiler tarafından zor kullanılarak yapılan hırsızlık.
4) Denize mal atılması.
5) Korsanlık.
6) Kara taşıtları, rıhtım veya liman düzen veya donanımı ile temas.
7) Deprem, yanardağ püskürmesi veya yıldırım.
8) Yük veya yakıtın yüklenmesi, boşaltılması ya da yer değiştirmesi sırasında olan kazalar.
9) Bu sigorta, sigortalı edilen şeyin, aşağıdaki rizikolar nedeniyle uğradığı zıya veya hasarı kapsar.
9.1. Kazanların patlaması, şaftların kırılması veya makine ya da teknedeki herhangi bir gizli kusur.
9.1.2 Kaptanın, gemi zabitlerinin, gemi adamlarının ya da kılavuzların ihmali.
Burada sigortalı olmamaları koşulu ile onarımcılar ya da gemi kiracılarının ihmali.
9.1.3. Kaptan, gemi zabitleri veya gemi adamlarının barataryası.
9.1.4. Hava taşıtları, helikopter veya benzer nesnelerle ya da bunlardan düşen şeylerle temas.
Ancak zıya veya hasarın, sigortalı, gemi sahibi, gemi işletmecileri ya da gemi sahibinin eksperleri yahut idare merkezindeki [karadaki] her hangi müdürün gerekli özeni göstermemesinden kaynaklanmaması şarttır.
9.2. Kaptan, gemi zabitleri, gemi adamları veya kılavuzlar gemide pay sahibi olsalar bile bu rizikolar anlamı içinde gemi sahibi olarak düşünülemez. (Institute Time Clauses Hulls,1995)

Yukarıda belirtilen rizikolara ek olmak üzere belirli şartlar, sınırlamalar ve istisnalar dâhilinde (bu şartlar, sınırlamalar ve istisnalar sigorta sözleşmesi içerisinde açıkça beyan edilir); Kirletme Tehlikesi, ¾ Çatışma Sorumluluğu, Kardeş Gemi ile Çatışma ve/veya Kurtarma Hizmeti Alma, Müşterek Avarya ve Kurtarmadan doğacak hasarlarda teminat dâhilindedir.

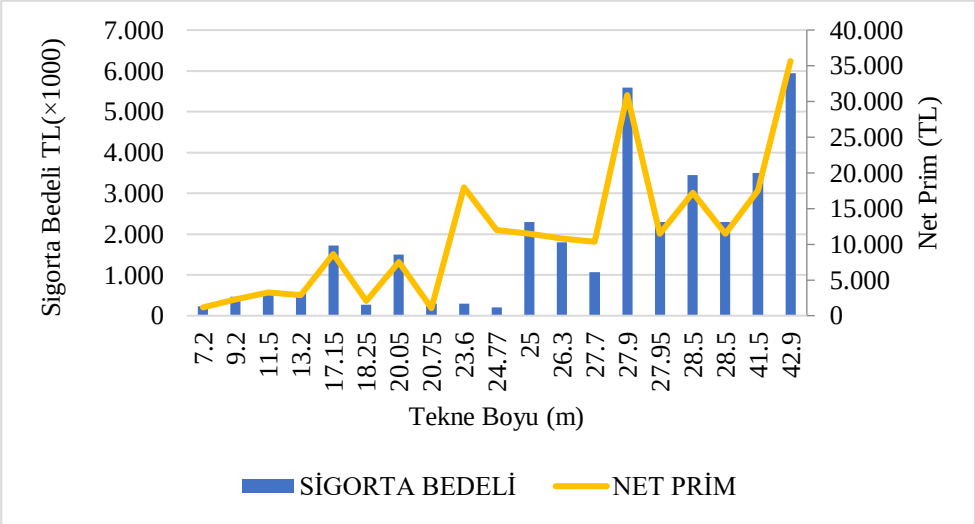
Muafiyet, sigortalı ve sigortacı arasında teklif aşamasında mutabık kalınan oran ve ya belirli bir miktar olarak sözleşmeye not edilir. Hasar oluştuktan sonra, bu oran veya miktar sigortalının üzerinde kalan kısımdır.

Hasar gerçekleştikten sonra sigortalı sözleşmede yer alan aşağıdaki belgeleri güncel olarak bulundurmalı ve ihtiyaç duyulduğu durumda sigortacıya vermekle yükümlüdür.
1) Denize Elverişlilik Belgesi: Gemiler, tahsis amacına, cinsine ve sefer bölgelerine göre; tekne, makine, genel donanım, can kurtarma, yangından korunma ve yangın söndürme durumu, seyir teçhizatı ve haberleşme sistemi, sağlık koşulları, yük ve yolcu taşıma kapasiteleri, deniz kirliliğini önleme donanımları ile diğer seyir emniyeti konuları bakımından İdare tarafından bu Yönetmelik hükümlerine göre belirli aralıklarla denetlenir (Ulaştırma ve Altyapı Bakanlığı, 2009). Bakanlık mevzuatında yer alan aynı maddenin(5) 2. Fıkrasında yer alan "Tam boyu 15 m ve üzerindeki su araçlarına formu Ek-2 de belirlenmiş olan "Su Aracı Uygunluk Belgesi" verilir. Tam boyu 15 m'nin altındaki su araçlarına ise donatanın talebi üzerine "Su Aracı Uygunluk Belgesi" verilir" ibaresi küçük ölçekli ve kabotaj sınırları dâhilinde balıkçılık yapan tekneler için alınması ve maliyet ve zaman yönetiminin balıkçı tarafından daha kolay yürütülmesini sağlayabilir.
2) Gemi Sicil Tasdiknamesi: Tonaj sınırı aranmaksızın yurt içinde inşa edilen gemiler, yatlar ve özel maksatlı, özel yapılı bütün gemiler Türk Uluslararası Gemi Siciline tescil edilir.
3) Liman Kayıt Belgesi: Bağlama kütüğü, gemi, deniz ve iç su aracının kaydedildiği, ana ve yardımcı kütüklerden oluşan elektronik kayıt sistemini ve bağlama kütüğü dosyasını ifade eder, en yakın Liman Başkanlığı'ndan çıkartılabilir.
4) Uluslararası Tonilato Belgesi: Ticari olarak faaliyette bulunan gemi, deniz ve iç su aracının belirleyici özelliklerini, teknik özelliklerini ve ölçüm sonuçlarını gösteren belgedir (Ulaştırma ve Altyapı Bakanlığı, 2014).
5) Yangın Söndürme Sistemleri veya Elemanları Muayene Test Sertifikası: Sertifikaya ihtiyacı olan geminin bulunduğu/bağlı olduğu Liman Başkanlıklarınca mevcut sistem ve ekipmanların, belli periyodlarda uygunluğunun ve çalışmasının kontrolü sonucu verilen belgedir.

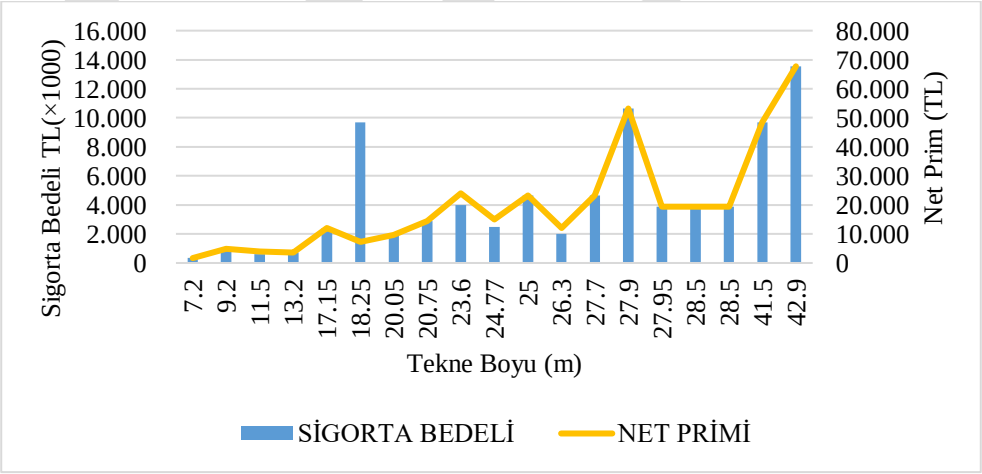
DRAFT

6) Gemi Adamı Donatımında Asgari Emniyet Belgesi: Ulusal bir belge olmak ile birlikte, (SOLAS V uyarınca) gereken yeterlilikleri yerine getirilmesini denetleyen belgedir. Gemilerin Genel Denetimi ve Belgelendirilmesi Hakkında Yönetmelik tarafından detayları yazılmıştır.
7) ISM sertifikası: IMO'nun A.741 (18) sayılı kararıyla Kasım 1993 tarihinde kabul edilmiş ve Mayıs 1994 tarihinde Denizlerde Can Emniyeti Uluslararası Sözleşmesi SOLAS 1974'ün 9. Bölümüne eklenmiş 13 maddelik bir kuraldır (Taylan, 1999).
Ancak burada önemli bir not olarak, ISM kodu alması zorunlu olan gemiler "bayrakları her ne olursa olsun uluslararası sularda çalışan bütün gemiler için zorunludur" ibaresi ile belirtilmiş, rekreasyonel balıkçılık icra eden küçük ölçekli balıkçılar için gerekli olup olmadığı muallaktır.
8) Klas-PI : IACS (Uluslararası Klas Kuruluşları Birliği) üyesi bir klas kuruluşuna veya Türk Loydu klasına sahip olması ve bu klas şartlarını poliçe süresi boyunca koruması gerekmektedir. Tıpkı ISM sertifikasında olduğu gibi balıkçı tekneleri için Klas Belgesi alma zorunluluğu "Gemi ve Su Araçlarının İnşa, Tadilat ve Bakım-Onarım Yönetmeliği" gereğince bulunmamaktadır. Bir balıkçı teknesinde bu belgenin olmayışı sigortacının ilk değerlendirme aşamasından riski reddetmesi için bir sebep olabilir.

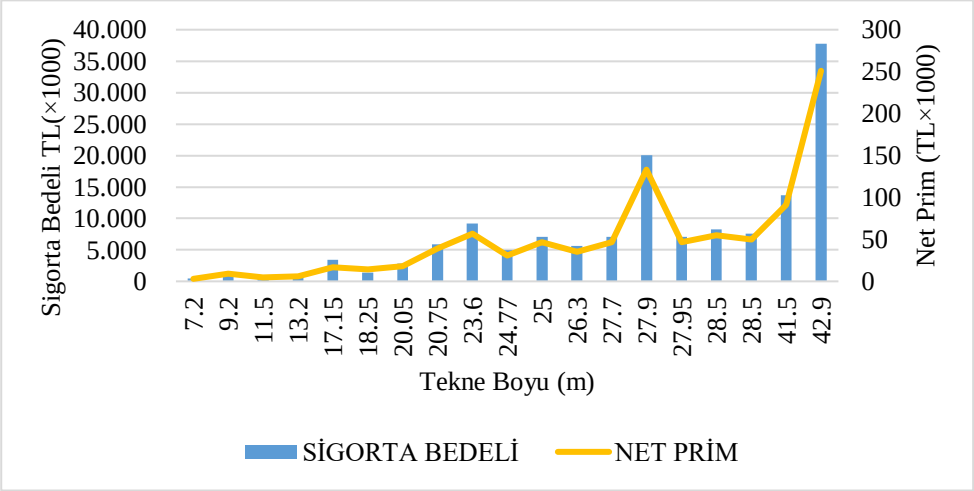
Bu bağlamda, üç yıldır düzenli olarak sigortası yapılan aynı firmaya ait 19 adet teknenin bedellere göre prim bilgileri Şekil 1, 2 ve 3'te verilmiştir.



Şekil 1. Tekne boyu ve bedeline göre net prim (2021)



Şekil 2. Tekne boyu ve bedeline göre net prim (2022)



Şekil 3. Tekne boyu ve bedeline göre net prim (2023)

Grafiklere baktığımızda, her yıl artan bir bedel ve prim oranı olduğu görülmektedir. Bunun en büyük sebebi denizcilik sektörünün dövize bağlı oluşu ve ülkemizdeki döviz kurunun dalgalı seyridir. Her geçen yıl artan döviz kuru, tekne bedellerini ve buna bağlı olarak sigorta primlerini de arttırmıştır. Bir diğer sebep ise tekne yaşlarının her geçen yıl artması ve yaşı artan teknenin sigortacı tarafından daha riskli görülmesi sebebi ile uygulanan fiyat çarpanın arttırılması. Ek olarak, endüstriyel balıkçılık, Uluslararası Çalışma Örgütü (ILO) tarafından dünyadaki en tehlikeli sektörlerden biri olarak gösterilmektedir. Bunun nedeni dünya genelinde açık deniz balıkçılığının yapılmasıdır. Ülkemizde her ne kadar açık deniz balıkçılığı yapılmıyor olsa da balıkçılık, 26.12.2012 tarihli Resmi Gazetede yayımlanarak yürürlüğe giren İş Sağlığı ve Güvenliğine İlişkin Tehlike Sınıfları Tebliği'nde tehlikeli sınıfta yer almaktadır (Abur, 2023). Bu sebeple, çoğu sigorta şirketinin risk kabul kriterlerine uymadığından dolayı balıkçı tekneleri için teminat bulmak oldukça zorlaşmaktadır.

Sigortalıların, sigorta poliçesi satın alma kararını vermelerindeki sürece bakıldığında, kredi ve fonlardan yararlanma durumunun oldukça etkili olduğunu söyleyebiliriz. Bankaların ya da fonları sağlayacak kuruluşların müşterilerinden poliçe talep etmeleri sigorta satın alınmasında ciddi bir etki yaratmaktadır.

Ming Shang (2020)'e göre, konuya özel sigorta şirketleri açısından bakıldığında, operasyonel süreçler karşılığında alınacak prim ile durum değerlendirildiğinde, maliyetin kazançtan fazla olması, risk yapısının karmaşık ve asimetrik bilgiler içermesi sebebi ile sigorta şirketlerinin bu riskleri göz ardı etmesine sebep olmaktadır. Küçük ölçekli, merkezi olmayan, yüksek riskli, düşük verimli üretim ile birlikte sınırlı ödeme kapasitesi ve karmaşık denetim ve kayıp tespit teknolojisi, sigorta şirketlerinin taahhüdünü sınırlamıştır. Kısıtlı arz ve yetersiz talep, balıkçılık sigortası piyasasında bir çelişki oluşturduğunu söylemişlerdir. Sigorta ve sigorta kapsamı hakkındaki eksik bilgiler, özellikle balıkçı ile sigortacı arasında herhangi bir köprü olmaması, sigortacılar tarafından balıkçılık faaliyetinin karmaşık ve riskli bulunması bu zamana kadar ülkemizde bu faaliyet alanına uygun bir ürün geliştirilememesine sebep olmuştur. Hali hazırda bazı sigorta şirketleri balıkçı teknelerine verdikleri teminatları "gezinti teknesi sigortası", "yat sigortası" başlıkları altında, birkaç ilave şart ile sunmaktadır. Ayrıca, ülkemiz genelinde de özellikle artan yakıt maliyetleri, gıda enflasyonu sebebi ile artan giderler arasına sigorta giderinin de eklenmesi imkansız hale gelmiştir.

Karadeniz'de yer alan balıkçıların eğitim durumlarının diğer bölgelere göre oldukça yüksek olduğu dikkat çekmektedir. Bunun yanı sıra üniversite mezunlarının küçük balıkçılığa yönelmeleri ülkemizde yaşanan işsizlik problemine de işaret etmektedir (Erdoğan-Sağlam ve ark., 2016) Akdeniz'de yer alan balıkçıların eğitim durumlarının diğer bölgelere göre daha düşük oranlarda olduğu görülmüştür. Özellikle Akdeniz bölgesinin en sosyal ve gelişmiş illerinden olan Hatay, Mersin ve Antalya'da balıkçılar arasında nadir de olsa üniversite mezunlarının çalıştığı dikkati çekmektedir. Yüksekokul mezunlarının bu illerde turizm sektörüne yönelmesi balıkçılığı iş alanı olarak görmelerini ikinci plana atmaktadır (Karadal, 2014). Gökçeada balıkçıların eğitim düzeyleri incelendiğinde ilkokul mezunu ile üniversite mezunu arasında değiştiği tespit edilmiştir. En yüksek oranın %54,2'ile ilköğretim mezunu oldukları ortaya konmuştur. Bu oran yapılan benzer çalışmalarda Ege Bölgesi'ndeki balıkçıların %2,30'u okur-yazar, %62,82'si ilkokul, %17,24'ü ortaokul, %10,73'ü lise ve %1,53'ü ise üniversite mezunu olduğu rapor edilmiştir (Çeliker ve ark., 2008). Türkiye

genelinde yapılan bu çalışmalardan anlaşıldığı üzere balıkçılık faaliyeti ile uğraşan kişilerin sosyo-ekonomik seviyesindeki yetersizlik sebebi ile balıkçılık idaresini oluşturan kurum ve kuruluşların balıkçılık faaliyeti ile ilgilenen tüm paydaşların hakkını gözeterek ve balıkçılık faaliyetinin devamlılığı ve sürdürülebilirliğini sağlamak üzere adımlar atması gerekmektedir. Sonuçta,

Balıkçılığa kayıtlı tüm teknelerin ve lisans sahibi tüm kişilerin balıkçılık faaliyeti yapılırken uygulanabilecek sigortalar hakkında bilgilendirilmesini sağlamak,
Ülkemizde 2005 yılında kurulmuş TARSİM'in (Tarım Sigortaları Havuzu A.Ş.) sağlamış olduğu mevcut sigortalara ek olarak tarım ve gıda üretiminin bir parçası olan avcılık yolu ile gerçekleştirilen balık üretimine yönelik sigorta poliçesi dizaynı ve mevcut poliçelere uygulandığı üzere %50 devlet desteği sağlamak,
2017 yılından itibaren kullanılmaya başlanan 12 m ve üzeri Balıkçı Gemileri İzleme Sistemi (BAGİS) yardımı ile sigortalı – sigortasız balıkçı teknelerinin takibini sağlamak, böylece balıkçı teknelerine özel bir prim havuz oluşumunu sağlamak,
Üniversitelerin ilgili bölümlerinde (Su Ürünleri Mühendisliği, Balıkçılık Teknolojisi Mühendisliği), balıkçılık sektöründe uygulanabilecek sigortalar konusu ile ilgili ders konularının müfredata eklenmesi, eğitimin niteliğinin ve sektör ihtiyaçlarına yönelik uzman yetiştirilmesini sağlamak gerekli ve önemlidir.

KAYNAKÇA

Abur, S.T., 2023, Balıkçı Gemilerinde İş Sağlığı ve Güvenliği, Yüksek Lisans Tezi, Katip Çelebi Üniversitesi, 41 s.

Çeliker, S. A., Korkmaz, Ş.A., Demir, A., Gül, U., Dönmez, D., Özdemir, İ., Kalkanlar, Ş., 2008, Ege Bölgesi'nde Su Ürünleri Avcılığı Yapan İşletmelerin Sosyo- Ekonomik Analizi Projesi. Tarımsal Ekonomi Araştırma Enstitüsü. Ankara. 107 s.

Erdoğan-Sağlam, N., Özbek G., Düzgüneş E., 2016, Doğu Karadeniz Bölgesi'nde Deniz Balıkçılarının Sosyo-Ekonomik Yapısı, Gaziosmanpaşa Üniversitesi Ziraat Fakültesi Dergisi, 33(3), 259-270.

FAO, 2022, The state of world fishries and aquaculture 2022. Towards blue transformation. Rome. 236 s. https://www.fao.org/documents/card/en/c/cc0461en

Institute Time Clauses Hulls, 1995, Joint Hull Committee

Karadal E., 2014 Akdeniz Bölgesi (Türkiye) Sahil Şeridi Deniz Balıkçılığının Sosyo-Ekonomik Durumu, Ordu Üniversitesi, Yüksek Lisans Tezi, 65s.

Shang M., Zhao X., Zheng H., 2020, Chinese policy on fishery insurance: Evolution, characteristics and challenges. Marine Policy Volume 119,104099.

Taylan M., 1999, Güvenli Gemi İşletmeciliği ve ISM Kodu. Gemi İnşaatı ve Deniz Teknolojisi Teknik Kongresi, İstanbul. Bildiriler Kitabı 347 s.

Ulaştırma ve Altyapı Bakanlığı, 2014, Bağlama Kütüğü Uygulama Yönetmeliği. https://www.mevzuat.gov.tr/mevzuat?MevzuatNo=20068&MevzuatTur=7&MevzuatTertip=5

Ulaştırma ve Altyapı Bakanlığı, 2009, Gemilerin Teknik Yönetmeliği. https://www.mevzuat.gov.tr/mevzuat?MevzuatNo=13556&MevzuatTur=7&MevzuatTertip=5

ORAL PRESENTATION

Üzümsü meyvelerin antioksidan kapasitesinin Diabetes Mellitus hastalığına etkisi

Gökçe Çakmak Kafadar[1*] (*ORCID: https://orcid.org/0000-0002-0364-1546*)

[*1]Kırklareli Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Kırklareli, Türkiye

*Sorumlu yazar e-mail: gokceckmk@gmail.com

Özet

İnsan sağlığı üzerindeki olumlu etkilerinin ortaya çıkmasıyla birlikte üzümsü meyvelerin önemi giderek artmaktadır. Meyve ve sebzeler içerisinde fenolik madde ve özellikle antosiyanince zengin olan üzümsü meyveler yüksek antioksidan kapasiteye sahiptir.Birçok bitki türü (nar, böğürtlen, yaban mersini, çilek vb.) içerdikleri fenolik bileşikler ve bu bileşiklere bağlı olarak sahip oldukları antioksidatif özelliklerden dolayı fonksiyonel gıdalar olarak değerlendirilmektedir.Üzümsü meyvelerin ise yüksek antioksidan kapasiteleri, askorbik asitten çok fenolik maddelerden, antosiyaninlerden, fenolik asit ve flavonoidlerden kaynaklanmaktadır. Antosiyanin alımının, Tip 2 diyabet modelinde kan şekeri seviyelerinin yükselmesini engellediği ve insülin duyarlılığını artırdığı tespit edilmiştir.Meyveler içerisinde özellikle üzümsü meyvelerin doğal antioksidanlar bakımından zengin olduğu ve yüksek antioksidan kapasitesi ile antosiyanin miktarına sahip oldukları bilinmektedir. Biyolojik olarak aktif fitokimyasallar, özellikle antosiyaninler ve proantosiyanidinler açısından zengin meyveler, kan şekeri seviyelerindeki artışı baskılayabilir, diyabet ve diğer metabolik bozuklukları iyileştirebilir. Meyvelerdeki antioksidanlar özellikle büyük ölçüde antosiyaninlerden kaynaklanmaktadır. Siyah, koyu kırmızı ve mavi renkler içeren meyvelerin antioksidan değerleri çok daha yüksektir. Ayrıca antosiyaninlerin adipozitler üzerinde etkili olabileceği ve adipozitokinler sergilediğini gösteren çeşitli çalışmalarda ise, antosiyanin alımının, tip 2 diyabet modelinde kan şeker tüketiminin yükselmesini engellediği ve insülin salgısını artırdığı tespit edilmiştir. Meyve ve sebzelerde antioksidanlar, insülin duyarlılığını arttırmak ve diyabetlere karşı koruma sağlamak için bireylere takviye denemeleri hipotez edilmiş ve bu çalışmalarda antioksidanların diyabet mellitusa karşı koruyucu etkisi olabileceği ileri sürülmektedir. Üzümsü meyvelerin içindeki antosiyaninlerin, temel fonksiyonlarından olan insülin direncinin gelişiminde, adiposit fonksiyonun etkilenerek postprandiyal glisemiyi tetiklediği gözlemlenmiştir. Ayrıca, adipositokinleri salgılanmasının artması hedef hücrelerin insülin duyarlılığını geliştirebileceği de tespit edilmiştir. Meyve ve sebzelerin sahip olduğu lif, magnezyum, folat, antioksidan içeriklerinin (C vitamini, E vitamini, karotenoidler) işlevsel yönlerinden dolayı diyabet gelişimi üzerinde belirli bir koruyucu etkileri olduğu ortaya konmuştur.

Anahtar Kelimeler: Üzümsü meyveler, Antioksidan, Antosiyanin, Diabetes Mellitus,

Effect of antıoxıdant capasıty of some berry fruıts on dıabetes mellıtus

Abstract

With the emergence of positive effects on human health, the importance of berry fruits is gradually increasing. Grape fruits, which are rich in phenolic substances and especially anthocyanins among fruits and vegetables, have high antioxidant capacity. Many plant species (pomegranate, blackberry, blueberry, strawberry, etc.) are considered as functional foods due to the phenolic compounds they contain and the antioxidant properties they have depending on these compounds. On the other hand, the high antioxidant capacity of berries is due to phenolic substances, anthocyanins, phenolic acids and flavonoids rather than ascorbic acid. Anthocyanin intake has been found to prevent the rise of blood sugar levels and improve insulin sensitivity in a Type 2 diabetes model. It is known that among fruits, especially berry fruits are rich in natural antioxidants and have high antioxidant capacity and anthocyanin content. Fruits rich in biologically active phytochemicals, especially anthocyanins and proanthocyanidins, can suppress the rise in blood sugar levels, improve diabetes and other metabolic disorders. The antioxidants in fruits are mainly due to anthocyanins. The antioxidant values of fruits containing black, dark red and blue colors are much higher. In addition, in various studies showing that anthocyanins can be effective on adipocytes and exhibit adipositokines, it has been determined that

anthocyanin intake prevents the increase in blood sugar consumption and increases insulin secretion in a type 2 diabetes model. Antioxidants in fruits and vegetables have been hypothesized to be supplemented with individuals to increase insulin sensitivity and protect against diabetes, and these studies suggest that antioxidants may have a protective effect against diabetes mellitus. It has been observed that anthocyanins in berries trigger postprandial glycemia by affecting adipocyte function in the development of insulin resistance, which is one of its basic functions. It has also been found that increased secretion of adipocytokines can improve the insulin sensitivity of target cells. It has been revealed that fruits and vegetables have a certain protective effect on the development of diabetes due to their functional aspects of fiber, magnesium, folate, antioxidant contents (vitamin C, vitamin E, carotenoids).

Keywords: Berries, Antioxidant, Anthocyanin, Diabetes Mellitus.

GİRİŞ

Meyveler, besin ve biyoaktif fitokimyasallar kaynağı olarak bilinen insan diyetinin önemli bileşenleridir. Antioksidan aktiviteleri ile bilinen C, A ve E vitaminlerini içerirler (Sławińska ve ark., 2023). birçok bitki türü (nar, böğürtlen, yaban mersini, çilek vb.) içerdikleri fenolik bileşikler ve bu bileşiklere bağlı olarak sahip oldukları antioksidatif özelliklerden dolayı fonksiyonel gıdalar olarak değerlendirilmektedir (İnan ve Kaya., 2015). Gıdalarda, özellikle sebzelerde bulunan antioksidanlar fenolik bileşikler (fenolik asitler ve flavonoidler), karotenoidler, tokoferol ve askorbik asittir. Bu bileşikler insan sağlığı için önemli koruyucu maddelerdir (Islam ve ak. 2017).Bütün bitki metabolizmalarında, sekonder metabolit olarak bulunan ve bitkilerin kendilerini bazı zararlılara karşı korumada rolleri olduğu bilinen çok sayıda farklı nitelik ve miktarlarda çeşitli fenolik bileşikler bulunmaktadır. Fenolik birleşikler, meyve ve sebzelerin tat, aroma, renk gibi meyve özelikleri üzerinde direkt etkileri olan ikincil metabolitlerdir. Antioksidanlar ise serbest radikallerin zararlı etkilerinden koruyan maddelerdir. Antioksidan maddeler bu serbest radikaleri tutarak ya da etkisiz hale getirerek oksidasyonu neden olduğu zararlanmaları hücresel bazda engelemekte ve hastalıkları oluşmasını önlemektedir (Şensoy., 2018).

Üzümsü meyveler grubunda yer alan çilek, ahududu, böğürtlen, frenküzümü, bektaşiüzümü, maviyemiş, turnayemişi gibi türler dünyada çok sevilen ve yaygın bir şekilde tüketilen meyve türleri arasında yer almaktadır. Özellikle bileşimlerinde yer alan vitamin, mineral madde ve fenolik maddelerin yüksek oranda bulunuşu bu türlerin önemini daha da artırmaktadır (Çelik., 2012). Üzümsü meyvelerin ise yüksek antioksidan kapasiteleri, askorbik asitten çok fenolik maddelerden, antosiyaninlerden, fenolik asit ve flavonoidlerden kaynaklanmaktadır (Eke., 2017). Meyveler içerisinde özellikle üzümsü meyvelerin doğal antioksidanlar bakımından zengin olduğu ve yüksek antioksidan kapasitesi ile antosiyanin miktarına sahip oldukları bilinmektedir (Engin ve ark., 2018). Antioksidan kapasite üzerinde en etkili bileşenlerin ise askorbik asit, antosiyaninler ve ellagitanninler olduğu belirtilmektedir (Erten ve Yılmaz., 2019). Üzümsü meyvelerde çok yoğun bir şekilde bulunan fenollerden ellagic asit (antikanserojen), flavon ve flavonoidlerden (antioksidan) antosiyanin, quercetin, kaempheol, myricetin en önemli "nutraceutical" veya "functional foods" değerine sahip bitkisel kimyasallardır (Pehluvan ve Güleryüz., 2004).

BULGULAR VE TARTIŞMA

Üzümsü meyvelerin tanımlanması

Üzümsü meyveler genel anlamda, üzüme benzeyen meyve türleri için kullanılmaktadır. Bu meyvelerin etli, sulu, yumuşak ve hoş kokulu olmaları ve aynı zamanda antioksidan kapasitelerinin diğer meyvelere göre çok daha yüksek olması en önemli özellikleridir. Üzümsü meyveler denildiği zaman daha çok üzüm (Vitis), çilek (Fragaria), ahududu ve böğürtlen (Rubus,) frenk üzümü ve bektaşi üzümü (Ribes), yaban mersini, kırmızı noktalı yaban mersini, kültür yaban mersini, bataklık yaban mersini (Vaccinium), kuşburnu (Rosa), kadıntuzluğu (Berberis), çakal eriği (Prunus) gibi cinsler ve bunlara bağlı türler akla gelmektedir (Özarda., 2009).

Üzümsü meyveler olarak da bilinen berry sınıfı fenolik bileşikler, organik asitler, taninler, antosiyaninler ve flavonoidler gibi biyoaktif bileşiklerin geniş bir çeşitliliğini içermesi ile bilinir (Çağlar ve Demirci., 2017).

Üzümsü sınıfının antioksidan içeriği

Berry sınıfı genel olarak dut, çilek, böğürtlen, ahududu, frenk üzümü, bektaşi üzümü, yaban mersini, mürver meyvesi gibi türleri içeren meyvelerin antioksidan kapasiteleri oldukça yüksektir. Böğürtlen (Rubus fructicosus), ahududu (Rubus ideaus L.), frenk Üzümü (Ribes sativum), kızılcık (Cornus mas cv. Vermio), çilek, siyah üzüm yüksek miktarlarda fenolik bileşik ve antosiyanin içeren meyvelerdir (Nizamlıoğlu ve Nas., 2010).

Çilek: Çilek meyvesi, doğal antioksidanların yanı sıra; vitamin, mineral, antosiyanin, flavonoidler ve fenolik asitler açısından da zengindir. Çileğe kırmızı rengini veren antosiyaninin, insan plazmasındaki antioksidan kapasiteyi arttırarak, düşük yoğunluklu lipoproteinler üzerindeki etkisi oldukça yüksektir. Diğer yandan, çileklerde bulunan yüksek miktarda askorbik asit, reaktif oksijen radikalleri üzerinde koruyucu özellik sağlamaktadır. Askorbik asidin, antosiyaninlerin ve toplam fenolik madde içeriğinin, nörotoksisiteye neden olan oksidatif stresi azalttığı bilinmektedir. Sağlam epidemiyolojik kanıtlar, çilek tüketiminin hiperlipidemi, insülin direnci, diyabet ve KVH riski/insidansı ile ters ilişkili olduğunu göstermiştir(Erdoğan ve ark., 2013).

Ahududu ve Böğürtlen: Ahududu, quercetin ve kaempheol flavonoidlerinin iyi bir kaynağıdır. Quercetin hem antikanserojen hem de antioksidan etkiye sahiptir. Quercetin ve Kaempferol gibi antioksidanlar, insan vücudundaki hücrelerde yıkım meydana getiren lipid peroksidasyonunu önleyebilmektedirler (Pehluvan ve Güleryüz., 2004). Ahududu ve böğürtlenler besin değeri bakımından oldukça önemli, sağlık için vazgeçilmez değerde yüksek oranlarda mineral maddeler ve vitaminler içermektedir. Az miktarda A, B, C vitaminleri ve diyet için lifli (çözülen veya çözülmeyen) yapıları çok büyük değere sahiptir (Çelik., 2012). Rubus cinsine ait bir meyve türü olan böğürtlen, polifenoller, özellikle antosiyaninler ve daha yüksek antioksidan aktivitesine katkıda bulunan diğer antioksidanlar açısından zengindir (Gowd ve ark.,2018). Böğürtlen meyvesi, antosiyanin-3- O -glukozit ve klorojenik asit başta olmak üzere yaklaşık 15 fenolik madde içerir (Li ve ark., 2022).

Yaban Mersini: Yaban mersini yüksek antioksidan kapasiteye sahip olup, bol miktarda fenolik bileşen içermektedir. Kanser, kardio ve serebrovasküler hastalıklar ve diyabet gibi bazı kronik hastalıklara karşı koruyucu etkisi yüksek antioksidan kapasitesine bağlanmaktadır. Küçük taneli birçok meyve üzerinde yapılan analizlerin sonuçlarına göre; yabani yaban mersinlerinin belirlenen oksijen radikallerini absorbe etme kapasitesiyle en yüksek antioksidan etkiye sahip oldukları saptanmıştır (Ceylan ve ark., 2017). Yaban mersini içinde bulunan en önemli polifenoller antosiyaninlerdir. Araştırmalarda yaban mersininin yüksek ORAC (Oxygen radical absorbance capacity) değerine sahip olduğu bulunmuştur. (Ayvaz., 2021). Yaban mersini antosiyaninlerin yanısıra antioksidan aktiviteye katkı sağlayan klorogenik asit, kuersetin, kamferol, mirisetin, prosiyanidin, kateşin, epikateşin, resveratrol gibi fenolik bileşikler ile vitamin C açısından da iyi bir kaynaktır. Kılcal damarların tıkanmasına neden olan düşük yoğunluktaki lipidlerin vücuttan atılması, kan şekerinin düzenlenmesi, kalp krizi riskinin azaltılması, içerdiği lif ile bağırsak metabolizmasının düzenlenmesi üzerine olan etkilerinin yanı sıra kanı temizlemek, kan kolestrolünü düşürmek gibi faydaları da bulunmaktadır (Yıldız., 2012).

Kızılcık: Kızılcık meyvesinin, zengin bir C vitamini ve polifenol kaynağı olduğu bilinmektedir. Hem meyvelerinde hem de yapraklarında önemli miktarda flavanoid, antosiyanin ve iridoid olduğu bildirilmiştir (Çelik., 2022). Biyoaktif bileşenler sayesinde antibakteriyel, antidiyabetik, antienflamatuvar etki göstererek bağırsak, mide kalp ve karaciğer sağlığının korunmasına yardımcı olmaktadır (Durgut ve ark., 2022). Başta meyveye rengini veren antosiyanin olmak üzere, fenolik bileşiklerden zengindir. Kızılcıkta baskın antosiyanin grupları siyanidin rutinosit ve pelargonidin galaktosittir. Bu bileşiklerin miktarı bitki genotipine, yetiştiriciliğine ve meyvenin olgunluğuna bağlıdır. Kızılcık meyvesi, çilek, limon gibi meyvelerden önemli ölçüde daha fazla C vitamini içermektedir. Kızılcık meyvesi, yüksek potasyum (K) ve magnezyum (Mg) içeriğine sahiptir (Çelik., 2022). Kızılcık içerisindeki antosiyanidin ve ursolik asitlerin aşırı kalorili beslenmenin devam ettiği yağlanma sürecinde yağ dokusunun oluşumunu %24 azalttığı, karaciğerde yağ birikiminin azaldığı ve trigliserol oranını düşürdüğü yönündedir (Bayoğlu., 2021). Kızılcık, Escherichia coli'nin idrar yoluna yapışmasını engelleyen proantosiyanidinler bakımından yüksek olan az sayıdaki meyveden biridir. Kızılcık kardiyovasküler hastalıkları tedavi etmeye, lipit profilini iyileştirmeye, düşük yoğunluklu lipoproteinleri azaltarak ateroskleroz olasılığını en aza indirmeye, kan basıncını düşürmeye ve metabolik sendromu önlemeye yardımcı olur. Kızılcık tüketimi, tip 2 diyabet riskini azaltır (Nemzer ve ark., 2022).

Goji Berry: Goji berry'de bulunan flavonoidler (rutin, myricetin, quercetin, kaempferol) ve fenolik asitler (kafeik asit, klorojenik asit ve kumarik asit), Goji berry'nin antioksidan ve antimikrobiyal aktivite sergilenmesine neden olan ana bileşenlerdir (Kalkan., 2019). Goji meyveleri karotenoidler (beta-karoten , lutein, likopen, zeaksantin, zeaksantin dipalmitat), polisakkaritler (posanın %30'unu oluşturur), vitaminler (askorbik asit glukopiranosil askorbik asit ve tokoferol), yağ asitleri, betain ve peptidoglikanlar içerir (Islam ve ak. 2017). Çalışmalar, L. barbarum'un yaşlanma, nöroproteksiyon, genel iyilik hali, yorgunluk/dayanıklılık, metabolizma/enerji harcaması, diyabetiklerde glukoz kontrolü, glokom, anti-tümör aktivite ve hücre koruma üzerindeki etkilerini göstermektedir (Amagase ve Farnsworth., 2011). Goji meyveleri, en güçlü antioksidanlardan biri olarak bilinen zeaksantin dipalmitatın bilinen en zengin doğal kaynağıdır ve anti-enflamatuar etkileri iyi tanımlanmıştır (Ramon ve ark., 2019).

Aronya: Antioksidan özelliği en yüksek ve süper meyve olarak adlandırılan aronya meyvesinin ORAC değerinin 16.062 gibi yüksek bir düzeyde olduğu bildirilmiştir (Anonim, 2021). Antioksidan açısında oldukça zengindir. Ayrıca vitamin ve mineral kaynağıdır. B12 ve D vitamini hariç tüm vitaminleri içermektedir. Bu yüzden aronya ya 'Süper Meyve' denilmektedir. Bilim adamları, aronya da kafeik asit, siyanidin-3 galaktozid, delfinidin, epikateşin, malvidin ve birçok özel maddeler saptamışlardır (Sosnowska, 2016, Bolling, 2017).
Bu özel maddeler, anti-bakteriyel, anti-viral ve anti-diyabetiktir. Aronya kan şekerini düşürür ve vücudun kendi doğal üretimi olan insülinin artmasına sağlar. Aronya içinde bulunan bazı bileşikler doğal kanser savaşçılarıdır (Özder., 2021).

Berry türü meyvelerin Diabetes Mellitus'a etkisi

Biyolojik olarak aktif fitokimyasallar, özellikle antosiyaninler ve proantosiyanidinler açısından zengin meyveler, kan şekeri seviyelerindeki artışı baskılayabilir, diyabet ve diğer metabolik bozuklukları iyileştirebilir. Postprandial glikozu azaltmak için varsayılan mekanizma, a-amilaz ve a-glukosidaz aktivitesini inhibe ederek glikoz emilimini sınırlamaktır (Golovinskaia ve Wang., 2021).
Goji berry içerisindeki sahip olduğu polifenolik bileşikler antioksidatif özellikler sayesinde düşük yoğunluklu lipoprotein (LDL) kolesterol fraksiyonunun ve trigliseritlerin kan seviyelerini düşürür ve metabolik hastalık (örn. diyabet, obezite) riskini en aza indirir. Yüksek biyolojik değeri nedeniyle kan basıncını düşürmek ve kandaki glikoz seviyesini düşürmek için kardiyovasküler hastalıkların tedavisinde uygulanır (Oszm iański ve Lachowicz., 2016).
Böğürtlen özleri ile kırmızı ahududu özleri karşılaştırıldığında; ahududu α-amilazın inhibe edilmesinde en etkili olanlardır. Bazı çalışmalar, antosiyaninlerin adipositlerin işlevini iyileştirdiğini ve insülin duyarlılığını artırdığını göstermektedir (Golovinskaia ve Wang., 2021).
Çilek, glukozidaz ve anjiyotensin-1-dönüştürücü enzimleri inhibe eder ve a-amilaz için daha az belirgin bir inhibe etme potansiyeline sahiptir (da Silva ve ark., 2010).
Frenk üzümü özleri, tip 2 diyabetik (T2D) farelerde ve insanlarda kan şekerini azaltabilir ve glikoz toleransını geliştirebilir.
Yaban mersini antosiyaninlerinin insülin duyarlılığını ve hiperglisemiyi azalttığı gösterilmiştir. Yaban mersini tozu ile takviye edilmiş diyet, farelerde glikoz toleransını artırır, obez farelerde glikoz metabolizması belirteçlerini normalleştirir ve insanlarda insülin duyarlılığını artırır (Elks ve ark., 2015). Ayrıca, antosiyaninlerin, insülin sekresyonunun indüklenmesinden sorumlu pankreatik hücrelerle birleşen peptit-1 gibi glukagon üretimini indüklediği doğrulanmıştır. (Golovinskaia ve Wang., 2021).
Kızılcık (Cornus mas L.; CM), başta meyveye rengini veren antosiyanin olmak üzere fenolik bileşiklerden zengindir. Antosiyaninlerin, α-glukozidaz enzim aktivitesini azalttığı ve inhibe ettiği gösterilmiştir. Alfa-glukozidaz, karbonhidratların hidrolizinde yer alan bir sindirim enzimidir. Bu enzimin inhibisyonunun karbonhidratların hidrolizini önlediği ve kan glikozu düzeylerini düşürdüğü görülmektedir. Antosiyanin içeriği zengin CM, glikozun azalmasına yol açan reseptörler aracılığıyla insülin etkisini iyileştiren G-protein reseptörlerini de aktive edici etkiye sahiptir (Çelik., 2022). Kızılcık flavonoidleri, glikozun bağırsak emilimini geciktirir ve glisemik yanıtı iyileştirir. Bazı araştırmalar, kızılcık ürünlerinin açlık glisemisini azaltarak, homeostaz modeli değerlendirmesi tahmini insülin direncini iyileştirerek, insülin duyarlılığını artırarak ve telafi edici insülin sekresyonunu önleyerek glikoz homeostazını destekleyebileceğini göstermektedir. Doğal bir polifenol ve lif kaynağı olan kızılcık tüketimi, T2D'li hastalarda daha olumlu bir glisemik tepki sağlayacaktır. Kızılcık, gastrik glikoz emilim oranındaki azalma ile ilgili olan çözünür lifler polidekstroz ve β-glukan içerir (Golovinskaia ve Wang., 2021).
Üzümsü meyvelerin içindeki antosiyaninlerin, temel fonksiyonlarından olan insülin direncinin gelişiminde, adiposit fonksiyonun etkilenerek postprandiyal glisemiyi tetiklediği gözlemlenmiştir. Ayrıca, adipositokinleri salgılanmasının artması hedef hücrelerin insülin duyarlılığını geliştirebileceği de tespit edilmiştir. Serum adiponektin protein seviyeleri sistemik insülin duyarlılığı ile ilişkilidir ve insüline dirençli, diyabetik ve obez kişilerde azaldığı gözlemlenmiştir. Obezite veya diyabette, glikoz taşıyıcı 4'ün (GLUT4) ekspresyonu ile adipositler azalmaktadır; azalmaya RBP4 ekspresyonunda ve kan sekresyonunda bir artış eşlik eder. Artış, iskelet kasında insülin sinyalinin bozulmasına neden olur ve karaciğerde glikoz üretimini uyarır. Bu değişiklikler kanda yüksek glikoz konsantrasyonlarına yol açar. Bu nedenle, adiposit GLUT4-RBP4 sisteminin düzensizliği tip 2 diyabet ile güçlü bir şekilde ilişkilidir. RBP4'ün düşürülmesi tip 2 diyabetin önlenmesi ve tedavisi için potansiyel olarak yeni bir hedeftir (Afacan ve Sönmezdağ., 2020).
Li ve arkadaşlarının 2015 yılında antosiyaninlerin tip 2 diyabetli hastalarda dislipidemi, oksidatif durum ve insülin duyarlılığı üzerindeki etkilerini araştırdığı çalışmada antosiyanin takviyesinin, tip 2 diyabetli hastalarda dislipidemiyi iyileştirerek, antioksidan kapasiteyi artırarak ve insülin direncini önleyerek yararlı metabolik etkiler gösterdiğini göstermektedir (Li ve ark., 2015).

Kalt ve arkadaşlarının yürüttüğü; obez, insüline dirençli yetişkinler üzerinde yapılan plasebo kontrollü bir çalışmada, insülin duyarlılığı 6 haftalık yaban mersini alımından sonra daha yüksek olduğu saptanmıştır. Yabanmersini ve siyah frenk üzümünden elde edilen antosiyanin özünün (günde 80 mg), plaseboya kıyasla 58 T2DM hastasında insülin duyarlılığını (HOMA-IR), plazma lipid profillerini iyileştirdiği ve oksidatif stresin plazma belirteçlerini azalttığı gözlemlenmiştir (Kalt ve ark., 2020).
Paralel, çift kör, randomize, kontrollü bir çalışmada, Curtis ve meslektaşları 45 kişiden oluşan bir grupta, enerjisi yoğun bir gıdayla (900 kcal; 500 g milkshake) birlikte tüketilen dondurularak kurutulmuş bir yaban mersininin (26 g, 150 g taze uzun boylu yaban mersine eşdeğer) postprandiyal kardiyometabolik tepki üzerindeki etkisini araştırdı. Yaban mersini yoğun gıda alımı, yemek sonrası glikoz ve insülin tepkisini ve toplam kolesterolü azaltırken, HDL-C, Apolipoprotein AI (APO-A1), yüksek yoğunluklu lipoprotein parçacık sayısını artırdığı gözlemlenmiştir (Venturi ve ark., 2023).
Lycium barbarum meyvelerinden elde edilen polisakkaritlerin antidiyabetik etkisi araştırılmıştır. Diyabetik farelere Lycium barbarum polisakkaritleri 6 hafta 200 mg/kg/gün dozda oral olarak verilmiş ve çalışma sonunda farelerin açlık kan şekeri seviyelerinde (%13.51 azalma) ve glikozillenmiş hemoglobin fonksiyonunda iyileşme gözlenmiştir. Ayrıca bu çalışma, LBP'lerin bağırsak mikrobiyotasını ve bağırsak bariyerini değiştirerek anti-diyabetik etki sağlayabileceğini göstermiştir. Lycium barbarum L.'den elde edilen su ekstraktının hipoglisemik etkilerini ve etki mekanizmasının değerlendirildiği bir çalışmada, Lycium barbarum L. özütünün diyabetik sıçanlarda kan şekeri ve lipit seviyelerini belirgin şekilde modüle edebileceğinin yanı sıra karaciğer, böbrek ve pankreastaki hasarları onarabileceği gösterilmiştir (Alp., 2023).
Sağlıklı yetişkinlerde, üzümsü meyve pürelerinin glikoz metabolizması, insülin direnci ve glisemik yanıtın diğer aracılar üzerindeki etkisini değerlendiren çalışmada; mor meyvelerin olduğu öğünün tüketilmesinden 15 ve 30 dakika sonra plazma glukoz konsantrasyonları, kontrol öğününün tüketildiği değerlere göre önemli ölçüde daha düşük tespit edilmiştir. Yapılan diğer çalışmada, mor meyveleri içeren öğünün, kontrol yemeğine kıyasla daha düşük ve uzun süreli insülin tepkisini uyardığı ve yüksek insülin tepkisi belirtisi görülmediği gözlemlenmiştir [36]. Bu iki çalışma, mor meyvelerin postprandiyal hiperglisemi üzerindeki etkisini göstermesine rağmen, sonuçların yorumlanması diğer faktörlerin de göz önünde tutulması gerekmektedir. Kontrol öğününün ve dut öğününün lif içeriği eşleşmemektedir. Başka bir çalışmada ise antosiyanin alımının, obez fare modellemesinde, yüksek yağlı bir diyetin neden olduğu kan şekeri seviyesinin yükselmesini engelleyebileceğini bildirmiştir. Sasaki ve ark. Yapmış olduğu tip 2 diyabet model çalışmasında, antosiyanin alımının kan şekeri seviyelerinin yükselmesini engellediği ve insülin duyarlılığını arttırdığı tespit edilmiştir. Bu etki, yaban mersini özündeki çeşitli C3G bileşiği ile ilişkilendirilmiştir (Afacan ve Sönmezdağ, 2020).
Yapılan bir çalışmada erken hafıza değişikliği geçiren yaşları ortalama 76 olan 9 yaşlı bireye 12 hafta boyunca 6-9 ml/kg yaban mersini suyunun etkisi araştırılmıştır. Sonuç olarak azalmış depresyon belirtileri (p=0.08) ve düşük glikoz düzeylerini (p=0.10) düşündüren eğilimler tespit edilmiştir (Özkan., 2020).

SONUÇ

Meyve ve sebzelerde bulunan mineraller, fitokimyasallar ve diğer biyoaktif bileşenler kronik hastalıkların önlenmesinde önemli rol oynamaktadırlar (Bulantekin ve ark., 2020).Nizamlıoğlu ve Nas (2010)' ın bildirdiğine göre, genel olarak frenküzümü, bektaşiüzümü, dut, çilek, böğürtlen, ahududu, maviyemiş, mürver yemişi gibi türleri içeren meyvelerin antioksidan kapasiteleri oldukça yüksektir (Çelik., 20212). Doğal halde yüksek oranlarda içerdiği; C vitamini, antioksidanlar, flavon ve flavonoidler (fenolik bileşenler) nedeniyle fonksiyonel özellik kazanan çilek ve diğer üzümsü meyveler, insan sağlığı açısından başka meyve gruplarıyla karşılaştırılamayacak kadar faydalı bir meyve grubudur (Erdoğan ve ark., 2013) Üzümsü meyveler üzerinde yapılan çalışmalarda adı geçen bitkisel kimyasallar bakımından bu grupta bulunan böğürtlen, ahududu ve maviyemişin en zengin meyveler arasında yer aldığı belirlenmiştir (Eke., 2017).
Antosiyaninler flavonoid grubunun bir bölümünü oluşturmaktadırlar. Özellikle böğürtlenlere kendilerine has karakteristik mavi rengi vermektedirler. Meyvelerdeki antioksidanlar özellikle büyük ölçüde antosiyaninlerden kaynaklanmaktadır. Siyah, koyu kırmızı ve mavi renkler içeren meyvelerin antioksidan değerleri çok daha yüksektir (Pehluvan ve Güleryüz., 2004). Ayrıca antosiyaninlerin adipozitler üzerinde etkili olabileceği ve adipozitokinler sergilediğini gösteren çeşitli çalışmalarda ise, antosiyanin alımının, tip 2 diyabet modelinde kan şeker tüketiminin yükselmesini engellediği ve insülin salgısını artırdığı tespit edilmiştir (Afacan ve Sönmezdağ., 2020). Yaban mersini, üzüm ve elma gibi spesifik meyvelerin tüketimi ile prospektif kohort çalışmaların bulguları temel alınarak bu meyveleri tüketen bireylerde diyabet riskinin daha düşük olduğu belirtilmektedir. Yapılan bir çalışmada flavonoid alımının veya flavonoid içeren gıdaların alımının, tip 2 diyabet riskinin azalmasıyla ilişkili olmadığı buna karşın kırmızı ve beyaz şarap tüketimiyle tip 2 diyabet

riskinin azaldığı ifade edilmiştir. Anavatanı Amerika kıtası olan saskatoon üzümü, aroniya, gilaburu, güz zeytini gibi üzümsü meyvelerin içerdikleri fitokimyasallar diyabet hastalığının ilerlemesini durdurduğu ifade edilmiştir (Bulantekin ve ark., 2020).

Birkaç çalışmada ortaya çıktığı gibi, meyveler ve meyve polifenolleri glikoz metabolizmasını etkileyebilir. Anti-diyabetik etkiler, pankreatik β-hücrelerini koruma, glikoz sindirimini, emilimini ve alımını etkileme ve glikoz/lipid metabolizma yollarını aktive etmedeki yararlı etkilerine bağlanıyor gibi görünmektedir. Ek olarak, berry-biyoaktif, glisemik kontrolün iyileştirilmesinden sorumlu inkretin sisteminin bir hormonu olan glukagon benzeri peptit-1'i (GLP-1) uyarabilir (Venturi ve ark., 2023).

Sonuç olarak, chokeberry meyve türevleri, metabolik sendromda meydana gelen bozuklukların ve bunların komplikasyonlarının önlenmesi ve tedavisinde umut verici bir tamamlayıcı tedavi seçeneği olabilir (Jurgoński ve ark., 2008). Berry sınıfının fonksiyonel bir besin olarak kan glikozunu düşürücü etkisi olduğu hücre ve hayvan çalışmalarıyla ispatlanmıştır. Ayrıca bu meyvelerin glikoz alımı üstünde konsantrasyona bağlı tesir ettiği kanıtlanmıştır (Alp., 2023).

KAYNAKLAR

Afacan FÖ, Sönmezdağ AS 2020. Antosiyaninlerin beslenmedeki önemi ve sağlik üzerine etkileri. Karya Journal of Health Science, 1(1): 19-24.

Alp E 2023. Goji Berry (Lycium Barbarum) ve Sağlık Üzerine Etkileri. Kapadokya Sağlık Bilimleri Dergisi, 1(2): 99-112.

Amagase H, Farnsworth NR 2011. A review of botanical characteristics, phytochemistry, clinical relevance in efficacy and safety of Lycium barbarum fruit (Goji). Food Research International, 44 (7): 1702-1717.

Anonim 2021. Türkiye İstatistik Kurumu (TUİK). Available at: www.tuik.gov.tr [20.07.2023]

Ayvaz Z 2021. Antioksidanlar. Ekoloji, 1: 26-33.

Bayoğlu Ş 2021. Farklı yörelerden Toplanan kızılcık (Cornus mas L.) Genotiplerinin Meyve özellikleri yönünden değerlendirilmesi. Bursa Uludağ Üniversitesi, Doktora Tezi, Türkiye.

Bulantekin Ö, Düzalan ÖB, Kuşçu A 2020. Meyve Sebze Tüketiminin Diyabette Önemi. Avrasya Sağlık Bilimleri Dergisi, 3(2): 55-61.

Ceylan Ş, Saral Ö, Mehmet Ö, Harşıt B 2017. Yaban mersininin (Vaccinium myrtillus L.) farklı çözücü ekstraktlarındaki antioksidan ve antimikrobiyal aktivitelerinin belirlenmesi. Artvin Çoruh Üniversitesi Orman Fakültesi Dergisi, 18(1): 21-27.

Çağlar, M., Demirci, M. (2017). Üzümsü Meyvelerde Bulunan Fenolik Bileşikler ve Beslenmedeki Önemi. A vrupa Bilim ve Teknoloji Dergisi, 7(11): 18-26.

Çelik E 2012. Organik olarak yetiştirilen frenküzümü ve bektaşiüzümü çeşitlerinin bazı özelliklerinin araştırılması. Ordu Üniversitesi Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi.

Çelik ZM, Sargin M, Tamer HG, Güneş FE 2022. İnsülin Direnci Olan Kadınların Tıbbi Beslenme Tedavisine Kızılcık (Cornus mas L.) Meyvesi Eklenmesinin Beslenme Durumuna Etkisi. 3.Uluslararası Sağlıklı Beslenme Kongresi, Edirne, Türkiye.

da Silva Pinto M, de Carvalho JE, Lajolo FM, Genovese MI, Shetty K 2010. Evaluation of antiproliferative, anti-type 2 diabetes, and antihypertension potentials of ellagitannins from strawberries (Fragaria× ananassa Duch.) using in vitro models. Journal of Medicinal Food, 13(5): 1027-1035.

Demir GM 2014. Streptozotocinle İndüklenen Diyabetli Ratlar Üzerinde Mersin Bitkisi (Myrtus Communis L.) Meyvesinin Su Ekstresi Etkilerinin İncelenmesi. Atatürk Üniversitesi Sağlık Bilimleri Enstitüsü, Yüksek Lisans Tezi.

Durgut S, Yılmaz E, Karabacak AÖ, Sinir GÖ 2022. Investigation of Physicochemical Properties, Antioxidant Activity and In vitro Bioaccessibility of Cornelian Cherry (Cornus mas. L.) Nectar Enriched with Herbal Teas. Turkish Journal of Agriculture-Food Science and Technology, 10(11), 2156-2164.

Eke İ 2017. Bazı yabani Vaccinium ve Rubus türlerinde antioksidan, fitokimyasal ve pomolojik özelliklerinin belirlenmesi. Niğde Ömer Halisdemir Üniversitesi Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi.

Engin SP, Boz Y, Mert C, Fidancı A, İkinci A. 2018, April. Growing aronia berry (Aronia melanocarpa (Michx.) Elliot). 1. International Gap Agriculture & Livestock Congress, Sanlıurfa, Türkiye.

Erdoğan Bayram S, Özeker E, Elmacı ÖL 2013. Fonksiyonel Gıdalar ve Çilek. Akademik Gıda, 11(2): 131-137.

Erten ES, Yılmaz FM 2019. Aydın ilinde yetiştirilen ticari çilek çeşitlerinin fiziksel, kimyasal, biyoaktif ve aroma özellikleri. Harran Tarım ve Gıda Bilimleri Dergisi, 23(2): 131-141.

Gazioğlu Şensoy, R. İ., Koç, H., Baş, E. Ö. (2018). Bazı yerli üzüm formlarının (vitis vinifera l) besin ve kalite özelliklerinin belirlenmesi. Ejons International Journal, *2*(4): 129-138.

Golovinskaia O, Wang CK 2021. Review of Functional and Pharmacological Activities of Berries. Molecules (Basel, Switzerland), 26(13): 3904.

Gowd V, Bao T, Wang L, Huang Y, Chen S, Zheng X, Cui S, Chen W 2018. Antioxidant and antidiabetic activity of blackberry after gastrointestinal digestion and human gut microbiota fermentation. Food chemistry, 269: 618–627.

Islam T, Yu X, Badwal TS, Xu B 2017. Comparative studies on phenolic profiles, antioxidant capacities and carotenoid contents of red goji berry (Lycium barbarum) and black goji berry (Lycium ruthenicum). Chemistry Central journal, 11(1): 59.

Kaya İ, Mavioğlu Kaya M 2015. Bazı Meyvelerin Antioksidan Özellikleri. 21. Yüzyılda Fen ve Teknik, 1(3): 41-46.

Jurgoński A, Juśkiewicz J, Zduńczyk Z 2008. Ingestion of black chokeberry fruit extract leads to intestinal and systemic changes in a rat model of prediabetes and hyperlipidemia. Plant foods for human nutrition (Dordrecht, Netherlands), 63(4): 176–182.

Kalt W, Cassidy A, Howard LR, Krikorian R, Stull AJ, Tremblay F, Zamora-Ros R 2020. Recent Research on the Health Benefits of Blueberries and Their Anthocyanins. Advances in nutrition (Bethesda, Md.), 11(2): 224–236.

Li D, Zhang Y, Liu Y, Sun R, Xia M 2015. Saflaştırılmış antosiyanin takviyesi, diyabetik hastalarda dislipidemiyi azaltır, antioksidan kapasiteyi artırır ve insülin direncini önler. Beslenme Dergisi, 145(4): 742-748.

Li J, Shi C, Shen D, Han T, Wu W, Lyu L, Li W 2022. Composition and Antioxidant Activity of Anthocyanins and Non-Anthocyanin Flavonoids in Blackberry from Different Growth Stages. Foods, 11(18): 2902.

Nemzer BV, Al-Taher F, Yashin, A, Revelsky I, Yashin Y 2022. Cranberry: Chemical Composition, Antioxidant Activity and Impact on Human Health: Overview. Molecules (Basel, Switzerland), 27(5): 1503.

Nizamlioğlu NM, Nas S 2010. Meyve ve Sebzelerde Bulunan Fenolik Bileşikler; Yapıları ve Önemleri. Gıda Teknolojileri Elektronik Dergisi, 5(1): 20-35.

Oszmiański J, Lachowicz S 2016. Chokeberry'den (Aronia melanocarpa L.) Kurutulmuş Meyve ve Meyve Suyu Üretiminin Biyoaktif Bileşiklerin İçeriği ve Antioksidatif Aktivitesi Üzerine Etkisi. Moleküller, 21(8): 1098.

Özarda Ö 2009. Üzümsü meyvelerden elde edilen ekstraktların antioksidan etkilerinin giderilmesi ve meyveli içeceklerdeki raf ömrüne etkisi. Gebze Yüksek Teknoloji Enstitüsü Mühendislik ve Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi.

Özder, A. (2021). Bazı Aronya Çeşitlerinin (Aronia Melanocarpa L.) Gelişme Performanslarının Belirlenmesi. Aydin Adnan Menderes Üniversitesi Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi.

ÖZKAN N 2020. Bazi nutrasötik meyvelerin nörodejeneratif etkileri. ERÜ Sağlık Bilimleri Fakültesi Dergisi, 7(1): 37-45.

Pehluvan M, Güleryüz M 2004. Ahududu ve böğürtlenlerin insan sağlığı açısından önemi. Bahçe, 33(1): 51-57.

Sá RR, da Cruz Caldas J, de Andrade Santana D, Lopes MV, Dos Santos WNL, Korn MGA, Júnior ADFS 2019. Multielementar/centesimal composition and determination of bioactive phenolics in dried fruits and capsules containing Goji berries (Lycium barbarum L.). Food Chemistry, 273: 15-23.

Sławińska N, Prochoń K, Olas B 2023. A Review on Berry Seeds—A Special Emphasis on Their Chemical Content and Health-Promoting Properties. Nutrients, 15(6): 1422.

Tosun İ, Yüksel S 2003. Üzümsü Meyvelerin Antioksidan Kapasitesi. Gıda, 28(3): 305-311.

Venturi S, Marino M, Cioffi I, Martini D, Del Bo' C, Perna S, Riso P, Klimis-Zacas D, Porrini M 2023. Berry Dietary Interventions in Metabolic Syndrome: New Insights. Nutrients, 15(8): 1906.

Yıldız S 2012. Ülkemizde doğal olarak yetişen ve kültüre alinan Vaccinium spp. türlerinin fenolik bileşiklerinin ve antioksidan kapasitelerinin araştırılması. Bursa Uludağ Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi.

ORAL PRESENTATION

İrritabl bağırsak sendromunda (IBS) besin ve beslenme çözümü

Gökçe Çakmak Kafadar[1*] (*ORCID: https://orcid.org/0000-0002-0364-1546*)

[*1]Kırklareli Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Kırklareli, Türkiye

*Sorumlu yazar e-mail: gokceckmk@gmail.com

Özet

İrritabl bağırsak sendromu (IBS) dünya genelinde oldukça sık rastlanan bir gastrointestinal fonksiyonel bozukluğudur. IBS hastalarında genel olarak karın ağrısı, şişkinlik karında rahatsızlık, ishal ya da kabızlık gibi belirtiler göstermektedir. Bu durum IBS'li hastaların yaşam kalitesini olumsuz etkilemektedir. Bu çalışmanın amacı IBS hastalığı ve bu hastalıkta etkili olabilecek besin ve beslenme çözümlerini sunmaktır. IBS hastalığında uygulanan bazı diyet çeşitleri bulunmaktadır. Bu diyet çeşitlerinden FODMAP diyeti olmak üzere, probiyotikler, prebiyotikler, sinbiyotikler ile mikrobiyotanın sağlığı, hastalığın tedavisinde önemli gözükmektedir. Yapılan çalışmalar FODMAP (fermente edilebilir oligosakkaritler, disakkaritler, monosakkaritler ve polioller) diyetinin diğer diyet çeşitlerine göre daha etkili olduğunu göstermektedir. Bununla birlikte bazı çalışmalarda IBS hastalarında bağırsak mikrobiyotasının zenginliğinde azalma olduğu saptanmıştır. Beyin-bağırsak ekseni nedeniyle probiyotik kullanımı IBS semptomlarına ve yaşam kalitesi üzerinde olumlu etkiler yaptığına dair çalışmalar mevcuttur. Mevcut veriler incelendiğinde IBS'nin etkileşim içinde olduğu çeşitli hastalıklar (depresyon vb.) mevcut besin ve beslenme çözümlerinin tamamen etkisinin görülmesini güçleştirmektedir. Bu sebeple bu alanda daha fazla randomize kontrollü çalışmalara gerek duyulmaktadır.

Anahtar Kelimeler: İrritabl bağırsak sendromu, Diyet, Besin, Beslenme

Food and nutrition solution for irritable bowel syndrome (IBS)

Abstract

Irritable bowel syndrome (IBS) is a very common gastrointestinal functional disorder worldwide. IBS patients generally show symptoms such as abdominal pain, bloating, abdominal discomfort, diarrhea or constipation. This situation negatively affects the quality of life of patients with IBS. The aim of this study is to present IBS disease and nutritional and nutritional solutions that may be effective in this disease. There are some types of diets applied in IBS disease. The health of the microbiota with probiotics, prebiotics, synbiotics, and the FODMAP diet, seem to be important in the treatment of the disease. Studies show that the FODMAP (fermentable oligosaccharides, disaccharides, monosaccharides and polyols) diet is more effective than other diet types. However, in some studies, it has been found that there is a decrease in the richness of the intestinal microbiota in IBS patients. There are studies showing that the use of probiotics has positive effects on IBS symptoms and quality of life due to the brain-intestinal axis. When the available data are examined, various diseases (depression, etc.) with which IBS interacts make it difficult to see the full effect of the existing food and nutrition solutions. Therefore, more randomized controlled studies are needed in this area.

Keywords: Irritable bowel syndrome, Diet, Food, Nutrition

GİRİŞ

Dünya çapında yaygın olarak görülen ve gastrointestinal fonksiyonel bir bozukluk olan irritabl bağırsak sendromu (IBS) hastaları, genellikle karın ağrısı, şişkinlik, karında rahatsızlık ve ishal/kabızlıktan şikâyetçi olmaktadır. Bu durumda hastanın yaşam kalitesini ciddi şekilde olumsuz etkilemektedir (Liu, Y. L.,& Liu, J. S., 2021).

Tedavi edilen en yaygın hastalıklardan biri olmasına rağmen IBS'li hastaların yalnızca %10 kadarı tıbbi destek almak istemektedir (Alammar, N.,& Stein, E., 2019). Batı popülasyonlarında yaklaşık %5 ile %15 arasında IBS'nin varlığı tahmin edilmektedir. Lovell ve Ford tarafından yapılan bir çalışmada IBS'nin çoğunlukla

kadınlarda görüldüğü ve 50 yaşından önce başlayarak genel toplumda %11.2 oranında görüldüğü bulunmuştur (Alammar, N.,& Stein, E., 2019).
IBS'nin patofizyolojisi belirsizdir. Değişen gastrointestinal motilite, visseral aşırı duyarlılık, enfeksiyöz sonrası reaktivite, beyin-bağırsak etkileşimleri, fekal mikro florada değişiklik, bakteriyel aşırı büyüme, gıda duyarlılığı, karbonhidrat malabsorpsiyonu ve bağırsak iltihabının tümü, IBS'nin patogenezini etkilemektedir. Buna ek olarak, bu mekanizmalardan algılanan belirtiler karın ağrısı ya da rahatsızlığı, şişkinlik, diyare ve konstipasyon oluşur. Bütün belirtiler gastrointestinal olmamakla birlikte yorgunluk çok yaygın bir şekilde görülebilir (Saha L., 2014).

BULGULAR ve TARTIŞMA

IBS tanı kriterleri

Roma IV son yayınlanan Roma kriterlerindendir ve IBS ile ilişkili karın semptomları olan hasta bireylerin daha iyi yönetilmesine yardımcı olmak için fonksiyonel bağırsak hastalığı uzmanları tarafından oluşturulmuştur. Semptomların tanıdan en az 6 ay önce kendini göstermesi ve son 3 ay içinde tanı kriterlerini doldurmalıdır. Bu bilgilere göre, son 3 ayda, haftada en az 1 gün tekrar eden karın ağrısı olmalıdır. Bununla birlikte, IBS, aşağıdaki kriterlerden 2 ya da daha fazlasıyla ilişkilidir.
Dışkılama ile ilgili
Dışkı sıklığındaki değişiklikle ilişkili
Dışkı formundaki değişiklikle ilişkili
Bu sınıflandırma kullanılarak IBS, baskın dışkı modeline göre sınıflandırılır.
1. Kabızlığın baskın olduğu IBS (IBS-C); %25'ten fazla sert dışkı ve %25'ten azından dışkı sulu ya da gevşek
2. İshalin baskın olduğu IBS (IBS-D): %25'ten fazlasında dışkı gevşek ve %25'ten azı sert dışkı
3. Karışık barsak alışkanlıklarına sahip IBS (IBS-M): %25'ten fazla gevşek dışkı ve %25'ten fazla sert dışkı
4. Sınıflandırılmamış IBS (IBS-U): %25'ten azı gevşek dışkı ve %25'ten azı sert dışkı (Alammar, N.,& Stein, E., 2019).
IBS belirtileri spesifik olmayabilir, fakat belirtilerin değerlendirilmesi anında "tehlike işareti" semptomlarının sorularak öğrenilmesi önem arz eder. Bunlar, IBS belirtileri olarak sayılmaz ve her zaman daha fazla değerlendirme gerektirir. Tehlike işareti belirtileri aşağıdakileri içerir (Alammar, N., & Stein, E., 2019).
Yaşın 50'den büyük olması
Gece belirtileri, özellikle ishal durumu
Kanlı dışkı olması
Ateş
Kiloda azalma
Laboratuvar testlerinde anormallik (anemi, yüksek inflamatuar belirteçler dahil)
Ailede inflamatuar bağırsak hastalığı ya da kolon kanseri hikayesi

İbs'de besin ve beslenme çözümü

IBS'li bireyler, sağlıklı insanlar ilekarşılaştırıldığında daha kötü hayat kalitesi bildirirler ve diyet, yaşam tarzlarını etkileyen önemli bir faktördü. Hastaların çoğu, IBS belirtilerini tükettikleri yiyeceklerle ilişkilendirdi. Bu durum, IBS'li bireylerde düşük hayat kalitesi ile ilişkili gıdadan kaçınma davranışı sergiler ve diyetin hastaların refahı üzerindeki olumsuz etkisini vurgular. Bazı yiyecekler IBS belirtilerini arttırabilse de, bazıları rahatlama sağlayabilir. IBS için etkili diyet yönetimi yaklaşımları tasarlamak için yapılan araştırmalarda çeşitli diyet stratejilerine odaklanılmıştır (Dimidi, E.,& Whelan, K., 2020).
IBS'li hasta bireylerden üçte ikisi kadarı belirtilerin yemeklerden sonra kötüleştiğini bildirmektedir. Hastalar, uzun süredir IBS semptomlarını belirli yiyeceklerin yenilmesi ya da bir öğünün kendisi ile ilişkilendirmiştir ve postprandiyal belirtiler artan hastalık düzeyi ve azalan hayat kalitesi ile ilişkili görmüşlerdir (Harer, K. N.,& Eswaran, S. L., 2021).
Eliminasyon diyetleri: Bazı özel gıdaların IBS belirtilerinin düzeyini arttırdığı düşüncesi, 1990'larda Cambridge grubunun öncülüğünü yaptığı ve zayıf dışlamaya odaklanan eliminasyon diyetlerinin temelini oluşturur. Bu düşünce modeli, hastaların genel olarak belirtilere sebep olduğunu gözlemlediği gıdaları ortadan kaldırarak başladığı daha ampirik bir yaklaşım olarak daha da geliştirildi. Bu ürün grubuna süt ürünleri, tahıllar, turunçgiller, patates, çay, kahve, alkol, katkı maddeleri ve koruyucular dahil edilmiştir. Diyet içeriği olarak çoğunlukla taze et, balık, pirinç, sebzeler ve keçi, koyun ya da soya sütüne dayanan bir çalışma yapılmıştır. Diyette dışlamalar yapılıp belirtiler düzeldikten sonra gıdalara tekrar başlandı.Her 2-3 günde bir yeni bir gıda grubu verildi ve bu şekilde gıdalar tolere edilen ya da semptomlarasebep olan gıdalar olarak

sınıflandırıldı. Semptomlar, eliminasyon diyeti ile hastaların 91/189'unda (%48) iyileşti ve yeniden tüketmeden sonra 73'ü bir ya da daha fazla gıda intoleransı tanımlayabildi ve 72'si 1 yıldan uzun takipte iyi durumda kaldığını ifade etti.

İsveç'te yapılan bir araştırma, IBS hastalarının %84'ünün belirtilerini tetikleyen en az bir gıdayı ifade ettiğini ortaya çıkardı. Bunlar arasında süt ürünleri (%49), fasulye/mercimek (%36), elma (%28), un (%24) ve erik (%23) ile biyojenik aminler, şarap/bira (%31), salam (%22) ve peynir (%20) yer aldı ve %52'si genel olarak kızarmış/yağlı yiyecekleri ifade etti.

Norveç'te nüfusa dayalı bir araştırmada, IBS hastalarının ortalama olarak 2,5 gıda maddesinden kaçındığı bildirildi. Buna sütten kaçınan %35, %14 peynir, %16 bakliyat, %24 soğan, %10 buğday unu, %26 kahve ve %12 bira dahil edildi. Tanımlanan gıdaların sayısı ile duygudurum bozuklukları ya da kas-iskelet ağrısı arasında bir ilişki olmadığı bulundu (Spiller R.,2021).

Düşük FODMAP diyeti: Saygılı bir hasta-hekim ilişkisi, başarılı IBS tedavisinin temel noktasıdır ve güvenli bir IBS tanısı koymak, hastanın tanıyı kabul etmesi için çok önemlidir. Tedavinin hedefleri, IBS belirtilerini kontrol etmenin dışında hayat kalitesini iyileştirmeyi ve stresi azaltmayı içerir. Özel olarak, FODMAP bakımından zayıf bir diyetin IBS popülasyonunda etkili ve güvenli olduğu düşünülmüştür. FODMAP'ler, muhtemelen çoklu mekanizmalar yoluyla GI belirtilerine katkıda bulunduğu düşünülen, ozmotik olarak aktif karbonhidratların çeşitli bir ailesidir.(Liu, J., Chey, W. D., Haller, E., & Eswaran, S., 2020).

Düşük FODMAP diyeti, kısa zincirli karbonhidratların, yani oligosakkaritlerin, disakkaritlerin, monosakkaritlerin ve poliollerin hariç tutulmasına dayanan bir diyet yaklaşımıdır. FODMAP'ler ortak hususlara sahip heterojen bir gruptur.İnce bağırsakta yavaş ve tamamen emilmezler, mikrobiyota tarafından hızla fermente edilebilirler ve yüksek ozmotik kapasiteye sahiptirler. Mevcut bilimsel kanıtlar, özellikle ağrı, şişkinlik ve diyare olmak üzere belirtilerin genel kontrolünde düşük FODMAP diyetinin (LFD) önemli bir etkinliğini göstermektedir. Yulaf, keten tohumu, psyllium gibi düşük FODMAP lif kaynakları ve kivi ve papaya gibi bazı meyve türleri bu durumlarda tercih edilecek yiyecek seçenekleri olabilir. Literatüre göre LFD, bağırsak mikrobiyotasında, yani Bifidobakterilerin bolluğunda olumsuz bir etkiye sahip olabilir; fakat bu negatif etki, probiyotik takviyelerin kullanımıyla kolayca kontrol altına alınabileceğini düşündürmektedir. Her vakanın kişisel ve ayrıntılı değerlendirmesi ve kişilerin terapötik sürece dahil edilmesi, her vaka için en uygun stratejinin tanımlanması içinçok önemlidir (Guerreiro, M., Sousa Guerreiro, C., & Cravo, M., 2019).

Tam tahıllı buğday ürünleri ya da yüksek FODMAP meyve ve sebzeleri uygun düşük FODMAP seçenekleri ile değiştirilmezse, FODMAP içeriğindeki azalma aynı zamanda lif alımında da eş zamanlı bir azalmaya sebebiyet verebilir. FODMAP'lerde bir azalma, kabızlık ihtimalini artırarak bağırsak lümenine ozmotik sıvı geçişini de azaltabilir. Bu etkiler, devam eden kabızlığı olan IBS hastaları için özellikle zararlı olabilir (Bellini, M.,ve diğerleri., 2020).

Buğday ve glutensiz diyetler: Buğdayın hasta anketlerinde ifade edildiği gibi, sürekli olarak IBS belirtilerini ağırlaştırdığı tespit edilmiştir. Mekanizması net değildir, fakat çölyak hastalarında görüldüğü gibi glütene karşı immün reaksiyon, IBS belirtileri ile bağlantılıdır.Artan bağırsak geçirgenliği ve artmış mukozal mast hücreleri ve azalmış serotonin taşıyıcı gibi IBS'de görülen önemli anormallikler de çölyak hastalarında glütensiz bir diyete başlamadan önce görülür. Bu gözlemler, birçok IBS'li bireyde, satışları son zamanlarda önemli ölçüde artan, daha çok talep edilen ve pahalı glütensiz diyeti benimsemesine ortaya çıkarmıştır. IBS yönetimine ilişkin kılavuzların çoğu, Roma III kriterlerini karşılayan seçilmemiş IBS hastalarında, kontroller ile karşılaştırıldığında çölyak hastalığına yakalanma riskinin 7 kat arttığından, çölyak hastalığını dışlamanın önemli olduğu konusunda uyarıda bulunur (Spiller R., 2021).

Laktoz ve fruktoz kısıtlaması: Bu, fermente edilebilir karbonhidratın kolona verilme hızına ek olarak malabsorpsiyon düzeyinin belirtilerin belirlenmesinde önemli olduğunu düşündürmektedir. Yapılan çalışmalar, devam eden laktoz alımına bir miktar adaptasyonun, diyette laktoza 3 hafta maruz kaldıktan sonra nefes hidrojen yanıtında azalma ve belirtilerde azalma ile gerçekleştiğini göstermektedir. Son dışkı inkübasyon çalışmaları ayrıca semptomatik olanların daha fazla laktat ve diğer kısa zincirli yağ asitleri ürettiklerini göstermektedir, bu da kolonik mikrobiyotanın belirtilerin gelişip gelişmemesi konusunda önemli belirleyiciler olduğunu düşündürmektedir. Diğer anahtar faktör, distansiyona ve bakteriyel metabolitlere karşı visseral aşırı duyarlılıktır.Çin'de yapılan veçoğunun genetik olarak belirlenmiş laktoz malabsorpsiyonuna sahip olduğu bir çalışmada, laktoz intoleransı olanların, asemptomatik laktoz malabsorpsiyonu olanlarla benzer seviyelerde laktoza karşı nefeste hidrojen tepkisine sahip olduğunu, fakat daha düşük dozlarda belirtileri bildirmede farklılık gösterdiğini göstermektedir. Laktoz intoleransı olan IBS hastaları da sıklıkla fruktozu zayıf bir şekilde emer ve bu, bazılarında sorbitol gibibelirtilere sebebiyet verebilir. Fruktoz ve sorbitol azaltılmış diyetler, kontrolsüz müdahalelerde yarar sağlamıştır (Spiller R., 2021).

DRAFT

TABLO 1: Bazı Gıdaların FODMAP İçeriği (Liu, J., Chey, W. D., Haller, E., & Eswaran, S., 2020)

FODMAP İÇERİĞİ	Tahıllar	Meyveler	Sebzeler	Süt Ürünleri Ve Bitki Bazlı Seçenekler	Proteinler	İçecekler
YÜKSEK	Buğday Çavdar Arpa	Elma Kayısı Böğürtlen Kiraz Hurma Greyfurt Mango Armut Karpuz	Enginar Kuşkonmaz Karnabahar Sarımsak Pırasa Mantarlar Arpacık soğan Şeker bezelye	Hindistan cevizi sütü (karton paket) Dondurma Süt Yumuşak peynir Soya sütü Yoğurt	Fasulye/ Baklagiller İşlenmiş Etler	Yüksek fruktoz içeren Gazlı içecekler ve Meyve suları İçki Çaylar: papatya, Rezene, Ve çay
DÜŞÜK	Mısır ekmeği/ Cipsi İrmik Glütensiz Makarnalar, Kraker ve Ekmekler Yulaf ezmesi Patates Patlamış mısır Pirinç Ekşi maya Ekmek Kinoa	Muz (olgunlaşmamış) Üzüm Kivi Limon Misket limonu Mandalina Portakal Papaya Ananas	Çin lahanası Brokoli Havuç Frenk soğanı Salatalık Patlıcan Lahana Marul İstiridye mantarı Zeytin Turp Ispanak Domates	Badem sütü Hindistan cevizi yoğurdu Laktozsuz Süt kreması, Yoğurt Peynir	Olgunlaşmamış soya fasülyesi Mercimek Konserve/ durulanmış nohut Biftek Tavuk Yumurtalar Balık/deniz ürünleri Domuz eti Hindi Tofu (sert)	Alkol: şarap (çoğu), bira, Kahve Sükroz tatlandırılmış veya diyet yumuşak İçecekler Çaylar (Yukarıda listelenen çaylar hariç) Su

Aloe Vera takviyesi: 360'tan fazla sulu bitki türü içeren aloe, sağlıkla ilgili özellikleri sebebiyle geniş çapta araştırılan türlerden biridir.Glukomannanlar, acemannan ve mannoz içermesi sebebiyle, aloe verain vitro mikrobiyota fermantasyonuanında gösterilmiş olan kısa zincirli yağ asitlerine ek olarak bifidobakterilerdeki artışlarla bir prebiyotik potansiyele de sahip olabileği düşünülmektedir. Bu fizyolojik etkiler dikkate alındığında, çalışmalar aloe vera bitkisinin IBS semptomlarını iyileştirebileceği olası gözükmektedir (Dimidi ve Whelan, 2020).

Nane yağı takviyesi: Mentha X piperita bitkisinden elde edilen nane yağının, bağırsaktaki histaminerjik ve kolinerjik reseptörleri modüle ettiği ve k-opioid agonist aktivitesi, serotonerjik antagonizm ve anti-inflamatuar etkiler sergilediği gösterilmiştir. Aktif içeriği L-mentol, bağırsak düz kaslarının gevşemesine sebep olduğu için potansiyel olarak karın rahatsızlığını azaltır. Ek olarak, alt özofagus sfinkterinin gevşemesinin de gastro-özofageal reflüye neden olabileceği gösterilmiştir.Bu sebeple, reflü belirtilerine engel olmak için üst gastrointestinal sistemdeki salınımı atlayan, fakat nane yağını düz kas gevşetici özelliklerini sergileyebileceği alt gastrointestinal kanalda serbest bırakan enterik kaplı nane formülasyonları bulunmaktadır. 835 IBS'li hastadan oluşan 12 RKÇ'nin sistematik bir incelemesi ve meta-analizi, nane yağının IBS belirtilerini iyileştirmede güvenli ve etkili olduğu sonucuna ulaşmıştır. Nane yağının genel semptom riski üzerinde plasebo ile karşılaştırıldığında bir faydası olduğu görülmüştür (iyileşme RR 2.4, %95 CI 1.9-3.0,P< .001) ve karın ağrısı (iyileşme RR 1.8, %95 GA 1.4-2.2,P< .001). Daha yakın tarihli, sistematik bir inceleme ve meta-analizi, IBS'de nane yağı, çözünür lif, antispazmodik ilaçlar ve bağırsak-beyin modülatörlerinin etkisini karşılaştırdı. Tüm araştırmalar içinde, nane yağı etkinlik bakımından ilk sırada yer aldı. Çalışmalar nane yağının güvenli olduğunu ve karın ağrısı da dahil olmak üzere IBS semptomlarını hafifletebileceğinin olası olduğunu ifade etmektedir (Dimidi ve Whelan, 2020).

Lif takviyeleri: Azalan lif alımı, genel toplumda kabızlığın artma riski ile ilişkilidir ve birkaç randomize çalışma, bazı IBS'li bireylere lif vermenin faydalı olduğunu düşündürmektedir. Buna ek olarak, iki meta-analiz, bu etkinin sadece psilyum denemelerinde görüldüğü ve buğday kepeği gibi parçacıklı lifin hiçbir yarar sağlamadığını ifade etmektedir. Son araştırmalar, psilyumun ince bağırsakta suyu hapsederek ve kolonik su

içeriğini artırarak daha yumuşak ve daha sık dışkıya sebep olarak nasıl davrandığını göstermektedir. Psilyumunneden kabızlıkta yararlı olduğu açıktır, fakat psilyum sindirimi yavaşlatmadığı ve hızlı fermantasyonu azaltmadığı sürece neden kabız olmayan hastalara da yarar sağladığı belirsizdir (Spiller, 2021).

SONUÇ

Dünya çapında yaygın olarak görülen ancak saptanması güç olan irritabl bağırsak sendromunun tanımlanması için ROMA IV kriterleri kullanılmaktadır. Bu kriterlerle tanı alan hastaların beslenmelerini düzenlemeleri önerilmektedir. IBS'de etkili olan çeşitli diyet, besin ve besin takviyeleri bulunmaktadır. Diyet şekli olarak FODMAP diyeti ile ilgili son yıllarda olumlu sonuçlar öne çıkarken aloe vera, nane yağı ve lif takviyelerinin de etkili olabileceği sonucuna varılmıştır. Beyin-bağırsak aksı sebebiyle depresyon gibi bir çok hastalıkla birlikte görülen IBS'de kesin olarak etkili diyet, besin veya besin takviyesi henüz saptanmamıştır. Bu sebeple dünya çapında yaygın görülen ve birçok bireyin yaşam kalitesini düşüren bu hastalıkta etkili olan besin ve beslenme çözümleri ile ilgili daha fazla randomize kontrollü çalışmalara ihtiyaç duyulmaktadır.

KAYNAKLAR

Alammar N, Stein E 2019. Irritable bowel syndrome: what treatments really work. Medical Clinics, 103(1): 137-152.

Bellini M, Tonarelli S, Nagy AG, Pancetti A, Costa F, Ricchiuti A, ... Rossi A 2020. Low FODMAP diet: evidence, doubts, and hopes. Nutrients, 12(1): 148.

Dimidi E, Whelan K 2020. Food supplements and diet as treatment options in irritable bowel syndrome. Neurogastroenterology & Motility, 32(8): e13951.

Guerreiro M, Guerreiro CS, Cravo M 2019. Irritable bowel syndrome: how can symptoms and quality of life be improved with diet?. Current Opinion in Clinical Nutrition & Metabolic Care, 22(5): 377-382.

Harer KN, Eswaran SL 2021. Irritable bowel syndrome: food as a friend or foe?. Gastroenterology Clinics, 50(1): 183-199.

Liu, J, Chey WD, Haller E, Eswaran S 2020. Low-FODMAP Diet for Irritable Bowel Syndrome: What We Know and What We Have Yet to Learn. Annual review of medicine, 71: 303–314.

Liu YL, Liu JS 2021. Irritable bowel syndrome in China: a review on the epidemiology, diagnosis, and management. Chinese medical journal, 134(12): 1396–1401.

Saha L 2014. Irritable bowel syndrome: pathogenesis, diagnosis, treatment, and evidence-based medicine. World journal of gastroenterology, 20(22): 6759–6773.

Spiller R 2021. Impact of Diet on Symptoms of the Irritable Bowel Syndrome. Nutrients, 13(2): 575.

ORAL PRESENTATION

Characterization of TALE genes in the sugar beet (*Beta vulgaris* L.) genome by bioinformatics methods

Özlem BAKIR BOĞA[*] *(ORCID: https://orcid.org* 0000-0002-1964-3271), Huzeyfe UDUN (*ORCID: https://orcid.org/* 0009-0000-9265-0251), Esabi Başaran KURBANOĞLU *(ORCID: https://orcid.org/0000-0002-7434-6309*)

[*1] Ataturk University, Science Faculty, Department of Biology, Erzurum, Turkey
[2,3] Ataturk University, Science Faculty, Department of Biology, Erzurum, Turkey

*Corresponding author e-mail: ozlembakir@atauni.edu.tr

Abstract

Sugar beet is one of the plant sugar sources, providing 30-35% of annual sugar production worldwide. The aim of this study was to identify and characterize members of the TALE gene family in silico at a genome-wide level in *Beta vulgaris*. TALE genes are distributed in chromosomes 1,2,3,4,5,6,7,8,9 and 10 in the sugar beet genome. TALE proteins were determined to have the amino acid sequence range of 171-1826. Among them, Bvul-TALE-23 with 1826 amino acids, has the longest amino acid sequence. In contrast, the shortest amino acid sequence was obtained in Bvul-TALE-9 with 171 amino acids. Their molecular weights are between 19583.09-203572.25 DA, 33 of these proteins have acidic character, and 19 of them have basic character. As a result of the phylogenetic analyses carried out to elucidate the evolutionary status of the conserved region structures of TALE genes, it was determined that they were clustered in 3 different groups. Accordingly, it can be said that those located in similar regions in the classification have amino acid structures close to each other and therefore are in the same group. The most motifs were obtained from Bvul-TALE-3, 14 (13 motifs), and the least motif was from Bvul-TALE-15 (1 motifs). Among the TALE proteins found in *B.vulgaris*, 5 TALE proteins with a reliability of 90% and above were determined. The highest exon and intron numbers were obtained in Bvul-TALE-39. We believe that the results obtained from this study may be useful for future functional studies on the TALE gene family in sugar beet.

Keywords: Bioinformatic, TALE, in silico analysis, *Beta vulgaris*

INTRODUCTION

Sugar beet (*Beta vulgaris* L.) belongs to Caryophyllales in basal taxa of seed dicots. The species is diploid with 2n=18 chromosomes and an estimated genome size of 714 758 megabases (Dohm et al., 2014). Today, sugar beet is one of two phyto sugar sources that provide 30-35% of the annual sugar production worldwide (Zhang et al., 2016). It is also used for cattle feed (Evans and Messerschmidt, 2017), bioethanol (Zabed et al., 2014), baker's yeast and industrial alcohol production. Green chemistry has promoted new uses of sugar beet, such as bioplastic and biojet production, which bring the same economic and environmental benefits as sugar and alcohol production, creating a promising future for the beet sector. However, the quality and yield performance of sugar beet is strongly affected by abiotic and biotic stress (Märländer et al., 2003; Tzilivakis et al., 2005). Sugar beet genomic research, including genome-wide computational characterization of gene families, will offer opportunities to create high-yielding sugar beet varieties independent of abiotic and biotic stresses. Homeobox genes encode a large family of transcription factors (TFs) essential for the growth and development of animals and plants common in eukaryotes. The first homeobox gene was found in fruit flies (McGinnis et al., 1984). A typical homeobox domain has a triple helix region of 60 amino acids. The first and second helix form a loop structure, and the second and third helix form a helical return structure (Billeter et al., 1993). Homeobox genes in plants have been classified in two different ways. Initially, homeobox genes were divided into seven classes: ZM-HOX, HAT1, HAT2, ATHB8, GL2, KNOTTED-like homeodomain (KNOX/KNAT) and BEL1-like homeodomain (BELL/BLH) (Mukherjee et al., 2009). In subsequent research, homeobox genes were divided into 11 classes, including HD-ZIP (I ~ IV), WOX, NDX, PHD, PLINC, LD, DDT, SAWADEE, PINTOX, KNOX/KNAT, and BLH/BELL (Burglin et al., 2016). The aim of this study is to identify TALE gene family members in the sugar beet genome for the first time, using in silico methods, at

a genome-wide scale. In this study, the phylogenetic relationships of the TALE gene family, their distribution in the genome, gene structure arrangement, composition of motifs, and 3-dimensional protein structures were characterized. Identification and functional analysis of the *Beta vulgaris* TALE gene family provide a basis for a better understanding of the genetic mechanism of tolerance to biotic and abiotic stresses in the sugar beet genome.

MATERIALS AND METHODS

Identification of TALE Genes in the *Beta vulgaris* Genome

The protein sequences of the TALE gene family in the *Beta vulgaris* genome were obtained from the Phytozome database v12.1 (Anonymous, 2018a) using the Pfam Access Number for TALE proteins (PF00046) obtained from the Pfam database. In order to identify all possible TALE proteins in the *Beta vulgaris* genome, both the Phptozome database's v12.1 blastp and latent Markov model (HMM) (Anonymous, 2018b) search were scanned in the *B.vulgaris* genome with assumed parameters. Unrelated arrays were obtained using the "decrease redundancy tool Anonymous (Anonymous, 2018c). Presence of TALE proteins in related sequences was investigated using HMMER (Anonymous, 2018d) database. The molecular weight and theoretical isoelectric point (pI) of the obtained TALE proteins were determined using the ProtParam tool Anonymous (Anonymous, 2018e).

Sequence Alignment, Phylogenetic Analyzes and Determination of Conserved Motifs

Sequence alignment was done using ClustalW via MEGA program. Sequence alignment was used for phylogenetic tree plotting. The phylogenetic tree was created using the Maximum Neigbour-joining method in the MEGA7 program and visualized using the iTOL database. iTOL database is used to create a new phylogenetic tree and to visualize the phylogenetic tree to make the relationship between evolutionary processes more evident. Exon-intron analysis was performed with the GSDS2.0 (http://gsds.cbi.pku.edu.cn/) database so that the phylogenetic tree could give as accurate results as possible. Exon-intron regions were created using the genomic sequences of TALE sequences and CDS sequences. This program was used because the upgraded GSDS 2.0 has more annotation features and formats with a newly designed face. MEME Suite (MEME 4.11.1; http:/meme-suite.org/) database was used to identify conserved motifs. While performing the analysis, care was taken to ensure that the motif widths were optimum ≥2 and ≤300 by taking the maximum number of 5 motifs to determine the motifs. The motifs determined in the MEME Suite program were taken and the Publisher program was used to combine the motifs, and thus the preserved motif regions were determined (Bailey et al., 2009).

Determination of Chromosome Distribution

Using the obtained data, chromosome distribution was found via MapChart and the results were determined (Voorrips, 2002).

Homology Modeling of TALE Proteins

Using previously identified protein sequences for homology modeling, the 3D structure of the proteins was predicted with Phyre2. The reliability of the model was verified with the specified numerical data (Kelley, 2009).

RESULTS and DISCUSSION

Bioinformatics Analysis of TALE Proteins

As a result of the analysis using the sequences of TALE proteins identified in sugar beet, 52 TALE genes were identified. These identified TALE genes were named Bvul-TALE using their Latin names. The TALE proteins identified in the sugar beet genome are 171-1826 amino acids long, their molecular weights are between 19583.09-203572.25 DA, and 33 of these proteins have acidic character, and 19 of them have basic character (Table 1).

ID	*Beta vulgaris* Genomic Database Identifier	Physical position on *Beta vulgaris* genome			Protein length (aa)	pI	Molecular weight (Da)	Instability index	Stable or unstable
		Chromosome	Start position (bp)	End Position (bp)					
Bvul-TALE-1	EL10Ac1g00783	1	9,945,997	9,948,263	270	9.06	30187.35	47.56	unstable
Bvul-TALE-2	EL10Ac7g16759	7	15,086,606	15,088,628	178	9,18	20599.45	48.01	unstable
Bvul-TALE-3	EL10Ac4g10133.1	4	57,931,394	57,936,200	701	7.99	78811.99	67.16	unstable
Bvul-TALE-4	EL10Ac4g10153.1	4	58,157,999	58,163,913	474	8.12	53325.19	66.94	unstable
Bvul-TALE-5	EL10Ac9g22528.1	9	41,912,603	41,920,744	870	6.06	94764.24	47.14	unstable
Bvul-TALE-6	EL10Ac6g15475.1	6	60,746,424	60,748,375	341	6,45	38506.07	54.89	unstable
Bvul-TALE-7	EL10Ac1g00511.1	1	6,259,172	6,266,623	846	5,94	91613.38	48.39	unstable
Bvul-TALE-8	EL10Ac7g17517.1	7	37,564,404	37,573,286	965	7,69	106630.36	52.63	unstable
Bvul-TALE-9	EL10Ac2g04794.1	2	53,421,955	53,423,032	171	8,36	19583.09	54.75	unstable
Bvul-TALE-10	EL10Ac2g03900.1	2	41,276,730	41,281,406	544	5,94	60485.34	51.17	unstable
Bvul-TALE-11	EL10Ac3g06895.1	3	43,790,312	43,798,213	427	5,83	46314.90	52.23	unstable
Bvul-TALE-12	EL10Ac9g22097.1	9	31,795,311	31,806,456	876	5,89	96735.03	50.94	unstable
Bvul-TALE-13	EL10Ac5g12332.1	5	48,159,075	48,161,340	371	8,93	41004.76	65.35	unstable
Bvul-TALE-14	EL10Ac4g10157.1	4	58,195,209	58,200,401	662	8,20	75065.04	64.02	unstable
Bvul-TALE-15	EL10Ac2g02609.1	2	3,255,386	3,259,425	317	4,53	35562.70	68.95	unstable
Bvul-TALE-16	EL10Ac4g08278.1	4	13,357,545	13,368,669	698	5,76	71861.13	75.19	unstable
Bvul-TALE-17	EL10Ac5g12711.1	5	54,129,452	54,136,000	279	6,32	31373.48	62.78	unstable
Bvul-TALE-18	EL10Ac9g22861.1	9	49,335,120	49,338,360	393	6,90	44180.69	72.22	unstable
Bvul-TALE-19	EL10Ac7g16294.1	7	5,973,643	5,978,985	747	7,15	81533.63	41.63	unstable
Bvul-TALE-20	EL10Ac6g13714.1	6	9,324,432	9,338,050	337	4,88	38630.02	49.03	unstable
Bvul-TALE-21	EL10As7g23761.1	Scaffold	542,53	549,831	201	9,02	22872.51	50.15	unstable
Bvul-TALE-22	EL10Ac6g13144.1	6	8,570,3	8,60932	325	4,90	36832.50	59.37	unstable
Bvul-TALE-23	EL10Ac9g20819.1	9	6,685,165	6,698,321	1826	5.16	203572.25	53.71	unstable
Bvul-TALE-24	EL10Ac2g04154.1	2	45,814,317	45,820,115	390	5.70	43122.87	54.40	unstable
Bvul-TALE-25	EL10As13g24064.1	Scaffold	310,548	313,928	332	5.03	37800.67	66.27	unstable
Bvul-TALE-26	EL10Ac4g10134.1	4	57,944,552	57,950,416	557	7.57	63152.34	73.99	unstable
Bvul-TALE-27	EL10Ac4g10128.1	4	57,862,566	57,873,906	410	6.63	45781.08	46.81	unstable
Bvul-TALE-28	EL10Ac7g15931.1	7	1,829,104	1,831,252	252	7.80	29224.13	52.27	unstable
Bvul-TALE-29	EL10Ac1g00603.1	1	7,483,607	7,494,119	671	6.22	74196.52	45.90	unstable
Bvul-TALE-30	EL10Ac8g20466.1	8	55,512,583	55,517,706	474	7,08	51508.52	49.80	unstable
Bvul-TALE-31	EL10Ac8g19616.1	8	32,257,212	32,257,845	210	6.50	23170.01	50.36	unstable
Bvul-TALE-32	EL10Ac7g15994.1	7	2,571,587	2,573,653	341	6.49	38661.93	57.44	unstable
Bvul-TALE-33	EL10Ac4g08849.1	4	33,899,363	33,915,146	1163	7.90	129834.71	50.86	unstable
Bvul-TALE-34	EL10Ac8g20447.1	8	55,263,491	55,266,584	176	9.51	21039.88	52.00	unstable
Bvul-TALE-35	EL10Ac8g20004.1	8	45,998,665	46,007,420	733	5.67	80271.01	51.08	unstable
Bvul-TALE-36	EL10Ac5g10942.1	5	7,606,682	7,609,368	336	4.61	37657.15	62.10	unstable
Bvul-TALE-37	EL10Ac1g02334.1	1	56,834,745	56,861,369	405	6.14	46002.93	49.04	unstable
Bvul-TALE-38	EL10Ac8g20128.1	8	49,346,086	49,351,811	1403	7.25	159009.99	53.88	unstable
Bvul-TALE-39	EL10Ac2g04000.1	2	43,109,604	43,116,683	871	6.29	96001.05	54.92	unstable
Bvul-TALE-40	EL10Ac6g14947.1	6	53,181,983	53,184,123	289	8.11	32333.39	57.81	unstable
Bvul-TALE-41	EL10Ac5g12426.1	5	49,787,812	49,788,715	233	5.68	26244.28	44.33	unstable
Bvul-TALE-42	EL10Ac9g22564.1	9	43,091,714	43,103,204	528	8.63	58844.08	53.47	unstable
Bvul-TALE-43	EL10Ac3g05205.1	3	3,564,471	3,566,001	245	5.30	28540.78	62.87	unstable
Bvul-TALE-44	EL10Ac6g14709.1	6	48,099,288	48,101,688	217	6.12	25215.28	69.00	unstable
Bvul-TALE-45	EL10Ac5g11720.1	5	31,757,329	31,761,079	308	6.40	33033.70	58.20	unstable
Bvul-TALE-46	EL10Ac6g15053.1	6	54,994,568	54,999,638	290	5.21	32363.10	53.76	unstable
Bvul-TALE-47	EL10Ac7g17149.1	7	21,839,302	21,870,386	940	5.81	104704.59	44.85	unstable
Bvul-TALE-48	EL10Ac4g09093.1	4	42,088,346	42,094,521	218	5.81	24756.72	53.49	unstable
Bvul-TALE-49	EL10Ac5g12025.1	5	41,006,840	41,013,826	828	5.78	91582.12	50.86	unstable
Bvul-TALE-50	EL10Ac9g22031.1	9	31,060,208	31,070,365	744	5.93	82337.25	46.15	unstable
Bvul-TALE-51	EL10Ac3g05718.1	3	9,931,779	9,938,042	751	5.71	83106.60	50.37	unstable
Bvul-TALE-52	EL10Ac3g05517.1	3	7,094,379	7,103,626	769	9.01	87865.37	49.07	unstable

Table 1. Information about the TALE genes found in the *B.vulgaris* genome

According to the results of the chromosomal distribution studies, it was determined that the chromosome with the highest number of TALE genes was the 4th chromosome with 8 genes, and the chromosomes with the least genes were the 1st and 3rd chromosomes with 4 genes.

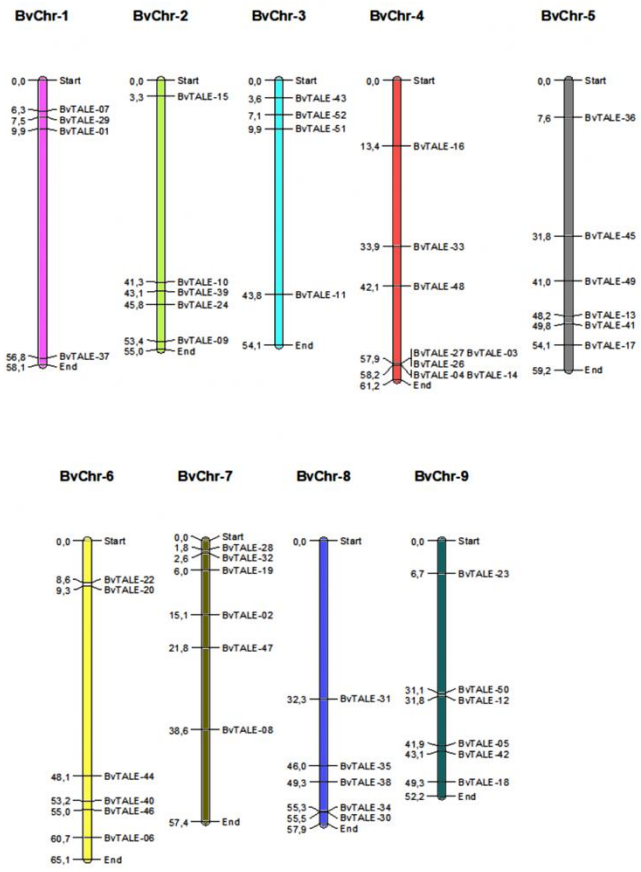


Figure 1. Distribution of TALE genes in *B.vulgaris* chromosomes

Phylogenetic Analyzes

Phylogenetic tree analysis was performed using the MEGA7 [MolecularEvolutionaryGenetic Analysis] program to determine the evolutionary relationships between TALE genes. Maximum neighbor-joining approach was used in this analysis. According to phylogenetic tree analysis, TALE genes were divided into 4 main groups. Sequences were aligned with ClustalOmega[Clustal W] in MEGA7 and an unrooted phylogenetic tree was constructed. The created phylogenetic tree was colored in a different color for each family using the ITOL database.

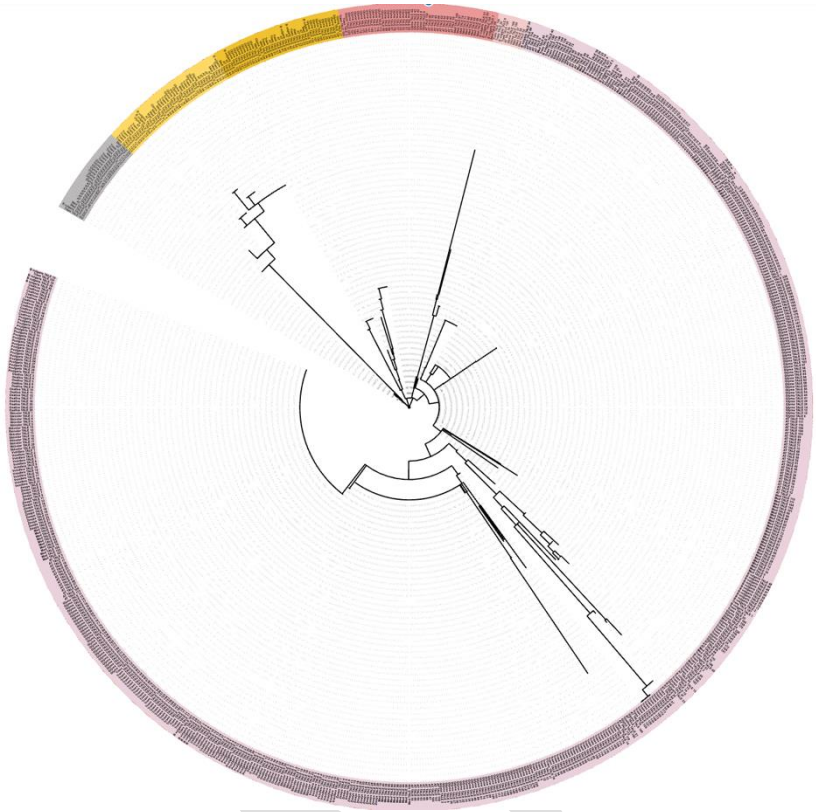


Figure 2. Phylogenetic tree drawn with TALE proteins of *P.vulgaris*, *A. thaliana*, *C.sativus*, *G.max*, *M. truncatula*, *O.sativa*, *S. bicolor*, *T. aestivum*, *V.vinifera* and *Z.mays plants*

Motif Regions of TALE Proteins

To test the reliability of the phylogenetic tree, motif compositions of TALE proteins were examined using the MEME database. Similar motif groups make it easy to determine a structural similarity between TALE proteins. According to this analysis, it was determined that proteins containing similar motif patterns in both species were in the same class in the phylogenetic tree.

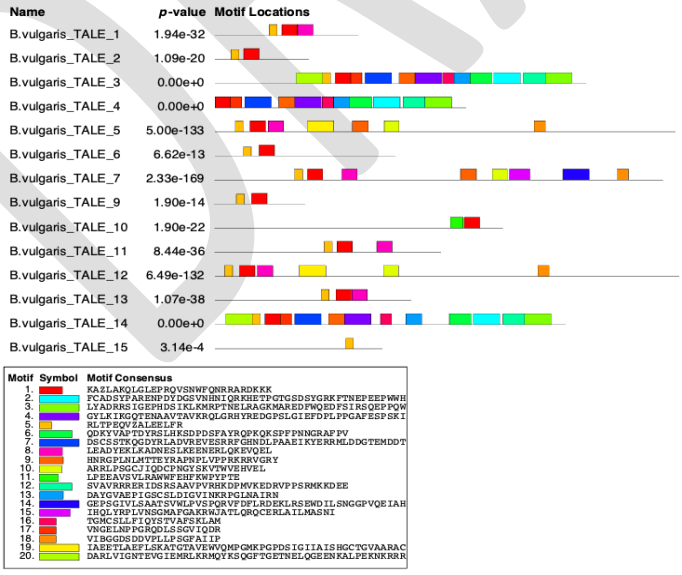


Figure 3. Distribution of predicted motifs in TALE genes

Exon and Intron Regions of TALE Proteins

Exon and intron regions of TALE proteins were determined from Gene Structure Server 2.0. Within the scope of the study, it was determined that most of these genes contain approximately equal amounts of exon and intron regions, but predominantly exon regions. Within the scope of the study, the most exon region was determined in Bvul-TALE-39.

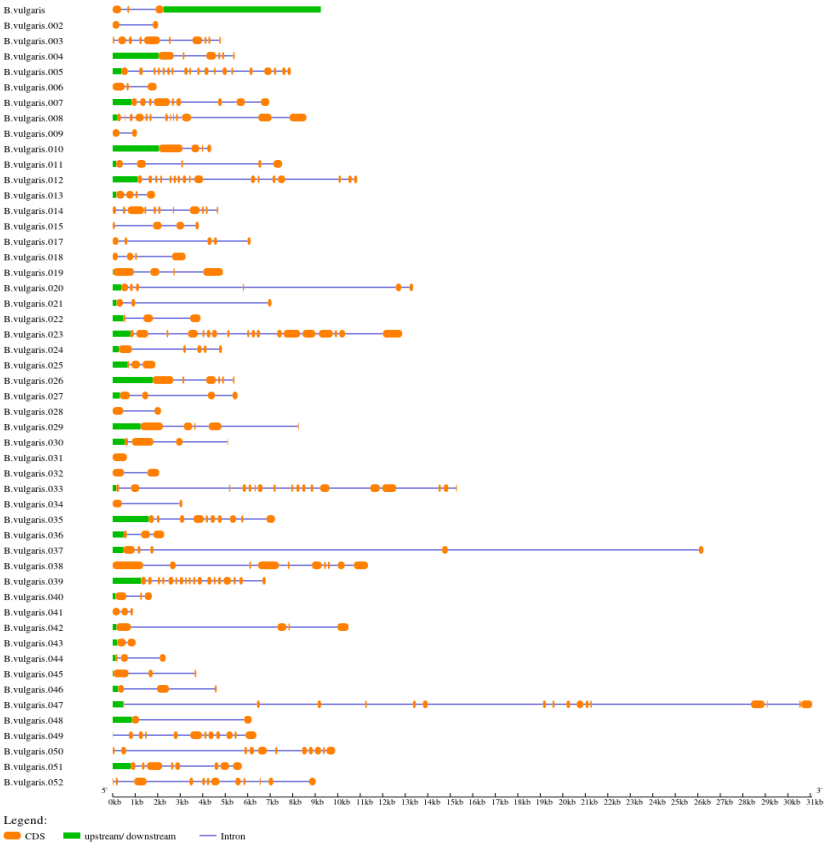


Figure 4. Number, length, and position of exon
s and introns in TALE genes

Homology Modeling of TALE Proteins

BLASTP scanning was performed with TALE proteins in Protein Data Bank[PDB] using the Phyre2 database and 3D homology modeling of these proteins was performed. This database was used to predict the structure and functions of TALE proteins. In this study, results with 90% and above reliability were taken as basis. Among the TALE proteins, the proteins with a reliability of 90% and above were determined.

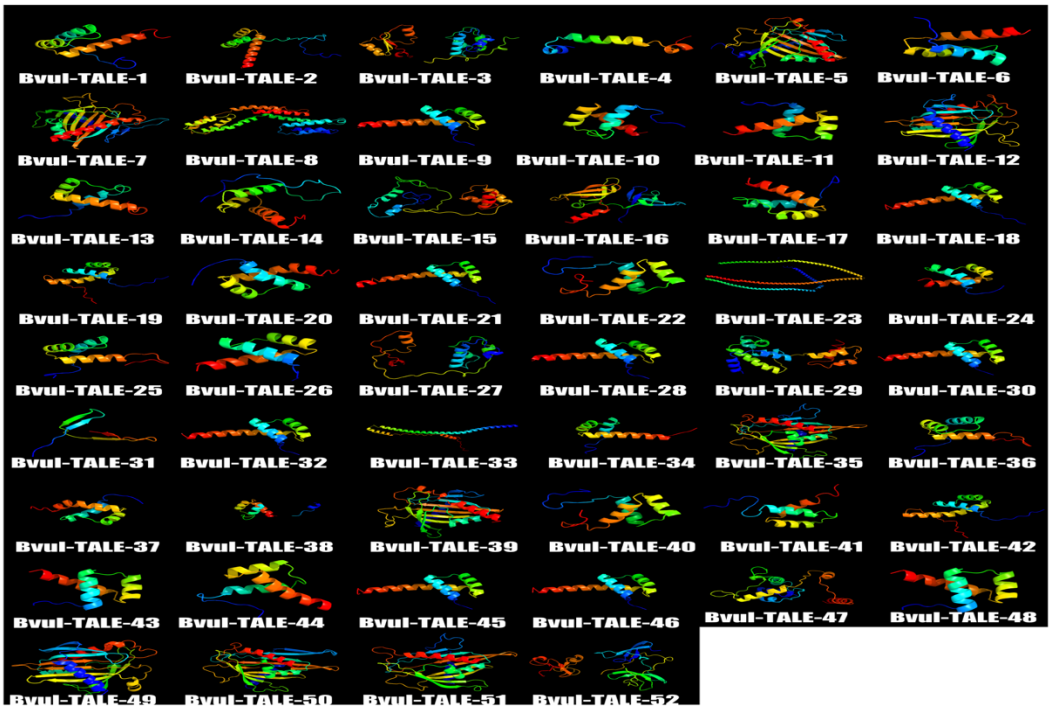


Figure 5. Homology images of TALE proteins

CONCLUSION

In this study, which was performed in silico with the TALE gene family in the *Beta vulgaris* genome, 52 TALE members were identified. The expression profiles of TALE genes revealed that these genes are expressed in different tissues and also showed that they can take part in various physiological functions during the development of the plant. It provides a basis for further functional research on the TALE gene family in *Beta vulgaris*. In conclusion, our study is shed light on future studies to study the functions of TALE genes and may make important contributions to the understanding of the evolution of TALE genes in different species.

ACKNOWLEDGEMENTS

This project was supported within the scope of TUBITAK 2209/A Support Program. We thank TÜBİTAK for their contribution.

REFERENCE

Anonymous, 2019a. Phytozome Database. (https:// phytozome.jgi.doe.gov/pz/portal.html), (Erişim tarihi: 18.09.2019).

Anonymous, 2019b. Hidden Markov Model (HMM). (http://www.ebi.ac.uk), (Erişim tarihi: 18.09.2019).

Anonymous, 2019c. Decrease Redundancy Tool. (http:// web.expasy.org/decrease_redundancy/), (Erişim tarihi: 18.09.2019).

Anonymous, 2019e. ProtParam. (http://web. expasy.org/protparam), (Erişim tarihi: 18.09.2019).

Bailey TL, Williams N, Misleh C, Li WW 2006. MEME: Discovering and analyzing DNA and Türkiye Tarımsal Araştırmalar Dergisi.

Belles-Boix, E., Hamant, O., Witiak, S.M., Morin, H., Traas, J., Pautot, V., 2006. KNAT6: an Arabidopsis homeobox gene involved in meristem activity and organ separation. Plant Cell, 18(8):1900–7.

Billeter M, Qian YQ, Otting G, Muller M, Gehring W, Wuthrich K., 1993. Determination of the nuclear magnetic resonance solution structure of an *Antennapedia* Homeodomain-DNA complex. J Mol Biology, 234(4):1084–94.

Burglin, T.R, Affolter M., 2016. Homeodomain proteins: an update. Chromosoma, 125(3):497–521.

Dohm JC, Minoche AE, Holtgräwe D, Capella-Gutiérrez S, Zakrzewski F, Tafer H, 2014. The genome of the recently domesticated crop plant sugar beet (Beta vulgaris). Nature, 505:546–9.

Evans, E., Messerschmidt, U., 2017. Sugar beets as a substitute for grain for lactating dairy cattle. Journal of animal science and biotechnology, 8(1), 1-10.

Goodstein, D. M., Shu, S., Howson, R., Neupane, R., Hayes, R. D., Fazo, J., Rokhsar, D. S., 2012. Phytozome: a comparative platform for green plant genomics. Nucleic acids research, *40*(D1), D1178-D1186.

Märländer, B., Hoffmann, C., Koch, H. J., Ladewig, E., Merkes, R., Petersen, J., Stockfisch, N., 2003. Environmental situation and yield performance of the sugar beet crop in Germany: heading for sustainable development. Journal of agronomy and crop science, 189(4), 201-226.

McGinnis W., Garber R.L., Wirz J., Kuroiwa A., Gehring W.J.,1984. A homologous protein-coding sequence in Drosophila homeotic genes and its conservation in other metazoans. Cell, 37(2):403–8.

Mukherjee K., Brocchieri L., Burglin T.R., 2009. A comprehensive classification and evolutionary analysis of plant Homeobox genes. Mol Biol Evol., 26(12):2775–94.

Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar S 2011. MEGA5: Molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Molecular Biology and Evolution, 28(10): 2731-2739.

Tzilivakis, J., Warner, D. J., May, M., Lewis, K. A., Jaggard, K., 2005. An assessment of the energy inputs and greenhouse gas emissions in sugar beet (Beta vulgaris) production in the UK. *Agricultural Systems*, *85*(2), 101-119

Voorrips RE 2002. MapChart: Software for the graphical presentation of linkage maps and QTLs. Journal of Heredity, 93(1): 77-78

Zabed, H., Faruq, G., Sahu, J. N., Azirun, M. S., Hashim, R., Nasrulhaq Boyce, A., 2014. Bioethanol production from fermentable sugar juice. *The scientific world journal*, *2014*.

Zhang Y, Nan J, Yu B, Yu B.2016. OMICS Technologies and Applications in Sugar Beet. Front Plant Sci., 7(900):1–11.

DRAFT

ORAL PRESENTATION

Characterization of expansin genes in alfalfa (*Medicago truncatula*) genome by bioinformatics methods

Özlem BAKIR BOĞA[*] *(ORCID: https://orcid.org* 0000-0002-1964-3271), Aylin KORKMAZ (*ORCID: https://orcid.org/* 0009-0000-3576-4607), Esabi Başaran KURBANOĞLU *(ORCID: https://orcid.org/0000-0002-7434-6309*)

[*1] Ataturk University, Science Faculty, Department of Biology, Erzurum, Turkey
[2,3] Ataturk University, Science Faculty, Department of Biology, Erzurum, Turkey

*Corresponding author e-mail: ozlembakir@atauni.edu.tr

Abstract

The aim of this study was to identify and characterize members of the expansin gene family in silico at a genome-wide level in *Medicago truncatula*. Expansin genes are distributed in chromosomes 1,2,3,4,5,6,7 and 8 in the *Medicago truncatula* genome. Expansin proteins were determined to have the amino acid sequence range of 123-457. Their molecular weights are between 13303.12-50362.36 DA, 33 of these proteins have acidic character, and 46 of them have basic character. As a result of the phylogenetic analyses carried out to elucidate the evolutionary status of the conserved region structures of expansin genes, it was determined that they were clustered in 5 different groups. Accordingly, it can be said that those located in similar regions in the classification have amino acid structures close to each other and therefore are in the same group. The most motifs were obtained from Med -Expansin-2,3,4 (10 motifs), and the least motif was from Med-Expansin-1 (1 motifs). Among the expansin proteins found in *Medicago truncatula.*, 5 Expansin proteins with a reliability of 90% and above were determined. The highest exon and intron numbers were obtained in Med-Expansin-24, 27. We believe that the results obtained from this study may be useful for future functional studies on the expansin gene family in *Medicago truncatula.*

Keywords: Bioinformatic, Expansin, in silico analysis, *Medicago truncatula*

INTRODUCTION

Expansins are proteins localized in the cell walls of plants and are quite small in size, with an approximate molecular weight of 25-30 kDa. They were first isolated from cucumber coleoptiles in 1992 (McQueen-Mason et al., 1992). These proteins can break the hydrogen bonds between cellulose microfibrils by acting on the pectin matrix, or it is hypothesized that they are involved in cell wall modification by making the carbohydrate polymers more accessible to the enzymes in the cell wall (Cosgrove, 1998). It is known that this family of proteins plays a role in plant cell growth and fruit softening. In a study on tomato, it was proven that the LeExp1 gene is expressed. Accordingly, there is a correlation between the degradation of expansin proteins and cell wall components (Rose et al., 1997). Therefore, when the fruit of the plant begins to soften, that is, when the cell wall begins to break down, this gene reaches its maximum level (Wei et al., 2011). According to another study to support all data; It was determined that the fruits in which the expansin protein was expressed were softer than the control samples, and these reactions were activated before the ripening stage (Zhang vd., 2017). Expansin transcripts were found in non-climacteric fruits such as grapes and strawberries, and it was determined that these fruits also participated in softening and carbohydrate degradation reactions (Backiyarani vd., 2022).In addition to all these effects of the expansin protein family, it is thought to be a factor in the emergence of root hair, meristem functions and many important developments. The aim of this study is to identify expansin gene family members in the *Medicago truncatula* genome for the first time, using in silico methods, at a genome-wide scale. In this study, the phylogenetic relationships of the expansin gene family, their distribution in the genome, gene structure arrangement, composition of motifs, and 3-dimensional protein structures were characterized. Identification and functional analysis of the *Medicago truncatula* expansin gene family provide a basis for a better understanding of the genetic mechanism of tolerance to biotic and abiotic stresses in the *Medicago truncatula* genome.

DRAFT

MATERIALS AND METHODS

Identification of Expansin Genes in the *Medicago truncatula* Genome

The protein sequences of the expansin gene family in the *M.turuncatula* genome were obtained from the Phytozome database v12.1 (Anonymous, 2018a) using the Pfam Access Number for expansin proteins (PF03330) obtained from the Pfam database. In order to identify all possible expansin proteins in the bean genome, both the Phptozome database's v12.1 blastp and latent Markov model (HMM) (Anonymous, 2018b) search were scanned in the *M.turuncatula* genome with assumed parameters. Unrelated arrays were obtained using the "decrease redundancy tool Anonymous (Anonymous, 2018c). Presence of expansin proteins in related sequences was investigated using HMMER (Anonymous, 2018d) database. The molecular weight and theoretical isoelectric point (pI) of the obtained expansin proteins were determined using the ProtParam tool Anonymous (Anonymous, 2018e).

Sequence Alignment, Phylogenetic Analyzes and Determination of Conserved Motifs

Sequence alignment was done using ClustalW via MEGA program. Sequence alignment was used for phylogenetic tree plotting. The phylogenetic tree was created using the Maximum Neigbour-joining method in the MEGA7 program and visualized using the iTOL database. iTOL database is used to create a new phylogenetic tree and to visualize the phylogenetic tree to make the relationship between evolutionary processes more evident. Exon-intron analysis was performed with the GSDS2.0 (http://gsds.cbi.pku.edu.cn/) database so that the phylogenetic tree could give as accurate results as possible. Exon-intron regions were created using the genomic sequences of expansin sequences and CDS sequences. This program was used because the upgraded GSDS 2.0 has more annotation features and formats with a newly designed face. MEME Suite (MEME 4.11.1; http:/meme-suite.org/) database was used to identify conserved motifs. While performing the analysis, care was taken to ensure that the motif widths were optimum ≥2 and ≤300 by taking the maximum number of 5 motifs to determine the motifs. The motifs determined in the MEME Suite program were taken and the Publisher program was used to combine the motifs, and thus the preserved motif regions were determined (Bailey et al., 2009).

Determination of Chromosome Distribution

Using the obtained data, chromosome distribution was found via MapChart and the results were determined (Voorrips, 2002).

Homology Modeling of Expansin Proteins

Using previously identified protein sequences for homology modeling, the 3D structure of the proteins was predicted with Phyre2. The reliability of the model was verified with the specified numerical data (Kelley, 2009).

RESULTS and DISCUSSION

Bioinformatics Analysis of Expansin Proteins

As a result of the analysis using the sequences of expansin proteins identified in *Medicago truncatula*, 79 expansin genes were identified. These identified expansin genes were named Med-Expansin using their Latin names. The expansin proteins identified in the *M.truncatula* genome are 123-457 amino acids long, their molecular weights are between 13303.12-50362.36 DA, and 33 of these proteins have acidic character, and 46 of them have basic character (Table 1).

DRAFT

ID	*Medicago truncatula* Genomic Database Identifier	Physical position on *Medicago truncatula* genome			Protein length (aa)	pI	Molecular weight (Da)	Instability index	Stable or unstable
		Chromosome	Start position (bp)	End Position (bp)					
Med-exp-1	Medtr3g107770.2	3	49724057	49730789	143	5.47	15466.65	46.00	unstable
Med-exp-2	Medtr7g013150.1	7	3744454	3745651	253	9.07	27955.02	23.91	stable
Med-exp-3	Medtr6g012520.1	6	3813981	3815849	244	9.38	26596.96	32.66	stable
Med-exp-4	Medtr5g075320.2	5	31998952	32001184	251	8.93	27089.50	33.30	stable
Med-exp-5	Medtr0899s0010.1	scaffold	1122	3500	271	5.74	28956.69	39.46	stable
Med-exp-6	Medtr5g079950.1	5	34206808	34208293	259	9.64	28046.79	33.25	stable
Med-exp-7	Medtr2g090255.1	2	38354583	38356267	252	8.26	28031.02	39.79	stable
Med-exp-8	Medtr0940s0140.1	scaffold	56286	58582	256	8.67	28477.73	33.12	stable
Med-exp-9	Medtr7g077050.1	7	29101738	29102299	186	6.92	20237.68	38.35	stable
Med-exp-10	Medtr5g011210.1	5	3183138	3184843	263	9.39	28956.22	34.29	stable
Med-exp-11	Medtr8g031150.1	8	11640536	11641359	146	9.61	16064.58	59.13	unstable
Med-exp-12	Medtr1g026020.1	1	8373565	8375989	255	8.89	27829.61	39.48	stable
Med-exp-13	Medtr2g095060.1	2	40610260	40611249	219	4.76	23306.87	41.62	unstable
Med-exp-14	Medtr7g076770.1	7	28956484	28957064	189	4.49	20598.79	55.63	unstable
Med-exp-15	Medtr8g031170.1	8	11654481	11655060	192	8.72	20898.11	34.74	stable
Med-exp-16	Medtr7g077010.1	7	29078042	29078612	189	6.07	20692.28	44.72	unstable
Med-exp-17	Medtr3g094040.1	3	42965157	42967247	253	8.37	27041.34	28.66	stable
Med-exp-18	Medtr2g095130.1	2	40627562	40628355	220	5.45	23140.81	52.94	unstable
Med-exp-19	Medtr7g011060.1	7	2865389	2868018	242	9.32	26138.29	38.04	stable
Med-exp-20	Medtr0008s0120.1	scaffold	44365	45786	199	9.03	22242.47	18.63	stable
Med-exp-21	Medtr4g028640.1	4	9628638	9630991	267	7.44	29082.95	41.38	unstable
Med-exp-22	Medtr6g083200.1	6	31171216	31175338	202	9.26	22345.12	19.40	stable
Med-exp-23	Medtr7g077090.1	7	29110793	29111363	189	7.98	20784.43	46.81	unstable
Med-exp-24	Medtr4g098930.1	4	40866851	40868963	254	4.46	27386.55	40.59	unstable
Med-exp-25	Medtr4g063975.1	4	23856594	23859315	258	9.64	28188.13	36.18	stable
Med-exp-26	Medtr2g090270.1	2	38369375	38371921	260	7.59	28424.21	36.04	stable
Med-exp-27	Medtr6g083270.1	6	31171216	31175338	457	6.22	50362.36	39.96	stable
Med-exp-28	Medtr6g083290.1	6	31189130	31192806	316	5.80	35170.36	38.04	stable
Med-exp-29	Medtr0118s0070.1	scaffold	38899	41967	275	5.25	29361.93	39.81	stable
Med-exp-30	Medtr7g077000.1	7	29072943	29073336	130	5.42	14114.53	46.16	unstable
Med-exp-31	Medtr4g036290.1	4	11849831	11850398	188	6.91	20614.11	40.58	unstable
Med-exp-32	Medtr7g111010.1	7	29078042	29078612	254	9.47	27988.06	36.52	stable
Med-exp-33	Medtr4g124340.1	4	51423187	51424794	284	9.22	30662.04	25.06	stable
Med-exp-34	Medtr7g012350.1	7	3617234	3617986	188	5.57	20376.86	40.81	unstable
Med-exp-35	Medtr4g099400.1	4	41244356	41245315	179	8.68	20196.96	37.63	stable
Med-exp-36	Medtr2g097030.1	2	41488820	41491699	260	9.63	28473.49	32.25	stable
Med-exp-37	Medtr8g031160.1	8	11645102	11645474	123	4.66	13303.12	36.62	stable
Med-exp-38	Medtr1g080800.1	1	35952384	35953845	146	5.57	15872.62	7.87	stable
Med-exp-39	Medtr7g012340.1	7	3618695	3619262	188	5.48	20456.73	37.75	stable
Med-exp-40	Medtr2g095170.1	2	40640889	40641393	167	4.67	17906.20	46.64	unstable
Med-exp-41	Medtr0122s0020.1	scaffold	5904	10805	266	8.31	29121.35	36.82	stable
Med-exp-42	Medtr6g068780.1	6	24724434	24725013	133	9.21	14601.88	63.13	unstable
Med-exp-43	Medtr7g076990.1	7	4550673	4550735	188	5.57	20496.97	45.07	unstable
Med-exp-44	Medtr3g107500.1	3	49581017	49581905	151	9.44	16657.28	60.58	unstable
Med-exp-45	Medtr4g102450.1	4	42465410	42467447	262	8.83	28520.47	30.72	stable
Med-exp-46	Medtr7g077040.1	7	29098071	29098536	128	5.01	13999.60	48.73	unstable
Med-exp-47	Medtr5g041700.1	5	18299810	18302055	248	9.50	26586.06	33.39	stable
Med-exp-48	Medtr4g036410.1	4	11854007	11854576	170	8.01	18695.90	53.29	unstable
Med-exp-49	Medtr2g095080.1	2	40616663	40617795	222	4.89	23351.96	40.32	unstable
Med-exp-50	Medtr5g013440.1	5	4262828	4265000	252	5.83	28204.92	32.86	stable
Med-exp-51	Medtr7g013140.1	7	3731568	3732586	251	5.85	27320.02	21.21	stable
Med-exp-52	Medtr1g080820.1	1	35952384	35953845	144	8.19	15899.83	12.09	stable
Med-exp-53	Medtr7g077110.1	7	29115836	29116668	188	5.92	20449.78	41.94	unstable
Med-exp-54	Medtr8g058990.1	8	20521997	20523500	253	5.10	28191.97	17.12	stable
Med-exp-55	Medtr8g031120.1	8	11621962	11622749	138	9.45	15116.43	59.24	unstable
Med-exp-56	Medtr4g099380.1	4	41237358	41240904	282	9.27	32010.63	33.82	stable
Med-exp-57	Medtr6g083310.1	6	31205474	31206675	167	8.14	18499.20	32.48	stable
Med-exp-58	Medtr3g435430.1	3	11593732	11593257	247	8.92	26302.76	29.57	stable
Med-exp-59	Medtr7g076960.1	7	29066427	29067196	185	6.85	20206.89	41.76	unstable
Med-exp-60	Medtr1g085460.1	1	38164771	38166135	257	9.35	27969.02	30.78	stable
Med-exp-61	Medtr1g080810.1	1	35946158	35948164	144	8.88	16210.42	14.19	stable
Med-exp-62	Medtr7g013120.1	7	3731568	3732586	253	8.36	27971.01	22.84	stable
Med-exp-63	Medtr3g466760.1	3	27478443	27479431	260	8.37	28072.60	30.53	stable
Med-exp-64	Medtr7g076920.1	7	29057950	29058722	188	5.57	20390.92	41.76	unstable
Med-exp-65	Medtr4g036300.1	4	11851795	11851862	188	5.57	20496.97	45.07	unstable
Med-exp-66	Medtr7g110990.1	7	45507353	45507759	239	9.54	26271.93	35.63	stable
Med-exp-67	Medtr2g090265.1	2	38363046	38365828	258	8.35	28249.04	35.67	stable
Med-exp-68	Medtr7g115220.1	7	47582959	47584201	202	5.73	21670.10	19.94	stable
Med-exp-69	Medtr7g076970.1	7	29068847	29069431	189	7.46	20738.40	47.00	unstable
Med-exp-70	Medtr7g061720.1	7	22311666	22315087	258	9.69	27702.58	28.70	stable
Med-exp-71	Medtr4g099370.1	4	41232062	41233559	251	5.02	28020.52	35.46	stable
Med-exp-72	Medtr4g081950.1	4	31830762	31833405	284	8.97	30617.64	36.27	stable
Med-exp-73	Medtr8g464350.1	8	22732659	22734252	254	6.57	28538.61	22.04	stable
Med-exp-74	Medtr5g076950.1	5	34206808	34208293	266	8.40	29121.94	44.37	unstable
Med-exp-75	Medtr4g075790.1	4	29007806	29009773	260	8.24	28436.35	39.30	stable
Med-exp-76	Medtr3g094060.1	3	42973625	42975332	253	8.57	27068.35	25.38	stable
Med-exp-77	Medtr1g080780.1	1	35929462	35929846	141	5.79	15448.45	38.32	stable
Med-exp-78	Medtr3g466790.1	3	27487699	27489977	254	7.55	27406.73	32.47	stable
Med-exp-79	Medtr3g064510.1	3	29068578	29070728	286	6.06	31165.12	46.18	unstable

Table 1. Information about the expansin genes found in the *M.truncatula* genome

According to the results of the chromosomal distribution studies, it was determined that the chromosome with the highest number of expansin genes was the 7th chromosome with 18 genes, and the chromosomes with the least genes were the 1st, 5th, and 6th chromosomes with 5 genes.

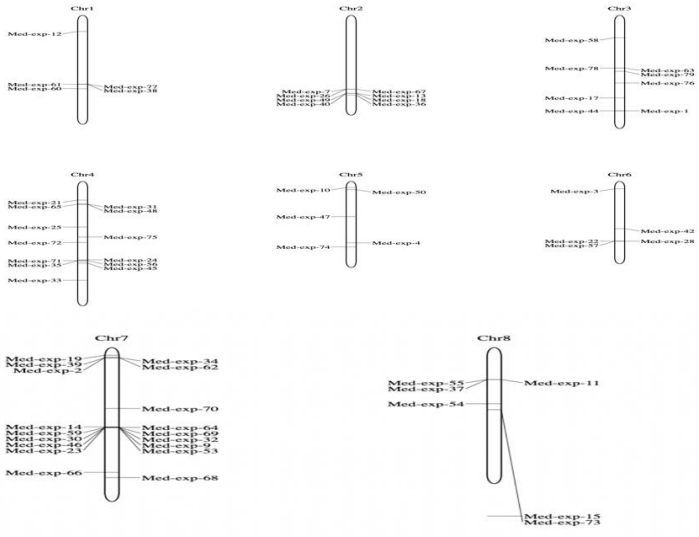


Figure 1. Distribution of expansin genes in *M.truncatula* chromosomes

Phylogenetic Analyzes

Phylogenetic tree analysis was performed using the MEGA7 [MolecularEvolutionaryGenetic Analysis] program to determine the evolutionary relationships between expansin genes. Maximum neighbor-joining approach was used in this analysis. According to phylogenetic tree analysis, expansin genes were divided into 5 main groups. Sequences were aligned with ClustalOmega[Clustal W] in MEGA7 and an unrooted phylogenetic tree was constructed. The created phylogenetic tree was colored in a different color for each family using the ITOL database (Tamura et al., 2011).

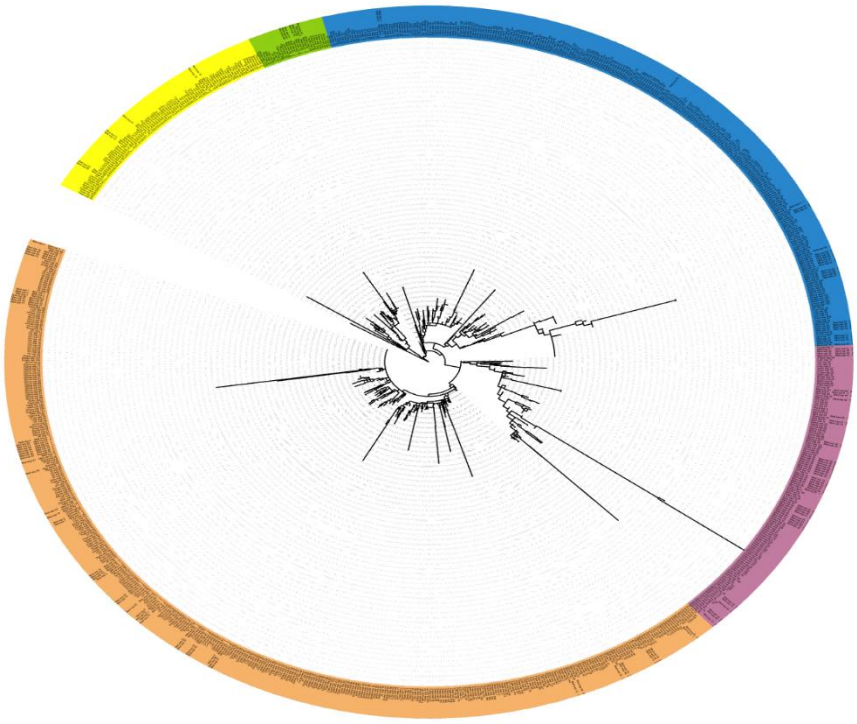


Figure 2. Phylogenetic tree drawn with expansin proteins of *P.vulgaris, A. thaliana, C.sativus, G.max, M. truncatula, O.sativa, S. bicolor, T. aestivum, V.vinifera* and *Z.mays plants*

Motif Regions of Expansin Proteins

To test the reliability of the phylogenetic tree, motif compositions of expansin proteins were examined using the MEME database. Similar motif groups make it easy to determine a structural similarity between expansin proteins. According to this analysis, it was determined that proteins containing similar motif patterns in both species were in the same class in the phylogenetic tree.

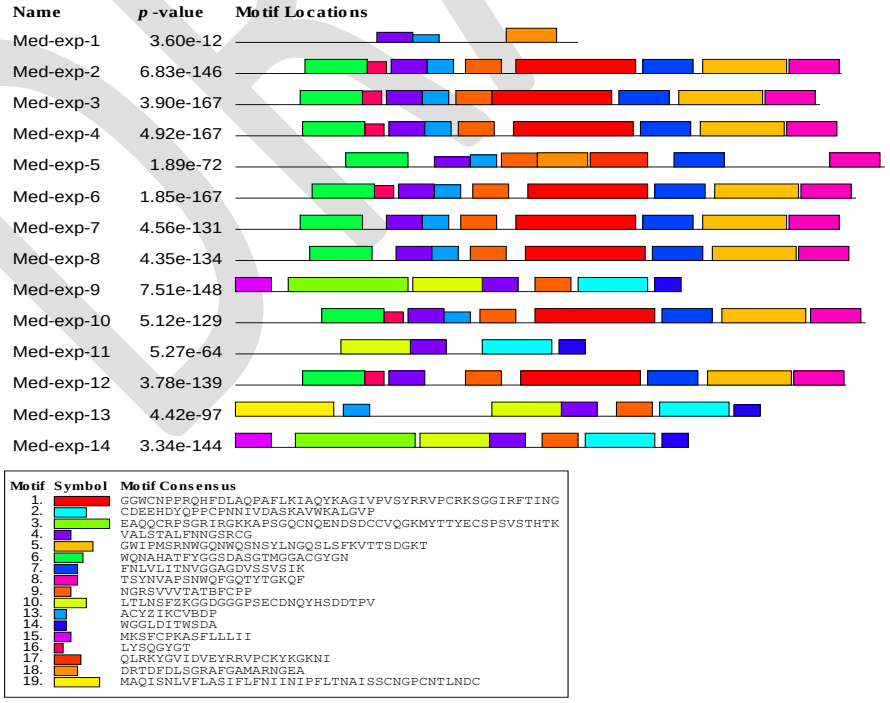


Figure 3. Distribution of predicted motifs in expansin genes

DRAFT

Exon and Intron Regions of Expansin Proteins

Exon and intron regions of expansin proteins were determined from Gene Structure Server 2.0. Within the scope of the study, it was determined that most of these genes contain approximately equal amounts of exon and intron regions, but predominantly exon regions. Within the scope of the study, the most exon region was determined in Med-Expansin-24, 27.

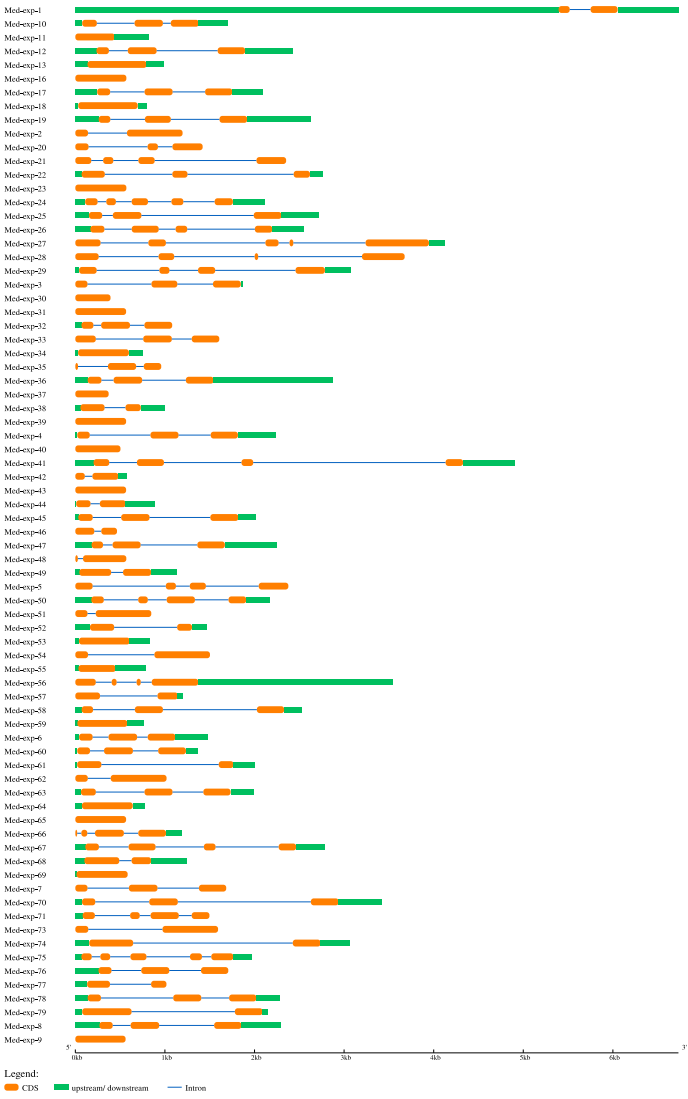


Figure 4. Number, length, and position of exon s and introns in expansin genes

Homology Modeling of Expansin Proteins

BLASTP scanning was performed with expansin proteins in Protein Data Bank[PDB] using Phyre2 database and 3D homology modeling of these proteins was performed. This database was used to predict the structure and functions of expansin proteins. In this study, results with 90% and above reliability were taken as basis. Among the expansin proteins, the proteins with a reliability of 90% and above were determined.



Figure 5. Homology images of expansin proteins

CONCLUSION

In this study, which was performed in silico with the expansin gene family in the *M.truncatula* genome, 79 expansin members were identified. The expression profiles of expansin genes revealed that these genes are expressed in different tissues and also showed that they can take part in various physiological functions during the development of the plant. It provides a basis for further functional research on the expansin gene family in *M.truncatula*. In conclusion, our study is shed light on future studies to study the functions of expansin genes and may make important contributions to the understanding of the evolution of expansin genes in different species.

ACKNOWLEDGEMENTS

This project was supported within the scope of TUBITAK 2209/A Support Program. We thank TÜBİTAK for their contribution.

REFERENCES

Anonymous, 2019a. Phytozome Database. (https:// phytozome.jgi.doe.gov/pz/portal.html), (Erişim tarihi: 18.09.2019).

Anonymous, 2019b. Hidden Markov Model (HMM). (http://www.ebi.ac.uk), (Erişim tarihi: 18.09.2019).

Anonymous, 2019c. Decrease Redundancy Tool. (http:// web.expasy.org/decrease_redundancy/), (Erişim tarihi: 18.09.2019).

Anonymous, 2019e. ProtParam. (http://web. expasy.org/protparam), (Erişim tarihi: 18.09.2019).

Backiyarani, S., Anuradha, C., Thangavelu, R., Chandrasekar, A., Renganathan, B., Subeshkumar, P., Uma, S., 2022. Genome-wide identification, characterization of ekspansin gene family of banana and their expression pattern under various stresses. *3 Biotech*, *12*(4), 1-21.

Bailey TL, Williams N, Misleh C, Li WW 2006. MEME: Discovering and analyzing DNA and Türkiye Tarımsal Araştırmalar Dergisi.

Cosgrove, D.J.,1998. Cell wall loosening by ekspansins. Plant Physiology, 118, 333–339.

McQueen-Mason, S.J., Durachko, D.M., Cosgrove, D.J.,1992. Two endogenous proteins that induce cell wall extension in plants. Plant Cell 4, 1425–1433.

Rose, J.K.C., Lee, H.H., Bennett, A.B.,1997. Expression of a divergent ekspansin gene is fruit-specific and ripening regulated. Proc Natl Acad Sci USA, 94:5955–5960.

Tamura, K., Peterson, D., Peterson, N., Stecher, G., Nei, M., Kumar, S., 2011. MEGA5: Molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Molecular Biology and Evolution, 28(10): 2731-2739.

Voorrips RE 2002. MapChart: Software for the graphical presentation of linkage maps and QTLs. Journal of Heredity, 93(1): 77-78

Wei, P.C., Zhang, X.Q., Zhao, P., Wang, X.C., 2011. Regulation of stomatal opening by the guard cell ekspansin AtEXPA1. Plant Signal Behav 6(5):740–742

Zhang, H., Cao, Y., Shang, C., Li, J., Wang, J., Wu, Z., Hu, B. 2017. Genome-wide characterization of GRAS family genes in Medicago truncatula reveals their evolutionary dynamics and functional diversification. *PLoS One*, *12*(9), e0185439.

DRAFT

ORAL PRESENTATION

Utilization of Non-Wood Forest Products (NWFPs) as a Food in Turkey and World

Levent Gülüm[1*] (*ORCID: https://orcid.org/0000-0001-8564-1664*), Orhan Kelleci[2] (*ORCID: https://orcid.org/0000-0003-4501-0854*) , Süheyla Esin Köksal[2] (*ORCID: https://orcid.org/0000-0001-7970-8412*)

[*1]Bolu Abant Izzet Baysal University, Mudurnu Sureyya Astarcı vocational School, Plant and Animal Production, Bolu, Turkey.
[2] Bolu Abant Izzet Baysal University, Mudurnu Sureyya Astarcı vocational School, Forestry, Bolu, Turkey.

*Corresponding author e-mail: leventgulum@ibu.edu.tr

Abstract

In this study, the potential utilization prospects of non-wood forest products (NWFPs) as food sources, their various forms of utilization, and their significance as sustenance for communities have been emphasized. In contemporary society, the assessment of NWFPs products as potential food sources tends to be underemphasized. These resources, inherent to their natural habitats, function as both nutritional provisions and sources of economic livelihood for local communities. To optimize the utility and output of non-wood forest products, it is imperative to advance their utilization and production opportunities.
Research efforts should be directed towards forest-related subjects, concentrating particularly on the advancement of nutritionally valuable forest products. This strategic focus extends to safeguarding the integrity and value of non-wood forest products. Encouragingly, organizations such as non-governmental entities, educational institutions, hospitals, and health centers ought to be motivated to champion the merits of non-wood forest products.
Creating collaborative platforms across sectors encompassing the environment, health, development, and agriculture is pivotal. This step is essential to seamlessly incorporate knowledge pertaining to nutritious forest foods and their conservation into the broader context of national nutrition strategies and programs. These platforms serve as conduits for the widespread dissemination of strategies aimed at harnessing the nutritional potential embedded within high-value tree biodiversity. In doing so, they contribute significantly to the domains of food security, nutrition, and conservation.

Keywords: Forest, Non-wood forest product, Food, Free fruit

INTRODUCTION

Non-wood forest products are special products or services naturally found on Earth that are distinct from timber raw materials. A portion of these products are cultivated and produced through agricultural activities due to their high demand. Thyme consumed as tea, and marigold used as an ornamental plant, are examples of plants that are both harvested from natural habitats and cultivated through agricultural practices in Turkey. However, the majority of non-wood forest products are still collected from nature and consumed as such (Ok & Tengiz, 2018).

Non-wood forest products" (NWFP) is a comprehensive term that encompasses various animal and plant resources, excluding timber, obtained from forests, wooded lands, and trees beyond forests. These products serve both market and subsistence needs, spanning from wild-harvested resources to cultivated plants in agroforestry systems, and are grouped into three main categories based on their functions and utilization purposes (Ozugurlu & Duzgun, 2000) (Table 1).

Due to its geographical location, Turkey possesses a rich flora compared to other European countries. There are around 9,500 species distributed in our country, with about 3,000 of these species displaying endemic characteristics. Non-wood forest products (NWFPs), which play a significant role in global production, also include some endemic species. As of 1990, approximately 97% of Turkey's forest product exports consist of

NWFPs. However, the sustainability of these resources is at risk, mainly due to improper utilization and lack of supervision (Türker et al., 2019).

Table 1. Some Non-wood forest products

Main category	Sub-categories	Examples of products
Non-wood plant products	Food	Wild and domesticated products, weeds, fruits, flowers,
	Forage	Food for livestock and wildlife, including birds, fishes,
	Pharmaceuticals	Drugs, salves, lotions, purgatives etc.
	Toxins	For hunting, ordeal poisons, salves, ointments, lotions,
	Aromatics	Essential oils for cosmetic and perfume industry, unguent,
	Biochemicals	Non-edible fats and oils, waxes, gums and latex, dyes,
	Fiber	Cloth, matting, cordage, basketry, brooms, cork etc.
	Wood	Wood for handicrafts
	Ornamentals	Plants for aesthetically pleasing horticulture and amenity
Wild Animal Products	Mammals	Meat, horn, skins, wool, bone etc.
	Birds	Meat, hides, skins, wool, bone pharmaceuticals etc.
	Fishes	Meat, eggs, feathers, edible nests, guano etc.
	Reptiles	Food, skins, shell, toxins, etc.
	Invertebrates	Plant exudates (manna), honey, wax, silk lac etc.
Services and Functions of Forests	Habitat	Grazing, shade, and shelter for animals
	Soil improvement and	Green manure, humus, N-fixation, hedges, soil
	Protected areas	Non-consumptive use (Tourism/recreation, wildlife
		Consumptive use (hunting, shooting, fishing etc.)
		Aesthetic, scenic, and historic sites etc.

Global initiatives aiming to enhance food security and well-being emphasize the significance of a well-rounded, micronutrient-dense diet. Incorporating adequate micronutrient-rich foods is vital for sustainable strategies that address micronutrient deficiencies, which can impede child development and heighten health risks, particularly among disadvantaged populations in developing nations (Black et al., 2013). This shift towards prioritizing dietary quality alongside yield and energy availability has prompted discussions on reevaluating agricultural policies, with forests and trees potentially playing a crucial role in enhancing nutrition within the food system and wider landscape (FAO, 2013; Ruel & Alderman, 2013; Vinceti et al., 2013).

Amidst the challenges of climate change, biodiversity loss, urbanization, and socioeconomic disparities, the capacity of the current global food system to sustainably provide for a projected population of 9 billion by 2050 is uncertain, underscoring the pressing need for collaborative efforts to ensure worldwide food and nutrition security (Hunter & Fanzo, 2013).

With around 40% of Indonesia's population, approximately 100 million people, experiencing micronutrient deficiencies, forests and trees in remote areas play a vital role in providing nutritious food. Examining data from the 2003 Indonesia Demographic Health Survey and the Indonesian Ministry of Forestry, the study explores the relationship between various tree-dominated land classes and the consumption of micronutrient-rich foods amidst the escalating loss of forests and tree-based systems.

Promoting the consumption of micronutrient-rich foods, including diverse fruits, vegetables, pulses, and certain animal products, is recognized as a viable, cost-effective, and sustainable approach to enhancing nutrient quality and dietary habits, while addressing multifaceted aspects of food production and consumption (Johns & Sthapit, 2004; Stephenson et al., 2010; Tontisirin et al., 2002). The challenge lies in creating food systems that are environmentally and economically sustainable, culturally sensitive, and nutritionally sound, while ensuring that both consumers and producers are equipped with the requisite information to make optimal dietary choices (Johns et al., 2013).

Forests and trees play a pivotal role in enhancing food security and nutrition in three key ways. Primarily, they indirectly bolster food security through ecosystem services benefiting agriculture (Foresight UK, 2011; Godfray et al., 2010; Sunderland, 2011). Additionally, smallholder farming practices reliant on trees, like swidden cultivation and agroforestry, tend to yield more diverse diets, indicative of better dietary quality (Kennedy et al., 2011; Pimentel et al., 1997; Ruel, 2003)(8-10). Lastly, individuals residing close to or within forested areas can directly access wild foods from these ecosystems, bypassing market intermediaries (C. Colfer, 2012; Dounias & Froment, 2011).

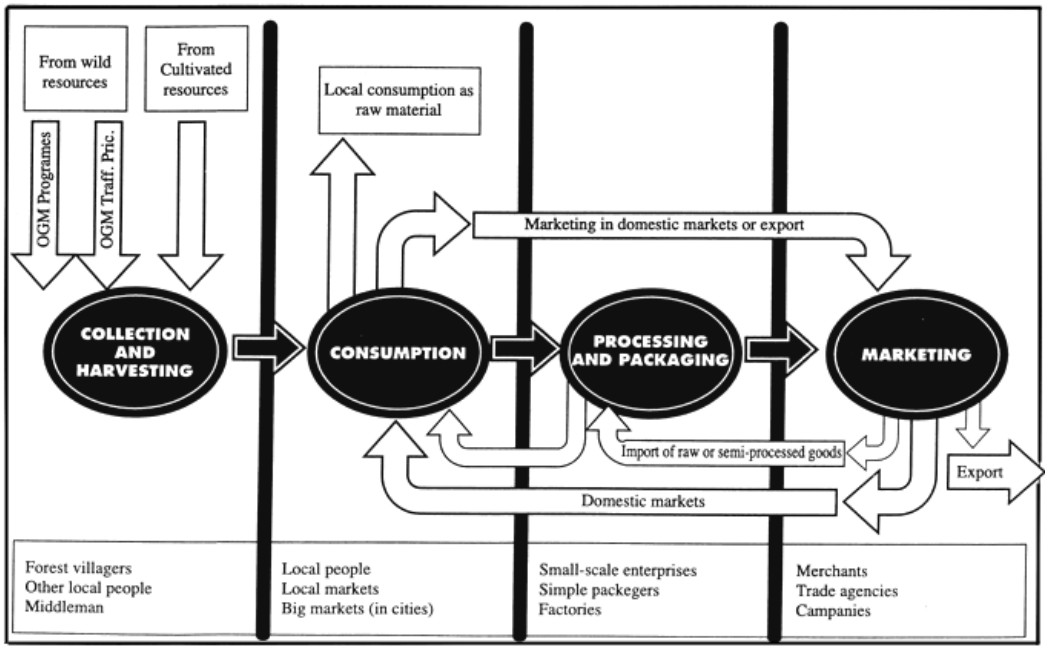


Figure 1. Flow chart of operation and treatment processes of NWFPs in Turkey (Ozugurlu & Duzgun, 2000)

Throughout history, forest inhabitants have exemplified how human lifestyles and genetic adaptations can harmonize with dynamic forest ecosystems. The concept of the "paleodiet" is often cited as a model for contemporary health promotion and disease prevention, even in industrialized nations. However, as forest ecosystems face mounting pressure from economic growth and industrialization, leading to deforestation, altered resource availability, and ecological shifts, forest dwellers encounter challenges in aligning their socioeconomic and cultural systems, ultimately affecting their ecological well-being and highlighting the complex trade-offs between modernity and traditional lifestyles (Dounias & Froment, 2011).

Pimentel et al. (1997) evaluated the contribution of forests and non-timber products to the food systems of developing nations, revealing that over 300 million individuals rely on forests for sustenance and livelihood each year. Additionally, forests safeguard land, water, and biodiversity, while supporting agricultural and environmental productivity, with an annual harvest value of around $90 billion attributed to non-timber products.

Sustainable diets

Sustainable diets prioritize low environmental impact, food security, and health across current and future generations, by being culturally appropriate, economically fair, nutritionally complete, and environmentally friendly while valuing biodiversity and optimizing resources (Burlingame & Dernini, 2012).

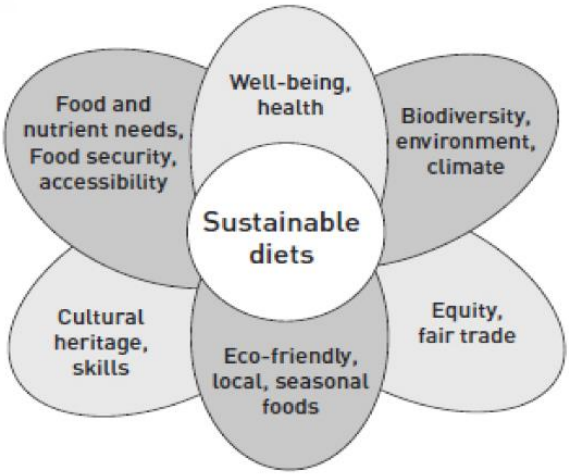


Figure 2. The many dimensions of a sustainable diet (Burlingame & Dernini, 2012)

Forest foods, encompassing items derived from trees, herbs, mushrooms, and animals, play a pivotal role in enhancing food security by offering accessible, cost-effective, and nutritionally rich sustenance (Dansi et al., 2008; Powell et al., 2011; Vinceti et al., 2008). Although few communities rely solely on forest foods for a comprehensive diet (Bailey et al., 1989; Butler, 2012), these resources provide essential micronutrients, supplementing caloric intake during lean seasons and times of agricultural scarcity (Faye et al., 2010; Humphry et al., 1993; Moreno-Black & Somnasang, 2000). By cultivating a diverse range of indigenous fruit tree species within agroforestry systems, aligned with fruit harvest seasons, a continuous supply of crucial nutrients can be maintained, contributing to sustained nutritional well-being (Jamnadass et al., 2011; Vinceti et al., 2013).

While forest foods are not a universal solution to global food security and nutrition challenges, they can play a significant role in specific geographic areas. Despite their contribution to the global food supply, the inclusion of wild foods in official economic statistics is recent and their reliability remains uncertain due to data primarily originating from the informal sector, and the lack of comprehensive inventories and standardized methodologies for documenting wild food consumption calls for caution in interpreting global estimates (Heywood et al., 2013; Penafiel et al., 2011; Scoones et al., 1992).

Forest Foods Quality

Forests and trees offer essential micronutrients often lacking in diets of developing nations, with significant health and developmental benefits. Nutrients like vitamin A, critical for immune function, are found in green leafy vegetables and orange-colored fruits; while iron, zinc, and vitamin B12, essential for growth and cognitive development, are present in animal source foods like bushmeat, forest-gathered leafy greens, and legumes, holding lifelong implications for health and socio-economic outcomes (Belem et al., 2009; Jansen van Rensburg et al., 2004; Kennedy et al., 2003; Nutrition, 2004).

CONCLUSION

In modern times, the evaluation of non-wood forest products (NWFPs) as food sources is often overlooked. NWFPs that naturally grow in their habitats serve as both a nutritional resource and an economic income for local communities. It is crucial to enhance and increase the utilization and production possibilities of non-wood forest products.

Amidst the uncertainties posed by a projected population of 9 billion by 2050, addressing global food demands is a complex challenge. "Sustainable diets" offer a comprehensive approach to ensure equitable access to nutritious food while navigating multifaceted issues such as poverty reduction, environmental preservation, well-being improvement, and cultural heritage preservation, aligning with the diverse challenges of the modern world. Forest foods, derived from trees, herbs, mushrooms, and animals, significantly contribute to food security by providing accessible and nutrient-rich sustenance, with their diverse nutritional benefits being particularly relevant during periods of scarcity and lean seasons, while the cultivation of indigenous fruit tree

species within agroforestry systems offers a continuous supply of essential nutrients to ensure ongoing nutritional well-being.

In order to benefit more effectively from non-wood forest products, analysis and documentation of their nutritional composition, digestibility and bioavailability, the effect on the nutritional value of certain forest foods should be determined. It is necessary to compare these systems with the nutrition and food systems of other countries and regions in terms of resilience, health, cost-effectiveness, and sustainability to explain and measure the sustainability of forest-based diets. Research on forest-related topics should be supported. Emphasis should be placed on the development of nutritionally valuable forest products, particularly targeting the protection of non-wood forest products to increase their value. NGOs, schools, hospitals, and health centers should be encouraged to promote the benefits of non-wood forest products. It is necessary to establish cross-sectoral policy platforms that bring together environmental, health, development, agriculture, and other sectors in order to better integrate information and knowledge about nutritious forest foods and their conservation into national nutrition strategies and programs. These platforms will facilitate the wider dissemination of strategies for utilizing high-nutrient-value tree biodiversity in food security, nutrition, conservation.

ACKNOWLEDGEMENTS

This study was conducted by searching academic research on non-wood forest products. We extend our gratitude to all academics who have contributed to research in this field.

REFERENCES

Bailey RC, Head G, Jenike M, Owen B, Rechtman R, and Zechenter E. 1989. Hunting and Gathering in Tropical Rain Forest: Is It Possible? *Am Anthropol*, *91*(1), 59–82.DOI: https://doi.org/10.1525/aa.1989.91.1.02a00040

Belem B, Sane BC, Ouattara ELY, Sama PG, and Boussim J. 2009. Wild leafy vegetables in the community of séguénéga, northern Burkina Faso and their contribution to food security and income generation. *Acta Hortic*, *806*, 121–128.DOI: https://doi.org/10.17660/ActaHortic.2009.806.13

Black RE, Victora CG, Walker SP, Bhutta ZA, Christian P, de Onis M, Ezzati M, Grantham-McGregor S, Katz J, Martorell R, and Uauy R. 2013. Maternal and child undernutrition and overweight in low-income and middle-income countries. *The Lancet*, *382*(9890), 427–451.DOI: https://doi.org/10.1016/S0140-6736(13)60937-X

Burlingame B, and Dernini S. 2012. Sustainable Diets and Biodiversity.

Butler CD. 2012. Human Health and Forests. In C. J. P. Colfer (Ed.), *A Global Overview of Issues, Practice and Policy*.

Colfer C. 2012. Human health and forests: a global overview of issues.

Dansi A, Adjatin A, Adoukonou-Sagbadja H, Faladé V, Yedomonhan H, Odou D, and Dossou B. 2008. Traditional leafy vegetables and their use in the Benin Republic. *Genet Resour Crop Evol*, *55*(8), 1239–1256.DOI: https://doi.org/10.1007/s10722-008-9324-z

Dounias E, and Froment A. 2011. From foraging to farming among present-day forest hunter-gatherers: consequences on diet and health. *International Forestry Review*, *13*(3), 294–304.DOI: https://doi.org/10.1505/146554811798293818

FAO. 2013. Forests and trees outside forests are essential for global food security and nutrition, Summary of the International Conference on Forests for Food Security and Nutrition.

Faye MD, Weber JC, Mounkoro B, and Dakouo J-M. 2010. Contribution of parkland trees to farmers' livelihoods: a case study from Mali. *Dev Pract*, *20*(3), 428–434.DOI: https://doi.org/10.1080/09614521003710013

Foresight UK. 2011. The future of food and farming. Final Project Report.

Godfray HCJ, Beddington JR, Crute IR, Haddad L, Lawrence D, Muir JF, Pretty J, Robinson S, Thomas SM, and Toulmin C. 2010. Food Security: The Challenge of Feeding 9 Billion People. *Science (1979)*, *327*(5967), 812–818.DOI: https://doi.org/10.1126/science.1185383

Heywood VH, Fanzo J, and Hunter D. 2013. Overview of agricultural biodiversity and its contribution to nutrition and health. In *Diversifying food and diets: Using agricultural biodiversity to improve nutrition and health* (pp. 35–67).

Humphry CM, Clegg MS, Keen CL, and Grivetti LE. 1993. Food diversity and drought survival. The Hausa example. *Int J Food Sci Nutr*, *44*(1), 1–16.DOI: https://doi.org/10.3109/09637489309017417

Hunter D, and Fanzo J. 2013. Agricultural Biodiversity, Diverse Diets and Improving Nutrition. In J. Fanzo, D. Hunter, T. Borelli, & F. Mattei (Eds.), *Diversifying Food and Diets: Using Agricultural Biodiversity to Improve Nutrition and Health* (pp. 1–14). Diversifying Food and Diets: Using Agricultural Biodiversity to Improve Nutrition and Health.

Jamnadass RH, Dawson IK, Franzel S, Leakey RRB, Mithöfer D, Akinnifesi FK, and Tchoundjeu Z. 2011. Improving livelihoods and nutrition in sub-Saharan Africa through the promotion of indigenous and exotic fruit production in smallholders' agroforestry systems: a review. *International Forestry Review*, *13*(3), 338–354.DOI: https://doi.org/10.1505/146554811798293836

Jansen van Rensburg WS, Venter SL, Netshiluvhi TR, van den Heever E, Vorster HJ, de Ronde JA, and Bornman CH. 2004. Role of indigenous leafy vegetables in combating hunger and malnutrition. *South African Journal of Botany*, *70*(1), 52–59.DOI: https://doi.org/10.1016/S0254-6299(15)30268-4

Johns T, Powell B, Maundu P, and Eyzaguirre PB. 2013. Agricultural biodiversity as a link between traditional food systems and contemporary development, social integrity and ecological health. *J Sci Food Agric*, *93*(14), 3433–3442.DOI: https://doi.org/10.1002/jsfa.6351

Johns T, and Sthapit BR. 2004. Biocultural Diversity in the Sustainability of Developing-Country Food Systems. *Food Nutr Bull*, *25*(2), 143–155.DOI: https://doi.org/10.1177/156482650402500207

Kennedy G, Ballard T, and Dop M. 2011. Guidelines for measuring household and individual dietary diversity.

Kennedy G, Nantel G, and Shetty P. 2003. The scourge of "hidden hunger": global dimensions of micronutrient deficiencies en revista. *Food, Nutrition and Agriculture*, *32*, 8–16.

Moreno-Black G, and Somnasang P. 2000. In times of plenty and times of scarcity: Nondomesticated food in northeastern Thailand. *Ecol Food Nutr*, *38*(6), 563–586.DOI: https://doi.org/10.1080/03670244.2000.9991597

Nutrition SCO. 2004. 5th Report on the World Nutrition Situation: Nutrition for Improved Development Outcomes.

Ok K, and Tengiz YZ. 2018. Türkiye'de Odun Dışı Orman Ürünlerinin Yönetimi. *Kahramanmaraş Sütçü İmam Üniversitesi Doğa Bilimleri Dergisi*.DOI: https://doi.org/10.18016/ksudobil.342303

Ozugurlu E, and Duzgun M. 2000. Policies to promote sustainable operations and utilization of non-wood forest products in Turkey.

Penafiel D, Lachat C, Espinel R, Van Damme P, and Kolsteren P. 2011. A Systematic Review on the Contributions of Edible Plant and Animal Biodiversity to Human Diets. *Ecohealth*, *8*(3), 381–399.DOI: https://doi.org/10.1007/s10393-011-0700-3

Pimentel D, McNair M, Buck L, Pimentel M, and Kamil J. 1997. The Value of Forests to World Food Security. *Hum Ecol*, *25*(1), 91–120.DOI: https://doi.org/10.1023/A:1021987920278

Powell B, Hall J, and Johns T. 2011. Forest cover, use and dietary intake in the East Usambara Mountains, Tanzania. *International Forestry Review*, *13*(3), 305–317.DOI: https://doi.org/10.1505/146554811798293944

Ruel MT. 2003. Operationalizing dietary diversity: a review of measurement issues and research priorities. . *J Nutr*, *133*(11), 3911–3926.

Ruel MT, and Alderman H. 2013. Nutrition-sensitive interventions and programmes: how can they help to accelerate progress in improving maternal and child nutrition? *The Lancet*, *382*(9891), 536–551.DOI: https://doi.org/10.1016/S0140-6736(13)60843-0

Scoones I, Melnyk M, and Pretty JN. 1992. The hidden harvest: wild foods and agricultural systems; a literature review and annotated bibliography. International instute for environment and development.

Stephenson K, Amthor R, Mallowa S, Nungo R, Maziya-Dixon B, Gichuki S, Mbanaso A, and Manary M. 2010. Consuming cassava as a staple food places children 2-5 years old at risk for inadequate protein intake, an observational study in Kenya and Nigeria. *Nutr J*, *9*(1), 9.DOI: https://doi.org/10.1186/1475-2891-9-9

Sunderland TCH. 2011. Food security: why is biodiversity important? *International Forestry Review*, *13*(3), 265–274.DOI: https://doi.org/10.1505/146554811798293908

Tontisirin K, Nantel G, and Bhattacharjee L. 2002. Food-based strategies to meet the challenges of micronutrient malnutrition in the developing world. *Proceedings of the Nutrition Society*, *61*(2), 243–250.DOI: https://doi.org/10.1079/PNS2002155

Türker MF, Öztürk A, and Tiryaki E. 2019**, May 15**. Ülkemiz ormancılık sektöründe odun dışı orman ürünleri kapsamında değerlendirilen odun dışı bitkisel ürünlerin işletmeciliği. *IIUlusal Karadeniz Ormancılık Kongresi*.

Vinceti B, Eyzaguirre P, and Johns T. 2008. The Nutritional Role of Forest Plant Foods for Rural Communities. In *Human Health and Forests* (1st ed., pp. 1–38).

Vinceti B, Termote C, Ickowitz A, Powell B, Kehlenbeck K, and Hunter D. 2013. The Contribution of Forests and Trees to Sustainable Diets. *Sustainability*, *5*(11), 4797–4824.DOI: https://doi.org/10.3390/su5114797

DRAFT

Remifentanil Genotoxicity Assessment via Comet Assay

Öge ARTAGAN (*https://orcid.org/0000-0001-9389-4450*)

Muğla Sıtkı Koçman University, Vacational School of Health Services, Medical Services and Techniques, Muğla, Turkey

Corresponding author e-mail: ogebasoglan@mu.edu.tr

Abstract

DNA is a fundamental molecule that can be a target for various molecules, hence it has a high probability of being damaged. Genotoxicity tests include rational test systems used to evaluate the safety of drugs, industrial chemicals, pollutants, and metabolites. Remifentanil is a potent synthetic opioid analgesic drug used to relieve pain during surgical operations and for sedation purposes. This study aimed to demonstrate the previously unrevealed genotoxic effects of remifentanil on healthy human lymphocytes. To accomplish this, scientists utilized a comet assay, which is a simple, quick, and highly sensitive method for detecting genotoxic harm arising from individual strand breaks in DNA. Importantly, there was a correlation between the observed genotoxic impact and the dosage. For comet assay, the human lymphocytes were isolated from healthy individuals and exposed to 50, 150, 250, 350 µg/mL of remifentanil for an hour. In human lymphocytes analyzed through the Comet assay, the greatest concentration of remifentanil led to maximum DNA migration (mean tail length). It showed that Remifentanil triggered genotoxicity by directly affecting chromosomes and DNA in human lymphocyte cells and may have genotoxicity for humans.
In genotoxicity assessments, the changes in tail length and tail intensity parameters were evaluated by comparing them with the values of spontaneous control and positive control, and the results were analyzed.
In conclusion, this study has demonstrated that remifentanil may potentially induce genotoxic effects on healthy human lymphocytes under in vitro conditions. The degree of genotoxicity may fluctuate depending on the dosage and duration of exposure.

Key Words: Comet assay, DNA damage, genotoxicity, lymphocyte culture, remifentanil

INTRODUCTION

With the discovery of modern general anesthesia, complex surgical and diagnostic procedures have advanced in patients of all age groups. Anesthetic and sedative agents interact with neurotransmitters and affect the central nervous system, disrupting the integration of neurons between different brain regions. (Liu et al., 2020). Anesthetic drugs are chemicals that temporarily induce a sleep-like state. They make painless or painless surgery possible. During general anesthesia, in other words anesthesia; consciousness, sensitivity to pain, defensive reflexes and muscle reflexes are significantly reduced. Anesthetics do not affect vital systems such as breathing and circulation, which are controlled by the spinal cord. (Wollweber, 2000).

Remifentanil is a potent synthetic opioid analgesic that has gained significant attention and utilization in the field of anesthesia and perioperative medicine. Remifentanil is characterized by its rapid onset, short duration of action, and precise titratability, making it an invaluable tool for anesthesiologists and healthcare professionals in the management of pain during surgical procedures and in critical care settings (Glass et al., 1993).

Living organisms have evolved to have the most suitable genetic structure for themselves. Genetic changes that occur in this genome are called mutations. If mutations are not corrected by DNA repair mechanisms, they can often lead to harmful or life-threatening consequences for living organisms. Damage to the genetic structure as a result of the interaction of genes with toxic subtances is called genotoxicity (Feretti et al., 2007). Substances that cause genotoxicity are called genotoxic substances or mutagens. All chemicals such as drugs, pesticides and cosmetics have toxic potential and these toxic potentials need to be evaluated. Various genotoxicity tests have been developed for this purpose. (Saygı, 2002).

DNA damage arises spontaneously during metabolism or due to the influence of certain environmental factors. Preserving the structure of DNA is crucial for the error-free transmission of genetic information. Techniques enabling the precise measurement of DNA damage are important in today's context. Single-cell gel

electrophoresis, commonly refered to as the "Comet Assay," is a non-invasive, rapid, and quite sensitive fluorescent microscopic method employed to measure DNA damage within the cell (Dinçer and Kankaya, 2010).

Based on this, it is aimed to perform the Comet test to determine the damage and strand breaks in the DNA where the genetic code is located. Evaluation of the data obtained as a result of clastogenic effects at the gene and chromosome level is important in terms of human health and its contribution to the literature.

MATERIALS AND METHODS

Blood saples were obtained from heathy volunteer aged 25-30 years old. Lymphocyte cells were isolated by using histopaque then cells resuspended in RPMI 1640. For an effective comet assay, cells exposed to an agent must have at least 80% viability (Kılıç and Ayaz, 2020). Based on this information, remifentanil concentrations of 50, 150, 250, and 350 µg/mL were used for the experimental procedure. The samples were subsequently placed in remifentanil concentrations and incubated for two hours at 37°C. At the end of the incubation time, tubes were centrifuged at 300 rpm for 5 minutes, the supernatant was removed. Next, 100 µL of a low-melting-point agar, mixed with the 100 µL lymphocyte sample. Afterwards soft agar and lymphocyte sample. dripped onto slides previously coated with high-melting-point agar. The slides were then covered with a coverslip (Tice et al., 2000). After incubating the slides 4+ °C for 15-20 minutes, the coverslips were separated and the slides placed in chalets containing a lysis buffer. They were incubated 4+ °C for 18 hours. End of the incubation period the slides were placed in horizontal tanks with an electrophoresis buffer (pH greater than 13) for 20 minutes to denature the DNA. Electrophoresis was performed at 25 V and 300 mA in running buffer. Following electrophoresis, the slides treated with a neutralizing buffer (pH 7.5) for 5 minutes. Subsequently, ethidium bromide (EtBr) was evenly applied over the slides, which were then covered with a coverslip. In a dark area, the fluorescence emitted by the stained DNA is easily detectable and provides clear visualization of the comets. As a result experiimental steps were performed in a dark area. For comet analysis, the slides were examined at 40x magnification using a fluorescent microscope. Image analysis and comet counting were conducted on 100 comets (Kassie, 2000). Ethidium bromide fluoresces intercalates into DNA molecules by inserting itself between base pairs. This property allows it to stain DNA effectively, making DNA fragments visible under a fluorescence microscope.under specific wavelengths. So EtBr staining was utilized to detect damage, and images acquired under a fluorescence microscope were analyzed using the Comet Image Processing and Analysis System Software (Comet IV).

DRAFT

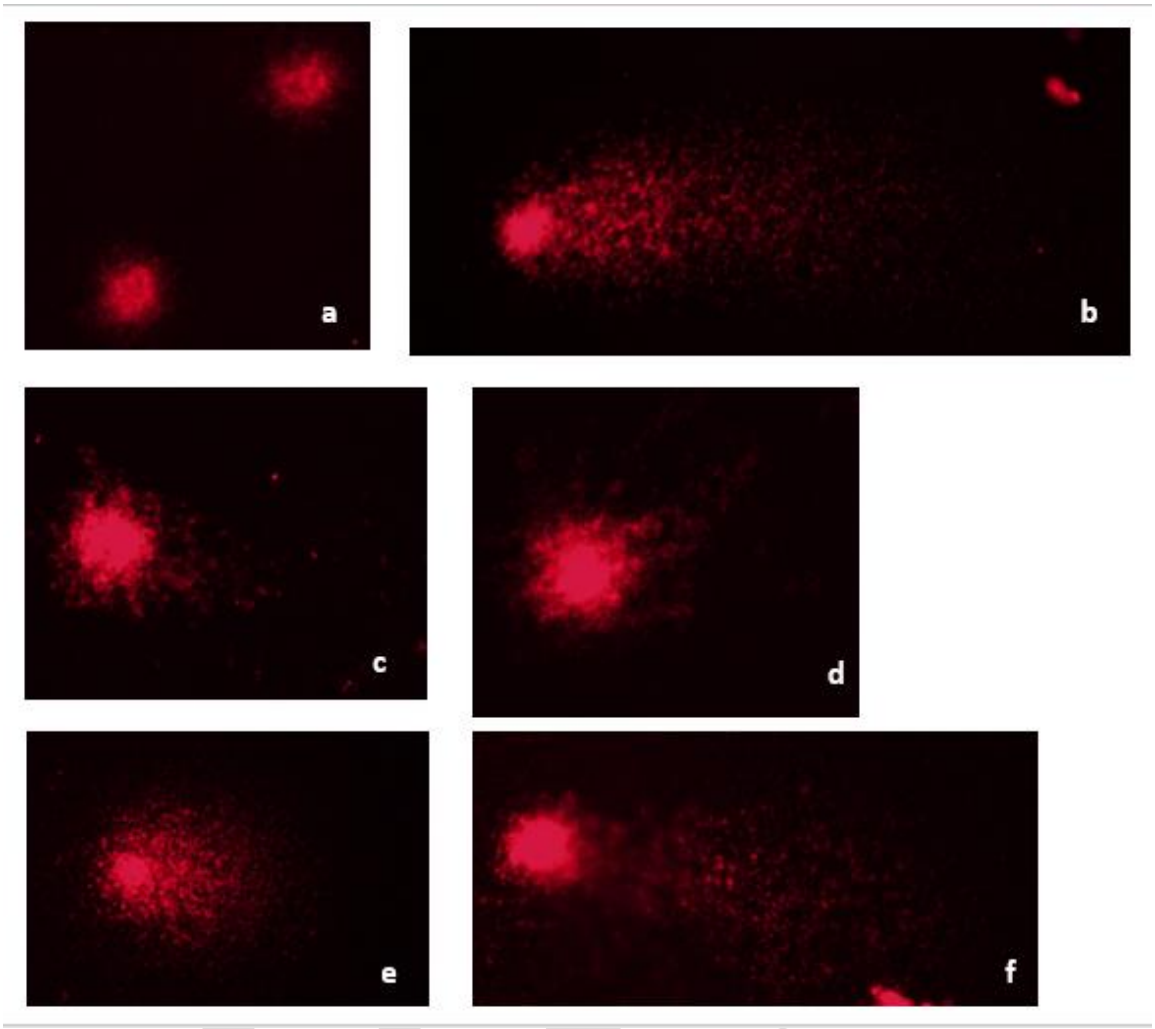


Figure 1: a) 40X magnification spontaneous control, b) 40X magnification positive(H_2O_2) control, c) 50 µg/mL remifentanil dose, d) 150 µg/mL remifentanil dose, e) 250 µg/mL remifentanil dose, g) 350 µg/mL remifentanil dose

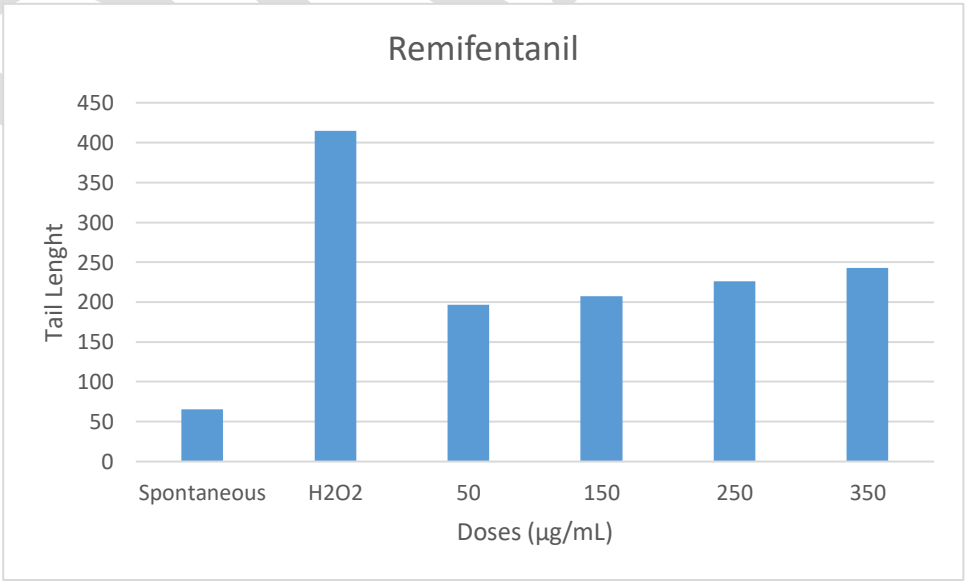


Figure 2. Tail length of DNA damage in human peripheral lymphocyte cells treated for 2h with doses of remifentanil.

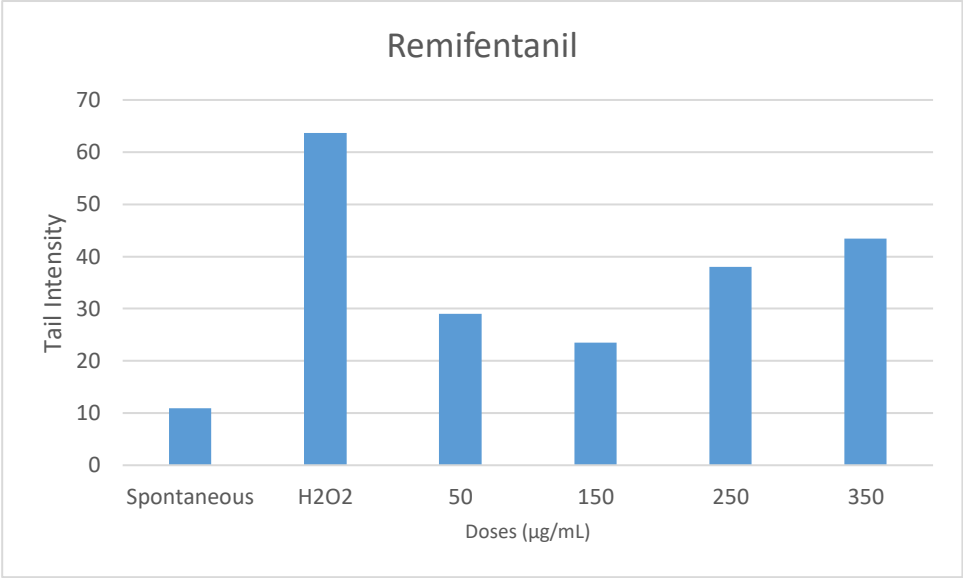


Figure 3. Tail intensity of DNA damage in human peripheral lymphocyte cells treated for 2h with doses of remifentanil.

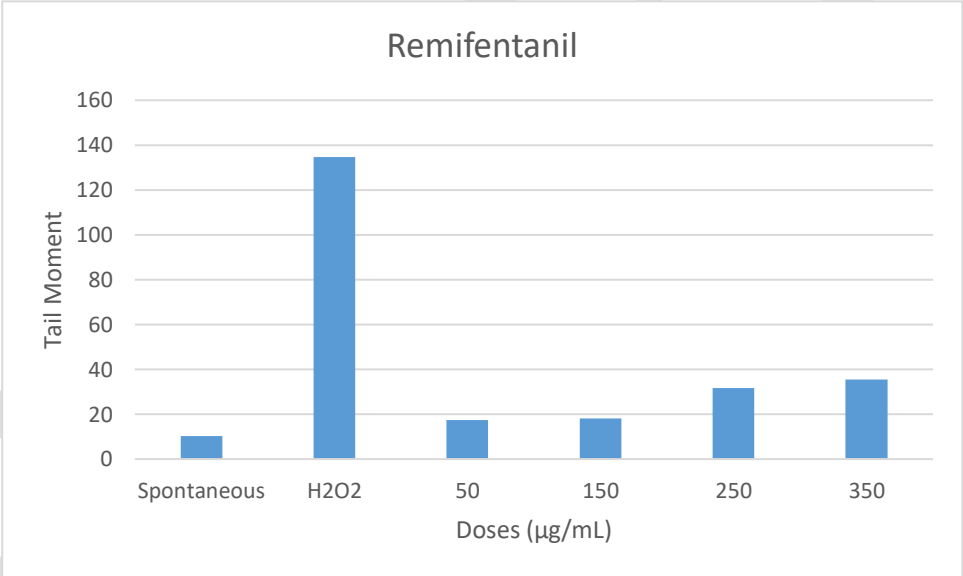


Figure 4. Tail moment of DNA damage in human peripheral lymphocyte cells treated for 2h with doses of remifentanil.

Table 1. DNA damage in human peripheral lymphocyte cells treated with doses of remifentanil for 2 hours.

	Treatment	Tail Length	Tail Intensity	Tail Moment
	Concentraion (µg/mL)	Mean ± SD	Mean ± SD	Mean ± SD
Spontaneous	-	69,00±4,629**	10,93±2,221**	9,91±4,177**
H_2O_2	50 µM	414,9±12,71	63,71±4,864	134,7±11,49
Remifentanil	50 µg/mL	194,8±45,26	29,57±19,94**	19,07±13,84
	150 µg/mL	204,7±57,06	23,24±18,37	17,17±14,30
	250 µg/mL	224,7±84,82	37,20±19,97**	31,31±29,09**
	350 µg/mL	235,1±68,85	42,55±17,49	32,91±17,46

H_2O_2: Hydrogen peroxide, SD: Standard deviation, ** P< 0.05 *** P<0.001, (Tamhane T2 Test), P value: Statistically significant difference

RESULTS and DISCUSSION

The comet assay was employed to assess DNA damage in human lymphocytes exposed to different doses of remifentanil. After a 2-hours exposure to remifentanil at concentrations of 50, 150, 250, and 350 µg/mL, the DNA tail length, tail intensity, and tail moment values in the treated cells were significantly higher than the observed in the spontaneous control group (Figure 1). Comet parameters, including comet mean, tail length, tail intensity, and tail moment, were examined in 100 cells for each concentration and compared statistically with the corresponding control data. The statistical analyses of comet parameters revealed significant differences between the control groups and the groups treated with the remifentanil doses (***p < 0.001). An increase in tail length and intensity is indicative of DNA damage resulting from single breaks in DNA molecules (Colins 2004). Notably, this effect was particularly pronounced in human peripheral lymphocytes exposed to remifentanil at the investigated concentrations (50, 150, 250, 350 µg/mL) (***p <0.001).

For all remifentanil doses and the control groups, a total of 100 samples were analyesed for comet parameters. Any sinificant differences in comet parameters (such as tail lenght, moment and density) recorded after 2 hours were evaluated by compairing the number of comets in the cells with the spontaneous and positive control. Comet parameters analysed via SPSS Statistics 22 (IBM). Since the differences between groups in the comet parameter were different, the Tamhane T2 test was applied and comparisons were evaluated with the control groups. The figures clearly demostrated that comet parameter were significantly increased in a dose dependent manner when compared to spontaneous and positive control groups (Table 1). This increase indicates the disruption of DNA structure leading to the formation of DNA damage. Comet parameters (Figure 2, Figure 3, Figure 4) which are associated with DNA damage were found to be significant when compared to spontaneous and positive control especially 250 µg/mL dose of remifentanil (**p<0.05).

The results of the research light on the previously undisclosed genotoxic effects of remifentanil on healthy human lymphocytes, highlighting potential concerns regarding its safety in clinical applications. Remifentanil, a powerful synthetic opioid analgesic, is widely employed for pain management during surgical procedures and sedation (Glass et al.,1993). While its clinical efficacy is well-established, this research underscores the importance of evaluating its genotoxic potential, particularly when administered at various dosages. Comet assay, a rational method for detecting DNA damage, was employed in this study to assess genotoxic harm arising from individual strand breaks in DNA. The results indicate a dose-dependent correlation between remifentanil exposure and genotoxic impact on human lymphocytes (Table 1). The highest concentration of remifentanil led to the maximum DNA migration, as evidenced by the mean tail length (Figure 1), signifying a direct effect on chromosomes and DNA within human lymphocyte cells. The evaluation of genotoxicity in this study considered changes in tail length and tail intensity parameters by comparing them with values obtained from spontaneous control and positive control groups. These assessments provide compelling evidence of remifentanil-induced genotoxicity under in vitro conditions. The genotoxic effects observed in this

study emphasize the need for cautious consideration when utilizing remifentanil, especially in cases involving prolonged or high-dose exposure. Future research should aim to further elucidate the mechanisms underlying this genotoxicity and explore strategies to minimize potential risks.

CONCLUSION

In conclusion, this study underscores the importance of evaluating the genotoxic potential of remifentanil, as it may induce genotoxic effects on lymphocytes culture. The extent of genotoxicity appears to vary with dosage and duration of exposure, emphasizing the need for careful consideration of remifentanil administration in clinical settings.

REFERENCES

Collins AR 2004. The comet assay for DNA damage and repair. Molecular Biotechnology, 26(3): 249-261.

Dinçer Y, Kankaya S 2010. DNA Hasarının Belirlenmesinde Comet Assay. Turkiye Klinikleri J Med Sci, 30(4):1365-73.

Feretti D, Zerbini I, Zani C, Ceretti E, Moretti M, Monarca S 2007. Allium cepa chromosome aberration and micronucleus tests applied to study genotoxicity of extracts from pesticide-treated vegetables and grapes. Food Add. Contam, 24(6): 561-572.

Glass PS, Hardman D, Kamiyama Y, Quill TJ, Marton G, Donn KH, Grosse CM, Hermann D 1993. Preliminary pharmacokinetics and pharmacodynamics of an ultra-short-acting opioid: remifentanil (GI87084B). Anesth Analg, 77(5):1031-40.

Kassie F, Parzefall W, Knasmuller S 2000. Single cell gel electrophoresis assay: a new technique for human biomonitoring studies. Mutat. Res, 463: 13–31.

Kılıç M, Ayaz Tüylü BA 2020. An in-vitro investigation of genotoxic effects of dexketoprofen trometamol on healthy human lymphocytes. Drug and Chemical Toxicology, 43(2): 174-181.

Koppen G, Azqueta A, Pourrut B, Brunbor G, Collins AR, Langie SA 2017. The next three decades of the comet assay: a report of the 11th International Comet Assay. Mutagenesis, 32(3): 397-427.

Liu X, Ji J, Zhao GQ 2020. General anesthesia affecting on developing brain: evidence from animal to clinical research. Journal of Anesthesia, 34(5): 765–772.

Saygı Ş 2003. Deneysel toksikolojide toksisite testleri ve test sonuçlarının önemi. Gülhane Tıp Dergisi, 45(3): 291-298.

Tice, R.R., et al., 2000. Single cell gel/comet assay: guidelines for in vitro and in vivo genetic toxicology testing. Environmental and Molecular Mutagenesis, 35, 206–221.

Wollweber H 2000. Anesthetics, General. Ullmann's Encyclopedia of Industrial Chemistry.

DRAFT

ORAL PRESENTATION

Hidrazin-Borandan Hidrojen Üretimi İçin MIL-53(Al) Yapısına Dekore Edilen Palladyum Nanokümelerinin Geliştirilmesi ve Tepkime Kinetiğinin İncelenmesi

Nazlı Karataş[1], Mehmet Gülcan[1]

[1]Van Yüzüncü Yıl Üniversitesi Fen Fakültesi, Kimya Bölümü, Van, Türkiye

Özet

Bu çalışmada MIL-53(Al) yapısına dekore edilen Pd (0) nanokümeleri ıslak emdirme-indirgeme yöntemiyle hazırlanarak HB'ın hidrolizinden hidrojen üretimindeki katalitik performansları ve katalitik etkinliği incelendi ve katalizör miktarları, substrat ve sıcaklık değişimlerinin etkileri ortaya konuldu. Hazırlanan MIL-53(Al) yapısına dekore edilen Pd (0) nanokümeleri, ICP-OES, SEM, SEM/EDX teknikleriyle tanımlandı.

Anahtar Kelimeler: Hidrazin-boran, Hidrojen, Hidroliz, MIL-53(AL) Nanoküme, Palladyum

Development of Palladium Nanoclusters Decorated to MIL-53(Al) Structure for Hydrogen Production from Hydrazine-Borane and Investigation of Reaction Kinetics

Abstract

In this study, Pd (0) nanoclusters decorated with MIL-53(Al) structure were prepared by the wet impregnation-reduction method, and their catalytic performance and catalytic efficiency in hydrogen production from the hydrolysis of HB were examined, and the effects of catalyst amounts, substrate and temperature changes were revealed. Pd (0) nanoclusters decorated on the prepared MIL-53(Al) structure were identified by ICP-OES, SEM, SEM/EDX techniques.

Keywords: Hydrazine-borane, Hydrogen, Hydrolysis, MIL-53(AL) Nanocluster, Palladium

GİRİŞ

Şu anda hidrojen en umut verici yenilenebilir enerji kaynağıdır. Bunun nedeni çoğunlukla hidrojen kullanımının fosil kaynakların kullanımından yenilenebilir enerjiye dayalı ekonomiye geçişi kolaylaştırmasıdır[1]. Ancak hidrojenin yaygın kullanımını sağlamak için güvenli, hafif ve verimli depolanması ve iletilmesi gibi birçok sorunun aşılması gerekmektedir [2-4]. Bu konular, uygun hidrojen içeriğine sahip kimyasalların depolanması ve uygulanmasıyla ilgili giderek daha yoğun bir araştırma çabasını motive etmiştir [5]. Bugüne kadar birçok katı hidrojen depolama malzemesi, hidrojen depolama uygulamalarında kullanılmak üzere test edilmiştir [6-10]. Son yıllarda amonyak-boran [11-22] ve hidrazin-boran [23-25] gibi kimyasallar, yüksek hidrojen içerikleri ve çoklu uygulamalarda kullanılabilme yetenekleri nedeniyle katı hidrojen depolama malzemeleri olarak kapsamlı bir şekilde incelenmiştir. Hidrojen bu kimyasallarda protik (N-H) veya hidridik (B-H) hidrojen olarak mevcuttur [26]. Amonyak-boranın yüksek hidrojen içeriğine (ağırlıkça %19,6) sahip olmasına ve basit bir B-N bağlı bileşik olmasına rağmen, NH_3 içeriği hidroliz koşullarında hidrolize edilmediğinden hidrojeni tam olarak salınamaz [27]. Ayrıca yüksek substrat konsantrasyonuna sahip hidrojen uygulamalarında amonyak (NH_3) kullanılan katalizör için zehirleyici etki yaratmaktadır [28]. Buna karşın hidrazin boranın ($N_2H_4BH_3$, HB) kullanımında durum böyle değildir [29-30]. Uygun bir katalizör kullanılarak, bir mol HB'nin hidrolitik dehidrojenasyonu, (1)'de anlatıldığı gibi 3 mol hidrojen gazı [24] verir.

$$N_2H_4BH_3 + 2H_2O \rightarrow N_2H_5BO_2\ (aq) + 3H_2\ (g) \qquad (1)$$

Bu çalışma da öncelikle MIL-53(AL) sentezlendi. Daha sonar MIL-53 (Al) destekli palladyum nanokümeleri (Pd@MIL-53(Al) ıslak emdirme-indirgeme yöntemiyle hazırlanmıştır. Ardından hazırlanmış Pd@MIL-53(Al) nanokatalizörü ICP-OES (İndüktif Eşleşmiş Plazma-Optik Emisyon Spektrometresi), SEM (Taramalı Elektron Mikroskopu), SEM/EDX (Taramalı Elektron Mikroskopu/Enerji Dağılımlı X-Işını Spektroskopisi

gibi karakterizasyon teknikleriyle tanımlanmıştır. Daha sonra Pd@MIL-53(Al) nanokatalizörünün katalitik performansı HB'nin hidrolizinden hidrojen üretimi tepkimesinde etkinlik ve dönüşüm açısından test edilmiştir.

MATERYAL

$Al(NO_3)_3 \cdot 9H_2O$, 1,4-benzendikarboksilik asid, $PdCl_2 \cdot xH_2O$ kimyasalları ticari olarak temin edilmiştir. HB ve MIL-53(Al) sentezlenmiştir.

METOT

Bu çalışma kapsamında gerçekleştirilmesi planlanan işlemler aşağıda maddeler halinde sunulmuştur:

MIL-53(Al) destek malzemesinin hazırlanması: Literatürde yer alan hidrotermal yöntemi kullanılarak sentezlenmiştir. Literatüre göre $Al(NO_3)_3 \cdot 9H_2O$ (2.60 g), 1,4-benzendikarboksilik asid (0.576 g) ve deiyonize su (10 mL) 50 mL Teflon kaplı çelik otoklava yerleştirilip ve 210°C'de 3 gün ısıtılacaktır. Reaksiyon sonunda beyaz toz süzülerek deiyonize su ve etanolle birkaç defa yıkandıktan sonra 60 °C de bir gece kuruması için bekletildi [31].

MIL-53(Al) yapısına dekore edilmiş Pd (0) nanokümelerinin hazırlanması: Çalışmada MIL-53(Al) yapısına dekore edilmiş Pd (0) nanokümeleri, çözelti fazında metal tuzunun destekleyici yüzeyine depolanması ve daha sonra $NaBH_4$ kullanılarak indirgenmesiyle hazırlandı. Bu amaçla, Pd metalinin başlangıç tuzu ($PdCl_2 \cdot xH_2O$) MIL-53(Al) katı destek malzemesinin yüzeyine emdirilmesi (3 saat boyunca 750 rpm karıştırma hızında karıştırılarak) ve daha sonra $NaBH_4$ varlığında indirgenmesi sağlandı.

Hidrazin-boran sentezi: HB, dihidrazin sülfatın ($(N_2H_5)_2SO_4$) kuru dioksan veya THF içerisinde sodyum borhidrür ($NaBH_4$) ile aşağıda verilen tepkimesinden sentezlendi. Sentezlenen HB'ın karakterizasyonu FT-IR, erime noktası ve ihtiyaç durumunda ^{1}H-NMR teknikleri ile yapıldı, sonuçların kaynakça ile uyumu olduğu tespit edilmiştir.

$$(N_2H_5)_2SO_4 + 2NaBH_4 \rightarrow \mathbf{2N_2H_4BH_3} + Na_2SO_4 + 2H_2$$

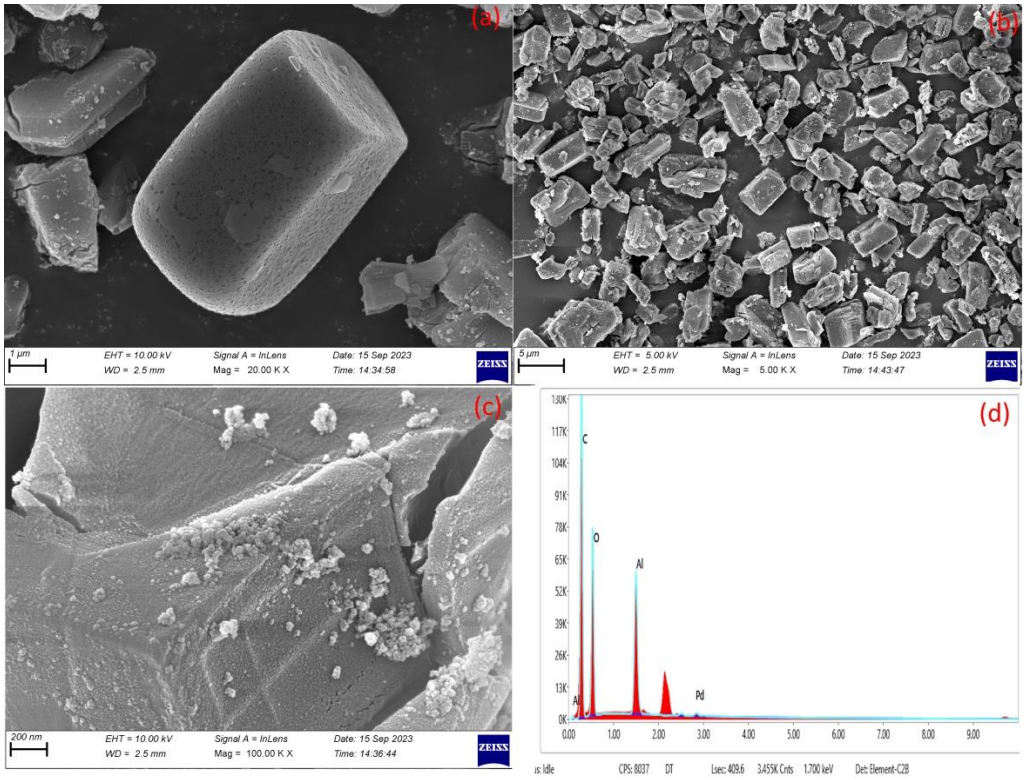
MIL-53(Al) yapısına dekore edilmiş Pd (0) nanokümelerinin HB'ın hidroliz tepkimesinde katalitik etkinliğinin incelenmesi: Çalışmanın bu kısmında hazırlanmış MIL-53 yapısına dekore edilmiş Pd (0) nanokümelerinin HB'ın hidrolizi tepkimesinde katalitik etkinliği, tepkime sonucu açığa çıkacak gazın hacimsel ölçümü ve katalitik etkinlik göz önüne alınarak incelendi. Hazırlanmış MIL-53 yapısına dekore edilmiş Pd (0) nanokümelerinin etkinlik, seçicilik, dönüşüm ve dayanıklılık yönünden en iyi katalitik performansa sahip tepkime koşulları belirlendi.

MIL-53(Al) ile dekore edilmiş Pd (0) nanokümelerinin tanımlanması: Proje çalışmasında MIL-53(Al) yapısına dekore edilmiş Pd (0) nanokümeleri ICP-OES, SEM, SEM/EDX gibi ileri analitiksel yöntemler kullanılarak tanımlandı. Bu analitiksel yöntemlerden ICP-OES; MIL-53(Al) yapısına dekore edilmiş Pd metal miktarının tespiti, SEM; Pd (0) nanokümelerinin parçacık boyut analizi ve morfolojilerinin belirlenmesi, SEM/EDX; SEM analizi sırasında seçili bölgelerde metal dağılımının elementel olarak incelenmesi amacıyla kullanılmıştır.

BULGULAR ve TARTIŞMA

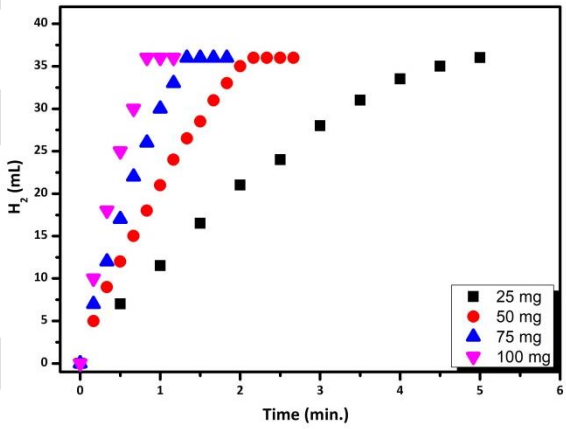
Çalışmanın ilk aşamasında MIL-53(Al) destek malzemesi kaynakçaya göre sentezlendikten sonra MIL-53(Al) destekli Pd (0) nanokümeleri ıslak emdirme-indirgeme yöntemiyle hazırlanarak HB'nin hidrolizi tepkimesi yoluyla hidrojen üretimindeki katalitik performanslar incelenmiştir.

İlk olarak Morfolojik olarak Pd@MIL-53(Al) katalizörünün incelenmesi için SEM analizleri yapılmıştır ve bu görüntüler Şekil 1'de verilmektedir. 1 µm, 5 µm ve 200 nm'de alınan SEM görüntülerinden Pd metallerinin destek malzemesinin yüzeyine homojen olarak dağıldığı görülmektedir (Şekil 1 (a,b,c)). Ayrıca SEM analizi sırasında yapılan haritalandırmada hem Pd metalinin (Şekil 1 (d)) ve destek malzemesinde bulunan Al, C, O ve Pd (Şekil 1 (b)) elementlerinin varlığı açıkça anlaşılmaktadır.



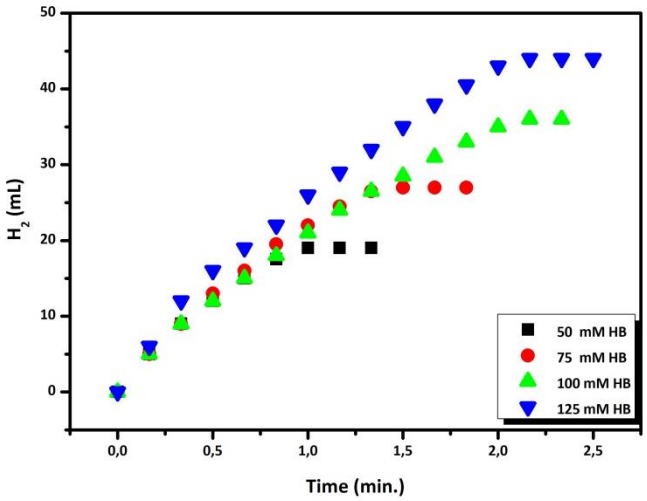
Şekil 1. Pd@MIL-53(Al) katalizörüne ilişkin (a) 1 µm, (b) 5 µm ve (c) 200 nm ölçekli SEM görüntüleri d) Taramalı Elektron Mikroskopu/Enerji Dağılımlı X-Işını Spektroskopisi görüntüsü

Pd@MIL-53(Al) nanokatalizörü ile katalizlenen HB'nin hidrolizi tepkimesinde, tepkime hızına katalizör miktarlarının etkisini incelemek için, HB derişimi (23 mg, 100 mM) sabit tutularak farklı miktarlarda Pd@MIL-53(Al) nanokatalizör kullanımıyla HB'nin hidroliz tepkimesi 298 K sıcaklıkta gerçekleştirilmiştir. Şekil 2'da farklı derişimlerde Pd@MIL-53(Al) nanokatalizörü (Pd@MIL-53(Al) = 25 mg, 50 mg, 75mg ve 100 mg) varlığında katalizlenen HB'nin hidroliz tepkimesinde açığa çıkan H_2 gazın HB gaz hacmine karşı zaman grafiği verilmektedir.



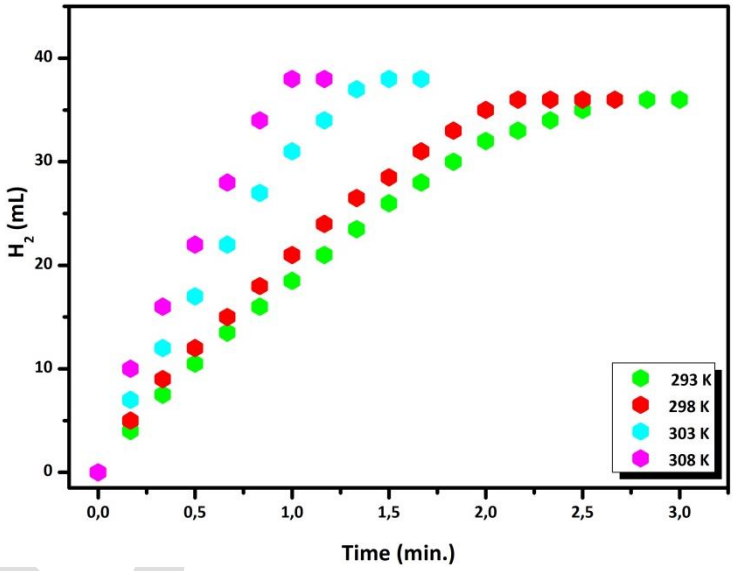
Şekil 2. Farklı miktarlarda Pd@MIL-53(Al) katalizörü ile katalizlenen HB'nin hidroliz tepkimesinde açığa çıkan gazın (H_2) zamana karşı grafiği.

Pd@MIL-53(Al) nanokatalizörü ile katalizlenen HB'nin hidroliz tepkimesinde tepkime hızına substrat derişiminin etkisini incelemek amacıyla, nanokatalizör derişimi (50 mg) sabit tutularak farklı derişimlerde HB kullanılarak katalitik hidroliz tepkimesi 298 K sıcaklıkta gerçekleştirilmiştir. Şekil 3'de farklı derişimlerde ([HB] = 50.0 mM, 75 mM, 100 mM, ve 125 mM) substrat kullanılarak katalizlenen HB'nin hidroliz tepkimesinde açığa çıkan H_2 gazın HB gaz hacmine karşı zaman grafiği verilmektedir.



Şekil 3. Farklı derişimlerde HB kullanılarak Pd@MIL-53(Al) nanokatalizörü ile katalizlenen HB'nin hidroliz tepkimesinde açığa çıkan gazın (H_2) zamana karşı grafiği.

Pd@MIL-53(Al) nanokatalizörü varlığında HB'nin hidroliz tepkimesine sıcaklığın etkisini belirlemek amacıyla katalizör ve substrat miktarları sabit tutularak sıcaklık değişimi etkisi incelenmiştir. Şekil 4'te farklı sıcaklıklarda(293K, 298 K, 303 K, 308 K) gerçekleştirilen hidroliz tepkimesinde açığa çıkan H_2 gazın HB gaz hacmine karşı zaman grafiği verilmektedir. Beklenildiği gibi katalitik hidroliz tepkime hızının sıcaklıkla doğru orantılı olarak arttığı görülmüştür.



Şekil 4. Pd@MIL-53(Al) nanokatalizörü ile katalizlenen HB'nin farklı sıcaklıklarda gerçekleştirilen hidroliz tepkimesinde açığa çıkan gazın (H_2) zamana karşı grafiği.

SONUÇ

Bu çalışma kapsamında elde edilen sonuçlar özetle şu şekilde sıralanabilir:

MIL-53(Al) kaynakçaya göre sentezlendi.
Pd@MIL-53(Al) nanokümeleri ıslak emdirme-indirgeme yöntemiyle sentezlendi.
Sentezlenen Pd@MIL-53(Al) nanokatalizörü SEM/SEM-EDX analizleri ile morfolojik olarak tanımlanması yapılmıştır.
Sentezlenen Pd@MIL-53(Al) nanokatalizörünün Hidrazin-boranın hidroliz tepkimesi yoluyla hidrojen üretimindeki katalitik etkinliği (katalizör miktarına, substrat derişimine ve sıcaklığa bağlı katalitik etkinliği) incelenmiştir.

DRAFT

TEŞEKKÜR

** 2209-A- Lisans Öğrencilerine Yönelik Araştırma Projesi Destek Programı kapsamındaki projemize (Proje No: 1919B012219607) maddi desteği için Türkiye Bilimsel ve Teknolojik Araştırma Kurumu'na teşekkür ederiz.*

KAYNAKLAR

[1] Niemann M.U., Srinivasan S.S., Phani A.R., Kumar A., Goswami D.Y., Stefanakos E.K., Nanomaterials for hydrogen storage applications: a review, J. Nanomater. 2008 (2008), https://doi.org/10.1155/2008/950967.

[2] Eberle U., Felderhoff M., Schüth F., Chemical and physical solutions for hydrogen storage, Angew. Chemie Int. Ed. 48 (2009) 6608–6630, https://doi. org/10.1002/anie.200806293.

[3] Sakintuna B., Lamari-Darkrim F., Hirscher M., Metal hydride materials for solid hydrogen storage: a review, Int. J. Hydrogen Energy 32 (2007) 1121–1140, https://doi.org/10.1016/j.ijhydene.2006.11.022.

[4] Russell G.J.T., Jones H., Materials for the Hydrogen Economy, 2007.

[5] Targets for Onboard Hydrogen Storage Systems for Light-Duty Vehicles, 2009 (Accessed January 9, 2019).

[6] Ogden J.M., Developing an infrastructure for hydrogen vehicles: a Southern California case study, Int. J. Hydrogen Energy 24 (1999) 709–730, https://doi. org/10.1016/S0360-3199(98)00131-1.

[7] Schlapbach L., Züttel A., Hydrogen-storage materials for mobile applications, Nature 414 (2001) 353–358, https://doi.org/10.1038/35104634.

[8] Züttel A., Materials for hydrogen storage, Mater. Today 6 (2003) 24–33, https:// doi.org/10.1016/S1369-7021(03)00922-2.

[9] Seayad A.M., . Antonelli D.M, Recent advances in hydrogen storage in metalcontaining inorganic nanostructures and related materials, Adv. Mater. 16 (2004) 765–777, https://doi.org/10.1002/adma.200306557.

[10] Bacsa R., Laurent C., Morishima R., Suzuki H., Le Lay M., Hydrogen storage in high surface area carbon nanotubes produced by catalytic chemical vapor deposition, J. Phys. Chem. B 108 (2004) 12718–12723, https://doi.org/10.1021/ jp0312621.

[11] Chandra M., Xu Q., A high-performance hydrogen generation system: transition metal-catalyzed dissociation and hydrolysis of ammonia-borane, J. Power Sources (2006), https://doi.org/10.1016/j.jpowsour.2005.05.043.

[12] Chandra M., Xu Q., Dissociation and hydrolysis of ammonia-borane with solid acids and carbon dioxide: an efficient hydrogen generation system, J. Power Sources 159 (2006) 855–860, https://doi.org/10.1016/J. JPOWSOUR.2005.12.033.

[13] Yamada Y., Yano K., Xu Q., Fukuzumi S., Cu/Co3O4 nanoparticles as catalysts for hydrogen evolution from ammonia borane by hydrolysis, J. Phys. Chem. C. 114 (2010) 16456–16462, https://doi.org/10.1021/jp104291s.

[14] Sen B., Demirkan B., Şimşek B., Savk A., Sen F., Monodisperse palladium nanocatalysts for dehydrocoupling of dimethylamineborane, Nano-Struct. Nano-Obj. 16 (2018) 209–214, https://doi.org/10.1016/j.nanoso.2018.07.008.

[15] Xu Q., Chandra M., Catalytic activities of non-noble metals for hydrogen generation from aqueous ammonia-borane at room temperature, J. Power Sources 163 (2006) 364–370, https://doi.org/10.1016/j.jpowsour.2006.09.043.

[16] Chandra M., Xu Q., Room temperature hydrogen generation from aqueous ammonia-borane using noble metal nano-clusters as highly active catalysts, J. Power Sources 168 (2007) 135–142, https://doi.org/10.1016/J. JPOWSOUR.2007.03.015.

[17] Günbatar S., Aygun A., Karatas Y., Gülcan M., Sen F., Carbon-nanotube-based rhodium nanoparticles as highly-active catalyst for hydrolytic dehydrogenation of dimethylamineborane at room temperature, J. Colloid Interface Sci. 530 (2018) 321–327, https://doi.org/10.1016/j.jcis.2018.06.100.

[18] Sen F., Karataş Y., Gülcan M., Zahmakıran M.; Amylamine stabilized platinum(0) nanoparticles: active and reusable nanocatalyst in the room temperature dehydrogenation of dimethylamine-borane. RSC ADVANCES , 2014. 4.(4), 1526-1531.

[19] Yurderi M., Bulut A., Zahmakıran M., Gülcan M., Özkar S.; Ruthenium(0) nanoparticles stabilized by metal-organic framework (ZIF-8): Highly efficient catalyst for the dehydrogenation of dimethylamine-borane and transfer hydrogenation of unsaturated hydrocarbons using dimethylamine-borane as hydrogen source. Applied Catalysis B-Environmental, 2014, 160, 534-541.

[20] Zhang X., . Kam L, Trerise R., Williams T.J., Ruthenium-catalyzed ammonia borane dehydrogenation: mechanism and utility, Acc. Chem. Res. 50 (2017) 86–95, https://doi.org/10.1021/acs.accounts.6b00482.

[21] Yan J.-M., . Zhang X.-B, Akita T., Haruta M., Xu Q., One-step seeding growth of magnetically recyclable Au@Co core-shell nanoparticles: highly efficient catalyst for hydrolytic dehydrogenation of ammonia borane, J. Am. Chem. Soc. 132 (2010) 5326–5327, https://doi.org/10.1021/ja910513h.

[22] Yan J.M., . Zhang X.B, Han S., Shioyama H., Xu Q., Synthesis of longtime water/ air-stable ni nanoparticles and their high catalytic activity for hydrolysis of ammonia-borane for hydrogen generation, Inorg. Chem. 48 (2009) 7389–7393, https://doi.org/10.1021/ic900921m.

[23] Hügle T., Kühnel M.F., Lentz D., Hydrazine borane: a promising hydrogen storage material, J. Am. Chem. Soc. 131 (2009) 7444–7446, https://doi.org/ 10.1021/ja9013437

[24] Demirkan B., Kuyuldar E., Karataş Y., Gülcan M., Sen F., Ex situ synthesis and characterization of a polymer-carbon nanotube-based hybrid nanocatalyst with one of the highest catalytic activities and stabilities for the hydrolytic dehydrogenation of hydrazine-borane at room temperature conditions, Journal of Colloıd And Interface Scıence. 552 (2019) 432–438.

[25] Çakanyıldırım Ç., Demirci U.B., Şener T., Xu Q., Miele P., Nickel-based bimetallic nanocatalysts in high-extent dehydrogenation of hydrazine borane, Int. J. Hydrogen Energy 37 (2012) 9722–9729, https://doi.org/ 10.1016/J.IJHYDENE.2012.03.054.

[26] Hamilton C.W., Baker R.T., Staubitz A., Manners I., B-N compounds for chemical hydrogenstorage, Chem. Soc. Rev. 38 (2009) 279–293, https://doi.org/10.1039/ B800312M.

[27] Sanyal U., Demirci U.B., Jagirdar B.R., Miele P., Hydrolysis of ammonia borane as a hydrogen source: fundamental issues and potential solutions towards implementation, ChemSusChem 4 (2011) 1731–1739, https://doi.org/10.1002/ cssc.201100318.

[28] Ramachandran P.V., Gagare P.D., Preparation of ammonia borane in high yield and purity, methanolysis, and regeneration, Inorg. Chem. 46 (2007) 7810– 7817, https://doi.org/10.1021/ic700772a.

[29] Vinh-Son N., Swinnen S., . Matus M.H, . Nguyen M.T, Dixon D.A., The effect of the NH2 substituent on NH3: hydrazine as an alternative for ammonia in hydrogen release in the presence of boranes and alanes, Phys. Chem. Chem. Phys. 11 (2009) 6339, https://doi.org/10.1039/b823358f.

[30] Demirkan B., Kuyuldar E., Karataş Y., Gülcan M., Şen F., Ex situ synthesis and characterization of a polymer-carbon nanotubebased hybrid nanocatalyst with one of the highest catalytic activities and stabilities for the hydrolytic dehydrogenation of hydrazine-borane at room temperature conditions, Journal of Colloid and Interface Science , 552 (2019) 432–438.

[31] Loiseau T., Serre C., Huguenard C., . Fink G, Taulelle F., Henry M., Bataille T., . Ferey G, Method For Preparing Metal-Organic Framework Crystallised And Porous Alumnium Aromatic Azocarboxylates. Chem Eur J 10 (2004) 1373-82.

DRAFT

ORAL PRESENTATION

Zirkonyum (IV) Oksit Nanotozu Kararlı Palladyum Nanokümelerinin Hazırlanması ve Metilamin-Boranın Hidrolizindeki Katalitik Performanslarının Araştırılması

Eren Ulaştan[1], Yaşar Karataş[2], Mehmet Gülcan[1]

[1]Van Yüzüncü Yıl Üniversitesi Fen Fakültesi, Kimya Bölümü, Van, Türkiye
[2] Van Yüzüncü Yıl Üniversitesi Muradiye Meslek Yüksekokulu, Kimya ve Kimyasal İşleme Teknolojileri Bölümü, Van, Türkiye

Özet

Bu çalışmanın ilk aşamasında önemli bir B-N bileşiği olan MeAB'nin hidroliz tepkimesinde kullanılmak üzere *nano*-ZrO_2 destekli Pd (0) nanokümeleri sentezlenerek tanımlandı. Bu amaç doğrultusunda hazırlanan *n*-ZrO_2 destekli Pd(0) nanokümeleri ICP-OES, XRD ve SEM gibi tekniklerle karakterize edildi. Hazırlanan Pd@*nano*-ZrO_2 katalizörü, MeAB'den hidroliz tepkimesi yoluyla hidrojen üretiminde katalizör olarak kullanılarak katalitik performansı araştırıldı. Hazırlanan Pd@*nano*-ZrO_2 katalizörünün MeAB'den hidroliz tepkimesi yoluyla hidrojen üretiminde sıcaklık ve derişim gibi parametreler göz önünde bulundurularak hidroliz tepkimeleri gerçekleştirildi.

Anahtar Kelimeler: Hidrojen, Hidroliz, Katalizör, Metilamin-Boran, Palladyum

Preparation of Zirconium (IV) Oxide Nanopowder Stable Palladium Nanoclusters and Investigation of their Catalytic Performance in the Hydrolysis of Methylamine-Borane

Abstract

In the first stage of this study, *nano*-ZrO_2 supported Pd (0) nanoclusters were synthesized and identified to be used in the hydrolysis reaction of MeAB, an important B-N compound. For this purpose, *n*-ZrO_2 supported Pd(0) nanoclusters prepared were characterized by techniques such as ICP-OES, XRD and SEM. The catalytic performance of the prepared Pd@*nano*-ZrO_2 catalyst was investigated by using it as a catalyst in the production of hydrogen from MeAB via hydrolysis reaction. Hydrolysis reactions were carried out by taking into account parameters such as temperature and concentration in the production of hydrogen through the hydrolysis reaction of the prepared Pd@nano-ZrO2 catalyst from MeAB.

Keywords: Hydrogen, Hydrolysis, Catalyst, Methylamine-Borane, Palladium

GİRİŞ

Son zamanlarda insan nüfusunun artması ve teknolojinin gelişmesiyle birlikte enerji tüketimi hızla artmıştır. İklim değişikliğinin olumsuz etkileri ve benzin, petrol, doğal gaz gibi fosil yakıt kaynaklarının tükenmesi, bilim adamlarını ve sanayicileri sera gazı emisyonlarını azaltmak ve yenilenebilir enerji kullanımını artırmak için ciddi adımlar atmaya yöneltmiştir [1,2]. Küresel ısınmanın neden olduğu sera gazları ozon tabakasına zarar vermekte ve zararlı ultraviyole ışık yoğunluğunu arttırmaktadır [3]. Bu nedenle güneş, hidrojen, rüzgar enerjileri gibi yeşil ve alternatif enerji kaynaklarının araştırılması araştırmacılar için öncelik haline gelmiştir [4]. Bu alternatif yeşil enerjiler arasında hidrojen enerjisi, yüksek yoğunluğu (120 MJ/kg), zengin rezervleri, düşük yoğunluğu (0,0899 g/L), sıfır emisyonu ve geniş uygulama alanları nedeniyle en önemli adaylardan biri olarak kabul edilmektedir [5]. H_2 üretimi, depolanması ve hidrojen rejenerasyonu, hidrojen üretiminin ana adımlarıdır. Amonyak-boran (AB, NH_3–BH_3) ve metilamin-boran (MeAB, CH_3NH_2–BH_3), hidrojen üretimi için en yaygın kullanılan amonyaklı bor (B-N) türevi bileşiklerdir [6,7,8]. AB, H_2 kapasitesi ρm = ağırlıkça %19,6 H olan oldukça kararlı B–N bileşiğidir [9]. AB ile karşılaştırıldığında MeAB ayrıca ağırlıkça %11,1 oranında önemli bir hidrojen içeriğine ve son derece stabil çalışma koşullarına sahiptir. Ayrıca bir AB türevi olan MeAB'nin AB'ye göre daha az uçucu olduğu ve dolayısıyla yakıt hücresi uygulamalarında daha avantajlı olduğu rapor edilmiştir. Önceki çalışmalar, oda sıcaklığında MeAB'nin molü başına 3 mol H_2 açığa çıkarabilen MeAB'nin hidrolizini bildirmiştir [10-12].

DRAFT

$$CH_3NH_2{-}BH_3\ (aq) + 2H_2O \rightarrow (CH_3NH_3)BO_2\ (aq) + 3H_2\ (g)$$

Bu proje kapsamında *nano*-ZrO_2 destekli Pd (0) nanokümeleri ıslak emdirme-indirgeme yöntemiyle hazırlandı ve detaylı karakterizasyon yapıldır. Ardından hazırlanmış olan nanokümeler MeAB'ın hidrolizi tepkimesinde ilk kez katalizör olarak kullanılarak ve katalitik performansları ortaya konulmuştur.

MATERYAL

Nano-ZrO_2,, $NaBH_4$, THF, metilamin hidroklorür, $PdCl_2 \times H_2O$, kimyasalları ticari olarak temin edilmiştir. MeAB ise sentezlenmiştir.

METOT

Bu çalışma kapsamında gerçekleştirilen işlemler aşağıda maddeler halinde sunulmuştur:

Metilamin-Boran ($CH_3NH_2BH_3$, MeAB) Sentezi ve Tanımlanması: Çalışmasının ilk aşamasında ilgili kaynakça kullanılarak MeAB sentezi gerçekleştirildi [13]. Bu amaçla, 250 mL iki boyunlu balon içerisine 3.88 g (0.1 mol) $NaBH_4$ konularak ve üzerine 200 mL önceden kurutulmuş THF eklendi. Oda sıcaklığında 30 dk karıştırıldıktan sonra karışım üzerine 6.752 g (0.1 mol) metilamin hidroklorür eklenerek ve tepkime kabının ağzı kapatılarak 24 saat boyunca oda sıcaklığında ve Ar atmosferinde karıştırıldır. 24 saat sonunda 3 numara kroze kullanılarak karışım süzüldü. Katı kısım izole edildikten sonra geriye kalan sıvı kısım evaporatöre edilerek ve bütün THF uzaklaştırıldı. THF tamamen uzaklaştırıldıktan sonra katı ürün üzerine 100 mL önceden kurutulmuş dietileter eklenip kriyostat ile sıcaklığı 0 ºC'ye ayarlanmış sistemde iki saat boyunca karıştırıldı. İki saatin sonunda katı kısım tekrar süzülerek izole edilecek ve eter fazındaki ürün oda sıcaklığında bir gece bekletilerek dietileterin tamamen uçması sağlandı ve oluşan beyaz renkli kristaller kullanılmak üzere amber renkli cam bir kapta saklandı.

$$CH_3NH_2{\cdot}HCl + NaBH_4 \rightarrow CH_3NH_2BH_3 + 2H_2 + 2NaCl$$

Elde edilecek MeAB'ı karakterize etmek için tanımlanması erime noktası tayini ile FT-IR tekniği kullanıldı.

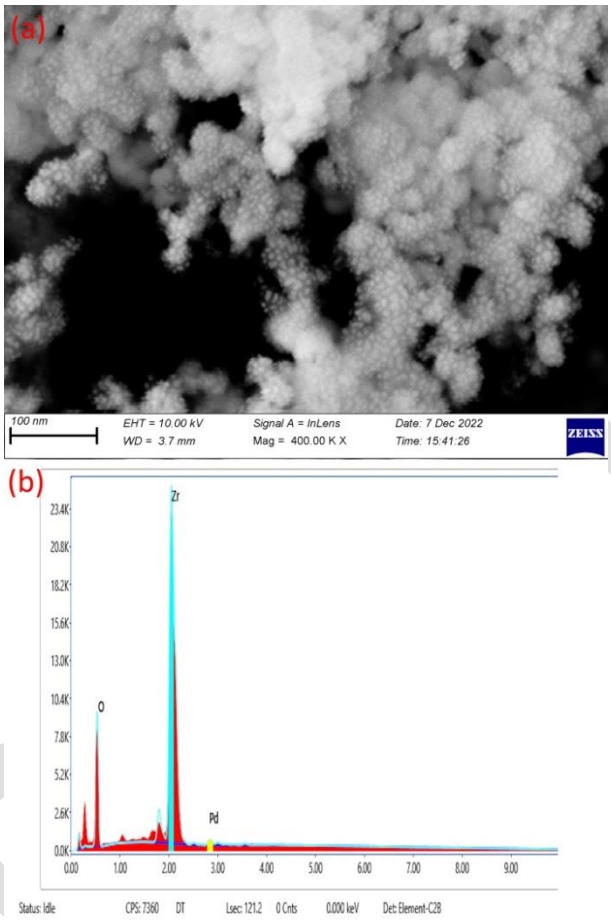
Pd@*nano*-ZrO_2 Nanokatalizörünün Hazırlanması ve Tanımlanması: Çalışmasında kullanılacak destek malzemesi olan *nano*-ZrO_2 ticari olarak temin edildi. Daha sonra *nano*-ZrO_2 destekli Pd (0) nanokümeleri, çözelti fazında metal tuzunun destekleyici yüzeyine depolanması ve daha sonra $NaBH_4$ kullanılarak indirgenmesiyle sentezldi. Bu amaçla, Pd metal başlangıç tuzu, *nano*-ZrO_2 malzemesinin yüzeyine emdirilmesi (3 saat boyunca 750 rpm karıştırma hızında karıştırılarak) ve daha sonra da kuvvetli bir indirgeyici ajan varlığında indirgenmesi sağlandı. Hazırlann bu katalitik malzemeler MeAB'nin hidroliz tepkimesinde katalizör olarak kullanılarak ve gaz çıkış hızları ile dönüşümleri incelendi. Son olarak elde edilen Pd@*nano*-ZrO_2 malzemesi; ICP-OES (İndüktif Eşleşmiş Plazma Optik Emisyon Spektrometresi), P-XRD (Toz X-Işınları Kırınımı), SEM (Taramalı Elektron Mikroskopu), gibi ileri analitiksel ve spektroskopik yöntemler kullanılarak tanımlandı. Bu analitiksel yöntemlerden ICP-OES; *nano*-ZrO_2 yüzeyine tutturulan Pd miktarının tespiti, XRD; metal (0) nanokümelerinin yüzeyde oluşumu sonucu destek malzemesinin kristal yapısının incelenmesi, SEM; metal (0) nanokümelerinin dağılım analizi ve katalitik tepkimeler sonrasında elde edilen ürünlerin analiz edilmesi ve muhtemel tepkime mekanizmasının belirlenmesi amacıyla kullanıldı.

Pd@*nano*-ZrO_2 Nanokatalizörünün Metilamin-Boran Hidroliz Tepkimesinde Katalitik Etkinliğin Belirlenmesi ve Tepkime Kinetiğinin İncelenmesi: Bu çalışma kısmında *nano*-ZrO_2 destekli Pd (0) nanokümeleri hazırlanarak, metilamin-boranın hidroliz tepkimesinde katalitik etkinliği, tepkime sonucu açığa çıkacak gazın hacimsel ölçümü ve katalitik etkinlik/katalitik dönüşüm göz önüne alınarak incelendi. *nano*-ZrO_2 destekli Pd(0) nanokümelerinin etkinlik, dönüşüm ve kararlılık yönünden katalitik performansın tespit edildiği koşullar ve hidroliz tepkimesi farklı çalışma koşullarında (katalizör ve substrat derişimi ve farklı sıcaklıklarda) gerçekleştirilerek tepkime kinetikleri incelendi.

BULGULAR ve TARTIŞMA

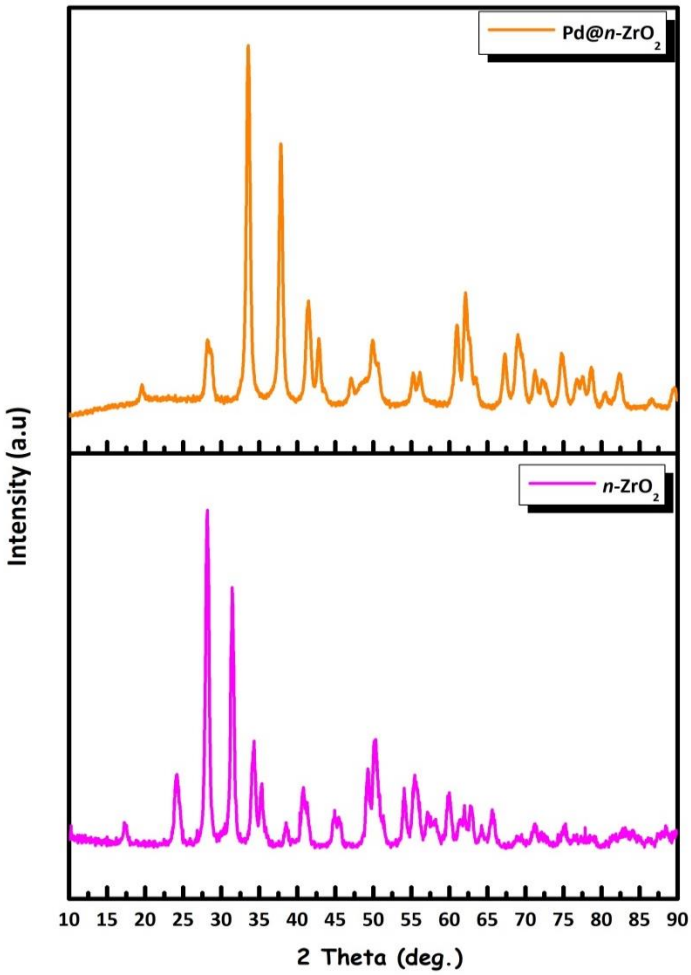
Çalışmanın ilk aşamasında *n*-ZrO_2 destekli palladyum nanokümeleri ıslak emdirme-indirgeme yöntemiyle hazırlanarak MeAB'nin hidrolizi tepkimesi yoluyla hidrojen üretimindeki katalitik performanslar incelenmiştir.

İlk olarak Morfolojik olarak Pd@*nano*-ZrO_2-katalizörünün incelenmesi için SEM analizleri yapılmıştır ve bu görüntüler Şekil 1'de verilmektedir. 100 nm'de alınan SEM görüntüsünden Pd metallerinin destek malzemesinin yüzeyine homojen olarak dağıldığı görülmektedir (Şekil 1 (a)). Ayrıca SEM analizi sırasında yapılan haritalandırmada hem Pd metalinin (Şekil 1 (b)) ve destek malzemesinde bulunan Zr, O ve Pd (Şekil 1 (b)) elementlerinin varlığı açıkça anlaşılmaktadır.



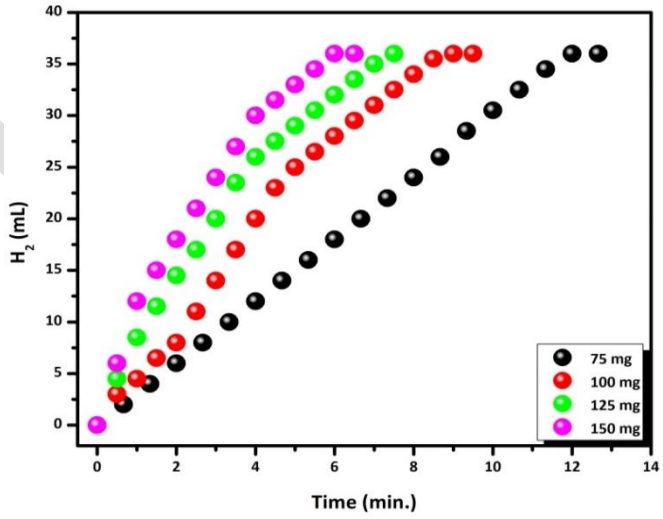
Şekil 1. (a) Pd@*nano*-ZrO_2 katalizörüne ilişkin 100 nm ölçekli SEM görüntüsü (b) Taramalı Elektron Mikroskopu/Enerji Dağılımlı X-Işını Spektroskopisi görüntüsü

Şekil 2'de *nano*-ZrO_2 ve Pd@*nano*-ZrO_2 örneklerinin XRD desenleri incelendiğinde Pd metalinin destek malzemesine yüklenmesi öncesi ve sonrası XRD desenleri incelendiğinde destek malzemesi olan *nano*-ZrO_2'nin kristal yapısında herhangi bir bozunmanın olmadığı tespit edilmiştir.



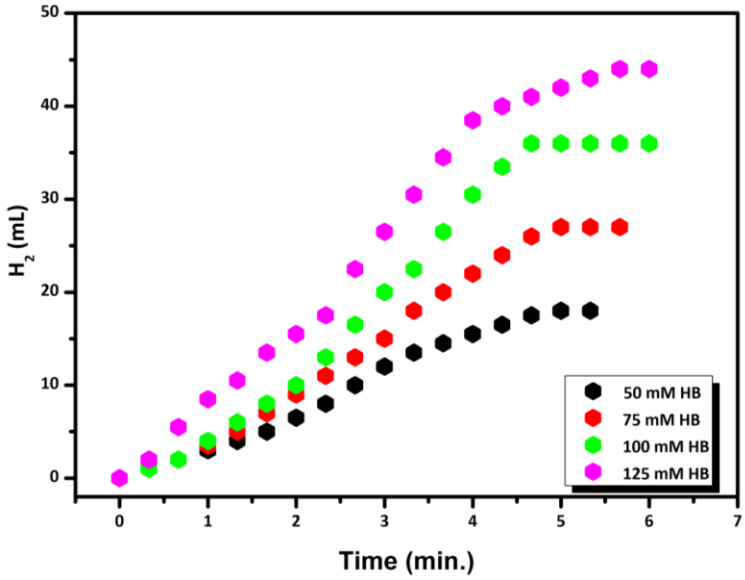
Şekil 2. *nano*-ZrO_2 ve Pd@*nano*-ZrO_2 örneklerine ait XRD desenleri.

Pd@*nano*-ZrO_2 nanokatalizörü ile katalizlenen MeAB'nin hidrolizi tepkimesinde, tepkime hızına katalizör miktarlarının etkisini incelemek için, MeAB derişimi (23 mg, 100 mM) sabit tutularak farklı miktarlarda Pd@*nano*-ZrO_2 nanokatalizör kullanımıyla MeAB'nin hidroliz tepkimesi 298 K sıcaklıkta gerçekleştirilmiştir. Şekil 3'da farklı derişimlerde Pd@*nano*-ZrO_2 nanokatalizörü (Pd@*nano*-ZrO_2 = 75 mg, 100 mg, 125mg ve 150 mg) varlığında katalizlenen MeAB'nin hidroliz tepkimesinde açığa çıkan H_2 gazın MeAB gaz hacmine karşı zaman grafiği verilmektedir.



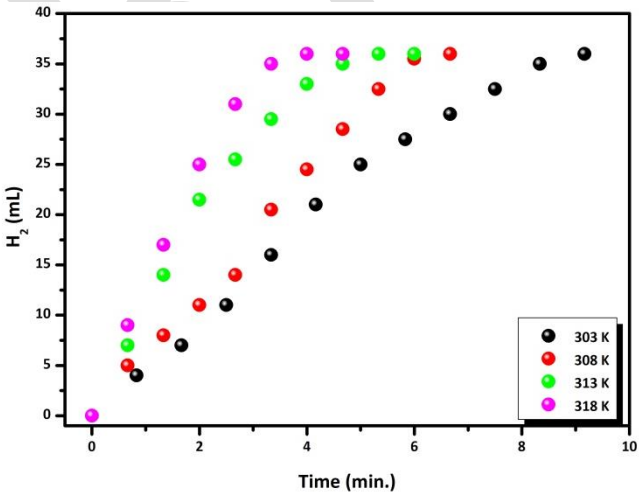
Şekil 3. Farklı miktarlarda Pd@*nano*-ZrO_2 katalizörü ile katalizlenen MeAB'nin hidroliz tepkimesinde açığa çıkan gazın (H_2) MeAB gaz hacmine karşı zaman grafiği.

Pd@*nano*-ZrO_2 nanokatalizörü ile katalizlenen MeAB'nin hidroliz tepkimesinde tepkime hızına substrat derişiminin etkisini incelemek amacıyla, nanokatalizör derişimi (100 mg) sabit tutularak farklı derişimlerde MeAB kullanılarak katalitik hidroliz tepkimesi 298 K sıcaklıkta gerçekleştirilmiştir. Şekil 4'de farklı derişimlerde ([MeAB] = 50.0 mM, 75 mM, 100 mM, ve 125 mM) substrat kullanılarak katalizlenen MeAB'nin hidroliz tepkimesinde açığa çıkan H_2 gazın MeAB gaz hacmine karşı zaman grafiği verilmektedir.



Şekil 4. Farklı derişimlerde MeAB kullanılarak Pd@*nano*-ZrO_2 nanokatalizörü ile katalizlenen MeAB'nin hidroliz tepkimesinde açığa çıkan gazın (H_2) gaz hacmine karşı zaman grafiği.

Pd@*nano*-ZrO_2 nanokatalizörü varlığında MeAB'nin hidroliz tepkimesine sıcaklığın etkisini belirlemek amacıyla katalizör ve substrat miktarları sabit tutularak sıcaklık değişimi etkisi incelenmiştir. Şekil 5'te farklı sıcaklıklarda gerçekleştirilen metanoliz tepkimesinde açığa çıkan H_2 gazın MeAB gaz hacmine karşı zaman grafiği verilmektedir. Beklenildiği gibi katalitik metanoliz tepkime hızının sıcaklıkla doğru orantılı olarak arttığı görülmüştür.



Şekil 5. Pd@*nano*-ZrO_2 nanokatalizörü ile katalizlenen MeAB'nin farklı sıcaklıklarda gerçekleştirilen hidroliz tepkimesinde açığa çıkan gazın (H_2) MeAB gaz hacmine karşı zaman grafiği.

SONUÇ

Bu çalışma kapsamında ilk olarak MeAB kaynakçaya göre sentezlendi. Daha sonra *nano*-ZrO_2 destekli palladyum nanokümeleri ıslak emdirme-indirgeme yöntemiyle sentezlendi. Sentezlenen Pd@*nano*-ZrO_2 nanokatalizörü SEM/SEM-EDX analizleri ile morfolojik olarak tanımlanması yapılmıştır. Sentezlenen

Pd@*nano*-ZrO_2 nanokatalizörünün metiamin-boranın (MeAB) hidroliz tepkimesi yoluyla hidrojen üretimindeki katalitik etkinliği (katalizör miktarına, substrat derişimine ve sıcaklığa bağlı katalitik etkinliği) incelenmiştir.

TEŞEKKÜR

2209-A- Lisans Öğrencilerine Yönelik Araştırma Projesi Destek Programı kapsamındaki projemize (Proje No: 1919B012207728) maddi desteği için Türkiye Bilimsel ve Teknolojik Araştırma Kurumu'na teşekkür ederiz.

KAYNAKLAR

[1] T. Liu, Z. Zhang, L. Yan, Z. Zhang, Y. Zhang, Y. Yin, Pd nanoparticles immobilized on aniline-functionalized MXene as an effective catalyst for hydrogen production from formic acid, Int. J. Hydrogen Energy 46 (66) (2021) 33098–33106, https:// doi.org/10.1016/j.ijhydene.2021.07.164.

[2] S.Y. Pang, W.F. Io, L.W. Wong, J. Zhao, J. Hao, Direct and in situ growth of 1T′ MoS_2 and 1T $MoSe_2$ on electrochemically synthesized MXene as an electrocatalyst for hydrogen generation, Nano Energy 103 (2022), 107835, https://doi.org/ 10.1016/j.nanoen.2022.107835.

[3] S. Suragtkhuu, S. Sunderiya, S. Purevdorj, M. Bat-Erdene, B. Sainbileg, M. Hayashi, A.S.R. Bati, J.G. Shapter, S. Davaasambuu, M. Batmunkh, Rhenium anchored Ti_3C_2Tx (MXene) nanosheets for electrocatalytic hydrogen production, Nanoscale Adv. 5 (2023) 349–355, https://doi.org/10.1039/D2NA00782G.

[4] V. Thirumal, R. Yuvakkumar, P.S. Kumar, G. Ravi, A. Arun, R.K. Guduru, D. Velauthapillai, Hetero structured two dimensional materials of MXene and graphene by hydrothermal method for efficient hydrogen production and HER activities, Int. J. Hydrogen Energy 48 (2021) 6478–6487, https://doi.org/ 10.1016/j.ijhydene.2021.12.045.

[5] F. Guo, H. Zou, Q. Yao, B. Huang, Z.H. Lu, Mono-dispersed bimetallic nanoparticles anchored on TiO_2-decorated titanium carbide MXene for efficient hydrogen production from hydrazine in aqueous solution, Renew. Energy 155 (2020) 1293–1301, https://doi.org/10.1016/j.renene.2020.04.047.

[6] M. Kanat, Y. Karatas ,, M. Gülcan, B. Anıl, Preparation and detailed characterization of zirconia nanopowder supported rhodium (0) nanoparticles for hydrogen production from the methanolysis of methylamine-borane in room conditions, Int. J. Hydrogen Energy 43 (50) (2018) 22548–22556, https://doi.org/10.1016/j. ijhydene.2018.10.130.

[7] S. Kaskun, Y. Akinay, M. Kayfeci, Improved hydrogen adsorption of ZnO doped multi-walled carbon nanotubes, Int. J. Hydrogen Energy 45 (60) (2020) 34949–34955, https://doi.org/10.1016/j.ijhydene.2020.06.304.

[8] J. Wang, Y. Yu, W. Xu, H. Yu, W. Zhang, H. Huang, G.-R. Zhang, D. Mei, Covalent triazine framework encapsulated Pd nanoclusters for efficient hydrogen production via ammonia borane hydrolysis, J. Catal. 411 (2022) 72–83, https://doi.org/ 10.1016/j.jcat.2022.05.009.

[9] L. Luconi, G. Tuci, G. Giambastiani, A. Rossin, M. Peruzzini, H2 production from lightweight inorganic hydrides catalyzed by 3d transition metals, Int. J. Hydrogen Energy 44 (47) (2019) 25746–25776, https://doi.org/10.1016/j. ijhydene.2019.08.017.

[10] N. Cao, J. Su, W. Luo, G. Cheng, Graphene supported Ru@ Co core–shell nanoparticles as efficient catalysts for hydrogen generation from hydrolysis of ammonia borane and methylamine borane, Catal. Commun. 43 (2014) 47–51, https://doi.org/10.1016/j.catcom.2013.09.003.

[11] Y Karataş.,, E Kuyuldar.,, H Acidereli.,, M Gülcan.,, F. Şen, Polypyrrole-multi walled carbon nanotube hybrid material supported Pt NPs for hydrogen evolution from the hydrolysis of MeAB at mild conditions, Scıentıfıc Reports, 9, 2019.

[12] Y Karataş .,T, Çetin.,, Y. Akinay, M Gülcan, Synthesis and characterization of Pd doped MXene for hydrogen production from the hydrolysis of methylamine borane: Effect of cryogenic treatment, Journal of the Energy Institute, 109, 2023.

[13] Gutowska, A.; Li, L.; Sin, Y.; Wang, C. M.; Li, X. S.; Linehan, J. C.; Smith, R. S.; Kay, B. D.; Schmid, B.; Shaw, W.; Gutowski, M.; Autrey, T. N., Angew. Chem., Int. Ed. 2005, 44, 3578−3582.

DRAFT

ORAL PRESENTATION

Rh/ZIF-67 Nanokatalizörünün Tepkime-İçi Sentezi, Tanımlanması ve Nitrofenollerin Aminofenollere İndirgenmesindeki Katalitik Performansının İncelenmesi*

Züleyha Kafi[1], Yaşar Karataş[2], Adem Rüzgar[2], Mehmet Gülcan[1]

[1]Van Yüzüncü Yıl Üniversitesi Fen Fakültesi, Kimya Bölümü, Van, Türkiye
[2] Van Yüzüncü Yıl Üniversitesi Muradiye Meslek Yüksekokulu, Kimya ve Kimyasal İşleme Teknolojileri Bölümü, Van, Türkiye

Özet

Su kaynaklarına ve su ekosistemine zarar veren kirleticilerin başında yeterince arıtılmadan çevreye verilen nitrofenol ve türevleri gelmektedir. Bu bileşiklerin arıtılması ve zararsız hale getirilmesi amacıyla uygulanan yöntemlerin başında katalitik indirgeme tepkimeleri gelmektedir. Nitrofenollerin indirgenmesi sonucu elde edilen aminofenoller farklı endüstriyel üretim aşamalarında yoğun olarak kullanılan zararsız ve önemli ara ürünlerdir. Bu çalışmada, 2-nitrofenol ve 4-nitrofenol gibi nitrofenol türevlerinin 2-aminofenol ve 4-aminofenol bileşiklerine katalitik indirgenmesi sürecinde kullanılmak üzere ZIF-67 destekli Rh (0) nanokümeleri hazırlanmış ve hazırlanan katalizörün yapısal ve morfolojik özellikleri XRD, SEM, SEM/EDX ve UV-vis teknikleriyle ortaya konulmuştur. Nitrofenollerin aminofenollere katalitik indirgenmesi oda koşullarında UV-vis spektrofotometre kullanılarak takip edilmiştir.

Anahtar Kelimeler: İndirgenme, Katalizör, Nanoparçacık, Nitrofenol, ZIF-67

Abstract

Nitrophenol and its derivatives, which are released into the environment without sufficient purification, are among the pollutants that harm water resources and the aquatic ecosystem. Catalytic reduction reactions are the primary methods used to purify and render harmless these compounds. Aminophenols obtained as a result of the reduction of nitrophenols are harmless and important intermediate products that are used extensively in different industrial production stages. In this study, ZIF-67 supported Rh (0) nanoclusters were prepared to be used in the catalytic reduction process of nitrophenol derivatives such as 2-nitrophenol and 4-nitrophenol to 2-aminophenol and 4-aminophenol compounds. Structural and morphological properties of the prepared catalyst were revealed by XRD, SEM, SEM/EDX and UV-vis techniques. Catalytic reduction of nitrophenols to aminophenols was monitored using a UV-vis spectrophotometer under room conditions.

Key Words: Reduction, Catalyst, Nanoparticle, Nitrophenol, ZIF-67

GİRİŞ

21. yüz yılda nüfusun hızla artması ve teknolojik gelişmeler üretim ve tüketim süreçlerini önemli ölçüde değiştirerek her alanda daha fazla ve daha tehlikeli atığın açığa çıkmasına neden olmuştur. Bu atıkların arıtılmadan çevreye verilmesi; temiz su kaynaklarına ve bu kaynaklardan etkilenen bütün ekosisteme telafisi çok zor zararlar vererek en önemli küresel sorunlardan biri haline gelmiştir. Gelişmiş ve gelişmekte olan bütün ülkelerin en önemli gündem maddelerinden biri haline gelen su kirliliğine neden olan kirleticilerin başında heterohalkalı aromatik bileşikler gelmektedir [1-5]. Petrokimya, kozmetik ve tekstil başta olmak üzere bir çok endüstri alanında açığa çıkan heterohalkalı aromatik bileşiklerin önemli bir kısmını nitrofenol ve türevleri oluşturmaktadır. Zehirli ve kanserojen yapıları nedeniyle çok düşük konsantrasyonlarda bile son derece toksik etkiler oluşturabilen bu bileşikler sahip oldukları kimyasal kararlılıkları nedeniyle doğal bozunma süreçlerinden etkilenmeden cok büyük bir alana yayılabilmekte ve uzun süre varlığını sürdürebilmektedir [6-8]. Bütün bu özellikleri nedeniyle insan sağlığı açısından son derece tehlikeli kimyasallar olarak kabul edilen nitrofenol ve türevleri ani solunum yolu rahatsızlıklarına deri altı iltihap birikimlerine ve görme kaybı oluşturabilecek derecede göz sorunlarına neden olabilmektedir. Bununla birlikte insan vücudunda protein dejenerasyonu sürecini de başlatabilen bu bileşikler böbrek, pankreas ve karaciğerde geri dönüşü mümkün olmayan zararlara neden olabilmektedir. İnsan vücudunda bu denli ciddi sorunlara yol açan nitrofenol ve türevleri Dünya Sağlık Örgütü tarafından yakından incelenmiş ve oluşturulan içme suyu standartlarına göre izin verilen fenol sınır değeri 0,002 mg/L olarak üye ülkelerin tamamına duyurulmuştur. Dünya sağlık örgütü

tarafından belirlenen sınır değer ABD Çevre Koruma Ajansı tarafından ABD'de uygulanmak üzere 0,001 mg/L'ye düşürülmüştür [9-11]. Bütün bu değerler nitrofenol ve türevlerinin verimli ve çevreci yöntemlerle arıtılmasını önemli bir çalışma alanı haline dönüştürmüş ve bu alanda birbirinden farklı birçok yöntem denenmiştir. Bütün bu yöntemler içerisinde nitrofenollerin aminofenollere indirgenmesi;
Tepkimenin çevreci koşullarda ve ekonomik olarak gerçekleşmesi
Tepkime veriminin sentezlenen yeni nanokatalizör sistemleri ile geliştirilebilmesi
Tepkime sonucu istenmeyen ara ürünlerin oluşmaması
İndirgenme sonucu elde edilen aminofenollerin nitrofenollere göre çok daha az toksik etki göstermesi ve çok farklı endüstri dallarında kullanılan önemli ve değerli bir ara ürün olması
gibi nedenlerle diğer arıtma yöntemlerine göre öne çıkmıştır.
Nitrofenollerin aminofenollere indirgenmesi amacıyla hidrojen kaynağı olarak ekonomik ve çevreci oluşu, kolay saklanabilirliği, su içindeki yüksek çözünürlüğü gibi avantajları nedeniyle çoğunlukla $NaBH_4$ kullanılır. $NaBH_4$ oldukça kolay sayılabilecek koşullar altında hidroliz olur ve hidroliz sonucu $NaOB_2$ ve H_2 elde edilir. Elde edilen H_2 gazının halkadaki nitro grubunu amin grubuna indirgemesi sonucu zararlı ve tehlikeli kimyasallar olan 2-nitrofenol (2-NP) ve 4-nitrofenol (4-NP) zararsız ve değerli bileşikler olan 2-aminofenol (2-AP) ve 4-aminofenole (4-AP) dönüştürülmüş olur. Hidroliz sonucu gerçekleşen indirgenme sürecinin bir diğer avantajı ise ara ürün olarak açığa çıkan çıkan$NaBO_2$' nin zararsız ve geri dönüşümü kolay olan bir bileşik olmasıdır [12-17].

$$NaBH_4 + 2H_2O \rightarrow NaBO_2 + 4H_2$$

Yukarıda değinilen avantajlı durumlara rağmen nitrofenollerin aminofenollere indirgenmesinde sadece $NaBH_4$'ün kullanıldığı çalışmalar verimli bir arıtma ve dönüştürme yöntemi olarak yaygınlaşamamıştır. Bunun en önemli sebebi $NaBH_4$'ün tek başına kullanıldığı indirgenme tepkimelerinin düşük verimle ve çok uzun sürede gerçekleşmesidir. Örneğin herhangi bir katalizör sisteminden faydalanılmadan sadece $NaBH_4$'ün kullanıldığı indirgenme tepkimelerinde 4-NP'ün 4-AP'e dönüşümü 229 dakika ve 2-NP'ün 2-AP'e dönüşümü 239 dakika sürmektedir [18,19]. Bu durum nitrofenollerin aminofenollere verimli bir şekilde dönüştürülebilmesinin ön şartının uygun nanokatalizör sistemlerinin geliştirilmesine bağlı olduğunu göstermektedir. Bu alanda yapılan çalışmalar verimli nanokatalizör sistemlerinin geliştirilmesinin;

ekonomik ve verimli metalin belirlenmesi
metalde zamanla gözlemlenen topaklaşma gibi katalizör performansını ve ömrünü olumsuz etkileyen sorunların giderilmesi amacıyla kullanılacak destek malzemelerinin belirlenmesi ve ihtiyaç durumunda sentezlenmesi
şeklinde iki aşamadan oluştuğunu ortaya koymuştur.

Çalışmada kullanılan nanokatalizör sistemlerinin sentezlenmesi aşamasında destek malzemesi olarak metal-organik çerçeve (MOF) ailesinin ilginç özelliklere sahip bir alt grubu olan zeolitik imidazol çerçeve (ZIF) kullanılmıştır. Yüksek ısısal/kimyasal kararlılık ve yüksek yüzey alanı gibi son derece önemli iki avantaja sahip olan zeolitik imidazol çerçeveler son yıllarda araştırmacıların ilgisini çeken destek malzemelerini başında gelmektedir.
Yapılan çalışmada Hazırlanan ZIF-67 destekli Rh (0) nanokümeleri nitrofenollerin aminofenollere indirgenmesinde kullanılmış ve yüksek katalitik etkinlikle birlikte yüksek tekrar kullanılabilirik performansı rapor edilmiştir.

MATERYAL

Yapılan çalışmada destek malzemesi olarak kullanılan ZIF-67 sentezlendikten sonra Rh@ZIF-67 nanokümeleri elde edildi. Katalizör sistemlerininin hazırlanması ve nitrofenollerin indirgenmesi süresince ihtiyaç duyulan Kobalt (II) nitrat hekzahidrat ($Co(NO_3)_2 \cdot 6H_2O$), 2-metilimidazol ,sodyum borhidrit, 2-nitrofenol (2-NP), 4- nitrofenol (p-nitrofenol, 4-NP) ve Rodyum (III) klorür hidrat ($RhCl_3 \cdot xH2O$) Sigma-Aldrich®ten temin edildi. Tüm cam eşyalar ve teflon kaplı manyetik karıştırma çubukları damıtılmış su ve aseton ile temizlendikten sonra 423 K'deki fırında kurutuldu.

YÖNTEM

Rh@ZIF-67 katalizörünün nitrofenol indirgemesindeki katalitik aktivitesi

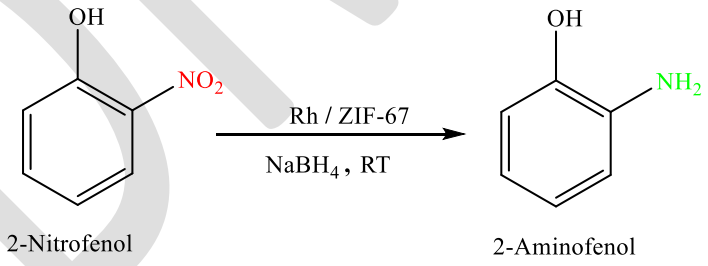
Rh^{+3}@ZIF-67 ön katalizörü iyon değiştirme yöntemiyle hazırlandı. Bu amaçla ilk olarak ZIF-67 sentezlendi. Daha sonra 5,0 mL sulu $RhCl_3 \cdot xH2O$ çözeltisi ile 100 mg ZIF-67 700 rpm'de 12 saat karıştırılarak metal tuzunun katı desteğe tutturulması sağlandı. Elde edilen katalizör (Rh^{+3}@ZIF-67) süzülerek izole edildi ve 373 K'de vakumda kurutuldu. Her substrat için, 5 mg Rh^{+3}@ZIF-67 ve 2,0 mM nitrofenol türevinin sulu çözeltisi (10 mL) (2-NP için 2,78 mg, 4-NP için 2,78 mg) alınarak sıcaklığı 298 K'e ayarlanmış ceketli schlenk içerisinde karıştırıldı. İndirgenmenin başlatılabilmesi için 1,0 ml suda çözülmüş 0,2 mmol $NaBH_4$ (77,2 mg) tepkime ortamına ilave edildi ve katalitik reaksiyon, 800 rpm'lik bir karıştırma hızında başlatıldı. Nitrofenol türevinin karşılık gelen aminofenole dönüşümü, belirli zaman aralıklarında 100 µL reaksiyon çözeltisi çekilerek 1 mL'ye seyreltildikten sonra Shimadzu UV-3600 UV-Vis spektrometresi ile analiz edildi. Nitrofenol türevlerinin optik absorpsiyon spektrumları 2-NP için 414 nm, 4-NP için 399 nm olarak ölçüldü.
Katalitik aktivitenin sıcaklıkla değişimini belirleyebilmek amacıyla 4 farklı sıcaklıkta (293 K, 298 K, 303 K, 308 K) deneyler yapıldı. Benzer şekilde katalizör miktarı ve substrat derişiminin katalitik performansa etkisini belirleyebilmek amacıyla farklı derişim (0,5 mM, 1 mM, 2 mM, 3 mM) ve farklı miktarlarda (1 mg, 3 mg, 5 mg, 7 mg) deneyler yapıldı ve elde edilen veriler kullanılarak katalitik etkinlikte meydana gelen değişimler hesaplandı.

Rh@ZIF-67 katalizörünün nitrofenol indirgemesinde tekrar kullanılabilirlik performansının araştırılması

Her substrat için katalitik reaksiyonun tamamlanmasından sonra Rh@ZIF-67 katalizörü, süzme yoluyla ayrıldı ve etanol-su karışımı ile temizlendi ve 373 K'de vakumda kurutuldu. İyice kurutulmuş katalizör miktarı tartıldıktan sonra reaksiyon kabına eşit miktarda substrat ilave edildi ve katalitik reaksiyon yeniden başlatıldı.

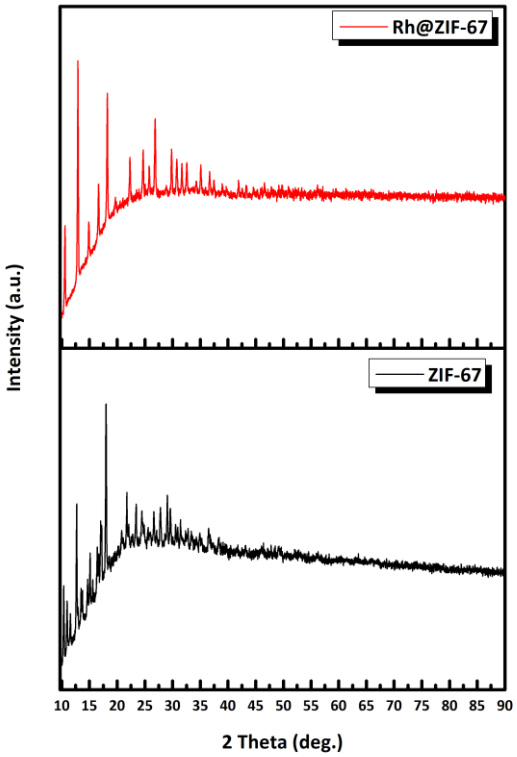
BULGULAR VE TARTIŞMA

Yapılan çalışmada oldukça yüksek yüzey alanı ve kararlılığa sahip bir ZIF üyesi olan ZIF-67'nin destek malzemesi olarak kullanıldığı Rh@ZIF-67 nanokümeleri klasik ıslak-emdirme yöntemi ile tepkime-içi koşullarda hazırlanmış, karakterize edilmiş ve indirgeyici olarak sodyum borhidrür ($NaBH_4$) varlığında yaygın olark karşılaşılan iki nitrofenolün (2-nitrofenol ve4-nitrofenol) katalitik indirgenmesinde katalizör olarak kullanılmıştır.



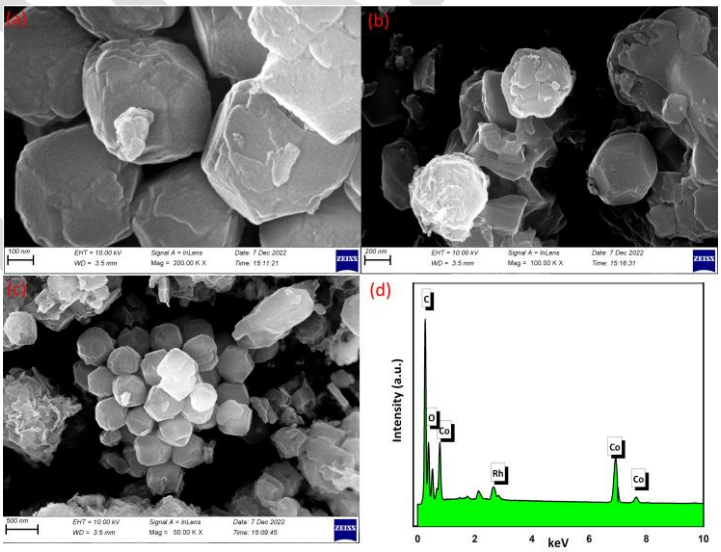
Şekil 1. 2-nitrofenolün 2-aminofenole indirgenmesi

Katalitik hidrojenasyonda kullanılan nanokatalizörde destek malzemesi olarak kullanılan ZIF-67'ye ve Rh@ZIF-67 nanokatalizörüne ait XRD analizi şekil 2'de gösterilmiştir. İncelenen XRD desenleri, metalin yüzeye tutunması sonucu destek malzemesinin yapısında herhangi bir bozulma olmadığını göstermektedir. Ayrıca Rh@ZIF-67 desenleri ile ZIF-67 desenlerinin yüksek oarnda benzerlik gösterdiği ve literatür ile uyumlu olduğu da belirlenmiştir [20].



Şekil 2. ZIF-67 destekli Rh nanokatalizörünün 2θ = 10- 90˚ aralığındaki XRD desenleri

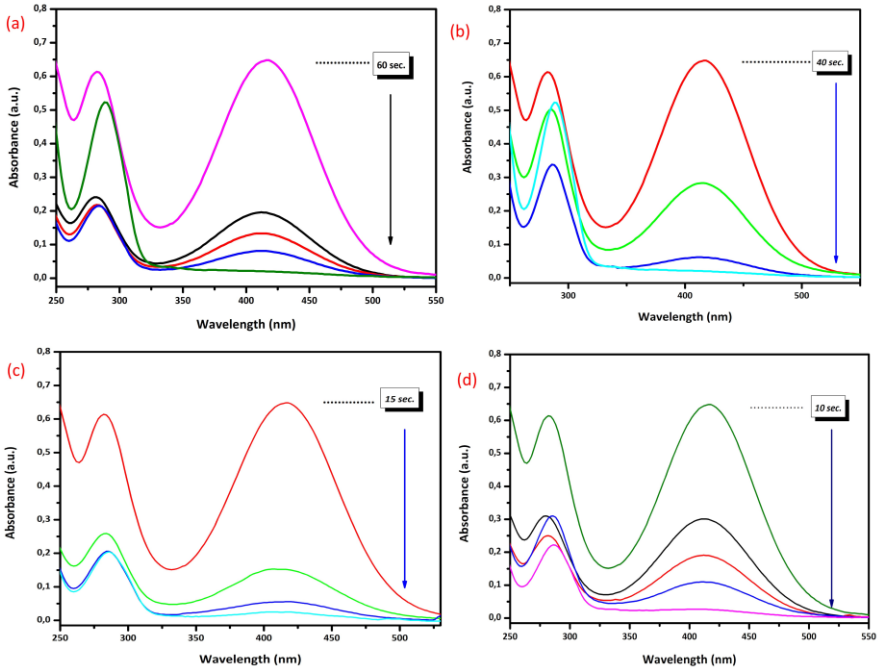
ZIF-67 destekli Rh nanokümelerin parçacık büyüklük dağılımları taramalı elektron mikroskobu (SEM) ile karakterize edildi. Şekil 3. (a-c), tepkime-içi koşullarda 4-nitrofenolün indirgenmesi sonucu elde edilen Rh@ZIF-67 örneklerine ait 100, 200 ve 500 nm büyüklükteki SEM görüntülerini göstermektedir. Görüntülerden de anlaşılacağı üzere Rh (0) nanokümeleri ZIF-67 yüzeyine düzgün bir şekilde tutunmuştur. Bununla birlikte yapılan SEM EDX analizinde de ZIF-67 yapısındaki elementler ve Rh metali görülmektedir (Şekil 3.d)



Şekil 3. Tepkime-içi koşullarda (a) 100 nm, (b) 200 nm, c) 500 nm 2- nitrofenolün indirgenmesi sonucu elde edilen Rh@ZIF-67 katalizörüne ait SEM görüntüleri, (d) SEM EDX analizi

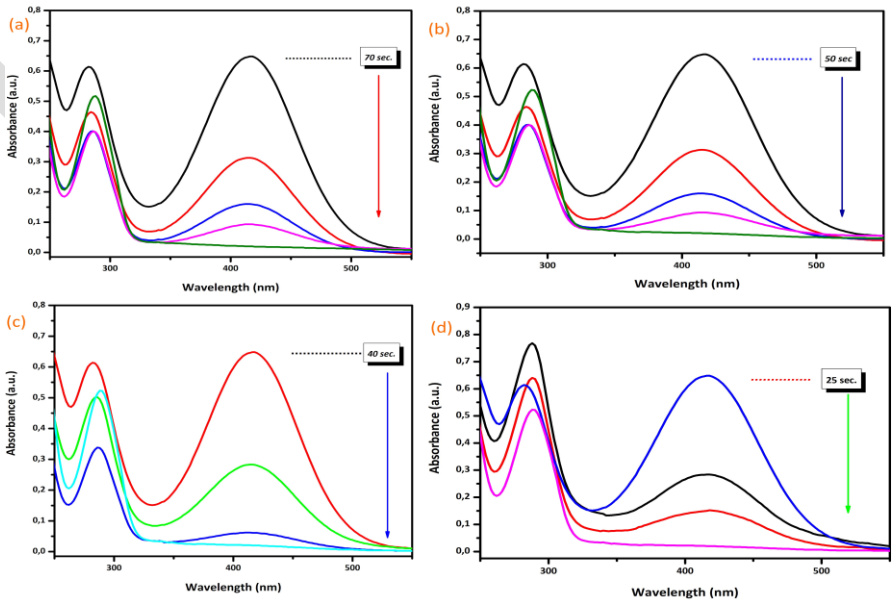
Alkali çözeltide en güçlü absorpsiyon bandı 2-nitrofenol için 414 nm, 4-nitrofenol için 399 nm olarak belirlendi, Absorbsiyon tepe noktasının değişiminin belirli zaman aralıklarında takip edilmesi sonucu katalitik

hidrojenasyon ile ilgili kinetik parametreler elde edildi. Sıcaklığın, nitrofenollerin Rh@ZIF-67 nanokatalizörü varlığında gerçekleştirilen katalitik indirgenme tepkimesi üzerindeki etkisini incelemek için 293, 298, 303 ve 308 K sıcaklıklarında deneyler gerçekleştirildi. Elde edilen UV-vis spektrumları incelendiğinde sıcaklık artışıyla indirgenme hızının doğru orantılı değiştiği belirlendi 2-nitrofenolün katalitik hidrojenasyonu 293 K'de 60 saniyede 308 K'de ise 10 saniyede gerçekleşmiştir (Şekil 4).



Şekil 4. Farklı sıcaklıklarda 2-nitrofenolün sulu $NaBH_4$ varlığında indirgenmesine ilişkin UV-Vis spektrumları.

Katalizör miktarının tepkime hızına etkisini belirlemek amacıyla yapılan deneylerde katalizör miktarının düşük olduğu durumlarda indirgenme reaksiyonunun beklenildiği gibi daha yavaş gerçekleştiği belirlendi . Ortamda bulunan katalizör miktarının artması etkinliğin ve tepkime hızının artmasına yol açmıştır. 2-nitrofenolün katalitik hidrojenasyonu katalizör miktarı 1 mg iken 70 saniye sürmekte iken katalizör miktarı 7 mg'a çıkarıldığında 25 saniye gibi çok daha kısa bir sürede gerçekleşmektedir (Şekil 5).



Şekil 5. Farklı katalizör miktarlarının 2-nitrofenolü sulu $NaBH_4$ varlığında indirgenmesine ilişkin UV-Vis spektrumları.

Katalitik performansın en önemli ölçütleri olarak kabul edilen izole edilebilirlik ve tekrar kullanılabilirlik, 298 K sıcaklıkta Rh@ZIF-67 nanokatalizörü varlığında nitrofenollerin katalitik indirgenmesinde test edildi. Yapılan çalışmada 2-nitrofenolün tamamen indirgenmesinden sonra, Rh@ZIF-67 nanokatalizörü süzülüp su ile yıkandıktan sonra kurutuldu. Elde edilen Rh@ZIF-67 nanokatalizörü tekrardan 2-nitrofenolün indirgenmesinde kullanıldı. Rh@ZIF-67 katalizörünün katalitik indirgenme tepkimelerinde yine aktif olduğu belirlendi. Beş katalitik çalışmanın sonunda elde edilen sonuçlar Rh@ZIF-67 nanokatalizörünün 2-nitrofenolün indirgenmesinde 5. katalitik yeniden kullanımda bile %100 oranında tam dönüşüm sağladığı ve %80 oranında etkinliğini koruduğu belirlendi.

SONUÇ

Bu çalışmada, ZIF-67 destekli Rodyum nanopartikülleri (Rh@ZIF-67) indirgeyici olarak $NaBH_4$ varlığında 2-nitrofenol ve 4-nitrofenolün katalitik hidrojenasyonunda kullanılmak üzere katalizör olarak sentezlendi, karakterize edildi ve test edildi. Bu çalışma sonucunda elde edilen en çarpıcı sonuçlar şu şekilde sıralanbilir.:

Rh@ZIF-67nanokatalizörü nitrofenollerin indirgenmesinde kullanılmak üzere klasik ıslak emdirme yöntemi ile tepkime-içi koşullarda hazırlandı.
Rh@ZIF-67 nanokatalizörü P-XRD, FTIR, SEM gibi çeşitli tekniklerle karakterize edildi Elde edilen sonuçlar Rh nanoparçacıklarının ZIF-67 yüzeyine homojen bir şekilde dağıldığını ortaya koymaktadır.
Rh@ZIF-67 nanokatalizörünün hem katalitik aktivitesi hem de tekrar kullanılabilirlik performansı, 2-NP ve 4-NP'un sulu $NaBH_4$ çözeltisi içindeki hidrojenasyonlarında araştırıldı. Rh@ZIF-67'nin nitrofenollerin karşılık gelen aminofenollere dönüşümünde oldukça aktif bir nanokatalizör olduğu tespit edildi.
Ayrıca Rh@ZIF-67 nanokatalizörü katalitik çalışmalar sırasında oldukça yüksek bir dayanıklılık sergileyerek 5. katalitik yeniden kullanımda bile %80 oranında etkinliğini korudu.

Teşekkür

** 2209-A- Lisans Öğrencilerine Yönelik Araştırma Projesi Destek Programı kapsamındaki projemize (Proje No:1919B012206248) maddi desteği için Türkiye Bilimsel ve Teknolojik Araştırma Kurumu'na teşekkür ederiz.)*

KAYNAKÇA

Lim, S., Shi, J. L., von Gunten, U., & McCurry, D. L. (2022). Ozonation of organic compounds in water and wastewater: A critical review. *Water research*, *213*, 118053.

Ilani, T., Schulz, E., & Chefetz, B. (2005). Interactions of organic compounds with wastewater dissolved organic matter: role of hydrophobic fractions. *Journal of Environmental Quality*, *34*(2), 552-562.

Koubaissy, B., Joly, G., Batonneau-Gener, I., & Magnoux, P. (2011). Adsorptive removal of aromatic compounds present in wastewater by using dealuminated faujasite zeolite. *Industrial & Engineering Chemistry Research*, *50*(9), 5705-5713.

Yang, L., Liu, Y., Li, C., Liu, Z., Liu, X., Wei, C., ... & Zhang, A. (2022). Biodegradation time series characteristics and metabolic fate of different aromatic compounds in the biochemical treatment process of coal chemical wastewater. *Bioresource Technology*, *361*, 127688.

Hojjati-Najafabadi, A., Mansoorianfar, M., Liang, T., Shahin, K., Wen, Y., Bahrami, A., ... & Vasseghian, Y. (2022). Magnetic-MXene-based nanocomposites for water and wastewater treatment: A review. *Journal of Water Process Engineering*, *47*, 102696.

Padervand, M., Ghasemi, S., Hajiahmadi, S., Rhimi, B., Nejad, Z. G., Karima, S., ... & Wang, C. (2022). Multifunctional Ag/AgCl/ZnTiO3 structures as highly efficient photocatalysts for the removal of nitrophenols, CO2 photoreduction, biomedical waste treatment, and bacteria inactivation. *Applied Catalysis A: General*, *643*, 118794.

Akhtar, T., Hill, A. J., Bhat, A., Schwank, J. W., Nasir, H., Bukhari, S. A. B., & Sitara, E. (2023). Fabrication of ruthenium doped Ag@ TiO2 core-shell nanophotocatalyst for the efficient reduction of nitrophenols. *Applied Surface Science*, *630*, 157491.

Lu, Y., Wan, X., Li, L., Sun, P., & Liu, G. (2021). Synthesis of a reusable composite of graphene and silver nanoparticles for catalytic reduction of 4-nitrophenol and performance as anti-colorectal carcinoma. *Journal of Materials Research and Technology*, *12*, 1832-1843.

Kiani, Z., Zhiani, R., Khosroyar, S., Motavalizadehkakhky, A., & Hosseiny, M. (2021). UiO-66/btb/Pd as a stable catalyst reduction of 4-nitrophenol into 4-aminophenol. *Inorganic Chemistry Communications*, *124*, 108382.

Dalla Nora, F. B., Lima, V. V., Oliveira, M. L., Hosseini-Bandegharaei, A., de Lima Burgo, T. A., Meili, L., & Dotto, G. L. (2020). Adsorptive potential of Zn–Al and Mg–Fe layered double hydroxides for the removal of 2–nitrophenol from aqueous solutions. *Journal of Environmental Chemical Engineering*, *8*(4), 103913.

Achille, Y. L., Engelbert, A. J., Gabin, A. A., Pauline, A., Parfait, D., Habib, G., & Marini Djang'eing'a, R. (2022). Quality Control of Paracetamol Generic Tablets Marketed in Benin and Search of Its Two Impurities P-Aminophenol and P-Nitrophenol by HPLC-UV/Visible. *American Journal of Analytical Chemistry*, *13*(11), 449-460.

Roy, D., Neogi, S., & De, S. (2021). Highly efficient reduction of p-Nitrophenol by sodium borohydride over binary ZIF-67/g-C3N4 heterojunction catalyst. *Journal of Environmental Chemical Engineering*, *9*(6), 106677.

Nguyen, T. B., Huang, C. P., & Doong, R. A. (2019). Enhanced catalytic reduction of nitrophenols by sodium borohydride over highly recyclable Au@ graphitic carbon nitride nanocomposites. *Applied Catalysis B: Environmental*, *240*, 337-347.

Wei, Y., Huang, X., Wang, J., Yu, H., Zhao, X., & Cheng, D. (2017). Synthesis of bifunctional non-noble monolithic catalyst Co-WP/carbon cloth for sodium borohydride hydrolysis and reduction of 4-nitrophenol. *international journal of hydrogen energy*, *42*(41), 25860-25868.

Gupta, V. K., Atar, N., Yola, M. L., Üstündağ, Z., & Uzun, L. (2014). A novel magnetic Fe@ Au core–shell nanoparticles anchored graphene oxide recyclable nanocatalyst for the reduction of nitrophenol compounds. *Water research*, *48*, 210-217.

Mandlimath, T. R., & Gopal, B. (2011). Catalytic activity of first row transition metal oxides in the conversion of p-nitrophenol to p-aminophenol. *Journal of Molecular Catalysis A: Chemical*, *350*(1-2), 9-15.

Soğukömeroğulları H. G., Karataş Y., Çelebi M., Gülcan M., Sönmez M., Zahmakıran M.(2019) Palladium nanoparticles decorated on amine functionalized graphene nanosheets as excellent nanocatalyst for the hydrogenation of nitrophenols to aminophenol counterparts. *Journal of Hazardous Materıals*, 369, 96-107.

Elfiad, A., Galli, F., Djadoun, A., Sennour, M., Chegrouche, S., Meddour-Boukhobza, L., & Boffito, D. C. (2018). Natural α-Fe2O3 as an efficient catalyst for the p-nitrophenol reduction. *Materials Science and Engineering: B*, *229*, 126-134.

Bhowmik, T., Kundu, M. K., & Barman, S. (2015). Ultra small gold nanoparticles–graphitic carbon nitride composite: an efficient catalyst for ultrafast reduction of 4-nitrophenol and removal of organic dyes from water. *RSC Advances*, *5*(48), 38760-38773.

Shao, Y., & Zeng, H. C. (2021). Pt, Ir, Ru, and Rh nanoparticles supported on ZIF-67 nanocubes for evaluation of hydrogen spillover ability of noble metals. *ACS Applied Nano Materials*, *4*(6), 6030-6044.

ORAL PRESENTATION

Dimetilamin-Borandan Hidroliz Yoluyla Hidrojen Üretimi için Amin Fonksiyonel Silika Destekli Rutenyum Nanokümelerinin Hazırlanması*

Halime Çiçek[1], Mehmet Gülcan[1]

[1]Van Yüzüncü Yıl Üniversitesi Fen Fakültesi, Kimya Bölümü, Van, Türkiye

Özet

Bu çalışmada amin-fonksiyonel silika (SiO_2-NH_2) destekli Ru (0) nanokümeleri hazırlanarak DMAB'ın hidrolizinden hidrojen üretimindeki katalitik performansları incelendi. Hazırlanan SiO_2-NH_2 destekli Ru (0) nanokümeleri, ICP-OES, XRD, SEM, SEM/EDX karakterizasyon teknikleriyle tanımlandı. Hazırlanan SiO_2-NH_2 destekli Ru (0) nanokümelerinin DMAB'ın hidroliz tepkimesinde tekrar kullanılabilirlik performansları da incelenerek farklı sıcaklıklarda hidroliz tepkimeleri gerçekleştirildi.

Anahtar Kelimeler: Amin fonksiyonel silika, Dimetilamin-boran, Hidrojen, Hidroliz, Nanoküme

Preparation of Amine Functional Silica Supported Ruthenium Nanoclusters for Hydrogen Production via Hydrolysis from Dimethylamine-Borane

Abstract

In this study, amine-functional silica (SiO_2-NH_2) supported Ru (0) nanoclusters were prepared and their catalytic performance in hydrogen production from the hydrolysis of DMAB was examined. The prepared SiO_2-NH_2 supported Ru (0) nanoclusters were identified by ICP-OES, XRD, SEM, SEM/EDX characterization techniques. The reusability performances of the prepared SiO_2-NH_2 supported Ru (0) nanoclusters in the hydrolysis reaction of DMAB were also examined and hydrolysis reactions were carried out at different temperatures.

Keywords: Amine functional silica, Dimethylamine-borane, Hydrogen, Hydrolysis, Nanocluster

GİRİŞ

Geleneksel fosil yakıtlar, modern toplumda çok yönlü kullanımlar için devasa enerjiler sağlasa da, bunların büyük ölçekli tüketimleri ciddi enerji krizlerine ve çevre kirliliğine yol açmaktadır. Sürdürülebilir kalkınma açısından hidrojen, yüksek enerji yoğunluğu ve temiz yanma özelliğinden dolayı fosil yakıtlara alternatif olarak kabul edilmektedir [1-3]. Ancak hidrojenin güvenli bir şekilde depolanması ve taşınması hala büyük ölçekli uygulamaları engellemektedir. Bu sorunu hafifletmek için, katı hidrojen depolama malzemelerinin, hidrojen kullanımlarındaki acil gereksinimi karşılamak için güvenli ve etkili bir strateji olduğu gösterilmiştir [4-8]. Buna göre dimetil amin-boran (DMAB), hidrojen üretimi için en çok araştırılan hidrojen depolama malzemelerinden biri olarak kabul edilmiştir [9]. Hızlı reaksiyon kinetiğini hızlandırmak için DMAB hidrolizinden hidrojen salınmasını tetikleyecek etkili bir katalizör gereklidir.

Heterojen katalizörler, katalizör ayırma ve geri dönüşümdeki faydalarından dolayı yoğun ilgi görmüştür [10-18]. Bununla birlikte Ru, DMAB dehidrojenasyonuna yönelik yüksek verimliliği nedeniyle artan araştırma ilgisini uyandırırken yüksek fiyatı ve sınırlı arzı büyük ölçekli uygulamaları kısıtlamaktadır. Düşük yükleme ve yüksek katalitik verime sahip Ru bazlı katalizörlerin geliştirilmesine ihtiyaç vardır. Önceki çalışmalar, katalitik verimliliğin büyük ölçüde soy metal parçacıklarının boyutlarına bağlı olduğunu göstermiştir.

Bu çalışma da öncelikle amin-fonksiyonel silika (SiO_2-NH_2) destekli rutenyum nanokümeleri (Ru@SiO_2-NH_2) ıslak emdirme-indirgeme yöntemiyle hazırlanmıştır. Ardından hazırlanmış Ru@SiO_2-NH_2 nanokatalizörü ICP-OES (İndüktif Eşleşmiş Plazma-Optik Emisyon Spektrometresi), XRD (X-Işınları Kırınımı), SEM (Taramalı Elektron Mikroskopu), SEM/EDX (Taramalı Elektron Mikroskopu/Enerji Dağılımlı X-Işını Spektroskopisi gibi karakterizasyon teknikleriyle tanımlanmıştır. Daha sonra Ru@SiO_2-NH_2

nanokatalizörünün katalitik performansı DMAB'ın hidrolizinden hidrojen üretimi tepkimesinde etkinlik ve dönüşüm açısından test edilmiştir.

MATERYAL

SiO_2-NH_2, DMAB, $RuCl_3 \cdot xH_2O$ kimyasalları ticari olarak temin edilmiştir.

METOT

Bu çalışma kapsamında gerçekleştirilmesi planlanan işlemler aşağıda maddeler halinde sunulmuştur:

SiO_2-NH_2 Destekli Ru (0) Nanokümelerinin Hazırlanması: Çalışmada SiO_2-NH_2 destekli Ru (0) nanokümeleri, çözelti fazında metal tuzunun destekleyici yüzeyine depolanması ve daha sonra $NaBH_4$ kullanılarak indirgenmesiyle hazırlandı. Bu amaçla, Ru metalinin başlangıç tuzu ($RuCl_3 \cdot xH_2O$) SiO_2-NH_2 katı destek malzemesinin yüzeyine emdirilmesi (3 saat boyunca 750 rpm karıştırma hızında karıştırılarak) ve daha sonra $NaBH_4$ varlığında indirgenmesi sağlandı.

SiO_2-NH_2 Destekli Ru (0) Nanokümelerinin DMAB'ın Hidroliz Tepkimesinde Katalitik Etkinliğinin İncelenmesi: Çalışmanın bu kısmında hazırlanmış SiO_2-NH_2 destekli Ru (0) nanokümelerin DMAB'ın hidrolizi tepkimesinde katalitik etkinliği, tepkime sonucu açığa çıkacak gazın hacimsel ölçümü ve katalitik etkinlik göz önüne alınarak incelendir. Hazırlanmış SiO_2-NH_2 destekli Ru (0) nanokümelerin etkinlik, seçicilik, dönüşüm ve dayanıklılık yönünden en iyi katalitik performansa sahip tepkime koşulları belirlendi.

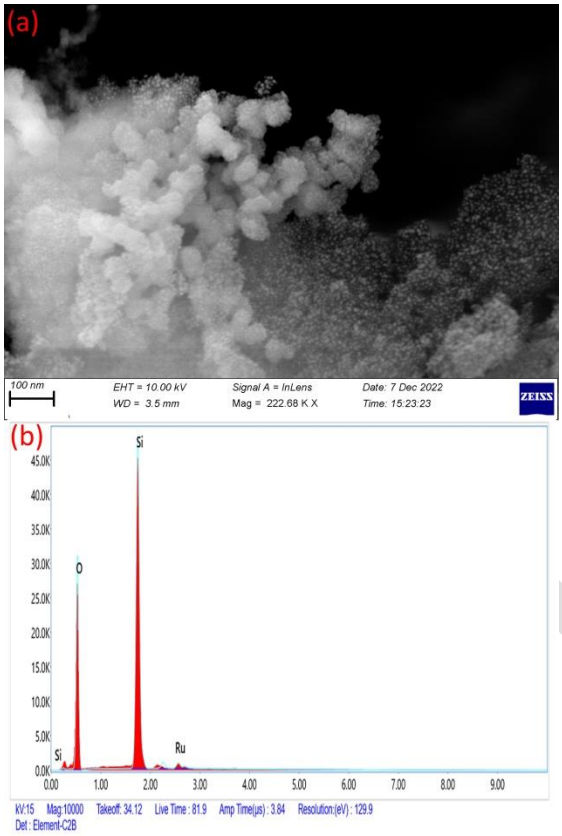
SiO_2-NH_2 Destekli Ru (0) Nanokümelerinin Tanımlanması: Çalışma da SiO_2-NH_2 destekli Ru (0) nanokümeleri ICP-OES, XRD, SEM, SEM/EDX gibi ileri analitiksel yöntemler kullanılarak tanımlanmıştır. Bu analitiksel yöntemlerden ICP-OES; SiO_2-NH_2 desteğine tutturulmuş Ru metal miktarının tespiti, XRD; metal nanokümelerinin oluşumu sonucu SiO_2-NH_2 destekl malzemesinin kristal yapısının incelenmesi, XPS; metalin indirgenme sonrası yükseltgenme basamağının incelenmesi ve yüzey kompozisyonunun belirlenmesi, SEM; Ru (0) nanokümelerinin parçacık boyut analizi ve morfolojilerinin belirlenmesi, SEM/EDX; SEM analizi sırasında seçili bölgelerde metal dağılımının elementel olarak incelenmesi amacıyla kullanılmıştır.

BULGULAR ve TARTIŞMA

Çalışmanın ilk aşamasında SiO_2-NH_2 destekli Ru (0) nanokümeleri ıslak emdirme-indirgeme yöntemiyle hazırlanarak DMAB'nin hidrolizi tepkimesi yoluyla hidrojen üretimindeki katalitik performanslar incelenmiştir.

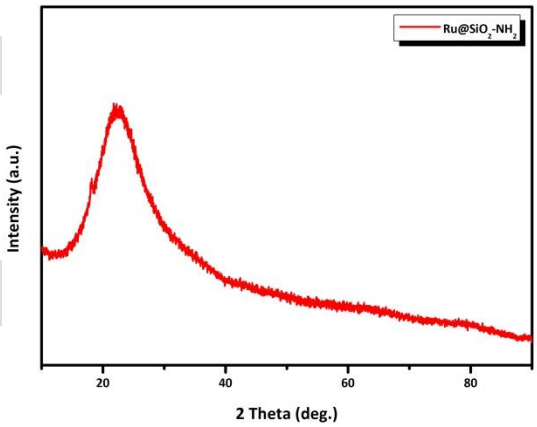
İlk olarak Morfolojik olarak Ru@SiO_2-NH_2 katalizörünün incelenmesi için SEM analizleri yapılmıştır ve bu görüntüler Şekil 1'de verilmektedir. 100 nm'de alınan SEM görüntüsünden Ru metallerinin destek malzemesinin yüzeyine homojen olarak dağıldığı görülmektedir (Şekil 1 (a)). Ayrıca SEM analizi sırasında yapılan haritalandırmada hem Ru metalinin (Şekil 1 (b)) ve destek malzemesinde bulunan Si, O ve Ru (Şekil 1 (b)) elementlerinin varlığı açıkça anlaşılmaktadır.

DRAFT



Şekil 1. (a) Ru@SiO_2-NH_2 katalizörüne ilişkin 100 nm ölçekli SEM görüntüsü (b) Taramalı Elektron Mikroskopu/Enerji Dağılımlı X-Işını Spektroskopisi görüntüsü

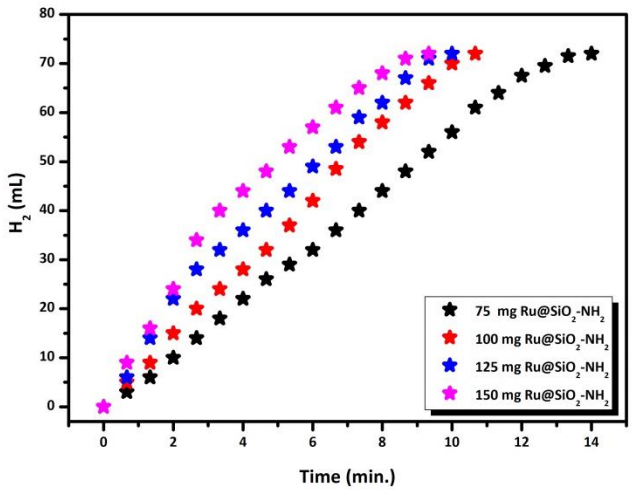
Şekil 2'de SiO_2-NH_2 ve Ru@SiO_2-NH_2 örneklerinin XRD desenleri incelendiğinde Ru metalinin destek malzemesine yüklenmesi önce ve sonrası XRD desenleri incelendiğinde destek malzemesi olan SiO_2-NH_2'nin kristal yapısında herhangi bir bozunmanın olmadığı tespit edilmiştir.



Şekil 2. SiO_2-NH_2 ve Ru@SiO_2-NH_2 örneklerine ait XRD desenleri.

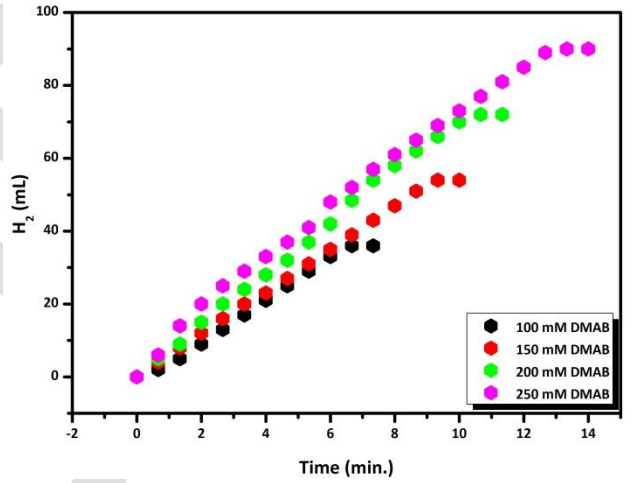
Ru@SiO_2-NH_2 nanokatalizörü ile katalizlenen DMAB'nin hidrolizi tepkimesinde, tepkime hızına katalizör miktarlarının etkisini incelemek için, MeAB derişimi (23 mg, 100 mM) sabit tutularak farklı miktarlarda Ru@SiO_2-NH_2 nanokatalizör kullanımıyla DMAB'nin hidroliz tepkimesi 298 K sıcaklıkta

gerçekleştirilmiştir. Şekil 3'da farklı derişimlerde $Ru@SiO_2$-NH_2 nanokatalizörü ($Ru@$ SiO_2-NH_2 = 75 mg, 100 mg, 125mg ve 150 mg) varlığında katalizlenen DMAB'nin hidroliz tepkimesinde açığa çıkan H_2 gazın DMAB gaz hacmine karşı zaman grafiği verilmektedir.



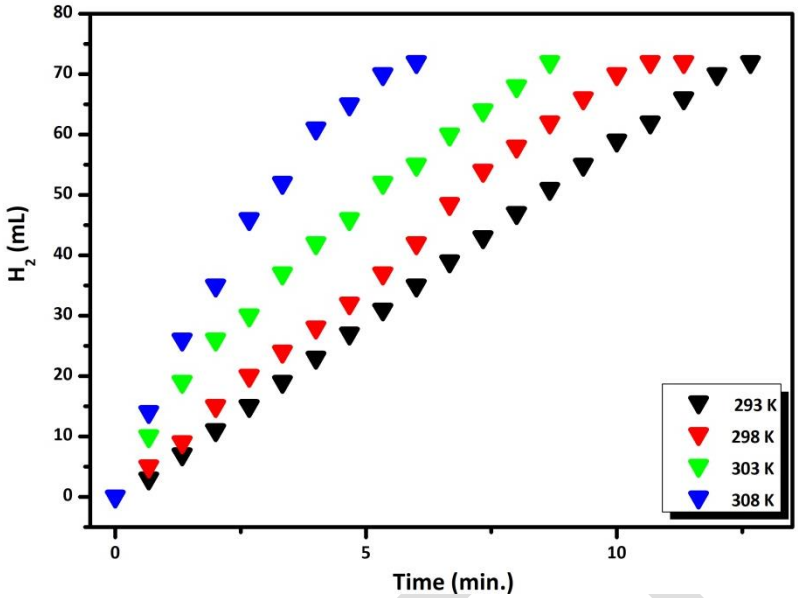
Şekil 3. Farklı miktarlarda $Ru@SiO_2$-NH_2 katalizörü ile katalizlenen DMAB'nin hidroliz tepkimesinde açığa çıkan gazın (H_2) DMAB gaz hacmine karşı zaman grafiği.

$Ru@SiO_2$-NH_2 nanokatalizörü ile katalizlenen DMAB'nin hidroliz tepkimesinde tepkime hızına substrat derişiminin etkisini incelemek amacıyla, nanokatalizör derişimi (100 mg) sabit tutularak farklı derişimlerde DMAB kullanılarak katalitik hidroliz tepkimesi 298 K sıcaklıkta gerçekleştirilmiştir. Şekil 4'de farklı derişimlerde ([DMAB] = 100.0 mM, 150 mM, 200 mM, ve 250 mM) substrat kullanılarak katalizlenen HB'nin hidroliz tepkimesinde açığa çıkan H_2 gazın DMAB gaz hacmine karşı zaman grafiği verilmektedir.



Şekil 4. Farklı derişimlerde DMAB kullanılarak $Ru@SiO_2$-NH_2 nanokatalizörü ile katalizlenen DMAB'nin hidroliz tepkimesinde açığa çıkan gazın (H_2) gaz hacmine karşı zaman grafiği.

$Ru@$ SiO_2-NH_2 nanokatalizörü varlığında DMAB'nin hidroliz tepkimesine sıcaklığın etkisini belirlemek amacıyla katalizör ve substrat miktarları sabit tutularak sıcaklık değişimi etkisi incelenmiştir. Şekil 5'te farklı sıcaklıklarda gerçekleştirilen hidroliz tepkimesinde açığa çıkan H_2 gazın DMAB gaz hacmine karşı zaman grafiği verilmektedir. Beklenildiği gibi katalitik hidroliz tepkime hızının sıcaklıkla doğru orantılı olarak arttığı görülmüştür.



Şekil 5. Ru@SiO_2-NH_2 nanokatalizörü ile katalizlenen DMAB'nin farklı sıcaklıklarda gerçekleştirilen hidroliz tepkimesinde açığa çıkan gazın (H_2) DMAB gaz hacmine karşı zaman grafiği.

SONUÇ

Bu çalışma kapsamında elde edilen sonuçlar özetle şu şekilde sıralanabilir:

SiO_2-NH_2 destekli rutenyum nanokümeleri ıslak emdirme-indirgeme yöntemiyle sentezlendi.
Sentezlenen Ru@SiO_2-NH_2 nanokatalizörü SEM/SEM-EDX analizleri ile morfolojik olarak tanımlanması yapılmıştır.

Sentezlenen Ru@SiO_2-NH_2 nanokatalizörünün Dimetil-aminboranın (DMAB) hidroliz tepkimesi yoluyla hidrojen üretimindeki katalitik etkinliği (katalizör miktarına, substrat derişimine ve sıcaklığa bağlı katalitik etkinliği) incelenmiştir.

TEŞEKKÜR

2209-A- Lisans Öğrencilerine Yönelik Araştırma Projesi Destek Programı kapsamındaki projemize (Proje No:1919B012206282) maddi desteği için Türkiye Bilimsel ve Teknolojik Araştırma Kurumu'na teşekkür ederiz.

KAYNAKLAR

1. Ding, R.; Chen, Q.; Luo, Q.; Zhou, L.; Wang, Y.; Zhang, Y.; Fan, G. Salt template-assisted in situ construction of Ru nanoclusters and porous carbon: Excellent catalysts toward hydrogen evolution, ammonia-borane hydrolysis, and 4-nitrophenol reduction. Green Chem. **2020**, 22, 835–842.
2. Ming, M.; Zhang, Y.; He, C.; Zhao, L.; Niu, S.; Fan, G.; Hu, J.S. Room-temperature sustainable synthesis of selected platinum group metal (PGM = Ir, Rh, and Ru) nanocatalysts well-dispersed on porous carbon for e_cient hydrogen evolution and oxidation. Small **2019**, 15, 1903057.
3. Yao, Q.; Lu, Z.H.; Huang, W.; Chen, X.; Zhu, J. High Pt-like activity of the Ni–Mo/graphene catalyst for hydrogen evolution from hydrolysis of ammonia borane. J. Mater. Chem. A **2016**, 4, 8579–8583.
4. Zhang, X.; Zhang, Q.; Xu, B.; Liu, X.; Zhang, K.; Fan, G.; Jiang,W. Efficient hydrogen generation fromthe NaBH4 hydrolysis by cobalt-based catalysts: Positive roles of sulfur-containing salts. ACS Appl. Mater. Interfaces **2020**, 12, 9376–9386.
5. Xu, C.; Ming, M.; Wang, Q.; Yang, C.; Fan, G.; Wang, Y.; Gao, D.; Bi, J.; Zhang, Y. Facile synthesis of e_ective Ru nanoparticles on carbon by adsorption-low temperature pyrolysis strategy for hydrogen evolution. J. Mater. Chem. A **2018**, 6, 14380–14386.
6. Mao, M.; Chen, Q.; Wu, J.; Fan, G. Anchoring and space-confinement e_ects to synthesize ultrasmall Pd nanoparticles for e_cient ammonia borane hydrolysis. Int. J. Hydrog. Energy **2020**.
7. Yao, Q.; Lu, Z.H.; Yang, Y.; Chen, Y.; Chen, X.; Jiang, H.L. Facile synthesis of graphene-supported

Ni-CeOx nanocomposites as highly e_cient catalysts for hydrolytic dehydrogenation of ammonia borane. Nano Res. **2018**, 11, 4412–4422.
8. Yao, Q.; Yang, K.; Hong, X.; Chen, X.; Lu, Z.H. Base-promoted hydrolytic dehydrogenation of ammonia borane catalyzed by noble-metal-free nanoparticles. Catal. Sci. Technol. **2018**, 8, 870–877.
9. Grochala, W.; Edwards, P.P. Thermal decomposition of the non-interstitial hydrides for the storage and production of hydrogen. Chem. Rev. **2004**, 104, 1283–1316.
10. Sen, B.; Kuzu, S.; Demir, E.; Okyay, T.O.; Sen, F. Hydrogen liberation from the dehydrocoupling of dimethylamine–borane at room temperature by using novel and highly monodispersed RuPtNi nanocatalysts decorated with graphene oxide. Int. J. Hydrog. Energy **2017**, 42, 23299–23306.
11. Chen, Q.; Yao, N.; Ming, M.; Fan, G.; Zhang, Y.; Hu, J.H. Sustainable synthesis of supported metal nanocatalysts for electrochemical hydrogen evolution. Chin. J. Catal. **2020**, 41, 1791–1811.
12. Xu, C.; Chen, Q.; Ding, R.; Huang, S.; Zhang, Y.; Fan, G. Sustainable solid-state synthesis of uniformly distributed PdAg alloy nanoparticles for electrocatalytic hydrogen oxidation and evolution. Chin. J. Catal. **2020**, 42, 251–258.
13. Kaya, M.; Zahmakiran, M.; Özkar, S.; Volkan, M. Copper(0) nanoparticles supported on silica-coated cobalt ferrite magnetic particles: Cost e_ective catalyst in the hydrolysis of ammonia-borane with an exceptional reusability performance. ACS Appl. Mater. Interfaces **2012**, 4, 3866–3873.
14. Dong, Y.; Chen, Q.; Qiu, C.; Ma, X.;Wang, Y.; Sun, T.; Fan, G. Synergistic catalysis of Pd–Ni (OH)2 hybrid anchored on porous carbon for hydrogen evolution from the dehydrogenation of formic acid. Int. J. Hydrog. Energy **2020**, 45, 12849–12858.
15. Ming, M.; Ren, Y.; Hu, M.; Zhang, Y.; Sun, T.; Ma, Y.; Li, X.; Jiang,W.; Gao, D.; Bi, J.; et al. Promoted e_ect of alkalization on the catalytic performance of Rh/alk-Ti3C2X2 (X=O, F) for the hydrodechlorination of chlorophenols in base-free aqueous medium. Appl. Catal. B **2017**, 210, 462–469.
16. Gunbatar S., Aygun A., Karataş Y., Gülcan M., ŞEN F.; Carbon-nanotube-based rhodium nanoparticles as highly-active catalyst for hydrolytic dehydrogenation of dimethylamineborane at room temperature. Journal Of Colloid and Interface Science, 2018, 530, 321-327.
17. Yurderi M., Bulut A., Zahmakıran M., Gülcan M., Özkar S.; Ruthenium(0) nanoparticles stabilized by metal-organic framework (ZIF-8): Highly efficient catalyst for the dehydrogenation of dimethylamine-borane and transfer hydrogenation of unsaturated hydrocarbons using dimethylamine-borane as hydrogen source. Applied Catalysis B-Environmental, 2014, 160, 534-541.
18. Sen F., Karataş Y., Gülcan M., Zahmakıran M.; Amylamine stabilized platinum(0) nanoparticles: active and reusable nanocatalyst in the room temperature dehydrogenation of dimethylamine-borane. RSC ADVANCES , 2014. 4.(4), 1526-1531.

DRAFT

ORAL PRESENTATION

Grafen Oksit Katkılı Metal Oksit Esaslı Katı Destek Yapısında Kararlılaştırılmış Manyetik Rutenyum (0) Nanokümelerinin Hidrazin-Boran Metanolizinden Hidrojen Üretiminde Katalitik Performanslarının İncelenmesi

Sidar Dündar[1], Mehmet Gülcan[1]

[1]Van Yüzüncü Yıl Üniversitesi Fen Fakültesi, Kimya Bölümü, Van, Türkiye

Özet

Bu çalışmada Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO kompoziti destekli Ru (0) nanokümeleri ıslak emdirme-indirgeme yöntemiyle hazırlanarak HB'ın metanolizinden hidrojen üretimindeki katalitik performansları (etkinlik, tekrar kullanlabilirlik ve kinetik çalışmalar açısından) araştırıldı. Hazırlanan Sn_3O_4-Co_3O_4- Fe_3O_4-*r*GO kompoziti destekli rutenyum nanokümeleri, ICP-OES, SEM, SEM/EDX gibi karakterizasyon teknikleriyle tanımlandı.

Anahtar Kelimeler: Hidrazin-boran, Hidrojen, Metanoliz, Nanoküme, Rutenyum

Investigation of Catalytic Performances of Stabilized Magnetic Ruthenium (0) Nanoclusters in Graphene Oxide Doped Metal Oxide Based Solid Support Structure in Hydrogen Production from Hydrazine-Borane Methanolysis

Abstract

In this study, Sn_3O_4-Co_3O_4-Fe3O_4-*r*GO composite supported Ru (0) nanoclusters were prepared by the wet impregnation-reduction method and their catalytic performance (in terms of efficiency, reusability and kinetic studies) in hydrogen production from the methanolysis of HB was investigated. The prepared Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO composite supported ruthenium nanoclusters were identified by characterization techniques such as ICP-OES, SEM, SEM/EDX.

Keywords: Hydrazine-borane, Hydrogen, Methanolysis, Nanocluster, Ruthenium

GİRİŞ

Yenilenebilirliği ve yüksek enerji yoğunluğu sayesinde hidrojen, çevre kirliliğini ve enerji kıtlığı engellerini azaltmak için umut verici bir temiz enerji taşıyıcısı olarak geniş çapta kabul görmektedir. Hidrojenin uygulanabilir bir enerji taşıyıcısı olarak kullanılabilmesi için güvenli ve verimli depolanması çok önemlidir[1,2]. Birçok hidrojen depolama yöntemi geliştirilmiş olmasına rağmen, metal borohidrür [3-5], amonyak-boran [6-8], gibi sıvı fazda kimyasal hidrojen depolama malzemeleri [6], sulu hidrazin [9], formik asit [10], etilendiamin-bisboran [11], metilamin-boran [12,13] ve hidrazin-boran, ($N_2H_4BH_3$, HB) [14,15], bunlar gaz veya sıvı hidrojen depolama sistemlerine göre daha yüksek enerji yoğunlukları, daha yüksek stabiliteleri ve daha düşük potansiyel riskleri nedeniyle hidrojeni depolamanın en etkili yolları olarak kabul edilmektedir. Ağırlıkça %15,4'lük hidrojen depolama kapasitesiyle HB, önemli bir bor-azot bileşiği olarak kabul edilir. HB'de depolanan hidrojen, termoliz, sulu olmayan çözücülerde dehidrojenasyon veya solvoliz (hidroliz veya metanoliz) yoluyla serbest bırakılabilir. Solvoliz, diğer yöntemlerin sert reaksiyon koşullarına sahip olması ve hidrojen üretim hızının yavaş olması nedeniyle tercih edilmektedir. HB'nin metanolizi ayrıca uygun katalizör varlığında 3.0 eşdeğer hidrojen açığa çıkarabilir [14].

$$N_2H_4BH_3 + 4CH_3OH \rightarrow N_2H_5B(OCH_3)_4 + 3H_2$$

Ayrıca katalitik reaksiyonun mekanik özellikleri göz önüne alındığında, HB'nin yapısındaki BH_3 grubunun hidrolizinden hidrojen üretmek mümkün olduğu gibi, seçici olarak gerçekleştirilen hidrojen üretim süreçlerinde N_2H_4 grubundaki hidrojenleri de ortaya çıkarmak mümkündür. Seçici olarak gerçekleştirilebilen bu reaksiyonlarda 1 mol HB'nin tamamen dehidrojenasyonu sonucunda 5:1 mol oranında H_2 ve N_2'nin oluşabileceği rapor edilmiştir [16-19].

DRAFT

Bu çalışmada öncelikle grafen oksit katkılı metal oksit esaslı kompozit katı destek malzemesi (Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO) sentezlendi. Ardından Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO kompoziti destekli Ru (0) nanokümeleri ıslak emdirme-indirgeme yöntemiyle hazırlanarak hazırlanmış olan Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO katalizörü ICP-OES (İndüktif Eşleşmiş Plazma-Optik Emisyon Spektrometresi), SEM (Taramalı Elektron Mikroskopu), SEM/EDX (Taramalı Elektron Mikroskopu/Enerji Dağılımlı X-Işını Spektroskopisi) gibi karakterizasyon teknikleriyle tanımlandı. Daha sonra Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO katalizörünün katalitik performansı HB'ın metanolizinden hidrojen üretimi tepkimesinde etkinlik ve dönüşüm açısından test edimiştir. Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO katalizörünün HB'ın metanolizi tepkimesindeki tekrar kullanılabilirlik performansları da incelendikten sonra farklı sıcaklıklarda katalitik metanoliz tepkimeleri gerçekleştirilerek tepkimenin aktivasyon parametreleri hesaplanmıştır.

MATERYAL

$SnCl_2{\cdot}2H_2O$ (kalay (II) klorür 2.hidrat), sodyum sitrat ($Na_3C_6H_5O_7{\cdot}2H_2O$), NaOH, $FeCl_2{\cdot}4H_2O$, $FeCl_3{\cdot}6H_2O$, $CoCl_2$, $NaBH_4$, Na_2CO_3, $RuCl_3$ $3H_2O$, dihidrazin sülfat kimyasalları ticari olarak temin edilmiştir.

METOT

Bu çalışma kapsamında gerçekleştirilen işlemler aşağıda maddeler halinde sunulmuştur:

Grafen Oksit Katkılı Metal Oksit Esaslı Katı Destek Yapısının Sentezi: Bu aşamada manyetit (Fe_3O_4), kalay (II,III) oksit (Sn_3O_4), kobalt (II,III) oksit (Co_3O_4), indirgenmiş grafen oksit (*r*GO) ve bu yapıların belirli oranlardaki kompozisyonlarından elde edilerek kompozit yapılı (Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO) katı destek malzemeleri sentezlenmiştir. Uygulanacak sentez protokolleri aşağıda detaylı olarak verilmiştir:

Kalay (II, III) oksit (Sn_3O_4) Sentezi: Sn_3O_4 yapısı hidrotermal yöntemle sentezlenmiştir. Bu amaçla kalay (II) klorür ($SnCl_2{\cdot}2H_2O$) ve sodyum sitrat ($Na_3C_6H_5O_7{\cdot}2H_2O$) 10 mL saf suda çözülerek bir karışım elde edildi. Bu karışım üzerine 0.2 M NaOH çözeltisi karıştırılarak eklendi ve karışım 100 mL hacimli bir otoklav içerisine alınarak ve 180 °C'de 12 saat boyunca bekletildi. Ardından otoklav oda sıcaklığına kadar soğutularak ve katı ürün izole edilerek yıkanıp (saf su ve 0.2 M NaOH çözeltisi ile) vakum etüvde 70 °C'de kurutulmuştur [20].

Kobalt (II, III) oksit (Co_3O_4) Sentezi: Co_3O_4 partiküllerinin sentezi için öncü olarak $CoCl_2$ ve NaOH kullanılmıştır. Öncelikle $CoCl_2$ ve NaOH'in ayrı beherlerde 25 mL 0.4 M'lık çözeltileri hazırlandı. NaOH çözeltisi, sürekli karıştırılarak $CoCl_2$ çözeltisine 80℃'de damla damla ilave edildi. Mavi çökeltilerin yavaş yavaş pembe çökeltilere dönüşümünün gözlenmesinin ardından, kuvvetlice karıştırarak yukarıdaki süspansiyonlara 5 mL %30'luk H_2O_2 çözeltisi eklendi. Pembe çökeltiler yavaş yavaş kahverengiye dönüşümü gözlendi. Renk değişimi, H_2O_2 eklenmesiyle Co (II)'nin Co (III)'e oksidasyonu olarak yorumlandı. Nihai çözelti, 100 mL iç hacme sahip paslanmaz çelik otoklava konularak ve 12 saat 150 °C'de bekletildi. Bu süre sonunda hidrotermal reaktör oda sıcaklığına soğutularak ve kahverengi ürün birkaç kez saf su ve etanol ile yıkandı. Yıkanan katı ürün 8000 rpm'de 20 dakika santrifüjlenerek ve vakum altında 60 °C'de 24 saat bekletilerek kurutuldu [21].

***İndirgenmiş Grafen Oksit (rGO) Sentezi*:** Grafen oksit (GO) sentezi, grafitten başlanarak kaynakçada iyi bilinen ve modifiye Hummers yöntemi [22] kullanılarak gerçekleştirildi. *r*GO da yine kaynakçada bildirilen prosedüre uygun biçimde hazırlanmıştır [23]. Bu amaçla, 0.15 g GO 150 mL distile suya konulacak ve yaklaşık 30 dakika ultrasonik banyoda işlemden geçirildi. 1.2 g $NaBH_4$, 30 mL saf su içinde çözündükten sonra GO karışımına damla damla ilave edildi. Ardından karışımın pH'ı %5 Na_2CO_3 ile 9-10'a ayarlanarak ve pH ayarlı karışım 80 °C'de 1 saat karıştırıldı. Koyu renkli karışım, 20 dakika boyunca 8000 rpm'de birkaç kez santrifüjlendikten sonra elde edilen *r*GO, 60 °C'de 24 saat vakum altında bekletilerek kurutuldu.

Grafen Oksit Katkılı Metal Oksit Esaslı (Sn_3O_4-Co_3O_4-Fe_3O_4-rGO) Katı Destek Malzemesinin Sentezi: Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO kompoziti, Sn_3O_4/Co_3O_4/Fe_3O_4:*r*GO'nun 10:1 ağırlık oranına uyacak şekilde hazırlandı. İlk olarak, 0.23 g hazırlanmış *r*GO, 20 mL distile su içinde dağıtılarak ve 30 dakika boyunca ultrasonik banyoda karıştırılarak iyice dağılması sağlandı. Ardından, önceden sentezlenmiş Sn_3O_4 ve Co_3O_4 partikülleri, 40 dakika arayla *r*GO karışımına ayrı ayrı ilave edilerek ve kuvvetlice karıştırıldı. Daha sonra 20 mL distile suya 1:2 molar oranda $FeCl_2{\cdot}4H_2O$ ve $FeCl_3{\cdot}6H_2O$ ilave edilecek ve bu çözelti *r*GO, Sn_3O_4 ve Co_3O_4 partikülleri içeren önceki çözeltiye eklenerek ve nihai karışım, 80 °C'de 30 dakika karıştırıldı. Ardından bu karışıma 20 mL distile su içerisinde 0.4 M NaOH damla damla ilave edildi. Çözeltinin pH'ı 9-10'a ulaştıktan sonra karışım, 80 °C'de 4 saat boyunca karıştırılarak ve bu süre sonunda oda sıcaklığına kadar soğutuldu.

Süzülerek ayrılan karışım birkaç kez etanol ve damıtılmış su ile yıkanarak, ardından 8000 rpm'de 20 dakika santrifüjlenerek ayrılarak ve vakum altında 24 saat 60°C'de tutularak kurutuldu [15].

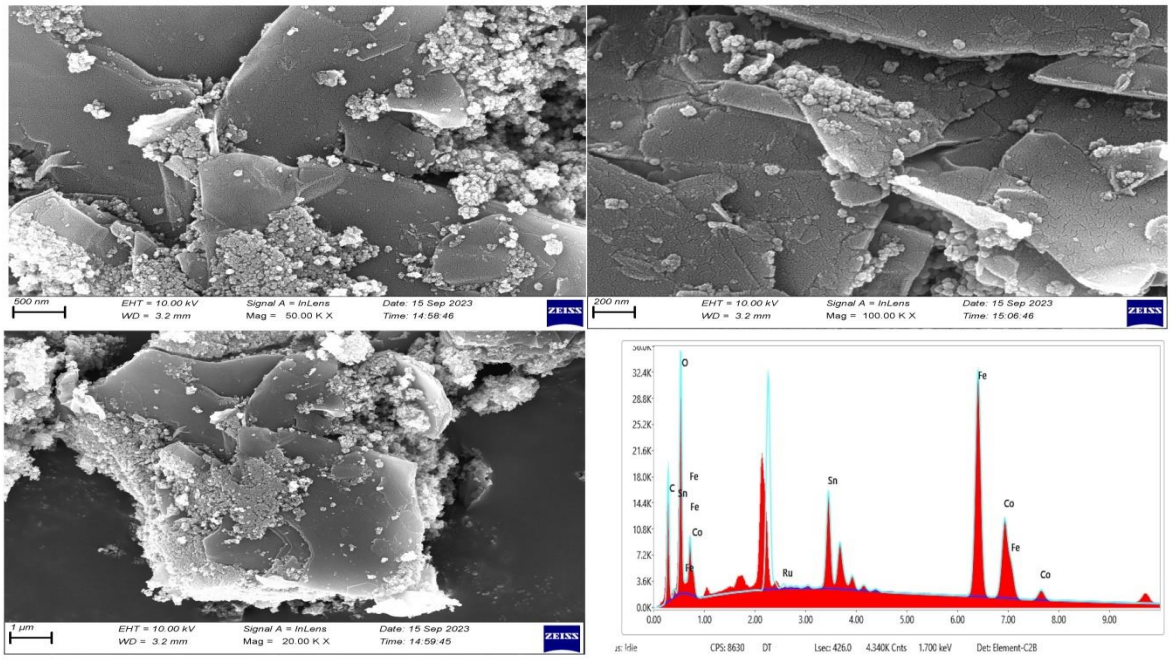
***Ru (0) Nanokümelerinin Hazırlanması ve Tanımlaması*:** Çalışmada grafen oksit katkılı metal oksit esaslı kompozit (Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO) katı destek yapısında kararlı manyetik Ru (0) nanokümeleri, çözelti fazında metal tuzunun destekleyici yüzeyine depolanması ve daha sonra $NaBH_4$ kullanılarak indirgenmesiyle tepkime dışı (*ex-situ*) olarak hazırlandı. Bu amaçla, Ru metalinin başlangıç tuzu Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO katı destek malzemesinin yüzeyine emdirilmesi (3 saat boyunca 750 rpm karıştırma hızında) ve daha sonra indirgeyici ajan varlığında indirgenmesi sağlandı. Hazırlanan katalitik malzeme (Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO) HB'nin metanolizi tepkimesinde katalizör olarak kullanılarak ve gaz çıkış hızları ve dönüşümleri incelenerek katalitik etkinliğin en yüksek olduğu optimum çalışma koşulları belirlendi. Son olarak elde edilen katalitik malzeme; ICP-OES (İndüktif Eşleşmiş Plazma-Optik Emisyon Spektrometresi), SEM (Taramalı Elektron Spektroskopis), SEM-Elementel Haritalama v gibi ileri analitiksel ve spektroskopik yöntemler kullanılarak tanımlanmıştır. Bu analitiksel yöntemlerden ICP-OES; grafen oksit katkılı metal oksit esaslı kompozit yapılı katı destek malzemesinin yüzeyine tutturulan Ru metal miktarının tespiti, SEM; Ru (0) nanokümelerinin morfolojilerinin belirlenmesi, SEM-Elementel Haritalama; seçili bölgelerde Ru (0) nanokümelerine ilişkin elementel bileşimin belirlenmesi amacıyla kullanılmıştır.

HB'nin Metanolizi Tepkimesinde Katalitik Performansın Belirlenmesi ve Tepkime Kinetiğinin İncelenmesi: Çalışmanın bu kısmında Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO katalitik malzemesinin HB'nin metanolizi tepkimesinde katalitik performansı, tepkime sonucu açığa çıkacak gazın hacimsel ölçümü ve katalitik etkinlik göz önüne alınarak incelendi. Ru (0) nanokümelerinin etkinlik, seçicilik, dönüşüm ve dayanıklılık yönünden en iyi katalitik performansın tespit edildiği koşullar için metanoliz tepkimesi farklı çalışma koşullarında (katalizör ve substrat derişimi ve farklı sıcaklıklarda) gerçekleştiridi.

BULGULAR ve TARTIŞMA

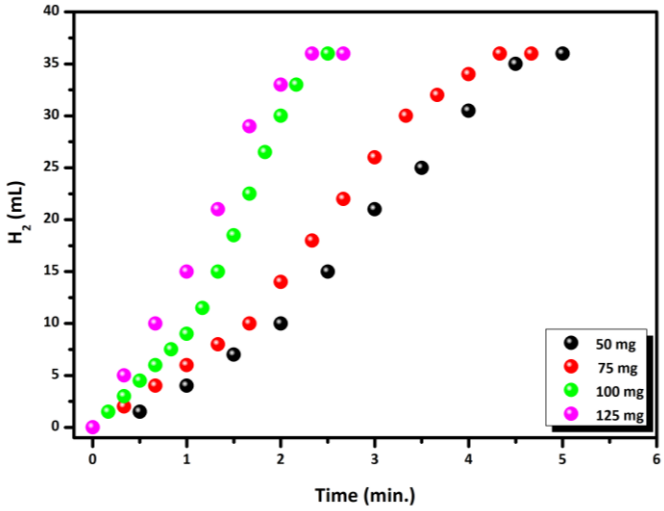
Çalışmanın ilk aşamasında grafen oksit katkılı metal oksit esaslı kompozit katı destek malzemesi (Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO) sentezlendi. . Ardından Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO kompoziti destekli rutenyum nanokümeleri ıslak emdirme-indirgeme yöntemiyle hazırlanarak HB'ın metanolizinden hidrojen üretimindeki katalitik performanslar incelenmiştir.

Morfolojik olarak Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO katalizörünün incelenmesi için SEM analizleri yapılmıştır ve bu görüntüler Şekil 1'de verilmektedir. Farklı çözünürlükte ve büyütme oranlarında alınan SEM görüntülerinden Ru metallerinin destek malzemesinin yüzeyine homojen olarak dağıldığı görülmektedir (Şekil 1 (a, b ve c)). Ayrıca SEM analizi sırasında yapılan haritalandırmada hem Ru metalinin (Şekil 1 (d)) ve destek malzemesinde bulunan C, Sn, Co, Fe ve O (Şekil 1 (d)) elementlerinin varlığı açıkça anlaşılmaktadır.



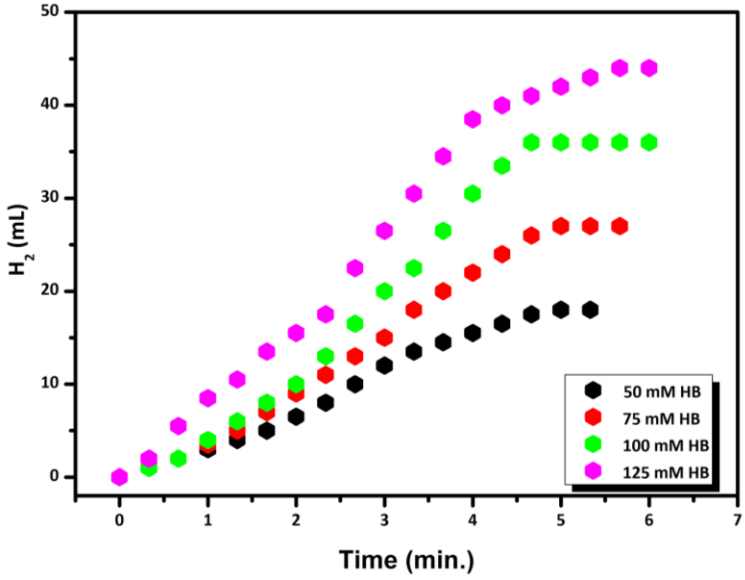
Şekil 1. Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO katalizörüne ilişkin (a) 500 nm ölçekli SEM görüntüsü (b) 200 nm ölçekli SEM görüntüsü (c) 1 µm ölçekli SEM görüntüsü, (d) Taramalı Elektron Mikroskopu/Enerji Dağılımlı X-Işını Spektroskopisi görüntüsü

Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO nanokatalizörü ile katalizlenen HB'nin metanoliz tepkimesinde, tepkime hızına katalizör miktarlarının etkisini incelemek için, HB derişimi (23 mg, 100 mM) sabit tutularak farklı miktarlarda Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO nanokatalizör kullanımıyla HB'nin metanoliz tepkimesi 298 K sıcaklıkta gerçekleştirilmiştir. Şekil 2'da farklı derişimlerde Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO nanokatalizörü (Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO = 50 mg, 75 mg, 100 mg ve 125 mg) varlığında katalizlenen HB'nin hidroliz tepkimesinde açığa çıkan H_2 gazın HB gaz hacmine karşı zaman grafiği verilmektedir.



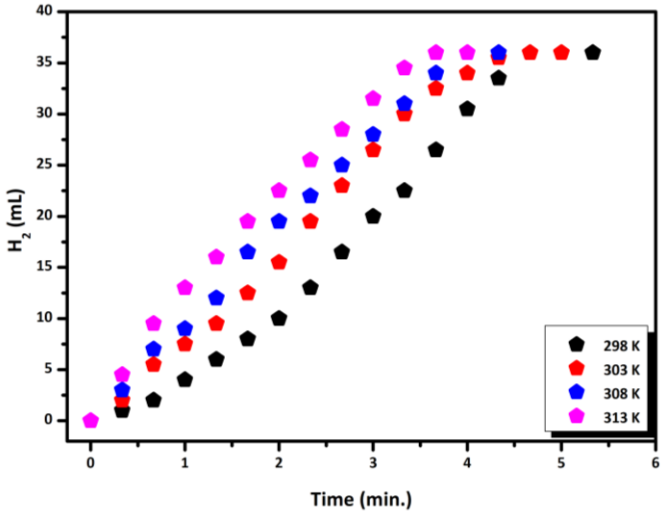
Şekil 2. Farklı miktarlarda Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO katalizörü ile katalizlenen HB'nin metanoliz tepkimesinde açığa çıkan gazın (H_2) HB gaz hacmine karşı zaman grafiği.

Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO nanokatalizörü ile katalizlenen HB'nin metanoliz tepkimesinde tepkime hızına substrat derişiminin etkisini incelemek amacıyla, nanokatalizör derişimi (50 mg) sabit tutularak farklı derişimlerde HB kullanılarak katalitik metanoliz tepkimesi 298 K sıcaklıkta gerçekleştirilmiştir. Şekil 3'de farklı derişimlerde ([EDB] = 50.0 mM, 75 mM, 100 mM, ve 125 mM) substrat kullanılarak katalizlenen HB'nin metanoliz tepkimesinde açığa çıkan H2 gazın HB gaz hacmine karşı zaman grafiği verilmektedir.



Şekil 3. Farklı derişimlerde HB kullanılarak Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO nanokatalizörü ile katalizlenen HB'nin metanoliz tepkimesinde açığa çıkan gazın (H_2) gaz hacmine karşı zaman grafiği.

Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO nanokatalizörü varlığında HB'nin metanoliz tepkimesine sıcaklığın etkisini belirlemek amacıyla katalizör ve substrat miktarları sabit tutularak sıcaklık değişimi etkisi incelenmiştir. Şekil 4'te farklı sıcaklıklarda gerçekleştirilen metanoliz tepkimesinde açığa çıkan H_2 gazın HB gaz hacmine karşı zaman grafiği verilmektedir. Beklenildiği gibi katalitik metanoliz tepkime hızının sıcaklıkla doğru orantılı olarak arttığı görülmüştür.



Şekil 4. Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO nanokatalizörü ile katalizlenen HB'nin farklı sıcaklıklarda gerçekleştirilen metanoliz tepkimesinde açığa çıkan gazın (H_2) HB gaz hacmine karşı zaman grafiği.

SONUÇ

Bu çalışma kapsamında elde edilen sonuçlar özetle şu şekilde sıralanabilir:

Çalışmanın ilk aşamasında grafen oksit katkılı metal oksit esaslı kompozit katı destek malzemesi (Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO) başarıyla sentezlendi.
Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO kompoziti destekli rutenyum nanokümeleri ıslak emdirme-indirgeme yöntemiyle sentezlendi.
Sentezlenen Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO nanokatalizörü SEM/SEM-EDX analizleri ile morfolojik olarak tanımlanması yapılmıştır.
Sentezlenen Ru@Sn_3O_4-Co_3O_4-Fe_3O_4-*r*GO nanokatalizörünün hidrazin-boranın (HB) metanoliz tepkimesi yoluyla hidrojen üretimindeki katalitik etkinliği (katalizör miktarına, substrat derişimine ve sıcaklığa bağlı katalitik etkinliği) incelenmiştir.

TEŞEKKÜR

** 2209-A- Lisans Öğrencilerine Yönelik Araştırma Projesi Destek Programı kapsamındaki projemize (Proje No: 1919B012220946) maddi desteği için Türkiye Bilimsel ve Teknolojik Araştırma Kurumu'na teşekkür ederiz.*

KAYNAKLAR

[1] T. He, P., Pachfule, H. Wu, Q. Xu, P. Chen Hydrogen carriers Nat. Rev. Mater., 1 (12) (2016), Article 16059, 10.1038/natrevmats.2016.59
[2] Z.-X. Yang, X.-G. Li, Q.-L. Yao, Z.-H. Lu, N. Zhang, J. Xia, K. Yang, Y.-Q. Wang, K. Zhang, H.-Z. Liu, L.-T. Zhang, H.-J. Lin, Q.-J. Zhou, F. Wang, Z.-M. Yu, J.-M. Ma Roadmap on hydrogen energy from production to utilizations Rare Met., 41 (10) (2022), pp. 3251-3267, 10.1007/s12598-022-02029-7 2022
[3] L. Sun, X.F., Gao, X. Ning, Z.M. Qiu, L.W. Xing, H. Yang, D.C. Li, J.M. Dou, Y.Y. Meng Cobalt-nickel bimetal carbon sphere catalysts for efficient hydrolysis of sodium borohydride: the role of synergy and confine effect Int. J. Hydrogen Energy, 48 (2023), pp. 3413-3428, 10.1016/j.ijhydene.2022.10.214
[4] Ö. Şahin, A. Bozkurt, M. Yayla, H.Ç. Kazıcı, M.S. İzgi As a highly efficient reduced graphene oxide-supported ternary catalysts for the fast hydrogen release from NaBH4 Graphene Technol., 5 (2020), pp. 103-111, 10.1007/s41127-020-00036-y

[5] Ö. Şahin, M.S. İzgi, S. Tayboğa, H.Ç. Kazıcı Effect of plasma pretreatment of Co–Cu–B catalyst on hydrogen generation from sodium borohydride methanolysis React. Kinet. Mech. Catal., 133 (2021), pp. 851-861, 10.1007/s11144-021-02004-w

[6] H. Durak, M. Gülcan, M. Zahmakiran, S. Ozkar, M. Kaya Hydroxyapatite-nanosphere supported ruthenium(0) nanoparticle catalyst for hydrogen generation from ammonia-borane solution: kinetic studies for nanoparticle formation and hydrogen evolution RSC Adv., 4 (2014), pp. 28947 28955, 10.1039/C4RA03213F

[7] H.Ç. Kazıcı, M.S. İzgi, Ö. Şahin A comprehensive study on the synthesis, characterization and mathematical modeling of nanostructured Co-based catalysts using different support materials for AB hydrolysis Chem. Pap., 75 (2021), pp. 2713-2725, 10.1007/s11696-021-01514-0

[8] H. Ç. Kazıcı, Mehmet Sait İzgi, Ömer Şahin Co-Mn-B nanoparticles supported on epoxy-based polymer as catalyst for evolution of H_2 from ammonia borane semi-methanolysis J. Electron. Mater., 51 (2022), pp. 2356-2368, 10.1007/s11664-022-09491-0

[9] Y. Karataş, A. Zengin, M. Gülcan Preparation and characterization of amine-terminated delafossite type oxide, $CuMnO_2$-NH_2, supported Pd (0) nanoparticles for the H_2 generation from the methanolysis of ammonia-borane Int. J. Hydrogen Energy, 47 (2022), pp. 16036-16046, 10.1016/j.ijhydene.2022.03.098

[10] Y. Karataş, M. Gülcan, M. Zahmakiran Silica supported ternary NiRuPt alloy nanoparticles: highly efficient heterogeneous catalyst for H_2 generation via selective decomposition of hydrous hydrazine in alkaline solution Int. J. Hydrogen Energy, 45 (2020), pp. 27098-27113, 10.1016/j.ijhydene.2020.07.048

[11] Y. Karatas, A. Bulut, M. Yurderi, I.E. Ertas, O. Alal, M. Gülcan, M. Celebi, H. Kivrak, M. Kaya, M. Zahmakiran PdAu-MnOx nanoparticles supported on amine-functionalized SiO_2 for the room temperature dehydrogenation of formic acid in the absence of additives Appl. Catal. B Environ., 180 (2016), pp. 586-595, 10.1016/j.apcatb.2015.06.060

[12] M. Celebi, A. Ruzgar, Y. Karatas, M. Gülcan Manganese oxide octahedral molecular sieves stabilized Rh nanoparticles for the hydrogen production from the ethylenediamine-bisborane hydrolysis Int. J. Hydrogen Energy, 47 (2022), pp. 16494-16506, 10.1016/j.ijhydene.2022.03.127

[13] Y. Karatas, E. Kuyuldar, H. Acidereli, M. Gülcan, F. Sen Polypyrrole-multi walled carbon nanotube hybrid material supported Pt NPs for hydrogen evolution from the hydrolysis of MeAB at mild conditions Sci. Rep., 9 (2019), Article 18553, 10.1038/s41598-019-55030-z

[14] R. Yıldırım, M. Gülcan H_2 production from the hydrolytic dehydrogenation of methylamine-borane catalyzed by sulfonated reduced graphene oxide-aided synthesis of ruthenium nanoparticles Int. J. Hydrogen Energy, 46 (2021), pp. 32523-32535, 10.1016/j.ijhydene.2021.07.096

[15] D. Özhava, N.Z. Kılıçaslan, S. Özkar PVP-stabilized nickel(0) nanoparticles as catalyst in hydrogen generation from the methanolysis of hydrazine borane or ammonia borane Appl. Catal. B Environ., 162 (2015), pp. 573-582, 10.1016/j.apcatb.2014.07.033

[16] B. Demirkan, E. Kuyuldar, Y. Karatas, M. Gulcan, F. Sen Ex situ synthesis and characterization of a polymer-carbon nanotube-based hybrid nanocatalyst with one of the highest catalytic activities and stabilities for the hydrolytic dehydrogenation of hydrazine-borane at room temperature conditions J. Colloid Interface Sci., 552 (2019), pp. 432-438, 10.1016/j.jcis.2019.05.075

[17] Q. Yao, Z.-H. Lu, R. Zhang, S. Zhang, X. Chen, H.-L. Jiang A noble-metal-free nanocatalyst for highly efficient and complete hydrogen evolution from $N_2H_4BH_3$ J. Mater. Chem., 6 (2018), pp. 4386-4393, 10.1039/C7TA10886A

[18] W. Wang, X. Hong, Q. Yao, Z.-H. Lu Bimetallic Ni–Pt nanoparticles immobilized on mesoporous N-doped carbon as a highly efficient catalyst for complete hydrogen evolution from hydrazine borane J. Mater. Chem., 8 (2020), pp. 13694-13701, 10.1039/D0TA05322H

[19] J. Long, Q. Yao, X. Zhang, H. Wu, Z.-H. Lu Defects engineering of metal-organic framework immobilized Ni-La$(OH)_3$ nanoparticles for enhanced hydrogen production Appl. Catal. B Environ., 320 (2023), Article 121989, 10.1016/j.apcatb.2022.121989.

[20] T. Tanabe, M. Hashimato, K. Mibu, T. Tanikawa, T. Gunji, S. Kaneko, H.Abe, M.Miyauchi, F. Matsumoto, Synthesis of Single Phase Sn_3O_4: Native VisibleLight-Sensitive Photocatalyst with High Photocatalytic Performance for Hydrogen Evolution. *J* Nanosci Nanotech. 17 (2017) 3454-3459.

[21] Z. Huang, Y. Zhao, H. Xu, Zhao., Surfactant-free synthesis, photocatalytic and electrochemical property study of Co_3O_4 nanoparticles. J., *Mater Res Bull* 100 (2018) 83-90.

[22] M. Kaur, H. Kaur, D. Kukkar, Synthesis and characterization of graphene oxide using modified Hummer's method. *AIP Conference Proceedings* 1953 (2018) 030180.

[23] E. G. Söğüt, Y. Karataş, Gülcan M., . Kılıç N. Ç, Enhancement of adsorption capacity of reduced graphene oxide by sulfonic acid functionalization: Malachite green and Zn (II) uptake. Mater Chem Phys. 256 (2020) 123662.

DRAFT

ORAL PRESENTATION

Polivinilpirolidon Kararlı Tek veya İki Metalli Nanokümeler: Hazırlanması, Karakterizasyonu ve Etilendiamin-Bisboran'dan Hidrojen Üretiminde Katalizör Etkilerinin Araştırılması*

Bilal Özçiçek[1], Yaşar Karataş[2], Adem Rüzgar[2], Mehmet Gülcan[1]

[*1]Van Yüzüncü Yıl Üniversitesi Fen Fakültesi, Kimya Bölümü, Van, Türkiye
[2] Van Yüzüncü Yıl Üniversitesi Muradiye Meslek Yüksekokulu, Kimya ve Kimyasal İşleme Teknolojileri Bölümü, Van, Türkiye

Özet

Molekül ağırlığının yaklaşık onda biri hidrojenden oluşan etilendiamin-bisboran ($BH_3NH_2CH_2CH_2NH_2BH_3$; EDB), hidrojen depolama malzemesi olarak son yıllarda ilgi odağı haline gelen ve birçok çalışmaya konu olan bir bileşik olarak ön plana çıkmaktadır. EDB, hidrojen üretimi üzerine yapılan araştırma/uygulama çalışmalarında yoğun bir biçimde kullanılan amonyak borana (AB) göre daha stabil bir yapıya sahiptir. Etilen diamin bisboranın bir diğer avantajı ise amonyak borandan hidrojen üretimi sürecinde açığa çıkan uçucu ve istenmeyen boran türevi bileşiklere etilendiamin-bisborandan hidrojen üretimi sırasında rastlanılmamasıdır. EDB, uygun bir katalizör varlığında termoliz ve hidroliz yoluyla yapısındaki hidrojeni serbest bırakır. Termoliz işleminin yakıt hücresi uygulamaları için dezavantajlara sahip olması hidroliz reaksiyonunu ön plana çıkarmaktadır. EDB'nin hidroliz reaksiyonu sonucunda her mol EDB için 6 mol H_2 açığa çıkmaktadır.

$$BH_3NH_2CH_2CH_2NH_2BH_3\ (aq) + 4H_2O \rightarrow BO_2NH_3CH_2CH_2NH_3BO_2\ (aq) + 6H_2\ (g)$$

Bu çalışmada polimerik bir malzeme olan polivinilpirolidon (PVP) kararlı tek (Rh ve Ru) veya iki metalli (Ru-Rh) nanokümeler alkol-indirgeme yöntemiyle hazırlanarak etilendiamin-bisboranın hidrolizinden hidrojen üretimindeki katalitik performansları incelendi. Hazırlanan PVP kararlı tek veya iki metalli nanokümelerin EDB'den hidroliz tepkimesi yoluyla hidrojen üretiminde tekrar kullanılabilirlik performansları da incelenerek farklı sıcaklıklarda hidroliz tepkimeleri gerçekleştirildi ve aktivasyon parametreleri hesaplandı.

Anahtar Kelimeler: Etilendiamin-bisboran, Hidrojen, Hidroliz, Nanoküme, PVP

Absract

Ethylenediamine-bisborane ($BH_3NH_2CH_2CH_2NH_2BH_3$; EDB), which consists of approximately one tenth of its molecular weight of hydrogen, stands out as a compound that has become the focus of attention in recent years as a hydrogen storage material and has been the subject of many studies. EDB has a more stable structure than ammonia borane (AB), which is used extensively in research/application studies on hydrogen production. Another advantage of ethylene diamine bisborane is that volatile and undesirable borane-derived compounds, which are released during the hydrogen production process from ammonia borane, are not encountered during the hydrogen production from ethylene diamine bisborane. EDB releases the hydrogen in its structure through thermolysis and hydrolysis in the presence of a suitable catalyst. The fact that the thermolysis process has disadvantages for fuel cell applications brings the hydrolysis reaction to the fore. As a result of the hydrolysis reaction of EDB, 6 moles of H2 are released for each mole of EDB.

$$BH_3NH_2CH_2CH_2NH_2BH_3\ (aq) + 4H_2O \rightarrow BO_2NH_3CH_2CH_2NH_3BO_2\ (aq) + 6H_2\ (g)$$

In this study, single (Rh and Ru) or bimetal (Ru-Rh) nanoclusters stabilized by polyvinylpyrrolidone (PVP), a polymeric material, were prepared by alcohol-reduction method and their catalytic performance in hydrogen production from the hydrolysis of ethylenediamine-bisborane was examined. The reusability performances of the prepared PVP-stabilized mono- or bi-metallic nanoclusters in hydrogen production through hydrolysis reaction from EDB were also examined, hydrolysis reactions were carried out at different temperatures and activation parameters were calculated.

Key Words: Ethylenediamine-bisborane, Hydrogen, Hydrolysis, Nanocluster, PVP

DRAFT

DRAFT

GİRİŞ

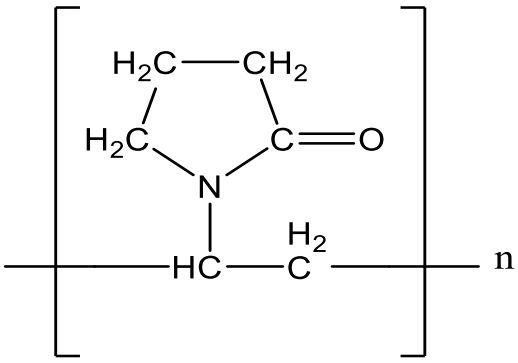
Endüstri devriminden itibaren dünyamızda gözlemlenen ekonomik, sosyal ve teknolojik gelişmelerin en kritik parametresi olarak kabul edilen enerji her geçen gün artan önemiyle küresel gündem maddelerinden biri haline gelmiştir. Yaşam tarzında meydana gelen değişiklikler ve hızla artan nüfus, enerjiye olan talebi artırmış ve bu talep uzun bir süre yoğunluklu olarak fosil yakıtlardan karşılanmıştır. Fosil yakıtlardan enerji üretim süreci sonucu açığa çıkan ve atmosfere yayılan zararlı ve zehirli gazların miktarının kabul edilebilir oranların çok üzerine çıkması başa çıkılması oldukça zor birçok soruna neden olmuştur. Bu sorunlar; küresel ısınma, bitki örtüsünün bozulması, sağlık sorunları, asit yağmurları, iklim değişiklikleri, temiz su kaynaklarının ve toprağın kirletilmesi şeklinde sıralanabilir. Bu denli büyük ve küresel sorunlar alternatif enerji kaynaklarına yönelimi ve bu alanda yapılan araştırma çalışmalarının sayısını artırmıştır. Güneş enerjisi, rüzgar enerjisi, jeotermal enerji, hidroelektrik enerjisi, biyoenerji, dalga enerjisi, okyanus termal enerjisi ve hidrojen enerjisi bilim insanlarının çalışmalarına konu olan önemli alternatif enerji kaynakları olarak sıralanabilir. Bu enerji kaynakları sürdürülebilirlik, kaynak çeşitliliği, verimlilik ve çevre temizliği gibi kriterlere göre değerlendirildiğinde diğer alternatif enerji kaynaklarına göre öne çıkan enerji kaynağı
hidrojen enerjisi olmaktadır [1-3]. Enerji sistemleri içerisinde enerji aktarıcısı, taşıyıcısı veya değiştiricisi olarak değerlendirilebilen hidrojen, evrenin en basit ve en çok bulunan elementi olup, renksiz, kokusuz, havadan 14,4 kez daha hafif ve tamamen zehirsiz bir gazdır. Yanma tepkimesi sonucu hiçbir zararlı veya zehirli atığın oluşmadığı hidrojen, bütün yakıtlar içerisinde birim kütlede en yüksek enerji içeriğine sahip gaz olarak dikkat çekmektedir. Yukarıda sayılan avantajlarına rağmen, hidrojenin güvenli, verimli ve ekonomik depolama ve nakliye koşullarının oluşturulmasında yaşanan zorluklar, hidrojen enerjisinin yaygınlaşmasının önündeki en büyük engel olarak görülmektedir. Hidrojenin çok küçük bir miktarının bile gaz halinde çok fazla hacim kaplaması hidrojenin gaz halinde depolanmasını zorlaştırmaktadır. Hidrojenin sıvı halde depolanması için ise yüksek basınçlı ve dayanıklı kaplara ihtiyaç duyulmaktadır. Bu tür yüksek basınçlı kapların üretiminde yaşanan zorluklar ve bu kapların oldukça pahalı malzemeler olması hidrojenin sıvı halde depolanmasını da zorlaştırmaktadır. Sıvı ve gaz fazında karşılaşılan güvenlik ve maliyet eksenli bu zorluklar katı fazda hidrojen barındıran katı maddelere yönelik çalışmaların artmasına neden olmuştur. [4-6]. Katı fazda yapısında hidrojen barındıran malzemelerin başında amin boran türevleri gelmektedir. Diğer hidrojen kaynaklarına oranla daha fazla çalışmaya konu olmuş bu bileşiklerin en önemli avantajları kararlı yapıları, yüksek hidrojen içerikleri ve düşük toksisiteleri şeklinde sıralanabilir. Hidroliz, termoliz veya hidrotermoliz yöntemleri ile hidrojen elde edilebilen bu bileşiklerden amonyak boran ağırlıkça %19,6, hidrazin boran ağırlıkça %15,4 ve dimetilamin boran ağırlıkça %16,9 ve etilendiamin-bisborandağırlıkça %11,4 hidrojen kapasitesine sahiptir. Bu bileşiklerden etilendiamin-bisboran hidrojen üretimi üzerine yapılan çalışmaların uzunca bir süre yoğunlaştığı bileşik olan amonyak borana oranla daha kararlı bir yapıya sahiptir. Etilendiamin-bisboranın bir diğer avantajı ise amonyak borandan hidrojen üretimi sürecinde açığa çıkan zararlı ve istenmeyen bileşiklere etilen etilendiamin-bisborandan hidrojen üretimi sırasında rastlanılmamasıdır [7-9].



Etilendiamin-bisboran ılımlı koşullar altında ve sulu ortamda yapısındaki hidrojeni salabilmektedir. Ancak bu tepkimenin hedeflenen hızda ve yüksek verimle gerçekleştirilebilmesi için ihtiyaç duyulan en temel faktör uygun destekleyici ve metallerin bir araya getirilmesi sonucu katalitik aktivitesi yüksek nanokatalizörlerin hazırlanmasıdır [10,11].

Yapılan çalışmada katalizör ömrünü kısaltan ve katalizör performansını düşüren en temel sorun olan metal topaklaşmasına karşı oldukça yüksek stabiliteye sahip organik bir polimer olan Poli(N-vinil-2-pirolidon) (PVP) kullanıldı.

DRAFT



Şekil 1. Poli (N-vinil-2-pirolidon) bileşiğinin molekül yapısı

Birçok polar çözücüde çözünebilen ve oldukça non-iyonik bir karaktere sahip olan PVP NC=O fonksiyonel yapılarının çoklu koordinasyonu yoluyla elde edilen oldukça geniş bir yüzey alanına sahiptir [12-17]. Geniş yüzey alanı sayesinde metal nanopartiküllerin mükemmel bir şekilde dağıtılabilmesine olanak sağlayan PVP'nin destek malzemesi olarak kullanılması sonucu sentezlenen nanokatalizörlerin etilendiamin-bisborandan hidroliz yoluyla hidrojen üretiminde oldukça verimli bir katalizör olduğu belirlendi.

MATERYAL

Rutenyum (III) klorür hidrat ($RuCl_3$•xH_2O), Rodyum(III) klorür hidrat ($RhCl_3$•xH_2O), poli(N-vinil-2-pirolidon) (PVP) ve etanol Sigma-Aldrich'ten ticari olarak temin edildi. Saf su, su arıtma sistemi (Milli Q-pure WS) ile damıtıldı. Çalışmada kullanılan cam malzemeler ve manyetik karıştırma çubukları yıkandıktan sonra saf su ve aseton ile durulandı, ardından 160 °C'de etüvde kurutuldu.

YÖNTEM

EDB Sentezi ve Karakterizasyonu

Yapılan çalışmanın ilk aşamasında EDB sentezi gerçekleştirildi. Bu amaçla, 500 mL iki boyunlu balon içerisine 2.9 g (76.66 mmol) $NaBH_4$ konulduktan sonra üzerine 300 ml THF ilave edildi. Oda sıcaklığında 30 dk karıştırılan karışım üzerine 4.87 g (36.61 mmol) etilendiamin hidroklorür eklendikten sonra tepkime kabının ağzı kapatılarak 24 saat boyunca oda sıcaklığında ve Ar atmosferinde karıştırıldı. Katı kısım izole edildikten sonra geriye kalan sıvı kısımdan bütün THF uzaklaştırıldı. Elde edilen katı ürün üzerine 100 mL önceden kurutulmuş dietileter eklenip 0 ºC'de iki saat boyunca karıştırıldı. İki saatin sonunda katı kısım tekrar süzüldü ve eter fazındaki ürün (EDB) oda sıcaklığında bir gece bekletildi. Dietileterin tamamen uçması sonucu elde edilen beyaz renkli kristaller kullanılmak üzere amber renkli cam bir kapta saklandı.

PVP Kararlı Tek Metalli (Rh ve Ru) Nanokümelerin Hazırlanması

PVP kararlı tek metalli (Rh ve Ru) nanokümeleri (Rh@PVP ve Ru@PVP) alkol indirgeme yöntemine göre sentezlendi. Bu sentez protokolüne göre, etanol-su karışımı içinde 0.25 mmol $RhCl_3$·xH_2O ve $RuCl_3$·xH_2O tuzlarıyla ayrı ayrı hazırlanan çözeltilere 2.5 mmol (200 mg) PVP ilave edildi ve iki saat kaynatıldı. İki saatin sonunda renk değişiminin gözlemlendiği çözeltide PVP hem kararlılaştırıcı ve hem de indirgen rolü oynamaktadır.

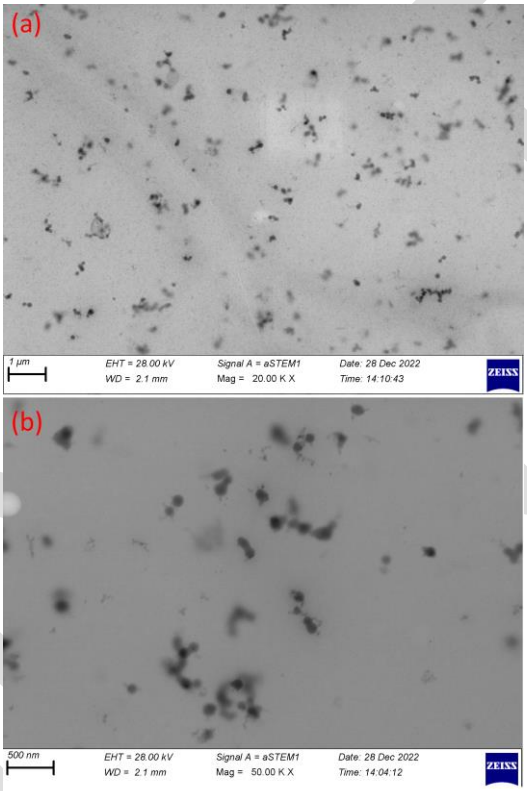
PVP Kararlı İki Metalli (Rh-Ru) Nanokümelerin Hazırlanması

PVP kararlı iki metalli (Rh-Ru) nanokümeleri (Rh-Ru@PVP) alkol indirgeme yöntemine göre sentezlendi. Bu amaçla etanol-su karışımı içinde 0.125 mmol $RhCl_3$·xH_2O ve 0.125 mmol $RuCl_3$·xH_2O tuzlarının birlikte bulunduğu çözeltiye 2.5 mmol (200 mg) PVP ilave edildi ve iki saat kaynatıldı. Çözeltide hedeflenen renk değişimi gerçekleştikten sonra çözelti kullanılmak üzere soğumaya bırakıldı.

BULGULAR VE TARTIŞMA

Poli(N-vinil-2-pirolidon) ile kararlılaştırılmış tek metalli (Rh ve Ru) nanokümeleri (Rh@PVP ve Ru@PVP) ve iki metalli (Rh-Ru) nanokümeleri (Rh-Ru@PVP) alkolle indirgenme yöntemine göre, uygun rodyum ve rutenyum tuzlarının PVP varlığında eşzamanlı olarak indirgenmesiyle sentezlendi. Bu yöntemde, PVP hem kararlılaştırıcı hem de indirgen olarak kullanıldı. Çözeltinin 2 saat boyunca kaynatılması sonucu renginin siyaha dönmesi, çözeltideki Rh^{3+} ve Ru^{3+} iyonlarının indirgenerek Rh^0 ve Ru^0 'a dönüştüğünü ve hedeflenen nanokümelerinin elde edildiğini göstermektedir.

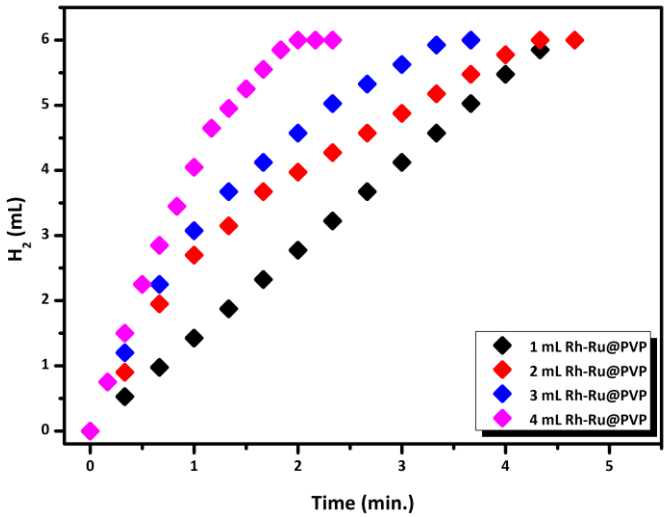
Poli(N-vinil-2-pirolidon) ile kararlılaştırılmış rodyum-rutenyum nanokümelerindeki (Rh-Ru@PVP) parçacık boyutunun belirlenebilmesi amacıyla taramalı elektron mikroskobu (SEM) kullanıldı. Rh-Ru@PVP örneklerine ait 500 nm ve 1 μm büyüklükteki SEM görüntüleri incelendiğinde Rh ve Ru parçacıklarının homojen olarak dağıldığı ve herhangi bir topaklaşmanın söz konusu olmadığı net bir şekilde anlaşılmaktadır (Şekil 2).



Şekil 2. Rh-Ru@PVP katalizörüne ait SEM görüntüleri

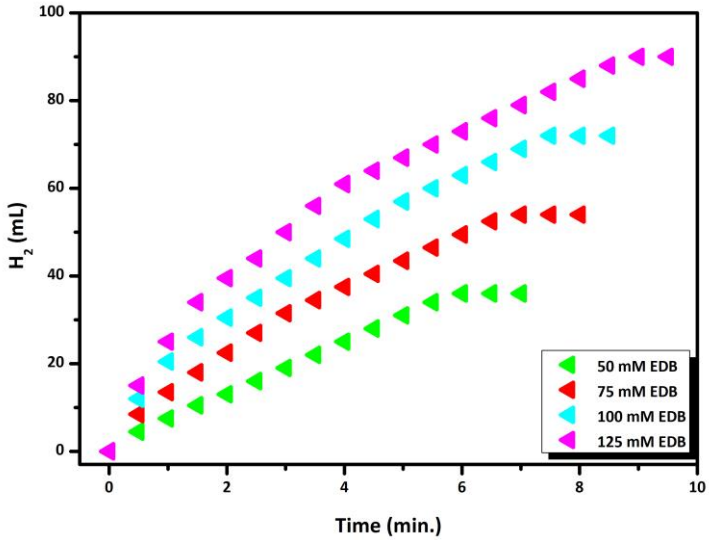
Yapılan kontrol deneyleri sonucunda Poli(N-vinil-2-pirolidon) ile kararlılaştırılmış rodyum-rutenyum nanokümelerinin (Rh-Ru@PVP) etilendiamin-bisboran'dan hidroliz tepkimesiyle hidrojen eldesinde oldukça aktif katalizörler oldukları belirlendi.

Şekil 3, etilendiamin-bisboran'dan Rh-Ru@PVP nanokümeleri katalizörlüğündeki hidroliz tepkimesine ilişkin (25 °C'de) farklı katalizör hacimlerinde (1 ml, 2ml, 3 ml, 4ml)) tepkime süresine karşı açığa çıkan H_2 grafiklerini göstermektedir. Tepkimenin başından itibaren çok hızlı bir hidrojen gazı çıkışının gözlemlendiği hidroliz tepkimesinde hidrojen oluşum hızları ile katalizör miktarı beklenildiği gibi doğru orantılı bir şekilde değişmektedir.



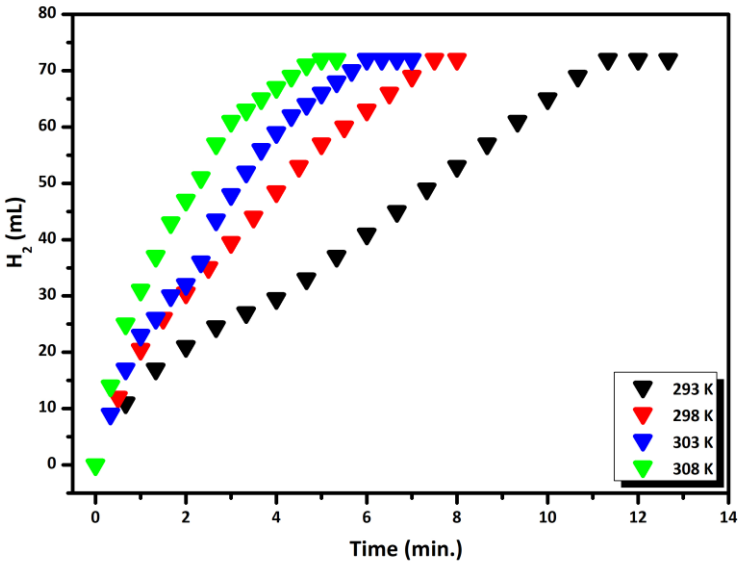
Şekil 3. Etilen di amin bisboranın farklı Rh-Ru@PVP nanokümeleri miktarlarındaki hidroliz tepkimesine ait tepkime süresine karşı açığa çıkan H_2 grafiği.

Rh-Ru@PVP nanokatalizörü ile katalizlenen EDB'nin hidroliz tepkimesinde tepkime hızına substrat derişiminin etkisini incelemek amacıyla, nanokatalizör derişimi sabit tutulup farklı derişimlerde EDB kullanılarak katalitik hidroliz tepkimesi gerçekleştirildi. Şekil 4'te farklı derişimlerde ([EDB] = 50 mM, 75 mM, 100 mM ve 125 mM) substrat kullanılarak katalizlenen EDB'nin hidroliz tepkimesinde açığa çıkan H_2 gazının zamana karşı grafiği verilmektedir. Grafikler incelendiğinde substrat derişimindeki artışın tepkime hızını artırdığı görülmektedir.



Şekil 4. Farklı derişimlerde EDB kullanılarak Rh-Ru@PVP nanokatalizörü ile katalizlenen EDB'nin hidroliz tepkimesinde açığa çıkan gazın (H_2) zamana karşı grafiği

Rh-Ru@PVP nanokatalizörü varlığında EDB'nin hidroliz tepkimesine sıcaklığın etkisini belirlemek ve tepkimeye ait termodinamik parametreleri hesaplamak amacıyla farklı sıcaklıklarda katalitik hidroliz tepkimeleri gerçekleştirildi. Şekil 5'te farklı sıcaklıklarda gerçekleştirilen hidroliz tepkimesinde açığa çıkan gazın zamana karşı grafiği verilmiştir. Katalitik hidroliz tepkime hızının sıcaklıkla doğru orantılı olarak arttığı görülmektedir.



Şekil 5. Rh-Ru@PVP nanokatalizörü ile katalizlenen EDB'nin farklı sıcaklıklarda gerçekleştirilen hidroliz tepkimesinde açığa çıkan gazın (H_2) zaman karşı grafiği.

SONUÇ

Yapılan çalışmada, poli(N-vinil-2-pirolidon) ile kararlılaştırılmış bimetalik rodyum-rutenyum nanokümeleri (Rh-Ru@PVP) sentezlendi, tanımlandı ve etilendiamin-bisboran'ın hidrolizinde katalizör olarak kullanıldı. Gerçekleştirilen deneysel çalışmaların sonuçları genel olarak şu şekilde özetlenebilir:

Poli(N-vinil-2-pirolidon) ile kararlılaştırılmış rodyum-rutenyum nanokümeleri (Rh-Ru@PVP), uygun rodyum ve rutenyum tuzlarının etanol-sulu karışımında poli(N-vinil-2-pirolidon) varlığında kaynatılması ve eşzamanlı indirgenmesi sonucu sentezlendi. Bu yöntemde, poli(N-vinil-2-pirolidon) hem kararlılaştırıcı hem de indirgen rolü oynamaktadır. Sentezlenen Rh-Ru@PVP nanokümelerinin tanımlanmasında ileri spektroskobik ve analitiksel yöntemler kullanıldı.

Verimli ve çevreci bir şekilde sentezlenen ve tanımlanan Rh-Ru@PVP nanokümeleri önemli amin boran bileşiklerinden biri olan etilendiamin-bisboran'ın hidroliz tepkimesinde katalizör olarak kullanıldı. Gerçekleştirilen kinetik çalışmalar sonucunda Rh-Ru@PVP nanokümelerinin etilendiamin-bisboran'ın hidrolizinde yüksek katalitik etkinliğe sahip oldukça etkin katalizörler oldukları tespit edildi.

Etilendiamin-bisboran'ın hidroliz tepkimesindeki dayanıklılık deneyleri Rh-Ru@PVP nanokümelerinin bu hidroliz tepkimesinde birbirini takip eden beşinci katalitik çevrim sonunda bile hala etkin olduklarını göstermektedir.

Teşekkür

** 2209-A- Lisans Öğrencilerine Yönelik Araştırma Projesi Destek Programı kapsamındaki projemize (Proje No: 1919B012206294) maddi desteği için Türkiye Bilimsel ve Teknolojik Araştırma Kurumu'na teşekkür ederiz.*

KAYNAKÇA

1-Jiang, H. L., & Xu, Q. (2011). Catalytic hydrolysis of ammonia borane for chemical hydrogen storage. *Catalysis Today*, *170*(1), 56-63.

2-Liu, Y., Wen, H., Zhou, D., Huang, X., Wu, X., Jiang, J., ... & Li, B. (2021). Tuning surface d charge of Ni-Ru alloys for unprecedented catalytic activity towards hydrogen generation from ammonia borane hydrolysis. *Applied Catalysis B: Environmental*, *291*, 120094.

3-Mao, M., Chen, Q., Wu, J., & Fan, G. (2020). Anchoring and space-confinement effects to synthesize ultrasmall Pd nanoparticles for efficient ammonia borane hydrolysis. *International Journal of Hydrogen Energy*, *45*(51), 27244-27253.

4-Zhang, J., Li, J., Yang, L., Li, R., Zhang, F., & Dong, H. (2021). Efficient hydrogen production from ammonia borane hydrolysis catalyzed by TiO2-supported RuCo catalysts. *International Journal of Hydrogen Energy*, *46*(5), 3964-3973.

5-Asim, M., Zhang, S., Maryam, B., Xiao, J., Shi, C., Pan, L., & Zou, J. J. (2023). Pt loading to promote hydrogen evolution from ammonia-borane hydrolysis of Ni2P under visible light. *Applied Surface Science*, *620*, 156787.

6-Sun, D., Mazumder, V., Metin, O., & Sun, S. (2011). Catalytic hydrolysis of ammonia borane via cobalt palladium nanoparticles. *ACS nano*, *5*(8), 6458-6464.

7- Çelebi M., Rüzgar A., Karataş Y., Gülcan M. Manganese oxide octahedral molecular sieves stabilized Rh nanoparticles for the hydrogen production from the ethylenediamine-bisborane hydrolysis. Int. J. Hydrogen Energy. 47 (2022) 16494-16506.

8- Ozay H., Ilgin P., Sezginturk M.K., Ozay O. Pd nanoreactors with excellent catalytic activity supported in p(SPA) hydrogel networks for hydrogen production from ethylenediamine bisborane Renew Energy. 155 (2020) 500-512.

9- Onder A., Ozay O. Fly ash as catalyst support material in the hydrolysis of ethylenediamine bisborane for hydrogen production: the use of coal-fired power plant waste. Int J Hydrogen Energy, 45 (2020) 11651-11661.

10-Xu, C., Wang, H., Wang, Q., Wang, Y., Zhang, Y., & Fan, G. (2019). Ruthenium coordinated with triphenylphosphine-hyper-crosslinked polymer: An efficient catalyst for hydrogen evolution reaction and hydrolysis of ammonia borane. *Applied Surface Science*, *466*, 193-201.

11- Chu, H., Li, N., Qiu, X., Wang, Y., Qiu, S., Zeng, J. L., ... & Sun, L. (2019). Poly (N-vinyl-2-pyrrolidone)-stabilized ruthenium supported on bamboo leaf-derived porous carbon for NH3BH3 hydrolysis. *International Journal of Hydrogen Energy*, *44*(55), 29255-29262.

12-Wang, X., Sun, S., Huang, Z., Zhang, H., & Zhang, S. (2014). Preparation and catalytic activity of PVP-protected Au/Ni bimetallic nanoparticles for hydrogen generation from hydrolysis of basic $NaBH_4$ solution. *International journal of hydrogen energy*, *39*(2), 905-916.

13-Du, J. T., Niu, H., Wu, H., Zeng, X. F., Wang, J. X., & Chen, J. F. (2021). PVP-stabilized platinum nanoparticles supported on modified silica spheres as efficient catalysts for hydrogen generation from hydrolysis of ammonia borane. *International Journal of Hydrogen Energy*, *46*(49), 25081-25091.

14-Zhao, N., Peng, J., Wang, J., & Zhai, M. (2022). Novel carboxy-functionalized PVP-CdS nanopopcorns with homojunctions for enhanced photocatalytic hydrogen evolution. *Acta Phys. Chim. Sin.*, *38*(4), 2004046.

15-Rakap, M. (2014). Hydrogen generation from hydrolysis of ammonia borane in the presence of highly efficient poly (N-vinyl-2-pyrrolidone)-protected platinum-ruthenium nanoparticles. *Applied Catalysis A: General*, *478*, 15-20.

16. Karataş Y., Gülcan M. (2017). Synthesized polyvidone-stabilized Rh(0) nanoparticles catalyzed the hydrolytic dehydrogenation of methylamine-borane in ambient conditions. *New Journal of Chemıstry*, 39 (2), 269-277.

17. Tacyildiz S., Demirkan B., Karataş Y., Gülcan M., Şen F. (2019). Monodisperse Ru-Rh bimetallic nanocatalyst as highly efficient catalysts for hydrogen generation from hydrolytic dehydrogenation of methylamine-borane. *Journal of Molecular Lıquids*, 285, 1-8.

ORAL PRESENTATION

MIL-101 Yapısında Kararlılaştırılmış Altın (0) Nanokümelerinin Hazırlanması, Karakterizasyonu ve Nitrofenol Türevlerinin İndirgenmesinde Katalitik Performanslarının Araştırılması*

Berfin Pehlivan[1], Adem Rüzgar[2], Mehmet Gülcan[1]

[1]Van Yüzüncü Yıl Üniversitesi Fen Fakültesi, Kimya Bölümü, Van, Türkiye
[2]Van Yüzüncü Yıl Üniversitesi Muradiye Meslek Yüksekokulu, Kimya ve Kimyasal İşleme Teknolojileri Bölümü, Van, Türkiye

Özet

Endüstriyel üretim süreçlerinin önemli bir kısmında ortaya çıkan nitrofenol ve türevlerinin verimli, çevreci ve ekonomik bir şekilde arıtılmaları veya dönüştürülmeleri önemli bir çalışma alanı olarak karşımıza çıkmaktadır. Yapılan araştırmalar kimyasal indirgeme (katalitik hidrojenasyon) yönteminin maliyet, verimlilik, zaman ve uygulanabilirlik açısından oldukça etkili bir arıtma yöntemi olduğunu göstermiştir. Bu yöntemin bir diğer avantajı da indirgenme sonucu elde edilen aminofenollerin nitrofenollere göre çok daha düşük toksisiteye sahip olmasıdır. Bununla birlikte, aminofenoller ve türevleri birçok analjezik ve antipiretik ilacın hazırlanmasında ara ürün veya ilaç etken maddesi olarak kullanılmaktadır. Boya ve kozmetik sanayiinde de yaygın olarak kullanılan aminofenoller yakıtlarda korozyon önleyici ve yağlayıcı olarak da kullanılmaktadır. Yapılan çalışmada oldukça zararlı ve tehlikeli kimyasallar olan 2-nitrofenol ve 4-nitrofenolün, 2-aminofenol ve 4-aminofenole verimli bir şekilde indirgenebilmesi amaçlandı. Bu amaç doğrultusunda MIL-101 destekli Au (0) nanokümeleri tepkime içi hazırlanarak bu katalizörlerin farklı sıcaklık ve derişimlerde indirgenme tepkimelerindeki katalitik performansları incelendi. Hazırlanan katalizörün yapısal ve morfolojik özellikleri XRD, SEM, SEM/EDX ve UV-vis teknikleriyle ortaya konuldu. Nitrofenollerin aminofenollere katalitik indirgenmesi oda koşullarında UV-vis spektrofotometre kullanılarak takip edildi.

Anahtar Kelimeler: Nitrofenol, Hidrojenasyon, Katalizör, Nanoparçacık, MIL-101

Abstract

Efficient, environmentally friendly and economical purification or transformation of nitrophenol and its derivatives, which occur in a significant part of industrial production processes, emerges as an important field of study. Research has shown that the chemical reduction (catalytic hydrogenation) method is a very effective treatment method in terms of cost, efficiency, time and applicability. Another advantage of this method is that the aminophenols obtained as a result of reduction have much lower toxicity than nitrophenols. However, aminophenols and their derivatives are used as intermediate products or drug active ingredients in the preparation of many analgesic and antipyretic drugs. Aminophenols, which are widely used in the paint and cosmetics industry, are also used as corrosion inhibitors and lubricants in fuels. The aim of the study was to efficiently reduce 2-nitrophenol and 4-nitrophenol, which are very harmful and dangerous chemicals, to 2-aminophenol and 4-aminophenol. For this purpose, MIL-101 supported Au (0) nanoclusters were prepared for the reaction and the catalytic performances of these catalysts in reduction reactions at different temperatures and concentrations were examined. Structural and morphological properties of the prepared catalyst were revealed by XRD, SEM, SEM/EDX and UV-vis techniques. The catalytic reduction of nitrophenols to aminophenols was monitored using a UV-vis spectrophotometer under room conditions.

Key Words: Nitrophenol, Hydrogenation, Catalyst, Nanoparticle, MIL-101

GİRİŞ

Son yıllarda sıkça gündeme gelen “atık su” kavramı; genel olarak başta sanayi olmak üzere tarım, hayvancılık ve konutlarda kullanıldıktan sonra çevreye aktarılan ve yapısındaki zararlı kimyasallar ile insan yaşamını doğrudan ve dolaylı yollarla tehdit eden küresel bir sorun olarak tanımlanmaktadır. Hızlı nüfus artışı, iletişim teknolojilerindeki gelişmeler ve kentleşme gibi birçok parametreye bağlı olarak değişen ve çeşitlenen üretim ve tüketim süreçleri hem çevreye aktarılan atık su miktarını hem de atık suların yapısında bulunan ve toksik etki gösteren bileşiklerin çeşitliliğini ve sayısını artırmaktadır. Bu zararlı bileşiklerin önemli bir kısmını oluşturan organik bileşikler canlılar üzerindeki hayati etkileri nedeniyle öncelikle incelenmesi, arıtılması veya dönüştürülmesi gereken bileşikler olarak değerlendirilmektedir. Bu amaçla yapılan çalışmalar, incelen organik

DRAFT

bileşiklerin önemli bir yüzdesinin nitrofenol ve türevlerine ait olduğunu ortaya koymuştur [1-4]. Yüksek toksisiteye, korrozif ve uyuşturucu özelliğe sahip olan nitrofenoller ve türevleri çok düşük konsantrasyonlarda bile temiz su kaynaklarını kirleterek tat ve koku değişikliğine neden olmakta ve suyun biyokimyasal yapısını bozabilmektedir. Bu nedenle Dünya Sağlık Örgütü (WHO) her türlü üretim tesisinden çevreye aktarılan atık suların yapısındaki nitrofenol ve türevlerinin arıtılmasını veya zararsız bileşiklere dönüştürülmesini talep etmektedir [5-9].

Nitrofenolleri atık sulardan giderme üzerine yapılan çalışmalarda test edilen yöntemlerin başında biyolojik arıtma, membran filtreleme, mikrobiyal parçalanma, elektrokimyasal oksidasyon, iyon değiştirme, adsorpsiyon ve kimyasal indirgeme gelmektedir. Nitrofenollerin arıtılmasını veya zararsız bileşiklere dönüştürülmesini hedefleyen bu yöntemler maliyet, etkinlik, uygulanabilirlik ve yüksek dönüşüm verimliliği gibi faktörler göz önünde bulundurularak karşılaştırıldığında öne çıkan ve birçok araştırma/uygulama çalışmasına konu olan yöntem kimyasal indirgeme (katalitik hidrojenasyon) yöntemi olmaktadır [10-14]. İlaç, petrokimya ve gıda sanayiinde yaygın olarak kullanılan katalitik hidrojenasyon yönteminin nitrofenollere uygulanması sonucu, bir yandan her yönüyle insan sağlığı açısından son derece tehlikeli kimyasallar olan nitrofenollerin ekonomik ve çevreci olarak arıtılması/dönüştürülmesi; diğer yandan birçok endüstriyel üretim sürecinde ihtiyaç duyulan ve oldukça önemli ara ürünler olarak değerlendirilen aminofenollerin eldesi mümkün olmaktadır [15,16].

Yapılan araştırmalar $NaBH_4$ gibi üstün özelliklere sahip bir “hidrojen kaynağı” ile aktif metal ve destekleyicilerin uygun şartlarda bir araya getirilmesi sonucu elde edilen “katalizör sistemlerinin” birlikte kullanılması durumunda katalitik hidrojenasyon sürecinin son derece verimli ve hızlı bir şekilde gerçekleştirilebildiğini ortaya koymuştur. Metal oksitler, biyoseramik malzemeler, karbon bazlı bileşikler (karbon nanolifler, karbon nanotüpler, grafen oksit, ve grafen), metal-organik çerçeveler (MOF), zeolitik imidazol çerçeveler (ZIF) ve polimerik yapılar, hem katalitik etkinliği hem de tekrar kullanılabilirlik performansını artırmak ve aktif metal nanopartiküllerin agregasyonundan kaynaklanan olumsuzlukları ortadan kaldırmak için kullanılan destek malzemelerinin başında gelmektedir [17-20]. Bu malzemelerden metal organik çerçevelerin yapısı nanoteknoloji alanında sağlanan teknolojik gelişmeler sonucu tam olarak aydınlatılmış ve bu malzemelerin koordinasyon bağları ile üç boyutta genişleyen gözenekli bir yapıya sahip olduğu belirlenmiştir. Ayarlanabilir gözenek boyutu, geniş spesifik yüzey alanı ve çok sayıda aktif alan mevcudiyeti gibi son derece ilginç özelliklere sahip olan MOF’lar gaz depolama ve gaz karışımlarının ayrılması başta olmak üzere katalizör uygulamaları, sensör teknolojisi, ilaç salınımı, piller ve süperkapasitörler gibi bir çok alanda yaygın olarak kullanılmaya başlanmıştır. Metal-organik çerçeve ailesinin önemli bir üyesi olan ve literatürde "gözenekli krom tereftalat" olarak bilinen MIL-101 oldukça yüksek yüzey alanı ve kararlı yapısı ile birçok çalışmaya konu olmuş ve umut verici sonuçlar elde edilmiştir [21-27].
Yapılan çalışmada nitrofenollerin (2-nitrofenol, 4-nitrofenol) aminofenollere verimli, çevreci ve uygulanabilir bir şekilde indirgenebilmesi amacıyla MIL-101 destekli Au nanokümeleri hazırlandı. Sodyum borhidrürün indirgeyici ajan olarak kullanıldığı çalışmalarda çözelti derişimi, sıcaklık, katalizör miktarı gibi farklı parametrelerde katalitik hidrojenasyon süreçleri kinetik ve termodinamik olarak incelendi. Sentezlenen nanokatalizörlerin yapısı FTIR, XRD, XPS, SEM gibi spektroskobik yöntemlerle ayrıntılı bir şekilde karakterize edildi.

MATERYAL

Yapılan çalışmada gerçekleştirilen iki aşamalı sentez çalışmalarının ilk aşamasında destek malzemesi olarak kullanılan MIL-101, ikinci aşamasında ise Au@MIL-101 nanokümeleri sentezlenmiştir. Sentez çalışmalarında ve katalitik indirgenme tepkimelerinde ihtiyaç duyulan krom (III) nitrat nonahidrat, tereftalik asit, sodyum borhidrit, 2-nitrofenol (2-NP), 4- nitrofenol (p-nitrofenol, 4-NP) ve altın (III) klorür trihidrat Sigma-Aldrich®ten temin edildi. Hem sentez hem de indirgenme tepkimelerinde kullanılan cam ve diğer malzemeler kromik asit temizleme solüsyonu, su ve aseton ile yıkanıp kurutularak kullanıldı.

YÖNTEM

Au@MIL-101 katalizörünün nitrofenol indirgemesindeki katalitik aktivitesi

Au^{+3}@MIL-101 ön katalizörünün sentezlenebilmesi amacıyla ilk aşamada krom (III) nitrat nonahidrat ve tereftalik asit kullanılarak MIL-101 sentezlendi. Destek malzemesinin sentezlenmesinden sonra hedeflenen

nanokatalizörlerin sentezi için 5,0 mL sulu $AuCl_3 \cdot 3H_2O$ çözeltisi ile 100 mg MIL-101 700 rpm'de 12 saat karıştırıldı. Çözeltide oluşan katı kısım süzülerek distile su ile yıkandı. Elde edilen nihai ürün (Au^{+3}@MIL-101) 100 C^{O}'de vakumda kurutuldu. Daha sonra çalışmada kullanılacak olan 4-nitrofenol ve 2-nitrofenol 2 mM (27.8 mg, 100 mL) stok çözeltisi hazırlandı. Bu çözeltiden 10 mL alınıp oda sıcaklığına ayarlanmış ceketli bir schlenk içerisine konuldu. Üzerine 5 mg hazırlanmış olan ön katalizör (Au^{+3}@MIL-101) eklenerek karışım 15 dakika boyunca termal dengeye gelmesi için karıştırıldı. Sonrasında tepkime ortamına 1,0 ml suda çözünmüş 0,2 mmol $NaBH_4$ (77,2 mg) ilave edilerek 700 rpm karıştırma hızında katalitik tepkime başlatıldı. Katalitik tepkime devam ederken belirli zaman aralıklarında tepkime kabından numune alınarak Shimadzu UV-3600 UV–vis spektrofotometre ile 2-nitrofenol ve 4-nitrofenole ait karakteristik absorbsiyon piklerindeki değişim kaydedildi. Nitrofenollerin indirgenmesindeki katalitik performans absorbsiyon spektrum pikleri aracılığıyla hesaplandı.

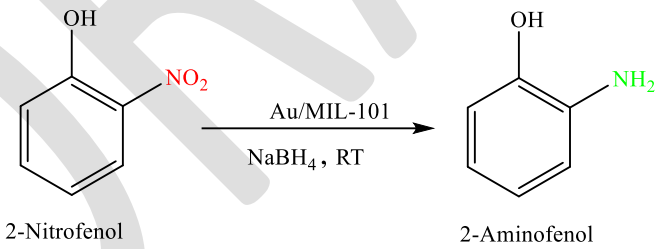
Katalizör miktarı, sıcaklık ve substrat derişimi gibi parametrelerin katalitik performansa etkisini belirleyebilmek amacıyla farklı sıcaklık (293 K, 298 K, 303 K, 308 K), farklı derişim (0,5 mM, 1 mM, 2 mM, 3 mM) ve farklı katalizör miktarlarında (1 mg, 3 mg, 5 mg, 7 mg) deneyler yapıldı ve elde edilen veriler kullanılarak katalitik etkinlikte meydana gelen değişimler hesaplandı.

Au@MIL-101 katalizörünün nitrofenol indirgemesinde tekrar kullanılabilirlik performansının araştırılması

Katalitik hidrojenasyonda kullanılan her bir substrat için indirgenmenin tamamlanmasından sonra tekrar kullanılabilirlik performansının belirlenebilmesi amacıyla Au@MIL-101 katalizörü süzme yoluyla elde edildi. Etanol-su karışımı ile temizlenen katalizör kurutularak tekrar kullanım için hazır hale getirildi. Kullanılacak katalizör miktarı tartıldıktan sonra reaksiyon kabına eşit miktarda substrat ilave edildi ve katalitik reaksiyon yeniden başlatıldı.

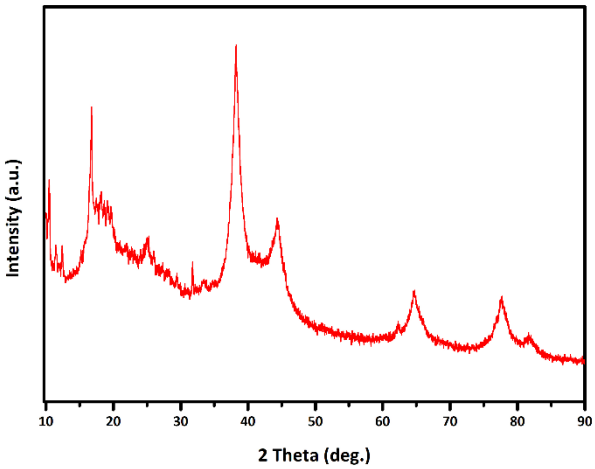
BULGULAR VE TARTIŞMA

2-nitrofenol ve 4-nitrofenolün 2-aminofenol ve 4- aminofenole indirgenebilmesi amacıyla öncelikli olarak reaksiyonda kullanılacak Au@MIL-101 katalizörü hazırlandı. Hazırlanan katalizör $NaBH_4$ varlığında indirgenme tepkimesinde kullanıldı. Daha sonra sıcaklık, derişim, miktar gibi faktörlerin katalitik aktiviteye etkisini belirleyebilmek amacıyla bir dizi deney yapıldı. En son aşamada katalizörün tekrar kullanılabilirlik performansı belirlendi.



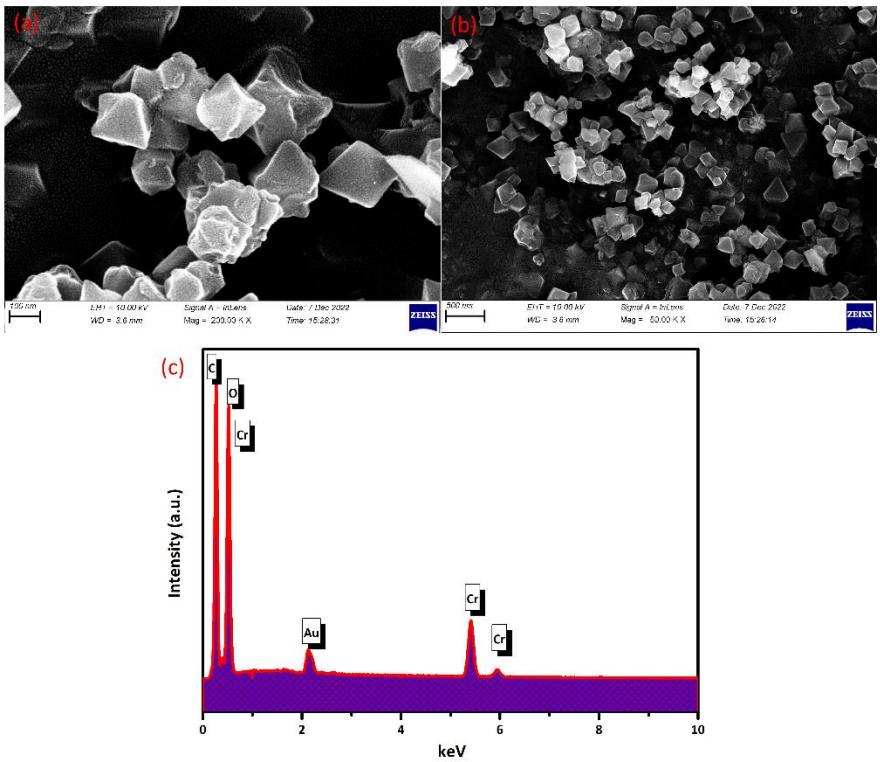
Şekil 1. 2-nitrofenolün 2-aminofenole indirgenmesi

Hazırlanan nanokümelerin yapısı ileri analitik ve spektral yöntemler kullanılarak aydınlatıldı. Bu amaçla yapılan XRD analizi ile Au parçacıklarının MIL-101'in yüzeyine tutunup tutunmadığı ve yüzeyde herhangi bir bozunma gerçekleşip gerçekleşmediği incelendi. MIL-101 destekli Au nanokümeleri için elde edilen XRD desenlerin $2\theta =$ 64.68° ve 76.3°'de olduğu ve literatür ile uyumlu olduğu belirlendi [26].



Şekil 2. MIL-101 destekli Au nanokatalizörünün 2θ = 10- 90˚ aralığındaki XRD desenleri

MIL-101 destekli Au nanokümelerin nitrofenollerin $NaBH_4$ varlığında indirgenmesinden tepkime-içi elde edilen Au@MIL-101 nanokatalizörünün parçacık büyüklük dağılımları taramalı elektron mikroskobu (SEM) ile karakterize edildi. Şekil 2(a-b)'de görüldüğü gibi Au@MIL-101 örneklerine ait 100 ve 500 nm büyüklükteki SEM görüntüleri incelendiğinde Au parçacıklarının MIL-101 yüzeyine düzgün bir şekilde tutunduğu ve homojen olarak dağıldığı görülmektedir. Şekil 2 c'de görülen SEM EDX analizinde ise hem destek malzemesinde yer alan elementlerin hem de Au metalinin varlığı çok net bir şeklide anlaşılmaktadır.

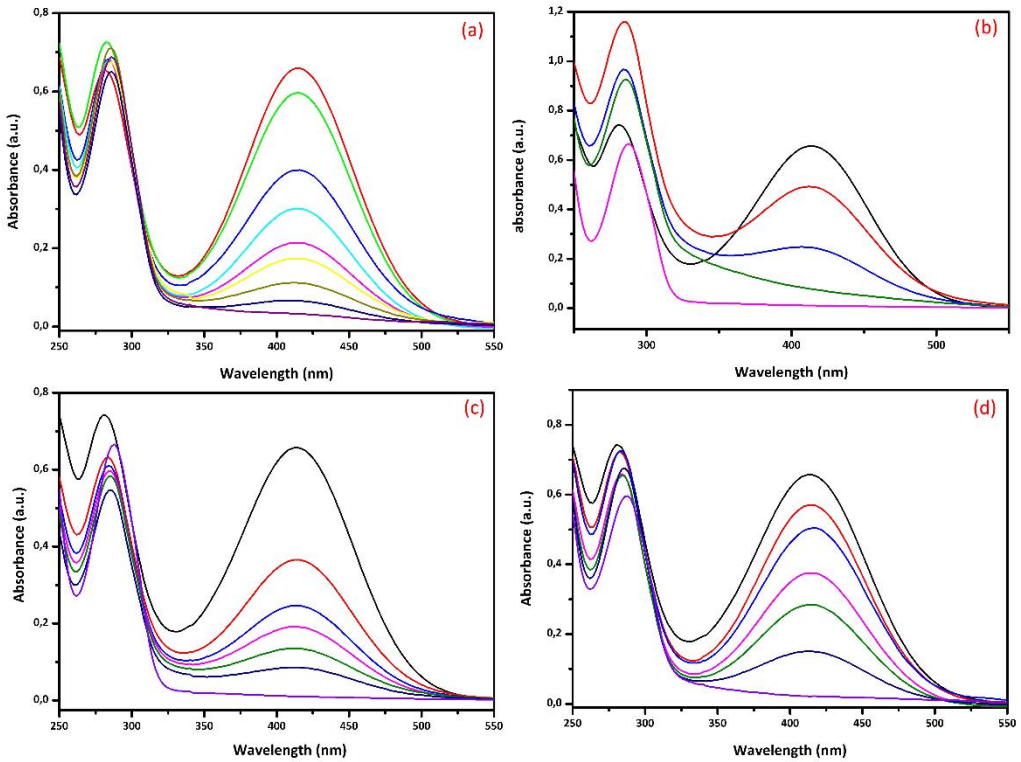


Şekil 3. Tepkime-içi koşullarda (a) 100 nm, (b) 500 nm 2- nitrofenolün indirgenmesi sonucu elde edilen Au@MIL-101 katalizörüne ait SEM görüntüleri, (c) SEM EDX analizi

Metal oranının nitrofenollerin katalitik indirgenmesi üzerindeki etkisini belirleyebilmek amacıyla destek malzemesi olarak kullanılan MIL-101 üzerine farklı oranlarda (%1, %2, %4 ve %8) metal yüklemesi yapılarak nitrofenollerin karşılık gelen aminofenollere indirgemesi çalışması yapılmıştır. Deneysel sonuçlar metal oranı ile tepkime hızının doğru orantılı olarak değiştiğini, metal oranı azaldıkça katalitik hidrojenasyonun

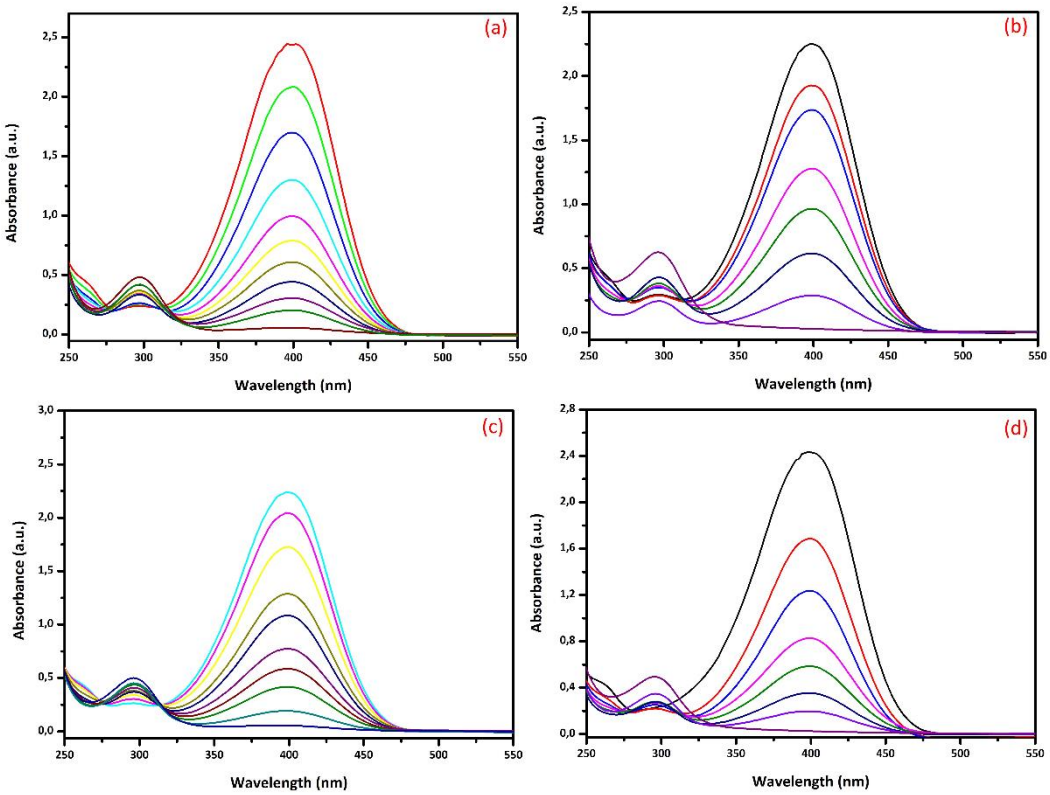
yavaşladığını ortaya koymuştur. Literatür araştırmaları da göz önünde bulundurularak %2 oranında metal yüklemesi yapılan Au@MIL-101 nanokatalizörü ile çalışmaların yapılmasına karar verilmiştir.

Bir sonraki aşamada sıcaklığın, nitrofenollerin Au@MIL-101 nanokatalizörü varlığında gerçekleştirilen katalitik indirgenme tepkimesi üzerindeki etkisini incelemek için 293, 298, 303 ve 308 K sıcaklıklarında deneyler gerçekleştirildi. Elde edilen UV-vis spektrumları incelendiğinde sıcaklık artışıyla indirgenme hızının doğru orantılı değiştiği belirlendi. 2-nitrofenolün katalitik hidrojenasyonu 293 K'de 90 saniye, 298 K'de 50 saniye, 303 K'de 40 saniye 308 K'de ise 25 saniye sürmüştür (Şekil 4).

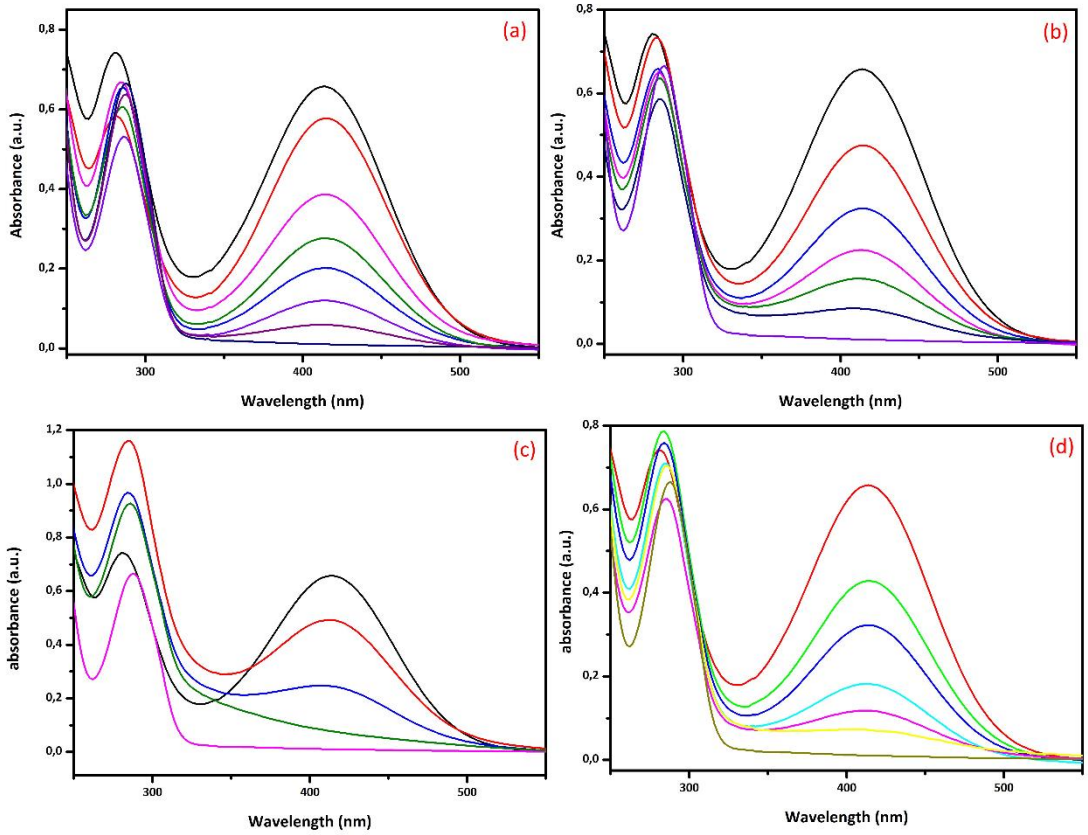


Şekil 4. Farklı sıcaklıklarda 2-nitrofenolün sulu $NaBH_4$ varlığında indirgenmesine ilişkin UV-Vis spektrumları a)293 K b) 298 K c) 303 K d) 308 K

Sıcaklığın tepkime hızına olan etkisinin belirlendiği çalışmalardan sonra katalizör miktarının indirgenme hızına etkisini belirleyebilmek amacıyla yapılan deneylerde katalizör miktarının artırılmasının tepkime hızını doğru orantılı etkileyerek artırdığı belirlendi. 4-nitrofenolün katalitik hidrojenasyonunda 7 mg Au@MIL-101 nanokatalizörü kullanıldığında 40 saniyede gerçekleşen dönüşüm, 1 mg Au@MIL-101 nanokatalizörü kullanıldığında 240 saniyede gerçekleşmektedir (Şekil 5). Benzer şekilde 1 mg katalizör kullanıldığında 180 saniye süren 2-nitrofenolün indirgenmesi katalizör miktarı 7 mg'a çıkarıldığında 30 saniye sürmektedir (Şekil 6).



Şekil 5. Farklı katalizör miktarlarının 4-nitrofenolü sulu $NaBH_4$ varlığında indirgenmesine ilişkin UV-Vis spektrumları a) 1mg b) 3 mg c) 5 mg d) 7 mg



Şekil 6. Farklı katalizör miktarlarının 2-nitrofenolü sulu $NaBH_4$ varlığında indirgenmesine ilişkin UV-Vis spektrumları a) 1mg b) 3 mg c) 5 mg d)7 mg

DRAFT

MIL-101 destekli Au@MIL-101 katalizörünün farklı konsantrasyonlarda (0.5mM, 1 mM, 2 mM ve 4 mM) hazırlanan 4-nitrofenol çözeltilerinin katalitik hidrojenasyon tepkimelerindeki etkinliğini belirleyebilmek amacıyla yapılan çalışmalarda konsantrasyonunun düşük olduğu durumda indirgenme reaksiyonunun daha hızlı gerçekleştiği belirlendi. Ortamda bulunan 4-nitrofenol miktarı arttıkça indirgenme süresinin artması doğal bir sonuç olarak değerlendirildi. Çözelti derişiminin 0.5 mM olduğu indirgenme tepkimesi 25 saniyede gerçekleşirken, derişimin 3 mM olduğu tepkime 90 saniye sürmektedir.

SONUÇ

Yapılan çalışmada oldukça yüksek yüzey alanı ve kararlı yapısı nedeniyle destekleyici olarak MIL-101 kullanıldı ve Au@MIL-101 nanokatalizörü tepkime-içi hazırlandı. Sentezlenen nanokatalizörlerin yapısı FTIR, XRD, SEM gibi spektroskobik yöntemlerle ayrıntılı bir şekilde karakterize edildi. İndirgeyici olarak sodyum borhidrürün kullanıldığı tepkimelerde 4-nitrofenolün 4-aminofenole; 2-nitrofenolün 2-aminofenole indirgenmesi UV-Vis spektrumları ile takip edildi. Elde edilen veriler yorumlandığında elde edilen sonuçlar özetle şu şekilde sıralanabilir:

Au@MIL-101 nanokatalizörü nitrofenollerin katalitik hidrojenasyonunda oldukça yüksek aktiflik göstermektedir.
Tekrar kullanılabilirlik ve dayanıklılık açısından da oldukça yüksek performans gösteren Au@MIL-101 katalizörü, nitrofenollerin indirgenmesinde 5. çevrimden sonra bile %85 oranında etkinlik göstermektedir.
MIL-101'in yüksek yüzey alanı ve kararlı yapısı atomların homojen olarak dağılmasını sağlamakta ve topaklaşmayı önemli ölçüde engellemektedir.

Teşekkür

**2209-A- Lisans Öğrencilerine Yönelik Araştırma Projesi Destek Programı kapsamındaki projemize (Proje No:1919B012206274) maddi desteği için Türkiye Bilimsel ve Teknolojik Araştırma Kurumu'na teşekkür ederiz.*

KAYNAKÇA

Naushad, M., Ahamad, T., & Khan, M. R. (2022). Remediation of wastewater containing 4-nitrophenol using ionic liquid stabilized nanoparticles: Synthesis, characterizations and applications. *Chemosphere*, *303*, 135173.

Hu, C., Hu, S., Fang, P., Tang, Z., Xiao, X., & Wu, H. (2022). Waste-tire-derived activated carbon as efficient adsorbent of p-Nitrophenol from Wastewater. *Journal of Chemistry*, *2022*.

Wang, Q., Wang, P., Xu, P., Hu, L., Wang, X., Qu, J., & Zhang, G. (2021). Submerged membrane photocatalytic reactor for advanced treatment of p-nitrophenol wastewater through visible-light-driven photo-Fenton reactions. *Separation and Purification Technology*, *256*, 117783.

Wang, H., Li, Z., Zhang, F., Wang, Y., Zhang, X., Wang, J., & He, X. (2021). Comparison of Ti/Ti_4O_7, Ti/Ti_4O_7-PbO_2-Ce, and Ti/Ti_4O_7 nanotube array anodes for electro-oxidation of p-nitrophenol and real wastewater. *Separation and Purification Technology*, *266*, 118600.

Ahmaruzzaman, M., & Gayatri, S. L. (2010). Activated tea waste as a potential low-cost adsorbent for the removal of p-nitrophenol from wastewater. *Journal of Chemical & Engineering Data*, *55*(11), 4614-4623.

Chen, J., Sun, X., Lin, L., Dong, X., & He, Y. (2017). Adsorption removal of o-nitrophenol and p-nitrophenol from wastewater by metal–organic framework Cr-BDC. *Chinese journal of chemical engineering*, *25*(6), 775-781.

Dong, X., Gan, Z., Lu, X., Jin, W., Yu, Y., & Zhang, M. (2015). Study on catalytic and non-catalytic supercritical water oxidation of p-nitrophenol wastewater. *Chemical Engineering Journal*, *277*, 30-39.

Luan, J., & Plaisier, A. (2004). Study on treatment of wastewater containing nitrophenol compounds by liquid membrane process. *Journal of Membrane Science*, *229*(1-2), 235-239.

Koubaissy, B., Joly, G., & Magnoux, P. (2008). Adsorption and competitive adsorption on zeolites of nitrophenol compounds present in wastewater. *Industrial & engineering chemistry research*, *47*(23), 9558-9565.

Zhao, P., Feng, X., Huang, D., Yang, G., & Astruc, D. (2015). Basic concepts and recent advances in nitrophenol reduction by gold-and other transition metal nanoparticles. *Coordination Chemistry Reviews*, *287*, 114-136.

Pachfule, P., Kandambeth, S., Díaz, D. D., & Banerjee, R. (2014). Highly stable covalent organic framework–Au nanoparticles hybrids for enhanced activity for nitrophenol reduction. *Chemical Communications*, *50*(24), 3169-3172.

Zhang, P., Sui, Y., Xiao, G., Wang, Y., Wang, C., Liu, B., ... & Zou, B. (2013). Facile fabrication of faceted copper nanocrystals with high catalytic activity for p-nitrophenol reduction. *Journal of Materials Chemistry A*, *1*(5), 1632-1638.

Huang, C., Ye, W., Liu, Q., & Qiu, X. (2014). Dispersed Cu2O octahedrons on h-BN nanosheets for p-nitrophenol reduction. *ACS applied materials & interfaces*, *6*(16), 14469-14476.

Huang, Y., Zheng, K., Liu, X., Meng, X., & Astruc, D. (2020). Optimization of Cu catalysts for nitrophenol reduction, click reaction and alkyne coupling. *Inorganic Chemistry Frontiers*, *7*(4), 939-945.

Mandlimath, T. R., & Gopal, B. (2011). Catalytic activity of first row transition metal oxides in the conversion of p-nitrophenol to p-aminophenol. *Journal of Molecular Catalysis A: Chemical*, *350*(1-2), 9-15.

Vaidya, M. J., Kulkarni, S. M., & Chaudhari, R. V. (2003). Synthesis of p-aminophenol by catalytic hydrogenation of p-nitrophenol. *Organic process research & development*, *7*(2), 202-208.

Gangu, K. K., Maddila, S., Mukkamala, S. B., & Jonnalagadda, S. B. (2019). Characteristics of MOF, MWCNT and graphene containing materials for hydrogen storage: A review. *Journal of energy chemistry*, *30*, 132-144.

Yan, J. M., Wang, Z. L., Gu, L., Li, S. J., Wang, H. L., Zheng, W. T., & Jiang, Q. (2015). AuPd–MnOx/MOF–graphene: an efficient catalyst for hydrogen production from formic acid at room temperature. *Advanced Energy Materials*, *5*(10), 1500107.

Zhong, R. Q., Zou, R. Q., Nakagawa, T., Janicke, M., Semelsberger, T. A., Burrell, A. K., & Del Sesto, R. E. (2012). Improved hydrogen release from ammonia–borane with ZIF-8. *Inorganic Chemistry*, *51*(5), 2728-2730.

Ertas, I. E., Gulcan, M., Bulut, A., Yurderi, M., & Zahmakiran, M. (2016). Metal-organic framework (MIL-101) stabilized ruthenium nanoparticles: Highly efficient catalytic material in the phenol hydrogenation. *Microporous and Mesoporous Materials*, *226*, 94-103.

Zorainy, M. Y., Alalm, M. G., Kaliaguine, S., & Boffito, D. C. (2021). Revisiting the MIL-101 metal–organic framework: design, synthesis, modifications, advances, and recent applications. *Journal of Materials Chemistry A*, *9*(39), 22159-22217.

Gómez-Paricio, A., Santiago-Portillo, A., Navalón, S., Concepción, P., Alvaro, M., & Garcia, H. (2016). MIL-101 promotes the efficient aerobic oxidative desulfurization of dibenzothiophenes. *Green Chemistry*, *18*(2), 508-515.

Yang, C. X., & Yan, X. P. (2011). Metal–organic framework MIL-101 (Cr) for high-performance liquid chromatographic separation of substituted aromatics. *Analytical chemistry*, *83*(18), 7144-7150.

Somayajulu Rallapalli, P. B., Raj, M. C., Patil, D. V., Prasanth, K. P., Somani, R. S., & Bajaj, H. C. (2013). Activated carbon@ MIL-101 (Cr): a potential metal-organic framework composite material for hydrogen storage. *International journal of energy research*, *37*(7), 746-753.

Wen, M., Mori, K., Kamegawa, T., & Yamashita, H. (2014). Amine-functionalized MIL-101 (Cr) with imbedded platinum nanoparticles as a durable photocatalyst for hydrogen production from water. *Chemical Communications*, *50*(79), 11645-11648.

Wang, Y., Zhang, Y., Jiang, Z., Jiang, G., Zhao, Z., Wu, Q., ... & Xu, C. (2016). Controlled fabrication and enhanced visible-light photocatalytic hydrogen production of Au@ CdS/MIL-101 heterostructure. *Applied Catalysis B: Environmental*, *185*, 307-314.

Soğukömeroğulları H. G., Karataş Y., Çelebi M., Gülcan M., Sönmez M., Zahmakıran M.(2019) Palladium nanoparticles decorated on amine functionalized graphene nanosheets as excellent nanocatalyst for the hydrogenation of nitrophenols to aminophenol counterparts. *Journal of Hazardous Materıals*, 369, 96-107.

ORAL PRESENTATION

The determination of genetic diversity for insecticide resistance management strategies in major agricultural pests: the case of *Ceratitis capitata*

Ersin DOĞAÇ[1*] (*ORCID: https://orcid.org/0000-0003-4426-2187*)

[*1] Muğla Sıtkı Koçman University, Faculty of Science, Department of Molecular Biology and Genetics, Muğla, Turkey

*Correspondence e-mail: ersindogac@mu.edu.tr

Abstract

The Mediterranean fruit fly (medfly), *Ceratitis capitata* Wied. (Diptera: Tephritidae), is an important citrus pest that negatively affects yield and quality, causing significant economic losses in citrus-producing countries. Although the medfly is economically important, studies on the genetic structure of this species are very limited. In this study, RAPD markers were used to provide information about the genetic structure of medfly populations in Turkey, was examined. which represents a significant part of the Eastern Mediterranean basin. The mean observed number of alleles (n_a) for all populations was 1.44 ± 0.49 and the mean effective allele number (n_e) was 1.21 ± 0.32. The rate of polymorphic loci for all populations ranged from 23.65% to 59.46%, and the gene diversity (h) of Nei ranged from 0.0897 to 0.1740. Gene flow level (Nm) was 1.12 and genetic differentiation value (G_{st}) was 0.31. According to the UPGMA dendrogram, no East-West grouping was observed among the studied populations. There are significant opportunities to enhance medfly control by harnessing the knowledge of the pest's genetic diversity and population dynamics. A deeper genetic understanding of this invasive insect can enable more precise, efficient, and long-lasting management practices.

Keywords: *Ceratitis capitata*, population genetics, RAPD, insecticide resistance management

INTRODUCTION

Citrus fruits are the most produced group in the world with a production of approximately 143 million tons (FAO, 2019). Considering the possible climate and water crises in the future, it is important that agricultural production, which will provide adequate nutrition for the increasing population, can be carried out with the least loss (Eckardt, 2009). Various pests cause economic product losses in agricultural products. Among these pests, the Mediterranean fruit fly (*Ceratitis capitata* Wiedemann, 1824), also known as the medfly, is one of the world's most destructive agricultural pests. It has been reported that medfly causes damage to more than 260 plants, including tropical and subtropical fruit, vegetable, and ornamental plants in more than 70 countries around the world (Magana et al., 2007). Medfly is a polyphagous species as it feeds on multiple crops, making it one of the most important quarantine species. It is native to the African Sahara but has spread to many other parts of the world, including South and North Africa, Central and South America, Western Australia, and many countries neighboring the Mediterranean such as Lebanon, Israel, Jordan, and Turkey (Thomas et al., 2013). The increase in trade routes around the world, the development of global trade policies, and the ability of the organism to feed on many hosts in many habitats are the most important factors in the spread of this organism. The worldwide distribution process of this organism was reported by Malacrida et al., (1998). Accordingly, the western Mediterranean basin is shown as the possible source of the spread of this pest to the Eastern Mediterranean basin, where our country is located (Malacrida et al., 1998). As in many species, the distribution of the medfly from its point of origin to other regions is characterized by lower genetic diversity values in newly invaded regions (Malacrida et al., 1992; Gomuliski et al., 1998; Malacrida et al., 1998). Because of its adeptness at hitchhiking on human transportation routes, the medfly has been able to successfully colonize new geographic regions, despite its relatively limited flight capabilities (Malacrida et al., 1998; Malacrida et al., 2007). In many parts of the world, studies to better understand the genetic structure and colonization process of this important pest have been carried out with the help of biochemical and molecular markers (Gasperi et al., 1991; Malacrida et al., 1992; Baruffi et al., 1995; Gasparich et al., 1997; Gomulski et al., 1998; Malacrida et al., 1998; Reyes and Ochando, 1998a; He and Haymer, 1999; Bonizzoni et al., 2000; Gasperi et

al., 2002; Bonizzoni et al., 2004; Reyes and Ochando, 2004; Barr et al., 2009; Karsten et al., 2013; Karsten et al., 2015; Güler et al., 2019; Kurd et al., 2020; Nikolouli et al., 2020; Güler et al., 2022) and the results showed that genetic variation does not show a homogeneous distribution. Results from the studies indicate that Kenya is the point of origin for this organism, as it has the highest genetic diversity (Baruffi et al., 1995; Gasparich et al., 1997; Gomulski et al., 1998; Malacrida et al., 1998; Reyes and Ochando, 2004; Malacrida et al., 2007).

Medfly is responsible for economic losses in horticultural production. Therefore, it is the target of early detection, control, and eradication programs (Beroiz et al., 2012). In the fight against this important pest; (i) cultural, (ii) mechanical, (iii) biological, and (iv) chemical control methods were used. As a result of chemical control, resistance development occurs in related species (Vontas et al., 2011). Considering all these, it is of great importance to clarify the gene flow levels, population genetic structures, and colonization processes among the populations of these species in terms of demonstrating the adequacy of current control methods (Collins and Schlipalius, 2018). Overall, while pesticide and bait development initially relies on basic knowledge of medfly genetics and biology, the application and management of these control methods benefit from a genetic understanding of target populations. Customizing and monitoring the genetic interactions between flies and control measures results in a more sustainable strategy that minimizes resistance and has maximum impact. Genetics-based approaches enable an adaptive management response to this invasive pest (Kadoić et al., 2020).

The aims of this study are twofold: first, investigate the genetic structure of medfly populations to elucidate the colonization process, and second determination of gene flow levels that are important in the spread of resistance alleles.

MATERIALS AND METHODS

Sample collection

Medfly samples were collected from the provinces of Muğla, Antalya, Aydın, İzmir, Yalova, Adana, and Mersin (Figure 1). Adult collections were carried out by hanging “last fly” pheromone traps on trees at a certain distance in citrus orchards. Samples were stored at -80 °C for morphological identification and further experimental studies.



Figure 1. Distribution collection sites

DNA isolation and amplification of RAPD loci

Morphological species identification of medfly samples was performed under the binocular microscope, using adult identification keys (White and Elson-Harris, 1994). DNA isolation of the collected samples was carried out by modifying the Lifton method used by Bender et al,. (1983). In this study, seven RAPD primers (OPA-03, OPA-09, OPA-18, OPE-07, OPE-19, OPM-10, and OPM-20) were used. The PCR conditions used in this study were applied to the studied species based on Göçmen, (2001).

Data analysis

The analyses of the data file created with these values were performed using computer-based computer programs such as Popgene Version 1.32 (Yeh et al., 1999) and GenAlex (Peakall and Smouse, 2006).

RESULTS

Genetic Structures and Population Genetic Diversity

As a result of the analysis of seven primers applied to a total of 105 individuals (OPA-03, OPA-09, OPA-18, OPE-07, OPE-19, OPM-10, and OPM-20), a total of 148 loci were detected and all of them were polymorphic. The highest number of loci was obtained from the OPA-18 primer with 24 and the least number of loci from the OPM-20 primer with 17. The overall mean polymorphism rate was 40.93% (Table 1). In Figure 2, RAPD band patterns of individuals belonging to the Antalya, and Yalova populations are presented.

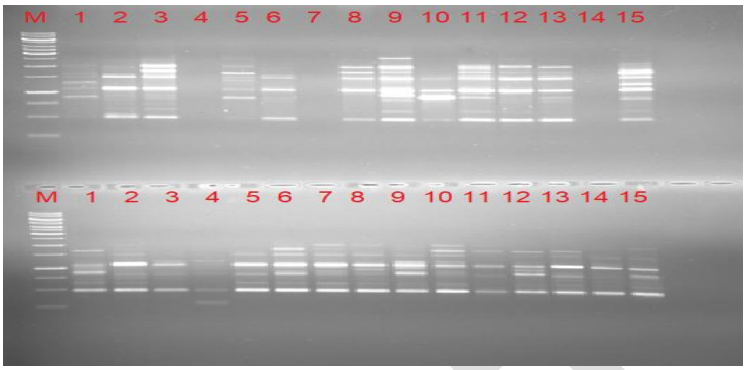


Figure 2. RAPD band patterns of Antalya and Yalova samples with OPA-18

The amount of genetic variation was dispersed homogeneously between studied populations. The mean expected heterozygosity (*He*) values varied from 0.17 (Aydın) to 0.08 (Izmir). Aydın population indicated the highest levels of genetic diversity consisting of the mean, effective number of alleles, number of bands, and number of private alleles. Compared to the other populations, lower diversity amounts were observed in the Izmir population. The mean observed allele number (n_a), which is one of the components of genetic diversity, was found to be 1.44 ± 0.49 for all populations, and the mean effective allele number (n_e) was 1.21 ± 0.32. In the studied populations, Izmir was the population with the lowest n_a value with 1.28 ± 0.45, while the population with the highest n_a value with 1.59 ± 0.49 was the Aydın population. Again in the studied populations, Izmir was the population with the lowest n_e value with 1.115 ± 0.31, while the population with the highest n_e value was the Aydın population with 1.29 ± 0.34. The mean rate of polymorphic loci in the studied populations was found to be 40.93%. The population with the lowest polymorphic locus ratio was Izmir with a value of 23.65%. Aydın population, on the other hand, has the highest rate of polymorphic loci with a value of 59.46%. In this study, the gene diversity value (*h*) of Nei was found to be an average of 0.13. The highest gene diversity was seen in the Aydın population with 0.17. The lowest value was observed in the Izmir population with 0.09 (Table 1).

Table 1. Genetic diversity parameters of *C. capitata*. Sample size (N), Observed Number of Alleles (n_a), Effective Number of Alleles (n_e), Gene Diversity (*h*), Percentage of Polymorphic Band (*%P*)

Populations	N	n_a	n_e	*h*	*%P*
Antalya	13	1.49±0.50	1.23±0.35	0.14	49.32 %
Aydın	13	1.59±0.49	1.29±0.34	0.17	59.46 %
İzmir	12	1.28±0.45	1.15± 0.31	0.09	23.65 %
Muğla	15	1.42±0.49	1.20±0.32	0.12	36.49 %
Adana	11	1.47±0.50	1.18±0.26	0.12	40.54 %
Mersin	13	1.38±0.49	1.19±0.32	0.12	37.84 %
Yalova	15	1.44±0.50	1.21±0.32	0.13	39.19 %
Avarage	**13.14**	**1.44±0.49**	**1.21±0.32**	**0.13**	**40.93 %**

According to band patterns across populations, Izmir was the population with the lowest number of bands with 41, while the population with the highest number of bands with 97 was the Aydın population. Similarly, Izmir was the population with the lowest number of common bands with 5, while Aydın was the population with the highest number of common bands with 36. In total, 32 new private alleles were determined. Antalya and Izmir were the populations with the highest number of private alleles with 8, while the population with the lowest number of private alleles with 2 was the Mersin population. Even though the number of private alleles is connected with sample size, all the studied populations had private alleles (Table 2).

Table 2. Total band patterns for binary data by populations

Population	Antalya	Aydın	İzmir	Muğla	Adana	Mersin	Yalova
No. Bands	78	97	41	63	63	65	66
No. Bands Freq. >= 5%	78	97	41	63	63	65	66
No. Private Bands	8	8	4	3	4	2	3
No. L Comm Bands (<=50%)	19	36	5	21	22	16	19
Mean He	0,14	0,17	0,08	0,10	0,10	0,12	0,11
SE of Mean He	0,02	0,02	0,01	0,01	0,01	0,01	0,01

According to the G-statistics, the total genetic diversity (H_T) was found to be 0.17 ± 0.03 at 148 loci. Of this, 0.12 is intra-population genetic diversity (H_S) and 0.05 is inter-population genetic diversity (D_{st}). Also, the mean genetic differentiation coefficient (G_{st}) was found to be 0.31. The gene flow level (N_m), obtained as the average of 300 loci in this study, was found to be 1.13.

The level of genetic variation between populations was determined by calculating Nei's (1978) standard genetic distance (*DN*) values for the studied population pairs. *DN* values based on 148 loci are given in Table 5. Considering the values, although the genetic differentiation values of all populations are low, the lowest genetic distance was found between Aydın and Antalya with 0.12, while the highest genetic distance was found between Muğla and Yalova with 0.46 (Table 3).

Table 3. Genetic distance values between populations (Nei 1978)

Populations	Antalya	Aydın	İzmir	Muğla	Adana	Mersin	Yalova
Antalya	*****						
Aydın	0.1221*	*****					
İzmir	0.2850	0.2953	*****				
Muğla	0.2414	0.3024	0.1893	*****			
Adana	0.2426	0.2265	0.4319	0.3667	*****		
Mersin	0.1570	0.1664	0.3010	0.3096	0.2062	*****	
Yalova	0.2927	0.2699	0.4152	0.4640*	0.3374	0.1755	*****

The visual expression of genetic distances between populations is given in the UPGMA (Unweighted Pair-Group Method with Arithmetic Average) dendrogram, which was created by using DN values (Figure 3). As can be seen from the dendrogram, there is no clustering of medfly populations due to their geographical location. According to the dendrogram, two groups stand out. The first group is composed of Muğla and İzmir populations. The second group consists of Antalya, Aydın, Yalova, Adana, and Mersin. According to the results of the AMOVA analysis, it was observed that 54% of the genetic variation calculated for 148 loci originates from within the population, and 46% originates from among the populations.

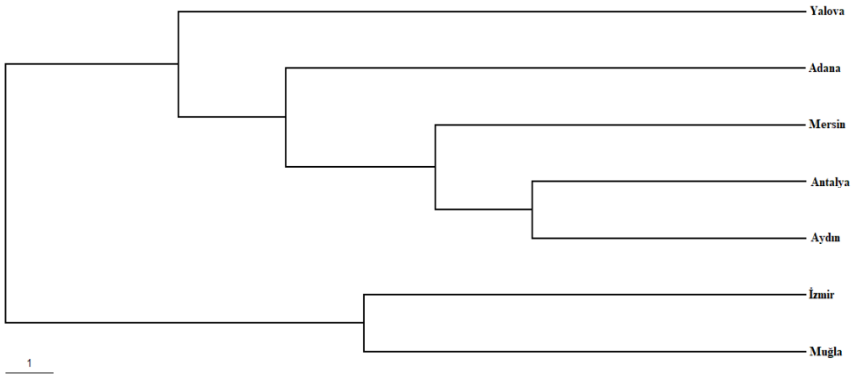


Figure 4. UPGMA dendrogram based on Nei 1978 genetic distance matrix

DISCUSSION

The genetic structure of medfly populations has been analyzed at macrogeographic and microgeographic levels in previous studies (Baruffi et al., 1995; Bonizzoni et al., 2001; Bonizzoni et al., 2002; Gasperi et al., 2002; Bonizzoni et al., 2004; Malacrida et al., 2007; Barr, 2009; Alaoui et al., 2010; Elfekih et al., 2010; Karsten et al., 2013; Rajabiyan et al., 2015; Güler et al., 2019; Kurd et al., 2020; Nikolouli et al., 2020; Güler et al., 2022). Turkey's geographical location and agricultural activities suggest that the medfly may have played a role in the invasion of the Mediterranean basin. However, there are very few studies conducted in our country with this organism to clarify this situation.

In this study, genetic diversity, population genetic structure, and inter-population relationships of medfly (*C. capitata*), which is the most important international agricultural pest, were determined in Turkey. For this reason, RAPD markers were used to elucidate the genetic structures of Mediterranean fruit fly populations. As a result of the study, genetic similarity was observed in all seven provinces in terms of both allele number and allele frequencies. As a result of RAPD analyses; it has been observed that there is a low level of genetic variation in medfly populations in Turkey and that there is no significant degree of differentiation between populations when evaluated geographically.

A low level of genetic diversity was observed in Mediterranean fruit fly populations sampled from different regions of Turkey and analyzed with seven polymorphic RAPD markers. This result is inconsistent with results of high genetic variation at the regional geographic scale (Gasperi et al., 2002; Alaoui et al. 2010). Among the possible reasons for this situation, can be considered that the researchers took samples from the hosts where chemical control was not carried out intensively, the use of African samples, and the use of laboratory strains. In our study, the average number of alleles (*na*) was 1.44 and the average effective allele number (*ne*) was 1.21 for 7 provinces. These values were reported as (*na*) 4.50 for Italy and (*na*) 3.3 for Greece in the studies conducted by Gasperi et al., (2002). Contrary to the results obtained from this study and the results obtained in previous studies using microsatellite and ISSR markers (Kurd et al., 2020; Güler et al., 2022), a lower level of genetic diversity was observed than the results in the literature.

Molecular analysis of variance indicated that the vast majority of the genetic variation (54% for *C. capitata*) was conserved within populations. The analysis of molecular variance (AMOVA) indicated that there is no genetic structure variation in *C. capitata* species populations. A mechanism of mixing of these agricultural pest populations from different geographical areas and probable gene flow rate is evident by these observations although a high geographical distance exists among these study sites. For this reason, the sites of the present study are attached by insect habitats, which authorize gene flow among these locations. Relatively high within-population genetic variation and migration rates can ensure the occasion for novel phenotypes to arise in pest populations (Zhou et al., 2000). In addition, the area, climatic alterations, and resistance to ecological factors (Willi et al., 2006) can also produce selection pressure on insects. Population genetic researches ensures helpful information concerning the potential for wide-ranging pest control, especially in species with wide geographical spread (Alphey and Bonsall, 2018). Comprehensing the phylogenetic relationships between insect populations is very important for implementing sustainable and effective medfly management.

The UPGMA dendrogram (Figure 4) showed that medfly populations in Turkey did not exhibit geographic clustering. Gene flow between populations (Nm = 1.13) appears to be sufficient to homogenize medfly populations in Turkey.

CONCLUSION

There are a couple of ways in which genetic knowledge can guide bait and insecticide selection for medfly control. These are; (i) targeting attractive volatiles and pheromones, (ii) targeting susceptible genotypes, (iii) mitigating resistance development, and (iv) monitoring for resistance alleles (Vontas et al., 2011; Karlsson Green et al., 2020; Rizvi et al., 2021). The mobility of medfly populations facilitates their ability to infest new areas of land, despite control efforts. A thorough understanding of the medfly's genetic population structure can help in developing more targeted and effective control strategies, especially for recent invasions. Continued monitoring of genetic patterns can also provide early detection of new colonization events, allowing for rapid response to limit spread. Overall, exploring the medfly's genetic diversity and population dynamics provides significant insights into both its invasion biology as well as options for management and control (Karsten et al., 2013; Arias et al., 2018). The information obtained from the studies will contribute to the

development of more effective and efficient control strategies against the medfly, which is the most important agricultural pest in the Mediterranean basin, including Turkey.

ACKNOWLEDGMENTS

This study was financially supported by the Scientific Research Project Office of Muğla Sıtkı Koçman University, project grant number MUBAP 17/082.

REFERENCES

Alaoui A, Imoulan A, Alaoui-Talibi ZE, El Meziane A 2010. Genetic structure of Mediterranean fruit fly (*Ceratitis capitata*) populations from Moroccan endemic forest of *Argania spinosa*. International Journal of Agriculture and Biology, 12:291–298.

Alphey N, Bonsall MB 2018. Genetics-based methods for agricultural insect pest management. Agricultural and Forest Entomology, 20(2): 131- 140.

Arias MB, Elfekih S, Vogler AP 2018. Population genetics and migration pathways of the Mediterranean fruit fly *Ceratitis capitata* inferred with coalescent methods. PeerJ, 6, e5340. doi: 10.7717/peerj.5340.

Barr NB 2009. Pathway analysis of *Ceratitis capitata* (Diptera: Tephritidae) using mitochondrial DNA. Journal of Economic Entomology, 102(1): 401-411.

Baruffi L, Damián G, Guglielmino CR, Bandi C, Malacrida AR, Gasperi G 1995. Polymorphism within and between populations of *Ceratitis capitata*: comparison between RAPD and multilocus enzyme electrophoresis data. Journal of Heredity, 74(4): 425-437.

Bender W, Spierer P, Hogness DS 1983. Chromosomal walking and jumping to isolate DNA from the *Ace* and *rosy* loci and bithorax complex in *D. melanogaster*. Journal of Molecular Biology, 168(1): 17-33.

Beroiz B, Ortego F, Callejas C, Hernandez-Crespo P, Castanera P, Ochando MD 2012. Genetic structure of Spanish populations of *Ceratitis capitata* revealed by RAPD and ISSR markers: implications for resistance management. Spanish Journal of Agricultural Research, 10: 815-825.

Bonizzoni M, Malacrida AR, Guglielmino CR, Gomulski LM, Gasperi G, Zheng L 2000. Microsatellite polymorphism in the Mediterranean fruit fly, *Ceratitis capitata*. Insect Molecular Biology, *9*(3): 251-261.

Bonizzoni M, Zheng L, Guglielmino CR, Haymer DS, Gasperi G, Gomulski LM, Malacrida AR 2001. Microsatellite analysis of medfly bio infestations in California. Molecular Ecology, 10(10): 2515-2524.

Bonizzoni M, Katsoyannos BI, Marguerie R, Guglielmino CR, Gasperi G, Malacrida A, Chapman T 2002. Microsatellite analysis reveals remating by wild Mediterranean fruit fly females, *Ceratitis capitata*. Molecular Ecology, 11(10):1915-1921.

Bonizzoni M, Guglielmino CR, Smallridge CJ, Gomulski M, Malacrida AR, Gasperi G 2004. On the origins of medfly invasion and expansion in Australia. Molecular Ecology, 13(12): 3845-3855.

Collins PJ, Schlipalius DI 2018 Insecticide resistance. In: Athanassiou CG, Arthur FH (eds) Recent advances in stored product protection. Berlin, Springer, pp. 169–182.

Eckardt NA, Cominelli E, Galbiati M, Tonelli C 2009. The future of science: food and water for life.

FAO. (2021). Citrus Fruit Statistical Compendium 2020. Rome.

Elfékih S, Makni M, Haymer DS 2010. Detection of novel mitochondrial haplotype variants in populations of the Mediterranean fruit fly, *Ceratitis capitata*, from Tunisia, Israel and Morocco. Journal of Applied Entomology, 134(8): 647-651.

Gasparich GE, Silva JG, Han HY, Mcpheron BA, Steck GJ, Sheppard WS 1997. Population genetic structure of Mediterranean fruit fly (Diptera: Tephritidae) and implications for worldwide colonization patterns. Entomology Society of America, 90(6): 790-797.

Gasperi G, Guglielmino CR, Malacrida AR, Milani R 1991. Genetic variability and gene flow in geographical populations of *Ceratitis capitata*. Journal of Heredity 67(3): 347-356.

Gasperi G, Bonizzoni M, Gomulski LM, Murelli V, Torti C, Malacrida AR, Guglielmino CR 2002. Genetic differentiation, gene flow and the origin of infestations of the medfly, *Ceratitis capitata*. Genetica, 116(1): 125-135.

Gomulski LM, Bourtzis K, Brogna S, Morandi PA, Bonvicini C, Sebastiani F, Torti C, Guglielmino CR, Savakis C, Gasperi G, Malacrida AR 1998. Intron size polymorphism of the Adh1 gene parallels the

worldwide colonization history of the Mediterranean fruit fly, *Ceratitis capitata*. Molecular Ecology, 7(12): 1729-1741.

Göçmen B 2001. Genetic characterization of 150 F6-Inbred durum wheat lines derived from kunduru-1149 x cham-1 cross using molecular markers and economically important traits.

Güler A, Karakoç E, Gökdere G, Doğaç E, Taşkin V 2019. Genetic structure of Mediterranean fruit fly (Diptera: Tephritidae) populations from Turkey revealed by mitochondrial DNA markers. Journal of Genetics, 98(2): 1-11.

Güler A, Taşkin V, Taşkin BG, Doğaç E 2022. Microsatellite-based genetic diversity of Mediterranean fruit fly (*Ceratitis capitata*, Diptera: Tephritidae) populations from Southwest Turkey. Turkish Journal of Zoology, 46: 278-288.

He M, Haymer DS 1999. Genetic relationships of populations and the origins of new infestations of the Mediterranean fruit fly. Molecular Ecology, 8(8): 1247-1257.

Kadoić Balaško M, Bažok R, Mikac KM, Lemic D, Pajač Živković I 2020. Pest management challenges and control practices in codling moth: A review. Insects, 11(1): 38.

Karlsson Green K, Stenberg J A, Lankinen Å 2020. Making sense of Integrated Pest Management (IPM) in the light of evolution. Evolutionary Applications, 13(8): 1791-1805.

Karsten M, Van Vuuren BJ, Barnaud A, Terblanche JS 2013. Population genetics of *Ceratitis capitata* in South Africa: Implications for dispersal and pest management. PLoS ONE, 8(1): e54281.

Karsten M, Van Vuuren BJ, Addison P, Terblanche JS 2015. Deconstructing intercontinental invasion pathway hypotheses of the Mediterranean fruit fly (*Ceratitis capitata*) using a Bayesian inference approach: are port interceptions and quarantine protocols successfully preventing new invasions?. Diversity and Distributions, 21(7): 813-825.

Kurd Ö, Doğaç E, Taşkin V, Taşkin BG 2020. Issr marker based population genetic study of Mediterranean fruit fly *Ceratitis capitata* (Diptera: Tephritidae). Genetica, 52(1): 311-322.

Magana C, Hernandez-Crespo P, Ortega F, Castanera P 2007. Resistance to malathion in field populations of *Ceratitis capitata*. Journal of Economic Entomology, 100(6): 1836-1843.

Malacrida AR, Guglielmino CR, Gasperi G, Baruffi L, Milani R 1992. Spatial and temporal differentiation in colonizing populations of *Ceratitis capitata*. Journal of Heredity, 69(2): 101-111.

Malacrida AR, Marinoni F, Torti C, Gomulski LM, Sebastiani F, Bonvicini C, Gasperi G, Guglielmino CR 1998. Genetic aspects of the worldwide colonization process of *Ceratitis capitata*. Journal of Heredity, 89(6): 501–507.

Malacrida AR, Gomulski LM, Bonizzoni M, Bertin S, Gasperi G, Guglielmino CR 2007. Globalization and fruitfly invasion and expansion: the medfly paradigm. Genetica, 131(1): 1-9.

Nikolouli K, Augustinos AA, Stathopoulou P, Asimakis E, Mintzas A, Bourtzis K, Tsiamis G 2020. Genetic structure and symbiotic profile of worldwide natural populations of the Mediterranean fruit fly, *Ceratitis capitata*. BMC Genetics, 21(2): 1-13.

Peakall ROD, Smouse PE 2006. GENALEX 6: genetic analysis in Excel. Population genetic software for teaching and research. Molecular Ecology, Notes. 6(1): 288-295.

Rajabiyan M, Shayanmehr M, Sharif MM 2015. The Mediterranean fruit fly (*Ceratitis capitata*) in Iran: genetic diversity and comparison with other countries. Journal of Entomological and Acarological Research, 47(1): 20-25.

Reyes A, Ochando MD 1998a. Genetic differentiation in Spanish populations of *Ceratitis capitata* as revealed by abundant soluble protein analysis. Genetica, 104(1): 59-66.

Reyes A, Ochando MD 2004. Mitochondrial DNA variation in Spanish populations of *Ceratitis capitata* (Wiedemann) (Tephritidae) and the colonization process. The Journal of Applied Entomology, 128(5):358-364.

Rizvi SAH, George J, Reddy GV, Zeng X, Guerrero A 2021. Latest developments in insect sex pheromone research and its application in agricultural pest management. Insects, 12(6): 484.

Thomas MC, Heppner JB, Woodruff RE, Weems HV, Steck GJ, Fasulo TR 2013. Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann) (Insecta: Diptera: Tephritidae).

Vontas J, Hernández-Crespo P, Margaritopoulos JT, Ortego F, Feng HT, Mathiopoulos, KD, Hsu, JC 2011. Insecticide resistance in Tephritid flies. Pesticide Biochemistry and Physiology, 100(3): 199-205.

White IM, Elson-Harris MM 1994. Fruit Flies of Economic Significance. Their identification and bionomics. Wallingford, UK: CAB International

Willi Y, Van Buskirk J, Schmid B, Fischer M 2006. Genetic isolation of fragmented populations is exacerbated by drift and selection. The Journal of Evolutionary Biology, 20:534- 542.

Yeh FC, Yang RC, Boyle T 1999. POPGENE Version 1.32: Microsoft Window-Based Freeware for Population Genetics Analysis. University of Alberta, Edmonton.

Zhou X, Faktor O, Applebaum SW, Coll M 2000. Population structure of the pestiferous moth *Helicoverpa armigera* in the eastern Mediterranean using RAPD analysis. Journal of Heredity, 85:251– 256.

DRAFT

ORAL PRESENTATION

Pleopod Üzerinde İnkübasyona Tabi Tutulan Kerevit (*Astacus leptodactylus* Eschscholtz, 1823) Yumurtalarının Açılma Oranı Üzerine Etkileri

Gülşen UZUN GÖREN[1,*] *(ORCİD 0000-0001-9109-2921)*, Şennan YÜCEL[2] (*ORCİD 0000-0002-0522-9846)*

[1] *Sinop University, Fisheries Faculty, 57000, Sinop, Turkey*

*Sorumlu yazar e-mail:gulsenuzn@hotmail.com

Özet

Bu çalışmada *Astacus leptodactylus*'un üretiminde yumurtalı dişi kerevitlerin yumurtaları açılıncaya kadar tanklarda tutularak yumurtaların açılması ile yavru alımı amaçlanmıştır. Araştırma materyalini oluşturan kerevitler *(Astacus leptodactylus*); Bafra Balık Gölleri'nden pinterlerle avlanmıştır. Avlanan yumurtalı kerevitler Sinop Üniversitesi Su Ürünleri Araştırma ve Uygulama Birimi'ne getirildi. Pleopodlar üzerinde yumurtaların inkübasyonunu sağlamak için yumurtalı kerevitler 96 x 196 x 50 cm dikdörtgen fiberglas tankların içine on litrelik plastik saklama kaplarına bireysel olarak koyuldu. Çalışmada su sıcaklığı 11.74±0.5 ^{o}C, pH 8.08, oksijen ise 5.13 mg/lt de yürütüldü. Çalışmada kerevit yumurtaları günlük olarak kontrol edildi. Yumurtalarda aynı anda kerevitlerin yaşam alanları olarak belirlenen her bir saklama kutusundaki yumurtaların üzerindeki mantarlaşma durumları çıplak gözle gözlendi. Bu gözlem sonucunda her bir yaşam alanındaki kerevitlerde farklı zamanlarda mantarlaşmanın başladığı anlaşıldı. İlk mantarlaşma 10. gününde görüldü. Belli bir peryot takip edilmemekle beraber, mantarlaşma diğer yaşam alanlarındaki kerevitlerde de görüldü. 45 gün içerisinde bütün kerevitlerin yumurtalarını mantar kapladı. Ayrıca bu süreçte mantarlı yumurtalar arasında sağlıklı gözüken yumurtalar da pleopodlar üzerinden düştü. Bu sürecin sonucunda anaçlarda ölmüştür. Gerek mantarlı yumurtalar gerekse pleopodlar üzerinden düşen yumurtalardan yavru elde edilemeyeceğinden dolayı yumurta sayıları ve yumurta çapları ölçülememiştir. Kerevit üretiminde yumurtaların inkübasyonu, üretimin en önemli aşamasıdır. Hedeflenen üretimin elde edilebilmesi için; F1 anaçlarının kontrollü üretim ile elde edilerek sağlıklı olması ve bunların yumurtalarından yavru elde edilmesi üretimin sürekliliği açısından önemli bir tedbir olarak düşünülmelidir.

Anahtar kelimeler: Kerevit, *Astacus leptodactylus*, inkübasyon, açılma oranı

Effects of Crayfish (*Astacus leptodactylus* Eschscholtz, 1823) Eggs Incubated on Pleopod on Hatch Rate

Abstract

In this study, in the production of *Astacus leptodactylus*, the eggs of female crayfish with eggs were kept in tanks until they hatch, and it was aimed to get offspring by opening the eggs. Crayfish (*Astacus leptodactylus*) that make up the research material; Fished with pinters from Bafra Fish Lakes. Crayfish with eggs caught were brought to Sinop University Fisheries Research and Application Unit. To allow incubation of eggs on pleopods, egg-laying crayfish were individually housed in ten-liter plastic storage containers inside 96 x 196 x 50 cm rectangular fiberglass tanks. In the study, the water temperature was 11.74±0.5 ^{o}C, the pH was 8.08, and the oxygen was 5.13 mg/lt. In the study, crayfish eggs were checked daily. At the same time, the fungus on the eggs in each storage box, which was determined as the habitat of the crayfish, was observed with the naked eye. As a result of this observation, it was understood that the fungus started at different times in the crayfish in each habitat. The first mushrooming was seen on the 10th day. Although a certain period was not followed, fungal growth was also seen in crayfish in other habitats. Within 45 days, the eggs of all the crayfish were covered with fungus. In addition, during this process, the eggs that looked healthy among the mushroom eggs were also dropped from the pleopods. As a result of this process, the broodstocks died. Egg numbers and egg diameters could not be measured because offspring could not be obtained from eggs that fell on either mushroom eggs or pleopods. Incubation of eggs in crayfish production is the most important stage of production. Achieving the targeted production; It should be considered as an important precaution in terms of

the continuity of production that F1 broodstock are healthy by means of controlled production and that offspring are obtained from their eggs.

Keywords: Crayfish, *Astacus leptodactylus*, incubation, hatching rate

Giriş

Kerevitler alt tür ile birlikte dünyada 737 türe sahiptir (Crandal ve De Grave, 2017) ve yaklaşık olarak sadece 15 tür ekonomik açıdan önemlidir (Ackerfors, 2000). Dünya genelinde ekoloji ve ekonomi bakımından önemli bir tür olan göl kerevitleri (*Astacus leptodactylus* Eschscholtz, 1823) ülkemizde yaygın bir şekilde iç sulardan elde edilen bir türdür ve aynı zamanda önemli ekonomik değeri olan bir ihraç ürünüdür. (Kozák vd., 2015). Yaşam alanlarının iklim şartlarına göre dönemsel üreme süreleri değişiklik göstermektedir. Üreme sezonu su sıcaklığının düştüğü sonbahar aylarında başlar. Ekim ve kasım ayları (7-12 ^{o}C) su sıcaklığı ile çiftleşmenin gerçekleştiği dönemdir. Bu dönemin 4-6 hafta sonrasında su sıcaklığı 6-11 ^{o}C olmaktadır ve böylece yumurta bırakma zamanı başlamaktadır. Bu aşamadan sonra yumurtalar için kuluçka süresinin kış ve ilkbahar mevsimi boyunca devam ettiği bildirilmiştir. Dişi bireyler ılıman iklim kuşağında 5-6 ay süresince, soğuk iklimler kuşaklarında 6-7 ay ya da daha uzun süre yumurtalarını taşırlar (Mazlum ve Yılmaz, 2012). Yapılmış çalışmalar *A. leptodactylus* dişilerinin çoğunun pleopodlarında 200-400 arası yumurtanın var olduğu bildirilmiştir (Köksal, 1988; Harlıoğlu vd., 2004).
Dünya'da kerevitler ticari ürün olarak 1830'lu yıllarda değerlendirilip ekonomiye kazandırılmıştır. Fakat bu süreç ülkemizde 1970 ve 1985 yılları arasında gelişmeye başlamıştır. Ancak Türkiye'de 1984 yılında Çivril Gölü'nde kerevit vebası (*Aphanomyces astaci*) hastalığı ilk kez gözlenmiş olmasının ardından, hastalık hızla diğer su kaynaklarındaki kerevitlere bulaşmış, yüksek ölümler dolayısıyla stok yoğunluğunda hızlı azalma tespit edilmiştir. Avcılık yoluyla 2022 yılında elde edilen kerevit üretimi 1011 ton olarak gerçekleşmiştir (TÜİK, 2022). Türkiye'de bu türün kültüre alınarak, doğal stok varlığının korunması ve desteklenerek takviye edilmesi ciddiye alınması gereken önemli bir konudur.
Su ürünlerinin tümünde olduğu gibi kerevitlerinde üretim aşamasında en önemli konuların başında yumurtaların inkübasyonu aşaması öne çıkmaktadır. İnkübasyon sürecindeki gidişat üretimin geleceğini etkileyen en önemli faktördür. Bu maksatla yapılan çalışmalarda; araştırma aşamasında kerevitlerin yumurta inkübasyonunda zuger şişeleri, elek sistemleri, tanklar yada annelerinin pleopodlarında yumurtaların açılmasını beklemek gibi bir çok metot kullanılabileceği bildirilmiştir. (Ackefors 1989; Carral vd., 1988; Leonard vd.. 2001). Köksal (1988), *Astacus leptodactylus* üretim aşamasında yumurta taşıyan dişi bireyin yumurtalarında açılma meydana gelinceye kadar havuzlarda kalması yada dişi bireyden ayrılan yumurtaların inkübatörler üzerinde inkübe edilmesi gibi iki türlü metot kullanılabildiği belirtilmiştir. Çalışmamızda ise *Astacus leptodactylus* üretim sürecinde yumurta taşıyan dişi bireylerin yumurtaları açılma süresince tanklarda tutulmuş ve yumurtaların açılması takip edilmiştir.

Materyal Ve Yöntem

Araştırma materyalini oluşturan kerevitler (*Astacus leptodactylus*); Bafra Balık Gölleri'nden Şubat ayında pinterlerle avlanan yumurtalı kerevitler (10 adet) Sinop Üniversitesi'nin Su Ürünleri Araştırma ve Uygulama Birimi'ne getirilmiştir. Pleopodlar üzerinde yumurtaların inkübasyonunu sağlamak için yumurtalı kerevitler; 96 x 196 x 50 cm dikdörtgen fiberglas tankların içine 10 lt'lik plastik saklama kaplarına bireysel olarak koyuldu. Plastik saklama kaplarında kerevitlerin gizlenme yeri olarak plastik boru parçaları konuldu. Çalışmada su kaynağı olarak kuyu suyu kullanıldı.
Her gün su sıcaklıkları, haftada bir kere çözünmüş oksijen miktarları olmak üzere, çalışma süresince, su sıcaklığı 11.74±0.5 ^{o}C, pH 8.08, oksijen ise 5.13 mg/lt olarak ölçülmüştür. Kerevitlerde yemleme günde 2 kez ve ad-libitum olarak yapılmıştır. Yenmeyen yemler ve artıkları günlük olarak sifonlanarak temizlendi.
Araştırmada kerevit üretiminde yavru alınması için yumurtaların açılıncaya kadar tanklarda tutulması hedeflenmiştir.

Bulgular

Çalışmada kerevit yumurtaları günlük olarak kontrol edildi. Yumurtalarda aynı anda kerevitlerin yaşam alanları olarak belirlenen her bir saklama kutusundaki yumurtaların üzerindeki mantarlaşma durumları çıplak gözle gözlendi. Bu gözlem sonucunda her bir yaşam alanındaki kerevitlerde farklı zamanlarda mantarlaşmanın

başladığı anlaşıldı. İlk mantarlaşma 10. gününde görüldü. Belli bir peryotta takip edilmemekle ve yaşam alanları arasında su geçişleri olmamakla beraber mantarlaşma diğer yaşam alanlarındaki kerevitlerde de görüldü. Bir kerevitte yumurtaların üzerinde mantar görüldüğünde hızla diğer yumurtalara da yayıldığı gözlendi. 45 gün içerisinde bütün kerevitlerin bütün yumurtalarını mantar kapladı. Ayrıca bu süreçte mantarlı yumurtalar arasında sağlıklı gözüken yumurtalarda pleopodlar üzerinden düştü. Bu sürecin sonucunda anaçlarda öldü. Gerek mantarlı yumurtalar gerekse pleopodlar üzerinden düşen yumurtalardan yavru elde edilmeye uygun olmadığından, yumurta sayıları ve yumurta çaplarını ölçülmemiştir.

Tartışma Ve Sonuç

Astacidae türlerinin, yumurta inkübe sürecinin çok uzun (7-9 ay) olması, kerevit vebası hastalığının olması ile yaşama oranı (% 0-70) düşüktür. Embriyonik gelişim periyodu boyunca, anaçları etkileyen herhangi bir faktör yavrularda büyük kayıplara yol açabilir. Agresif etkileşimler, el ile müdahale, pleopodlara yumurtanın zayıf olarak yapışması ve yumurtaların pleopodlardan atılması mortalite nedenleri olarak kabul edilmektedir (Carral vd., 1992). Kerevit yumurtalarının doğal koşullar altında %10-20'sinin çıkma vasfına sahip olduğu, başka bir deyişle bir kerevitin bıraktığı 200 adet yumurtadan 10-20 adet arasındaki bir sayıda larvanın geliştiği belirtilmiştir (Alpbaz, 2005).

Başarılı bir yumurtlama süreci yumurtaların döllenmesi ve pleopodlara bağlanması evreleri kerevitlerin hayatta kalma oranlarının yüksek olması için önemli bir süreçtir. Ancak yumurtlamayı takiben uzun inkübasyon periyodu süresince belki de yaygın olarak Saprolegnia ve diğer mantar türlerinin sebep olduğu enfeksiyonlar ya da inkübasyon sürecinde yumurta sapının incelmesi sebebiyle yumurta sayıları çeşitli oranlarda azalmaya devam etmektedir. Bazı dişilerde, yumurtlamayı takiben %40-100 oranlarında yumurta kayıpları meydana gelebilir (Reynolds vd., 1992). Leonard vd. (2001) yapmış oldukları araştırmada benzer sonuç elde ederek; bentik ve süspansiyon olmaksızın inkübe sistemlerde *Cherax destructor* yumurtaları %0 oranında yaşamışlardır.

Yapay inkübasyon sırasında, ölü yumurtalar genellikle çevredeki sağlıklı yumurtalara yayılma kabiliyetine sahip mantarlar tarafından istila edilir. Cansız yumurtaların periyodik olarak uzaklaştırılması yoluyla mantar çoğalması kontrol edilebilse de (Carral vd. 2004; Policar vd., 2006), mantar önleyici tedavilerin uygulanması tavsiye edilir. Fakat çalışmamızda pleopodlar üzerinde yumurtaların açılmasında herhangi bir dezenfeksiyon yöntemi kullanılan inkübasyon tekniği nedeniyle uygulanmaması dezavantaj yaratmaktadır.

Yürüttüğümüz araştırmada mantar ve pleopodlardan yumurtaların düşmesi sonucunda yumurta kayıplarının %100 olduğu tespit edilmiştir. Bu durum anaçların doğal ortamdan alınmış olmasından ve de muhtemelen bu anaçların vebalı olduğu düşünülmektedir. Bu cümleden olarak, yukarıda belirtilen olumsuz sonucun elde edilmemesi için yumurtası alınacak anaçların hastalığa yakalanmamış olması önemli bir tedbir olarak düşünülmektedir. Bunun için ilk tedbir olarak F1 anaçlarının kontrollü üretim ile elde edilerek sağlıklı olması ve bunların yumurtalarından yavru elde edilmesi üretimin sürekliliği açısından önemli bir tedbir olarak düşünülmelidir.

Kaynaklar

Ackefors GEH 1989. Intensification of European Freshwater Crayfish Culture in Europe. Special Session of Crayfish Culture of Aquaculture 89 World Aquaculture Society. Los Angeles, USA, February 13, 29 pp.

Ackefors H 2000. Intensification of European Freshwater Crayfish Culture in Europe (J.V.Huner editör) Freshwater Crayfish Aquaculture in North America, Europe and Australia: Families Astacidae, Cambaridae and Parastacidae. Food Products Press, New York, USA, s 200.

Alpbaz A 2005. Su ürünleri yetiştiriciliği. Alp Yayınları, 548, İzmir.

Carral JM, Pe´rez JR, Celada JD, Sa´ez-Royuela M, Melendre PM, Aguilera A 2004. Effects of dead egg removal frequency on stage 2 juvenile production in artificial incubation of *Austropotamobius pallipes* Lereboullet. Bull Fr Peche Piscicult 372-373: 425-430. doi:10.1051/kmae:2004015.

Carral JM, Celada JD, Gaudisio VR, Temino C, Fernandez R 1988. Artificial ıncubation ımprovement of crayfish eggs (*Pacifastacus leniusculus* Dana) under low temperature during embryonic development, Freshwater Crayfish 7: 230-250.

Carral JM, Celada J, Gonzalez J, Gaudioso VR, Ferniindez R, Lopez-Baisson C 1992. Artificial incubation of crayfish eggs (*Pacifastacus leniusculus* Dana) from early stages of embryonic development. Aquaculture, 105: 261-269. doi: 10.1016/0044-8486(92)90208-3.

Crandall KA, De Grave S 2017. An updated classification of the freshwater crayfishes (Decapoda: Astacidea) of the world, with a complete species list. Journal of Crustacean Biology, 1-39.

Harlıoğlu MM, Barım Ö, Türkgülü İ, Harlıoğlu AG 2004. The potential fecundity of an introduced population of freshwater crayfish, *Astacus leptodactylus* (Eschscholtz, 1823). Aquaculture 230: 189-195.

Kozák P, Füreder L, Kouba A, Reynolds J, Souty-Grosset C 2015. Current conservation strategies for European crayfish. Knowledge Managament Aquatic Ecosystems, 401, 01.

Köksal G 1988. *Astacus leptodactylus* in Europe. In: D. M. Holdich and R. S. Lowery (Editors), Freshwater Crayfish. Biology, Management and Exploitation. Croom Helm, London, pp. 365-400.

Leonard BV, Lennard WA, Kildea D 2001. A method for testing the effectiveness of artificial incubation of eggs maternal brooding in the freshwater crayfish *Cherax destructor* (Decapoda: Parastacidae). Aquaculture, 195: 299-309. doi: 10.1016/S0044-8486(00)00561-5.

Mazlum Y, Yılmaz E 2012. Kerevitlerin Biyolojisi ve Yetiştiriciliği. Mustafa Kemal Üniversitesi Yayınları: 34, Hatay, 120 p.

Policar T, Koza´k P, Martı´n J 2006. Effects of egg bath and daily removal of dead eggs on hatching success and production of stage 2 juveniles during artificial incubation in noble crayfish (*Astacus astacus* L.). Bull Fr Peˆche Piscic 380-381: 1197-1206.

Reynolds JD, Celada JD, Carral JM, Matthews MA 1992. Reproduction of astacid crayfish in captivity-current developments and implications for culture, with special reference to Ireland and Spain. Invertebrate Reproduction Development, 22 (1-3): 253-265. doi:10.1080/07924259.1992.9672278.

TÜİK 2022. Türkiye İstatistik Kurumu. https://www.tuik.gov.tr. (Erişim tarihi: 10.06.2022).

DRAFT

ORAL PRESENTATION

Peynir ve peynir teknolojisinde yeni uygulamalar

Fadime SEYREKOĞLU* *(ORCID: https://orcid.org/ 0000-0001-9787-4115)*

*Amasya Üniversitesi, Suluova Meslek Yüksekokulu, Gıda İşleme Bölümü, Amasya, Türkiye

*Sorumlu yazar e-mail: fadime.tokatli@amasya.edu.tr

Özet

Süt ve ürünleri içerdiği değerli besin ögeleri sayesinde toplumun tüketmesi gereken besin grupları arasında yer alır. Peynir özellikle kahvaltılarda sıklıkla tüketilen süt ürünleri içerisindedir. Ülkemizde ve dünyada çok çeşitli peynir türleri üretilmektedir. Farklı hammaddeler, farklı üretim metodları birçok peynir türünün ortaya çıkmasını sağlamaktadır. Özellikle geleneksel yöntemler üretim esnasında bazı kayıpların artmasına verimde, kalitede kayıplara sebep olmaktadır. Süte uygulanan veya peynir üretim prosesinde uygulanan yeni teknolojiler kayıpları azaltıp kalite ve mikrobiyal güvenilirliğin artmasını sağlamaktadır. Son yıllarda imitasyon peynirlerin üretimi ile ilgili çalışmalar hız kazanmaktadır. Bitkisel kaynakların kullanımı sayesinde daha ekonomik, fonksiyonel özellikler gösteren peynirlerin üretimi bu teknoloji ile sağlanmaktadır. Ayrıca üretim prosesinin daha hızlı olması istenen biyoaktif ve bitkisel bileşenlerin ilave edilebilmesi bu peynirlerin özel tüketici gruplarına hitap etmesi imitasyon peynirlerin avantajları arasında yer almaktadır. Proses esnasında uygulanan ısıl işlem tat- aromada bazı istenmeyen değişikliklere sebep olmaktadır. Isısal olmayan yöntemler arasında yer alan hidrostatik basınç uygulamaları, basınç uygulaması ile gıda güvenliğini sağlayarak raf ömrünü arttırır. Böylece verim ve raf ömründe artış olgunlaşma süresinde ise kısalma sağlar. Yeni teknolojilerden biri olan membran teknolojisi de peynir altı suyu proteinleri konsantresi, süt proteinlerinin fraksiyonlanması, sütte mikrobiyal yükün azaltılmasını sağlar. Süte ısıl işlem uygulanmadığı için sütte istenmeyen değişiklikler meydana gelmez. Sonuçta yüksek kuru madde oranına sahip peynir üretilmesi sağlanır. Ultrasound uygulamaları da ısıl olmayan inovatif gıda muhafaza metodları arasında yer almaktadır. Özellikle süt ve ürünlerinde mikrobiyal yükün azaltılmasında oldukça etkili bir metoddur. Bahsedilen metodlar yeni peynir türlerinin üretilmesini sağlarken verimde, kalitede de artış sağlar. Yapılan çalışmalar arttırılıp, endüstriye aktarılabilme olanakları değerlendirilmelidir.

Anahtar Kelimeler: Peynir, imitasyon, ultrasound, membran, hidrostatik basınç.

New applications in cheese and cheese technology

Abstract

Milk and products contain valuable nutrients, so among the food groups that the society should consume. Cheese is among the dairy products that are frequently consumed especially for breakfast. Various types of cheese are produced in our country and in the world. Different raw materials and different production methods lead to the emergence of many types of cheese. In particular, traditional methods cause some losses to increase during production, resulting in losses in efficiency and quality. New technologies applied to milk or applied in cheese production process reduce losses and increase quality and microbial reliability. In recent years, studies on the production of imitation cheeses have gained momentum. Owing to the use of vegetable sources, the production of cheese with more economical and functional properties is provided by this technology. In addition, the addition of bioactive and herbal components, which are desired to make the production process faster, is among the advantages of imitation cheeses, making these cheeses appealing to special consumer groups. The heat treatment applied during the process causes some undesirable changes in taste and aroma. Hydrostatic pressure applications, which are among the non-thermal methods, increase the shelf life by providing food safety with pressure application. Thus, it provides an increase in yield and shelf life and a shortening of ripening time. Membrane technology, which is one of the new technologies, provides whey protein concentrate, fractionation of milk proteins and reduction of microbial load in milk. Since no heat treatment is applied to the milk, undesirable changes do not occur in the milk. As a result, cheese with high dry matter content is produced. Ultrasound applications are also among the non-thermal innovative food preservation methods. It is a very effective method in reducing the microorganisms, especially in milk and its

products. While the mentioned methods enable the production of new cheese types, they also increase the yield and quality. The possibilities of increasing the studies and transferring them to the industry should be evaluated.

Keywords: Cheese, imitation, ultrasound, membrane, hydrostatic pressure

GİRİŞ

Dünyada ve ülkemizde süt ve ürünleri tüketimi her geçen yıl artarak devam etmektedir. Sağlık üzerindeki etkileri süt ve ürünlerinin üretimini ve çeşitliliğini de etkilemektedir. Süt ürünleri grubunda yer alan ve ülkemizde kahvaltılarımızda vazgeçilmez olan peynir bu sektörde önemli besin grupları arasındadır. Peynir çeşitliliğine baktığımızda her yörede üretilen peynir çeşitleri bulunmakla birlikte aynı yörede farklı özelliklere sahip olan peynirlerde mevcuttur. Peynir üretiminde kullanılan hammadde koyun, keçi, inek sütü yada bu sütlerin karışımı olabilir. Çok farklı üretim metotları, kullanılan farklı starter kültürler ve olgunlaşma aşamasındaki farklılıklar son ürünün fiziksel, kimyasal, mikrobiyolojik ve duyusal özelliklerini etkilemektedir. Peynir üretiminde farklılıklar olmasına rağmen özellikle beyaz peynir üretiminde kayıplar ve verimde azalmalar görülebilir. Ayrıca geleneksel yöntemlerle üretilen peynirlerin raf ömrü kısıtlı olup kısa sürede bozulmalar görülebilir. Mevcut teknolojilere ilave yapılan bazı uygulamalar peynir üretiminde oluşan kayıpları azaltıp, verimi arttırıp daha sağlıklı ürünlerin üretilmesini sağlamaktadır. Bu aşamada kullanılan hammadde olan süte bazı işlemler uygulanmakta, yada proses esnasında farklı teknikler geliştirilmekte ve yeni teknolojilerin kullanımları değerlendirilerek endüstriye aktarımı amaçlanmaktadır.
Son üründe istenen özellikleri sağlamak için mevcut ingrediyentlere farklı bileşenler eklenmekte sağlık açısından daha yararlı peynirler üretilmektedir. Bu araştırmada da dünyada ve ülkemizde uygulanan peynir prosesleri ve imitasyon peynir üretimine değinilecektir. Her geçen gün peynir ve çeşitleri ile ilgili literatürde yapılan çalışmaların sayısı artmaktadır. Yapılan bu çalışmalar ve yeni teknolojiler ile gerçekleştirilen peynir üretimi, kişi başına düşen peynir tüketimini arttırarak peynirdeki kayıpları azaltacak ve peynir çeşitliliğine katkı sağlayacaktır.

PEYNİR

Günümüzde insanlar sağlıklı besinlere yönelmekte ve temel ihtiyaçlarını bu besin gruplarından karşılamaktadır. Süt ve ürünleri bu anlamda oldukça değerli besin gruplarıdır ve yapılarında vitamin (A, B2, B6 ve B12), protein ve mineralleri muhteva ederler. Sağlıklı bir diyette süt ve ürünleri mutlaka bulunmalıdır. Ülkemizde süt ve ürünleri insanlar tarafından sevilerek tüketilen besin gruplarının başında yer almaktadır. Ülkemizde süt üretimi hayvan ağırlığı ve laktasyon artışı ile artış göstermektedir (Çınar, 2018). Süt ve ürünleri içerisinde peynir tüketimi yılda ortalama %1.6 artış göstererek ilk sırada yer almaktadır. Yaşam standartlarının değişmesi, tüketici talepleri peynir çeşitlerinin artmasına daha fonksiyonel özellikler gösteren yeni peynir türlerinin ortaya çıkmasına ve tüm bunlar peynirle ilgili inovasyon çalışmalarının artmasına sebep olmaktadır. Doğal peynir çeşitleri yanında faklı ikameler eklenmiş peynirler, çeşitli teknolojilerle zenginleştirilmiş peynirler ve son zamanlarda ortaya çıkan imitasyon peynirler pazarda yer almaktadır (Ersan ve Gizem, 2020).

Dünyada peynir üretimi %1,5 oranında artış göstererek 2019 yılında 24 milyon tona ulaşmıştır. Dünyada peynir üretiminde ilk sırayı 5,9 milyon ton üretimle ABD alırken, önemli peynir üreticilerinden olan Brezilya, Rusya ve Türkiye de ise peynir üretimi 600-800 bin tona ulaşmıştır. Ülkemizde peynir üretimi ise 2019 yılında %6,5 azalmıştır ve 707 bin tondur. Toplam arzda%6,6 artış görülmüştür ve 714 bin tona ulaşmıştır. Aynı yıl görülen peynir ihracatı 50 bin tona ulaşmıştır. Dünyada üretilen süt özellikle peynir, tereyağı ve peynir altı suyu tozuna işlenmektedir. Dünya genelinde peynir tüketimi her geçen gün artış göstermektedir. AB, ABD, Yeni Zelanda ve Kanada da peynir tüketimi oldukça fazladır (Tepge, 2020). Ülkemizde süt ürünleri arasında en çok tercih edilen ürünlerden biri peynirdir. Ülkemizde üretimi ve tüketimi en fazla olan ve en büyük pazara sahip olan peynir beyaz peynirdir. USK 2019 yılı verilerine göre ülkemizdeki kişi başı yıllık peynir tüketim miktarı 17,5 kg'dır (USK, 2020).

Peynir çeşitli oranlarda yağ içeren sütün proteolitik enzimler yada çeşitli asitlerle pıhtılaştırılması sonrasında peynir altı suyunun ayrılması ve oluşan pıhtının şekillendirilmesi sonucunda elde edilen bir süt ürünüdür (Üçüncü, 2008). Protein ve yağ bakımından oldukça zengin olan peynir çok farklı metodlarla üretilebilir. Taze ve tuzsuz olabileceği gibi tuzlama ve olgunlaştırma işlemleri ile peynire çok çeşitli tat-aroma ve görünümde kazandırılabilir. Tuzlama işlemi peynirde ortaya çıkan proteolizi ve bu durumda yapı-kıvam, tat-aroma özelliklerini etkilemektedir (Çavuş, 2020).

Sütten elde edilen ve oldukça lezzetli olan peynir asırlardır yapılmakta ve her geçen gün çeşitliliği artış göstermektedir. Yapıldığı bölgeye kullanılan hammaddeye ve üretim metoduna göre farklılıklar göstermektedir. Ülkemizde farklı bölgelerde farklı peynir çeşitleri bulunmaktadır. Sıcaklık, yer, kullanılan hammaddeler uygulanan farklı üretim prosesleri farklı peynirlerin ortaya çıkmasını sağlamaktadır. Peynirlerin üretiminde farklılıklar bulunmasına rağmen peynir yapımında temel aşamalar vardır. Proses kısaca şu şekildedir; sütün pıhtılaştırılması, pıhtının kesilmesi ve peynir altı suyunun ayrılması, pıhtının toplanması, birleştirilmesi ve şekillendirilmesi olarak belirtilebilir. Bunlara ek olarak tuzlama, olgunlaştırma, fermentasyon, haşlama gibi farklı işlem basamakları da peynirin çeşidine göre proses aşamaları olarak uygulanmaktadır.

2. PEYNİR TEKNOLOJİSİNDEKİ YENİ GELİŞMELER

2. 1. İmitasyon Peynir Üretimi

Değişen yaşam şartları, fonksiyonel özelliklere sahip olan yeni peynir çeşitleriyle ilgili yapılan inovasyon çalışmaları, peynir kullanımının giderek artış göstermesi peynir endüstrisinde yeniliklere sebep olmaktadır. Bu alanda doğal peynirlere yakın özelliklere sahip olan peynir benzeri ürünlerin oluşumu karşımıza çıkmaktadır. Ulusal ve uluslararası pazarda üretimi ve tüketimi giderek artış gösteren; düşük maliyetli ve farklı ingrediyentlere sahip olan imitasyon peynirler önem kazanmaktadır. İşlenmiş peynir grubunda yer alan imitasyon peynirler peynir analoğu, taklit peynir ve ikame peynir olarak da isimlendirilirler (Ersan ve Gizem, 2020). Süt yağının tamamının veya bir kısmının diğer yağlar ile yer değiştirilerek oluşturulan peynir benzeri ürünler Kodeks Alimentarius'a göre imitasyon peynirler olarak tanımlanmaktadır (Kodeks Alimentarius, 2016). FAO ise imitasyon peynirleri besin değeri daha düşük olan doğal peynirlere benzeyen peynir benzeri ürün ve peynir ikameleri olarak 2 grupta incelemektedir (FAO, 2017). Ülkemizde ise bu peynirlerle ilgili herhangi bir yasal tanımlama bulunmamaktadır. Üretimlerinde; homojen bir matriks içinde yer alan su, proteinler, tuzlar, sıvı yada katı formda bulunan yağlar, hidrokolloidler, asitleştiriciler, koruyucu ajanlar ve diğer katkı maddeleri kullanılmaktadır. Üretimlerinde kullanılan bileşenlere göre; süt bazlı, kısmi süt bazlı ve süt bazlı olmayan imitasyon peynirler olarak sınıflandırılmaktadır (Ersan ve Gizem, 2020).

Krem peynir, kaşar ve mozzarella'ya benzeyen peynir türleri kullanılarak imitasyon peynirler üretilebilmektedir. Üretimde kullanılan bileşenler, miktarları, proseste kullanılma sıraları, proses ve son ürün özelliklerine göre farklılıklar göstermektedir. Öncelikle ürün formülasyonu oluşturulur, sonra bu bileşenler karıştırılır ve ısıl işlem uygulanır, sıcak olarak paketleme ve soğutma prosesleri imitasyon peynir üretiminde kullanılan genel prosestir (Ersan ve Gizem, 2020). Toz şeklinde bulunan tuz, kazein, kazeinatlar ve hidrokolloidler suyla karıştırılıp 50 ^{o}C'ye kadar ısıtılmakta ve karıştırılmaktadır. Üretimde yer alan bitkisel yağlar koyulduktan sonra 85°C'ye kadar ısıtma ve karıştırma yapılmaktadır. En son asitleştiriciler ve tat- aroma maddeleri ilave edilip soğutma aşamasına geçilmektedir. Uygulanan proses parametreleri ürünün özelliklerini ve muhafaza süresini etkilemektedir. Ayrıca kullanılan peynir bileşenlerinin türü, kimyasal ve fonksiyonel özellikleri, aralarındaki interaksiyonlar son ürünün reolojisini ve bütün özelliklerini belirleyen faktörlerdendir (Ersan ve Gizem, 2020).

İmitasyon peynirler
Tüketiciler tarafından satın alımlarda,

Dondurulmuş ürünlerde,

Gastronomi alanında çeşitli gıdaların hazırlanmasında,

Cips, çerez gibi atıştırmalık ürünlerin hazırlanmasında kullanılmaktadır (Ersan ve Gizem, 2020).

Bitkisel yağların süt yağı yerine kullanılması, protein olarak da hidrokolloidlerin yada çeşitli nişastaların kullanılması üretim maliyetlerini düşürerek daha uygun fiyatlı ürünlerin ortaya çıkmasını sağlamaktadır. Ayrıca kullanılan alet- ekipmanın az olması, iş gücünün az olması, üretim prosesinin diğer peynirlere göre kısa olması ve olgunlaşma aşamasının olmaması da maliyeti düşüren diğer faktörler arasındadır. Soya, hurma ve hindistan cevizi yağı gibi bitkisel ürünler bu alanda değerlendirilerek laktozsuz, düşük kalorili özel peynirler üretilebilir (Ersan ve Gizem, 2020). Hem bitkisel kaynaklar değerlendirilirken hem de özel diyette olan insanların gereksinimleri imitasyon peynirler ile karşılanabilir. Hastalıkların önlenmesi ve tedavi edilmesinde kullanılan fonksiyonel zenginleştirici ajanlar sayesinde çok çeşitli ürünler üretilerek insan sağlığına ve topluma

faydalı olan imitasyon peynirler üretilebilir. Örneğin; düşük fenialanin ve düşük protein ile beslenmeleri gereken fenilketonuri hastaları bileşimleri farklılaştırılarak oluşan imitasyon peynirleri tüketebilir (Ersan ve Gizem, 2020). Farklı teknikler ile imitasyon ve doğal peynirlerin ayrımı yapılmaktadır. Örneğin lizinoalanin analizi ile imitasyon peynirlerin süt proteini bulundurup bulundurmadığı kontrol edilebilir. Ayrıca taramalı elektron mikroskobu ile lipidlerin durumu tespit edilir. İmitasyon peynirlerde lipidlerde topaklanma, doğal peynirde ise düzgün yağ kürecikleri oluşmaktadır. Yine peynirde bulunan bitki pigmentleri ve fitosteroller için kromotografik yöntemler kullanılarak ayrım yapılabilir (Ersan ve Gizem, 2020).

2.2. Peynir Üretiminde Yüksek Hidrostatik Basınç Uygulamaları

Süt ürünlerinin üretiminde tat-aromada oluşan olumsuzlukları ve ısıl işlemlerden kaynaklanan diğer olumsuzlukları gidermek için ısısal olmayan yöntemler üzerinde durulmaktadır. Bu uygulamaların başında yüksek hidrostatik basınç uygulaması gelmektedir (Çınar, 2018). Katı ve sıvı gıdalara 100 ile 1000 MPa arasında basınç uygulayarak gıdayı güvenli hale getirip raf ömrünü arttıran uygulamalara yüksek hidrostatik basınç uygulamaları denir. Peynir üretiminde hammadde olarak kullanılan süt çevresel etkenlerden, sıcaklık ve pH'dan etkilendiği için bazı işlemlerden geçirilmesi gerekmektedir. Bu uygulamanın peynir teknolojisinde raf ömrünü arttırma, verimi arttırma oluşturduğu yağ kristalleri sayesinde olgunlaştırmayı hızlandırma gibi avantajları mevcuttur (Çınar, 2018).

Yeni ısısal olmayan teknolojiler gıdanın tekstür, yapı- kıvam, lezzet, tat-aroma, renk, besin değeri gibi özellikleri üzerinde sıcaklığın sebep olduğu olumsuz değişiklikleri önleyerek oda sıcaklığında ve benzer sıcaklıkta mikroorganizmaları etkisiz hale getirmektir (Munoz-Cuevas ve ark., 2013; Barba ve ark., 2015).

Gıdanın boyutu, yapısı, şekli fark etmeksizin her noktasında aynı etkiyi gösteren bu uygulamanın ısıl işleme göre daha kısa sürmesi yöntemin avantajları arasındadır (Sezer ve İnanç, 2013). Isıl işlem uygulamaları ile kombine edilerek uygulanan yöntem sterilizasyon ile aynı etkileri göstermektedir. Isıl işleme göre mikroorganizmaları daha düşük sıcaklıklarda inaktive ederek gıdanın fizikokimyasal özelliklerinde daha iyi koruma sağlar (Heinz ve Buckow, 2009; Devi ve ark., 2015). İlk olarak süt ve ürünlerinde uygulanan yöntemle ilgili çalışmalar artmaktadır. Soğuk sterilizasyon olarak da isimlendirilen bu yöntem 400-600 MPa basınç uygulaması ile ortamdaki zararlı bakterileri yok etmektedir (Sfakianakis ve Tzia, 2014). Yöntemin temel prensibi kapalı bir ortamda sıvıya bir basınç uygulanması ve sıvının her yerine homojen olarak iletilmesine dayalı pascal prensibine dayanır. Ambalajlı yada ambalajsız katı veya sıvı gıda sistemin basınç ünitesine yerleştirilir ve genel olarak su (yada hidrokarbonlar, hidrolik yağlar) yardımıyla basınç uygulanır (Oğuzhan, 2013).

Laboratuvar ölçekli yada az miktarlardaki üretimde piston vasıtasıyla basınç iletilirken, büyük ölçekli endüstriyel üretimlerde ise pompa basınç iletimini sağlamaktadır (Oğuzhan, 2013). Ayarlanan sıcaklık, basınç ve süre sayesinde istediğimiz özellikteki gıda maddelerini bu sistemle üretebiliriz. Elektrik enerjisi kullanıldığı için atık oluşmamaktadır (Sayın ve Tamer, 2014; Bruschi ve ark., 2017). Kurulum ve yatırım maliyetlerinin yüksek olması, dilimli veya tüm halde olan gıdalarda istenmeyen tekstür değişimleri oluşturabilmesi ve enzimlerin bazılarının basınca karşı direnç göstermesi sistemin dezavantajları arasında yer almaktadır (Oğuzhan, 2013). Isıl işlem protein moleküllerinde bulunan kovalent olmayan bağları parçalamakta molekül içi ve moleküller arasında farklılıklara sebep olmaktadır (Devi ve ark., 2013). Yüksek basınç uygulaması ise kovalent bağları etkilememekte ve proteinlerin birincil yapılarında değişiklik olmamaktadır (Sezer ve İnanç, 2013).

Koyun sütüyle yapılan peynirlere 200-500MPa ve 12°C sıcaklıkta 10 dakika boyunca yüksek hidrostatik basınç uygulanmış ve olgunlaşma sonrasında kontrol grubu ile kıyaslama yapıldığında kontrol ile benzerlikler görülürken mikrobiyal yük ve enzim aktivitelerinde farklılıklar görülmüştür (Martinez-Rodriguez ve ark., 2012). Yapılan bir çalışmada peynir üretiminde farklı sütler; çiğ süt, pastörize süt ve 15 dakika 500 MPa basınç uygulanan süt kullanılmıştır. Peynirlerin lipoliz miktarlarına bakıldığında, yüksek basınç uygulanan süt ve çiğ sütten elde edilen peynirlerde benzer sonuçlar elde edilirken pastörize sütten elde edilenlerde düşük çıkmıştır (Sezer ve İnanç, 2013).

Geleneksel (%5.3), ve farklı miktarlarda tuz içeren (%2.5,%1.9, %0.2) çedar peynirlerinde 405MPa hidrostatik basınç uygulanmıştır, mikrobiyolojik yüklerinde 1, 2.5, 3, 4 log azalma gözlenirken kontrol grubuyla duyusal açıdan benzer sonuçlar elde edilmiştir (Ozturk ve ark., 2013). Yüksek hidrostatik basınç uygulamalarının mikroorganizmalar üzerindeki etkileri ile ilgili çalışmalar giderek artış göstermektedir. *Bacillus cereus* sporları üzerindeki etkisini gözlemlemek için farklı basınç uygulamaları (200, 400, 600MPa) ve ısıl işlem (70°C) uygulaması yapılmıştır (Silva, 2015). Uygulanan basınç miktarındaki artış ile mikrobiyal yük azalmıştır. 600 MPa basınç uygulaması 40 dakika süre ile uygulandığında bütün mikroorganizmaların inaktif olduğu tespit

edilmiştir. Sıcaklık uygulaması 70 ºC'ye ulaştığında ise 3.5 log seviyesinde azalma tespit edilmiştir. Uygulanan ısıl işlem daha çok enerji tüketmekte iken yüksek hidrostatik basınç uygulamasının *Bacillus cereus* ve sporlarını yok etmede daha çok avantaja sahip olduğu sonucuna varılmıştır (Silva, 2015).
Peynir üretiminde uygulanan bu yöntem sayesinde verim artmakta, olgunlaşması esnasında gerçekleşen proteolizde biyokimyasal reaksiyonları etkilemekte ve olgunlaşma süresi azalmaktadır (Liepa ve ark., 2016). Tüketiciler az işlenmiş, fizikokimyasal ve duyusal açıdan güvenli gıdalara yönelmektedir. Yüksek hidrosatik basınç uygulamaları tüketicilerin bu isteğine cevap verecek özellikteki sağlıklı ve mikrobiyal anlamda güvenilir gıda üretimine olanak tanımaktadır. Özellikle süt ve ürünlerinden peynir teknolojisinde kullanımı istenmeyen kayıpları önleme, mikrobiyolojik açıdan güvenli hale getirme, olgunlaşma süresini kısaltma gibi avantajları yapılan çalışmalarla kanıtlanmıştır (Çınar, 2018).

2.3. Peynir Teknolojisinde Membran Uygulamaları

Süt endüstrisinde membran teknolojisi uygulamaları mikrofiltrasyon, ultrafiltrasyon (UF), nanofiltrasyon ve tersozmoz olarak ayrılacak olan bileşenin molekül ağırlığı ve boyutuna bağlı olarak değişkenlik göstermektedir. Membran teknolojisi ile; peynir altı suyu proteinleri konsantrasyonu, süt proteinlerinin fraksiyonlanması, süt yağının fraksiyonlanması, peynir yapımında kullanılacak olan sütten bakteri sporlarının ve mikroorganizmaların uzaklaştırılması, yüksek kurumadde oranına sahip olan peynir üretiminin gerçekleştirilmesi sağlanır. Peynir üretiminde kullanılacak olan süte ısıl işlem uygulanmadığı için sütün yapısında ve bileşenlerinde herhangi bir değişiklik olmaz ve direk üretimde kullanılabilir (Henning, 2006).
Endüstriyel anlamda ultrafiltrasyon uygulamaları peynir yapımında kullanılacak sütte, peynir altı suyundan ve sütten bakterilerin uzaklaştırılmasında, peynir altı suyu ve süt proteini konsantresi ve izolatında, salamura arıtmada kullanılabilir ve daha az enerji, yer ve sermayeye ihtiyaç duyulur (Henning, 2006).
İstenilen maddenin geçişini sağlamak için ayırma ve taşınmanın yapıldığı engeller sistemine membranlar denilmektedir. Genel prensibi membranın yüzeyinde olan çözeltinin yapısında bulunan bileşenlerin özelliklerine göre ayrılması işlemine dayanmaktadır (De Morais Countinho ve ark., 2009). Membran kısmından geçip süzülen kısıma filtrat çözelti (permeat), geçemeyip kalan kısıma ise derişik çözelti (retentat) adı verilmektedir. Membranın fiziksel, kimyasal yapısı ayırma prosesinde etkilidir. Ayrıca çözeltinin derişimi, uygulanan basınç, elektriksel potansiyel ve sıcaklık farkı da ayırma işlemindeki itici güç üzerinde etkilidir (Henning, 2006).
Peynir üretiminde ana basamaklardan biri kazein misellerinin pıhtılaşmasıdır. Enzimler kullanılarak sınırlı proteoliz, starter kültür veya asit ilavesi ile asitleştirme ve sıcaklık uygulamaları yada bu metodların kombinasyonu ile peynir oluşumu için gerekli olan pıhtılaşma yapılır. Bu pıhtılaşma zayıf jel kütlesinin kesme, pişirme, haşlama gibi çeşitli metodlarla taze peynir formundan istenilen forma dönüştürülmesini sağlar (Henning, 2006). Uygulanan metodlar peynir çeşidine ve istenilen fonksiyonel özelliklere bağlıdır. Peynirlerin türleri ve üretim metodları, sütün bileşimi, oranı ve asitlik gelişimi, nem içeriği pıhtı oluşumu ve olgunlaşma şartlarına bağlıdır (Henning, 2006). UF gibi yenilikçi teknolojiler peynir altı suyu proteinlerinin geri kazanılması yoluyla verimi arttırmaktadır. Peynir altı suyu proteinlerinin geri kazanılması yoluyla verimi arttırmaktadır. Peynir yapımında kullanılan ultrafiltrasyon 3'e ayrılır (Henning, 2006).
*Düşük konsantrasyon faktörü

*Orta konsantrasyon faktörü

*Yüksek konsantrasyon faktörü
Ancak bazı peynir türlerinde kesme işlemi zordur ve üretimde kayıplar artmaktadır. Membran teknolojisilor-feta gibi taze peynirler ve olgunlaştırılmış peynirlerde kayıpları azaltıp, verimi arttırarak başarıyla uygulanabilir (Henning, 2006).

2.4.Peynir Teknolojisinde Ultrasound Uygulamaları

Ultrasound uygulamaları gıdalarda kalitenin ve doğallığın korunduğu ısıl olmayan inovatif gıda muhafaza metodlarından biridir. Kurutma teknolojisi, ekstraksiyon uygulamaları, mikroorganizmaların ve enzimlerin inaktivasyonu gibi alanlarda kullanılmaktadır. Yapılan çalışmalar yöntemin basınçla veya ısıl işlemle birlikte uygulandığında mikroorganizmaların inaktivasyonunda artış olduğunu tespit etmiştir (Tokuşoğlu, 2013).
Yöntemin çalışma prensibi ses enerjisinin dalgalar oluşturması sonucu oluşan hareketin dalgalar oluşturması ve ortamda bulunan materyaller üzerinde sıkışma ve gevşeme meydana getirmesidir. Mekanik titreşim, akustik dalgalanma ve kavitasyon oluşumu etki mekanizmalarındandır (Rastogi, 2011). Ultrasound uygulamaları düşük güçlü- yüksek frekanslı ve yüksek güçlü düşük frekanslı olarak 2 şekilde uygulanmaktadır. Düşük güçlü- yüksek frekanslı ultrasound uygulamaları 100 kHz frekanslarda ve 10 W/cm^2'nin altındaki enerjilerde, yüksek güçlü düşük frekanslı ultrasound uygulamaları ise 20- 100 kHz frekanslarında ve 10-1000 W/cm^2'nin

enerjilerinde uygulanmaktadır. Düşük güçlü- yüksek frekanslı ultrasound uygulamaları gıda maddelerinde önemli fizikokimyasal değişikliklere sebep olmamakta ve oldukça hızlı, ucuz ve basit bir metoddur (Rastogi, 2011). Yüksek güçlü düşük frekanslı ultrasound uygulamaları ise gıda maddelerinde biyokimyasal ve fizikokimyasal özelliklerde modifikasyonlara sebep olmakta ve gıda sistemlerinin kalitesinde artış sağlamaktadır.

Sıcaklık, basınç ve uygulama süresi işlem parametreleridir. Süt endüstrisinde düşük frekanslı ultrasound işlemleri tercih edilmektedir (Türkmen, 2012). Süt teknolojisinde ultrasound uygulamaları ile ilgili yapılan çalışmalar da giderek artış göstermektedir. Çeşitli sütlere ultrasound uygulamasının (24 kHz frekansta, 200 W) yapıldığı çalışmada (çiğ süt, düşük sıcaklıkta kısa süre ısıl işlem uygulanan süt, homojenize pastörize süt) toplam canlı bakteri ve psikrotrof suşlar araştırılmıştır. Uygulanan işlem süresi arttıkça bakteri sayısında azalma ve raf ömründe artış görülmüştür. Mikrobiyal yükün azaltılması sağlanırken tat- aromada yanık ve yabancı tat oluşumu gözlemlenmiştir (Chouliara, 2010).

Çiğ sütün bileşimi, oksidasyonu ve peynir teknolojisindeki özellikleri üzerinde ultrasound uygulamalarının etkisinin incelendiği çalışmada ortaya çıkan kötü tat- aromayı azaltmak için HCl ve CO_2 ilavesi yapılmıştır. Uygulanan yöntem ile somatik hücre sayısında azalma, serbest yağ asidi miktarında artma, oksidasyon ve pH da artma gözlemlenmiştir (Marchesini, 2012). Duyusal analizlerde ise yabancı- yanık tat oluşumu görülürken ilave edilen CO_2 ile yabancı tat ve oksidasyon ürünlerinde azalma, ekşi tatta artış görülmüştür. Çalışmada koagülasyon artışı ile verim artışı tespit edilmiştir. Ultrasound uygulamalarında CO_2 kullanımının süt ve ürünlerinde kötü tat gelişimini engelleme, proteolitik aktiviteyi düşürme, oksidasyonu azaltmada etkili olduğu tespit edilmiştir (Marchesini, 2012). Latin Amerika ve Amerika'nın bazı bölgelerinde termosonikasyon uygulanan süt ile üretilen Queso Fresco peynirinde ultrasound uygulamanın etkisi araştırılmıştır. Geleneksel olarak elde yapılan bir peynir çeşidi olduğu için verimi düşük, mikrobiyal yükü fazla ve raf ömrü düşüktür. Uygulanan sonikasyon işlemi sayesinde pıhtılaşma süresi kısalmış, verimde yaklaşık %20 artış görülmüştür. Peynirin parlaklığı, mikroyapısı ve tekstürel özelliklerinde gelişme tespit edilmiştir. Üretilen peynirlerin kalitesi ve raf ömrü önemli ölçüde artmıştır (Bermudez, 2010).

Ultrasound işlemi ısıl işlemle birlikte kullanıldığı zaman ortaya çıkan termosonikasyon, basınç ile uygulandığı zaman manosonikasyon, ultrasound işlemi, ısıl işlem ve basınç birlikte kullanıldığı zaman manotermosonikasyon uygulamaları da gıda alanında ve süt ve ürünlerinde kullanılabilir (Demirdöven, 2009).

Peynir teknolojisinde her geçen gün değişen teknolojiler ve uygulamalar vardır. Belirtilen bu teknolojilerden farklı olarak yapılan bazı uygulamalarda mevcuttur. Peynirde olgunlaştırmayı hızlandırmak için; kullanılan sıcaklığın yükseltilmesi, enzim ilave edilmesi, peynir bulamacı kullanımı, kullanılan starter kültürlerde uygulanan modifikasyonlar (yardımcı kültür, zayıflatılmış kültür ve mutant kültür) gibi uygulamalarda kombinasyon halinde yada tek başına kullanılmaktadır (Doğan ve ark., 2022).

Peynir teknolojisinde modifiye atmosfer paketleme yöntemi kullanarak kimyasal kullanmadan mikrobiyolojik olarak ürün güvenli hale getirilip raf ömrü arttırılmaktadır (Doğan ve ark., 2022). Peyniri biyoaktif bileşenlerce zenginleştirmek için peynire hardal tohumu özü, kırmızı biber, çeşitli mineraller ve bazı probiyotik kültürler eklenmiştir. Bazı peynir çeşitlerinde ambalaj malzemeleri farklılaştırılarak ürün özellikleri ve raf ömrü arttırılmaya çalışılmıştır. Kaşar peynirinde ambalaj malzemesi olarak balmumu ve yenilebilir filmlerin kullanımı (Yılmaz, 2011)., gouda ve kuark peynirinde nanokompozit film ve gümüş nanopartiküllerin kullanımı gibi çalışmalar bu konuda literatüre ışık tutmaktadır (Pluta-Kubica, 2021).

SONUÇ

Dünyada ve ülkemizde peynir üretim sürecinde karşılaşılan problemler, yaşanılan zorluklar ve kayıplar yapılan çalışmalarla belirlenip geliştirilen teknolojilerle bu sorunlara çözümler üretilmelidir. Ekonomik olarak peynirden daha uygun maliyetli olan imitasyon peynir üretim teknolojileri geliştirilmeli ve herkesin alabileceği düzeyde ürünler üretilmelidir. Yüksek hidrostatik basınç uygulamaları, ultarasound uygulamaları ve membran teknolojileri ile peynirde kullanılan hammaddede gereken özellikler sağlanarak kayıplar azaltılmaktadır. Bu teknolojilere alternatif olan yeni teknolojiler belirlenmeli ve bu teknolojilerin birlikte kullanımları ile ilgili çalışmalar yapılmalıdır. Sağlık üzerinde etkileri kanıtlanan bitkisel bileşenler ve farklı starter ve probiyotik kültürler peynir üretiminde kullanılarak sağlık açısından yararlı ürünler üretilmektedir. Ayrıca muhafaza şartlarında nanokompozit filmler ve farklı atmosfer şartları sağlanarak raf ömrü arttırılmaktadır. Bu alanda yapılan çalışmaların sayısı arttırılmalı ve endüstriye aktarımı gerçekleştirilmelidir. Böylece ülkemizde ve dünyada peynir tüketimi arttırılabilir. Yapılan arge çalışmaları ile sağlıklı, fonksiyonel özellikler kazandırılmış, raf ömrü arttırılmış yeni peynirler üretilebilir.

KAYNAKLAR

Anonim 2017. Food and Agriculture Organization of United Nations, (FAO), Available at: ftp://ftp.fao.org/docrep/fao/008/a0400e [10.09.22]

Barba F J , Terefe N S , Buckow R, Knorr D, Orlien V 2015. New opportunities and perspectives of high pressure treatment to improve health and safety attributes of foods: A review. FoodResearch International, 77(1): 725-742.

Bermudez D, Canovas G 2010. Processing of softhispaniccheese ("quesofresco") using thermo-sonicated milk: A study of physico chemical characteristics and storage life. Journal of FoodScience, 75: 548-558.

Bruschi C, Komora N, Castro S M, Saraiva J, Ferreira VnB, Teixeira P 2017. High hydrostatic pressure effects on *Listeria monocytogenes* and *L. innocua:* evidence for variability in inactivation behaviour and in resistance to pediocin bacHA-6111-2. Food Microbiology, 64: 226-231.

Chouliara E, Georgogianni K G, Kanellopoulou N, Kontominas M G 2010. Effect of ultrasonication on microbiological, chemical and sensory properties of raw, thermized and pasteurized milk. International Dairy Journal, 20: 307–313.

Çavuş, M. 2020. Peynir tuzlamada yeni bir yöntem: Jel salamura tekniği (Doktora Tezi), Erciyes Üniversitesi Fen Bilimleri Enstitüsü, Kayseri.

Çınar İ, 2018. Peynir üretiminde yüksek hidrostatik basınç uygulamaları. Kahramanmaraş Sütçü İmam Üniversitesi Mühendislik Bilimleri Dergisi, 21(1): 86-91.

Demirdöven A, Baysal T 2009. The use of ultrasound and combined technologies in food preservation. Food Reviews International, 25: 1-11.

De Morais Coutinho C, Chiu M C, Basso R C, Ribeiro A PnB , Gonçalves L A G, Viotto L A 2009. State of art of the application of membrane technology to vegetable oils: a review. Food Research International, 42: 536-550.

Devi A F, Buckow R, Hemar Y, Kasapis S 2013. Structuring dairy systems through high pressure processing. Journal of Food Engineering, 114(1): 106-122.

Devi A F, Buckow R, Singh T, Hemar Y, Kasapis S 2015. Colour change and proteolysis of skim milk during high pressure thermal-processing. Journal of Food Engineering. 147: 102-110.

Doğan E, Demir P, Arslan A 2022. Peynir teknolojisinde modifiye atmosfer paketleme. Current Perspectives on Health Sciences, *3*(1): 16-24.

Ersan L Y, Gizem S 2020. Peynir Benzeri Ürünlerde İnovatif Yaklaşımlar: İmitasyon Peynir. Türk Bilimsel Derlemeler Dergisi, 13(1): 23-31.

Heinz V, Buckow R 2009. Food preservation by high pressure. Journal of Consumer Protection and Food Safety, 5(1): 73-81.

Henning D R, Baer R J, Hassan A N, Dave R 2006. Major advances in concentrated and dry milk products, cheese, and milk fat-based spreads. Journal of Dairy Science, 89(4): 1179-1188.

Liepa M , Zagorska J, Galoburda Rn 2016. High-Pressureprocessing as noveltechnology in dairyindustry: A Review. FoodSciences, 1: 76-83.

Marchesini G, Balzan S, Montemurro F 2012. Effect of ultrasound aloneor ultrasound coupled with COR2R on the chemical composition, cheese-making properties and sensory traits of raw milk. Innovative Food Science and Emerging Technologies, 16:391-397.

Martinez-Rodriguez Y, Acosta-Muniz C, Olivas G I, Guerrero-Beltran J, Rodrigo-Aliaga D, Sepulveda D R 2012. High hydrostatic pressure processing of cheese. Comprehensive Reviews in Food Science and Food Safety, 11: 399-416.

Munoz- Cuevas M, Guevara L, Aznar A, Martinez A, Periago P M, Fernandez P S 2013. Characterisation of the resistance and the growth variability of *Listeria monocytogenes* after high hydrostatic pressure treatments. Food Control, 29(2): 409-415.

Oğuzhan P 2013. Yüksek hidrostatik basınç teknolojisinin gıda endüstrisinde kullanımı. Fen Bilimleri Enstitüsü Dergisi, 6(2): 205-219.

Ozturk M, Govindasamy-Lucey S, Jaeggi J J, Johnson M E, Lucey J A 2013. Theinfluence of highhydrostaticpressure on regular, reduced, lowand no salt added Cheddarcheese. International Dairy Journal, 33: 175-183.

Pluta-Kubica A, Jamróz E, Khachatryan G, Florkiewicz A, Kopel P 2021. Application of furcellaran nanocomposite film as packaging of cheese. Polymers, 13(9): 1428.

Rastogi N 2011. Opportunities and challenges in application of ultrasound in food processing. Critical Reviews in Food Science and Nutrition, 51: 705–722.

Sayın L, Tamer C E 2014. Yüksek hidrostatik basınç ve ultrasonun gıda koruma yöntemi olarak kullanılması. Uludağ Üniversitesi Ziraat Fakültesi Dergisi, 28(1): 83-94.

Sezer E, İnanç A L 2013. Gıda sanayinde yüksek basınç uygulamalarındaki besin kayıpları. Kahramanmaraş Sütçü İmam Üniversitesi Doğa Bilimleri Dergisi, 16(4): 36-43.

Sfakianakis P, Tzia C 2014. Conventional and ınnovative processing of milk for yogurt manufacture; development of texture and flavor: a review. Foods, 3: 176-193.

Silva F V 2015. High pressure processing of milk: Modeling the inactivation of psychrotrophic Bacillus cereus spores at 38–70° C. Journal of Food Engineering, 165: 141-148.

Tepge. Durum ve Tahmin. Sütve Süt Ürünleri. 2020. Dr. Zeliha Yasan Ataseven.TepgeYayın NO: 321. ISBN: 978-605-7599-48-3. Ekim: 2020.

Tokuşoğlu Ö 2013. Ultrasound processing in foods: fundamentals, applications and advances in commercialization. ınadvances in commercialization of nonthermal processing. Short Course-IftPreannual Meeting. 12-13Haziran, Hilton-Chicago, USA

Türkmen F 2012. Yüksek güçlü ultrasound işleminin sütün fizikokimyasal ve jelleşme özelliklerine etkisi (Yüksek Lisans Tezi). İstanbul Teknik Üniversitesi Fen Bilimleri Enstitüsü, İstanbul

USK, 2020. 2019 Süt Raporu, Dünya ve Türkiye'de Süt Sektörü İstatistikleri. Ulusal Süt Konseyi, Ankara.

Üçüncü M 2008. A"danZ"ye Peynir Teknolojisi.Meta Basım, İzmir.

Yılmaz F 2011. Kaşar peyniri üretiminde balmumunun kaplama materyali olarak kullanılabilirliği ve peynir kalitesine etkisi (Yüksek Lisans Tezi). Atatürk Üniversitesi, Fen Bilimleri Enstitüsü, Erzurum

DRAFT

ORAL PRESENTATION

Morphological and molecular systematics of *Anopheles* (Diptera: Culicidae) species sampled from Çanakkale and Muğla provinces

Aleyna Çağan[1] (*ORCID: https://orcid.org/0000-0001-7688-6379*), Sezer Yalçın[1] (*ORCID: https://orcid.org/0000-0003-2848-7239),* Ersin Doğaç[2*] (*ORCID: https://orcid.org/0000-0003-4426-2187*)

[1] Muğla Sıtkı Koçman University, Institute of Science and Technology, Department of Molecular Biology and Genetics, Muğla Turkey
[*2] Muğla Sıtkı Koçman University, Faculty of Science, Department of Molecular Biology and Genetics, Muğla, Turkey

*Correspondence: ersindogac@mu.edu.tr

Abstract

Malaria is one of the most important vector-borne diseases that has long had a worldwide impact and has killed thousands of people. The most important vector organisms in the spread of malaria are mosquito species in the genus *Anopheles*. For this reason, mosquitoes of the genus *Anopheles* are one of the most important species to be controlled all over the world. Since the identification of the target species is of great importance in the fight against malaria and malaria vectors, systematic studies on these species have gained momentum. For this purpose, both morphological and molecular systematic studies have been carried out depending on the developing techniques, and *Anopheles* species and malaria vectors have been identified. In this study, *Anopheles* species in Çanakkale and Muğla provinces were determined using both morphological and molecular methods. In this context, morphological examination of adult samples of *Anopheles* species sampled between 2018-2019 was carried out and then the species diagnoses were confirmed using molecular identification methods. In Muğla province, 263 *An. superpictus*, 62 *An. hyrcanus*, 16 *An. maculipennis*, 191 *An. sacharovi*, 208 *An. claviger* species were identified from 740 individuals. In Çanakkale province, 126 *An. superpictus*, 156 *An. sacharovi*, 353 *An. maculipennis*, 191 *An. claviger* species were identified from 826 individuals. No *An. hyrcanus* species were found in Çanakkale province. Regular monitoring of *Anopheles* species that vector malaria is very important for control efforts. These monitoring studies contribute to the provision of information, planning, and resources necessary to control malaria epidemics and ultimately eradicate malaria worldwide.

Keywords: *Anopheles*, ITS2, Morphological and Molecular systematics.

INTRODUCTION

Mosquitoes are members of the Diptera family, which includes the subfamilies Anophelinae and Culicinae, and harbor about 3,500 different species (Harbach and Kitching, 2005). About 200 of these species are carriers of diseases that pose a serious threat to humans (Reinert, 2001). They pose a health threat by playing a role in the spread of diseases such as malaria, dengue fever, yellow fever, encephalitis, West Nile fever, and filariasis worldwide (Lehane, 1991; Kettle, 1995; Beaty and Marquart, 1996). Among these diseases, malaria is the most destructive, especially in the past and present. Approximately 500 million people worldwide are infected by malaria parasites carried by mosquitoes each year, and more than 1 million of them die. The World Health Organization (WHO) recognizes malaria as one of the three most important infectious diseases today, along with tuberculosis and AIDS (WHO 2009a, b; Özbilgin 2011). Therefore, mosquitoes, which are in an extremely critical position in terms of human health, represent a situation that requires increased efforts to combat them. From a global public health perspective, *Anopheles* species, in particular, are of great importance as they transmit malaria parasites to humans, making them a focal point in the fight against malaria. For effective control of malaria, the WHO emphasizes that the fight against *Anopheles* species, the main vector of malaria, plays a vital role (WHO, 2012). Accordingly, it has been emphasized that a better understanding of *Anopheles* population density is essential for a successful fight against malaria in the future (malERA, 2011). Controlling diseases such as malaria by combating these species remains a critical public health priority. In our country, a series of morphological examinations to identify mosquito species have been carried out with specimens

collected from malaria-endemic areas (Parris, 1959; Postiglione et al., 1970; Postiglione et al., 1972; Kasap and Kasap, 1983; Merdivenci, 1984; Alten et al., 2000; Simsek, 2006). In this regard, studies determining insecticide resistance levels in vector species (Kasap et al., 1992, 2000; Lüleyap et al., 2002; Alten et al., 2003), studies in which laboratory colonies were established (Kasap and Kasap, 1983b; Simsek et al., 2005; Yurttaş and Alten, 2006), ecological (Kasap, 1986; Simsek, 2006) and etiological (Demirhan and Kasap, 1995; 1996) studies were conducted. These studies have helped us to understand the distribution and diversity of *Anopheles* species in Turkey. However, despite all these studies, a complete fauna of *Anopheles* species in our country has not yet been definitively revealed, especially when compared to mosquito systematic studies in other countries (Köşlüoğlu, 2017). This situation shows that the mosquito fauna in our country requires more detailed and comprehensive research. In the light of recent studies, 64 mosquito species belonging to 8 different genera have been found in our country, although the exact number has not yet been determined. Worldwide, as of 2004, 444 species belonging to the genus *Anopheles* and species complexes including 40 species that have not yet been named have been identified. In Turkey, this genus is represented by 10 different species. Under the genus *Anopheles*, *An. algeriensis*, *An. claviger*, *An. hyrcanus*, *An. maculipennis*, *An. marteri*, *An.* melanoon, *An.* pulcherrimus, *An. plumbeus*, *An. sacharovi*, and *An. superpictus* species constitute the *Anopheles* fauna of our country (Ramsdale et al., 2000; Şimşek et al., 2011; Günay, 2015). These data help us to understand the diversity of mosquito species in our country and the distribution of these species worldwide. This study aimed to identify *Anopheles* species sampled from Muğla and Çanakkale provinces using both morphological and molecular methods. In line with this goal, the project numbered 117 Z 847, supported by TUBITAK, includes adult *Anopheles* samples collected from Muğla and Çanakkale provinces between 2018 and 2019. These samples were examined morphologically under laboratory conditions and then evaluated for molecular species identification.

MATERIALS AND METHODS

The study material consists of samples collected from Muğla and Çanakkale provinces between 2018 and 2019 within the scope of TUBITAK 1001 project number 117Z847. In each of the 8 locations sampled for the two provinces, 4 locations were sampled to best sample the location. The locations of the samples taken from each location are given on the map in Figure 1.

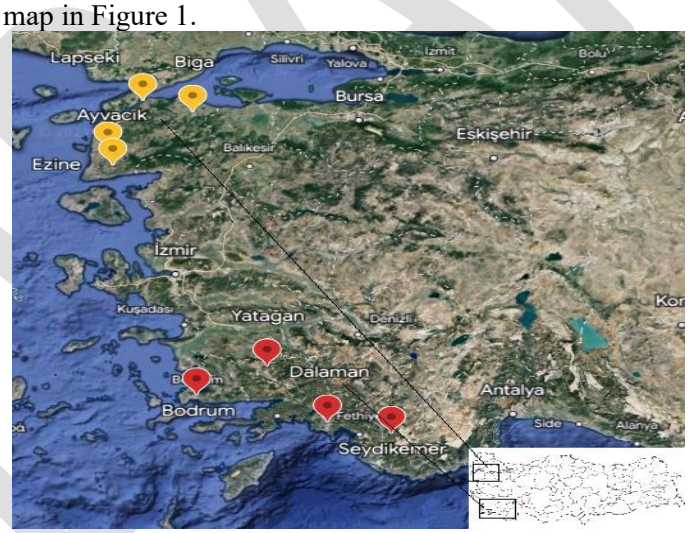


Figure 1. The locations of 8 locations belonging to 2 provinces sampled within the scope of the project are indicated on the map.

Morphological Description

The collected adult mosquitoes were identified using adult identification keys (Glick, 1992; Becker et al. 2010) and the MosKeyTool program (https://www.medilabsecure.com/moskeytool.html).

Genomic DNA Isolation and Analysis of rDNA ITS2 Region for Molecular Species Identification

Genomic DNA isolations were performed on the species, 5 individuals each from morphologically different species for each province. Genomic DNA isolations were performed using the Zymo Research Quick DNA Miniprep Plus Kit (Catalog No: D4068 & D406). rDNA ITS2 region was analyzed according to Djadid et al. (2007) and forward 5.8 S 5'-TGTGAACTGCAGGACACATG-3' and reverse 28 S 5'-TATGCTTAAATTCAGGGGGT-3' primers were used for amplification.

Data Analysis

To compare the obtained nucleotide sequence with the same organism or with different organisms, it was queried with BLAST application (https://blast.ncbi.nlm.nih.gov/Blast.cgi?PAGE_TYPE=BlastSearch). The DNA base sequences obtained were compared with DNA sequences registered in NCBI.

RESULTS

As a result of the morphological examination of the material obtained as a result of the field study in the laboratory environment, 5 *Anopheles* species (*An. sacharovi*, *An. superpictus*, *An. claviger*, *An. hyrcanus*, *An. maculipennis*) were morphologically determined. Of the 740 individuals collected from Muğla province, 263 were *An. superpictus*, 208 were *An. claviger*, 16 were *An. maculipennis*, 191 were *An. sacharovi* and 62 were *An. hyrcanus* (Table 1). Out of 826 individuals collected from Çanakkale province; 126 were identified as *An. superpictus*, 191 as *An. claviger*, 353 as *An. maculipennis*, and 156 as *An. sacharovi* (Table 1). The numbers and distributions of the identified species are given in Table 1.

Table 1. Number of samples collected from Çanakkale and Muğla provinces.

	An. claviger		*An. maculipennis*		*An. sacharovi*		*An. superpictus*		*An. hyracanus*	
	Muğla	Çanakkale	Muğla	Çanakkale	Muğla	Çanakkale	Muğla	Çanakkale	Muğla	Çanakkale
1. Dönem	110	85	0	60	48	41	116	42	0	0
2. Dönem	44	44	0	247	45	44	104	43	62	0
3. Dönem	54	62	16	46	98	71	43	41	0	0
Toplam	**208**	**191**	**16**	**353**	**191**	**156**	**263**	**126**	**62**	**0**

Molecular confirmation of the morphologically identified species was performed with 5 individuals each. In this context, ABI-formatted chromatogram files were opened and visually examined in FinchTV (Geospiza, Inc.; Seattle, WA, USA; http://www.geospiza.com) and queried with BLAST application to compare nucleotide sequences with the same organism or with different organisms (https://blast.ncbi.nlm.nih.gov/Blast.cgi?PAGE_TYPE=BlastSearch).

The obtained MEGA 7 package program (Kumar et al. 2016) was aligned using the Clustal W multiple sequence alignment program (Clustal W multiple sequence alignment program). As a result of the alignment, the relationships and proximity of the species with each other are given in the Maximum-Likelihood dendrogram in Figure 2.

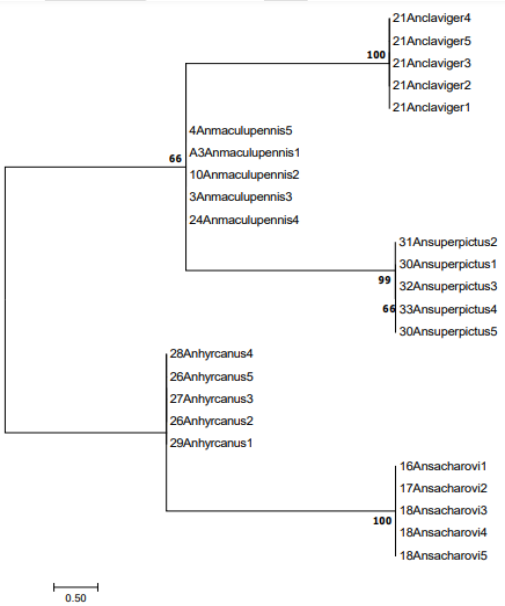


Figure 2. Maximum-Likelihood dendrogram of *Anopheles* species collected from Muğla and Çanakkale provinces.

NETWORK analysis was performed to visualize and examine the relationships between the haplotypes obtained and the graph is given in Figure 3 (ver. 4.6, Bandelt et al. 1999; Polzin and Daneschmand 2003).

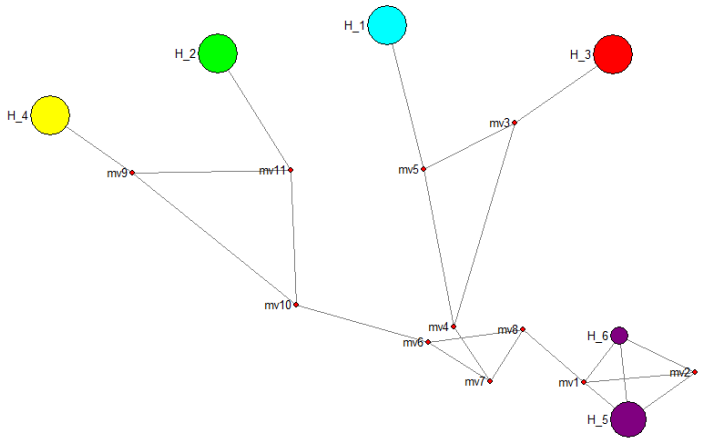


Figure 3. Network graph obtained as a result of base sequence analysis for ITS gene region of *Anopheles* species (H_1: *An. claviger*, H_2: *An. hyrcanus*, H_3: *An. maculipennis*, H_4: *An. sacharovi*, H_5 and H_6: *An. superpictus*).

DISCUSSION

The World Health Organization (WHO) has highlighted the importance of vector control of *Anopheles* species (WHO, 2012). In this context, it has been emphasized that a better understanding of *Anopheles* populations is a critical factor in achieving success in the fight against malaria in the future (malERA, 2011). To reduce or prevent malaria cases in Turkey, it is necessary to determine the *Anopheles* fauna in Turkey. Many studies based on the morphological examination have been carried out for species identification of *Anopheles* mosquitoes sampled from malaria-endemic regions of our country. In this study, *Anopheles* species collected from Çanakkale and Muğla provinces were first identified by morphological methods and then by molecular methods. In total, 740 samples from Muğla province and 826 samples from Çanakkale province were examined and species identification was made. As a result of the study, 5 different species (*An. superpictus*, *An. sacharovi*, *An. hyrcanus*, *An. maculipennis*, *An. claviger*) from Muğla province and 4 different species (*An. superpictus*, *An. sacharovi*, *An. maculipennis*, *An. claviger*) from Çanakkale province were identified. After the morphological species determinations, the ITS2 gene region was confirmed by molecular identification analysis. The presence of various species belonging to the *An. maculipennis* Meigen, (1818) complex in our country started with the work of Akalın (1936), and in the following years, *Anopheles* species continued to be identified and the *Anopheles* fauna of Turkey began to be established. Parrish (1959), in his publication on the *Anopheles* fauna of Turkey, identified 13 different *Anopheles* species (*An. algeriensis, An. claviger, An. hyrcanus, An. maculipennis, An. marteri, An. multicolor, An. plumbeus, An. sacharovi, An. sergenti, An. sinensis, An. superpictus*) and subspecies. Gökberk (1961) determined that the *Anopheles* fauna of Turkey consisted of *An. algeriensis*, *An. claviger*, *An. hyrcanus*, *An. marteri*, *An. sergenti*, *An. maculipennis*, *An. sacharovi*, *An. superpictus* and *An.* multicolor species and reported the zoogeographic distribution of these species in Turkey. This study conducted in Muğla and Çanakkale provinces revealed that 5 different *Anopheles* species were encountered. These species are *An. superpictus*, *An. sacharovi*, *An. hyrcanus*, *An. maculipennis* and *An. claviger*. Compared to other studies, *An. algeriensis*, *An. marteri*, *An. sergenti*, *An.* multicolor, *An. plumbeus*, *An. sinensis* and *An. melanoon* were not found among 13 different *Anopheles* species and subspecies previously identified in the fauna of Turkey. Şimşek et al. (2011) carried out species identification using ITS2 sequence data of *An. maculipennis* complex specimens collected from 70 localities in 10 provinces in southern Turkey (Adana, Antalya, Burdur, Hatay, Isparta, Kayseri, Mersin, Muğla, Niğde, and Osmaniye). They found *An. sacharovi* species in the samples collected from Gökova and Ortaca locations in Muğla province. As a result of this study, *An. maculipennis* s.s, *An. melanoon* and *An. sacharovi* species were identified in the Mediterranean Region. With this study, the presence of *An. melanoon* species in Turkey was confirmed for the first time at the molecular level. In our study, *An. maculipennis* and *An. sacharovi* species were found in Muğla and Çanakkale provinces and *An. melanoon* species, which was found in the study of Şimşek et al. (2011), was not observed. Sevgili and Şimşek (2012) were sampled *An. maculipennis* samples from 12 provinces (Adana, Aydın, Burdur, Çankırı, Edirne, Iğdır, Kırklareli, Konya, Mersin, Muğla, Samsun and Şanlıurfa) and 70 different locations from different regions of Turkey and compared the 200 ITS2 region sequences obtained with the existing sequences in GenBank. According to the results of the study, 106 of the samples were

identified as *An. maculipennis* s.s, 5 as *An. melanoon* and 89 as *An. sacharovi*. While *An. sacharovi* species was observed in the samples collected from Dalaman basin, this species was not observed in the samples from Northern and Central Anatolia. In our study, *An. superpictus*, *An. sacharovi*, *An. hyrcanus*, *An. maculipennis* and *An. claviger* species were observed as a result of the identification of samples taken from Bodrum, Yatağan, Seydikemer and Dalaman locations in Muğla province. Although the sub-locations were different, *An. sacharovi* species were observed in common studies. Günay (2015) morphologically identified 6318 individuals from 34 different provinces of Turkey, including Muğla and Çanakkale, between 2011 and 2015. Based on the results obtained, it was determined that the mosquito species list in Turkey consists of 64 taxa in total. Samples were collected from Datça and Fethiye locations in Muğla province and Ayvacık location in Çanakkale province and only 5 *An. hyrcanus* and 10 *An. sacharovi* species were identified in Muğla province. The researcher did not find any *Anopheles* species in Çanakkale province. He observed 76 individuals of *An. claviger* from 17 different provinces, 1382 individuals of *An. maculipennis* from 22 different provinces and 68 individuals of *An. superpictus* from 3 different provinces. Günay did not find any *Anopheles* species in Çanakkale province in his study conducted in 2011-2015, and in our study with the samples collected in 2018-2019, 4 *Anopheles* species were identified in Çanakkale province. While *An. superpictus*, *An. sacharovi*, *An. maculipennis* and *An. claviger* were detected in Çanakkale province (Ayvacık, Biga, Lapseki, Ezine), *An. superpictus*, *An. hyrcanus*, *An. sacharovi*, *An. maculipennis* and *An. claviger* were detected in Muğla province (Bodrum, Yatağan, Seydikemer, Dalaman). *An. hyrcanus* and *An. sacharovi* species were commonly detected in Muğla province. In their study, Sevgili and Şimşek (2012) found a total of 5 *An. sacharovi* species from Muğla location (Akçapınar, Kapıkargın) as a result of field and molecular studies. Tüzün (2010), detected *An. algeriensis*, *An. claviger* and *An. superpictus* species in samples sampled from the Datça peninsula. Köşlüoğlu (2017) morphologically identified 7 Anopheles species including *An. sacharovi*, *An. melanoon*, *An. superpictus*, *An. claviger*, *An. hyrcanus*, *An. maculipennis* and *An. plumbeus* in a study conducted throughout Turkey including Muğla province (Kapıkargın). In common with the previous studies, *An. superpictus*, *An. hyrcanus*, *An. sacharovi*, *An. maculipennis* and *An. claviger* species were observed in Muğla province, while *An. algeriensis*, *An.* melanoon and *An. plumbeus* species were not observed. In our study, 126 *An. superpictus*, 156 *An. sacharovi*, 353 *An. maculipennis*, 191 *An. claviger* species were detected from 826 individuals in Çanakkale province. In the face of these data, *An. maculipennis* was determined as the most abundant and dominant species for Çanakkale province. In previous studies conducted in Çanakkale province, the dominant and rare species of these species were not fully determined. In Muğla province, 263 *An. superpictus*, 62 *An. hyrcanus*, 16 *An. maculipennis*, 191 *An. sacharovi*, 208 *An. claviger* species were identified from 740 individuals. Due to the high number of *An. superpictus* individuals in Muğla province, it was concluded that this species is the dominant species. In addition, according to the data obtained, *An. hyrcanus* was determined as a rare species (Table 1). Possible reasons for the differences in the dominant and most abundant species between localities may be the different habitats of the species and their exposure to different ecological and climatic conditions.

CONCLUSION

The ITS2 region of ribosomal DNA has been proven to be useful in species identification by Linton et al. 2003, and successfully in the identification of *Anopheles* species by Djadid et al. 2007, Oshaghi et al. 2008, Şimşek et al. 2011. In our country, studies on the classification of species in the *An. maculipennis* complex by base sequence analysis of the rDNA-ITS2 region is also available in the literature. However, these studies were conducted on Datça, Gökova, Ortaca, and Dalaman regions in Muğla and there is not enough data for other regions of Muğla. The fauna of Çanakkale province has not been studied so far. In our study, *Anopheles* species were identified in Muğla and Çanakkale provinces using both morphological and molecular methods. Considering the studies conducted above, it is seen that different *Anopheles* species are denser and these dominant species change in different years in similar locations. Therefore, more studies are needed to determine the population densities of these important vector organisms. In conclusion, the data obtained revealed that both morphological and molecular studies should be carried out together for the determination of the *Anopheles* fauna of Turkey. In particular, more detailed and comprehensive sampling studies should be carried out for species groups, the samples obtained should be compared with the samples belonging to species in our nearby geography and analyzed by using ITS2 and other molecular markers together. Since these species are vectors of malaria, regular monitoring and analysis of these species is necessary for control efforts.

ACKNOWLEDGMENTS

This study was financially supported by the Scientific and Technological Research Council of Turkey (TUBITAK) under the 2209/A University Students Research Projects Support Program.

REFERENCES

Alten B, Çaglar SS, Özel O 2000. Malaria and its vectors in Turkey. European Mosquito Bulletin, 7: 27–33.

Alten B, Çaglar SS, Simsek FM, Kaynas S 2003. Effect of insecticide treated bednets for malaria control in southeast Anatolia-Turkey. Journal Vector Ecology, 28: 97–107

Bandelt HJ, Forster P, Rohl A 1999. "Median-joining networks for inferring intraspecific phylogenies", Molecular Biology and Evolution, 16: 37–48.

Beaty BJ, Marquardt WC 1996. The Biology of Disease Vectors. University Press of Colorado, pp. 632, USA.

Becker N, Petric D, Zgomba M, Boase C, Madon MB, Dahl C, Kaiser A 2010. Mosquitoes and Their Control.Springer-Verlag Berlin Heidelberg. 577 pp.

Demirhan O, Kasap M 1995. Bloodfeeding behavior of *Anopheles sacharovi* in Turkey. Journal of the American Mosquito Control Association, 2: 478– 481

Demirhan O, Kasap M 1996. Blood-sucking and parity rates of *Anopheles sacharovi* adults and seasonal densities of adult and pre-adult population in Tarsus (Icel) region. Tr. J. Zool., 20: 21–26.

Glick J 1992. "Illustrated key to the female *Anopheles* of Southwestern Asia and Egypt (Diptera: Culicidae)", Mosquito Systematics, 24(2), 125-153.

Gökberk C 1961. *Anopheles sacharovi* (Favre 1903) in Turkey. Mosquito News, 21(2).

Günay F 2015. Türkiye'de Sivrisinek Faunası Üzerine DNA Barkodlama Yöntemiyle Moleküler Analizler. Doktora Tezi. Hacettepe Üniversitesi, Fen Bilimleri Enstitüsü, Ankara, Türkiye, 120 s.

Harbach RE, Kitching IJ 2005. Reconsideration of Anopheline phylogeny (Diptera: Culicidae: Anophelinae) based on morphological data. Systematics and Biodiversity, 3: 345–374.

Kasap H, Kasap M 1983. Türkiye Anophelinae (Diptera: Culicidae) türleri. Türkiye Hijyen Derneği Biyoloji Dergisi, 40(1): 39-52.

Kasap H, Kasap M, Akbaba M, Alptekin D, Demirhan O, Luleyap U, Pazarbası A, Akdur R, Wade J 1992. Residual efficacy of pirimiphos methyl (Actellic) on *Anopheles sacharovi* in Cukurova. Turkey. Journal of the American Mosquito Control Association, 8: 47–51.

Kasap H, Kasap M, Alptekin D, Lüleyap Ü, Herath PRJ 2000. "Insecticide resistance in *Anopheles sacharovi* Favre in Southern Turkey", Bulletin of World Health Organization, 78, 687–692.

Kasap M 1986. Seasonal variation in populations of *Anopheles maculipennis*, *Anopheles claviger* and *Culex pipiens* in Turkey. Journal of the American Mosquito Control Association, 11: 11–14.

Kasap M, Kasap H 1983b. Laboratory colonization of *Anopheles sacharovi*, the principal vector of human malaria in Turkey. Mosquıto News, 43: 489–499.

Kettle DS 1995. Medical and Veterinary Entomology. CAB International, pp. 725, UK

Köşlüoğlu Ç 2017. Türkiye'de Yayılış Gösteren *Anopheles* Türlerinin (Diptera: Culicidae) Morfolojik ve Moleküler Sistematiği. Adnan Menderes Üniversitesi Yüksek Lisans Tezi. 73 syf. Aydın.

Kumar NP, Rajavel AR, Natarajan R, Jambulingam P 2007. "DNA Barcodes can distinguish species of Indian mosquitoes (Diptera: Culicidae)", Journal of Medical Entomology, 44, 1–7.

Lehane MJ 1991. Biology of Blood-sucking Insects. Harper Collins Academic, pp. 288, UK.

Luleyap H, Alptekin D, Kasap H, Kasap M 2002. Detection of knockdown resistance mutations in *Anopheles sacharovi* (Diptera: Culicidae) and genetic distance with *Anopheles gambiae* (Diptera: Culicidae) using cDNA sequencing of the voltage-gated sodium channel gene. Journal of Medical Entomology, 39: 870–874.

malERA. Consultative Group on Vector Control A research agenda for malaria eradication: vector control. PLoS Med. 2011;8(1):e1000401.

Merdivenci A 1984. Türkiye Sivrisinekleri. Yurdumuzda varlığı bilinen sivrisineklerin biyomorfolojisi, biyoekolojisi, yayılışı ve sağlık önlemleri. İstanbul Üniversitesi Cerrahpaşa Tıp Fakültesi, Yay. Rek. No.3215, Tas Matbaası, pp. 136-354, İstanbul.

Özbilgin A, Topluoglu S, Es S, İşlek E, Mollahaliloglu S, Erkoç Y 2011. "Malaria in Turkey: Successful control and strategies for achieving elimination", Acta Tropica, 120, 15-23.

Polzin T, Daneschmand SV 2003. "On Steiner trees and minimum spanning trees in hypergraphs", Operations Research Letters, 31, 12–20.

Postiglione M, Smiraglia BC, Lavagnino A, Gökberk C, Ramsdale C 1970. A preliminary note on the occurance in Turkey of the *An. subalpinus* form of the *An. maculipennis* complex. Rivista Di Parassitologia, 31: 155-159.

Postiglione M, Tabanli S, Ramsdale CD 1972. *Anopheles claviger* in Turkey. Rivista di Parassitologia, 33: 219-230.

Ramsdale C, Snow K 2000. Distribution of the genus *Anopheles* in Europe, European Mosquito Bulletin, 7, 1–26.

Reinert JF 2001. Revised list of abbreviations for genera and subgenera of Culicidae (Diptera) and notes on generic and subgeneric changes. Journal of the American Mosquito Control Association, 17(1): 51-55.

Sevgili E, Şimşek FM 2012. Distribution pattern and molecular identification of *Anopheles maculipennis* complex in eight river basins of Anatolia, Turkey. North-Western Journal of Zoology, 8 (2): 223-231.

Şimşek FM 2006. Seasonal frequency and relative density of larval populations of mosquito species (Diptera: Culicidae) in the Sanlıurfa province, Turkey. Turkish Journal of Zoology, 30: 383–392.

Şimşek FM, Kaynas S, Alten B, Caglar SS 2005. Laboratory colonization of the malaria vector *Anopheles* (Cellia) *superpictus* from Sanliurfa, Turkey. European Mosquıto Bulletin, 19: 26–30.

Şimşek FM, Ulger C, Akiner MM, Tuncay SS, Kiremit F, Bardakci F 2011. Molecular identification and distribution of *Anopheles maculipennis* complex in the Mediterranean region of Turkey. Biochemical Systematic Ecology, 39, 4–6, 258– 265.

Tüzün N, Sivrisinek, Datça Yarımadası'ndaki. "Türleri ve Üreme Alanları." İzmir: Ege Üniversitesi Fen Bilimleri Enstitüsü (2010). 7s

WHO. WHO malaria report. In: WHO, editor. WHO global malaria program. Geneva 2012. p. 260.

World Health Organization. 2009. "Guidelines for the treatment of malaria", 2nd ed. Geneva, Switzerland, pp. 13–56.

World Health Organization. 2009. "World Malaria Report 2009", Geneva, Switzerland, pp. 27–44.

Yurttas H, Alten B 2006. Geographic differantion of life table attributies among Anopheles sacharovi (Diptera: Culicidae) populations in Turkey. Journal of Vector Ecology, 31: 275–284.forest. New York: Springer, pp. 66–84.

DRAFT

ORAL PRESENTATION

Mucizevi meyve: Hünnap (*Ziziphus jujuba* Mill.)

Büşra Yılmaz [1*] (ORCID: https://orcid.org/0009-0006-2304-4729), Mustafa Kenan Geçer [1](ORCID: https://orcid.org/0000-0003-1959-9909)

[1] Bolu Abant İzzet Baysal Üniversitesi, Ziraat Fakültesi, Tohum Bilimi ve Teknolojisi Bölümü, Bolu, Türkiye

*Sorumlu yazar e-mail: yilmazbusra824@gmail.com

Özet

Hünnap anavatanı Çin olup 4000 yıldan fazla bir sürede yetiştirilen bir meyvedir. Ülkemizde ise; Ege Bölgesi'nde, Akdeniz Bölgesi'nde ve Marmara Bölgesi'nde bazı illerde hünnap bitkisinin doğal yayılış alanlarını oluşturmaktadır. Bölgeye göre 6 ila 9 m arasında değişiklik gösteren sert gövdeye sahip çalı formunda bir ağaçtır. Hünnap bir ılıman iklim bitkisi olup, Deniz seviyesinden 1700 metrelere kadar çıkabilir. -20ºC'ye kadar dayanır. Süzek ve verimli topraklarda aşırı yağışlara karşı dayanıklı ve kuraklıktan etkilenmez. Hünnap eldesi ve üretimi sayesinde meyve, meyve suyu, çay, sirke ve bakım ürünlerinde kullanılmaktadır. Bitkinin kabuk, meyve, tohum ve yaprak gibi kısımları besin içeriği açısından zengindir. İnsan sağlığına faydalı bileşenleri sayesinde insan sağlığı üzerine etkileri çok büyüktür.

Anahtar Kelimeler: Hünnap, hünnap üretimi, meyve özellikleri, sağlıklı yaşam

The miracle fruit: Jujube (*Ziziphus jujuba* Mill.)

Abstract

Jujube is a fruit that is native to China and has been grown for more than 4000 years. In our country; It forms the natural distribution areas of the jujube plant in some provinces in the Aegean Region, the Mediterranean Region and the Marmara Region. It is a shrub-shaped tree with a hard trunk that varies between 6 and 9 m depending on the region. Jujube is a temperate climate plant and can reach 1700 meters above sea level. Withstands down to -20ºC. Resistant to excessive precipitation in drained and fertile soils and not affected by drought. Jujube is used in fruit, fruit juice, tea, vinegar and care products thanks to its extraction and production. Parts of the plant such as bark, fruit, seeds and leaves are rich in nutrients. Thanks to its beneficial components for human health, its effects on human health are enormous.

Keywords: Jujube, jujube production, fruit characteristics, healty life

GİRİŞ

Hünnap (*Ziziphus jujuba* Mill.) Rhamnaceae familyasına ait olup 5-10 m yüksekliğinde ağaç ya da çalı formunda bir bitkidir(Nur Gül,2021).Hünnap, Ünnap, Hinnap, İnnap, Çiğde, Kuran İğdesi gibi yerel isimleri bulunan şifalı bir bitkidir. Birçok faydaları olan Hünnap ülkemizde yeterince tanınmamaktadır. Genellikle bazı bölgelerimizde doğal bitki örtüsü içerisinde kendiliğinden çoğalmaktadır. Üretimi ise oldukça sınırlıdır(Erdemir Gündoğmuş,2017).Ülkemizde ise; Ege Bölgesi'nde Çanakkale ve Denizli, Akdeniz Bölgesi'nde Burdur, Isparta, Hatay ve Antalya; İç Anadolu Bölgesi'nde Kayseri, Marmara Bölgesi'nde ise Bursa illeri hünnap bitkisinin doğal yayılış alanlarını oluşturmaktadır(Gün Sefa, 2017). Rakımı 0-1500 m, yıllık ortalama sıcaklık kışın 7-13°C ve yazın 37-48°C, yıllık ortalama yağış miktarı 120-2200 mm olan bölgelerde, kumlu-tınlı nötr veya hafif alkali topraklarda iyi yetişebilir (Kavas İlknur 2014).

Hünnap ağaçları morfolojik olarak dik ve tırmanıcı olup, bitkiler ağaç ve çalı formunda, boyları 7-10 metreyi bulmaktadır. Bu türler içerisinde *Ziziphus jujuba* ve *Ziziphus mauritiana'*nın meyveleri tüketim için yaygın olarak yetiştirilmektedir. Üretilen meyveler çoğunlukla kurutulmuş ve taze olarak tüketilmektedir(Gün Sefa, 2017).Faydaları ise; düşük kalorili bir meyve olduğu aynı zamanda da enerji verdiği için kilo verme konusunda

yardımcı olmaktadır, içindeki A ve bolca C vitamini sayesinde bağışıklık sistemini güçlendirir, vücudu hastalıklara karşı korur.

Hünnap, içinde bulunan ve niyasin olarak da bilinen B3 vitamini sayesinde kan dolaşımının düzenli bir şekilde devam etmesini sağlar. Yine B3 vitaminin etkisiyle hünnap sinir sistemi fonksiyonlarının da sağlıklı bir şekilde sürdürülmesine katkı sağlar. Güçlü antioksidan özelliği nedeniyle, karaciğer rahatsızlıklarından çeşitli kanser türlerine kadar birçok hastalığın iyileşme sürecine olumlu etkilerde bulunmaktadır. Hünnap meyvesinin antioksidan özelliği, cilt sağlığını koruma ve cildi güzelleştirme konusunda da önemli katkılar sağlanır. Çarpıntı, uykusuzluk, anemi, halsizlik, ateş, karaciğer hastalıkları vb. hastalıklarda kullanılır(Sunar Yunus, 2022).

HÜNNAPIN TARİHÇESİ

Hünnap, 4000 yıldan fazla bir süredir yetiştirildiği ve 400'ün üzerinde çeşidinin olduğu Çin orijinli bir meyvedir. Hünnap bitkisi asırlar önce Asya'dan dünyanın diğer bölgelerine dağılım göstermiş ve bugün Rusya, Kuzey Afrika, Güney Avrupa, Orta Doğu ve Güneybatı Birleşik Devletleri'nde yetiştirilmeye 5 başlanmıştır. Hıristiyanlık döneminin başlangıcında Avrupa'da tanınmış ve 1837'de Amerika'ya getirilmiştir(Koyuncu Büşra,2021).Hünnap vadilerde ve dağlık bölgelerde yaygın olarak dağılım gösteren Rhamnaceae familyasının yabani bir türüdür(Koyuncu Büşra,2021).Hünnap bitkisinin anavatanı Çin'in Yunan eyaleti olup, Çin'de 7000 yıldan beri gıda ve tıbbi amaçlı kullanılmak için yetiştirilmektedir. Bitkinin doğal yayılma alanı Rusya, Ortadoğu, Hindistan, Güney Avrupa, Anadolu ve Kuzey Afrika'dır(Kemeç Hürkan, 2019).

Hünnap, Hinnap, İnnap, Çiğde ve Kuran İğdesi gibi çok yerel isimleri bulunan şifalı bir bitkidir (Sunar Yunus, 2022).

HÜNNAPIN DÜNYADA VE TÜRKİYEDE ÜRETİM MİKTARI

Hünnap meyvesi yaklaşık 50 ülkede yetiştirilmektedir. Toplam hünnap üretiminin %90' undan fazlası Çin'de yetiştirilmektedir. Son 30 yıl içerisinde Çin'de, hünnap üretimi hızlı bir şekilde artış göstermiştir(Sunar Yunus, 2022).

Tablo1. Türkiye'de hünnap üretim alanı ve miktarı (TÜİK,2023)

Yıllar	Üretim Alanı(da)	Üretim miktarı Hünnap(ton)
2022	2921	2248
2021	2681	1760
2020	2439	1229
2019	1772	960
2018	1621	792
2017	1306	602
2016	1028	315
2015	673	302
2014	604	248
2013	458	142

TÜİK istatistiklerine göre, Ülkemizde, 2013 yılında 142 ton olan üretim miktarı giderek artmıştır. 2018 yılından 2022 yılına kadar hünnap üretiminde %27.7 artış sağlanmıştır.

Tablo 2.2022 yılı itibariyle hünnap üretim alanı ve miktarı bakımından başlıca illerimiz (TÜİK,2023)

İller	Üretim Alanı(da)	Üretim Miktarı Hünnap(ton)
Amasya	512	522
Manisa	473	252
Denizli	310	233
Balıkesir	150	187
Çanakkale	245	170
Antalya	250	137
Muğla	75	129
Hatay	33	128
Ankara	89	105
Bursa	127	86

İllere göre 2022 yılı hünnap üretim miktarlarına bakıldığında, 522 ton ile Amasya ilk sırada yer almaktadır. Hünnap üretiminde,2022 yılında ikinci sırada 252 tonla Manisa yer almaktadır. Denizli, Balıkesir, Çanakkale, Antalya, Muğla, Hatay, Ankara, Bursa hünnap üretiminde ön sırada olan diğer illerimizdendir.

HÜNNAP AĞACININ GENEL ÖZELLİKLERİ

Hünnap (*Ziziphus jujuba* Mill.), Rhamnaceae ailesine ait olup Ziziphus türlerinin en önemlisidir. Bölgeye göre 6 ila 9 m arasında değişim gösteren güçlü ve sert gövdeye sahip bir ağaçtır. Ağaçları, yarı yaprak döken ve çok dallı yapıdadır. Kabuğu derin boylamasına oluklara sahiptir ve grimsi kahverengi ya da kırmızımsı renklidir. Hünnap, 4 tür filiz yapısına sahiptir; birincil filiz (uzayan), ikincil filizler (yan dallar), ana taşıyıcı filizler (meyveli mahmuz) ve meyve taşıyan sürgünler (dalcık). Dalcıklar, özellikle de genç zamanlarında, yoğun beyaz tüylü ve zigzag şekline eğilimlidir; daha sonra dik yayılarak gri kahverengi ve esnek bir hal alır. Meyve veren dalları, yaprak döken özellikte değildir. Yaprakları, parlak ve oval şekilde olup dal yapısına sahip değildir. Yapraklar 2,5 ila 5,5 cm. uzunluğunda ve 2 ila 4 cm. genişliğindedir. Çiçekleri kokulu, soluk yeşilimsi veya sarı renkte ve 4 ila 8 mm. arasında değişen çaplarda olup küçüktür. Bir hünnap çiçeği, 5 çanak yaprak (sepal), 5 taç yaprak (petal), 5 başçık (anter) içeren 2 yumurtalıklı yapıdadır. Çiçekler, tek başlarına veya her yaprak aksilinde küme halinde bulunabilir. Çiçeklenme, ağacın çeşidine ve bulunduğu kısma göre değişim gösterir. Hünnap, sert çekirdekli bir meyvedir ve ortasındaki tek çekirdekli kısımda, 2 adete kadar tohum içerir. Meyvesi, yumurtalık ve nektar diskten meydana gelir. Meyve büyüklüğü, çeşidine bağlı olarak başparmak büyüklüğünden golf topuna kadar değişir. Meyve şekli de yuvarlak, oval, elma benzeri veya anormal şekilli olabilir. Meyve dokusu; asidik ve tatlı, rengi yeşilimsi, sarı veya bazen de kırmızımsıdır(Korkut Serkan,2022).Hünnap, çok çeşitli toprak tiplerine ve hava koşullarına iyi uyum sağlar. 0 ila 2.000 m arasındaki rakımda ve pH'sı 5,5 ila 8,5 arası toprakta büyürler. Yazın 48,9 °C sıcaklığı bile tolere edebilir ve kışın da -30°C soğuğa dayanabilirler(Korkut Serkan, 2022).

HÜNNAPIN BİLEŞİMİ VE BESİN İÇERİĞİ

Genel olarak makro besinler, tükettiğimiz besinlerdeki proteinler, yağlar, karbonhidratlar ve lifler gibi ana besinlerdir. Hünnap meyveleri zengin makro besin kaynaklarıdır. Mikro besinler, besinlerde küçük miktarlarda bulunan genellikle mineraller ve vitaminler olarak belirtilen önemli maddelerdir. Mikro besinler, sağlıklı büyüme, gelişme ve hastalıkların önlenmesi için hayati unsurlardır. Önceki çalışmalar, hünnap meyvelerinin vitaminler, makro elementler (N, K, Mg, Ca, P vb.) ve mikro elementler (Fe, Mn, Cu, Zn vb.) dahil olmak üzere zengin mikro besin kaynakları olduğunu göstermiştir(İkinci Ali, 2022).

Hünnap meyvesi diğer meyvelerle karşılaştırıldığında nispeten daha yüksek şeker içeriğine sahiptir. Bileşimindeki temel monosakkaritler ramnoz, ksiloz, mannoz, arabinoz, glukoz ve galaktoz olarak rapor edilmiştir(Koyuncu Büşra,2021).

Hünnap, esansiyel yağ asitlerinin önemli bir kaynağıdır; dolayısıyla meyvesi de doymamış yağ asitleri bakımından zengindir. Kurutulmuş meyvesinden elde edilen 33 yağ asidi bulunmaktadır. Yoğun bulunan yağ

asitleri; oleik, linoleik, palmitik ve palmitoleik asitlerdir. Yüksek C vitamini içeriğine sahip olması, bir diğer dikkat çekici özelliğidir. Daha az olmakla birlikte hünnap meyvesi tiamin, riboflavin, niasin, B6 ve A vitaminleri gibi vitaminleri de içermektedir. Hünnap meyvesi; magnezyum, fosfor, potasyum, sodyum ve çinko açısından da iyi bir kaynaktır(Korkut Serkan,2022).

HÜNNAP ÜRETİMİ

Hünnap meyvesi yaş veya kurutulmuş olarak üretimi ve pazarı olabilecek bir potansiyele sahiptir. Kurutulmuş hünnap, Çin'de yerel ve ihracat pazarlarında hâkim bir üründür. Hünnap meyveleri tamamen kırmızı ya da olgunlaşmış olarak toplandığında, meyve olgunluğundan ötürü şeker içeriği ve kurutma kalitesi daha yüksek olmaktadır. Toplandıktan sonra meyveler geleneksel yöntemeler veya kurutma fırınında kurutulabilir.

Isıyla kurutma en uygun yöntemdir. C vitamini kaybının az olması ve daha iyi meyve kalitesi vermesi açısından tercih edilmektedir. Kurutulmuş hünnaplar doğrudan atıştırmalık olarak tüketilebilir. Yulaf lapası, güveç, çorba veya çayda kullanılır veya hünnap ezmesi olarak da işlenebilir (Koyuncu Büşra,2021).Kurutulmuş meyvelerin çay içeriği, atıştırmalık, ekmek, kekler gibi gıda endüstrilerinde aktif bileşen olarak kullanılabilmesine ek olarak keçi sütü yoğurdu, kırmızı hünnap yoğurdu gibi ürünlerin besin değerini ve kalitesini artırmak için diğer yiyeceklere de eklenebildiği belirtilmektedir(Çıtıl Rıza,2022).Çin'de, hünnap meyvesinden üretilen şaraplar “hong zao jiu” olarak isimlendirilmektedir. Meyveler tamamen kırmızı renge ulaştığı zaman toplanır. Böylece meyvenin %60-70 olan C vitamini içeriği korunur. Hünnap meyve parçaları, özellikle kış boyunca taze tutabilmeleri için likör içine konur ve tamamen likör ile kaplanır. Daha sonra kavanoz veya kilitli torbalara konularak ve 6 ay ile 1 yıl arasında saklanabilirler. Ayrıca küçük paketler halinde de hazırlanarak ve birkaç ay sonra doğrudan bu ambalajlarda satılabilirler(Koyuncu Büşra,2021).Meyve, çay veya sirke olarak farklı şekillerde tüketilebilmektedir(Çıtıl Rıza.,2022).

Hünnabın su ve hidroalkolik ekstrakları, cilt bakım ürünlerinde antiinflamatuvar, nemlendirici, yatıştırıcı, kırışıklık karşıtı ve güneşten koruma özellikleri için kullanılmaktadır Kapsül, tablet veya sıvı formda bitkisel tonik formüllerinde bir bileşen olarak hünnap ekstraktları kullanılmaktadır. Ayrıca hünnabın ham hali çorba karışımlarında da kullanılır. Meyve suyu, şarap ve sirke yapımında da hünnaplar kullanılabilir. Taze hünnap meyveleri, dondurma veya meyve salatası olarak da tüketime sunulabilir. Kurutulmuş hünnaplar doğrudan meyve ve fındık karışımlarında kullanılabilir veya kek, tart ya da fırın ürünlerinin üretiminde, pişirmede kuru üzüm ya da hurmaya ikame edilir(Koyuncu Büşra,2021).

HÜNNAPIN SAĞLIK ÜZERİNE ETKİLERİ

Hünnap, tıbbi bitkilerden biri olarak kabul edilmektedir. Hünnap meyvesinin biyolojik aktiviteleri ve insan sağlığı üzerine faydaları üzerinde uzun süre çalışılmaktadır. Doğal şifalı otlar, özellikle yenilebilir ve şifalı çift amaçlı bitkiler, pişirme işlemleri sırasında malzeme olarak kullanılarak tedavi edici işlevlerin gerçekleştirilmesine yönelik bir uygulamadır. Hünnap, binlerce yıla yayılan sağlık özellikleri nedeniyle günlük hayatta favori bir meyve olarak kabul edilmiştir. Bahsedilen hücresel ve hayvansal bulgulara göre hünnabın anemi, iltihaplanma ve demir/vitamin eksikliğine karşı korunma için tıbbi gıda ve takviye geliştirmede umut verici bir potansiyel taşımaktadır(İkinci Ali,2022).Antik çağlardan beri kanser tedavisi için bitkisel ilaçlar kullanılmıştır. Amerika Birleşik Devletleri’nde ölüme yol açan en önemli 4 hastalık olan diyabet, kanser, hipertansiyon ve kardiyovasküler hastalıkların önlenmesi ve tedavisinde fitokimyasal maddelerden yararlanılmaktadır. Bitkisel ilaçlar, ABD’de ve tüm dünyada yaygın olarak kullanılmaya devam etmekte ve şu anda tamamlayıcı ve alternatif tıp adı altında değerlendirilmektedir. Hünnap bitkisi de içeriğinde var olan fitokimyasallar sayesinde bazı ülkelerde alternatif tıpta kullanılan başlıca bitkiler arasında yer almaktadır(Kemeç Hürkan,2019).Hünnap meyvesi sağlıklı bir gıda seçimini temsil eder. Her şeyden önce, diyet lifi ve fruktoz13 içeriği, sindirimi yavaşlatarak kan şekeri seviyelerinin düzenlenmesine katkıda bulunabilir; lif içeriği ayrıca doyurucu etkisi ile kalori alımının kontrol edilmesine de katkıda bulunabilir (Gao Qing-Han,2013).

Hünnap, lezzetli olmasının yanı sıra etkili bir bitkisel ilaç olarak görülmektedir. Kilo alımına katkı sağlamakta, kas gücünü ve dayanıklılığı artırmaktadır. Çin tıp alanında karaciğer fonksiyonunu güçlendirmek için bir tonik olarak reçetelendirilmektedir. Japon araştırmaları hünnap meyvesinin bağışıklık sistemi direncini artırdığını

göstermiştir. Çin'de yapılan bir klinik denemede karaciğer şikâyeti olan 12 hastaya hünnap, yer fıstığı ve esmer şeker verilmiş ve dört hafta sonrasında karaciğer fonksiyonlarının iyileştiği görülmüştür. Hünnap, idrar ve balgam söktürücü olarak da kullanılmaktadır. Kontrollü bir klinik çalışmada hünnap meyvelerinin kronik kabızlık için yararlı olduğu tespit edilmiştir. Ayrıca hünnap boğaz ağrısını tedavi etmek için eczacılıkta da kullanılmıştır(Koyuncu Büşra,2021).Birçok şifalı bitkinin, bağışıklık sistemini düzenleyici özelliklere sahip olduğu ve vücudun bağışıklık sistemini yeniden düzenleyerek enfeksiyonlara karşı koruduğu bilinmektedir. Alkaloidler, terpenoidler, steroidler, proteinler ve polisakkaritler gibi fitokimyasal bileşenlerin bağışıklık sistemini düzenleyici özelliği olduğu düşünülmektedir(Kemeç Hürkan,2019). Hünnap bitkisinin çeşitli kısımlarının antikanser, antiinflamatuvar, anti-obezite, antihelminthic, antioksidan, hepatoprotektif ve gastrointestinal koruyucu aktiviteler gibi biyolojik etkiler ortaya çıkardığı rapor edilmiştir (Goswami Priya,2019).

SONUÇ

Hünnap (*Ziziphus jujuba* Mill.), Rhamnaceae ailesine ait olup Ziziphus türlerinin en önemlisidir. Meyve şekli de yuvarlak, oval, elma benzeri veya anormal şekilli olabilir**.** Zengin besin içeriği sayesinde aktivite gösteren hünnabın sağlık üzerine de birçok olumlu etkisi bulunmaktadır**.** Hünnap meyvelerinin vitaminler, makro elementler (N, K, Mg, Ca, P vb.) ve mikro elementler (Fe, Mn, Cu, Zn vb.) dahil olmak üzere zengin mikro besin kaynakları olduğunu göstermiştir. Özellikle, yapılan çalışmalar sonucunda karaciğer fonksiyonunu güçlendirme, idrar ve balgam söktürücü olarak, boğaz ağrısı enfeksiyonu için bitkisel bir ilaç olarak kullanılabilirliği belirtilmiştir. Yulaf lapası, güveç, çorba veya çayda kullanılır veya hünnap ezmesi olarak da işlenebilir. Taze hünnap meyveleri, dondurma veya meyve salatası olarak da tüketime sunulabilir. Kurutulmuş hünnaplar doğrudan meyve ve fındık karışımlarında kullanılabilir veya kek, tart ya da fırın ürünlerinin üretiminde, pişirmede kullanılabilir. Ülkemizde Hünnabın üretimi yıllar geçtikçe artmaktadır ve daha da çok artırılması gerekmektedir. Hünnabın sağlık ve beslenme açısından önemi büyüktür. Bu araştırma ile hünnap meyvesi hakkında yapılacak çalışmalara ön ayak olunması hedeflenmekte ve bu meyvenin üreticilere sağlayacağı gelir ve tüketicilere de sağlıklı yaşama katkı sunması açısından öneminin vurgulanması amaçlanmıştır.

KAYNAKLAR

Çıtıl R., Sorhan S., Önder Y., EğriM. (2022). Fitoterapide Gelecek Vadeden Bir Drog; Hünnap (Ziziphus Jujuba Mill.). Bütünleyici ve Anadolu Tıbbı Dergisi,3(2),51-62.

Gao Q., Wu S., Wang M. (2013). The Jujube (Ziziphus Jujuba Mill.) Fruit: A Review of Current Knowledge of Fruit Composition and Health Benefits. Journal of Agricultural and Food Chemistry 2013;61,14,3351-3363.

Goswami P., Banerjee R., Mukherjee A. (2019). Potential antigenotoxicity assessment of Ziziphus jujuba fruit. Heliyon 2019;(5),11.

Gül E., Altuntaş E., Özgöz E. (2021). Hünnap Meyveleri ve Çekirdeklerinin Hasat Sonrası Uygulamalarla İlgili Bazı Mühendislik Özelliklerinin Belirlenmesi. Gaziosmanpaşa Bilimsel Araştırma Dergisi,10(2),1-14.

Gündoğmuş E., Taşçı M., (2017). Hünnap (Zizysphus jujube mill.) Bahçelerinde Gelir Yöntemine Göre Değerleme: Denizli İli Çivril İlçesi Örneği. Tekirdağ Ziraat Fakültesi Dergisi,14(02),42-53.

Gün S (2017). Hünnap meyvesinin (Ziziphus jujuba mill.) soğukta muhafaza performansı üzerine farklı olgunluk safhası ve modifiye atmosfer paketlemenin(map) etkisi. Ordu Üniversitesi basılmamış,70.

İkinci A, Ekinci H (2022). Hünnap (Ziziphus jujuba mill.) meyvesinin besin içeriği ve insan sağlığı açısından önemi. ASES Uluslararası Sağlık, Mühendislik ve Bilimler Kongresi,365-370.

Kavas İ (2014). Bazı hünnap genotiplerinin morfolojik, fenolojik ve pomolojik özelliklerinin belirlenmesi ve melezleme olanaklarının araştırılması. Adnan Menderes Üniversitesi basılmamış,47.

Kemeç Hürkan Y., (2019). Hünnap (Ziziphus jujuba mill.) Meyvesi: Geçmişten Günümüze Tıbbi Önemi. Iğdır Üniversitesi Fen Bilimleri Enstitüsü Dergisi,9(3),1271-1281.

Korkut S., Hişmioğulları Ş., Hişmioğulları A. (2022). Hünnap (Ziziphus jujuba) Meyvesinin Biyolojik Etkinliği ve Kimyasal Bileşimi.Balıkesir Sağlık Bilimleri Dergisi,11(1),44-50.

DRAFT

Koyuncu B (2021). Bisküvi ve kek üretiminde farklı prosesler ile kurutulmuş hünnap meyvelerinin kullanımı. Necmettin Erbakan Üniversitesi basılmamış,90.

Sunar Y (2022). Hünnap meyvesi üretim ve pazarlama yapısı: Amasya ili örneği. Ondokuz Mayıs Üniversitesi basılmamış,51.

TÜİK 2023.Bitkisel Üretim İstatistikleri, Meyve Üretim Miktarları. Erişim linki: https://biruni.tuik.gov.tr/medas/?kn=92&locale=tr [28.08.2023].

DRAFT

ORAL PRESENTATION

Referans serum panellerinin oluşturulmasının önemi

Beyhan Sareyyüpoğlu[1*] (*ORCID: https://orcid.org/0000-0002-0279-1673*)

[*1]Şap Enstitüsü Müdürlüğü, Aşı Kalite ve Kontrol Bölümü, Ankara, Türkiye

*Sorumlu yazar e-mail:beyhan.sar@gmail.com

Özet

Antikor taramasına dayalı serolojik çalışmalar, popülasyonda hastalık dinamiklerinin izlenmesinde gerek beşeri gerekse hayvan hastalıklarının kontrolünde çok önemli bir yer tutar. Dünya üzerinde farklı hastalık etkenleri için çok sayıda in-house ve ticari serolojik test kiti mevcuttur. Her bir kitin tanısal değerinin incelenmesinde pek çok parametre (sensitivite, spesifite, pozitif ve negatif prediktif değeri, tekrar edilebilirlik, intra assay-gün içi ve interassay-günler arası tutarlılığı vb.) bulunmaktadır. Test sensitivite ve spesifitesi; test sonuçlarının örneklenen hasta grubunu doğru (negatif, pozitif veya şüpheli) olarak belirleyebilmesi için öncelikle bakılması gereken iki önemli parameteredir. Serolojik antikor tarama ticari test kitlerinin ya da yeni laboratuvarda geliştirilen bir in-house test kitinin tanısal değerinin doğru olarak tespit edilmesinde referans serum panellerinin hazırlanması önemlidir. Bu bildiri de beşeri ve hayvan sağlığında antikor tanı kitlerinin değerlendirilmesinde kullnılacak referans serum paneli hazırlanmasında nasıl yol izleneği üzerinde durulmuştur.

Anahtar Kelimeler: Beşeri, hayvan, seroloji, serum, panel

Abstract

Serological studies based on antibody screening play a very important role in monitoring the dynamics of diseases in the population both in the control of human and animal diseases. There are many in-house and commercially available serological tests for different disease agents around the world. There are many parameters (sensitivity, specificity, positive and negative predictive value, repeatability, intra and interassay consistency, etc.) for evaluation for kit diagnostic value. Test sensitivity and specificity are two important parameters that should be looked at first in order for the test results to determine the sampled patient group correctly (negative, positive or suspicious). In order to accurately determine the diagnostic value of serological antibody screening commercial test kit or a newly developed in-house test, it is to prepare reference serum panels. In this paper, how to prepare a reference serum panel to be used in the evaluation of antibody diagnostic kits in human and animal health was discussed.

Keywords: Human, animal, serology, serum, panel

GİRİŞ

Antikor tarama serolojik testleri, hastalık dinamiklerinin izlenmesinde insan ve hayvan hastalıklarının kontrolünde çok önemli bir yer tutar (Burgu ve Akça, 1983; Gürhan ve ark, 1984; Presta, 1992; Haselbeck ve ark., 2022). COVID-19 pandemisi de serolojik testlerin önemini tekrar vurdulamıştır (Winter ve Hedge, 2020; Kaya ve ark., 2022). Hastalık etkenine yönelik antikor tespiti, popülasyonda hastalık taraması ve izlenmesi yanında ayrıca aktif hastalık etkeninin tespit edilemediği durumlarda anahtar rol oynayan tanı enstümanlarından biridir.

Dünyada sadece önemli enfeksiyoz ajanları dikkate aldığımızda farklı hastalık etkenleri için gerek beşeri gerekse hayvan sağlığı açısından çok sayıda ticari ve in house antikor dayalı serolojik test test kiti mevcuttur (Öztürk, 1997; Pinto ve ark., 2011; 2019[b]; Infantino ve ark., 2020). Her bir tanı kitinin değerlendirilmesinde olduğu gibi serolojik kitlerin de değerlendirilmesinde temel parametreler testin sensitivite ve spesifitesi yanında, test tekrarlanabilirliği, pozitif/negative prodüktif değeri, intra-inter assay (gün içi ve günler arası

tutarlılık) gibi pek çok parametreye bağlıdır (Parikh ve ark., 2008; Mallett ve ark., 2012; Trevethan, 2017; 2019[b]). Testin tanısal değerinin yüksek olması, test sonuçlarının örneklenen hasta grubunu doğru (negatif, pozitif veya şüpheli) olarak belirleyebilmesi için elzem gereksinimdir. Serolojik antikor tespit edici ticari test kitlerinin ya da yeni geliştirilen in house test kitinin tanısal değerinin doğru olarak tespit edilmesinde, referans serum panellerinin hazırlanması önemlidir. Bu bildiri de sadece referans serum paneli hazırlanmasında dikkat edilecek noktalar üzerinde durulmuştur.

Hayvan sağlığında, Dünya Bulaşıcı Hayvan Hastalıkları Manualinde (OIE Manual), referans panellerin seçimi ve hazırlanması ile ilgili genel kısımda; serum örnekleri için yer verilmiş kısımlar yer almaktadır (OIE 2019[a]). Brucella, Tüberculoz gibi zoonoz hastalık etkenleri için OIE referans Laboratuvarı (ANSES)' de etkene yönelik serum referansları koleksiyon halinde sunulmuştur. OIE' nin ayrı bir bölümünde "antikor belirlemeye yönelik serolojik testlerinin optimizasyonu" kısmında referans serumlarının hazırlanma koşullarından bahsedilmektedir (OIE 2019[c])

Öncelikle "referans serumun" tanımı ardından serum panelini toplarken kullanılacak referans popülasyonlardan ve serumun porsiyonlanma ve saklanmasından bahsedilecektir. **Referans serum**, bir pathogen etkenin ilgili referans suşuna karşı hazırlanan anti-serumdur (Wright ve ark., 1993; OIE 2019[a]). Bir seroloijk test ilk defa geliştirildiğinde, testin değerlendirilmesinde **referans serum veya referans standart kullanılır.** Referans serum bilinen düzeyde ve aktivitede aranılan antikoru içermesi gerekir. Bu genelde antikor titresi tespit edilerek yapılır. Bu amaçla serumu tanımlamak için (titresine göre kuvvetli, zayıf, negatif serum gibi) etkene yönelik bilinen altın standart testler kullanılmalıdır (Virus Nötralizasyon Testi-VNT, ELISA vd). Çoğu enfeksiyoz ajan için internal referans standart serumlar ANSES'de (OIE referans laboratuvarı) bulunmaktadır. Daha önce de belirtildiği gibi uluslararası koleksiyonlarda bulunmayan referansların, özellikle ilgili enfeksiyoz ajan için hastalık dinamiği yüksek endemik ülkelerde, ülke tarafından referans serumlarının hazırlanması gerekmektedir.

Pozitif ve negatif referans serum setleri; serolojik test kitinin amaçlanan çalışma aralığı veya diğer ifadeyle dinamik aralığın üzerindeki veya altındaki antikor konsantrasyonlarını içeren bu serumlar (kuvvetli pozitif veya negatif), bir antikor testinin geliştirilmesi ve/veya standardizasyonu boyunca kullanılmalıdır. Kitin doğrulamanın çeşitli yönlerinde kullanılabilmeleri (tekrar edilebilirlik, analik limit vb.) için yeterli miktarlarda hazırlanmaları önerilmektedir. pozitif örnekler; hedef popülasyonda bilinen enfekte ve enfekte olmamış (naif) negatif hayvanları temsil etmelidir. Tercihen tek tek hayvanlardan toplanmalıdırlar, fakat birkaç hayvandan da örnek havuzu (örnek pool) oluşturulabilir (OIE 2019[b], 2019[c])

Bilinen enfekte ve enfekte olmamış referans hayvanlardan toplanan serum setleri-eşit olmayan referans standart; tanısal sensitivite ve spesifiteyi belirlemek için ideal referans serum örnekleridir. Bu serum panelleri, "altın standart" olarak tanımlanan testler ile öncelikle doğrulaması yapılmış serumlardır. Bununla birlikte, bu tür örneklerin oluşturulması zordur. Çünkü, bazı etkenler için altın standart test yoktur (Abiha ve Kanık, 2016). Bu panel tanısal sensitivenin (konfirme enfekte hayvanları içeren serum paneli yani pozitif serum paneli) ve spesifitenin (konfirme enfekte olmamış hayvanları içeren serum paneli yani negatif serum paneli) belirlenmesi için kullanılır. Bu panellerin hazırlanmasında bazı durumlar dikkatlice irdelenmelidir. Bu tip serum örneklerinin toplanmasında sahada hayvan/insan klinik belirti veriyordur ancak organizmada antikor şekillenmemiştir. Enfeksiyonun erken döneminde toplanmış olabilir. Aksine durum olarak, enfeksiyonun ilerleyen döneminde kronik süreçte toplanan örnekler çok yüksek düzeyde antikor içeriyor olabilir. Değerlendirilen test kitinin yanlış-yüksek düzeyde sensitivite değerinde görünmesine neden olabilir. Bu nedenle, enfeksiyonun pozitif olduğu bir popülasyonda; hastalığı değişik düzeyde geçiren (eşit olmayan antikor düzeyine sahip bireyler), özellikle zayıf pozitif bireylerden de yeterli miktarda serum toplanmalıdır. Genelde toplanmasının ve belirlenmesinin güçlüğü nedeniyle serum panellerinde zayıf pozitif serum panelleri atlanmaktadır, üretilen bazı kitler içinde zayıf pozitif serumlar maalesef referans olarak yer almayabilir (Jacobson, 1998; OIE 2019[c] ; Edouard ve ark., 2021).

Naif veya enfeksiyona maruz kalmamış hayvanlardan toplanan serum paneli (negatif serum paneli)
Bu amaçla, ilgili enfeksiyonun olmadığı coğrafyalardan serum toplanır. Bu panel için toplanan birey ya da bir kaç bireyden oluşturulan-pool serumda spesifik antikor olmadığı; popülasyon klinik gözlem öyküsü,

önceki test sonuçları incelenerek altın standart test ile gösterilmelidir. Tanısal spesifite tespitinde kullanılan temel serum panelidir (Jacobson, 1998; OIE 2019[c]).

Rölatif referans standart; Bu standart, aynı numuneler üzerindeki başka bir serolojik tahlilin test sonuçlarına kıyaslanarak, enfeksiyon durumlarına göre sınıflandırılmış referans hayvanlarla karakterize edilir. Genellikle yeni bir serolojik testin değerlendirilmesi için mevcut olan tek pratik referans materyal kaynağıdır. Ancak ortaya çıkan tanısal sensitivite ve spesifite tahminleri, yalnızca referans testin belgelenebilir, belirlenmiş ve kabul edilebilir performans özelliklerine sahipse kabul edilebilir, yani görecelidir. Göreceli referans standardın getirdiği doğal önyargı tanı kitini değerlendirirken dikkate alınmalıdır (OIE, 2008; OIE 2019[c]).

Deneysel enfeksiyon veya aşılama ile oluşturulmuş serum paneli (Ek referans standart). Sahada enfeksiyon olmayabilir ya da daha hızlı ve kontrol edilebilir (enfeksiyon sonrası ve öncesi sürelerin hesaplanması sorunu) pozitif örnekler elde etmenin tek yolu deneysel enfeksiyondur. Bu yaklaşım, teorik olarak çok iyi bilinen enfeksiyonun dinamiklerini modellemek için uygundur. Ancak aynı hayvanlardan tekrarlanan gözlemleri temsil eden veriler, sensitivite ve spesifite hesaplanmasında kullanılamaz. Tanısal sensitivite ve spesifiteyi oluşturmak için kullanılan istatistiksel modeller bağımsız gözlemler gerektirir (her hayvandan yalnızca bir örnek). Deney hayvanlarının kaynakları ve tarihçesi, enfeksiyon sonra serumun kaçıncı gün toplandığı çok iyi bilinmelidir. Deneysel enfeksiyonun hiçbir zaman sahadaki doğal enfeksiyonu tam olarak ifade edemeyeceği bilinmelidir. Bu nedenle kitin tanısal değerine deneysel olarak toplanmış serum paneliyle karar verilmemelidir (OIE 2019[c] Ringa ve Bauch , 2014) .

Yukarıda serum setlerinin hazırlanmasında kullanılabilecek hedef popülasyon örnekleri, ağırlıklı olarak hayvan sağlığı referans serum panelleri için önerilmiştir. İnsan sağlığında referans serum hazırlanmasında; primer ve sekonder setler olarak tanımlamalar bulunur. Primer referans setleri, Dünya Sağlık Teşkilatı (WHO) oluşturmaktadır. Özellikle COVID-19 pandemisinin de etkisiyle sekonder serum referans panelleri adıyla WHO bir manual hazırlamıştır (WHO, 2022). Manual içindekiler kısmı incelendiğinde, aday serumun seçiminden, serumların kalibrasyon ve etiketlenip stoklanmasına kadar detaylı hazırlanmış bir manual bulunur.

Gerek insan gerekse hayvan sağlığında hazırlanan serum panellerinin, hazırlandıktan sonra uzun yıllar (en az 3-5 yıl arası) kullanılabilmesi için mümkün olduğunca yüksek miktarda toplanması, uygun biçimde porsiyonlanması ve stabilitesinin sağlanması en önemli konudur. Genelde serum porsiyonlandıktan sonra -20 ^{0}C'de stoklandığında uzun süre etkinliğini kaybetmez. Porsiyonlar test kitlerinin değerlendirilmesi için kullanılırken dondur-çözdür yapılmaması önerilir. O nedenle referans serumlar genel olarak -20 ^{0}C'den çıkarıldıktan sonra bekletilmemesi, kullanıldaktan sonra atılması ya da kısa süreler (max 1 hafta) +4 ^{0}C'de bekletilebilir. Çoğunlukla liyofilize paneller de hazırlanabilir. Ancak liyofilizasyon işlemi sırasında ürün hacim ve etkinlik kaybı olabileceğinden bu konu detaylı bir biçimde irdelenmelidir (Marcovovina ve ark., 1990; OIE 2019[a], OIE 2019[c] ,WHO 2020)

SONUÇ

Sonuç olarak, ülkemizde ve dünyada var olan enfeksiyonlar ve hızla yeni enfeksiyonların gözlenmesi, hastalık tarama stratejisinin önemli bir aracı olarak serolojik antikor tarama testlerinin daha doğru değerlendirilmesini gerektirmektedir. Bu nedenle enfeksiyoz hastalıklara yönelik referans serum setlerinin hazırlanması önemlidir. Bu çalışmada gerek hayvan gerekse insan sağlığına yönelik kullanılan serolojik tanı kitlerinin değerlendirilmesinde referans serum panellerinin hazırlanmasında nasıl bir yol izleneceği derlenmiştir.

KAYNAKLAR

Burgu İ, Akça Y 1983. Sığırlarda rotavirus enfeksiyonlarının dağılımı üzerine araştırmalar. Ankara Üniversitesi Veteriner Fakültesi Dergisi, 30 (1): 35-44.

Abiha ve Kanık 2016. Tanısal mikrobiyolojide altın standart testin yokluğunda gizli sınıf analizinin kullanımı. Arşiv Kaynak Tarama Dergisi, 2016; 25(4):467-488.

Crowther JR, Unger H, Viljoen GJ 2006. Aspects of kit validation for tests used for the diagnosis and surveillance of livestock diseases: producer and end-user responsibilities. Revue Scientifique Technical Offiice International Epizootic, 25 (3): 913– 935.

Edouard S, Colson P, Melenotte C, Pinto CF, Thomas L, La Scola B, Million M, Tissot-Dupont H, Gautret P, Stein A, Brouquil P, Parola P, Lagier JC, Raoult D, Drancourt M 2021. Evaluating the serological status of COVID-19 patients using an indirect immunofluorescent assay, France. European Journal of Clinical Microbiology Infectious Diseases, 40:361–371.

Gürhan B, Şenel E, Dalkılır G, Öztürkmen H 1984. Şap aşılı sığırlarda mikronötralizasyon ve ELISA ile antikor düzeylerinin saptanması. Journal of Etlik Veterinary Microbiology, 7(5): 99-108.

Haselbeck AH, Im J, Prifti K, Marks F, Holm M, Zellweger RM 2022. Serology as a tool to assess infectious disease landscapes and guide public health policy. Pathogens, 11(7):732.

Infantino M, Damiani A, Gobbi FL, Grossi V, Lari B, Macchia D 2020. Serological assays for SARS-CoV-2 infectious disease: benefits, limita- tions and perspectives. Israel Medical Assocciation Journal, 22(4):203–10.

Kaya S, Akbayırlı U, Arkalı T 2022. Fabrika çalışanlarının COVID-19 antikor tarama sonuçlarının değerlendirilmesi. Türk Mikrobiyoliji Cemiyeti Dergisi, 52(4):310-315.

Jacobson RH 1998. Validation of serological assays for diagnosis of infectious diseases. Revue Scientifique Technical Offiice International Epizootic, 17: 469–486.

Mallett S, Halligan S, Ompson M, Collins GS, Altman DG 2012. Interpreting diagnostic accuracy studies for patient care. BMJ. 345: e3999.

Marcovina SM, Adolpson Jl, Parlavecchia M, Alberts JJ 1990. Effects of lyophilization of serum on the measurement of apolipoproteins A-I And B. Clinical Chemistry, 36(2):366-9.

OIE 2008. Standard for management and technical requirements for laboratories conducting tests for ınfectious diseases. In: OIE Quality Standard and Guidelines for Veterinary Laboratories: Infectious Diseases. OIE, Paris, 1–31.

OIE 2019[a]. Validation guideline, selection and use of reference samples and panels. Available at: https://www.oie.int/fileadmin/Home/eng/Health_standards/aahm/current/GUIDELINE_3.6.6_REFERENCE_SAMPLES.pdf [03.05.19]

OIE 2019[b]. Registration of diagnostic kits. Available at: http://www.oie.int/our-scientific - expertise/registration-of-diagnostic-kits/background-information. Accessed September 2013 [12.03.22]

OIE 2019[c]. Development and optimization antibody detection assays. Available at: https://www.woah.org/fileadmin/Home/fr/Health_standards/tahm/2.02.01_ANTIBODY_DETECT.

Öztürk S 1997. İmmunolojik tanı kitlerinin geliştirilmesi, Doktora Tezi. Availbale at: https://www.proquest.com/docview/2606903615?pq-origsite=gscholar&fromopenview=tru

Parikh R, Mathai A, Parikh S, Sekhar GC, Omas R 2008. Understanding and using sensitivity, specificity and predictive values. Indian Journal Ophthalmology, 56:45–50.

Pinto LM, Greiner J, Schumacher SG, Denkinger CM, Steingart KR, Pai M 2011. Immunodiagnosis of tuberculosis: state of the art. Medical Principles Practice, 21 (1): 4–13.

Presta LG 1992. Antibody engineering. Current Opinion in Structural Biology, 2(4):593-596.

Ringa, N, Bauch, CT 2014. Dynamics and control of foot-and-mouth disease in endemic countries: a pair approximation model. Journal Theorotical Biology 357: 150-9.

Trevethan R 2017. Sensitivity, specificity and predictive values: foundations, Pliabilities, and Pitfalls in Research and Practice. Frontiers, 5: 00307

WHO 2022. WHO Guidelines On Secondary Standards For Antibody Testing. Available at: (Https://Cdn.Who.İnt/Media/Docs/Default-Source/Biologicals/Annex-2---Who-Guidelines-On-Secondary-Standards-For-Antibody-Testing---11-May-2022.Pdf?Sfvrsn=C0d1c8ce_1&Download=True) [06.08.22]

Winter AK, Hedge S 2020. The important role of serology for COVID-19 control. Lancet Infectious Disease, 20(7): 758–759.

Wright PF, Nilsson E, Van Rooıj EM, Lelenta M, Jeggo MH 1993. Standardization and validation of enzyme linked immunosorbent assay techniques for the detection of antibody in infectious disease diagnosis. Revue Scientifique Technical Office International Epizootic, 12: 435–450.

DRAFT

ORAL PRESENTATION

Example of sustainable approach in the textile industry: Olive mill wastewater

Sabri Can TUZ[1] (https://orcid.org/0000-0002-8650-946X), Aslı DEMİR[1] (https://orcid.org/0000-0002-6642-1604),
Murat ELİBOL[2] (https://orcid.org/0000-0002-6756-6290)

[1]Ege University, Faculty of Engineering, Departmant of Textile Engineering, Izmir, Türkiye
[2]Ege University, Faculty of Engineering, Departmant of Bioengineering, Izmir, Türkiye

sabricantuz1999@gmail.com

Abstract

Olive oil consumption has been increasing in recent years due to its beneficial effects on health (Yılmaz and Özcan; Bayram and Özçelik, 2012). Our country is one of the most important olive producing countries in the world. According to the International Olive Council data, olive oil production in Turkey in the 2021-2022 season is determined as 227,500 tons (Özkan, 2022). In olive oil production enterprises, three phases emerge at the end of the process: olive oil, solid waste and liquid waste (olive mill wastewater). Approximately 1 ton of liquid waste is discharged for 1 ton of olives (Beccari et al., 1996; Elibol et al., 2008). The composition of olive mill wastewater includes oil, tannins, polyphenols, polysaccharides and proteins. In addition to the high chemical and biological oxygen demand, the olive mill wastewater formed during olive oil extraction creates a significant environmental burden, especially with the oil in its composition, dark color and bad odor. Direct discharge of olive mill wastewater into soil or streams without any treatment brings about significant environmental problems (Elkacmi and Bennajah, 2019). However, there are components in the composition of olive mill wastewater that can be recovered and have economic value. Thus, it can be a resource with economic value instead of waste. In this study, the possibilities of using olive mill wastewater, which is abandoned to nature as waste with high environmental burden, in the textile industry were examined. By reusing waste, an alternative sustainable method has been introduced to coloring which is done using water and various chemicals in classical production. An attempt was made to reveal a sustainable method by performing physical and biological characterization of the dyed textile materials. Additionally, the environmental impact (carbon and water footprint, etc.) of dyeing made in this way was analyzed and compared with the conventional method. Thus, by utilizing olive mill wastewater, which creates a significant environmental burden, in the textile industry, a waste product will be converted into different resources, contributing to the sustainable production approach.

Keywords: Olive mill wastewater, waste management, textile, sustainability

INTRODUCTION

Olives are an important and traditional product in Mediterranean countries since ancient times and hold an important place in the economy of their countries. The most important product of the olive plant is olive oil (Aslan, 2017). The production of olive oil has been increased rapidly in response to growing demand of olive oil (Doğan, 2021). 90% of the world's olive cultivation is done in Mediterranean countries (Aslan, 2017). Turkey, with its geographical location and Mediterranean climate characteristics, is one of the world's leading olive and olive oil producers, along with other Mediterranean countries such as Spain, Italy, Greece and Tunisia (Apohan, 2007). Olive oil is produced from olive trees that yield between 15 and 40 kg of olives per year from each olive tree (Paraskeva and Diamadopoulos, 2006; Tunç and Ünlü, 2015; Avcı, 2016). Considering that 1 liter of olive oil is extracted from an average of 5 kg of olives, 1 olive tree can produce an average of 3 L or 4 L of olive oil per year (Apohan, 2007).
The production of olive oil yields a considerable amount of waste water, which is a powerful pollutant and is currently discarded (Mulinacci et al., 2001).

DRAFT

Two different methods are used in olive oil production: batch (press) and continuous (centrifuge). In both methods, two by-products such as pomace and olive mill wastewater are formed as a result of production (Elibol et al., 2008). The amount and characteristics of olive mill wastewater released during olive oil production; It varies depending on the olive oil production process, the type of olive processed, the type of pesticide and fertilizer used during olive cultivation, the climate of the growing region and the years (Değirmenbaşı, 2016).
Mediterranean countries produce 30 million m^3 of wastewater annually during the extraction of olive oil from olives. More than 400,000 tons of olive mill wastewater are produced annually in Turkey (Apohan, 2007).

OMWW is characterized by a brownish black color, slightly acidic nature (pH 3-6), strong specific olive oil odor, high electrical conductivity, and high content of polyphenols (0.4-5.45 %), reducing sugar (1.5-19.3 %), potassium as a major inorganic element (4 g/L), organic load (0.8-220 g/L COD; 0.3-110 g/L BOD; 25-45 g/L TOC) and suspended solids (1-9 g/L) (Çelik, 2018).

Olive mill wastewater covers the surface in environments where it is discharged, preventing the oxygen exchange necessary for living things (Avcı, 2016).

The most important reasons for the difficulties experienced in olive mill wastewater treatment are; this water contains toxic substances such as high organic matter and polyphenols, seasonal production is made and a production season lasts about 3-4 months (Elibol et al., 2008; Rozzi and Malpei, 1996). Many studies have been carried out and different treatment technologies have been developed to eliminate or reduce the polluting effect of this wastewater. Various methods have been applied to purify and dispose of olive mill wastewater (Elibol et al., 2008; Kasırga, 1988). Since olive mill wastewater contains components that can be recycled and have economic value, wastewater should be seen as a resource with economic value, not a waste, in disposal scenarios (Elibol et al., 2008; Vlyssides et al., 2004).

Based on this point and some preliminary studies, it has been seen that olive oil wastewater can also be considered as a natural dyestuff for dyeing textile materials. Utilizing olive mill wastewater in this way results in finding a zero-cost, high-quantity and renewable alternative natural dye.

MATERIALS AND METHODS

Some physical properties of olive mill wastewater are examined. The values were given in Table 1.

Table 1. Some physical properties of olive mill wastewater (Elibol et al., 2008)

pH	8
Chemical Oxygen Demand (g/l)	80
Biochemical Oxygen Demand (g/l)	55
Suspended Solids (g/l)	15
Oil-Grease (mg/l)	400
Phenol (Caffeic Acid) (mg/l)	3500

100% cotton woven fabric was used in this study. The properties of the cotton fabric is given in Table 2.

Table 2. The Properties of The Cotton Fabric

Fabric	Weight	Density	Yarn Count
Woven Cotton	126,1 g/m^2	Weft: 30 tel/cm Warp: 47 tel/cm	Weft: Ne 39/1 Warp: Ne 41/1

Before being treated with olive mill wastewater, cotton fabrics were pre-treated to increase color yield. The results of different pretreatmned were compared each other. The fabric samples were pretreated with chitosan and mordanted by potassium aluminium sulfate (KAl(SO4)2).

For pre-treatment with chitosan, cotton fabrics were impregnated with chitosan solutions at 1% and 2% concentration, prepared by dissolving in 1% acetic acid (CH3COOH), to AF = 100%, and then dried at 40°C.

For the mordanting process, cotton fabrics were treated with distilled water containing 10% and 20% potassium aluminium sulfate at boiling temperature for 1 hour.

All dyeing processes were carried out in a water bath at a liquor ratio of 1:60, according to the specified dyeing conditions.

The color yield (K/S) measurement of the dyed fabrics was determined on a spectrophotometer. K/S values obtained as a result of the analysis are calculated according to the Kubelka Munk equation given below. At least 3 measurements were made for each sample and the average of these measurements was taken. K/S values at the maximum absorption wavelength of dyed fabrics were taken into account.

Kubelka Munk: K/S = (1-R)2 / 2R

K: Light absorption

S: Light reflection

R: Remission value at maximum absorption wavelength

The Fourier transform infrared (FTIR) spectra of cotton fabrics were determined by means of a Perkin Elmer 100 FTIR spectrometer in the attenuated total reflectance (ATR) mode using a diamond/zinc selenide crystal.

RESULTS AND DISCUSSION

FTIR Results

The FTIR of cotton fabrics that have undergone different treatments results are shown in Figure 9.

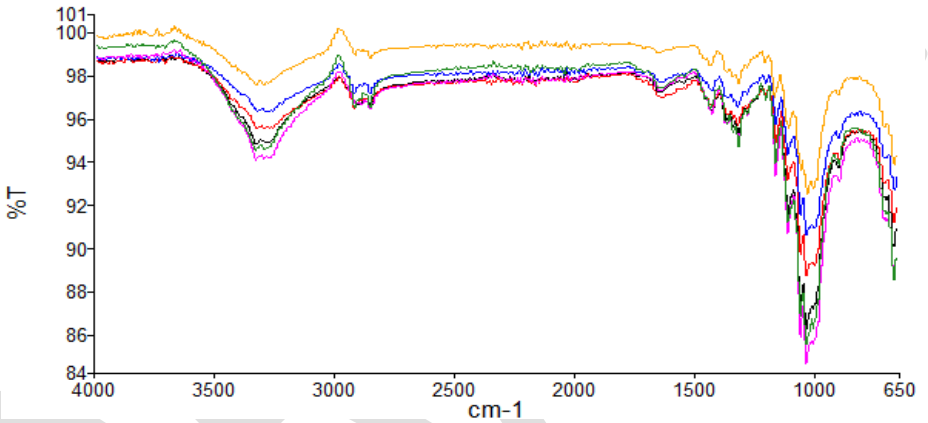


Figure 9. The FTIR of cotton fabrics that have undergone different treatments

The peaks in the range of 3300-3100 cm-1 in cotton fibers belong to the hydroxyl (-OH) groups of cellulose. The C-H stretching peak in the 2800-3000 cm-1 region, and the peaks at 1600-1650 cm-1 belong to the asymmetric COO- or C=O peak. The absorption band around 1050-1030 cm-1 originates from different functional groups of cellulose (C-C, C-O and C-O-C vibrations) (Dilsiz, 2016; Gündüz Balpetek, 2017; Hartzell-Lawson and Hsieh, 2000; Uğur and Sarıışık, 2015; Türemen, 2017).

From the FTIR spectra, it can be seen that there are characteristic peaks related to the cellulose structure in cotton fibers (around 3300, 1050 and 1035 cm-1) (Dilsiz, 2016; Hartzell-Lawson and Hsieh, 2000; Türemen, 2017).

Dyeing Results

pH Optimization: The pH value of olive mill wastewater was measured as 8. To see the effect of pH change in the dye bath, the pH value was changed to 4-5-6, and dilute formic acid solution was used to adjust the pH. During the dyeing process, the temperature was kept constant at 100°C and the duration was 60 minutes.

	pH	**Temperature**	**Time**
pH Optimization	**4-5-6**	100°C	**60 Minutes**

K/S values of cotton fabrics after the above mentioned dyeing conditions are given in Figures 1 and 2. Measurements were carried out at a wavelength of 360 nm.

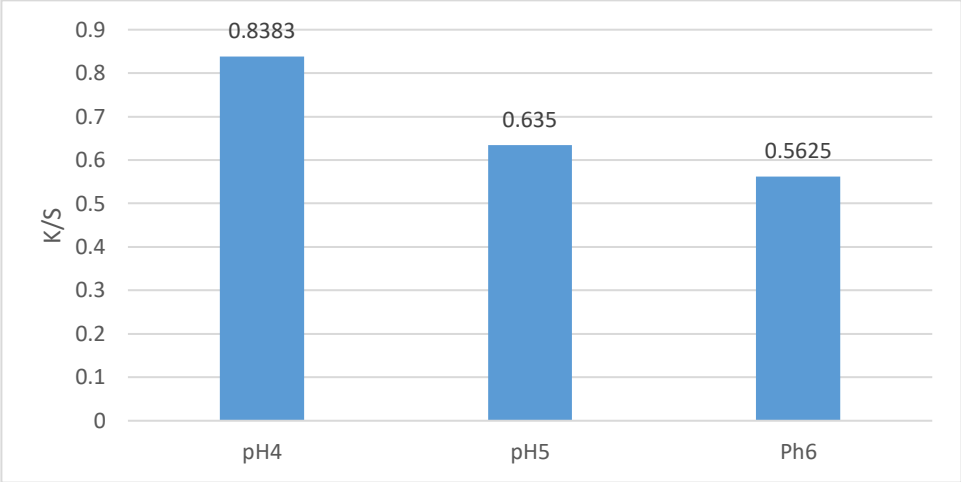


Figure 1. The effect of pH change on K/S values of cotton fabrics

The color tones of cotton fabrics dyed at different pH values are shown in Figure 2.

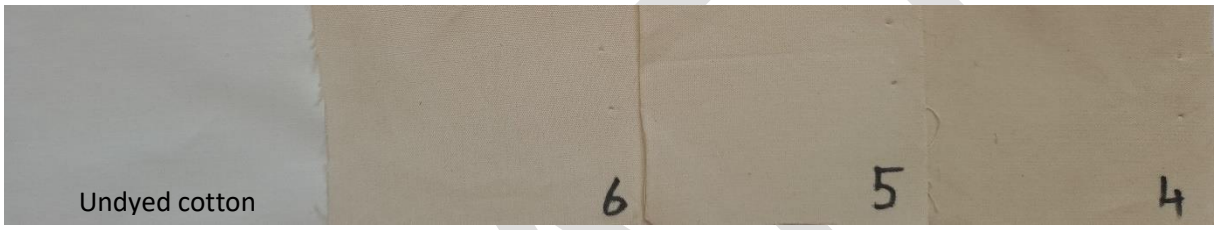


Figure 2. The color tones of cotton fabrics dyed at different pH values

Time Optimization: In order to see the effect of the time change, 3 different periods were chosen: 30-60-90. pH was kept constant at 4 and temperature at 100°C.

	pH	Temparature	Time
Time Optimization	4	100°C	30-60-90 m

K/S values of cotton fabrics after the above mentioned dyeing conditions are given in Figures 3 and 4. Measurements were carried out at a wavelength of 360 nm.

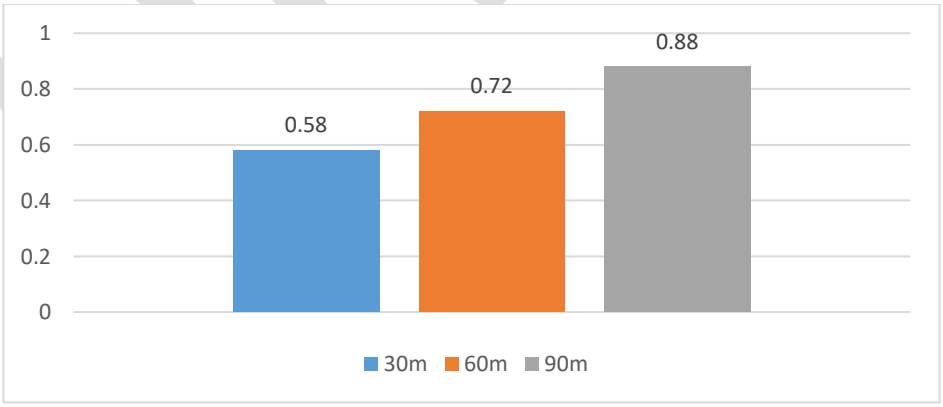


Figure 3. The effect of time change on K/S values of cotton fabrics

The color tones of cotton fabrics dyed for different periods of time are shown in Figure 4.

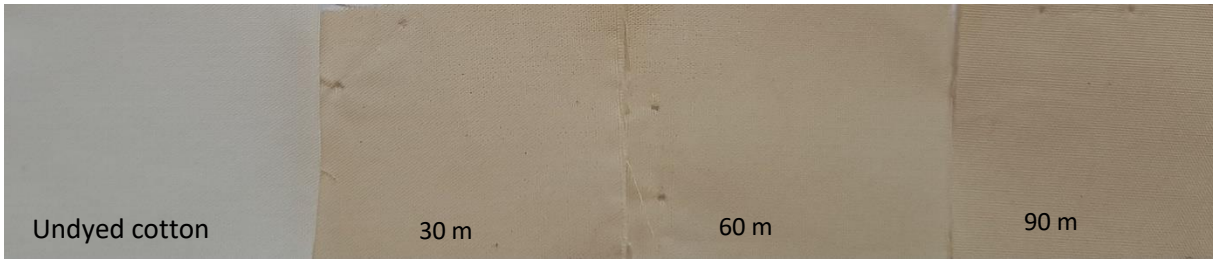


Figure 4. The color tones of cotton fabrics dyed at different times

Temperature optimization: To see the effect of temperature change, three different temperatures were chosen: 60-80-100°C. pH was kept constant at 4 and time at 60 minutes.

	pH	Temparature	Time
Temparature Optimization	4	60-80-100°C	60 Minutes

K/S values of cotton fabrics after the above mentioned dyeing conditions are given in Figure 5 and 6. Measurements were carried out at a wavelength of 360 nm.

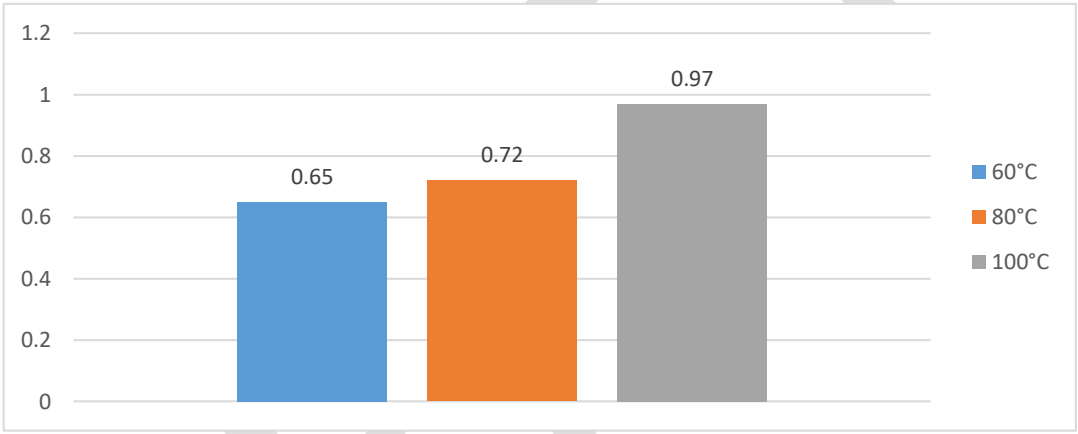


Figure 5. The effect of temperature change on K/S values of cotton fabrics

The color tones of cotton fabrics dyed at different temperature values are shown in Figure 6.



Figure 6. The color tones of cotton fabrics dyed at different temperatures

As a result of the optimization made by changing pH, temperature and time in dyeing cotton fabrics with olive mill wastewater, it was determined that the best color values were obtained in dyeing at pH 4, 100°C temperature and 60 minutes.

The K/S values of the dyeing of cotton fabrics dyed in different processes according to optimization conditions are given in Figures 7 and 8. Measurements were made at 360 nm wavelength.

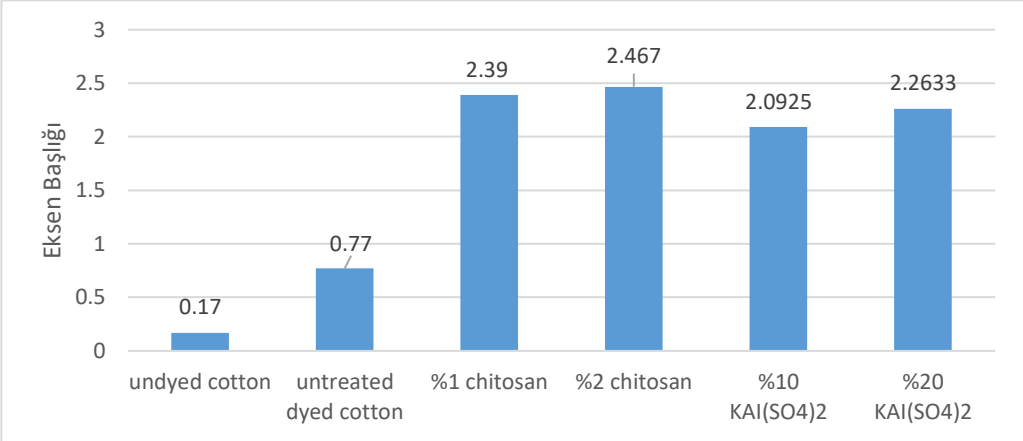


Figure 7. The effect of temperature change on K/S values of cotton fabrics

The color tones of dyed cotton fabrics that have undergone different treatments values are shown in Figure 8.



Figure 8. The color tones of dyed cotton fabrics that have undergone different treatments

It has been determined that cotton fabric can be dyed without using salt or any other chemicals after pre-treatment with chitosan. Thus, the use of chitosan can reduce the salt load mixed into agricultural lands and groundwater by reducing the pollution of wastewater originating from the textile industry.

As a result, dyeing of cotton fabrics with olive mill wastewater with the help of chitosan; by utilizing the shell waste left over from creatures consumed as food, such as crayfish and shrimp, it will provide many advantages such as reducing the amount of waste and turning it into a value-added product, reducing dyeing process costs and dyeing time, consuming less energy, and reducing the use of auxiliary chemicals used in dyeing (Yüce et al., 2020).

In the use of products obtained from cotton fiber in natural dyeing technology, cotton fiber has some structural limitations such as weak dye interaction, lower dye uptake and low color fastness. There are two reasons why cotton fibers are difficult to dye with natural dyes compared to protein fibers. The first of these reasons is that cotton fiber is cellulose-based and has a high crystalline region ratio, which reduces the cellulose dye population. The other reason is D-glucopyranose, the monomer of cotton fiber, which contains dehydrated hydroxyl groups and ether bonds with negative surface density. This high negative charge density makes it unsuitable for dyeing with negatively charged polyphenolic dyes.

Mordanting is one of the processes used to overcome these disadvantages in dyeing cotton fiber with natural dyes (Merdan and Eyüpoğlu, 2023). The process of binding dyestuffs to textile fibers is called mordanting, and the substances used for this purpose are called mordant substances. Some of the natural dyestuffs change their colors after complexing with certain metallic cations (Aluminium (Al+3), Iron (Fe+2), Calcium (Ca+2) and Tin (Sn+2)). Most natural dyestuffs have low light and washing fastness. Therefore, in order to increase fastness, it is subjected to another process with appropriate chemicals before, during or after dyeing. Water-soluble metal salts and substances with weak acid or base properties are used as mordant substances (Avcı, 2016).

In addition, the mordanting process applied to cotton fiber aims to minimize the negative repulsive force exerted by the cotton fiber monomer on the natural dyestuff molecules (Merdan and Eyüpoğlu, 2023).

Potassium aluminium sulfate, an ecological mordant, was used in this study. The experiment results are given in Figure 7. From the experiment results, it is seen that as a result of pre-treatment with 10 and 20% mordant and 1 and 2% chitosan, the color yield values increased by 172%, 220%, 210% and 194%, respectively, compared to the untreated dyed fabric.

CONCLUSION

In the industry, some waste materials that are generated when raw materials are processed for a specific purpose and cause environmental pollution need to be eliminated or their pollution load should be reduced. However, the economic evaluation of these wastes, if possible, has become even more important, especially in recent years.

Based on this understanding, in this study, studies were carried out on the use of olive mill wastewater as a dyestuff in textiles. In the studies, the best dyeing conditions for woven fabric containing 100% cotton were first determined by optimization (temperature, time and pH). Then, cotton fabrics that had undergone different treatments (1% and 2% chitosan, 10% and 20% potassium aluminium sulfate (KAl(SO4)2)) were dyed under the best conditions and their dyeing efficiencies were compared.

As a result of the optimization made by changing the pH, temperature and time in dyeing the fabrics with olive mill wastewater, the test results with the best color values were obtained at pH 4, 100°C and 60 minutes. As a result of pre-treatment with mordant and chitosan, it is seen that even the use of low amounts increases the dyeing efficiency by 172% and 210%, respectively.

It can be seen from color measurements that olive mill wastewater, which is a waste and brings with it significant environmental problems, can be evaluated in the textile industry as an alternative natural dyestuff.

The study is a part of the project and still ongoing. The experiments about biological properties will be carried out in the future.

ACKNOWLEDGEMENTS

This study was supported by Ege University Scientific Research Projects Coordination Unit (Project number 29729).

REFERENCES

Apohan E 2007. Vinas ve zeytin yağı fabrikası atık suyunun değerlendirilmesi ve biyolojik iyileştirilmesinde beyaz çürükçül fungus peletlerinin kullanımı. T.C. İnönü Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi, Malatya.

Aslan Ç 2017. Zeytinyağı Atık Ürünü Zeytin Kara Suyunun Bacillus Subtilis ATCC 6633'ün Gelişiminde Besi Ortamı Olarak Kullanılabilirliğinin Araştırılması. T.C. Mustafa Kemal Üniversitesi Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi, Hatay.

Avcı BB 2016. Zeytinyağı Üretimi Atıklarının Tekstil Boyacılığında Kullanım Olanaklarının Araştırılması. Dokuz Eylül Üniversitesi Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi, İzmir.

Bayram B, Özçelik B 2012. Zeytinyağının Biyoaktif Bileşenleri ve Sağlık Üzerine Yararları. Akademik Gıda, 10(1): 77-84.

Beccari M, Bonemazzi F, Majone M, Riccardi C 1996. Interaction between acidogenesis and methanogenesis in the anaerobic treatment of olive oil mill effluents. Water Research, 30(1): 183-189.

Çelik G 2018. Multistage recovery process of biophenolic antioxidants with focus on hydroxytyrosol from olive mill wastewater concentrates. Boğaziçi University Institute of Environmental Sciences, Master's Thesis, Istanbul.

Değirmenbaşı D 2016. Zeytinyağı fabrikası sıvı atığından antioksidan enzimler üretimi, Ankara Üniversitesi Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi, Ankara.

Dilsiz YF 2016. Pamuğa Kimyasal Modifikasyon Uygulayarak Multicolor Efektine ve Antibakteriyellik Özelliğine Sahip Fonksiyonel Gömleklik Kumaş Eldesi. Namık Kemal Üniversitesi Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi, Tekirdağ.

Doğan S 2021. Recovery and Purification of Natural Antioxidants from Olive Mill Wastewater Concentrates by A Sustainable Process. Boğaziçi University Institute of Environmental Science, Master's Thesis, Istanbul.

Elibol M et al 2008. Zeytinyağı işletmeleri katı(pirina) ve sıvı(karasu) atıklarından mikrobiyal lipaz üretimi, Tübitak Project, Project No: 106M464.

Elkacmi R, Bennajah, M 2019. New techniques for treatment and recovery of valuable products from olive mill wastewater. Handbook of Environmental Materials Management, pp. 1-20.

Gündüz Balpetek F 2017. Yeni teknoloji deterjanların farklı tekstil yüzeylerinde performanslarının araştırılması. Ege Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi, İzmir.

Hartzell-Lawson MM, Hsieh YL 2000. Characterizing the Noncellulosics in Developing Cotton Fibers. Textile Research Journal, 70(9): 810-819.

Kasırga E 1988. Zeytinyağı Endüstrisi Atıksularının Anaerobik Biyolojik Stabilizasyon Yöntemi ile Arıtılması ve Kinetik Model Geliştirilmesi. Dokuz Eylül Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi, İzmir.

Merdan N, Eyüpoğlu Ş 2023. Meşe Palamudundan Elde Edilen Doğal Boyarmadde ile Pamuk Kumaşın Boyanmasına Farklı Mordan Maddelerin Etkisinin Araştırılması. İstanbul Ticaret Üniversitesi Fen Bilimleri Dergisi, 22(43):33-43.

Mulinacci N, Romani A, Galardi C, Pinelli P, Giaccherini C, Vincieri FF 2001. Polyphenolic content in olive oil waste waters and related olive samples. Journal of agricultural and food chemistry, 49(8): 3509-3514.

Özkan Z 2022. Ürün Raporu Zeytinyağı 2021, Tarımsal Ekonomi ve Politika Geliştirme Enstitüsü (TEPGE), https://arastirma.tarimorman.gov.tr/tepge/Menu/37/Urun-Raporlari [15.06.2023]

Paraskeva P, Diamadopoulos E 2006. Technologies for olive mill wastewater (OMW) treatment: a review. Journal of Chemical Technology & Biotechnology: International Research in Process, Environmental & Clean Technology, 81(9): 1475-1485.

Rozzi A, Malpei F 1996. Treatment and disposal of olive mill effluents. International Biodeterioration & Biodegradation, 38(3-4): 135-144.

Tunç M, Ünlü A 2015. Zeytinyağı Üretim Atıksularının Özellikleri, Çevresel Etkileri ve Arıtım Teknolojileri. Nevşehir Bilim ve Teknoloji Dergisi, 4(2): 44-74.

Türemen M 2017. Tekstil Terbiye İşlemlerinde Nanokitosan Kullanımı. Ege Üniversitesi Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi, İzmir.

Uğur ŞS, Sarıışık AM 2015. Deposition of Nanoparticle Multilayers to Improve Mechanical Properties of Denim Fabrics. Th Journal of the Textile Institute, 106(7): 718-724.

Vlyssides AG, Loizides M, Karlis PK 2004. Integrated strategic approach for reusing olive oil extraction by-products. Journal of Cleaner Production, 12(6): 603-611.

Yılmaz H, Özcan MM. Zeytinyağı Endüstrisi Atık Ürünlerinden Zeytin Posası (Pirina), Zeytin Yaprağı ve Zeytin Çekirdeğinin Değerlendirilmesi.

Yüce İ, Erdogan S, Becenen N, Şen H 2020. Kitosan ile Ön İşlem Görmüş Pamuklu Kumaşın Asit Boyalarla Tek Adımda Renklendirilmesi, Trakya Üniversitesi Mühendislik Bilimleri Dergisi, 21(1): 1-14.

ORAL PRESENTATION

Predatör akar *Neoseiulus californicus* (mcgregor) (Acari:Phytoseiidae)'da tebufenpyrad direnci ile bazı detoksifikasyon enzimleri arasındaki ilişki

Arzu YEŞİL[1] (*ORCID: 0009-0003-6399-7258*), Gizem BERBER TORTOP[2*] (*ORCID: 0000-0003-3090-3705),* Sibel YORULMAZ[1] *(ORCID: 0000-0003-3836-5673)*

[1]Isparta Uygulamalı Bilimler Üniversitesi, Ziraat Fakültesi, Bitki Koruma Bölümü, Isparta, Türkiye
[*2]Bilecik Şeyh Edebali Üniversitesi, Ziraat ve Doğa Bilimleri Fakültesi, Bitki Koruma Bölümü, Bilecik, Türkiye

*Sorumlu yazar e-mail: gizem.berber@bilecik.edu.tr

Özet

Phytoseiidae familyasında yer alan *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae), fitofag akar mücadelesinde etkinliği ve alternatif besinlerle beslenebilmesi ile bilinen bir akardır. Zararlıları kontrol altına almak için yapılan akarisit/insektisit uygulamaları ile *N. californicus* gibi hedef dışı organizmalarda da direnç gelişimi gözlenebilmektedir. Bu çalışmada, sabit koşullar altında, predatör akar *N. californicus* (McGregor) popülasyonunda seleksiyon baskısı sonucunda tebufenpyrad direnç gelişimi araştırılmış ve 9 kez selekte edilen popülasyonda LC_{50} değeri 53.9 olarak belirlenmiştir. Tebufenpyrad seleksiyonu sonucu elde edilen dirençli ve hassas popülâsyonlarda sinerjistlerin akarisit üzerindeki etkinliğini anlamak ve yapılan biyokimyasal çalışmalar sonucu enzimlerle olan ilişkilerini ortaya koymak amacıyla PBO ve IBP sinerjistleri ile çalışmalar yapılmıştır. S9 popülasyonlarında PBO ve IBP sinerjistlerinin etki oranları sırasıyla 2.61 ve 1,17 kat olarak belirlenmiştir. Popülâsyonlarda esteraz ve monooksigenaz enzimlerinin aktiviteleri de uygun metotlarla belirlenmiştir. Esteraz enzim seviyesinde tebufenpyrad dirençli S9 popülasyonu ile başlangıç popülasyonu arasında önemli bir fark bulunmazken, monooksigenaz enzim aktivitesi için istatiksel olarak fark önemli bulunmuştur. Bu çalışma, pestisitlere dirençli *N. californicus*'un laboratuvar koşullarında kısa sürede direnç geliştirmesi nedeniyle arazi şartlarında kullanımları da dahil olmak üzere entegre bir akar yönetimi programının daha iyi tasarlanmasına katkıda bulunacaktır.

Anahtar Kelimeler: akarisit, direnç, METI’s, phytoseiid

The relationship between tebufenpyrad resistance and some detoxification enzymes in predator mite *Neoseiulus californicus* (McGregor) (Acari:Phytoseiidae)

Abstract

Neoseiulus californicus (McGregor) (Acari: Phytoseiidae), belonging to the Phytoseiidae family, is a mite known for its effectiveness in combating phytophagous mites and its ability to feed on alternative foods. Insecticide applications used to control pests cause side effects on non-target organisms such as *N. californicus.* In this study, the development of tebufenpyrad resistance as a result of selection pressure in the predator mite *N. californicus* (McGregor) under constant conditions was investigated and the LC50 value was determined as 53.9 in the population selected 9 times. The effect ratios of PBO and IBP synergists in S9 populations were determined as 2.61 and 1.17 fold, respectively, in order to understand the effectiveness of synergists on acaricide in resistant and sensitive populations obtained as a result of Tebufenpyrad selection and to reveal their relationship with enzymes as a result of biochemical studies. The activities of esterase and monooxygenase enzymes in the populations were also determined by appropriate methods. While there was no significant difference between the tebufenpyrad resistant S9 population and the initial population in esterase enzyme level, the difference was statistically significant for monooxygenase enzyme activity. This study will contribute to the better design of an integrated mite management program, including the use of pesticide-resistant *N. californicus* in field conditions, as it develops resistance in a short time under laboratory conditions.

Keywords: acaricides, METI's, Phytoseiid, resistance

GİRİŞ

Dünya genelinde tarım ürünlerinde en önemli zararlılardan biri olan, iki noktalı kırmızı örümcek, *Tetranychus urticae* Koch (Acari: Tetranychidae) polifag bir zararlıdır. 70'ten fazla farklı bitki cinsine ait 1100'ün üzerinde konukçuya sahip kozmopolit zararlıdır (Bolland ve ark., 1998; Grbić ve ark., 2011). Yapraklarda beslenme sonucu küçük noktacıklara, sararmaya, erken yaprak dökülmesine, bodur büyümeye ve nihayetinde tüm bitkinin ölümüne neden olurlar (Patil ve ark., 2014). İki noktalı kırmızı örümceğin kontrolü biyolojik, kimyasal veya entegre mücadele gibi yöntemler ile sağlanabilmektedir. Fakat genellikle kısa sürede etki göstermeleri sebebiyle insektisitlerin ve akarisitlerin kullanımı tercih edilmektedir. *T. urticae*'nin kimyasal mücadelesi, kısa yaşam döngüsüne ve yüksek üreme potansiyeline sahip olduğundan, pestisit direncinin hızla gelişmesi nedeniyle kolay değildir (Stumpf ve ark., 2001; Adesanya ve ark., 2021).

Kırmızı örümceklerin biyolojik mücadelesinde en çok kullanılan avcı grubu, Phytoseiidae familyası içerisinde yer alan akarlardır (Pickett ve ark., 1987; McMurtry ve Croft, 1997). Bu familyadaki türler genellikle predatör akarlar olarak bilinir ve bazıları dünya genelinde fitofag akarların kontrolünde aktif olarak kullanılırlar. Phytoseiidae familyasında yer alan *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae), fitofag akar mücadelesinde etkinliği ve alternatif besinlerle beslenebilmesi ile bilinen bir predatördür (Rhodes ve Liburd 2006; Sato ve ark., 2007; El Taj ve Jung 2012). Ayrıca tetranychid türlerinde en etkili biyolojik mücadele etmenlerinden biridir. Dünya'nın farklı ülkelerinde yaygın olarak dağılmakta ve ticari olarak da kullanılmaktadır (Gotoh ve ark., 2004; Canlas ve ark., 2006; McMurtry ve ark., 2013).

Entegre Zararlı Yönetim (IPM) programları içerisinde kullanılan spesifik insektisitlerin BCAs'lar üzerindeki etkilerinin belirlenmesi programın sağlıklı bir şekilde yürütülebilmesi açısından önem taşımaktadır (Desneux ve ark., 2007). Çünkü insektisitler ve akarisitler phyotesiid akarlar üzerinde yan etkilere sebep olabildiği gibi seleksiyon baskısı sonucunda direnç gelişimine de neden olabilmektedirler (Bostanian ve Akalach 2006; Duso ve ark., 2008; Cong ve ark., 2016; Ditillo ve ark., 2016; Wu ve Hoy 2016). Pestisit direnci, kimyasal mücadeleyi etkisiz kılması nedeniyle genellikle olumsuz bir özellik gibi görülmesine karşın, faydalı böcek ve akarlarda olumlu bir özellik olarak öne çıkmaktadır (Ghazy ve ark., 2016).

METI-I direnci genellikle artan P450 aktivitesi ile ilişkilendirilmiştir ve bu, farklı METI-I bileşikleri arasında yaygın olarak gözlemlenen çapraz direnç modellerinin nedeni olarak ileri sürülmüştür (Stumpf ve ark., 2001; Kim ve ark., 2004, 2006; Van Pottelberge ve ark., 2009). İçerisinde tebufenpyrad etken maddesinin de yer aldığı METI I akarisitleri (IRAC grup 21) olarak adlandırılan grup, NADH:ubikinon oksidoredüktaz'ı inhibe ederek mitokondriyal elektron taşıma sistemini bozmaktadır. METI I akarisitleri dünyada 1990 yılların başından beri tetranychid, tarsonemid ve eriophyd akarların tüm dönemlerine karşı kullanılmaktadır (Tomlin, 2003). METI I akarisitlerinin yoğun olarak uygulandığı alanlarda bulunabilecek olan phytoseiid akarların da bu akarisitlerden etkilenme olasılığı oldukça yüksektir. Özellikle tarla koşullarında akarisit seleksiyon baskısına maruz kalan predatör akarların çeşitli mekanizmalarla direnç geliştirebildiği bilinmektedir. METI I akarisitleri için Phytoseiidae akarlarda direnç gelişimi ve mekanizmasının belirlenmesine yönelik çalışmalar oldukça sınırlıdır.

Bu çalışmada önemli bir predatör akar olan *N. californicus*'da seleksiyon baskısı sonucunda tebufenpyrad direnç gelişimi araştırılmıştır. Ayrıca direncin önemli detoksifikasyon enzimleri olan esteraz ve monoksigenaz enzimleri ile olan ilişkisi kinetik olarak belirlenmiştir. Bununla birlikte tebufenpyrad direncinin detoksifikasyon enzimleri ile olan bağlantısını daha iyi açıklayabilmek adına sinerjist çalışmaları da yapılmıştır.

MATERYAL VE METOT

Predatör akarların orjini ve yetiştirilmesi

Neoseiulus californicus'un Türkiye orijini 2008 yılında organik elma yetiştiriciliği yapılan bahçeden toplanmıştır (Salman ve Recep 2013) ve günümüze kadar iklim kabinlerinde herhangi bir pestisite maruz kalmadan yetiştirilmiştir. *N. californicus* ve ona besin olarak verilen *T. urticae* popülasyonunun üretimi 26±2°C sıcaklık, %60±5 orantılı nem ve 16 saat aydınlatma koşulları sağlanan iklim odalarında ve fasulye (*Phaseolus vulgaris* L. var. Barbunia) bitkisi üzerinde yapılmıştır.

İnsektisit/Akarisit

Çalışmada tebufenpyrad etkili maddeye sahip Croshe 20 WP (Hektaş) ticari preparat kullanılmıştır. Bu ticari preparat elma bahçelerinde Tetranychidae familyasına ait zararlı kırmızıörümcek türleri için yaygın olarak kullanılmaktadır.

Seleksiyon çalışmaları

N. californicus'un 2008 yılından bu yana iklim odasında yetiştirilen popülasyonu seleksiyon için başlangıç popülasyonu olarak kullanılmıştır. Ancak seleksiyon işlemlerine başlamadan önce *N. californicus* popülasyonunun tebufenpyrad için hassasiyet kontrolleri yapılmıştır. Bu amaçla etken maddede *T. urticae* için de önerilen tarla uygulama dozu kullanılarak *N. californicus* popülasyonunda LC_{50} denemeleri kurulmuştur.

Tebufenpyrad için predatör akarda LC_{50} denemeleri şu şekilde yapılmıştır: Denemeler 1 kontrol+7 doz, her doz için 3 tekerrür olacak şekilde kurulmuştur. Tebufenpyrad uygulama dozları belirlenirken ilk dozda %90'dan az, kontrol grubunda ise %10'dan fazla ölüm olmaması dikkate alınmıştır (Van Leeuwen ve ark., 2010). 9 cm petri içerisine yaprağın nem ihtiyacını karşılayabilmesi amacıyla su agarı dökülerek soğutulmuştur. 3 cm barbunya yaprak disklerin etrafı Tangle Trap yapışkanı ile çevrelenerek predatör akarların kaçması önlenmiştir ve bu yaprak diskler agar ortamına konulmuştur. Her tekerrür için petri içerisine 25±2 adet predatör akar ergini dişisi eklenmiştir. Her doz için %50 seyreltilerek hazırlanan insektisit konsantrasyonları ile petriler spraytower yardımıyla yaprak yüzeyine 1 bar basınçta 2 mL ilaç gelecek şekilde ilaçlama yapılmıştır. Ölü-canlı sayımları 24 saat sonunda yapılarak LC_{50} değerleri POLO (probit analizi) bilgisayar programı ile belirlenmiştir. LC_{50} denemesi yapıldıktan ve tebufenpyrad için *N. californicus*'un hassasiyet düzeyi yüksek olarak belirlendikten sonra seleksiyon işlemlerine başlanmıştır. Predatör akar popülasyonu için seleksiyon dozu olarak POLO programından belirlenen LC_{60} değerleri kullanılarak başlanılmıştır.

Seleksiyon işlemi, tabanında agar ortamı bulunan 9 cm çapındaki petriler üzerinde etrafı Tangle Trap yapışkanı ile çevrilmiş olan 3 cm barbunya yaprak diskleri üzerine 50±2 adet predatör akar ergin dişisi aktarılmıştır. Seleksiyon çalışmaları 15 tekekrrür olarak yapılmıştır. LC_{60} dozu petrilere ilaçlama kulesinde 1 bar basınç altında yaprak üzerine 2 mL olacak şekilde uygulanmıştır. Petriler 26±1ºC sıcaklıkta %60-65 nem ve 16:8 (A/K) fotoperiyot koşullarında 24 saat bırakılmıştır. Bu süre içerisinde *N. californicus* bireylerinin besinsizlikten ölmelerini engellemek amacıyla petriler içerisine her avcı akar için yaklaşık 3 adet ergin dişi *T. urticae* (petri içerisine yaklaşık 150 tane olacak şekilde) bireyleri aktarılmıştır. Seleksiyon uygulamasından 24 saat sonra petri içerisinde canlı kalan *N. californicus* bireyleri (kendi boyu kadar hareket edebilen birey canlı olarak sayılacaktır) temiz barbunya bitkisi üzerine aktarılmıştır. Ayrıca bu bitki üzerine seleksiyon sonrası canlı kalan *N. californicus* bireylerinin beslenebilmesi amacıyla yeteri kadar (her predatör akar için yaklaşık olarak 3 adet ergin *T. urticae* bireyi) aktarılmıştır.

Sinerjist + tebufenpyrad çalışmaları

Seleksiyon aşamalarından sonra tebufenpyrad dirençli *N. californicus*'da monooksigenaz enzim inhibitörü piperonyl butoxide (PBO) (2000µl/l) ve esteraz enzim inhibitörü S-Benzyl-O,O-diisopropyl phosphorothioate (IBP) (200 µl/l) kullanılmıştır (Stumpf ve Nauen 2002; Kim ve ark., 2004; Van Leeuwen ve ark., 2004). Sinerjist çalışmalarında *N. californicus*''un ergin dişi bireyleri kullanılmıştır. Denemeler 1 kontrol+7 doz, 3 tekerrür olacak şekilde kurulmuştur. Sinerjistler 1:1 oranında aseton: saf su içerisinde çözdürülmüştür. Hazırlanan sinerjist çözeltileri ilaçlama kulesinde tabanında ıslatılmış pamuk bulunan petriler üzerine yerleştirilen yapraklar üzerine 1 bar basınç altında 1 ml olarak püskürtülerek petriler 24 saat boyunca 26±1 ºC sıcaklıkta %60-65 nem ve 16:8 fotoperiyot koşullarında tutulmuştur. Sinerjist uygulamasından 24 saat sonra her doz için %50 seyreltilerek hazırlanan tebufenpyrad konsantrasyonları ile petriler spray tower yardımıyla yaprak yüzeyine 2 ml ilaç gelecek şekilde ilaçlanmıştır. Ölü-canlı sayımları 24 saat sonunda yapılmıştır. Bireylerin besinsizlikten ölmelerini engellemek amacıyla her gün petri içerisine av aktarılmıştır. Kontrolde ise sadece sinerjist kullanılmıştır.

Sinerjistik etki oranı= Sinerjistsiz LC_{50} değeri/ Sinerjistli LC_{50} değeri formülü ile hesaplanmıştır (Kim ve ark., 2004).

Esteraz ve P450 Monoksigenaz Enzim Aktiviteleri

Esteraz aktivitesinin kinetik olarak belirlenmesinde substrat olarak α–naphtylacetate ve (Stumpf ve Nauen 2002)'in geliştirdikleri yöntem kullanılmıştır. 20 adet ergin dişi 100 µl sodyum fosfat buffer (0.1M, pH 7.5) (%0.1 Triton X-100 içeren) içinde homojenize edilmiştir. Bu homojenat 10000 g, +4 º C'de ve 5 dk santrifüj edildikten sonra enzim kaynağı olarak kullanılmıştır. Enzim kaynağı olarak kullanılan supernatant 10 kat seyreltilmiştir. Mikroplakanın hücrelerine 25 µl supernatant + 25 µl fosfat buffer (0.2 M, pH:6) konulmuştur. Çalışma hücrelere 200 µl substrat solüsyonunun eklenmesiyle başlatılmıştır. Substrat solüsyonu 30 mg

fastblue RR tuzunun 50 ml 0.2 M sodyum fosfat buffer'da çözülmesi ve bu karışıma 500 µl 100 Mm α - naphtylacetate'ın eklenmesiyle elde edilmiştir. Enzim aktivitesi 23 º C, 450 nm'de 10 dk süreyle okunmuştur. Sitokrom P450 monooksigenaz enziminin belirlenmesinde substrat olarak *p*-nitroanisole (PNOD) ve (RosE ve ark., 1995) yöntemi uyarlanarak kullanılmıştır. 50 adet dişi birey 100 µl homojenizasyon buffer'da (0.05 M Tris-HCl + %1.15KCl + 1mM EDTA pH (7.7)) plastik ezici ile ezilerek +4 °C 10000 g'de 20 dk santrifüj edilmiştir. Mikroplaka hücrelerine 45 µL homojenizasyon buffer + 45 µL supernatant+100 µL 2mM PNOD eklenerek karışım 30 °C'de 5 dk inkübe edilmiştir. Reaksiyon mikroplaka hücrelerine 10 µL 9.6 mM NADPH eklenerek başlatılmıştır. P450 enzim aktivitesi Versamax kinetic microplate reader'da (Molecular Devices) 405 nm 30 °C'de 15 dk süreyle ölçülmüştür.

Biyokimyasal çalışmalarda, kontrol hücreleri ise homojenatsız olarak okunmuştur. Enzim okumaları dört tekerrürlü olarak yapılmıştır. Tüm enzim aktiviteleri Softmax PRO software programında analiz edilerek sonuçlar mOD min-1 mg-1 protein olarak verilmiştir. Örneklerin toplam protein miktarlarının belirlenmesinde Bradford (1976)'un total protein tayin yöntemi kullanılarak ve Bovine Serum Albumine (BSA) standart olarak alınmıştır. Enzim sonuçlarından elde edilen veriler tek yönlü varyans analizi tekniği ile (One-Way ANOVA) analiz edilmiş ve popülasyonlar arasındaki farklılıkların belirlenmesinde Tukey testi kullanılmıştır.

BULGULAR

Tebufenpyrad ile seleksiyon sonrası elde edilen popülasyonların $LC_{50, 60}$ değerleri ve direnç oranları tablo 1'de verilmiştir.

Tablo 1. *Neoseiulus californicus* popülasyonlarında tebufenpyrada karşı belirlenen LC, df, x^2 değerleri ve direnç oranları

Popülasyon	**n***	**Eğim±SE**	**LC_{50} (mga.i. L^{-1}) (95% CL)**	**LC_{60} (mga.i. L^{-1}) (95% CL)**	**df**	**x^2**	**R****
Başlangıç popülasyonu	602	1.432±0.207	0.020 0.012-0.025	0.026 0.018-0.038	6	5.8	
S1	598	1.419±0.171	0.078 0.056-0.107	0.11 0.086-0.165	6	6.8	3.9
S2	593	1.464±1.164	0.095 0.069-0.126	0.14 0.106-0.190	6	5.5	4.75
S3	601	1.142±1.145	0.160 0.111-0.224	0.267 0.191-0.387	5	4.8	8
S4	590	1.279±0.144	0.223 0.165-0.295	0.352 0.266-0.477	6	7.5	11.15
S5	594	1.546±0.161	0.252 0.148-0.388	0.367 0.232-0.583	6	6.5	12.6
S6	603	1.330±0.146	0.403 0.201-0.716	0.625 0.349-1.226	5	7.3	20.15
S7	604	1.761±0.176	0.458 0.303-0.655	0.638 0.441-0.934	6	7.5	22.9
S8	584	1.547±0.157	0.802 0.262-1.984	1.170 0.486-3.770	6	6.8	40.1
S9	598	1.720±0.221	1.079 0.743-1.466	1.372 1.111-1.478	5	7.2	**53.9**

*: Toplam birey sayısı
**: Direnç oranı

S9 popülasyonunda PBO ve IBP sinerjistlerinin etkileri belirlenmiş ve sonuçlar tablo 2'de verilmiştir.

DRAFT

Tablo 2. *Neoseiulus californicus* popülasyonlarında tebufenpyrad ve tebufenpyrad + sinerjistlerin etki oranları

Kimyasal	**n***	**Eğim±SE**	**LC_{50} (mga.i. L^{-1}) (95% CL)**	**df**	**x^2**	**SR****
			S9 popülasyonu			
tebufenpyrad (Sinerjistsiz)	598	1.720±0.221	1.079 0.743-1.466	5	7.2	-
tebufenpyrad +IBP	603	1.587±0.171	0.916 0.725-1.139	6	6.5	1.17
tebufenpyrad +PBO	569	1.274±0.134	0.413 0.316-0.521	6	7.5	**2.61**
			Başlangıç popülasyonu			
tebufenpyrad (Sinerjistsiz)	602	1.432±0.207	0.020 0.012-0.025	6	5.8	
tebufenpyrad + IBP	595	1.854±0.224	0.020 0.016-0.033	6	6.5	1.00
tebufenpyrad + PBO	585	2.021±0.252	0.019 0.013-0.043	6	5.5	1.05

*: Birey Sayısı
**: Sinerjistik etki oranı

Popülasyonların esteraz enzim aktiviteleri Tablo 3'de verilmiştir.

Tablo 3. Esteraz enzim aktiviteleri

Populasyon	**N***	**mOD/min/mg protein (±SE)**
S9	4	**7,85** (±0.03) a
Başlangıç	4	**6,32** (±0.02) a

N: tekerrür sayısı

Popülasyonların monoksigenaz enzim aktiviteleri Tablo 4'de verilmiştir.

Tablo 4. Monooksigenaz enzim aktivitesi

Popülasyon	**N***	**mODmin^{-1}mg^{-1} protein (±SE)**
S9	4	0.01794 (±0.004) a
Başlangıç	4	0.00780 (±0.001) b

N: tekerrür sayısı

TARTIŞMA

Zararlı akarlar ile mücadelede özellikle Phytoseiidae familyasının önemli bir yer aldığı uzun zamandır bilinmektedir (Solomon ve ark., 2000; Duso ve ark., 2009). Phytoseiidae familyasında yer alan *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae), fitofag akar mücadelesinde etkinliği ve alternatif besinlerle beslenebilmesi ile bilinen bir akardır (Rhodes ve Liburd, 2006; Sato ve ark., 2007; El Taj ve Jung, 2012). Zararlıların kontrolünde tek bir mücadele yönteminin yeterli olmaması sebebiyle entegre mücadele genellikle tercih edilmektedir. Bu kapsamda pestisitlerin doğal düşmanlar üzerindeki etkinliği oldukça önemlidir. Yapılan çalışmada, *N. californicus*'da tebufenpyrad direnç oranı, 9 seleksiyon sonrası 53.9 kata kadar çıkmıştır. Ülkemizde 1990'lı yıllarda ruhsat alan ve günümüze kadar yoğun bir şekilde kullanılan tebufenpyradın da içerisinde yer aldığı METI I akarisitlerinde fitofag akarlarda belirlenen çok sayıda direnç çalışması

yayınlanmıştır (Croft ve Van de Baan, 1988; Van Leeuwen ve ark., 2009, 2010; Kwon ve ark., 2010a, b, c, 2012; Osakabe ve ark., 2010; Bajda ve ark., 2017). Ancak phytoseiid akarlarda METI I akarisit direnci üzerine yapılan çalışma sayısı oldukça sınırlıdır. Yakın zamanda yapılan bir çalışmada, laboratuvar koşullarında 7 kez pyridaben ile selekte edilen *N. californicus*'da direnç 270,0 kat olarak belirlenmiştir (Özkan ve Yorulmaz, 2022). Albayrak ve ark., 2022, in vitro koşullarda 5 seleksiyon ile *N. californicus'*da 69 kat pirimicarb direnci elde etmişlerdir. Salman ve Keskin (2019), laboratuvar şartlarında *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae)'de milbemectine karşı beş seleksiyon sonrasında 31 kat, spirodiclofene karşı ise 108 kat direnç geliştiğini belirlemişlerdir. Assis ve ark., 2018'de, yaptıkları çalışmada *N. californicus*'da METI I akaristi olan fenpyroximate karşı 113,96 kat direnç belirlemişlerdir. Yorulmaz-Salman ve Ay (2014), *N. californicus*'ta on üç seleksiyon sonrasında 52 kat spiromesifen direnci belirtmişlerdir. Salman ve Recep (2013), 14 defa hexythiazox uygulaması yapılan *N. californicus* populasyonunda 64,04 kat direnç geliştiğini tespit etmişlerdir. Poletti ve Omoto (2012), Brezilya'da yaptıkları bir çalışmada 3500 kat deltamethrin dirençli *Phytoseiulus macropilis* Banks (Acari: Phytoseiidae) popülasyonu belirlemişlerdir. Auger ve ark., 2005'de, mancozeb ile 10 kere seleksiyon yaptıkları *Typhlodromus pyri* Scheuten (Acari: Phytoseiidae)'in popülasyonunda hassas popülasyona göre direncin 73 kat arttığını belirlemişlerdir. Sato ve ark., 2000, *Amblyseius womersleyi* Schicha (Acari: Phytoseiidae)'de methidathion ile 4 seleksiyon sonrası 311 kat dirençli popülasyon elde etmişlerdir. Görüldüğü üzere, başlangıç popülasyonunun genetik çeşitliliği, zararlı türü, başlangıç popülasyonunun direnç durumu gibi biyolojik etmenler ve ayrıca pestisitlerin bulunduğu sınıf veya pestisit türü laboratuvar seleksiyonlarında elde edilen direnç oranını etkilemektedir. Yapılan çalışmada ve literatürde de phytoseiidae familyası içerisinde yer alan predatör akarların, hem tarla hem de laboratuvar koşullarında seleksiyon baskısı sonucunda kimyasallara karşı direnç geliştirebilmektedir.

Tebufenpyrad seleksiyonu sonucu elde edilen dirençli ve hassas popülâsyonlarda sinerjistlerin akarisit üzerindeki etkinliğini anlamak ve yapılan biyokimyasal çalışmalar sonucu enzimlerle olan ilişkilerini ortaya koymak amacıyla PBO ve IBP sinerjistleri ile çalışmalar yapılmıştır. S9 popülasyonlarında PBO ve IBP sinerjistlerinin etki oranları sırasıyla 2.61 ve 1,17 kat olarak belirlenmiştir. Popülâsyonlarda esteraz ve monooksigenaz enzimlerinin aktiviteleri de uygun metotlarla belirlenmiştir. Esteraz enzim seviyesinde tebufenpyrad dirençli S9 popülasyonu ile başlangıç popülasyonu arasında önemli bir fark bulunmazken, monooksigenaz enzim aktivitesi için istatiksel olarak fark önemli bulunmuştur. Literatür incelendiğinde, *T. urticae*'deki METI Kompleks I akarisitlerine direnç mekanizmalarını araştıran birçok çalışmada, genellikle P450 monooksijenaz aracılı metabolizmanın ana direnç sağlayan faktör olduğunu ve muhtemelen farklı METI-Is bileşikleri arasında gözlemlenen geniş çapraz direnç modelinden de sorumlu olduğu konusunda fikir birliğine varılmıştır (Hollingworth ve Ahammadsahib, 1995; Y. Kim ve ark., 2006; Y.-J. Kim ve ark., 2004; Motoba ve ark., 2000; Stumpf ve Nauen, 2001; Sugimoto ve Osakabe, 2014; Van Pottelberge ve ark., 2008, 2009). Fenpyroximate direncinde hem esteraz hem de P450 enzimlerinin (Ay ve Kara, 2011; Khanjani ve ark., 2020) rol aldığını belirten çalışmalar da yer almaktadır. Literatürde diğer predatör akarlar üzerine farklı pestisitlerle ilişkili enzimler ve sinerjistleri ile ilgili çalışmalar şu şekilde yer almaktadır; Fournier ve ark., 1987, *P.persimilis*'te methidathion direncinin gelişmesinde GST enziminin etkili olduğunu ileri sürmüşlerdir. Sato ve ark., 2001, 2006 methidathion dirençli *A. womersleyi* popülasyonu üzerinde monooksigenaz aktivitesinin oldukça sinerjistik bir etkiye sahip olduğunu belirlemişlerdir. Detoksifikasyon sürecinde yer alan farklı enzim gruplarının akar türüne ve sinerjiste göre etkilerinin değiştiği görülmektedir. Çalışma sonuçlarına göre, *N. californicus*'un tebufenpyrad direncinde monoksigenaz enziminin rol oynama potansiyeli olduğu düşünülmektedir.

SONUÇ

Predatör akarları pestisitlerden korumanın yöntemlerinden biri, IPM topluluğunun sürekli ilgisini çeken, pestisitlere dirençli predatör popülasyonların meydana getirilmesidir. Literatürde ve yapılan çalışmada da görüldüğü üzere seleksiyonlar ile fitofag akarlar gibi hızlı direnç gelişimi predatör akarlarda da gerçekleşmektedir. Seleksiyon çalışmaları ile pestisitlerin phytoseiid akarlar üzerindeki etkilerini anlamak, çoğaltma veya koruma programlarına başarılı bir şekilde entegre olmaları için oldukça önemlidir ayrıca bir pestisitin kalıcılığı ve sinerjistler ile etkileşimleri bir IPM programındaki yerlerinin belirlenmesine yardımcı olacaktır. Kimyasallara karşı direnç geliştiren doğal düşmanlar, pestisitlerin yoğun olarak kullanıldığı alanlarda daha uzun süre canlı kalabilmektedir. Bu nedenle entegre zararlı yönetim programlarına dahil edilebilirler. Ancak dayanıklı doğal düşmanlar saha şartlarında test edilmeli ve adaptasyon çalışmaları da yapılmalıdır.

DRAFT

TEŞEKKÜR

Çalışmayı maddi yönden destekleyen TÜBİTAK 2209-A Üniversite Öğrencileri Yurt İçi Araştırma Projeleri Destek Programı'na teşekkür ederiz.

KAYNAKLAR

Adesanya AW, Lavine MD, Moural TW, et al 2021. Mechanisms and management of acaricide resistance for *Tetranychus urticae* in agroecosystems. Journal of Pest Science, 94:639–663.

Albayrak T, Yorulmaz S, İnak E, et al 2022. Pirimicarb resistance and associated mechanisms in field-collected and selected populations of *Neoseiulus californicus*. Pesticide Biochemistry and Physiology, 180:104984.

Assis CP, Gondim Jr MG, Siqueira HA 2018. Synergism to acaricides in resistant *Neoseiulus californicus* (Acari: Phytoseiidae), a predator of *Tetranychus urticae* (Acari: Tetranychidae). Crop Protection, 106:139–145.

Auger P, Bonafos R, Kreiter S, Delorme R 2005. A genetic analysis of mancozeb resistance in *Typhlodromus pyri* (Acari: Phytoseiidae). Experimental & applied acarology, 37:83–91.

Ay R, Kara FE 2011. Toxicity, inheritance of fenpyroximate resistance, and detoxification-enzyme levels in a laboratory-selected fenpyroximate-resistant strain of *Tetranychus urticae* Koch (Acari: Tetranychidae). Crop Protection, 30:605–610. https://doi.org/10.1016/j.cropro.2010.11.012

Bajda S, Dermauw W, Panteleri R, et al 2017. A mutation in the PSST homologue of complex I (NADH: ubiquinone oxidoreductase) from *Tetranychus urticae* is associated with resistance to METI acaricides. Insect biochemistry and molecular biology, 80:79–90

Bolland HR, Gutierrez J, Flechtmann CH 1998. World catalogue of the spider mite family (Acari: Tetranychidae). Brill

Bostanian NJ, Akalach M 2006. The effect of indoxacarb and five other insecticides on *Phytoseiulus persimilis* (Acari: Phytoseiidae), *Amblyseius fallacis* (Acari: Phytoseiidae) and nymphs of *Orius insidiosus* (Hemiptera: Anthocoridae). Pest Management Science: formerly Pesticide Science, 62:334–339.

Canlas LJ, Amano H, Ochiai N, Takeda M 2006. Biology and predation of the Japanese strain of *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae). Systematic and Applied Acarology, 141–157.

Cong L, Chen F, Yu S, et al 2016. Transcriptome and difference analysis of fenpropathrin resistant predatory mite, *Neoseiulus barkeri* (Hughes). International Journal of Molecular Sciences, 17:704.

Croft BA, Van de Baan HE 1988. Ecological and genetic factors influencing evolution of pesticide resistance in tetranychid and phytoseiid mites. Experimental & Applied Acarology, 4:277–300.

Desneux N, Decourtye A, Delpuech J-M 2007. The sublethal effects of pesticides on beneficial arthropods. Annu Rev Entomol, 52:81–106.

Ditillo JL, Kennedy GG, Walgenbach JF 2016. Effects of insecticides and fungicides commonly used in tomato production on *Phytoseiulus persimilis* (Acari: Phtyoseiidae). Journal of economic entomology, tow234

Duso C, Fanti M, Pozzebon A, Angeli G 2009. Is the predatory mite Kampimodromus aberrans a candidate for the control of phytophagous mites in European apple orchards? BioControl, 54:369–382. https://doi.org/10.1007/s10526-008-9177-6

Duso C, Malagnini V, Pozzebon A, et al 2008. Comparative toxicity of botanical and reduced-risk insecticides to Mediterranean populations of *Tetranychus urticae* and *Phytoseiulus persimilis* (Acari Tetranychidae, Phytoseiidae). Biological Control, 47:16–21. https://doi.org/10.1016/j.biocontrol.2008.06.011

El Taj HF, Jung C 2012. Effect of temperature on the life-history traits of *Neoseiulus californicus* (Acari: Phytoseiidae) fed on *Panonychus ulmi*. Experimental and Applied Acarology, 56:247–260.

Fournier D, Cuany A, Pralavorio M, et al 1987. Analysis of methidathion resistance mechanisms in *Phytoseiulus persimilis* AH. Pesticide Biochemistry and Physiology, 28:271–278.

Ghazy NA, Osakabe M, Negm MW, et al 2016. Phytoseiid mites under environmental stress. Biological control, 96:120–134.

Gotoh T, Yamaguchi K, Mori K 2004. Effect of temperature on life history of the predatory mite *Amblyseius (Neoseiulus) californicus* (Acari: Phytoseiidae). Experimental & Applied Acarology, 32:15–30.

Grbić M, Van Leeuwen T, Clark RM, et al 2011. The genome of *Tetranychus urticae* reveals herbivorous pest adaptations. Nature, 479:487–492.

DRAFT

Hollingworth RM, Ahammadsahib KI 1995. Inhibitors of respiratory complex I. Mechanisms, pesticidal actions and toxicology. Reviews in pesticide toxicology (USA)

Khanjani M, Saberfar F, Mirzaie-Asl A, Sheikhi Garjan A 2020. Resistance mechanisms of the two-spotted spider mite, *Tetranychus urticae* (Acari: Tetranychidae) populations to fenpyroximate. Journal of Crop Protection, 9:337–345.

Kim Y, Park H, Cho J, Ahn Y 2006. Multiple resistance and biochemical mechanisms of pyridaben resistance in *Tetranychus urticae* (Acari: Tetranychidae). Journal of economic entomology, 99:954–958.

Kim Y-J, Lee S-H, Lee S-W, Ahn Y-J 2004. Fenpyroximate resistance in *Tetranychus urticae* (Acari: Tetranychidae): cross-resistance and biochemical resistance mechanisms. Pest Management Science: formerly Pesticide Science, 60:1001–1006.

Kwon DH, Choi JY, Je YH, Lee SH 2012. The overexpression of acetylcholinesterase compensates for the reduced catalytic activity caused by resistance-conferring mutations in *Tetranychus urticae*. Insect biochemistry and molecular biology, 42:212–219.

Kwon DH, Clark JM, Lee SH 2010a. Cloning of a sodium channel gene and identification of mutations putatively associated with fenpropathrin resistance in *Tetranychus urticae*. Pesticide biochemistry and physiology, 97:93–100.

Kwon DH, Im JS, Ahn JJ, et al 2010b. Acetylcholinesterase point mutations putatively associated with monocrotophos resistance in the two-spotted spider mite. Pesticide biochemistry and physiology, 96:36–42.

Kwon DH, Yoon KS, Clark JM, Lee SH 2010c. A point mutation in a glutamate-gated chloride channel confers abamectin resistance in the two-spotted spider mite, *Tetranychus urticae* Koch. Insect molecular biology, 19:583–591.

McMurtry JA, Croft BA 1997. Life-styles of phytoseiid mites and their roles in biological control. Annual review of entomology, 42:291–321.

McMurtry JA, de Moraes GJ, Sourassou NF 2013. Revision of the lifestyles of phytoseiid mites (Acari: Phytoseiidae). Syst Appl Acarol, 18: 297–320.

Motoba K, Nishizawa H, Suzuki T, et al 2000. Species-specific detoxification metabolism of fenpyroximate, a potent acaricide. Pesticide biochemistry and physiology, 67:73–84.

Osakabe M (Mh), Uesugi R, Goka K 2010. Evolutionary Aspects of Acaricide-Resistance Development in Spider Mites. Psyche: A Journal of Entomology, 2009:e947439. https://doi.org/10.1155/2009/947439

Patil DL, Patel KA, Toke NR, Ambule AT 2014. Biology of *Tetranychus urticae* Koch (Acarina: Tetranychidae) on carnation under laboratory conditions. International Journal of Plant Protection, 7:334–338.

Pickett CH, Gilstrap FE, Morrison RK, Bouse LF 1987. Release of predatory mites (Acari: Phytoseiidae) by aircraft for the biological control of spider mites (Acari: Tetranychidae) infesting corn. Journal of economic entomology, 80:906–910.

Poletti M, Omoto C 2012. Susceptibility to deltamethrin in the predatory mites *Neoseiulus californicus* and *Phytoseiulus macropilis* (Acari: Phytoseiidae) populations in protected ornamental crops in Brazil. Experimental and applied acarology, 58:385–393.

Rhodes EM, Liburd OE 2006. Evaluation of predatory mites and Acramite for control of twospotted spider mites in strawberries in north central Florida. Journal of Economic Entomology, 99:1291–1298.

Rose RL, Barbhaiya L, Roe RM, et al 1995. Cytochrome P450-associated insecticide resistance and the development of biochemical diagnostic assays in *Heliothis virescens*. Pesticide Biochemistry and Physiology, 51:178–191.

Salman SY, Keskin C 2019. The effects of milbemectin and spirodiclofen resistance on *Phytoseiulus persimilis* AH (Acari: Phytoseiidae) life table parameters. Crop Protection, 124:104751.

Salman SY, Recep AY 2013. Analysis of hexythiazox resistance mechanisms in a laboratory selected predatory mite *Neoseiulus californicus* (Acari: Phytoseiidae). Turk entomol derg, 37:409–422.

Sato ME, Da Silva MZ, De Souza Filho MF, et al 2007. Management of *Tetranychus urticae* (Acari: Tetranychidae) in strawberry fields with *Neoseiulus californicus* (Acari: Phytoseiidae) and acaricides. Experimental and Applied Acarology, 42:107–120.

Sato ME, Miyata T, Kawai A, Nakano O 2000. Selection for resistance and susceptibility to methidathion and cross resistance in *Amblyseius womersleyi* Schicha (Acari: Phytoseiidae). Applied Entomology and Zoology, 35:393–399.

Sato ME, Miyata T, Kawai A, Nakano O 2001. Methidathion resistance mechanisms in *Amblyseius womersleyi* schicha (Acari: Phytoseiidae). Pesticide Biochemistry and Physiology, 69:1–12.

Sato ME, Tanaka T, Miyata T 2006. Monooxygenase activity in methidathion resistant and susceptible populations of *Amblyseius womersleyi* (Acari: Phytoseiidae). Experimental & applied acarology, 39:13–24.

Solomon MG, Cross JV, Fitzgerald JD, Campbell CA 2000. M.; Jolly, RL; Olszak, RW; Niemczyk, E.; Vogt, H. Biocontrol of pests of apples and pears in Northern and Central Europe—3. Predators Biocontrol Sci Technol, 10:91–128.

Stumpf N, Nauen R 2002. Biochemical markers linked to abamectin resistance in *Tetranychus urticae* (Acari: Tetranychidae). Pesticide biochemistry and physiology, 72:111–121.

Stumpf N, Nauen R 2001. Cross-resistance, inheritance, and biochemistry of mitochondrial electron transport inhibitor-acaricide resistance in *Tetranychus urticae* (Acari: Tetranychidae). Journal of Economic Entomology, 94:1577–1583.

Stumpf N, Zebitz CP, Kraus W, et al 2001. Resistance to organophosphates and biochemical genotyping of acetylcholinesterases in *Tetranychus urticae* (Acari: Tetranychidae). Pesticide Biochemistry and Physiology, 69:131–142.

Sugimoto N, Osakabe M 2014. Cross-resistance between cyenopyrafen and pyridaben in the twospotted spider mite *Tetranychus urticae* (Acari: Tetranychidae). Pest management science, 70:1090–1096.

Tomlin C 2003. The e-Pesticide Manual, Version 3.0., BCPC. Crop Protection Publication: Cambridge, UK, CD-ROM

Van Leeuwen T, Stillatus V, Tirry L 2004. Genetic analysis and cross-resistance spectrum of a laboratory-selected chlorfenapyr resistant strain of two-spotted spider mite (Acari: Tetranychidae). Experimental & applied acarology, 32:249–261.

Van Leeuwen T, Vontas J, Tsagkarakou A, et al 2010. Acaricide resistance mechanisms in the two-spotted spider mite *Tetranychus urticae* and other important Acari: a review. Insect biochemistry and molecular biology, 40:563–572.

Van Leeuwen T, Vontas J, Tsagkarakou A, Tirry L 2009. Mechanisms of acaricide resistance in the two-spotted spider mite *Tetranychus urticae*. Biorational Control of Arthropod Pests: Application and Resistance Management, 347–393.

Van Pottelberge S, Van Leeuwen T, Nauen R, Tirry L 2009. Resistance mechanisms to mitochondrial electron transport inhibitors in a field-collected strain of *Tetranychus urticae* Koch (Acari: Tetranychidae). Bulletin of entomological research, 99:23–31.

Van Pottelberge S, Van Leeuwen T, Van Amermaet K, Tirry L 2008. Induction of cytochrome P450 monooxygenase activity in the two-spotted spider mite *Tetranychus urticae* and its influence on acaricide toxicity. Pesticide biochemistry and physiology, 91:128–133.

Wu K, Hoy MA 2016. The glutathione-S-transferase, cytochrome P450 and carboxyl/cholinesterase gene superfamilies in predatory mite *Metaseiulus occidentalis*. PLoS One, 11:e0160009.

Yorulmaz-Salman S, Ay R 2014. Determination of the inheritance, cross-resistance and detoxifying enzyme levels of a laboratory-selected, spiromesifen-resistant population of the predatory mite *Neoseiulus californicus* (Acari: Phytoseiidae). Pest management science, 70:819–826.

ORAL PRESENTATION

Growing of stem cells and extarcting their signslling molecules in a three dimensinal cellulose environment extracted from *Musa* sp.

Moyassar Basil Hadi Al-Shaibani[1*] (https://orcid.org/0000-0003-4380-9945)

*[1] Al-Nahrain University, College of Biotechnology, Department of Molecular and Medical Biotechnology, Baghdad, Iraq

Corresponding author Email: moyassar.basil@nahrainuniv.edu.iq

Abstract

Banana peels are the most waste products of fruits in the world and represent a rich source of cellulose. Cellulose could be used as a valuable material for various therapeutic and medical applications. Cellulose is a polymer could be applied as scaffold for growing human stem cells in 3-dimensional cell culture (3DCC) to collect their secretions for further therapeutic applications instead of MSC secretions collected of human stem cells (hSCs) in traditional 2-dimensional cell culture (2DCC). Therefore, this study has developed cellulose scaffold (CS) from banana peels. Banana peels were collected and dried using heating and cooling rounds and used as a source for cellulose using standard protocols. The collected cellulose was defatted, purified from proteins and bleached by hydrogen peroxide solutions, then dried and powdered. Previously isolated and characterized human mesenchymal stem cells (MSCs) (N=4) were grown in tissue culture plates in traditional 2DCC as well as growing on CS to from MSC 3D culture. MSC sections from both cultures were collected after 24 h and 48 h and analyzed using ELISA kits. MSC secretions collected from both 2DCC and 3DCC contained stromal derived factor-1 (SDF-1) and interleukin 1 (IL-1). Concentrations of biomolecules secreted by MSCs in 3DCC were significantly higher than in 2DCC. It could be concluded that CS could be used as 3D culture scaffold to grow MSCs and collect their secretions for medical and therapeutic applications.

Keywords: banana peels, cellulose, 3D scaffold, 3D cell culture, MSC secretions.

INTRODUCTION

One of the most widely consumed fruits in the world, bananas account for 40% of all fruit trade globally. The peel, which accounts for between 30% and 40% of the weight of the entire fruit is the primary by-product of the banana processing industry. This banana peel waste is an issue for the environment because it has high levels of nitrogen and phosphate as well as a lot of water, which is conducive to the growth of microorganism. Making wheat into a more valuable commodity is one method to lessen the issue. cellulose and other polymers can be found in banana peel (Hariani et al., 2016). Making cellulose from banana peels, a more valuable product that may be utilized more widely in the food sector (Iravani and Varma., 2022). The primary structural element of plants, cellulose, is a glucose polymer linked in a -1, 4 linkage arrangement. The cellulose polymer's high intermolecular hydrogen bonding and ability to crystallize in a linear structure due to the -1, 4 linkage give it significant shear and tensile strength. cellulose can be refined for usage as a food ingredient due to its chemical make-up. The least soluble of all the fiber components, cellulose is insoluble in both hot and cold water as well as hot dilute acids and alkalis (Singanusong et al., 2013). Pharmaceutical derivatives such as microcrystalline cellulose and powdered cellulose, which are utilized as inert fillers and thickening agents in the reinforcing of drugs, are examples of the many uses for cellulose drugs (Cherian et al., 2008). Additionally, cellloluse can be employed as a potential biopolymer to construct three-dimensional (3D) scaffolds instead of using non-renewable polymers for tissue culture and tissue engineering (Firmanda et al., 2022). Utilization of 3D scaffold is a promising technique to grow cells and collect their secretions for further uses in therapeutic applications (Al-Shaibani 2022). Human mesenchymal stem cells (MSCs) are the pivotal player for coordinating the developmemr and regeneration of all body organs by differentiation and secreting biologically active substances (Zahorec et al., 2015). According to Teixeira and Salgado (2013), the secretome of MSCs is described as a complex mixture of soluble products made up of a vesicular fraction made up of microvesicles

and exosomes that are involved in the transference of proteins and genetic material (such as microRNAs) to other cells. Due to its homing abilities, the MSCs secretome is now being recognized as a potential active pharmaceutical component, and its vesicular portion has been demonstrating promising characteristics to be used as a drug delivery system. This creates an ideal opportunity for the release of specific and targeted substances (drugs, proteins, etc.) into damaged lesions. For instance, MSCs modulate the immune response (Blaber et al., 2012) by enhancing the synthesis of anti-inflammatory cytokines including IL-10 and IL-4 (Aggarwal and Pittenger, 2005). Moreover, MSCs produce bioactive substances to act as inhibitors for fibrosis and apoptosis and to act as inducers for angiogenesis, mitosis and / or differentiation of progenitor cells, activating target cells or neighbouring cells to release biologically active substances (Caplan and Dennis, 2006). All in all, MSCs impact the various events during regeneration such as, cell migration, inflammation, proliferation and tissue remodelling (Hocking and Gibran, 2012). Many recent studies suggest that MSCs are the main candidates for cell mediated therapies and tissue regeneration (Sasaki et al., 2008) not only for their differentiation ability but also for their ability to produce active biomolecules (Caplan and Dennis, 2006). To avoid ethical issues related to applying animal material to human therapies, this sudy aimed to collect and analyze proteomic content of MSC secretions on 3D scafoold prepared from plant cellulose for further appplications for human therapies.

MATERIALS AND METHODS

Extraction of Cellulose

This process was achieved as described by Singanusong et al., (2013). Breifly, fresh banana yellow peels were obtained and immediately cut into pieces 0.3×2.5 cm in size and dried at 55°C for 10 h to reach a moisture content of 7.5% (dry basis) using a hot air convection oven. After cooling to room temperature, the peel was weighed, ground, and passed through a 35 mesh testing to obtain banana peel powder (BPP) and kept in a polyethylene plastic bag then placed in a refrigerator at 4°C until analysis. Extraction of fat was achieved by soaking BPP in gradual ethanol concentrations i.e., 90%, 95%, and 99% for 8h, 16h, and 24 h. Twenty grams of BPP were mixed with 200 ml of ethanol 10% (w/v) in a water bath at 50°C and incubated in a shaker at a shaking speed of 150 rpm. Then it was washed 3 times with distilled water and filtered with Whatman paper No. 4 and the defatted banana peel powder was dried in a hot air oven at 80°C for 7 h at a velocity 304.8 mm/h. The sample soaked in distilled water served as a control. Extraction of protein was done by soaking BPP in sodium hydroxide (1:10 w/v) at 3 pH levels of 11.6, 11.8, and 12.0 for 8, 16, and 24 h and that soaked in distilled water served as a control. The experiment was conducted in a water bath at 50°C with a shaking speed of 150 rpm. The samples were washed 3 times with distilled water, filtered with Whatman paper No. 4, and dried in the hot air oven at 80+2oC for 7 h. The defatted and protein-removed BPP was soaked in hydrogen peroxide solutions of 10, 15, 20, and 30% for 1.5, 3.0, 4.5, 6.0, and 7.5 h. The bleached samples were washed 3 times with distilled water, filtered with Whatman paper No. 4, and dried in the hot air oven at 60+2°C for 10 h.

Preparation and Evaluation of the 3D Cellulose Scaffold

A circular pieces of thin polystyrene paper of a diameter of 2 cm was coated with 0.01 mg/ml collagen followed by spreading BPP to prepare 3D cellulose scaffold and left overnight at 37°C to settle the kept at 4°C until use. The cellulose scaffold was characterized by scanning electronic microscope (SEM) to assess thickness and porosity. Upon use, cellulose scaffold was fixed between two circular rubbers with a diameter of 1 cm in a sandwich panel. The cells are grown on the upper surface of the scaffold and culture will be exposed to media from top and bottom.

Cell Culture

All cell culture was performed in class II cabinets to reduce the risk of infection and contamination. Then, all the cells and 3D-BEM models were incubated at standard culture conditions (SCC) which were provided by humidified incubator adjusted to 37ºC, ≈20% O_2 and 5% CO_2. Unless otherwise stated, all culture conditions set out in this study for cell culture and cell experiments were SCC. MSCs were previously isolated form

adipose tissue form donors undergone plastic surgery and grown in StemMACS MSC Expansion Media, human (UK) supplemented with foetal calf serum and L-Glutamine. Before use, it was supplemented with 5 ml of 100IU/ml penicillin - 100 µg/ml streptomycin (MSC Media) as described by Al-Shaibani (2022). To split cells, old media was aspirated, and cells were rinsed with phosphate buffered saline (PBS), trypsinised with trypsin 1X and incubated for 10 minutes. A quantity of 10 ml of MSC Media was added to prepare the cell suspension and centrifuged at 1500 rpm for 5 minutes. The supernatant was discarded, and cells were counted and a cell number of 5×10^4 was seeded into a T175 flask. A volume of 15-20 ml of fresh MSC Media were added to the flask and incubated under SCC. Trypan blue stain was used to assess viability of cells. A volume of 10 µl trypan blue was mixed with 10 µl of cell suspension. A quantity of 10 µl of this mixture was transferred to a Neubauer chamber and covered with a cover slip prior to counting under the microscope (Sandell and Sakai, 2008). (Equation 1) and (Equation 2) were used to calculate cell count and cell viability.

$$Total\ Cell\ Count = No\ of\ cells\ in\ 25\ small\ square \times Dilution\ Fcator \times 10^4 \qquad Equation\ 1$$

$$Cell\ Viability = \frac{No\ of\ viable\ cells\ in\ 25\ small\ square}{Total\ cell\ count\ (Live + dead\ cells)} \times 100\ \% \qquad Equation\ 2$$

Construction and Evaluation of MSC 3D culture

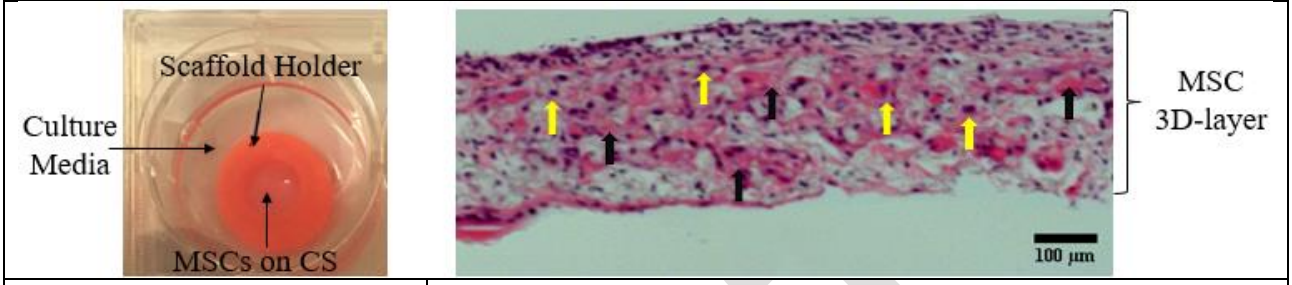
Prior seeding cells, the cellulose scaffold (CS) was activated with 70% ethanol for 30 seconds, treated with MSC media for one minute, fixed in Petri dish (30×15 mm) and incubated under SCC until required. To establish MSC 3D-culture, 3×10^6 cells were suspended in 200 µl MSC media, seeded onto the CS and incubated at SCC for 90 minutes. Then, 20 ml of MSC media was added to the Petri dish and covered the entire scaffold, and the culture was incubated at SCC for 7 days, with regular feeding of the cells with MSC media every two days. Feeding the culture included withdrawal of old medium from the well using a micropipette and then 20 ml of fresh MSC medium was gently added on the inside wall of the petri dish without disturbing the cells on the CS. Sections of MSC 3D-culture were stained with haematoxylin-eosin (HE) to evaluate the formation of 3D structure of MSCs and to assess cell penetration through the CS. Briefly, paraffin embedded sections of MSC 3D-culture were always kept on ice during sectioning to ensure effective sectioning. The microtome was adjusted to 4 µm and sections were kept in a water bath adjusted at 40°C for 2-3 minutes before transferring to slides. The slides were then heated in an oven at 60°C overnight and the next day, dehydrated by washing for 5 minutes in each xylene, 100% ethanol, 95% ethanol, 70% ethanol and distilled water. Slides were then stained with hematoxylin-eosin (HE) to evaluate the formation of 3D-tissue of MSC 3D-culture. Sections were then washed four times in TBS for 2 minutes, covered with DPX, left in the dark at room temperature overnight, and analysed the next day using fluorescent microscope (Leica).

Collection of MSC Secretome and Analysing Its Content

The MSC secretome (MSC-CM) from MSC 3D-culture was collected in standard DMEM serum free supplemented with 4% of 200 mM L-Glutamine, 1% of 100 IU/ml penicillin, and 100 lg/ml streptomycin (PS) (Sigma/UK). In the case of 2D culture, when cells reached 80% confluence, it was considered day zero time point, old media was discarded, and a confluent monolayer of MSC culture was washed three times with PBS and fresh DMEM serum free was added to the culture. MSC secretions were collected from three samples (N = 3) seeded in 2DCC on day one and day two and are referred to as 24-2D and 48-2D. On the other hand, in 3D culture, MSCs formed a tissue like structure within 7 days, which is considered the day zero time point. On day zero, the old media was discarded and 3D MSC model was washed three times with PBS and fresh DMEM serum free was added to the culture. Following days, MSC secretome was collected from three samples (N = 3) of MSC 3D culture at day one, and day two, and were referred to as 24-3D and 48-3D. The MSC-secretions from 2DCC and 3D MSC model were collected and filtered using a 0.2 lm filter unit before being analyzed by ELISA for growth factors and cytokines such as stromal derived factor-1 (SDF-1: Cat No. EHCXCL12A) and interleukin 1 (IL-1: Cat No. EH254RB). ELSA kits were purchased from (Life Technologies Thermo Fisher Scientific/US) and assays were performed according to protocols stated by the manufacturer company.

Results

MSC 3D culture generated in this study was prepared by seeding MSCs on the CS for 7 days. As shown in figure (1a), the structure of MSC 3D culture is composed MSCs grown on the CS which was fixed by a plastic holder (orange part) and fed with MSC medium for 7 days. The scaffold now bearing MSCs and formed cellular layer of MSCs could be used to generate MSC secretions. The MSC 3D cellular layer upon staining with hematoxylin– eosin revealed that MSCs distributed within the CS and formed a tissue-like structure in which cells are distributed in a 3D environment that mimics an *in vivo* niche as illustrated in figure (1b).



(a) (b)

Figure 1. Three-dimensional structure of MSCs on CS.

(a) MSCs grown on CS in tıssue culture plate and fed with MSC medium for 7 days. (b) Hematoxylin–eosin (HE) staining shows 3D culture of MSCs at day 7 after seeding the MSCs over the CS, revealing the formation of a cellular layer of MSCs that mimics an *in vivo* niche. Cells spread and deeply penetrated the scaffold and formed 3D tissue-like structure of MSC dressing. Yellow arraows are pointing out to MSCs, red arrows are pointing out to cytoplasm.

MSCs secreted detectable levels of SDF-1 and IL-1. Multiple comparisons were investigated between MSC secretions using Tukey's multiple comparisons test. The first one compared concentrations of SDF-1 and IL-1collected from 2DCC at different time points i.e., 24 h and 48 h. MSC secretions contained different concentrations of SDF-1 and IL-1 collected from their culture at 24 h, with consistent secretion of these growth factors within this time until 48 h. Interestingly, there was a decrease in growth factor concentrations at time point 48 h. Two-way ANOVA revealed significant differences in concentrations of SDF-1 and IL-1 at time points of 24 h and 48 h. As shown in (Figure 2A), SDF-1 level decreased from 188 ± 6 ng/ml to 109 ± 6 ng/ml ($P = 0.02$), IL-1 concentration depleted from 173 ± 13 ng/ml to 85 ± 7 ng/ml ($P = 0.01$). The second compared between SDF-1 and IL-1collected from 3DCC at different time points. MSC secretions collected from 3DCC contained consistent levels of SDF-1 and IL-1 within time change. Two-way ANOVA revealed no significant differences ($P > 0.05$) in concentrations of SDF-1 and IL-1 at time points 24 and 48 as shown in (Figure 2B). The third comparison investigated differences between concentrations of SDF-1 and IL-1 collected from 2DCC and 3DCC at the same time points. Two-way ANOVA showed significant variations between concentrations of MSC secretions collected from both 2DCC and 3DCC. As shown in (Figure 3C), at time point 24 h concentrations of SDF-1 and IL-1 collected from 3DCC were significantly higher than those collected from 2DCC. Additionally, at time point 48 h, concentrations of SDF-1 and IL-1 collected from 3DCC were also significantly higher than those collected from 2DCC as shown in and (Figure 2D).

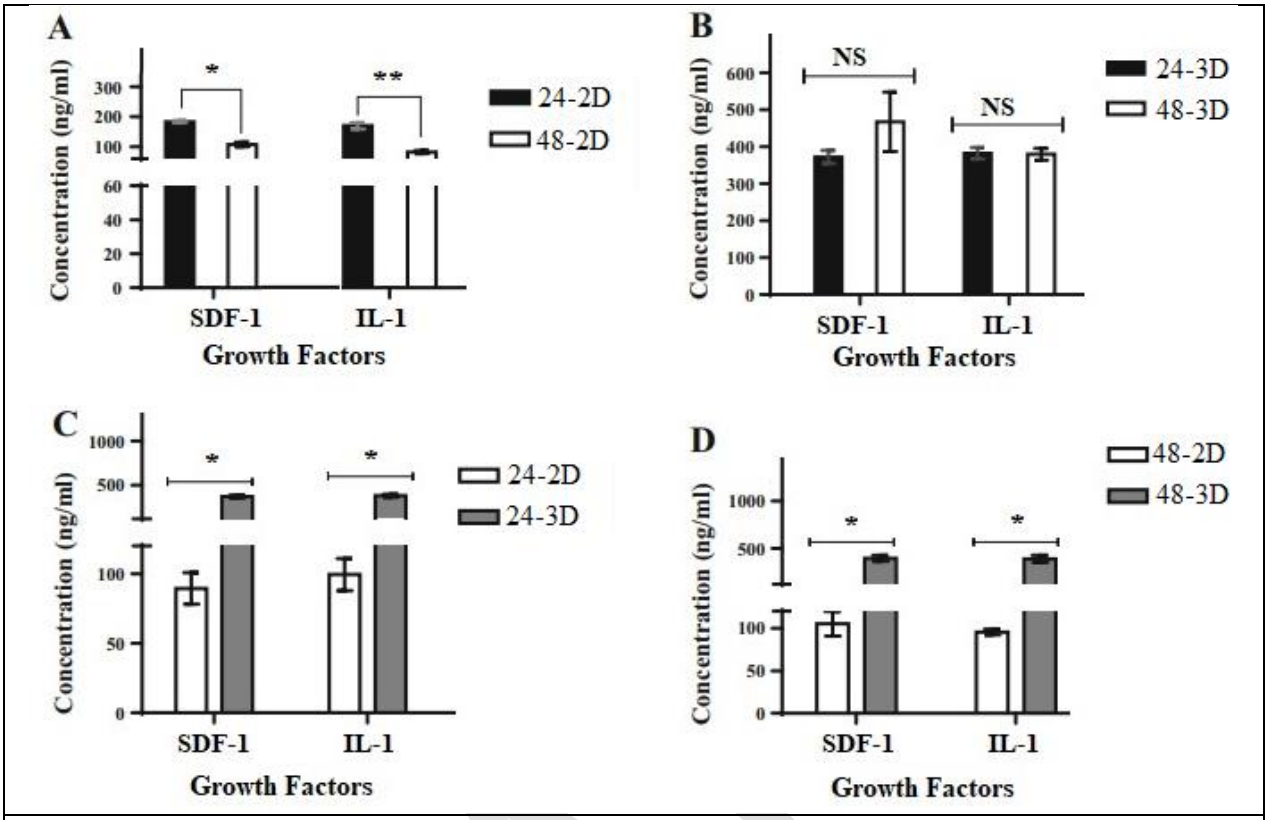


Figure 2. Analysis of MSC secretions by ELISA. Concentrations of SDF-1 and IL-1 secreted by MSCs in both 2DCC and 3DCC at 24 h and 48 h.

A- Concentrations of SDF-1 and IL-1 collected from 2DCC at different time points show a significant decrease in concentrations of SDF-1 and IL-1at 48 h compared to 24h. B- Concentrations of SDF-1 and IL-1 collected from 3DCC at different time points show no significant differences in concentrations of all target growth factors and cytokines at the different time points (24h and 48h). (C and D) Statistical comparisons show that concentrations of SDF-1 and IL-1 collected from 3DCC are significantly higher than their counterparts collected from 2DCC at 24h and 48h, respectively. The data is represented as a mean of the concentration of SDF-1 and IL-1. N = 4, error bars = standard error of the mean (SEM).

Discussion

It has been reported that MSC secretions could be applied for various therapeutic appliactions (James *et al*., 2006). Therefore, this study has investigated the ability of MSCs to secrete two important cytokines which are SDF-1 and IL-1. It has been previously reported that MSCs in 3D cell culture secrete many growth factors and cytokines including platelet derived growth factor (PDGF-AB), transforming growth factor-1 (TGF-1), hepatocyte growth factor (HGF), stromal derived factor-1 (SDF-1), interleukin 1 (IL-1), and interleukin 6 (IL-6) (Al-Shaibani, 2022). Therefore, MSCs has been used in this study to establish MSC culture in 3DCC and further collect and analyse their secretions using CS prepared from banana peels. MSCs has formed 3D tissue like structure in 3D cell culture and secreted SDF-1 and IL-1 which are collectively or separately play vital roles in regeneration and development (Alfaro *et al.*, 2013). SDF-1 plays a role in regulating skin homeostasis and tissue remodelling (Werner, 2011). It also promotes wound closure by inducing cell migration. Many of which play an integral role in inflammation, proliferation, migration, differentiation, collagen regulation, angiogenesis and the remodelling phases of the healing process (Cowin et al., 2001). The 3D microenvironment of the living organism is primarily responsible for the biological behavior of cells in vivo, which differs dramatically from their behavior in vitro (Edmondson et al. 2014). According to research by Qiu et al. (2018), MSCs that are grown on scaffolds made of extracellular matrix (ECM) promote tissue regeneration. In contrast to 2DCC cells, which lack the

proper ECM and hence are unable to exercise their normal behavior as in vivo and further impact their secretome, ECM play a crucial part in the successful proliferation of cells in vivo and allow the cells to connect with one another. As a result, the 3D scaffold might be employed instead of the ECM to depict a region that offers mechanical support for cells to establish their proper niche for further cell-cell or cell-ECM communication, in order to establish the appropriate spatial spreading (Lv et al. 2017). Consequently, regular growth factor and cytokine secretions. Since the 3D architecture is later supported by the secretion of ECM representing glycosaminoglycan secreted by MSCs, it is possible to credit the consistent secretions of growth factors and cytokines gathered from 3DCC culture to the 3D architecture. Additionally, the ECM released by MSCs serves as a link between cells, allowing cells to communicate with one another, allowing MSCs to remain stationary and function without becoming immobile while conserving energy and improving performance (Qiu et al. 2018). However, selecting the right scaffold is one of the most important aspects that affects cell biology and behavior. Collectively, the 3D culture of MSCs on CS enhance them to secrete high concentrations of these bioactive molecules that guide and judge the healing process in acute and chronic wounds by coordinating numerous cellular responses of different cell types resulting in the re-establishment of the damaged barrier (Barrientos et al., 2008).

CONCLUSION

The created 3D-MSC model secretes more growth factors and cytokines than those obtained using conventional 2DCC, indicating that banana peels is a good source for extraction cellulosea for further using in 3D cell culture to grow cells of various types and collect their secretions.

CONFLICT OF INTEREST

No conflict of interest

REFERENCES

Aggarwal S, Pittenger MF (2005). 'Human mesenchymal stem cells modulate allogeneic immune cell responses', *Blood*, 105(4), pp. 1815-22. https://doi.org/10.1182/blood-2004-04-1559

Alfaro MP, Deskins DL, Wallus M, DasGupta J, Davidson JM, Nanney LB, Gannon M, Young PP 2013. 'A physiological role for connective tissue growth factor in early wound healing', *Laboratory Investigation*, 93(1), pp. 81-95. https://doi.10.1038/labinvest.2012.162

Al-Shaibani MBH (2022). Three-dimensional cell culture (3DCC) improves secretion of signaling molecules of mesenchymal stem cells (MSCs). *Biotechnology Letters*, 44, 143–155. https://doi.org/10.1007/s10529-021-03216-9

Barrientos S, Stojadinovic O, Golinko MS, Brem H, Tomic-Canic M 2008. 'Growth factors and cytokines in wound healing', *Wound Repair Regeneration*, 16(5), pp. 585-601. https://doi.org/10.1111/j.1524-475X.2008.00410.x

Blaber S, Webster R, Hill C, Breen E, Kuah D, Vesey G, Herbert, B 2012. 'Analysis of in vitro secretion profiles from adipose-derived cell populations', *Journal of Translational Medicine*, 10(1), p. 172. https://doi.org/10.1186/1479-5876-10-172

Caplan AI, Dennis, JE (2006). 'Mesenchymal stem cells as trophic mediators', *Journal of Cellular Biochemistry*, 98(5), pp. 1076-84. https://doi.org/10.1002/jcb.20886

Cherian BM, Pothan LA, Nguyen-Chung T, Mennig G, Kottaisamy M and Thomas S. (2008). A novel method for the synthesis of cellulose nanofibril whiskers from banana fibres and characterisation. Journal of Agricultural and Food Chemistry, 56(14), 5617–5627. https://doi.org/10.1021/jf8003674

Cowin AJ, Kallincos N, Hatzirodos N, Robertson JG, Pickering KJ, Couper J, Belford DA 2001. 'Hepatocyte growth factor and macrophage-stimulating protein are upregulated during excisional wound repair in rats', *Cell Tissue Research*, 306(2), pp. 239-50. https://doi.org/10.1007/s004410100443

Edmondson R, Broglie JJ, Adcock AF, Yang L (2014) Threedimensional cell culture systems and their applications in drug discovery and cell-based biosensors. Assay Drug Dev Technol 12(4):207–218. https://doi.org/10.1089/adt.2014.573

Firmanda A, Syamsu K, Widya Sari Y, Cabral J, Pletzer D, Mahadik B, Fisher J and Fahma F. (2022). 3D printed cellulose based product applications. Mater. Chem. Front. 2022, 6, 254–279. https://doi.org/10.1039/D1QM00390A

Hariani PL, Riyanti F. Asmara RD. (2016). Extraction of cellulose from kepok banana peel (Musa parasidiaca L.) for adsorption procion dye. Molekul, 1(1), 135. https://doi.10.20884/1.jm.2016.11.1.202

Hocking AM (2012). 'Mesenchymal Stem Cell Therapy for Cutaneous Wounds', *Advanced Wound Care (New Rochelle)*, 1(4), pp. 166-171. https://doi.10.1089/wound.2011.0294

Iravani S and Varma RS. (2022). Cellulose-Based Composites as Scaffolds for Tissue Engineering: Recent Advances. Molecules, 27, 8830. https://doi.org/10.3390/molecules27248830

James D, Noggle SA, Swigut T, Brivanlou AH 2002. Contribution of human embryonic stem cells to mouse blastocysts, Developmental Biology,295 (1): 90-102. https://doi.org/10.1016/j.ydbio.2006.03.026

Lv D, Hu Z, Lu L et al (2017) Three-dimensional cell culture: A powerful tool in tumor research and drug discovery (Review). Oncol Lett 14(6):6999–7010. https://doi.org/10. 3892/ol.2017.7134

Qiu X, Liu S, Zhang H et al (2018) Mesenchymal stem cells and extracellular matrix scaffold promote muscle regeneration by synergistically regulating macrophage polarization toward the M2 phenotype. Stem Cell Res Ther 9(1):88. https://doi.org/10.1186/s13287-018-0821-5

Sandell L. and Sakai D. (2008) 'Mammalian Cell Culture', in *Current Protocols Essential Laboratory Techniques*. John Wiley & Sons, Inc.

Sasaki M, Abe R, Fujita Y, Ando S, Inokuma D. and Shimizu H. (2008). 'Mesenchymal Stem Cells Are Recruited into Wounded Skin and Contribute to Wound Repair by Transdifferentiation into Multiple Skin Cell Type', *The Journal of Immunology*, 180(4), pp. 2581-2587. https://doi.org/10.4049/jimmunol.180.4.2581

Singanusong R, Tochampa W, Kongbangkerd T. and Sodchit C. (2013). Extraction and properties of cellulose from banana peels. Suranaree J. Sci. Technol. 21(3):201-213. https://doi.10.14456/sjst.2014.1

Teixeira FG and Salgado AJ. (2020). Mesenchymal stem cells secretome: current trends and future challenges. Neural Regeneration Research 15(1): p 75-77. | https://doi.10.4103/1673-5374.264455

Werner S 2011. 'A Novel Enhancer of the Wound Healing Process: The Fibroblast Growth Factor-Binding Protein', *The American Journal of Pathology*, 179(5), pp. 2144-2147. https://doi.10.1016/j.ajpath.2011.09.001

Zahorec P, Koller J, Danisovic L, Bohac M 2015. 'Mesenchymal stem cells for chronic wounds therapy', *Cell Tissue Bank*, 16(1), pp. 19-26. https://doi.10.1007/s10561-014-9440-2

ORAL PRESENTATION

Effect of temperature on rheologic properties of pasteurized cow's milk

Anisa Dhroso[1*], Hasime Manaj[2], Laura Shabani[3], Ilirjan Malollari[4]

[*1, 2, 3, 4] University of Tirana, Faculty of Natural Sciences, Department of Industrial Chemistry, Tirana, Albania.

*anisa.dhroso@fshn.edu.al

Abstract

The physico-chemical and rheological properties of pasteurized cow's milk available in Albania market depend on temperature and time storage were determined, which can evaluate the quality of food product. In the first phase, fat, solid non-fat (SNF) and density were determined by Lactoscan apparatus with different temperature. In the second phase of the study, dynamic viscosity was measured using the Falling Ball Viscosimeter, temperature range -20°C to 150°C, viscosity range 0.5 mPas to 10^5 mPas, reproducibly <0.5%. All the samples were measured in temperature range 5-30°C. The density and dynamic viscosity value of milk are used to calculate kinematic viscosity. Dependence of density, dynamic viscosity, kinematic viscosity and fluidity related to temperature were described by exponential functions. It was found that, viscosity of milk decreases non-linearly with increasing temperature. Therefore, temperature is one of the most important factors, which determine quality of milk.

Keywords:rheological properties, temperature, pasteurized milk

INTRODUCTION

Albania is a small European country located in the south-eastern part of the continent, with population amounts to 2.9 M inhabitants. The total land size of Albania is 2,875,000 ha, from which 24% is agriculture land, 36% forestry, 15% pastures and meadows, and 25% is classed as "others". (INSTAT, 2019) Milk production activities have a long tradition in Albania due to the favourable natural resources and is one of the most developed agricultural sub-sectors in the country. Over the past five years, the dairy processing industry has undergone important changes. The milk and dairy industry are second largest industry in terms of number of enterprises (12.8% of total), after flour and bakery industry. The region with the largest milk production is Fier, contributing to 19.4% of total milk production, and together with the regions of Elbasan, Korçe and Tirane make together up to 52.5% of the total milk production of the country. The same regions are the largest producers of cow's milk (54.7%). (Eurostat, 2019) Liquid milk category includes semi-pasteurised milk (a semi-processed product), pasteurised milk in bulk and retail packaging and UHT milk Pasteurized milk is raw milk that has been heated to a specified temperature and time to kill pathogens without the product being damaged. Pathogens are microorganism such as bacteria that make us sick. Raw milk can contain pathogens such as Campylobacter, E. coli, Salmonella, Listeria and other bacteria. (Janzen, J. J., et al., 1982) Raw milk includes milk from cows, goats, sheep and other dairy animals. An optimum mixture of carbohydrates, proteins, and fats in these products dictates the rheological properties. Interactions of three basic ingredients (carbohydrates, proteins and fats) along with water and other minor additives during shearing, mixing and heating interfere in product formulations. (Ahmed, J., et al.., 2006) Milk is a complex fluid consisting of seven main components: water, fat, protein, sugar (lactose), minerals, vitamins and enzymes. For the quality evaluation of food materials, it is important to know their physical properties particularly, mechanical, rheological, and thermo physical. (Manzocco L., et al, 1998) In this article, the results of measurement of physicochemical and rheological parameters are presented for a sample of pasteurised cow's milk. The dynamic viscosity of milk was experimentally determined as a function of temperature (5 to 30°C). The variation of the viscosity of cow's milk with the temperature is analyzed applying the two and multi constant equations.

DRAFT

DRAFT

MATERIALS AND METHODS

The physico-chemical and rheological properties of pasteurized cow's milk available in Albania market depend on temperature and time storage were determined. The milk sample during experimental part was storage at the temperature 4C. All the measurement were made in Chemical Process Engineering laboratory in collaboration with Food Chemistry laboratory. The measurement were performed in temperature range 5-30C, at the beginning of storage (first day of bottle opening) and after one week of storing (last day of bottle expiration). In the first phase, physicochemical properties (fat, solid non fat (SNF), lactosa, add water, freezing point, salts and density) were determined by Lactoscan apparatus in different temperature. The device is electronic and quite simple to use. After placing the milk sample in the device, in a certain amount the button is pressed to start the analysis and after a short period of time the physico-chemical data appear on the screen. The function of the milk analyzer Lactoscan is to make quick analysis of milk. All measurements were performed in three repetitions. The density of milk is used to calculate kinematic viscosity. The dynamic viscosity measurement can be done by various types of viscometers: falling ball viscometers, rotational viscometers, capillary flow viscometers and orifice type viscometers. (Sahin and Sumnu, 2006) Cow's milk dynamic viscosity was measured using the Falling Ball Viscosimeter, temperature range -20°C to 150°C, viscosity range 0.5 mPa.s to 10^5 mPa.s, reproducibly <0.5%. Viscosity of cow's milk is important in determining the flow conditions in dairy processes. (Kessler, H.G., 2002) Kinematic viscosity and fluidity were determined according to the definitions. Office Excel software was used to carry out the effect of temperature on dynamic viscosity of milks by different mathematical models. Where, the dynamic viscosity of milk was represented as a function of temperature by using two and multiconstant proposed mathematical models. Equation 1, include the Arrhenius model that is commonly used to model temperature dependence of a property (Clements L. D. et al., 1982):

$$\mu = \mu_{\infty,T} \exp\left(\frac{E_a}{RT}\right) \quad (1)$$

Where μ is the dynamic viscosity in mPa.s, $\mu_{\infty,T}$ is the viscosity at infinite-temperature in mPa.s, E_a is the exponential constant that is known as activation energy (J/mol); R is the gas constant (J/mol.K) and T is the absolute temperature Kelvin. (Munson, B. R., et al, 1994) Multi-constant formula known as Andrade (three constant) and Clements (four constant) models that are represented in the following equations (Abramovic H., Klofutar G. 1998) (Clements L. D., Noureddini H. and Teoh B. C. 1982):

$$Ln\mu = A + \frac{B}{T} + \frac{C}{T^2} \quad \text{and} \quad Ln\mu = A + \frac{B}{T} + \frac{C}{T^2} + \frac{C}{T^3} \quad (2)$$

Where μ is the dynamic viscosity in mPa.s, T is the temperature in Kelvin. A, B and C are constants. Kinematic viscosity ($m^2 \cdot s^{-1}$) is defined as a ratio of dynamic viscosity to density of fluid ρ ($kg \cdot m^{-3}$) at the same temperature.

$$\eta = \frac{\mu}{\rho} \quad (3)$$

Reciprocal value of dynamic viscosity is called fluidity and unit of fluidity is $mPa^{-1} \cdot s^{-1}$ (Hlaváč P., et. al 2016).

$$\varphi = \frac{1}{\mu} \quad (4)$$

The mean absolute percentage error (MAPE), which indicates the deviance of the observed values from the calculated, was calculated using the following formula:

$$MAPE^a = \frac{\left|\sum_{1=1}^{n}\left(\frac{A_0 - A_C}{A_0}\right)\right|}{n} \cdot 100 \quad (5)$$

Where A_0 is the observed value, AC is the calculated value, and n represents the number of pairs of samples.

RESULTS AND DISCUSSION

The physico-chemical and rheological properties of pasteurized cow's milk available in Albania market depend on temperature were determined. Measured values of physicochemical properties (fat %, non fat solid %, protein %, lactosa %, add water %, freezing point %, salts %, density %) at the first and last day of storage

are presented on Figure 1 (a) and (b). By comparing our experimental data with standard value, we can see that they are roughly the same.

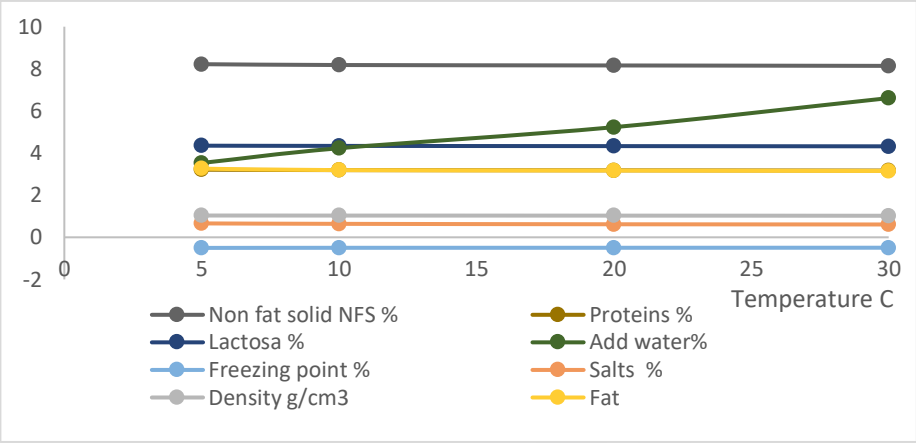


Figure 1 (a) Physicochemical properties of pasteurized cow's milk on the first day of bottle opening

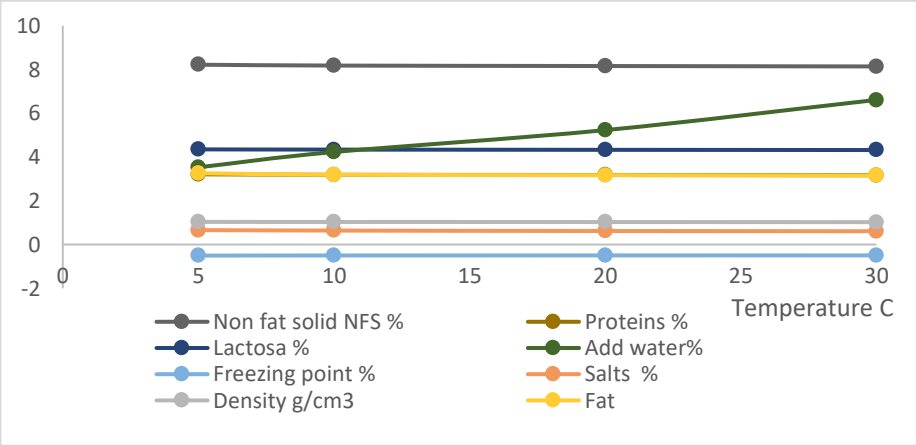


Figure 1 (b) Physicochemical properties of pasteurized cow's milk on the last day of bottle expiration

Milk fat is one of the most important parts of milk and depends on the type of dairy animal, breed and feeding of the animal. We see from the results obtained that the fat content in the samples taken for analysis ranges from 3.12 - 3.24% for samples before the expiration date and 3.15-3.26% for the samples after the expiration date. The determination of milk fat is of great importance to evaluate the stability and quality of milk during the storage period. Today, these analyses are useful, because on the basis of their results, the control of the milk fat of individual cows is ensured. Total milk solid (fat + SNF) contains all the constituent parts of milk such as: proteins, fats, lactose, mineral salts, vitamins with the exception of water. Total milk solid is within standard value 11-13%. Milk proteins are of great importance in the nutritional value and in the technological process. The protein value on the last day of storage is lower due to the hydrolysis of heat-resistant enzymes of bacterial origin (psychrophilic bacteria). The sample gives lower values ranging from 3.13-3.25% before expiration and 3.17-3.21% after expiration. Density of milks is also influenced by amount of fat content. The highest fat content had caused the lowest density. (Hlaváč P. and Božiková M., 2016) In milk, the main ingredient is lactose. Lactose is a combination of glucose and galactose and has a light sweet taste. Lactose loses its quantity through fermentation. Before and after the expiration date, we see that the values of added water are higher. The increase in the values of added water reflects the decrease in density and SNF. Regarding the amount of salts, we see that there are no big differences between the samples.

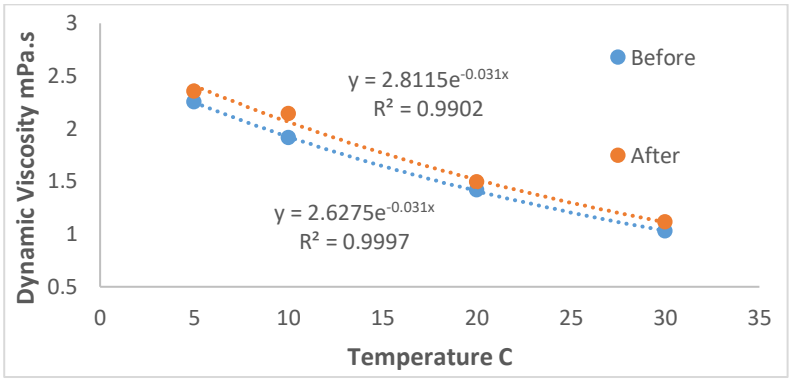


Figure 2 (a) *Effect of temperature on dynamic viscosity*

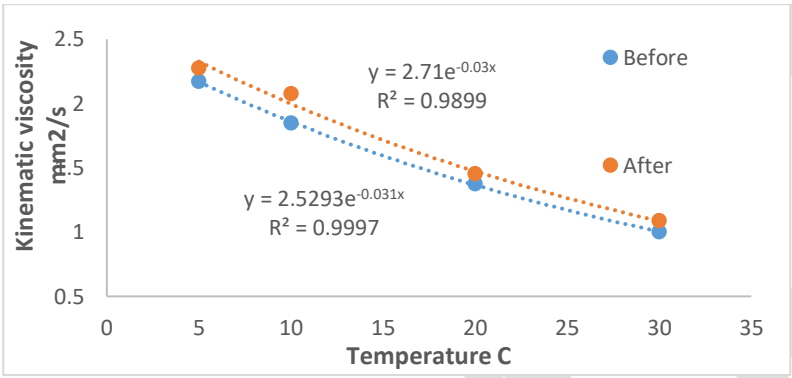


Figure 2 (b) *Effect of temperature on dynamic viscosity*

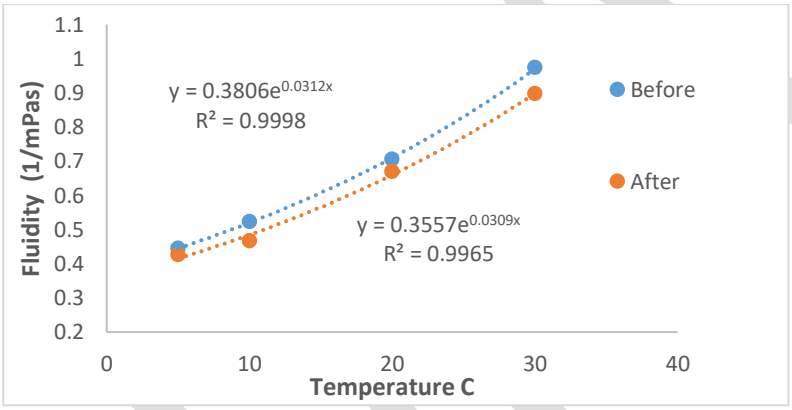


Figure 2. (c) *Effect of temperature on dynamic viscosity*

The experimental data of cow's milk before and after expiring date, for dynamic viscosity (mPa.s) versus temperature is shown in Figure 2 (a). The dynamic viscosity is decreasing with increasing of temperature. The values of dynamic viscosities are a bit higher after one week of storing, because of water loosening.

The kinematic viscosity can be calculated from the dynamic viscosity and the density (equation 3). On Figure 2 (b) is presented kinematic viscosity decreasing versus temperature range 5 – 30°C. The values at the beginning of storing are a bit higher than after.

While fluidity is increasing with increasing of the temperature, is presented in Figure 2 (c). The values of the estimated constants and correlation coefficients are shown in each equation, set in the graphs.

The experimental data, for dynamic viscosity fitting by Arrhenius, Andrade and Clements models are presented in Fig. 3 (a-c), by using Equation 1 and 2 respectively.

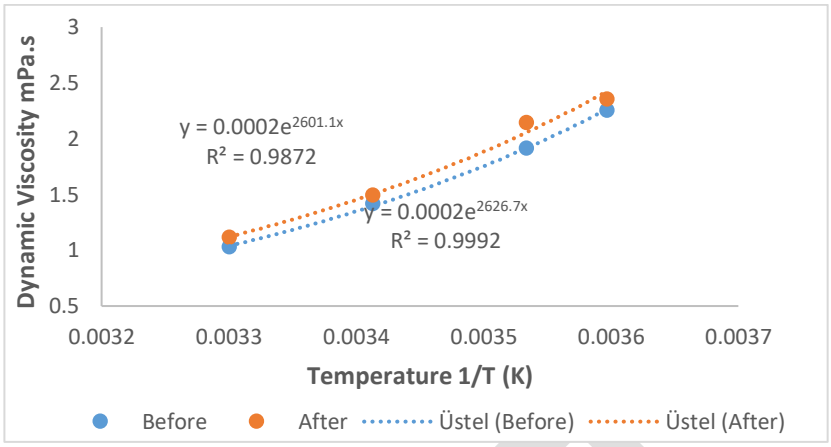


Figure 3. (a) *Effect of temperature on Arrhenius model*

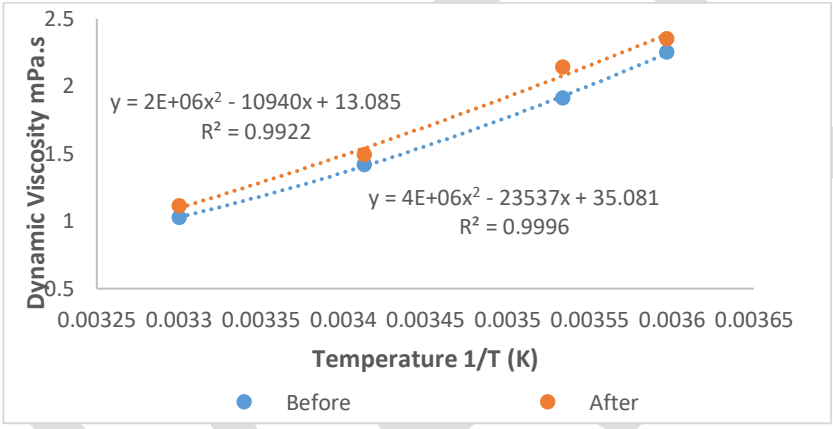


Figure 3. (b) *Effect of temperature on Andrade model*

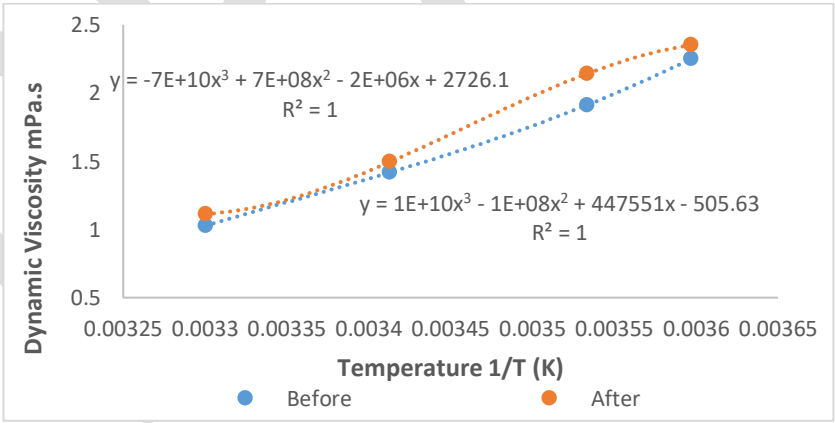


Figure 3. (c) *Effect of temperature on Clements model*

The values of the estimated constants are shown in each equation, set in the graphs. In all cases the determination coefficient (R^2) exceeded values >0.99. The mean absolute percentage errors (MAPEa) were below 10%: 7.3% Arrhenius model, 2.1% Andrade model and 0.38% Clements model. All of the mean absolute percentage errors were low, which means that the viscosity values obtained were very stable. However, comparisons of the calculated data indicate that the temperature-dependence of viscosity for the sample was best described by the Clements model. An equation with lower MAPE values gives a better fit to experimental data compared to an equation with higher MAPE values.

DRAFT

CONCLUSIONS

The article is focused in physicochemical parameters determination and on influence of temperature on rheological parameters of cow's milk made in Albania. The physicochemical parameters results depend on milk nature and can be used as a way of characterizing. The limit values of physicochemical and rheological parameters in milk were within the Albanian and European standard values. The dynamic viscosity versus temperature was measured and described by different mathematical models. The value of correlation coefficient and mean absolute percentage error indicates that the models fit satisfactorily to experimental data. However, comparisons of the results obtained indicate that the temperature dependence of viscosity was best described by the Clements model.

REFERENCES

Abramovic H., Klofutar G. (1998): ″The temperature dependence of dynamic viscosity for some vegetables oils ″, Acta Chim. Slov. 45(1), 69-77.

Ahmed, J., Ramaswamy, H.S., & Pandey, P.K. (2006). Dynamic rheological and thermal characteristics of caramels. *Food Science and Technology - Lebensmittel-Wissenschaft und - Technologie (lwt)*, 39, 216–224.

Clements L. D., Noureddini H. and Teoh B. C. (1992): ″Viscosity of vegetables oils and fatty acids ″, J. Am. Oil Chem. Soc., 69 (12), 1189-1191.

Eurostat (2020). EUROSTAT database. Available at https://ec.europa.eu/eurostat/data/database?node_code=earn_ses_monthly

Hlaváč P., Božiková M., Hlavačova1 Z., Kubik L. (2016),.: Influence of temperature and storing time on selected red wine physical properties, Acta Universitatis Agriculturae et Silviculturae mendelianae brunensis, volume 64, number 2, pg 433-439.

Hlaváč P., Božiková M., (2016): Temperature effect on milk selected physical properties: 186–190.

INSTAT (2019). The population of Albania. Retrieved from http://www.instat.gov.al/en/themes/demography-and-social-indicators/population/

INSTAT (2019). Land structure by Land structure, Type and Year (2017). Retrieved from http://databaza.instat.gov.al/pxweb/en/DST/START__BU__AGI/NewBU0009/table/tableViewLayout2/?rxid=243b6be0-5b40-4ba3-a237-f32b68f64a3c

Janzen, J. J., Bishop, J. R., Bodine, A. B., Caldwell, C. A. (1982). Shelf-life pasteurized fluid milk as affected by age of raw milk, Journal of Diary Science 65, Lemon University, SC, USA, 2233 – 2236.

Kessler, H.G. 2002. Food and Bio Process Engineering: Dairy Technology Edited by: Kessler, Verlag A. MÜnchen, , Germany

Manzocco L., Maltini E., Lerici C.R, (1998).: Changes of some thermal and physical properties in model system simulating an alcoholic fermentation. Journal of Food Processing and Preservation, 22: 1–12.

Munson, B. R., Young, D. F., Okiishi, T. H. (1994): Fundamentals of fluid mechanics, John Wiley & Sons, New York.

Sahin, S., Sumnu, S. G. (2006): Physical properties of foods, Springer, USA.

DRAFT

ORAL PRESENTATION

Maviyemiş (*Vaccinium corymbosum L.)* odun çeliklerinin köklenmesi üzerine IBA (Indole-3-Butyric Asit) uygulamalarının etkisi

Özgün KALKIŞIM[1](https://orcid.org/0000-0002-6129-8539), Mehmet Zahit AYDIN[2](https://orcid.org/0000-0002-7833-521X)

[1]Recep Tayyip Erdoğan Üniversitesi, Ziraat Fakültesi, Bahçe Bitkileri Bölümü 53100 Rize, Türkiye

[2]Recep Tayyip Erdoğan Üniversitesi, Ziraat Fakültesi, Bahçe Bitkileri Bölümü 53100 Rize, Türkiye

Sorumlu yazar e-mail: ozgun.kalkisim@erdogan.edu.tr

Özet

Bu çalışma maviyemiş bitkisinin vejetatif çoğaltma yöntemlerinden biri olan odun çelikleriyle çoğaltılması kapsamında, dinlenme döneminde yıllık sürgünlerden alınan odun çeliklerine 4 farklı Indole-3-Butyric Asit hormon dozlarının köklenme üzerine etkilerini araştırmak amacıyla yapılmıştır. Bu amaçla Rize ilinin Fındıklı ilçesinde bulunan kapama maviyemiş bahçesinde yetiştirilen Bluecrop maviyemiş çeşidinden alınan odun çelikleri kullanılmıştır. Odun çeliklerine Indole-3-Butyric Asit'in dört farklı dozu (Kontrol, 2500, 3500, 4500, 5500 ppm) uygulanmıştır. Çalışma alt ısıtmasız perlit ortamlı beton yastıkların bulunduğu polycarbon serada yürütülmüştür. Çalışma tesadüf parsellerinde faktöriyel deneme desenine göre 4 tekerrürlü olarak planlanmıştır. Odun çeliklerinde en yüksek köklenme oranı % 45 ile 3500 ppm dozundaki Indole-3-Butyric Asit uygulamasından elde edilmiştir. En düşük köklenme oranı ise % 10 ile kontrol grubu çeliklerinden elde edilmiştir. Köklenme oranı bakımından en iyi sonucu veren 3500 ppm dozundaki Indole-3-Butyric Asit uygulamasındaki canlı çelik sayısı oranı % 60, ortalama kök sayısı 2,22 adet ve ortalama kök uzunluğu ise 27,51 mm olarak tespit edilmiştir.

Tüm sonuçlar dikkate alındığında Bluecrop maviyemiş çeşidinin odun çeliklerinin köklenme ve kök gelişimi üzerine, 3500 ppm dozundaki Indole-3-Butyric Asit hormonu uygulamasının en iyi sonucu verdiği ve bu uygulamanın üreticilere önerilebileceği kanaatine varılmıştır.

Anahtar Kelimeler: Maviyemiş, çoğaltma, Indole-3-Butyric Asit

Abstract

This study was carried out to investigate the effects of 4 different Indole-3-Butyric Acid hormone doses of hardwood cuttings taken from annual shoots on rooting during the resting period, within the scope of propagation of blueberry plant with hardwood cuttings, which is one of the vegetative propagation methods. For this purpose, hardwood cuttings taken from Bluecrop blueberry variety grown in the orchard blueberry garden in Fındıklı district of Rize province were used. Four different doses of Indole-3-Butyric Acid (Control, 2500, 3500, 4500, 5500 ppm) were applied to hardwood cuttings. The study was carried out in a polycarbon greenhouse with perlite environment concrete pads without bottom heating. The study was planned according to the factorial experimental design in randomized with 4 replications. The highest rooting rate in hardwood cuttings was obtained from the application of Indole-3-Butyric Acid at a dose of 45% to 3500 ppm. The lowest rooting rate was obtained from the control group cuttings with 10%. The rate of viable cuttings in the application of 3500 ppm Indole-3-Butyric Acid, which gave the best results in terms of rooting was 60%, the average root number was 2.22 and the average root length was 27.51 mm.

Considering all the results, it was concluded that 3500 ppm dose of Indole-3-Butyric Acid hormone application gives the best results on rooting and root development of hardwood cuttings of Bluecrop blueberry variety and this application can be recommended to producers.

Keywords: Blueberry, propagation, Indole-3-Butyric Acid

1.GİRİŞ

Dünya üzerinde ılıman iklim kuşağında yetiştiriciliği yapılan maviyemiş ilk olarak 1900'lü yılların başında anavatanı olan ABD'de kültüre alınmıştır. Asitli topraklarda yetişen maviyemiş Fundagiller (*Ericaceae*) familyasının Vaccinium cinsi içerisinde yer almaktadır. Maviyemiş bitkisinin 450' ye yakın türünün olduğu bildirilmektedir. Maviyemiş bitkisinin kültüre alınmış türleri Vaccinium corymbosum L. (yüksek boylu), Vaccinium angustifolium (Alçak boylu), Vaccinium myrtilloides (Alçak boylu), Vaccinium ashei Reade (Tavşangözü) olarak adlandırılmaktadır. (Strik ve ark., 1993; Gough, 1994; Çelik, 2012). Son yılların popüler bir meyvesi olan maviyemiş sağlık açısından oldukça yararlı olmasının yanında birim alandan alınan verim oldukça yüksektir (Strik ve ark., 1993; Gough, 1994; Himelrick ve ark., 2002; Çelik, 2009a, 2009b ve 2009c; Çelik, 2012a).

Maviyemiş meyvesinin kullanım alanı oldukça fazladır. Taze meyve olarak, meyve suyu sanayisinde, ilaç sanayisinde, süt ve süt ürünlerinde, kuru meyve, meyveli ekmek, çörek, kek, puding, pasta, baharat sanayisinde, meyve salatalarında, reçel, marmelat, konserve sanayisinde, çay, diyet menüleri ile şarap gibi birçok alanda değerlendirilmektedir (Çelik, 2005).

FAO istatikliklerine göre 2020 yılı dünya maviyemiş üretiminde 294 bin ton ile ABD ilk sırada, 146 bin ton ile Kanada 2. sırada, 180 bin ton ile Peru üçüncü sırada yer almaktadır. Dünya maviyemiş üretimi 1961 yılında 26.091 ton iken, son 60 yılda 32 kata yakın üretim artışı gerçekleşmiştirb(FAO,2020).

Türkiye'de maviyemiş (Vaccinium corymbosum L.) bitkisinin üretimi her geçen gün artmaktadır. Ege ve Akdeniz bölgelerinde ağırlıklı olarak topraksız tarım ile yetiştirilmekle birlikte, Karadeniz ve Marmara bölgelerinde kapama bahçelerde de yetiştirilmektedir (Çelik, 2004; Çelik, 2006; Çelik ve Ağaoğlu, 2013).

Maviyemiş bitkisi vejetatif çoğaltma yöntemlerinden en yaygın olarak yapraklı yeşil çelik, odun çeliği, daldırma, dip sürgünü, ayırma ve doku kültürü (in vitro) yöntemleri ile çoğaltılabilmektedir (Abolins ve ark., 2003; Çelik, 2006; Çelik, 2007; Krewer ve Cline, 2006; Çelik ve Ateş, 2009; Çelik, 2012a; Çelik, 2012b; Çelik ve Ağaoğlu, 2013; Ruter, 2015; Çelik, 2016; Çelik, 2017; Colombo ve ark., 2018; Karabulut ve Çelik, 2019). Vejetatif çoğaltma yöntemlerinden biri olan aşı yöntemi ile; makineli hasat sırasındaki meyve kayıplarını önlemek için yüksek toplama tablası oluşturmak üzere çoğaltma çalışmaları da yapılmaktadır (Wei, 2011; Wei ve ark., 2013; Marino ve ark., 2014).

Türkiye'de ilk maviyemiş çoğaltma çalışmaları Çelik tarafından 2004 yılında başlatılmıştır. Çelik,2006 Temmuz ayında bazı maviyemiş çeşitlerinden aldığı yapraklı yeşil çeliklere 1000 ppm IBA uyguladıktan sonra sera ortamında alttan ısıtmalı (25°C) olarak Rekord çeliklerin de % 100.00 köklenme sağlamış, ısıtmasız Bluejay çeliklerindeki köklenmenin % 38,89 ile en düşük düzeyde kaldığını tespit etmiştir. Yapılan başka bir çalışmada Akbulut ve ark. 10 farklı maviyemiş çeşidinden kış döneminde aldıkları odun çeliklerine IBA, NAA ve GA3 'in 4 farklı dozunu (0, 250, 500, 750 ve 1000 ppm) uygulamışlardır. Maviyemiş odun çeliklerinde en yüksek köklenme oranını Nortland çeşidinde (% 52,5), en düşük köklenme oranını ise Golden Traube (% 2,5) ve Chandler (% 3,2) çeşitlerinden elde etmişlerdir.

Çeliğin alıdınığı sürgünün odunlaşma durumu da önemli olup odun çelikleri için yıllık sürgünler tercih edilmektedir. Yeteri kadar odunlaşmış ya da ikinci yıl sürgünleri, dinlenme döneminden sonra uyanan gözlerden çıkan sürgünler kadar iyi köklenmemektedir (Çelik, 2007; Çelik, 2012b, Pacholczak ve Nowakowska, 2015; Çelik, 2016). Çelik (2006), yaz ayında aldığı yapraklı yarı odun çeliklerinden alttan ısıtma (25°) uygulaması ile % 100 (Rekord) başarı sağlarken, alttan ısıtma uygulanmayan Bluejay yarı odun çeliklerinden ise % 39 başarı sağlayabilmiştir.

Doku kültürü tekniği, kısa sürede kitlesel üretim imkânı sağlandığı için son yıllarda tercih edilmektedir. Ancak, bu yöntem diğer yöntemlere kıyasla pahalıdır. Özel cihazlar ve ekipmanlar, laboratuvar ve bu alanda bilgili elemana ihtiyaç olmaktadır (Orlikowska, 1986; Brissette ve ark., 1990, Austin, 1994). Maviyemişte doku

kültürü ve çelikle çoğaltım metodlarını karşılaştıran Smolarz ve Chebelebowska (1998); büyüme, gelişme ve olgunlaşmada herhangi bir farklılık göstermez iken, dördüncü ve beşinci yıllardaki verimlerinin ise doku kültürü ile elde edilenlerde daha düşük olduğunu saptamışlardır.

DRAFT

2. Materyal ve Metod

2.1. Materyal

Bu araştırma; 2021-2022 dinlenme döneminde Recep Tayyip Erdoğan Üniversitesi Ziraat Fakültesinin deneme ve uygulama seralarında yürütülmüştür. Araştırmada Rize ilinin Fındıklı ilçesinde bulunan kapama maviyemiş bahçelerinden alınan odun çelikleri kullanılmıştır. Çalışmada Karadeniz bölgesinde oldukça yaygın yetiştiriciliği yapılan Bluecrop maviyemiş çeşidinin odun çelikleri kullanılmıştır. Odun çelikleri dinlenme döneminde bir yıllık dallardan 20-25 cm uzunluğunda ve 2-3 göz bulunduracak şekilde hazırlanmıştır. Odun çeliklerine dört farklı IBA (Indole-3-Butyric Asit) dozu (Kontrol-0, 2500, 3500, 4500, 5500 ppm) uygulaması yapılmıştır. Çalışma alt ısıtmasız beton yastıkların bulunduğu polycarbon serada yürütülmüştür. Köklendirme ortamı olarak perlit kullanılmıştır. Sera içi sisleme süresinin uzunluğu ile aralığı hava sıcaklığı ve oransal neme göre ayarlanmıştır. Çalışma tesadüf parsellerinde faktöriyel deneme desenine göre 4 tekerrürlü olarak planlanmıştır (Resim 1).



Şekil 1. Serada bulunan galvanize sac yastıklı perlit köklendirme ortamındaki odun çelikleri

2.2. Metod

Odun çeliklerinin, perlit ortamına dikim tarihinden itibaren 120 gün sonra sökülerek canlı çelik sayısı (adet) belirlenmiş, köklenmiş odun çeliği sayıları belirlenerek oranları (%) tespit edilmiş, ayrıca kök sayıları (adet) belirlenmiş ve kök uzunlukları cetvel yardımıyla ölçülerek tespit edilmiştir. Çalışma, 4 tekerrürlü ve her tekerrürde 10 çelik olacak şekilde tesadüf parselleri deneme desenine göre planlanmıştır. İstatistiki analizler JMP 13 paket programı kullanılarak yapılmıştır. F testine göre önemli bulunan ortalamalar arasındaki farklar, LSD testine göre farklı harfler kullanılarak ifade edilmiştir.

3.Bulgular ve Tartışma

Maviyemiş çeliklerinin köklenmesi ile ilgili elde edilen sonuçlar Tablo 1'de sunulmuştur. Yapılan hormon uygulamaları sonucunda en yüksek canlı çelik sayısı ve köklenme oranı Indole-3-Butyric Asidin 3500 ppm dozundan elde edilmiştir (Resim 2). Elde edilen sonuçlar değerlendirildiğinde en yüksek köklenme oranı % 45 ile 3500 ppm dozunda gerçekleşmiştir. Çelik ve Ateş (2009) ise maviyemiş mini çeliklerinde 2000 ppm IBA'nın köklenmeyi artırdığını saptamışlardır.

Araştırma kapsamındaki maviyemiş çeliklerinin ortalama kök sayıları üzerine farklı hormon uygulamalarının etkisi incelendiğinde, en yüksek değer yine IBA 3500 ppm dozu uygulamasında (2,22 adet), en düşük değer ise kontrol grubunda (0,30 adet) saptanmış olup, diğer uygulamalar arasındaki fark istatistiksel olarak önemli

bulunmuştur. Benzer konuda yapılan çalışmalarda Turna ve ark. (2013) hormon uygulanan çeliklerin hormon uygulanmayan çeliklere göre daha iyi köklenme meydana getirdiklerini tespit etmiştir. IBA hormonunun 1000 ppm ve 5000 ppm dozlarındaki köklenme oranı diğer hormon ve dozlarına göre en yüksek bulunmuştur. Braha ve ark., 2015 maviyemiş (V. *Corymbosum* L.) odun çeliklerine çeşitli konsantrasyonlarda (1500, 3000, 4500 mg/l) uygulamalar yapmışlar ve 3000 mg/l dozunda IBA uygulamasının NAA ile karşılaştırıldığında en yüksek köklenme yüzdesine sahip olduğunu belirlemişlerdir.



Şekil 2. Köklenmiş maviyemiş çeliklerinin görünüşü

Odun çeliklerinde en yüksek ortalama kök uzunluğu 3500 ppm IBA uygulamasında 27,51 mm, en düşük ortalama kök uzunluğu ise 3,63 mm ile kontrol grubunda saptanmış olup istatistiksel fark önemli bulunmuştur.

Odun çeliklerinde en yüksek canlı çelik sayısı da yine % 60 ile 3500 ppm dozundaki IBA uygulamasından, en düşük canlı kök sayısı ise % 17,5 ile kontrol grubundan elde edilmiştir. Martinussen ve ark. (2006) Maviyemiş gibi Vaccinium cinsi içinde yer alan kekreyemişin sert veya yumuşak odun çelikleri ile çoğaltılabileceğini saptamışlar ve 6000 ppm IBA ile % 85 başarı elde etmişlerdir.

Tablo 1. Maviyemiş odun çeliklerinde köklenme değerleri

UYGULAMA	Canlı Çelik Oranı (%)	Köklenme oranı %	Çelik başına ortalama Kök Sayısı	Çelik başına ortalama Kök Uzunluğu
Kontrol	17,5	10 b	0,30 c	3,63 c
2500 ppm	47,5	37,5 ab	1,47 ab	26,81 ab
3500 ppm	60	45 a	2,22 a	27,51 a
4500 ppm	25	15 b	0,65 bc	13,01 c
5500 ppm	35	17,5 ab	0,75 bc	13,83 bc

Aynı harfle gösterilen ortalamalar arasında fark yoktur (p<0.05).

SONUÇ

Anavatanı ABD olan maviyemiş ülkemizde Akdeniz ve Ege bölgesi ağırlıklı olmakla birlikte, Karadeniz bölgesindede yetiştirilmektedir. 1996 yılında ilk defa Türkiye'de başlayan maviyemiş yetiştiriciliği, Rize ilinin Güneysu ilçesinde yabani olarak yetişen ve maviyemiş ile aynı cins içinde yer alan çayüzümü (*Vaccinium arctostaphylos* L.) tiplerinin selekte edilerek kültüre alınması ile olmuştur. Sağlık açısında da oldukça faydalı bir meyve olan maviyemiş; kan şekerini düşürür, lifli yapısından dolayı bağırsak metabolizmasını düzenler ve kan kolesterolünü düşürür. Gece görüş kabiliyetini artıran maviyemiş, göz yorgunluğunu giderir, miyopluk ve şeker hastalığından kaynaklanan görme bozukluklarını gidermesi gibi insan sağlığına birçok fayda sağlamaktadır. Bu sebeplerden ötürü Karadeniz bölgesinde çaydan sonra en çok tercih edilen meyvelerden biri olan maviyemiş oldukça farklı alanlarda değerlendirilmektedir. Karadeniz bölgesindeki kapama bahçelerde Bluecrop maviyemiş çeşidi oldukça yaygın olarak yetiştirilmektedir. Bölge de bahçe tesisinde kullanılmak üzere fidan temininde sorunlar ortaya çıkmakta gerekli olan fidan temininde zorluklarla karşılaşılmaktadır. Bluecrop çeşidinin çelikle hızlı bir şekilde çoğaltılması ile ilgili yeterli çalışma olmaması nedeniyle bu çalışma Bluecrop çeşidinde kitlesel üretimde geliştirilecek tekniklere ışık tutacaktır. Rize ili Recep Tayyip Erdoğan Üniversitesi'nde gerçekleştirilen bu çalışmada dinlenme döneminde yetiştiricilik yapılan bahçelerden alınan yıllık odun çelikleri kullanılmıştır. Dinlenme döneminde alınan çelikler alt ısıtmasız perlit ortamına dikilmiştir.

Araştırma sonucunda IBA uygulamasının maviyemiş odun çeliklerinin köklenmesi üzerine etkisi olduğu saptanmıştır. Dinlenme döneminde alınan odun çeliklerinde en yüksek canlı çelik sayısı %60 ile 3500 ppm dozunda IBA uygulamasından, en düşük canlı çelik sayısı ise % 17,5 ile kontrol grubunda meydana geldiği saptanmıştır.

Köklenme oranı kontrole kıyasla en yüksek 3500 ppm dozundaki IBA uygulamasında % 45 olarak; en düşük köklenme oranı ise %10 ile kontrol grubunda odun çeliklerinden elde edilmiştir. Köklenme oranında belirli doza kadar artan IBA dozlarına paralel olarak genelde bir artış gözlenmiştir. 4500 ppm IBA uygulamasının odun çeliklerinin köklenmesi üzerine daha düşük oranda etki gösterdiği tespit edilmiştir. Odun çeliklerinde en yüksek ortalama kök sayısı 2.22 adet olarak 3500 ppm dozundaki IBA uygulamasında, en düşük ortama kök sayısı ise 0,30 adet ile kontrol grubundaki odun çeliklerinde saptanmış olup oluşan farklılık istatistiksel olarak önemli bulunmuştur. Odun çeliklerinin ortalama kök uzunlukları ise en yüksek 27,51 mm olarak 3500 ppm dozundaki IBA uygulamasından elde edilmiş olup bu uygulamanın diğer uygulamalara göre daha önerilebilir olduğu tespit edilmiştir.

Elde edilen sonuçlara göre Bluecrop maviyemiş çeşidinin çelikle çoğaltılmasında, dinlenme döneminde alınan odun çeliklerinin köklenme yüzdesi ve kök sayısı bakımından en yüksek değerlerin elde edildiği 3500 ppm dozundaki IBA uygulaması önerilebilir bulunmuştur.

KAYNAKLAR

Abolins M, Liepniece M & Gurtaja L 2003. Propagation of highbush blueberries by softwood cuttings in Latvia. Horticulture and Vegetable Growing, 22(2), 65-73.
Akbulut M, Bakoğlu N, Baykal H, Şavşatlı Y 2015. Maviyemişlerde (*Vaccinium corymbosum* L.) çelikle üretimde farklı hormon dozlarının köklenme üzerine etkisinin incelenmesi. Tarım Bilimleri Araştırma Dergisi 8(2):52- 56
Austin ME 1994. Rabbiteye Blueberries. Development, Production and Marketing. AGSCIENCE Inc., Florida, USA.160p.
Braha S, Rama P & Kukali E 2015. Determining Rooting Ability of Ennobled Blueberry Wood Pieces (*Vaccinium corymbosum* L.) with Presence of Growth Regulators IBA and NAA. Albanian Journal of Agricultural Sciences, 14(4), 355.
Brissette L, Tremblay L, Lord D 1990. Micropropagation of lowbush blueberry from mature fieldgrown plants. HortScience, 25(3): 349-351
Colombo RC, Carvalho DU, Cruz MA & Roberto SR 2018. Blueberry propagation by mini cuttings in response to substrate and indolebutryic acid application methods. Journal of Agricultural Sciences, 10(9), 450- 458.
Çelik H 2009b. Yield and berry characteristics of some northern highbush blueberries grown at different altitudes in Turkey. ISHS Acta Horticulturae, 838: 63-67.
Çelik H 2006. Kuzey orijinli yüksek çalı maviyemiş yumuşak odun çeliklerinde köklenme üzerine alttan ısıtma sıcaklığının etkisi. II. Ulusal Üzümsü Meyveler Sempozyumu, Gaziosman Paşa Üniversitesi Ziraat Fakültesi, Tokat.
Çelik H 2007. Northland kuzey orijinli yüksek çalı maviyemiş (*Vaccinium corymbosum* L.) yeşil çeliklerinin köklenmesi üzerine farklı ortamların etkisi. V. Ulusal Bahçe Bitkileri Kongresi, Atatürk Üniversitesi Ziraat Fakültesi, Erzurum.
Çelik H 2012a. Yüksek Boylu Maviyemiş (Highbush Blueberry) Yetiştiriciliği. Gifimey Mesleki Yayınlar Serisi-III, İstanbul.
Çelik H 2012b. Yüksek boylu maviyemiş çeşitlerinde köklenme üzerine çelik tipi, çelik alma zamanı ve köklenme ortamının etkisi. IV. Ulusal Üzümsü Meyveler Sempozyumu. Akdeniz Üniversitesi Ziraat Fakültesi, Antalya.
Çelik H 2016. Yüksek boylu maviyemiş (*Vaccinium corymbosum* L.) çeşitlerinden alınan yapraklı yumuşak odun mikro çeliklerde köklenme üzerine ortamların etkisi. Bahçe, 45(1), 1-6.
Çelik H 2016. Yüksek boylu maviyemiş (*Vaccinium corymbosum* L.) çeşitlerinden alınan yapraklı yumuşak odun mikro çeliklerde köklenme üzerine ortamların etkisi. Bahçe, 45(1), 1-6.
Çelik H 2017. Yüksek boylu maviyemiş çeşitlerinden alınan yapraklı yarı odunsu çeliklerde köklenme üzerine çelik alma zamanı ve IBA uygulamalarının etkisi. Bahçe, 46 (Özel Sayı 1), 63-72.
Çelik H 2005. Yabanmersini (Likapa) Yetiştiriciliği. Hasat Yayıncılık. 128 sayfa.
Çelik H & Ağaoğlu YS 2013. Maviyemiş. In Y.S. Ağaoğlu & R. Gerçekçioğlu (Eds.), Üzümsü Meyveler (pp. 245-377). Ankara, Türkiye: Tomurcukbağ Ltd. Şti., Eğitim Yayını.
Çelik H & Ateş S 2009. Maviyemiş (*Vaccinium corymbosum* L.) yumuşak odun çeliklerinde yaprak miktarı ve bazaldaki kesim yerinin köklenme üzerine etkileri. III. Ulusal Üzümsü Meyveler Sempozyumu, Sütçü İmam Üniversitesi, Ziraat Fakültesi, Kahramanmaraş.
Çelik H, 2009a. The performance of some northern highbush blueberry (*Vaccinium corymbosum* L.) varieties in North Eastern part of Anatolia. Anadolu J. Agric. Sci., 24(3):141-146.
Çelik H, Ateş S 2009c. Maviyemiş (*Vaccinium corymbosum* L.) yumuşak odun çeliklerinde yaprak miktarı ve bazaldaki kesim yerinin köklenme üzerine etkileri. III. Ulusal Üzümsü Meyveler Sempozyumu, 10-12 Haziran 2009, Kahramanmaraş 139-146.
FAO 2020. http://www.fao.org/faostat/en/#data/QC/visualize, Erişim Tarihi [19.04.2023]
Gough RE 1994. The Highbush Blueberry and Its Management. Food Product Pres. 272p.
Himelrick DG, Powell AA, Dozier WA 2002. Commercial Blueberry Production Guide for Alabama. Alabama A&M And Auburn Universities, Alabama Cooperative Extension System, ANR-904.
Karabulut B & Çelik H 2019. Maviyemişin çoğaltılması, Türkiye ve dünyada yapılan çalışmalar. Bahçe, 48 (Özel Sayı-1), 197- 206.

Krewer G & Cline B 2006. Blueberry propagation suggestions.
Marino SR, Williamson JG & Olmstead JW 2014. Vegetative growth of three southern highbush blueberry cultivars obtained from micropropagation and softwood cuttings in two Florida locations. HortScience, 49(5), 556-561.
Martinussen I, Nilsen H, Rothe G, Lund L & Rapp K 2006. Seasonal variations in rooting of Lingonberry (*Vaccinium vitisidea* L.) cuttings. *Acta Horticulturae*, 715, 439-442.
Orlikowska T 1986. Micropropagation of highbush blueberry. Fruit Science Reports. 13(3): 105-115
Pacholczak A & Nowakowska K 2015. The ex vitro rooting of blueberry (*Vaccinium corymbosum* L.) microcuttings. Folia Horticulturae, 27(2), 145-150.
Ruter JM 2015. Cloning Plants by Rooting Stem Cuttings. In A.C. Beyl & R.N. Trigiano (Edts.) Plant Propagation Concepts and Laboratory Exercises, (pp. 219-229), CRC Press.
Smolarz K, Chlebowska D 1998. Growth vigour and yielding of highbush blueberry cv. Bluecrop propagated from semi-woody cuttings and in-vitro. Hort. Abstr. 68(9):7544
Strik B, Fisher G, Hart J, Ingham R, Kaufman D, Penhallegon R, Pscheidt J, William R, Brun C, Ahmedullah M, Antonelli A, Askham L, Bristow P, Havens D, Scheer B, Shanks C and Barney D 1993. Highbush Blueberry Production Guide. Oregon State University. Department ofExtension and Experiment Station Station communication.
Turna İ, Kulaç Ş, Güney D & Seyis E 2013. Boylu Maviyemiş (*Vaccinium corymbosum* L.)'in Çelikle Üretilmesinde Hormon ve Ortamın Etkisi. Düzce Üniversitesi Orman FakültesiOrmancılık Dergisi,9(2),93-104. Retrieved from https://dergipark.org.tr/en/pub/duzceod/issue/4819/290874.
Wei QY 2011. Field performance of blueberry trees using Vaccinium arboreum as a rootstock. ASHS Annual Conference Paper. Waikoloa, Hawaii.
Wei QY, Basey A & Sturman P 2013. Effect of rootstock and soil amendment on threehighbush blueberry cultivars. ASHS Annual Conference Paper. Palm Desert, California.

DRAFT

ORAL PRESENTATION

Klimbazolyum tuzları ve onların paladyum komplekslerinin sentezi: Antimikrobiyal aktivite çalışmaları

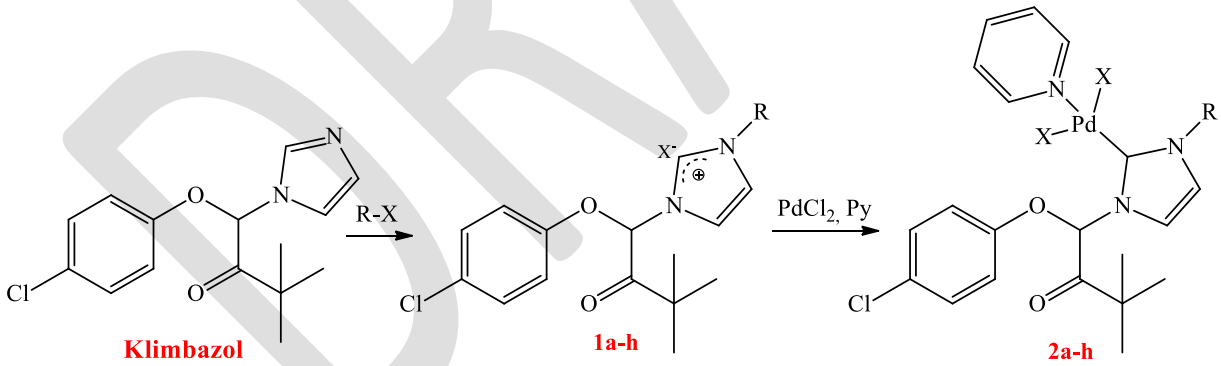
Simge Nazife YILMAZ[1]* (0009-0009-3134-6593), Hayati TÜRKMEN[1] (0000-0001-7411-2652)

[1]Ege Üniversitesi, Fen Fakültesi, Kimya Bölümü, İzmir, Türkiye

yzsimge@outlook.com

Özet

İmidazol, yapısında iki azot bulunduran, oda sıcaklığında genellikle beyaz kristal formda bulunan aromatik heterosiklik yapıda organik bir bileşiktir. Aynı zamanda, bilinen çeşitli farmakolojik etkilerinden kaynaklı ilaç kimyası ve tıp biliminin vazgeçilmez bileşiklerinden biridir. Klimbazol ise, antifungal imidazol türevidir. Kepek, sedef, egzama ve bazı fungal enfeksiyonların tedavisinde ayrıca benzer dezenfeksiyon işlemlerinde kullanılır. Kuvvetli çözünürlüğe, uygun maliyete ve düşük genotoksisiteye sahip olduğu için, ilaç sanayiide kullanımının yaygınlaşması ön görülebilir. Bu çalışmada, çeşitli alkil ve benzilhalojenürlerin klimbazol etkileştirilerek farklı yapıda klimbazolyum tuzları elde edilmiştir. Ayrıca tuzların PEPPSI tipi paladyum kompleri sentezleniştir. Sentezlenen tuzların ve komplekslerin yapı karekterizasyonları spektroskopik yöntemlerle aydınlatılmıştır. Tuzların ve paladyum kompleksleri farklı patojenik mikroorganizmalara karşı *in vitro* antimikrobiyal testleri yapılmıştır. Bu zamana dek yapılan çalışmalar, klimbazolün çözünürlüğü, ekotoksik ve genotoksik etkisi, hali hazırda kullanımı ve potansiyel kullanım alanları gibi konuları inceleme üzerine yapılmıştır. Çalışma sayesinde, klimbazolyum tuzları ve metal kompleksleri karakterize edilmiş, aynı zamanda fizyolojik ve farmakolojik etkileri incelenmiştir. Ayrıca çalışma, literatürdeki klimbazol türevleri eksikliğini gidermek ve literatüre yeni klimbazol türevleri kazandırmak, tıp ve ilaç kimyası alanında birtakım potansiyel yara iyileştirme ürünleri geliştirmek, aynı zamanda akademiye yeni ve geliştirilmesi gereken bir konu kazandırmayı amaçlamaktadır.



R: C_3H_7 (**a**), C_6H_{13}(**b**), $C_{10}H_{21}$(**c**), $CH_2C_6H_5$ (**d**), $CH_2C_6H_2Me_3$(**e**), $CH_2C_6Me_5$ (**f**), $CH_2C_6H_4Cl$ (**g**), $CH_2C_6H_3Cl_2$(**h**)

X: Cl, Br

Şekil 1. Reaksiyon Akış Şeması

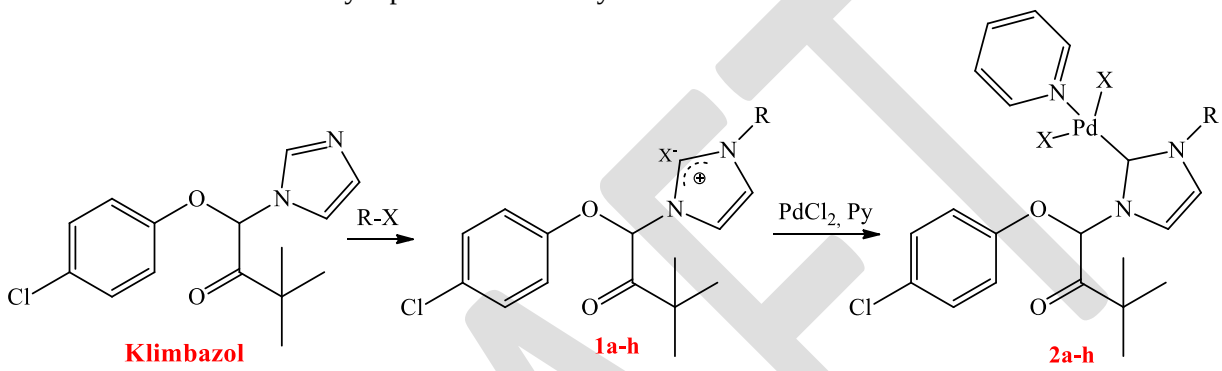
Anahtar Kelimeler: Klimbazol, Klimbazolyum tuzları, Paladyum kompleksleri, Antimikrobiyal Aktivite

Synthesis of climbazolium salts and their palladium complexes: Antimicrobial activity studies

Abstract

Imidazole is an organic compound with an aromatic heterocyclic structure containing two nitrogen compounds in its structure, which is usually found in white crystalline form at room temperature. At the same time, it is one of the indispensable compounds of pharmaceutical chemistry and medical science due to its various known

pharmacological effects. Climbazole, on the other hand, is a derivative of the antifungal imidazole. It is also used in the treatment of dandruff, psoriasis, eczema and some fungal infections, as well as in similar disinfection processes. Since it has strong solubility, affordable cost and low genotoxicity, its widespread use in the pharmaceutical industry can be predicted. In this study, climbazole salts of various alkyl and benzyl halides were obtained by activating climbazole in different structures. In addition, palladium complexes of the PEPPSI type of salts were synthesized. The structure characterizations of the synthesized salts and complexes have been elucidated by spectroscopic methods. Antimicrobial tests of salts and palladium complexes against different pathogenic microorganisms have been performed *in vitro*. Studies conducted so far have been conducted on the solubility of climbazole, its ecotoxic and genotoxic effects, its current use and potential uses. Thanks to the study, climbazolium salts and metal complexes were characterized, at the same time, their physiological and pharmacological effects were studied. In addition, the study aims to eliminate the lack of climbazole derivatives in the literature and introduce new climbazole derivatives into the literature, develop a number of potential wound healing products in the field of medicine and pharmaceutical chemistry, as well as introduce a new and necessary topic to the academy.



R: C_3H_7 (**a**), C_6H_{13}(**b**), $C_{10}H_{21}$(**c**), $CH_2C_6H_5$ (**d**), $CH_2C_6H_2Me_3$(**e**), $CH_2C_6Me_5$ (**f**), $CH_2C_6H_4Cl$ (**g**), $CH_2C_6H_3Cl_2$(**h**)

X: Cl, Br

Scheme 1. Reaction Flow Scheme

Keywords: Climbazole, Climbazolium salts, Palladium complexes, Antimicrobial activity

GİRİŞ

İyonik sıvılar, 100°C' nin altında sıvı olabilen tuzlardır (Tavanaie, 2013). İyonik sıvılar, tıpkı NaCl gibi anyon ve katyon çiftlerinden oluşsa da, buradaki iyonlar atomik değil asimetrik moleküler iyonlardır. Bu nedenle iyonlar daha hızlı bir araya gelip kristal ayrımı oluşturamazlar, bu nedenle daha düşük erime sıcaklıklarına sahiptirler (Aslanov, 2011). İyonik sıvıların en önemli özelliklerinden birinin ihmal edilebilir bir buhar basıncı seviyesine sahip olmaları ve bu nedenle atmosfere karışmalarının oldukça düşük olması ve yeşil kimya için bir çözüm olarak kabul edilmeleri olduğu söylenebilir (Postleb ve ark., 2013). Aynı zamanda iyonik sıvıların birçok kullanımı vardır. Son zamanlarda, iyi bilinen güçlü farmasötik etkileri nedeniyle tıpta kullanım alanlarından biri haline gelmiştir (Ferraz ve ark., 2015). İyonik sıvıların sitotoksik özellikleri ve antimikrobiyalleri, ilaç sentezindeki olası uygulamalar açısından tıp bilimcilerinden büyük ilgi görmüştür.
Klimbazol, egzama ve kepek gibi insan derisi enfeksiyonlarının tedavisinde ve dezenfeksiyon işlemlerinde kullanılan antifungal imidazolün bir türevidir. Kimyasal yapısı ve özellikleri incelendiğinde climbazol, ketokonazol ve mikonazol gibi diğer mantar öldürücülere benzer denebilir (Couteau ve ark., 2000). Diklorometan, aseton, kloroform, alkoller gibi çözücülerde oldukça yüksek çözünürlüğe sahip iken suda az çözünürlük gösterir. Alkollerde zincir uzunluğu arttıkça, klimbazolün hidrojen bağı yapmasından kaynaklı çözünürlük azalır fakat buna rağmen yine de iyi çözünürlük göstermektedir. Toksisitesine bakacak olursak, yapılan denemeler sonucu sucul canlılarda ve bitkilerde oldukça toksik olduğu gözlemlenmiştir (Richter ve ark., 2013). Ancak, fareler üzerind yapılan genoktoksisite çalışmalarında tam tersi sonuç saptanmıştır. 4 saatlik maruziyet sonucu farelerde mutasyon saptanmazken, 24 saatlik maruziyet sonucunda oldukça hafif bir artış göstermiştir (Pérez-Rivera ve ark., 2009).

DRAFT

MATERYAL VE METOT

Klimbazolyum Tuzları Sentezi Genel Prosedür:

1:1 oranda klimbazol, alkil-benzil halojenür önceden argonla doldurulmuş bir reaksiyon balonunda karıştırılır. İnce tabaka kromatografisi ile kontrol edilerek reaksiyon takip edilir ve gerektiğinde sonlandırılır. Ardından DCM/Eter difüzyonunda 1 gün boyunca bekletilir. Saf ürün filtre edilerek ayrılır.

Paladyum Kompleksleri Sentezi Genel Prosedür:

1:1 oranda **1a-h** ve $PdCl_2$,1:5 oranda K_2CO_3 ve 1-2 damla piridin 100°C sıcaklıkta 3 saat boyunca karıştırılır. Reaksiyon sonlandırıldıktan sonra vakumlu destilasyon ile piridin ortamdan uzaklaştırılır. Kalan turuncu katı, DCM ile çözülüp filtre edilir ve tuz ortamdan uzaklaştırılır. Kalan süzüntü, DCM ile yapılan kolon kromatografisi ile saflandırılır. Sarı kristal ürün elde edilir.

Tablo 1. Tuz ve Komplekslerin Karakter Analizi Sonuçları

Örnek	Verim (%)	Tuz piki (ppm)	Piridin piki (ppm)	C=O piki (cm^{-1})
1a	68	10.90	-	1728
1b	62	10.78	-	1759
1c	67	8.84	-	1728
1d	80	10.89	-	1721
1e	51	10.62	-	1726
1f	56	10.66	-	1725
1g	69	11.08	-	1728
1h	82	11.05	-	1728
2a	24	-	8.99	1728
2b	25	-	8.99	1729
2c	35	-	8.98	1731
2d	36	-	8.99	1729
2e	21	-	9.03	1732
2f	23	-	9.05	1730
2g	15	-	8.92	1729
2h	27	-	8.92	1724

SONUÇ

Elde ettiğimiz tuzların ^{1}H ve ^{13}C-NMR analizlerine baktığımız zaman, en asidik protonun tuz pikine ait olan proton olduğu görülmektedir. Tuzlarda, **1b** ve **1c**'den yola çıkılarak zincir uzunluğu arttıkça bileşiğin iyonik sıvı olma meyilinin arttığı yorumu yapılabilir. Ancak, benzilik gruplarda böyle bir şey söz konusu değildir. Hepsi beyaz katı madde olarak elde edilmiştir. Ayrıca, **1e** ve **1f**'deki verimin diğer tuzlara nazaran düşüklüğünün sebebi benzil gruplarındaki sübstitüe grubun artması ile maddenin daha kararsız hale gelmesi olarak değerlendirilebilir. Komplekslesmelerde ise, düşük verimde bileşik eldesi, klimbazolün rasemik yapısından kaynaklı yan ürün oluşma ihtimalinin olmasından kaynaklandığı düşünülmektedir.

TEŞEKKÜR

Çalışmam boyunca bana her zaman öncülük eden ve manevi desteklerini esirgemeyen sayın hocam Prof. Dr. Hayati TÜRKMEN'e, ihtiyacım olduğu her zaman bilgilerini benimle paylaşan ve yol gösteren değerli hocam Arş. Gör. Serdar Batıkan KAVUKÇU'ya ve laboratuvarda bulunduğum süreçte kendi deneyimlerini

ve bilgilerini benimle paylaşan ve yardımcı olan sevgili meslektaşlarım Aslıhan KARAER TUNÇAY, Sinem ÇAKIR ve Vedat Tolga ÖZDEMİR'e teşekkürlerimi sunarım.

KAYNAKLAR

Tavanaie M.A., 2013, Ionic Liquids as New Solvents for Textile Fiber Formation and Modification, Chem. Eng. Technol., 36: 1823–1837.

Aslanov L.A., 2011, Ionic liquids: Liquid structure, Journal of Molecular Liquids 162: 101–104

Giernoth R., Stefanik D., Postleb F., 2013, BIOnic Liquids: Imidazolium-based Ionic Liquids with Antimicrobial Activity, Universität zu Köln, 1-6

Branco L.C., Costa-Rodrigues J., Ferraz R., Fernandes M.H., Marrucho I.M., Noronha J.P., Rebelo L.P.N., Santos M.M., Prudencio C., Petrovski Z., 2015, Antitumor Activity of Ionic Liquids Based on Ampicillin, Chem Med Chem, 10: 1480 – 1483

Couteau C., Coiffard L.J.M., Jadaud M., Peigne F., 2000, Influence of pH on the photodegradation kinetics under UV light of climbazole solutions, Analusis, 28: 557-560

Coors A., Richter E., Ternes T.A., Wick A., 2013, Ecotoxicity of Climbazole, A Fungicide Contained in Antidandruff Shampoo, Environmental Toxicology and Chemistry, 12: 2816–2825

Aardema M.J., Hu T., Nash J.F., Pérez-Rivera A.A., 2009, Evaluation of the genotoxicity of the imidazole antifungal climbazole: Comparison to published results for other azole compounds, Mutation Research, 672: 27–39

DRAFT

ORAL PRESENTATION

Different techniques used for the recovery of pesticides in organic farming

Yasemin TORLAK[1*] (*ORCID: https://orcid.org/0000-0001-5964-2532*)

[*1]Pamukkale University, Cal Vocational High School, 20700, Çal, Denizli,Turkey

*Corresponding author e-mail: ytorlak@pau.edu.tr

Abstract

Pesticides have become very preferred in organic farming systems in order to make agricultural products more productive and with longer storage life. However, due to the increase in population and urbanization, pesticide use is increasing day by day. The main cause of pollution of the environment and crops stems from the excessive and uncontrolled use of pesticides. This situation disrupts both human health and ecosystem balance. Therefore, strict regulations have been developed and regulated for the pursuit of these compounds. Especially in the last decade, various techniques, from traditional detection techniques to advanced detection techniques, have been investigated for the detection, extraction and recovery of pesticides. Thanks to these techniques, time and economic savings can be achieved in the management of pesticide pollution. In this study, different techniques used in the analysis of pesticide residues were investigated by devices such as pretreatment with polymer-coated magnetic nanoparticles, gas chromatography combined with various detectors, and high-performance liquid chromatography. As a result of such studies, the measures to be taken in areas where pesticides are used heavily will be easily determined, and the control of pesticide pollution will become easier.

Keywords: Pesticides, organic farming, different techniques, pollution management

INTRODUCTION

Pesticides are very important chemical compounds in in farming, horticulture forestry, parks industrial vegetation control, sport fields, grass management ,educational facilities, etc due to many advantages (Marican and Durán-Lara 2018). These compounds have strong and rapid impacts for repulsion, prevention, mitigation, or destruction of weeds or pests. Thus, various active ingredients have been used as pesticides to protect agricultural products (Saleh, Zouari, and Al-Ghouti 2020) Depending on the target, pesticides can be classified as insecticides, herbicides, bactericides, fungicides, miticides and molluscicides, and some as nematicides, wood preservatives and rodenticides.

Depending on the chemical structure, pesticides are divided into the following classes: organophosphate group pesticides (compounds containing fenitrothion, chlorpyrifos, malathion, diclofenthion and diazinon), organochlorine pesticides (chlorothalonil, dichloro-diphenyltrichloroethane, lindane and endosulfan), carbamate pesticides (formethanate, carbetamide, carbofuran; chlorophenol). pesticides), chlorophenol pesticides (2-,3-,4-chlorophenols, 2,5-,2,6-dichlorophenols and 2,3,5-trichlorophenol; and synthetic pyrethroids).(Foo and Hameed 2010)(Marican and Durán-Lara 2018)(Aktar, Sengupta, and Chowdhury 2009). It has been observed that pesticide residues in agricultural products have increased with the increased use of these chemicals. These residues have been reported to cause various short- or long-term adverse health effects such as carcinogenicity in humans. (Wallace and Buha Djordjevic 2020), teratogenic (Sharma et al. 2020), hepatotoxic (Alarcan et al. 2020) and several other effects. For this reason, decreasing pesticide residues from extensively soil or consumed food materials is crucial. (Hassaan and El Nemr 2020) (Tang et al. 2021)

Pollutants enter the environment directly from disposal sites, landfills, spills during transportation, or as a result of accidents such as leaching from industrial facilities. Pesticides can reach plants in different ways, such as through soil, rain, wind and irrigation water. They then mix with soil, groundwater, surface waterways and surface runoff from wastewater treatment plants. Different techniques are being developed to separate pesticides from liquid streams and purify mixtures of substances.(Iorhemen, Hamza, and Tay 2016) (Jariyal et

al. 2015). And these techniques have been efficiently applied to different matrices. (Cengiz et al. 2021)(Marican and Durán-Lara 2018) Different techniques used for the recovery of pesticides as shown in Table 1.

Table 1. Recovery of pesticides, using different technique

Pesticide	Technique used in recovery	Matrix	Reference
tricyclic antidepressants (TCAs)	Extraction	in biological fluids	(Mofazzeli, Asaadi Shirvan, and Mohammadi 2018)
fungicides, insecticides and herbicide residues in hops	liquid chromatography–mass spectrometry/mass spectrometry (LC-MS/MS).	various adsorbent mixtures for dispersion solid-phase extraction	(Dušek, Jandovská, and Olšovská 2018)
Triazine (atrazine), chloroacetamide (alachlor), and phenoxy (2,4-dichlorophenoxyacetic acid)	a method for herbicide quantification	in environmental aqueous samples	(Suárez et al. 2018)
polycyclic aromatic hydrocarbons (PAHs)	high-performance liquid chromatography-tandem mass spectrometry (HPLC-MS/MS)	in environmental aqueous samples	(Feng et al. 2018)
organochlorine species	dielectric barrier discharge ionization (DBDI) as LC-MS ionization interface and electrospray ionization (ESI) method		(Lara-Ortega et al. 2018)
organophosphorus pesticides	zero valent Fe-reduced graphene oxide quantum dots (rGOQDs@Fe) as a new sorbent for magnetic dispersive solid- phase microextraction (MDSPME) coupled with gas chromatography–mass spectrometry method (GC-MS)	in water and fruit juice	(Akbarzade et al. 2018)
alachlor, atrazine, metolachlor, and simazine	dispersive pipette extraction and gas chromatography-mass spectrometry method (GC-MS)	in surface water	(Guan et al. 2018)
Cardiac troponin	an anti-passivation electrode (rGO/IL/Chi) by writing a graphene–	in environmental aqueous samples	(Zheng et al. 2018)

DRAFT

	ionic liquid–chitosan composite directly on a single-sided conductive gold strip		
phenoxy acid herbicides (fenoprop, mecoprop, dichlorprop, 2-(4-chlorophenopropionic acid (4-CPPA), 2-(3-chlorophenoxy) propionic acid (3-CPPA), and 2-phenoxypropionic acid (2-PPA))	reverse-phase/strong anionic exchange mixed-mode or strong anionic exchange retention mechanism	in river samples and effluents from wastewater treatment plants	(Valimaña-Traverso et al. 2018)
carcinogenic pesticides in milk	a cloud point methodology and spectrophotometry	in environmental aqueous samples (or milk)	(Mohd et al. 2018)
organophosphorus (OP) pesticides	Biochemical Assays	a plasmon resonance immunosensor for development of a broad-specific monoclonal antibody (mAb) for	(Jiao et al. 2018)
3- phenoxybenzoic acid (3-PBA) based on nanobody (Nb)-alkaline phosphatase (AP) fusion protein	a test method using direct competitive fluorescence enzyme immunoassay	3-phenoxybenzoic acid in urine	(Jiska Cohen-Mansfield, Maha Dakheel-Ali, MDb, Marcia S. Marx, PhDb, Khin Thein, MDb, and Natalie G. Regier and Waage et al. 2017)

Pesticides have disadvantages such as high levels of contaminants in water and soil and their natural breakdown. There are various treatment techniques discussed in detail in this review to remove pesticides from different matrices such as soil and water. This review article gives general information about the adsorption and recovery capacities of different adsorbent methods for the recovery of pesticides harmful to human health in matrices such as water and soil, and each technique is tried to be explained by taking into account the advantage and disadvantage of each technique. Due to these disadvantages, it has led to the development of different techniques for the recovery of pesticides from water and soil by biological, physical and chemical methods, as shown in Figure 1. (Marican and Durán-Lara 2018)

DRAFT

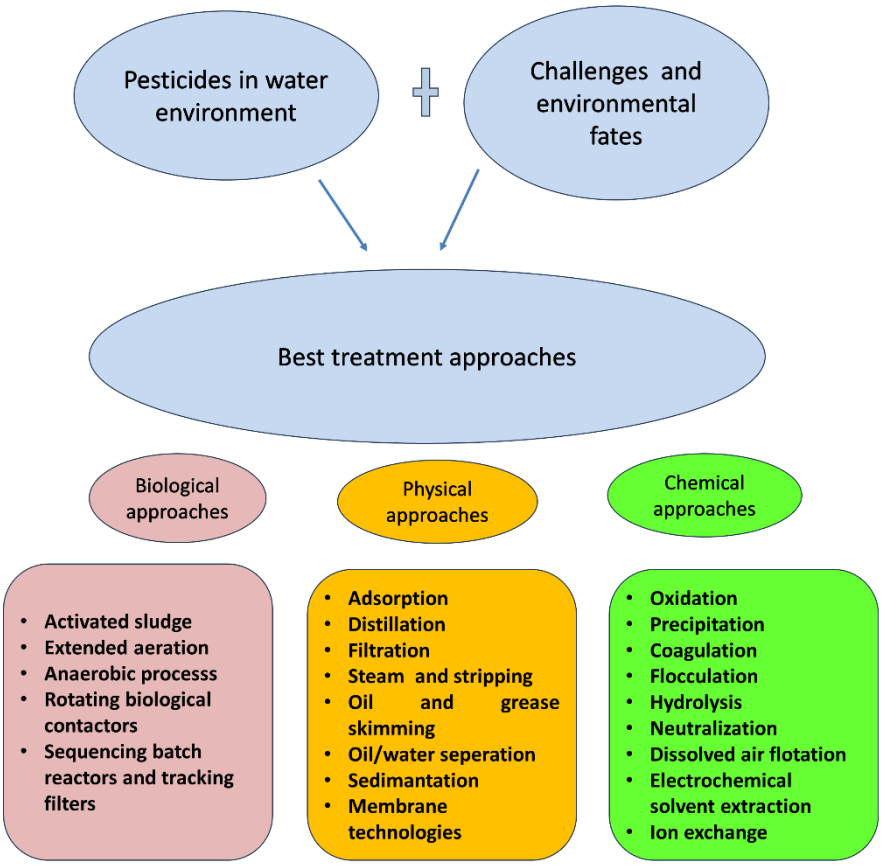


Figure 1. Best treatment approaches as chemical, physical, and biological wastewater treatment methods (Singh et al. 2018)(Saleh et al. 2020) (L et al. 2018a).

The aim of this review is to compare different analyses that can lead to future scientific research on the techniques and materials with the most promising and successful results for the removal of pesticides.

Biological processes of recovery

Biological recovery is an attractive technology achieved by completely converting organic compounds into products such as CO_2 and H_2O that may be less harmful to nature and human health. It is considered low-cost and environmentally friendly in removing pollutants compared to other methods. (Nwankwegu and Onwosi 2017). There are three methods in biological recovery with microorganisms. The first is improvement through enhanced natural attenuation, achieved by utilizing the natural capacities of the microorganisms present in the matrix. The second method is bioaugmentation, which is achieved by taking advantage of the natural capacities of the microorganisms present in the matrix. The other is biological processes achieved by adding non-native or genetically modified microorganisms and electroreceptors or nutrients. (Javaid, Ashiq, and Tahir 2016) There are two different techniques in biological recovery: aerobic and anaerobic recovery. Pesticides containing dichlor are digested through an aerobic process. This aerobic process involves oxidation and cleavage of the ether bond and hydroxylation of chlorophenol to form chloro-catechol. Additionally, when the aromatic ring in the compound is opened, water and carbon dioxide can be easily digested through bacterial metabolism.(Musbah et al. 2018) Via reductive dehalogenation, dichlorinated pesticides can also be digested anaerobically. When the ratio of chlorine atoms in the compound is less, it will decompose into CH_4 and CO_2.

Pesticides are difficult to biodigest. However, once the biodegradation system is established, it can be easily maintained. In addition, some pretreatments may be necessary to accelerate enzymatic reactions, degradation of some pesticides by photochemical degradation, or biological digestion reactions. (Huang et al. 2018). In another study, they stated that the degradation of norfloxacin, as an antibacterial agent, from wastewater

through the enzymatic reaction via chloroperoxidase was facilitated by the addition of H_2O_2. (Zhao et al. 2017). There is no need for a large land area in the treatment method using biologically enriched activated sludge. Lower cost is required compared to the advanced oxidation process. It is also more eco-friendly than chlorination. The disadvantage of this technique is that it requires a disposal area for the sludge and manual operation is required for this. (Luo et al. 2014) Degradation of pesticides by adding ligninolytic fungi that secrete ligninolytic enzymes to the targeted biomixture is one of the preferred techniques. Ligninolytic fungi added to this mixture depolymerize and mineralize lignin. (Castillo Mdel et al., 2008).

Hexachlorocyclohexane (HCH) is an organochlorine pesticide made from a mixture of more than eight stereoisomers with an environmental half-life of up to ten years. The HCH compound is difficult to remove from water and soil. In recent years, there has been research on developing a bacterial consortium that can digest HCH. Actively used bacterial consortia include strains of *Pseudomonas, Burkholderia, Flavobacterium, and Vibrio.*(Murthy and Manonmani 2007)

Physical processes of recovery

Physical recovery is the technique of correcting the problem by physical means. One of these techniques is membrane filtration techniques, which are widely used especially in wastewater. (Chiam and Sarbatly 2011)

Karimi et al. (2016) obtained a thin film composite poly (piperazine amide) nanofiltration membrane using the interfacial polymerization technique and triethylamine as an accelerator. They reported that nanofiltration membranes containing poly (piperazine amide) provided a significant improvement in the removal of pesticides.(Karimi, Rahimpour, and Shirzad Kebria 2016).

Qin et al. (2020) reported that ZrO_2 nanoparticles were efficiently produced by reverse micelle-mediated sol-gel process to prepare the nanofiltration membrane for the removal of carbofuran pesticides from wastewater. (Qin et al. 2020)

In another study, Musbah et al. (2018) reported that polypiperazine amide nanofiltration and OPMN-K membrane achieved successful results in the removal of different pesticides such as atrazine, simazine and diuron in the presence of humic acid. (Musbah et al. 2018).

Chemical processes of recovery

Recovery of pesticides through chemical techniques consists of various chemical reactions that help hydrolyse contaminants in water or soil. Some of the most commonly used chemical techniques are: Fenton treatment is an advanced oxidation process that also includes coagulation and ozonation. Many of these are used in conjunction with photocatalysis and membrane techniques.(Saleh et al. 2020)(Meghwal et al. 2019)

Vela et al. (2019) in their study with free radical have studied the removal of sodium persulfate ($Na_2S_2O_8$) from the matrix to which different pesticides such as pemetrozine, heksythiazox, spiromesifen, folpet, akrinatrininflonikamid, imidacloprid, acetamiprid, cymoxanil, tiacropride, spinosad, tebukazol, difokazol, difokazol, difokiliprid are bound.

Persulphate photolysis converts pesticides into less harmful substances by producing free radicals capable of oxidizing ($SO_4^{\bullet-}$). Because the pesticide causes mineralization into CO_2 and H_2O. With this study, it was observed that initially dissolved organic carbon was reduced by 87%. Therefore, it is thought that oxidation with sulphate radicals may be a promising method for the recovery of pesticides.(Vela et al. 2019)

One of the chemical techniques used is photochemical degradation. Ultraviolet (UV) light is used as electromagnetic radiation used for the photodegradation of pollutants in soil and wastewater. This UV light used includes UV-A (315–400 nm), UV-B (280–315 nm), UV-C (180–280 nm). nm), and vacuum UV light (10–180 nm). However, since UV light alone is generally not sufficient to degrade pollutants, it is also used in combination with different photochemical degradation techniques. (L et al. 2018b) (Rani and Sud 2015)

In their ozonation technique studies, Meijers et al. (1995) investigated the recovery rates of nearly 30 pesticides using this technique. As a result, it was concluded that different pesticides such as isoproturon, methoxuron, dimethoate, chlortoluron, diuron, and vinclozolin were removed. (Meijers et al. 1995)

Fenton reaction is the oxidation process of Fe^{2+} and H_2O_2 compounds with organic substrates. This oxidation process is one of the most effective chemical techniques in the recovery of pesticides. (Barbusiński 2009). With the Fenton reaction, a result was obtained in the recovery of an organophosphate insecticide known as fenitrothion at a rate of 98.5%, another organophosphorus pesticide such as orfenvinphos at a rate of 97.1%, and a rate of 90% of organochlorine pesticides from wastewater contaminated with pesticides (Barbusiński and Filipek). 2001).(Barbusiński and Filipek 2001) (Barbusiński 2009).

Saritha et al. (2007) showed advanced oxidation process as the best result in the recovery of 4-chloro-2-nitrophenol (4C-2-NP), which is known as a pollutant. It showed that a concentration much lower than the initial concentration was reached. (Saritha et al. 2007)

Adsorption technique is one of the processes that gives very effective results in the recovery of pollutants as a low-cost water purification method. The success of the adsorbent technique depends on the surface area of the contaminant, its porosity, and its interaction with the target substance. (Al-Ghouti and Da'ana 2020)

Ultrasound waves are waves created by obtaining high vibration using very high frequency sound waves. In the ultrasound technique, aviation bubbles are formed in the matrix to which they are attached, and these bubbles burst and particles are formed with micro turbulence. Then, these particles are disrupted and the chemical reaction is accelerated. These ultrasound waves used for the recovery of pesticides are the degradation of the diazinon compound in water. (Kadam et al. 2015) (Zhang et al. 2010)

CONCLUSION

This review article evaluates biological, physical, and chemical techniques for removing specific pesticides from water and soil, describing alternatives for pesticide recovery based on the unique characteristics of each technique. Each pesticide has different biological, physical and chemical properties. In other words, it supports the revision of appropriate techniques and materials for the removal of certain pesticides from water and soil. Choosing an inappropriate recovery technique may result in more toxic by-products. Therefore, comprehensive scientific analysis and laboratory studies are required to purify the pesticide.

The biological recovery technique of pesticides involves biological processes using fungal and bacterial species that enable the degradation of pesticides. Although this technique is widely used due to its low cost and high efficiency, it depends on many factors such as pH, temperature and oxygen level of water and soil, moisture content and nutrient availability. Within the physical remediation, there are good adsorption materials for pesticides with low cost of production that can be used effectively to treat wastewater containing pesticides. The common feature of pesticides is their poor water solubility. This feature turns them into complex compounds at the point of recovery. Therefore, the solubility of pesticides in water must be increased for subsequent removal.

As a result, choosing the best technique and material for the recovery of some pesticides is extremely important for subsequent research. amount of water and soil, investment cost, pH, matrix type, temperature, solubility of pesticides, etc. all variables such as are taken into account.

REFERENCES

Akbarzade, Samaneh, Mahmoud Chamsaz, Gholam Hossein Rounaghi, and Mahdi Ghorbani. 2018. "Zero Valent Fe-Reduced Graphene Oxide Quantum Dots as a Novel Magnetic Dispersive Solid Phase Microextraction Sorbent for Extraction of Organophosphorus Pesticides in Real Water and Fruit Juice Samples Prior to Analysis by Gas Chromatography-Mass Spectrom." *Analytical and Bioanalytical Chemistry* 410(2):429–39. doi: 10.1007/s00216-017-0732-9.

Aktar, Wasim, Dwaipayan Sengupta, and Ashim Chowdhury. 2009. "Impact of Pesticides Use in Agriculture: Their Benefits and Hazards." *Interdisciplinary Toxicology* 2(1):1–12. doi: 10.2478/v10102-009-0001-7.

Al-Ghouti, Mohammad A., and Dana A. Da'ana. 2020. "Guidelines for the Use and Interpretation of Adsorption Isotherm Models: A Review." *Journal of Hazardous Materials* 393(November 2019):122383. doi: 10.1016/j.jhazmat.2020.122383.

Alarcan, Jimmy, Julia Waizenegger, Marize de Lourdes Marzo Solano, Dajana Lichtenstein, Claudia Luckert, Ad Peijnenburg, Geert Stoopen, Raju Prasad Sharma, Vikas Kumar, Philip Marx-Stoelting, Alfonso Lampen, and Albert Braeuning. 2020. "Hepatotoxicity of the Pesticides Imazalil, Thiacloprid and Clothianidin – Individual and Mixture Effects in a 28-Day Study in Female Wistar Rats." *Food and Chemical Toxicology* 140(December 2019):111306. doi: 10.1016/j.fct.2020.111306.

Barbusiński, K., and K. Filipek. 2001. "Use of Fenton's Reagent for Removal of Pesticides from Industrial Wastewater." *Polish Journal of Environmental Studies* 10(4):207–12.

Barbusiński, Krzysztof. 2009. "Fenton Reaction - Controversy Concerning the Chemistry." *Ecological Chemistry and Engineering S* 16(3):347–58.

Cengiz, Mehmet Fatih, Onur Basançelebi, Mehmet Başlar, and Muharrem Certel. 2021. "A Novel Technique for the Reduction of Pesticide Residues by a Combination of Low-Intensity Electrical Current and Ultrasound Applications: A Study on Lettuce Samples." *Food Chemistry* 354(February):129360. doi: 10.1016/j.foodchem.2021.129360.

Chiam, Chel-ken, and Rosalam Sarbatly. 2011. "Purification of Aquacultural Water: Conventional and New Membrane-Based Techniques." *Separation & Purification Reviews* 40(2):126–60. doi: 10.1080/15422119.2010.549766.

Dušek, Martin, Vladimíra Jandovská, and Jana Olšovská. 2018. "Analysis of Multiresidue Pesticides in Dried Hops by LC-MS/MS Using QuEChERS Extraction Together with DSPE Clean-Up." *Journal of the Institute of Brewing* 124(3):222–29. doi: 10.1002/jib.490.

Feng, Jiaqing, Min Sun, Xiuqin Wang, Yu Tian, Chuannan Luo, and Juanjuan Feng. 2018. "Barium Sulfate Nanoparticles as a Coating for Solid-Phase Microextraction of Polycyclic Aromatic Hydrocarbons in Aqueous Samples." *Chromatographia* 81(9):1287–92. doi: 10.1007/s10337-018-3568-3.

Foo, K. Y., and B. H. Hameed. 2010. "Detoxification of Pesticide Waste via Activated Carbon Adsorption Process." *Journal of Hazardous Materials* 175(1–3):1–11. doi: 10.1016/j.jhazmat.2009.10.014.

Guan, Sharon Hongxia, Michelle Wenlin Huang, Xiaoping Li, and Qingsong Cai. 2018. "Determination of Atrazine, Simazine, Alachlor, and Metolachlor in Surface Water Using Dispersive Pipette Extraction and Gas Chromatography–Mass Spectrometry." *Analytical Letters* 51(4):613–25. doi: 10.1080/00032719.2017.1341904.

Hassaan, Mohamed A., and Ahmed El Nemr. 2020. "Pesticides Pollution: Classifications, Human Health Impact, Extraction and Treatment Techniques." *The Egyptian Journal of Aquatic Research* 46(3):207–20. doi: 10.1016/j.ejar.2020.08.007.

Huang, Yichen, Lijuan Xiao, Feiyu Li, Mengshi Xiao, Derong Lin, Xiaomei Long, and Zhijun Wu. 2018. "Microbial Degradation of Pesticide Residues and an Emphasis on the Degradation of Cypermethrin and 3-Phenoxy Benzoic Acid: A Review." *Molecules* 23(9):2313. doi: 10.3390/molecules23092313.

Iorhemen, Oliver, Rania Hamza, and Joo Tay. 2016. "Membrane Bioreactor (MBR) Technology for Wastewater Treatment and Reclamation: Membrane Fouling." *Membranes* 6(2):33. doi: 10.3390/membranes6020033.

Jariyal, Monu, V. K. Gupta, Vikas Jindal, and Kousik Mandal. 2015. "Isolation and Evaluation of Potent Pseudomonas Species for Bioremediation of Phorate in Amended Soil." *Ecotoxicology and Environmental Safety* 122:24–30. doi: 10.1016/j.ecoenv.2015.07.007.

Javaid, Muhammad Kashif, Mehrban Ashiq, and Muhammad Tahir. 2016. "Potential of Biological Agents in Decontamination of Agricultural Soil." *Scientifica* 2016(Figure 1):1–9. doi: 10.1155/2016/1598325.

Jiao, Shasha, Pengyan Liu, Ying Liu, Rubing Zou, Ying Zhao, Yihua Liu, Guonian Zhu, and Yirong Guo. 2018. "Binding Properties of Broad-Specific Monoclonal Antibodies against Three Organophosphorus Pesticides by a Direct Surface Plasmon Resonance Immunosensor." *Analytical and Bioanalytical Chemistry* 410(28):7263–73. doi: 10.1007/s00216-018-1337-7.

Jiska Cohen-Mansfield, Maha Dakheel-Ali, MDb, Marcia S. Marx, PhDb, Khin Thein, MDb, and Natalie G. Regier, PhDb, and Waage et al. 2017. "乳鼠心肌提取 HHS Public Access." *Physiology & Behavior* 176(1):139–48. doi: 10.1021/acs.jafc.8b04521.Development.

Kadam, Shekhar U., Brijesh K. Tiwari, Carlos Álvarez, and Colm P. O'Donnell. 2015. "Ultrasound Applications for the Extraction, Identification and Delivery of Food Proteins and Bioactive Peptides." *Trends in Food Science & Technology* 46(1):60–67. doi: 10.1016/j.tifs.2015.07.012.

Karimi, Hayde, Ahmad Rahimpour, and Mohammad Reza Shirzad Kebria. 2016. "Pesticides Removal from

Water Using Modified Piperazine-Based Nanofiltration (NF) Membranes." *Desalination and Water Treatment* 57(52):24844–54. doi: 10.1080/19443994.2016.1156580.

L, Goodwin, Carra I, Campo P, and Soares A. 2018a. "Treatment Options for Reclaiming Wastewater Produced by the Pesticide Industry." *International Journal of Water and Wastewater Treatment* 4(1). doi: 10.16966/2381-5299.149.

L, Goodwin, Carra I, Campo P, and Soares A. 2018b. "Treatment Options for Reclaiming Wastewater Produced by the Pesticide Industry." *International Journal of Water and Wastewater Treatment* 4(1):1–15. doi: 10.16966/2381-5299.149.

Lara-Ortega, Felipe J., José Robles-Molina, Sebastian Brandt, Alexander Schütz, Bienvenida Gilbert-López, Antonio Molina-Díaz, Juan F. García-Reyes, and Joachim Franzke. 2018. "Use of Dielectric Barrier Discharge Ionization to Minimize Matrix Effects and Expand Coverage in Pesticide Residue Analysis by Liquid Chromatography-Mass Spectrometry." *Analytica Chimica Acta* 1020:76–85. doi: 10.1016/j.aca.2018.02.077.

Luo, Yunlong, Wenshan Guo, Huu Hao Ngo, Long Duc Nghiem, Faisal Ibney Hai, Jian Zhang, Shuang Liang, and Xiaochang C. Wang. 2014. "A Review on the Occurrence of Micropollutants in the Aquatic Environment and Their Fate and Removal during Wastewater Treatment." *Science of The Total Environment* 473–474(January):619–41. doi: 10.1016/j.scitotenv.2013.12.065.

Marican, Adolfo, and Esteban F. Durán-Lara. 2018. "A Review on Pesticide Removal through Different Processes." *Environmental Science and Pollution Research* 25(3):2051–64. doi: 10.1007/s11356-017-0796-2.

Meghwal, Kiran, Reema Agrawal, Srishti Kumawat, Nirmala Kumari Jangid, and Chetna Ameta. 2019. *Chemical and Biological Treatment of Dyes*.

Meijers, R. T., E. Oderwald-Muller, P. A. N. M. Nuhn, and J. C. Kruithof. 1995. "Degradation of Pesticides by Ozonation and Advanced Oxidation." *Ozone: Science & Engineering* 17(6):673–86. doi: 10.1080/01919512.1995.10555778.

Mofazzeli, Farideh, Homeira Asaadi Shirvan, and Fatemeh Mohammadi. 2018. "Extraction and Determination of Tricyclic Antidepressants in Real Samples Using Air-dispersed Liquid–Liquid Microextraction Prior to Gas Chromatography and Flame Ionization Detection." *Journal of Separation Science* 41(23):4340–47. doi: 10.1002/jssc.201800359.

Mohd, N. I., N. N. M. Zain, M. Raoov, and S. Mohamad. 2018. "Determination of Carcinogenic Herbicides in Milk Samples Using Green Non-Ionic Silicone Surfactant of Cloud Point Extraction and Spectrophotometry." *Royal Society Open Science* 5(4):171500. doi: 10.1098/rsos.171500.

Murthy, H. M. Rajashekara, and H. K. Manonmani. 2007. "Aerobic Degradation of Technical Hexachlorocyclohexane by a Defined Microbial Consortium." *Journal of Hazardous Materials* 149(1):18–25. doi: 10.1016/j.jhazmat.2007.03.053.

Musbah, Ibrahim, Delphine Cicéron, Abdellah Saboni, and Silvia Alexandrova. 2018. "Removal of Pesticides and Desethylatrazine (DEA) by Nanofiltration: Effects of Organic and Inorganic Solutes on Solute Rejection." *Journal of Chemical Technology and Metallurgy* 53(4):657–63.

Nwankwegu, Amechi S., and Chukwudi O. Onwosi. 2017. "Bioremediation of Gasoline Contaminated Agricultural Soil by Bioaugmentation." *Environmental Technology & Innovation* 7:1–11. doi: 10.1016/j.eti.2016.11.003.

Qin, Hang, Wenming Guo, Xin Huang, Pengzhao Gao, and Hanning Xiao. 2020. "Preparation of Yttria-Stabilized ZrO2 Nanofiltration Membrane by Reverse Micelles-Mediated Sol-Gel Process and Its Application in Pesticide Wastewater Treatment." *Journal of the European Ceramic Society* 40(1):145–54. doi: 10.1016/j.jeurceramsoc.2019.09.023.

Rani, Sunita, and Dhiraj Sud. 2015. "Role of Enhanced Solar Radiation for Degradation of Triazophos Pesticide in Soil Matrix." *Solar Energy* 120:494–504. doi: 10.1016/j.solener.2015.07.050.

Saleh, Iman A., Nabil Zouari, and Mohammad A. Al-Ghouti. 2020. "Removal of Pesticides from Water and Wastewater: Chemical, Physical and Biological Treatment Approaches." *Environmental Technology & Innovation* 19:101026. doi: 10.1016/j.eti.2020.101026.

Saritha, P., C. Aparna, V. Himabindu, and Y. Anjaneyulu. 2007. "Comparison of Various Advanced Oxidation Processes for the Degradation of 4-Chloro-2 Nitrophenol." *Journal of Hazardous Materials* 149(3):609–14. doi: 10.1016/j.jhazmat.2007.06.111.

Sharma, Rajnesh Kumar, Priyanka Singh, Aarzoo Setia, and Aman Kumar Sharma. 2020. "Insecticides and Ovarian Functions." *Environmental and Molecular Mutagenesis* 61(3):369–92. doi: 10.1002/em.22355.

Singh, N. B., Garima Nagpal, Sonal Agrawal, and Rachna. 2018. "Water Purification by Using Adsorbents: A

Review." *Environmental Technology & Innovation* 11:187–240. doi: 10.1016/j.eti.2018.05.006.

Suárez, Ruth, Sabrina Clavijo, Alba González, and Víctor Cerdà. 2018. "Determination of Herbicides in Environmental Water Samples by Simultaneous In-Syringe Magnetic Stirring-Assisted Dispersive Liquid-Liquid Microextraction and Silylation Followed by GC-MS." *Journal of Separation Science* 41(5):1096–1103. doi: 10.1002/jssc.201700875.

Tang, Fiona H. M., Manfred Lenzen, Alexander McBratney, and Federico Maggi. 2021. "Risk of Pesticide Pollution at the Global Scale." *Nature Geoscience* 14(4):206–10. doi: 10.1038/s41561-021-00712-5.

Valimaña-Traverso, Jesús, Sonia Morante-Zarcero, Damián Pérez-Quintanilla, María Ángeles García, Isabel Sierra, and María Luisa Marina. 2018. "Cationic Amine-Bridged Periodic Mesoporous Organosilica Materials for off-Line Solid-Phase Extraction of Phenoxy Acid Herbicides from Water Samples Prior to Their Simultaneous Enantiomeric Determination by Capillary Electrophoresis." *Journal of Chromatography A* 1566:146–57. doi: 10.1016/j.chroma.2018.06.042.

Vela, Nuria, José Fenoll, Isabel Garrido, Gabriel Pérez-Lucas, Pilar Flores, Pilar Hellín, and Simón Navarro. 2019. "Reclamation of Agro-Wastewater Polluted with Pesticide Residues Using Sunlight Activated Persulfate for Agricultural Reuse." *Science of The Total Environment* 660:923–30. doi: 10.1016/j.scitotenv.2019.01.060.

Wallace, David R., and Aleksandra Buha Djordjevic. 2020. "Heavy Metal and Pesticide Exposure: A Mixture of Potential Toxicity and Carcinogenicity." *Current Opinion in Toxicology* 19:72–79. doi: 10.1016/j.cotox.2020.01.001.

Zhang, Yuanyuan, Wenjia Zhang, Xiaojun Liao, Jianing Zhang, Yaxi Hou, Zhiyong Xiao, Fang Chen, and Xiaosong Hu. 2010. "Degradation of Diazinon in Apple Juice by Ultrasonic Treatment." *Ultrasonics Sonochemistry* 17(4):662–68. doi: 10.1016/j.ultsonch.2009.11.007.

Zhao, Ruinan, Xiaohong Li, Mancheng Hu, Shuni Li, Quanguo Zhai, and Yucheng Jiang. 2017. "Efficient Enzymatic Degradation Used as Pre-Stage Treatment for Norfloxacin Removal by Activated Sludge." *Bioprocess and Biosystems Engineering* 40(8):1261–70. doi: 10.1007/s00449-017-1786-y.

Zheng, Qi-qi, Yuan-chao Lu, Zun-zhong Ye, Jian-feng Ping, Jian Wu, and Yi-bin Ying. 2018. "An Anti-Passivation Ink for the Preparation of Electrodes for Use in Electrochemical Immunoassays." *Journal of Zhejiang University-SCIENCE B* 19(9):726–34. doi: 10.1631/jzus.B1700510.

DRAFT

ORAL PRESENTATION

Effect of progesterone on the physiological and molecular responses of tomato seedlings under drought and salt stress

Ufuk Çelikkol Akçay (*ORCID: https://orcid.org/0000-0003-1260-3813*)

Isparta University of Applied Sciences, Faculty of Agriculture, Department of Agricultural Biotechnology, Isparta, TURKEY

Corresponding author e-mail: ufukakcay@isparta.edu.tr

Abstract

Drought and salinity stresses limit agricultural production worldwide. Stress mitigation by external biostimulator molecules has been an active research topic in recent years. In this study, the effect of the steroidal hormone progesterone, present in both animal and plant tissues, was investigated in terms of morphological (tissue length and weights), physiological (relative water content, ion leakage, MDA and proline levels) and enzymatic (ACS2, P5CS, FeSOD, CAT2, GR1 and APX1) stress indicators under both drought and salinity stresses. Both stresses were alleviated to some extent by progesterone treatment in tomato seedlings.

Keywords: *Solanum lycopersicum* L., abiotic stress, progesterone

INTRODUCTION

Drought and salinity are the most common abiotic stresses restricting current agricultural production worldwide. Both stresses are associated with low osmotic potential, which results in stomatal closure to prevent further water loss at the expense of reduced photosynthetic efficiency. Both stresses usually result in reduced growth and negatively affect physiological processes by triggering ROS production (Gupta et al., 2017). Salinity stress exerts further destructive effects by causing ion toxicity and nutritional imbalances (Haj-Amor et al., 2022). Various molecules that regulate growth and development in plants are being investigated for their ameliorative potential under various stresses (Zulfiqar and Ashraf, 2020). One of the molecules is progesterone, a well-known animal steroidal hormone essential for ovulation, luteinization and the continuation of pregnancy in mammals.

Progesterone was detected in plants in the 1960s and its presence has been demonstrated in a wide variety of plant species and in various plant tissues (Lino et al., 2007). Similar to animals, its roles in flowering and generative development have been characterized (Janeczko and Filek, 2002; Janeczko et al., 2003). Lino et al. (2007) proposed that progesterone acts as a growth hormone in plant tissues and that its role and function is different from those of brassinosteroids, which are another group of plant steroidal hormones.

Progesterone was also shown to decrease necrosis and ion leakage and improve photosystem II efficiency in *Arabidopsis* under a biotic stress, *Pseudomonas syringae* infection (Janeczko et al., 2013a). Progesterone was also shown to play a role in various abiotic stresses. Erdal (2012a) showed that progesterone protects germinating maize seeds under salt stress. The molecule also protected wheat and chickpea seedlings from salt stress by both enzymatic and non-enzymatic antioxidative defense mechanisms and by increasing the anabolism of carbon compounds (Erdal, 2012b; Erdal and Dumlupınar, 2011). Progesterone mitigates drought, heat and light stress in wheat by enhancing antioxidant defense and photosystem II activity (Janeczko et al., 2013b; Su et al., 2014; Xue et al., 2017). Progesterone was also shown to protect chickpea and maize seedlings from chilling damage and to enter the lipid bilayer and similar to other sterols, it plays a role in membrane fluidity and frost tolerance in winter wheat (Genisel et al., 2013; Erdal and Genisel, 2016; Filek et al., 2017; Janeczko et al., 2019).

Tomato (*Solanum lycopersicum* L.) is a model plant for the study of fruits. It is also an economically important crop exhibiting many health benefits due to its high vitamin and lycopene content. In this study, the effects of progesterone were investigated through observation of common physiological stress indicators such as growth

parameters, membrane damage status and osmoprotectant proline production as well as transcript levels of the enzymatic antioxidative defense system including superoxide dismutase (FeSOD), catalase (CAT2), glutathione reductase (GR1), ascorbate peroxidase (APX1), 1-aminocyclopropane-1-carboxylic acid synthase (ACS2) and delta 1-pyrroline-5-carboxylate synthase (P5CS).

MATERIALS AND METHODS

Plant growth and treatment

Tomato seeds (Kayra F1 tomato variety from Anamas Tohum Ltd. Şti., Antalya) were placed in polypropylene containers with a volume of 400 ml containing sterile perlite as one seed in each container. Germination was achieved in all pots at the end of a week by irrigation with sterile Hoagland solution (Hoagland and Arnon, 1950) every other day. Plant growth was continued for 21 days in a 16-hour light/8-hour dark cycle, at 24°C in a plant growth cabinet containing 50% humidity. Drought stress started at the end of the 21st day, 0 water potential was reached on the 3rd day, and at the end of the 5th day, the tissue samples were collected for analysis. Salt stress was initiated with Hoagland solution containing 200 mM NaCl at the end of the 21st day and applied for 5 days as in drought stress.

Progesterone was applied by adding 1 µM progesterone (Progestan, Koçak Farma, İstanbul) into Hoagland solution starting from seed sowing and the application continued throughout stress applications. Progesterone concentration was determined by pre-testing with 10 plants that received treatments at three different concentrations for each hormone. Root and stem lengths of the plants and ion leakage measurements were evaluated and the concentration with the best anatomical and physiological response was used throughout the study. Salt stress was applied at a concentration of 200 mM NaCl based on the observation that it induced a severe stress response for the same tomato variety within a week in previous laboratory studies.

Determination of physiological plant stress indicators

In the presence or absence of progesterone application and stress, after 21 days of development and 5 days of stress application, all plants were removed from perlite and washed under tap water, and root/stem lengths were determined as cm. Root and stem tissues were separated and weighed, and after drying at 60 °C for 48 h, they were weighed again and their dry weight (g) was determined.

Relative water contents (BSU) were determined according to the formula; BSU (%) = (Wet weight-Dry weight)/(Turgid weight-Dry weight) X 100, as was specified in Smart and Bingham (1974). Turgid weight was determined by keeping the leaves in distilled water at room temperature for 24 h.

The levels of osmoprotectant amino acid proline were determined by the method of Bates et al. (1973) without any modifications.

Cellular membrane damage was determined by measuring the phospholipid peroxidation product malondialdehyde (MDA) level according to Ohkawa et al. (1979) without any modifications.

Another indicator of cell wall and membrane damage, ion leakage levels, were determined according to Nanjo et al. (1999) without any modifications.

Determination of gene expression

Total RNA isolation was performed according to the instructions of Thermo GeneJET plant mini kit (Thermo, USA), followed by treatment with Thermo RNase-free DNase I (Thermo, USA) in accordance with the instructions for quantitative RT-PCR. The amounts of total RNA obtained were determined spectrophotometrically using Nanodrop 2000. The quality of total RNAs was also determined by separation and imaging with 1% agarose gel electrophoresis followed by cDNA synthesis, which was performed according to Thermo RevertAid FirstStrand cDNA synthesis kit instructions (Thermo, USA).

In this study, *ACS2, P5CS, FeSOD, CAT2, GR1*, and APX1 gene expressions in tomato leaves were normalized using the EF-1 reference gene, and the relative quantification of genes was compared with the control group using a Biorad CFX Connect Real-Time PCR device and CFX Maestro software (Biorad, USA). All amplification reactions were performed using Biorad iTaq Universal SYBR Green Supermix as specified in the user manual. The primers used in the study were optimized with PrimerPremier 5.0 software (Premier

Biosoft International, USA) to provide optimum amplification conditions. PCR conditions included initial denaturation of 30 s at 95°C, followed by amplification for 5 seconds at 95°C and 30 seconds at 54°C, repeating 40 cycles. RT-PCR reactions included two biological and two technical replicates. Relative expression of genes was determined according to the $2^{-\Delta\Delta C_T}$ method. $\Delta\Delta C_T$ values were calculated by subtracting the mean ΔC_T values of the samples from the mean ΔC_T values of the controls, and these values were used to determine the $2^{-\Delta\Delta C_T}$ differences.

Statistical Analysis

The study was conducted in triplicate and each replicate included 20 plants. The data obtained in the research were evaluated using the SPSS 16.0 program. The differences between the means were determined by the one-way ANOVA and Tukey's test.

RESULTS

Neither stress nor progesterone treatment had no significant effect on shoot and root lengths. Drought stress and sole progesterone application slightly reduced shoot dry weights, whereas salinity stress significantly reduced both shoot and root dry weights. Although not significant, progesterone treatment increased shoot dry weights under drought and both shoot and root dry weights under salinity stress. Both stress slightly reduced the water content, and progesterone did not show any significant effect.

Table 1. Effect of different stress and hormone treatments on plant morphological parameters. C, T, K, P, P+T and P+K indicate control, salt, drought, progesterone, progesterone+salt and progesterone+drought applications, respectively.

Treatment	Tissue Length (cm)	Dry Weight (gr)	Water Content (%)
		SHOOT	
C	27,90 ± 0,56 a	0,29 ± 0,047 a	93,37 ± 0,38 a
T	20,57 ± 0,32 b	0,15 ± 0,021 b	92,14 ± 0,61 a, c
K	21,5 ± 0,98 b	0,23 ± 0,012 a, b	90,75 ± 0,55 b, c
P	27,30 ± 0,60 a	0,22 ± 0,032 a, b	93,50 ± 1,11 a
P+T	20,43 ± 0,38 b	0,15 ± 0,021 b	91,95 ± 0,91 a, c
P+K	21,80 ± 0,35 b	0,29 ± 0,045 a	89,14 ± 0,34 b
		ROOT	
C	24,73 ± 2,57 a, b	0,041 ± 0,007 a, c	93,43 ± 0,91 a, b
T	23,17 ± 0,12 a	0,022 ± 0,002 b	92,34 ± 0,34 a, b
K	25,47 ± 1,06 a, b	0,042 ± 0,008 a, c	91,87 ± 0,87 b
P	26,10 ± 1 a, b	0,027 ± 0,007 b, c	94,13 ± 1,01 a
P+T	26,57 ± 0,38 a, b	0,027 ± 0,004 b, c	92,39 ± 0,50 a, b
P+K	23,47 ± 0,32 a, b	0,054 ± 0,004 a	91,89 ± 0,90 b

Relative water content also decreased upon stress application and no significant effect of progesterone was observed on the parameter. Stress applications did not change ion leakages significantly; however, the value was significantly lower than every other treatment, including control, upon progesterone treatment under salt stress. Unlike ion leakage, both stress increased MDA contents, whereas progesterone treatment reduced the level back to the control level under salinity stress. The proline content was the most reactive parameter to both drought and salt stresses, with 7- and 20-fold increases, respectively. The presence of progesterone resulted in 9% and 38% decrease in proline content under salinity and drought stresses, respectively.

Table 2. Effect of different stress and hormone treatments on physiological stress indicators. C, T, K, P, P+T and P+K indicate control, salt, drought, progesterone, progesterone+salt and progesterone+drought applications, respectively.

Treatment	Relative Water Content (%)	Ion Leakage (%)	MDA Content (nmol/g)	Proline Content (nmol/g)
C	85,48 ± 0,40 a	5,02 ± 0,26 a	5,54 ± 0,72 a	40,54983 ± 16,07 a
T	69,96 ± 6,42 b	4,97 ± 0,72 a	7,38 ± 0,41 a, b	810,4811 ± 146,39 b
K	78,91 ± 3,92 a, b	5,07 ± 0,51 a	9,98 ± 0,94 b, c	291,5808 ± 33,35 c
P	81,74 ± 3,31 a	6,20 ± 0,32 a	4,01 ± 0,88 a	41,06529 ± 5,75 a
P+T	70,71 ± 2,86 b	3,30 ± 0,37 b	5,38 ± 0,32 a	737,8007 ± 62,46 b
P+K	77,09 ± 1,87 a, b	5,56 ± 0,85 a	13,14 ± 2,59 c	179,2096 ± 24,97 a, c

The antioxidative defense system genes *FeSOD* and *CAT2* were upregulated under salinity and drought stresses as well as stress-related *ACS2* and *P5CS* genes. *APX1* and *GR1* gene expressions did not significantly change under stress treatments. Progesterone treatment alone usually did not change gene expression except for FeSOD and *CAT2*, which increased with progesterone treatment. Progesterone treatment under drought stress increased *FeSOD*, *CAT2*, *GR1* and *P5CS* gene expressions significantly, compared to drought stress treatments alone. *GR1* and *P5CS* gene expressions also increased with progesterone treatment under salinity stress in addition to *ACS2* gene expressions, while *FeSOD*, *CAT2* and *APX1* expressions were unaffected.

DISCUSSION

Although limited in number, previous studies have reported oxidative stress mitigation under exogenous progesterone treatment. Our study showed enhancements in root length and dry weight upon progesterone application under salinity stress. The same observation was also performed for maize seedlings under salt stress (Erdal, 2011). The results of our study also showed that the level of MDA and ion leakage was reduced with progesterone treatment under salinity stress. Similar effects were also reported by other researchers for wheat, chickpea, and maize under heat, drought, salt, and cold stresses (Erdal, 2011; Genisel et al., 2013; Su et al., 2014; Erdal and Genisel, 2016; Xue et al., 2017). Our results showed that the effects of progesterone were more prominent under salt stress compared with drought stress in terms of morphological and physiological stress parameters.

In chickpea plants, progesterone application alone increased SOD and CAT enzyme activities and decreased lipid peroxidation levels, which is also in agreement with our findings (Erdal and Dumlupınar, 2011). The abovementioned literature also reported increments in antioxidative enzyme activities, which were supported by gene expressions investigated in this study. *P5CS*, *FeSOD*, *CAT2* and *GR1* expressions were significantly higher in progesterone-treated plants under drought stress, whereas only *P5CS* and *GR1* expressions were higher under salt-stressed plants. Because plant morphology and physiological indicators show better protection under salinity stress, some other parameters not investigated in this study might explain the better protection status of salt-stressed plants upon progesterone treatment. According to Su et al. (2014) and Xue et al. (2017), the main reason for plant tolerance in progesterone-treated plants under stress is the facilitation of D1 protein phosphorylation, which plays a role in photosystem II and therefore increases photosynthetic efficiency, which was not investigated in our study.

CONCLUSION

Although we have observed stress mitigation effects of progesterone under drought and salinity stresses, our results are not sufficient to explain the molecular mechanisms of protection and better protection of tomato seedlings under salt stress conditions. Further studies including transcriptomics technologies are necessary to decipher the mechanism of action of progesterone under oxidative stress conditions.

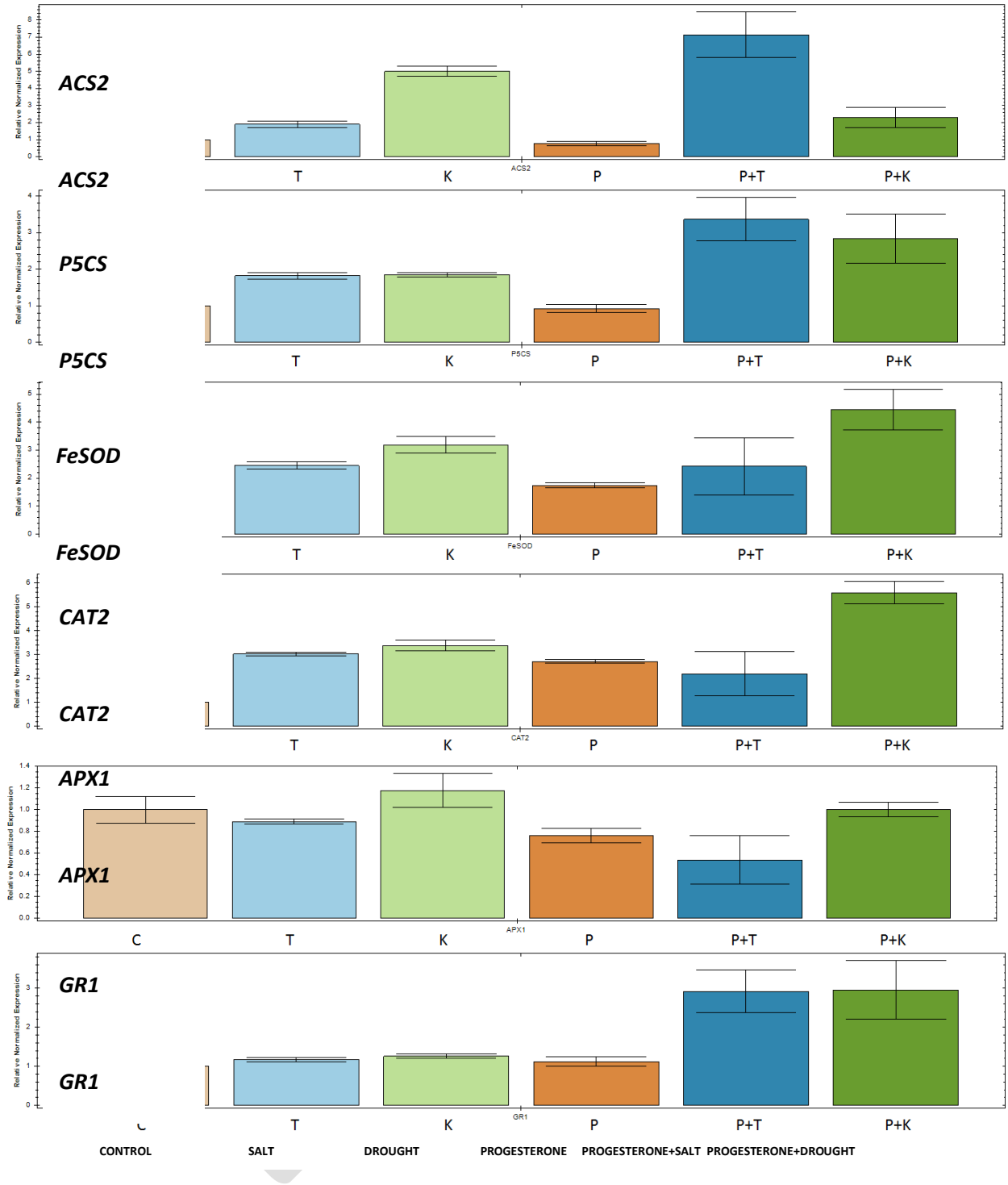


Figure 1. Graph of relative normalized gene expressions for different treatment groups. C, T, K, P, P+T and P+K indicates control, salt, drought, progesterone, progesterone+salt and progesterone+drought applications, respectively.

REFERENCES

Erdal S, Dumlupınar R 2011. Mammalian sex hormones stimulate antioxidant system and enhance growth of chickpea plants. Acta Physiol Plant, 33: 1011-1017.

Erdal S 2012a. Exogenous mammalian sex hormones mitigate inhibition in growth by enhancing antioxidant activity and synthesis reactions in germinating maize seeds under salt stress. J Sci Food Agric, 92: 839-843.

Erdal S 2012b. Alleviation of salt stress in wheat seedlings by mammalian sex hormones. J Sci Food Agric, 92: 1411-1416.

Erdal S, Genisel M 2016. The property of progesterone to mitigate cold stress in maize is linked to a modulation of the mitochondrial respiratory pathway. Theor. Exp. Plant Physiol, 28:385-393.

Filek M, Skorska ER, Sieprawaska A, Kvasnica M, Janeczko A 2017. Regulation of the membrane structure by brassinosteroids and progesterone in winter wheat seedlings exposed to low temperature. Steroids, doi:10.1016/j.steroids.2017.10.002.

Genisel M, Turk H, Erdal S 2013. Exogenous progesterone application protects chickpea seedlings against chilling-induced oxidative stress. Acta Physiol Plant, 35: 241-251.

Gupta P, Srivastava S, Seth CS 2017. 24-Epibrassinolide and sodium nitroprusside alleviate the salinity stress in *Brassica juncea* L. cv. Varuna through cross talk among proline, nitrogen metabolism and abscisic acid. Plant Soil, 411:483–498.

Haj-Amor Z, Araya T, Kim DG, Bouri S, Lee J, Ghiloufi W, Yang Y, Kang H, Jhariya MK, Banerjee A, Lal R 2022. Soil salinity and its associated effects on soil microorganisms, greenhouse gas emissions, crop yield, biodiversity and desertification: a review. Sci Total Environ, 843:156946.

Janeczko A, Filek W 2002. Stimulation of generative development in partly vernalized winter wheat by animal sex hormones. Acta Physiol Plant, 24: 291–295.

Janeczko A, Filek W, Biesaga-Koscielniak J, Marcinska I, Janeczko Z 2003. The influence of animal sex hormones on the induction of flowering in *Arabidopsis thaliana*: comparison with the effect of 24-epibrassinolide. Plant Cell, Tissue and Organ Culture, 72: 147–151.

Janeczko A, Oklestkova J, Siwek A, Dziurka, Pociecha E, Kocurek M, Novak O 2013. Endogenous progesterone and its cellular binding sites in wheat exposed to drought stress. Journal of Steroid Biochemistry and Molecular Biology, 138: 384– 394.

Janeczko A, Pociecha E, Dziurka M, Jurczyk B 2019. Changes in content of steroid regulators during cold hardening of winter wheat - Steroid physiological/biochemical activity and impact on frost tolerance. Plant Physiology and Biochemistry, 139: 215-228.

Iino M, Nomura T, Tamaki Y, Yamada Y 2007. Progesterone: Its occurrence in plants and involvement in plant growth. Phytochemistry, 68: 1664-1673.

Su X, Wu S, Yang L, Xue R, Li H, Wang Y, Zhao H 2014. Exogenous progesterone alleviates heat and high light stress induced inactivation of photosystem II in wheat by enhancing antioxidant defense and D1 protein stability. Plant Growth Regul, 74: 311-318.

Xue RL, Wang SQ, Xu HL, Zhang PJ, Li H, Zhao HJ 2017. Progesterone increases photochemic efficiency of photosystem II in wheat under heat stress by facilitating D1 protein phosphorylation. Photosynthetica, 55: 664-670.

Zulfiqar F, Ashraf M 2020. Bioregulators: unlocking their potential role in regulation of the plant oxidative defense system. Plant Mol Biol, 105:11–41.

DRAFT

ORAL PRESENTATION

Investigating Effects of Chenopodium Album L. Extracts on Alanine and Aspartate Aminotransferase Levels in Acute Gentamicin Toxicity

Mustafa Makav[1*] (*ORCID: https://orcid.org/0000-0003-1879-8180*), Cemre Aydeğer[2] (*ORCID: https://orcid.org/0000-0003-1654-6406)*, Hüseyin Avni Eroğlu[2] *(ORCID: https://orcid.org/0000-0002-1040-3255)*

[*1] Kafkas University, Faculty of Veterinary, Department of Veterinary Physiology, Kars, Turkiye
[2] Çanakkale Onsekiz Mart University, Faculty of Medicine, Department of Physiology, Çanakkale, Turkiye

* Corresponding author e-mail: mustafamakav@gmail.com

Abstract

Gentamicin, an antibiotic, is commonly used in many infectious diseases and has lots of advantages although it has some complications. It is determined that gentamicin's side effects on the liver could impair the process of the disease. Chenopodium Album L. containing flavonoids and protecting the liver is a plant. Current studies presented that Chenopodium Album L. extract ameliorates liver toxication occurred from different reasons. In light of this information, the aim of the study was that present the effects of the extract on alanine and aspartate aminotransferase (ALT and AST) in acute gentamicin toxicity. A total of 32 mature 223-250 g Wistar albino rats were divided into four groups: Control, Toxication, Extract, and Treatment. The toxication model was realized with one dose of 80 mg/kg gentamicin intraperitoneally. 7 days later, the extract was applied subcutaneously at 0.45 mg/kg. On the eighth day of the study, animals were sacrificed, and blood tissues were collected. Obtained serums were used for the determination of ALT and AST levels via the UV- P5P method. As a result of the examinations, it was found that ALT and AST levels were not changed significantly ($p<0.05$). Low damage from acute toxication or insensitivity to ALT and AST values could be the possible reason for the results. Thus, to establish the *Chenopodium album L.* effects on acute gentamicin, there is needed to comprehensive approaches.

Keywords: Gentamicin, Chenopodium album, Alanine Aminotransferase, Aspartate Aminotransferase

INTRODUCTION

Gentamicin commonly used in infections originates from gram-negative bacteria and is an antibiotic structured the aminoglycoside. Given patients' ages, symptoms, and vitals, it could be applied and be treat local microbial resistance including bacterial septicemia, meningitis, urinary system infections, and soft tissue infections (Chaves and Tadi, 2020). Despite the clinical advantages of gentamicin like the other aminoglycoside, it has critical complications. The most seen complication is nephrotoxicity, and it could be seen in approximately %30 of patients following the 7 days of the gentamicin treatment. Current studies indicated that gentamicin could cause acute kidney injury even if used in only one dose. Although kidney toxicity due to gentamicin comes to the forefront, it is more important that side effects on the liver have a role detoxification of medication and toxins (Babaeenezhad et al., 2021; Galaly et al., 2014). According to some researchers, it results from long-distance organ injury following acute kidney damage (Babaeenezhad et al., 2021). On the other hand, gentamicin directly increases superoxide anion, hydrogen peroxide, and hydroxyl radicals produced by mitochondria. The free radicals start some cellular activity including phospholipid membrane peroxidation, breaking of DNA chain, and protein denaturation. Therefore, it can result in cellular damage (Najaran et al., 2014). So, the process causes oxidative stress and increases alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkalin phosphatase (ALP). Additionally, an increase in reactive oxygen species triggers inflammation and suppresses the endogen antioxidant system (Babaeenezhad et al., 2021; Najaran et al., 2014). With all, studies should be focused on the antioxidant and anti-inflammatory agents to prevent gentamicin-related toxicity.

Chenopodium album L., is a fast-growing and annual plant called *sirken* in our country (Atalay and Kamalak, 2019). This plant also grows on different continents containing different contents like fatty acids, flavonoids,

and β-carotene. So that it has antibacterial, anti-fungal, anti-nociceptin, anti-inflammatory, anti-diabetic, and anti-hyperlipidemic properties (Chamkhi et al., 2022) exhibiting also hepatoprotective effects (Ata et al., 2015; Baldi and Choudhary, 2013; Jain & Singhai, 2012; Nayak et al., 2012; Parkash & Patel, 2015). Nayan et al. (2012) reported that Chenopodium album L. protected the liver against CCl4-induced hepatotoxicity decreasing serum glutamate oxaloacetate transaminase (SGOT), serum glutamate pyruvate transaminase (SGPT) levels (Nayak et al., 2012). Jain and Sighai (2012) experimented with Chenopodium album L. effects in vivo and in vitro on CCl4-induced hepatotoxicity. The in vitro and in vivo results indicated that AST, ALT, and lactate dehydrogenase (LDH) levels were decreased. (Jain & Singhai, 2012). Considering, Chenopodium album L. extract has alleviating effects on the liver. Moreover, there is a lack of the effects of Chenopodium album L. extract on gentamicin-related hepatotoxicity. Therefore, in this study, we aimed to investigate the effects of Chenopodium album L. extract on gentamicin-related acute hepatotoxicity, ALT, and AST levels.

MATERIALS AND METHODS

2.1. Animals

Male Wistar albino rats (32 animals, 225-250 g) were obtained from Kafkas University Experimental Research Centre. Rats were housed at 40-60% humidity, 22±2 ˚C temperature, and 12 h/12 h light/dark cycle. During the experiment, rats were fed with standard food pellets and water *ad libitum*. All rats were examined in terms of behavioural, respiratory, and cardiovascular conditions. Distressed rats were not counted in the study. The whole protocol was proper with the 1964 Helsinki Declaration.

Rats were randomly divided equally into 4 experimental groups as represented below:

- **Group 1 (Control, n=8)**: A vehicle of extract was applied to rats orally by gavage.
- **Group 2 (Toxicity, n=8):** Acute toxicity was established by Gentamicin.
- **Group 3 (Extract, n=8):** Only extract was applied to rats orally by gavage.
- **Group 4 (Treatment, n=8):** Acute toxicity was established by Gentamicin after the extract was applied to rats orally by gavage.

2.2. Establishment of Gentamicin Toxicity

Toxicity was established according to the previous model of Sule and Arhoghro (2016). Briefly, only one dose of 80 mg/kg gentamicin was injected intraperitoneally (i.p.) (Sule and Arhoghro, 2016).

2.3. Application of *Chenopodium album L.* Extract

Chenopodium Album L. was given orally at a dose of 0,45 mg/kg by gavage after 7 days of acute toxicity.

2.4. Euthanasia and Harvesting

Animals were sacrificed under general anaesthesia (ketamine hydrochloride (75mg/kg) and xylazine (15 mg/kg) i.m.) on the 8th day of the study. Blood samples were harvested, and serum ALT-AST levels were evaluated by the UV - P5P method.

2.6. Statistics

Data was determined with IBM SPSS (Version 26). Normality was performed by the Shaphiro-Wilk test. Statistical differences were examined by one-way ANOVA and post-hoc Tukey test. The significance is represented as $p< 0.05$.

DRAFT

RESULTS

Our results indicated that there were differences between experimental groups in ALT and AST levels, but results were statistically insignificant ($p<0.05$, Figure 1).

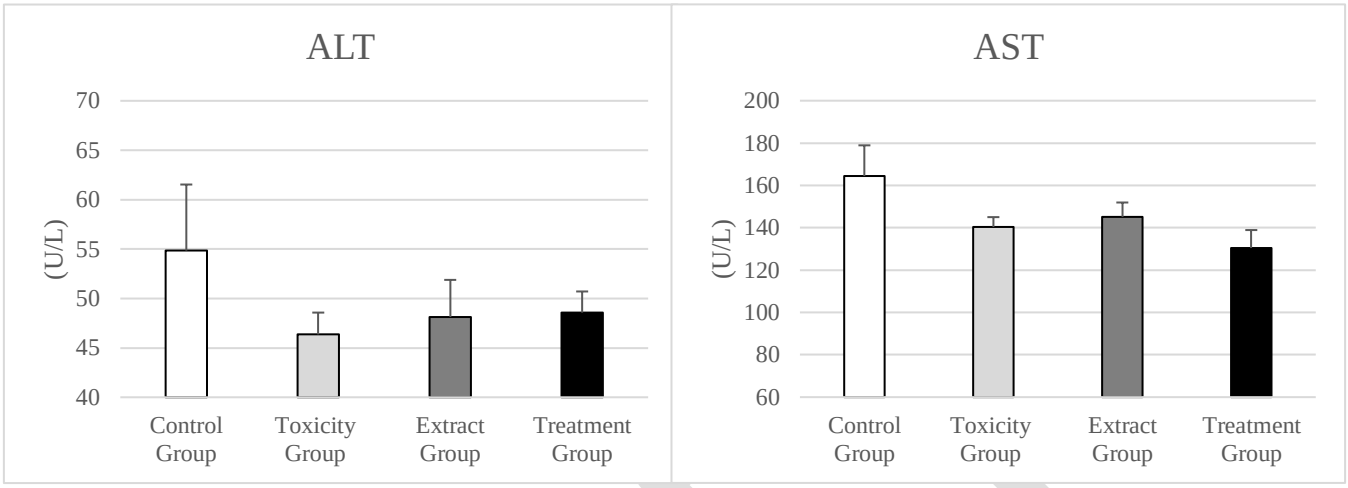


Figure 1: Serum ALT and AST levels of experimental groups.

DISCUSSION

In the present study, we aimed to investigate the effects of Chenopodium album L. extract on ALT and AST levels of gentamicin with one dose-induced hepatotoxicity in rats. To do that Sule and Arhoghro's (2016) acute toxicity model was established in rats.

ALT is an enzyme composed of 496 amino acids with a half-life life 47±10 h. ALT has three isoforms called ALT1, ALT2 and ALT2_2. Among them, ALT2_2 is enzymatically inactive. ALT is mainly located in the cytosol of hepatocytes but is also placed minimally in muscle, adipose tissue, colon, prostate, and brain. ALT located in hepatocytes is 3000 times more active than in serum ALT. (Åberg et al., 2021; Ndrepepa and Kastrati, 2019). At clinical practice, serum ALT levels are cheap and reachable tests for the measurement of liver function. The release of ALT following tissue injuries could be able to diagnose a range of pathologies like viral and toxic hepatitis, muscular dystrophy, and other muscular atrophy diseases. Specifically, serum ALT levels increase following liver injury (Liu et al., 2014). In our study, serum ALT levels of rats were examined. According to the biochemistry results, there was not a significant difference between groups in ALT levels. It was suggested that there could be a variety of reasons for this result. Previous studies indicated that serum ALT levels might not always increase in all liver injury cases (Liu et al., 2014). Thus, our outcomes indicated differences in serum ALT levels but were statistically insignificant. Additionally, ALT levels in circulation have many origins. Due to a lack of knowledge about underlying mechanisms, ALT crosses to the intracellular space and circulation by cytoplasmic budding or blow-ups (Lindblom et al., 2007). Therefore, in our study gentamicin was applied in only one dose, so it is possible that this application wouldn't have as much damage to the hepatocytes. Also, ALT does not show up in the diurnal rhythm. Serum ALT levels are 45% higher during daytime while between different days the serum ALT levels differ from 10% to 30% (Cordoba et al., 1998; Fraser and Harris, 1989).

AST has a key role in amino acid metabolism and there are cytoplasmic and mitochondrial isoforms. AST is expressed in almost every tissue. Initially, the heart and liver then skeletal muscle, kidneys, pancreas, spleen, lung, and erythrocytes. AST is wiped out from circulation by hepatic sinusoidal cells. The total serum AST half-life is 17 h. From the early 1950s serum AST levels were commonly used for diagnosis and prognosis of liver diseases. Serum ALT levels increase was reported after inflammatory liver disease, alcoholic liver disease, cirrhosis, and drug toxicity (Åberg et al., 2021; Ndrepepa, 2021). So, in this study, we examined serum AST levels in addition to ALT levels. There was no significant difference between groups in serum AST levels. We suggested that the severity of gentamicin toxicity might be minimal. Previous studies reported that median

injuries exhibit lower enzymatic fraction (Wieme and Demeulenaere, 1970). In addition, hepatocyte injury and plasma aminotransferase correlation are also weakened (Pratt and Kaplan, 2000).

CONCLUSION

Chenopodium album L. did not show any effects on serum ALT and AST levels. So, we could declare that Chenopodium album L. does not have hepatotoxic effects.

In this study, histopathological and genetic analyses were not performed. The powerful side of our study is to examine the effects of Chenopodium album L on ALT and AST levels which are routine tests. Further studies are required to investigate the effects of Chenopodium album L on gentamicin cytotoxicity.

REFERENCES

Åberg,F, Danford CJ, Thiele M, Talbäck M, Rasmussen DN, Jiang ZG, Hammar N, Nasr P, Ekstedt M, But A, Puukka P, Krag A, Sundvall J, Erlund I, Salomaa V, Stål P, Kechagias S, Hultcrantz R, Lai M, Afdhal N, Jula A, Männistö S, Lundqvist A, Perola M, Färkkilä M, Hagström, H 2021. A Dynamic Aspartate-to-Alanine Aminotransferase Ratio Provides Valid Predictions of Incident Severe Liver Disease. Hepatology Communications, 5(6): 1021–1035.

Ata M, Jahangir S, Islam M, Akter R, Sarkar TH, Uddin G, Chowdhury ZA, Biswas RSR 2015. Hepatoprotective Effect of Chenopodium Album Linn. Whole Plant on Biochemical Parameters Against Paracetamol Induced Hepatotoxicity in Rats. Chattagram Maa-O-Shishu Hospital Medical College Journal, 14(2); 1-4.

Atalay Aİ, Kamalak A 2019. Olgunlaşma Dönemlerinin Sirken (Chenopodium album) Otunun Kimyasal Kompozisyonuna, Besleme Değerine ve Metan Üretimine Etkisi. Türk Tarım ve Doğa Bilimleri Dergisi, 489–493.

Babaeenezhad E, Nouryazdan N, Nasri M, Ahmadvand H, Moradi Sarabi M 2021. Cinnamic acid ameliorate gentamicin-induced liver dysfunctions and nephrotoxicity in rats through induction of antioxidant activities. Heliyon, 7(7); e07465.

Baldi A, Choudhary NK 2013. In vitro antioxidant and hepatoprotective potential of chenopodium album extract. International Journal of Green Pharmacy, 7(1); 50–56.

Chamkhi I, Charfi S, El Hachlafi N, Mechchate H, Guaouguaou FE, El Omari N, Bakrim S, Balahbib A, Zengin G, Bouyahya A 2022. Genetic diversity, antimicrobial, nutritional, and phytochemical properties of Chenopodium album: A comprehensive review. Food Research International, 154; 110979.

Chaves BJ, Tadi P 2020. Gentamicin, StatPearls Publishing, pp. 964–991.

Cordoba T, O'Riordan K, Dupuis J, Borensztajn J, Blei AT, Poynard T, Piton A, Bismuth FI, Pelissier E, Sansonetti N, Opolon P 1998. Diurnal variation of serum alanine transaminase activity in chronic liver disease. Hepatology (Baltimore, Md.), 28(6); 1724–1725.

Fraser GG, Harris EK 1989. Generation and application of data on biological variation in clinical chemistry. Critical Reviews in Clinical Laboratory Sciences, 27(5); 409–437.

Galaly SR, Ahmed OM, Mahmoud MA 2014. Thymoquinone and curcumin prevent gentamicin-induced liver injury by attenuating oxidative stress, inflammation and apoptosis. J Physiol Pharmacol, 65(6); 823-832.

Jain N, Singhai A 2012. Hepatoprotective activity of Chenopodium album Linn: in vitro and in vivo studies. Journal of Experimental and Integrative Medicine, 2(4); 331.

Lindblom P, Rafter I, Copley C, Andersson U, Hedberg JJ, Berg AL, Samuelsson A, Hellmold H, Cotgreave, I, Glinghammar B 2007. Isoforms of alanine aminotransferases in human tissues and serum--differential tissue expression using novel antibodies. Archives of Biochemistry and Biophysics, 466(1), 66–77.

Liu Z, Que S, Xu, J, Peng T. 2014. Alanine Aminotransferase-Old Biomarker and New Concept: A Review. International Journal of Medical Sciences, 11(9); 925.

Najaran M, Mokaber H, Pourahmadi M, Farzam M, Jahromi HK. 2014. Pathological changes of gentamicin in liver tissue and antioxidant property of cinnamon extract on wistar rats. Biomedical and Pharmacology Journal, 7(1); 341–347.

Nayak PD, Dinda SC, Swain PK, Kar B., Patro VJ 2012. Hepatoprotective activity against CCL 4-induced hepatotoxicity in rats of Chenopodium album aerial parts. Journal of Phytotherapy and Pharmacology, 4(2); 33–41.

Ndrepepa G 2021. Aspartate aminotransferase and cardiovascular disease - A narrative review. In Journal of Laboratory and Precision Medicine 6(6).

Ndrepepa G, Kastrati A 2019. Alanine aminotransferase—a marker of cardiovascular risk at high and low activity levels. In Journal of Laboratory and Precision Medicine, 4(4); 29.

Parkash J, Patel KR 2015. Hepatoprotective Activity of Chenopodium Album Leaves Extract in CCl4 Induced Hepatotoxicity in Rats. Journal of Drug Delivery & Therapeutics, 5(2); 88

Pratt DS, Kaplan MM 2000. Evaluation of abnormal liver-enzyme results in asymptomatic patients. The New England Journal of Medicine, 342(17); 1266–1271.

Sule OJ, Arhoghro ME 2016. Biochemical Effect of Ethanolic Extract of Phyllantus Amarus (L.) On Gentamicin-Induced Liver and Kidney Damage in Rats. Journal of Medical and Biological Science Research, 2(7); 114–117.

Wieme RJ, Demeulenaere L 1970. Enzyme assays in liver disease. Journal of Clinical Pathology. Supplement (Association of Clinical Pathologists), 4(1); 51–59.

DRAFT

ORAL PRESENTATION

Exercise model for experimental animals

Cemre Aydeğer[1*] (*ORCID: https://orcid.org/0000-0003-1654-6406)*, Hüseyin Avni Eroğlu[1] *(ORCID: https://orcid.org/0000-0002-1040-3255)*, Mustafa Makav[2] (*ORCID: https://orcid.org/0000-0003-1879-8180*),

[*1] Çanakkale Onsekiz Mart University, Faculty of Medicine, Department of Physiology, Çanakkale, Turkiye
[2] Kafkas University, Faculty of Veterinary, Department of Veterinary Physiology, Kars, Turkiye

* Corresponding author e-mail: cemre.ayd@hotmail.com

Abstract

Exercise is a drug that provides a healthy and quality lifestyle, prevents pathologies, and is a treatment method. Exercise is the most common application with its benefits and has no side effects. Considering the beneficial effects of exercise, studies about exercise gained attention in recent years. The underlying molecular mechanisms of exercise are still lacking. Therefore, animal models established on exercise are required. Several exercise models exerting advantages and disadvantages have been used in experimental studies. With all, this study aimed to determine the experimental models in exercise. To that, professional experimental studies in this field have been examined which were published in Pubmed and Google Scholar databases between the years 2003-2023. Results have indicated that experimental models in exercise studies are classified as volunteered, non-volunteered, compulsory, and specific models. It has been pointed out in those studies that all classified exercise models both have positive and negative effects and were not ideal types of exercise. It was also suggested that the options for exercise models should have been established according to the main hypothesis. Additionally, the target population should have been considered during exercise model selection.

Keywords: exercise, exercise therapy, experimental animal models

INTRODUCTION

Exercise is a type of drug that provides a healthy quality of life for individuals and prevents diseases. The importance of exercise elevating day by day due to its mental and physical enhancing properties. Therefore, the studies based on exercise effects gained attract recently (Ruegsegger and Booth, 2018; Thompson et al., 2020). Due to the systemic effects of exercise, studies mostly were established in vivo not in vitro. So exercise studies are generally established on humans or animals (Feng et al., 2019). Human studies are mostly comprised of epidemiological research. In addition, tissue harvesting for underlying molecular mechanisms of exercise effects from humans is not ethically approved. Thus, animal models are used extensively in exercise studies (Chu-Tan et al., 2022).

In animal models for exercise studies, generally, dogs, cats, and rabbits are used (Feng et al., 2019). But rats and mice are the most common types of animals that are used in exercise animal models with a 95 % rate (Hickman et al., 2017; Seo et al., 2014). Here we summarized in this review the exercise models that are established on mice/rats and, the advantages and disadvantages of these models.

MATERIALS AND METHODS

The present study is based on the studies which were published between the years 2003-2023 by professionals of the field with the keywords; exercise, experimental models, exercise physiology, and experimental exercise models. The related studies were analyzed and most related of the research were contributed to the review.

DRAFT

RESULTS and DISCUSSION

Experimental exercise models can be categorized as volunteered exercise, non-volunteered exercise, forced exercise, and specified exercise (Seo et al., 2014).

The voluntary exercises are a type of exercise that does not activate a systemic response. So that general parameters do not generally change (Naderi et al., 2015). This type of exercise is commonly performed on wheels set up inside a cage. The animals are allowed to the mechanism ad-libitum for 24 hours. Therefore, a force is not applied to the animals. The mechanism can be customized with additive equipment if required. For example, the laps can be increased by the addition of extra wheels. Also, the distance that the animal made can be evaluated. The mechanism also can be standardized by locking some of the wheels (Leasure and Jones, 2008). Further, volunteer exercise also has some disadvantages. Although this type of exercise is an aerobic exercise, it is limited in exhaustive experiments. Considering that, it is more appropriate for senescence, behavioral research, ischemia, cancer, diabetes, emotional conditions, muscle physiology, neuromuscular diseases, and stroke cases. Additionally, because the animals are on their own during the whole exercise process, their paws may stuck to the mechanism (Seo et al., 2014).

One of the other exercise animal models is involuntary exercise. The muscle contraction and exercise process are applied to the animal by an electrical force from outside. The common method in this process is functional electrical stimulation (FES). To achieve this method, the electrodes should have been implanted under general anesthesia in the muscles of the animals. Following implantation, the stimulation threshold is determined with the application of different voltages. The exercise begins with this electrical threshold voltage application. The most attractive advantage of this method is that animal motivation and coordination are not required. That means this method is quantitative and repeatable. Differing from conscious exercise, all motor units can be activated with electrical stimulation (Ke et al., 2011). The disadvantages of this method involve anesthesia requirements and implantation can cause neuropathy. Under higher frequency conditions, the muscle fibers may be exposed to transmission. Besides, the muscle contraction is not physiological (Seo et al., 2014).

In forced exercise models, animals are forced to get on the exercise platform. The treadmill, lifting weights, squads, swimming, and stairs up are subtypes of forced exercise. The treadmill is a common exercise model which is used for measuring time and speed. The treadmill is nearly the same as a human treadmill, so the slope and speed of the tool can be changed as required. The treadmill has a school gride behind it. The animals are forced to run on the treadmill with the effect of the shock grill. Running exercise also consists of an adaptation process. Additionally, rats are nocturnal animals so this type of exercise can be applied under red light in the dark (Burghardt et al., 2004; Seo et al., 2014; Shankarappa et al., 2011; Yoo et al., 2000; Zhang et al., 2013).

The mechanism of weightlifting exercise was developed by Klitgaar in 1988, and it has been used since then with different modified models. The animals were forced to reach the food above them with load bearing on their shoulders. During the exercise, the plantar flexors and knee and ankle extensor muscles work. This type of exercise is similar to human progressive resistance exercise and quantitative. In addition, the exercise forces to work bilateral extremities, so multi-muscle comparison can be available. Further, the exercise requires an adaptation process for two weeks and fasting. The fasting procedure provides minimal muscle hypertrophy (Klitgaard, 1988; Krutki et al., 2017; Legerlotz et al., 2007; Seo et al., 2014).

Squad with pulling weight is a forced exercise, differing from weightlifting exercise with making the exercise at the horizontal plane. An award and punishment system is applied. Food is award and miliamper electrical disturbance is used as punishment. So, the movement is provided (Seo et al., 2014).
Stairs up exercise is one of the forced exercise types and also a resistance exercise. The mechanism consists of a start point, stairs, and arrival cage. At the start point, the animal is forced to start the route with electrical stimulation at a milliampere level. The food is placed in the arrival cage and the animal is forced to climb up stairs. According to the selected protocol, a weight can be loaded on animal tails or backs. The exercise procedure requires an adaptation process. The time of adaptation decreases animal stress (Kelty et al., 2019; Krutki et al., 2017; Zhai et al., 2020).

Swimming is known as aerobic and anaerobic forced and resistance exercise. The required equipment for this exercise is cheaper than a water tank. During the procedure, a different weighed lifting was loaded on animals' tails such as 5-10% of animal weights (adapting load- performance test load). Moreover, it is suggested that animals are taken after the exercise for the body and water temperature (Campos-Ferraz et al., 2013; Falavigna et al., 2012; Seo et al., 2014).

The specified exercises are the specifically aimed models which are used in experimental exercise models. The legs of animals are lifted and elevated to establish muscle atrophy in space and flight physiology studies. Tounge exercises are required for dysphagia pathologies. Bipedalled downstream running (eccentric) exercises are proper for Achille's tendinosis, muscle damage, or musculoskeletal pain models (Hinks et al., 2022; Seo et al., 2014; Williamson et al., 2021). It is clear from that, there are specific exercise models for specific patterns.

CONCLUSION

There are a bunch of exercise models for investigating the prevention and treatment effects of exercise. These models include aerobic, anaerobic, and resistance exercises. In addition, it is possible to select different exercise times and speeds. Specified exercises are also appropriate for specialized types of exercise models. Considering that, it can be suggested that every exercise model can be modified according to the selected study topic and the main object of the study.

REFERENCES

Burghardt PR, Fulk LJ, Hand GA, Wilson MA 2004. The effects of chronic treadmill and wheel running on behavior in rats. Brain Research, 1019(1–2); 84–96.

Campos-Ferraz PL, Bozza T, Nicastro H, Lancha AH 2013. Distinct effects of leucine or a mixture of the branched-chain amino acids (leucine, isoleucine, and valine) supplementation on resistance to fatigue, and muscle and liver-glycogen degradation, in trained rats. Nutrition, 29(11–12); 1388–1394.

Chu-Tan JA, Kirkby M, Natoli R 2022. Running to save sight: The effects of exercise on retinal health and function. Clinical & Experimental Ophthalmology, 50(1); 74–90.

Falavigna G, de Araújo Junior JA, Rogero MM, Pires ISdeO, Pedrosa RG, Martins Junior E, de Castro IA, Tirapegui J 2012. Effects of Diets Supplemented with Branched-Chain Amino Acids on the Performance and Fatigue Mechanisms of Rats Submitted to Prolonged Physical Exercise. Nutrients, 4(11); 1767–1780.

Feng R, Wang L, Li Z, Yang R, Liang Y, Sun Y, Yu Q, Ghartey-Kwansah G, Sun Y, Wu Y, Zhang W, Zhou X, Xu M, Bryant J, Yan G, Isaacs W, Ma J, Xu X 2019. A systematic comparison of exercise training protocols on animal models of cardiovascular capacity. Life Sciences, 217; 128–140.

Hickman DL, Johnson J, Vemulapalli TH, Crisler JR, Shepherd R 2017. Commonly Used Animal Models. Principles of Animal Research for Graduate and Undergraduate Students, 117.

Hinks A, Jacob K, Mashouri P, Medak KD, Franchi MV, Wright DC, Brown SHM, Power GA 2022. Influence of weighted downhill running training on serial sarcomere number and work loop performance in the rat soleus. Biology Open, 11(7).

Ke Z, Yip SP, Li L, Zheng XX, Tong KY 2011. The Effects of Voluntary, Involuntary, and Forced Exercises on Brain-Derived Neurotrophic Factor and Motor Function Recovery: A Rat Brain Ischemia Model. PLOS ONE, 6(2); e16643.

Kelty TJ, Schachtman TR, Mao X, Grigsby KB, Childs TE, Dylan Olver T, Michener PN, Richardson RA, Roberts CK, Booth FW 2019. Resistance-exercise training ameliorates LPS-induced cognitive

impairment concurrent with molecular signaling changes in the rat dentate gyrus. Journal of Applied Physiology, 127(1); 254–263.

Klitgaard H 1988. A model for quantitative strength training of hindlimb muscles of the rat. Journal of applied physiology, 64(4); 1740-1745.

Krutki P, Mrówczyński W, Baczyk M, Łochyński D, Celichowski J 2017. Adaptations of motoneuron properties after weight-lifting training in rats. Journal of Applied Physiology, 123(3); 664–673.

Leasure JL, Jones M 2008. Forced and voluntary exercise differentially affect brain and behavior. Neuroscience, 156(3); 456–465.

Legerlotz K, Schjerling P, Langberg H, Brüggemann GP, Niehoff A 2007. The effect of running, strength, and vibration strength training on the mechanical, morphological, and biochemical properties of the Achilles tendon in rats. Journal of Applied Physiology, 102(2); 564–572.

Naderi R, Mohaddes G, Mohammadi M, Ghaznavi R, Ghyasi R, Vatankhah AM 2015. Voluntary Exercise Protects Heart from Oxidative Stress in Diabetic Rats. Advanced Pharmaceutical Bulletin, 5(2); 231.

Ruegsegger GN, Booth FW 2018. Health Benefits of Exercise. Cold Spring Harbor Perspectives in Medicine, 8(7); a029694.

Seo DY, Lee SR, Kim N, Ko KS, Rhee BD, Han J 2014. Humanized animal exercise model for clinical implication. Pflugers Archiv European Journal of Physiology, 466(9); 1673–1687.

Shankarappa SA, Piedras-Rentería ES, Stubbs EB 2011. Forced-exercise delays neuropathic pain in experimental diabetes: effects on voltage-activated calcium channels. Journal of Neurochemistry, 118(2); 224–236.

Thompson WR, Sallis R, Joy E, Jaworski CA, Stuhr RM, Trilk JL 2020. Exercise Is Medicine. American Journal of Lifestyle Medicine, 14(5); 511–523.

Williamson PM, Freedman BR, Kwok N, Beeram I, Pennings J, Johnson J, Hamparian D, Cohen E, Galloway JL, Ramappa AJ, DeAngelis JP, Nazarian A 2021. Tendinopathy and tendon material response to load: What we can learn from small animal studies. Acta Biomaterialia, 134; 43–56.

Yoo HS, Bunnell BN, Crabbe JB, Kalish LR, Dishman RK 2000. Failure of neonatal clomipramine treatment to alter forced swim immobility: chronic treadmill or activity-wheel running and imipramine. Physiology & Behavior, 70(3–4); 407–411.

Zhai L, Liu Y, Zhao W, Chen Q, Guo T, Wei W, Luo Z, Huang Y, Ma C, Huang F, Dai X. 2020. Aerobic and resistance training enhances endothelial progenitor cell function via upregulation of caveolin-1 in mice with type 2 diabetes. Stem Cell Research & Therapy, 11(1).

Zhang P, Yu H, Zhou N, Zhang J, Wu Y, Zhang Y, Bai Y, Jia J, Zhang Q, Tian S, Wu J, Hu Y 2013. Early exercise improves cerebral blood flow through increased angiogenesis in experimental stroke rat model. Journal of NeuroEngineering and Rehabilitation, 10(1); 1–10.

DRAFT

ORAL PRESENTATION

A step beyond probiotics as new concept: postbiotic

Seda SEYİRT[1*](*ORCID: https://orcid.org/0000-0002-4975-0561*), Pınar ŞANLIBABA[2] (*ORCID: https://orcid.org/0000-0003-4638-6765)*

[*1]Ankara University, Engineering Faculty, Department of Food Engineering, Ankara, Turkey
[2]Ankara University, Engineering Faculty, Department of Food Engineering, Ankara, Turkey

*Corresponding author e-mail: sseyirt@ankara.edu.tr

Abstract

The term 'postbiotics' refers to soluble factors secreted by live bacteria, or released after bacterial lysis, such as enzymes, peptides, teichoic acids, peptidoglycan-derived muropeptides, polysaccharides, cell surface proteins, and organic acids that give a health advantage to the host. Postbiotics take attention for their clear chemical structure, safety dose parameters, long shelf life and ingredient of various signaling molecules which may have anti-inflammatory, immunomodulatory, anti-obesogenic, antihypertensive, hypocholesterolemic, anti-proliferative, and antioxidant activities. Postbiotics imitate the evidenced health benefits of probiotics without the safety risks of probiotics, which include microbial translocation, infection or reducing enhanced inflammatory responses. Additionally, postbiotics do not be necessary tight manufacturing or storage conditions, and also to being more stable than the living bacteria and having a longer shelf life, which these new concept may be a good alternative to probiotics.

Keywords: Postbiotics, Probiotics, Health benefits

INTRODUCTION

In the early XX century, Metchnikoff observed that fermented dairy products such as yoghurt were associated with better health and longevity among people living in Bulgarian villages and caused to the development of the probiotic concept (Metchnikoff, 1907). According to FAO/WHO (2001) defined that probiotics are “live microorganisms that, when administered in adequate amounts, confer a health benefit on the host”. Probiotics mainly comprise of *Lactobacillus* species, *Bifidobacterium* species, and *Streptococcus* species, and also *Lactococcus lactis* and some *Enterococcus* species. Probiotics are reported many beneficial health effects such as increasing nutrients bioavailability, enhancement of intestinal health, improvement of immune response, lowering serum cholesterol levels, prevention of cancer, reduce lactose intolerance, prevention or reduction of allergies in sensitive individuals, reduction of the risk of gastrointestinal diseases (Argyri et al., 2013, Kechagia et al., 2013). The beneficial effect of probiotics depends on the able to survive passage through the GIT and colonise the GIT. Thus, viability of probiotics are important property to ensure the beneficial effects to the host’s health and this complicate use of probiotics in some foods. The many factors such as the composition of the food matrix, the processing and storage conditions can reduce the probiotic cell viability (Russell et al., 2011). On the other hand, a new approach such as paraprobiotic and postbiotic were constituted which imply bacterial viability is not an essential requirement for health benefits, supplying a broad dimension to the probiotic concept (Barros et al., 2020).

The term postbiotics, also known as either metabiotics, biogenics, or simply metabolites/CFS (cell-free supernatants); refers to soluble factors (products or metabolic byproducts) secreted by live bacteria or released after bacterial lysis that give a health advantage to the host. On the other hand, paraprobiotics, also known as “non-viable probiotics”, “inactivated probiotics” or “ghost probiotics”, is defined inactivated (non-viable) microbial cells, which, when administered in sufficient amounts confer benefits to consumers (Cuevas-González et al., 2020). Postbiotics and paraprobiotics are more stable and safer for industrial uses when compared to the viable microorganisms because of their viability is not necessary either during consumption or large-scale production thus, its applications in food products may offer several technological advantages for the food manufacturers (Barros et al., 2020). In addition to, nonviable microbial cells can have safety

benefit over probiotics by decreasing the risk of microbial translocation, infection or rised inflammatory responses (Taverniti and Guglielmetti, 2011).

Postbiotics

The word postbiotic is derived from the Greek for 'post', meaning after, and 'bios', meaning life. (Vinderola, 2022). Postbiotics is a recently arised concept that states to a broad range of bioactive molecules including non-viable/inactivated microbial cells, cell compounds, and any soluble products or metabolic by products resulting from probiotics, which, when applied to consumers, directly or indirectly mediate positive biological activity (Bourebaba et al., 2022). Currently used definition of this concept was "preparation of inanimate microorganisms and/or their components that administer a health benefit to the host" (Salminen et al., 2021). Therefore, postbiotics have been accepted to imitate probiotics functions and activities, and do not be necessary tight manufacturing or storage conditions (Patel and Denning, 2013). Postbiotics have several alluring properties that including clear chemical structures, safety dose parameters, and longer shelf life. The use of postbiotics can be a current and safer alternative to avoids risk related to viable probiotic bacteria (Aguilar-Toalá et al., 2018)

Lactobacillus species (*L. rhamnosus*, *L. bulgaricus*, *L. acidophilus*, *L. reuteri*, *L. casei*, *L. fermentum*, *L. paracasei*, *L. helveticus*, *L. johnsonni*, and *L. gasseri*) and *Bifidobacterium* species (*B. bifidum*, *B. longum* subspecies *infantus*, *B. breve*, and *B. longum*) have been reported the most common source of postbiotics. High effective postbiotics which include cytoplasmic extracts and cell wall components were detected in several species relating to *Lactobacillus*, *Bifidobacterium*, *Saccharomyces*, *Bacillus*, *Streptococcus*, or *Faecalibacterium* genera (Bourebaba et al., 2022). The majority of postbiotics have been acquired by using cell disruption techniques, such as heat and enzymatic treatment, solvent extraction, as well as sonication (Aguilar-Toalá et al., 2018).

Classification of postbiotics and its therapeutic benefits

There are various types of postbiotics molecules resulting from extracellular and intracellular probiotic cells. The postbiotics molecules contain peptidoglycans-derived muropeptides, exopolysaccharide (EPS), teichoic acids, and surface protruding molecules like fimbriae, pili or flagella that constitute cell wall components, secreted proteins/peptides, bacteriocins such as acidophilin, reuterin, and bifidin, cell-free supernatant, organic acids such as lactic acid and acetic acid, vitamins, short-chain fatty acids like butyric acid and propionate, neurotransmitters, biosurfactants, etc. (Bourebaba et al., 2022).

Because of their established chemical structures and long storage stability, postbiotics have been found to have health benefits by demonstrating local effects on specific tissues of the gut epithelium and by influencing multiple others organs and tissues. Postbiotics objective to prevent the risk of live microorganisms application to altered intestinal barriers or compromised immune defenses by imitate the therapeutic effect of probiotics and also to being more stable and having a longer shelf life (Patel and Denning, 2013; Sharma and Shukla, 2016).

Many potential activities of postbiotics which include anti-inflammatory and antibacterial properties, immunomodulatory, anti-obesogenic, anticarcinogenic, antihypertensive, hypocholesterolemic, antiproliferative, and antioxidant activities, boosting the immune system and strengthening the function of the gut barrier have been reported (Aguilar-Toalá et al., 2018; Sharma and Shukla, 2016). In addition to, postbiotics have been detected having antiproliferative specific activity to colon cancer cells (Tiptiri-Kourpeti et al., 2016). Studies support that beneficial effects of postbiotic may depend on secreted-derived factors (Compare et al., 2017).

In vitro and in vivo anti-cancer, anti-inflammatory, anti-proliferative, and immunomodulatory effect of peptidoglycan obtained from probiotic bacteria have been reported (Bourebaba et al., 2022). Effecty of Cell-free supernatants (CFS) such as oxidative stress reduction, anti-inflammatory, anti-tumoral, antibacterial effects as well as biofilms formation suppression have been detected (De Marcoet al., 2018; Escamilla et al., 2012). It has been found that cell-free extracts produced from *B. longum* and *L. acidophilus* indicated a significant antioxidative activity (Lin and Chang, 2000).

Posbiotics was identified as pathogenic bacteria inhibitors towards pathogens which include *Listeria monocytogenes* L-MS, *Salmonella enterica* S-1000, *Escherichia coli* E−30 and vancomycin resistant *Enterococci*, when using cell-free supernatants culture got from *L. plantarum* strains RG11, RG14, RI11, UL4,

TL1 and RS5 strains. (Kareem et al., 2014). Specific SCFAs have been demonstrated to conduce to plasma cholesterol homeostasis in rodents and humans (den Besten et al., 2013). In addition, another producers of postbiotics are yeast. Canocini et al. (2011) detected that culture supernatants, when get from *Saccharomyces boulardii*, developed wound healing capacity and epithelial cells migration via the activation of α2β1 integrin collagen receptors in vitro models.

It has been showed that the culture supernatant obtained from *Lactobacillus paracasei* B21060 may preserve healthy tissue towards the inflammatory properties of invasive *Salmonella* in a human mucosa explant of colon (Tsilingiri et al., 2012), and also culture supernatant when got from *Lactobacillus casei* DG, may alleviate the inflammatory response in ileac and colonic mucosa cultures from post-infectious bowel syndrome patients (Compare et al., 2017). Xu et al. (2011) reported that exopolysaccharides (EPS) from *Bifidobacterium animalis* indicated in vitro inhibition of lipid peroxidation and radical scavenging activity (hydroxyl and superoxide radicals).

Applications of postbiotics

Postbiotics is more stable than the living bacteria they are derived from (Venema, 2013) and also their positive safety profile as there is no require for the uptake of billions of living microbes (Shigwedha et al., 2014). Another major advantage of postbiotics may be applied in a controlled and standardized way (Gabriele, 2016).

Individual postbiotic has many food safety functions such as in food biopreservation and packaging, control and eradication of foodborne pathogen's biofilm, biodegradation of harmful chemicals contaminants. The effectivenes sof postbiotics in the food systems is attached to many factors include the LAB strain from which the postbiotics is prepared, the type of target microorganism or contaminant, concentration and form of application, and the food matrix features. Postbiotics indicate many antimicrobial activities both against pathogenic microorganisms and on spoilage microorganisms in vitro conditions(Moradi et al., 2020).

Cell-free preparations devired from the metabolic products of different beneficial bacteria have been offered with potential pharmaceutical applications in the prevention or treatment of diseases (Klein et al., 2013). For example, Colibiogen® (Laves-Arzneimittel GmbH, Schötz, Switzerland) a commercially protein-free filtrate from cultures of Escherichia coli (strain Laves 1931), including amino acids, peptides, polysaccharides and fatty acids, has been indicated to be effective in inhibiting in vitro both antibiotic resistant and susceptible Salmonella isolates (Zihler et al., 2009).

Some postbiotics have been deliberately implemented certain foods. For instance, cell free supernatant from probiotic *L. plantarum* YML 007 strain having biopreservative effect on soybeans obtained that developed shel life of unshelled soybeans until 2 months (Rather et al., 2013). Another study observed that the direct addition of bifidin bacteriocin from *Bifidobacterium lactis* Bb-12 advanced the shelf life of ground meat until 3 months at 4 ◦C (Mokhtar et al., 2016). It has been observed that postbiotics from *Lactobacillus rhamnosus* EMCC 1105 at 100 mg/g concentration annihilated the *Clostridium perfringens* on chicken mince on the 4th day of storage at 6 ◦C (Hamad et al., 2020), and also lyophilized postbiotics obtained from *Leuconostoc* spp. reduced the growth of background microorganisms, and E coli O157:H7 on the ground meat (Koo et al., 2015). Exopolysaccharide got from *L. rhamnosus* indicated that 8.2% raise in Cheddar cheese yield with *L. lactis* (Torino et al., 2015). Postbiotics can be helpful as microbial-free food supplements, fermented functional foods, and prophylactic drugs, as complementary treatment for several diseases (Chaluvadi et al., 2016).

CONCLUSION

The postbiotics concept refers to non-viable microorganisms and bacterial-free extracts that can ensure advantages to the host by offering additional bioactivities to probiotics. The results from studies indicate that postbiotics exhibit several bioactivities such as anti-inflammatory, immunomodulatory, anti-proliferative, antioxidant, and antimicrobial. In this regard, adding of postbiotics in foods can be a well alternative with high therapeutic value and rised shelf-life of food products. Despite the scientific evidence present, the mechanisms of action and the signaling pathways involved have not yet been fully explained, therefore supplementary in-depth investigations are requested.

REFERENCES

Aguilar-Toalá JE, Garcia-Varela R, Garcia HS, Mata-Haro V, González-Córdova AF, Vallejo-Cordoba B, Hernández-Mendoza A 2018. Postbiotics: An evolving term within the functional foods field. Trends in food science & technology, 75: 105-114.

Argyri AA, Zoumpopoulou G, Karatzas KAG, Tsakalidou E, Nychas GJE, Panagou EZ, Tassou CC 2013. Selection of potential probiotic lactic acid bacteria from fermented olives by in vitro tests. Food Microbiology, 33 (2): 282-291.

Barros CP, Guimaraes JT, Esmerino EA, Duarte MCK, Silva MC, Silva R, ... Cruz AG 2020. Paraprobiotics and postbiotics: Concepts and potential applications in dairy products. Current opinion in food science, 32: 1-8.

Bourebaba Y, Marycz K, Mularczyk M, Bourebaba L 2022. Postbiotics as potential new therapeutic agents for metabolic disorders management. Biomedicine & Pharmacotherapy, 153: 113138.

Canocini A, Siret C, Oellegrino E, Pontier-Bres R, Pouyet L, Montero MP, et al. 2011. *Saccharomyces boulardii* improves intestinal cell restitution through activation of the α2β1 integrin collagen receptor. PLoS One, 6(3): e18427.

Chaluvadi S, Hotchkiss AT, Yam KL 2016. Gut microbiota: Impact of probiotics, prebiotics, synbiotics, pharmabiotics, a postbiotics on human health. In R. R. Watson, & V. R. Preedy (Eds.). Probiotics, prebiotics and synbiotics. Bioactive foods in health promotion. London: Elsevier, pp. 515–523.

Compare D, Rocco A, Coccoli P, Angrisani D, Sgamato C, Iovine B, et al. 2017. *Lactobacillus casei* DG and its postbiotic reduce the inflammatory mucosal response: An ex vivo organ culture model of post-infectious irritable bowel syndrome. BMC Gastroenterology, 17(53).

Cuevas-González PF, Liceaga AM, Aguilar-Toalá JE 2020. Postbiotics and paraprobiotics: From concepts to applications. Food research international, 136: 109502.

De Marco S, Sichetti M, Muradyan D, Piccioni M, Traina G, Pagiotti R, Pietrella D 2018. Probiotic cell-free supernatants exhibited anti-inflammatory and antioxidant activity on human gut epithelial cells and macrophages stimulated with LPS. Evidence-Based Complementary and Alternative Medicine, 2018.

den Besten G, van Eunenm K, Groen AK, Venema K, Reijngoud DJ, Bakker BM 2013. The role of short-chain fatty acids in the interplay between diet, gut microbiota, and host energy metabolism. Journal of Lipid Research, 54: 2325–2340.

Escamilla J, Lane MA, Maitin V 2012. Cell-free supernatants from probiotic *Lactobacillus casei* and *Lactobacillus rhamnosus* GG decrease colon cancer cell invasion in vitro. Nutrition and cancer, 64(6): 871-878.

Food and Agriculture Organization/World Health Organization (FAO/WHO). 2001. Health and nutritional properties of probiotics in food including powder milk with live lactic acid bacteria, report of a joint (FAO/WHO) expert consultation on evaluation of health and nutritional properties of probiotics in food including powed milk with live lactic acid bacteria. Cordoba, Argentina. Available at: http://www.who.int/ Foodsafety/plubications/fs_management/en/probiotics.pdf [01.09.2023]

Gabriele H 2016. Requirements for a successful future of probiotics. In P. Foerst, & C. Santivarangkna (Eds.). Advances in probiotic technology. New York: Taylor & Francis-CRC Press, pp. 139–145.

Hamad GM, Abdelmotilib NM, Darwish AMG, Zeitoun AM 2020. Commercial probiotic cell-free supernatants for inhibition of Clostridium perfringens poultry meat infection in Egypt. Anaerobe, 62: 102181.

Kareem KY, Ling FH, Chwen LT, Foong OM, Asmara SA 2014. Inhibitory activity of postbiotic produced by strains of *Lactobacillus plantarum* using reconstituted media supplemented with inulin. Gut Pathogens, 6(23).

Kechagia M, Basoulis D, Konstantopoulou S, Dimitriadi D, Gyftopoulou K, Skarmoutsou N Fakiri EM 2013. Health Benefits of Probiotics: A Review. ISRN Nutrition, 2013: 1-7.

Klein G, Schanstra JP, Hoffmann J, Mischak H, Siwy J, Zimmerman K 2013. Proteomics as a quality control tool of pharmaceutical probiotic bacterial lysate products. PLoS One, 8(6): e66682.

Koo OK, Kim SM, Kang SH 2015. Antimicrobial potential of *Leuconostoc* species against *E. coli* O157:H7 in ground meat. Journal of the Korean Society for Applied Biological Chemistry, 58(6): 831–838.

Lin MY, Chang FJ 2000. Antioxidative effect of intestinal bacteria *Bifidobacterium longum* ATCC 15708 and *Lactobacillus acidophilus* ATCC 4356. Digestive diseases and sciences, 45: 1617-1622.

Metchnikoff II 1907. The prolongation of life: Optimistic studies. Springer Publishing Company.

Mokhtar M, Mostafa GA, Eldeeb GS, Taha RA 2016. Effect of bacteriocins (from B*ifidobacterium* spp.) on prevalence of some *Aeromonas* and *Pseudomonas* species in minced meat during cold storage. Journal of Food and Nutritional Disorders, 5(1).

Moradi M, Kousheh SA, Almasi H, Alizadeh A, Guimarães JT, Yılmaz N, Lotfi A 2020. Postbiotics produced by lactic acid bacteria: The next frontier in food safety. Comprehensive reviews in food science and food safety, 19(6): 3390-3415.

Patel RM, Denning PW 2013. Therapeutic Use of Prebiotics, Probiotics, and Postbiotics to Prevent Necrotizing Enterocolitis, Clinics in Perinatology, 40: 11–25

Rather IA, Seo BJ, Kumar VJR, Choi UH, Lim JH, Park YH 2013. Isolation and characterization of a proteinaceous antifungal compound from *Lactobacillus plantarum* YML007 and its application as a food preservative. Letters in Applied Microbiology, 57: 69-76.

Russell DA, Ross RP, Fitzgerald GF, Stanton C 2011. Metabolic activities and probiotic potential of bifidobacteria. International Journal of Food Microbiology, 149(1): 88-105

Salminen S, Collado MC, Endo A, Hill C, Lebeer S, Quigley EM, ... Vinderola G 2021. The International Scientific Association of Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of postbiotics. Nature Reviews Gastroenterology & Hepatology, 18(9): 649-667.

Sharma M, Shukla G 2016. Metabiotics: one step ahead of probiotics; an insight into mechanisms involved in anticancerous effect in colorectal cancer. Frontiers in microbiology, *7*: 1940.

Shigwedha N, Sichel L, Jia L, Zhang L 2014. Probiotical cell fragments (PCFs) as "novel nutraceutical ingredients". Journal of Biosciences and Medicines, 2: 43–55.

Taverniti V, Guglielmetti S 2011. The immunomodulatory properties of probiotic microorganisms beyond their viability (ghost probiotics: Proposal of paraprobiotic concept). Genes & Nutrition, 6: 261–274.

Tiptiri-Kourpeti A, Spyridopoulou K, Santarmaki V, Aindelis G, Tompoulidou E, Lamprianidou EE, et al. 2016. *Lactobacillus casei* exerts anti-proliferative effects accompanied by apoptotic cell death and up-regulation of TRAIL in colon carcinoma cells. PLoS One, 11(2): e0147960

Torino MI, de Valdez GF, Mozzi F 2015. Biopolymers from lactic acid bacteria. Novel applications in foods and beverages. Frontiers in Microbiology, 6: 834.

Tsilingiri K, Barbosa T, Penna G, Caprioli F, Sonzogni A, Viale G 2012. Probiotic and postbiotic activity in health and disease: Comparison on a novel polarised ex-vivo organ culture model. Gut, 61: 1007–1015

Venema K 2013. Foreword. Beneficial Microbes, 4(1): 1–2.

Vinderola G, Sanders ME, Salminen S 2022. The concept of postbiotics. Foods, 11(8): 1077.

Xu R, Shang N, Li P 2011. In vitro and in vivo antioxidant activity of exopolysaccharide fractions from *Bifidobacterium animalis* RH. Anaerobe, 17(5): 226–231.

Zihler A, Le Blay G, de Wouters T, Lacroix C, Braegger CP, Lehner A, et al. 2009. In vitro inhibition activity of different bacteriocin-producing Escherichia coli against *Salmonella* strains isolated from clinical cases. Letters in Applied Microbiology, 49: 31–38.

DRAFT

ORAL PRESENTATION

Türkiye kıyılarında dağılım gösteren zargana *Belone svetovidovi* (Collette ve Parin, 1970) popülasyonlarının morfolojisi

Dilruba SEYHAN ÖZTÜR* (*ORCID: https://orcid.org/0000-0001-6060-6483*

İzmir Kâtip Çelebi Üniversitesi, Su Ürünleri Fakültesi, İzmir/Türkiye

*Sorumlu yazar e-mail:seyhandilruba@gmail.com

Özet

On cins ve 44 türden oluşan Belonidae familyası üyeleri hem amatör hem de ticari balıkçılık faaliyetlerinde talep gören ekonomik değere sahip türler barındırır. Türkiye deniz balıkları listesinin verildiği son çalışmada ise Türkiye kıyılarında *Belone belone* (Linnaeus, 1761), *Belone svetovidovi* (Collette ve Parin, 1970) ve *Tylosurus acus* (Lacepède, 1803) olmak üzere 3 türün varlığından söz edilmektedir. *B. svetovidovi* türünün ise Türkiye'nin Ege ve Marmara Denizi Kıyıları'nda dağılım gösterdiği bilinmektedir. Ege ve Marmara Denizi'nde toplamda 7 farklı istasyonda gerçekleştirilen örnekleme çalışmalarında Ege Denizi'nden 145, Marmara Denizi'nden 19 birey temin edilmiştir. Vücut bütünlüğünde herhangi bir deformasyon bulunmayan toplam 114 adet *B. svetovidovi* bireyi morfolojik analizlere tabii tutulmuş ve toplamda 32 farklı karakter kullanılarak morfolojik analizler gerçekleştirilmiştir. *B. svetovidovi* bireylerinin 26.1-41.5 cm (ortalama 32.67 cm) standart boya sahip oldukları tespit edilmiştir. Kafa boyu 5.7-12.5 cm (ortalama 10.02 cm) arasında değişirken, göz çapının 0.76-1.12 cm (ortalama 0.93 cm) arasında değiştiği saptanmıştır. Bireylerin meristik karakterlerinde ise, dorsal yüzgeç ışın sayısının 14-19 (ortalama 16.86) arasında değiştiği, anal yüzgeç ışın sayısının 18-25 (ortalama 21.18) arası değişkenlik gösterdiği, pektoral yüzgeç ışın sayısının 11-15 (ortalama 12.48) arasında değiştiği gözlemlenmiştir. Solungaç diken sayısı 31-50 (ortalama 41.42) arası değişirken, maksil diş sayısının 106-312 (ortalama 189.28) ve mandibul diş sayısının ise 105-440 (ortalama 286.68) arasında değişkenlik gösterdiği saptanmıştır. Hesaplanan Bray Curtis benzerlik değerlerine göre en yüksek benzerlik Orta Ege (OE) ve Güney Ege (GE) (%99.4), en düşük benzerlik oranının ise Kuzey Ege (KE) ve Marmara (MD) popülasyonları (%98.6) arasında olduğu hesaplanmıştır. Yapılan SIMPER analizine göre popülasyonlar arasındaki arasında en yüksek farklılık Marmara Denizi ve Kuzey Ege Denizi popülasyonları arasında (%1.40) en düşük farklılık değerinin ise Orta Ege ve Güney Ege Denizi popülasyonları arasında (%0.52) bulunmuştur. Buna ek olarak popülasyonlar arasında farklılığa sebep olan karakterlerin başında "İnterorbital/HL; alt çene diş sayısı/mandibul uzunluğu; kafa derinliği/HL" gibi genelde baş ile ilgili oranların yer aldığı tespit edilmiştir.

Anahtar Kelimeler: Belonidae, Morphology, *Belone svetovidovi*, Akdeniz, Ege Denizi, Marmara Denizi.

Abstract

Members of the Belonidae family, consisting of ten genera and 44 species, contain species of economic value that are in demand in both amateur and commercial fishing activities. In the last study, which lists the marine fishes of Turkey, it is mentioned that there are three species distributed in Türkiye coasts: *Belone belone* (Linnaeus, 1761), *Belone svetovidovi* (Collette and Parin, 1970) and *Tylosurus acus* (Lacepède, 1803). It is known that *B. svetovidovi* is distributed in the Aegean and Marmara Sea coasts of Türkiye. Sampling studies carried out in 7 different stations in the Aegean and Marmara Seas, 145 individuals from the Aegean Sea and 19 individuals from the Marmara Sea were obtained. A total of 114 *B. svetovidovi* individuals without any deformation in their body integrity were subjected to morphological analysis, and morphological analyses were carried out using a total of 32 different characters. *B. svetovidovi* individuals were found to have a standard length of 26.1-41.5 cm (average 32.67 cm). While head length varied between 5.7-12.5 cm (average 10.02 cm), eye diameter varied between 0.76-1.12 cm (average 0.93 cm). In the meristic characters of individuals, the number of dorsal fin rays varied between 14-19 (average 16.86), the number of anal fin rays varied between

DRAFT

18-25 (average 21.18), the number of pectoral fin rays varied between 11-15 (average 12.48). Gill rakers varied between 31-50 (average 41.42), while the number of maxillary teeth varied between 106-312 (average 189.28) and the number of mandibular teeth varied between 105-440 (average 286.68). According to Bray Curtis similarity values, the highest similarity rate is between the Central Aegean (OE) and Southern Aegean (GE) (99.4%), and the lowest similarity rate is between the Northern Aegean (KE) and Marmara (M) populations (98.6%). According to the SIMPER analysis, the highest difference between populations was found between the Marmara Sea and North Aegean Sea populations (1.40%), and the lowest difference value was found between the Central Aegean and South Aegean Sea populations (0.52%). In addition, the characters that cause differences between populations are "Interorbital/HL; number of mandibular teeth/mandible length; It has been determined that there are generally head-related ratios such as "head depth/HL".

Keywords: Belonidae, Morfoloji, *Belone svetovidovi*, Mediterranean Sea, Aegean Sea, Sea of Marmara

GİRİŞ

On cins ve 44 türden oluşan Belonidae familyası üyeleri (Fricke ve ark., 2023) hem amatör hem de ticari balıkçılık faaliyetlerinde talep gören ekonomik değere sahip türler barındırır. Tropikal ve subtropikal denizlerin genellikle sahil bölgelerinde epipelajik ve neritik olarak yaşamlarına devam eden *Belone* cinsine ait türlerin hepsinde ovipar üreme görülür ve yumurtaları demersaldir (Akşıray, 1987; Collette, 2016). Yumurtalarının üzerinde bulunan ipliksi ve yapışkan uzantılar yumurtanın bitkilere ve sert yapılara tutunmasını sağlar (Yüce, 1975; Collette, 2016) Zarganalar uzun mesafe göçleri yapmazlar ancak kışın derin sularda geçirdikleri, ilkbahar aylarından itibaren ise yüzey sularına çıktıkları bilinmektedir (Akşıray, 1987).

Ülkemiz sularında "Zargana balığı–Sargana" olarak bilinen ve yayılış gösterdiği denizlerin kıyılarındaki ülkeler için ekonomik değere sahip olan bu türler ülkemizde de balıkçılık ekonomisi açısından oldukça önemlidir. Türkiye deniz balıkları listesinin verildiği son çalışmada ülkemiz kıyılarında *Belone belone* (Linnaeus, 1761), *Belone svetovidovi* (Collette ve Parin, 1970) ve *Tylosurus acus* (Lacepède, 1803) olmak üzere 3 türün varlığından söz edilmektedir (Bilecenoğlu ve ark.,2014). *Belone belone* türünün tüm kıyılarımızda dağılım gösterdiği bilinmekle beraber, *Belone svetovidovi* ve *Tylosurus acus* türlerinin Ege ve Akdeniz kıyılarımızda dağılım gösterdiği bildirilmiştir (Bilecenoğlu ve ark.,2014; Dalyan ve Eryılmaz, 2006) Ancak güncel çalışmalar *Belone svetovidovi* türünün Marmara Denizi'nde de yerleşik popülasyonlara sahip olduğunu göstermektedir (Seyhan Öztürk, 2023).

Collette ve Parin (1970) tarafından Galiçya/Vigo/İspanya kıyılarından örneklenmiş müze örnekleri üzerinden tanımlanan *B. svetovidovi* daha sonrasında Akdeniz'in çeşitli noktalarından rapor edilmiştir (Dorman, 1984; Collette ve Parin, 1986; Golani, 1996; Meriç ve Altun, 1999; Dalyan ve Eryılmaz, 2006). Gerek dış morfolojileri, gerek iç içe geçmiş meristik karakterleri ile sıklıkla *B. belone* ile karıştırılan *B. svetovidovi* hakkında Akdeniz havzasında yapılan çalışmalar daha çok sınırlı morfolojik analizlerin yer aldığı yeni dağılım alanlarını işaret eden çalışmalar olmuştur (Dorman, 1984; Collette ve Parin, 1986; Golani, 1996; Meriç ve Altun, 1999; Dalyan ve Eryılmaz, 2006).

Bu çalışma ile Ege Denizi ve Marmara Denizi'nden örneklenen *B. svetovidovi* popülasyolarının detaylı morfolojik analizlerinin yapılması amaçlanmış ve hakkında oldukça sınırlı çalışma bulunan bu tür ile ilgili literatüre katkı sağlanması hedeflenmiştir.

MATERYAL VE METOT

Örnekleme Çalışmaları

Örnekleme çalışmaları Türkiye'nin Ege ve Marmara Denizi kıyılarında toplamda 7 farklı istasyonda gerçekleştirilmiştir. (Tablo 1). Örneklerin temini istasyon doğrulamasına mahal bırakmayacak şekilde, küçük ölçekli kıyı balıkçılığı faaliyetlerinin gerçekleştirildiği balıkçı barınaklarından ve kooperatiflerden doğrudan (aracı olmaksızın) satın alma yoluyla yapılmıştır

Morfolojik Analizler

Bireylerinin morfoloji analizlerinde Zorica ve Čikeš Keč (2011)' den yararlanılmış olup morfolojik incelemelerin detaylandırılması adına ilave karakter kullanılmıştır. Morfometrik ölçümler ve meristik sayımlar için 32 farklı karakter kullanılmış olup vücut bütünlüğünde herhangi bir deformasyon bulunmayan örnekler çalışılmıştır.

Popülasyonlar arasında benzerlik ve farklılığa neden olan karakterler Primer7 ile belirlenmiştir (Clarke ve Gorley, 2015). Bray-Curtis Benzerlik matrisi uygulanan verilerde MDS (non-Metric Multidimensional Scaling – Metrik Olmayan Çok Yönlü Ölçümleme) ve SIMPER (Similarity Percentages –Benzerlik Yüzdesi) yöntemleri kullanılmıştır. Popülasyonları gruplandırmak içinse bir kümeleme yöntemi olan UPGMA (Aritmetik Ortalamalı Ağırlıksız Çift Grup Yöntemi) uygulanmıştır

Tablo 1. *Belone svetovidovi* bireylerinin örneklendiği istasyonlar:

Örnekleme Bölgesi	İstasyonlar	Örnek sayısı
Marmara Denizi	Boğaz/BGZ – Gemlik/GML	19
Kuzey Ege	Çanakkale/CNK – Altınoluk/ALT	35
Orta Ege	İzmir/IZM	73
Güney Ege	Bodrum/BDR – Didim/DM	37

BULGULAR ve TARTIŞMA

Türkiye'nin Ege ve Marmara kıyılarında ki 7 farklı istasyondan örneklenen toplam 114 adet *B. svetovidovi* bireyi morfolojik analizlere tabii tutulmuştur. Toplamda 32 farklı karakter kullanılarak gerçekleştirilen morfolojik analizlerde *B. svetovidovi* bireylerinin 26.1-41.5 cm (ortalama 32.67 cm) standart boya sahip oldukları tespit edilmiştir. Kafa boyu 5.7-12.5 cm (ortalama 10.02 cm) arasında değişirken, göz çapının 0.76-1.12 cm (ortalama 0.93 cm) arasında değiştiği saptanmıştır.

Bireylerin meristik karakterlerinde ise, dorsal yüzgeç ışın sayısının 14-19 (otalama 16.86) arasında değiştiği, anal yüzgeç ışın sayısının 18-25 (ortalama 21.18) arası değişkenlik gösterdiği, pektoral yüzgeç ışın sayısının 11-15 (ortalama 12.48) arasında değiştiği gözlemlenmiştir. Solungaç diken sayısı 31-50 (ortalama 41.42) arası değişirken, maksil diş sayısı 106-312 (ortalama 189.28) ve mandibul diş sayısının ise 105- 440 (ortalama 286.68) arasında değişkenlik gösterdiği saptanmıştır. Tüm meristik karakterlerin istasyonlarımıza göre dağılımı ise Tablo 2'de verilmiştir. Detaylı morfometrik değerlerler ise Tablo 3de sunulmuştur.

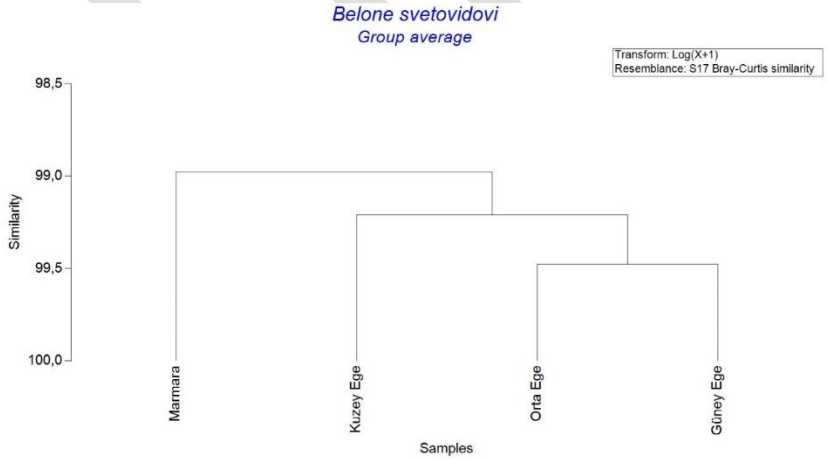
MERİSTİK ORTALAMALAR	Güney ege			Kuzey Ege			Orta Ege			Marmara			Genel Ortalama		
Belone svetovidovi	**Değişim aralığı (min-mak)**	**Ortalama**	**std**	**Değişim aralığı (min-mak)**	**Ortalama**	**std**	**Değişim aralığı (min-mak)**	**ortalama**	**std**	**Değişim aralığı (min-mak)**	**ortalama**	**std**	**Değişim aralığı (min-mak)**	**ort**	**std**
Dorsal Y. ışın sayısı	16-19	17,43	0,87	15-19	16,68	1,17	14-19	16,70	1,29	15-19	16,64	1,21	14-19	16,86	0,4
Anal Y.ışın sayısı	24-18	21,14	1,56	20-23	21,41	1,14	18-25	20,73	1,61	20-23	21,45	1,04	18-25	21,18	0,3
PektoralY. ışın sayısı	12-15	13,05	0,86	11-14	12,32	0,89	11-15	12,28	1,67	11-13	12,27	0,79	11-15	12,48	0,4
Pelvik Y. ışın sayısı	6-7	6,10	0,30	6	6,00	0,00	6-7	6,03	0,18	6-7	6,09	0,30	6-7	6,05	0,0
Solungaç dikeni sayısı	34-48	42,48	3,59	36-47	41,73	3,81	25-50	42,57	4,76	33-44	38,73	3,69	31-50	41,42	1,8
Üst çene diş sayısı	138-248	203,52	30,99	106-261	167,77	40,22	130-312	194,90	40,59	179-211	190,91	14,71	106-312	189,28	15,3
Alt çene diş sayısı	150-378	287,95	60,83	115-436	261,32	90,69	105-440	298,80	79,56	220-390	298,64	46,30	105-440	286,68	17,7

Tablo 2. Meristik karakterlerin istasyonlara göre dağılımı

Tablo 3. *Belone svetovidovi* türüne ait morfometrik oranlar

	Belone svetovidovi							
	MARMARA		**EGE DENİZİ**					
			Güney		Orta		Kuzey	
Birey sayısı (n)	11		21		60		22	
	ort	std ±	ort	std ±	ort	std ±	ort	std ±
HL/SL	29.5	2.1	29.3	1	28.6	1.8	27.6	3.6
Predorsal/SL	80.5	1.3	81.3	1.7	80.9	1.3	80.6	5.4
Prepelvik/SL	66.8	1.5	68.1	4	66.8	1.6	65.8	3.6
Preanal/SL	78.4	1	78.7	0.6	78.6	1.3	77.8	3.4
Prepektoral/SL	32.2	2.5	32.2	2.8	31.8	2.6	31	5
Pelvik-anal/SL	11.5	0.7	11.5	0.4	11.9	1	12	0.8
Pektoral Y. uzunluğu/SL	5.8	0.4	5.3	0.3	5.8	0.4	5.7	0.4
Pelvik Y. uzunluğu/SL	4.3	0.3	4	0.2	4.5	0.6	4.4	0.4
Kaudal Y. uzunluğu/SL	10	0.4	9.4	1	10.2	0.6	10.1	0.9
Kuyruk sapı yüksekliği/SL	2	0.1	1.7	0.1	1.8	0.2	2	0.4
Dorsal Y. kaidesinden vücut yüksekliği/SL	5.4	0.4	5.5	0.3	5.5	0.5	5.2	0.6
Pelvik yüzgeç kaidesinden vücut yüksekliği /SL	5.5	0.4	5.4	0.3	5.5	0.7	5.2	0.5
Anal Y. kaidesinden vücut yüksekliği /SL	5.4	0.4	5.3	0.3	5.4	0.5	5.1	0.5
Dorsal Y. taban uzunluğu/SL	12.8	0.8	12.1	0.5	12.1	0.7	12.3	1.1
Anal Y. taban uzunluğu/SL	14.9	0.6	14.6	0.7	14.8	0.9	15.3	1.5
Kafa yüksekliği/HL	14.9	1.6	13.6	0.8	13.7	2.2	12.6	2.9
Pre orbital/HL	66.7	15	71.1	0.9	69.8	5.8	65.9	7.7
Post orbital/HL	22.7	2.8	20.4	0.6	21.5	2.9	23.2	5.3
Göz çapı/HL	9.7	1.1	8.6	0.4	9.4	1	10.5	2.5
Gözler arası mesafe/HL	7.9	1.1	7.2	0.4	8.1	0.9	9.9	2.5
Maksil diş sayısı/maksil uzunluğu	35.4	2.7	31.2	4.9	34.5	6.8	37.5	5.3
Mandibul diş sayısı/mandibul uzunluğu	50.6	7.8	39.2	8.1	47	12.7	50.7	10.8
Maksil diş sayısı/göz çapı	212.3	24.1	225.9	38.3	202.1	41.7	188.8	45.8
Mandibul diş sayısı/göz çapı	333.5	63	320.1	71.2	309.5	82.2	295.7	107
Toplam diş sayısı/ SL	15.9	2.7	15	2.9	14.7	3.4	14.4	4

Hesaplanan Bray Curtis benzerlik değerlerine göre en yüksek benzerlik Orta Ege (OE) ve Güney Ege (GE) (%99.4), en düşük benzerlik oranının ise Kuzey Ege (KE) ve Marmara (M) popülasyonları (%98.6) arasında olduğu hesaplanmıştır (Şekil 1 ve Tablo 4).



Şekil 1. *B. svetovidovi* popülasyonlarının morfolojik benzerliklerinin Bray-Curtis benzerlik dendogramı

Tablo 4. *B. svetovidovi* popülasyonları arası Bray-Curtis benzerlik değerleri

	Marmara	Kuzey Ege	Orta Ege
Marmara			
Kuzey Ege	98,601		
Orta Ege	99,266	99,233	
Güney Ege	99,066	99,186	99,478

Yapılan SIMPER analizine göre popülasyonlar arasındaki arasında en yüksek farklılık Marmara Denizi ve Kuzey Ege Denizi popülasyonları arasında (%1.40) en düşük farklılık ise Orta Ege ve Güney Ege Denizi popülasyonları arasında (%0.52) bulunmuştur (Şekil 2).

Groups MD & KE
Average dissimilarity = 1,40

Species	Group MM Av.Abund	Group KE Av.Abund	Av.Diss	Contrib%	Cum.%
İnterorbital/HL	2,10	2,39	0,19	13,38	13,38
Lower jaw teeth/mandibul	3,69	3,95	0,17	11,83	25,21
Eye diamet/HL	2.26	2.44	0.12	8.49	33.70

Groups MD & GE
Average dissimilarity = 0,93

Species	Group MM Av.Abund	Group GE Av.Abund	Av.Diss	Contrib%	Cum.%
Lower jaw teeth/mandibul	3,69	3,94	0,16	17,54	17,54
upper jaw teeth/maxil	3,47	3,59	0,08	8,61	26,15

Groups MD & OE
Average dissimilarity = 0,73

Species	Group MM Av.Abund	Group OE Av.Abund	Av.Diss	Contrib%	Cum.%
Lower jaw teeth/mandibul	3,69	3,87	0,12	15,89	15,89
up jaw teeth/eye diameter	5,42	5,31	0,07	9,93	25,82
İnterorbital/HL	2,10	2,21	0,07	9,33	35,16

Groups KE & GE
Average dissimilarity = 0,81

Species	Group KE Av.Abund	Group GE Av.Abund	Av.Diss	Contrib%	Cum.%
İnterorbital/HL	2,39	2,19	0,13	16,26	16,26
Head depth/HL	2,61	2,77	0,10	12,53	28,79
lower jaw teeth/eye diameter	5,69	5,81	0,08	9,62	38,41

Groups KE & OE
Average dissimilarity = 0,77

Species	Group KE Av.Abund	Group OE Av.Abund	Av.Diss	Contrib%	Cum.%
İnterorbital/HL	2,39	2,21	0,12	15,41	15,41
Eye diamet/HL	2,44	2,34	0,07	8,58	23,99
upper jaw teeth/maxil	3.65	3.57	0.05	6.92	30.91

Groups OE & GE
Average dissimilarity = 0,52

Species	Group OE Av.Abund	Group GE Av.Abund	Av.Diss	Contrib%	Cum.%
Head depth/HL	2,69	2,77	0,05	9,82	9,82
lower jaw teeth/eye diameter	5,74	5,81	0,05	9,32	19,13
total teeth number/ SL	2,75	2,83	0,05	9,22	28,35

Şekil 2. Popülasyonlar arasında morfolojik farklılığa neden olan karakterler

Tür ile ilgili gerçekleştirilen morfoloji çalışmaları incelendiğinde sadece 2 çalışma dikkat çekmekte ve bu çalışmaların özellikle 5 adet meristik karakter üzerine yoğunlaştığı görülmüştür (Meriç ve Altun, 1999; Dorman, 1987). Kullanılan birey sayısının az olduğu çalışmalardan elde edilen sonuçlar ile bu çalışmadan elde edilen sonuçlarla benzerlik göstermekle beraber maksimum ve minumum sınırlarda farklılıklar saptanmıştır (Tablo 5). Çalışmada incelenen popülasyonların Bray Curtis benzerlik değerleri Kuzey Ege- Marmara popülasyonu hariç popülasyonların %99 üzeri benzerlik gösterdiğini ortaya koymuştur. Dolayısıyla morfolojik olarak farklı bir popülasyon yapısı gözlenmemiştir. SIMPER analizine göre ise saptanan en yüksek farklılık ise Marmara Denizi ve Kuzey Ege Denizi popülasyonları arasında (%1.40) tespit edilmiştir. Buna ek olarak, popülasyonlar arasında farklılığa sebep olan karakterlerin başında "İnterorbital/HL; Alt çene diş sayısı/mandibul uzunluğu; Head depth/HL" gibi genelde baş ile ilgili oranların yer aldığı tespit edilmiştir. Tekerrürlü gerçekleştirilen örnekleme çalışmalarında *B. svetovidovi* türünün popülasyon yoğunluğunun Orta Ege Denizi'nde diğer örnekleme bölgelerine göre daha yoğun olduğu, bu yoğunluğun ise güneye doğru azaldığı gözlenmiştir. Güney Ege ve Levantin kıyılarımızda yapılan tekrarlı örnekleme çalışmalarında *B. svetovidovi* türüne sadece Didim'de ulaşılabilmiştir.

Tablo 5. *Belone svetovidovi* ile gerçekleştirilen morfoloji çalışmaları

	Dorman, 1987	Meriç ve Altun, 1999	Bu çalışma
Solungaç Diken Sayısı	41-53 (ort. 48.7) N=25	39-53 (Ort 45.20) N=54	31-50 (Ort. 41.42) N=114
Dorsal Yüzgeç Işın sayısı	16-18 (Ort 17.06) N=30	15-19 (Ort. 16.84) N=54	14-19 (Ort 16.87) N=114
Anal Yüzgeç Işın Sayısı	20-23 (Ort 21.6) N=30	19-23 (Ort. 23.39) N=54	18-25 (Ort. 21.18) N=114
Pektoral Yüzgeç Işın Sayısı	11-13 (Ort. 12) N=30	11-13 (Ort. 12.11) N=54	11-15 (Ort. 12.48) N=114

Collette ve Parin (1970) tarafından Galiçya/Vigo/İspanya kıyılarından örneklenmiş müze örnekleri üzerinden tanımlanan *B. svetovidovi* türünün Doğu Akdeniz'in kuzey enlemlerindeki soğuk sularda daha yoğun dağılım gösterdiği dikkat çekmektedir (Froese ve Pauly, 2023). Levantin kıyılarımızdaki su sıcaklığının diğer denizlerimize nazaran daha fazla olmasının, bu türün güneydeki dağılımını sınırlayan faktör olabileceğini düşündürmektedir. Bununla beraber, besin tuzları bakımından oldukça oligotrof yapıda olan Levantin Denizi'ndeki yüksek sıcaklık değerleri (Salihoğlu ve ark., 1990) demersal yumurtalayan bu türlerin yumurtlama substratları olabilen deniz çayırlarının da dağılımını sınırlamaktadır (Çelebi, 2007). İstilacı türlerin bölgedeki varlığı ve yerli türlerle olan rekabet ilişkileri gibi uzun dönemli ekolojik değişikliklerin türün güneydeki dağılımını sınırlandıran diğer faktörler olabileceği değerlendirilmektedir.

SONUÇ

Gerçekleştirilen çalışmada *B. svetovidovi* türünün kıyılarımızdaki popülasyon yoğunluğunun Orta Ege Denizi'nde diğer örnekleme bölgelerine göre daha yoğun olduğu, bu yoğunluğun ise güneye doğru azaldığı gözlenmiştir. Levantin kıyılarımızdaki su sıcaklığının diğer denizlerimize nazaran daha fazla olmasının, bu türün güneydeki dağılımını etkileyen faktör olabileceği ve Levantin Denizi'ndeki yüksek sıcaklık değerlerinin demersal yumurtalayan bu türlerin yumurtlama substratları olabilen deniz çayırlarının da dağılımını da etkileyerek türün güneydeki dağılımını sınırladığı kantine varılmıştır.

Balıkçılık kaynaklarının sürdürülebilirliğinin ekosistem temelli yaklaşımlarla sağlanabileceği, yalnızca biyolojik çeşitliliğin korunmasının yeterli olmadığının anlaşıldığı, mevcut tür ve popülasyonların genetik varyasyon düzeyinin ve popülasyonların değişim süreçlerinin bilinmesinin önemli olduğunun farkına varılmıştır.

Gerçekleştirilen bu çalışmada, Ege ve Marmara denizlerindeki *Belone svetovidovi* türüne ait popülasyonların detaylı morfometrik ve meristik analizleri yapılmış ve literatüre katkıda bulunulmuştur.

TEŞEKKÜR

Bu çalışma TÜBİTAK 121 Y 553 No'lu proje tarafından desteklenmiştir.

KAYNAKLAR

Akşıray F. 1987. Türkiye Deniz Balıkları ve Tayin Anahtarı. İstanbul: İstanbul Üniversitesi Rektörlüğü Yayınları.

Clarke KR ve Gorley RN. 2015. Getting Started with PRIMER v7 Plymouth Routines In Multivariate Ecological Research. Revista Mexicana de Biodiversidad, 89(3): 898-909.

Collette BB, Parin NV. 1970. "Needlefishes (Belonidae) of the Eastern Atlantic Ocean". Atl. Rep., 11,1–60.

Collette BB, Parin NV. 1986. Fishes of the North-eastern Atlantic and the Mediterranean. Vol. II, UNESCO Editörler: Whitehead, P.J.P., M.-L. Bauchot, J.-C. Hureau, J. G. Nielsen and E. Tortonese.

Collette BB. 2016. Order Beloniformes. Belonidae (Pp. 2118-2126). Scomberesocidae (Pp. 2127-2130). Hemiramphidae (Pp: 2156-2162). The living marine resources of the Eastern Central Atlantic. v.3.

Çelebi B. 2007. A study on *Posidonia oceanica* (L.) Delile, 1813 Seagrass Meadows in the Levant Sea. Yüksek Lisans Tezi. M.E.T.U. (İngilizce).

Dalyan C, Eryılmaz L. 2006. Two new fish records from Turkish coast of the eastern Mediterranean: the garfish, *Belone svetovidovi* Collette and Parin, 1970; the spiny gurnard, *Lepidotrigla dieuzeidei* Audoin in Blanc and Hureau, 1973. J.Black Sea Med Env., 12:155–158.

Dorman JA. 1984. *Belone svetovidovi* (Atheriniformes, Belonidae): A Species of Garfish New to Ireland, with Records of Other Unusual Fish from Courtmacsherry Bay. The Irish Naturalists' Journal, 21(8): 356-357.

Dorman JA. 1987. *Belone svetovidovi* Collette and Parin: a species of garfish new to northern Europe. Journal of the Marine Biological Association of the United Kingdom, 67(3): 679-685.

Fricke R, Eschmeyer, WN, van der Laan R. (eds) 2023. Eschmeyer's Catalog Of Fıshes: Genera, Specıes, References. Available at:(http://researcharchive.calacademy.org/research/ichthyology/catalog/fishcatmain.aspElectronic version [03/2023]

Froese R, Pauly D. Editors. 2023. FishBase. *World Wide Web electronic publication*. Available at: www.fishbase.org, [03/2023]

Golani D. 1996. The marine ichthyofauna of the Eastern Levant—history, inventory, and characterization. Israel Journal of Ecology and Evolution, 42(1): 15-55.

Meriç N, Altun Ö. 1999. The Garfish, *Belone svetovidovi* Collette and Parin, 1970, new to the Aegean Sea. Israel Journal of Zoology, 45: 423-426

Meriç N, Altun Ö. 1999. The Garfish, *Belone svetovidovi* Collette and Parin, 1970, new to the Aegean Sea. Israel Journal of Zoology, 45: 423-426.

Salihoğlu I, Saydam C, Baştürk 0, Yilmaz K, Göçmen D, Hatipoğlu E, Yilmaz A. 1990. Transport and distribution of nutrients and chlorophyll-a by meso-scale eddies in the northeastern Mediterranean. Marine Chemistry, 29: 375-39

Seyhan Öztürk D. 2023. Molecular and Phenotypic Characteristics of Short-Beaked Garfish *Belone svetovidovi* Collette and Parin, 1970 in a New Location, the Sea of Marmara. Marine Science and Technology Bulletin, 12 (2): 156-161 . DOI: 10.33714/masteb.1254345

Yüce Y. 1975. Zargana balığı *Belone belone* (L)'nın Biyolojisi. Istanbul University, Hidrobiology Institue, (2): 1-25.

Zorica B, Čikeš Keč V. 2011. Phenotypic characteristics of garfish *Belone belone* (Linnaeus, 1761) in the Adriatic Sea. Acta Adriatica, 52(2): 269 – 278.

DRAFT

ORAL PRESENTATION

Effect of dopamine on the physiological and molecular responses of tomato seedlings under drought and salt stress

Ufuk Çelikkol Akçay (*ORCID: https://orcid.org/0000-0003-1260-3813*)

Isparta University of Applied Sciences, Faculty of Agriculture, Department of Agricultural Biotechnology, Isparta, TURKEY

Corresponding author e-mail: ufukakcay@isparta.edu.tr

Abstract

Drought, heath and salt stresses are the most common abiotic stresses resulting in yield reduction or complete loss of agricultural production in recent years. This study investigated the effects of the biostimulator molecule dopamine in tomato seedlings under drought and salt stress, by examining various morphological and physiological parameters, as well as various enzymes that play a role in the antioxidative defense system. The results showed that dopamine alleviated drought stress mainly by increasing superoxide dismutase and catalase antioxidative enzyme gene expression, decreasing ethylene production and improving cell membrane and tissue integrity in tomato seedlings. Although responses of the seedlings were mostly similar, unlike drought stress, the defense mechanisms excluded antioxidative defense enzyme gene expression under salt stress.

Keywords: *Solanum lycopersicum* L., abiotic stress, dopamine

INTRODUCTION

According to the latest FAO agricultural production statistics (2020), tomato (*Solanum lycopersicum* L.) is the most widely produced vegetable crop worldwide. In addition to its high economic value and health benefits, tomato is a model plant for fleshy fruit research. The high water requirement of tomato makes it very vulnerable to both drought and soil salinity (Battilani et al., 2012).

In recent years, unexpected droughts and extreme heat conditions have become very common throughout the world due to climate change (Zomer et al., 2017). High salt concentrations in the soil also restrict agricultural production due to intensive usage of synthetic fertilizers and use of irrigation systems depending on groundwater, which contains high amounts of dissolved salts (Haj-Amor et al., 2022). The common denominators of both stresses are osmotic and oxidative stress resulting from stomatal closure, decreased photosynthesis efficiency and accumulation of reactive oxygen and nitrogen species (Abdelaal et al., 2022).

In recent years, the stress mitigation potential of various biostimulator compounds has been investigated. Among them, a promising substance is a biogenic amine or catecholamine dopamine. It is biosynthesized from tyrosine and is a very common active molecule in both plants and animals. Although the roles of dopamine in animals as a neurotransmitter regulating metabolism, hormone secretion, and control of movement, reward and addiction processes are widely studied, its roles in plant tissues are largely unknown (Howe and Dombeck, 2016). The recognition of the stress-mitigating effects of dopamine is a promising new field of study. A recent study reported increased levels of dopamine in apple leaves under drought and salt stress (Zhang et al., 2022). It has also been shown that, exogenous dopamine application alleviated stress symptoms in plants under drought, salt, nutrient deficiency and pathogen infection. Commonly observed effects of dopamine under stress conditions include increased photosynthetic rates, higher water use efficiencies and improved antioxidant capacity (Li et al., 2015; Yogendra et al., 2016; Liang et al., 2017; Gao et al., 2020; Lan et al., 2020). Dopamine and its oxidation product melanin also show strong antioxidant capacities similar to those of ascorbic acid or catechin and play a direct role in the scavenging of reactive oxygen species (Rosel et al., 1994; Kanazawa, 2000). This study was performed to investigate the effect of dopamine under drought and salt stress by observing common physiological stress indicators such as morphological responses, membrane damage and osmoprotectants as well as components of the enzymatic antioxidative defense system, including superoxide dismutase (FeSOD), catalase (CAT2), glutathione reductase (GR1), ascorbate peroxidase (APX1), 1-aminocyclopropane-1-carboxylic acid synthase (ACS2) and delta 1-pyrroline-5-carboxylate synthase (P5CS).

DRAFT

DRAFT

MATERIALS AND METHODS

In this study, the Kayra F1 tomato variety (Anamas Tohum Ltd. Şti., Antalya) was used as plant material.

Plant growth and treatments

Tomato seeds were placed in polypropylene containers with a volume of 400 ml containing sterile perlite, as one seed in each container. Germination was achieved in all pots at the end of a week by irrigation with sterile Hoagland solution (Hoagland and Arnon, 1950) every other day. Plant growth was continued for 21 days, in a 16-hour light/8-hour dark cycle, at 24˚C in a plant growth cabinet containing 50% humidity. Drought stress was started at the end of the 21st day, 0 water potential was reached on the 3rd day, and at the end of the 5th day the tissue samples were collected for analysis. Salt stress was initiated with Hoagland solution containing 200 mM NaCl at the end of the 21st day and applied for 5 days as in drought stress.

Dopamine was applied by adding 100 µM dopamine hydrochloride (Dopasel, Haver Farma, Istanbul) into Hoagland solution starting from seed sowing and the application continued throughout stress applications. Dopamine concentration was determined by pre-testing with 10 plants that, received treatments at three different concentrations for each hormone. Root and stem lengths of the plants and ion leakage measurements were evaluated and the concentration with the best anatomical and physiological response was used throughout the study. Salt stress was applied at a concentration of 200 mM NaCl based on the observation that it induced a severe stress response for the same tomato variety within a week in previous laboratory studies.

Determination of physiological plant stress indicators

In the presence or absence of dopamine application and stress, after 21 days of development and 5 days of stress application, all plants were removed from perlite and washed under tap water, and root/stem lengths were determined as cm. Root and stem tissues were separated and weighed, and after drying at 60˚C for 48 h, they were weighed again and their dry weight (g) was determined.

Relative water contents (BSU) were determined according to the formula; BSU (%) = (Wet weight-Dry weight)/(Turgid weight-Dry weight) X 100 as specified in Smart and Bingham (1974). Turgid weight was determined by keeping the leaves in distilled water at room temperature for 24 h.

The levels of osmoprotectant amino acid proline were determined by the method of Bates et al. (1973) without any modifications.

Cellular membrane damage was determined by measuring the phospholipid peroxidation product malondialdehyde (MDA) level according to Ohkawa et al. (1979) without any modifications.

Another indicator of cell wall and membrane damage, ion leakage levels were determined according to Nanjo et al. (1999) without any modifications.

Determination of gene expression

Total RNA isolation was performed according to the instructions of Thermo GeneJET plant mini kit (Thermo, USA), followed by treatment with Thermo RNase-free DNase I (Thermo, USA) in accordance with the instructions for quantitative RT-PCR. The amounts of total RNA obtained were determined spectrophotometrically using Nanodrop 2000. The quality of total RNAs was also determined by separation and imaging with 1% agarose gel electrophoresis followed by cDNA synthesis, which was performed according to Thermo RevertAid FirstStrand cDNA synthesis kit instructions (Thermo, USA).

In this study, *ACS2*, *P5CS*, *FeSOD*, *CAT2*, *GR1* and *APX1* gene expressions in tomato leaves were normalized using the *EF-1* reference gene and the relative quantification of genes was compared with the control group using a Biorad CFX Connect Real-Time PCR device and CFX Maestro software (Biorad, USA). All amplification reactions were performed using Biorad iTaq Universal SYBR Green Supermix as specified in the user manual. The primers used in the study were optimized with PrimerPremier 5.0 software (Premier Biosoft International, USA) to provide optimum amplification conditions. PCR conditions included initial denaturation of 30 s at 95°C, followed by amplification for 5 seconds at 95°C and 30 s at 54°C, repeating 40 cycles. RT-PCR reactions included two biological and two technical replicates. Relative expression of genes was determined according to the $2^{-\Delta\Delta C_T}$ method. $\Delta\Delta C_T$ values were calculated by subtracting the mean ΔC_T values of the samples from the mean ΔC_T values of the controls, and then these values were used to determine the $2^{-\Delta\Delta C_T}$ differences.

DRAFT

Statistical Analysis

The study was conducted in triplicate and each replicate included 20 plants. The data obtained in the research were evaluated using the SPSS 16.0 program. The differences between the means were determined by the one-way Anova and Tukey's test.

RESULTS

Salt and drought stress significantly reduced the length, dry weight and water contents of shoot tissues, whereas dopamine application did not have any significant effect on the morphological parameters except for slight improvements in dry weights (Table 1). Salt and drought stress did not have any significant effect on root length, dry weight or water content, except for 50% reduction in root dry weight under salt stress. Dopamine application improved dry weights under both salt and drought stresses. Dopamine application also significantly increased the root length by 26% under salt treatment.

Table 1. Effect of different stress and hormone treatments on plant morphological parameters. C, T, K, D, D+T and D+K indicate control, salt, drought, dopamine, dopamine+salt and dopamine+drought applications, respectively.

Treatment	Tissue Lenght (cm)	Dry Weight (gr)	Water Content (%)
		SHOOT	
C	27,90 ± 0,56 a	0,29 ± 0,047 a	93,37 ± 0,38 a
T	20,57 ± 0,32 b	0,15 ± 0,021 b	92,14 ± 0,61 a, c
K	21,5 ± 0,98 b	0,23 ± 0,012 c, d	90,75 ± 0,55 b, c
D	27,20 ± 0,10 a	0,31 ± 0,020 a	92,21 ± 0,21 a, c
D+T	22,83 ± 0,68 b	0,17 ± 0,012 b, c	91,83 ± 0,81 a, c
D+K	21,50 ± 1 b	0,26 ± 0,010 a, d	89,96 ± 0,89 b
		ROOT	
C	24,73 ± 2,57 a	0,041 ± 0,007 a, c	93,43 ± 0,91 a
T	23,17 ± 0,12 a	0,022 ± 0,002 b	92,34 ± 0,34 a
K	25,47 ± 1,06 a, b	0,042 ± 0,008 a, c	91,87 ± 0,87 a
D	25,27 ± 1,60 a	0,044 ± 0,007 a, c	92,81 ± 0,70 a
D+T	29,33 ± 1,21 b	0,032 ± 0,002 b, c	92,87 ± 0,88 a
D+K	22,00 ± 0,76 a	0,050 ± 0,006 a	92,59 ± 0,60 a

All physiological stress indicators were measured using tomato leaf tissues (Table 2). Under both abiotic stress, leaf relative water contents decreased and dopamine had no improvement effect. Although stress treatments did not significantly change leaf ion leakage levels, the value was at its minimum under drought stress/dopamine co-treatment, which was significantly lower than the control treatment. MDA contents also increased under abiotic stresses; however, dopamine treatment effectively reduced it back to control levels under salt treatment. The proline contents showed 1925% and 628% increments under salt and drought stresses, respectively. However, dopamine treatments slightly reduced proline levels under both stresses.

Table 2. Effect of different stress and hormone treatments on physiological stress indicators. C, T, K, D, D+T and D+K indicate control, salt, drought, dopamine, dopamine+salt and dopamine+drought applications, respectively.

Treatment	Relative Water Content (%)	Ion Leakage (%)	MDA Content (nmol/g)	Proline Content (nmol/g)
C	85,48 ± 0,40 a	5,02 ± 0,26 a	5,54 ± 0,72 a, b	40,54983 ± 16,07 a
T	69,96 ± 6,42 b	4,97 ± 0,72 a	7,38 ± 0,41 b	810,4811 ± 146,39 b
K	78,91 ± 3,92 a, b	5,07 ± 0,51 a	9,98 ± 0,94 c	291,5808 ± 33,35 c
D	73,55 ± 2,96 b	4,24 ± 0,93 a	5,28 ± 0,46 a	49,48454 ± 16,2 a
D+T	71,05 ± 5,11 b	4,04 0,95 a	4,90 ± 0,28 a	629,5533 ± 139,68 b
D+K	78,83 ± 2,01 a, b	2,98 ± 0,39 b	10,99 ± 1,22 c	241,0653 ± 52,07 a, c

Transcript levels of ACS2, which is the key enzyme in stress hormone ethylene synthesis and P5CS, which is the rate-limiting enzyme in proline synthesis, increased significantly under drought stress and reduced back to control levels upon dopamine application (Figure 1). Both drought and salt stress treatments, as well as dopamine application alone increased *FeSOD* and *CAT2* gene expressions significantly compared to control treatment. However, under drought stress, dopamine application significantly increased *FeSOD* and *CAT2* expressions compared with drought treatments alone. The only treatment that significantly changed *APX1* and *GR1* gene expression was drought. Unlike *FeSOD* and *CAT2* gene expressions, both expressions were reduced back to control levels under dopamine treatment.

DISCUSSION

In this study, no significant effect of dopamine was reported on the morphological parameters of tomato plants, as also supported by the related literature. There are two contradictory studies which have reported root growth inhibition in soybean seedlings and root growth promotion in *Acmella oppositifolia* and *Nicotiana tabacum* by dopamine treatment, indicating plant-specific responses that cannot be generalized (Protacio et al., 1992; Guidotti et al., 2013). Both drought and salt stresses reduced tissue lengths, weights and water contents and shoot tissue was more sensitive to stress treatments than root tissues as also supported by the literature (Li, 2009). In this study, dopamine application slightly improved the morphological parameters under stress treatments. The application also reduced ion leakage and MDA levels as well as proline levels. Limited literature data also support these observations. Wang et al. (2020) reported that overexpression of the tyrosine decarboxylase gene (*MdTyDC*), which is responsible for the synthesis of dopamine derivatives, resulted in higher dopamine levels, increased fresh weight and reduced MDA levels in apple callus under salt stress. Apple *MdTyDC* overexpression lines also exhibited lower ion leakage levels and water loss rates in addition to enhanced photosynthetic performance under drought stress (Gao et al., 2021).

In this study, *ACS2* and *P5CS* gene expressions increased significantly with drought stress and decreased with presence of dopamine under stress. *ACS2* is the rate-limiting enzyme in ethylene synthesis and its expression results in the production of ethylene hormone, which plays a role in aging and senescence followed by necrosis. To the best of our knowledge, there is no report on dopamine application/*ACS2* expression interaction in the literature. The findings of this study showed potential low levels of ethylene under drought stress in the presence of dopamine. *P5CS*, which is responsible for proline production, also exhibited a similar expression pattern and the transcript levels were also in agreement with tissue proline levels, which showed reduction under stress upon dopamine application. Proline levels and P5SC expression results of this study contradict those of Wang et al. (2020, 2021) who reported increased levels with dopamine presence, which possibly shows plant, tissue and/or dose-dependent effects of dopamine.

The results of this study also showed that the transcript levels of *FeSOD* and *CAT2* increased with the presence of dopamine, whereas *APX1* and *GR1* gene expressions decreased with the presence of dopamine under drought stress. SOD and CAT are the two main antioxidative defense enzymes that detoxify superoxide radicals and hydrogen peroxide (H_2O_2), respectively. Catalase enzymes function independently from reducing equivalents, whereas APX1, which is also an H_2O_2 scavenging enzyme, requires a steady supply of NADPH. This requirement may result in the sensitivity of APX1 to severe stress and decreased transcript levels. Similar to our study, Guidotti et al. (2013) and Gomes et al. (2014) also reported increased SOD and decreased or

unaffected POD activity upon dopamine application. Although salt stress upregulated *FeSOD* and *CAT2* gene expressions, dopamine presence did not change the expressions under salt stress. Unlike drought, salt stress also has an ion toxicity component in addition to osmotic stress. Ion toxicity is destructive to antioxidative defense enzymes and the antioxidative potential of dopamine might be insufficient under this additional stress component.

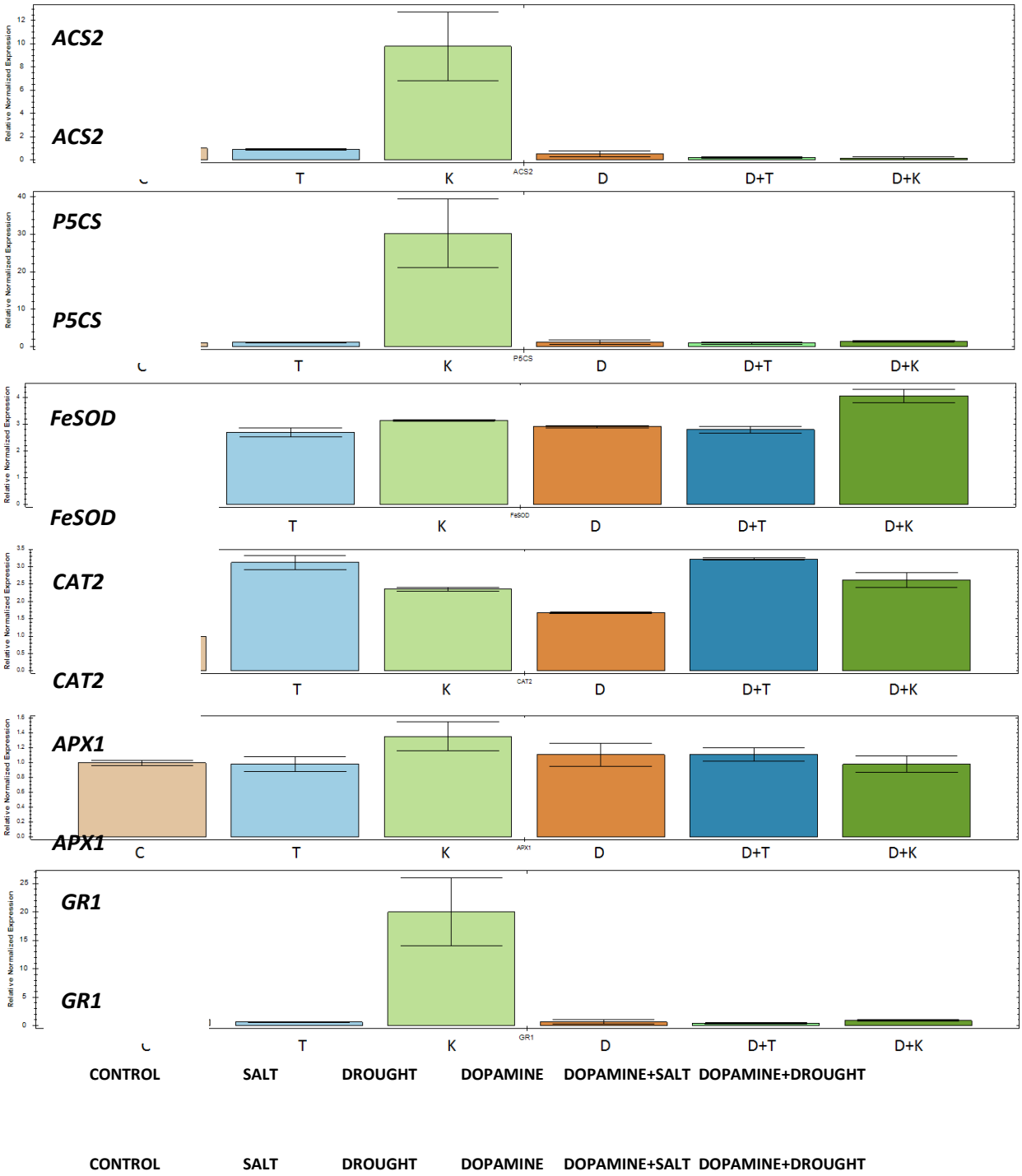


Figure 1. Graph of relative normalized gene expressions for different treatment groups. C, T, K, D, D+T and D+K indicates control, salt, drought, dopamine, dopamine+salt and dopamine+drought applications, respectively.

CONCLUSION

External dopamine applications improve the morphological and physiological responses of tomato plants to drought and salt stress. Dopamine also boosts *FeSOD* and *CAT2* gene expression under drought stress. Reduced tissue ethylene levels and decreased levels of reactive oxygen species, superoxide and H_2O_2 in turn result in cell membrane integrity and improved tissue lengths and weights in tomato plants. The results showed that external application or seed treatment with dopamine has a promising effect on abiotic stress mitigation for agricultural production.

REFERENCES

Abdelaal K, Alsubeie MS, Hafez Y, Emeran A, Moghanm F, Okasha S, Omara R, Basahi MA, Darwish DBE, Ibrahim MFM, El-Yazied AA, Rashwan EA, Elkelish A, Mady MA, Ibraheem F 2022. Physiological and biochemical changes in vegetable and field crops under drought, salinity and weeds stresses: control strategies and management. Agriculture, 12:2084.

Battilani A, Prieto MH, Argerich C, Campillo C, Cantore V 2012. Tomato. In: Steduto P, Hsiao TC, Fereres E, Raes D (eds), Crop Yield Response to Water, Irrigation and Drainage Paper No. 66. FAO, Rome, Italy, pp.192–198.

Bates LS, Waldren RP, Teare ID 1973. Rapid determination of free proline for water-stress studies. Plant Soil, 39:205–207.

Gomes BR, Soares RCS, Santos WD, Marchiosi R, Soares AR, Filho OF 2014. The effects of dopamine on antioxidant enzymes activities and reactive oxygen species levels in soybean roots. Plant Signaling and Behavior, 9:12, e977704.

Guidotti BB, Gomes BR, de Cassia Siqueira-soares R, Soares AR, Ferrarese-Filho O 2013. The effects of dopamine on root growth and enzyme activity in soybean seedlings. Plant Signaling Behav, 8 (9):e25477. doi:10.4161/psb.25477.

Haj-Amor Z, Araya T, Kim DG, Bouri S, Lee J, Ghiloufi W, Yang Y, Kang H, Jhariya MK, Banerjee A, Lal R 2022. Soil salinity and its associated effects on soil microorganisms, greenhouse gas emissions, crop yield, biodiversity and desertification: a review. Sci Total Environ, 843:156946.

Hoagland DR, Arnon DI 1950. The water-culture method for growing plants without soil. Cal Agric Exp Sta Ciru, 347:1–32

Howe MW, Dombeck DA 2016. Rapid signalling in distinct dopaminergic axons during locomotion and reward. Nature, 535: 505–510.

Kanazawa K, Sakakibara H 2000. High content of dopamine, a strong antioxidant, in cavendish banana. J Agr Food Chem, 48: 844–848.

Lan GP, Jiao CJ, Wang GQ, Sun YH, Yan S 2020. Effects of dopamine on growth, carbon metabolism, and nitrogen metabolism in cucumber under nitrate stress. Sci Hortic, 260:108790.

Li C, Sun X, Chang C, Jia D 2015. Dopamine alleviates salt-induced stress in *Malus hupehensis*. Physiologia Plantarum, 153: 584–602.

Li Y 2009. Physiological responses of tomato seedlings (*Lycopersicon esculentum*) to salt stress. Mod Appl Sci, 3(3):171–176.

Nanjo T, KobayashiM, Yoshiba Y, Kakubari Y, Yamaguchi-Shinozaki K, Shinozaki K 1999. Antisense suppression of proline degradation improves tolerance to freezing and salinity in *Arabidopsis thaliana*. FEBS Lett, 461:205–210.

Ohkawa H, Ohishi N, Yagi K 1979. Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Anal Biochem, 95: 351–358.

Protacio CM, Dai YR, Lewis EF, Flores HE 1992. Growth stimulation by catecholamines in plant tissue/organ cultures. Plant Physiol, 98:89–96.

Rosel MA, Mosca L, Foppoli C, Blarzino C, Coccia R 1994. Lipoxygenase-catalyzed oxidation of 5-S-substituted catecholamines. Melanoma Res, 200:344–350.

Smart RE, Bingham GE 1974. Rapid estimates of relative water content. Plant Physiol, 53:258–260.

DRAFT

Yogendra KN, Dhokane D, Kushalappa AC, Sarmiento F, Rodriguez E, Mosquera T 2016. *StWRKY8* transcription factor regulates benzylisoquinoline alkaloid pathway in potato conferring resistance to late blight. Plant Sci, 256: 208–216.

Wang Y, Gao T, Zhang Z, Yuan X, Chen Q, Zheng J, Chen S, Ma F, Li C 2020. Overexpression of the tyrosine decarboxylase gene *MdTyDC* confers salt tolerance in apple. Environmental and Experimental Botany, 180: 104244.

Wang Y, Chen Q, Zheng J, Zhang Z, Gao T, Li C, Ma F 2021. Overexpression of the tyrosine decarboxylase gene *MdTyDC* in apple enhances long-term moderate drought tolerance and WUE. Plant Science, 313: 111064.

Gao T, Wang Y, Liu Y, Ma M 2021. Overexpression of tyrosine decarboxylase (*MdTYDC*) enhances drought tolerance in *Malus domestica*. Scientia Horticulturae, 289: 110425.

Zhang Z, Zhang J, Tang Z, Wang Y, Gao T, Liu X, Feng MA, Chao L 2022. Tissue distribution and changes in dopamine during development and stress responses in *Malus* germplasm. Journal of Integrative Agriculture, 21: 710–724

Zomer RT, Bossio DA, Sommer R, Verchot LV 2017. Global sequestration potential of increased organic carbon in cropland soils. Sci Rep, https://doi.org/10.1038/s41598-017-15794-8

DRAFT

ORAL PRESENTATION

Dual therapies combined with magnetic hyperthermia in cancer treatments

Melek Acar[1]* (*ORCID: https://orcid.org/0000-0001-5931-7799*), Yağmur Ünver[2] (*ORCID: https://orcid.org/0000-0003-1497-081X*)

*[1]Atatürk University, Graduate School of Natural and Applied Sciences, Department of Molecular Biology and Genetics, Erzurum, Turkey
[2]Atatürk University, Faculty of Science, Department of Molecular Biology and Genetics, Erzurum, Turkey

*Corresponding author e-mail: melek.acar12@ogr.atauni.edu.tr

Abstract

According to the report published by IQVIA, the world pharmaceutical market reached 1.5 trillion USD in 2022. For this reason, domestic pharmaceutical production, which has both economic and strategic importance, is a priority area. In addition, when world prescription drug sales in 2022 are examined, it is seen that the highest share is in oncology drugs. 193 billion USD were spent in 2022 for the treatment of cancer, which is the second cause of death in the world. Therefore, more effective, more economical treatments need to be developed. The most commonly used methods in cancer treatment are surgery, radiotherapy and chemotherapy. But, new cancer treatment methods are being developed as an alternative to traditional methods for more effective treatment with fewer side effects. When we look at cancer treatment methods, it is seen that the therapeutic effectiveness of combined treatments is greater than a single method. For example, hyperthermia alone is rarely used. It is usually used together with chemotherapy or radiation to increase the effectiveness of the treatment. Magnetic hyperthermia, one of the methods frequently used in dual treatments, uses the heat generating abilities of magnetic nanoparticles (MNP) under the alternating magnetic field (AMF). Thus, it is possible to increase the temperature only in the tumor area. Many studies show that the therapeutic effect is greater in dual treatments applied together with MHT. The use of these dual therapies by creating personalized treatment combinations for a more effective treatment is the new goal of scientists.

Keywords: cancer treatment, magnetic hyperthermia, dual therapy

INTRODUCTION

Cancer, which is expected to become the first cause of death in the world in the near future, is a major threat to human health today. According to the report published by IQVIA, in 2022, the world pharmaceutical market reached 1.5 trillion USD (IQVIA, 2023). This number is increasing day by day due to the rapidly increasing number of cases. When world prescription drug sales in 2022 are examined, it is seen that the highest share is in oncology drugs. According to WHO (World Health Organization) 2020 data, breast cancer, lung cancer and colorectum cancer are the most common cancer types. When it comes to death rates, lung and colorectum cancer come first, with rates of 18% and 9.4%. The incidence of cancer types is shown in the Figure (World Health Organization, 2020).

DRAFT

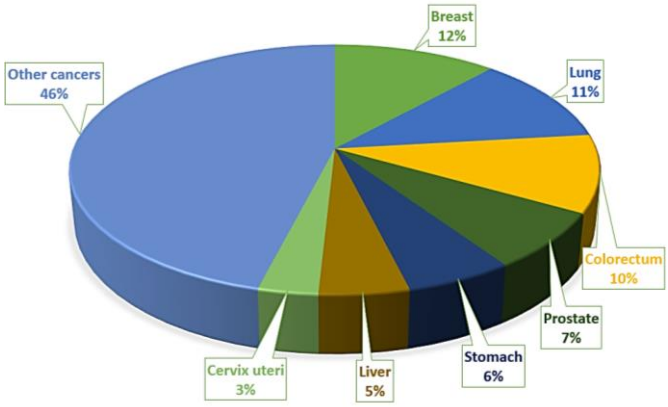


Figure 1. Estimated number of new cases in 2020, World, both sexes, all ages

The most commonly used methods in cancer treatment are surgery, radiotherapy and chemotherapy. Surgical treatment is the most effective treatment method, but many patients do not prefer surgical treatment. While other methods kill cancer cells, they also kill healthy cells and have severe side effects (Yaghoubi et al., 2020). According to the current data of the National Cancer Institute, new cancer treatment methods used as an alternative to traditional methods; hormone therapy, immunotherapy, photodynamic therapy, photothermal therapy, stem cell transplantation, targeted therapy and hyperthermia (National Cancer Institute, 2015). Considering cancer treatment methods, the therapeutic effectiveness of a single method is less than combined treatments (Gurunathan et al., 2018). Therefore, two or more methods are used together for a more effective treatment with less side effects (Table).

For example, hyperthermia, a treatment method to kill cancer cells by increasing the environment temperature, is rarely used alone (Behrouzkia et al., 2016). Hyperthermia is often used in combination with chemotherapy and radiation, thus providing a more effective treatment. Hyperthermia increases the sensitivity of cells to radiation and drugs. Hyperthermia, when used together with radiation, may make some of the tumor cells more prone to radiations. Additionally, free radicals generated from radiotherapy damage the DNA of tumor cells, and hyperthermia prevents repair (Jha et al., 2016). When used together with chemotherapy, it may increase the effects of anticancer agents. Thus, it provides a chemo-sensitizing and radio-sensitizing effect (Behrouzkia et al., 2016). Hyperthermia can also be commonly applied with other different forms of cancer therapy (Jha et al., 2016). However, if the tumor area cannot be targeted in hyperthermia applications, heating also occurs in healthy tissues (Rajan & Sahu, 2020). This is an undesirable situation in cancer treatments. Because while destroying the tumor area, it is desired to minimize the side effects by not damaging the healthy cells. In order to overcome this situation, another hyperthermia strategy called 'magnetic hyperthermia' has been developed.

Magnetic nanoparticles (MNPs) are used as heat mediators in magnetic hyperthermia. With an externally applied AC magnetic field (AMF), magnetic energy is converted into thermal energy only in the region of MNPs. Thus, it is possible to specifically increase the temperature of only the tumor area (Peiravi vd., 2022). By exposing cancer cells to a temperature higher (41-46 °C) than normal body temperature for a certain period of time, an effective cellular heat shock damage is induced (Acar et al., 2022). This heat shock alters cellular processes, protein structure and function, eventually leading to cellular death called apoptosis (Kumar & Mohammad, 2011). Biocompatible and biodegradable MNPs are pertinent materials for MHT applications. Metals such as iron, cobalt, nickel, manganese, zinc, gadolinium, magnesium, their alloys, and oxides are some of the explored MNPs for MHT. Iron oxide nanoparticles (IONPs) possessing excellent self-heating ability is the most used MNPs (Rajan & Sahu, 2020). The selection of MNP exhibiting a sufficient heating capacity (specific absorption rate) to achieve satisfactory temperatures is important. The specific absorption rate (SAR) is the measure of heat diffuse per unit mass of MNPs (Gupta & Sharma, 2021). To determine the SAR value, MNPs are exposed to AMF. Depending on the amount of iron content of the sample, heat production is determined (Ludwig et al., 2014). Then, SAR values are calculated using the specific formulation (Acar et al., 2022; Albarqi et al., 2019).

In addition, surface functionalization of MNPs enables selective targeting of cancer cells (Bañobre-López et al., 2013). For example, folic acid receptors are overexpressed on the surface of breast cancer cells. Due to their specificity to these receptors, folic acid conjugated nanocarriers are used for targeting purposes. In this way, the accumulation of the carrier in non-target organs is reduced (Tagde et al., 2020). Therefore, it is important to functionalize MNPs to specifically target different types of cancer.

Table 1. Combinatorial approaches for cancer

Combinatorial approaches	Type of cancer	Reference
Chemotherapy and Magnetic Hyperthermia	Breast cancer	(Petryk et al., 2013)
Chemotherapy and Photothermal Therapy	Cervix cancer	(T. Liu et al., 2014)
	Brain cancer	(Dash et al., 2021)
Chemotherapy, Hyperthermia and Ultrasound	Liver cancer	(Misra et al., 2015)
Hyperthermia and Radiation Therapy	Breast cancer	(Datta et al., 2016)
Gene Therapy and Magnetic Hyperthermia	Ovarian cancer	(Yin et al., 2016)
Gene Therapy and Photothermal Therapy	HeLa tumor	Liu vd. (2018)
		(Zhao et al., 2021)
Chemotherapy and Hyperthermia	Colon cancer	(Zheng et al., 2019)
Magnetic Hyperthermia and Immune Therapy	Breast cancer	(Pan et al., 2020)
Chemotherapy, Radiotherapy and Photothermal Therapy	Colon cancer	(Mirrahimi et al., 2020)
Magnetic Hyperthermia, Photothermal Therapy and Chemotherapy	Breast cancer	(Sharifi et al., 2020)
Chemotherapy and Photodynamic Therapy	Prostate cancer	(Ghosh et al., 2022)
Gene therapy and Chemotherapy	Breast cancer	(Jia et al., 2022)

Looking at the studies, it seems that MHT is more effective when used together with other therapies. In a study in which cisplatin (chemotherapy) and MHT were used together for breast cancer treatment, it was observed that dual therapy was 1.7 times more effective than MHT alone and 1.4 times more effective than cisplatin alone (Petryk et al., 2013). In another study, they used gene therapy and MHT together for the treatment of ovarian cancer. They controlled TRAIL expression by hyperthermia by adding the TRAIL gene, which is associated with the apoptotic pathway, behind the hsp70 promoter (a heat-induced promoter) sequence. Thus, tumor size was observed to decrease significantly in dual therapy (Yin et al., 2016). In a study where MHT was used together with immunotherapy, superparamagnetic $CoFe_2O_4$@$MnFe_2O_4$ nanoparticles were synthesized for MHT. Combined MHT and α-PD-L1 treatment was applied to breast cancer model mice. An effective anticancer activity was achieved by MHT-induced immunotherapy as a result of the formation of abundant tumor-associated antigens (Pan et al., 2020).

CONCLUSION

From past to present, efforts have been made to develop methods for cancer treatment that are both more effective and have fewer side effects. When we look at the studies, it is seen that the use of two or more therapies together is more effective than single therapies. For this reason, finding the most effective method by applying different combinations for different types of cancer is considered the best strategy. Creating personalized treatment combinations for even more effective treatment is the new target for cancer treatment.

DRAFT

ACKNOWLEDGEMENTS

M.A. is thankful for The Council of Higher Education (CoHE, 100/2000) PhD Scholarship Program, Turkey.

REFERENCES

Acar, M., Solak, K., Yildiz, S., Unver, Y., & Mavi, A. (2022). Comparative heating efficiency and cytotoxicity of magnetic silica nanoparticles for magnetic hyperthermia treatment on human breast cancer cells. *3 Biotech*, *12*(11), 313. https://doi.org/10.1007/s13205-022-03377-y

Albarqi, H. A., Wong, L. H., Schumann, C., Sabei, F. Y., Korzun, T., Li, X., Hansen, M. N., Dhagat, P., Moses, A. S., Taratula, O., & Taratula, O. (2019). Biocompatible Nanoclusters with High Heating Efficiency for Systemically Delivered Magnetic Hyperthermia. *ACS Nano*, *13*(6), 6383–6395. https://doi.org/10.1021/acsnano.8b06542

Bañobre-López, M., Teijeiro, A., & Rivas, J. (2013). Magnetic nanoparticle-based hyperthermia for cancer treatment. *Reports of Practical Oncology & Radiotherapy*, *18*(6), 397–400. https://doi.org/10.1016/j.rpor.2013.09.011

Behrouzkia, Z., Joveini, Z., Keshavarzi, B., Eyvazzadeh, N., & Aghdam, R. Z. (2016). Hyperthermia: How Can It Be Used? *Oman Medical Journal*, *31*(2), 89–97. https://doi.org/10.5001/omj.2016.19

Dash, B. S., Lu, Y. J., Chen, H. A., Chuang, C. C., & Chen, J. P. (2021). Magnetic and GRPR-targeted reduced graphene oxide/doxorubicin nanocomposite for dual-targeted chemo-photothermal cancer therapy. *Materials Science and Engineering C*, *128*. https://doi.org/10.1016/j.msec.2021.112311

Datta, N. R., Puric, E., Klingbiel, D., Gomez, S., & Bodis, S. (2016). Hyperthermia and Radiation Therapy in Locoregional Recurrent Breast Cancers: A Systematic Review and Meta-analysis. *International Journal of Radiation Oncology*Biology*Physics*, *94*(5), 1073–1087. https://doi.org/10.1016/j.ijrobp.2015.12.361

Ghosh, S., Gul, A. R., Xu, P., Lee, S. Y., Rafique, R., Kim, Y. H., & Park, T. J. (2022). Target delivery of photo-triggered nanocarrier for externally activated chemo-photodynamic therapy of prostate cancer. *Materials Today Chemistry*, *23*. https://doi.org/10.1016/j.mtchem.2021.100688

Gupta, R., & Sharma, D. (2021). Therapeutic response differences between 2D and 3D tumor models of magnetic hyperthermia. *Nanoscale Advances*, *3*(13), 3663–3680. https://doi.org/10.1039/D1NA00224D

Gurunathan, S., Kang, M. H., Qasim, M., & Kim, J. H. (2018). Nanoparticle-mediated combination therapy: Two-in-one approach for cancer. *International Journal of Molecular Sciences*, *19*(10), 1–37. https://doi.org/10.3390/ijms19103264

IQVIA. (2023). *The Global Use of Medicines*. https://www.iqvia.com/insights/the-iqvia-institute/reports/the-global-use-of-medicines-2023

Jha, S., Sharma, P. K., & Malviya, R. (2016). Hyperthermia: Role and Risk Factor for Cancer Treatment. *Achievements in the Life Sciences*, *10*(2), 161–167. https://doi.org/10.1016/j.als.2016.11.004

Jia, R., Wang, Y., Ma, W., Huang, J., Sun, H., Chen, B., Cheng, H., He, X., & Wang, K. (2022). Activatable Dual Cancer-Related RNA Imaging and Combined Gene-Chemotherapy through the Target-Induced Intracellular Disassembly of Functionalized DNA Tetrahedron. *Analytical Chemistry*, *94*(15), 5937–5945. https://doi.org/10.1021/acs.analchem.2c00364

Kumar, C. S. S. R., & Mohammad, F. (2011). Magnetic Nanomaterials for Hyperthermia-based Therapy and Controlled Drug Delivery. *Adv Drug Deliv Rev*, *63*(9), 789–808. https://doi.org/10.1016/j.addr.2011.03.008.Magnetic

Liu, T., Wang, C., Gu, X., Gong, H., Cheng, L., Shi, X., Feng, L., Sun, B., & Liu, Z. (2014). Drug Delivery with PEGylated MoS 2 Nano-sheets for Combined Photothermal and Chemotherapy of Cancer. *Advanced Materials*, *26*(21), 3433–3440. https://doi.org/10.1002/adma.201305256

Liu, Y., Shu, G., Li, X., Chen, H., Zhang, B., Pan, H., Li, T., Gong, X., Wang, H., Wu, X., Dou, Y., & Chang, J. (2018). Human HSP70 Promoter-Based Prussian Blue Nanotheranostics for Thermo-Controlled Gene Therapy and Synergistic Photothermal Ablation. *Advanced Functional Materials*, *28*(32), 1–12. https://doi.org/10.1002/adfm.201802026

Ludwig, R., Stapf, M., Dutz, S., Müller, R., Teichgräber, U., & Hilger, I. (2014). Structural properties of magnetic nanoparticles determine their heating behavior - an estimation of the in vivo heating potential. *Nanoscale Research Letters*, *9*(1). https://doi.org/10.1186/1556-276X-9-602

Mirrahimi, M., Beik, J., Mirrahimi, M., Alamzadeh, Z., Teymouri, S., Mahabadi, V. P., Eslahi, N., Ebrahimi Tazehmahalleh, F., Ghaznavi, H., Shakeri-Zadeh, A., & Moustakis, C. (2020). Triple combination of

heat, drug and radiation using alginate hydrogel co-loaded with gold nanoparticles and cisplatin for locally synergistic cancer therapy. *International Journal of Biological Macromolecules*, *158*, 617–626. https://doi.org/10.1016/j.ijbiomac.2020.04.272

Misra, S. K., Ghoshal, G., Gartia, M. R., Wu, Z., De, A. K., Ye, M., Bromfield, C. R., Williams, E. M., Singh, K., Tangella, K. V., Rund, L., Schulten, K., Schook, L. B., Ray, P. S., Burdette, E. C., & Pan, D. (2015). Trimodal Therapy: Combining Hyperthermia with Repurposed Bexarotene and Ultrasound for Treating Liver Cancer. *ACS Nano*, *9*(11), 10695–10718. https://doi.org/10.1021/acsnano.5b05974

National Cancer Institute. (2015). Types of Cancer Treatment - National Cancer Institute. In *National Cancer Institute* (p. 368). https://www.cancer.gov/about-cancer/treatment/types

Pan, J., Hu, P., Guo, Y., Hao, J., Ni, D., Xu, Y., Bao, Q., Yao, H., Wei, C., Wu, Q., & Shi, J. (2020). Combined Magnetic Hyperthermia and Immune Therapy for Primary and Metastatic Tumor Treatments. *ACS Nano*, *14*(1), 1033–1044. https://doi.org/10.1021/acsnano.9b08550

Peiravi, M., Eslami, H., Ansari, M., & Zare-Zardini, H. (2022). Magnetic hyperthermia: Potentials and limitations. *Journal of the Indian Chemical Society*, *99*(1), 100269. https://doi.org/10.1016/j.jics.2021.100269

Petryk, A. A., Giustini, A. J., Gottesman, R. E., Kaufman, P. A., & Hoopes, P. J. (2013). Magnetic nanoparticle hyperthermia enhancement of cisplatin chemotherapy cancer treatment. *International Journal of Hyperthermia*, *29*(8), 845–851. https://doi.org/10.3109/02656736.2013.825014

Rajan, A., & Sahu, N. K. (2020). Review on magnetic nanoparticle-mediated hyperthermia for cancer therapy. *Journal of Nanoparticle Research*, *22*(11), 319. https://doi.org/10.1007/s11051-020-05045-9

Sharifi, M., Hasan, A., Nanakali, N. M. Q., Salihi, A., Qadir, F. A., Muhammad, H. A., Shekha, M. S., Aziz, F. M., Amen, K. M., Najafi, F., Yousefi-Manesh, H., & Falahati, M. (2020). Combined chemo-magnetic field-photothermal breast cancer therapy based on porous magnetite nanospheres. *Scientific Reports*, *10*(1), 5925. https://doi.org/10.1038/s41598-020-62429-6

Tagde, P., Kulkarni, G. T., Mishra, D. K., & Kesharwani, P. (2020). Recent advances in folic acid engineered nanocarriers for treatment of breast cancer. *Journal of Drug Delivery Science and Technology*, *56*(December 2019), 101613. https://doi.org/10.1016/j.jddst.2020.101613

World Health Organization. (2020). *Estimated number of new cases*. https://www.iarc.who.int/

Yaghoubi, A., Khazaei, M., Avan, A., Hasanian, S. M., Cho, W. C., & Soleimanpour, S. (2020). p28 Bacterial Peptide, as an Anticancer Agent. *Frontiers in Oncology*, *10*(August), 1–10. https://doi.org/10.3389/fonc.2020.01303

Yin, P. T., Shah, S., Pasquale, N. J., Garbuzenko, O. B., Minko, T., & Lee, K.-B. (2016). Stem cell-based gene therapy activated using magnetic hyperthermia to enhance the treatment of cancer. *Biomaterials*, *81*, 46–57. https://doi.org/10.1016/j.biomaterials.2015.11.023

Zhao, Y., Zhao, T., Cao, Y., Sun, J., Zhou, Q., Chen, H., Guo, S., Wang, Y., Zhen, Y., Liang, X. J., & Zhang, S. (2021). Temperature-Sensitive Lipid-Coated Carbon Nanotubes for Synergistic Photothermal Therapy and Gene Therapy. *ACS Nano*, *15*(4), 6517–6529. https://doi.org/10.1021/acsnano.0c08790

Zheng, Y., Wang, W., Zhao, J., Wu, C., Ye, C., Huang, M., & Wang, S. (2019). Preparation of injectable temperature-sensitive chitosan-based hydrogel for combined hyperthermia and chemotherapy of colon cancer. *Carbohydrate Polymers*, *222*(January), 115039. https://doi.org/10.1016/j.carbpol.2019.115039

DRAFT

ORAL PRESENTATION

Investigation of phenolic compounds and elicited by jasmonate derivatives on *in vitro* samples of *Hypericum perforatum* L.

Şeyma ÖNLÜ*(*ORCID: https://orcid.org /0000-0003-2005-1019*)

* Department of Molecular Biology and Genetic Department, Science and Art Faculty, Muş Alparslan University, Muş, Turkey

* Corresponding author e-mail: s.sofuoglu@alparslan.edu.tr

Abstract

In this study, jasmonic acid (JA) and methyl jasmonate (MeJA) elicitation and LC-HRMS analyses were performed in *Hypericum perforatum in vitro* plantlets and callus. The highest shoot regeneration was obtained from LS (Linsmaier&Skoog)/B5 medium containing 1 mg/L benzylaminopurine (BAP). Callus was produced in LS/B5 media which contain 0.5 mg/L Thidiazuron (TDZ) and 0.5 mg/L Indole Butyric Acid (IBA). Elicitation was applied 0.01 mg/L JA callus, 0.01 mg/L JA *in vitro* plantlets, and 0.01 mg/L MeJA *in vitro* plantlets. Epicatechin, acacetin, fumaric acid, hispidulin, naringenin, naringin, chicoric, hyperoside, rhamnocitrin, chlorogenic acid, chrysin, isosakuranetin, apigenin-7-glucoside compounds were identified by Liquid chromatography-high-resolution mass spectrometry (LC-HRMS). While there was no production in calli; a five-fold increase in epicatechin compound in 0.01 mg/L JA *in vitro* plantlets and a significant decrease in 0.01 mg/L MeJA, compared to the control group. Acacetin was not detected in the control group, but it was detected in the same amount in all groups. Cichoric, fumaric acid, chlorogenic acid and hyperoside compounds were detected highest in plantlets containing 0.01 mg/L JA. Fumaric acid was detected in 0.01 mg/L JA calli, 2.4 times more than the control group. Fumaric acid and hyperoside were produced in both plantlets and calli, however, hispidulin was produced only in calli. Chlorogenic acid was not produced in calli but was produced in the highest amounts in other groups. Naringin, chrysin, isosakuranetin, and apigenin-7-glucoside compounds could not be detected in any of the groups.

Keywords: *H.perforatum*, LC-HRMS, *In vitro* plantlet, Callus, Jasmonic acid, Methyl Jasmonate

INTRODUCTION

H.perforatum L. (Clusiaceae) is a common species and there are around 482 species in the world. Due to its traditional usage, it has been widely studied by researchers (Crockett and Robson 2011). It has important medical effects such as antimicrobial, antioxidant, antiviral, antifungal, cell regenerative and antidepressant because of contains naphthodiantrone, phlorogucinol and other phenolic compounds which have been synthesized by the aerial parts of the plant (Gadzovska et al. 2007, Çırak et al. 2016). *In vitro* production of compounds with these medicinal effects is encouraged through plant tissue culture techniques. While the flowering period of plants in nature takes a whole year, this period is shortened with *in vitro* techniques; additionally, the production of desired metabolites can be achieved under biotic and abiotic stress conditions (Wang et al 2015). Many studies conducted on *Hypericum* have revealed variations in metabolite amounts with various elicitors such as-jasmonic acid, salicylic acid, and methyl jasmonate-under *in vitro* conditions. In addition, the content of compounds was analyzed with various analytical (HPLC, HPLC-MS, LC-MS, LCMS-MS, etc.) devices (Jeyasri et al. 2023).

Today, many exogenous elicitors create a stress response within plant cells and thus increase secondary metabolite production through plant tissue culture techniques. Jasmonic acid and methyl jasmonate, which are jasmonate derivatives obtained from jasmine, are elicitors used in *in vitro* techniques. Jasmonic acid is a hormone used biotically to stimulate the accumulation of alkaloids, terpenoids, and phenolic compounds in plants (Zhao et al. 2005, Jirakiattikul et al. 2021). In previous studies, it was stated that jasmonic acid increased the amount of total phenolic and flavonoids in plants such as *H. perforatum* (Wang et al. 2015), *Artemisia absinthium* (Ali et al. 2014), *Phyllanthus pulcher* (Danaee et al. 2015).

Methyl jasmonate is a volatile methyl ester of jasmonic acid and an important signaling molecule in biotic and abiotic stresses. It is considered an important plant hormone that regulates internal and external transmission in plants due to its volatility and easy passage through biological membranes (Wang et al. 2021, Jeyasri et al 2023). It has been reported that externally applied methyl jasmonate in *in vitro* culture activates the antioxidant system and increases the expression of defense-related genes (Murthy et al. 2014; Ho et al. 2020). In this study, *H.perforatum* plantlets and callus were obtained, then jasmonic acid and methyl jasmonate were added to separately the nutrient medium (0.01 mg/L) under *in vitro* conditions. Moreover; epicatechin, acacetin, fumaric acid, hispidulin, naringenin, naringin, chicoric, hyperoside, rhamnocitrin, chlorogenic acid, chrysin, isosakuranetin, apigenin-7-glucoside compounds were detected with the LC-HRMS devices.

MATERIALS AND METHODS

Plant Material and Seeds Surface Sterilization

Hypericum perforatum L. seeds were collected from Samsun-Ballıca, (41'27'97'01' and 36'33'60'67') Turkey. Seeds were washed for 15 minutes under tap water, surface sterilized with 70 % ethanol for 1 min, then NaOCl 15 % for 5 min, and rinsed 3 times in sterile deionized water. Seeds were cultured LS /B5 medium supplemented with 3 % sucrose and 0.7 % agar without plant growth regulators (Önlü 2019).

In vitro Plantlets and Callus Formation

For in vitro multiplication, 1 mg/L BAP was used and rooted 1 mg/L IBA at LS/B5 media. Callus cultures were started with axillary buds of sterile seedlings. The media contained LS/B5 including 3 % sucrose, and 0.7 % agar. Five different TDZ (0.1, 0.25, 0.5, 0.75, 1 mg/L) and IBA (1,0.75, 0.5, 0.25, 0.1 mg/L) concentrations were used for callus culture.

Elicitation of Elicitors

After 2 months of callus induction, the friable and green callus as equal fragments were transferred to new LS/B5 media containing jasmonic acid (0.01 mg/L) elicitor for 15 days. *In vitro* plantlets also were transferred to jasmonic acid (0.01 mg/L) and methyl jasmonate (0.01 mg/L) media, separately. Jasmonic acid (Duchefa, Netherland) and methyl jasmonate (Duchefa, Netherland) were dissolved in pure ethanol and filtered using a microfilter of 0.22 µ pore size. All treatments were incubated in the 16 h light/8 h dark photoperiod at 24 ± 2°C and photon flux of 90 µmol m^{-2} s^{-1} in the culture chamber.

Extraction and Quantification of Secondary Metabolites

The calli and plantlets which treatments with JA and MeJa were freeze-dried lyophilized and powdered. Plant materials were extracted with 80 % (v/v) methanol in an ultrasonic bath for 30 min at 4°C as previously reported (Gadzovska et al. 2007, Gadzovska et al. 2013).

Statistical Analyses

All the experiments were conducted using three biological replicates and all measurements were analyzed in triplicate. Significant differences in the amount of compounds between different plant extracts were evaluated with the Anova and Kruskal Wallis test. IBM SPSS Statistics 26.0 (IBM Corp. Released 2019. IBM SPSS Statistics for Windows, Version 26.0. Armonk, NY: IBM Corp.) and OriginPro 2018 (OriginLab Corporation, Northampton, MA, USA) were used for statistical analyses and calculations. The statistical significance level was accepted as $p<0.05$.

LC-HRMS and Chromatographic Conditions

The analyses of the extracts were made with LC-HRMS. Epicatechin, acacetin, fumaric acid, hispidulin, naringenin, naringin, chicoric acid, hyperoside, rhamnocitrine, chlorogenic acid, chrysin, isosacuranetin, apigenin 7-glucoside compounds were determined. LC-HRMS measurements were carried out on a Thermo ORBITRAP Q-EXACTIVE (Bremen, Germany) mass spectrometry-equipped ESI ion source and with a Dionex LC system. Identification of compounds was performed previously reported (İnal et al. 2022) by comparison of retention time of standard compounds in the range of purity 95-99% see section chemicals) and HRMS data of Bezmialem Vakıf University, Drug Application and Research Center Library-ILMER.

RESULTS and DISCUSSION

It was determined that the amount of chlorogenic acid was higher in plantlets containing 0.01 mg/L JA than in both control and methyl jasmonate. The hispidulin compound only was produced in calli containing 0.01 mg/L

JA. While fumaric acid was produced highest in calli 0.01 mg/L JA, its amount decreased in 0.01 mg/L MeJa. For hyperoside, methyl jasmonate and callus groups were decreased. Epicatechin was detected at the highest rate in *in vitro* plantlets of 0.01 mg/L JA, while it could not be detected in callus. Acacetin was determined at the same rate in all groups, except of the control. While naringenin was produced in small amounts in 0.01 mg/L MeJA plantlets, it could not be detected in other groups. Considering the overall composition of the compounds, jasmonic acid increased the amounts compared to methyl jasmonate (Table 1).

Table 1. Amounts of compounds analyzed by LC-HRMS in *in vitro* plantlets and calli of *H. perforatum* L.

	Compounds	**Retention Time**	**Quantification(mg/L)**				**Mother Ion (m/z)**	**Ion mode**	**LOD/LOQ (mg/L)**
			***in vitro* seedling_ control**	***in vitro* seedling_0.01 mg/L JA**	***in vitro* seedling_0.01 mg/L MeJA**	**Callus_0.01 mg/L JA**			
1	Epicatechin	2.66	0.52	2.64	0.04	n.d.	291.0863	Negative	0.23/0.76
2	Acacetin	7.88	n.d.	0.10	0.10	0.11	283.0612	Negative	0.13/0.42
3	Fumaric acid	2.49	0.63	1.21	0.71	1.55	115.0037	Negative	0.26/0.88
4	Hispidulin	6.60	n.d.	n.d.	n.d.	0.39	299.0561	Negative	0.14/0.46
5	Naringenin	5.89	n.d.	n.d.	0.04	n.d.	271.0612	Negative	0.2/0.67
6	Naringin	5.02	n.d.	n.d.	n.d.	n.d.	579.1719	Negative	0.23/0.77
7	Chicoric	3.66	0.21	0.35	0.09	n.d.	473.0725	Negative	0.14/0.48
8	Hyperoside	4.84	0.33	0.34	0.22	0.13	463.0882	Negative	0.33/1.09
9	Rhamnocitrin	6.60	0.1	0.1	n.d.	n.d.	299.0561	Negative	0.16/0.53
10	Chlorogenic acid	2.49	6.08	9.15	1.18	n.d.	353.0878	Negative	0.33/1.08
11	Chrysin	7.07	n.d.	n.d.	n.d.	n.d.	253.0506	Negative	0.21/0.69
12	Isosakuranetin	6.60	n.d.	n.d.	n.d.	n.d.	285.0768	Negative	0.23/0.77
13	Apigenin-7-Glucoside	4.60	n.d.	n.d.	n.d.	n.d.	433.1129	Positive	0.18/0.60

n.d.: Not detected

The metabolites desired to be produced may sometimes be at low levels in plant tissue culture samples. Since LC-HRMS has high resonance in liquid chromatograms, it can detect molecules of low mass. According to our LC-HRMS analysis results, also it can be compared either methyl jasmonate or jasmonic acid is more effective as an external elicitor (Figure 1). Although there are many studies on the effect of methyl jasmonate on metabolite production cell suspension culture of *H.perforatum* (Wang et al 2015), *Isatis tinctoria* (Gai et al. 2019); in our study, it had little effect on phenolic compounds. The elicitor of JA made a significant increase in the amount of epicatechin and chlorogenic acid, which also provided hispidulin production in calli. Franklin and Dias (2011) reported that the amount of chlorogenic acid was higher in *in vitro* plantlets of *H.perforatum.* It has been reported that chlorogenic acid, which has an important role in plant functions, accumulates in tissues after plant injuries and triggers organ formation (Ramamurthy et al. 1992; Campos-Vargas and Saltveit 2002).

Zhang et al (2014) reported that hyperoside, a flavonoid group, inhibits the proliferation of osteosarcoma cells by inducing G0/G1 arrest in the cell cycle without causing significant cell death. Catechin group phytochemicals are some of the alternative chemotherapeutics that have been extensively tested for their anti-tumor and anti-inflammatory properties (Kurbitz et al. 2011). Epicatechin is one of these derivatives and its production should be supported *in vitro* plant culture.

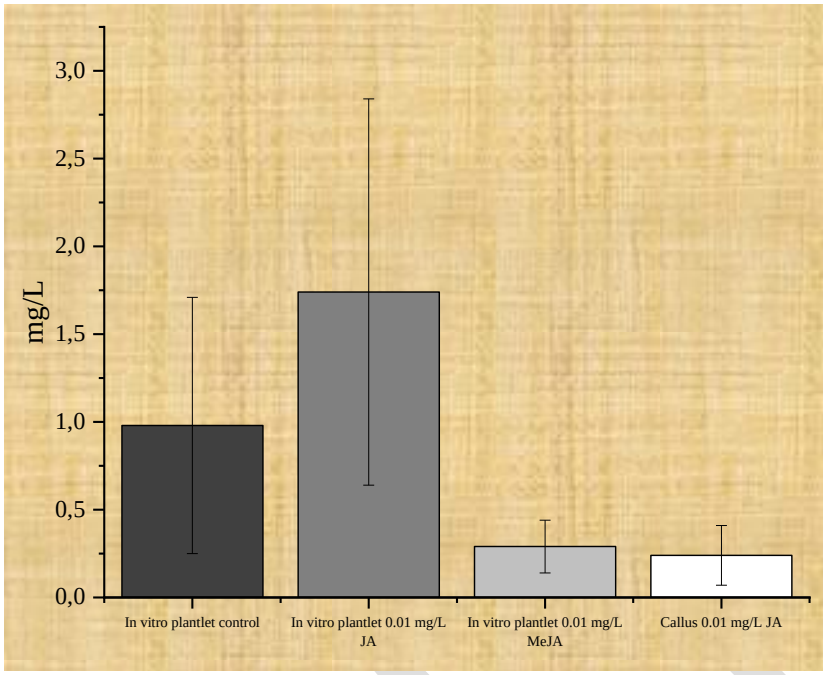


Figure 1. Comparison of total compounds from *in vitro* examples from *H.perforatum* L. Respectively; *In vitro* plantlets control, *In vitro* plantlets 0.01 mg/L JA, *In vitro* plantlets 0.01 mg/L MeJA, Callus 0.01 mg/L JA. Each bar represents the percentage ± standard deviation (SD) for indicated total compounds. Different letters show significant $p<0.05$ from ANOVA and KW.

CONCLUSION

This study demonstrated the role of stress in the stimulated production of phenolic compounds and the effect of JA in callus cultures of stimulated hispidulin. The productivity of phenolic compounds was found to be dependent on the elicitation period and elicitors. The results of the present study indicate that JA is the most effective elicitor for phenolic compounds in *in vitro* plantlets, especially for chlorogenic acid. The medical importance of phytochemicals induced by jasmonic acid in *H. perforatum* plantlets has been emphasized by the mentioned studies. It is important to produce, purify, and bring these valuable chemicals into the economy under *in vitro* conditions.

ACKNOWLEDGEMENTS

This research received financial support from Muş Alparslan University Research Foundation (Project No: BAP-18-FEF-4901-02).

REFERENCES

Ali M, Abbasi BH, Ali GS 2014. Elicitation of antioxidant secondary metabolites with jasmonates and gibberellic acid in cell suspension cultures of *Artemisia absinthium* L. Plant Cell Tiss Organ Cult, 120:1099–1106.

Campos-Vargas R, Saltveit ME 2002. Involvement of putative chemical wound signals in the induction of phenolic metabolism in wounded lettuce. Physiol. Plant, 114:73:84.

Crockett SL and Robson NKB 2011. Taxonomy and Chemotaxonomy of the Genus *Hypericum*. Medicinal and Aromatic Plant Science and Biotechnology, 5 (Special Issue 1), 1-13.

Çırak C, Radušienë, J, Jakstas V, Ivanauskas L, Yayla F, Seyis F, Camas N 2016. Secondary metabolites of *Hypericum* species from the Drosanthe and Olympia sections. South African Journal of Botany, 104: 82–90.

Danaee M, Farzinebrahimi R, Kadir MA, Sinniah UR, Mohamad R, Taha RM 2015. Effects of MeJA and SA elicitation on secondary metabolic activity, antioxidant content and callogenesis in *Phyllanthus pulcher*. Braz J Bot, 38:265–272.

Franklin G, Dias ACP 2011. Chlorogenic acid participates in the regulation of shoot, root, and root hair development in *Hypericum perforatum*. Plant Physiology and Biochemistry, 49:835-842.

Gadzovska S, Maury S, Delaunay A, Spasenoski M, Joseph C, Hagege D. 2007 Jasmonic acid elicitation of *Hypericum perforatum* L. cell suspensions and effects on the production of phenylpropanoids and naphtodianthrones. Plant Cell Tiss Organ Culture, 89:1–13.

Gadzovska S, Maury S, Delaunay A, Spasenoski M, Hagege D, Courtois D. and Joseph C. 2013. The influence of salicylic acid elicitation of shoots, callus, and cell suspension cultures on production of naphtodianthrones and phenylpropanoids in *Hypericum perforatum* L. Plant Cell Tissue Organ Culture, 113,25–39.

Ho TT, Murthy HN, Park SY 2020. Methyl jasmonate induced oxidative stress and accumulation of secondary metabolites in plant cell and organ cultures. Int J Mol Sci, 21(3):716.

İnal E, Ulusoy Ş, Algın Yapar E, Sönmez Gürer E, Yalçın Ş, Kartal M. 2022. Investigation of resveratrol and phenolic compounds of ethnomedicinal plant *Polygonum cognatum* Meissn. collected from Sivas. Journal of Research in Pharmacy, 26(6): 1752-1757.

Jeyasri R. Muthuramalingam P, Karthick K, Shin H, Choi SH, Ramesh M 2023. Methyl jasmonate and salicylic acid as powerful elicitors for enhancing the production of secondary metabolites in medicinal plants: an updated review. Plant Cell, Tissue and Organ Culture, 153:447–458.

Jirakiattikul Y, Rithichai P, Kwanthong P, Itharat A 2021. Effect of jasmonic acid elicitation period on enhancement of bioactive compounds and antioxidant activity in callus cultures of *Hibiscus sabdariffa* Linn. Horticulture, Environment and Biotechnology, 62:629–636.

Kurbitz C, Heise D, Redmer T, Goumas F, Arlt A, Lemke J, Rimbach G, Kalthoff H and Trauzold A 2011. Epicatechin gallate and catechin gallate are superior to epigallocatechin gallate in growth suppression and anti-inflammatory activities in pancreatic tumor cells. Cancer Science, 102: 728–734.

Murthy HN, Lee EJ, Paek KY 2014. Production of secondary metabolites from cell and organ cultures: strategies and approaches for biomass improvement and metabolite accumulation. Plant Cell Tissue Organ Culture, 118(1):1–16.

Önlü Ş 2019. *Hypericum* türlerinde *in vitro* bitki rejenerasyonu ve sekonder metabolit üretimi, Fen Bilimleri Enstitüsü, Doktora Tezi, Ankara Üniversitesi.

Ramamurthy MS, Maiti B, Thomas P, Nair P.M. 1992. High-performance liquid chromatography determination of phenolic acids in potato tubers (*Solanum tuberosum*) during wound healing. J. Agric. Food Chem, 40:569-572.

Wang J, Qian J, Yao L, Lu Y 2015. Enhanced production of flavonoids by methyl jasmonate elicitation in cell suspension culture of *Hypericum perforatum*. Bioresour Bioprocess, 2:1–9.

Wang Y, Mostafa S, Zeng W, Jin B 2021. Function and mechanism of jasmonic acid in plant responses to abiotic and biotic stresses. Int J Mol Sci 22(16):8568.

Zhao J, Davis LC, Verpoorte R 2005. Elicitor signal transduction leading to production of plant secondary metabolites. Biotechnol Adv, 23:283–333.

Zhang N, Ying MD, Wu YP, Zhou ZH, Ye ZM, Li H, Lin DS 2014. Hyperoside, a Flavonoid Compound, Inhibits Proliferation and Stimulates Osteogenic Differentiation of Human Osteosarcoma Cells. Plos One, 9(7):98973.

DRAFT

ORAL PRESENTATION

Effect on Electrochemical Performance of Polyaniline-based CZTS nanocomposite

Suleyman Gokhan Colak [1](ORCID 0000-0002-4978-1499), Ahmet Gungor [2](ORCID 0000-0002-8319-1652), Melis Ozge Alas Colak [3](ORCID 0000-0002-0546-087X), Rukan Genc [3,4](ORCID 0000-0002-9569-8776), Emre Erdem [1](ORCID 0000-0002-8395-0364)

[1] Department of Biomedical Engineering, Faculty of Engineering and Natural Sciences, Iskenderun Technical University, Hatay, Turkey
[2] Faculty of Engineering and Natural Sciences, Sabanci University, TR-34956, Istanbul, Turkey
[3] Sabanci University, SUNUM Nanotechnology Research Centre, TR-34956 Istanbul, Turkey.
[4] Department of Chemical Engineering, Faculty of Engineering, Mersin University, Mersin, Turkey.

*Corresponding E-mail: sgokhan.colak@iste.edu.tr
ahmet.gungor@sabaniuniv.edu

Abstract

In this paper, we offer a unique method for improving the electrochemical performance of supercapacitors by fabricating nanocomposites from a conductive polymer, polyaniline (PANI), and CZTS material. The paper describes the hydrothermal synthesis of CZTS and the subsequent formation of PANI: CZTS nanocomposites using in situ chemical oxidative polymerization. To create nanocomposites, 30% mass ratios of CZTS to aniline were used, and the results were characterized using methods such as X-ray diffraction (XRD), scanning electron microscopy (SEM), elemental mapping (EDX), FT-IR, and electron impedance spectroscopy and cyclic voltammetry. The addition of PANI with CZTS considerably improved the electrochemical characteristics of nanocomposites. PANI has specific capacitance, energy density, and power density values of 36 F/g, 5.1 Wh/kg, and $0.83x10^5$ W/kg in a symmetric supercapacitor design, respectively. Among of synthesized sample, the PANI: CZTS30 symmetric supercapacitor design displayed significant gains, with specific capacitance, energy density, and power density values of 168 F/g, 23.4 Wh/kg, and $1x10^5$ W/kg, respectively. The use of low-cost, readily manufactured materials such as PANI and PANI: CZTS nanocomposites offers promise for a variety of energy storage and conversion devices.

Keyword: CZTS, PANI, specific capacitance, energy storage, nanocomposite

INTRODUCTION

Polyaniline (PANI) and copper zinc tin sulfide (CZTS) are two fascinating materials in the field of energy storage, each having specific features and possible uses. PANI, a conducting polymer, has received substantial interest owing to its extraordinary electrical conductivity, as well as its capacity to undergo chemical and physical tuning, all of which make it appropriate for a broad variety of applications, especially in the area of energy storage.(Kumar, Sahoo and Panda, 2018) It belongs to the family of conducting polymers, distinguished by their propensity to conduct electricity after doping or oxidation. PANI contains many oxidation states, enabling diversity in its characteristics that make it well-suited for diverse applications, notably in the construction of batteries and supercapacitors. Its high electrode capacity and redox characteristics make it a good alternative for electrode materials, adding to its popularity in energy storage systems.(Wu *et al.*, 2021) However, PANI also confronts significant limitations, such as low cycle stability, particularly in battery applications, resulting to a steady loss in storage capacity with repeated charge and discharge cycles (Li *et al.*, 2008). This downside is linked to structural modifications that PANI suffers during cycling, resulting in mechanical strain and decreased performance. Additionally, PANI's inherent electrical conductivity is rather poor, needing doping to produce acceptable levels of conductivity, with the choice of dopant considerably impacting its electrochemical performance.

To overcome the difficulties associated with PANI in energy storage applications, current research has investigated numerous novel solutions. One of these ways is the integration of PANI with conductive materials to boost its conductivity and structural integrity while minimizing ion diffusion lengths. This coupling of PANI with other sophisticated materials, such as metal oxides or sulfides(Jadhav, Mane and Shinde, 2020; Lokhande, Chavan and Pandey, 2020), has led to the creation of hybrid systems, seeking to boost energy storage performance and stability. These hybrid systems use the unique features of both PANI and the selected additive material to achieve synergistic effects that result in increased overall performance. Transition metal chalcogenides, for instance, have attracted great interest in recent decades for their remarkable performance as electrodes in supercapacitors. These materials offer advantages such as enhanced redox chemistry, extended cycle lifespans, increased electronic conductivity, and a range of structural benefits, including flexibility, larger surface area, and denser active sites, all contributing to higher specific capacitance compared to their oxide and nitride counterparts(Wu and Ma, 2018).

Copper zinc tin sulfide (CZTS), among the main chalcogenides within the category of metal sulfide materials, has gained great interest owing to its exceptional features, notably in the context of thin film solar cells. CZTS boasts various positive qualities, including low toxicity, plentiful component elements, and attractive optoelectronic characteristics, giving it a very promising material for solar energy conversion. Its direct bandgap of roughly 1.5 eV places it as an effective absorber of light in the visible spectrum, making it a promising contender for solar energy absorption(Roy *et al.*, 2018). Furthermore, CZTS has a high absorption coefficient, significant light scattering, and prolonged carrier lifetime, all of which contribute to the possibility for high device efficiency. However, obstacles exist in the manufacture of high-quality CZTS films with regulated composition and shape, as well as in the construction of acceptable device topologies. Therefore, continuing research attempts concentrate on increasing CZTS synthesis and processing techniques while extending our grasp of its core optoelectronic features and the subtleties of device physics(Guo *et al.*, 2014).

Beyond its use in solar cells, CZTS has proven tremendous promise in the realm of energy storage, spanning batteries, capacitors, and supercapacitors. In the world of rechargeable batteries, CZTS may operate as both an anode and a cathode material, delivering a high theoretical capacity and outstanding cycle stability, which are key features for long-term energy storage solutions(Zhou *et al.*, 2013). Furthermore, CZTS-based capacitors and supercapacitors display high energy and power densities, respectively, due to their vast surface area and the ability to store charge efficiently at the interface between the electrode and the electrolyte. Importantly, CZTS materials have the potential to overcome difficulties related with the toxicity and restricted availability of certain metals often employed in conventional electrode materials, such as cobalt and lithium(Ito, 2013).

Despite these promising qualities, the performance of CZTS-based energy storage devices remains hampered by issues linked to the synthesis of high-quality CZTS materials and the design of acceptable device topologies. To increase the electrochemical properties of CZTS, researchers have investigated numerous approaches. Surface modification using conductive materials, such as carbon nanotubes or graphene, has proved beneficial in enhancing electrical conductivity(Permatasari *et al.*, 2021). Doping CZTS with materials, particularly iron, has been examined to increase its inherent electrical characteristics. Modifying synthesis parameters, such as temperature and precursor ratios, has shown promise in modifying the composition, crystallinity, and morphology of CZTS materials, consequently boosting their electrochemical capabilities. Nanostructuring techniques, including the use of nanoparticles or nanowires, have enlarged the surface area, giving extra active sites for electrochemical reactions.

Copolymerization of CZTS with conductive polymers has emerged as a viable technique to increase the electrochemical properties of CZTS-based products(Chen *et al.*, 2013). The incorporation of conductive polymers, such as polypyrrole or polythiophene, into CZTS matrices has the potential to boost electronic conductivity, hence increasing the electrochemical activity of the resultant nanocomposite material. Additionally, copolymerization of CZTS with conductive polymers may lead to greater mechanical stability and flexibility, making these materials appropriate for use in flexible and wearable energy storage devices. However, the synthesis of CZTS-conductive polymer composites demands careful control of reaction conditions to enable the uniform distribution of the conductive polymer inside the CZTS matrix while keeping the optimum electrochemical properties of both components. Therefore, more research efforts are necessary to completely understand the fundamental principles of copolymerization and to develop feasible synthesis procedures for CZTS conductive polymer composites that display superior electrochemical performance. In a pioneering investigation, the current research analyses the impacts of integrating CZTS into the conductive

organic polymer PANI within the context of energy storage applications, examining its possibilities beyond the world of solar cells. Specifically, the research examines the electrochemical performance of nanocomposite materials generated by altering the mass ratios of CZTS and PANI. To completely examine their performance and appropriateness for energy storage, symmetrical supercapacitors were fabricated and evaluated. In conclusion, this scholarly discourse underlines the varied potential of PANI and CZTS in energy storage applications, illuminating their particular advantages, present obstacles, and options for development. It underlines the continuous research initiatives aimed at leveraging the unique features of these materials and creating novel solutions for the creation of enhanced energy storage systems capable of addressing the expectations of future energy storage technologies.

MATERIALS AND METHODS

All chemicals used in the materials' production were utilized without being purified. Commercial supplies included $CuCl_2$ (Sigma Aldrich), $ZnCl_2$ (Across Organics), $SnCl_2$ (TCI Chemical), and Thiourea ($CS(NH_2)_2$) (Across Organics). The monomer was aniline hydrochloride (Thermo-scientific 99% purity, molecular weight-Mw-129.59 g/mol), while the initiator was ammonium persulfate (Thermo-scientific APS, 98% purity, molecular weight 228.19 g/mol). Sigma Aldrich supplied sulfuric acid (95-98 wt%, Mw=98.074 g/mol) as a solvent. A Millipore Milli-Q system (Millipore Inc., = 18 M cm) was utilized to purify the water used throughout the synthesis.

CZTS was generated utilizing a well-known solution-based traditional synthesis process. To make the initial solution, sufficient quantities (molar ratio=2:1:1:8) of copper chloride ($CuCl_2$), zinc chloride ($ZnCl_2$), tin chloride ($SnCl_2$), and thiourea ($CS(NH_2)_2$) were added in the combination DMSO solution with a solvent. The solution was then heated in a controlled environment (200°C) to generate the necessary kesterite phase. After that, the substance was pureed by washing it several times with ethanol and water, resulting in a black solid powder. In the production of PANI and PANI: CZTS 30 nanocomposite, a conventional in situ chemical oxidative copolymerization process was utilized (Figure 1). Firstly, aniline monomer (2.8 mmol) was dissolved in 10 mL of 1 M H_2SO_4 solution for 10 minutes. In another beaker, APS (1.85 mmol) is put in 5 mL of 1M H_2SO_4 and dissolved by stirring for 10 minutes. The APS solution was carefully added dropwise to the prepared aniline solution and stirred at room temperature for 10 minutes at a steady pace. With the addition of APS, the polymerization process commenced, PANI was produced, and the solution colour became dark green. The solution is maintained at 4°C for 24 hours to get pure PANI. The products were collected by centrifugation and rinsed with ethanol and deionized water. The finished product was dried at 70°C for one day. The same procedure was used to make PANI: CZTS 30 nanocomposite, where the ratio of aniline to CZTS was 30:1 (mass%). CZTS powder is added to a beaker containing 10 mL of 1 M H_2SO_4 and swirled in a magnetic stirrer for 30 minutes. The aniline solution made in another beaker is then added to the beaker holding the CZTS solution, and the ensuing stages are done in the same way as in the synthesis of pure PANI. The synthesized substance was designated PANI: CZTS30.

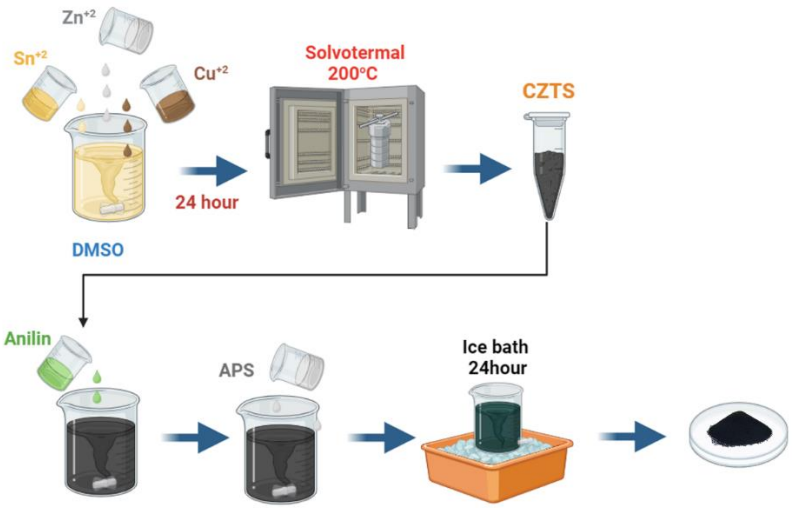


Figure 1. Synthesis procedure of PANI, CZTS, PANI: CZTS 30 materials

DRAFT

RESULTS and DISCUSSION

In the present research, a variety of sophisticated analytical methods was employed to fully analyze the structural, chemical, and electrochemical properties of materials under investigation, primarily focusing on PANI (Polyaniline) and its composites with CZTS (Copper Zinc Tin Sulfide) with potential implications for supercapacitor applications. X-ray Diffraction (XRD) investigation was undertaken utilizing a high-resolution PANalytical EMPYREAN model equipped with Cu Kα radiation source (λ = 1.5406 Å) to reveal the crystallographic characteristics and phase composition of CZTS. Raman spectroscopy, employing a Renishaw InVia Qontor apparatus with an excitation wavelength of 532 nm at room temperature, aided the examination of molecular vibrations, revealing insights into the structural features of the materials. Fourier Transform Infrared (FTIR) spectroscopy carried out using a Thermo Scientific iS50 equipment in the spectral region of 400-4000 cm^{-1}, permitted the recognition of surface functional groups and transmittance measurements. Scanning Electron Microscopy (SEM) examination, done using an FEI Quanta 650 model with gold-plated samples, was useful in visualizing the morphology and surface characteristics of PANI, CZTS, and PANI: CZTS composites. The electrochemical investigation, done utilizing BioLogic VMP 300 electrochemical devices with 6M KOH as the electrolyte and Whatman glass microfibers as dielectric separators, investigated the electrochemical performance of supercapacitors built using PANI and PANI: CZTS nanocomposites. Methods such as the use of cyclic voltammetry (CV), potentiostatic electrochemical impedance spectra (PEIS), were applied within a voltage range of 0 to +1 V at varying scan rate. Overall, this multimodal methodology allowed for a full assessment of the materials' feasibility for electrochemical energy storage applications.

Furthermore, we undertook a meticulous synthesis of PANI: CZTS (Polyaniline: Copper Zinc Tin Sulfide) nanocomposites utilizing a low-temperature oxidation-based polymerization method. CZTS was introduced into the polymer matrix at varying mass ratios, comprising 30% of the resulting composite. The overarching objective of this research endeavor was to facilitate the intercalation of CZTS within the PANI matrix, with the anticipated outcome of enhancing the electrochemical performance of the resultant composite material. This intercalation mechanism was based on the postulate of electrostatic and Van der Waals interactions occurring between PANI and CZTS constituents. The structural characterization of the synthesized materials was meticulously conducted through X-ray diffraction (XRD) analysis, a robust technique for extracting vital crystallographic information. We present below our findings from this analysis pertaining to PANI, CZTS, and the PANI: CZTS nanocomposites.

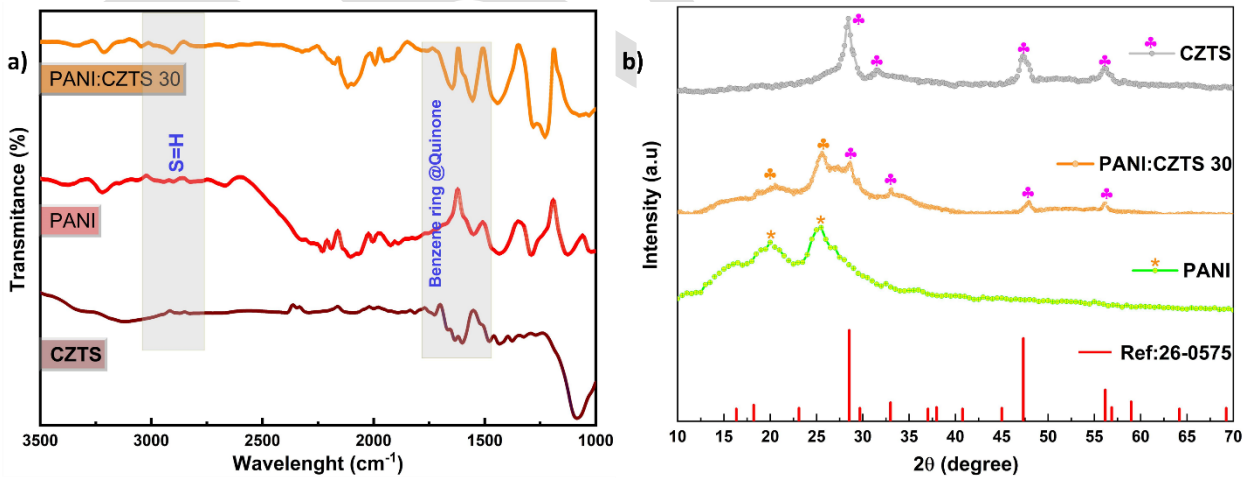


Figure 2. FTIR (a) and XRD (b) analysis graph of synthesized materials

Primarily, the XRD pattern for pure PANI revealed the presence of two distinctive, well-defined peaks at specific diffraction angles, unambiguously attributed to the predominant 113 and 322 hkl miller indices crystallographic planes. The sharpness of these peaks alluded to a substantial degree of polymer chain alignment within these crystallographic planes, while peak intensity correlated with the population of crystalline domains within these planes. Subsequently, the XRD pattern for CZTS exhibited a series of

discernibly sharp and well-aligned kesterite peaks at specific 2θ values, corresponding precisely to the (112), (200), (220), and (312) lattice planes(Chen *et al.*, 2013). This unequivocal alignment confirmed the tetragonal crystal phase of CZTS, aligning precisely with the JCPDS card reference, thereby attesting to the high purity of the synthesized material. Lastly, upon analyzing the XRD patterns of the PANI: CZTS nanocomposites, wherein CZTS constituted 30% of the composite mass, it was apparent that the characteristic peaks of both PANI and CZTS were distinctly present within the synthesized composite materials. This observation strongly indicates that the PANI and CZTS constituents retained their inherent crystal structures even within the composite framework. It is crucial to underscore that the synthesis methodology and subsequent structural characterizations presented herein adhere rigorously to established protocols. They collectively provide a solid foundation for further inquiries into the electrochemical properties and potential applications of PANI: CZTS nanocomposites, thus contributing to the burgeoning field of advanced materials science.

The Fourier-transform infrared (FTIR) spectroscopy analysis undertaken in this work has shown to be significant in clarifying the composition and structural properties of the synthesized materials. Notably, the FTIR spectrum of the raw PANI material displayed five conspicuous distinctive peaks. Specifically, the peaks at around 1556 and 1461cm^{-1} were linked to the C=C stretching vibrations associated with quinonoid and benzenoid rings, respectively. Furthermore, the appearance of a distinct peak at 1292 cm^{-1} showed the production of the PANI emeraldine salt phase. In addition, the peaks detected at 1049 and 800cm^{-1} were suggestive of the presence of sulfonate groups connected to aromatic rings, leading to an increased proton conductivity, and C-H out-of-plane bending vibrations inside the PANI structure, respectively. The observation of C–C bond stretching vibrations within the spectrum region of 1550 cm−1 to 1875 cm^{-1} further validated the presence of benzoid and quinoid units indicative of the emeraldine form inside the synthesized PANI material. Moreover, the FTIR analysis showed discrete infrared bands covering the range of 1150 cm^{-1} to 1490 cm^{-1}, corresponding to C-N and C-H bending vibrations occurring at the C1 and C4 locations of the benzene ring. Additionally, a faint band at 500 cm^{-1} was found, confirming the existence of C double bonds and the Zn–S bond, so conclusively proving the effective integration of CZTS nanoparticles into the FTIR spectrum of the nanocomposites. Notably, a faint vibration band at around 2900 cm^{-1}, due to the S-H vibration in CZTS nanoparticles, emphasized the presence of surface hydroxyl groups, possibly coming from ambient moisture adsorption (Dongyun *et al.*, 2014). In summation, the application of FTIR spectroscopy in this study has provided comprehensive insights into the molecular vibrations and functional groups within both PANI and CZTS constituents, thereby corroborating the effective synthesis of PANI: CZTS nanocomposites and affording valuable revelations concerning their structural composition.

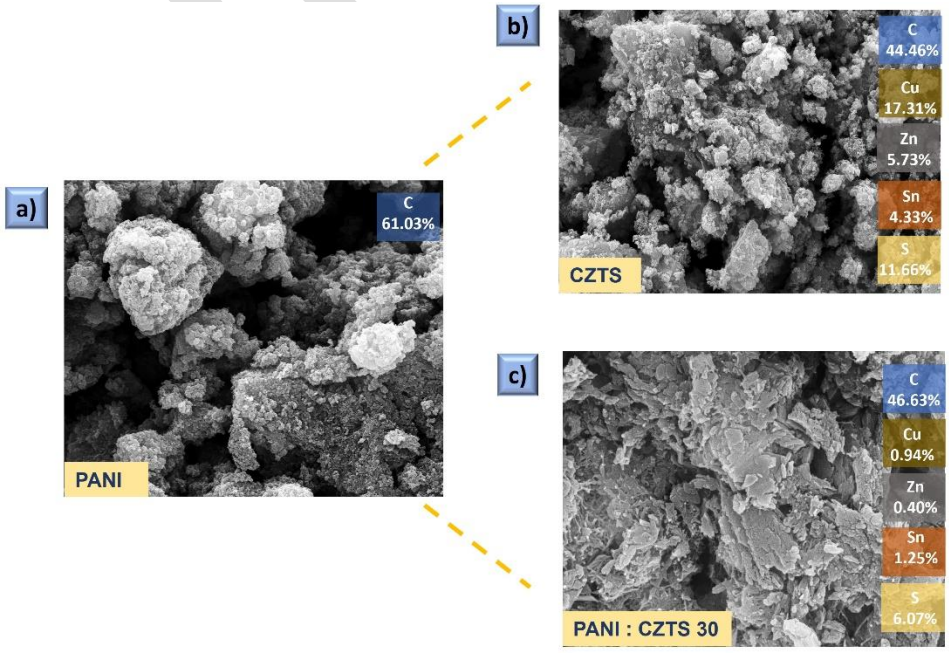


Figure 3. SEM image and EDX analysis all synthesized samples

Moreover, the morphological characteristics of several materials, including pure CZTS, PANI (Polyaniline), and PANI: CZTS nanocomposites at different mass ratios (referred to as PANI: CZTS 30) were comprehensively examined using scanning electron microscopy (SEM) and energy-dispersive X-ray (EDX) spectroscopy. These investigations gave useful insights into the composition, structural characteristics, and elemental distribution within the synthesized materials. The SEM images, as represented in Figure 3, were collected at a scale of 1 µm. A remarkable and constant finding across all samples was the uniform dispersion of particles, reflecting a good synthesis process. The photos verified the production of CZTS, PANI, and their nanocomposite structures with various PANI to CZTS ratios. This homogeneity in particle dispersion emphasized the efficiency of the synthesis approach applied in creating well-dispersed nanocomposites. In addition to SEM examination, EDX spectroscopy was performed to confirm the composition and production of the nanocomposite materials. This approach enabled for exact elemental analysis by measuring atomic percentages. Notably, the EDX spectra for the PANI: CZTS nanocomposites indicated the existence of PANI and CZTS ingredients in the samples.Specifically, the EDX examination indicated the following atomic percentages for the different materials: For pure PANI, carbon (C) was the dominating element, totaling 61.3% of the substance. In the instance of CZTS, the composition comprised of carbon (C: 44.46%), copper (Cu: 17.31%), zinc (Zn: 5.73%), tin (Sn: 4.33%), and sulfur (S: 11.66%). Turning to the PANI: CZTS nanocomposites, the EDX analysis characterised their compositions as follows: PANI: CZTS 30: Carbon (C: 46.63%), copper (Cu: 0.94%), zinc (Zn: 0.40%), tin (Sn: 1.25%), and sulfur (S: 6.07%). The EDX study therefore gave convincing evidence of CZTS incorporation into the conductive PANI matrix inside the nanocomposites. Furthermore, the presence of transition metals such as copper, zinc, and tin indicated the effective creation of the PANI: CZTS nanocomposite material.

In order to investigate the electrochemical properties of PANI, CZTS, and PANI: CZTS:30 composite materials, electrochemical impedance spectroscopy (EIS) and cyclic voltammetry (CV) analyses of different materials were conducted using a two-electrode system. PANI, CZTS, and PANI: CZTS30 samples were utilized as electrode materials in the electrochemical analyses. A 6M KOH solution was employed as the electrolyte. CV analysis was conducted in the range of 0-1 V. The designed supercapacitors were characterized through potentiostatic electrochemical impedance spectroscopy (PEIS) analyses and cyclic voltammetry (CV) measurements, employing a symmetric design, and the prepared designs are presented in Table X.

Table 1. The specific supercapacitor designs based on these synthesized nanocomposites.

Electrode 1	Electrode 2	Electrolyte	Separator	Type
PANI	PANI	6M KOH	Glass fiber	Symmetric
CZTS	CZTS	6M KOH	Glass fiber	Symmetric
PANI: CZTS30	PANI: CZTS30	6M KOH	Glass fiber	Symmetric

In energy storage systems, one of the most critical factors in evaluating the processes occurring at the electrode/electrolyte interface is the characterization of capacitive behavior. To examine the charge/discharge processes taking place at the electrode/electrolyte interface, electrochemical impedance spectroscopy (EIS) analysis is commonly employed. Through PEIS analysis, the impedance of the energy storage system is determined at different frequency values, allowing for the characterization of capacitive behavior.

As a result of the PEIS analysis conducted on PANI, CZTS, and PANI: CZTS:30 composite materials, Nyquist plots were obtained, as shown in Figure 4. The semi-circular shape of the Nyquist plots in the high-frequency region provides insights into the ion transfer rate of the electrode used and the charge transfer resistance occurring at the electrode/electrolyte interface. It is noteworthy that in the high-frequency region, a semi-circular shape is not observed in all three plots. Therefore, this result implies an increase in the ion transfer rate at the electrodes used during the measurements and a decrease in the charge transfer resistance at the electrode/electrolyte interface.

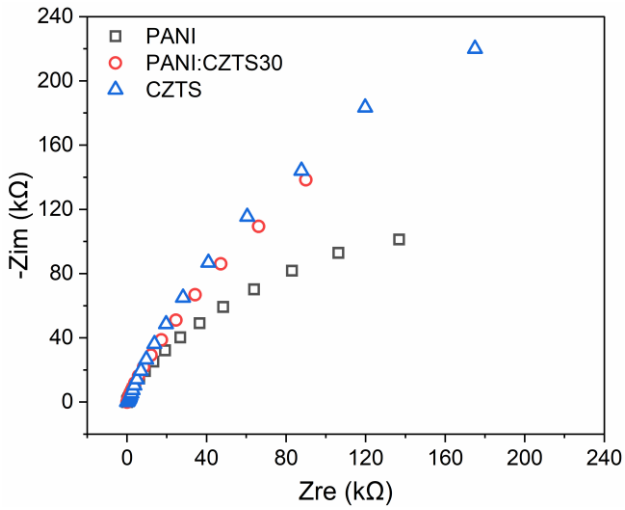


Figure 4. Nyquist plots of each design

Cyclic voltammetry (CV) analysis is another important method for determining the capacitive behavior of the electrodes used. Additionally, CV can be used to characterize the material's redox behavior. CV analysis provides information about the events occurring at the electrode/electrolyte interface during charge/discharge cycles. Figure 5 presents the CV curves of all synthesized samples at a scan rate of 10 mV/s. The reason for selecting a scan rate of 10 mV/s is that as the scan rate decreases, the area under the CV curve increases, resulting in a more consistent specific capacitance value. Therefore, CV analysis was conducted at a scan rate of 10 mV/s. Upon examining the CV curves of PANI, CZTS, and PANI: CZTS30 composites, it is apparent that all the curves are approximately rectangular in shape. This indicates that the process of charge storage in the electrode materials used in the designed supercapacitor primarily involves the formation of electrical double layers at the interface. Hence, it can be stated that the designed supercapacitor exhibits Electric Double-Layer Capacitor (EDLC) behavior based on the CV curves. Furthermore, the specific capacitance values calculated with the area under the CV curves for PANI, CZTS, and PANI: CZTS30 are 35.8, 67.6, and 168.1 F/g, respectively. This demonstrates a significant increase in specific capacitance as a result of the contribution of PANI to CZTS. Predictably, the synergistic interactions between PANI and CZTS lead to increased ion transfer rates, decreased interfacial charge transfer resistance, and enhanced electron diffusion rates.

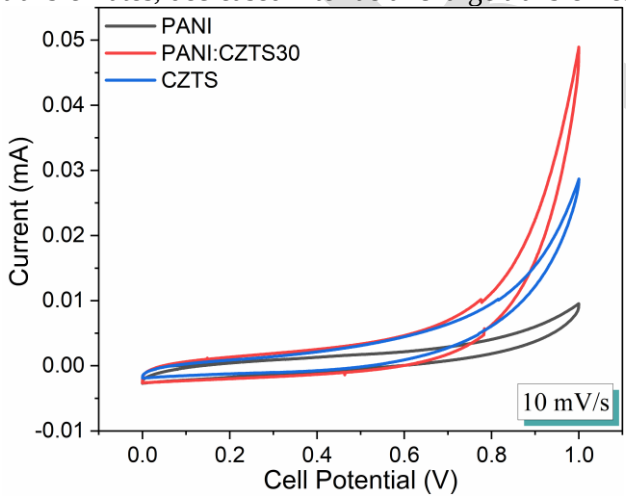


Figure 5. CV curves of PANI, CZTS and PANI: CZTS30 composites at 10 mV/s scan rate.

CONCLUSION

This study presents a novel approach for the synthesis of nanocomposites using the conductive polymer polyaniline (PANI) and CZTS material, aiming to enhance the electrochemical performance of supercapacitors. CZTS was synthesized via a hydrothermal method, while PANI: CZTS 30 nanocomposites were produced by in situ chemical oxidative polymerization with varying mass ratios of CZTS to aniline. The

materials were extensively characterized through X-ray diffraction, SEM, elemental mapping, FT-IR, and CV spectroscopic analyses, confirming the presence of CZTS in the synthesized PANI/CZTS nanocomposites. CV spectroscopy revealed the incorporation of CZTS particles, but significant changes in spectral intensity were observed. The electrochemical properties demonstrated substantial improvements when PANI was combined with CZTS. Specifically, PANI: CZTS 30 exhibited significantly enhanced specific capacitance value compared to pure PANI. In summary, this research offers valuable insights into the electrochemical performance of PANI and PANI: CZTS nanocomposites, highlighting their potential as cost-effective and efficient materials for energy storage applications. These materials hold promise for various energy-related devices, including high-performance supercapacitors, contributing to advancements in energy storage and harvesting technologies.

ACKNOWLEDGEMENTS

This study was partially supported by The Scientific and Technological Research Council of Türkiye (TUBITAK) (grant no. 121C416). Ahmet Güngör thanks TUBITAK for financial support.

REFERENCES

Chen S, Walsh A, Gong XG, and Wei SH, 2013. Classification of lattice defects in the kesterite Cu2ZnSnS4and Cu2ZnSnSe4earth-abundant solar cell absorbers. Advanced Materials, 25(11): 1522–1539.

Dongyun M, Guoying S, Hongzhi W, Qinghong Z, and Yaogang L, 2014. Controllable growth of high-quality metal oxide/ conducting polymer hierarchical nanoarrays with outstanding electrochromic properties and solar-heat shielding ability. Journal of Materials Chemistry A, 2(33): 13541–13549.

Guo BL, Chen YH, Liu XJ, Liu WC, and Li AD, 2014. Optical and electrical properties study of sol-gel derived Cu2ZnSnS4thin films for solar cells. AIP Advances, 4(9): 0–10.

Ito K, 2013. Copper Zinc Tin Sulfid- Based Thin Film Solar Cell. in Wiley, 561–563.

Jadhav V V., Mane RS, and Shinde P V., 2020. *Electrochemical Supercapacitors: History, Types, Designing Processes, Operation Mechanisms, and Advantages and Disadvantages* SpringerBriefs in Materials.

Kumar N, Sahoo PK, and Panda HS, 2018. Localize current burst in modified carbon nanotube/polyaniline composite fibers mat electrode miniaturized resistance and improved rate capability for solid-state supercapacitor. Journal of Materials Science: Materials in Electronics, 7562–7574.

Li Q, Wu J, Tang Q, Lan Z, Li P, Lin J, and Fan L, 2008. Application of microporous polyaniline counter electrode for dye-sensitized solar cells. Electrochemistry Communications, 10(9): 1299–1302.

Lokhande PE, Chavan US, and Pandey A, 2020. *Materials and Fabrication Methods for Electrochemical Supercapacitors: Overview* Electrochemical Energy Reviews. Springer Singapore.

Permatasari FA, Irham MA, Bisri SZ, and Iskandar F, 2021. Carbon-Based Quantum Dots for Supercapacitors: Recent Advances and Future Challenges. Nanomaterials, 11(1): 91.

Roy A, Devi PS, Karazhanov S, Mamedov D, Mallick TK, and Sundaram S, 2018. A review on applications of Cu2ZnSnS4 as alternative counter electrodes in dye-sensitized solar cells. AIP Advances, 8(070701).

Wu K, Fu P, Wang Z, Zhao Q, Guo J, Ruan B, and Wu M, 2021. The effect of polyaniline electrode doped with transition metal ions for supercapacitors. Polymers for Advanced Technologies, 32(5): 2082–2092.

Wu M, and Ma T, 2018. Transition Metal Compound Electrocatalysts for I-Mediated Dye-Sensitized Solar Cells 7 . 2 Transition Metal Compound Counter Electrode Catalysts for Iodide Redox Couple in DSSCs., 155–176.

Zhou H, Hsu WC, Duan HS, Bob B, Yang W, Song T Bin, Hsu CJ, and Yang Y, 2013. CZTS nanocrystals: A promising approach for next generation thin film photovoltaics. Energy and Environmental Science, 6(10): 2822–2838.

DRAFT

ORAL PRESENTATION

Covid-19 Hastalığında Bazı Sitokinlerin Rolleri

Elisha Apatewen AKANBONG[1] (*ORCID: https://orcid.org/0000-0002-2556-7236*), Alparslan Kadir DEVRİM[2] (*ORCID: https://orcid.org/0000-0002-3293-7290*), Ali ŞENOL[1*] (*ORCID: https://orcid.org/0000-0003-4080-7776*), Tuba DEVRİM[3] (*ORCID: https://orcid.org/0000-0002-5321-2002*)

[*1]Kırıkkale Üniversitesi, Veteriner Fakültesi, Biyokimya Anabilim Dalı, Kırıkkale, Türkiye
[2]İzmir Bakırçay Üniversitesi, Menemen Meslek Yüksekokulu, Veterinerlik Bölümü, İzmir, Türkiye
[3]İzmir Bakırçay Üniversitesi, Tıp Fakültesi, Patoloji Anabilim Dalı, İzmir, Türkiye

*Sorumlu yazar e-mail: alisenol712@gmail.com
alisenol@kku.edu.tr

Özet

Sitokin salınım sendromu (SSS) veya sitokin fırtınası, SARS-CoV-2 tarafından başlatılabilen, bağışıklık sisteminin orantısız tepkisinden kaynaklanan proinflamatuar sitokinlerin aşırı üretiminin bir sonucudur. SARS-CoV-2'nin neden olduğu COVID-19, sitokinlerle korelasyon göstermektedir. SARS-CoV-2, yaygın dağılım gösteren makrofajlar ve mast hücreleri sayesinde IL-1 üretimini tetiklemektedir. IL-1 ise, IL-6 ve TNF-α üretimlerini etkileme eğilimindedir. COVID-19 şiddetinin ilerlemesi, IL-6 gibi bazı sitokin düzeylerini etkiler. IL-6, SSS oluşumundan başlıca sorumlu olan sitokindir. SSS, COVID-19 ile ilgili komplikasyonların ve COVID-19 ile ilişkili ölümlerin ana nedenidir. Bu zamana kadar literatürde bildirilmiş verilere rağmen, SARS-CoV-2 ve sitokinler arasındaki ilişki tam olarak aydınlatılmış değildir. Bu çalışma ile söz konusu ilişkinin irdelenmesi amaçlamıştır. COVID-19 tedavisi sırasında sitokinlerin hedeflenmesi, hastaların hayatta kalma oranlarını artırma ve COVID-19 ile ilişkili ölümleri azaltma potansiyelini taşımaktadır. COVID-19 hastalığında, sitokin salınım mekanizmalarına ve salınan sitokinlerin etkilerine odaklanılmasının, özellikle T lenfositler üzerindeki etkilerinin ve IFN-γ üretiminin irdelenmesinin, hastalığın ölümcül etkilerini azaltmaya yardımcı olabileceği düşünülmektedir.

Anahtar Kelimeler: COVID-19, Sitokinler, Sitokin Salınım Sendromu, Sitokin Fırtınası

Roles of Some Cytokines in Covid-19 Disease

Abstract

Cytokine release syndrome (CNS), or cytokine storm, is the result of overproduction of pro-inflammatory cytokines resulting from a disproportionate response of the immune system, which can be initiated by SARS-CoV-2. COVID-19 caused by SARS-CoV-2 correlates with cytokines. SARS-CoV-2 triggers IL-1 production by widely distributed macrophages and mast cells. IL-1, on the other hand, tends to affect the production of IL-6 and TNF-α. The progression of COVID-19 severity affects the levels of some cytokines, such as IL-6. IL-6 is the cytokine primarily responsible for CNS formation. CNS is the leading cause of COVID-19-related complications and COVID-19-related deaths. Despite the data reported in the literature to date, the relationship between SARS-CoV-2 and cytokines has not been fully elucidated. This review aims to examine this relationship. Targeting cytokines during COVID-19 treatment holds the potential to increase patient survival rates and reduce COVID-19-related deaths. It is thought that focusing on the mechanisms of cytokine release and the effects of released cytokines in COVID-19 disease, especially examining their effects on T lymphocytes and IFN-γ production, may help reduce the fatal effects of the disease.

Keywords: COVID-19, Cytokines, Cytokine Release Syndrome, Cytokine Storm

DRAFT

GİRİŞ

Coronavirüs hastalığı 2019 (COVID-19), yeni insan koronavirüsü Şiddetli Akut Solunum Sendromu Coronavirüs 2'den (SARS-CoV-2) kaynaklanır. SARS-CoV-2, zarflı tek sarmallı bir RNA genomu olan Coronaviridae familyasına aittir (Li ve ark., 2020). Aralık 2019'da Wuhan-Çin'de ortaya çıkmasının ardından hızla 196'dan fazla ülkeye yayılarak Dünya Sağlık Örgütü (WHO) tarafından küresel sağlık acil durumu olarak ilan edilmiştir (Darif ve ark., 2020, Kunnumakkara ve ark., 2021). Araştırmalar SARS-CoV-2'nin zoonotik bir kökene sahip olduğunu ve yarasaların SARS-CoV-2'ye benzer şekilde önemli koronavirüs rezervuarları olduğunu öne sürmüşlerdir. SARS-CoV-2'nin insandan insana bulaşması, enfekte bir insandan öksürme ve hapşırma sırasındaki aerosol damlacıklarının doğrudan temas yoluyla olabilir. Kontamine yüzeyler de virüsün dolaylı olarak bulaşmasına katkıda bulunmaktadır (Karabacak ve Kırdemir, 2021). SARS-CoV-2 üst ve alt solunum yollarına saldırarak çoğalmaktadır. Enfekte kişilerde 2 hafta içinde semptomların görüldüğü kuluçka süresi bulunmaktadır (Chan ve ark., 2020). Enfekte kişilerin bağışıklık durumlarına bağlı olarak, hafiften, orta dereceye kadar değişen semptomlar bulunmaktadır. Bununla birlikte, yaşlı bireyler ve diyabet, kronik solunum yolu hastalığı, kanser ve kardiyovasküler hastalık gibi kronik hastalıkları olan hastalarda, zayıf bağışıklık sistemlerinin bulunmasından dolayı, ciddi semptomlar ortaya çıkabilmektedir. Bu hastalarda gelişen enfeksiyon, pnömoni ve akut solunum sıkıntısı sendromuna (ARDS) yol açarak organ yetmezliği ve ölümle sonuçlanabilmektedir (Karabacak ve Kırdemir, 2021).

Eskiden kaynak hücre ile ilişkilerini tanımlamak için lenfokinler ve monokinler olarak adlandırılan sitokinlerin, neredeyse tüm nükleuslu hücrelerce sentezlenebildikleri kesinleşince günümüzde 'sitokinler' olarak adlandırılmışladır (Dinarello, 2000). Sitokin terimi sito (hücre) ve kinos (hareket) olan iki Yunanca kelimeden köken almaktadır (dos Santos ve ark., 2020). İmmün hücreler, endotelyal hücreler, fibroblastlar ve diğer stromal hücreler (Fares ve ark., 2020) tarafından salgılanan, pleiotropik ve hücreye özgü fonksiyonlara sahip düşük molekül ağırlıklı protein olan sitokinler, hücresel iletişim ve hücresel yanıtların koordinasyonunda görev almaktadır (Chauhan ve ark., 2021, Devrim ve ark., 2020). Ayrıca sitokinler ilgili hücrelere büyüme, farklılaşma, inflamatuar veya antiinflamatuar sinyaller de sağlamaktadır ve çoğu zaman uyarılara yanıt olarak belirli periyotlarda salgılanırlar (Berraondo ve ark., 2019, Devrim ve ark., 2020). Sitokinlerin dolaşımdaki yarı ömürleri sınırlı olduğundan etki süreleri kısadır. Tipik olarak sitokinler otokrin veya parakrin etki gösterir (Berraondo ve ark., 2019, Simpson ve ark., 2020). Bununla birlikte uzak hücrelere de sinyal göndererek endokrin aktivite sergileyebilirler (Katze, 2012). Sitokin geninin ekspresyonu, bazal homeostatik koşullar altında sıkı bir şekilde düzenlenmekte olup çoğu zaman sürekli değildir. Sitokin geninin transkripsiyon ve translasyon oranları, büyüme faktörleri, yabancı uyaranlar ve/veya sitokinler (Fares ve ark., 2020) gibi çeşitli uyaranlardan (Chauhan ve ark., 2021) etkilenebilir. Sitokinler bağışıklık hücrelerinin aktivasyonunda esastır, yani bağışıklığa çok büyük katkı sağlarlar. Bununla birlikte salınımları düzenlenmediğinde birçok inflamatuar hastalığın (Fares ve ark., 2020) ve sitokin salınım sendromunun (CRS) veya sitokin fırtınasının (Shimabukuro-Vornhagen ve ark., 2018) ortaya çıkmasını etkilerler. CRS, SARS-CoV-2 tarafından tetiklenebilen bağışıklık sisteminin orantısız tepkisinden kaynaklanan proinflamatuar sitokinlerin aşırı üretiminin bir sonucudur. CRS'nin bazı hastalarda COVID-19'un ciddiyeti ve zararlı etkilerinin ortaya çıkmasında rol oynadığı gösterilmiştir (Darif ve ark., 2020, Tang ve ark., 2020, Ye ve ark., 2020). Bu çalışma ile SARS-CoV-2 ile sitokinler arasındaki ilişkinin değerlendirilmesi amaçlamıştır.

SARS-COV-2 TARAFINDAN SİTOKİN SALINIMININ BAŞLATILMASI

Anjiyotensin dönüştürücü enzim 2 reseptörü (ACE 2), solunum yolu epitel hücrelerinin hücre zarlarının yüzeyinde eksprese edilir. SARS-CoV-2'nin bu reseptöre bağlanması, proinflamatuar sitokinlerin salınmasını tetikler. Anjiyotensin II (Ang II) hormonu, vazokonstriktör olmasının yanı sıra pro-inflamatuar yanıtları da uyarır, böylece NF-κβ'yı yukarı düzenleyerek IL-6, IL-8 ve TNF-α dahil sitokinlerin salınmasını sağlar. Ang 1-7, renin-anjiyotensin sisteminin (RAS) ana modülatörü olan ACE 2'nin modülatör etkisi altında Ang II'den meydana gelen bir vazodilatördür. Bu nedenle, ACE2'nin inhibisyonu veya eliminasyonu sonucunda Ang 1-7 üretiminin bloke edilmesi, AngII seviyelerinde bir artışa yol açar ve bu da proinflamatuar sitokinlerin abartılmasına neden olur. Ayrıca Ang 1-7 hormonu hücreler tarafından antiinflamatuar sitokinlerin salınımını tetikleyerek akciğer hasarını önler (Mahmud-Al-Rafat ve ark., 2020). SARS-CoV-2 enfeksiyonu, ADAM Metallopeptidaz Alanı 17 (ADAM 18) aracılığıyla ACE 2 aktivitelerini inhibe eder. Bu sonuçta membrana bağlı ACE2'nin çözünür bir formda (sACE2) serbest bırakılmasıyla sonuçlanır. ADAM 17'nin aracılık ettiği SARS-CoV-2 tarafından ACE 2 üretiminin veya salınımının azaltılması, Ang II'den Ang 1-7 sentezini engeller. Bu nedenle, COVID-19 hastalarında Ang II seviyesi artar ve bu da vücuttaki pro-inflamatuar yanıtı artırma

veya şiddetlendirme eğilimindedir. Dolayısıyla, COVID-19 hastalarında Ang II düzeyinin artması, hastalardaki pro-inflamatuar yanıtı artırma veya şiddetlendirme eğiliminde olup, sonuçta COVID-19 hastalarında organ veya çoklu organ yetmezliğinin ortaya çıkmasına neden olmaktadır (Mahmud-Al-Rafat ve ark., 2020). Virüsün kendisinin yanı sıra bozunmuş ürünleri de CRS'nin ortaya çıkmasını tetikleyebilir. SARS-CoV-2'nin germline tarafından kodlanan konak sensörleri ve pattern tanıma reseptörleri (PRR'ler) tarafından tanınması, hem doğal hem de adaptif bağışıklık tepkilerini uyarmak için sitokinlerin salınmasında hayati bir rol oynar (Sallenave ve ark., 2020). CRS, COVID-19 hastalarında sık görülen bir durum olup IL-1 tarafından başlatılmaktadır. SARS-CoV-2, makrofajlar ve mast hücreleri (MC'ler) (Conti ve ark., 2020) tarafından IL-1 salgılanmasını indükler ve SARS-CoV-2 tarafından indüklenen IL-1 salgılanması, hem şiddetli hem de şiddetli olmayan hastalarda yüksek olarak bulunmaktadır (Huang ve ark., 2020). IL-1 salgılanmasını takiben TNF-α ve IL-6'nın salgılanmasını da tetikler (Conti ve ark., 2020). IL-6 büyük ölçüde CRS'nin nedenidir. IL-6'nın iki ana reseptörü vardır; IL-6R ve çözünür form olan SIL-6R. Aynı zamanda klasik ve trans-sinyalizasyon yolu olmak üzere iki ana sinyal yoluna sahiptir. COVID-19 hastalarında IL-6, SIL-6R'ye (trans-sinyal yolu) bağlanarak IL-6/SIL-6R kompleksini oluşturur ve bu da gp130'un aktivasyonuyla sonuçlanır. GP130'un aktivasyonu, Janus kinaz (JAK)/sinyal transdüserleri, transkripsiyon aktivasyonu (STAT) ve fosfatidilinositol-3 kinaz yolu gibi birçok hücre içi yolun tetiklenmesine yol açarak tüm hücreler tarafından proinflamatuar sitokinlerin salınmasına neden olur (Darif ve ark., 2020, Ebihara ve ark., 2011). IL-6 aynı zamanda sitokinlerin (CRS) aşırı ekspresyonunu tetikleyebilen serum amiloid A (SAA) gibi pozitif akut faz reaktanlarının sentezini ve üretimini de indükler ve bu da diyabet gibi kronik hastalıkları olan bireylerin COVID-19 şiddetiyle karşılaşma riski daha fazla olduğunu açıklamaktadır. IL-6 tarafından indüklenen SAA üretimi daha sonra eritrosit (RBC) aglütinasyonu yoluyla pulmoner ve doku gaz değişiminin azalması, fibrinojene bağlanma yoluyla atipik koagülopati ve yüksek yoğunluklu lipoprotein (HDL) fonksiyon bozukluğu veya lipoproteinlere bağlanarak ateroskleroz gibi kompliasyonlara neden olur (Chatterjee ve ark., 2020).

IL-6'nın pozitif akut faz reaktanları üzerindeki etkisine ek olarak, hepatositlerde C-reaktif protein (CRP) geninin transkripsiyonunu da tetiklemektedir. COVID-19'un erken evresinde CRP'deki yüksek seviyelerin akciğer hasarını ve hastalığın ciddiyetini göstermektedir. CRP aynı zamanda birkaç organ sisteminin etkilenen ciddi COVİD-19 olan hastalarda ve ölen kişilerde de yüksek seviyelerde bulunmuştur. CRP, aktivasyon komplemanı, pro-inflamatuar sitokin üretiminin indüksiyonu ve apoptoz yoluyla COVID-19'un ciddi sonuçlarının ortaya çıkmasına katkıda bulunabilir (Mosquera-Sulbaran ve ark., 2021).

COVID-19'da SİTOKİN FIRTINASI

Yeni insan koronavirüsü SARS-CoV-2 de dahil olmak üzere enfeksiyonlar, CRS'ye veya sitokin fırtınasına neden olan bağışıklık tepkisinin hiperaktivitesini tetiklemektedir. CRS, B hücreleri, T hücreleri, nötrofiller, makrofajlar, monositler, dendritik hücreler, NK hücreleri gibi birçok beyaz kan hücresinin ve proinflamatuar sitokinlerin aşırı miktarda bulunması olark tanımlanmaktadır (Azkur ve ark., 2020, Behrens ve ark., 2017). Bu anormallik, kemokinlerin, koloni uyarıcı faktörlerin (CSF'ler), interlökinlerin (IL'ler), interferonların (IFN'ler) ve tümör nekroz faktörü-alfa'nın (TNF-a) aşırı ekspresyonunu içerir. Sitokin fırtınasının ARDS'nin ana nedeni olarak kabul edilmektedir. Ayrıca sitokin fırtınası ve buna bağlı komplikasyonlar, COVID-19'a bağlı ölümlerin ana nedeni olarak tanımlanmaktadır (Karabacak ve ark., 2021, Ye ve ark., 2020). İmmün yanıt ile COVID-19 şiddeti arasında bir korelasyon bulunmaktadır. T lenfositlerin; CD4+ T hücreleri ve CD8+ T hücreleri hafif, orta ve şiddetli tüm COVID-19 hastalarında azalmakta olup bu durum ciddi hastalarda daha belirgin bir azalma şeklindedir. Bu durum bu hücreler ile hastalığın şiddeti arasında negatif bir korelasyon olduğu anlamına gelmektedir. Buna IL-6, IL10, IL-2R ve TNF-α düzeylerinde belirgin bir yükselme eşlik etmektedir. Aksine, IFN-y'nin (Chen ve ark., 2019) daha düşük bir ifadesi bulunmaktadır ve bu muhtemelen CD4+ T hücre sayısındaki azalmanın bir sonucudur. Yine şiddetli ve aşırı şiddetli COVID-19 hastalarında T lenfositlerin (CD4+ ve CD8+ T hücreleri) ve B hücrelerinin mutlak sayısının kademeli olarak azaldığı, ağır hastaların sayılarının aşırı ağır hastalara göre daha yüksek olduğu bildirildi. Buna rağmen, IFN-y üreten CD8+ T hücrelerinin yüzdesi hem şiddetli hem de aşırı şiddetli hastalarda hafif hastalara göre artmaktadır. Ayrıca aşırı şiddetli hastalarda IFN-y üreten CD4+ T hücrelerinin yüzdesi de artmaktadır. Sonuç olarak, COVID-19 hastalarında IFN-γ üretimini de içeren T lenfositlerin sayısı ve işlevi tutarsızlık bulunmaktadır. Ayrıca aşırı şiddetli hastalarda IL6, IL-10 ve IL-2R'nin ekspresyonu arttığı tespit edilmiştir (Wang ve ark., 2020). Yapılan bir çalışmada 50 hem ağır hem de kritik COVID-19 hastasında IFN-α üretimi ve aktivitesinin düşük olduğu, IFN-β üretiminin ise olmadığı bildirilmiştir. Bu varyasyonlar kalıcı kan viral yükü ve aşırı eksprese edilen inflamatuar yanıt ile ilişkilendirilmektedir (Hadjadj ve ark., 2020). COVID-19 hastalarında daha yüksek

serum IL-6, IL-10, IL-2, IL-4, TNF-α ve IFN-γ seviyeleri gözlenmiştir. Ancak çalışmanın kritik hasta grubunda serum IL-6 ve IL-10 düzeyleri anlamlı derecede yüksek bulunmuştur. Ayrıca serum IL-10 düzeyleri C-reaktif protein (CRP) miktarı ile pozitif korelasyon göstermiştir (Han ve ark., 2020). 43 yetişkin COVİD-19 hastası (28 hafif ve 15 ağır hasta) üzerinde yapılan bir araştırma, IL-6'nın 2 grup arasında önemli ölçüde farklılık gösterdiğini ve en yüksek IL-6 düzeyine şiddetli grubun sahip olduğunu bildirilmiştir. Ayrıca IL-6, yetişkin hastalarda COVID-19 şiddetinin ortaya çıkmasıyla yakından ilişkili olarak gösterilmektedir (Gao ve ark., 2020). Başlangıçta COVID-19 testi pozitif çıkan bir ölen hasta grubu ve taburcu edilmiş olan hasta grubu üzerinde yapılan bir çalışmada, ölüm grubunun IL-6 düzeyinin, ölen grupta önemli ölçüde daha yüksek olduğunu gösterilmiştir. Otopsi yapılarak sonuçların doğrulanmasının ardından SARS-CoV-2 ile aktive olan CRS'nin, COVID-19 ölümlerinin nedeni olabileceği öne sürülmüştür (Ruan ve ark., 2020). Liu ve ark. (2020), 12 COVID-19 hastasında ölçülen toplam plazma sitokinlerinden 38 sitokinin anlamlı düzeyde yükseldiğini bildirmiştir. Ayrıca SARS-CoV-2 yüküyle ilişkili 17 sitokin ve sitokinlerin 15'inin akciğer hasarıyla güçlü bir korelasyonu vardı. Buna M-CSF, IL-10, IFN-α2, IL-4, IP-10, IL-7, IL-1ra, G-CSF, IL-12, IFN-y, IL-1α, IL-2 dahildir. HGF, PDGF-BB ve IL-17. TH17 hücreleri, kendini kopyalamayı da içeren virüslere yanıt olarak akciğerlerde IL-17 üretir. Ayrıca sinerjizmleri de CD8+ T hücresi aracılı hedef yıkımını teşvik eder (Hou ve ark., 2014).

Nötrofil-lenfosit oranı (NLR), trombosit-lenfosit oranı (PLR) ve lenfosit-monosit oranı (LMR) gibi dolaşımdaki biyobelirteçler, COVID-19 hastalarının inflamatuar ve bağışıklık durumunu temsil edebilmektedir. NLR, PLR ve LMR; sistemik inflamasyonun prognozunda kullanılan yararlı belirteçler olup, viral pnömoninin prognozunda yaygın olarak kullanılmaktadır. NLR, ciddi hastalığı olan COVID-19 hastalarında yükselir. Dolayısıyla NLR; COVID-19 şiddetiyle ilişkili olup, kötü klinik seyir için bağımsız bir prognostik biyobelirteç olarak kullanılabilir (Yang ve ark., 2020). Ayrıca 29 çalışma üzerinde yapılan bir meta-analizde ağır hastalarda NLR düzeylerinin arttığı rapor edilmiştir (Feng ve ark., 2020). PLR seviyesinin, COVID-19 şiddeti için uygun maliyetli ve kolay ulaşılabilen bir prognostik biyobelirteç olduğu söylenebilir. Düzeyi, COVID-19'un ciddiyetine yanıt olarak artar ve bu, şiddetli grubun, şiddetli olmayan gruba göre daha yüksek PLR düzeyleri sunduğu bir meta-analizde gösterilmiştir (Simadibrata ve ark., 2020). LMR düzeyinin ise COVID-19'un ciddiyeti ile negatif bir korelasyon sergilediği düşünülmektedir. Şiddetli COVID-19 hastalarında daha yüksek NLR seviyeleri ve daha düşük LMR seviyeleri bildirilmiştir. Tersine, hafif veya orta şiddette COVID-19 vakası olan hastalarda daha yüksek LMR seviyeleri raporlanmıştır. Her ne kadar NLR, PLR ve LMR seviyeleri COVID-19'un prognozunda yararlı olsa da NLR, PLR ve LMR için normal referans aralıkları hala belirsizliğini korumaktadır (Kong ve ark., 2020). Yukarıdakilerden yola çıkarak, tedavi sırasında sitokinlerin hedeflenmesi, COVID-19 hastalarında COVID-19 hastalığının ilerleme hızını azaltabilir ve COVID-19'un neden olduğu ölümleri azaltabilir. Örneğin Tocilizumab, IL-6'nın reseptörüne bağlanmasını engelleyebilen rekombinant hümanize IL-6 reseptör antagonistidir. Tocilizumab esas olarak romatoid artrit tedavisinde kullanılmaktadır ancak KRS'nin büyük oranda IL-6'dan kaynaklanması nedeniyle COVID-19 tedavisi sırasında da kullanılabilir (Xu ve ark., 2020). Tocilizumab'ın ağır ve kritik 21 hastayı tedavi etmek için kullanıldığında cesaret verici sonuçlar elde edildiği bildirmiştir. Bunun dışında kortikosteroid tedavisi gibi başka alternatifler de COVID-19 tedavisinde benimsenebilir. Kortikosteroidler, anti-inflamatuar etkileri olan steroid hormonlardır ve yaygın olarak inflamasyonu baskılamak için kullanılır. Şiddetli SARS hastalarına glukokortikoidlerin uygun şekilde uygulanması, ateşin düşmesi, akciğerlere sızan radyasyonun hafifletilmesi ve oksijenasyonun iyileştirilmesi yoluyla durumlarını iyileştirdi. Ancak glukokortikoidlerin uygulanmasının zamanlaması doğru yapılmalı ve dozajı dikkatli bir şekilde alınmalıdır. Glukokortikoidlerin çok erken uygulanması vücudun savunma mekanizmasını engelleyerek viral yükün artmasına neden olur. Bu nedenle, glukokortikoidler esas olarak sitokin fırtınasından muzdarip, kritik durumdaki COVID-19 hastalarının tedavisinde kullanılmaktadır. Kritik durumdaki COVID-19 hastalarına doğru zamanda kortikosteroid uygulanması ARDS oluşumunu azaltmış ve etkilenen organları korumuştur (Ye ve ark., 2020). İntravenöz immünoglobulin tedavisi aynı zamanda immün ikame ve immün modülasyon açısından ikili bir potansiyele sahiptir. Ancak COVID-19 hastalarında uygulanmasının önemi doğrulanmamıştır (Chen ve ark. 2020). Klorokin'in TNF ve IL-6'nın üretimini ve salınımını inhibe ettiği, dolayısıyla klorokin'in COVID-19 hastalarında sitokin fırtınasını baskılayabildiği bildirilmiştir. Yine kök hücre tedavisi de hastalarda sitokin fırtınasını önleyebilmektedir. Mezenkimal kök hücreler (MSC) kendini yenileme potansiyellerinin yanı sıra güçlü bir anti-inflamatuar ve immün düzenleyici fonksiyona sahiptirler. MSC, T lenfositlerin ve makrofajların anormal aktivasyonunu inhibe edebilir ve ayrıca bunların sırasıyla düzenleyici T hücrelerine ve anti-inflamatuar makrofajlara farklılaşmasını da aktive edebilir. MSC ayrıca TNF-α, IL-1, IL-6, IL-12 ve IFN-γ gibi proinflamatuar sitokinlerin üretimini de inhibe edebilir (Ye ve ark., 2020).

DRAFT

SONUÇ

Tüm enfeksiyonlarda olduğu gibi COVID-19 enfeksiyonu da sitokin salınımını uyarmaktadır ancak bazı hasta grupları aşırı sitokin tepkisi göstererek sitokin fırtınasının oluşmasına neden olmaktadır. Ayrıca, COVID-19 şiddetinin artması, başta IL-6 olmak üzere bazı sitokin düzeyleri üzerinde etkiye sahiptir. Bu nedenle sitokinler, COVID-19'un teşhisinde önemlidir ve bunların tedavi sırasında doğru zamanda doğru hastada kullanılması daha olumlu sonuçlar verebilmektedir. Sitokin fırtınası, COVID-19'a bağlı komplikasyonlara neden olur ve COVID-19'a bağlı ölümlerin ana nedenidir. Literatürde COVID-19 enfeksiyonunun neden olduğu sitokin salınımına ilişkin bildirilen veriler dikkate alındığında, bu enfeksiyonun T lenfositler ve bunların ürettiği IFN-γ düzeyleri üzerindeki etkisine odaklanan çalışmaların önemli bulgular sağlayacağı sonucuna varılmıştır.

KAYNAKLAR

Azkur AK, Akdis M, Azkur D, Sokolowska M, van de Veen W, Brüggen MC et al 2020. Immune response to SARS-CoV-2 and mechanisms of immunopathological changes in COVID-19. Allergy. 75(7): 1564-81.

Behrens EM, Koretzky GA 2017. Cytokine storm syndrome: Looking toward the precision medicine era. Arthritis Rheumatol. 69(6): 1135–43.

Berraondo P, Sanmamed MF, Ochoa MC, Etxeberria I, Aznar MA, Pérez-Gracia JL et al 2019. Cytokines in clinical cancer immunotherapy. British journal of cancer; 120(1): 6-15.

Chan JF, Yuan S, Kok KH, To KK, Chu H, Yang J et al 2020. A familial cluster of pneumonia associated with the 2019 novel coronavirus indicating person-to-person transmission: a study of a family cluster. The lancet.; 395(10223): 514-23.

Chatterjee SK, Saha S, Munoz MNM 2020. Molecular Pathogenesis, Immunopathogenesis and Novel Therapeutic Strategy Against COVID-19. Front Mol Biosci 7:196.

Chauhan P, Nair A, Patidar A, Dandapat J, Sarkar A, Saha B 2021. A primer on cytokines. Cytokine. 155458.

Chen N, Zhou M, Dong X, Qu J, Gong F, Han Y et al 2020. Epidemiological and Clinical Characteristics of 99 Cases of 2019 Novel Coronavirus Pneumonia in Wuhan, China: a descriptive study. Lancet,; 395 (10223): 507-513.

Conti P, Caraffa A, Gallenga CE, Ross R, Kritas SK, Frydas I et al 2020. Coronavirus-19 (SARS-CoV-2) induces acute severe lung inflammation via IL-1 causing cytokine storm in COVID-19: a promising inhibitory strategy. J Biol Regul Homeost Agents. 34(6): 1971-5.

Darif D, Hammi I, Kihel A, El Idrissi Saik I, Guessous F, Akarid K 2021. The pro-inflammatory cytokines in COVID-19 pathogenesis: What goes wrong? Microb Pathog [Internet].; 153:104799.

Devrim T, Ataç F, Devrim AK, Balcı M. The concomitant use of USP28 and p53 to predict the progression of urothelial carcinoma of the bladder. Pathol Pract. 2020; 216(1): 152774.

Devrim T, Ekici H, Devrim AK, Sozmen M, Senol A, Bozkurt KM, Duru O 2020. Late effects of cutaneous 3-methylcholanthrene exposure on DNA damage-related pleiotropic growth factors and oxidative stress markers in mice. Bratisl Med J 121(5): 325-330.

Dinarello CA 2000. Impact of basic research on tomorrow's medicine. Chest.; 118(2): 503–8.

dos Santos G, Delay L, Yaksh TL, Corr M 2020. Neuraxial Cytokines in Pain States. Front Immunol. 10:3061.

Ebihara N, Matsuda A, Nakamura S, Matsuda H, Murakami A 2011. Role of the IL-6 classic-and trans-signaling pathways in corneal sterile inflammation and wound healing. Invest Ophthalmol Vis Sci. 52(12): 8549–57.

Fares J, Fares MY, Khachfe HH, Salhab HA, Fares Y 2020. Molecular principles of metastasis: a hallmark of cancer revisited. Signal Transduct Target Ther. 5(1): 1–17.

Fares J, Cordero A, Kanojia D, Lesniak MS 2021. The Network of Cytokines in Brain Metastases. Cancers (Basel).; 13(1): 142.

Feng X, Li S, Sun Q, Zhu J, Chen B, Xiong M, et al 2020. Immune-inflammatory parameters in COVID-19 cases: A systematic review and meta-analysis. Front Med.; 7: 1–14.

Gao Y, Li T, Han M, Li X, Wu D, Xu Y et al 2020. Diagnostic utility of clinical laboratory data determinations for patients with the severe COVID-19. Journal of medical virology. 92(7): 791-6.

Hadjadj J, Yatim N, Barnabei L, Corneau A, Boussier J, Smith N et al 2020. Impaired type I interferon activity and inflammatory responses in severe COVID-19 patients. Science. 369(6504): 718-24

Han H, Ma Q, Li C, Liu R, Zhao L, Wang W et al 2020. Profiling serum cytokines in COVID-19 patients reveals IL-6 and IL-10 are disease severity predictors. Emerging microbes & infections. 9(1): 1123-30.

Hou W, Jin Y-H, Kang HS, Kim BS 2014. Interleukin-6 (IL-6) and IL17 Synergistically Promote Viral Persistence by Inhibiting Cellular Apoptosis and Cytotoxic T Cell Function. Perlman S, editor. J Virol. 88(15): 8479 LP – 8489.

Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y et al 2020. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. The lancet. 395(10223): 497-506.

Karabacak P, Kırdemir P 2021. COVID-19 hastalarında akut solunum sıkıntısı sendromu yönetimi. Med J SDU. (özel sayı1): 51-56.

Katze M 2012. Into the eye of the cytokine storm. Microbiol Mol Biol Rev.; 76(1): 16–32.

Kong J, Wang T, Di Z, Shi B, Yu X, Huang C, et al 2020. Analysis of hematological indexes of COVID-19 patients from fever clinics in Suzhou, China. Int J Lab Hematol. 42(5): e204–6.

Kunnumakkara AB, Rana V, Parama D, Banik K, Girisa S, Sahu H et al 2021. COVID-19, cytokines, inflammation, and spices: How are they related?. Life sciences. 16:119201

Li X, Geng M, Peng Y, Meng L. Lu SH. 2020.Mol immune Pathog diagnosis COVID-19, J Pharm Anal.; 10(2):102–8.

Liu Y, Zhang C, Huang F, Yang Y, Wang F, Yuan J et al 2020. Elevated plasma levels of selective cytokines in COVID-19 patients reflect viral load and lung injury. National Science Review. 7(6): 1003-11.

Mahmud-Al-Rafat A, Asim MM, Taylor-Robinson AW, Majumder A, Muktadir A, Muktadir H et al. 2020. A combinational approach to restore cytokine balance and to inhibit virus growth may promote patient recovery in severe COVID-19 cases. Cytokine, 15:155228.

Mosquera-Sulbaran JA, Pedreañez A, Carrero Y, Callejas D 2021. C-reactive protein as an effector molecule in Covid-19 pathogenesis. Rev Med Virol. e2221.

Ruan Q, Yang K, Wang W, Jiang L, Song J 2020. Clinical predictors of mortality due to COVID-19 based on an analysis of data of 150 patients from Wuhan, China. Intensive Care Med.; 46(5): 846–8.

Sallenave J-M, Guillot L 2020. Innate immune signaling and proteolytic pathways in the resolution or exacerbation of SARS-CoV-2 in Covid-19: key therapeutic targets? Front Immunol.11.

Shimabukuro-Vornhagen A, Gödel P, Subklewe M, Stemmler HJ, Schlößer HA, Schlaak M et al 2018. Cytokine release syndrome. Journal for immunotherapy of cancer. 6(1):1-4.

Simadibrata DM, Pandhita BAW, Ananta ME, Tango T 2020. Platelet-to-lymphocyte ratio, a novel biomarker to predict the severity of COVID-19 patients: A systematic review and meta-analysis. J Intensive Care Soc.

Simpson S, Kaislasuo J, Guller S, Pal L 2020. Thermal stability of cytokines: A review. Cytokine. 125:154829.

Tang X, Wu C, Li X, Song Y, Yao X, Wu X et al 2020. On the origin and continuing evolution of SARS-CoV-2. National Science Review.; 7(6): 1012-23.

Wang F, Hou H, Luo Y, Tang G, Wu S, Huang M et al 2020. The laboratory tests and host immunity of COVID-19 patients with different severity of illness. JCI insight.; 5(10): e137799.

Xu X, Han M, Li T, Sun W, Wang D, Fu B et al 2020. Effective treatment of severe COVID-19 patients with tocilizumab. Proceedings of the National Academy of Sciences.; 117(20): 10970- 5.

Yang AP, Liu J ping, Tao W qiang, Li H ming. The diagnostic and predictive role of NLR, d-NLR and PLR in COVID-19 patients. Int Immunopharmacol. 2020; 84: 106504.

Ye Q, Wang B, Mao J 2020. The pathogenesis and treatment of the `Cytokine Storm' in COVID-19. J Infect.; 80(6): 607-613.

DRAFT

ORAL PRESENTATION

Obezite patogenezinde MAPK ve PI3K/AKT sinyal yolları

Adem Keskin[1*] (*ORCID: https://orcid.org/0000-0003-1921-2583*)

[*1] Aydın Adnan Menderes Üniversitesi, Sağlık Bilimleri Enstitüsü, Biyokimya (Tıp) Bölümü, Aydın, Türkiye.

*Sorumlu yazar e-mail: ademkeskin78@gmail.com

Özet

Dünya genelinde önemli bir sağlık sorunu olan ve vücutta aşırı yağ birikmesiyle karakterize edilen obezite tip 2 diyabet, kardiyovasküler hastalıklar ve alkolsüz yağlı karaciğer hastalığı gibi bir dizi kronik hastalıkların riskini keskin bir şekilde artırır. Diyet ve egzersiz gibi yaşam tarzı müdahalelerinin obezite ile mücadelede kayda değer etkileri olmasına rağmen, kilo vermede uzun vadeli başarıya ulaşmak son derece zordur ve obezite prevalansı dünya çapında artmaya devam etmektedir. Geçtiğimiz birkaç on yılda, obezitenin patofizyolojisi kapsamlı bir şekilde araştırılmış ve artan sayıda sinyal yolu obezite ile ilişkilendirilmiş, obeziteyle daha etkili ve kesin bir şekilde mücadele etmek için sinyal yollarına odaklanılmıştır. Bu makalede, obezitenin patogenezi ve tedavisi üzerine odaklanılan iki ana sinyal yolunun rolü incelenmiştir: mitojenle aktifleşen protein kinaz (MAPK) ve fosfatidilinositol 3-kinaz (PI3K)/AKT sinyal yolları. MAPK sinyal yolu, iştah kontrolü, glukoz seviyelerinin düzenlenmesi ve yağ hücresi oluşumuna katkıda bulunurken, aynı zamanda insülin direncinin gelişimine de yol açabilir. Özellikle hipotalamusta ve yağ dokusunda MAPK'nin rolü vurgulanmıştır. PI3K/AKT sinyal yolu ise hücre büyümesi ve çoğalmasının düzenlenmesinde kritik bir rol oynar. Bu yolun anormal aktivasyonu, obezitenin gelişimini destekleyebilir ve insülin direncine katkıda bulunabilir. Ayrıca, iştah düzenlemesi ve enerji dengesi üzerinde de etkilidir. Obezitenin toplumsal maliyeti yüksektir ve sağlık sorunlarını artırır. Bu nedenle obezitenin önlenmesi ve tedavisi büyük önem taşır. Araştırmalar, sinyal yollarının iştah düzenlemesi, yağ dokusunun metabolizması ve enerji dengesi arasındaki ilişkilerin daha iyi anlaşılması gerektiğini göstermektedir. Sonuç olarak, obezitenin yönetimi için yenilikçi ve güvenli önlemlerin araştırılması gerekmektedir. Sinyal yolları, obezitenin patogenezinde önemli bir rol oynar ve gelecekteki çalışmalar bu konuya daha fazla odaklanmalıdır.

Anahtar Kelimeler: Obezite, Sinyal yolu, MAPK sinyal yolu, PI3K/AKT sinyal yolu

MAPK and PI3K/AKT signaling pathways in the pathogenesis of obesity

Abstract

Obesity, a major health problem worldwide and characterized by excess fat accumulation in the body, sharply increases the risk of a number of chronic diseases, such as type 2 diabetes, cardiovascular diseases and non-alcoholic fatty liver disease. Although lifestyle interventions such as diet and exercise have significant effects in combating obesity, long-term success in weight loss is extremely difficult to achieve and the prevalence of obesity continues to increase worldwide. Over the past few decades, the pathophysiology of obesity has been extensively investigated and an increasing number of signaling pathways have been associated with obesity, focusing on signaling pathways to combat obesity more effectively and precisely. This article focuses on the pathogenesis and treatment of obesity by examining the role of two major signaling pathways: mitogen-activated protein kinase (MAPK) and phosphatidylinositol 3-kinase (PI3K)/AKT signaling pathways. While the MAPK signaling pathway contributes to appetite control, regulation of glucose levels and fat cell formation, it may also lead to the development of insulin resistance. The role of MAPK has been particularly emphasized in the hypothalamus and adipose tissue. The PI3K/AKT signaling pathway plays a critical role in regulating cell growth and proliferation. Abnormal activation of this pathway may promote the development of obesity and contribute to insulin resistance. It is also effective on appetite regulation and energy balance. The social cost of obesity is high and increases health problems. Therefore, prevention and treatment of obesity is of great importance. Research shows that there is a need for a better understanding of the relationships between signaling pathways, adipose tissue metabolism, and energy balance in the regulation of appetite. As

a result, innovative and safe measures for obesity management need to be investigated. Signaling pathways play an important role in the pathogenesis of obesity, and future studies need to focus more on this issue.
Keywords: Obesity, Signaling pathway, MAPK signaling pathway, PI3K/AKT signaling pathway

GİRİŞ

Yağ dokusundaki aşırı artış, hem anormal yağ hücresi lipit birikiminin (adiposit hipertrofisi) hem de hiperplazi olarak bilinen öncüllerinin katılımıyla yeni adipositlerin gelişmesinin bir sonucudur. Yağ dokusundaki bu artış sonucu ortaya çıkan obezite, tip 2 diyabetus melltus, kardiyovasküler hastalıklar, metabolik sendrom , alkolsüz yağlı karaciğer hastalığı gibi bulaşıcı olmayan hastalıklarla sıklıkla birlikte görülen, gün geçtikçe kötüleşen bir sağlık sorunudur (Gustafson ve ark., 2019), (Shen ve ark., 2022). Obeziteye eşlik eden komorbiditeler, hastaların yaşam kalitesini kötüleştirir (Kivimäki ve ark., 2022). Bu da vücut kitle indeksi ile orantılı olarak sağlık bakım maliyetlerinde artışa karşılık gelir (Kent ve ark., 2019). Yakın zamanda obezite ve aşırı kilo prevalansındaki mevcut eğilim değişmezse, yaklaşık ekonomik maliyetlerin küresel gayri safi yurt içi hasılanın %3,29'una yükseleceği rapor edilmiştir (Okunogbe ve ark., 2021). Toplu olarak, obezitenin geç ve geri döndürülemez sonuçlarının tedavisiyle ilişkili olarak dünya nüfusuna gelen bu önemli sosyo-ekonomik yükler, dikkatleri obezitenin yönetiminde yenilikçi ve güvenli önleme ve erken müdahalelerin araştırılmasına çekmiştir (Müller ve ark., 2022).

Tüm canlı sistemler çevreleri hakkında bilgi edinir. Hücresel düzeyde bunu sinyal yolları aracılığıyla yaparlar. Bu tür yollar, hücre dışı bir işaretin veya sinyalin varlığını algılayan ve hücrenin iç kısmına ileten moleküller arasındaki tersine çevrilebilir bağlanma etkileşimlerine dayanır (Azpeitia ve ark., 2020). Bu etkileşimlerin oluşturduğu sinyal yollarının modüle edilmesinde yer alan eylemlerin anlaşılması, hastalıkların önlenmesi ve tedavisi için yararlı bilgiler sağlamaktadır (Keskin, 2023). Sinyal iletimine ilişkin gelişen bilgi, kişiselleştirilmiş tıpta yeni bir çağa girerken obezite araştırmalarının gelecekteki yönüne ışık tutabilmektedir. Sinyal iletim yolları ile obezite arasındaki ilişkiye ilişkin giderek artan sayıda kanıt bulunmaktadır; Obezitenin patofizyolojisi kapsamlı bir şekilde araştırılmıştır ve bu da obeziteyle daha etkili ve kesin bir şekilde mücadele edilmesini mümkün kılmaktadır (Müller ve ark., 2022).

Bilim ve teknolojideki gelişmelerin yanı sıra ilaç sektörünün de hızla büyümesiyle obeziteyle mücadelede büyük başarılar elde edilmiş; kalori kısıtlaması, yaşam tarzı yönetimi, farmakoterapi ve bariatrik cerrahi gibi çeşitli stratejiler obezite karşıtı çareler olarak önerilmiştir (Trepanowski ve ark., 2017), (Wolfe ve ark., 2016). Bununla birlikte, bu müdahaleler küresel boyuttaki tıbbi ihtiyaçları karşılama kapasitesine sahip değildir. Son zamanlarda iştahın düzenlenmesi ve periferik enerji emilimi, depolanması ve tüketimi ile ilgili çok sayıda faktör/sinyal ortaya çıkarılmıştır (Huang ve ark., 2019), (Xu ve ark., 2021). Bu ilerlemeler obezitenin ortaya çıkışının anlaşılmasına ışık tutmaktadır. Bu sinyal yolllarının modüle edilmesinde polifenollerin rolü, yakın zamanda yapılan birçok çalışmanın konusu olmuştur (Keskin, 2023). Buna ek olarak, bu sinyalleri hedef alan bazı bileşikler klinik kullanıma dönüştürülmüştür. Örneğin, obezite karşıtı araştırmaların sıcak noktası olan iştah düzenlemesi, hem merkezi melanokortin yolu hem de leptin ve bağırsak hormonları gibi periferik sinyaller tarafından düzenlenir. Kan şekeri düzeylerini düşürebilen ve siklik adenosin monofosfat'a (cAMP) bağımlı sinyal yollarıyla insülin salgılanmasını teşvik ederek glikoz toleransını geliştirebilen bağırsaktan türetilen bir hormon olan glukagon benzeri peptid 1 (GLP-1), aynı zamanda anoreksijenik nöronları doğrudan uyararak iştahı da azaltabilir. Öte yandan oreksijenik nöronları γaminobütirik asit (GABA) bağımlı sinyal yoluyla baskılayabilir (Huang ve ark., 2019).

Obezitenin patofizyolojisi hala tam olarak anlaşılmamış olmasına rağmen, birçok yolla kontrol edilen heterojen bir sağlık sorunu olduğu yaygın olarak kabul edilmektedir. Obezitenin ortaya çıkması ve gelişmesinde yer alan sinyal yollarının giderek daha iyi anlaşılması, obeziteyle daha kesin bir şekilde mücadele etmemizi sağlar. Bu derlemede, obezitenin patogenezinde, özellikle iştahın düzenlenmesinde, yağ dokusu metabolizmasında ve fonksiyonunda, glikoz hemostazında ve enerji harcamasında yer alan bazı sinyal yollarının incelenmesi amaçlandı.

MATERYAL VE METOT

Obezite sağlık sorununun patogenezinde sinyal yollarının öneminin daha iyi anlaşılmasını sağlamak amacıyla PubMed, Scopus, Web of Science ve Google Scholar veritabanlarını kullanılarak literatür taraması yapıldı. Anahtar kelime olarak, obezite, sinyal yolu, mitojenle aktifleşen protein kinaz (MAPK) sinyal yolu ve fosfatidilinositol 3-kinaz (PI3K)/AKT sinyal yolu, kelimeleri kullanıldı. Alıntılama sayıları ve yayın tarihleri dikkate alınarak 35 makale seçildi. Ayrıca bu seçim, obezite patogenezi esasına dayalı olarak yapıldı.

Literatür taramasında en çok çalışma yapılan iki sinyal yolu, ana başlıklar altında kategorize edildi. Referans olarak değerlendirmeye alınan çalışmalarda ilgili sinyal yolu ile iştahın düzenlenmesi, yağ dokusu metabolizması ve fonksiyonu, glukoz hemostazı ve enerji harcaması arasındaki ilişkiye odaklanıldı.

MEMELİ HÜCRELERİNDE SİNYAL İLETİMİNİN KRİTİK ARACI: MAPK SİNYAL YOLU

MAPK sinyali, hücre dışı uyarıları hücre içi sinyallere bağlayan bir kinaz kademesi içerir (Pudewell ve ark., 2021). MAPK tarafından gerçekleştirilen fosforilasyon ile, gen ekspresyonuna aracılık etmek ve proliferasyon, inflamasyon, farklılaşma ve apoptoz gibi hücresel olayları başlatmak için aşağı yöndeki transkripsiyon faktörleri aktive edilir (Sun ve ark., 2015). Hücre dışı sinyalle düzenlenen kinaz (ERK) 1/2, c-Jun N-terminal kinaz (JNK) ve p38 MAPK dahil olmak üzere MAPK sinyal yolu üyeleri iştah, adipogenez, glukoz homeostazisi ve termojenezin düzenlenmesinde önemli bir rol oynar (Kassouf ve Sumara, 2020).

MAPK aracılı iştah düzenlemesinin yanı sıra merkezi sinir sistemindeki diğer MAPK fonksiyonları da obezitenin patogenezine katkıda bulunur. ERK1/2, hipotalamik nöronlarda glukozla uyarılan proopiomelanokortin ekspresyonunu arttırır ve anoreksijenik etkiye katılır (Zhang ve ark., 2015). Buna ek olarak MAPK sinyal yolu merkezi sinir sistemindeki JNK1 nakavtı, hipotalamik-hipofiz-tiroid ekseninin negatif geri bildirimini bloke ederek gıda alımını azaltır ve enerji harcamasını artırır (Sabio ve ark., 2010). p38 MAPK alt birimi nakavt veya inhibisyonu olan embriyonik farelerden alınan primer embriyonik fibroblastlarda ve yetişkin farelerden alınan preadipositlerde, PPARγ ekspresyonu artar, bu da p38 MAPK'nin adipogenezi baskıladığını gösterir (Aouadi ve ark., 2006). Tersine, in vitro insan preadiposit farklılaşması sırasında p38 MAPK aktivitesinde artış gözlenir ve bu hücrelerde p38 MAPK'nin farmakolojik inhibisyonu, trigliseritlerin birikimini ve diğer adipogenez belirteçleriyle birlikte PPARγ ekspresyonunu azaltır (Aouadi ve ark., 2007). İnsan beyaz yağ dokusunda, artan hipertrofik adiposit sayısı aynı zamanda yukarı regüle edilmiş p38 MAPK sinyalleriyle de ilişkilidir ve fosforile edilmiş p38 MARK, açlık trigliserid, insülin ve glukoz düzeyleriyle bağlantılıdır (Bashan ve ark., 2007). Bu bulgular p38 MAPK'nın adiposit farklılaşması ve adipogenez üzerinde iki işlevli etkileri olduğunu göstermektedir.

MAPK sinyal yolu, insülin direncinin gelişiminde yakından rol oynar. MAPK sinyal yolunun birden fazla noktadan defosforilasyonu ve deaktivasyonu, insülin reseptör substrat-1'in tirozin fosforilasyon seviyesini ve insülin sinyal transdüksiyonuna aracılık etme kapasitesini eski haline getirir (Khoubai ve Grosset, 2021). Benzer şekilde, MAPK sinyal yolunun endojen bir aktivatörü olan kaspaz 9'un eksikliği, yüksek yağlı diyetin neden olduğu insülin direncini ve adiposit büyümesini azaltır (Zeng ve ark., 2018). Yüksek yağlı diyet ile beslenen farelerden alınan yağ dokularında, entegre multiomik analiz, makrofajlardaki MAPK sinyal yollarında inflamatuar genlerin zenginleştiğini göstermektedir (Wang ve ark., 2021).

HÜCRE BÜYÜMESİ VE ÇOĞALMASININ ÖNEMLİ BİR DÜZENLEYİCİSİ: PI3K/AKT SİNYAL YOLU

Hücre büyümesi ve çoğalmasının önemli bir düzenleyicisi olan PI3K/AKT sinyal yolunun anormal aktivasyonu, obezitenin gelişimini teşvik etmektedir (Sun ve ark. 2019). PI3K ve AKT, bu yoldaki hormonlar ve büyüme faktörleri gibi yukarı yönlü sinyaller tarafından etkinleştirilen iki ana düğümdür. PI3K aktivasyonu, fosfatidilinositol 4,5-bifosfatı fosfatidilinositol 3,4,5-trisfosfata dönüştürür, fosfoinositid bağımlı kinazları ve AKT'yi aktive eder ve ardından glikojen sentaz kinaz3, sırasıyla glikojen sentezini, glukoz alımını ve adipogenezi düzenleyen forkhead box protein (Fox) ailesini aktive eder (Hemmings ve Restuccia, 2015).

PI3K/AKT sinyal yolunda katalitik bir alt birim olarak bulunan rapamisinin memeli hedefi (mTOR) çok çeşitli önemli hücresel süreçleri kontrol etme konusundaki olağanüstü yeteneğinden dolayı hücresel yaşlanmada önemlidir (Keskin ve ark., 2023). mTOR, mTORC1 ve mTORC2 olmak üzere iki farklı kompleks oluşturur. mTORC1 ve mTORC2 PI3K/AKT/mTOR sinyal yolunda farklı hareket eder ve her ikisi de obezitenin patogeneziyle yakından ilişkilidir. mTOR ayrıca merkezi ve periferik sistemlerde iştahın düzenlenmesine de katkıda bulunur. Hipotalamusta mTOR'un uyarılması, proopiomelanokortin nöronlarını aktive ederek gıda alımını azaltır ve hayvan çalışmalarında yaşa bağlı obeziteyi iyileştirir. PI3K/AKT yolu, merkezi sinir sistami ve periferik dokular yoluyla iştahı düzenler. Leptinin hipotalamusun mediobazal kısmına etki ederek kısmen PI3K-AKT-FoxO1 yoluyla gıda alımını baskılamaktadır (Kwon ve ark., 2016). PI3K'nın seçici inhibisyonunun leptinin etkisini ortadan kaldırdığı rapor edilmiştir (Hill ve ark., 2008).

PI3K/AKT yolu, insülin sinyal yolu için vazgeçilmezdir. Bu sinyalin düzensizliği obezitenin ciddiyeti ve insülin direnciyle ilişkilidir (Li ve ark., 2017). AKT aktivitesi ile vücut yağ yüzdesi arasında negatif korelasyon hem hayvan modellerinde hem de insanlarda bulunmuştur ve obez popülasyondaki insülin direncinden AKT sorumlu olabilir (Friedrichsen ve ark., 2010), (Mackenzie ve Elliot,. 2014). PI3K/AKT sinyallemesinin inhibisyonu, glikoz taşıyıcı 4 (GLUT4) depolama keseciklerinin bir elemanı olan Sort1'in bozulmasına yol açar ve insülin duyarlılığını azaltır (Li ve ark., 2017).

SONUÇ

Sonuç olarak MAPK sinyal yolu, iştah kontrolü, glukoz seviyelerinin dengelemesi ve vücut ısısının ayarlanması gibi işlevlerin yanı sıra yağ hücrelerinin oluşumu ve gelişimi üzerinde önemli etkilere sahiptir. Aynı zamanda insülin direncinin gelişiminde de rol oynar, bu da obezitenin patogeneziyle yakından ilişkilidir. PI3K/AKT sinyal yolu da, anormal aktivasyonunda obezitenin gelişimini teşvik etmesinin yanı sıra obez popülasyondaki insülin direncinden AKT sorumlu olabilmektedir. Aynı zamanda merkezi sinir sistemi ve periferik dokular yoluyla iştahı düzenlemesine ve leptin salınımına etki ederek gıda alımını baskılamasına katkıda bulunarak obezitenin patogenezinde önemli rolü olan diğer sinyal yoludur. Obezitenin ve neden olduğu birçok sağlık sorununun tedavi maliyetini en düşük düzeylere indirmek ve toplumdaki sağlıklı birey oranlarını en yüksek düzeylere yükseltmek amacıyla obezitenin önlenmesine yönelik çalışmalar artarak devam edecektir. Bu çalışmalarda, sinyal yollarının iştah düzenlemesi, yağ dokusunun metabolizması ve fonksiyonu, glukoz dengelemesi ve enerji harcaması arasındaki ilişkiler öncelikli bir öneme sahip olacaktır.

KAYNAKLAR

Aouadi M, Laurent K, Prot M, Le Marchand-Brustel Y, Binétruy B, Bost F. 2006. Inhibition of p38MAPK increases adipogenesis from embryonic to adult stages. Diabetes. 55(2): 281-289.

Aouadi M, Jager J, Laurent K, Gonzalez T, Cormont M, Binétruy B, Le Marchand-Brustel Y, Tanti JF, Bost F. 2007. p38MAP Kinase activity is required for human primary adipocyte differentiation. FEBS Lett., 581(29): 5591-5596.

Azpeitia E, Balanzario EP, Wagner A. 2020. Signaling pathways have an inherent need for noise to acquire information. BMC Bioinformatics. 21(1): 462.

Bashan N, Dorfman K, Tarnovscki T, Harman-Boehm I, Liberty IF, Blüher M, Ovadia S, Maymon-Zilberstein T, Potashnik R, Stumvoll M, Avinoach E, Rudich A. 2007. Mitogen-activated protein kinases, inhibitory-kappaB kinase, and insulin signaling in human omental versus subcutaneous adipose tissue in obesity. Endocrinology. 148(6): 2955-2962.

Friedrichsen M, Poulsen P, Richter EA, Hansen BF, Birk JB, Ribel-Madsen R, Stender-Petersen K, Nilsson E, Beck-Nielsen H, Vaag A, Wojtaszewski JF. 2010. Differential aetiology and impact of phosphoinositide 3-kinase (PI3K) and Akt signalling in skeletal muscle on in vivo insulin action. Diabetologia. 53(9): 1998-2007.

Gustafson B, Nerstedt A, Smith U. 2019. Reduced subcutaneous adipogenesis in human hypertrophic obesity is linked to senescent precursor cells. Nature communications, 10(1): 2757.

Hemmings BA, Restuccia DF. 2015. PI3K-PKB/Akt pathway. Cold Spring Harb Perspect Biol. 2012 Sep 1;4(9):a011189. doi: 10.1101/cshperspect.a011189. Erratum in: Cold Spring Harb Perspect Biol. 7(4). pii: a026609.

Hill JW, Williams KW, Ye C, Luo J, Balthasar N, Coppari R, Cowley MA, Cantley LC, Lowell BB, Elmquist JK. 2008. Acute effects of leptin require PI3K signaling in hypothalamic proopiomelanocortin neurons in mice. J Clin Invest., 118(5): 1796-1805.

Hu E, Kim JB, Sarraf P, Spiegelman BM. 1996. Inhibition of adipogenesis through MAP kinase-mediated phosphorylation of PPARgamma. Science. 274(5295): 2100-2103.

Huang R, Ding X, Fu H, Cai Q. 2019. Potential mechanisms of sleeve gastrectomy for reducing weight and improving metabolism in patients with obesity. Surg Obes Relat Dis. 15(10): 1861-1871.

Kassouf T, Sumara G. 2020. Impact of Conventional and Atypical MAPKs on the Development of Metabolic Diseases. Biomolecules., 10(9): 1256.

Kent S, Jebb SA, Gray A, Green J, Reeves G, Beral V, Mihaylova B, Cairns BJ. 2019. Body mass index and use and costs of primary care services among women aged 55–79 years in England: a cohort and linked data study. International Journal of Obesity, 43(9): 1839-1848.

Keskin A. 2023. Impact of Polyphenolic Compounds on the MAPK Signaling Pathway against Carcinogenesis. Journal of Clinical Practice and Research, 45(3): 217-221.

Keskin A. 2023. Hücre yaşamı ile ilgili sinyal yolakların düzenlenmesinde diyet polifenollerin rolü Turk J Health S. 4(1): 32-36.

Keskin A, Aci R, Ari M, Duran U. 2023. Cellular senescence, mTOR signaling pathway and polyphenol. International Journal of Advanced Biochemistry Research, 7(1): 01-04.

Kivimäki M, Strandberg T, Pentti J, Nyberg ST, Frank P, Jokela M, Ervasti J, Suominen SB, Vahtera J, Sipilä PN, Lindbohm JV, Ferrie JE. 2022. Body-mass index and risk of obesity-related complex multimorbidity: an observational multicohort study. Lancet Diabetes Endocrinol, 10 (2022): 253-263.

Khoubai FZ, Grosset CF. 2021. DUSP9, a Dual-Specificity Phosphatase with a Key Role in Cell Biology and Human Diseases. Int J Mol Sci. 22(21):11538.

Kwon O, Kim KW, Kim MS. 2016. Leptin signalling pathways in hypothalamic neurons. Cell Mol Life Sci., 73(7): 1457-1477.

Li J, Chen C, Li Y, Matye DJ, Wang Y, Ding WX, Li T. 2017. Inhibition of insulin/PI3K/AKT signaling decreases adipose Sortilin 1 in mice and 3T3-L1 adipocytes. Biochim Biophys Acta Mol Basis Dis., 1863(11): 2924-2933.

Li T, Zhang Z, Kolwicz SC Jr, Abell L, Roe ND, Kim M, Zhou B, Cao Y, Ritterhoff J, Gu H, Raftery D, Sun H, Tian R. 2017. Defective Branched-Chain Amino Acid Catabolism Disrupts Glucose Metabolism and Sensitizes the Heart to Ischemia-Reperfusion Injury. Cell Metab. 25(2): 374-385.

Mackenzie RW, Elliott BT. 2014. Akt/PKB activation and insulin signaling: a novel insulin signaling pathway in the treatment of type 2 diabetes. Diabetes Metab Syndr Obes., 7:55-64.

Müller TD, Blüher M, Tschöp MH, DiMarchi RD. 2022. Anti-obesity drug discovery: advances and challenges. Nature Reviews Drug Discovery, 21(3): 201-223.

Okunogbe A, Nugent R, Spencer G, Ralston J, Wilding J. 2021. Economic impacts of overweight and obesity: current and future estimates for eight countries. BMJ global health, 6(10): e006351.

Pudewell S, Wittich C, Kazemein Jasemi NS, Bazgir F, Ahmadian MR. 2021. Accessory proteins of the RAS-MAPK pathway: moving from the side line to the front line. Commun Biol., 4(1): 696.

Sabio G, Cavanagh-Kyros J, Barrett T, Jung DY, Ko HJ, Ong H, Morel C, Mora A, Reilly J, Kim JK, Davis RJ. 2010. Role of the hypothalamic-pituitary-thyroid axis in metabolic regulation by JNK1. Genes Dev. 24(3): 256-264.

Shen H, Huang X, Zhao Y, Wu D, Xue, K, Yao J, Wang Y, Tang T, Qiu Y. 2022. The Hippo pathway links adipocyte plasticity to adipose tissue fibrosis. Nature Communications, 13(1): 6030.

Sun F, Wang J, Sun Q, Li F, Gao H, Xu L, Zhang J, Sun X, Tian Y, Zhao Q, Shen H, Zhang K, Liu J. 2019. Interleukin-8 promotes integrin β3 upregulation and cell invasion through PI3K/Akt pathway in hepatocellular carcinoma. J Exp Clin Cancer Res. 38(1): 449.

Sun Y, Liu WZ, Liu T, Feng X, Yang N, Zhou HF. 2015. Signaling pathway of MAPK/ERK in cell proliferation, differentiation, migration, senescence and apoptosis. J Recept Signal Transduct Res., 35(6): 600-604.

Trepanowski JF, Kroeger CM, Barnosky A, Klempel MC, Bhutani S, Hoddy KK, Gabel K, Freels S, Rigdon J, Rood J, Ravussin E, Varady KA. 2017. Effect of Alternate-Day Fasting on Weight Loss, Weight Maintenance, and Cardioprotection Among Metabolically Healthy Obese Adults: A Randomized Clinical Trial. JAMA Intern Med. 177(7): 930-938.

Xu Q, Ding H, Li S, Dong S, Li L, Shi B, Zhong M, Zhang G. 2021. Sleeve Gastrectomy Ameliorates Diabetes-Induced Cardiac Hypertrophy Correlates With the MAPK Signaling Pathway. Front Physiol. 12: 785799.

Wang Z, Zhu M, Wang M, Gao Y, Zhang C, Liu S, Qu S, Liu Z, Zhang C. 2021. Integrated Multiomic Analysis Reveals the High-Fat Diet Induced Activation of the MAPK Signaling and Inflammation Associated Metabolic Cascades via Histone Modification in Adipose Tissues. Front Genet. 12: 650863.

Wen X, Zhang B, Wu B, Xiao H, Li Z, Li R, Xu X, Li T. 2022. Signaling pathways in obesity: mechanisms and therapeutic interventions. Signal Transduct Target Ther. 7(1): 298.

Wolfe BM, Kvach E, Eckel RH. 2016. Treatment of Obesity: Weight Loss and Bariatric Surgery. Circ Res. 118(11): 1844-1855.

Zeng X, Du X, Zhang J, Jiang S, Liu J, Xie Y, Shan W, He G, Sun Q, Zhao J. 2018. The essential function of CARD9 in diet-induced inflammation and metabolic disorders in mice. J Cell Mol Med., 22(6): 2993-3004.

Zhang J, Zhou Y, Chen C, Yu F, Wang Y, Gu J, Ma L, Ho G. 2015. ERK1/2 mediates glucose-regulated POMC gene expression in hypothalamic neurons. J Mol Endocrinol., 54(2): 125-135.

DRAFT

ORAL PRESENTATION

Investigation of Biochemical Changes in Mice Exposed to Monocrotaline

Ali Şenol[*1] (*ORCID:https://orcid.org/0000-0003-4080-7776)*, Alparslan Kadir Devrim[2] (*ORCID: https://orcid.org/0000-0002-3293-7290*), Miyase Çınar[1] (*ORCID:https://orcid.org/*0000-0003-3806-9938), Mahmut Sözmen[3] (*ORCID: https://orcid.org/0000-0001-7976-4051*)

*[1] Kırıkkale University, Faculty of Veterinary Medicine, Department of Biochemistry, Kırıkkale, Turkey
[2] Izmir Bakırçay University, Menemen Vocational School, Veterinary Health Program, Izmir, Turkey
[3] Ondokuz Mayıs University, Faculty of Veterinary Medicine, Department of Pathology, Samsun, Turkey

* Corresponding author e-mail: alisenol712@gmail.com
alisenol@kku.edu.tr

Abstract

Monocrotaline (MCT) is a pyrrolizidine alkaloid that causes hepatotoxicity in animals and humans. Humans are exposed to toxic PAs through consumption of PA-containing plants, PA-contaminated foods, herbal medicines, tea, and honey. The aim of this study is to examine biochemical changes in mice administered intragastric MCT. For this purpose, 30 male BALB/c mice were divided into three groups: control, acute toxicity and subacute toxicity. The control group received 0.9% saline three times in total, with an interval of 5 days. While a single dose of MCT was administered at a dose of 120 mg/kg to the acute toxicity group, MCT at a dose of 120 mg/kg was administered to the subacute toxicity group three times in total with an interval of 5 days. At the end of the trial, plasma AST, ALT, GGT, TBIL, TCHO and TPro levels were determined. Among the biochemical parameters evaluated within the scope of the study, AST, ALT, GGT, TBIL and TCHO levels were found to be higher in the trial groups. However, while there was no statistical difference between the groups in AST enzymatic activity, one of the hepatocellular damage markers, ALT enzymatic activity was found to be significantly higher in the acute toxicity group than in the control group. When GGT and TBIL levels, which are hepatobiliary damage markers, were examined, they were found to be higher and more significant in the acute toxicity group than in the control and subacute toxicity groups. Unlike other parameters, it was found that there was a decrease in TPro levels in the trial groups receiving MCT compared to the control group, and this decrease was statistically significant in all groups. As a result, it was observed that MCT administered intragastricly to mice played a role in the formation of hepatocellular and hepatobiliary damage.

Keywords: Monocrotaline, hepatotoxicity, biochemical parameter, mouse

INTRODUCTION

At the meeting of the joint expert committee (JECFA) formed by the World Health Organization (WHO) and the Food and Agriculture Organization (FAO), held in 2020, the toxicity of pyrrolizidine alkaloids (PA) was emphasized and of critical importance for health (WHO, 2020).

Pyrrolizidine alkaloids (PAs) are toxins of plant origin, produced by approximately 6000 plant species, consisting of a nesine base combined with one or two nesic acids via ester bonds (Xu et al., 2019; WHO, 2020). It is known that more than 600 unsaturated PAs and a large portion of their nitrogen oxides (N-oxides) produced by some plant species are hepatotoxic (Fu et al., 2004; Edgar et al., 2015).

People are exposed to toxic PAs through consumption of PA-containing plants, PA-contaminated foods, herbal medicines, tea and honey (Mulder et al., 2015; Zhu et al., 2018; He et al., 2020). Various plant products are frequently contaminated with PAs during mechanical harvesting processes. This is a possible source of exposure not only for humans but also for animals (Edgar et al., 2011; Dusemund et al., 2018; He et al., 2020). In particular, the exposure of farm animals such as cattle, sheep and goats to PA causes the presence of PA in animal products such as meat, milk or dairy products and increases the PA exposure of humans (EFSA, 2011; Mulder et al., 2016)

Hepatic sinusoidal obstruction syndrome (HSOS), liver damage, liver cirrhosis, pneumotoxicity and pulmonary hypertension have been reported to occur when exposed to PA due to various reasons (Ward et al., 2008; Rubbia-Brandt, 2010; Agostini et al., 2012). When the animals used in the experimental studies were examined, it was stated that hepatocellular necrosis was detected as a major and consistent finding, and similar findings were observed in humans after the consumption of wild herbs contaminated with seeds containing PA (Stewart and Steenkamp, 2001).

Numerous cases of human and animal poisoning by pyrrolizidine alkaloids were reported around the world (Karagöz et al., 2013). More than 4000 people were hospitalized in Tajikistan in 1992 due to consumption of grain contaminated with pyrrolizidine alkaloids (Chauvin et al., 1993; Mayer and Luthy, 1993; He et al., 2021a), and in Ethiopia, as of 2011, a total of 1033 people were reported to be affected, including 314 deaths (CDC, 2012; Robinson et al., 2014; Chiu et al., 2016). In China, where traditional herbal medicine is extensively practiced, a total of 2214 PA-induced liver injuries, including more than 200 deaths, have been reported due to the consumption of a PA-containing plant (Gynura japonica) (Zhu et al., 2021).

The liver, located in the metabolic center of the living body, is a very important organ for removing toxic substances from the body. Starting from this point, the main purpose of the study is to determine the effect of unintentionally exposed MCT, one of the pyrrolizidine alkaloids, on some biochemical parameters that play a critical role in the hepatic damage process. In this context, alanine aminotransferase (ALT), aspartate aminotransferase (AST), gamma-glutamyl transferase (GGT), total bilirubin (TBIL), total cholesterol (TCHO) and total protein (TPro) values will be examined. In this way, differences between control and experimental (acute and subacute toxicity) groups can be determined.

MATERIALS AND METHODS

Research and Publication Ethics:

The study design was approved by the Animal Experiments Local Ethics Committee of the University of Kırıkkale (Decision no. 2023/01).

Material:

Study material: 30 male BALB/c mice, 6-8 weeks old, weighing 20-25 g were created. The animals were housed in plastic cages at a controlled room temperature (22-25 °C) and 55-60 % humidity with 12:12-h light-dark cycle in Kırıkkale University Hüseyin AYTEMIZ Experimental Research and Application Center.

Experimental design:

They were fed with commercial standard dry pellet rat food and water ad libitum during the study and acclimated for two weeks before the experiment. Afterwards, they were randomly divided into three groups, including control, acute toxicity and subacute toxicity and the practices given in Table 1 were performed.

Table 1. Experimental Groups and Practices

Group name	Practices	Number of mice (n)
Control Group (Group 1)	Only intragastric saline was administered to the mice. The application started on day 0 of the experiment and continued 3 times in total, with 5-day intervals. The experiment was terminated at the 48th hour following the last application.	10
Acute Toxicity Group (Group 2)	MCT was administered intragastricly to mice at a dose of 120 mg/kg. The application was made once and on the 0th day of the experiment. The experiment was terminated at the 48th hour following the application.	10
Subacute Toxicity Group (Group 3)	MCT was administered intragastricly to mice at a dose of 120 mg/kg. The application started on day 0 of the experiment and continued 3 times in total, with 5-day intervals. The experiment was terminated at the 48th hour following the last application.	10

At the end of the experimental periods specified in Table 1, the animals were anesthetized and sample collection was carried out. For this purpose, xylazine (Rompun®) was administered intraperitoneally at a dose of 10 mg/kg as preanesthetic (Özden et al., 2020), and ketamine (Ketasol®) was administered intraperitoneally at a dose of 90 mg/kg to ensure deep anesthesia (Akbudak et al., 2021).

Then, the abdominal and chest cavities of all mice were opened, blood samples were collected into the heparinized test tubes for biochemical analysis, via cardiac puncture and plasma was removed after centrifugation at 3000 rpm for 10 minutes (Çınar et al., 2021). Blood plasma samples were aliquoted into eppendorf tubes and stored in the deep freezer (–80 °C) until analysis.

Biochemical analysis:

The plasma samples were analyzed for AST, ALT, GGT, total bilirubin, total protein and total cholesterol with biochemistry autoanalyzer for biochemical evaluation. Measurements were carried out on the Mindray BS-400 Biochemistry autoanalyzer and with commercially purchased test kits.

Statistical analyses:

Descriptive analysis and normality tests of all data obtained from each group were performed with SPSS 20.0 package program. Results were presented as mean ± standard error mean (Mean±SE'). One-way analysis of variance (ANOVA) was applied to show the difference between groups. The lower P value than 0.05 was considered as statistically significant. When the P value was significant, Duncan's Multiple Range test was performed.

RESULTS

The levels of some serum biochemical parameters of the control and experimental groups are given in Table 2.

Table 2. Some Biochemical Parameters of Control and Experimental Groups (x ± SH) (n=10)

Parameters	Control	Acute Toxicity	Subacute Toxicity	P
AST (U/L)	90,60±7,36	137,72±19,04	112,33±9,76	>0,05
ALT (U/L)	90,96±4,96^{b}	124,20±12,60^{a}	106,08±7,45^{ab}	≤0.05
GGT (U/L)	2,46±0,37^{b}	6,22±0,70^{a}	2,65±0,28^{b}	<0.001
Total bilirubin (mg/dl)	1,31±0,32^{b}	3,69±0,27^{a}	1,75±0,40^{b}	<0.001
Total cholesterol (mg/dl)	36,80±2,40	40,40±1,57	39,90±1,62	>0,05
Total protein (mg/dl)	5,87±0,10^{a}	4,93±0,14^{c}	5,48±0,07^{b}	<0.001

Among the biochemical parameters evaluated within the scope of the study, AST, ALT, GGT, total bilirubin and total cholesterol levels were observed to be higher in the trial groups. However, while there was no statistical difference between the groups in AST enzymatic activity, one of the hepatocellular damage markers, ALT enzymatic activity was found to be significantly higher in the acute toxicity group than in the control group. When GGT and total bilirubin levels, which are hepatobiliary damage markers, were examined, they were found to be higher and more significant in the acute toxicity group than in the control and subacute toxicity groups. There was no statistical difference in total cholesterol levels, which were similar in all groups. Unlike other parameters, decreased total protein levels were found to be statistically significant among all groups.

DISCUSSION

Determination of biochemical parameters provides important information in terms of reflecting the functions of organs and metabolic events (Etim, 2014). In this way, it is possible to confirm clinical diagnoses, evaluate the course of the disease, evaluate the results, and apply the appropriate treatment (Karagül et al., 2000). In the presented study, the effects of monocrotaline, one of the pyrrolizidine alkaloids (PA) that can be heavily exposed to many types of plant and animal foods, on some biochemical parameters were evaluated. In particular, AST, ALT, GGT, total bilirubin, total protein and total cholesterol levels were examined because PAs, which pose a risk to public health, have protoxin properties and develop toxicity after metabolic activation in the liver.

In a study conducted by Ren et al (2023), at a dose of 90 mg/kg given MCT intragastricly, statistically significant higher serum AST, ALT, and total bilirubin levels, lower serum Alb levels were observed in the MCT-treated group than those in the control group. Similarly, when Yamashita et al (2002) administered MCT intragastricly at a dose of 100 mg/kg, they demonstrated the hepatic damage caused by MCT with high serum AST, ALT and total bilirubin levels. Srinivasan and Liu (2012), who investigated acute monocrotaline poisoning, they reported that AST and ALT activity and hepatic injury had peaked 24 hours after acute monocrotaline poisoning.

In the study in question, it was observed that AST, ALT, GGT enzymatic activities and total bilirubin levels, which are the criteria for determining hepatocelluler and hepatobiliary, were increased in the experimental groups compared to the control group.

However, it was determined that AST was not statistically significant, while ALT, GGT enzymatic activities and total bilirubin levels were statistically significant in the acute toxicity group that was euthanized at the 48th hour. Additionally, lower serum total protein levels were observed in the MCT-treated groups than those in the control group. Looking at the total protein levels in Table 2, it can be seen that there is a statistically significant difference between both the control group and the MCT-administered trial groups. No statistically significant difference was found in total cholesterol levels, one of the biochemical parameters evaluated within the scope of the study.

It was observed that the data obtained were compatible with the literature and that MCT application at a dose of 120 mg/kg caused changes in hepatocellular and hepatobiliary system parameters, which are criteria for demonstrating liver damage. When a comparison was made between the experimental groups included in the study, it was observed that the biochemical values of the acute toxicity and subacute toxicity groups showed a similar increase or decrease compared to the control group. While there was no difference in AST, ALT and Total cholesterol levels between the two groups in which MCT was applied, GGT, total bilirubin and total protein levels were found to be statistically significant.

CONCLUSION

As a result, it was determined that MCT administration at a dose of 120 mg/kg to BALB/c mice affected the levels of AST, ALT, GGT, total bilirubin and total protein, which are the criteria for detecting hepatic damage. It was concluded that presenting new studies in which advanced techniques will be used and different doses and administration routes will be experienced will contribute to the literature in order to examine the damage caused by pyrrolizidine alkaloids exposed to various food contaminations from a broad perspective.

ACKNOWLEDGEMENT

This study was funded by the Scientific and Technological Research Council of Turkey (TUBITAK 1002-A Project No. 222O862) and was produced from a part of the first author's doctoral thesis.

REFERENCES

Agostini J, Benoist S, Seman M, Julié C, Imbeaud S, Letourneur F, Cagnard N, Rougier P, Brouquet A, Zucman-Rossi J, Laurent-Puig P 2012. Identification of molecular pathways involved in oxaliplatin-associated sinusoidal dilatation. Jornal of Hepatology, 56, 869-876.

Akbudak IH, Kılıç Erkek O, Tuzcu EB, Pakyurek H, Bor Kucukatay M 2021. Ketamine/xylazine anesthesia is safe in hemorheological point of view: a preliminary report. Pamukkale Tıp Dergisi, 14(2), 444-450.

Centers for Disease Control and Prevention (CDC) 2012. Investigating liver disease in Ethiopia. CDC Web Archive, https://www.cdc.gov/nceh/stories/ethio pia.html. [12.03.2022]

Chauvin P, Dillon JC, Moren A, Talbak S, Barakaev S 1993. Heliotrope poisoning in Tadjikistan. Lancet, 341, 1663

Chiu C, Martin C, Woldemichael D, W/Selasie G, Tareke I, Luce R, G/Libanos G, Hunt D, Bayleyegn T, Addissie A, Buttke D, Bitew A, Vagi S, Murphy M, Seboxa T, Jima D, Debella A 2016. Surveillance of a chronic liver disease of unidentifed cause in a rural setting of Ethiopia: a case study. Ethiopian Medical Journal, 54(1), 27-32.

Çınar Y, Kabakçı R, Şenol A 2021. Effect of Gingko Biloba Extract (Egb761) Supplementation on Hemato-Biochemical Parameters Following Acute Treadmill Exercise in Rats. F.U. Vet. J. Health Sci. 2021; 35(2), 98-102.

Dusemund B, Nowak, N, Sommerfeld C, Lindtner O, Schäfer B, Lampen A 2018. Risk assessment of pyrrolizidine alkaloids in food of plant and animal origin. Food and Chemical Toxicology, 115, 63-72.

Edgar JA, Colegate SM, Boppré M, Molyneux RJ 2011. Pyrrolizidine alkaloids in food: a spectrum of potential health consequences. Food Additives & Contaminants: Part A, 28(3), 308-324.

Edgar JA, Molyneux RJ, Colegate SM 2015. Pyrrolizidine alkaloids: potential role in the etiology of cancers, pulmonary hypertension, congenital anomalies, and liver disease. Chemical research in toxicology, 28(1), 4-20.

Etim N. 2014. Haematological Parameters and Factors Affecting Their Values. Agricultural Science, 2, 37-47.

European Food Safety Authority (EFSA) 2011. Scientific opinion on pyrrolizidine alkaloids in food and feed. EFSA Journal, 9(11), 2406.

Fu PP, Xia Q, Lin G, Chou MW 2004. Pyrrolizidine alkaloids-genotoxicity, metabolism enzymes, metabolic activation, and mechanisms. Drug metabolism reviews, 36(1), 1-55.

He X, Xia Q, Shi Q, Fu PP 2020. Efects of glutathione and cysteine on pyrrolizidine alkaloid-induced hepatotoxicity and DNA adduct formation in rat primary hepatocytes. J Environ Sci Health C Toxicol Carcinog, 38(2), 109-123.

He Y, Zhu L, Ma J, Lin G 2021a. Metabolism-mediated cytotoxicity and genotoxicity of pyrrolizidine alkaloids. Archives of Toxicology, 95(6), 1917-1942.

Karagoz Ş, Ilgın S, Atlı O, Perk BO, Burukoglu D, Ergun B, Sırmagul B 2013. Is N-acetyl cysteine protective against monocrotaline-induced toxicity? Toxin Reviews, 32(3), 47-54.

Karagül H, Altıntaş A, Fidancı UR, Sel T, 2000. Klinik Biyokimya, 1. Baskı, Medisan Yayınevi, Dışkapı-ANKARA, s.1-419.

Mayer F, Lüthy J 1993. Heliotrope poisoning in Tadjikistan. The Lancet, 342(8865), 246-247.

Mulder PP, de Witte SL, Stoopen GM, van der Meulen J, van Wikselaar PG, Gruys E, Groot MJ, Hoogenboom RL 2016. Transfer of pyrrolizidine alkaloids from various herbs to eggs and meat in laying hens. Food Additives & Contaminants: Part A, 33(12), 1826-1839.

Mulder PPJ, Lopez Sánchez P, These A, Preiss-Weigert A, Castellari M 2015. Occurrence of pyrrolizidine alkaloids in food. EFSA Supporting Publications, 12(8), 859E

Özden H, Kilitçi A, Şahin Y, Karaca G, Umudum H, Yıldız A, Dulkadiroğlu E 2020. Healing effects of single-dose triptolide in rats with severe acute pancreatitis. Düzce Medical Journal, 22(2), 105-108.

Ren X, Xu K, Xu J, Mei Q 2023. Melatonin attenuates monocrotaline-induced hepatic sinusoidal obstruction syndrome in rats via activation of Sirtuin-3. J Biochem Mol Toxicol, e23422.

Robinson O, Want E, Coen M, Kennedy R, van den Bosch C, Gebrehawaria Y, Kudo H, Sadiq F, Goldin RD, Hauser ML, Fenwick A, Toledano MB, Thursz MR 2014. Hirmi Valley liver disease: a disease associated with exposure to pyrrolizidine alkaloids and DDT. Journal of hepatology, 60(1), 96-102.

Rubbia-Brandt L, Lauwers GY, Wang H, Majno PE, Tanabe K, X Zhu, A, Brezault C, Soubrane O, Abdalla E, Vauthey JN, Mentha G, Terris B 2010. Sinusoidal obstruction syndrome and nodular regenerative hyperplasia are frequent oxaliplatin-associated liver lesions and partially prevented by bevacizumab in patients with hepatic colorectal metastasis. Histopathology, 56(4), 430-439.

Stewart MJ, Steenkamp V 2001. Pyrrolizidine poisoning: a neglected area in human toxicology. Therapeutic drug monitoring, 23(6), 698-708.

Ward J, Guthrie JA, Sheridan MB, Boyes S, Smith JT, Wilson D, Wyatt JI, Treanor D, Robinson PJ 2008. Sinusoidal obstructive syndrome diagnosed with superparamagnetic iron oxide–enhanced magnetic resonance imaging in patients with chemotherapy-treated colorectal liver metastases. Journal of clinical oncology, 26(26), 4304-4310.

World Health Organization (WHO) 2020. Safety evaluation of certain food additives and contaminants; Pyrrolizidine alkaloids. FAO/WHO Expert Committe on Food Additives (JECFA), Geneva, 2020, Volume 71-S2.

Xu J, Wang W, Yang X, Xiong A, Yang L, Wang Z 2019. Pyrrolizidine alkaloids: An update on their metabolism and hepatotoxicity mechanism. Liver Research, 3(3-4), 176-184.

Yamashita Y, Fujise N, Imai E, Masunaga H 2002. Reduction of monocrotaline-induced hepatic injury by deleted variant of hepatocyte growth factor (dHGF) in rats. Liver 22:302–307.

Zhu L, Wang Z, Wong L, He Y, Zhao Z, Ye Y, Fu PP, Lin G 2018. Contamination of hepatotoxic pyrrolizidine alkaloids in retail honey in China. Food Control, 85, 484-494.

Zhu L, Zhang CY, Li DP, Chen HB, Ma J, Gao H, Yang Y, Wang JY, Fu PP, Lin G 2021. Tu-San-Qi (Gynura japonica): the culprit behind pyrrolizidine alkaloid-induced liver injury in China. Acta Pharmacologica Sinica, 42(8), 1212-1222.

DRAFT

ORAL PRESENTATION

Climate Change's Effects on the Sustainable Food Biosecurity

Başar UYMAZ TEZEL[1]* *(https://orcid.org/ 0000-0002-4156-8861)*, Pınar ŞANLIBABA[2] *(https://orcid.org/0000-0003-4638-6765)*

[*1] Çanakkale Onsekiz Mart University, Bayramiç Vocational School, Laboratory Technology Program, BAYRAMİÇ/ÇANAKKALE/TURKEY
[2] Ankara University, Department of Food Engineering, Faculty of Engineering, GÖLBAŞI/ANKARA/TURKEY

*Corresponding author e-mail: buymaz@comu.edu.tr

Abstract

Anthropogenic disturbances, dating back millennia and increasingly intensifying, have caused the devastating increase in greenhouse gases and the Earth's climate to deviate from its natural cycle. Climate change has been an ongoing fact since the Swedish scientist Svante Arrhenius first articulated in 1896 that the amount of carbon dioxide released into the atmosphere due to human activities could lead to global warming. The literature defines climate change as a long-term change in the statistical properties of the climate system. It is demonstrated by an unusual distribution around the recorded mean over an average period of 30 years. The Intergovernmental Panel on Climate Change "Special Report on the Impacts of Global Warming of 1,5 °C" warned that "climate-related risks to health, livelihoods, food security, water supply, human security, and economic growth will escalate at 1,5°C and worsen beyond 2°C. Climate change exerts its influence not solely on crop production and food security but also extends its impact on food safety and the incidence and prevalence of food-borne diseases.

Climate change's influence on microorganisms is in alterations in metabolic activity, shifts in mortality rates, fluctuations in biomass, heightened biodiversity, and adverse or favourable effects on their physiological functions. Within the context of Sustainable Food Biosecurity, alterations in climatic factors are poised to exert significant impacts on *(i)* sources and modes of transmission, *(ii)* growth and survival, and *(iii)* the ecological dynamics of pathogens within the food supply chain. In the most optimistic scenario, it has been proposed that coupled with climate change, pathogenic microorganisms could potentially lead to food contamination at any stage of the production process, spanning from the farm to the consumer's table. Climate change has been posited to influence the prevalence of climate-sensitive infectious diseases categorised as water-borne, food-borne, and vector-borne.

Keywords: Climate Change, Sustainable Food Biosecurity, Pathogenic Microorganisms

Introduction

In the current discourse, we may say that climate change has gained increased prominence, standing out as one of the foremost threats confronting both Earth's ecosystems and human societies. The United Nations 1994 formally delineated climate change as "*a change of climate which is attributed directly or indirectly to human activity that alters the composition of the global atmosphere and which is, in addition to natural climate variability, observed over comparable periods.*" (United Nations, 1994). This phenomenon was initially articulated in 1896 by the Swedish scientist Svante Arrhenius, who posited that carbon dioxide emissions to the atmosphere by human activity could precipitate global warming (Arrhenius, 1896). From the point of literature perspective, climate change is characterised as a long-term change in statistical properties of the climate system and is demonstrated by an unusual distribution around the recorded mean over an average period of 30 years (Delcour et al., 2015). By the onset of the Industrial Revolution at the end of the eighteenth, anthropogenic destructions, such as agricultural activities, burning of fossil fuel, and deforestation have culminated in global climate change (Grace et al., 2019). These anthropogenic endeavours significantly contribute to the release of greenhouse gases (GHG), notably carbon dioxide (CO_2), methane (CH_4), and

nitrous oxide (N_2O), which serve as the primary agents responsible for instigating climate change (Filho et al., 2022). The nomenclature "greenhouse' originated from the capability to retain these gases' solar heat, thereby making the atmosphere warmer (IPCC, 2007).

Let us delve into the historical context and the ominous trajectory of climate change. Throughout the last 650 millennia, there has been a pronounced escalation in the atmospheric concentration of carbon dioxide (Bergot et al., 2004). Notably, within the preceding two decades, the expansion rate in greenhouse gas concentrations, particularly encompassing carbon dioxide, methane, and nitrous oxide, have accelerated compared to prior decades (Boland et al., 2004; Grace et al., 2019). Furthermore, it is noteworthy that the average global surface temperature has demonstrated an ominous increase, registering a rise of 0.2°C per decade over the past three decades (Boland et al., 2004; Grace et al., 2019). The climatic factors subject to influence encompass temperature, relative humidity, precipitation, and ultraviolet (UV) radiation. These influences give rise to climate variability or alterations in climate conditions, thereby impacting ecosystems. These alterations are discernible through persistent temperature increments, shifts in precipitation quantity and patterns, elevated levels of carbon dioxide and ozone, modifications in the water cycle, and instances of drought. Notably, these transformations are anticipated to persist, even in the event of greenhouse gas concentrations reaching a state of stabilisation. This continued impact can be attributed to the thermal inertia of the system and the protracted timeframe required for the system to return to a lower equilibrium state (Boland et al., 2004; Grace et al., 2019).

When we direct our attention toward apprehensive future scenarios about climate change, the Intergovernmental Panel on Climate Change (IPCC) (driven by four methods for population, economic growth and carbon use, global climate models)have predicted global warming ranging from 1.7 to 4.8 ºC and a rise in the mean global precipitation of 5 to 15% by the end of the century. (IPCC, 2014). The researchers have suggested that a significant shift in the variables may induce meteorological hazards (extreme weather events, with weather variables and frequency below or above the fixed mean threshold) as well as natural calamities, which are sudden localised extreme hydrological, geophysical, meteorological, or climatological events (Duchenne-moutien and Hudaa Neetoo 2021).

Amid the array of climate change-related hazards, it is discerned that *(i)* mycotoxin and marine biotoxin contamination, *(ii)* environmental residues stemming from diverse anthropogenic activities, and *(iii)* zoonotic diseases stand out as emergent risks to human life influenced by climatic factors. This paper aims to provide a comprehensive overview of the prospective ramifications of climate change on both the food industry and food safety, particularly concerning microbial spoilage throughout various stages of the food supply chain.

Climate Change and Food Industry

Food Production and Climate Change

It is advantageous to approach the ramifications of climate change on the food industry from diverse perspectives because climate change does not only mean an increase in average global temperature. The domino effect of climate change includes extreme weather events and natural disasters such as floods, droughts, hurricanes, tsunamis, etc., increased frequency of heavy rainfall events, prolonged dry periods, water acidification and potential rise in sea level. FAO (2008) states that these climate change-related disasters may primarily affect agriculture, fisheries and animal husbandry (FAO, 2008). It is inescapable that the food industry, which relies heavily on agricultural, livestock, and fisheries inputs, will experience direct and/or indirect impacts attributable to climate change on a global scale. Researchers anticipate that the agricultural sector will be most vulnerable to climate change when compared to livestock production or fisheries, as noted by Misiou and Koutmanis (2022). The impact of climate change will manifest prominently for both biotic populations and abiotic factors that play a role in agriculture. Climate changes will influence microbial populations, including insects, pests, vectors, and soil microorganisms, subsequently impacting the prevalence of microorganisms like fungi and viruses. In addition, abiotic factors, such as air pollution, nutrient deficiencies, and extreme temperatures, will affect soil quality, plant health, and crop productivity, as indicated by the Food and Agriculture Organization (FAO, 2008). Nelson et al. (2009) briefly encapsulate the impact of climate change on agriculture as follows:

(i) Climate change is anticipated to diminish yields for critical crops in developing nations.

(ii) The effects of climate change on irrigated yields will exhibit variability.

(iii) Climate change will trigger price escalations for essential agricultural staples, including rice, wheat, maise, and soybeans.

(iv) Elevated feed costs are likely to translate into higher meat prices, resulting in a modest slowdown in meat consumption growth and a more substantial decline in cereal consumption.

(v) Relative to 2000 levels, the availability of food energy in 2050 is projected to decrease.

Soil properties are vulnerable to the effects of climate change. Nevertheless, due to the intricate and uncertain interplay among minerals, chemicals, soil microbial communities, and meteorological phenomena, assessing the consequences of climate change on soil quality and plant growth remains challenging. A more comprehensive comprehension of these interactions, necessitating a multidisciplinary approach, is imperative to enable a pragmatic assessment of climate change on soil quality (Misiou and Koutmanis, 2022).

Soil microbes play a pivotal role in facilitating various essential processes in crop production, including biological nitrogen fixation, nutrient accumulation and utilisation, root and shoot development, disease regulation, and soil quality improvement. The forecasted implications of climate change include alterations in soil microbial composition, given that temperature is one of the pivotal determinants influencing the growth rate of organisms. Higher temperatures trigger consequential alterations in the lipid compositions of cell membranes as part of a physiological response. This temperature increase initiates a cascade of effects, including the loss of labile carbon in the soil, a reduction in the abundance of fungi and Actinobacteria, and an increase in the prevalence of Oligotrophic bacteria, leading to significant shifts within the ecosystem (Jansson et al., 2019). It is estimated that a vast microbiome comprising 40,000 to 50,000 species exists within a gram of soil. In response to climatic changes, bacterial, archaeal, and fungal hyphal communities within the soil undergo modifications, resulting in changes in community structure, a decline in their numbers, and a transition to a dormant state. These alterations also manifest in modifying the soil microbiome composition and reducing its quality, ultimately impacting soil fertility (Berhe et al. 2018). Anticipations suggest that changes in soil microbe composition will lead to modified functional profiles among soil microbes. One notable alteration involves the promotion of Oligotrophic Taxa, which are microbes with a slower growth rate and an ability to thrive in nutrient-poor conditions, over Copiotrophic Taxa, which are microbes adapted for nutrient-rich environments. This shift is attributed to soil warming.

Consequently, this change is expected to influence the cycling of essential elements such as phosphorus, sulphur, and nitrogen in the soil. As warming increases soil inorganic nitrogen levels and expands the plant nitrogen pool size, it concurrently suppresses microbial decomposition rates and alters nitrogen cycling dynamics (Naylor et al., 2020). Furthermore, climate change impacts the microbiome's respiration, affecting soil carbon and nitrogen levels.

Cultivated crops exhibit a heightened susceptibility to fluctuations in climatic conditions. While one might anticipate positive outcomes regarding enhanced CO_2 concentrations and elevated temperatures concerning crop yields, in the context of rice production, the fact that temperature extremes emerge as a significant constraining factor. Furthermore, the evidence underscores that the impact of CO_2 is more pronounced in C3 plants than C4 plants, owing to inherent physiological distinctions (Mirón et al., 2023).

Climate change may have deleterious impacts on food quality, particularly with regard to critical factors such as protein and starch concentrations, which can significantly influence taste and mineral concentrations that play a pivotal role in consumer nutrient intake. This phenomenon finds support in evidence indicating that heightened levels of CO_2 are associated with diminished protein content and reduced quantities of essential minerals, including calcium, sulphur, magnesium, iron, and other trace elements, within the grain (Mirón et al., 2023).

Conversely, factors impacting fodder production can exert profound effects on animal nutrition, thereby influencing the production of food products of animal origin. Empirical findings from studies examining the repercussions of climate change on husbandry reveal the following (Mirón et al., 2023)

(i) Genetically selected animals are more sensitive to environmental changes

(ii) Lower dairy production and greater heat-related mortality

(iii) Lower growth index

(iv) Changes in fertility, increases in mortality due to heat, and economic losses

Both fishing and aquaculture face significant vulnerability in the current climate change scenario, as global trends reveal a noteworthy elevation in ocean temperatures and widespread ocean acidification. Based on empirical findings from a study, the acidification of oceanic waters resulting from heightened atmospheric CO_2 concentrations has been observed to have immediate and direct consequences on calcareous marine species, particularly molluscs such as oysters, clams, and sea snails (Kornder et al., 2018).

Climate Change's Effect on Sustainable Food Biosecurity

Climate change has been recognised as a potential catalyst for the increased contamination of water and food by bacteria, viruses, and pathogens. Several climatic factors, including temperature, humidity, and extreme weather events, can alter the conditions affecting these agents' survival and transmission patterns. These encompass alterations in the occurrence, persistence, dominance, and toxicity of marine and freshwater algal blooms, bacteria, fungi, viruses, parasites, and vectors pathogenic to plants and animals. Within the context of Sustainable Food Biosecurity, alterations in climatic factors are poised to exert significant impacts on *(i)* sources and modes of transmission, *(ii)* growth and survival, and *(iii)* the ecological dynamics of pathogens within the food supply chain. For instance, shifts in temperature can potentially disrupt the reproductive cycles of pathogens, leading to changes in their distribution and proliferation. In certain regions, this may even give rise to the emergence of novel pathogens specifically adapted to particular hosts (Duchenne-moutien and Hudaa Neetoo 2021).

Consequently, more significant outbreaks caused by highly persistent pathogens in the environment, temperature or pH-resistant strains, or those with low infectious thresholds become more likely. Such pathogens include *Mycobacterium avium*, *Salmonella* spp., *Shigella* spp., enterohaemorrhagic *Escherichia coli* (*E. coli* O157:H7), enteric viruses, and parasitic protozoa. As a result, the risks associated with water and food-borne infectious diseases may escalate, potentially leading to the emergence of new risks (FAO, 2020; EFSA, 2020).

Temperature and precipitation are two pivotal climatic factors significantly impacting the incidence of food-borne diseases. In the most optimistic scenario, it has been postulated that temperature escalation may result in *(i)* Augmented prevalence of parasites in freshwater fish and aquatic plants, *(ii)* Identification of novel mycotoxin-producing fungal species in European maise crops, *(iii)* Elevated incidence of mastitis in cattle, *(iv)* Enhanced prevalence of *Salmonella* in poultry, and *(v)* A heightened abundance of *Vibrio* cells in seafood. In the event of a temperature decline, it is predicted that there will be an elevated risk of Norovirus and hepatitis A contamination in fruits.

In the event of Internalization of an increase in precipitation, it is predicted that there will be *(i)* internalisation of pathogenic *E. coli* and *Salmonella* in leafy green vegetables, *(ii)* increased contamination of seafood by faecal indicator organisms due to water runoffs, *(iii)* increase risk of splash dispersal and aerosolised *Salmonella* infecting tomatoes due to increased frequency of short period of heavy rainfall (Duchenne-moutien and Hudaa Neetoo 2021).

Mycotoxins represent secondary metabolites synthesised predominantly by filamentous fungi, primarily belonging to the genera *Aspergillus*, *Fusarium*, *Penicillium*, and *Alternaria*. Among these mycotoxins, the most profoundly toxic include aflatoxins, zearalenone, trichothecenes, fumonisins, and ochratoxins, which are notable for their carcinogenic and immunosuppressive properties, which have adverse effects on both humans and animals. Mycotoxigenic fungi have the potential to infect a diverse range of staple crops, dried fruits, feed crops, fruits, and vegetables and have the potential to pose threats to the safety of these food items. The primary climatic determinants influencing the occurrence and prevalence of mycotoxigenic fungi encompass temperature, humidity, and precipitation. Additionally, a subtle increase in carbon dioxide (CO_2) levels has been recorded to exert an influence, particularly when interacting with temperature and water availability. Variations in these crucial factors may engender food safety apprehensions with regard to mycotoxigenic fungi (Duchenne-moutien and Hudaa Neetoo 2021).

Conclusion

Climate change exerts its influence not solely on crop production and food security but also extends its impact on food safety and the incidence and prevalence of food-borne diseases. Global warming exerts an effect on all microorganisms, encompassing spoilage bacteria and fungi. Consequently, an augmented risk of microbial spoilage is anticipated for food items, including dried products and non-refrigerated processed foods. In the most optimistic scenario, it has been proposed that coupled with climate change, pathogenic microorganisms could potentially lead to food contamination at any stage of the production process, spanning from the farm to the consumer's table. Among the various facets of this complex issue, the most critical and overlooked aspect, the microbiological spoilage of foods, warrants a heightened level of preparedness from both the food industry and policymakers.

It is conspicuously necessary to grapple with the implications of climate change and adopt multidisciplinary approaches, fostering a comprehensive understanding and identification of latent emerging risks.

REFERENCES

Arrhenius S 1896. XXXI. On the influence of carbonic acid in the air upon the temperature of the ground. The London, Edinburgh, and Dublin Philosophical Magazine and Journal of Science, 41(251), 237–276.

Bergot M, Cloppet E, Perarnaud V, Deque M, Marcais B, Desprez-Loustau ML 2004. Simulation of potential range expansion of oak disease caused by Phytophthora cinnamomic under climate change. Global Change Biology, 10: 1539-1552.

Berhe AA, Barnes RT, Six J, Marín-Spiotta E 2018. Role of soil erosion in biogeochemical cycling of essential elements: carbon, nitrogen, and phosphorus. Annu. Rev. Earth Planet Sci. 46 (1) 521–548.

Boland GJ, Melzer MS, Hopkin A, Higgins V, Nassuth A 2004. Climate change and plant diseases in Ontario. Canadian Journal of Plant Pathology-Revue Canadienne de Phytopathologie, 26: 335-350.

Delcour I, Spanoghe P and Uyttendaele M. 2015. Literature review: impact of climate change on pesticide use. Food Res. Int. 68:7–15.

Duchenne-Moutien RA, Neetoo H. 2021. Climate Change and Emerging Food Safety Issues: A Review, Journal of Food Protection, Vol. 84, No. 11, 1884–1897.

EFSA. 2020. Climate Change as a Driver of Emerging Risks for Food and Feed Safety, Plant, Animal Health and Nutritional Quality. EFSA supporting publication 2020.

FAO. 2008. Climate change: Implications for food safety. Retrieved October 21, 2020, from http://www.fao.org/3/i0195e/i0195e00.htm.

FAO. 2020. Climate change: Unpacking the burden on food safety. https://doi.org/10.4060/ca8185en.

Filho WL, Setti AFF, Azeiteiro UM, Lokupitiya E, Donkor FK, Etim NA, Matandirotya N. 2022. An overview of the interactions between food production and climate change. Science of the Total Environment 838, 156438.

Grace MA, Achick TE, Bonghan BE, Bih ME, Ngo NV, Ajeck MJ, Bobyiga GT, et al. 2019. An Overview of the Impact of Climate Change on Pathogens, Pest of Crops on Sustainable Food Biosecurity. International Journal of Ecotoxicology and Ecobiology, 4(4): 114-124.

IPCC, 2007. Climate Change 2007: Working Group I: The Physical Science Basis. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

IPCC. 2014. Summary for policymakers. In: Edenhofer O, Pichs-Madruga R, Sokona Y, et al., eds. Climate Change 2014, Mitigation of Climate Change. Contribution of Working Group III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge, United Kingdom and New York, NY, USA: Cambridge University Press.

Jansson JK, Hofmockel KS. 2019. Soil microbiomes and climate change, Nat. Rev.Microbiol. 18 (1) 35–46.

Kornder NA, Riegl BM, Figueiredo J. 2018. Thresholds and drivers of coral calcification responses to climate change. Global Change Biol. 24 (11), 5084–5095.

Mirón IJ, Linares C, Díaz J. 2023. The influence of climate change on food production and food safety. Environmental Research 216, 114674.

Misiou O, Koutsoumanis K. 2022. Climate change and its implications for food safety and spoilage. Trends in Food Science & Technology 126, 142–152.

Naylor D, Sadler N, Bhattacharjee A, Graham EB, Anderton CR, McClure R, Lipton M, Hofmockel KS, Jansson JK. 2020. Soil microbiomes under climate change and implications for carbon cycling. Annu. Rev. Environ. Resour. 45 (1) 29–59.

Nelson GC, Rosegrant MW, Koo J, Robertson R, Sulser T, Zhu T, Ringler C, Msangi S, Palazzo A, Batka M, Magalhaes M, Valmonte-Santos R, Ewing M, Lee D. 2009. Climate Change: Impact on Agriculture and Costs of Adaptation. International Food Policy Research Institute, Washington.

United Nations 1994. *United Nations framework convention on climate change* (New York).

Uyttendaele M, Liu C, and Hofstra N. 2015. Special issue on the impacts of climate change on food safety. Food Res. Int. 68:1–6.

DRAFT

ORAL PRESENTATION

Biosorption, an efficient process for removing heavy metals from wastewater: A mini review

Derya Altintas[1*] *(ORCID: https://orcid.org/0000-0001-5374-0123)*,
Yesim Yesiloglu[2*] *(ORCID: https://orcid.org/0000-0002-1733-4790)*
Yuksel Bayrak[2*] *(ORCID: https://orcid.org/0000-0002-5008-456X)*

[*1]Trakya University, Arda Vocational College, Department of Pharmacy Services, Edirne, Turkey.
[*2]Trakya University, Faculty of Pharmacy, Department of Biochemistry, Edirne, Turkey.
[*2]TrakyaUniversity, Faculty of Science, Department of Chemistry, Edirne, Turkey.

*Corresponding author e-mail: deryaaltintas@trakya.edu.tr

Abstract

The heavy metal pollution constitutes a pressing environmental problem. Even if heavy metals have entered the organism in trace amounts by respiratory or skin, they are slow to be removed from metabolism and therefore reach dangerous dose as a result of accumulation. The effects that this dangerous dose will create in the body vary depending on the accumulation concentration, chemical properties, ion status, redoxing ability, solubility and complexization of heavy metal. This problem has stimulated great efforts to develop treatment sustainable techniques for the heavy metal removal from wastewater including chemical oxidation, ion exchange, precipitation, adsorption, coagulation and flocculation, filtration, reverse osmosis, biosorption and membrane seperation. This review focuses on experimental studies in the literature of using several biological materials for the biosorption of heavy metals from wastewater and we concluded that biosorption is successful techniques because of its high removal activity, economic effectiveness and enviroment friendly.

Keywords: Biosorption, wastewater, biomass, heavy metal.

INTRODUCTION

Heavy metals; It is defined as metallic elements with high specific gravity, which can have a toxic effect at low concentrations (Velarde et al., 2023). It is known that heavy metals with carcinogenic effects cause damage to DNA. Unprotected exposure in the same environment as some heavy metals accumulating in the brain and all other tissues causes serious poisoning. Different methods have been proposed for the removal of the mentioned pollution, including chemical precipitation, chemical oxidation or reduction, filtration, ion exchange, reverse osmosis, biosorption, electrochemical treatment, evaporation, advanced oxidation processes and membrane technologies (Fei and Hu, 2023). Although many methods have been used in the treatment of wastewater containing heavy metals, the use of biological materials has increased in the removal of heavy metals from wastewater due to their lower cost. The biosorption method, which provides the removal of heavy metals using biosorbents, can be given as an example. Live or non-living biological materials can be used as biosorbents and the reason why the method is preferred is that the biological materials used in biosorption are generally cheap and abundant (Beni and Esmaeili, 2020). In this study, the usability of the biosorption method was examined and studies in the literature were mentioned.

HEAVY METALS AND THEIR HARMFUL EFFECTS ON HUMAN HEALTH

‘‘Heavy metals’’ refer to metal and metalloids that have an atomic density greater than 5 g/cm^3 and its pollution has become a global enviromental problem (Aryal et al., 2022). Heavy metals with serious toxic hazards to human body include lead (Pb), mercury (Hg), cadmium (Cd), chromium (Cr), nickel (Ni) and arsenic (As) (Wang and Matsushita, 2021). Heavy metal accumulation in the human body can cause serious health issues such as cellular poisoning, brain damage, cancer, neurotoxicity, renal damage, genotoxic abnormalities, pulmonary diseases, birth defects, diabetes, intestinal irritation, allergic reaction on the skin, chronic anemia, asthma, diarrhea and cardiovascular diseases (Mishra et al., 2021) (Figure 1). Therefore it is a necessity to develop sustainable techniques for the removal of heavy metals from wastewater.

DRAFT

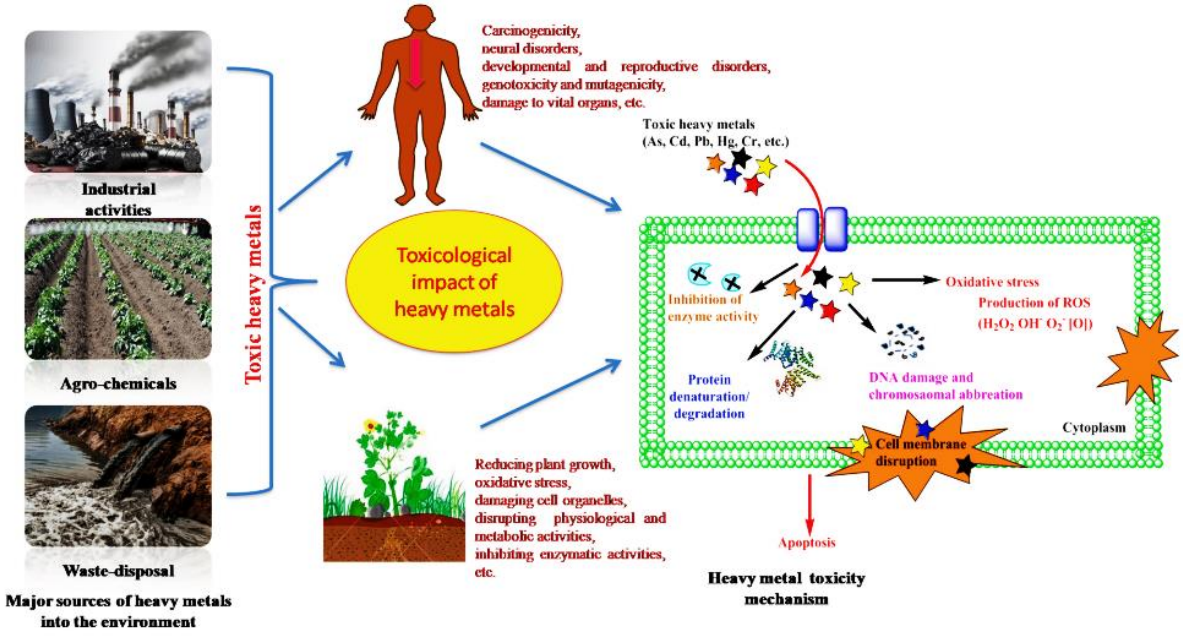


Figure 1. Toxicological impact of heavy metals (Mishra et al., 2021).

CONVENTIONAL METHODS FOR HEAVY METAL REMOVAL FROM WASTEWATER

In the last decade, several processes treat water containing heavy metals including chemical oxidation, ion exchange, precipitation, adsorption, coagulation and flocculation, filtration, reverse osmosis, catalysis, biosorption and membrane seperation (Jyoti et al., 2022). Performance comparison of these methods for heavy metal removal are given in Table 1.

Table 1. Performance comparison of conventional methods for heavy metal removal (Fei et al., 2023; Jyoti et al., 2022).

Method	Advantage	Disadvantage
Reverse osmosis	Resistance to high temperatures	High pressure requirement
Ion exchange	High efficiency	Low capital cost, high maintenance cost
Membrane separation	Simple operation	Membrane fouling
Catalysis	High efficiency for chelating wastewater	High operation cost
Flotation	High efficiency and selectivity	High capital, operation, and maintenance cost

DRAFT

The flotation method is widely used in heavy metal removal due to its high metal selectivity. In this method, which is intended to be removed from the wastewater phase of heavy metal particles, high cost is undesirable. In the reverse osmosis method, it separates the dissolved salts of heavy metals from a semi-permeable membrane and wastewater and fresh water. In the method, a certain concentration of salt is added to the compartment where the wastewater is present and the formation of osmotic pressure is ensured. With a higher-pressure application than osmotic pressure, the passage through the membrane is ensured and heavy metal cations can be separated from the water in which they are located (Fei et al., 2023). Adsorption has been the most used method for removal of heavy metals from wastewater. Physical, exchange and chemical adsorption are carried out in the removal of pollution. It starts with the transfer of the heavy metal ions from the solution to the surface of the adsorbents, followed by the binding between the heavy metal ions and the surface through physical or chemical interactions. Activated carbon is an adsorbent commonly used in the removal of heavy metal ions (Jyoti et al., 2022). Common causes of use include a large surface area and porous structure. Due to the cost of activated carbon, biological materials have started to be preferred in heavy metal removal from wastewater.

BIOSORPTION

Biosorption is the method of removal of heavy metals using biosorbents. Heavy metal ions adsorb through surface precipitation, electrostatic and Van der Waals interactions or by variation of these processes through physical adsorption. Figure 2 shows the different mechanisms involved in the biosorption (Elgarahy et al., 2021). A range of biomass products, such as pepper seed, walnut leaf, tea pulp, apple peel, cranberry pulp, banana peel, lemon pulp, marine algae and bacterial cells are effective as metal biosorbent (Priya et al., 2022). This method is used not only for the removal of heavy metals by biosorption, but also for the removal of toxic dyes.

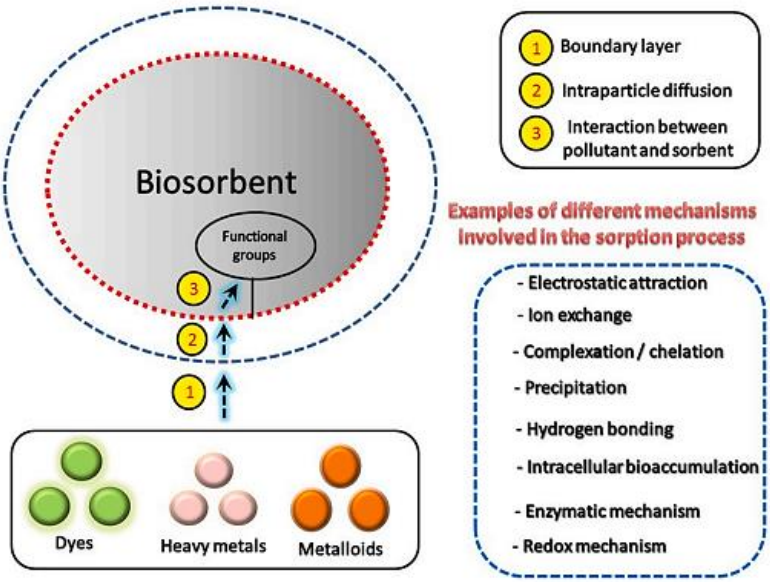


Figure 2. Examples of different mechanisms involved in the biosorption (Elgarahy et al., 2021).

In addition, the fact that biological materials to be used in biosorption are generally cheap and abundant is the reason for the method to be preferred. The advantages of the biosorption method are shown in Figure 3. Waste formation after biosorption is considerably less than after adsorption. The possibility of losing the characteristic of biological material or its rapid access to saturation and its inability to be easily controlled are the disadvantages of the biosorption method (Priya et al., 2022). However, these disadvantages started to be overcome by physical or chemical modifications.

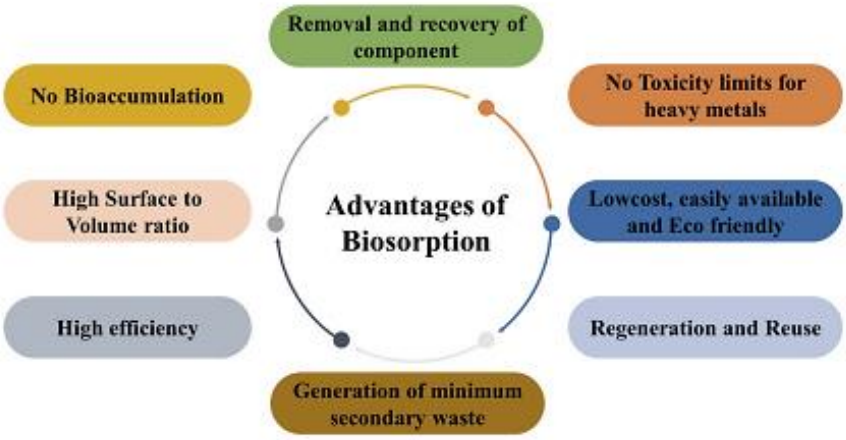


Figure 3. The advantages of the biosorption method (Priya et al., 2022).

The biosorption method can proceed as an active process or passive process. Biological material that is alive in the active process is used and there is accumulation of metabolic activities and pollution on the living surface. In contrast, in the passive process, biosorption takes place before metabolic activities are engaged. In heavy metal biosorption, one or more of its interactions between heavy metal ion and biosorbent, such as ion exchange, micro-level precipitation, biosorption to the cell surface and cell membrane transport can be found in the same mechanism (Syeda et al., 2022). In the biosorption on the cell surface, interaction occurs between functional groups on the surface and heavy metal ions, and physical adsorption occurs. In the process of micro-level precipitation, heavy metal ion accumulates on the surface of the biosorbent cell in metal hydroxide, resulting in precipitation. Finally, in the process of ion exchange, cations (K^+, Na^+, Ca^{+2}, Mg^{+2}) naturally linked to functional groups on the biosorbent surface are replaced by metal cations in the solution (Elgarahy et al., 2021). Apart from these, it is necessary to look at the factors affecting the biosorption process. The first of the factors affecting the biosorption process is pH changes in solution and it affects the surface charge of biomass and the ionization degree of the functional group (aldehydes, carboxyl, amino, glycoproteins and polysaccharides) linked with it. In addition, the temperature of the solution with heavy metal ions affects the biosorption process. Finally, it is necessary to mention the biosorption isotherm equations. Freundlich and Langmuir are the two most used models to define the biosorption of heavy metal ions by biomass. Among these 2 models, the model that best describes the process of biosorption between the biosorbent and biosorbate system is the Langmuir isotherm model (Liu et al., 2023).

The Langmuir isotherm is presented in Eqs. (1) and (2).

Eq. (1): $q_e = (q_{max}\, b\, C_e) / (1 + b\, C_e)$

Eq. (2): $C_e / q_e = 1 / (b\, q_{max}) + (C_e / q_{max})$

q_{max} (mg/g): Biosorption capacity

b (L/mg): Langmuir constants related to the energy.

The Freundlich isotherm is presented in Eqs. (3) and (4).

Eq. (3): $q_e = K_F\, C_e^{1/n}$

Eq. (4): $\log C_e/ q_e = (1 - 1/n) \log C_e - \log k_f$

K_F (mg/g) and n: Freundlich constants related to the temperature and intensity of the biosorbent.

In addition to these summaries, biosorption thermodynamics also provides important information. Gibbs free energy change in the biosorption method (ΔG°) that is, the degree of spontaneous occurrence of the method, the thermodynamic parameters such as enthalpy change (ΔH°) and entropy change (ΔS°) provide information about the thermodynamics of the biosorption (Liu et al., 2023).

The Gibbs free energy change is presented in Eqs. (5) and (6).

Eq. (5): $\Delta G^0 = -RT \ln K_C$

Eq. (6): $\Delta G^0 = \Delta H^0 - T\Delta S^0$

ΔG°: Gibbs free energy change

Kc: Equilibrium constant (unitless)

ΔH°: Enthalpy change

ΔS°: Entropy change

STUDIES ON BIOSORPTION OF HEAVY METALS FROM WASTEWATER

In a study conducted by Atılgan, the biosorption potential of the biosorbent prepared with fish scales was examined for the removal of cadmium (Cd^{2+}) from wastewater. The highest removal efficiency was recorded as 99.02% when fish scale biochar was 0.35 g (Atılgan M, 2021).

Ayob et al. modified pineapple waste using NaOH for the removal of Pb^{+2} from wastewater and biosorption capacity increased from 52.57% to 85.88% after modification (Ayob et al., 2020).

In another study Ozturk et al. conducted by the biosorption of Ni^{+2} ion was examined using waste tea. When using 0.025 g of waste tea in biosorption, the removal efficiency was calculated as 15.80%, while the yield increased to 48.40% when using 0.20 g of waste tea. This study shows that tea waste can be an alternative in the biosorption of heavy metals (Ozturk et al., 2020).

In a study conducted by Gurel et al., the biosorption of Ni^{+2} ions from the wastewater were investigated using the pepper seed. The maximum biosorption capacity was recorded as 4.2 mg/g under conditions determined as the most appropriate pH value of 5.5. It has been demonstrated by this study that raw pepper seeds have the potential to remove Ni^{+2} ions (Gurel et al., 2020).

In a study conducted by Mohamed et al., the authors modified banana peel for the removal of Pb^{2+} and Fe^{2+} ions. The results showed that the maximum biosorption capacity reached 100% and 64%, respectively (Mohamed et al., 2020).

In another study, Ozudogru conducted by the biosorption of thorium metal from wastewater using *Cystoseira barbata* an algae. It determined that the biosorption efficiency at optimum pH value increased by 95% and the maximum biosorption capacity was 36.45 mg/g in the experiments performed (Ozudogru et al., 2019).

CONCLUSION

This review focuses on studies in the literature of using several biological materials for the biosorption of heavy metals from wastewater. Different methods have been proposed for the removal of the mentioned pollution, including chemical precipitation, chemical oxidation or reduction, filtration, ion exchange, reverse osmosis, biosorption, electrochemical treatment, evaporation, advanced oxidation processes and membrane technologies. Although many methods have been used in the treatment of wastewater containing heavy metals, the use of biological materials has increased in the removal of heavy metals from wastewater due to their lower cost. In addition, the fact that biological materials to be used in biosorption are generally abundant is the reason for the method to be preferred. Finally, we concluded that biosorption is successful techniques because of its high removal activity, economic effectiveness and enviroment friendly.

REFERENCES

Aryal LR, Thapa A, Poudel BR, Pokhrel MR, Dahal B, Paudyal H, Ghimire KN, 2022. Effective biosorption of arsenic from water using La (III) loaded carboxyl functionalized watermelon rind. Arabian Journal of Chemistry, 15:103674.

Atılgan M, 2021. Cd^{2+} Biosorption Potential of Biochar Obtained from Fish Scales. Master of Science Thesis, Eskişehir Osmangazi University.

Ayob A, Zamre NM, Izzati N, Ariffin M, Hidayu N, Rani A, Mohamad NF, 2020. Pineapple waste as an adsorbent to remove lead from synthetic wastewater. International Journal of Latest Research in Engineering and Management, 04:1-8.

Beni AA, Esmaeili A, 2020. Biosorption, an efficient method for removing heavy metals from industrial effluents: A Review. Environmental Technology & Innovation, 17:100503.

Elgarahy AM, Elwakeel KZ, Mohammad SH, Elshoubaky G.A, 2021. A critical review of biosorption of dyes, heavy metals and metalloids from wastewater as an efficient and green process. Cleaner Engineering and Technology, 4:100209.

Fei Y, Hu YH, 2023. Recent progress in removal of heavy metals from wastewater: A comprehensive review. Chemosphere, 335:139077.

Gurel L, Mese T, Seker A, Kısırkaya A, Yıldırım AS, 2020. The nickel removal from wastewaters by biosorption using capia pepper residual. Nigde Omer Halisdemir University Journal of Engineering Sciences, 9(1):84-95.

Jyoti D, Sinha R, Faggio C, 2022. Advances in biological methods for the sequestration of heavy metals from water bodies: A review. Environmental Toxicology and Pharmacology, 94:103927.

Liu J, Zhang C, Tao B, Beckerman J, 2023. Revealing the roles of biomass components in the biosorption of heavy metals in wastewater by various chemically treated hemp stalks. Journal of the Taiwan Institute of Chemical Engineers, 143:104701.

Mishra S, Lin Z, Pang S, Zhang Y, Bhatt P, Chen S, 2021. Biosurfactant is a powerful tool for the bioremediation of heavy metals from contaminated soils. Journal of Hazardous Materials, 418:126253.

Mohamed RM, Hashim N, Abdullah S, Abdullah N, Mohamed A, Daud MA, Muzakkar KF, 2020. Adsorption of heavy metals on banana peel bioadsorbent, American Physical Society Sites, 1532.

Ozturk M, Yıldız S, Aslan S, 2020. Biosorption of nickel (II) ions onto tea waste: equilibrium. Journal of Engineering Sciences and Design, 8(4):985-998.

Ozudogru Y, 2019. Biosorption of radioactive thorium by Cystoseira barbata. DEU Journal of Science and Engineering, 62:461-468.

Syeda HI, Sultan I, Razavi KS, Yap PS, 2022. Biosorption of heavy metals from aqueous solution by various chemically modified agricultural wastes: A review. Journal of Water Process Engineering, 46:102446.

Velarde L, Nabavi MS, Escalera E, Antti ML, Akhtar F, 2023. Adsorption of heavy metals on natural zeolites: A review. Chemosphere, 328:138508.

Wang H, Matsushita MT, 2021. Heavy metals and adult neurogenesis. Current Opinion in Toxicology, 26:14-21.

DRAFT

ORAL PRESENTATION

Investigation of physical and chemical properties of carbon dots doped with metal ions

Melis Özge ALAŞ ÇOLAK[1] (*ORCID: 0000-0002-0546-087X*), Pınar KARACABEY (*ORCID: 0000-0002-7875-5847)*[2], Rükan GENÇ ALTÜRK[1,2*] (*ORCID: 0000-0002-9569-8776*)

[1] Sabanci University, SUNUM Nanotechnology Research Centre, Istanbul, Turkey.
[2] Department of Chemical Engineering, Faculty of Engineering, Mersin University, Mersin, Turkey.

*Corresponding author e-mail: rukan.genc@sabanciuniv.edu

Abstract

Doping has proven to be an effective approach for enhancing the physicochemical properties of carbon dots (CDs) and has garnered increasing attention in recent years. The incorporation of metal ions into the CD structure is a straightforward strategy for imparting additional physicochemical properties to CDs. Metal ions possess more electrons, vacant orbitals, and larger atomic radius. When these ions are doped into CDs, they can modify the electron density distribution and energy gap, resulting in improvements in their optical, electronic, and magnetic properties. In this study, thermal synthesis was employed to refine the one-pot synthesis of CDs, including the preparation of metal-doped CDs. Fluorescence emission, FT-IR, and Zeta potential analyses were performed to examine the effects of different metals (Sr^{+2}, Cu^{+}, Na^{+2} and Ca^{+2}) on the physical and chemical properties of CDs. Our observations revealed that metal doping influenced the fluorescence color and the maximum emission peak of CDs. Furthermore, metal-doped CDs exhibited a more positive surface charge compared to bare CDs. This suggests that Sr^{+2}, Cu^{+}, Na^{+2} and Ca^{+2} ions are predominantly located inside the carbon dots, particularly on or near the surface. These improved physicochemical properties make CDs promising and competitive materials for a wide range of applications, including detection, imaging, phototherapy, optoelectronics, and catalysis.

Keywords: carbon dots, metal doping, fluorescence, thermal synthesis

INTRODUCTION

Carbon dots (CDs) are emerging as competitive zero-dimensional nanomaterials with a hemispherical structure, featuring a crystalline sp2 carbon core and abundant amino, carboxyl, and hydroxyl groups on their surface (Mansuriya and Altintas, 2021.). CDs can exhibit excitation wavelength-dependent, and independent fluorescence emissions (Gallareta-Olivares *et al.*, 2023.). In comparison to other photoluminescent (PL) materials, such as organic dyes or conventional semiconductor quantum dots (QDs), these new CDs offer several advantages, including ease of preparation, surface modification capabilities, excellent biocompatibility, low cytotoxicity, good water dispersibility, and tunable fluorescence properties (Alaş *et al.*, 2022.). The unique physicochemical properties of CDs make them suitable for a wide range of applications, including energy storage and production, chemical sensing, biological imaging, phototherapy, and optoelectronic devices (Kandasamy, 2019). Recently, methods such as thermal, hydrothermal, and microwave/microwave-assisted synthesis, known for being faster, more cost-effective, and environmentally friendly, have gained preference (Simsek *et al.*, 2019.). In CD synthesis, natural resources like apples, coffee, honey, and molasses are utilized as carbon sources to minimize chemical usage and reduce process costs (Alas and Genc, 2017.; Vinoth Kumar *et al.*, 2021.). Researchers are trying to produce higher-quality CDs for practical applications regarding quantum efficiencies, stability, and narrow or tuned emission wavelengths. Therefore, two effective approaches, surface passivation and chemical doping, have been developed recently to improve CDs' fluorescence properties and performance.

Doping is a valuable method for controlling the physicochemical properties of CDs, and it has garnered significant attention from researchers in recent years (Yan *et al.*, 2019.). The introduction of heteroatoms/metals into the nanostructure of CDs can induce substantial changes in their chemical composition, electronic structures, and nanostructure. This occurs due to the push/pull electron effect of

heteroatoms/metals, resulting in the emergence of various intrinsic properties. Consequently, the photoluminescent (PL) properties of CDs can undergo alterations owing to the varying HOMO-LUMO band gaps in heteroatom/metal-doped CDs. In comparison to non-metallic atoms, metal ions possess larger atomic radii, more electrons, and vacant orbitals. Therefore, when metal ions are doped, there is a shift in electron density distribution and energy levels, enabling the adjustment of the physical and chemical properties of CDs (Dang and Pham, 2022.; Li *et al.*, 2022.; Gallareta-Olivares *et al.*, 2023.). n heteroatom doping on the surface functional groups of carbon dots (CDs), researchers have employed heteroatoms such as nitrogen (N), fluorine (F), sulfur (S), phosphorus (P), silicon (Si), and boron (B) to fine-tune emission properties and enhance quantum yield (QY) (Zhao *et al.*, 2022.). Most of the studies conducted thus far have focused on nitrogen doping (N@CDs), which has consistently led to improved fluorescence in CDs (Yan *et al.*, 2019.). Additionally, recent reports have highlighted the ability to modulate the color emission, QY, and decay lifetime of CDs to some extent through the addition of metal elements (Yan *et al.*, 2019.; Li *et al.*, 2022.; Gallareta-Olivares *et al.*, 2023.). For instance, Liu *et al*. produced yellow fluorescent manganese-doped CDs (Mn-CDs). In comparison to pure CDs, Mn-CDs exhibited increased coverage of conjugated π-domains and surface states, resulting in a redshifted emission wavelength. The fluorescence QY and lifetime of Mn-CDs reached 46% and 8.11 ns, respectively, demonstrating higher QY (68.6%) and longer lifetime (8.75 ns) compared to pure CDs. These improvements were attributed to the development of passivated defects on the surface of Mn-CDs due to manganese doping (Liu *et al.*, 2018.). In another study, transition metal ion (Mn^{2+}, Fe^{2+}, Co^{2+} and Ni^{2+}) doped CDs (TMCDs) were synthesized from lemon juice using a one-pot microwave method. TMCDs exhibited excitation wavelength-dependent emission with brighter photoluminescence at 460 nm. When compared to pure CDs with a 48.31% PL QY and 3.6 ns fluorescence lifetime, synthesized Mn/C, Fe/C, Co/C, and Ni/CDs exhibited QY values of 35.71%, 41.72%, 75.07%, and 50.84%, respectively, along with enhanced fluorescence lifetimes of 9.4, 8.6, 9.2, and 8.9 ns, respectively (Murugan *et al.*, 2018.).

In this current study, metal-doped CDs (Sr^{+2}, Cu^{+}, Na^{+2} and Ca^{+2}) were synthesized from lemon salt using a one-pot thermal synthesis method to enhance the optical and physical properties of CDs. The characterization included fluorescence emission spectroscopy, Zeta Potential measurements, and Fourier Transform Infrared Spectroscopy (FTIR) to determine the physical properties of the particles and surface functional groups. Additionally, the metal-doped CDs emitted greenish-yellow or blue fluorescence under UV light, with photoluminescence quantum efficiency (QY) ranging from approximately 3% to 32%.

MATERIALS AND METHODS

Lemon salt was procured from local supermarket. Urea and ethanol were purchased IsoLab. Ultrapure water used throughout all the experiments was purified by the Milli-Q system (Millipore Inc., Ω=18 MΩ cm). All chemicals used in the synthesis of the materials were used without purification. Strontium nitrate, copper (I) chloride, sodium oxalate, calcium chloride dihydrate, and potassium chlorate ($KClO_3$) were purchase Sigma-Aldrich.

Metal doped CDs were synthesized using the same method we applied before (Alaş *et al.*, 2019., 2022.). In the synthesis, citric acid was used as a carbon source and urea was used as a nitrogen additive. First, citric acid, urea and metal ion precursor (2:2:1 w/w) were dissolved in water/ethanol (1:1 v/v). The resulting solution mixture was taken into a ceramic container and heat treated at 250 °C for 45 minutes. After the reaction, final material was dissolved in Milli-Q water and centrifuged at 8000 rpm for 20 minutes to precipitate aggregates. The supernatant was collected and the final product was dried at 90 ˚C overnight. In addition, metal-free CDs were synthesized by the same procedure without metal. The schematic representation of metal doped CD synthesis is shown in Figure 1.

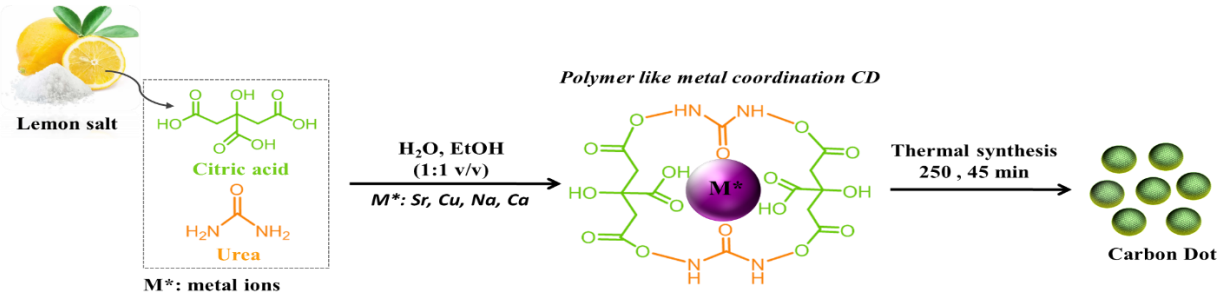


Figure 1. The schematic representation of metal doped CD synthesis.

DRAFT

RESULTS and DISCUSSION

Metal-doped carbon dots (CD) have been synthesized from lemon salt which nature carbon source by one-pot thermal synthesis (Figure 1). The UV-Vis spectrum is given in Figure 2a. CDs and metal-doped CDs exhibited peaks at 270 nm attributed to the π-π* transition of the C=C bond (Alaş *et al.*, 2019.). Also, the peaks centered at 336 nm and 400 nm are due to the n-π* electronic transition of the C=N and C=O bond (Cheng *et al.*, 2018.; Alaş *et al.*, 2019.). CD and metal-doped CDs dissolved in water display greenish yellow (CD, Sr-CD, Cu-CD, and Ca-CD) and blue (Na-CD) fluorescence under 365 nm UV light (Figure 2b inset). When the emission spectra (λ_{nm}:365 nm) of each CD are examined, CD, Sr-CD, Cu-CD, Na-CD, and Ca-CD have maximum emission maxima of 490, 471, 500, 456, and 498 nm, respectively. The PL spectra of Cu-CD and Ca-CD showed two overlapping peaks attributed to the π-π* transition of C-C over the sp^2 domain of CDs (high electron transition) and the n-π* transition as the edgeband transition of the surface or doped groups (low electron transition). Depending on the interaction of metal ions acting as additives with the CD surface, PL is increased or quenched. The fluorescent emission intensity of metal-doped CDs was slightly lower than that of pure CDs. The decrease in fluorescence intensity and the change in fluorescence colors are due to metal doping in CDs (Kumar *et al.*, 2019.). The PL quantum yield (PL QY) was calculated to be 32.26, 12.14, 2.79, 7.13, and 6.23% for CD, Sr-CD, Cu-CD, Na-CD, and Ca-CD, respectively.

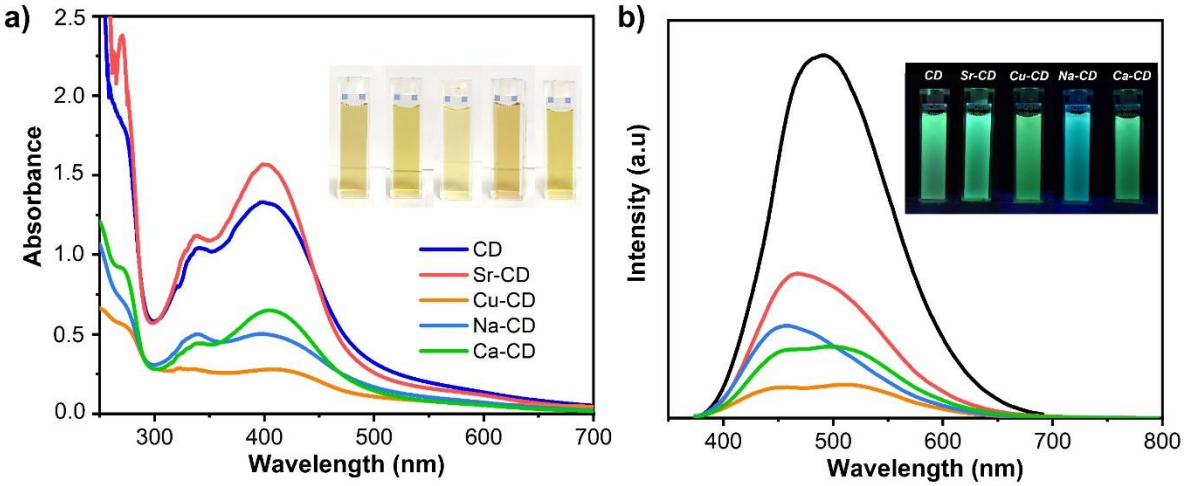


Figure 2. UV/Vis and fluorescence emission spectrums of metal-doped CDs.

Table 1. Physical properties of synthesized CDs. Fluorescence emission maxima (λm) and Quantum Yield (QY) measured at λ_{exc}:365 nm. Hydrodynamic radius (*Rh*), and Zeta Potential (ζ-Pot) of the samples measured in Milli-Q water.

Sample	λ_{em} (nm)	QY (%)	ζ-Pot (mV)	*Conductivity* (mS/cm)
CD	490	32.26	-13.57 ± 1.57	0.05 ± 0.003
Sr-CD	471	12.14	-11.70 ± 1.07	0.11 ± 0.004
Cu-CD	500	2.79	-5.45 ± 0.86	0.49 ± 0.018
Na-CD	456	7.13	-13.43 ± 0.83	0.15 ± 0.0005
Ca-CD	498	6.23	-11.87 ± 0,40	0.17 ± 0.009

The surface net charges and conductivity values of the synthesized CDs were determined by dynamic light scattering method (DLS) using Zeta Sizer device. The surface charge of metal-doped CDs was analyzed through zeta potential measurements and compared to that of pure CDs. When Table 1 is examined, it is seen that all of the samples have negative zeta potentials. Metal doping into CDs showed increased positive zeta potentials (ζ-Pot) of CDs. The positive shifting zeta potentials prove that metallic Sr^{+2}, Cu^{+}, Na^{+2} and Ca^{+2} ions are located inside the carbon dots, especially above or near the surface (Kumar *et al.*, 2019.).

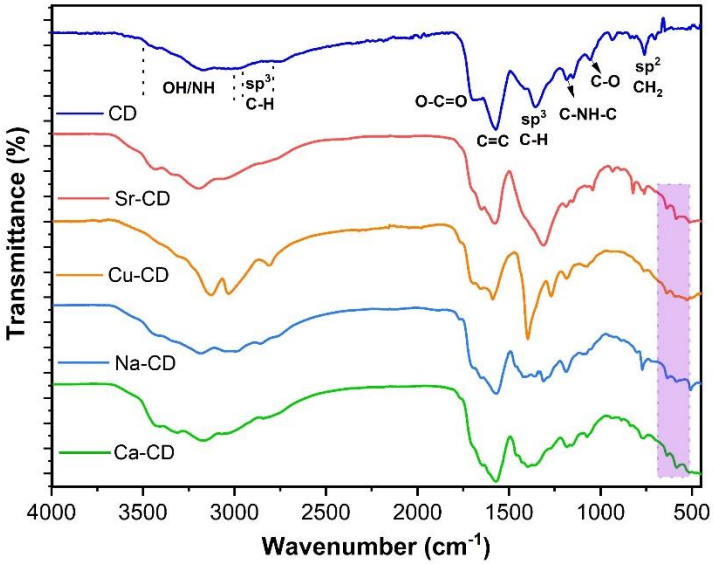


Figure 3. FT-IR spectrums of CD and metal-doped CDs.

The functional groups on the surface of pure CDs and metal-doped CDs were examined using FT-IR spectroscopy and are given in Figure 3. In their spectra, all CDs showed broad absorption bands. Peaks, which is attributed to the OH/N-H stretching vibration show at between 3500-3100 cm^{-1}. Peaks of aldehyde C-H stretching vibration at 2700-2760 cm^{-1}, C=O stretching vibration indicating the formation of aromatic carbonyl bonds at 1715-1700 cm^{-1}, and asymmetric and symmetrical C-O-C vibration peaks at 1000-1350 cm^{-1} were observed. Also, the peaks at about 1600-1500 cm^{-1} and 1200-1100 cm^{-1} are attributed to vibrations of the C-NH-C bond of aromatic C=C and amine groups on the surface of CDs, respectively. The vibrations between 1460-1380 cm^{-1} and 770-730 cm^{-1} are attributed to the sp^3 C-H and aromatic sp^2 C-H bands, respectively (Alaş, 2022.; Alaş *et al.*, 2022.). Spectra of metal-doped CDs showed peaks not in the pristine CD spectrum at below 750 cm^{-1}, confirming the formation of metal oxide traces on the CD surface (Murugan *et al.*, 2018.).

CONCLUSION

In summary, one-pot thermal "green" synthesis of metal-doped CDs have been successfully demonstrated using lemon salt. Both Pure CD and metal-doped CDs revealed characteristic absorbance peaks due to π-π* electronic transition, excitation-wavelength dependent PL behavior and PL-QY values. The decreased PL emission intensity and the resulting QY for metal-doped CDs were attributed to non-radiative electronic transition caused by complex formation between metal ions and CDs surface functional groups. CDs exhibited negative ζ potential due to the rich abundance of oxo-functional groups explaining the better colloidal stability of CDs in aqueous medium. With metal doping, the surface charge of CDs shifted to positive, indicating that metal ions were present in CDs. The chemical composition and surface functional groups of the CD samples and the presence of trace amounts of additive metal ions in the CD were determined by FT-IR analysis. The present study provides an easy strategy for the synthesis of fluorescent metal-doped CDs whose physicochemical properties can be improved for use in applications such as detection, imaging, phototherapy, optoelectronics, and catalysis.

REFERENCES

Alaş MO, Güngör A, Genç R, and Erdem E, 2019. Feeling the power: Robust supercapacitors from nanostructured conductive polymers fostered with Mn2+ and carbon dots. Nanoscale, 11(27): 12804–12816.

Alaş MÖ, 2022. *Enerji, optik sensör ve aktif gida paketleme alanlarinda kullanilmak üzere yüzeyi işlevselleştirilmiş floresans karbon noktalarin geliştirilmesi.*

Alaş MÖ, Doğan G, Yalcin MS, Ozdemir S, and Genç R, 2022. Multicolor Emitting Carbon Dot-Reinforced PVA Composites as Edible Food Packaging Films and Coatings with Antimicrobial and UV-Blocking

Properties. ACS Omega, 7(34): 29967–29983.

Alas MO, and Genc R, 2017. An investigation into the role of macromolecules of different polarity as passivating agent on the physical , chemical and structural properties of fluorescent carbon nanodots. Journal of Nanoparticle Research, 19(5): 185–199.

Cheng W, Xu J, Guo Z, Yang D, Chen X, Yan W, and Miao P, 2018. Hydrothermal synthesis of N,S co-doped carbon nanodots for highly selective detection of living cancer cells. Journal of Materials Chemistry B, 6(36): 5775–5780.

Dang DK, and Pham TT, 2022. Metal Ions Doped-Carbon Dots: Synthetic Approaches, Physicochemical Properties and Their Applications. Journal of Technical Education Science, (73): 54–63.

Gallareta-Olivares G, Rivas-Sanchez A, Cruz-Cruz A, Hussain SM, González-González RB, Cárdenas-Alcaide MF, Iqbal HMN, and Parra-Saldívar R, 2023. Metal-doped carbon dots as robust nanomaterials for the monitoring and degradation of water pollutants. Chemosphere, 312(October 2022).

Kumar VB, Kumar R, Gedanken A, and Shefi O, 2019. Fluorescent metal-doped carbon dots for neuronal manipulations. Ultrasonics Sonochemistry, 52(November): 205–213.

Li X, Fu Y, Zhao S, Xiao JF, Lan M, Wang B, Zhang K, Song X, and Zeng L, 2022. Metal ions-doped carbon dots: Synthesis, properties, and applications. Chemical Engineering Journal, 430(P4): 133101.

Liu Y, Chao D, Zhou L, Li Y, Deng R, and Zhang H, 2018. Yellow emissive carbon dots with quantum yield up to 68.6% from manganese ions. Carbon, 135: 253–259.

Mansuriya BD, and Altintas Z, 2021. Carbon dots: Classification, properties, synthesis, characterization, and applications in health care-an updated review. Nanomaterials, 11: 2525.

Murugan AV, Kumar Y, Periyasamy L, and Santhakumar M, 2018. Transition Metal Ion (Mn2+, Fe2+, Co2+, and Ni2+)-Doped Carbon Dots Synthesized via Microwave-Assisted Pyrolysis: A Potential Nanoprobe for Magneto-fluorescent Dual-Modality Bioimaging. ACS Biomaterials Science & Engineering, 4: 2582–2596.

Simsek S, Ozge Alas M, Ozbek B, and Genc R, 2019. Evaluation of the physical properties of fluorescent carbon nanodots synthesized using Nerium oleander extracts by microwave-assisted synthesis methods. Journal of Materials Research and Technology, 8(3): 2721–2731.

Vinoth Kumar J, Kavitha G, Arulmozhi R, Arul V, Singaravadivel S, and Abirami N, 2021. Green Sources Derived Carbon Dots for Multifaceted Applications. Journal of Fluorescence, 31(4): 915–932.

Yan F, Sun Z, Zhang H, Sun X, Jiang Y, and Bai Z, 2019. The fluorescence mechanism of carbon dots, and methods for tuning their emission color: a review. Microchimica Acta, 186(8): 583.

Zhao Y, Zhu X, Liu L, Duan Z, Liu Y, Zhang W, Cui J, Rong Y, and Dong C, 2022. One-Step Synthesis of Nitrogen/Fluorine Co-Doped Carbon Dots for Use in Ferric Ions and Ascorbic Acid Detection. Nanomaterials, 12(14): 2377.

DRAFT

ORAL PRESENTATION

Rett sendromunda nöronal hücre farklılaşmasını etkileyebilecek hedef genlerin in silico analizi

Yusuf Çağlar Odabaşı* (https://orcid.org/0000-0003-0690-5575), Özlem Yeşil Çeliktaş (https://orcid.org/0000-0003-4509-2212)

Ege Üniversite, Mühendislik Fakültesi, Biyomühendislik, İzmir, Türkiye

*odabasi98yusuf@gmail.com

Özet

Sinir sistemi gelişiminde nöron hücrelerinin sağlıklı şekilde farklılaşması oldukça önemlidir. Birçok nörolojik hastalığın temelinde de bu süreç yatmaktadır. Rett sendromu çoğunlukla kadınlarda görülen nörogelişimsel bir hastalık olup nadir görülen ve karmaşık bir yapıya sahip olduğu için klinik olarak çalışılması oldukça güçtür. Bu nedenle in silico gibi klinik öncesi çalışmalar oldukça önemlidir. Rett sendromunun nöronal hücre farklılaşmasındaki etkisini incelemek için sağlıklı ve MECP2 mutasyonlu tek hücre rna sekans verisi kullanılmıştır. Gerçekleştirilen transkriptomik analizler ile SOX11 ve SOX4 başta olmak üzere 10 gen belirlenmiştir. Bu genlerin gelecek çalışmalarda çok daha detaylı incelenmesi gerekmektedir.

Anahtar Kelimeler: Rett sendromu, nöronal farklılaşma, hücre gidişatı, ortak ekspresyon modül

In silico analysis of target genes that may affect neuronal cell differentiation in Rett syndrome

Abstract

Healthy differentiation of neuron cells is very important in the development of the nervous system. This process lies at the basis of many neurological diseases. Rett syndrome is a neurodevelopmental disease mostly seen in women, and it is very difficult to study clinically because it is rare and has a complex structure. Therefore, preclinical studies such as in silico are very important. To examine the effect of Rett syndrome on neuronal cell differentiation, single-cell-RNA-seq data from healthy and MECP2 mutations were used. With the transcriptomic analysis, 10 genes, especially SOX11 and SOX4, were identified. These genes need to be examined in more detail in future studies.

Keywords: Rett syndrome, neural differentiation, cell trajectory, co-expression module

GİRİŞ

Nörogelişimsel hastalıklar (NGH), karmaşık moleküler mekanizmaları nedeniyle anlaşılmasını ve tedavi edilmesini zor olan karmaşık bozukluklardır. Özellikle kesin bir tedavisi olmayan NGH'ler için klinik öncesi modeller oluşturarak genetik temellerini incelemek, hastalık mekanizması, hedef genlerin belirlenmesi ve potansiyel ilaç denemeleri için çok önemlidir (Christopher ve ark., 2021) Etkili terapötik hedefleri ortaya çıkarmak için genomik verileri kullanarak gen-hastalık ilişkilerinin analiz edildiği in silico sistem biyolojisi gibi klinik öncesi yaklaşımlar gereklidir (Finlay-Jones ve ark., 2019) X kromozomunda bulunan MECP2 geninde meydana gelen mutasyonların yol açtığı Rett sendromu (RTT) nadir bir NGH'dir ve kadınlarda daha sık görülür (Amir ve ark., 1999; Neul ve ark., 2010). Elektriksel ve kimyasal sinyalleri ileterek nöronlar arası iletişimin sağlandığı bölge olan sinapların RTT durumunda işlev bozukluğu meydana gelmektedir. Bu bozulma, nöronlar arasındaki iletişimin zayıflamasına neden olarak sinir ağlarında bilgi aktarımını etkilemenin yanında hastalarda gözlenen bilişsel ve motor eksikliklerine sebebiyet vermektedir (Boggio ve ark., 2010). Sinapslara ek olarak RTT durumunda, nöronlar üzerinde bulunan ve gelen sinyalleri algılayanodendritlerin gelişiminin ve dallanmasının azaldığı görülmektedir. Dendritlerde meydana gelen gelen bu gelişim bozukluğu sinir sistemini işlevsizleşmesine neden olmaktadır (Nerli ve ark., 2020). RTT'de sinaptik bağlantılar ve dendrit gelişiminde görülen bozulmalar direkt olarak nöronların olgunlaşma ve farklılaşma sürecini etkilemektedir (Kim ve ark., 2011) Sinir sisteminde bulunan nöronal olamayan ancak nöronların işlevini destekleyen ve düzenleyen gila hücrelerinde de RTT durumunda anormallikler gözlemlenmektedir. Glia hücrelerinin nöral

fonksiyon bozukluğu üzerindeki etkileri ve mekanizmaları halen araştırılmaktadır (Kahanovitch ve ark., 2019). Bu çalışmada, birleştirilmiş/kaynaştırılmış (fused) serebral korteks + ganglionik eminens organoidine ait tek hücre rna-sekans verileri kullanılarak RTT'nin nöral hücre farklılaşması üzerindeki etkisi in silico olarak incelenmektedir. Bu kapsamda hücre gidişatı (cell trajectory) ve ortak ekspresyon modül (co-expression module) analizleri gerçekleştirilerek gelecek çalışmalarda kullanılabilecek hedef genler belirlenmesi amaçlanmaktadır.

MATERYAL VE METOT

Veri Kaynağı ve Verinin Hazırlanması

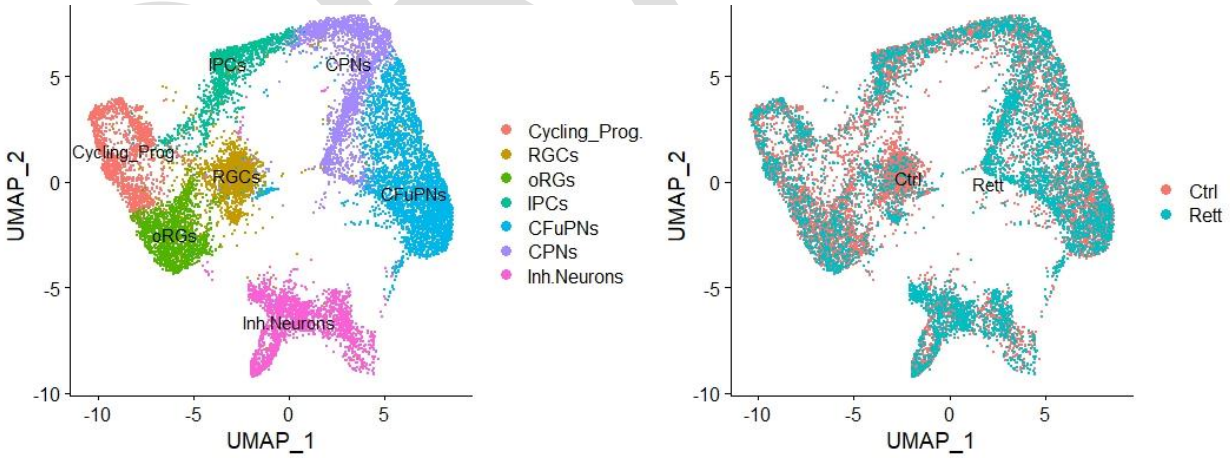
Daha önce yayınlanmış, birleştirilmiş/kaynaştırılmış serebral korteks ve medial ganglionik eminens organoidlerine ait tek hücre rna sekans verisi GSE165577 numarası ile Gene Expression Omnibus veritabanından indirilmiştir (Samarasinghe ve ark., 2021). R paketi 'Seurat' (Seurat v4.3.0.1) kullanılarak, toplam 10 hücre kümesinden oluşan 70. gün verilerden "Cycling Progenitor" (Cyc.Prog.), "Outer Glial Cells" (oRGs), "Radial Glial Cells" (RGCs), "Induced Pluripotent Stem Cells" (IPCs), "Callosal Projection Neurons" (CPNs), "Corticofugal Projection Neurons" (CFuPNs) ve "Inhibitory Interneurons" (Inh.Neurons) hücre kümeleri seçilip alt grup oluşturulmuş. Bu alt grup sağlıklı kontrol (Ctrl) ve 705delG MECP2 mutasyonu (Rett) olmak üzere ikiye ayrılmıştır (Şekil 1).

Hücre Gidişatı Analizi

R paketi 'monocle3' (monocle3 v1.3.4) kullanılarak 'pseudotime cell trajectory' oluşturuldu. Boyutları azaltmak için 'UMAP' yöntemi seçilerek 'cluster_cells' ve 'learn_graph' fonksiyonları. Görselleştirme için 'plot_cells' fonksiyonu kullanıldı. Hücre sıralamaları 'order_cells' fonksiyonu kullanılarak oluşturuldu ve 'ggplot' fonksiyonu ile görselleştirildi. Rett alt grubu için bu naliz yapılmamış olup Ctrl alt kümesi için belirlenen hücre sırlaması referans alınarak Rett alt grubu incelenmiştir.

Ortak Ekspresyon Modül Analizi

R paketi 'hdWGCNA' (hdWGCNA v0.2.23) ile Ctrl ve Rett verileri için ayrı ayrı ortak ekspresyon modülleri belirleyip oluşturulan hücre sıralaması gidişatlarının bu modüllerdeki değişimleri incelenmiştir. Ölçeksiz topoloji modeli uyumunun eşiği $>0{,}80$ olarak ayarlandıktan sonra en iyi bağlantı için yumuşak eşik (soft treshold) Ctrl için 5 Rett için 7 olarak seçilmiştir. Görselleştirmeler için 'plotDendrogram', 'DotPlot' ve 'PlotModuleTrajectory) fonksiyonları kullanılmıştır.

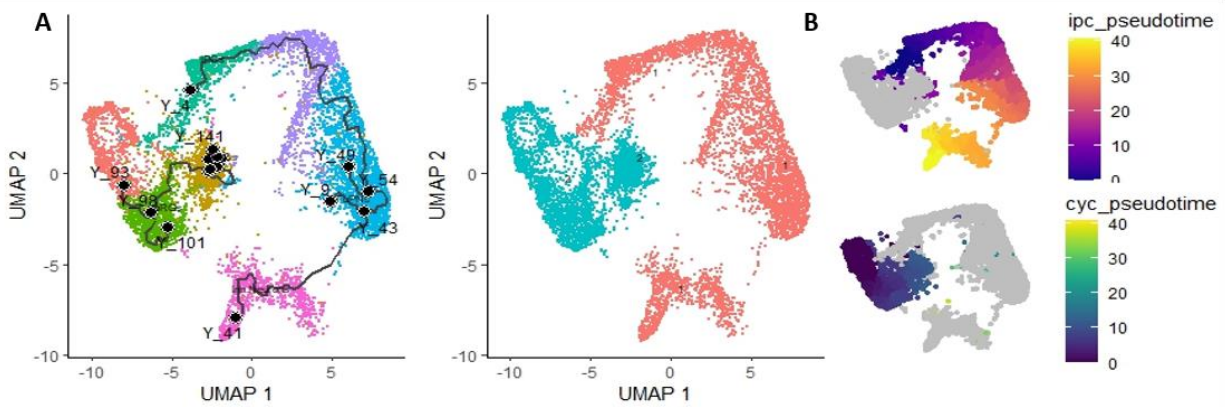


Şekil 1. Hücre kümeleri ve genotipe göre oluşturulan alt grupların UMAP grafikleri.

BULGULAR ve TARTIŞMA

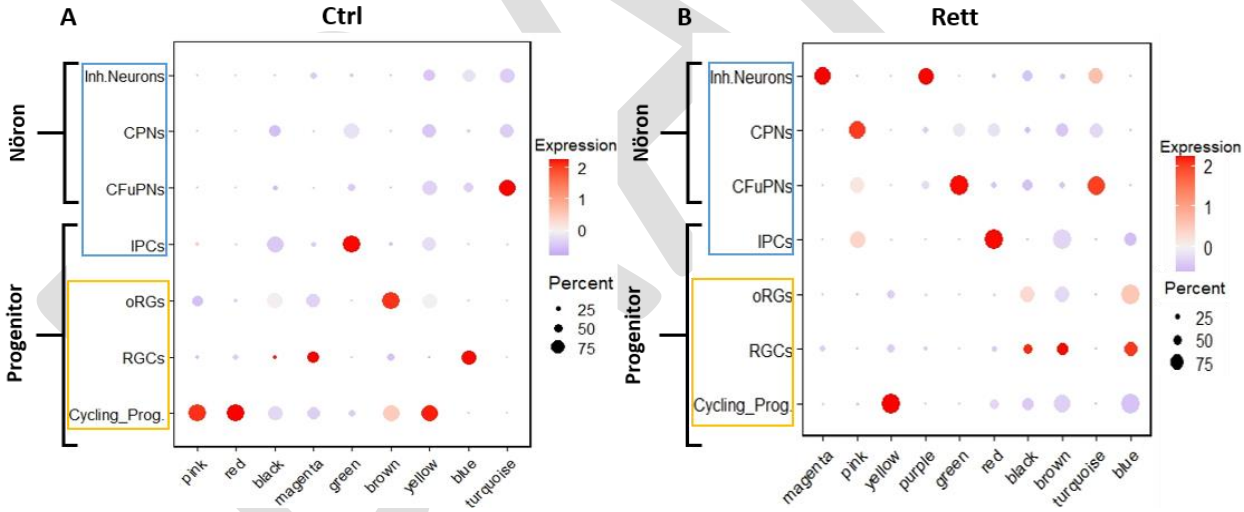
Ctrl ve Rett alt grupları için oluşturulan veriler 7 hücre tipinden oluşmaktadır. oRGs, RGCs, Cyc.Prog. ve IPCs hücre kümeleri 'progenitor' olarak tanımlanırken CPNs, CFuPNs ve Inh.Neurons hücre kümeleri 'nöron' olarak tanımlanmıştır. Rett verisinde nöron grubunun progenitor grubuna oranı Ctrl verisine göre yükseliş göstermektedir. Bu durum verilerin alındığı orijinal çalışmada da gösterilmiştir. Hücre gidişatı analizi sonucunda Ctrl alt grubu için iki hücre yol haritası elde edilmiştir (Şekil 2A). İlk hücre yol haritası (ipc_pseudotime) nöron grubunun yanında IPCs'yi de barındırıyorken ikinci hücre yol haritası (cyc_pseudotime) Cyc.Prog., oRGs ve RGCs hücre kümelerinden oluşmaktadır. İlk yol için IPCs, ikinci yol

için ise Cyc.Prog. başlangıç noktası olarak seçilmiş ve hücre gidişatı oluşturulmuştur (Şekil 2B).Ctrl için oluşturulan bu yol haritaları referans alınarak Rett verileri içinde uygulanmış ve değişimler incelenmiştir.



Şekil 2. A) Hücre gidişatı sonucunda ortaya çıkan yol haritasının UMAP grafikleri üzerindeki gösterimi. **B)** Elde edilen iki yol haritasına başlangıç noktaları belirtilerek gidişat yöneliminin gösterimleri.

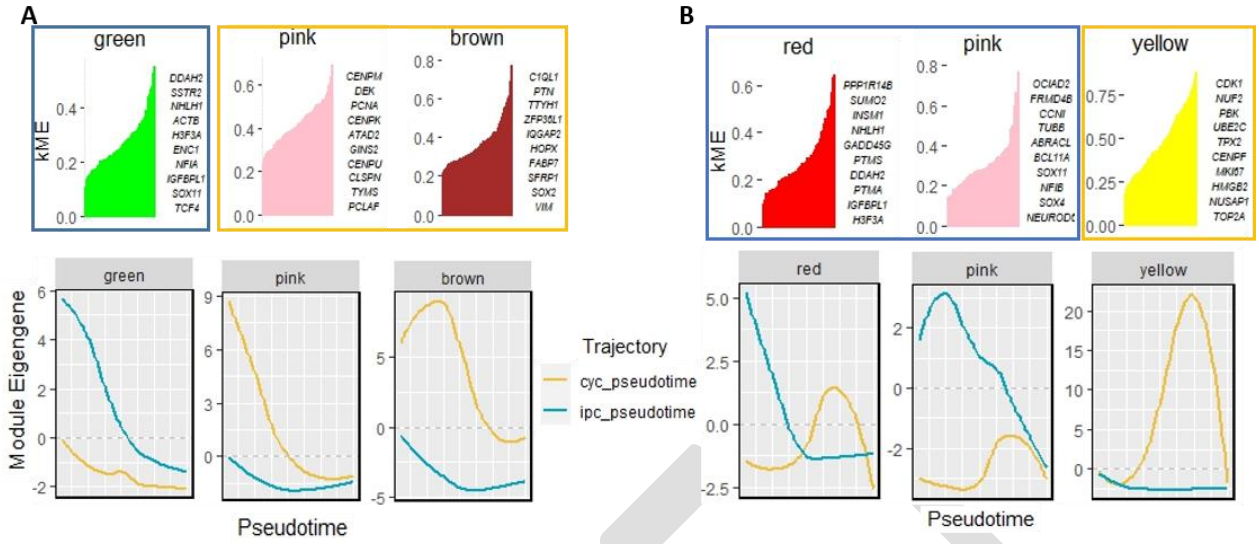
Hücre yol haritaları belirlendikten sonra hücre kümelerine özel genlerin incelenmesi için Ctrl ve Rett alt gruplarına ayrı ayrı ortak ekspresyon analizi gerçekleştirilmiştir. Oluşturulan gen modüllerinin hangi hücre kümelerine ait oldukları ve ekspresyon seviyeleri nokta grafikleri ile gösterilmiştir. Ctrl alt grubunda 'turqoise' modülü harici diğer modüllerin progenitor hücre grubunda ekspresyon gösterdiği görülmektedir. Ayrıca, 'green' modülü ipc_pseudotime'ye özel iken 'pink' ve 'brown' modülleri cyc_pseudotime'ye ait olarak görülmektedir (Şekil 3A). Rett alt grubunda ise Ctrl grubuna göre daha fazla modül nöron grubunda ekspresyon göstermektedir. 'pink' ve 'red' modülleri ipc_pseudotime'ye ait iken 'yellow' modülü cyc_pseudotime'ye ait olduğu görülmüştür (Şekil 3B).



Şekil 3. A) Ctrl alt grubuna ait modül-hücre kümesi ortak ekspresyon nokta grafiği. **B)** Ctrl alt grubuna ait modül-hücre kümesi ortak ekspresyon nokta grafiği. x ekseni modülleri, y ekseni hücre kümelerini ifade etmektedir. Mav kutucuk ipc_pseudotime yol haritasını, turuncu kutucuk cyc_pseudotime yol haritasının göstermektedir.

RTT durumunda, oluşturulan iki hücre yol haritasında meydana gelen değişimleri incelenmek istediğinden bu yol haritalarına ait modüllerin ilk 10 merkez genlerini belirledik ve yol haritalarının bu modüllerdeki eğilimini gösterdik. Ctrl alt grubunda 'green' modülündeki eğilimlere göre Rett alt grubundaki 'red' ve 'pink' modüllerinindeki eğilimler bir takım farklılıklar göstermektedir. Özellikle cyc_pseudotime yolunda bu farklılıklar oluşurken 'pink' modülünde ips_pseudotime yolunda da ufak farklılıklar görülmektedir. Ctrl grubunda 'pink' ve 'brown' modüllerindek eğilimler Rett alt grubunda 'yellow' modülü ile karşılaştırıldığında ipc_pseudotime yolunda bir farklılık görülmezken cyc_pseudotime yolunda eğilim oldukça farklıdır (Şekil 4A, B). Ctrl 'green' modülünün merkez genleri ile Rett 'pink' modüllerinin merkez genleri incelendiğinde SOX11 geninin ortak olduğu görülmektedir. Rett 'pink' modülünde Ctrl 'green' modülünden farklı olarak

OCIAD2, FRMD4B, CCNI, TUBB, ABRACL, BCL11A, NFIB, SOX4 ve NEUROD6 genleri görülmekte. Ctrl 'pink' ve 'brown' modülleri ile Rett 'yellow' modülü merkez genleri karşılaştırıldığında hepsinin farklı olduğu görülmekte.



Şekil 4. A) Ctrl alt grubuna seçilen modüllerin merkez ilk 10 genleri ve oluşturulan iki hücre yol haritasının bu modüllerdeki eğilimleri. **B)** Rett alt grubuna seçilen modüllerin merkez ilk 10 genleri ve oluşturulan iki hücre yol haritasının bu modüllerdeki eğilimleri. Mavi kutucuk ipc_pseudotime, turuncu kutucuk cyc_pseudotime yol haritasına ait modülleri temsil etmekte.

TARTIŞMA

Nörogenez, nöron farklılaşması ve hücre kaderi belirlenmesinde rolü olan SOX11 transkripsiyon faktörünün fare serebral korteks gelişimi sırasında dendritik morfogenezin baskılanmasında da gerekli olduğu gösterilmiştir. Bunun yanında SOX11 ekspresyonununda meydana gelen azalmaların radyal göçün son bulması ile arttığı tespit edilmiştir (Hoshiba ve ark., 2016). SOX11 gibi nöronal gelişimde rol oynayan SOX4'ün glial hücre olan oligodendrositlerin farklılaşmasında etkili olduğu belirtilmiş, bu etkinin temel helix-loop-helix transkripsiyonel baskılayıcısı olan HES5'in SOX4 tarafından indüklenerek nöronal kök hücrelerin oligodendrosite farklılaşması baskılanmaktadır (Bergsland ve ark., 2006; Braccioli ve ark., 2018). Başta sinaptik hedefleme ve nöron motor trafiği olmak üzere nöronal göç ve farklılşmada önemli rol oynayan mikrotübül yapılarından sorumlu tübülin genlerinde RTT durumunda azalış ve mikrotübül yapısında bozulmalar görülmüştür. Ayrıca, tübülin deasetilazı olan HDAC6' da artış ve bu artışa bağlı olarak asetillenmiş alfa-tübülin de azalmalar raporlanmıştır (Gold ve ark., 2015; Sferra ve ark., 2020). Kortikal projection nöronlarda farkılaşmasının incelenmesi için progenitor ve projection hücrelerde eksprese olan BCL11A ile projection hücrelerde eksprese olan BCL11B mutasyonlarına sahip fareler ile yapılan çalışmada BCL11A/B eksikliğinde kortikal nöronlarda proliferasyonda bozulma ve erken farklılaşma tespit edilmiştir (Du ve ark., 2022). Bu sonuçlara ek olarak, OCIAD2, NFIB, ABRCL ve NEUROD6' da nöronal sistem gelişiminde, hücre faklılşması ve proliferasyonunda etkil oldukları çeşitli çalışmalar ile gösterilmiştir (Uittenbogaard ve ark., 2010; Hargus ve ark., 2014; Betancourt ve ark., 2014; Fan ve ark., 2021)

SONUÇ

Bu çalışmada Ctrl verilerine kıyasla Rett verilende meydana gelen değişimler in silico olarak araştırılmıştır. Hücre gidişatı analizi ile belirlediğimiz hücre yol haritalarının, ortak modül analizi ile belirlediğimiz modüller üzerindeki eğilimleri incelenmiş. Bu kapsamda Ctrl için 'green', Rett için 'pink' modülleri seçilmiş olup bu modüllere ait ilk 10 merkez genlerindeki ortak olmayan genler belirlenmiştir (SOX11, OCIAD2, FRMD4B, CCNI, TUBB, ABRACL, BCL11A, NFIB, SOX4 ve NEUROD6). Bu genlerin nöronal farklılaşma ile ilişkileri kısaca verilmiş olup, ileride yapılacak klinik çalışmalar için referans olabileceğini düşünmekteyiz.

KAYNAKLAR

Amir, R. E. *et al.* 1999Rett syndrome is caused by mutations in X-linked MECP2, encoding methyl-CpG-

binding protein 2. *Nat Genet* **23**, 185–188.
Bedogni, F. *et al.* 2014. Rett syndrome and the urge of novel approaches to study MeCP2 functions and mechanisms of action. *Neuroscience & Biobehavioral Reviews* **46**, 187–201.
Bergsland M., Werme M., Malewicz M., Perlmann T., and Muhr J. 2006. The establishment of neuronal properties is controlled by Sox4 and Sox11. Genes & Development, 20:3475–3486.
Betancourt, J., Katzman, S. & Chen, B. 2014. Nuclear factor one B regulates neural stem cell differentiation and axonal projection of corticofugal neurons: NFIB regulates cortical development. *J. Comp. Neurol.* **522**, 6–35.
Boggio. 2010. Synaptic determinants of Rett syndrome. *Front.Syna.Neurosci.* doi:10.3389/fnsyn.2010.00028.
Braccioli, L., Vervoort, S. J., Puma, G., Nijboer, C. H. & Coffer, P. J. 2018. SOX4 inhibits oligodendrocyte differentiation of embryonic neural stem cells in vitro by inducing Hes5 expression. *Stem Cell Research* **33**, 110–119.
Choy, K. H. C. *et al.* 2021. Cognitive behavioral markers of neurodevelopmental trajectories in rodents. *Transl Psychiatry* **11**, 556.
Du, H. *et al.* 2022. Transcription factors *Bcl11a* and *Bcl11b* are required for the production and differentiation of cortical projection neurons. *Cerebral Cortex* **32**, 3611–3632.
Fan, S., Chen, P. & Li, S. 2021. miR-145-5p Inhibits the Proliferation, Migration, and Invasion of Esophageal Carcinoma Cells by Targeting ABRACL. *BioMed Research International* **2021**, 1–10.
Finlay-Jones, A. *et al.* 2019. Very Early Identification and Intervention for Infants at Risk of Neurodevelopmental Disorders: A Transdiagnostic Approach. *Child Dev Perspectives* **13**, 97–103.
Gold, W. A., Lacina, T. A., Cantrill, L. C. & Christodoulou, J. 2015. MeCP2 deficiency is associated with reduced levels of tubulin acetylation and can be restored using HDAC6 inhibitors. *J Mol Med* **93**, 63–72.
Hargus, G. *et al.* 2014. Origin-Dependent Neural Cell Identities in Differentiated Human iPSCs In Vitro and after Transplantation into the Mouse Brain. *Cell Reports* **8**, 1697–1703.
Hoshiba, Y. *et al.* 2016. Sox11 Balances Dendritic Morphogenesis with Neuronal Migration in the Developing Cerebral Cortex. *J. Neurosci.* **36**, 5775–5784.
Jeffrey L. Neul *et al.* 2010. Rett syndrome: Revised diagnostic criteria and nomenclature. *Ann Neurol.* **68**, 944–950.
Kahanovitch, U., Patterson, K. C., Hernandez, R. & Olsen, M. L. 2019. Glial Dysfunction in MeCP2 Deficiency Models: Implications for Rett Syndrome. *IJMS* **20**, 3813.
Kim, K.-Y., Hysolli, E. & Park, I.-H. 2021. Neuronal maturation defect in induced pluripotent stem cells from patients with Rett syndrome. *Proc. Natl. Acad. Sci. U.S.A.* **108**, 14169–14174.
Nerli, E., Roggero, O. M., Baj, G. & Tongiorgi, E. 2020. In vitro modeling of dendritic atrophy in Rett syndrome: determinants for phenotypic drug screening in neurodevelopmental disorders. *Sci Rep* **10**, 2491.
Samarasinghe, R. A. *et al.* 2021. Identification of neural oscillations and epileptiform changes in human brain organoids. *Nat Neurosci* **24**, 1488–1500.
Sferra, A. *et al.* 2020. TUBB Variants Underlying Different Phenotypes Result in Altered Vesicle Trafficking and Microtubule Dynamics. *IJMS* **21**, 1385.
Uittenbogaard, M., Baxter, K. K. & Chiaramello, A. 2010. NeuroD6 genomic signature bridging neuronal differentiation to survival via the molecular chaperone network. *J. Neurosci. Res.* **88**, 33–54.

DRAFT

ORAL PRESENTATION

Aromaterapide kullanılan *Laurus nobilis* L. (defne) bitkisinin uçucu yağ içeriği, toprak yapısı ve bitki elementlerine çevresel koşulların etkisinin araştırılması

H. Furkan ERCAN (*0009-0003-4418-4817*), Ömer ELKIRAN* (*0000-0003-1933-4003*),

*Sinop Üniversitesi, Sağlık Hizmetleri MYO, Tıbbi Hizmetler ve Teknikler Bölümü, Çevre Sağlığı Programı, Sinop, Türkiye
*omer_elkiran@hotmail.com

Özet

Bu çalışmada, aromaterapide uçucu yağları kullanılan ve farklı lokasyonlardan alınan *Laurus nobilis* L. (defne) (Lauraceae) bitkisinin yaprak ve meyvelerinin uçucu yağ içeriklerine, örneklerin alındığı bölgedeki toprak yapısına ve bitki elementlerine çevresel faktörlerin etkisi araştırılmıştır. Çalışma kapsamında Hatay (Samandağ), Hatay (Yayladağ) ve Sinop'tan örnekler alınmıştır. Defne bitkisinin yaprak ve meyve örneklerinin uçucu yağları su distilasyonu yöntemiyle çıkarılarak, kimyasal bileşenler gaz kromatografisi-kütle spektrometresinde (GC-MS) tanımlanmıştır. Çalışma sonuçlarına göre, 1,8-sineol yaprak ve meyve uçucu yağlarında, farklı miktarlarda olmak üzere ana bileşen olarak tanımlanmıştır. Toprak örneklerinin analizinde saturasyon, pH, iletkenlik (Tuz), kireç, yarayışlı potasyum, fosfor ve organik madde değerlerine bakılmıştır. İncelenin parametreler farklı lokasyonlarda değişkenlik göstermişlerdir. Ayrıca yaprak ve meyvelerin içerisindeki kimyasal elementler ve miktarları, indüktif eşleşmiş plazma-kütle spektrometresi (ICP-MS) cihazında tanımlanmıştır. Kimyasal element analiz sonuçlarına göre Mg, P, K, Ca ve Mn en yüksek oranda çıkan elementlerdir. Yaptığımız literatür taramasında, çalışmamızdaki lokasyonların toprak özellikleri ve bu lokasyonlardan alınan bitki örneklerinin farklı organlarının kimyasal içerikleri çalışmalarına rastlanmamıştır. Dolayısıyla çalışmamızda kullandığımız parametreler ilk kez yapılmıştır. *Bu çalışma, ikinci yazarın danışmanlığında birinci yazar tarafından yüksek lisans tezinden üretilmiştir.

Anahtar Kelimeler: *Laurus nobilis*, GC-MS, 1,8-Sineol, Toprak, ICP-MS

Investigation of the effect of environmental conditions on essential oil content, soil structure and plant elements of *Laurus nobilis* L.

Abstract

In this study, the effects of environmental factors on the essential oil content of leaves and fruits of *Laurus nobilis* L. (Lauraceae) plant, the essential oils of which are used in aromatherapy and taken from different locations, the soil structure and plant elements in the area where the samples were taken, were investigated. Within the scope of the study, samples were taken from Hatay (Samandağ), Hatay (Yayladağ), and Sinop. The essential oils of the leaf and fruit samples of the laurel plant were extracted by water distillation method, and the chemical components were identified in gas chromatography-mass spectrometry (GC-MS). According to the results of the study, 1,8-cineol was identified as the main component in leaf and fruit essential oils, with different amounts. In the analysis of soil samples, saturation, pH, conductivity (Salt), lime, useful potassium, phosphorus, and organic matter values were examined. The parameters examined varied in different locations. In addition, chemical elements and their amounts in leaves and fruits were defined in an inductively coupled plasma-mass spectrometer (ICP-MS) device. According to the results of chemical element analysis, Mg, P, K, Ca and Mn are the elements that come out at the highest rate. In our literature review, the soil properties of the locations in our study and the chemical contents of different organs of the plant samples taken from these locations were not found. Therefore, the parameters we used in our study were made for the first time.
*This study was conducted by the first author as a part of a master's thesis under the supervision of the second author.

Keywords: *Laurus nobilis*, GC-MS, 1,8-Cineole, Soil, ICP-MS.

DRAFT

DRAFT

GİRİŞ

Lauraceae, Embriyofitlerin, trakeofitler (damarlı bitkiler) sınıfında bulunur. Trakeofitlerin Euphyllophytes alt dalında olan lauraceae, spermatofit yani tohumla üreyen kısmında bulunması ile dikkat çeker. Aynı zamanda bilimin katkılarıyla oldukça derinlere inilen sınıflandırmada lauracea kapalı tohumlu bir bitki familyasıdır (Qiu ve ark., 2006).

Lauraceae familyasının bir üyesi olan *Laurus* cinsinin; *Laurus nobilis* L. *ve L. canariensis* Willd olmak üzere iki türünün bulunduğunu belirten çalışmalar bulunmaktadır. Bunun yanında *Laurus nobilis* L. dar yapraklı "*angustifolia*" ve kenarları dalgalı "*crispa*", *aurea* ve *undula* olmak üzere 4 alt türünün olduğunu söylemek mümkün hale gelmiştir. Ülkemizde bu dört türden sadece bir türünün bulunduğu yapılan araştırmalar sonucunda görülmektedir (Baydar, 2009).

Ülkemizde Akdeniz Bölgesi'ne özgü bir bitki olarak bilinen defne bitkisinin genel olarak 2 ila 6 metre boylarında, çalı formunda olabildiği gibi ağaç formunda da olduğunu yer yer defne bitki boyunun 10 metreyi bulabildiğini görmekteyiz (Baytop, 1999).

Laurus nobilis yaprağının kalite özelliklerini etkileyen oldukça farklı faktörler bulunmaktadır. Yapılan araştırmalar sonucu: bitki türü, çeşidi (Türk defnesi, Fas defnesi vs.), yetiştiği lokasyonun özellikleri (Karaburun, Bodrum vs.), kültürel uygulama farklılıkları (gübre, ilaç vs. işlemler ile doğal veya kültür ürünü olması), kurutma yöntemleri (kapalı alanda, gölgede veya suni kurutma), depolama ve ambalajlama, temizlik, lezzet, koku (aromatik madde miktarı), renk (doğal koyu renk tercih edilmekte olup renk açıldıkça eterik yağ miktarı azalmaktadır) bu faktörler arasında sayılmaktadır (Göker ve Acar, 1983).

MATERYAL VE METOT

Çalışma Materyallerinin Toplanması

Bitkiler ve toprak örnekleri Sinop merkezden, Hatay (Yayladağı) ve Hatay (Samandağı)'dan doğal ortamlarında Kasım 2021 tarihinde toplanmıştır. Bitkilerin tür tespitleri Flora of Turkey (Davis, 1982) kullanılarak yapılmıştır.

Uçucu Yağların Elde Edilmesi Ve Analizi

Toplanan bitkiler oda sıcaklığında kurutulmuştur. Klevenger aparatı kullanılarak su distilasyonu yöntemiyle, kurutulmuş organlardan ortalama 200g, 3 saat kaynatılarak yaklaşık 0.5ml uçucu yağ elde edilmiştir. (Görsel 1) Elde edilen uçucu yağlar 4°C'de tutulmuştur. Uçucu yağın kompozisyonundaki kalitatif ve kantitatif değişiklik, uçucu yağların GC-MS (Gaz Kromatografisi- Kütle Spektroskopisi) analizleri sonucu belirlenmiştir. Kimyasal bileşenler W9N11.L ve MPW2011.L kütüphaneleri kullanılarak tanımlanmıştır.



Görsel 1: Bitki yaprak ve meyvelerinden uçucu yağ elde etmek için laboratuvar çalışmaları

ICP-MS Ölçümleri

Bitki yaprak ve meyvelerinin element ölçümleri Sinop Üniversitesi Bilimsel ve Teknolojik Araştırmalar Uygulama ve Araştırma Merkezinde yapılmıştır. Analizler için kurutulan örnekler toz haline getirildikten sonra (Görsel 2) ICP-MS cihazıyla ölçümler yapılmıştır.



Görsel 2: ICP-MS çalışmaları için ön hazırlıklar (örneklerin kurutularak toz haline getirilmesi)

Toprak Analizleri

Toprak örnekleri bitki örneklerinin toplanması sırasında, toprak numunesi alma standartları dikkate alınarak paketlenmiş ve analiz için Amasya Merkezi Araştırma laboratuvarına gönderilmiştir (Başaran, 2011). Toprak örnekleri içerisinde; Saturasyon, Ph, İletkenlik (Tuz), Kireç, Yarayışlı Potasyum, Fosfor, Organik Madde ölçümleri yapılmıştır.

BULGULAR ve TARTIŞMA

Toprak Analizleri

Çalışmamız kapsamında Hatay (Yayladağ), Hatay (Samandağ) ve Sinop (Merkez)'tan alınan toprak örneklerinin Saturasyon, Ph, İletkenlik (Tuz), Kireç, Yarayışlı Potasyum, Fosfor ve Organik Madde parametreleri açısından analizleri yapılmış, literatürdeki referans değerlere göre ayrı ayrı yorumlanmıştır (Anonim, 2018).

Hatay (Yayladağ)'dan alınan toprak örneği analiz sonuçları Tablo 1'de verilmiş, referans değerler dikkate alınarak literatürdeki diğer çalışmalarla karşılaştırılıp yorumlanmıştır.

Toprak Parametreleri	Metot	Birim	Deney Sonucu
Saturasyon	Saturasyonda	%	72,6
pH	Saturasyonda (pH metre)	pH	7,41
İletkenlik (Tuz)	Saturasyonda (EC metre)	µs/cm	347
Kireç	Kalsimetre	%	55,85
Yarayışlı Potasyum	Amonyum Asetat/Flame	K_2O kg/da P_2O_5	22,4
Fosfor	Olsen Spektrofotometrik	kg/da	4,07
Organik Madde	Walkey Black	%	1,8

Tablo 1. Hatay (Yayladağ) toprak örneği analiz sonuçları

Toprak pH değeri, pH metre kullanılarak saturasyon işlemi ile 7,41 bulunmuştur. Bu değer, toprağın pH değerine göre hafif alkali toprak grubunda olduğunu göstermektedir (Anonim, 2018). Toprak iletkenlik (tuz) değeri Saturasyonda (EC metre) kullanılarak 347 µs/cm yani 0,347 mS cm-1 bulunmuştur. Bu değer, toprağın iletkenlik (tuz) açısından tuzsuz toprak olduğunu yani iletkenliğinin olmadığını veya zayıf olduğunu göstermektedir. (Anonim, 2018). Toprak kireç oranı kalsimetre kullanılarak %55,85 bulunmuştur. Bu değer, literatürdeki referans değerlere göre toprağın fazla kireçli olduğunu göstermektedir. (Ülgen ve Yurtsever, 1974). Topraktaki yarayışlı potasyum oranı, Amonyum Asetat/Flame metodu kullanılarak 22,4 K2O kg/da P2O5 bulunmuştur. Bu sonuç, topraktaki yarayışlı potasyum miktarı olarak, çok düşük yarayışlı potasyum

oranına sahip olduğunu göstermektedir. (FAO,1990). Topraktaki fosfor oranı Olsen Spektrofotometrik metodu kullanılarak 4,07 kg/da bulunmuştur. Bu değer, toprağın sahip olduğu fosfor miktarı açısından düşük fosfor oranına sahip olduğunu göstermektedir. (Ülgen ve Yurtsever, 1974). Topraktaki organik madde oranı, Walkey Black metodu kullanılarak %1,8 bulunmuştur. Bu değer literatürdeki değerlere göre, toprak örneğinin yüksek organik madde oranına sahip olduğunu göstermektedir (Anonim, 2018)

Hatay (Samandağ)'dan alınan toprak örneği analiz sonuçları Tablo 2'de verilmiş, referans değerler dikkate alınarak literatürdeki diğer çalışmalarla karşılaştırılıp yorumlanmıştır.

Toprak Parametreleri	Metot	Birim	Deney Sonucu
Saturasyon	Saturasyonda	%	77
pH	Saturasyonda (pH metre)	pH	7,45
İletkenlik (Tuz)	Saturasyonda (EC metre)	µs/cm	642
Kireç	Kalsimetre	%	38,8
Yarayışlı Potasyum	Amonyum Asetat/Flame	K_2O kg/da P_2O_5	75,6
Fosfor	Olsen Spektrofotometrik	kg/da	76,02
Organik Madde	Walkey Black	%	2,7

Tablo 2. Hatay (Samandağ) toprak örneği analiz sonuçları

Toprak pH değeri, pH metre kullanılaraksaturasyon işlemi ile 7,45 bulunmuştur. Bu değer, toprağın pH değerine göre hafif alkali toprak grubunda olduğunu göstermektedir (Anonim, 2018; Çepel, 1983). Toprak iletkenlik (tuz) değeri Saturasyonda (EC metre) kullanılarak 642 µs/cm yani 0,642 mS cm-1 bulunmuştur. Bu değer, toprağın iletkenlik (tuz) açısından tuzsuz toprak olduğunu yani iletkenliğinin olmadığını veya zayıf olduğunu göstermektedir (Anonim, 2018). Toprak kireç oranı kalsimetre kullanılarak %38,8 bulunmuştur. Bu değer, literatürdeki referans değerlere göre toprağın çok fazla kireçli olduğunu göstermektedir (Ülgen ve Yurtsever, 1974). Topraktaki yarayışlı potasyum oranı, Amonyum Asetat/Flame metodu kullanılarak 75,6 K2O kg/da P2O5 bulunmuştur. Bu sonuç, topraktaki yarayışlı potasyum miktarı olarak, düşük yarayışlı potasyum oranına sahip olduğunu göstermektedir (FAO,1990). Topraktaki fosfor oranı Olsen Spektrofotometrik metodu kullanılarak 76,02 kg/da bulunmuştur. Bu değer, toprağın sahip olduğu fosfor miktarı açısından yüksek fosfor oranına sahip olduğunu göstermektedir (Ülgen ve Yurtsever, 1974). Topraktaki organik madde oranı, Walkey Black metodu kullanılarak %2,7 bulunmuştur. Bu değer literatürdeki değerlere göre, toprak örneğinin yüksek organik madde oranına sahip olduğunu göstermektedir (Anonim, 2018).

Sinop'tan alınan toprak örneği analiz sonuçları Tablo 3'de verilmiş, referans değerler dikkate alınarak literatürdeki diğer çalışmalarla karşılaştırılıp yorumlanmıştır.

DRAFT

Toprak Parametreleri	Metot	Birim	Deney Sonucu
Saturasyon	Saturasyonda	%	52,8
pH	Saturasyonda (pH metre)	pH	7,42
İletkenlik (Tuz)	Saturasyonda (EC metre)	µs/cm	439
Kireç	Kalsimetre	%	19,4
Yarayışlı Potasyum	Amonyum Asetat/Flame	K_2O kg/da P_2O_5	30,85
Fosfor	Olsen Spektrofotometrik	kg/da	3,69
Organik Madde	Walkey Black	%	1,5

Tablo 3. Sinop toprak örneği analiz sonuçları

Toprak pH değeri, pH metre kullanılarak saturasyon işlemi ile 7,42 bulunmuştur. Bu değer, toprağın pH değerine göre hafif alkali toprak grubunda olduğunu göstermektedir (Anonim, 2018). Toprak iletkenlik (tuz) değeri Saturasyonda (EC metre) kullanılarak 439 µs/cm yani 0,439 mS cm-1 bulunmuştur. Bu değer, toprağın iletkenlik (tuz) açısından tuzsuz toprak olduğunu yani iletkenliğinin olmadığını veya zayıf olduğunu göstermektedir (Anonim, 2018). Toprak kireç oranı kalsimetre kullanılarak %19,4 bulunmuştur. Bu değer, literatürdeki referans değerlere göre toprağın fazla kireçli olduğunu göstermektedir (Ülgen ve Yurtsever, 1974). Topraktaki yarayışlı potasyum oranı, Amonyum Asetat/Flame metodu kullanılarak 30,85 K2O kg/da P2O5 bulunmuştur. Bu sonuç, topraktaki yarayışlı potasyum miktarı olarak, çok düşük yarayışlı potasyum oranına sahip olduğunu göstermektedir (FAO,1990). Topraktaki fosfor oranı Olsen Spektrofotometrik metodu kullanılarak 3,69 kg/da bulunmuştur. Bu değer, toprağın sahip olduğu fosfor miktarı açısından düşük fosfor oranına sahip olduğunu göstermektedir (Ülgen ve Yurtsever, 1974). Topraktaki organik madde oranı, Walkey Black metodu kullanılarak %1,5 bulunmuştur. Bu değer literatürdeki değerlere göre, toprak örneğinin orta düzeyde organik madde oranına sahip olduğunu göstermektedir (Anonim, 2018).

Defne Bitkisinin Meyve Ve Yapraklarının Kimyasal Analizleri

Bölgelere göre defnenin yaprak ve meyvesinde en fazla bulunan elementlere sırasıyla bakacak olursak;
Mg miktarına göre yapraklar:
Hatay Yayladağı Yaprağında 3107909 ppb,
Hatay Samandağ'ı Yaprağında 2556563 ppb,
Sinop Yaprağında 1771661 ppb miktarında Mg bulunur.
Mg miktarına meyveler:
1. Hatay Samandağ'ı Meyvesinde 949237 ppb
2. Hatay Yayladağı Meyvesinde 748325 ppb
3. Sinop Meyvesinde 568928 ppb

P miktarına göre yapraklar:
Hatay Samandağ'ı yaprağında 1863096 ppb
Sinop yaprağında 1760360 ppb
Hatay Yayladağı yaprağında 1695099 ppb
P miktarına göre meyveler:
Hatay Samandağ'ı Meyvesinde 2332026 ppb
Hatay Yayladağı Meyvesinde 1813188 ppb
Sinop Meyvesinde 1801037 ppb
K miktarına göre yapraklar:
Hatay Yayladağı yaprağında 12970883 ppb
Sinop yaprağında 10985434 ppb
Hatay Samandağ'ı yaprağında 8892027 ppb

K miktarına göre meyveler
Hatay Samandağ'ı Meyvesinde 11641157 ppb
Sinop Meyvesinde 9770916 ppb
Hatay Yayladağı Meyvesinde 8998593 ppb
Ca miktarına göre yapraklar
Hatay Samandağ'ı yaprağında 15097444 ppb
Sinop yaprağında 12892055 ppb
Hatay Yayladağı yaprağında 11492966 ppb
Ca miktarına göre meyveler
Hatay Samandağ'ı Meyvesinde 1496656 ppb
Hatay Yayladağı Meyvesinde 1198474 ppb
Sinop Meyvesinde 731640 ppb
Mn miktarına göre yapraklar
Sinop yaprağında 198658 ppb
Hatay Samandağ'ı yaprağında 56758 ppb
Hatay Yayladağı yaprağında 11669 ppb
Mn miktarına göre meyveler
Sinop Meyvesinde 16309 ppb
Hatay Samandağ'ı Meyvesinde 11562 ppb
Hatay Yayladağı Meyvesinde 7600 ppb
Literatür taramamızda çalıştığımız lokasyonlara ait defne bitki elementleri konusunda çalışmaya rastlanılmamıştır. Diğer bölgelerden yapılan çalışma sonuçları, bulduğumuz çalışma sonuçları ile karşılaştırılmıştır. Dolayısıyla tezimizde yer alan bölgeler için Defne organlarının kimyasal içerik çalışması ilk kez yapılmıştır. Yaşar ve ark., (2012) Bartın'dan üç farklı lokasyondan aldıkları defne yapraklarının analiz sonuçlarında Mg elementini ortalama 21,4 olarak, K elementini ortalama 14,9 olarak ve Ca elementini ortalama 9,9 olarak bulmuşlardır (Yaşar ve ark., 2012).

Defne Bitkisi Meyve Ve Yapraklarının Uçucu Yağ Analizleri

Hatay (Yayladağ) yaprak uçucu yağ analizinde 11 kimyasal bileşen (% ≥ 1) tespit edilmiştir. 1,8-Sineole (66.4%), Sabinene (9.4%), 3-Cyclohexen-1-ol (4.2%) ve α-Pinene (3.6%) ana bileşenler olarak bulunmuştur. Hatay (Yayladağ) meyve uçucu yağ analizinde 11 kimyasal bileşen (% ≥ 1) tespit edilmiştir. 1,8-Sineole (45%), α-Pinene (9.9%) Sabinene (8.4%), 1-Phellandrene (6.9%) ve β- Elemene (3.8%) ana bileşenler olarak bulunmuştur.

Hatay (Samandağ) alınan yaprak uçucu yağ analizimizde 10 kimyasal bileşen (% ≥ 1) tespit edilmiştir. 1,8-Sineole (61.5%), Sabinene (10.6%), Camphene (4.6%) 3-Cyclohexen-1-ol (4.%) ve α-Terpineol (3.5%) ana bileşenler olarak bulunmuştur. Hatay (Samandağ) meyve uçucu yağ analizinde 11 kimyasal bileşen (% ≥ 1) tespit edilmiştir. 1,8-Sineole (40.9%), β-Elemene (13.7%), α-Pinene (9.8%) Sabinene (9.3%) ve1-Phellandrene (8%) ana bileşenler olarak bulunmuştur.

Sinop'tan alınan yaprak uçucu yağ analizinde 11 kimyasal bileşen (% ≥ 1) tespit edilmiştir. 1,8-Sineole (57.3%), Sabinene (11%), Camphene (5.4%) α-Pinene(4.2%) 3-Cyclohexen-1-ol (3.6%) ana bileşenler olarak bulunmuştur. Sinop meyve uçucu yağ analizinde 15 kimyasal bileşen (% ≥ 1) tespit edilmiştir. 1,8-Sineole (27.6%),1-Phellandrene (10.1%), 1,3,6-Octatriene (8.5%), Sabinene (7.3%) ve β-Elemene (6.8%), ana bileşenler olarak bulunmuştur.

Çalışma sonuçlarımıza göre defne bitkisinin tıbbi ve ekonomik değerini gösteren, çalışılan organların hepsinde ana bileşen olarak çıkan 1,8-Sineole (66.4%) en yüksek oranda Hatay (Yayladağ)'dan alınan defnenin yapraklarında, en düşük oranda 1,8-Sineole (27.6%) Sinop'tan alınan örneklerin meyvesinde çıkmıştır. Ana bileşenler genel olarak benzerlik göstermektedir. Hatay (Yayladağ)'dan alınan örnekte ana bileşen olan 1,8-Sineole, coğrafik şartlara bağlı olarak en yüksek oranda bulunmuştur diyebiliriz.

Kılıç ve ark., (2004) tarafından yapılan çalışmada Karadeniz bölgesinden toplanan defne bitkisinin yaprak ve meyve uçucu yağlarına bakılmıştır (Kılıç ve ark., 2004). Yapraklarda 1,8-sineole, α-terpinyl acetate, sabinene, α-pinene, meyvelerde (E)-β-ocimene, biclyclogermacrene, ana bileşenlerdir. Bizim yaprak sonuçlarımızla benzerlik gösterirken, meyve sonuçlarımızla farklılık bulunmaktadır. Karadeniz (2001), tarafından Hatay'dan alınan defne yaprak ve meyveleriyle yapılan uçucu yağ çalışmasında yapraklarda bizim çalışmamızda olduğu gibi 1,8-sineole ana bileşen, meyve uçucu yağ çalışmasında ana bileşen α-pinen'dir. α-pinen bizim çalışma

sonuçlarımızda da ana bileşenler arasındadır (Karadeniz, 2001). Karık ve ark., (2015) tarafından Türkiye'de yayılış gösteren defne bitkileriye yaptıkları yaprak uçucu yağ çalışmasında 1,8-sineol bizim çalışmamızda olduğu gibi ana bileşen olarak tespit edilmiştir (Karık ve ark., 2015).

Literatürde Türkiye'de yayılış gösteren defne bitkilerine ait uçucu yağ çalışmalarında genel olarak, bizim çalışmamızda olduğu gibi 1,8-sineole ana bileşen olarak tespit edilmiştir. Diğer ana bileşenlerdeki ve 1,8-sineole miktarındaki farklılık bitkinin toplanma yerinin coğrafik özellikleri, toplanma zamanı ve kullanılan yöntemlerinden kaynaklanmaktadır.

SONUÇ

Yapılan çalışmada konumuz olan defne bitkisi, ülkemiz için tıbbi ve ekonomik değeri yüksek ve insanlar tarafından gıda, şifa amaçlı kullanılan bir bitkidir. Çalışmamızda, farklı ortamlardan alınan bitki örneklerinde uçucu yağ miktarları ve kimyasal bileşenlerin farklı olduğu görülmüştür. Bu sonuçlar, defne konusunda yapılan ve farklı parametrelerin kullanıldığı çalışmaların yeni sonuçlar ortaya çıkarabileceğini göstermektedir. Çalışma sonuçlarımız, bitkinin farklı organlarının uçucu yağ ve kimyasal içerik bakımından farklı olabileceğini göstermekte, coğrafik şartların bitkinin verimini değiştirebileceği konusundaki çalışmaları doğrulamaktadır. Bitkinin yetiştiği ortamların toprak özelliklerine ait verilerimiz, literatürde bu konuda sınırlı çalışmalardan dolayı, yeni çalışmalara kaynak oluşturacaktır. Toprak konusunda yeni çalışmalar yapılarak bitkinin farklı özelliklerinin (Ör: Fitoremediasyon) tespiti gerekmektedir. Elde ettiğimiz verilerin, ülkemizde doğal olarak yayılış gösteren bu bitkiye ait yeni çalışmalara veri olacağını ve bilim dünyasına katkı sağlayacağını düşünmekteyiz.

KAYNAKLAR

Anonim 2018. www.naturalresources.sa.gov.au/.../140916-standard-tests-a... [10.01.2023]

Baydar H 2009. Tıbbi ve Aromatik Bitkiler Bilimi ve Teknolojisi. Süleyman Demirel Üniversitesi Ziraat Fakültesi Yayınları No. 51: 234-235. Isparta.

Baytop T 1999. Türkiye'de bitkiler ile tedavi: geçmişte ve bugün. Nobel Tıp Kitabevleri.

Davis PH 1982. Flora of Turkey, Vol. 7, Edinburg Universty Pres, Edinburg, pp. 947.

FAO, 1990. Micronutrient. Assessment at the country level: an international study. FAO soil bulletin by Mikko Sillanpaa. Rome.

Göker Y, Acar İ 1983. Orman Yan Ürünlerinden Akdeniz Defnesi. Journal of the Faculty of Forestry Istanbul University, 33(1): 124-140.

Karadeniz H 2001. Hatay bölgesi defne yaprağı ve meyvası uçucu yağının özelliklerinin belirlenmesi (Master's thesis, Fen Bilimleri Enstitüsü).

Karık Ü, Çiçek F, Erdinç O, Tutar M, Fırat A 2015. Türkiye defne (*Laurus nobilis* L.) populasyonlarının uçucu yağ bileşenleri. Anadolu Ege Tarımsal Araştırma Enstitüsü Dergisi, 25:1-16.

Kılıç A, Hafizoglu H, Kollmannsberger H, Nitz S 2004. Volatile constituents and key odorants in leaves, buds, flowers, and fruits of *Laurus nobilis* L. Journal of agricultural and food chemistry, 52: 1601-1606.

Ülgen N, Yurtsever N 1974. Türkiye gübreler ve gübreleme rehberi. Toprak ve Gübre Araştırma Enstitüsü Müdürlüğü, Teknik Yayınlar, (28).

Qiu YL, Li L, Wang B, Chen Z, Knoop V, Groth-Malonek M, ... Davis C C 2006. The deepest divrgences in land plants inferred from phylogenomic evidence. Proceedings of the National Academy of Sciences, 103(42): 15511-15516.

Yaşar Ü., Özyiğit İİ, Yalçın İE, Doğan İ, Demir G 2012. Determination of some heavy metals and mineral nutrients of bay tree (*Laurus nobilis* L.) in Bartin city, Turkey. Pakistan Journal of Botany, 44: 81-89.

ORAL PRESENTATION

Antalya ili nar üreticilerinin bitki koruma uygulamaları

Mustafa Kemal Özen[1] (*ORCID: https://orcid.org/0009-0008-5296-609X*), Gizem Berber Tortop[2*] (*ORCID: https://orcid.org/0000-0003-3090-3705*), Sibel Yorulmaz[1] *(ORCID: https://orcid.org/0000-0003-3836-5673)*

[1]Isparta Uygulamalı Bilimler Üniversitesi, Ziraat Fakültesi, Bitki Koruma Bölümü, Isparta, Türkiye
[*2]Bilecik Şeyh Edebali Üniversitesi, Ziraat ve Doğa Bilimleri Fakültesi, Bitki Koruma Bölümü, Bilecik, Türkiye

*Sorumlu yazar e-mail: gizem.berber@bilecik.edu.tr

Özet

Nar (*Punica granatum*), Lythraceae familyasından, içinde küçük çekirdekleri ve tanecikleri bulunan, hafif ekşi tadı olan, ılıman iklimlerde yetişen bir meyve türüdür. Nar üretiminde önde gelen ülkeler İran, Hindistan, Çin, Türkiye, ABD, Irak, İspanya, Suriye, Azerbaycan, Afganistan, Mısır, Özbekistan ve Pakistan'dır. Türkiye'de 2002 yılında 3 milyon nar ağacı ve 50-60 bin ton nar üretimi varken olumlu piyasalar ve devlet desteği ile 2010 yılına gelindiğinde 13 milyon ağaç ve 315 bin ton üretime ulaşmıştır. Ülkemizde bölge olarak Akdeniz, Ege ve Güneydoğu Anadolu bölgeleri, il olarak ise Antalya, Muğla, Mersin, Adana nar yetiştiriciliğinde ön plana çıkmaktadır. Tarımsal ürünlerde girdi kullanım miktarlarının, maliyet ve gelirlerin belirlenmesi mikro düzeyde üreticiler ve ekonomi politikasını yürütenler açısından büyük önem taşımaktadır. Bu amaçla Antalya ilinde en fazla nar üretimi yapılan Döşemealtı, Serik ve Aksu ilçelerindeki nar üreticileri nar üretiminde karşılaşılan hastalık ve zararlılar ile mücadele yöntemleri hakkında anket çalışması yapılmıştır. Anket sonuçlarına göre o bölgede en sık rastlanılan zararlılar nar yaprakbiti (*Aphis punicae)*, nar beyazsineği (*Siphoninus phillyreae*), turunçgil unlubiti (*Planococcus citri*) iken en çok rastlanılan hastalıklar ise kahverengi leke (*Alternaria alternata*), kök ve kök boğazı çürüklükleri (*Phytophthora spp.*) olarak ifade edilmiştir.

Anahtar Kelimeler: Nar, Üretim, Bitki Koruma

Plant protection practices of pomegranate producers in Antalya province

Abstract

Pomegranate (*Punica granatum*) is a type of fruit from the Lythraceae family, with small seeds and grains, a slightly sour taste, and grows in temperate climates. The leading countries in pomegranate production are Iran, India, China, Turkey, USA, Iraq, Spain, Syria, Azerbaijan, Afghanistan, Egypt, Uzbekistan and Pakistan. While there were 3 million pomegranate trees and 50-60 thousand tons of pomegranate production in Turkey in 2002, with positive markets and government support, 13 million trees and 315 thousand tons of production were reached in 2010. In our country, the Mediterranean, Aegean and Southeastern Anatolia regions, and Antalya, Muğla, Mersin, Adana come to the forefront in pomegranate cultivation. Determining the amount of input use, costs and incomes in agricultural products is of great importance for producers and economic policy makers at the micro level. For this purpose, pomegranate producers in Döşemealtı, Serik and Aksu districts, where the most pomegranate is produced in Antalya province, were surveyed about the diseases and pests encountered in pomegranate production and control methods. According to the survey results, the most common pests in that region are pomegranate aphid (*Aphis punicae*), pomegranate whitefly (*Siphoninus phillyreae*), citrus mealybug (*Planococcus citri*), while the most common diseases are brown spot (*Alternaria alternata*), root and root collar rots (Phytophthora spp.).

Keywords: pomegranate, production, plant protection

GİRİŞ

Nar (*Punica granatum* L.), Lythraceae familyasının Punica cinsinden çok yıllık bir bitki olup ticari değeri kadar kültürel olarak da önemli bir meyvedir. Bu meyvenin ticari türü olan *Punica granatum* L. Ortaçağ'da çekirdekli elma anlamına gelen "*Pomuni granatum*"dan adını almıştır. Bir Fenike kolonisi olan Kartacalılar Akdeniz havzasında nar ticaretini başlattıkları için eski kaynaklarda "Kartaca (Fenike) Elması" (The apple of Carthage / Carthaginianapple) olarak da yer almaktadır. Günümüzde A.B.D.'de çekirdekli elma (Seedy apple) olarak da bilinmektedir (Kurt ve Şahin 2013).

Çalı formunda olan nar bitkisinin boyları 2 ila 5 metre arasındaki değişkenlik göstermektedir. Meyveleri çok taneli ve etli tohumlardan oluşan, kırmızıdan beyaza kadar değişik tonlarda renklere sahiptir. Özellikle potasyum ve karbonhidrat açısından oldukça zengin bir Akdeniz meyvesidir. Genellikle taze ve meyve suyu olarak tüketilen, çeşitli ve değerli maddelerin elde edildiği bir bitkidir. Son yıllarda yetiştirme tekniği, gıda teknolojisi, depolama ve taşıma alanlarında görülen gelişmeler sonucu daha çok tanınan, yetiştiriciliğine ilgi duyulan bir meyve haline gelmiştir (Özgüven ve ark., 2015). Ayrıca günümüzde AIDS için kullanılan yiyecekler sınıfına alınmış ve Japon patentli ilaçlarda yer alan 9 bitkiden biri olmuştur. Narın içerdiği flavanoidlerin güçlü bir antioksidan olduğu belirlenmiştir. Yine, nar meyve suyu ve yağının ömrü uzattığı, kalp hastalıkları ile kanseri önlediği bildirilmiştir (Lansky ve ark., 1998).

Son yıllarda, Türkiye'de nar yetiştiriciliğinin artmasında, insan sağlığına olan faydalarının anlaşılması ve ekonomik olarak da değerinin artması gibi nedenler yer almaktadır. Ülkemizde uzun yıllardan beri bahçe kenarlarında, çit bitkisi ve süs bitkisi olarak yetiştirilen nar bitkisi, son zamanlarda kapama bahçeler şeklinde yetiştirilmeye başlanmıştır. Nar bitkisi, çok çeşitli iklim ve toprak koşullarına kolayca adapte olabilmesi, çoğaltılmasının kolay olması, birim alanda yüksek verim vermesi ve erken meyveye yatması gibi avantajlara sahiptir. Ayrıca, kuru hava şartlarına, yarı çöl ve hatta çöl iklimine antepfıstığı ile birlikte en dayanıklı meyve türü olarak bilinmektedir (Dursun ve ark., 2019).

Nar yetiştiriciliği, yaygın olarak birçok ülkede yapılmasına rağmen dünyada büyük bir potansiyele sahip değildir. Türkiye, narın anavatanı olarak kabul edilen yetiştirici ülkeler arasında ilk sıralarda yer almaktadır. Türkiye'de yetiştiriciliği yapılan diğer meyve çeşitlerinde olduğu gibi, narlarda da yetiştiricilik sorunlarının yanında birçok bitki koruma sorunları ile karşılaşılmaktadır. Bu sorunların başında da, zararlılarla mücadele gelmektedir. Gerek yurtiçinde gerekse yurtdışında nar zararlıları ile doğal düşman türlerinin belirlenmesi amacıyla az sayıda faunistik çalışma bulunmaktadır. Türkiye'de sadece Güneydoğu Anadolu Bölgesi ile Antalya İlinde olmak üzere iki faunistik çalışma yapılmış, yurtdışında ise İspanya ve Hindistan'da benzer konuda çalışmalar gerçekleştirilmiştir (Öztürk ve ark., 2005).

Dünya'da ve Türkiye'de özellikle 2000'li yıllardan sonra nar üretiminde önemli artışlar gözlenmiştir (Kurt ve Şahin 2013). Nar, Türkiye'de tüm bölgelerde yetişen bir meyve olmasının yanında özellikle Ege ve Akdeniz bölgelerinde ve Güneydoğu Anadolu bölgesinde yetiştirilmektedir (Gölükcü ve ark., 2008). Ülkemiz nar üretiminde ilk beş sırada yer alan iller sırasıyla Antalya (107.237 ton), Mersin (45.594 ton), Denizli (39.715 ton), Adana (20.769 ton) ve Hatay (20.769 ton)'dır. Bölgelere göre nar üretim miktarı incelendiğinde 234.609 ton ile Akdeniz Bölgesi birinci, 146.080 ton ile Ege Bölgesi ikinci ve 51.790 ton ile Güneydoğu Anadolu Bölgesi üçüncü sırada yer almaktadır Güneydoğu Anadolu Bölgesinin toplam nar üretim payı içeresinde ki değeri %11.62'dir. Güneydoğu Anadolu bölgesinde nar üretimi bakımından Gaziantep birinci, Kilis ikinci, Şanlıurfa üçüncü, Adıyaman dördüncü ve Siirt beşinci sırada yer alırken Batman ili son sırada yer almaktadır (Şimşek ve Gülsoy 2017).

Tarımsal ürünlerde girdi kullanım miktarlarının, maliyet ve gelirlerin belirlenmesi mikro düzeyde üreticiler ve ekonomi politikasını yürütenler açısından büyük önem taşımaktadır. Tarımsal ürün maliyetleri ile ilgili araştırmaların sonuçları, hükümetlerin fiyat politikalarını saptamalarında başvurabilecekleri önemli bir araç olmaktadır. Ayrıca tarımsal ürün maliyetleri, işletmelerde özellikle fiziki üretim girdilerinin kullanım düzeylerinin belirlenmesi, işgücü planlaması, finansman programlarının yapılması, ürün bütçelerinin ve yatırım projelerinin hazırlanması vb. planlama faaliyetlerinde yaygın olarak kullanılmaktadır (Özalp ve Yılmaz, 2013). Bu sebeple üreticilerle yapılan anket çalışmaları oldukça önem kazanmaktadır.

MATERYAL VE METOT

Bu çalışma da 2020-2021 yılında Antalya ilinde en fazla nar üretimi yapılan Döşemaltı, Serik ve Aksu ilçelerindeki nar üreticileri ile görüşülerek yapılmıştır.

Çalışma kapsamında üretim büyüklükleri dikkate alınarak Döşemealtı'nda 25, Serik'te 15 ve Aksu'da 10 üretici olacak şekilde toplamda 50 nar üreticisi ile görüşülmüştür. Üreticilere 25 sorudan oluşan görüşme formları sunulmuştur. Anket sonucunda elde edilen bilgiler değerlendirilmiştir.

1. Kaç dekar alanda nar üretimi yapıyorsunuz?

%6'sı 1-10 dekar .

%24'ü 10-25 dekar

%8'i 25-40 dekar

%40'ı 40-50 dekar

%22'si 50+ dekar

2. Nar üretiminde en fazla karşılaştığınız hastalık ve böcekler nelerdir?

Hastalık

%76'sı Kahverengi Leke Hastalığı (*Alternaria alternata*)

%20'si Kök ve Kök Boğazı Çürüklüğü (Phytophthora spp.)

%4'ü Diğer

Böcekler

%40'ı Nar Yaprak Biti (*Aphis punicae*)

%12'si Nar Beyaz Sineği (*Siphoninus phillyreae*)

% 38'i Turunçgil Unlubiti (*Planococcus citri*)

%10'u Akdeniz Meyve Sineği (*Ceratitis capitata*)

3.Herhangi bir hastalık ya da zararlı ile karşılaşmak düşüncesiyle bitkileri kontrol etmek için düzenli aralıklarla ilaç atar mısınız?

%60'ı Evet

%20'si Bazen

%20'si Hayır

4.Bir hastalığı ya da zararlıyı görür görmez ilaç atar mısınız?

%92'si Evet

%8'i Hayır

Hayır ise atmamanızın nedeni nedir?

%0 Masraflı olduğu için

%50'si Hastalık ya da zararlı az olduğu için

%50'si Hastalık ya da zararlı önemsiz olduğu için

%0'ı Diğer (Belirtiniz)

5.Kullandığınız tarım ilaçlarını nereden temin ediyorsunuz?

%82'si Zirai İlaç Bayii

%6'sı Tarım Kredi Kooperatifi

%10'u Ziraat Odası

%2'si Diğer

6.Kullandığınız ilaçları seçerken nelere dikkat edersiniz?

%28'i Kullanacağım ürün için ruhsatlı olmasına

DRAFT

%34'ü Etkili Olmasına

%10'uKarışabilir olmasına

%8'i Ekonomik olmasına

%20'si Tanınmış bir ilaç olmasına

%0'ı Diğer (Hasat süresinin kısa olmasına)

7.Bitki koruma ürünleri uygulamaya yönelik kurs ve eğitim aldınız mı?

%60'ı Evet

%40'ı Hayır

8.Sezon boyunca kaç ilaçlama yapıyorsunuz?

%0'ı 1-3ilaçlama

%10'u 4-7 ilaçlama

%84'ü 8-15 ilaçlama

%6'sı 15>ilaçlama

9.İlaçlamayı ne zaman yapıyorsunuz?

%26'sı Sabah

%2'si Öğle

%64'ü Akşam

%8'i Zaman Gözetmeksizin

10.İlaç uygulamasını nasıl yapıyorsunuz?

%0'ı Yağmurlama sulama ile

%0'ı Damlama sulama ile

%100'ü İlaçlama makinesi ile

%0'ı Diğer

11.İlaçlama dozunu ayarlamada nelere dikkat edersiniz?

%18'i Etikete göre uygulama yaparım

%44'ü Bayinin önerisine göre ayarlarım

%12'u Etiketten biraz fazla uygularım

%18'i Kendi tecrübeme göre

%8'i Diğer üreticilerin yaptıklarını uygularım

12.Tarım ilacını hazırlarken suyun pH"sına bakıyor musunuz?

%98'i Hayır

%2'si Evet

13.Kimyasal mücadelede birden fazla ilacı karıştırarak kullanıyor musunuz?

%2'si Hayır

%98'i Evet

Evet ise nedeni;

%82'si Bir ilaçlamada birden fazla hastalık ve zararlıyı yok etmek için

%2'si Birden fazla ilaç kullanarak tek bir hastalık ve zararlıyı yok etmek için

DRAFT

%16'sı İlaçlama maliyetini azaltmak için

14.Size göre narda mücadelesi en zor zararlı nedir?

%38'iTurunçgil Unlu Biti (*Planococcus citri*)

%44'ü Nar Yaprak Biti (*Aphis punicae*)

%12'si Nar Beyazsineği (*Siphoninus phillyreae*)

%6'sı Harnup Güvesi (*Ectomyelois ceratoniae*)

15. Kullandığınız ilaç size göre etkili oluyor mu?

%56'sı Evet

%28'i Hayır

%16'sı Bazen

16.Tarım ilaçlarını hazırlarken hangi tedbirleri alıyorsunuz?

%12'si İlaç ambalajındaki gerekli bilgileri okuyorum

%4'üİlaçların hazırlanmasında eldiven, gözlük kullanıyorum

%4'üSüspansiyon ve emilsiyon ilaçların çok iyi karışmasına

%26'üSigara içmemeye, yemek yememeye uyuyorum

%54'ü Hiçbir önlem almıyorum

17. Tarım ilaçlarını atarken hangi tedbirleri alıyorsunuz?

%50'ıYemek yememeye, sigara içmemeye uyuyorum

%2'si Koruyucu elbise kullanıyorum

%48'i Hiçbir önlem almıyorum

%0'ı Diğer (Belirtiniz)

18.Artan ilaçları ve ilaçlama tankının temizlenmesi sırasında oluşan ilaçlı suyu nereye boşaltıyorsunuz?

%18'i Bahçenin bir kenarına döküyorum

%24'ü Boş bir araziye püskürtüyorum

%0'ı Sulama kanalına veya akarsuya boşaltıyorum

%58'i Başka ağaç ve sebzelerde kullanıyorum

19.İlaçlamalardan önce ve sonra ilaçlama alet ve ekipmanları hakkında aşağıdakilerden hangisini yapıyorsunuz?

%40'ı Kalibrasyon ayarlarını yapmak

%60'ı Her uygulamadan sonra ilaç deposu temizliği

%0'ı Koruyucu giysi ve ekipmanları ilaçlardan ayrı yere koymak

20. Kullanmış olduğunuz tarım ilaçlarını nerede depoluyorsunuz?

%8'i Evin herhangi bir yerinde

%44'ü Özel bir dolapta

%28'i Özel bir odada rafta

%0'ı Ahır veya hayvan barınağında

%20'si Ayrı bir binada ya da katta

21.İlaçlı mücadele çevre kirliliğine yol açıyor mu?

DRAFT

%0'ı İlaçların çevreyi kirlettiği doğru değil

%38'ı İlaçlar çevre kirliliğine neden oluyor, fakat abartıyorlar

%62'si İlaçlı mücadele ile çevreyi kirletiyoruz, ama mücadele gerekli

22.İlaçlar çevreyi nasıl kirletiyor?

%60'ı İlaçlar gereğinden fazla atılarak

%20'si Kullanılması sakıncalı ilaçlar gereksiz yerlerde kullanılarak

%10'u İlaçlama esnasında hedefin dışına taşarak

%10'u İlaç artıklarını ortada bırakarak

23.Kimyasal mücadelede kalıntı sorunu sizce önemli mi?

%20'si İlaçlamada önemli olan hastalığı ortadan kaldırmak

%20'si Önemli, fakat ilacın yararı daha önemlidir

%20'si Çok önemli, ilacın yararından daha önemli

%40'ı İlaç atarken özellikle kalıntısının oluşturacağı sorunları dikkate alırım

24.Boşalan ilaç kutularına ne yapıyorsunuz?

%50'si Tarlanın bir kenarına atıyorum

%0'ı Sulama kanalı veya akarsuya atıyorum

%20'si Genel çöp kutusuna atıyorum

%30'u Bir yerde depolayıp, sonra yakarak imha ediyorum

25.İlaçlama ile hasat arasında olması gereken bekleme süresine dikkat ediyor musunuz?

%36'sı Mahsulun olgunlaşmasına göre hasat ederim

%14'ü İlaçlamadan sonra gerekli bekleme süresine uyarım

%50'si Pazar koşullarına göre hareket ederim

%0'ı Bekleme süresine dikkat etmem

BULGULAR

Antalya ilinde en fazla nar üretimi yapılan Döşemaltı, Serik ve Aksu ilçelerindeki nar üreticileri ile yapılan anket çalışmasına göre narda en çok görülen hastalığın kahverengi leke (*Alternaria alternata*) en çok görülen zararlının ise Nar Yaprak Biti (*Aphis punicae*) olduğu belirlenmiştir. Üreticilerin %60'nın düzenli olarak ilaç attığı, genellikle pestisitleri zirai ilaç bayilerinden temin ettikleri ve ilaç dozunu ayarlarken de bayilerin önerilerine göre yaptıkları ayrıca pestisitlerin etkili olmasına özen gösterdikleri ifade edilmiştir. Üreticilerin %84'ü sezon boyunca 8-15 kez ilaçlama yapmaktadırlar. Tarım ilaçlarını hazırlarken % 54'ü hiçbir önlem almazken, ilaç uygularken %50'si yemek yememeye, sigara içmemeye dikkat etmektedir. Genellikle ilaçlamayı akşam saatlerinde yaptıklarını, ilaçlama yaparken suyun ph'ına bakmadıklarını, birden fazla hastalık ve zararlı ile mücadele etmek için ilaçları karıştırarak kullandıklarını, üreticilerin tamamının ilaçlamayı ilaçlama makinası ile yaptıklarını belirtmişlerdir. Ayrıca anket çalışması yapılan üreticilerden %60'ı bitki koruma ürünleri uygulamaya yönelik kurs ve eğitim aldıklarını söylemişlerdir.

SONUÇ

Antalya genelinde üreticiler zararlı ve hastalıklarla mücadelede pestisit kullanımını tercih etmektedirler. Özellikle nar üreticileri sezon boyunca ürün ve ağaç kaybı yaşamamak için yüksek oranda insektisit ve fungisit kullanmaktadır. Yapılan anket çalışmasına göre Antalya ilinde nar üretiminde tarımsal faaliyetlerin en önemli kısmını oluşturan çiftçilerin yetiştiricilik ile ilgili bilgi ve sorumlulukları açısından eksiklikler tespit edilmiştir. Bu eksikliklerin giderilmesi amacıyla çeşitli önerilerde bulunulabilir. Üreticilerin gereğinden fazla pestisit kullanma istekleri vardır ve satın aldıkları ilaçları yetersiz bulmaktadırlar. Çiftçilere özellikle pestisitler

konusunda Tarım İl ve İlçe Müdürlükleri tarafından doğru pestisit kullanımı ve uygulamaları hakkında bilgi verilmelidir. Bakanlık tarafından zaman zaman pestisitler, çevre sağlığı, doğal denge vb. konularında eğitim ve seminerler verilmelidir. Genel olarak bitki koruma uygulamaları ve pestisit kullanım sorumluluğu konusunda yasal düzenlemelerin getirilmesi ve uygulanmasının sağlanması yararlı olacaktır.

DRAFT

KAYNAKLAR

Dursun E, Ikinci A, Bolat I 2019. Türkiye ve Şanlıurfa İlinde Nar Yetiştiriciliğinin Bugünkü Durumu ve Geleceği & Present Situation and Future of Pomegranate Cultivation in Turkey and in Şanlıurfa City.

Gölükcü M, Tokgöz H, KIRALAN M 2008. Ülkemizde yetiştirilen önemli nar (Punica granatum) çeşitlerine ait çekirdeklerin bazı özellikleri. Gıda 33:281–290.

Kurt H, Şahin G 2013. Bir Ziraat Coğrafyası Çalışması: Türkiye'de Nar (Punica granatum L.) Tarımı. e-Marmara Coğrafya Dergisi (elektronik)

Lansky E, Shubert S, Neeman I 1998. Pharmological and therapeutic properties of pomegranate, 231-235. In: Proceedings of the I. International Symposium of Pomegranate [15-17 October 1998, Orihuela (Alicante) Spain], 389pp

ÖZALP A, YILMAZ İ 2013. Antalya ili nar üretiminde girdi kullanımı ve kârlılık analizi. Akdeniz University Journal of the Faculty of Agriculture 26:19–26.

Özgüven A, yılmaz C, Yılmaz M, et al 2015. NAR YETİŞTİRİCİLİĞİ-Kıbrıs Ekolojik Koşullarında Değişik Nar Çeşitlerinin Adaptasyonu.

Öztürk N, Ulusoy MR, Bayhan E 2005. Doğu Akdeniz Bölgesi nar alanlarında saptanan zararlılar ve doğal düşman türleri. Türk. entomol. derg., 29 (3): 225-235.

Şimşek M, Gülsoy E 2017. Güneydoğu Anadolu Bölgesinin nar (Punica granatum L.) potansiyeli konusunda bir araştırma. Journal of the Institute of Science and Technology 7:31–41.

DRAFT

ORAL PRESENTATION

Combined efforts in tissue engineering and organ-on-chips to study cancer metastasis

Reyhan Coban (*https://orcid.org/0009-0007-9297-9867*), Yusuf Caglar Odabasi (https://orcid.org/0000-0003-0690-5575), Ozlem Yesil-Celiktas* (https://orcid.org/0000-0003-4509-2212)

Ege University, Faculty of Engineering, Bioengineering Department, Izmir, Turkey

*Corresponding author e-mail: ozlem.yesil.celiktas@ege.edu.tr

Abstract

Cancer metastasis is a complex phenomenon driven by a plethora of mechanical and molecular factors. Although animal models have been widely used for testing of drug candidates to elicit anti-metastatic drug candidates, these *in vivo* models can not fully recapitulate the pathology. Organ-on-chip (OoC) platforms designed to address this unmet need stands out as an advance approach. Organ-on-chips are microfluidic devices consisting of perfused channels and tissue constructs to emulate physiology at the tissue and organ level thus providing favorable environmental conditions for living cells. In the scope of this paper, an in-depth exploration of the key mechanisms underlying cancer metastasis, emphasizing the interplay between mechanical and molecular forces from tissue engineering perspective is provided. Furthermore, the studies related to organ-on-chip platforms that are used to investigate cancer metastasis are critically discussed.

Keywords: organ-on-chip, tissue engineering, cancer metastasis, extracellular matrix

INTRODUCTION

Cancer is the major cause of death worldwide, posing a serious public health problem. However, tumor metastasis continues to be the main cause of cancer-related deaths, which is the spread of cancer cells from the primary tumor to distant organs or tissues. Although animal models have been used for elicitation of potential drug candidates for many years, the specie differences results in bias (Perrin, 2014; Day et al., 2015) in terms of translation of the data to the clinical setting. Therefore, effective experimental models are required to understand the molecular drivers of metastatis. *In vitro* cell culture models provide an understanding of cellular mechanisms, which include cell differentiation, migration, and growth, which are influenced by chemical and mechanical microenvironments (Duval et al., 2017). 3D *in vitro* dynamic tumor models are known as highly suitable models for cancer research due to their closeness to in vivo models (Ma et al. 2012; Moshksayan et al. 2018). These models mostly consist of proliferating, quiescent, apoptotic, hypoxic and necrotic cell types, rendering them suitable for the investigation of intricate metastatic stages including invasion, intravasation, and extravasation (Azimian-Zaraveh et al. 2022). Simultaneously, the integration of microfluidic systems with 3D cultures, allows important cell-cell interactions and spatio-temporal changes to be imitated by both physical and mechanical forces (Saglam-Metiner et al., 2023). This integration closing the gap between traditional tissue culture studies and human systems (Portillo-Lara R et al. 2016; Yesil-Celiktas et al. 2018). To achieve more accurate replication of the complex stages of metastasis and establishment of the tumor microenvironment, it becomes imperative discussion on extracellular matrices (ECM). The ECM, provides structural stability and functional flexibility to cells, tissues, and organs owing to specific macromolecules and enzymes (Kyriakopoulou K et al, 2023). They also consist of proteins and polysaccharides that contain signals not only for the physical environment but also for tissue development, homeostasis, and repairment of the tissue structure. (Gregory and Annette, 2009; Tsang et al., 2010) This makes them prime candidates for the generation of a more verisimilar tumor microenvironment.

Within the scope of tissue engineering aimed at emulating ECM-like tissue, bioinspired materials are being systematically developed that mimic hierarchical structures/patterns in nature with their unique properties and functions (chemical and physical properties, structural hierarchy, composition, and more) (Arslan et al, 2023). Synthetic and naturally derived bioinspired materials cannot fully recapitulate the biological, chemical and mechanical properties of the human tissue microenvironment (Nguyen and Camci-Unal, 2020). Plant-based

scaffolds have emerged in recent years to mimic many properties, especially microstructural, of natural ECM. Plant-based scaffolds are promising due to their high surface area, natural vascular networks, varying porosity ranges, hardness, mechanical properties and water transport (Adamski et al, 2018; Hickey and Pelling, 2019). When mimicking on-chip metastasis models, it is important to create vascularized systems similar to those in the body to better reflect the mechanical and physicochemical process. Instead of the materials and tissue scaffolds used for this purpose, plant-derived vascular systems can be used in metastasis modeling due to their various advantages.

Here we summarize reproducible models of cancer metastasis process in the circulatory system by focusing on *in vitro* microfluidic systems, which have become rapid trend in recent years.

RESULTS and DISCUSSION

Understanding of cancer metastasis helps to design and fabricate advance organ-on-chip platforms. Indeed, the metastatic cascade is a stepwise process comprising several distinct stages (**Fig. 1**). The first step involves the invasion of cancer cells into the surrounding tissue. This process is facilitated by the loss of cell adhesion, increased cell motility, and the secretion of enzymes that degrade the extracellular matrix (ECM) (Martin et al, 2013). Mechanically, this involves cell contractility and the ability to penetrate tissue barriers. Cancer cells that have invaded locally may enter blood vessels (intravasation) or lymphatic vessels. This step relies on both molecular interactions, such as adhesion molecules like integrins, and mechanical forces generated by cell contractility (Fujimoto et al, 2021).

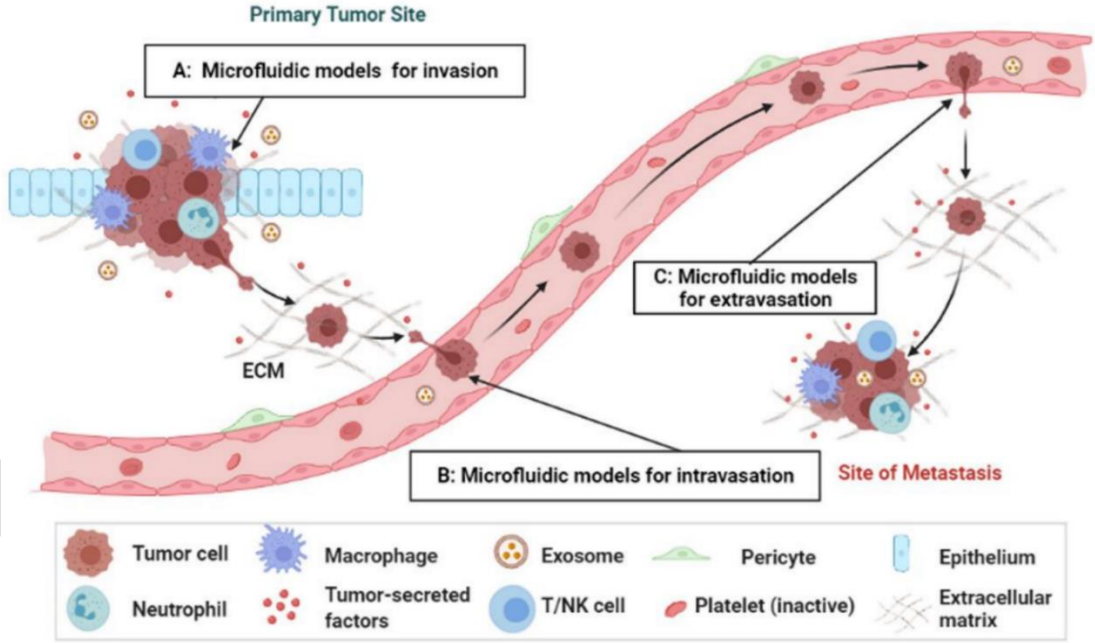


Figure 1. Microfluidic models involving metastatic steps such as invasion/intravasation (A,B) and extravasation (C) processes, by Zhang et al. 2022, Open access publication under CC-BY licence

This is followed by survival in the circulation. Circulating tumor cells (CTCs) face mechanical challenges in the bloodstream, including shear forces. Molecular mechanisms like immune evasion and resistance to anoikis (detachment-induced cell death) are crucial for their survival. Upon reaching distant organs, CTCs must extravasate, or exit the circulation, and enter the parenchyma of the target organ. This process involves interactions with endothelial cells, molecular signals, and mechanical squeezing through tight spaces (Joosse et al, 2015). CTCs that successfully extravasate may form micrometastases in the target tissue (Dujon et al, 2021). This requires the activation of dormancy-associated molecular pathways and the establishment of a suitable microenvironment. To become clinically detectable, micrometastases require sustained growth and angiogenesis, driven by molecular factors like vascular endothelial growth factor (VEGF) and mechanical expansion (Balayan et al., 2022). Eventually, some micrometastases evolve into macro-metastases, causing overt clinical symptoms and complications (Chu et al., 2022). In addition to the metastatic cascade, understanding of mechanical and molecular factors are imperative as well. Cancer metastasis is a multi-step

process orchestrated by a myriad of mechanical and molecular factors. An in-depth exploration of the key mechanisms underlying cancer metastasis, emphasizing the interplay between mechanical and molecular forces from tissue engineering perspective is provided.

Mechanical and molecular factors in metastasis

Cancer cells exhibit enhanced motility driven by actin polymerization, focal adhesion turnover, and membrane protrusions. Mechanotransduction pathways, such as Rho GTPases, play a role in this process. Cancer cells produce matrix metalloproteinases (MMPs) to degrade the ECM, allowing them to infiltrate neighboring tissues. The balance between cell contractility and ECM stiffness influences invasion (Martin et al., 2013). There are circulatory challenges to consider. CTCs experience shear forces, collisions with blood cells, and interactions with the endothelium during circulation. Their ability to withstand these mechanical stresses is crucial for metastasis. Finally, mechanical factors influence the ability of CTCs to extravasate and adapt to the microenvironment at distant sites. This involves interactions with the ECM, stromal cells, and the vasculature. Epithelial-Mesenchymal Transition (EMT) is a critical molecular process in metastasis, involving the downregulation of epithelial markers and upregulation of mesenchymal markers, enhancing cell motility and invasion (Ribatti et al., 2020). Molecular signaling via chemokines and cytokines guides the homing of CTCs to specific organs. The 'seed and soil' hypothesis suggest that interactions between tumor cells (seeds) and the microenvironment (soil) are crucial for metastatic colonization. Molecular signals, including VEGF, promote the formation of new blood vessels within metastatic lesions, ensuring a nutrient supply and facilitating growth. Metastatic cells can evade immune surveillance through various mechanisms, including downregulation of major histocompatibility complex (MHC) molecules and expression of immune checkpoint molecules.

Cancer metastasis-on-chip

Despite the highly controllable *in vitro* 2D models, reproducing the complexity of the tumor microenvironment poses a significant challenge. The adaptation of cancer metastasis to organ-on-chip technologies are promising for understanding the disease. Specifically, within the realm of 'metastasis-on-chip,' the targeted platforms involve distinct microfluidic chips designed to establish connections between cancer tissues, both within the same chip and across multiple chips, while also interfacing with vascular systems lined with endothelial cells. Metastasis-on-chip has numerous advantages over its *in vivo* counterparts in terms of clinical applications. There are many studies in the literature where it is used to examine the contribution of mechanical and biochemical cues in spread tumor (Carvalho et al., 2015; Lee et al., 2016; Portillo-Lara and Annabi, 2016). In parallel, the spread of invasion, intravasion, extravasion and angiogenesis in the metastasis process has been remodeled (Caballero et al., 2017).

Invasion models

The invasion process involves the reduction of the capacity of adhesion force in cell-cell interactions and subsequent dispersal of invasive tumor cells from the primary tumor and into the surrounding stroma. During the process, number of substances are secreted to degrade the extracellular matrix and the degradation of the expression of proteins are involved in the control of migration (Martin et al., 2013). In recent years, numerous "metastasis-on-chip" models have been developed with the aim to gain better understanding of the phenomena, elucidate the origin of cancer cells, and examine the differences in the composition of the cellular microenvironment both from mechanical and physicochemical perspectives. In a study an on-chip invasion model, the so-called Petri dish-based liquid feeding system was designed to examine the long-term invasion of gastric cancer, which is known to have high metastatic potential **(Fig. 2A).** The system was characterized by its simplicity, cost-effectiveness, and replicability. It also represented a prospective model for anticancer drug screening, metastasis mechanism research, and even the discovery of new biomarkers associated with metastasis through the investigation of molecular differences such as proteins and microRNA differences between the host and invading cells (Chen et al., 2016).

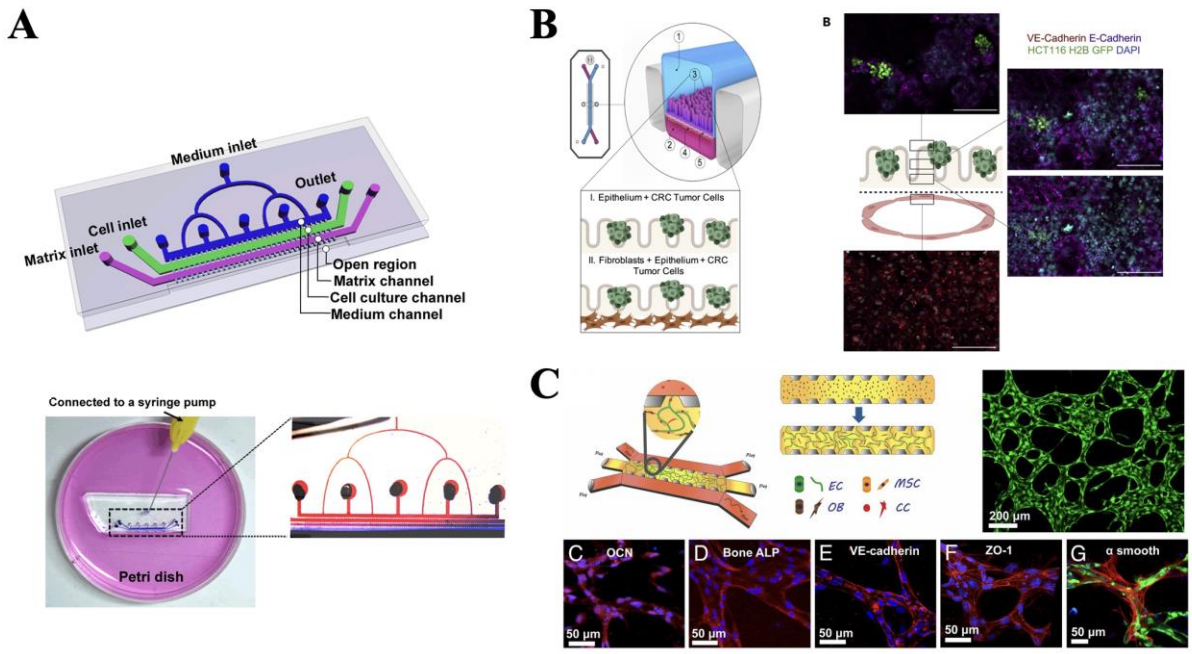


Figure 2. Cancer metastasis-on-chip models. **(A)** A schematic illustration of invasion-on-chip model (Top). A depiction of the microfluidic system comprising a PDMS-glass chip and a Petri dish with culture medium (Bottom left). An image showing a continuous chemical gradient by red and blue dye inside the device using the designed liquid supply system (Bottom right). (Chen et al. 2016, Open access publication under CC-BY license) **(B)** The organ-on-chip intravasation platform (schematic courtesy of Emulate, Inc.) consists of an epithelial channel (1) comprising epithelial and cancerous cells (3) and an endothelial channel (2) comprising HUVEC cells (4) separated by a porous membrane (5). Confocal fluorescence images of a chip cross-section spanning 106 µm from the top of the endothelial channel into the epithelial channel, highlighting the endothelial: epithelial tissue:tissue interface. Scale bar is 100 µm (Strelez et al., 2021, Open access publication under CC-BY license). **(C)** Schematic illustration of the extravasation model. Two side media channels allow addition of cancer cells, biochemical factors, and flow across the vasculature formed in the gel channel. Endothelial cells (ECs), MSCs, and osteoblast-differentiated cells (OBs) are initially seeded in the gel. ECs form vasculature, whereas MSCs and OBs create a microenvironment. Cancer cells introduced in the vessel extravasate into the organ-mimicking gel (Jeon et al., 2015, Open access publication under CC-BY license).

Intravasation models

The carcinoma cells originated from epithelial cells, locally invade tissues and gain access to the circulatory system through lymphatic or blood vessels. This cascade of events is referred as intravasation (Marciel and Hoffmann, 2017). Examining these critical steps in cancer metastasis is vital to understand the whole process. Within a study, a microfluidic model was devised to understand intravasation, known as the early metastatic period of colorectal cancer, and identify new therapeutic targets in the tumor microenvironment. When examined from an alternative standpoint within the same study, the other objective in developing this system was to create a chip that integrates various cell types and mechanical effects, as well as to examine the effect of the tumor microenvironment with respect to the spread of primary colon tumor **(Fig 2B)**. The platform was suitable for researching the colorectal cancer intravasation process by emulating the spread of tumor cells into the blood vessel (Strelez et al., 2021).

Extravasation models

As previously elucidated, intravasation is a process in which primary tumor cells move beyond the endothelial membrane of blood vessels to join the circulation. Extravasation is the process of tumor cells leaving the circulation and entering a metastatic area or organ. After the extravasation process, secondary tumor formation begins (Zhang et al., 2022). Similar to intravasation, numerous "metastasis-on-chip" devices have been developed to examine extravasation. In a research aimed at elucidating the microenvironment specific to the human vascularized organ was created to investigate the extravasation process of metastatic tumor cells. Within this context, they developed a microfluidic 3D *in vitro* model (**Fig. 2C**) to examine the invasion of

human breast cancer cells into an environment that mimics the bone and muscle microenvironment (Jeon et al., 2015).

Angiogenesis models

Tumor angiogenesis, known as the formation of new blood vessels in the tumor stroma, provides oxygen and nutrients to cancer cells, preparing the ground for the initiation of metastasis (Stroka, 2014). Physiological angiogenesis involves receptor activation of endothelial cells with angiogenic growth factors and subsequent disruption of the basement membrane by endothelial cells (Caplin et al., 2015). Furthermore, tumor angiogenesis has differences compared to physiological angiogenesis. For example, tumor vasculature is characterized by irregular sprouting, tortuous capillary networks, and leaky barrier properties (Portillo-Lara et al. 2016; Lee et al., 2016). However, this angiogenic vascular structure growing around the tumor is abnormal and leaky (Munn et al., 2003).

CONCLUSION

Cancer metastasis is a multifaceted process, driven by intricate interactions between mechanical and molecular factors. Understanding these mechanisms is crucial for the development of targeted therapies aimed at inhibiting metastatic progression and improving cancer patient outcomes. Future research should continue to elucidate the complex interplay between mechanical and molecular forces in metastasis, offering new avenues for intervention and treatment. The utilization of cancer metastasis-on-chip platforms can accelerate the translation of potential drug candidates to the clinical setting.

ACKNOWLEDGEMENTS

The financial support provided by TUBITAK through grant no. 120N422 is highly appreciated.

REFERENCES

Arslan Y, Alessia Paradiso, Naz Celiktas, Tolga Erdogan, Ozlem Yesil-Celiktas, Wojciech Swieszkowski 2023. Bioinspired microstructures through decellularization of plants for tissue engineering applications. European Polymer Journal, 198, 112415.

Azimian Zavareh, V., Rafiee, L., Sheikholeslam, M., Shariati, L., Vaseghi, G., Savoji, H., & Haghjooy Javanmard, S., 2022. Three-Dimensional in Vitro Models: A Promising Tool To Scale-Up Breast Cancer Research. *ACS Biomaterials Science & Engineering*, *8*(11): 4648-4672.

Balayan, V., & Guddati, A. K., 2022. Tumor dormancy: Biologic and therapeutic implications. World Journal of Oncology, 13(1): 8.

Caballero, D., Kaushik, S., Correlo, V. M., Oliveira, J. M., Reis, R. L., & Kundu, S. C., 2017. Organ-on-chip models of cancer metastasis for future personalized medicine: From chip to the patient. Biomaterials, 149: 98-115.

Caplin, J. D., Granados, N. G., James, M. R., Montazami, R., & Hashemi, N., 2015. Microfluidic organ-on-a-chip technology for advancement of drug development and toxicology. Advanced healthcare materials, 4(10): 1426-1450.

Carvalho, M. R., Lima, D., Reis, R. L., Correlo, V. M., & Oliveira, J. M., 2015. Evaluating biomaterial-and microfluidic-based 3D tumor models. Trends in biotechnology, 33(11): 667-678.

Chen, Z. Z., Li, W. M., Zhang, Y., Yu, M., Shan, L. F., Yuan, D. Z., ... & Fang, J., 2016. Establishment of a gastric cancer subline with high metastatic potential using a novel microfluidic system. Scientific reports, 6(1): 38376.

Chu, Y., Sun, T., & Jiang, C., 2022. Emerging landscapes of nanosystems based on pre-metastatic microenvironment for cancer theranostics. Chinese Chemical Letters, 33(9): 4157-4168.

Day CP, Merlino G, Van Dyke T 2015. Preclinical Mouse Cancer Models: A Maze of Opportunities and Challenges. Cell, 163(1): 39–53

Duval, K., Grover, H., Han, L.-H., Mou, Y., Pegoraro, A. F., Fredberg, J. J., & Chen, Z., 2017, Modeling Physiological Events in 2D vs. 3D Cell Culture. *Physiology*, *32*(4): 266–277.

Hickey, R. J., & Pelling, A. E., 2019. Cellulose biomaterials for tissue engineering. Frontiers in bioengineering and biotechnology, 7, 45.

Jeon, J. S., Bersini, S., Gilardi, M., Dubini, G., Charest, J. L., Moretti, M., & Kamm, R. D., 2015. Human 3D vascularized organotypic microfluidic assays to study breast cancer cell extravasation. Proceedings of the National Academy of Sciences, 112(1): 214-219.

Joosse, S. A., Gorges, T. M., & Pantel, K. 2015. Biology, detection, and clinical implications of circulating tumor cells. EMBO molecular medicine, 7(1): 1-11.

Kyriakopoulou, K., Koutsakis, C., Piperigkou, Z., & Karamanos, N. K. 2023. Recreating the extracellular matrix: novel 3D cell culture platforms in cancer research. FEBS J.

Lee, E., Song, H. G., & Chen, C. S., 2016. Biomimetic on-a-chip platforms for studying cancer metastasis. Current opinion in chemical engineering, 11: 20-27.

Martin, T. A., Ye, L., Sanders, A. J., Lane, J., & Jiang, W. G., 2013. Cancer invasion and metastasis: molecular and cellular perspective. In Madame Curie Bioscience Database [Internet]. Landes Bioscience.

Martin, T. A., Ye, L., Sanders, A. J., Lane, J., & Jiang, W. G., 2013. Cancer invasion and metastasis: molecular and cellular perspective. In Madame Curie Bioscience Database [Internet]. Landes Bioscience.

Munn, L. L. 2003. Aberrant vascular architecture in tumors and its importance in drug-based therapies. Drug discovery today, 8(9): 396-403.

Nguyen, M. A., & Camci-Unal, G. 2020. Unconventional tissue engineering materials in disguise. *Trends in biotechnology*, *38*(2): 178-190.

Perrin, S., 2014, Preclinical research: Make mouse studies work. Nature, 507(7493): 423–425

Portillo-Lara R, Annabi N, 2016. Microengineered cancer-on-a-chip platforms to study the metastatic microenvironment. Lab Chip 16(21): 4063–4081

Portillo-Lara, R., & Annabi, N., 2016. Microengineered cancer-on-a-chip platforms to study the metastatic microenvironment. Lab on a chip, 16(21): 4063-4081.

Ribatti, D., Tamma, R., Annese, T., 2020. Epithelial-mesenchymal transition in cancer: A historical overview. Translational Oncology, 13, 100773.

Saglam-Metiner, P., Devamoglu, U., Filiz, Y., Akbari, S., Beceren, G., Goker, B., Yaldiz, B., Yanasik, S., Biray-Avci, C., Erdal, E., Yesil-Celiktas, O., 2023 Spatio-temporal dynamics enhance cellular diversity, neuronal function and further maturation of human cerebral organoids. Communications Biology, 6(1), 173.

Schultz, G. S., & Wysocki, A., 2009. Interactions between extracellular matrix and growth factors in wound healing. Wound repair and regeneration, 17(2): 153-162.

Sharifi F, Ozlem Yesil-Celiktas, Aslihan Kazan, Sushila Maharjan, Saghi Saghazadeh, Keikhosrow Firoozbakhsh, Bahar Firoozabadi, Yu Shrike Zhang 2020. A hepatocellular carcinoma-bone metastasis-on-a-chip model for studying thymoquinone loaded anti-cancer nanoparticles. Bio-design and Manufacturing, 3: 189-202.

Strelez, C., Chilakala, S., Ghaffarian, K., Lau, R., Spiller, E., Ung, N., ... & Mumenthaler, S. M., 2021. Human colorectal cancer-on-chip model to study the microenvironmental influence on early metastatic spread. Iscience, 24(5).

Stroka, K. M., & Konstantopoulos, K. 2014. Physical biology in cancer. 4. Physical cues guide tumor cell adhesion and migration. American Journal of Physiology-Cell Physiology, 306(2): C98-C109.

Tsang, K. Y., Cheung, M. C., Chan, D., & Cheah, K. S., 2010. The developmental roles of the extracellular matrix: beyond structure to regulation. Cell and tissue research, 339(1): 93-110.

Yaldiz B, Pelin Saglam-Metiner, Sefa Burak Cam, Petek Korkusuz, Ozlem Yesil-Celiktas, 2021. Effect of sterilization methods on the mechanical stability and extracellular matrix constituents of decellularized brain tissue. Journal of Supercritical Fluids, 175, 105299.

Yesil-Celiktas O, Hassan S, Miri AK, Maharjan S, Al-kharboosh R, Quiñones-Hinojosa A, Zhang YS, 2018 Mimicking human pathophysiology in organ-on-chip devices. Adv Biosyst 2:1800109

Yildiz-Ozturk E, Pelin Saglam-Metiner, Ozlem Yesil-Celiktas, 2021. Lung carcinoma spheroids embedded in a microfluidic platform. Cytotechnology, 73(3): 457-471.

Zhang, X., Karim, M., Hasan, M. M., Hooper, J., Wahab, R., Roy, S., & Al-Hilal, T. A., 2022. Cancer-on-a-Chip: Models for studying metastasis. Cancers, 14(3): 648.

Zhang, X., Karim, M., Hasan, M. M., Hooper, J., Wahab, R., Roy, S., & Al-Hilal, T. A., 2022. Cancer-on-a-Chip: Models for studying metastasis. Cancers, 14(3): 648.

ORAL PRESENTATION

Steroid Hormonlarından Östrojenin Kontrollü Salımı İçin Biyouyumlu İmplantın Hazırlanması Ve Salım Profilinin Çıkartılması

Selda Şimşek[1] (0000-0002-1585-7379), Sema Çetin[2] (0000-0001-8442-4019), Hesna Ural Ulucan[3] (0000-0003-1885-6766)

[1]Kırıkkale Universitesi, Fen Edebiyat Fakültesi, Biyoloji Bölümü,
[2]Kırıkkale Universitesi, Mühendislik ve Doğa Bilimleri Fakültesi, Biyoloji Bölümü,
[3]Ufuk üniversitesi, SHMYO Tıbbi Hizmetler ve Teknikleri Bölümü, Tıbbi Laboratuvar Programı

susimsek77@hotmail.com

Özet

Bu çalışmamızla, çok fazla kullanım alanı olan ve yararlı etkilerinin yanında yan etkileri de bulunan steroidlerden östrojen hormonunun eksikliğine bağlı görülen şikayetlerin, geliştireceğimiz biyouyumlu implant ile salım profili çıkartılarak ortadan kaldırılacağı ve kontrollü salım sistemlerinin avantajlarından da yararlanılarak daha uygun bir tedavi oluşturulabileceği düşüncesindeyiz.İlaç salım sistemleri oluşturulurken ilacın en kısa sürede istenilen kan konsantrasyonuna ulaşması ve ilacın kontrollü bir şekilde belirlenen süre içerisinde devam ettirilmesi amacıyla HEMA monomeri ve/veya MMA monomerinin UV fotopolimerizasyon yöntemi ile kopolimerizasyonu gerçekleştirdi ve ilaç salım sisteminde implant olarak kullanılacak bu kopolimerler Fourier Transform Infrared Spectroscopy (FTIR), Scanning Electron Microscope (SEM), Differential Scanning Calorimetry (DSC), yüzey temas açısı ölçümü ve biyouyumluluk testleriyle de biyouyumlulukları incelenip karakterize edildi. FTIR spektroskopisinden elde edilen bulgular hedeflenen polimer yapısını gösterdi.Termal kararlılıkları belirlemek amacıyla termogravimetrik analiz yapıldı. Ölçümler 100-500°C sıcaklıklar arasında, hava ve azot atmosferinde 10°C/d hız ile gerçekleştirildi. Termal kararlılığın, pHEMA hidrojelinin yapısına MMA komonomerinin katılması ile azaldığı görüldü. P(HEMA-MMA) kopolimer hidrojelin fizyolojik fosfat tamponundaki denge suyu içeriğinin HEMA'ya göre daha düşük olduğu gözlendi. Elde edilen SEM mikrograflarının görüntüsünden, hidrojel yapıdaki pHEMA ve p(HEMA-MMA)'nın gözeneksiz ve düzgün bir yüzey morfolojisine sahip olduğu görüldü. İlaç salım deneyleri için, östrojenin 0,01mg/ml, 0,1mg/ml ve 0,2 mg/ml üç farklı dozunun salım kinetikleri, fizyolojik fosfat tamponu pH 7.4 PBS içerisinde sürekli akış salım sistemi ile değerlendirildi ve spektrofotometrik metot ile, 260 nm dalga boyundaki spektrumları kaydedildi. pHEMA hidrojellerine yüklenen östrojenin salımı 288 saatte yani 12 günde, p(HEMA-MMA) hidrojellerine yüklenen östrojenin salımı ise 440 saatte yani yaklaşık 18 günde tamamlandı ve bu süre boyunca kontrollü salım sisteminin bazal östrojen seviyesi için başarıyla uygulanabileceği gösterildi. Bu çalışmamızla, pHEMA ve p(HEMA-MMA) hidrojellerinin zamana bağlı kümülatif salım yüzdesi sonuçlarına baktığımızda, geliştirdiğimiz biyouyumlu taşıyıcı implantın hem daha yavaş salım, hem de daha uzun salım profili gösterdiği ve salımın %100 gerçekleştiği görüldü.

Anahtar Kelimeler: Biyomalzeme, Hidrojel, Kontrollü salım sistemleri, Östrojen

Preparation And Release Profile Of Biocompatible Implant For Controlled Release Of Estrogen From Steroid Hormones

Abstract

With this study, we think that the complaints related to the deficiency of estrogen hormone, which has many areas of use and has side effects as well as beneficial effects, will be eliminated by determining the release profile with the biocompatible implant we will develop, and a more suitable treatment can be created by taking advantage of the advantages of controlled release systems. While creating drug delivery systems, copolymerization of HEMA monomer and/or MMA monomer was carried out with the UV

photopolymerization method in order to reach the desired blood concentration in the shortest time and to continue the drug within a determined period of time in a controlled manner, and these copolymers to be used as implants in the drug delivery system were examined by Fourier Transform Infrared Spectroscopy (FTIR), Scanning Electron Microscope (SEM), Differential Scanning Calorimetry (DSC), surface contact angle measurement and was also examined and characterized their biocompatibility using biocompatibility tests. Findings from FTIR spectroscopy showed the targeted polymer structure. Thermogravimetric analysis was performed to determine thermal stabilities. Measurements were carried out at temperatures between 100-500°C, in air and nitrogen atmosphere, at a speed of 10°C/d. It was observed that thermal stability decreased with the incorporation of MMA comonomer into the structure of pHEMA hydrogel. It was observed that the equilibrium water content of the P(HEMA-MMA) copolymer hydrogel in the physiological phosphate buffer was lower than HEMA. From the images of the obtained SEM micrographs, it was seen that pHEMA and p(HEMA-MMA) in the hydrogel structure had a non-porous and smooth surface morphology. For drug release experiments, the release kinetics of three different doses of estrogen, 0.01mg/ml, 0.1mg/ml and 0.2mg/ml, were evaluated by a continuous flow release system in physiological phosphate buffer pH 7.4 PBS and by spectrophotometric method, at 260 nm. wavelength spectra were recorded. The release of estrogen loaded into pHEMA hydrogels was completed in 288 hours, (in 12 days), and the release of estrogen loaded into p(HEMA-MMA) hydrogels was completed in 440 hours (18 days), and during this period, it was shown that the controlled release system could be applied successfully for the basal estrogen level. When we look at the results of time-dependent cumulative release percentage of pHEMA and p(HEMA-MMA) hydrogels in this study, it was seen that the biocompatible carrier implant we developed showed both a slower release and a longer release profile and the release was 100% realized.

Keywords: Biomaterial, Hydrogel, Controlled release systems, Estrogen

GİRİŞ

Günümüzde "Biyomalzeme Bilimi" büyük gelişmelerin olduğu bir bilim dalıdır. Biyomalzemeler temel olarak tıbbi uygulamalarda kullanılmakla birlikte biyoteknoloji alanında da kullanılmakdır.(1) Kontrollü salım, etkin maddenin bir sistem içerisinden istenilen sürede, belirlenmiş bir hızla ve gereken miktarda salımını sağlayacak şekilde tasarımının yapıldığı bir yöntem olan kontrollü salımda iki ana bileşen vardır; aktif madde ve aktif maddenin salımını düzenleyen polimerik destek maddedir. Kontrollü ilaç serbestleşmesinde kullanılan doğal ya da sentetik polimer , ilaç ile birlikte kullanıldığında etken madde daha önceden tasarlanan şekilde salınır.(2)Bilimsel çalışmalarda oldukça sık kullanılan biyomalzemelerden biri olan hidrojeller, yumuşak ve esnek yapıda olmaları ,suyu yapılarında tutabilme özellikleri ile canlı dokulara benzerlik göstermektedirler. Biyolojik olarak parçalanan, toksisitesi olmayan ve biyolojik uyum gösterebilen yapılarından dolayı, hidrojeller biyomalzeme olarak kullanılması giderek güvenli hale gelmektedir. Ayrıca, ilacın salınım hızını düzenleyen geleneksel dozaj yöntemleri karşısında hidrojellerin kontrollü serbest bırakma mekanizması, daha fazla avantaj sağlamaktadır.(3)

Kontrollü salım çalışmalarında kullanılan hidrojellerden p(HEMA) ve/veya p HEMA -MMA sentetik ve mekanik dayanımı yüksek, , hidrofilik yapıda, biyolojik olarak uyumlu ve antibakteriyel özelliğe sahip, biyolojik ve kimyasal degredasyona karşı dirençli bir materyaldir ve akrilat kökenlidir. Akrilat kökenli olan diğer bir sentetik polimer de poli (metil metakrilat) -poli(MMA) 'dır . P(MMA) biyomateryal alanında kullanılan bir destek materyalidir.

Çalışmamızda üretilen hidrojellerin kimyasal gruplarını incelemek için, Fourier Dönüşümlü Infrared Spektrofotometre (FTIR) kullanılırken, malzemelerin yüzey morfolojisini incelemek için Taramalı Elektron Mikrokobu (SEM) kullanılacaktır.

Bu çalışmamızla, çok fazla kullanım alanı olan ve yararlı etkilerinin yanında yan etkileri de bulunan steroidlerden östrojen hormonunun eksikliğine bağlı görülen vajinal kuruluk, cinsel isteksizlik, saç dökülmesi , sıcak basması, çarpıntı, gece terlemesi, halsizlik ,sinirlilik, depresyon, gibi şikayetlerin, geliştireceğimiz biyouyumlu implant ile salım profili çıkartılarak ortadan kaldırılacağı ve kontrollü salım sistemlerinin avantajlarından da yararlanılarak daha uygun bir tedavi oluşturulabileceği düşüncesindeyiz.

2.MATERYAL VE METOT

2.1 Materyaller

Tez çalışmamızda kullanılan 2-Hydroxyethilmetakrilat (HEMA), metil methacrylate (MMA), Dr. Zeydanlı Hayat Bilimleri Ltd. Şti firmasından temin edildi. İnsan serum albumini, fibrinojen, γ-globulin, sığır serum albumini Sigma-Aldrich'dan elde edildi. Kullanılan diğer tüm kimyasallar analitik saflıkta olup, Merck AG (Darmstadt, Almanya) firmasından temin edildi.

Çalışmamızın aşamalarının tamamında kullanılan su, Barnstead (Dubuque, IA, USA) ROpure LP marka ters ozmoz, Barnstead D3804 NANOpure organik/kolloid uzaklaştırıcı yüksek akışlı selüloz asetat membran (Barnstead D2731) üniteleri ve iyon-değişimi dolgulu yatak kolonundan oluşan ultra-saf su sisteminden elde edildi.

2.2 Biyomateryallerin Sentezi

Farklı oranlarda denemesi yapılan ve son monomer oranı 1.00:1.00 (v/v) olacak şekilde kullanılan HEMA ve MMA monomerleri homojen olacak şekilde karıştırıldı. Yapıya çapraz bağlayıcı olarak kullanılacak olan N,N,metilenbisakrilamit (0,01 mg) ve tepkime başlatıcısı olarak amonyum persülfat (0,01 mg) eklendi. Sentezlenecek taşıyıcı implantın biyouyumluluğunu artırmak amacıyla 0,01mg HSA ve 0,02 mg PEG yapıya eklendi ve 2 dakika süresince polimer çözeltisinden, azot gazı geçirildi. Polimerizasyon karışımına hızlandırıcı olarak %10'luk 250µl TEMED eklendi ve 1 dakika boyunca azot gazından geçirilerek, 0.4 cm çapında ve 10 cm uzunluğundaki silindir kalıplara dökülerek oda sıcaklığında UV ışığı fotopolimerizasyon yöntemi ile sentezlendi. Polimerik taşıyıcı implantın (biyomateryalin) farklı miktarda östrojen hormonu yüklü eşlenikleri, yukarıda verilen aynı koşullarda UV ışığı fotopolimerizasyonu ile kontrollü ilaç salım sisteminde kullanılmak üzere sentezlendi. Polimerizasyon sonucunda oluşan silindirik biçimdeki polimerik taşıyıcılar damıtık su ile yıkanarak kullanılana kadar 4 oC sıcaklıkta muhafaza edildi.

2.3. Biyomateryalin Karakterizasyonu

Kontrollü salım için tasarlanan sistemler , uygun yüzey morfolojisi sahip olmalıdır. İlaç salım oranını belirleyen en önemli faktörlerden birisi yüzey alanıdır.(4) Çalışmamızda geliştirğimiz hidrojellerin salım sisteminin yüzey morfolojisini incelemek için, taşıyıcı sistemlerinin SEM mikrografları JEOL marka (Model, JSM 5600, Japonya) Taramalı Elektron Mikroskobu ile elde edilirken, BET yöntemi ile,gözenekliliği ve spesifik yüzey alanı belirlendi. Yoğunluğu , piknometre ile polimerin çözünmediği bir sıvı içerisinde belirlendi. Dijital kumpas ile de hidrojelin ıslak haldeki kalınlığı ölçüldü.

Piknometreler ,sıvı ve katı numunelerin yoğunluklarının, hassas şekilde ölçülmesi için kullanılan etkin test cihazlarımız içinde yer alır. Toprak dahil olmak üzere sıvı ve katıların yoğunluklarının dışında özgül ağırlıklarını ölçmek için de kullanılır.Genellikle camdan yapılan ve hava kabarcıklarının dışarı çıkması için kılcal tüplü buzlu cam tıpa aparatı bulunur.

Yüzey Analiz ve Porozite Cihazı (BET) , katı (toz) maddelerin çok hızlı, doğru ve hassas olarak yüzey alanı ve gözenek (por) boyut dağılımı bilgilerini elde etmeyi sağlar. Yüzey gözenekliliği, malzeme biliminde araştırmacıların ilgilendiği önemli bir özelliktir. Farmasötik, medikal implant, seramik, katalizör, boya ve kaplamalar, kozmetik, yakıt hücreleri, yerbilimi, adsorbanlar, yapı malzemeleri, elektronikler, maden, plastik ve polimerler, gibi çalışmalarda yaygın olarak kullanılmaktadır.

FTIR Spektroskopisi (Fourier Transform Infrared Spectroscopy)(Bruker, Vertex 70V,USA) ile birçok grup için karakteristik pikler alınmaktadır. Bu sayede spektrumu alınan maddede hangi karakteristik grupların olduğu belirlenerek maddenin yapısı yorumlanır.Özellikle bir kimyasal tepkimeyi izlemede önemli rol oynamaktadır. Bu amaçla; tarım, gıda, ilaç, polimer, petrol, metal, sanayilerindekullanım alanı bulmuştur.(5)

FTIR spektrofotometresi ile taşıyıcı polimerlerin FTIR spektrumu alındı. Kuru hidrojel, KBr ile karıştırılarak pelet hale getirildi. FTIR spektrumları alındı. MMA'in yapıya katılması, kontrol amaçlı kullanılan pHEMA spektrumundan oluşan farklı fonksiyonel gruplara ait bantların yardımı ile tespit edildi. Dijital kumpas ile P(HEMA-MMA) membranların ıslak durumdaki kalınlığı belirlendi. DSC (DifferentialScanningCalorimetry) (Model DSC-60-DTG-60H, Shimadzu, Japan) analizi ile pHEMA ve p(HEMA-MMA) taşıyıcı implantlarının mekanik dayanımı belirlendi.

Hidrojel yapıdaki biyomateryalin şişme özelliği, serum fizyolojik ve distile su içerisinde, oda sıcaklığında tampon sistemi içerisinde (pH2.5-8.5) gravimetrik yöntemle belirlendi. Çözelti pH'sı direkt olarak ölçüldü ve iyonik şiddet 0.2'ye ayarlanarak sabit tutuldu. Örnekler ilk olarak şişme ortamına yerleştirildi. Denge anına ulaşıldıktan sonra ortam değiştirildi. Biyomateryalin şişme oranı aşağıdaki eşitlik kullanılarak hesaplandı.

2.4. Serum Proteinleri Adsorpsiyonu

pHEMA ve p(HEMA-MMA) biyomateryallerinin kan uyumluluğunu saptamak için, 1/5 oranında fosfat

tamponunda seyreltilmiş insan kan serumu içerisine aktarıldı (7.5 ml, 50 mM, pH 7.4) ve 37˚C'de 120 dakika manyetik karıştırmalı hücrelerde kan serumu ile temasları sağlandı. Serum proteinlerinin adsorpsiyonu kesikli sistemde çalışıldı. Her bir protein için belirli başlangıç konsantrasyonunda çalışıldı. Taşıyıcı implanta adsorplanan protein miktarı flurosans spektrofotometre kullanılarak (Jasco, FP-750, Tokyo, Japonya) belirlendi.(6)

2.5. Kan Uyumluluk Analizi

pHEMA, p(HEMA-MMA) taşıyıcı implantları, 0.5 cm boyunda kesilerek, %0.85 NaCl çözeltisi içinde dengeye getirildi. Sağlıklı bir bireyden alınan venöz kan örneği, 1/9 oranında sodyum sitratla karıştırıldı ve 3000 rpm'de 10 dakika santrifüjlenerek plazması elde edildi. Sodyum sitratlı plazmadan, 300 µl alınarak, polimer tüpleriyle temas ettirildi ve 1 saat inkübe edildi. Polimerlerle temas etmemiş plazma kontrol olarak kullanıldı.(7)

2.6. İnvitro Salım Çalışmaları

Östrojen kontrollü salımının incelenmesi için , taşıyıcı destek materyaline hazırlanan matriks içi tutuklama yolu ile östrojen yüklendi. PEO(polietilen oksit) ve albumin içeren farklı monomer oranlarına sahip taşıyıcı polimerik implantlanta, farklı miktarda östrojen 0,01mg/ml, 0,1mg/ml ve 0,2 mg/ml yüklemesi yapılarak sistem parametrelerinin salım profiline ve salım kinetiğine etkisi araştırıldı. Taşıyıcı implanta yüklenen östrojen miktarı, standart BSA kalibrasyon eğrisinin elde edildiği Bradford yöntemi kullanılarak belirlendi. Östrojen yüklü biyomateryaller sürekli sistem reaktörüne yerleştirilerek sisteme sabit akış hızında, peristaltik pompa ile (Ismatec, IPG Model, Almanya) fizyolojik tampon çözeltisi girişi sağlandı. Belirli zaman aralılıklarında alınan örneklerle salınan ilaç miktarı spektrofotometre ile (Labo Med.Inc. Spectro UV-Vısrs Spectrophotometer), 260 nm dalga boyunda spektrofotometrik olarak takip edilerek belirlendi.

3. BULGULAR

İlaç alanındaki yapılan çalışmalarda asıl hedef, ilaç dozunu en az seviyeye indirmek, doz aralığını uzatmak, hastanın zararlı ve yan etkilerden etkilenmemesini sağlayarak biyoyararlanımını en üst seviyeye çıkartmaktır. Buna bağlı olarak tedavi sürecini kısaltarak hastanın yaşam kalitesini arttırmaktır. Kontrollü salım sistemleri ise bu beklentilere yanıt veren en iyi sistemdir. İlaç salım sistemleri çok kısa zamanda kardiyoloji, onkoloji, oftalmoloji, endokrinoloji ve immünoloji dahil olmak üzere tıbbın birçok dalında etkili olmuştur. Kontrollü salım çalışmalarında kullanılan hidrojellerden p(HEMA) ve/veya pHEMA -MMA sentetik ve mekanik dayanımı yüksek, biyolojik ve kimyasal degredasyona karşı dirençli bir materyaldir, hidrofilik yapıdadır, biyolojik olarak uyumludur. biyouyumlu olan pHEMA ve p(HEMA-MMA), silindir yapıda sentezlenerek karakterizasyon tayininin yapılması hedeflenmiştir. Bu tez kapsamında, protein adsorpsiyonuna karşı dirençli ve kan uyumluluk özelliği arttırılarak östrojen hormonunun kontrollü salımında kullanılmak üzere pHEMA temelli yeni bir taşıyıcı implant sisteminin geliştirilmesi tasarlandı.

3.1. Biyomateryalin Karakterizasyonu

Kontrollü salım çalışmamızda kullanılacak olan taşıyıcı implantların polimerizasyonunda ilk olarak çapraz bağlayıcı olarak kullanılan bisakrilamit oranı (0.01mg) ve redoks başlatıcısı olarak kullanılan amonyum persülfat (0.01mg) miktarları en uygun hale getirildi. En uygun değer bisakrilamit ve APS için 0.01mg olarak belirlendi. Bu değerlere göre ko-polimer membranı oluşturan HEMA:MMA ko-monomer oranı 1:0 ile 0:1 (v/v) arasında değiştirilerek hidrojel oluşumu, hazırlanan kompozisyondaki membranların yeterli mekanik güce etkisi, deneysel koşullarda incelendi ve elde edilen sonuçlarda p(HEMA-MMA) membranının mekanik gücünün ko-monomer oranına bağlı olarak değiştiği gözlendi (Çizelge 3.1). Monomer oranı 1:1 olan p(HEMA-MMA) membran kompozisyonu, yeterli mekanik güce sahip olduğundan çalışmanın daha sonraki aşamalarında kullanıldı.

Çizelge 3.1. Farklı HEMA:MMA oranlarında sentezlenen kompozit Hidrojelinin mekanik dayanıklılığa etkisi

HEMA:MMA oranı (v/v)	Polimerizasyon	Mekanik dayanıklılık
A)1.00:0.00	+	Orta
B) 1.00:0.25	+	Orta
C) 1.00:0.50	+	Yeterli
D) 1.00:1.00	+	Yeterli
E) 0.00:1.00	–	Yetersiz
F) 0.25:1.0	-	Yetersiz
G) 0.5:1.0	-	Yetersiz

Dijital kumpas yardımı ile ölçülen destek materyalinin kalınlığı 2.63mm olarak bulundu. Membran yapıdaki taşıyıcı implantın yoğunluğu Gay Lussac piknometresi ile materyal için çözücü olmayan bir sıvı (n-dekan) kullanılarak yapıldı. Hidrojelin yoğunluğunun 1.06 g/cm3 olduğu bulundu. Vakumlu etüvde, 30°C sıcaklıkta kurutulan kompozit hidrojel, azaltılmış basınç altında paladyum ile kaplandı ve taramalı elektron mikroskobu kullanılarak membranların elektron mikrografları elde edildi.

Elde edilen SEM mikrograflarının görüntüsünden, Hidrojel yapıdaki pHEMA ve p(HEMA-MMA)'nın gözeneksiz ve düzgün bir yüzey morfolojisine sahip olduğu görülmektedir. Biyomateryalin bu özelliği ilaç salım hızının yavaş ve kontrollü olmasını sağlayacaktır.

Sıcaklık, pH, çözücü bileşimi, ışık ve basınç gibi değişimlere karşı şişme veya büzüşme davranışı gösterebilen hidrojeller, ağırlıklarının 40-50 katı kadar şişme kapasitesine sahiptirler. Yapılarında tuttukları fazla su miktarından dolayı canlı dokulara benzer özellikler gösterdiğinden hidrojellerin biyomedikal alanlarda kullanımı oldukça dikkat çekmektedir.

Termal kararlılıkları belirlemek amacıyla termogravimetrik analiz yapılmıştır. Ölçümler 50°C -250°C sıcaklıkları arasında, hava ve azot atmosferinde 10°C/d hız ile gerçekleştirilmiştir.Termal kararlılığın, pHEMA hidrojelinin yapısına MMA komonomerinin katılması ile azaldığı görülmüştür.

Elde edilen polimerlerin karakterizasyonlarında FTIR spektroskopisinden yararlanılmış.

Hidrofobisite ile serbest yüzey enerjisi arasında ters bir ilişki vardır ve biri artarsa diğeri azalmaktadır. Polimerin hidrofobik karakterinin artması serbest yüzey enerjisinin azalması demektir. Polimerlerin serbest yüzey enerjileri farklı yöntemler (Zisman, Fowkes ve Van Oss Asit – Baz) kullanılarak hesaplanmıştır.

Fowkes Wu yöntemlerine göre hesaplanan yüzey enerjilerinin birbirlerine yakın olduğu, ancak Wu yönteminin Fowkes göre, daha düşük bir yüzey enerjisi polar bileşenine (γp) sahip olduğu gözlendi. Her iki yöntemde de, bütün test edilen membranlar için, dispersif bileşenler, toplam yüzey serbest enerjisine ana katkıyı yaptığı görüldü.

Toplam serbest yüzey enerjisi (γ^{Toplam}), van Oss yöntemi kullanılarak hesaplandı. van Oss yöntemi, araştırılan tüm membranlara farklı değerlerde uygulanan, Lifshitz-van der Waals (γ^{LW}) ve asit-baz bileşenlerinin (γ^{AB}) toplamıdır. Membranların baz bileşenlerinin (γ^{-}), asit bileşenlerine (γ^{+}) kıyasla daha yüksek olduğu görüldü.

3.2. Biyomateryalin Kan Uyumluluk Analizleri

Kan plazma proteinleri, albümin ,globülin ve fibrinojendir.Plazma proteinlerinden albümin kanın ozmotik basıncının ayarlanmasından sorumludur ve plazma proteinlerinin %60 'ını oluşturur. Plazma proteinlerinin %38'ini oluşturan globülinler α ve β globulinler yağları ve yağda çözünen vitaminleri taşırken γ globulin antikorları taşır. Plazma proteinlerinin %2'sini oluşturan fibrinojen ise pıhtılaşma faktörüdür.

Yapılan birçok araştırmada, biyomateryal olarak kullanılan polimerik biyomateryallerin, kan ile teması sonucunda plazma proteinlerini hızlı bir şekilde adsorpladığı rapor edilmiştir.(8) Sentezlenen pHEMA ve/veya pHEMA-MMA taşıyıcı implant sistemlerinde albumin adsorpsiyonu, biyouyumluluk açısından önemlidir.Fakat, biyomateryal yüzeyine fibrinojen adsorpsiyonu hazırlanan biyomateryalin biyolojik uyumluluğunuda azaltmaktadır. Albumin trombosite dirençli bir özellik göstermesi trombositlerin biyomateryal yüzeyine yapışmasını engeller. Fibrinojenin ise trombositlerin biyomateryal yüzeyine yapışmasını başlatıcı özelliği vardır. Bu sebeple, matriks içi tutuklama yöntemiyle hazırlanan biyomateryale albumin ve polietilen glikol(PEG) yerleştirilerek, biyomateryalin kan uyumluluğunda artış sağlanması hedeflenmiştir.(9)

Yaptığımız çalışmada yapısında albumin ve PEG bulunan pHEMA ve p(HEMA-MMA) taşıyıcı implantlarının adsorplanan kan serum protein miktarlarının ihmal edilebilir düzeyde olduğu görülmüştür.

3.3. Östrojen Hormonunun Kontrollü Salımı

İlaçların daha etkili ve daha pratik olarak hedefe iletilmesini hedeflendirilmiş ilaç taşıyıcı sistemler ile sağlanmaktadır. Kandaki ilaç konsantrasyonu, istenilen tedavi edici düzeyde uzun süre sabit tutulur ve böylece ilaçtan sağlanacak fayda artırılır. pHEMA ve p(HEMA-MMA) hidrojellerine yüklenen östrojen salımı, kontrollü
salım sisteminde belirli zaman aralıklarında alınan örneklerin analizi ile belirlenmiştir.

İlaç salım deneyleri için, Östrojenin 0,01mg/ml, 0,1mg/ml ve 0,2 mg/ml üç farklı dozunun salım kinetikleri, fizyolojik fosfat tamponu pH 7.4 PBS içerisinde sürekli akış salım sistemi ile değerlendirildi ve spektrofotometrik metot ile, 260nm dalga boyundaki spektrumları kaydedildi.

pHEMA hidrojellerine yüklenen östrojenin salımı 288 saatte yani 12 günde , p(HEMA-MMA) hidrojellerine yüklenen östrojenin salımı ise 440 saatte yani yaklaşık 18 saatte tamamlandı .Bu süre boyunca kontrollü salım sisteminde bazal östrojen seviyesi için başarıyla uygulanabileceğini gösterdik.

İlaç:polimer oranı, salınım özelliğini etkilediği düşünülen bir diğer parametredir. Hazırlanan taşıyıcı implantın hidrojel yapıda olması östrojenin destek materyalinin içinden dış ortama difüzyonunu kısıtlayan bir engel yoktur. Salım çalışmalarında, östrojenin daha yavaş salımının, taşıyıcı implanttaki ilaç: polimer oranına, gözeneksiz yüzey yapısına bağlı olduğu görüldü.

Bu çalışmamızda, pHEMA ve p(HEMA-MMA) hidrojellerinin zamana bağlı kümülatif salım yüzdesi sonuçlarına baktığımızda geliştirdiğimiz biyouyumlu taşıyıcı implant, hem daha yavaş salım hemde daha uzun salım profili gösterdi ve %100 salım gerçekleşti.

4. TARTIŞMA VE SONUÇ

Uzun yıllar çeşitli hastalıkları tedavi edici yeni bir molekül geliştirmek amacıyla, ilaç alanında yapılan çalışmaların ekonomik yükü ,araştırmaların uzun zaman alması ve her zaman istenilen sonucu vermemesi ilaç sanayinin karşılaştığı önemli sorunlar haline gelmiş ve yeni arayışları beraberinde getirmiştir. Bu çalışmalarla hastanın tedavisinin yanı sıra yaşam kalitesini arttırmakta amaçlanmaktadır.İlaç dozunu azaltma, dozlama aralığını uzatma, vücuda yan etki ve zararlı etkilerini ortadan kaldırma, ilacı hedef bölgeye gönderme çalışmaları son yıllarda önem kazanmıştır. Kontrollü salım sistemleri, bu beklentilere en iyi yanıt veren uygulama şeklidir.

Kontrollü salım sistemlerinde sıklıkla doğal veya yapay polimerlerden oluşan veya kompozit halde elde edilen polimerik malzemeler kullanılmaktadır. İlaç taşıyıcı sistemlerin hazırlanmasında daha çok polimerik biyomateryal kullanımı tercih edilir.

Gerçekleştirilen bu çalışmamızda; hidrojel yapıdaki pHEMA ve p(HEMA-MMA), literatür taraması ve denemeler sonucu belirlenen monomer oranlarında hazırlandı. Hazırlanan biyomateryale albumin ve polietilen glikol(PEG) yerleştirilerek, biyomateryalin kan uyumluluğunda artış sağlandı.

Östrojen hormonunun azalmasına bağlı görülen menopozda tedavinin amacı eksik olan östrojen hormonunu yerine koymaktır. Menopoz sonrası dönemde görülen kemik erimesinin önlenmesi ,kalp-damar hastalıkları gibi birçok şikayetin giderilmesini sağlayarak, menopoz öncesi, menopoz ve menopoz sonrası dönemde kadınların hayat standartları hormon replasman tedavisi ile artırılacaktır.

Çalışmamızda, taşıyıcı implant yüzey polaritesinin belirlenmesi için temas açısı ölçümleri gerçekleştirilmiştir. Su, gliserol ve diiyodometan (DIM) test sıvıları kullanılarak kompozit membranların temas açıları ölçümleri yapılmıştır. Östrojenin 0,01mg/ml, 0,1mg/ml ve 0,2 mg/ml üç farklı dozunun salım kinetikleri, fizyolojik fosfat tamponu (PBS, pH 7.4) içinde sürekli akış salım sistemi ile değerlendirildi ve spektrofotometrik metod ile 260 nm dalga boyunda polimerik silindirlerinden östrojenin salım miktarı tayin edildi. pHEMA hidrojellerine yüklenen östrojenin salımı 288 saatte yani 12 günde , p(HEMA-MMA) hidrojellerine yüklenen östrojenin salımı ise440 saatte yani yaklaşık 18 saatte tamamlandı .Bu süre boyunca kontrollü salım sisteminde bazal östrojen seviyesi için başarıyla uygulanabileceğini gösterdik.

Sonuç olarak yapmış olduğumuz analizlerin verileri sonucunda pHEMA, P(HEMA-MMA) nın kontrollü ilaç salımında taşıyıcı implant olarak başarıyla uygulanabileceğini ve biyolojik olarak en uyumlu hidrojelin p(HEMA-MMA) olduğu gösterilmiştir. Ayrıca toksisiteye yol açmaması, kolay sterilizasyon, organik çözücülerin kullanılmaması, numune dozunun ayarlanması gibi pek çok farklı avantajda sağladığını göstermektedir.

DRAFT

KAYNAKLAR

1.Ş.Y. Güven, Biyouyumluluk Ve Biyomalzemelerin Seçimi , Makine Mühendisliği Bölümü, Süleyman Demirel Üniversitesi, 32260, Isparta, Türkiye, Süleyman Demirel Üniversitesi Mühendislik Bilimleri ve Tasarım Dergisi 2(3),ÖS:BiyoMekanik2014, 303-311, 2014

2.H. Ural, İnsulin Hormonunun Kontrollü Salımında Kullanılmak Üzere Biyouyumlu Taşıyıcı İmplantın Hazırlanması ve Uygulaması, Doktora Tezi. Kırıkkale Üniversitesi, Kırıkkale, 2007

3.A. Ulusoy, N. Dikmen, Hidrojellerin Tıpta Uygulamaları, Cilt 29, Sayı 2, 129 - 137, 30.06.2020.

4. R. Blanco, A. Arai, N. Grinberg, D.M. Yarmush, B.L. Karger. Role of association on protein adsorption isotherms. β-lactoglobulin a adsorbed on a weakly hydrophobic surface, Journal of chromatography. 482, 1(1989).

5. P. Erdem, Kontrollü İlaç Salım Sisteminde Kullanılacak biyouyumlu Taşıyıcı İmplant materyallerinin Karakterizasyon Tayini, Kırıkkale Üniversitesi, Fen *Bilimleri Anabilim Dalı,(2014)*

6. Arıca MY. Epoxy-Derived pHEMA Membrane for Use Bioactive Macromolecules Immobilization: Covalently Bound Urease in a Continuous Model System. Journal of Applied Polymer Science. 2000;77:2000-2008. DOI: 10.1002/1097-4628(20000829)77:93.0.CO;2-M.

7. M.Yılmaz, Polihidroksi Etilmetakrilat Kökenli Yapay Damarların Hazırlanması Ve Biyo-Uyumluluk Özelliklerinin Arttırılması Ve Karakterizasyonu, Kırıkkale Fen Bilimleri Enstitüsü, Doktora Tezi, 2006.

8. Hong J., Andersson J., Ekdahl, K.N., Elgue G., Axen N, Larsson R., Contact between a polymer and whole blood: Sequence of events leading to thrombin generation, J. Lab. Clin. Med., 138, 139-45, (1999). Hsiue G.H., Yang J.M., Wu R.L., Preparation and properties of a biomaterial: HEMA grafted SBS by γ-ray irradiation, J. Biomed. Mat. Res., 22, 405-9 (1988).

9.Lopez, M.J., Moreno, J., Ramos-Cormenzana, A., Xanthomonas campestris strain selection for xanthan production from olivemill waste waters, Water Research, 35, 1828–1830, 2001.

DRAFT

ORAL PRESENTATION

Kinoa ve Gıda Endüsrisinde Kullanım Alanları

İrem BAĞCI[1*](*https://orcid.org/0000-0002-1376-5657*), Cem Baltacıoğlu[1] *(https://orcid.org/0000-0001-8308-5991)*

[1] Niğde Ömer Halisdemir Üniversitesi, Mühendislik Fakültesi, Gıda Mühendisliği Bölümü, Niğde, Türkiye

[*]rmbgc.99@gmail.com

Özet

Kinoa (Chenopodium quinoa Willd.) geleneksel olarak Güney Amerika kültürleri tarafından binlerce yıldır tüketilen ve günümüzde dünya genelinde işlevsel bir gıda olarak dikkat çeken tahıl benzeri (pseudocereal) bir bitkidir. Günden güne kinoaya ilginin artmasıyla insan ve hayvan beslenmesinde kullanımı yaygınlaşmıştır. Üzerine çalışmalar yapılması ve birçok gıda bileşimine katılması da popülerliğini arttırmıştır. Ülkemizde de yetiştiriciliği ve denemeleri yapılmakta olan bu bitkiye ilgi her geçen gün daha da artmaktadır. 5-6 yıl öncesine kadar Türkiye bu bitkiyi ithal ederken, bugün ihraç eden bir ülke konumuna gelmiştir. Beslenme değerinin yanı sıra olumlu sağlık etkileri ile de ön plana çıkmaktadır. Gıda sektörü son zamanlarda doğal kaynaklardan elde edilen bitkisel materyalleri kullanarak ürün geliştirme çalışmalarına ağırlık vermiştir. Üreticiler bu malzemelerin içerisinde bulunan bileşenlerin fonksiyonel özelliklerinden yararlanarak insan sağlığına uygun alternatif ürünler ortaya koymaya çalışmaktadırlar. Günden güne kinoaya ilginin artmasıyla insan ve hayvan beslenmesinde kullanımı yaygınlaşmıştır. Üzerine çalışmalar yapılması ve birçok gıda bileşimine katılması da popülerliğini arttırmıştır. Bu incelemede kinoanın besin değerleri, ülkemizdeki yeri ve gıdalarda kullanımının nasıl ve ne yönde etki ettiği hususunda, kinoa ve çeşitli özellikleri hakkında genel bilgiler vermek amaçlanmaktadır.

Anahtar Kelimeler: Kinoa, Besin Değeri, Gıdalarda Kullanımı

Quinoa And Uses In Food Industry

Abstract

Quinoa (Chenopodium quinoa Willd.) is a grain-like (pseudocereal) plant that has been traditionally consumed by South American cultures for thousands of years and today attracts attention as a functional food worldwide. As the interest in quinoa increases day by day, its use in human and animal nutrition has become widespread. Studies on it and its inclusion in many food compositions have increased its popularity. Interest in this plant, which is grown and tested in our country, is increasing day by day. While Turkey was importing this plant until 5-6 years ago, today it has become an exporting country. In addition to its nutritional value, it also stands out with its positive health effects. The food industry has recently focused on product development using plant materials obtained from natural sources. Manufacturers are trying to produce alternative products suitable for human health by taking advantage of the functional properties of the components contained in these materials. As the interest in quinoa increases day by day, its use in human and animal nutrition has become widespread. Studies on it and its inclusion in many food compositions have increased its popularity. In this review, it is aimed to give general information about quinoa and its various properties, its nutritional values, its place in our country, how and in what direction its use in foods affects.

Keywords: Quinoa, Nutritional Value, Properties, Use in Foods

DRAFT

DRAFT

GİRİŞ

Kinoa, Amerika kıtasının en eski mahsullerinden biridir. Kuzey Şili'deki arkeolojik bulgular, kinoanın MÖ 3000'den önce kullanıldığını göstermiştir. Ayacucho, Peru'da kinoanın MÖ 5000'den önce yetiştirildiğine dair kanıtlar elde edilmiştir (Jancurova, Minarovicova, & Dandar, 2009). Kinoa (*Chenopodium quinoa* Willd.) kökeni Güney Amerika olan ve çok farklı tipteki topraklarda ve iklimlerde tarımı yapılabilen, *Chenopodiaceae* familyasına ait tek yıllık bir bitkidir (Özgören & Yapar, 2022). Yetiştirilen ve toplanan Chenopodium türleri, Tiahuanacotan ve İnka kültürlerinin bir parçası olmuştur. Kinoa bu ata kültürlerde çeşitli roller üstlenmiş, insan ve hayvan beslenmesindeki rolünün yanı sıra kutsal bir öneme sahip olmuştur (Valencia, Mari, & Serna, 2011). Kinoa, 1–2 m. boyunda, derinlere nüfuz eden köklere sahip, geniş yapraklı bir bitkidir ve deniz seviyesinden 3800 m. yüksekliğe kadar yetiştirilebilir. Bitki donmaya, tuzluluğa ve kuraklığa tolerans gösterir ve alışılagelmişin dışındaki topraklarda gelişme kabiliyetine sahiptir. Kinoa tanesi, olağanüstü protein kalitesi, yüksek orandaki mineral ve vitamin mevcudiyeti sayesinde oldukça besleyicidir (Sevindik, Gültekin, & Uran, 2021). Farklı şekil, büyüklük, renk ve tane kompozisyonuna sahip kinoa çeşitleri bulunmakta ve kinoa türlerinin sınıflandırılmasında genellikle beyazdan siyaha değişen renk karakteristiklerinden yararlanılmaktadır. Kinoa türlerinin renklerindeki bu farklılık kinoa yapısında yer alan betalainlere atfedilmektedir (Alaşalvar, Erinç, & Çolakoğlu, 2019). Kinoa ıspanakgiller familyasından olup strese dayanıklı bir bitkidir. Botanik yapısı nedeniyle yalancı tahıl olarak da adlandırılmaktadır (Yağan, 2019).

Tahıl ana olarak adlandırılan kinoa son derece besleyicidir. Kinoa tohumu protein, kalsiyum, magnezyum, demir, çinko gibi mineraller, diyet lifi ile E ve B grubu vitaminlerince nispeten iyi bir kaynaktır. Ayrıca esansiyel aminoasitlerin tamamını içeren kinoa lisin, sistein ve methionin aminoasitleri acısından da zengin olduğundan çok iyi bir protein kaynağı olarak kabul edilmektedir. Buğday, çavdar, yulaf, darı, mısır ve pirinçten çok daha fazla protein içermektedir. Yağ oranı ise yağlı tohumlardan az ancak tahıllardan daha yüksektir. Kinoa gluten içermediği için glutene duyarlılığı olan çölyak hastaları ve veganların (hayvansal ürün yemeyen) protein ve karbonhidrat ihtiyaçlarını karşılayan besleyici bir besindir. Bunların yanı sıra kinoa kolesterol de içermemektedir (Keskin & Kaplan Evlice, 2015). Kinoa modern dünyanın gündemine 2000'li yıllarda gelmiştir. Kinoanın popüler olmasında iki önemli gelişme rol oynamıştır. Bunlardan birincisi Birleşmiş Milletler Gıda ve Tarım Örgütü (FAO)'nün 2013 yılını kinoa yılı ilan etmesidir. Teşkilat bu uygulama ile kinoanın dünyada gıda sıkıntısı yaşayan toplumlar için bir umut ışığı olduğuna dikkat çekmek istemiştir. Kinoanın tanınmasına yardımcı olan diğer bir gelişme ise NASA tarafından astronotların beslenmesinde kullanılmaya başlanmasıdır. Bitkinin tohumları küçük hacimde yüksek besleme değerine sahiptir. Bu nedenle uzay araştırmalarına da konu olmaktadır. Bu iki önemli gelişme kinoaya olan ilgiyi artırmış, dünya üzerindeki ticaret hacmi yükselmiştir. Ülkemizde de yetiştiriciliği ve denemeleri yapılmakta olan bu bitkiye ilgi her geçen gün daha da artmaktadır. 5-6 yıl öncesine kadar Türkiye bu bitkiyi ithal ederken, bugün ihraç eden bir ülke konumuna gelmiştir (Tan & Temel, 2019). Gıda sektörü son zamanlarda doğal kaynaklardan elde edilen bitkisel materyalleri kullanarak ürün geliştirme çalışmalarına ağırlık vermiştir. Üreticiler bu malzemelerin içerisinde bulunan bileşenlerin fonksiyonel özelliklerinden yararlanarak insan sağlığına uygun alternatif ürünler ortaya koymaya çalışmaktadırlar. Özellikle çeşitli tohumlar veya bunlardan elde edilen proteinler ve karbonhidratlar bunlara örnek olarak gösterilebilirler. Bu malzemelerin kullanımı ile sağlıklı beslenmeye uygun ürünler ortaya koymanın yanı sıra, gıdalarda ortaya çıkabilen duyusal ve kimyasal problemlerin de önüne geçilmeye çalışılmaktadır (Meral & Kılınççeker, 2022). Bu çalışma kinoanın besin değerleri, ülkemizdeki yeri ve gıdalarda kullanımının nasıl ve ne yönde etki ettiği hususunda bilgi vermeyi amaçlamaktadır.

Kinoa'nın Ülkemizdeki Yeri

Ülkemizde ise kinoa üretimi, 2010 yılında bu ürünü ilk kez duymuş çiftçiler tarafından başlatılmıştır. Bu süre zarfında üniversiteler ve araştırma enstitüleri tarafından yürütülen araştırmalar ile kinoanın ülkemiz

koşullarına adapte olan çeşitleri belirlenmeye, verim odaklı bitki besleme programları ve ekim dönemleri tespit edilmeye başlanmıştır. 2015 yılına gelindiğinde aradan geçen 4 yıllık süreçte, kinoa üretimini başarıyla sonuçlandıran profesyonel üreticiler ortaya çıkmaya başlamıştır ve 2015 yılından itibaren, ticari boyutta üretim modellerine geçilmeye başlanmıştır (Bayram, Pekacar, & Deliorman Orhan, 2018).

Kinoa'nın Besin Değeri

Kinoa bitkisinin içerisinde bulunan protein; özellikle histidin (%3,2) ve lisin (%6,1) yönünden zengindir. Yağının yüksek kalitesi sayesinde ve bazı çeşitlerinin % 9,5'e kadar yağ konsantrasyonlarına sahip olmasından dolayı; potansiyel olarak değerli yeni bir yağ maddesi olarak ta görülmektedir. Kinoa bitkisi 1 kg kuru ağırlık bazında, diğer tahıllara kıyasla daha fazla Ca (1487 mg), Fe (132 mg), K (9267 mg), Mg (2496 mg), Cu (51 mg), Mn (100 mg) ve Cl (1533 mg) barındırmaktadır ve Na:K oranı 1:76 çıkmaktadır. RDA(önerilen günlük besin alım miktarı)'lar yani tavsiye edilen diyet içeriği bakımından ve yaş ve cinsiyete bağlı olarak 100 g yenilebilir kinoa porsiyonu %27-40 Fe, %23-76 Mg, %47-200 Cu, %11-16 P, %15-19 K, %10-15 Zn ve sadece %1-2 Na sağlamaktadır. Kinoanın içerisinden ayrılması gereken madde saponinlerdir. Yıkama ya da aşındırıcı kabuk ayırma yoluyla çıkarılabilir; 100 g yenilebilir porsiyonda 0,01 g'dan fazla saponin olamaz (Yağan, 2019).

Kinoadaki ana bileşen karbonhidratlardan oluşur ve kuru maddenin %67 ila %74'ü arasında değişir. Nişasta yaklaşık %52-60 yapar. Nişasta bileşiği tohumlarının perisperminde bulunur. Nişasta, basit birimler veya küresel agretatlar olarak mevcut olabilir. Amiloz içeriği, pirinç (%17), buğday (%22) veya arpa (%26) gibi tahıllardan daha düşük olarak yaklaşık %11'dir. Kinoa %2 ile %10 arasında yağ içerir. Kinoa ve soya yağları, benzer yağ asidi bileşimleri sergiler; bu nedenle kinoa, linolenik ve linolenik gibi esansiyel yağ açısından zengin bir kaynaktır (Jancurova, Minarovicova, & Dandar, 2009).

KİNOA'NIN FİZİKSEL ÖZELLİKLERİ

Tohumların fiziksel, gravimetrik, sürtünme ve aerodinamik özellikleri, temizleme, sınıflandırma, taşıma, havalandırma, kurutma ve depolama gibi farklı hasat sonrası işlemlerin tasarlanmasında faydalıdır. %4,6 ile %25,8 (kuru bazda) arasında değişen nem içeriğine sahip kinoa tohumu numunelerinin bu özelliklerini belirlemiştir. %15'lik bir ortalama nem içeriğine sahip kinoa tohumunun boyu 1,7 ila 2 mm arasında değişirken, %72 tanede %5'lik tane bulunurken, %27,4'lük tohum boyutu Tablo 1'de verilen 2 mm'den büyük olmuştur. Diğer Özellikler Aşağıdaki gibi tablolaştırılmıştır (Sharma, Chandra, Dwivedi, & Parturkar, 2015).

Tablo 1: Kinoa Tohumlarının Fiziksel Özellikleri

Özellik		Nem %4,6 d.b.	Nem %25 d.b.
Gravmetrik	1000 Tohum Ağırlığı	2,53 gr	3,11 gr
	Gerçek Yoğunluk	928 kg m^{-3}	1188 kg m^{-3}
	Kütle Yoğunluğu	747 kg m^{-3}	667 kg m^{-3}
	Yatak Gözenekliliği	0.19	0.44
Sürtünme	Durma Açısı Statik	18 ^{0}	25 ^{0}
	Sürtünme Katsayısı	0,14	0,27
Aerodinamik	Terminal Hızı	0,6 ms^{-1}	1,02 ms^{-1}

DRAFT

KİNOA'NIN KİMYASAL VE BESİN ÖZELLİKLERİ

Tablo 2: Kinoa Tohumlarının Kimyasal Bileşimi

Diğer Tahılların Karşılaştırılması				
Enerji Değeri	Kinoa	Buğday	Pirinç	Mısır
Kcal/100g	350,00	309,00	353,00	338,00
Protein/100g	13,81	11,50	7,40	9,20
Yağ/100g	5,01	2,00	2,20	3,80
Karbonhidratlar mg/100g	59,74	59,40	74,60	65,20
Kalsiyum mg/100g	66,60	43,70	23,00	15,00
Fosfor mg/100g	408,30	406,00	325,00	256,20
Magnezyum mg/100g	204,20	147,00	157,20	120,00
Potasyum mg/100g	1040,00	502,00	150,00	330,00
Demir mg/100g	10,90	3,30	2,60	-
Manganez mg/100g	2,47	3,40	1,10	0,48
Çinko mg/100g	7,47	4,10	-	2,50

Rezervlerin yakın bileşimi, proteinlerin kimyasal karakterizasyonu, yağların yağ asidi bileşimi, mineral içeriği ve fonksiyonel ve besin değerleri gibi hususları kapsayan kinoa hakkında kapsamlı literatür mevcuttur. And bölgesinde yetiştirilen dört farklı kinoa çeşidinin yakın bileşimi, yani. Blanca de Juli, Kancolle, La Molina 89 ve Sajama Valencia tarafından belirlenmiş olup,(2011) analiz sonuçları Tablo 3'te sunulmuştur (Jancurova, Minarovicova, & Dandar, 2009).

Tablo 3: Dört Kinoa Çeşidinin Yaklaşık Bileşimi (% kuru bazda)

Bileşen	Blanca de Juli	Kcancolla	La Molina 89	Sajama
Nem	11,39	10,78	12,03	12,62
Kül	3,38	3,52	5,46	3,04
Protein	13,96	15,17	15,47	14,53
Doymuş Yağ	5,51	5,77	6,85	4,69
Ham Lif	2,00	3,07	3,38	1,92
Karbonhidratlar	75,15	72,47	68,84	75,82

Ekstrüzyonla pişirme yöntemiyle kinoadan üretilen hazır unun yaklaşık bileşimi, bileşimi şu şekilde göstermiştir: nem %4,8, protein %12,2, lipitler %5,6, kül %2,3, toplam karbonhidrat %74,9 ve lif %4,1 (Jancurova, Minarovicova, & Dandar, 2009). Kinoa unu, düşük prolamin ve glutamin içeriği nedeniyle glüten açısından düşüktür. Genellikle bisküvi, erişte ve hamur işlerinin hazırlanmasında pişirme unlarını zenginleştirmek ve fırınlanmış gıdaların hazırlanmasında nemi korumak ve hoş bir tat vermek için kullanılır. Kinoa ve diğer temel gıdaların besin içerikleri Tablo 4'te karşılaştırılmıştır (Jancurova, Minarovicova, & Dandar, 2009).

Tablo 4: Diğer Temel Gıdalarla Karşılaştırıldığında Kinoanın Besinsel Bileşimi (%)

Bileşenler	Kinoa	Et	Yumurtalar	Peynir	İnek Sütü	İnsan Sütü
Proteinler	3,4 ve	30,00	14,00	18,00	3,50	1,80
Yağlar	6,10	50,00	3,20	-	3,50	3,50
Karbonhidratlar	71,00	-	-	-	-	-
Şeker	-	-	-	-	4,70	7,50
Demir	5,20	2,20	3,20	-	2,50	-
100 g başına kalori	350,00	431,00	200,00	24,00	60,00	80,00

Besleyici bileşenlerin yanı sıra kinoa tohumu perikarpı, toksik ve acı tadı olan bileşenler olan saponinler içerir ve bu da, gıda ürünlerinin üretimi için yemeden veya işlenmeden önce ortadan kaldırılmasını gerekli kılar. Biyopolimerler tanelerin belirli kısımlarında da bulunur, nişasta perisperm hücrelerini işgal ederken, lipit gövdeleri, fitinin globoid kristallerine sahip protein gövdeleri ve fitoferritin birikintileri olan proplastitler endosperm ve embriyo dokularının depolama bileşenleridir (Jancurova, Minarovicova, & Dandar, 2009).

KİNOA'NIN PROTEİN İÇERİĞİ

Kinoanın protein miktarı ve kalitesi genel olarak tahıl tanelerinden daha üstün olup, glütensiz olma özelliği ve yüksek sindirilebilirlik sunar. Kinoanın toplam protein içeriği (%12,9 ila %16,5), arpa (%10,8 ila %11,0), yulaf (%11,6), pirinç (%7,5 ila %9,1) ve mısırdan (%10,2 ila %13,4) ve toplam protein içeriğine eşit buğday (%14,3 ila %15,4). Kinoanın depolama proteinleri çoğunlukla globulin ve albüminden oluşur ve pek çok tahıl ürününde ana depolama proteinleri olan prolaminler çok az bulunur veya hiç bulunmaz. Buğdaydan gliadin, çavdardan sekalin ve arpadan hordein (toplu olarak "glutenler" olarak anılır) gibi prolaminler, çölyak hastalarında otoimmün tepkileri indükler (Jancurova, Minarovicova, & Dandar, 2009). 15 kinoa çeşidi üzerinde yakın zamanda yapılan bir in vitro çalışma, yalnızca 2 çeşidin çölyak için toksik prolamin epitoplarının herhangi bir saptanabilir seviyesini göstermiştir. Bağışıklık tepkileri ise in vitro tam gıda tüketimi ile kopyalanmamıştır. Bu sonuçlar, kinoanın tahıl taneleri için güvenli bir glütensiz ikame olduğunu göstermektedir. toplam proteininin %37'sini, FAO (Abugoch James) tarafından lösin, izolösin ve fenilalanin ve tirozin için bir referans kaynağı haline gelen globulin 11S tipi bir protein olan chenopodin oluşturur (Jancurova, Minarovicova, & Dandar, 2009).

Literatürde kinoa tohumu için bildirilen ortalama protein içeriği kuru bazda (db) %16,3'tür. Kinoa tohumlarının protein içeriği arpa (%11 db), pirinç (%7,5 db) veya mısırdan (%13,4 db) daha yüksektir ve buğdayınkiyle (%15,4 db) karşılaştırılabilir. Kinoa tohumu nispeten daha az protein içerir. baklagil tohumlarına kıyasla proteinler (Tablo 5). Kinoa proteininin besin değeri süt proteininkiyle karşılaştırılabilir. Acısı alınmış ham kinoadaki protein verimlilik oranı (PER), kazeininkinin %78-93'üdür. Bu rakamlar kinoa pişirildiğinde artmakta ve kazeinin %102-105'i olmaktadır (Jancurova, Minarovicova, & Dandar, 2009).

Tablo 5: Kinoa ve Bazı Tahıl ve Baklagillerin Kimyasal Bileşimi (g/100g kuru ağırlık)

Bileşenler	Kinoa	Arpa	Mısır	Pirinç	Buğday	Fasulye	Acı Bakla	Soya
Protein	16,5	10,8	10,22	7,6	14,3	28,0	39,1	36,1
Yağ	6,3	1,9	4,7	2,2	2,3	1,1	7,0	18,9
Lif	3,8	4,4	2,3	6,4	2,8	5,0	14,6	5,6
Kül	3,8	2,2	11,7	3,4	2,2	4,7	4,0	5,3
Karbonhidratlar	69,0	80,7	81,1	80,4	78,4	61,2	35,3	34,1
kcal/100g	399	383	408	372	392	367	361	451

Lizin, çoğu tahıl tanesinde sınırlayıcı bir amino asittir. Kinoanın amino asit bileşimi Koziol (1992) tarafından incelenmiştir, kinoa tohum proteinindeki esansiyel amino asit dengesi, tahıllar ve baklagil proteinlerinden daha yüksek lizin (%5.1-6.4) ve metiyonin (%0.4-1) içeriği nedeniyle mükemmeldir. Kinoa proteinleri (QP), mısır, pirinç ve buğday proteinlerinden daha yüksek histidin içeriğine sahiptir. (Tablo 6) (Jancurova, Minarovicova, & Dandar, 2009).

Kinoanın metiyonin artı sistin içeriği de çocuklar (2-12 yaş) ve yetişkinler için yeterlidir. FAO/WHO'nun 10 yaşındaki çocuklar için önerdiği gereksinimlere göre, kinoa proteini yeterli düzeyde aromatik amino asitlere (fenilalanin ve tirozin) ve benzer şekilde histidin, izolösin, treonin, fenilalanin, tirozin ve valin içeriğine sahiptir. Karşılaştırıldığında, QP'lerdeki lizin ve lösin, 2-5 yaşındaki bebekler veya çocuklar için amino asitleri sınırlandırırken, FAO/WHO'ya göre bu proteinin tüm temel amino asitleri yeterlidir (Tablo 6) (Jancurova, Minarovicova, & Dandar, 2009).

Tablo 6: Kinoa ve Diğer Besinlerdeki Esansiyel Amino Asitler (g/100 g protein)

Amino Asitler	Kinoa	Mısır	Pirinç	Buğday	Fasulye	Süt	FAO[a]
Histidin	3,2	2,6	2,1	2,0	3,1	2,7	2,6
İzolösin	4,9	4,0	4,1	4,2	4,5	10,0	4,6
Lösin	6,6	12,5	8,2	6,8	8,1	6,5	9,3
Lizin	6,0	2,9	3,8	2,6	7,0	7,9	6,6
Metiyonineb	5,3	4,0	3,6	3,7	1,2	2,5	4,2
Fenilalaninek	6,9	8,6	10,5	8,2	5,4	1,4	7,2
Treonin	3,7	3,8	3,8	2,8	3,9	4,7	4,3
Triptofan	0,9	0,7	1,1	1,2	1,1	1,4	1,7
Valline	4,5	5,0	6,1	4,4	5,0	7,0	5,5

KİNOA'NIN KARBONHİDRAT İÇERİĞİ

Karbonhidratlar, kinoa tohumunun kuru maddesinin başlıca kısımlarıdır; ana bileşenler olarak nişasta ve diyet lifleri içerir. Kinoa tohumlarındaki karbonhidrat içeriği, kuru maddenin %67 ila %74'ü arasında değişir; bunun dışında nişasta yaklaşık %55-65'ini oluşturur. Kinoa tohumlarında başlıca nişasta bileşiği, perispermde basit birimler veya 3 µm'den küçük çok küçük tane boyutuna sahip küresel kümeler halinde bulunur. Amiloz içeriği ile karşılaştırıldığında kinoa nişastası, pirinç

(%17), buğday (%22) veya arpa (%26) gibi diğer tahıllardan daha az olan %11 amiloz içerir (Jancurova, Minarovicova, & Dandar, 2009).

Lamothe ve diğerleri, (2015)'e göre kinoa, %10 toplam diyet lifi içerir. Lif, ince bağırsakta enzimatik sindirime ve emilime dirençli, kalın bağırsakta genellikle tam veya kısmi fermantasyona uğrayan karbonhidrat fraksiyonudur. Diyet lifi, optimal sindirim sağlığı için gerekli kabul edilir ve ayrıca çeşitli fonksiyonel faydalar sağlar (Jancurova, Minarovicova, & Dandar, 2009). Kinoa nişastasının jelatinleşme sıcaklığı nispeten düşüktür, 57-64°C'dir. Kinoa nişastası, buğday nişastasından daha yüksek viskozite sergiler, ancak mısır nişastası kadar yüksek değildir. Mumsu mısır nişastası gibi kinoa nişastası da normal mısır nişastasına göre üstün donma-çözülme stabilitesine sahiptir. Bulunan diğer karbonhidratlar pentosanlar (%2,9-3,6), disakkaritler (%2,3), ham lif (%2,5-3,9) ve monosakkaritlerdir (Jancurova, Minarovicova, & Dandar, 2009).

KİNOA'NIN YAĞ İÇERİĞİ

Kinoa %2 ila %9 yağ içerir, ortalama yağ içeriği yaklaşık %6 USDA'dır, sahte yağ ürünü olarak kabul edilebilir. Bu yüzdeler kabaca mısırın yağ içeriğine benzer, ancak soya fasulyesinden (%20-25 yağ) çok daha azdır. Kinoa, Mısır ve Soya yağları benzer yağ asidi bileşimleri sergiler. Soya fasulyesi ve çoğu tahılda olduğu gibi, linoleik asit baskın yağ asididir, benzer şekilde kinoa, linolenik (18:2n-6: %52) ve linolenik (18:3n-6: %40) gibi zengin bir esansiyel yağ asitleri kaynağıdır. Tüm kinoa tohumlarının genel yağ asidi bileşimi, mevcut ana asitler olarak linoleik, oleik ve palmitik asitlerle diğer tahıl tanelerine benzerdir. Tablo 7'de farklı tahıllardaki yağ asidi kompozisyonları verilmiştir (Jancurova, Minarovicova, & Dandar, 2009).

Tablo 7: Kinoa ve Genel Tahıl Tanelerinin Yağ Kalitesi (Değerler Toplam Yağın Yüzdesi Olarak İfade Edilmiştir)

Yağ Asitleri	Kinoa	Buğday	Arpa	Pirinç	Mısır
Baskın Asit	18:2	18:2	18:2	18:1	18:2
Doymuş	13%	23%	26%	32%	16%
Tekli Doymamış	34%	22%	16%	37%	31%
Çoklu Doymamış	53%	55%	58%	31%	53%

Kinoa tohumundan ve tohum fraksiyonlarından izole edilen lipidler, Przybylski tarafından lipit sınıfları ve yağ asidi kompozisyonu için karakterize edildi, analiz edildiğinde, kinoa tohumu lipitlerinin diğer tohum fraksiyonları arasında en fazla miktarda nötr lipit içerdiği, tüm kinoa tohumunda daha yüksek miktarlarda SYA tespit edildiği ve toplam lipidlerin sırasıyla %18.9'u ve %15.4'ü kabuğunda bulunduğu, nötr lipitlerin %50'si trigliseridler; digliseritler, nötr lipit fraksiyonlarının %20'sine katkı sağladığı ve tüm tohumda mevcut olduğu gözlenmiştir (Jancurova, Minarovicova, & Dandar, 2009). Fosfolipitler arasında lizofosfatidil etanolamin bütün tohum fosfolipidlerinin %45'ini ve fosfatidil kolin %12'sini oluşturmuştur (Sharma, Chandra, Dwivedi, & Parturkar, 2015).

KİNOA İÇİNDEKİ MİNERALLER

Kinoa iyi bir mineral kaynağıdır; Arpa, yulaf, pirinç, mısır veya buğday gibi yaygın temel gıdalar ile karşılaştırıldığında, kinoa tohumları yüksek kaliteli proteinler, daha yüksek seviyelerde enerji, kalsiyum, fosfor, demir, lif ve B vitaminleri içerir. Kinoa tohumlarının mineral içeriğine ilişkin

karşılaştırmalı veriler Tablo 8'de sunulmaktadır (Jancurova, Minarovicova, & Dandar, 2009). Daha yüksek miktarlarda kalsiyum (874 mg kg-1), fosfor (5,3 g kg-1), magnezyum (2,6 mg/100 g), demir (81 mg kg-1), çinko (36 mgkg-1) , potasyum (12) g kg-1) ve bakır (10 mg kg-1) yaygın tahıl tanelerinin çoğundan daha fazla olduğu, tohumların cilalanması ve yıkanması mineral içeriğini bir dereceye kadar azalttığı, demir, çinko ve potasyumda %12-15, bakırda %27 ve magnezyumda %3 oranlarında azalma olduğu saptanmıştır. (Sharma, Chandra, Dwivedi, & Parturkar, 2015).

Tablo 8: Kinoa ve Bazı Tahıllarda Mineral Bileşimi (mg kg-1 kuru ağırlık)

Mineraller	Kinoa	Buğday	Pirinç	Arpa
Kalsiyum	1487	503	69	430
Magnezyum	2496	1694	735	1291
Potasyum	9267	5783	1183	5028
Fosfor	3837	4677	1378	3873
Demir	132	38	7	32
Bakır	51	7	2	3
Çinko	44	47	6	35

KİNOA'NIN VİTAMİN İÇERİĞİ

A vitamini, E vitamini gibi yağda çözünen vitaminler ve tiamin, riboflavin, niasin ve askorbik asit gibi suda çözünen vitaminlerin aralığı Ruales ve diğerleri (1992) tarafından ölçülmüş ve Tablo 9'da ifade edilmiştir. Kinoa iyi bir kaynak olarak kabul edilir. Tiamin (0,4 mg/100 g), folik asit (78,1 mg/100 g) ve C vitamini (16,4 mg/100 g) gibi vitaminler içermektedir. Saponinlerin tohumlardan uzaklaştırılması işlemi, vitamin ve mineral içeriklerini bir dereceye kadar azaltmaktadır. Potasyum, demir ve manganez durumunda kayıp önemlidir ($P < 0.001$) (Jancurova, Minarovicova, & Dandar, 2009).

Tablo 9: Kinoa Tanesinin Vitamin İçeriği (mg/100g kuru madde)

S. No.	Vitaminler	Aralık
1.	A Vitamini (Karoten)	0,12 - 0,53
2.	E Vitamini	4,60 - 5,90
3.	Tiamin	0,05 - 0,60
4.	Riboflavin	0,20 - 0,46
5.	Niasin	0,16 - 1,60
6.	Askorbik Asit	0,00 - 8,50

DRAFT

KİNOA'NIN FONKSİYONEL ÖZELLİKLERİ

Antioksidan Özelliği: Seçilmiş sözde tahıllar amaranth ve kinoa tohumlarının ve filizlerinin antioksidan potansiyeli, Plazmanın Ferrik İndirgeme Yeteneği (FRAP) testi, radikal süpürme aktivitesi (ABTS) ve radikal süpürme aktivitesi (DPPH) gibi yöntemlerle yapıldı. Toplam polifenoller ve antosiyaninler de belirlendi. Bu çalışmada, kinoada bulunan iyi bir antioksidan kaynağı olarak filizin besin değerine odaklanılmıştır. Filiz değerleri de ölçülmüş olup, kinoa tohumlarının ve filizlerinin önemli ölçüde yüksek antioksidan aktivite gösterdiği saptanmıştır (Jancurova, Minarovicova, & Dandar, 2009).
Araştırma sonucunda, filizlerin tohumlardan önemli ölçüde daha yüksek bir antioksidan aktiviteye sahip olduğunu göstermiştir. Bu polifenoller, antosiyaninler ve diğer bileşiklerin içeriğindeki farklılığın bir sonucu olabilir ve Amarant ve kinoa filizleri, "yeni" sebzelerdir; veganların ve vejeteryanların beslenmesinde ortak bir diyet olarak da kullanılabileceği anlaşılmıştır (Jancurova, Minarovicova, & Dandar, 2009).

Antimikrobiyal Özellik: Şili'nin üç farklı coğrafi bölgesinden (her bölgeden iki adet) altı farklı kinoa tohumu çeşidi (Ancovinto, Cancosa, Cahuil, Faro, Regalona ve Villarrica) antimikrobiyal özellikleri açısından incelenmiştir. Tohumun kuru etanol ekstraktı suda çözüldü (30.0 mg ekstrakt/mL damıtılmış su) ve disk difüzyon testi tekniği kullanılarak Staphylococcus aureus (ATCC 25923) (Gram pozitif) ve Escherichia coli (ATCC 25922) (Gram negatif) olmak üzere iki mikroorganizmaya karşı inhibisyon bölgesini test etmek için kullanılır. Tüm kinoa örneklerinin ekstraktları, E. coli için 8,3-14,8 mm inhibisyon bölgesi ve S. aureus için 8,5-15,0 mm inhibisyon bölgesi aralığında antimikrobiyal aktivite göstermiştir. Cancosa tohumları en yüksek antimikrobiyal etkiye sahipti (Jancurova, Minarovicova, & Dandar, 2009).

Altı çeşidin tamamında Toplam Fenolik İçerik (TPC), Toplam Flavonoid İçeriği (TFC), Toplam Saponin İçeriği (TSC) ve inhibisyon bölgesi arasındaki Pearson katsayısı korelasyonları yapılmıştır. TFC ile E. coli ve S. aureus'a karşı antimikrobiyal aktiviteler arasındaki ilişki orta(sırasıyla $r = 0.60$ ve 0.43) ancak E. coli için pozitif olarak anlamlı ($p < 0.05$) bulundu , bu da flavonoid bileşiklerin antimikrobiyal aktivitelere katkıda bulunabileceğini gösteriyor E. coli ve S. aureus'a karşı (Jancurova, Minarovicova, & Dandar, 2009).

KİNOA'NIN GIDALARDA KULLANIMI

Makarna

Makarnanın zenginleştirilmesi amacıyla yürütülen çalışmalarda makarnanın ana bileşeni olan durum buğdayının bileşiminde bulunmayan ya da az miktarda bulunan besleyici/fonksiyonel elementlerinin takviyesi hedeflenerek, makarna formülasyonlarına bitkisel ya da hayvansal kaynaklı ilaveler yapılmaktadır. Bitkisel kaynaklı olarak farklı tahıl, pseudo-tahıl(kinoa, amarant, karabuğday vb.) ve baklagil unları ile çeşitli kaynaklardan elde edilen meyve sebze lifleri ve posaları kullanılabilmektedir (Demir, 2018).

Pseudo-tahıllar yalancı tahıl (tahıl benzeri) olarak adlandırılmakta, gluten içermediğinden çölyak hastalarının beslenmesi için de alternatif hammadde olarak düşünülmektedir. Bunlar arasında karabuğday, kinoa ve amarant en fazla dikkat çekenler olup son zamanlarda gıda formülasyonlarında yaygın olarak kullanılmaya başlanmışlardır (Demir, 2018).

Erişte

% 30 kame oranı esas alınarak erişte formülasyonuna, 10 farklı kombinasyonda pseudo-tahıl (amarant, kinoa, karabuğday) ikameleri gerçekleştirilmiştir. Üretilen erişte örneklerinde bazı fiziksel kimyasal, besinsel ve duyusal özellikler incelenmiştir. Erişte kombinasyonlarına pseudo-tahıl ikamesi ile son ürünlerin L^* değeri azalırken a^* ve b^* değeri artmıştır. Ayrıca ikameler ile eriştenin mineral madde, toplam fenolik madde, fitik asit, ham protein, ham yağ ve kül miktarlarında şahit (%100 buğday unu ile yapılan erişte) erişte örneğine göre artışlar tespit edilmiştir. Tüm değerlendirme kriterleri göz önünde bulundurularak %20 amarant + %10 kinoa ikameli 5 numaralı erişte kombinasyonunun en uygun kombinasyon olduğu sonucuna varılmıştır (Öncel, 2017). Erişte örneklerindeki kinoa unu oranlarının artışı, duyusal olarak görünüş, doku ve lezzet ortalama puanlarının azaltmıştır. %20 oranında kinoa unu kullanılarak elde edilen erişte, en iyi örnek olarak belirlenmiştir (Argan, 2019).

Bisküvi

Bu çalışmada farklı boyutlarda kinoa kepeği kullanılarak, laboratuvar ortamında bisküvi üretimi yapılmış ve farklı boyutlarda kinoa kepeğinin bisküvilerin fiziksel, tekstürel ve duyusal özelliklerine olan etkisi incelenmiştir. Elde edilen verilere göre, kinoa kepek unu boyutuna bağlı olarak bisküvilerin karakteristiklerinde önemli değişimler meydana getirmektedir. Çalışma sonucunda kinoa kepeğinin sağlıklı beslenen bireyler için yeni bir alternatif hammadde olarak bisküvi üretiminde kullanılabileceği sonucuna varılmıştır (Erinç, 2020).

Kek

Farklı boyutlarda kinoa kepek unları değirmende öğütülmüş ve 5 farklı boyutta buğday ununda yapılmış keklere %25 oranında katılmıştır. Elde edilen sonuçlara göre kinoa kepek ununun boyutları küçüldükçe örneklerin kuru madde oranı artış göstermiştir. Yine kepek unu boyutlarının küçülmesi ile kek iç ve dış parlaklık değerlerinde azalma gözlenmiştir. Keklerin tekstür ve duyusal özelliklerinde kinoa katılımı sonucun düşüş belirlenmiştir ve bu olumsuzluk kepek boyutu küçüldükçe daha fazla olmuştur. Elde edilen verilere göre, kinoa kepek unu boyutuna bağlı olarak keklerin karakteristiklerinde değişimler meydana getirmektedir (Alaşalvar, Erinç, & Çolakoğlu, 2019).

Muffin Kek

%100 buğday ununda üretilen kontrol grubu örnek ve buğday ununa farklı oranlarda (%25 ve 50) kinoa unu ikame edilmesi ile üretilen muffin keklerde meydana gelen fizikokimyasal ve duyusal değişimler araştırılmıştır. Muffin kek formülasyonunda kinoa unu ikame oranı arttıkça örneklerin protein, yağ, kül ve amino asit içerikleri ile tekstür profil analizi sonucunda elde edilen sertlik değerlerinde artış meydana gelirken, karbonhidrat içeriğinde ve spesifik hacimde azalma meydana gelmiştir. Duyusal özellikler bakımından %50 oranında kinoa unu ikame edilmiş örnekte gözeneklilik, tekstür, tat ve genel beğeni puanlarında anlamlı düzeyde azalma ($p<0.05$) meydana gelmiştir. Kinoa ikame edilme oranı %25 olan muffin kek örneğinin ise kontrol örneği ile benzer duyusal ve tekstürel özelliklere sahip olduğu belirlenmiştir. Tüm sonuçlar göz önüne alındığında, kinoanın protein ve yağ gibi önemli gıda bileşenlerini yüksek oranda bulundurması ve zengin amino asit içeriğine sahip olması nedeniyle iyi bir zenginleştirme materyali olarak kullanılabileceği belirlenmiştir (Özgören & Yapar, 2022).

Yapılan bu çalışma ile kinoa ununun buğday ununa kıyasla daha yüksek protein, kül ve yağ içeriğine sahip olduğu belirlenmiştir. Bunun yanında dengeli ve yüksek oranda amino asit içeriğine sahip olması sayesinde özellikle unlu mamullerde iyi bir zenginleştirme kaynağı olarak kullanılabileceği kanaatine varılmıştır (Özgören & Yapar, 2022). Kinoa unu ikame edilmiş muffin kek örneklerinin yalnızca buğday ununda üretilen örneklere kıyasla gıda bileşenleri açısından daha zengin olduğu belirlenmiştir. Kinoa unu ikame oranının artışı belli bir seviyeye kadar kalite özelliklerini olumsuz etkilememiştir. %25 oranında kinoa unu ikame edilmiş muffin kek örneği duyusal ve tekstürel özellikler açısından kontrol örneği ile benzer bulunmuştur. Ayrıca protein, yağ, kül miktarı ve amino asit kompozisyonu açısından kontrol örneğine kıyasla daha zengin bir içeriğe sahip olduğu belirlenmiştir. Kinoa unu ikame edilen örneklerin kalori değerinde ise belirgin bir değişim

tespit edilmemiştir (Özgören & Yapar, 2022). Tüm bu nedenlerden dolayı %25 oranına kadar kinoa unu ikamesinin muffin keklerin kalite özelliklerinde olumsuz etkiye neden olmadan kullanılabileceği belirlenmiştir (Özgören & Yapar, 2022).

Probiyotik Yoğurt

Probiyotik yoğurtlarda kinoa ununun prebiyotik katkı olarak kullanım olanakları ve farklı oranlarda kinoa unu ilavesinin probiyotik yoğurtların fizikokimyasal, tekstürel, mikrobiyolojik ve duyusal özellikleri üzerine etkisi araştırılmıştır. Farklı oranlarda kinoa unu (A:kontrol(%0.0), B:%0.5, C:%1.0, D:%2.0 ve E:%4.0) ilave edilerek üretilen probiyotik yoğurtlar 4±2°C'de 21 gün süreyle depolanmıştır. Depolamanın 1., 11. ve 21. günlerinde yoğurtların fizikokimyasal, tekstürel, mikrobiyolojik ve duyusal özelliklerini belirlemek amacıyla analizler yapılmıştır (Hafif, 2019). Elde edilen analiz sonuçlarına göre probiyotik yoğurt örneklerine kinoa unu ilavesinin fizikokimyasal özelliklerden kuru madde, titrasyon asitliği, pH, kül, protein, yağsız kuru madde, serum ayrılması, viskozite, su tutma kapasitesi değerleri; tekstürel özelliklerden sertlik, konsistens(kıvam), iç yapışkanlık ve viskozite indeksi değerleri; mikrobiyolojik özelliklerden *Lactobacillus acidophilus, Streptococcus thermophilus* ve *Bifidobacterium animals subsp. lactis (BB-12)* sayıları; duyusal özelliklerden renk ve görünüş, yapı ve kıvam, tat ve aroma puanları üzerine etkisi önemli ($p<0.01$) bulunmuştur. Depolama süresinin de probiyotik yoğurtların bu özellikleri üzerine etkisi benzerlik göstermiştir (Hafif, 2019).

Farklı oranlarda kinoa unu ilavesinin probiyotik yoğurtlarda prebiyotik etkisinin araştırıldığı bu çalışmada, mikrobiyolojik özellikler değerlendirildiğinde kinoa ununun prebiyotik etki gösterdiği görülmüştür. Ayrıca fizikokimyasal, tekstürel ve duyusal özellikler değerlendirildiğinde kinoa ununun probiyotik yoğurtlara ilavesinin bu özelliklere olumlu katkılar yaptığı ve özellikle %0.5 ile %1.0 kinoa unu ilavesinin probiyotik yoğurtlarda rahatlıkla kullanılabileceği kanısına varılmıştır. Yüksek oranlarda (%2.0 ve %4.0) kinoa unu ilavesinin probiyotik yoğurtların duyusal özelliklerini olumsuz etkilemesi sebebiyle bu oranlardaki (%2.0 ve %4.0)kinoa ununun meyveli/aromalı yoğurtlarda denenebileceği düşünülmektedir (Hafif, 2019).

Fırın Ürünleri

Çeşide bağlı olarak değişmekle birlikte, yaklaşık % 10 - 18 protein, % 4.50 - 8.75 yağ, % 54,1 - 64,2 karbonhidrat, % 2.40 - 3.65 kül ve % 2,1 - 4,9 lif içermektedir. Ayrıca, E vitamini, B grubu vitaminler, lisin aminoasidi ve esansiyel yağ asitleri açısından da zengin bir bitkidir. Bu nedenle buğday ununa ilave edildiğinde elde edilen ürünlerin besin değerini artırmaktadır. Ancak kinoanın bileşiminde acılığa sebep olan saponinlerin bulunması ve buğday gluten proteinlerinin eksikliği fırın ürünlerinde kullanımını sınırlandırmaktadır. Yapısında bulunan saponinler uzaklaştırıldıktan sonra, kinoa zenginleştirme amacı ile ekmek, kek, bisküvi gibi fırın ürünlerinin üretiminde tat, aroma, tekstür, görünüş gibi özellikler dikkate alınarak, buğday ununa belli oranlarda katılabilmektedir. Bunun yanı sıra çölyak hastaları için ekmek yapımında kinoa nişasta kaynağı olarak da kullanılabilmektedir (Keskin & Kaplan Evlice, 2015). Kinoa yapısında gluten proteinlerini içermediğinden, buğday ununa belli oranlarda katılarak yüksek proteinli ekmek, kek ve bisküvi gibi fırın ürünlerinin üretiminde kullanılmaktadır. Böylece hem besin değeri artırılmakta hem de kabul edilebilir aromaya sahip ürünler elde edilmektedir. Ayrıca, çölyak hastalarının kullanabileceği alternatif bir bitki konumundadır (Keskin & Kaplan Evlice, 2015)

Ekmek

Karabuğday, kinoa ve amarant unları iki farklı yöntemle fermente edilerek glutensiz ekmek üretiminde farklı oranlarda (%0, 15, 30 ve 45) kullanılmıştır. Pseudo-tahıl fermente hamur ilavesinin, ekmeklerin bazı, fiziksel, kimyasal ve duyusal özellikleri üzerine etkisi incelenmiştir. Ekmeklerde en düşük parlaklık ve en yüksek kırmızılık değerleri karabuğday içeren örneklerde belirlenmiştir. Formülasyonda pseudo-tahıl fermente hamur oranının artması ekmeklerde kül, protein, fitik asit, toplam fenolik madde, antioksidan aktivite ve mineral madde (Ca, P, K, Fe, Mg ve Zn) miktarının yükselmesine neden olmuştur Kinoa ve amarant katkılı ekmekler 48. ve 72. Saatte karabuğday katkılı ekmeklerden daha düşük sertlik değerlerine sahip olmuştur. Maya

kullanılarak fermente edilen ekşi hamurdan hazırlanan ekmeklerde hacim ve spesifik hacim değerleri daha yüksek, sertlik değerleri ise daha düşük belirlenmiştir. Amarant katkılı ekmekler simetri, gözenek yapısı ve görünüş açısından yüksek puanlar almasına rağmen tat-koku açısından tüketiciler tarafından daha az beğenilmiştir. Pseudo-tahılların %15-30 fermente hamurları ile hazırlanan ekmekler, kontrole yakın genel kabul edilebilirlik puanları almıştır (Yeşil, 2021).

Pseudo-tahıl içeren ekmeklerde kontrolden daha yüksek gözenek yapısı, tat-koku, görünüş ve çiğnenebilirlik puanlarının elde edilmesini sağlayarak glutensiz ekmeklerde hem teknolojik hem de duyusal kalitenin geliştirilmesinde en uygun emülgatör uygulamaları olmuştur. Pseudo tahılların yüksek kullanım oranlarında meydana gelebilecek teknolojik ve duyusal kalite kayıplarının önlenmesi için emülgatörlerin yanı sıra farklı enzimler kullanılabilir. Ekmek formülasyonunda, pseudo-tahılların kullanımı ile artan fitik asit miktarının azaltılması amacıyla farklı yöntemlerin denenmesi besinsel kalitenin yükseltilmesi açısından faydalı olacaktır (Yeşil, 2021).

Glutensiz Ekmek

Farklı oranlarda (%5, 10, 20 ve %30) kinoa unu içeren ve kısmi pişirilerek dondurma yöntemiyle üretilen glutensiz ekmeklerin 0, 5, 10, 15, 30, 45 gün depolanması sonrasında. bazı kimyasal, fiziksel, tekstürel ve duyusal kalite kriterlerinin incelenmesi amaçlanmıştır. Glutensiz un formülasyonu olarak; %53.15 pirinç unu, %24.53 nohut unu, %12.32 patates nişastası, %5 mısır unu ve %5 mısır nişastası karışımı kullanılmıştır. Analiz sonuçlarına göre; kinoa unu ilavesi ekmek örneklerinin protein ve toplam diyet lif miktarlarını etkilemezken, nem, kül, kalsiyum (Ca), magnezyum (Mg) ve fosfor (P) miktarları ile sertlik, sakızımsılık ve çiğnenebilirlilik gibi tekstürel değerlerini arttırmıştır. Renk değerlerinden parlaklık (L*) değeri azalmış, kırmızılık (a*) değeri artmış, sarılık (b*) değeri ise değişiklik göstermemiştir.

Depolanan glutensiz ekmeklerde sertlik ve sakızımsılık artarken, elastikiyet ve esneklik azalmıştır. Soğuk depolama ile renk değerlerinde önemli bir değişiklik olmadığı belirlenmiştir. Duyusal analiz sonuçlarına göre; kinoa unu ilavesi ve kısmi pişirerek dondurulup depolama işlemi glutensiz ekmeklerin kabul edilebilirliğini olumlu yönde etkilemiştir. Sonuç olarak kinoa ununun %30 oranına kadar formülasyona ilave edilmesinin uygun olduğu ve glutensiz ekmeklerin 45 güne kadar kısmi pişirilerek dondurulup depolanabileceği tespit edilmiştir (Hayıt & Gül, 2019). Glutensiz ekmek formülasyonuna kinoa unu ilave edilip, elde edilen ekmekler kısmi pişirilerek dondurulmuş ve üretilen ekmeklerin ekmek özellikleri incelenmiştir. Kinoa ilavesi glutensiz ekmeklerin mineral madde değerini ve kül içeriğini arttırmıştır. Tekstürel, renk ve duyusal özelliklere olumsuz bir etki göstermemiştir (Hayıt & Gül, 2019). Bu yüzden glutensiz ekmeklerin besin içeriğini arttırmak için formülasyona %30 oranına kadar kinoa ilavesinin yapılabileceği sonucuna varılmıştır. Ayrıca çalışma sonucunda kısmi pişirilerek dondurulan glutensiz ekmeklerin 45 güne kadar kalitelerinde büyük problemler olmadan depolanabileceğine duyusal değerlendirme sonucunda karar verilmiştir. Hastaların en büyük ve önemli sorunlarından olan taze ekmeğe ulaşım probleminin kısmi pişirerek dondurma yöntemi ile çözülebileceği sonucuna varılmıştır (Hayıt & Gül, 2019).

Lavaş Ekmeği

Kinoa tohumu biyolojik değeri yüksek proteinleri, düşük glisemik indeksli karbonhidratlar, fitosteroidler, esansiyel amino asitler, mineral maddeler, vitaminler ve biyoaktif bileşenler bakımından zengindir. Kinoanın diğer tahıl taneleri gibi gluten içermemesi, gluten hassasiyeti olan insanlar için önemli bir gıda kaynağı olduğunu göstermektedir. Buğday ununa kinoa unu katılarak lavaş yapılması, lavaşı protein, yağ, mineral madde ve esansiyel amino asitler bakımından zenginleştirmektedir. Böylece besin madde bakımından zenginleştirilmiş fonksiyonel lavaş üretilebilecektir. Kinoa ununu %20 düzeyinde kullandığımızda ekmek özellikleri bakımından herhangi bir olumsuzlukla karşılaşılmadığından lavaş yapımında %20'ye kadar kullanılabileceği kanısına varılmıştır (Akturfan, 2018).

DRAFT

Kinoa ve Kayısı Karışımı Sostan Elde Edilen Puding

Kinoa ve kayısı karışımı ile hazırlanan sosun (%30 kayısı, %3 kinoa, aroma verici, şeker, su modifiye nişasta, siyah havuç suyu konsantresi, karragenan ve beta karoten) %3, %5 ve %7 oranlarda eklendiği pudingler hazırlanmış ve protein, yağ, kuru madde analizleri yapılarak; tekstür analizi ve pH değerleri ile toplam mezofilik aerob bakteri (TMAB) sayısı 5 gün boyunca takip edilmiştir. Tüm analizler üç tekerrürlü olacak şekilde yürütülmüştür.

Kontrol grubu olarak; sos içermeyen puding hazırlanmıştır. Pudinglerin hepsi aynı formülasyonla hazırlanmışlardır. Ayrıca, 9'lu hedonik skalanın kullanıldığı duyusal analiz 9 panelist tarafından analizin 1., 3. ve 5. günlerinde değerlendirilmiştir. Çalışmanın sonuçları incelendiğinde; pH değerleri; sade, %3, %5 ve %7 sos içerikli pudinglerde sırasıyla ortalama olarak 5 gün için 6.71, 6.47, 6.41 ve 6.36 olarak ölçülmüştür. Yağ değerleri; sade, %3, %5 ve %7 sos içerikli pudinglerde ortalama olarak sırasıyla 2.37, 2.34, 2.28 ve 2.26 olarak ölçülmüştür (Çolak Şaşmazer, Özoğlu, & Korukluoğlu, 2019).

Yine aynı şekilde protein değerleri 2.37, 2.15, 2.18 ve 2.10 olarak ölçülürken; kuru madde değerleri %21.59, %23.39, %24.32 ve %25.55 olarak ölçülmüştür. TMAB sayıları tüm örneklerde ilk gün 1 log'dan az iken 5.günde 2 log değerlerine ulaşmıştır. Duyusal analiz sonuçları incelendiğinde ise genel olarak puding beğeni seviyesinin 7 (beğendim) seviyelerinde olduğu, beğeninin zaman geçtikçe düşüş gösterdiği ve soslu pudingler içerisinde %5 orana sahip olanın daha yüksek beğeni aldığı gözlemlenmiştir. Sonuç olarak %5 sos içeriğine sahip puding tüketiciye alternatif ve fonksiyonel bir tatlı olarak sunulabilir (Çolak Şaşmazer, Özoğlu, & Korukluoğlu, 2019)**.**

Pişmemiş Tavuk Köftesi

Buğday unu ve kinoa ununun hazırlanan karışımların soğukta depolanan tavuk köftelerin tekstür özellikleri üzerine etkisi araştırılmıştır. Bu karışımlar ile hazırlanan pişmemiş tavuk köfteler, soğukta muhafazanın 1, 3, 7 ve 10'uncu günlerinde tekstür analizine tabii tutulmuştur. Çalışma sonucunda köfte bileşiminde kinoa unu kullanımının tekstür değerlerini değiştirebildiği belirlenmiştir (Meral & Kılınççeker, 2022). Araştırma sonucunda et ve ürünlerinde pişmiş ürünlerde olduğu kadar pişmemiş ürünlerde de tekstürel profilinin değişim gösterebileceği ve satış esnasında tercihi etkileyebileceği anlaşılmıştır. Çalışmada kullanılan ve fonksiyonel özelliğe sahip bileşenler bakımından zengin kinoa ununun ise bu değişimleri etkileyebileceği gözlenmiştir (Meral & Kılınççeker, 2022). Köftelerin depolanması sürecinde zamana bağlı olarak tekstür profil değerlerinde bir azalma belirlenirken, bu değişimleri azaltmada özellikle % 30 ve % 50 kinoa unu karışımlarının kullanımının daha avantajlı olabileceği ve tavsiye edilebileceği sonucuna varılmıştır (Meral & Kılınççeker, 2022).

Et Ürünleri

Kinoa (*Chenopodium quinoa* wild.)'nın gıda sanayii için önemli olan bazı özellikleri ve et ürünlerinde sağlayabileceği faydalar üzerinde durulmuştur. Özellikle kimyasal yapısı ele alınarak, et sektöründeki uygulamalarından örnekler verilmiştir (Kılınççeker & Karahan, 2019). Kinoa tohumunun fonksiyonel özellikteki yüksek karbonhidrat ve protein içeriği, esansiyel yağ asitleri profili, antioksidan bileşikler ve yüksek demir içeriği nedeniyle et ürünleri üretiminde önemli faydalar sağlayabileceği düşünülmektedir. Ancak çalışmalara bakıldığında, et ürünlerinde kinoa ile ilgili araştırmaların yetersiz olduğu gözlenmiştir. Dolayısı ile bu derlemede kinoanın sahip olduğu yapısal özelliklerden dolayı et ürünlerinin kalitesini geliştirmede katkısının olabileceği ve daha fazla deneysel çalışmaya ihtiyaç olduğu ortaya konulmuştur (Kılınççeker & Karahan, 2019). Kinoanın et ürünlerini hazırlamada alternatif bir bitkisel katkı olabileceği anlaşılmaktadır. Özellikle sahip olduğu renk özelliği, içerdiği protein, nişasta, lif oranları ve diğer bileşenler ile bunların fonksiyonel özelliklerinden dolayı ürün kalitesine önemli katkılar sağlayabileceği düşünülmektedir. Ancak yapılan çalışmalara bakıldığında özellikle kırmızı ve beyaz et ürünlerinde kullanımı ile ilgili verilerin yetersiz olduğu ve daha fazla uygulamaya ihtiyaç duyulduğu anlaşılmıştır (Kılınççeker & Karahan, 2019).

Ek Gıda

Kinoa (*Chenopodium quinoa* Willd.) unu ilave edilmiş ek gıda formülasyonlarının (CF) fizikokimyasal, reolojik, moleküler, termal ve duyusal özelliklerini değerlendirmektir. Bu çalışmada, ek gıda (CF) formülasyonuna kinoa unu (QF) ilavesinin, ek gıdanın fizikokimyasal ve reolojik özelliklerini önemli ölçüde etkilediği, üründeki protein, lif miktarını arttırırken, şeker miktarının algılanmasının azalmasına neden olduğu tespit edilmiştir. Kinoa ununun (QF), 15K örneği haricindeki ek gıdalarda partikül boyutunda azalmaya neden olduğu gözlenmiştir (Ayseli, 2020). Kinoa unu (QF) ilavesiyle camsı geçiş sıcaklığı düşmüştür. FT-IR analizlerine ait piklerde aromatik-amino asitler gözlenmiştir. GC, GC-MS ve GC-O analizleri sonucunda önemli bir kısmını aldehit, keton ve alkollerin oluşturduğu 50 adet aroma ve 23 adet aroma-aktif bileşik tespit edilmiştir. Duyusal test sonuçlarına göre en fazla beğeniyi 8K örneği almıştır. Sonuç olarak kinoa unu (QF) ilavesinin bebek ve küçük çocuk ek gıdalarının fizikokimyasal ve duyusal özelliklerini olumlu yönde geliştirdiği gözlemlenmiştir (Ayseli, 2020). Yalnızca kinoa değil ülkemizde hali hazırda yetişen organik ve konvansiyonel tahılların ek gıda üretimi amacıyla işlenmesinde kaynak olarak kullanılacağını, bu sayede de ithal ikamesi tahıl bazlı bebek ve küçük çocuk ek gıdası üretim faaliyetlerinin orta vadede artacağı öngörülmektedir (Ayseli, 2020).

Türk Mantısı

Buğday unu ile kombine edilmiş en uygun kinoa ve amarant kullanım oranının saptanması, elde edilen hamurun ürünün niteliklerine etkisi araştırılmıştır. Bu amaçla, %0, %5, %10, %15 ve %20 oranında kinoa ve amarant unları kullanılarak 9 farklı mantı hamuru elde edilmiştir. Hamurların pişme (pişme süresi, hacim ve ağırlık artışları, suda çözünen madde miktarı), fiziksel (renk), kimyasal (yağ, protein, toplam fenolik madde, antioksidan aktivite) ve duyusal özellikleri incelenmiştir. Kinoa ve amarant ilaveli mantı numunelerinde protein ve yağ oranının arttığı gözlemlenmiştir. En yüksek protein değeri %20 kinoalı mantıda %12,86, en yüksek yağ oranı ise %20 amarantlı mantı örneğinde %2,87 olarak tespit edilmiştir. Toplam fenolik madde miktarının ikame unların ilavesi ile arttığı görülmüş ve %20 kinoa içeren örnekte 31,34 mg GAE/100g, %20 amarant içeren mantı örneğinde ise 27,93 mg GAE/100g, olarak bulunmuştur. Antioksidant aktivite kinoa içeren mantı örneklerinde daha fazla gözlemlenmiş ve %20 kinoa içeren mantı örneğinde %44,67 olarak elde edilmiştir. Pişme testlerinde ise pişme süresi ikame unların miktarı ile artmış ve kinoa unu içeren mantı örneklerinde daha fazla olduğu görülmüştür. Son olarak gerçekleştirilen duyusal analizde tüm örnekler kabul sınırları içinde değerlendirilmiştir (Aygün, 2021).

Kinoa ve amarant unu ilavesi ile mantı örneklerinde pişme sonrası ağırlık artışı meydana gelmekle beraber en yüksek artış olan kontrol numunesine ulaşamamışlardır. Pişme sonrası ölçülen hacimsel artış sonuçlarına göre ise en yüksek hacim artışı %20 oranında kinoa ve amarant içeren mantı örneklerinde gözlemlenmiştir. Bu durum, kinoa ve amarant nişasta yapısının ile şişme kapasitesinin daha fazla olmasının bir sonucu olabileceğini göstermektedir. Mantı örneklerinin suya geçen madde miktarı istatiki olarak incelendiğinde mozaik yapı göstermesi pişme işleminde oluşan madde kaybının kontrol numunesi ile benzer olduğunu göstermektedir. Mantı örneklerinin pişme sürelerine bakıldığında, kinoa ve amarant miktarı arttıkça pişme sürelerinin de arttığı saptanmıştır (Aygün, 2021). Kinoa içeren mantı örneklerinin istatiki olarak daha fazla pişme süresine sahip olduğu, en düşük pişme süresinin kontrol örneğinde gözlenmiştir. Pişme analizleri göz önünde bulundurulduğunda kinoa ve amarant katkısı mantı örneklerinin pişme özelliklerine olumsuz yansımamıştır (Aygün, 2021).

Pseudo-tahıllardan olan kinoa ve amarant, yüksek fenolik madde, doymamış yağ asitlerince zengin olması mantı örnekleriniinin fonksiyonelliği arttırmıştır. Bu çalışma sonucunda endüstriyel ya da ev yapımı mantılarda kinoa ve amarant ilavesinin mantı örneklerinin besleyicilik ve kalite özelliklerini arttırması nedeniyle tüm kinoa ve amarant oranlarının mantı formulasyonlarında kullanılabileceği görülmüştür (Aygün, 2021).

Tarhana

Tarhana üretiminde kinoa, karabuğday ve lüpen unları 5 farklı ikame oranında (% 0, 10, 20, 30 ve 40) kullanılmıştır. Tarhana üretimi (3 x 5) x 2 deneme desenine göre iki tekerrürlü olarak yürütülmüştür. Üretilen tarhana örneklerinde fiziksel, kimyasal, besinsel ve duyusal özellikler belirlenmiştir. Duyusal açıdan incelendiğinde tarhanaya ilave edilen unlar arasında en fazla tat, koku, ekşilik ve genel beğeni skorları % 20 kinoa unu ilavesi ile sağlanmış, % 20 lüpen unu ilavesinin ise renk üzerine olumlu etkiler gösterdiği panelistler tarafından belirtilmiştir. Ülkemizde endüstriyel ve ev yapımı olarak üretilen tarhanaların besinsel, fonksiyonel ve duyusal açıdan zenginleştirilmesinde kinoa ve lüpen ununun rahatlıkla tarhana formulasyonuna ilave edilebileceği, karabuğday ununun ise besinsel ve fonksiyonel özellikleri artırmada oldukça etkili olduğu fakat duyusal açıdan karabuğdayın kendine özgü tadını tarhanada da hissettirdiği için panelistler tarafından fazla beğeni almadığı kanaati uyanmıştır (Çevik, 2016).

Sonuç ve Öneriler

Birçok bireyde gıda alerjisi ve intoleransı görülmektedir. Çölyak, gluten ataksi ve gluten intolerans gibi rahatsızlar söz konusu olduğunda gluten tüketiminden kaçınılmalıdır. Gluten içermeyen darı, mısır ve pirinç gibi tahıllar, karabuğday, amarant ve kinoa gibi pseudo-tahıllar iyi bir mineral, vitamin ve lif kaynağı olmaktadır. Bu çalışmada, kinoanın besin değerlerinden, ülkemizdeki yerinden ve gıdalarda kullanımından bahsedilmiştir. Çalışma sonucunda kinoanın çölyak gibi glutenden kaynaklanan hastalıklardan dolayı, gluten içeren ürünleri tüketemeyen kişiler için alternatif bir tahıl olduğu görülmüştür. Ülkemizde gluten içeren unlardan üretilen ekmek, makarna, bisküvi, kek gibi birçok gıdanın kinoa unu ile alternatifinin geliştirilmeye başlandığı ve kinoanın daha da yaygınlaştığı fark edilmiştir. Ancak toplumumuzda kinoa, amarant ve karabuğday gibi alternatif pseudo-tahılların farkındalığı oluşturulması gerektiği, kinoa konusunda çalışmaların daha da artırılması gerektiği öngörülmüştür.

KAYNAKÇA

Akturfan, M. (2018). Lavaş Ekmeğinee Farklı Düzeylerde Kinoa Tohumu Unu Katılmasının Hamur ve Ekmek Özellikleri İle Kimyasal Bileşim Üzerine Etkisi. Konya: Selçuk Üniversitesi Sağlık Bilimleri Enstitüsü.

Alaşalvar, H., Erinç, H., & Çolakoğlu, A. S. (2019). FARKLI BOYUTLARDA KİNOA KEPEK UNU KULLANIMININ KEKLERİN FİZİKSEL, DUYUSAL VE TEKSTÜREL ÖZELLİKLERİ ÜZERİNE ETKİSİ. *Kahramanmaras Sutcu Imam University Journal of Engineering Sciences* , 22.

Argan, B. E. (2019). *Ekmeklik Buğday Ununa Kinoa Unu Karıştırılarak Üretilen Eriştelerin Bazı Kalite Özelliklerinin Belirlenmesi.* İzmir: Ege Üniversitesi Fen Bilimleri Enstitüsü.

Aygün, G. (2021). *Kinoa ve Amarant Unu İlavesinin Türk Mantısının Kalite Özelliklerine Etkilerinin Belirlenmesi.* Tekirdağ: Tekirdağ Namık Kemal Üniversitesi Fen Bilimleri Enstitüsü.

Ayseli, M. T. (2020). *Kinoa Unu İlavesiyle Üretilen Bebek Ek Gıdasının Fizikokimyasal Ve Teknolojik Özelliklerinin Belirlenmesi.* İstanbul: Yıldız Teknik Üniversitesi Fen Bilimleri Enstitüsü.

Bayram, M., Pekacar, S., & Deliorman Orhan, D. (2018). Kinoa ve Sağlık Üzerine Etkileri. *Gümüşhane University Journal Of Health Sciences* , 47-57.

Çevik, A. (2016). *Tarhananın Besinsel Zenginleştirilmesinde Kinoa, Karabuğday ve Lüpen Unlarının Kulllanımı.* Konya: Necmettin Erbakan Üniversitesi Fen Bilimleri Enstitüsü.

Çolak Şaşmazer, R., Özoğlu, Ö., & Korukluoğlu, M. P. (2019). Kayısı ve kinoa KArışım Soslu Puding Üretiminin Ve Bazı Kalite ÖZelliklerinin Araştırılması. *Uluslararası Gıda, Tarım ve Hayvancılık Kongresi* (s. 43-52). Gaziantep: İksad Yayınevi.

Demir, B. (2018). *Çimlendirilmiş Kinoa Ununun Glutenli ve Glutensiz Makarna Üretiminde Kullanım İmkanları.* Konya: Necmettin Erbakan Üniversitesi Fen Biimleri Enstitüsü.

Erinç, H. (2020). Farklı Boyutlarda Kinoa Kepek Unu Kullanımının Bisküvilerin Fiziksel, Duyusal ve Tekstürel Özellikleri Üzerine Etkisi. *The Journal Of Food* , 1121-1133.

Hafif, O. (2019). *Farklı Oranlarda Kinoa (Chenopodium quinoa Willd.) Unu İlavesinin Probiyotik Yoğurtların Fizikokimyasal, Tekstürel, Mikrobiyolojik ve Duyusal Özellikleri Üzerine Etkisi.* Şanlıurfa: Harran Üniversitesi Fen Bilimleri Enstitüsü.

Hayıt, F., & Gül, H. (2019). Kinoa Ununun ve Kısmi Pişirilerek Dondurma Yönteminin Glutensiz Ekmek Kalitesi Üzerine Etkisi. *Karadeniz Fen Bilimleri Dergisi* , 406-427.

Jancurova, M., Minarovicova, L., & Dandar, A. (2009). Quinoa - a Review. *Czech Journal of Food Sciences* , 21-79.

Keskin, Ş., & Kaplan Evlice, A. (2015). Fırın Ürünlerinde Kinoa Kullanımı. *Tarla Bitkileri Merkez Araştırma Enstitüsü Dergisi* , 150-156.

Kılınççeker, O., & Karahan, A. M. (2019). Kinoa (Chenopodium quinoa Wild.)'nın Bazı Özellikleri ve Et Ürünlerinde Kullanımı. *Türkiye Tarımsal Araştırmalar Dergisi* , 237-241.

Meral, R., & Kılınççeker, O. (2022). KİNOA (CHENOPODIUM QUINOA WILD.) UNUNUN SOĞUKTA DEPOLANAN PİŞMEMİŞ TAVUK KÖFTELERİNİN TEKSTÜR PROFİLİNE ETKİSİ. *ADYUTAYAM* , 46-52.

Öncel, E. (2017). *Erişte Üretiminde Farklı Oran ve Kombinasyonlarda Karabuğday Amarant ve Kinoa Unlarının Kullanım İmkanları.* Konya: Necmettin Erbakan Üniversitesi Fen Bilimleri Enstitüsü.

Özgören, E., & Yapar, A. (2022). Kinoa (Chenopodium quinoa Willd) Unu ile Zenginleştirilen Muffin Keklerin Bazı Fizikokimyasal ve Duyusal Özellikleri. *Dergi Park* , 244-250.

Sevindik, A., Gültekin, R. N., & Uran, H. (2021). ALTIN TAHIL : KİNOA. *Kırklareli University Journal of Engineering and Science* , 155-165.

Sharma, V., Chandra, S., Dwivedi, P., & Parturkar, M. (2015). QUINOA (CChenopodium quinoa Willd): A Nutritional Healthy Grain. *International Journal of Advanced Research* , 725-736.

Tan, M. P., & Temel, S. D. (2019). *Her Yönüyle Kinoa Önemi, Kullanılması ve Yetiştiriciliği.* Ankara: İKSAD Publishing House .

Valencia, R. C., Mari, R. A., & Serna, L. A. (2011). Quinoa (Chenopodium quinoa, Willd) as a source of dietary fiber and other fuctional components. *Food Science and Technology* , 225-230.

Yağan, O. K. (2019). *Adana Koşullarında Yetiştirilen Kinoa (Chenepodium quinoa Willd) Azotlu Gübre İsteğinin Belirlenmesi.* Konya: Selçuk Üniversitesi Fen Bilimleri Enstitüsü Toprak Bilimi ve Bitki Besleme Anabilim Dalı.

Yeşil, S. (2021). *Fermente Edilmiş Karabuğday, Kinoa ve amarant Unlarının ve Farklı Emülgatörlerin Glutensiz Ekmek Kalitesi Üzerine Etkileri.* Karaman: Karamanoğlu Mehmetbey Üniversitesi Fen Bilimleri Enstitüsü.

DRAFT

ORAL PRESENTATION

Investigation of the Existence and Seroprevalence of Canine Herpesvirus-1 (CHV-1) in Türkiye: A PCR and ELISA-Based Study

Mehmet Cevat Temizkan[1*] (*ORCID: https://orcid.org/0000-0002-4353-6759*), Secil Sevinc Temizkan[2] (*ORCID: https://orcid.org/0000-0002-2427-3877*), Mustafa Eren Catal[3] (*ORCID: https://orcid.org/0009-0000-1005-2304*)

[*1]Yozgat Bozok University, Faculty of Veterinary Medicine, Department of Genetics, Yozgat, Türkiye.
[2]Yozgat Bozok University, Faculty of Veterinary Medicine, Department of Virology, Yozgat, Türkiye.
[3]Yozgat Bozok University, Sefaatli Vocational College, Department of Veterinary, Yozgat, Türkiye.

*Corresponding author e-mail: m.cevat.temizkan@yobu.edu.tr

Abstract

Canine herpesvirus-1 (CHV) is one of the primary causes of the disease known as kennel cough in dogs. The virus leads to respiratory and genital system infections in adult dogs and fatal hemorrhagic diseases in newborns. CHV-1 has been studied worldwide for many years using methods such as PCR and ELISA. In this study, blood samples were collected from a total of 192 dogs in the provinces of Izmir and Yozgat in Türkiye, and these samples were analyzed using PCR and ELISA. Our results revealed no presence of CHV-1 antigen in the Yozgat province. The detection of the disease from the blood is only feasible during the viremia stage. Therefore, our inability to identify the antigen from the collected samples indicates not the absence of the disease, but rather the absence of the dog being in the viremia stage. In Izmir province, the CHV-1 seroprevalence was found to be 2.7% (2/74), while no CHV-1 antibodies were detected in Yozgat province. Taking into account all studies conducted in Türkiye, CHV-1 seroprevalence was determined to be 39.01%. In conclusion, the future molecular detection and characterization of the disease are also of importance.

Keywords: Canine herpesvirus, Dog, ELISA, PCR, Prevalence, Türkiye

INTRODUCTION

Canine herpesvirus-1 (CHV-1), a 125171 bp enveloped double-stranded DNA virus, is a member of the Varicellovirus genus, Alphaherpesvirinae subfamily, and Herpesviridae family. It causes kennel cough (Day et al., 2020; Gracin et al., 2023; Matsuu et al., 2020; Kaczorek et al., 2016) with significant mortality rates, particularly in puppies (Gracin et al., 2023; Krogenaes et al., 2014; Nöthling et al., 2008). CHV-1 causes respiratory and genital system infections in adult dogs and fatal hemorrhagic diseases in newborn puppies (Bottinelli et al., 2015; Day et al., 2020; Nöthling et al., 2008; Sahna and Aslan, 2014). In adult dogs, the infection usually follows a subclinical course or localizes in the upper respiratory tract and eyes. In puppies, the infection generally leads to the loss of the majority of the litter (Erles et al., 2004; Gracin et al., 2023). The severity of CHV-1's impact on dog health reveals the need for further research into the virus and its potentially serious consequences.

Numerous studies on CHV-1 have been conducted in many countries around the world (Bottinelli et al., 2015; Cobzariu et al., 2018; Dahlbom et al., 2009; Day et al., 2020; Erles and Brownlie, 2005; Erles et al., 2004; Gracin et al., 2023; Kaczorek et al., 2016; Krogenaes et al., 2014; Lavan and Knesl, 2015; Matsuu et al., 2020; Musayeva et al., 2013; Nöthling et al., 2002; Okonkowski et al., 2021; Pratelli et al., 2014; Rezaei et al., 2020; Rota et al., 2020). However, the studies conducted in Türkiye are quite limited (Acar et al., 2009; Gur and Acar, 2007; Sahna and Aslan, 2014; Yapici et al., 2018; Yesilbag et al., 2012). The majority of global studies focus on antigen detection using PCR (Bottinelli et al., 2015; Day et al., 2020; Erles and Brownlie, 2005; Erles et al., 2004; Gracin et al., 2023; Kaczorek et al., 2016; Lavan and Knesl, 2015; Matsuu et al., 2020; Okonkowski et al., 2021; Pratelli et al., 2014; Rezaei et al., 2020) and antibody detection through serum neutralization or ELISA (Bottinelli et al., 2015; Cobzariu et al., 2018; Dahlbom et al., 2009; Erles and Brownlie, 2005; Gracin et al., 2023; Krogenaes et al., 2014; Musayeva et al., 2013; Nöthling et al., 2002; Pratelli et al., 2014; Rota et al., 2020). Studies conducted in Türkiye have been carried out using ELISA and

similar methods for antibody detection. In Türkiye, no study has been reported targeting antigen detection using ELISA, PCR, or any other method.

For antigen detection, PCR is the most widely accepted method worldwide (Bottinelli et al., 2015; Day et al., 2020; Erles and Brownlie, 2005; Erles et al., 2004; Gracin et al., 2023; Kaczorek et al., 2016; Lavan and Knesl, 2015; Matsuu et al., 2020; Okonkowski et al., 2021; Pratelli et al., 2014; Rezaei et al., 2020). Additionally, studies have shown that ELISA provides more sensitive results than other tests for the detection of CHV-1 antibodies (Nöthling et al., 2008; Ronsse, et al., 2002; Yesilbag et al., 2012).

In this study, PCR analysis was performed for the molecular detection of CHV-1 in dogs. CHV-1 IgG ELISA analysis was conducted to determine seroprevalence. In Izmir, previously collected serum samples were examined with ELISA. Samples collected from Yozgat were assessed with PCR and ELISA for the detection of CHV-1 antigen and antibody presence. Our hypothesis is the low level or no detection of CHV-1 antigen presence and the detection of antibody presence in dogs in Izmir and Yozgat.

MATERIALS AND METHODS

Ethics Statement

The research protocol of the current study was approved by the Erciyes University Ethics Committee for the Local Use of Animals in Experiments (No. 23/017 and 23/018).

Sample Collection and Preparation

During the course of the study, a total of 192 dog samples were collected from the provinces of Izmir and Yozgat in Türkiye.

For Izmir, samples were collected from a total of 74 dogs in 2020, comprising 46 owned dogs and 28 shelter dogs, all over the age of 1.5 years, in 2 different shelters. All sampled dogs were unvaccinated against CHV-1. Approximately 3 mL of blood samples were collected in EDTA-containing tubes, placed on an ice pack within a styrofoam container, and swiftly transported to the mobile laboratory located within the animal shelter for plasma separation. Within 3 hours after the samples were collected, they were centrifuged at 3000 g for 20 minutes. Carefully avoiding the mixing of layers, 1 mL of the blood plasma was transferred into 1.5 ml tubes. The plasma samples were subsequently transported to the Yozgat Bozok University, Faculty of Veterinary Medicine laboratory in dry ice. Plasma samples were stored at -20°C until they were analyzed using ELISA.

For Yozgat, samples were collected from 118 mixed-breed shelter dogs, all over the age of 1.5 years, in 3 different shelters within Yozgat in 2023. All dogs were vaccinated against rabies only. Approximately 3 mL of blood samples were collected in EDTA-containing tubes, placed on an ice pack within a styrofoam container, and promptly transported to the Yozgat Bozok University, Faculty of Veterinary Medicine laboratory. Upon arrival at the laboratory, 400 µl of 100 samples were transferred to 1.5 ml tubes for genetic analysis and stored at -20°C until DNA isolation. To perform the ELISA analyses, the 118 blood samples were centrifuged at 3000 g for 20 minutes. Without mixing with other layers, 1 mL of the blood plasma was carefully transferred to 1.5 ml tubes. The plasma samples were preserved at -20°C until the ELISA analyses were conducted.

DNA Isolation

For the Yozgat samples, due to the isolation kit's capacity for 100 samples, 18 samples were randomly excluded from the isolation process. A spin column-based blood DNA isolation kit (Macherey Nagel, Germany) was utilized for DNA isolation. The isolation procedure was performed following the kit's protocol. The DNA isolations of the Yozgat samples were conducted at the Yozgat Bozok University, Faculty of Veterinary Medicine laboratory. The obtained DNA samples were stored at -20°C in a freezer until they were used in the PCR analysis.

PCR Analysis

The primers implemented in the PCR analysis possess the following sequences: Forward: CCTAAACCTACTTCGGATGA and Reverse: GGCTTTAAATGAACTTCTCTGG (Ronsse et al., 2005). These primers are specific to CHV-1 and amplify a 443 base-pair region. For each sample, the PCR mixture contained 2.5 µl of 10X DreamTaq Buffer, 1 µl of 10 mM dNTP mix, 1 µl of each primer, 0.25 µl of Dream Taq DNA polymerase (5 U/µl) (Thermo Scientific, USA), and 18.25 µl of nuclease-free water. Individually, 6

µl of the isolated DNA samples were added to each mixture. The tubes containing the PCR mixtures were placed in a Thermal Cycler (T100, Biorad, Germany). The applied thermal cycling conditions for the PCR were as follows: an initial denaturation step at 96°C for 4 minutes, followed by 35 cycles of 1 minute at 51°C, 1 minute at 72°C, and 1 minute at 94°C, with a final step of 1 minute at 50°C and 10 minutes at 72°C.

To visualize the PCR products, a 100 ml 1% agarose gel was prepared. Agarose, TBE (Tris-Boric Acid-EDTA), and ethidium bromide were used in the gel preparation. In each well of the prepared gel, 5 µl of Gel Loading Dye and 5 µl of the PCR sample were added for each sample. A 100 bp DNA Ladder was used as a control, and the electrophoresis was carried out at 100 V and 50 mA. Following electrophoresis, the gel was visualized using a gel imaging system.

ELISA Analysis

CHV-1 ELISA Antibody Kit (Shanghai Coon Koon Biotech, China) was used for the 74 Izmir and 118 Yozgat samples. The kit is based on the detection of CHV-1 IgG. All ELISA procedures were conducted following the kit's protocol. The plates were then read using spectrophotometry with a 450 nm filter on an automatic ELISA reader (SPECTROstarnano, BMG Labtech, Germany) according to the manufacturer's protocol. The ELISA procedures were conducted at the Yozgat Bozok University, Faculty of Veterinary Medicine laboratory.

RESULTS and DISCUSSION

As a result of the PCR analyses, CHV-1 presence was not detected in 100 samples from Yozgat. Similarly, no antibody presence was detected in 118 samples collected from Yozgat. In Izmir, out of a total of 74 dogs, including 46 owned dogs and 28 shelter dogs, CHV-1 antibody presence was detected in two dogs (2.7%), one owned dog, and one shelter dog (Table 1). Until now, the presence of CHV-1 antigen has not been reported in Türkiye. Considering all the studies conducted in Türkiye to date, the seroprevalence of CHV-1 is seen as 39.01% (Figure 1; Table 1). Global studies on CHV-1 generally focus on PCR for antigen detection, ELISA, and serum neutralization studies for antibody detection. The results of these studies vary greatly from one another.

Sampling Year	Sample Number	Positive	Negative	Location	References
Antibody Prevalence					
2007 2010	560	39.3%	60.7%	Balıkesir Bursa	Yesilbag et al., 2012
2007	94	48.8%	51.2%	Afyonkarahisar	Gur and Abay, 2007
2009	110	71.8%	28.2%	Afyonkarahisar	Acar et al., 2009
2010 2011	141	68.8%	31.2%	Antalya Konya	Yapici et al., 2018
2014	95	25.2%	74.8%	Kayseri Sivas	Sahna and Aslan, 2014
2020	74	2.70%	97.3%	Izmir	This study
2023	118	0%	100%	Yozgat	
Total	1192	39,01%	60,99%	All	
PCR Results					
2023	100	0%	100%	Yozgat	This study
Total	118	0%	100%	All	

Table 1. Temporal Variation of CHV-1 Antibody Prevalence and PCR Results in Türkiye

In England, in one of two different studies, 80 dogs with canine infectious respiratory disease (CIRD) were examined by PCR, but no positive samples were found (Erles and Brownlie, 2005). In another study, 12.8% of 211 tracheal samples taken from dogs were found to be PCR-positive (Erles et al., 2004). In Europe, between 2011-2015, PCR analysis was performed on different types of samples taken from 1,354 dogs with and without canine infectious respiratory disease complex in Germany, Italy, and Finland, but CHV-1 presence was not detected (Bottinelli et al., 2015; Day et al., 2020; Pratelli et al., 2014). In a study conducted in Poland between 2014-2015, 40 dogs with canine infectious respiratory disease complex were examined, and a positivity rate of 80% was detected. However, this rate was found to be 40-50% one week later (Kaczorek et al., 2016). In Croatia, no CHV-1 antigen presence was found in the samples collected from 203 dogs (Gracin et al., 2023). In Japan, between 2017-2018, CHV-1 was detected by PCR in 1.2% to 8.2% of samples taken from owned and shelter dogs, respectively (Matsuu et al., 2020). In a study in the USA, 503 dogs were examined by PCR, and CHV-1 presence was found in only 0.8% of the dogs (Lavan and Knesl, 2015). In another study in the same country, 133 owned dogs and 295 shelter dogs were examined for CHV-1 presence by PCR; CHV-1 presence was detected in 1.5% of owned dogs and 2.7% of shelter dogs. The same study found no difference in the risk of contracting CHV-1 between owned dogs and shelter dogs (Okonkowski et al., 2021). In a study conducted with 140 dogs in Iran, CHV-1 presence was detected in 15% of dogs by PCR (Rezaei et al., 2020). In Türkiye, there is no other study aimed at detecting CHV-1 presence by PCR (Figure 1). As can be understood from past studies, the detection of CHV-1 antigen by PCR is quite challenging in situations where almost the entire shelter is not infected. Moreover, the detection of the disease from the blood is only feasible during the viremia stage. Therefore, our inability to identify the antigen from the collected samples indicates not the absence of the disease, but rather the absence of the dog being in the viremia stage.



Figure 1. CHV-1 Distribution in Türkiye

In a study conducted in South Africa, the CHV-1 seropositivity rate was found to be 22% in samples taken from 328 dogs across 38 different shelters, with 18 shelters showing no presence of CHV-1 (Nöthling et al., 2002). In a study carried out in England, dog populations in two shelters were examined over a year, and CHV-1 seroprevalence was reported to be 7.9% (Erles and Brownlie, 2005). In a 2014 study in Italy, the CHV-1 seropositivity rate was found to be 14.6% in 865 dogs (Pratelli et al., 2014). In the same country, in a 2015 study conducted in a shelter, 243 dogs were examined for CHV-1, but no antibody presence was detected (Bottinelli et al., 2015). In another study in Italy in 2020, 370 dogs were examined, and 50.3% of the dogs were found to have CHV-1 antibodies. However, the seropositivity rate was zero in 10 of the 33 shelters (Rota et al., 2020). The CHV-1 seropositivity rate was found to be 81.5% in Finland in 2009 (Dahlbom et al., 2009), 26.8% in Lithuania in 2010 (Musayeva et al., 2013), 85.5% in Norway in 2014 (Krogenaes et al., 2014), 86.36% in Romania in 2018 (Cobzariu et al., 2018), and 32.02% in Croatia in 2023 (Gracin et al., 2023). The very low detection rate of CHV-1 in Izmir province presents a similar situation to the study conducted by Erles and Brownlie (2005). In addition, as in previous studies (Matsuu et al., 2020; Okonkowski et al., 2021), the same number of positive samples were detected in owned and shelter dogs. The study conducted in Yozgat province reveals similar results to the study carried out by Bottinelli et al. (2015). Although the sampled shelters represent the entire province of Yozgat, the number of shelters is limited to 3, and in previous studies, even when samples were taken from many shelters, CHV-1 was detected in only half of them. Furthermore, seroprevalence values are found at very different levels in different countries (Cobzariu et al., 2018; Dahlbom et al., 2009; Gracin et al., 2023; Krogenaes et al., 2014; Musayeva et al., 2013; Nöthling et al., 2002), they can change over the years within the same country, as in the case of Italy (Bottinelli et al., 2015; Pratelli et al., 2014; Rota et al., 2020). Therefore, not detecting CHV-1 in Yozgat province does not necessarily imply that the disease has never been present in this region.

Upon reviewing the research conducted in Türkiye, among the 560 serum samples collected from dogs in Balıkesir and Bursa provinces between the years 2007 and 2010, 39% of these samples were found to be seropositive for CHV-1 (Yesilbag et al., 2012). In different studies conducted in Afyonkarahisar province, CHV-1 seroprevalence was determined to be 48.8% in stray dogs in 2007 (Gur and Acar, 2007), and 71.8% in Kangal breed dogs in 2009 (Acar et al., 2009). Before 2011, 141 samples collected from Antalya and Konya provinces were examined, and 68.79% of the dogs were found to have CHV-1 antibodies (Yapici et al., 2018). In a study conducted in Kayseri and Sivas provinces in 2014, research was carried out to detect CHV-1 antibodies in 95 dogs, 40 of which were German Shepherds and 55 were Sivas Kangals. In total, 25.2% of the dogs were found to be positive for CHV-1 antibodies. However, it was observed that only 2.5% of the German Shepherd dogs were seropositive, while 41.8% of the Sivas Kangal dogs were seropositive. This situation has been discussed as possibly being due to breed susceptibility to infection (Sahna and Aslan, 2014). Since human mobility is relatively lower in Yozgat province compared to many other cities, it is thought that animal mobility may also be limited. It can also be considered that the dogs in Yozgat, which mate with each other continuously, may create a situation similar to breed-specific conditions (Sahna and Aslan, 2014; Yesilbag et al., 2012).

CONCLUSION

In our findings, a seropositivity rate of 2.7% was observed in the province of Izmir. Neither antigen nor antibody presence was found in Yozgat. The presence and molecular characterization of CHV-1 antigen in Türkiye have not yet been reported. Considering all studies related to CHV-1 conducted in Türkiye over the last 15 years, the seropositivity rate is 39.01%. For a detailed understanding of CHV-1 in Turkey, future molecular detection and characterization of the disease are crucial.

ACKNOWLEDGEMENTS

This work was supported by TUBITAK (Scientific and Technological Research Council of Türkiye), [Grant Number: 1919B012204302].

REFERENCES

Acar A, Gur S, Dogan I, Akca Y 2009. A serologic investigation of Canine Herpesvirus Type 1 infection in Kangal dogs. Journal of Animal and Veterinary Advances, 8(7): 1377-1380.

Bottinelli M, Rampacci E, Stefanetti V, Marenzoni ML, Malmlov AM, Coletti M, Passamonti F 2016. Serological and biomolecular survey on canine herpesvirus-1 infection in a dog breeding kennel. Journal of Veterinary Medical Science, 78(5): 797-802.

Carmichael LE, Squire RA, Krook L 1965. Clinical and pathologic features of a fatal viral disease of newborn pups. American Journal of Veterinary Research, 26(113): 803-814.

Cobzariu D, Necula GA, Bărăităreanu S, Stefan G, Danes D 2018. Canine herpesvirus-1 specific seroconversion and clinical aspects in kennel dogs from Romania. Scientific Works. Series C, pp. 23-28.

Dahlbom M, Johnsson M, Myllys V, Taponen J, Andersson M 2009. Seroprevalence of canine herpesvirus-1 and Brucella canis in Finnish breeding kennels with and without reproductive problems. Reproduction in Domestic Animals, 44(1): 128-131.

Day MJ, Carey S, Clercx C, Kohn B, MarsilIo F, Thiry E, Freyburger L, Schulz B, Walker DJ 2020. Aetiology of canine infectious respiratory disease complex and prevalence of its pathogens in Europe. Journal of comparative pathology, 176: 86-108.

Erles K, Brownlie J 2005. Investigation into the causes of canine infectious respiratory disease: antibody responses to canine respiratory coronavirus and canine herpesvirus in two kennelled dog populations. Archives of virology, 150(8): 1493.

Erles K, Dubovi EJ, Brooks HW, Brownlie J 2004. Longitudinal study of viruses associated with canine infectious respiratory disease. Journal of Clinical Microbiology, 42(10): 4524-4529.

Gracin K, Maljkovic MM, Barbic L, Staresina V, Lojkic M, Stevanovic V 2023. A retrospective cohort study of canid herpesvirus-1 seroprevalence and epidemiology in kennel dogs in Croatia. Research in Veterinary Science, 158: 117-123.

Gur S, Acar AA 2007. Serologic investigation of Canine Herpesvirus-1 (CHV-1) infection in stray dogs in Afyon Province. Firat University Health Sciences Journal, 21(1): 37-40.

Kaczorek E, Schulz P, Małaczewska J, Wójcik R, Siwicki AK, Stopyra A, Lew-Kojrys S, Pomianowski A, Hryniewicka K, Mikulska-Skupień E 2017. Prevalence of respiratory pathogens detected in dogs with kennel cough in Poland. Acta Veterinaria Brno, 85(4): 329-336.

Krogenæs A, Rootwelt V, Larsen S, Renström L, Farstad W, Lund A 2014. A serological study of canine herpesvirus-1 infection in a population of breeding bitches in Norway. Acta Veterinaria Scandinavica, 56(1): 1-7.

Lavan R, Knesl O 2015. Prevalence of canine infectious respiratory pathogens in asymptomatic dogs presented at US animal shelters. Journal of Small Animal Practice, 56(9): 572-576.

Matsuu A, Yabuki M, Aoki E, Iwahana M 2020. Molecular detection of canine respiratory pathogens between 2017 and 2018 in Japan. Journal of Veterinary Medical Science, 82(6): 690-694.

Musayeva K, Šengaut J, Petkevičius S, Malakauskas A, Gerulis G, Šalomskas A 2013. Seroprevalence of canine herpes virus in Lithuanian dog population. Veterinarija ir Zootechnika, 61(83): 48-52.

Nöthling JO, Hüssy D, Steckler D, Ackermann M 2008. Seroprevalence of canine herpesvirus in breeding kennels in the Gauteng Province of South Africa. Theriogenology, 69(3): 276-282.

Okonkowski LK, Szlosek D, Ottney J, Coyne M, Carey SA 2021. Asymptomatic carriage of canine infectious respiratory disease complex pathogens among healthy dogs. Journal of Small Animal Practice, 62(8): 662-668.

Pratelli A, Colao V, Losurdo M 2014. Serological and virological detection of canine herpesvirus-1 in adult dogs with and without reproductive disorders. The Veterinary Journal, 200(2): 257-260.

Rezaei M, Jajarmi M, Alizadeh R, Khalili M, Babaei H 2020. First molecular study of Canine Herpesvirus-1 in reproductive specimens of adult dogs in southeast of Iran. Comparative Immunology, Microbiology and Infectious Diseases, 71: 101487.

Ronsse V, Verstegen J, Onclin K, Guiot AL, Aeberlé C, Nauwynck HJ, Poulet H 2002. Seroprevalence of canine herpesvirus-1 in the Belgian dog population in 2000. Reproduction in domestic animals, 37(5): 299-304.

Ronsse V, Verstegen J, Thiry E, Onclin K, Aeberlé C, Brunet S, Poulet H 2005. Canine herpesvirus-1 (CHV-1): clinical, serological and virological patterns in breeding colonies. Theriogenology, 64(1): 61-74.

Rota A, Dogliero A, Biosa T, Messina M, Pregel P, Masoero L 2020. Seroprevalence of canine herpesvirus-1 in breeding dogs with or without vaccination in Northwest Italy. Animals, 10(7): 1116.

Sahna KC, Aslan O 2014. Seroprevalence of canine herpesvirus-1 in Anatolian Shepherd dogs and German Shepherd dogs. Firat University Health Sciences Veterinary Journal, 28(3): 107-110.

Sambrook J, Fritsch EF, Maniatis T 1989. Molecular cloning: a laboratory manual (No. Ed. 2). Cold Spring Harbor Laboratory Press.

Yapici O, Avci O, Levent O, Hasircioglu S 2018. Detection of canine herpesvirus infection on dogs. Microbiology Research Journal International, 24(1): 1-6.

Yesilbag K, Yalçın E, Tuncer P, Yılmaz Z 2012. Seroprevalence of canine herpesvirus-1 in Turkish dog population. Research in veterinary science, 92(1): 36-39.

DRAFT

ORAL PRESENTATION

Study of environmental state in Vlora Bay, Albanian coast

Vanela Prifti[1*],Sonila Kane[2],Pranvera Lazo[2]

[1] National Environmental Agency, Sector of Sampling and Laboratory Analyses, Tirana, Albania.
[2] University of Tirana, Faculty of Natural Sciences, Department of Chemistry, Tirana, Albania.

*Corresponding Author: Vanela.Gjeci@akm.gov.al

Abstract

Vlora Bay is the most popular and frequented tourist destination in the south of Albania. The assessment of ecological status involves the assessment of the quality of the structure and functioning of surface water ecosystems. To reach good ecological and chemical status of surface waters according to WFD, the reduction of water pollution is crucial and a main topic in water management. Those pollutants are caused by diffuse sources of pollution and point sources of pollution. Point source pollution is mainly caused by urban inhabitants with discharges from waste water treatment plants, the activity along the coastal area.(3) In our study we have monitored three sampling sites positioned at a distance of 10 m from the coastal line, one site positioned at the delta of the Vjosa River estuary discharging site, and a control site positioned 100 m away from the coastal line have been selected to assess the water quality of the Vlora Bay estuary. Seawater quality is assessed based on physicochemical parameters such as: water temperature, pH, total suspended solids, dissolved oxygen, COD, BOD_5 and total phosphorous (according to Albanian Standard DCM 177, dated March 31, 2005). The main significant pressures on surface water bodies are untreated discharge in the receiving area, nutrient enrichment, chemical pollution and altered habitats due to morphological changes.

INTRODUCTION

Water is vital for the three pillars of Europe's sustainable growth: its society, its economy, and its environment. All three pillars depend on the adequate availability and supply of water of sufficient quality at the right time and in the right location hr eutrophication of marine ecosystems, including coastal and estuarine areas, is affected by nutrient over-enrichment from nitrogen and phosphorus coming from land-based sources, marine activities, and atmospheric deposition, as well as fluxes from neighbouring water bodies. The resulting excessive availability of nitrogen and phosphorus accelerates the growth of microalgae and higher forms of plant life, with indirect effects on aquatic organisms and water quality.
Vlora is very rich in historical and archaeological landmarks and from natural beauty point of view, one of the most attractive tourist centres in the Eastern part of the Mediterranean Sea. In the last 10 years the city economy has been marked by a construction booming that was directed to build new residences for an increasing population and to accommodate tourists, and it was the main destinations over the recent years. Due to the high urbanization and development in Vlora there are many problems associated with unmanaged urbanization, such as pollution, weak sewerage infrastructure and waste disposal, poor water supply, presence of illegal settlements and traffic congestion.
This study aims at assessing the quality of the coastal water in Vlora Bay, and the Water Framework Directive (WFD) 2000/60/EC of the European Parliament and the Council of October 23, 2000 and for assessing the quality of the Vjosa Estuary sampling point is classified according to the Water Framework Directive 2000/60/EC OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 23 October 2000) there are five classes of classification where Class III – Moderate state is the minimum level of water quality acceptance.

MATERIALS AND METHODS

Three sampling points in the coastal area of the Bay of Vlora, which monitor the impact of urban discharges, were selected as monitoring sites based on the scheme where the discharge collector represents the pollution source. Monitoring sites located after discharge in the receiving area, at a distance of 10 m, to assess its impact on the quality of the coastal area.The urban discharge in Vlora city are realised through the pumping station. The sampling stations of this study are shown in the table below (Table 1).

Table 1. Sampling stations in Vlora Bay

No.	Laboratory code of the sample	Area	Sampling point	Coordinates
1.	V1	Vlorë	After the pumping station	N 40.470928 E 19.458578
2.	V3	Vlorë	Plazhi i Vjeter - Kabinat	N40.453422 E 19.478732
3.	V4	Vlorë	Marine School	N 40.442729 E 19.494879
4.	V2	estuary	Vjosa Estuary	N 40.383400 E 19.192000
5.	Vcontrol	Vlore	Controlling site, marine sample	N 40.399285 E 19.419600

The observed data were compared with the Albanian Standard DCM 177, dated March 31, 2005, "The Permitted Values of Liquid Discharges and Zoning Criteria of the Water Receiving Environment." It revealed that the measured parameters did not go above the allowed limits. The water quality classification for the Vjosa estuary is done according to Water Framework Directive (WFD) 2000/60/EC of the European Parliament and the Council of October 23, 2000. The monitoring data, compared to the requirements of WFD for the nutrients, classifies the Vjosa estuary sample as in a good status (Class I). Also, the controlling site that is the sampling station Vcontrol is classified in good quality, where all the physico – chemical parameters are in low concentrations (limit of detection of the methods) as there is no negative impact and we can use this sampling site as a reference station.

Water temperature

Temperature is an important parameter for assessing water quality as it affects other water parameters and can alter the physical and chemical properties of water. This parameter is measured on site with the multimeter. The water temperatures measured varied from 13˚C to 38.8˚C, as low temperatures are measured on winter expedition and high temperature on summer measurement.

Dissolved Oxygen

Dissolved oxygen, DO, is a key parameter to determine the quality of the water and refers to the level of free, non-compound oxygen present in water. It has a strong effect to the on the organisms living within a body of water, by making it an important parameter in assessing the quality of the water bodies.
Dissolved oxygen is necessary to various forms of life including fish, invertebrates, bacteria and plants, which use oxygen in their respiration process. Fish and crustaceans obtain oxygen for respiration through their gills, while plant life and phytoplankton require dissolved oxygen for respiration when there is no light for photosynthesis. Bottom feeders, need minimal amounts of oxygen (1-6 mg/l), while shallow water fish need higher levels (4-15 mg/l). The DO is measured on site and the values are presented in the graph below assessing that in all the stations the water is saturated in oxygen and all above the WFD limit values (red line) for the “moderate” class of water.

pH

Measurement of pH is one of the most important tests in water chemistry, usually determined on site. This parameter is proportional with the logarithmic value of H+ concentration in water. pH ranged between 0 and 14 and defines how acidic or basic a body of water is along a logarithmic scale. In the coastal sampling stations the values measured are 7.24 – 8.1, and at Vjosa River estuary pH is measured 7.85.

Biochemical Oxygen Demand - BOD_5

Biochemical oxygen demand is a key parameter to determine the scale of the quality of water and is determined by a respirometric method with oxi-top system inhibited for 5 days in the incubator. The results for the three monitoring stations in the coastal samples is assessed higher of the allowed values only in station V1 – the pumping station as shown in the graph below(Fig.1).
At Vjosa estuary the BOD_5 is estimated below the detection limit (< 1 mgO/l) classifing in High State (Class I).
High BOD_5 values are as as a result of the urban discharges without any treatment that are domestic and industrial sewage, where the process of decomposition of organic matter requires oxygen, the amount of organic matter in river can be measured in terms of oxygen demand (biochemical oxygen demand BOD and chemical oxygen demand COD) concluding in high rates of BOD and COD.

Suspended solids

Suspended solids refers to small particles which remains in suspension in water as a colloid or due to motion of the water, where the suspended particles that are not dissolved in a sample water and is determined by gravimetric method. As can be seen from the graph (Fig.5) the suspended solids determination is within the limits.

Ammonia

Ammonium determination is performed with the spectrophotometric method. Ammonium concentrations are normally raised as a result of organic pollution, caused by discharges from wastewater, industrial effluents and agricultural runoff. It exerts a demand on oxygen in water as it is transformed to oxidised forms of nitrogen. In addition it is toxic to aquatic life at certain concentrations in relation to water temperature, salinity and pH.

Nitrite/Nitrate

Nitrite concentration originates from fertilizers through run-off water, sewage and mineral deposits.
Nitrate in water in the most common cases originates primarily from fertilizers, septic systems, and manure storage or manure spreading operations.

Phosphorous

Phosphorus is an essential element for plant life, but when there is too much of it in water, it can speed up eutrophication of the aquatic environment. Phosphorus originates from a variety of sources, many of which are related to human activities where major sources include human and animal wastes, detergents, and agricultural runoff. Phosphorus is measured in two ways, as ortho-phosphate (soluble reactive phosphorus) and total phosphorus.
According to the measured values we have higher concentration of total phosphorous in V1 – pumping station in Vlora, in two other stations it is within the limits. The monitoring value for Vjosa estuary is at very low concentration assessed as limit of detection.

RESULTS AND DISCUSSION

At Table 2 and 3 are presented the monitoring data of the five sampling stations at Vlora Bay, and in Figures 1,2 and 3 are shown the monitoring data in a graphical form.

DRAFT

Table 2. The monitoring data of the parameters monitored in situ.

Parameters determined	Water temperature	pH	Salinity	Conductivity	Dissolved Oxygen
	°C		mg/l	mS/cm	mg/l
V1	20.4	7.2	0.9	1913	4.6
V3	21.4	8.1	35.2	53.7	10.5
V4	20.6	8.1	35.4	54.4	8.7
V2(Vjosa)	18.4	7.9	37.1	57	13
Vcontrol	19.6	8.2	36.3	67.8	11.2

Table 3. The monitoring data of the parameters monitored in the laboratory.

Parameters determined	Alkalinity	Conductivity	TSS	COD	BOD_5	NH_4^+	NO_2	NO3	$P\text{-}PO_4$	P_{total}
	mg/l	mS/cm	mg/l	mg/l	mg/l	mg/l	mg/l	mg/l	mg/l	mg/l
V1	328	1913	33	24	14	6.55	0.0088	0.871	0.526	0.6
V3	141	53.7	21.7	4	2	0.35	0.013	0.427	0.019	0.023
V4	131	54.4	23.3	3	1	0.40	0.0096	0.411	0.021	0.025
V2(Vjosa)	152	57	108	<1	< 1	0.009	0.002	0.423	0.003	0.0062
Vcontrol	147	67.8	28.4	< 1	0.12	0.014	0.005	0.018	0.004	0.0058

Table 4. Statistical processing data using descriptive statistic (* expressed in mg/l, ** in µS/cm).

Variable	Mean*	StDev	CV%	Min*	Q1*	Median*	Q3*	Max*
Water temperature	20.08	1.137	5.66	18.4	19	20.4	21	21.4
pH	7.9	0.406	5.14	7.2	7.55	8.1	8.15	8.2
Salinity	28.98	15.72	54.23	0.9	18.05	35.4	36.7	37.1
Alkalinity	179.8	83.2	46.28	131	136	147	240	328
Conductivity**	429	829	**193.28**	54	54	57	990	1913
TSS	42.9	36.7	**85.53**	21.7	22.5	28.4	70.5	108
DO	9.6	3.19	33.24	4.6	6.65	10.5	12.1	13
COD	10.33	11.85	**114.64**	3	3	4	24	24
BOD_5	4.28	6.53	**152.46**	0.12	0.34	1.5	11	14
NH4	1.46	2.85	**194.5**	0.01	0.01	0.35	3.48	6.55
NO2	0.00768	0.00426	55.51	0.002	0.0035	0.0088	0.0113	0.013
NO3	0.43	0.302	70.22	0.018	0.214	0.423	0.649	0.871
P-PO4	0.115	0.23	**200.81**	0.003	0.004	0.019	0.274	0.526
Ptotal	0.132	0.262	**198.31**	0.006	0.006	0.023	0.313	0.6

DRAFT

Table 4 gives us the information about the dispersion to have a better understanding of our environmental monitoring data at Vlora Bay. Based on coefficient of variation data, it is shown that water temperature, pH, salinity, alkalinity, DO, nitrite, and nitrate are relatively stable along river basin (CV% ranged between 25 to 75%). Other parameters, conductivity, TSS, COD, BOD_5, and NH_4^+ revealed high variability by indicating high anthropogenic inputs in Ishmi River (Figure 4)

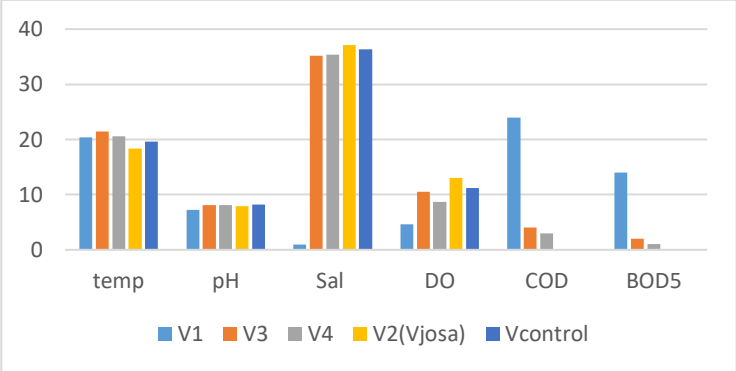


Figure 1. Temp, pH, salinity, DO, COD and BOD

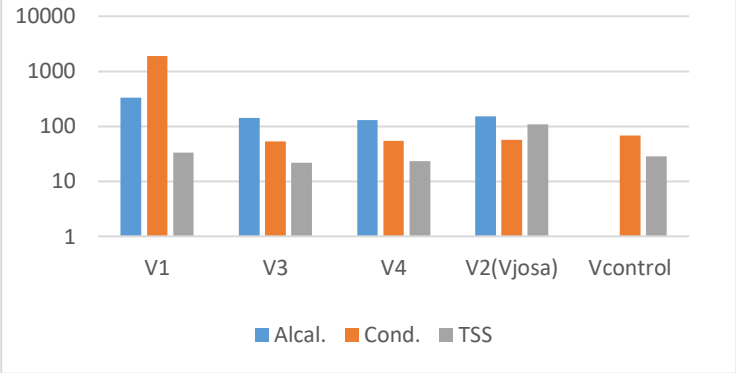


Figure 2. Alkalinity, conductivity and TSS parameters

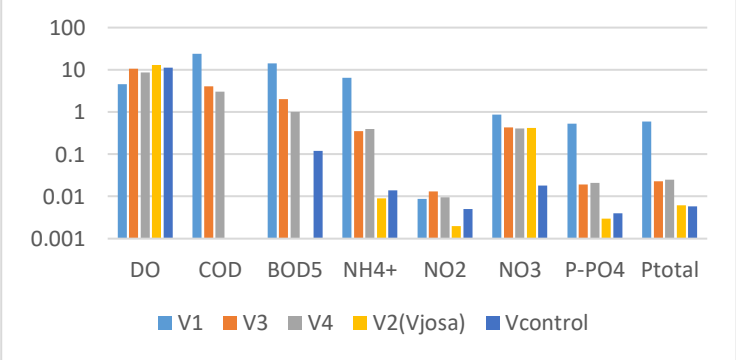
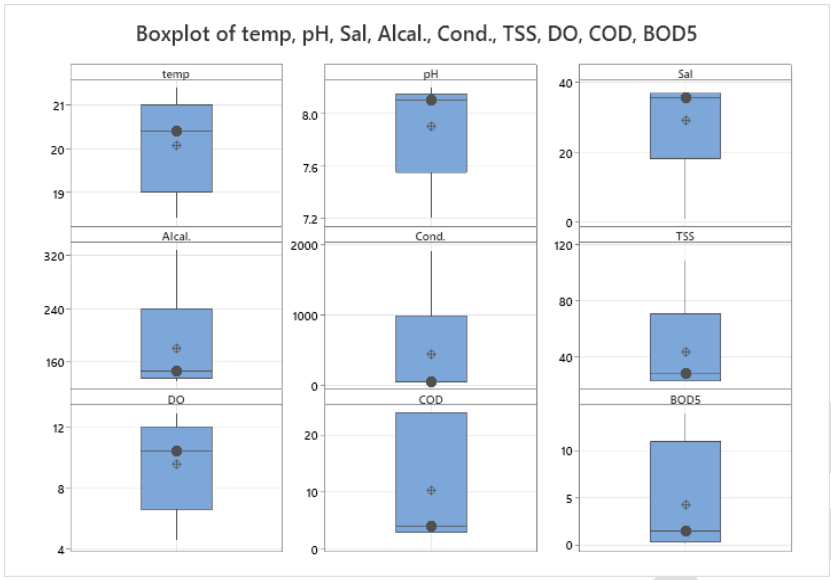


Figure 3. DO, COD, BOD, and the nutrients.

As it is expected, the highest values of BOD5 and COD measured in station V1 – After the pumping station indicates for organic pollution resulting in poor quality of the monitored station. Also, high concentration of total suspended solids and total phosphorous are measured. The monitoring results indicate the presence of urban discharges above the allowed limits.



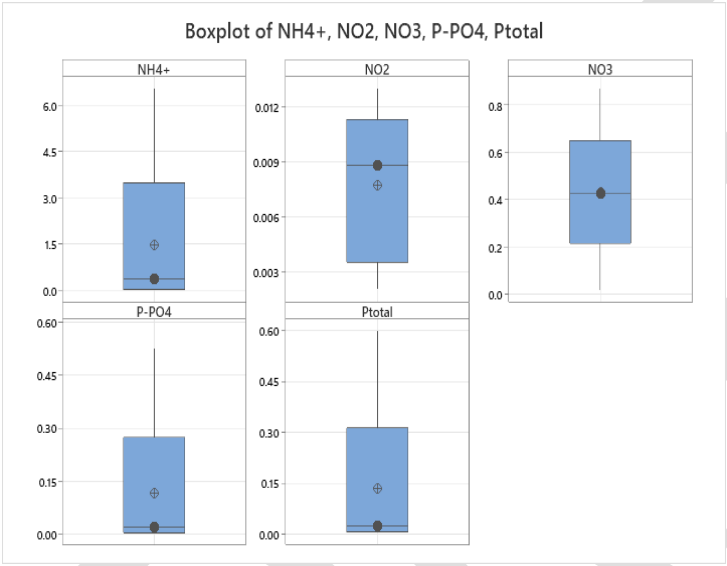
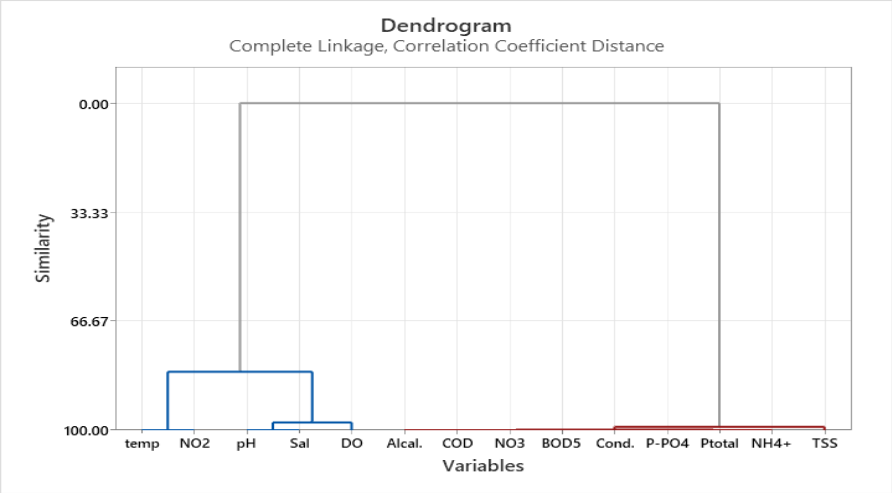


Figure 4: Boxplots of the monitoring data in Vlora Bay

Table 5: The correlation of the monitoring data in Pairwise Pearson Correlations

Sample 1	Sample 2	N	Correlation	P-Value
NO2	temp	5	0.991	0.001
Sal	pH	5	0.955	0.011
Alcal.	pH	5	-0.970	0.006
Cond.	pH	5	-0.962	0.009
COD	pH	3	-0.999	0.027
BOD5	pH	4	-0.998	0.002
NH4+	pH	5	-0.957	0.011
NO3	pH	5	-0.890	0.043
P-PO4	pH	5	-0.960	0.010
Ptotal	pH	5	-0.961	0.009
Alcal.	Sal	5	-0.991	0.001
Cond.	Sal	5	-0.999	0.000
DO	Sal	5	0.895	0.040
COD	Sal	3	-0.999	0.024
BOD5	Sal	4	-0.996	0.004
NH4+	Sal	5	-1.000	0.000
P-PO4	Sal	5	-1.000	0.000
Ptotal	Sal	5	-1.000	0.000
Cond.	Alcal	5	0.996	0.000
COD	Alcal	3	1.000	0.002
BOD5	Alcal	4	0.988	0.012
NH4+	Alcal	5	0.988	0.002
P-PO4	Alcal	5	0.992	0.001
Ptotal	Alcal	5	0.992	0.001
COD	Cond	3	0.999	0.027
BOD5	Cond	4	0.992	0.008
NH4+	Cond	5	0.998	0.000
P-PO4	Cond	5	0.999	0.000
Ptotal	Cond	5	0.999	0.000
NH4+	DO	5	-0.900	0.037
P-PO4	DO	5	-0.890	0.043
Ptotal	DO	5	-0.889	0.044
BOD5	COD	3	1.000	0.017
NH4+	COD	3	0.999	0.031
NO3	COD	3	1.000	0.007
P-PO4	COD	3	0.999	0.029
Ptotal	COD	3	0.999	0.029
NH4+	BOD_5	4	0.997	0.003
P-PO4	BOD_5	4	0.995	0.005
Ptotal	BOD_5	4	0.995	0.005
P-PO4	NH_4^+	5	1.000	0.000
Ptotal	NH_4^+	5	1.000	0.000
Ptotal	$P\text{-}PO_4^{3-}$	5	1.000	0.000



Variables	
Cluster 1	temp pH Sal DO NO2
Cluster 2	Alcal. Cond. TSS COD BOD5 NH4+ NO3 P-PO4 Ptotal

Figure 6: The dendogram Cluster1 and Cluster 2

Figure 6 presents the Cluster 1 and 2 analysis which is done based on Pearson correlation coefficients (Figure 5), as the variables shows that the parameters under monitoring are clustered into 2 groups (Complete Linkage; Correlation Coefficients Distance).
Cluster 1 has grouped together in a similarity level between the parameters: water temperature, pH, salinity, dissolved oxygen and nitrite. Cluster 2 has grouped together in a similarity level between the parameters like: alkalinity, conductivity, total suspended solids, COD, BOD, ammonia, nitrate, ortho – phosphorous and total phosphorous.

For the classification of the sampling station V2 - Vjosa estuary we have compared the monitoring data with the maximum permitted values of the EU WFD 2000/60/EC referring to the classification of the surface water, which classify the water in 5 classes.
Class I – high state;
Class II – good state;
Class III – moderate state;
Class IV – poor quality; and
Class V – bad quality/immediate measures.
The quality status of Vjosa estuary sample according to EU WFD resulted as follows:

Table 5: Classification of Vjosa estuary sample according to EU WFD

No.	Parameter	Values measured	Norm of WFD	Classification
1.	pH	7.85	9	Alkaline
2.	Dissolved oxygen*	13	>5	High oxygen saturation
3.	BOD_{5*}	< 1	< 7	Class I – high state
4.	Ammonia*	0.009	< 0.6	Class I – high state
5.	Nitrite*	0.002	< 0.12	Class I – high state
6.	Nitrate	0.423	< 4	Class I – high state
7.	Ortho-phosphorous*	0.003	< 0.2	Class I – high state
8.	Total phosphorous*	0.0062	< 0.4	Class I – high state

* mg/l

The measurements indicate that Vjosa estuary monitoring station is in high state – excellent water quality (Table 5).

All the data monitored in three sampling stations in Vlora Bay gives us the information that in the first station V1 – which is the pumping station of the city for the parameters monitored classifies this station not in a good quality as there is low dissolved oxygen, high in BOD, suspended solids and total phosphorous as a result of the urban discharges without any treatment that are domestic and industrial sewage.
The chemical oxygen demand is high but within the limited values. The controlling site indicates good quality as there is no pressure and no activity in the sampling point.

CONCLUSION

Assessing all the monitoring data we face that there is a high contamination from anthropogenic factors (domestic and activities operating in the area) like urban wastewater dischargement that goes directly into the receiving environment without the right treatment contributing in poor quality of the water, as the requirements for reaching a good environmental status is not respected.
We must admit that the environment pollution is health risk for the population and in coastal area is dangerous for the bathers.
Main sources of pollution are:

Direct dischargement of wastewater and high scale of urbanization
Rapid development of tourism
Hotels and other activities that operates along the coast which do not respect the environmental permit requirements

ACKNOWLEDGEMENTS

All the analytical procedure, sampling and analyses are carried out at the National Environmental Agency in Albania depending from the Ministry of Tourism and Environment.

REFERENCES

1.Albanian Standard DCM 177, dated March 31, 2005, "The Permitted Values of Liquid Discharges and Zoning Criteria of the Water Receiving Environment
2. Drivers and pressures of selected key water management challenges – European Environmental Agency Overview – 9 November 2020 - Author: Eleftheria Kampa1, Jeanette Völker2, Ulf Stein1, Volker Mohaupt2
3. Directive 2000/60/EC of the European Parliament and of the Council of 23 October 2000 establishing a framework for Community action in the field of water policy. European Commission PE-CONS 3639/1/100 Rev 1, Luxembourg.
4.Directive 2008/105/EC of The European Parliament and of the Council on environmental quality standards in the field of water policy, amending and subsequently repealing Council Directives 82/176/EEC, 83/513/EEC, 84/156/EEC, 84/491/EEC, 86/280/EEC and amending Directive 2000/60/EC of the European Parliament and of the Council.
5.EU Directive 2000/60/EC 2004 - 2006. (2005): Common implementation strategy for the Water Framework Directive (2000/60/EC). Guidance on the intercalibration process 2004-2006. 2005: 14: 31.
6. National Environment Agency State Report 2022; www.akm.gov.al
7. Standard methods for the examination of water and wastewater 23th Edition Baird, R., & Bridgewater, L. (2017)

ORAL PRESENTATION

Avokadonun (*Persea americana*) bakırın asidik ortamdaki korozyonuna etkisinin araştırılması

Ece Altunbaş Şahin[1*] (*ORCID* https://orcid.org/ 0000-000232010487), Yeşim Aydın Dursun[2] (*ORCID* https://orcid.org/0000-0003-2052-8817)[1], İbrahim Halil Geçibesler[3] (*ORCID* https://orcid.org/ 0000-0002-4473-2671), Ramazan Solmaz[3] (*ORCID* https://orcid.org/ 0000-0002-9295-1203)

[1] Bingöl Üniversitesi, Genç MYO, Sivil Savunma ve İtfaiyecilik Programı, 12000, Bingöl/Türkiye

[2] Bingöl Üniversitesi, Fen Edebiyat Fakültesi, Kimya Bölümü, 12000, Bingöl/Türkiye

[3]Bingöl Üniversitesi, Sağlık Bilimleri Fakültesi, İş Sağlığı ve Güvenliği Bölümü, 12000, Bingöl/Türkiye

*Ece Altunbaş Şahin (ealtunbassahin@gmail.com, easahin@bingol.edu.tr

Özet

Bakır ve alaşımları, insanlık tarihinin gelişmesinde ve ilerlemesinde; önemli ve belirleyici role sahiptir. Kolay ve düşük maliyetle üretilmesi, yüksek termal ve elektriksel iletkenliğe sahip olması, bakır metaline, başta kimya ve mikro elektronik uygulamaları olmak üzere çok sayıda farklı alanda kullanım alanı oluşturmuştur. Bakır malzemenin kullanımını kısıtlayan en önemli faktör ise korozyon eğiliminin fazla olmasıdır. Korozyonu tamamen engellemek mümkün olmamakla birlikte; farklı uygulamalar ile hızını azaltmak mümkündür. Organik inhibitörler kullanılarak korozyon hızını yavaşlatmak mümkündür. Organik inhibitörlerin yüksek maliyetleri doğa ve çevreye verdiği zarar, onların kullanımını sınırlandırmaktadır İnhibitörler kullanıldıkları ortamlarda önemli sorunlar oluşturmazlarken, kullanıldıkları sistemlerde ortaya çıkacak kaçaklar, boşaltma ya da temizlik gibi işlemler inhibitörlerin doğaya karışmasına neden olmaktadır. Bunun için inhibitör seçimi yaparken çok titiz davranmak gerekir. Bu durum, tamamen doğal ürünler olan ve çevreye herhangi bir zararları bulunmayan, bitki ve bitki atıklarını (kök, gövde, yaprak, çekirdek, kabuk ve çiçek) korozyon inhibitörü uygulamalarında ön plana çıkartmaktadır. Yapılan çalışmalar, bitki ekstrelerin, inhibisyon etkinliği yüksek doğal olarak sentezlenmiş, amino asitler, organik asitler, alkaloidler, polifenonller ve tananler açısından oldukça zengin kaynaklar olduğu belirlenmiştir. Bu anlamda ekonomik, çevresel fayda sağlayan, çevresel olarak güvenli tabir edilen bitki ekstrelerinin inhibitör olarak kullanılması yaygınlaşmaktadır. Bu çalışmada farklı derişimlerde hazırlanan avokado (metanol içerisinde) ekstarktının, bakırın korozyon hızına etkisi 0,5 M H_2SO_4 çözeltisinde, E_{ocp}-zaman, polarizasyon eğrileri ve elektrokimyasal impedans spektroskopisi ile değerlendirilmiştir. İnhibitörün karakterizasyonu SEM ve temas açısı ölçümleri ile belirlenmiştir. Sonuçlar, avokado ektraktının, bakırın korozyonunu yavaşlattığı ve oluşan filmin yüzeye sıkıca tutunduğunu göstermektedir.

Anahtar Kelimeler: Korozyon, yeşil inhibitör, avokado

Investigation of the effect of avocado (*Persea Americana*) on the corrosion of copper in acidic environment

Abstract

Copper and its alloys play a significant and defining role in the development and progress of human history. Their ease of production at low cost and their high thermal and electrical conductivity have given copper metal a wide range of applications, particularly in fields such as chemistry and microelectronics. The most significant factor limiting the use of copper material is its high susceptibility to corrosion. While it is not possible to completely eliminate corrosion, it is possible to reduce its rate through various applications. The corrosion rate can be slowed down by using organic inhibitors. However, the high cost of organic inhibitors and the environmental damage they cause restrict their usage. Inhibitors themselves do not pose significant issues in the environments where they are used, but leaks, discharge, or cleaning procedures in the sistem where they are applied can lead to the introduction of inhibitors into the environment. Therefore, it is crucial to be extremely careful when selecting inhibitors. This situation highlights the prominence of corrosion inhibitors derived from entirely natural sources, such as plant and plant residues (roots, stems, leaves, seeds, bark, and flowers), which have no adverse effects on the environment. Studies have shown that plant extracts are rich

sources of naturally synthesized compounds with high inhibition efficacy, including amino acids, organic acids, alkaloids, polyphenols, and tannins. In this context, the use of plant extracts as inhibitors is becoming more widespread, offering economic and environmental benefits and being considered environmentally safe. In this study, the effect of avocado extract prepared at different concentrations (in methanol) on the corrosion rate of copper was evaluated in a 0.5 M H_2SO_4 solution using E_{ocp}-time, polarization curves, and electrochemical impedance spectroscopy. The characterization of the inhibitors was determined through SEM and contact angle measurements. The results indicate that avocado extract slows down the corrosion of copper and tightly adheres to the surface, forming a protective film.

Keywords: corrosion, green inhibitor, avocado

GİRİŞ

Bakır, dünyanın bir çok yerinde rezerv olarak bulunan ve üretimi bölgesel anlamda sorun oluşturmayan bir metaldir. Bakır ve alaşımları; mükemmel termal iletkenlik, nisbeten iyi süneklik, kimyasal kararlılık ve iyi elektrik iletkenliklerinden dolayı başta, ısı değişim malzemeleri, kablo gibi elektriksel aksamlarda, inşaatlarda ve yapı malzemelerinde yaygın olarak tercih edilirler [1]. Kullanımları süresince geçen zamanla birlikte, bakır doğrudan yada dolaylı olarak korozyona uğrayabilir. Korozyon olayı bakır metal ekipmanın hizmet ömrünün büyük ölçüde kısalmasına neden olur. Ömrü kısalmış ve hatta hurdaya çıkarılmış metalik malzemeler büyük ekonomik kayıplara yol açacaktır. Bu durum, üretimin durmasına, çalışma ortamının bozulmasına, kaynak tüketimine, ürün kalitesinin düşmesine, çevre kirliliğine neden olacak, yeni teknolojilerin gelişimini büyük ölçüde etkileyecektir [2]. Bakır metalinin, korozyona karşı korunmasında, inhibitör kullanımı en yaygın ve popüler yöntemlerden bir tanesidir. İnhibitörler, korozif etkiyi azaltmak veya önlemek için korozyon ortamına katılan maddelerdir. Son yıllarda dünyanın her yerinde özellikle ekolojik çevreye ve insan güvenliğine daha fazla önem verilmektedir. Bu nedenle, korozyon inhibitörleri ile yapılan çalışmalar yeni, yeşil ve yüksek verimliliğe sahip çevre dostu maddelerin tercih edilmesine doğru ilerlemektedir. Bu kapsamda özellikle bitki ekstraktlarının doğal ortamdan kolayca elde edilmeleri, parçalanabilir olması ve yenilenebilir olması araştırmacıların ilgisini çekmektedir [3,4]. Bu çalışmada avokadonun (*Persea Americana*) (A) asidik ortamda bakırın korozyonuna inhibisyon yetkisi, değişik elektrokimyasal teknikler ile araştırılmıştır. Bakır yüzeylerinin yapısı SEM ve temas açısı teknikleri ile aydınlatılmıştır.

MATERYAL VE METOT

Bu çalışmada kullanılan avokado meyvesinin etli bölümleri diğer kısımlarından ayrılmıştır. Bu etli bölümlerin kurutulması, baskısız kağıt üzerinde güneş ışığı almayan havada bir odada gerçekleştirilmiştir. Kurutulan materyaller laboratuvar tipi değirmende öğütülerek (10 mesh) tanecik boyutu küçültülmüştür. Her bir ekstrakt materyalinden 20 g tartılarak amber cam flasklara alınmıştır. Ekstrakt materyalinin üzerine 400 mL metanol:kloroform (1:1;v:v) ilave edilerek laboratuvar koşullarında ekstrakte edilmştir. Ekstraksiyon çözücüsü döner buharlaştırıcı kullanılarak alçak basınç altında 40°C'yi aşmayan sıcaklıklarda uzaklaştırılmıştır.

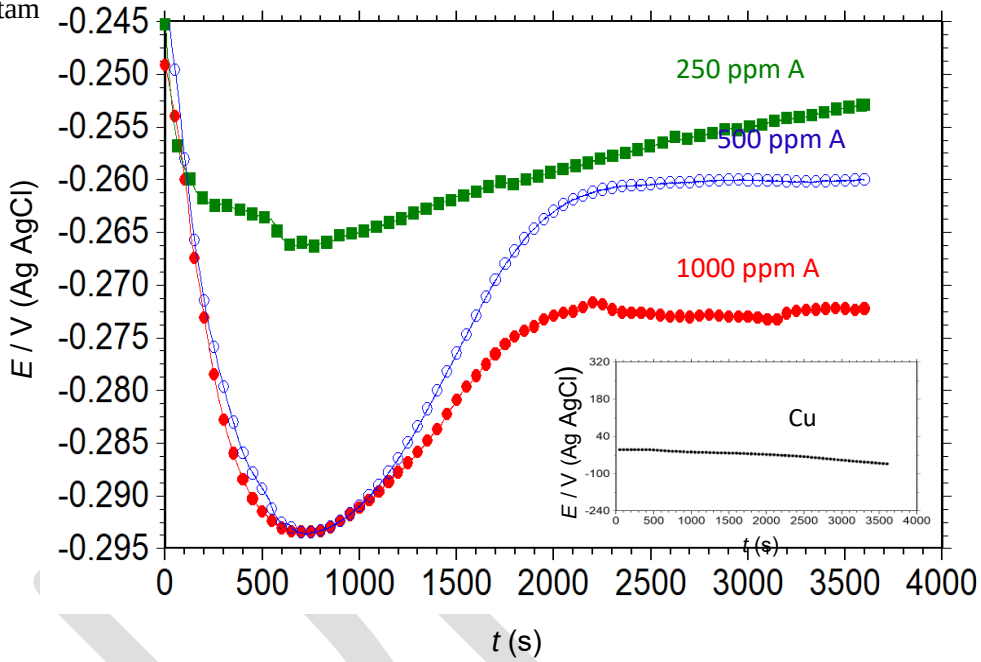
A'nın 0,5 M H_2SO_4' de bakır metalinin korozyonuna etkisi üç elektrot tekniği kullanılarak belirlenmiştir. Çalışma elektrotu olarak 0,283 cm^2 yüzey alanı silindirik bakır (%99,9), karşı elektrot olarak Pt levha (1 x 1 cm^2) ve referans elektrot olarak Ag/AgCl, KCl (3,0 M KCl) kullanılmıştır. Çalışma elektrotları 5 cm uzunluğunda kesilerek, bir yüzüne iletkenliği sağlamak için bakır tel geçirilmiştir. Diğer yüzü açıkta kalacak şekilde poliester blok içerisinde gömülmüştür. Tüm elektrokimyasal deneyler 273 K de ve atmosfere açık koşullarda, en az üç tekrar olacak şekilde CHI 6096E (Electrochemical Analyzer, Seri No. R0679) marka elektrokimyasal analizör cihazı kullanılarak, inhibitör içermeyen ve farklı derişimlerde (1000 – 50 ppm) A içeren 0,5 M H_2SO_4 çözeltilerinde gerçekleştirilmiştir. İnhibitörlü ve inhibtörsüz ortamlarda, çalışma

elektrotlarının açık devre potansiyelleri ve elektrot potansiyellerinin dengeye geldiğini belirlemek için E_{ocp}-t eğrileri 1 saat boyunca kaydedilmiştir.

E_{ocp}-*t* eğrileri 1 saat boyunca kaydedildikten hemen sonra elektrokimyasal impedans (EIS) ölçümleri, okunan açık devre potansiyelinde 100 kHz frekanstan başlayarak 5 mV genlik ile yapılmıştır. EIS ölçümleri biter bitmez aynı sistemde lineer polarizasyon (LPR) ölçümleri yapılmıştır (1 mV s^{-1}). Elde edilen akım-potansiyel eğrilerinin eğiminden polarizasyon dirençleri (R_p) hesaplanmıştır. Potansiyodinamik polarizasyon (PPE) ölçümleri, LPR ölçümleri biter bitmez aynı sitemde yapılmıştır. Ölçümler açık devre potansiyelinden başlanarak, anodik ve katodik yönlere doğru 1 mV s^{-1} tarama hızı ile yapılmıştır. İnhibitörlü ve inhibitörsüz ortamda bakır elektrodun yüzey analizleri taramalı elektron mikroskopu (SEM) ve temas açısı ölçümleri ile yapılmıştır.

BULGULAR ve TARTIŞMA

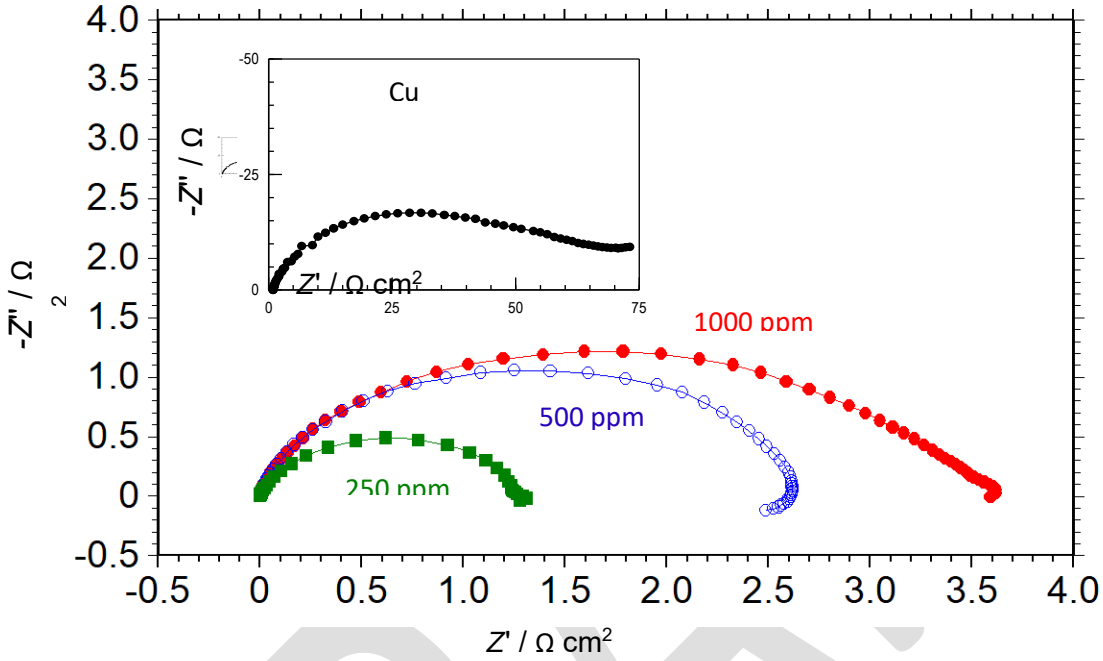
Bakır elektrot, inhibitor içermeyen ve farklı derişimlerde A inhibitörü içeren 0,5 M H_2SO_4 çözeltisinde 1 saat boyunca daldırılarak, E_{ocp}-t eğrileri takip edilmiştir (Şekil 1). Şekil 1'de görüldüğü gibi, inhibitör içeren ortam e olduğu belirlenmiştir.



Şekil 1. Bakır elektrotun 0,5 M H_2SO_4 ve farklı derişimlerde A içeren 0,5 M H_2SO_4 çözeltilerinde elde edilen E_{ocp}-t eğrileri

Bakır için inhibitor içermeuen ve farklı derişimlerde (1000 ppm, 500 ppm ve 250 ppm) A içeren 0,5 M H_2SO_4 çözeltisinde 1 saat daldırma süresi sonunda elde edilen Nyquist eğrileri Şekil 2'de verilmiştir. Şekil 2'de verilen Nyqusit eğrileri incelendiğinde; inhibitör içeren ortamlarda, her derişim için eğrinin şekli değişmemekle birlikte inhibitör içermeyen ortamla kıyaslandığında direncin önemli ölçüde artığı görülmektedir. İnhibitör içeren ortamlarda R_p değerleri artan derişimle birlikte artmaktadır. Bakır için Nyquist eğrilerinden hesaplanan R_p değeri 77 ohm cm^2'dir. En yüksek derişim için belirlenen R_p değeri 3578 ohm cm^2'dir. Bu artış, inhibitörün metal yüzeyine adsorplanıp koruyucu bir film oluşturduğunu göstermektedir Ayrıca Tablo 1'de görüldüğü gibi LPR ölçümlerinden elde edilen R_p değerleri, Nyquist eğrilerinden elde edilen değerler ile uyumludur. Metal/çözelti ara yüzeyinde inhibitör moleküllerinin adsorpsiyonu, R_p değerlerini arttırmaktadır. R_p'nin artması, bakırın korozyon hızının azaldığını göstermektedir.

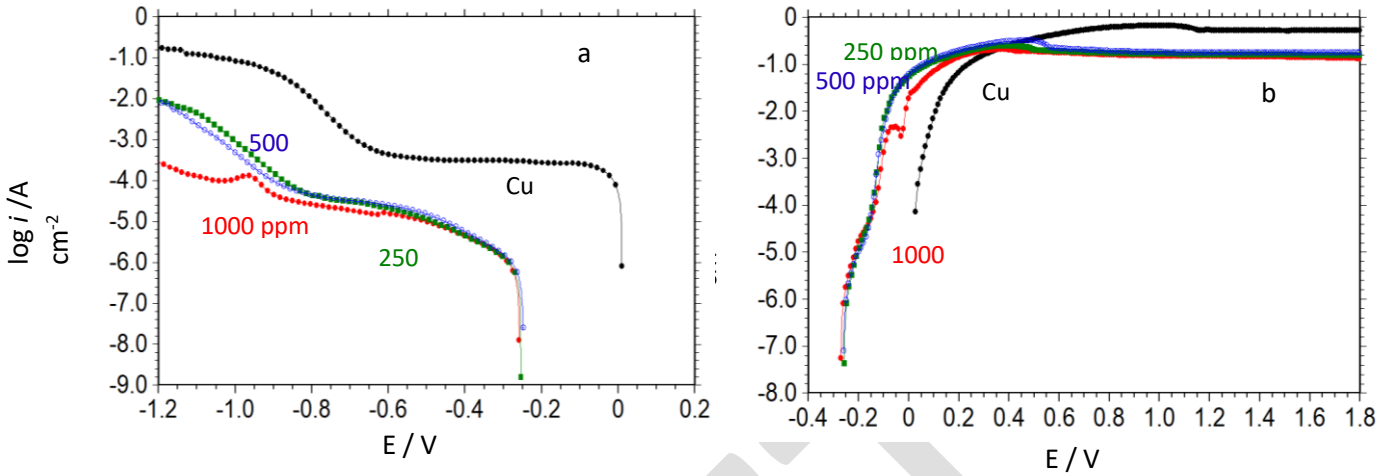
İnhibitör içermeyen ve farklı derişimlerde (1000 ppm, 500 ppm ve 250 ppm) A içeren 0,5 M H_2SO_4 çözeltisinde bakırın elektrokimyasal davranışları yarı logaritmik akım potansiyel eğrilerinden faydalanarak belirlenmiştir. Elde edilen eğriler Şekil 3'de, bu eğrilerden hesaplanan korozyon hızı (i_{kor}) ve korozyon potansiyeli (E_{kor}) sonuçları Tablo 2'de verilmiştir. Bakırın, inhibitör içermeyen 0,5 M H_2SO_4 çözeltisinde belirlenen E_{kor} değeri 0,020 V'dur. Aynı ortamda ve aynı koşullarda ortama farklı derişimler de inhibitör eklenmesi ile eğrilerinin karakteristiğinin değişmediği, bununla birlikte E_{kor} değerlerinin daha negatif potansiyellere kaydığı gözlemlenmiştir. E_{kor} en yüksek derişim olan 1000 ppm için -0,271 V olarak hesaplanmıştır



Şekil 2 Bakır elektrotun inhibitör içermeyen ve farklı derişimlerde A içeren 0,5 M H_2SO_4 çözeltisinde elde edilen Nyquist eğrileri

Tablo 1. Bakır elektrotun inhibitör içermeyen ve değişik derişimlerde A içeren 0,5 M H_2SO_4 çözeltilerinde EIS ve LPR ölçümlerinden belirlenen elektrokimyasal parametreler

		EIS		LPR	
	Derişim ppm	R_p (Ω cm^2)	*E*%	R_{p*} (Ω cm^2)	*E*%
Cu	-	77	-	77	-
	50	345	77,60	351	78,06
	100	655	88,24	676	88,60
	250	1281	93,98	1333	94,22
	500	2487	96,90	2504	96,92
	1000	3578	97,84	3303	97,66



Şekil 3. Bakır elektrotun inhibitor içermeyen ve farklı derişimlerde A içeren 0,5 M H_2SO_4 çözeltilerinde elde edilen katodik (a) ve anodik (b) akım-potansiyel eğrileri.

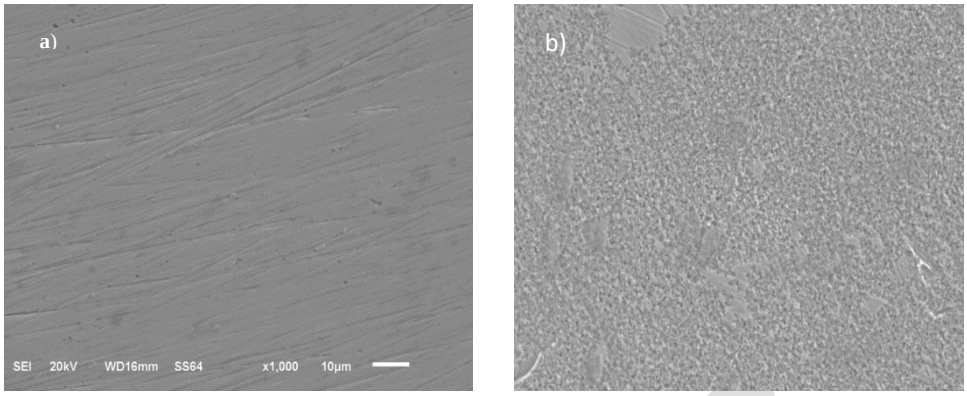
Tablo 2. Farklı derişimlerde A içeren ve içermeyen 0,5 M H_2SO_4 çözeltilerde yarı logaritmik akım-potansiyel eğrilerinde elde edilen korozyon parametreleri

	Derişim (ppm)	E_{kor} (V,Ag/AgCl)	i_{kor} (µA cm^{-2})
Cu	-	0,020	4,78
A	50	-	-
	100	-	-
	250	-0.252	0,997
	500	-0.255	0,870
	1000	-0.271	0,830

Tablo 2'den görüleceği gibi inhibitör içermeyen asidik ortamda bakır için i_{kor} değeri 4,78 µA cm^{-2}'dir. 1000 ppm A'nın varlığında ise i_{kor} değer, 0,830 µ Acm^{-2} dir. Bu rakamsal verilerden de anlaşılacağı gibi, asidik ortama A inhibitörünün farklı derişimlerde eklenmesi, anodik metal çözünmesi ve katodik bölgede hidrojen oluşum reaksiyonunu azaltmaktadır. 1000 ppm A içeren asit çözeltisinde bakır için potansiyodinamik ölçümlerden belirlenen etkinlik değerleri % 82,6 dır.

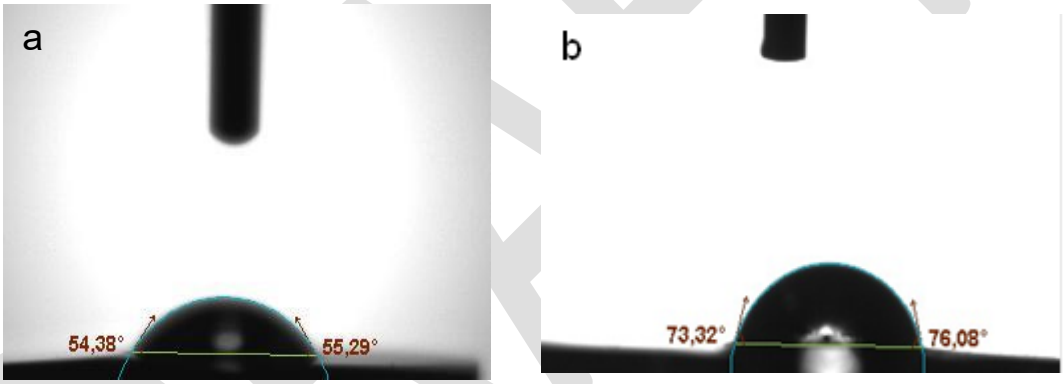
Şekil 4'de Bakırın inhibitör içermeyen ve 1000 ppm A içeren 0,5 M H_2SO_4 çözeltilerinde 1 saat bekletildikten sonra elde edilen SEM görüntüleri verilmektedir. Asidik ortamda bakırın SEM görüntüleri incelendiğinde, 1 saat sonunda yüzeyde çok fazla değişme meydana gelmediği sadece zımpara izlerinin olduğu görülmektedir. İnhibitör içeren ortama bakıldığında ise, bakır yüzeyinde A 'nın dikkat çekici bir şekilde film oluşturduğu görülmektedir.

DRAFT



Şekil 4. İnhibitör içermeyen (a) ve 1000 ppm A içeren (b) 0,5 M H_2SO_4 çözeltisinde daldırılan bakır elektrodun SEM görüntüleri

Şekil 5'de inhibitör içermeyen ve 1000 ppm A içeren asidik ortamda bakır için elde edilen temas açısı sonuçları görülmektedir. Şekil den de görüleceği gibi, inhibitörlü ortamda bakır yüzeyi hidrofobik özelliğe sahiptir.



Şekil 5. İnhibitör içermeyen (a) ve 1000 ppm A içeren (b) 0,5 M H_2SO_4 çözeltisinde daldırılan bakır elektrodun temas açıları

SONUÇ

Ekonomik değeri olan çevre dostu doğal ürün A' nın 0,5 M H_2SO_4 çözeltisindeki bakırın korozyonuna inhibisyon etkisi araştırılmıştır. Bu amaçla elektrokimyasal yöntemlerden yararlanılmıştır. Elde edilen sonuçlar aşağıda maddeler halinde özetlenmiştir.

Yapılan deneysel çalışmalar sonucunda A' nın bakırın asidik ortamda korozyon hızını yavaşlattığı belirlenmiştir.
A'nin bakırı koruma etkinliği asidik ortamdaki artan derişimi ile artmaktadır. İnhibisyon etkinliği 1000 ppm A derişiminde %97,84 olup pratik uygulamalar için son derece etkilidir
EIS ve LPR sonuçları birbirleri ile uyum içinedir.
Potansiyometrik polarizasyon çalışmaları, A'nın hem bakırın anodik çözülme ve hidrojen oluşum reaksiyonunun hızını azalttığını göstermektedir. Dolayısı ile organik türler karma inhibitör olarak sınıflandırılabilir

TEŞEKKÜR

Bu çalışma Bingöl Üniversitesi Bilimsel Araştırma Projeleri Koordinasyon Birimine (BÜBAP) (BAP-GMYO.2021.001) sunulan projeden üretilmiştir. BÜBAP birimine, Bingöl Üniversitesi Merkezi Araştırma Laboratuvarına ve İş Sağlığı ve Güvenliği Bölümüne teşekkür ederiz.

KAYNAKLAR

[1] Koçak H., Bakır Alaşımları El Kitabı, Sağlam Metal, İstanbul, 2006.
[2] Erbil M.,.Korozyon (İlkeler- Önlemler), Ankara, 2012
[3] Şahin EA, Solmaz R, Gecibesler IH, Kardaş G. Adsorption ability, stability and corrosion inhibition mechanism of phoenix dactylifera extrat on mild steel. Mater Res Express. 2020;7(1)
[4] Zhang J., Li H. 2-(2-chlorophenyl)-1H-benzimidazole as a new corrosion inhibitor for copper in sulfuric acid. International Journal of Electrochemical Science, 2020:15;5362 – 5372.

DRAFT

ORAL PRESENTATION

Artificial Neural Network Modeling for Drying Kinetics of Mentha Piperita L.

Burcu Taştan[1] (*ORCID:0000-0001-7008-4746*), Eda Göz[2] (*ORCID: 0000-0002-3111-9042)*, Mehmet Yüceer[1*] (*ORCID: 0000-0002-2648-3931*)

[*1]İnönü University, Engineering Faculty, Chemical Engineering Department, Malatya, Turkey.
[2]Ankara University, Engineering Faculty, Chemical Engineering Department, Ankara, Turkey.

*Corresponding author e-mail: mehmet.yuceer@inonu.edu.tr

Abstract

The drying kinetics of *Mentha piperita* L. leaves grown under the ecological conditions of Malatya province in Turkey were investigated. The leaves were dried both in the shade and in the oven. To understand the drying kinetics and predict the moisture ratio (MR, %), an Artificial Neural Network (ANN) model with a three-layer feed-forward backpropagation network structure was developed. Drying time (min.) and drying methods (shade drying, oven drying at 35°C, and 50°C) were selected as input parameters to predict the MR. The performance of the model was evaluated using statistical criteria, including the correlation coefficient (R), mean absolute percentage error (MAPE, %), root mean square error (RMSE), and mean square error (MSE). For the test phase, these values were calculated as 0.9999 (R), 0.9223% (MAPE), 0.0034 (RMSE), and 1.14 x 10^{-5} (MSE), respectively. Additionally, these values were determined as 1.0000 (R), 0.5422 (MAPE, %), 0.0012 (RMSE), and 1.42 x 10^{-6} for the training phase. According to these criteria, the ANN model performs excellently in predicting the MR of *Mentha piperita* L. MR under different drying conditions.

Keywords: *Mentha piperita* L., Drying Process, Modelling, Artificial Neural Network models

INTRODUCTION

Fresh and dried spearmint samples have been used as ingredients in food, therapeutics, flavor and fragrance additives and pharmaceuticals. Many spearmint samples are dried to enhance the quality of dried peppermint leaves, including improvements in polyphenol content, antioxidant capacity, and yield of peppermint essential oils and extend their shelf life. Drying is a complex process where heat transfer and mass transfer co-occur. Various drying processes are employed, including sun drying, shade drying, hot air drying, vacuum drying, freeze drying, microwave drying, osmotic drying, and infrared drying. Determining the drying kinetics is essential to analyze and understand the drying behavior of a food product. Thin-layer drying models are commonly used to describe the drying kinetics of many plants. These models can be categorized as theoretical, semi-theoretical, and empirical models. Thin-layer models are considered semi-theoretical models, as they are derived by simplifying the general series solution of Fick's second law, and they do not consider the material's geometry, conductivity, and diffusivity.

Alternatively, some artificial intelligence (AI) techniques, such as Artificial Neural Networks (ANN), can be employed to predict the moisture ratio of plants. When developing thin-layer drying models, separate optimization is necessary for each drying condition, and the calibrated model is valid only under specific conditions. In contrast, AI models are developed using data from all drying conditions. AI models can be considered more advantageous, as a single ANN model is sufficient to explain all drying processes, making it a more versatile option than kinetic models.

The ANN model is one of the most popular AI models used in drying studies, with numerous instances in the literature where drying processes have been modeled using ANN. For instance, Sarımeşeli and Yüceer (2014) conducted a study investigating the microwave drying kinetics of thyme leaves, employing ANN methods to model the experimental data. The statistical performance metrics were impressive, with R-square, MAPE (%), and RMSE values calculated as 0.9999, 4.0937, and 0.025, respectively. Furthermore, Sarımeşeli and Yüceer

explored the infrared drying of spinach leaves in another study (Sarımeşeli and Yüceer, 2015). They utilized sample amount, power, and drying time as input variables to estimate the moisture ratio (MR), achieving high-quality results with R-square, MAPE%, and RMSE values of 0.9999, 0.95262, and 0.00284, respectively.

In a separate study conducted by Jafari and colleagues (Jafari et al., 2016), onions were subjected to drying using a pilot plant fluidized bed dryer. In addition to ANN, fuzzy logic and nonlinear regression methods were developed and assessed in this research. Notably, the ANN model outperformed the other two methods regarding performance.

Furthermore, Beigi and Ahmadi (Beigi and Ahmadi, 2019) employed the ANN method to predict the drying curves of celeriac (*Apium graveolens* L.). They used chamber pressure, air temperature, and time as input variables to estimate the moisture content.

Kaveh and colleagues (Kaveh et al., 2018) employed both Adaptive Neuro-Fuzzy Inference System (ANFIS) and ANN structures to predict the moisture-specific energy consumption and diffusivity of potatoes, cantaloupe, and garlic. Interestingly, the ANFIS model demonstrated superior performance compared to the ANN approach.

In another study conducted by Omari and colleagues (Omari et al., 2018), both static and dynamic ANN models were developed for drying mushrooms. The input variables selected for this study included temperature, microwave power density, and moisture content. The ANN network utilized two hidden layers. Notably, the dynamic model exhibited superior performance compared to the static model.

Furthermore, in a study conducted by Karakaplan and colleagues (Karakaplan et al., 2019), an ANN model was developed to predict the moisture content of *Mentha spicata*. The model's performance was assessed using the coefficient of determination (R^2), RMSE, and MAPE (%).

Selvi and colleagues (Selvi et al., 2022) developed an ANN model to characterize the drying kinetics of linden leaves with different thicknesses, ranging from 0.210 mm to 0.230 mm. This study used infrared drying at three different temperatures (50°C, 60°C, and 70°C). The validation results indicated that the predicted moisture ratio by the ANN model aligned well with the experimental moisture ratio data.

In the current study, the drying kinetics of *Mentha piperita* L. samples were investigated using the ANN model, with drying time and drying methods defined as input parameters. Model performance was assessed using four statistical criteria: R, MAPE%, RMSE, and MSE. According to these statistical criteria, the ANN model strongly predicted the moisture ratio (MR) of *Mentha piperita* samples.

MATERIALS AND METHODS

Drying Experiments

The plant samples were subjected to two different drying methods: shade drying and oven drying. The drying time was defined as the duration required for the mint samples to reach an approximate moisture level of 10% from their initial moisture content. A constant initial weight of 20g (±0.5g) was maintained for all experiments.

The shade drying experiments were conducted in a closed environment at an ambient temperature of approximately 23°C, while the oven drying experiments were carried out at two different temperatures, namely 35°C and 50°C. Weight loss measurements were recorded at various time intervals during the drying process. Each drying temperature experiment was conducted in triplicate.

Artificial Neural Network (ANN) Design

The Artificial Neural Network (ANN) is employed for solving nonlinear problems. A simple network structure has three layers: the input layer, the hidden layer, and the output layer. The input layer receives data from external sources and passes it on to the other layers. In this layer, information is transmitted to the hidden layers without processing. The data received from the input layer is processed within the hidden layer and then transmitted to the output layer. While some artificial neural networks lack a hidden layer, others can have multiple intermediate layers. The number of neurons in these layers is independent of the number of inputs, output variables, and the complexity of the process. The output layer combines information from the hidden layers with the network's outputs.

In this study, two input parameters (drying methods and drying time) were used to predict the moisture ratio of *Mentha piperita* L. samples. A feed-forward neural network structure was selected, with the hyperbolic tangent sigmoid (‘*tansig’*) activation function used in the hidden layer and the linear function (*‘purelin’*) employed in the output layer. The Levenberg-Marquardt algorithm (*‘trainlm’*) was chosen to train the network structure. A total of 115 data points were collected from experiments, and these data were randomly divided into a training set (70%) and a testing set (30%). The network structure is illustrated in Figure 1.

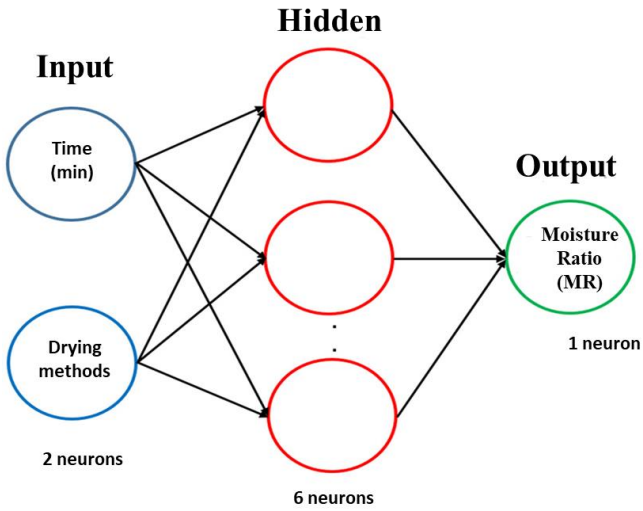


Figure 1. ANN structure

RESULTS

Artificial Neural Network (ANN) Model

In this study, experimental data were modelled using ANN. Training and test phase graphs can be found in Figure 2.

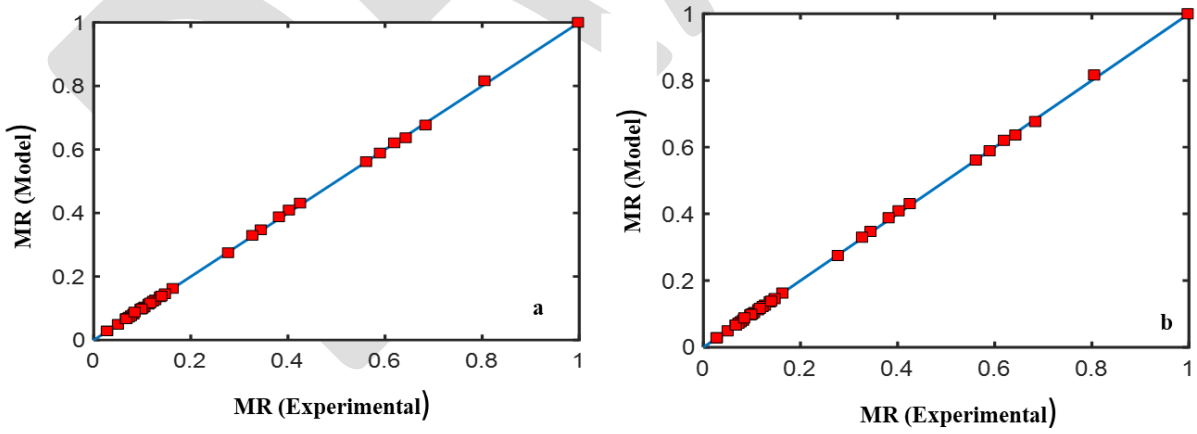


Figure 2. The experimental-model fit graph (a: training phase b: test phase)

The statistical evaluation criteria for the ANN models are presented in Table 1.

Table 1. Performance criteria values of ANN models

Training phase			
R	**%MAPE**	**RMSE**	**MSE**
1.0000	0.5422	0.0012	1.42×10^{-6}
Test Phase			
R	**%MAPE**	**RMSE**	**MSE**
0.9999	0.9223	0.0034	1.14×10^{-5}

According to Table 1, the ANN model demonstrates excellent performance due to its high correlation coefficient (R≅1) and low values for MAPE%, RMSE, and MSE.

CONCLUSION

Drying is a highly complex process where heat and mass transfer occur simultaneously. It has become imperative to employ drying processes to enhance the activity and properties of medicinal plants. This study employed three different drying methods: shade drying, oven drying at 35°C, and oven drying at 50°C. In recent years, numerous kinetic models have been developed to comprehend drying kinetics. However, modeling drying kinetics using thin-layer models can be time-consuming due to the need for optimization steps to calculate kinetic parameters. On the other hand, the development of artificial intelligence (AI) models offers a more straightforward way to understand the drying mechanism. Additionally, an AI model with good performance can reduce the experimental requirements in various studies.

REFERENCES

Beigi M, Ahmadi, I 2019. Artificial neural networks modeling of kinetic curves of celeriac (Apium graveolens L.) in vacuum drying. Food Science and Technology, 39(supl.1): 35–40.

Jafari SM, Ganje M, Dehnad D, Ghanbari V 2016. Mathematical, fuzzy logic and artificial neural network modeling techniques to predict drying kinetics of onion. Journal of Food Processing and Preservation, 40(2):329–339.

Karakaplan N, Göz E,Tosun E, Yüceer M 2019. Kinetic and artificial neural network modeling techniques to predict the drying kinetics of *Mentha spicata L.* Journal of Food Processing and Preservation, 43:e14142.

Kaveh M, Sharabiani V, Chayjan R, Taghinzdah E, Gilandeh Y, Golpour I 2018. ANFIS and ANNs model for prediction of moisture diffusivity and specific energy consumption potato garlic and cantaloupe drying under convective hot dryer. Information Processing in Agriculture, 5(3):
372–387.

Omari A, Khazei N, Sharifian F 2018. Drying kinetic and artificial neural network modelling of mushroom drying process in micro- wave-hot air dryer. Journal of Food Process Engineering, 41(4): 1–10.

Sarimeseli A, Coskun MA, Yuceer M 2014. Modelling micro-wave drying kinetics of thyme (*Thymus Vulgaris* L.) leaves using ANN methodology and dried product quality. Journal of Food
Processing and Preservation, 38: 558–564.

Sarimeseli A, Yuceer M 2015. Investigation of infrared drying be-haviour of spinach leaves using ANN Methodology and dried product quality. Chemical and Process Engineering, 36(4): 425–436.

Selvi KÇ, Alkhaled AY, Yıldız T 2022. Application of artificial neural network for predicting the drying kinetics and chemical attributes of Linden (*Tilia platyphyllos Scop.*) during the infrared drying process. Processes. 10(10):2069.

ORAL PRESENTATION

Yabani yılkı atında ön ve arka ayak parmaklarının morfometrisi

Sema Özkadif[1*] *ORCID: https://orcid.org/*0000-0002-5398-9874,

Ayşe Halıgür[1] *ORCID: https://orcid.org/*0000-0002-3668-4286

[1*]Çukurova Üniversitesi, Ceyhan Veteriner Fakültesi, Anatomi Anabilim Dalı, Adana, Türkiye

*semaerten80@gmail.com

Özet

Yılkı atları Türkiye'nin doğusundan batısına kadar farklı bölgelerde bulunmaktadır. Kış şartlarına uzun süre dayanabilen yılkı atları, bazen soğuğa ve açlığa yenik düşerek ölmektedirler. Vücudun tüm ağırlığını taşıyan ayaklar, buzları da kırarak suya ulaşımı sağlarlar. Klinik değerlendirmeler için ayak anatomisi ve morfometrisi oldukça önemlidir. Bu çalışma, yılkı atlarında parmak kemikleri (phalanx proximalis, phalanx media ve phalanx distalis)'nin ön ve arka ayak ile sağ ve sol taraf arasındaki morfometrik farklılıklarını ortaya koymak amacıyla gerçekleştirilmiştir. Çalışmada Adana'nın Pozantı ilçesinin yüksek dağlarında yoğun kış şartları nedeniyle ölen ve Ceyhan Veteriner Fakültesi Anatomi anabilim dalına getirilen 8 yılkı atı kullanılmıştır. Yılkı atlarının tüm ayaklarının parmaklarından bazı morfometrik ölçümler alınmıştır. Elde edilen ölçüm değerlerine göre yapılan analiz sonucunda ön ve arka ayak ile sağ ve sol taraf arasında istatistiki olarak fark olmadığı tespit edilmiştir. Phalanx media'nın uzunluğu phalax proximalis'in uzunluğunun yaklaşık yarısı kadar olduğu görülmüştür. Bu çalışmadan elde edilen morfometrik verilerin yılkı atlarının anatomik bilgi birikimine katkı sağlamasının yanı sıra, klinik araştırmalarda ve ayak biyomekaniğinin anlaşılmasında katkı sağlayacağı düşünülmektedir.

Anahtar Kelimeler: Yılkı atı, parmak, anatomi

Morphometry of phalanxes of forelimb and hindlimb in wild yılkı horse

Abstract

Yılkı horses are found in different regions from east to west of Turkey. Yılkı horses, which can withstand winter conditions for a long time, sometimes die by succumbing to cold and starvation. The feet, which carry the entire weight of the body, break the ice and provide access to water. Foot anatomy and morphometry are very important for clinical evaluations. This study was carried out in order to reveal the morphometric differences of the foot bones (phalanx proximalis, phalanx media and phalanx distalis) between the forelimb and hindlimb and the right and left sides of the horses. In the study, 8 horses that died in the high mountains of Adana's Pozantı district due to intense winter conditions and were brought to the Ceyhan Veterinary Faculty Anatomy Department were used. Some morphometric measurements were taken from the bones of all foot of the yılkı horses. As a result of the analysis made according to the measurement values obtained, it was determined that there was no statistical difference between the forelimb and hindlimb and the right and left sides. The length of the phalanx media was found about half of the length of the phalax proximalis. It is thought that the morphometric data obtained from this study will contribute to the anatomical knowledge of yılkı horses, as well as to clinical research and understanding of foot biomechanics.

Keywords: Yılkı horse, phalanx, anatomy

GİRİŞ

Yılkı atları Türkiye'de Raman dağından Manisa'ya kadar Türkiye'nin çeşitli bölgelerinde görülür (Eyidogan, 2022). Yabani yılkılar, sert doğa şartlarına adapte olmuş, son derece dayanıklı ve güçlü atlardır (Kandır, 2013). Kış şartlarına karşı doğada birkaç nesil direnen hayvanlar araziye uyum sağlarlar. Ancak çoğu da kış şartlarının üstesinden gelemez ve sert geçen kış dönemleri yılkı atlarının ölümüne neden olur. Ayrıca artan çevre sorunları, özellikle su kaynaklarının atıklarla kirlenmesi, yılkı atlarının yaşam mücadelesini zorlaştırır (Kutukcu, 2017).

Yılkı atlarının ayakları güçlü bir yapıya sahip olduklarından donmuş göletleri kırarak, diğer canlıların da sudan faydalanmasına imkan sağlarlar (Hacan ve ark., 2018). Ekstremitelerin distal kısımlarında oldukça fazla yaralanmalar meydana gelir. Phalanx distalis ise atın vücudunun tüm ağırlığını taşır (Pasko ve ark., 2017). Bu nedenle anatomisi ve şekli, hareketin biyomekaniğinde son derece önemli bir rol oynar (Dzierzecka ve ark, 2016).

Ayak anatomisi bilgisi, ayakta meydana gelen anatomik değişikliklerin teşhisi için oldukça önemlidir. Ayağın klinik değerlendirilmesinde, anatomik değişikliğin etkili bir şekilde saptanması ve miktarının belirlenmesi gerekmektedir. Bu klinik açıdan önemlidir, çünkü tedavi sonucu ve iyileşme beklentileri anatomik derece ile ilişkilidir (Vosugh ve ark., 2017).

Literatür araştırmalarında; saf İran Arap atının, Nijarya atının ve eşeğin ön ayaklarının phalanx distalis'inin normal radyografik morfometrisinin belirlendiği ((Vosugh ve ark., 2017; Ogbanya ve ark., 2018; Elrashidy ve ark., 2022), Holstein sığırların ön ve arka ekstremitelerinin tüm phalanx'larına ait ölçüm değerlerinin ortaya konulduğu (Ocal ve ark., 2004; Gundemir ve ark., 2020) çalışmaların olduğu görüldü. Bu çalışmada, yılkı atlarında parmak kemikleri (phalanx proximalis, phalanx media ve phalanx distalis)'nin ön ve arka ayak ile sağ ve sol taraf arasında morfometrik farklılıkları ortaya koymak amaçlandı.

MATERYAL VE METOT

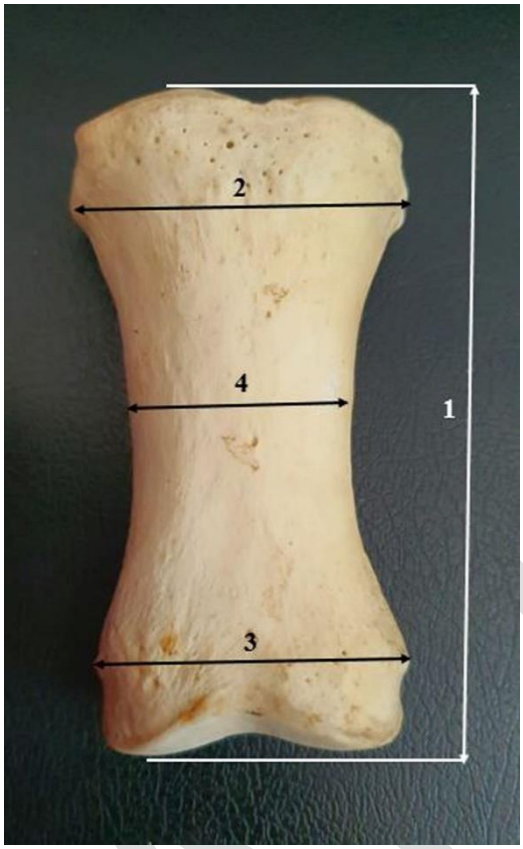
Çalışma Orman ve Su İşleri Bakanlığı 26.08.2016 tarih ve 27006244-445.01.01-173572 sayılı izni ile gerçekleştirilmiştir. Çalışmada materyal olarak Adana'nın Pozantı ilçesinin yüksek dağlarında yoğun kış şartlarına dayanamayıp ölen ve Ceyhan Veteriner Fakültesi Anatomi Anabilim Dalı'na getirilen 8 adet yetişkin yılkı atı kullanıldı. Atın ön ve arka ayaklarına ait tüm phalanx proximalis, phalanx media ve phalanx distalis'inden digital kumpas ile bazı morfometrik ölçümler literatür (Ocal ve ark., 2004; Dzierzecka ve ark, 2016; Gundemir ve ark., 2020)'e uygun olarak alındı.

Phalanx proximalis'ten; phalanx uzunluğu, phalanx proximal uç genişliği, phalanx distal uç genişliği, phalanx diaphysis genişliği'nin ölçümleri alındı (Şekil 1).

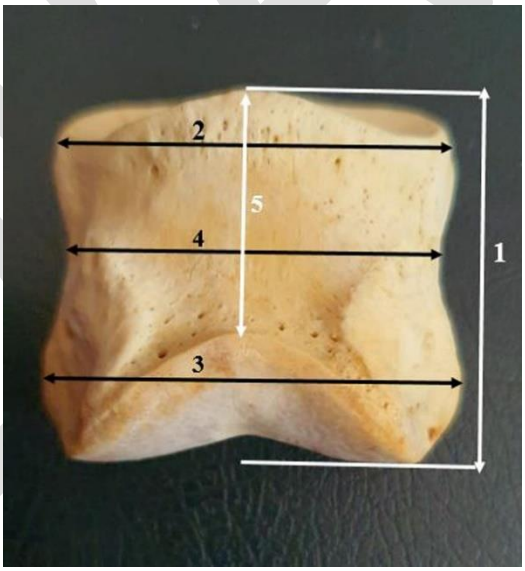
Phalanx media'dan; phalanx uzunluğu, phalanx proximal uç genişliği, phalanx distal uç genişliği, phalanx diaphysis genişliği, phalanx dorsal uzunluğu'nun ölçümleri alındı (Şekil 2).

Phalanx distalis'ten; phalanx dorsal uzunluğu, phalanx proc. extenserius'da yüksekliği, phalanx proc. palmaris medialis'in yüksekliği, phalanx proc. palmaris lateralis'in yüksekliği, phalanx medialinde plantar uzuluğu, phalanx lateralinde plantar uzuluğu, phalanx facies solearis'in orta genişliği'nin ölçümleri alındı (Şekil 3).

DRAFT



Şekil 1. Phalanx proximalis üzerinden alınan ölçüm yerleri. 1. phalanx uzunluğu, 2. phalanx proximal uç genişliği, 3. phalanx distal uç genişliği, 4. phalanx diaphysis genişliği



Şekil 2. Phalanx media üzerinden alınan ölçüm yerleri. 1. phalanx uzunluğu, 2. phalanx proximal uç genişliği, 3. phalanx distal uç genişliği, 4. phalanx diaphysis genişliği, 5. phalanx dorsal uzunluğu

DRAFT



Şekil 3. Phalanx distalis üzerinden alınan ölçüm yerleri. 1. phalanx dorsal uzunluğu, 2. phalanx proc. extenserius'da yüksekliği, 3. phalanx proc. palmaris medialis'in yüksekliği, 4. phalanx proc. palmaris lateralis'in yüksekliği, 5. phalanx medialinde plantar uzuluğu, 6. phalanx lateralinde plantar uzuluğu, 7. phalanx facies solearis'in orta genişliği

İstatistiksel analizler SPSS 15.0 paket programı ile yapıldı. Bağımsız t-testi ile phalanx proximalis, phalanx media ve phalanx distalis'e ait ölçümlerin ortalama ve standart sapma değerleri verilerek, ön ayak ile arka ayak, ayrıca sağ taraf ile sol taraf arasındaki farklar belirtildi. İstatistiksel anlamlılık P<0.05 olarak kaydedildi.

BULGULAR

Yılkı atlarının phalanx'ları üzerinden alınan morfometrik ölçüm değerlerinden elde edilen istatistiksel işlemler sonucunda hiçbir ölçüm değerinde anlamlı olmadığı görüldü.

Tablo 1. Yılkı atında phalanx proximalis'den elde edilen morfometrik ölçümler. (ortalama ± SD)

Ölçümler	Ön ayak		Arka ayak	
	Sağ (n=8)	Sol (n=8)	Sağ (n=8)	Sol (n=8)
Phalanx uzunluğu (mm)	81.29±8.77	80.63±6.97	80.97±10.16	80.20±8.19
Phalanx proximal uç genişliği (mm)	49.28±7.24	48.34±6.50	49.41±6.96	48.90±7.31
Phalanx distal uç genişliği (mm)	40.92±4.79	39.41±4.91	40.30±6.70	39.43±6.06
Phalanx diaphysis genişliği (mm)	30.29±4.79	29.11±4.11	29.54±5.09	29.30±4.73

*Aynı satırda istatistiksel olarak anlamlı (P<0.05)

Tablo 1'e göre phalanx proximalis'e ait morfometrik ölçüm değerlerinin istatistiki olarak ön ayak ile arka ayak arasında, ayrıca sağ ve sol taraf arasında farklılık göstermediği tespit edildi. Phalanx proximalis'in proksimal ucunun distal ucundan daha geniş olduğu görüldü.

DRAFT

Tablo 2. Yılkı atında phalanx media'dan elde edilen morfometrik ölçümler. (ortalama ± SD)

Ölçümler	Ön ayak		Arka ayak	
	Sağ (n=8)	Sol (n=8)	Sağ (n=8)	Sol (n=8)
Phalanx uzunluğu (mm)	42.78±4.52	43.28±4.45	44.25±5.24	41.36±4.66
Phalanx proximal uç genişliği (mm)	46.68±7.02	47.41±6.68	47.20±7.24	45.37±5.76
Phalanx distal uç genişliği (mm)	44.30±6.93	44.12±6.77	43.70±6.57	43.77±4.86
Phalanx diaphysis genişliği (mm)	40.38±5.77	38.79±6.67	40.01±6.26	40.59±5.87
Phalanx dorsal uzunluğu (mm)	26.61±4.20	26.20±3.13	27.39±3.27	24.58±2.37

*Aynı satırda istatistiksel olarak anlamlı (P<0.05)

Tablo 2'e göre phalanx media'dan alınan morfometrik ölçüm değerlerinden elde edilen istatistiki işlem sonuca göre, hem sağ ve sol taraf arasında hem de ön ve arka ayak arasında fark olmadığı tespit edildi. Ayrıca proximal ucunun distal ucundan daha geniş olduğu görüldü.

Phalanx media'nın uzunluğunun, phalanx proximalis'in uzunluğunun yaklaşık yarısı kadar olduğu görüldü. Fakat phalanx media'nın diaphysis genişliğinin phalanx distalis'ten daha geniş olduğu fark edildi.

Tablo 3. Yılkı atında phalanx distalis'den elde edilen morfometrik ölçümler. (ortalama ± SD)

Ölçümler	Ön ayak		Arka ayak	
	Sağ (n=8)	Sol (n=8)	Sağ (n=8)	Sol (n=8)
Phalanx dorsal uzunluğu (mm)	44.68±6.50	45.58±6.71	45.23±8.63	48.25±7.39
Phalanx proc. extenserius'da yüksekliği (mm)	34.95±5.22	37.96±7.06	37.95±6.13	37.98±4.84
Phalanx medialinde plantar uzuluğu (mm)	62.49±12.91	60.37±13.33	59.16±14.21	59.10±12.79
Phalanx lateralinde plantar uzuluğu (mm)	60.00±12.02	56.83±14.22	57.67±12.49	56.86±12.12
Phalanx proc. palmaris medialis'in yüksekliği (mm)	19.80±3.19	21.13±4.30	18.21±3.01	18.13±2.44
Phalanx proc. palmaris lateralis'in yüksekliği (mm)	19.81±3.10	21.22±5.24	18.38±2.98	17.76±3.03
Phalanx facies solearis'in orta genişliği (mm)	61.12±9.88	60.51±11.01	55.21±8.97	56.00±9.80

*Aynı satırda istatistiksel olarak anlamlı (P<0.05)

Tablo 3'de phalanx distalis'ten elde edilen morfometrik verilerin istatistiki sonucuna göre ön ve arka ayak ile sağ ve sol taraf arasında istatistiki fark olmağı görüldü.

DRAFT

TARTIŞMA

Yılkı atlarının phalanx (phalanx proximalis, phalanx media ve phalanx distalis)'ları üzerinde yapılan morfometrik çalışmada, hem ön hem de arka ayakda ölçülen parametrelerde sağ ve sol taraf arasında istatistiki olarak fark olmadığı tespit edilmiştir. Saf İran atlarının phalanx'larının radyografik görüntüleri üzerinde yapılan morfometrik çalışmada da ön ayakta sağ ve sol taraf arasında istatistiksel olarak anlamlı olmadığı belirtilmiştir (Vosugh ve ark, 2017). Akhal-teke atlarının phalanx distalis'lerinin ön sağ ve sol ayakları arasında herhangi bir istatistiki farkın olmadığı radyolojik görüntüler üzerinden elde edilen ölçümlerle ifade edilmiştir (Masoudifard ve ark., 2014). Holstein sığırların da yılkı atlarında olduğu gibi phalanx proximalis, phalanx media ve phalanx distalis'leri üzerinden alınan ölçüm değerlerine göre sağ ve sol taraf arasında istatistiki olarak anlamlı olmadığı ifade edilmiştir (Ocal ve ark., 2004).

Yapılan çalışmada ön ayak ile arka ayak arasında tüm phalanx'lara ait ölçümlerin istatistiki olarak anlamlı olmadığı görülmüştür. Fakat Holstein sığırların ön ayaktaki phalanx proximalis ve phalanx media'nın değerleri, arka ayaktan daha küçük değerlerde olduğu bildirilmiştir (Ocal ve ark., 2004; Gundemir ve ark., 2020). Phalanx distalis'in ön ayağın medialinde bulunan en yüksek değere sahipken, arka ayakta lateralinde bulunanın ise küçük değerlere sahip olduğu belirtilmiştir (Ocal ve ark., 2004). Ayrıca Holstein sığırlarda ön ile arka ayakların medialdeki phalanx proximalis'in proximal uç genişliği ve phalanx media'nın distal uç genişliği arasında istatistiki olarak anlamlı olduğu, diğerlerinde ise yılkı atlarında olduğu gibi ön ve arka ayak parmakları arasında fark olmadığı bildirilmiştir (Gundemir ve ark., 2020).

Yılkı atının phalanx proximalis uzunluğu ortalama sağ ön ayakta 81,29 mm, sağ arka ayakta ise 80,97 mm olarak; phalanx media uzunluğu ortalama sağ ön ayakta 42,78 mm, sağ arka ayakta 44,25 mm; phalanx distalis uzunluğu ise ortalama sağ ön ayakta 44,68 mm, sağ arka ayakta 45,23 mm olarak ölçülmüştür. Eşekte ortalama phalanx proximalis'in uzunluğu 67,4 mm, phalanx media uzunluğu 36,2 mm ve phalanx distalis uzunluğu ise 28,2 mm olduğu bildirilmiştir (El-Shafaey ve ark., 2017). Buna göre yılkı atının phalanx'larının, eşek phalanx'larından daha uzun olduğu görülmektedir.

Yılkı atında phalanx media'nın proximal ucunun genişliği ortalama ön ayağın sağ tarafında 46,68 mm, sol tarafında 47,41 mm; arka ayağın ise sağ tarafında 47,20 mm, sol tarafında ise 45,37 mm olarak ölçülmüştür. Eşeğin radyografik görüntülerinde ise ortalama 30,5 mm olduğu ifade edilmiştir (Elrashidy ve ark., 2022). Buna göre yılkı atının phalanx media'sı eşekten daha büyük bir yapıya sahiptir.

SONUÇ

Sonuç olarak, yılkı atlarının ön ve arka ayak ile sağ ve sol taraftaki phalanx'larının istatistiki olarak farklı olmadığı gözlendi. Bu çalışmadan elde edilen morfometrik verilerin yılkı atlarının klinik araştırmalarında ve ayak biyomekaniğinin anlaşılmasına katkı sağlayacağı düşünülmektedir.

KAYNAKLAR

Dzierzecka M, Purzyc H, Charuta A, Barszcz K, Komosa M, Hecold M, Klosinska D 2016. Evaluation of distal phalanx formation and association with front hoof conformation in coldblooded horses. Biologia, 71(3): 337-342.

Elrashidy MH, Al-lethie AA, Attaai A, El-Hawari SF 2022. Radiographic morphometry of the foot in clinically normal donkeys (*Equus asinus*). International Journal of Veterinary Sciences, 5(4): 66-74.

El-Shafaey EA, Salem MG, Mosbah E, Zaghloul AE 2017. Morphometric evaluation of relevant radiographic parameters of the forefeet of clinically normal donkeys (*Equus asinus*). Journal of Hellenic Veterinary Medical Society, 68: 467-478.

Eyidogan D 2022. Anadolu'nun Yılkı atları. Available at: https://www.dogadergisi.com/en/anadolunun-yilki-atlari/ [16.05.23]

Gundemir O, Ozkan E, Mutus R 2020. Morphometric study on the digital bones in the domestic cattle. Kafkas Universitesi Veteriner Fakültesi Dergisi, 26(1): 75-82.

Hacan O, Kocak S, Çelikeloglu K, Bozkurt Z, Erdogan M, Tekerli M 2018. The Independent Spirit of Turkey: Wild Horse. International Journal of Veterinary and Animal Research, 1(1): 16-18.

Kandır EH 2013. Esaretten özgürlüğe Yılkı atlarının öyküsü. Göller Bölgesi Aylık Hakemli Ekonomi ve Kültür Dergisi Ayrıntı, 13(1): 1-4.

Kutukcu A E 2017. Anadolu'da özgür atlar zamanı. Available at: https://www.hurriyet.com.tr/kelebek/hayat/anadoluda-ozgur-atlar-zamani-40492876 [16.05.23]

Masoudifard M, Vajhi AR, Mansouri SH, Molazem M, Bahonar AR, Zehtabvar O 2014. Radiographic measurements of front feet of the sound Akhal-Teke horses. Iranian Journal of Veterinary Medicine, 8: 21-25.

Ocal MK, Sevil F, Parin U 2004. A quantitative study on the digital bones of cattle. Annals of Anatomy, 186: 165-168

Ogbanya KC, Eze CA, Ihedioha JI 2018. Radiographic morphometry of the hoof and third phalanx of apparently healthy Nigerian horses. Indian Journal of Animal Research, 829: 1-5.

Pasko SB, Dzierzwcka M, Purzyc H, Charuta A, Barszcz K, Bartyzel BBJ, Komosa M 2017. The Osteometry of equine third phalanx by the use of three-dimensional scanning: New Measurement Possibilities. Scanning. Hindawi Scanning, Volume 2017, Article ID 1378947, 6 pages.

Vosugh D, Nazem MN, Hooshmand AR 2017. Radiological anatomy of distal phalanx of front foot in the pure Iranian Arabian horse. Folia Morphologica, 76(4): 702–708.

DRAFT

ORAL PRESENTATION

Obtaining modified activated carbon from Kochia Scorparia L. plant

Esra Kulaksız Alayont[1*] (*ORCID: https://orcid.org/0009-0000-1604-038X*),
Halil Durak[2] (*ORCID: https://orcid.org/0000-0003-3052-6751*),
Yüksel Bayrak[3] (*ORCID: https://orcid.org/0000-0002-5008-456X*),

[*1]Van Yüzüncü Yıl Üniversitesi, Fen Bilimleri Enstitüsü, Kimya Bölümü, Van, Türkiye
[2] Van Yüzüncü Yıl Üniversitesi, Sağlık Hizmetleri Meslek Yüksekokulu, Sağlık Teknikerliği Bölüm, Van, Türkiye
[3] Trakya Üniversitesi, Fen Fakültesi, Kimya Bölüm, Edirne, Türkiye

*Sorumlu yazar e-mail: esra_kulaksiz@hotmail.com

Özet

Yapılan bu çalışmada; Kochia Scorparia L. bitkisinden modifiye aktif karbon elde edilerek optimum şartlar belirlenmiştir. Doğada atıl durumda bulunan Kochia Scorparia L. bitkisi aktif karbon olarak değerlendirilmiş ve adsorpsiyon kapasitesinin arttırılması için $ZnCl_2$ ile aktive edilmiştir. Bu işlem sırasında $ZnCl_2$ farklı oranlarda kullanılmış ve karbonizasyon sıcaklığı 400°C – 500 °C ve 600 °C olarak belirlenmiştir. Yapılan BET yüzey alanı analizi sonuçlarına göre; 1:2 oranında $ZnCl_2$ ile muamele edilen ve 400°C sıcaklık değerinde karbonizasyon işlemine tabii tutulan Kochia Scorparia L. bitkisinin yüzey alanı değeri 599,57 m^2xg^{-1} olarak bulunmuştur.

Anahtar Kelimeler: Kochia Scorparia L. bitkisi, aktif karbon, $ZnCl_2$

Obtaining modified activated carbon from Kochia Scorparia L. plant

Abstract

In this study; optimum conditions were determined by obtaining modified activated carbon from the Kochia Scorparia L. plant. Kochia Scorparia L. plant, which is dormant in nature, was evaluated as activated carbon with $ZnCl_2$ to increase its adsorption capacity. During this process, $ZnCl_2$ was used in different proportions and carbonization temperature was applied at 400 °C - 500 °C and 600 °C. According to the results of BET surface area analysis; It was concluded that the surface area of the Kochia Scorparia L. plant, which was treated with $ZnCl_2$ at a ratio of 1:2 and subjected to carbonization at 400°C, was 599,57 m^2xg^{-1}.

Keywords: Kochia Scorparia L. plant, activated carbon, $ZnCl_2$

GİRİŞ

Su tüm canlıların ve insanların temel fonksiyonlarını yerine getirebilmesi için hayati önem taşır. Su kaynaklarının azalması, su kirliliğinin artması ve arıtım sonucu su geri kazanımının daha az verimle gerçekleşmesi gibi nedenlerden dolayı su kaynaklarının korunması gittikçe daha da önem kazanmaktadır. Su içerisinde bulunan bazı kirlilikler beklenmedik kirleticiler olarak adlandırılır. Bu maddelerin arıtımı veya uzaklaştırılması için alınan önlemler birçok ülkede az ilgi görmüş ve çeşitli nedenlere bağlı olarak su kaynakları insan sağlığını tehdit edecek zararlı maddeler bulundurmaya başlamış ve bu maddeler su kaynakları içerisinde yayılmıştır.

Adsorpsiyon yönteminde biyokütleye uygulanan kimyasal işlemler neticesinde aktif karbonların yapısı değişerek modifiye edilmektedir. Bu aktif karbonlar adsorban olarak adlandırılırlar ve bir çok çalışmada modifiye edilmiş selülozik veya farklı yapıdaki adsorbanların modifiye edilmemiş selüloz ve ya adsorbanlara göre daha yüksek adsorpsiyon kapasitesi sahip olduğu gösterilmiştir (Yahya ve ark.,2015).

Canlılık faaliyetleri esnasında ortaya çıkan organik maddeler, inorganik anyonlar, toksik ağır metaller, zehirli gazlar ve birçok kirletici çevreye bırakılmakta ve özellikle sularda yüksek oranda kirlilik meydana getirmektedir. Yakın gelecekte insanlığı bekleyen en büyük tehdit canlılığı sürdürmek için gerekli ve temiz su kaynaklarının bulunamayacak olmasıdır. Canlılık için hayati önem taşıyan suyun geri kazanılması amacıyla yapılan bilimsel çalışmalar son yıllarda artış göstermektedir.

Bu amaçla çevre dostu, uygun maliyetli arıtma teknolojilerine ihtiyaç büyük önem arz etmektedir.

Atık sulardan çeşitli kirlilikleri uzaklaştırmak için kullanılan adsorpsiyon işlemi çevre dostu arıtma teknolojilerinden biridir. Bu işlemde çeşitli doğal kaynaklardan elde edilebilen selülozik maddeler adsorplayıcı madde olarak kullanılabilmektedir.

Gaz veya çözünmüş maddelerin katı yüzeyinde yoğunlaşması olayına adsorpsiyon adı verilir. Yüzeyde konsantrasyonu artmış maddeye adsorplanmış madde, adsorplayan maddeye de adsorplayıcı madde veya adsorban denir (Berkem ve ark., 2020.)

Aktif karbonların adsorpsiyon özelliklerinin geliştirilmesi için $ZnCl_2$, H_3PO_4, H_2SO_4, K_2S, KCNS, HNO_3, H_2O_2, $KMnO_4$ $(NH_4)_2$ NaOH, KOH ve K_2CO_3 gibi kimyasal maddeler ile aktive edilmektedir bu süreç kimyasal aktivasyon olarak adlandırılır (Petroviç ve ark.,2003).

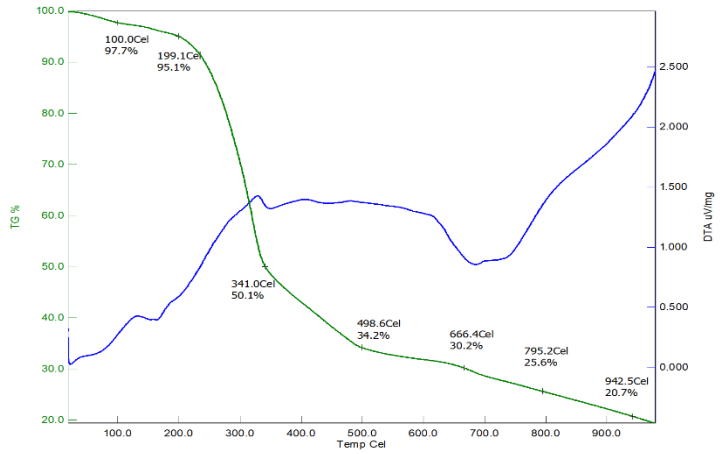
Bu kimyasal aktive edici ajanlar arasında $ZnCl_2$ nin istenen mikro gözenekli yapıyı ve daha büyük bir yüzey alanını üretmede etkili olduğu için yaygın olarak tercih edilmektedir (Berkem ve ark., 2020).

MATERYAL VE METOT

Aktif karbon kaynağı olarak Kochia scoparia L bitkisi yaz döneminde toplanıp gölgede kurutularak ve uygun boyutlara getirilerek aktif karbon olarak kullanıma hazırlanmıştır. Morfolojik yapının gözlenmesi adına TGA analizi yapılmış maddenin ısıl davranışları ve bozunumu göz önüne alınarak karbonizasyon sıcaklık değerlerine karar verilmiştir. Kochia scoparia L bitkisinin adsorpsiyon kapasitesini anlayabilmek için BET yüzey alanı ölçümü yapılmış. Daha sonra Kochia scoparia L bitkisi $ZnCl_2$ kimyasal maddesi varlığında yaş emdirme işlemine tabii tutulmuştur.

$ZnCl_2$ literatürde aktif karbon üretiminde en etkili aktive edici madde olarak bilindiğinden bu bilgi doğrultusunda Kochia scoparia L. bitkisine 1:1, 1:2,1:3, ve 1:4 oranlarında ortama $ZnCl_2$ eklenerek yaş emdirme işlemi gerçekleştirilmiştir.

Kochia scoparia L . bitkisinin hem ham haline BET analizi yapılmış, hem de farklı oranlarda $ZnCl_2$ ile yaş emdirme işleminden sonra 400°C – 500 °C ve 600 °C karbonize edilerek BET analizleri yapılmıştır.



Şekil 1. Ham maddeye uygulanan TGA analiz sonucu

Tablo 1. Kochia scoparia L. bitkisi $ZnCl_2$ ile yaş emdirilip 400 °C 'ta korbonize edildiği BET analizi

BET parameters	400-1	400-2	400-3	400-4
Surface Area/$m^2 \times g^{-1}$	283.61	599.57	491.22	553.02
Pore Volume/$cm^3 \times g^{-1}$	0.020	0.088	0.042	0.069
Average pore width by BET/nm	2.65	2.88	2.84	1.96

BULGULAR ve TARTIŞMA

Kochia scoparia L bitkisinin ham haline uygulanan TGA analizindeki amaç; bitkinin yapısının hangi sıcaklıklarda bozunduğunu görebilmek, aktif karbon üretirken karbonizasyon işleminin kaç °C sıcaklıkta gerçekleşeceğine karar verebilmek ve BET yüzey ölçümü yaparken ortama verilecek inert gazın kaç °C sıcaklıkta gideceğini belirleyebilmektir. Bu sonuçlara göre Kochia scoparia L bitkisinin yapısı 400 °C değerine yaklaşırken kayda değer bozunum göstermiş ve yapısının 600°C'tan sonra kaybetmeye başlamıştır. Bu analiz sonucuna göre karbonizasyon sıcaklıklarının 400°C – 500 °C ve 600 °C olması uygun görülmüştür.

Kochia scoparia L . bitkisinin ham haline BET Analizi yapıldığında yüzeyinin azot adsorpsiyonu yapmadığı görüldü. Yapısında bulunan kuvvetli selülozik yapılardan dolayı azot adsorplamadığı yorumu yapıldı. Fakat Kochia scoparia L . bitkisi $ZnCl_2$ ile modifiye edilip 400 °C 'ta korbonize edildikten sonra yüzey morfoojisi değişerek adsorpsiyonu yapabildiği gözlemlenmiştir

SONUÇ

Bu sonuçlar doğrultusunda ham madde ile 1:2 oranında $ZnCl_2$ ile 400 °C 'ta korbonize edilmiş aktif karbonun en yüksek yüzey alanı ve por hacmi değerlerini verdiği sonucuna ulaşılmıştır.

Literatürle karşılaştırıldığında ham madde ile 1: 1,5 - 2 oranında aktive edici madde ile elde edilmiş aktif karbonun en yüksek yüzey alanı ve por hacmi değerlerini verdiği görülmektedir. Elde edilen deneysel veriler literatürde yapılmış olan çalışmaları desteklemektedir.

Yapılan çalışmadan çıkartılacak en önemli sonuç ise; ham haliyle hiçbir şekilde adsorpsiyon yapma kapasitesi bulunmayan maddenin aktive edici maddeler ile muamelesi sonucunda adsorpsiyon özelliği kazanabilmesidir. Ayrıca elde edilen aktif karbonun doğadan uygun maliyetli ve ulaşılabilir bir maddeden elde edilmiş olması çevre dostu bir proses olduğuna işaret etmektedir.

KAYNAKLAR

Arslan, R., Tozluoğlu, A.,, Sertkaya, S.,, Fidan, H., ve Küçük S.,2021. Functionalized Nanocellulose Based Adsorbents for Dye Removal From Wastewater. Journal of Forestry Faculty. 22,148-60.

Berkem, A.R., Baykut, S., and Berkem, M.L., 2020. Fizkokimya, İstanbul Üniversitesi Yayın No. 3628, II.Cilt, İstanbul.

Petrovic, M., Gonzalez, M. ve Barceló, D., 2003. Analysis and Removal of Emergin Contaminants in Wastewater and Drinking Water, Trends Analyt. Chem., 22, 685-696.

Yahya MA, Al-Qodah Z, Zanariah Ngah CW (2015) Aktif karbon üretimi için kullanılan potansiyel sürdürülebilir öncüler olarak tarımsal biyo-atık malzemeler: Bir inceleme. Yenilenebilir ve Sürdürülebilir Enerji İncelemeleri *46: 218–235.*

DRAFT

ORAL PRESENTATION

Sustainable nanobubble water technology for dyeing of cotton fabrics

Tuba-Toprak-Cavdur[1*] *(ORCID: https://orcid.org/0000-0001-8475-3197)*

*[1]Bursa Uludag University, College of Engineering, Department of Textile Engineering, Bursa, Turkey

*Corresponding author e-mail:tubatoprak@uludag.edu.tr

Abstract

It is a crucial issue for the processes to become sustainable in the textile industry as one of the sectors polluting the environment the most and consuming high volumes of water. It is noteworthy that the most preferred natural fiber, cotton and the reactive dye used in its dyeing require very high amount of chemicals and water. For this reason, in this study, the usability of nanobubble water technology, an innovative technology, in reactive dyeing was examined. The results of the study were analyzed basically in terms of colorimetry and fastness. According to the colorimetry results, it was revealed that nanobubble technology effected color coordinates of dyed fabrics. Morover, nanobubbles had an effect on the color strength, causing a decrease compared to conventional ones. Although good level of fastness values were achieved with this technology, the fact that their values were half a point lower than the those of the conventional ones revealed the need to investigate the use of this technology in detail, especially in cleaning processes such as washing.

Keywords: Sustainability, Environmentally-friendly, Nanobubble, Colorimetry, Washing fastness

INTRODUCTION

The nanobubbles (NBs) history began in 1950 with the Epstein-Plesset theory (Zhou et al. 2021). Nanobubbles are fine bubbles with less than 1 μm in diameter (Haris et al. 2020). Although many studies have been done with nanoscale gas bubbles (Zhou et al. 2021), there are still debates about NBs short-life paradox and their longevity in water (Weijs et al. 2012, Yasui et al. 2016, 2018, Meegoda et al. 2019). NBs are also characterized by mass transfer efficiency, high zeta potential, producing high dissolved oxygen concentration, and large specific surface area (Takahashi et al. 2007, Qian et al. 2019, Haris et al. 2020). The nanobubble (NB) technology has been used in health protection, aquaculture, agricultural and many other fields in recent years (Azevedo et al. 2019, Zhou et al. 2019, Endo-Takahashi and Negishi 2020, Minamikawa and Makino 2020, Ma et al. 2022). The NB technology is generally used in wastewater treatment in textile (Bui and Han 2020, Wu et al. 2021, Anis et al. 2022, Rojviroon and Rojviroon 2022) because NBs decompose organic substances by generating free radicals (Sakr et al. 2022). NB technology has started to be used in textile dyeing and finishing processes as a new application area alongside water treatment (Mohsin et al. 2020, 2022)

Reactive dyes are used in more than 50% of cotton dyeing (Grancarić et al. 2013), so they are the most popular dye class for cotton dyeing (Gopalakrishnan et al. 2019). Some of the reasons why reactive dyes are preferred are that their high washing fastness, ease of applications, and wide bright color gamut. Contrary to these advantages, high amounts of salt and alkali are required for exhaustion and fixation, respectively (Varadarajan and Venkatachalam 2016, Arivithamani and Giri Dev 2018). That is, reactive dyeing is characterized by high electrolyte and alkali concentrations (Khatri et al. 2013, Zhang et al. 2022). Moreover wastewater is highly colored because of hydrolyzed/unfixed dyes (Burkinshaw et al. 2000, Ramasamy and Kandasaamy 2005). Because of removing hydrolyzed/unfixed dyes after dyeing, high volumes of water is used (Khatri and White 2016). Consequently, in terms of environmental impact, reactive dyeing is a water-intensive dyeing process and its wastewater discharged is highly polluted.

As in all sectors, sustainability has gained great importance in textiles. In textile as one of the sectors polluting the environment the most, one of the most critical issues that is emphasized with sensitivity is the environmentally-friendly production processes. In this concern this study attempted to investigate the probability of cotton in the presence of NBs. The results were analyzed in terms of colorimetry and color fastnesses.

DRAFT

DRAFT

MATERIALS AND METHODS

In the study, woven cotton fabrics were used and supplied from Yürek Tekstil. The black, red and blue reactive dyes were purchased from Archroma. Alkali (sodium carbonate) and salts (sodium chloride) as dyeing auxiliaries were of analytical grade and provided by Merck.
The dissolved oxygen level was controlled and the water taken from the NB generator was used in the dyeing processes. Reactive dyeing of woven fabrics was carried out 80°C in 60 minutes at a concentration of 40 g/L salt (NaCl) and 20 g/L sodium carbonate (Na_2CO3) with %2 o.w.f. dye concentrations. The reactive washing cycle consisted of cold (20 °C-30°C, 10') and warm (70 °C-80 °C, 10') washing, soaping (90 °C-95 °C, 20'), and warm (70 °C-80 °C, 10') and cold (20 °C-30 °C, 10') rinsing, respectively (Toprak-Cavdur et al. 2022). Dyeing and washing were carried out at 10:1 liquor ratio. Duration-temperature diagram of reactive dyeing is given in Fig. 1.

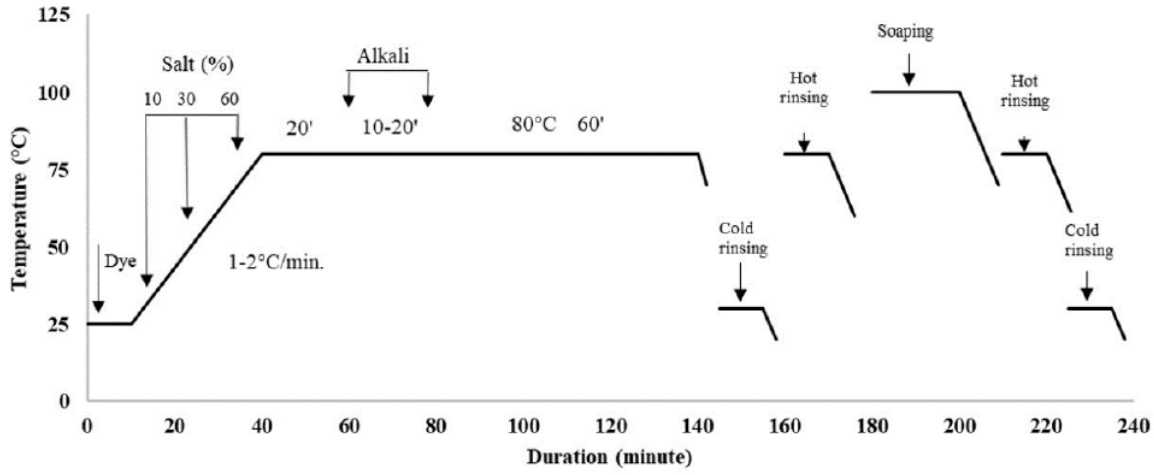


Figure 1. Duration-temperature diagram of reactive dyeing

The CIELAB (1976) L*, a*, b*, C* and h° color coordinates were measured and color strength values were calculated using a Konica Minolta CM-3600D spectrophotometer (under D65 illuminant and 10° standard observer) in specular component included (SCI) mode. Four measurements were taken from each sample with a 90° rotation to reduce possible experimental errors.

The color strength (K/S) values were calculated using the Kubelka-Munk equation. The K/S formula (Akgun et al. 2014) is presented in Equation 1.

$$K/S = \frac{(1-R)^2}{2R} \quad (1)$$

In the Equation 1, *R* is the decimal fraction of the reflectance of fabric, *K* is the absorption coefficient, and *S* is the scattering coefficient.

The washing fastness measurements of the dyed fibers were carried out by a TEST color fastness tester, using the color fastness to domestic and commercial laundering standard test method (EN ISO 105-C06/A1M).

RESULTS

Colorimetry

Color coordinates of cotton fabrics are given in Table 1. a*-b* plot of the samples are presented in Figure 2.

Table 1. Color coordinates of the dyed samples

	L*	a*	b*	C*	hº
Black	14.73	-1.03	-1.84	2.11	240.69
Black-NBs	17.77	-0.48	-0.79	0.94	237.37
Blue	21.32	-0.68	-14.30	14.19	267.20
Blue-NBs	21.63	0.02	-16.10	16.10	270.07
Red	34.04	50.68	26.19	57.04	27.32
Red-NBs	36.63	50.55	27.19	57.41	28.27

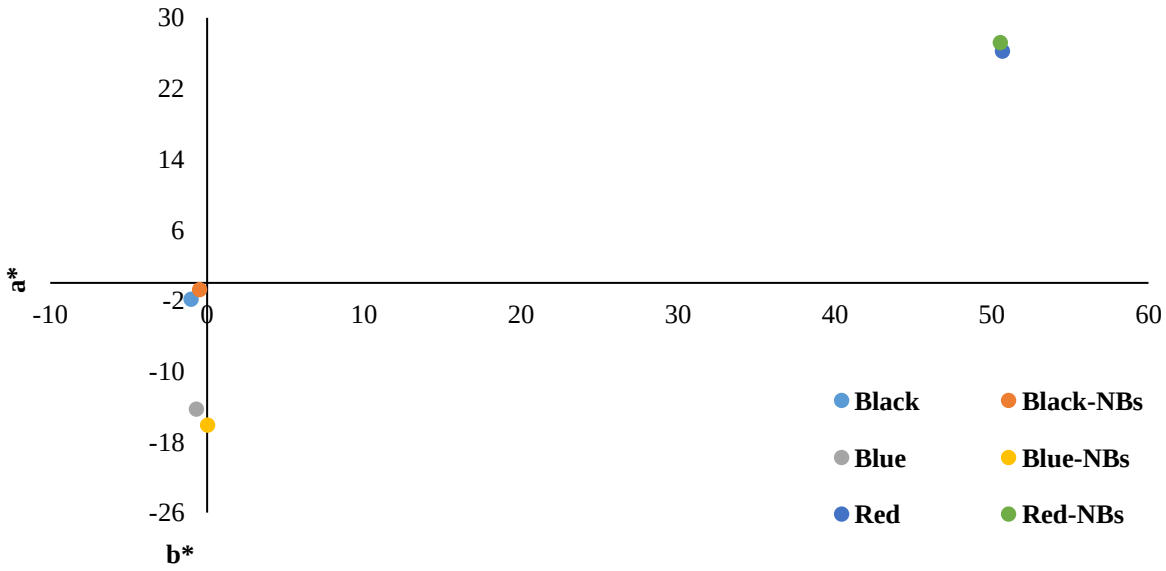


Figure 2. a*-b* plot of the dyed samples

NBs resulted in lighter colors in all dyeing processes in which they were used except in the one for the blue color. The effects of NBs on the coordinates of colors on the red-green (a*) and yellow-blue (b*) axis were different. For example, while NBs reduced greenness and blueness in black, it decreased greenness and increased blueness in blue. Moreover, NBs also increased the yellowness of the red color. The results of Table 1, especillay increased L* values, could be explained by the possibility that reactive oxygen species, the proliferation of which was promoted by NBs (Yasui et al. 2018, Wang et al. 2020), could have degraded the dye molecules (Chen et al. 1999, Ismail and Sakai 2022, Mahanna and El-Bendary 2022). This degradation could also have been caused by NBs increasing the oxidation reaction by increasing the dissolved oxygen level (Rojviroon and Rojviroon 2022). These could explain the changes in the a*-b* coordinates of the colors. Figure 3 shows color strenghs of dyed fabrics.

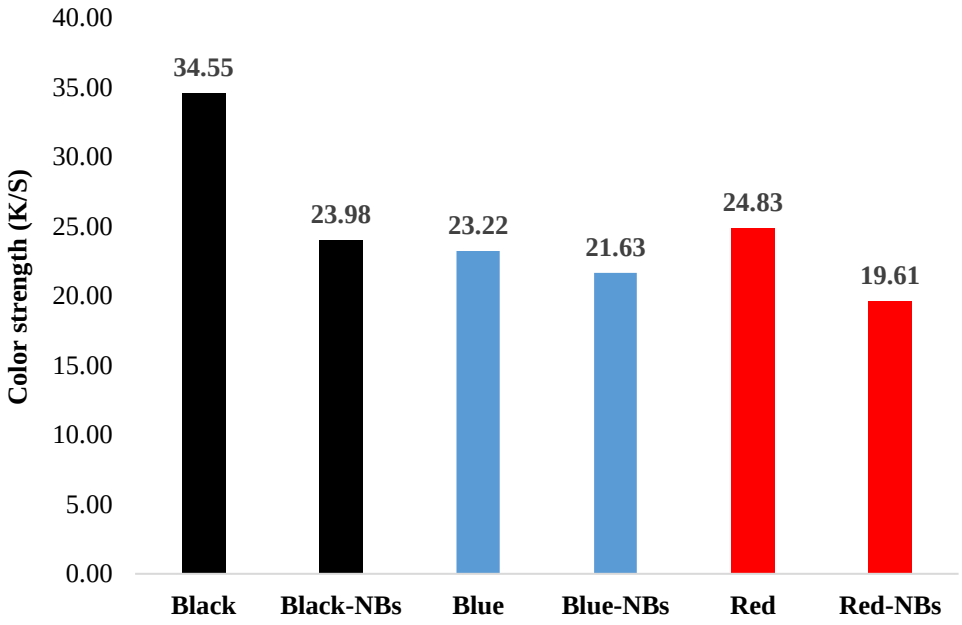


Figure 3. Color strengths of the dyed samples

The presence of NBs in the dyeing bath caused lower color strengths, especially in black and red. Although the effect of NBs on the color strength of the blue color was not as significant as those of the others, it still caused a slight decrease. It has also been revealed in previous studies that the use of NBs causes a decrease in color strength (Anis et al. 2022, Toprak-Cavdur 2023). This situation could have explained by (i) the NBs deterioration the structure of dye molecules (Chen et al. 1999, Ismail and Sakai 2022, Mahanna and El-Bendary 2022) by increasing the dissolved oxygen level (Rojviroon and Rojviroon 2022), (ii) the zeta potential of the fiber surface cannot be reduced due to the adsorption of some of the Na^+ ions in the dyeing medium onto the NBs (Atkinson et al. 2019, Yasuda 2023), and (iii) the interfacial free energy and high surface area of NBs (Atkinson et al. 2019, Ma et al. 2022) could have prevented the dye aggregates from accumulating on the fiber surface.

Washing Fastness

Washing fastness values of fabrics are given in Table 2.

Table 2. Washing fastness of samples

	Staining					
	AC	**CO**	**PA**	**PES**	**AC**	**WO**
Black	4/5	4/5	4/5	4/5	4/5	4/5
Black-NBs	4/5	4	4/5	4/5	4/5	4/5
Blue	4/5	4/5	4/5	4/5	4/5	4/5
Blue-NBs	4/5	4	4/5	4/5	4/5	4/5
Red	4/5	4/5	4/5	4/5	4/5	4/5
Red-NBs	4/5	3/4	4/5	4/5	4/5	4/5

The washing fastness of the dyed samples with/without NBs were good to excellent. The use of NBs in dyeing did not cause changes staining of different fabrics, except cotton. NBs, like other substances (Wu et al. 2008, Liu and Craig 2009, Ushida et al. 2012), could have effectively removed unfixed/hydrolyzed dye molecules from the fiber surface, resulting in half point lower CO staining values. In general, the reasons for the high washing fastness values of the fabrics after dyeing could be explained by the strong covalent bonding of dye

anions to cotton (Lewis 2011).

CONCLUSION

In this study, an environmentally-friendly textile process was designed using innovative NB water technology. According to the colorimetry results, the NBs effected L*-a*-b* values. Obtaining lighter shades compared to the traditional ones when using NBs in dyeing could have been explained by the deterioration of reactive dye molecules (Chen et al. 1999, Ismail and Sakai 2022, Mahanna and El-Bendary 2022). This situation was also supported by the lower color strengths obtained with using NBs compared to conventional dyeings. The effective removal of unfixed/hydrolyzed dye molecules from the fiber surface, could have been effective in obtaining a half point lower cotton staining values with NBs compared to the conventional ones. According to these results, it is concluded that it could be beneficial to investigate the use of NB technology in detail, especially in washing.

REFERENCES

Akgun M, Becerir B, Alpay HR 2014. Reflectance Prediction of Colored Polyester Fabrics by A Novel Formula. Fibers Polym, 15:126–137.

Anis P, Toprak-Cavdur T, Çalışkan N 2022. Oxygen-enriched Nanobubbles for a Green Reactive Washing Process. AATCC J Res, 9:152–160.

Arivithamani N, Giri Dev VR 2018. Characterization and Comparison of Salt-free Reactive Dyed Cationized Cotton Hosiery Fabrics with that of Conventional Dyed Cotton Fabrics. J Clean Prod, 183:579–589

Atkinson AJ, Apul OG, Schneider O, Garcia-Segura S, Westerhoff P 2019. Nanobubble Technologies Offer Opportunities to Improve Water Treatment. Acc Chem Res, 52:1196–1205.

Azevedo A, Oliveira H, Rubio J 2019. Bulk Nanobubbles in the Mineral and Environmental Areas: Updating Research and Applications. Adv Colloid Interface Sci, 271:101992.

Bui TT, Han M 2020. Decolorization of Dark Green Rit Dye Using Positively Charged Nanobubbles Technologies. Sep Purif Technol, 233:116034.

Burkinshaw SM, Mignanelli M, Froehling PE, Bide MJ 2000. The Use of Dendrimers to Modify the Dyeing Behaviour of Reactive Dyes on Cotton. Dye Pigment, 47:259–267

Chen G, Lei L, Yue PL 1999. Wet Oxidation of High-Concentration Reactive Dyes. Ind Eng Chem Res, 38:1837–1843.

Endo-Takahashi Y, Negishi Y 2020. Microbubbles and Nanobubbles with Ultrasound for Systemic Gene Delivery. Pharmaceutics, 12:1–14.

Gopalakrishnan M, Punitha V, Saravanan D 2019. Water Conservation in Textile Wet Processing. In: Muthu SS (ed), Water in Textiles and Fashion: Consumption, Footprint, and Life Cycle Assessment. Cambridge: Woodhead Publishing, pp 135–153.

Grancarić AM, Ristić N, Tarbuk A, Ristić I 2013. Electrokinetic Phenomena of Cationised Cotton and Its Dyeability with Reactive Dyes. Fibres Text East Eur, 21:106–110

Haris S, Qiu X, Klammler H, Mohamed MMA 2020. The Use of Micro-Nano Bubbles in Groundwater Remediation: A Comprehensive Review. Groundw Sustain Dev, 11:100463.

Ismail GA, Sakai H 2022. Review on Effect of Different Type of Dyes on Advanced Oxidation Processes (AOPs) for Textile Color Removal. Chemosphere, 291:132906.

Khatri A, Padhye R, White M 2013. The Use of Trisodium Nitrilo Triacetate in the Pad-steam Dyeing of Cotton with Reactive Dyes. Color Technol, 129:76–81.

Khatri A, White M 2016. Sustainable Dyeing Technologies. In: Blackburn R (ed), Sustainable Apparel: Production, Processing and Recycling. Cambridge: Woodhead Publishing, pp 135–160.

Lewis DM 2011. The Chemistry of Reactive Dyes and Their Application Processes. In: Clark M (ed), Handbook of Textile and Industrial Dyeing Volume 1. Cambridge: Woodhead Publishing, pp 303–364.

Liu G, Craig VSJ 2009. Improved Cleaning of Hydrophilic Protein-Coated Surfaces Using The Combination

of Nanobubbles and SDS. ACS Appl Mater Interfaces, 1:481–487.

Ma P, Han C, He Q, Miao Z, Gao M, Wan K, Xu E 2022. Oxidation of Congo Red by Fenton Coupled with Micro and Nanobubbles. Environ Technol, 44:2539-2548.

Mahanna H, El-Bendary N 2022. Enhanced Catalytic Oxidation of Reactive Dyes by Reuse of Adsorption Residuals as A Heterogeneous Catalyst with Persulfate/UV Process. Int J Environ Sci Technol, 1–12.

Meegoda JN, Hewage SA, Batagoda JH 2019. Application of the Diffused Double Layer Theory to Nanobubbles. Langmuir, 35:12100–12112.

Minamikawa K, Makino T 2020. Oxidation of Flooded Paddy Soil through Irrigation with Water Containing Bulk Oxygen Nanobubbles. Sci Total Environ, 709:136323.

Mohsin M, Sardar S, Hasan M, Akhtar K S, Anama W, Ijaz S, Hassan A 2022. Water Efficient, Eco-friendly and Effluent Free Nano bubble Finishing of Cotton Fabric. J Nat Fibers, 19:12586-12595.

Mohsin M, Sardar S, Hassan M, Akhtar N, Hassan A, Sufyan M 2020. Novel, Sustainable and Water Efficient Nano Bubble Dyeing of Cotton Fabric. Cellulose, 27:6055–6064.

Qian J, Craig VSJ, Jehannin M 2019. Long-Term Stability of Surface Nanobubbles in Undersaturated Aqueous Solution. Langmuir, 35:718–728.

Ramasamy M, Kandasaamy P V 2005. Effect of Cationization of Cotton on It's Dyeability. Indian J Fibre Text Res, 30:315–323.

Rojviroon O, Rojviroon T 2022. Photocatalytic Process Augmented with Micro/Nano Bubble Aeration for Enhanced Degradation of Synthetic Dyes in Wastewater. Water Resour Ind, 27:100169.

Sakr M, Mohamed MM, Maraqa MA, Hamouda M A, Hassan A A, Ali J, Jung J 2022. A Critical Review of the Recent Developments in Micro–nano Bubbles Applications for Domestic and Industrial Wastewater Treatment. Alexandria Eng J, 61:6591–6612.

Takahashi M, Chiba K, Li P 2007. Free-radical Generation from Collapsing Microbubbles in the Absence of A Dynamic Stimulus. J Phys Chem B, 111:1343–1347.

Toprak-Cavdur T 2023. A Novel and Integrated Approach to Dyeing Recycled Cotton and Chitosan Yarns Blended Fabrics with Innovative Nanobubble Technology. Fibers Polym.

Toprak-Cavdur T, Uysal S, Anis P 2022. Dyeing Behavior and Characterization of Recycled Cotton and Chitosan Blended Fabrics. J Nat Fibers, 19:13243-13257.

Ushida A, Hasegawa T, Takahashi N, Nakajima T, Murao S, Narumi T, Uchiyama H 2012. Effect of Mixed Nanobubble and Microbubble Liquids on The Washing Rate of Cloth in An Alternating Flow. J Surfactants Deterg, 15:695–702.

Varadarajan G, Venkatachalam P 2016. Sustainable Textile Dyeing Processes. Environ Chem Lett, 14:113–122.

Wang L, Ali J, Wang Z, Oladoja NA, Cheng R, Zhang C, Mailhot G, Pan G 2020. Oxygen Nanobubbles Enhanced Photodegradation of Oxytetracycline under Visible Light: Synergistic Effect and Mechanism. Chem Eng J, 388:124227.

Weijs JH, Seddon JRT, Lohse D 2012. Diffusive Shielding Stabilizes Bulk Nanobubble Clusters. ChemPhysChem, 13:2197–2204.

Wu J, Zhang K, Cen C, Wu X, Mao R, Zheng Y 2021. Role of Bulk Nanobubbles in Removing Organic Pollutants in Wastewater Treatment. AMB Express 11:96.

Wu Z, Chen H, Dong Y, Mao H, Sun J, Chen S, Craig V S J, Hu J 2008. Cleaning Using Nanobubbles: Defouling by Electrochemical Generation of Bubbles. J Colloid Interface Sci, 328:10–14.

Yasuda K 2023. Characteristics of Ultrafine Bubbles (Bulk Nanobubbles) and Their Application to Particle-Related Technology. KONA Powder Part J, 1–14

Yasui K, Tuziuti T, Kanematsu W 2018. Mysteries of Bulk Nanobubbles (Ultrafine Bubbles); Stability and Radical Formation. Ultrason Sonochem, 48:259–266.

Yasui K, Tuziuti T, Kanematsu W, Kato K 2016. Dynamic Equilibrium Model for a Bulk Nanobubble and a Microbubble Partly Covered with Hydrophobic Material. Langmuir, 32:11101–11110.

Zhang Y, Shahid-ul-Islam, Rather LJ, Li Q 2022. Recent Advances in the Surface Modification Strategies to Improve Functional Finishing of Cotton with Natural Colourants - A review. J Clean Prod, 335:130313.

Zhou L, Wang S, Zhang L, Hu J 2021. Generation and Stability of Bulk Nanobubbles: A Review and Perspective. Curr Opin Colloid Interface Sci, 53:101439.

Zhou Y, Li Y, Liu X, Wang K, Muhammad T 2019. Synergistic Improvement in Spring Maize Yield and Quality with Micro/Nanobubbles Water Oxygation. Sci Rep, 9:1–10.

DRAFT

ORAL PRESENTATION

Kuşlarda tüylerin yaratıcı ve ilginç kullanımı

Sema Özkadif[1*] (*ORCID: https://orcid.org/*0000-0002-5398-9874)

[1*]Çukurova Üniversitesi, Ceyhan Veteriner Fakültesi, Anatomi Anabilim Dalı, Adana, Türkiye

*semaerten80@gmail.com

Özet

Kuşlar özellikle kanatları ve tüyleri ile karakterize olan, Aves sınıfına giren sıcakkanlı omurgalı hayvanlardır. Bazı kuşlar uçma yeteneklerini kaybetmişlerdir. Tüyler derinin epidermal tabakasından üretilmiş beta-keratinden oluşan yapılardır. Göz alıcı renk ve yapıya sahip olan tüyler sadece uçma fonksiyonuna sahip olmayıp, çeşitli işlevlere sahiptirler. Bunlar; uçma, sıcak tutmaya yardımcı olma, vücut sıcaklığı kontrolü, yüzme ve dalış, karda yürüme, kızakla kayma, denge ve destek, işitme, yiyecek arama ve yeme, temiz tutma, his, sindirime yardımcı olma, ses çıkarma, yuva yapma, su taşıma, avcılardan kaçma, görsel sinyaller gönderme, kamuflaj, koku, çiçeklerin döllenmesine yardımcı olma ve rüzgâr, nem ve güneşten koruma olarak ifade edilebilir. Tüm kuşların ayırt edici bir özelliği olan tüyler, küçük ama dayanıklı yapılarıyla kuşları yaralanmaktan da korurlar. Birçok fonksiyona sahip olan tüyler olmadan kuşların yaşaması pek de mümkün görünmemektedir.

Anahtar Kelimeler: kuş, tüy, fonksiyon

Creative and interesting use of feathers in birds

Abstract

Birds are warm-blooded vertebrate animals belonging to the Aves class, characterized especially by their wings and feathers. Some birds have lost their ability to fly. Feathers are structures composed of beta-keratin produced from the epidermal layer of the skin. Feathers with eye-catching color and structure do not only have the function of flight, but also have various functions. These can be expressed as; flying, helping to keep warm, controlling body temperature, swimming and diving, walking on snow, sledding, balance and support, hearing, foraging and eating, keeping clean, feeling, aiding digestion, making sounds, building nests, carrying water, escaping from predators, sending visual signals, camouflage, scent, helping pollinate flowers and protecting from wind, moisture and sun. Feathers, a distinctive feature of all birds, also protect birds from injury with their small but durable structure. It does not seem possible for birds to survive without feathers, which have many functions.

Keywords: bird, feather, function

GİRİŞ

Kuşlar sıcakkanlı omurgalılar olup, Aves sınıfına girerler. Kuşlar tüm dünyada her ortamda ortaya çıkmıştır. Pek çok kuş uçmanın yanı sıra koşabilir, engelleri aşabilir ve yüzebilir. Penguenler gibi bazı kuşlar kanatlarını korumuş, ancak uçma yeteneklerini kaybetmişlerdir (Murakami, 2022).

Tüyler, bugün yalnızca kuşlarda bulunan, epidermal beta-keratinden oluşan boru şeklindeki yapılardır (Prum, 1999). Tüyler, kuş biyolojisi için son derece önemlidir ve deriden üretilmiş en önemli yapılarından biridir (Murakami, 2022). Kuşların derileri diğer omurgalı hayvanlara göre oldukça incedir. Eğer vücut tüylerle örtülü olmasaydı ağaç ve diğer sert cisimlerle sık sık yaralanırdı (Kuru, 1999).

Bazı kuş tüylerinin harika renkleri vardır. Bu renkler, ışığın kırılması ile oluşturulur. Yanardönerlik denilen bu ışık türü bir nesneye baktığınız açıya bağlı olarak renk değiştirebilir (Kazilek, 2009). Yanardönerlik, düzenli hava boşluklarının, melanin tüplerinin, keratin liflerinin düzenlenmesinin vb. üretiminden kaynaklanan ışık

girişimiyle üretilir ve tondaki değişiklikler, tüylere farklı açılardan bakıldığında aralık değişikliklerinden kaynaklanır (Yu ve ark., 2004).

TÜYLERİN İŞLEVLERİ

Son dönem kuşlarının tüylerinin bir takım işlevleri ve biyolojik rolleri vardır (Bock, 2000). Karmaşık bir yapıya sahip olan tüylerin görevleri; uçma, sıcak tutmaya yardımcı olma, vücut sıcaklığı kontrolü, yüzme ve dalış, karda yürüme, kızakla kayma, denge ve destek, işitme, yiyecek arama ve yeme, temiz tutma, his, sindirime yardımcı olma, ses çıkarma, yuva yapma, su taşıma, avcılardan kaçma, görsel sinyaller gönderme, kamuflaj, koku, çiçeklerin döllenmesine yardımcı olma ve rüzgâr, nem ve güneşten koruma şeklinde sıralanabilir (Stettenheim, 2006; Terrill ve Shultz, 2023).

1. Uçma

Uçma tüyleri, kuşların kanatlarında bulunan çok güçlü ve sert tüylerdir (Stettenheim, 2006). Kuşlar, kanatlardan ve kuyruktan elde edilen kaldırma gücü sayesinde güçlü uçuş ve süzülme elde ederler (Maybury ve ark., 2001). Uçan kuşların kanatları ve kuyrukları tarafından oluşturulan kaldırma kuvvetinin sağladığı manevra kabiliyetine ek olarak, bazı kuş türlerinin havada manevra yapmaya yardımcı olan uzun kuyrukları vardır (Terrill ve Shultz, 2023). Kuyruk tüyleri yönlendirme için kullanır (Murakami, 2022).

Suda uçma, havada uçmaya kıyasla kanadın yapısal bileşenlerine ve tüylerine çok farklı mekanik kuvvetler uygular. Bu ikilik, hava ve suyun fiziksel özelliklerindeki önemli farklılıklardan kaynaklanmaktadır (Johnson, 2012).

Uçamayan kuşlar genellikle üç kategoriye ayrılabilir; (a) uçmayan ancak su altında 'uçmak' için ilgili yapıları kullanan kanatlı dalgıçlar, (b) tamamen uçamayan kuşlar ve kanatlı olmayan dalgıçlar ve (c) yıllık döngüleri sırasında uçuşlarını kaybeden ve yeniden kazanan türler (Kulp et al., 2018).

2. Sıcak tutmaya yardımcı olma: Tüyler, kuşun vücuduna yakın hava ceplerini hapsederek, onu sıcak tutmaya yardımcı olur. Kuşlar soğukta tüylerini kabartarak, vücut ısısını hapseder ve daha sıcak kalmak için fazladan hava ekler (Stettenheim, 2006). Genel olarak, soğuk ortamlarda yaşayan kuşların daha uzun tüyleri vardır ve daha fazla havayı hapsetmek için diken yoğunlukları azdır (Pap ve ark, 2017). Bununla birlikte, kısmi su geçirgenliğine izin vererek kaldırma kuvvetini azaltan tüyler bile vücudun yakınında küçük bir hava tabakasını hapseder ve bu da çok soğuk suda bile etkili bir yalıtkan görevi görür (Gremillet ve ark, 2005).

Tüyler, kuşların şiddetli rüzgârlara maruz kaldıkları ve buzlu kutup sularında dalış yaptıkları gibi en zorlu ortamlarda hayatta kalmalarını ve ortam sıcaklıkları -40°C'nin altına düştüğünde bile sabit bir yüksek vücut sıcaklığını sürdürmelerini sağlar (Pap ve ark, 2020).

3. Vücut sıcaklığı kontrolü: Tüyler ayrıca sıcak ortamlarda ısı düzenleyici bir rol oynar. Tüylerin termoregülasyondaki rolü, kuşları serin tutmak için güneş radyasyonunu yansıtmasıdır (Medina ve ark, 2018). Kuşlar vücut ısısını sabit tutmak için, çok ısınmışsa soğuması için başlarını ve ayaklarını havaya veya suya maruz bırakabilir veya ısınmaya yardımcı olmak için tüylerinin arasına sıkıştırabilir (Stettenheim, 2006).

4. Yüzme ve dalış: Pek çok kuş, yapısal değişikliklerle tüylerini su geçirmez hale getirir (Terrill ve Shultz, 2023). Kuşlar, tüylerinde hava tutma oranını artırarak kaldırma kuvvetini artırabilir (Lovvorn ve Jones, 1991). Ördek gibi su kuşları, tüylerde hapsolmuş havayı kullanarak suda yüzebilir ve soğuk sudan koruma sağlayabilir (Dove ve Agreda, 2007). Bazı suda yaşayan kuşların tüylerinde ayrıca daha yüksek nodus yoğunluğu vardır, bu da kuş tüyünün kohezyonunu (yapışkanlık) artırabilir ve bu da onu sudan gelen basınca karşı daha dirençli hale getirir (D'Alba ve ark, 2017). Yağlar suyun yüzey gerilimini azaltır ve bu nedenle kuşlar çevrelerinde petrolle karşılaştıklarında su geçirmezlikleri ciddi şekilde tehlikeye girebilir (Whitmer ve ark., 2018).

Tüm kanatlı dalgıçlar yüzerken tüy kullanırlar. Bu kuşlar tüylerinde çeşitli morfolojik adaptasyonlar gösterirler, bu da genellikle kanatların yüzmek ve uçmak için kullanılması arasındaki ödünleşmeyi yansıtır. Havadan uçuş yeteneklerini koruyan dalış kuşları, uzun, ince birincil ve ikincil uçlara sahip olma eğilimindedir (Spear ve Ainley, 1997). Penguenler kanatlarını geliştirerek onları harika yüzücüler yapan sert, düz yüzgeçlere dönüştürmüşlerdir (Stettenheim, 2006).

5. Karda yürüme: Sıra dışı tüy kullanımlarından biri de karda yürüyüş yapmaktır. Karla kaplı bölgelerde yaşayan tavuğu andıran orman tavuğu kuşlarının, kışın tıpkı kar ayakkabısı gibi ayak boyutunu artıran tüylerle kaplı ayakları vardır. Bu, kuşların kara batmasını önler (Stettenheim, 2006).

6. Kızakla kayma: Antartika kuşları, karınlarının pürüzsüz tüylerinin üzerine çökerler ve palet benzeri kanatlarını ayaklarıyla birlikte kullanarak karda ve buzda kızak gibi hareket ederler (Stettenheim, 2006).

7. Denge ve Destek: Pek çok kuş, uçmadığı zamanlarda, ağaçkakanlarda görüldüğü gibi, yerdeyken veya ağaçların kenarlarına tırmanırken kuyruk tüylerini destek olarak kullanır (Stettenheim, 2006). Akrobatik olarak yiyecek arama eğiliminde olan kuşlar, kendilerini desteklemek için kuyruklarını kullanırlar ve baş aşağı veya kısaca dikey yüzeylere karşı yiyecek ararken denge sağlarlar (Manegold, 2008). Ayrıca kuyruk tüyleri, dikey bir yüzeye yapışarak uyuyan kuşlar için de yararlı olabilir (Van Els ve Whitney, 2011).

8. İşitme: Bazı yırtıcı hayvanlar, özellikle baykuşlarda görülür (Hausmann et al., 2009). Baykuşlar sesleri toplamak ve kulaklarına yönlendirmek için yüz tüylerini iki tabak (yüz diskleri) gibi düzenler, böylece karanlıkta avlarını daha doğru bir şekilde bulabilirler (parabolik reflektör) (Stettenheim, 2006).

9. Yiyecek arama ve yeme: Göl ve akarsu sularında balık avlayan balıkçıllar gibi bazı kuşlar, bazen tüylerini başlarının üzerinde bir şemsiye oluşturmak için kullanırlar. Bu, suda balıkları görmelerini kolaylaştırabilir. Örneğin karabalıkçılı, avını çeken gölge oluşturmak için kanatlarını kullanır. Birçok böcekçil kuşun kanatlarında veya kuyruğunda yiyecek arama sırasında düzenli ve aniden ortaya çıkan gizli beyaz noktalar vardır (Jablonski, 2001).

Tüyler, gözleri ve burun deliklerini koruyarak, bıyık benzeri dokunsal sensörler olarak hareket ederler (Terrill ve Shultz, 2023). Bazı böcek yiyen kuşların ağızlarının çevresinde riktal kıllar adı verilen özel uzun tüyler bulunur. Bunlar havadaki böceği yakalamak için bir huni görevi görebilir veya böceği yakalarken gözleri koruyabilir. Diğer kuşlar, meyveleri seçmek için ağızlarının yan tarafındaki tüyleri kullanır (Stettenheim, 2006).

10. Temiz tutma: Tüyler, deri üzerinde fiziksel bir bariyer oluşturarak veya kimyasal caydırıcılar üreterek kuşları parazitlerden korur (Terril ve Shultz, 2023). Birçok kuş türü, sivrisinekleri caydırmak için çoğunlukla baş ve ayaklarının hareketine ve konumuna güvenir (Darbro ve Harrington, 2007). Balıkçıllar gibi bazı kuşların, normal tüylere sürtmek ve onları uygun durumda tutmak için gagaları ve ayaklarıyla ezdikleri, toz tüyü adı verilen küçük tüyleri vardır. Bu pudra tüyü ayrıca akarlar gibi tüy parazitlerini kontrol etmeye de yardımcı olabilir (Stettenheim, 2006). Kuşlar, kimyasal savunmalar ve iç tüy pigmentleri kullanarak tüylerini bu bakterilere karşı korurlar (Ruiz-Rodriguez ve ark., 2009). Kuşun diğer vücut parçalarının yanı sıra tüylerinde de zehirli kimyasalların bulunduğu ve birçok amfibi derisinde bulunana benzer bir kimyasal savunma görevi görebilir (Dumbacher ve Pruett-Jones, 1996).

11. His: Tüylerin sinirleri yoktur, ancak tüyün kuşa bağlandığı yeri çevreleyen sinirleri uyarırlar. Kuşlar, bu sinirlerin uyarılmasına bağlı olarak tüylerinin pozisyonunu ve duruşunu ayarlayabilirler (Stettenheim, 2006).

12. Sindirime yardımcı olma: Bazı balık yiyen kuşlar kendi tüylerini yerler. Bu, kuşu keskin balık kemiklerinden korumaya yardımcı olur (Stettenheim, 2006). Bununla birlikte, kimyasal sindirim için midede yiyecek tutma ve sindirilmemiş yiyeceklerin bağırsağa girmesini filtreleme işlevi gördüğü de belirtilmektedir (Jehl, 2017). Tüy yeme, tutsak kuşlarda, özellikle papağanlarda ve tavuklarda da bilinmektedir, ancak bu durumda işlevsel olmayan bir davranış bozukluğu gibi görünmektedir. (McKeegan ve Savory, 1999).

13. Ses çıkarma: Kuş seslerini şarkı ya da uğultu olarak düşünürüz, ancak bazı kuşlar tüylerini kullanarak uğultu, davul ve ıslık gibi birçok farklı ses çıkarabilirler. Baykuş gibi geceleri avlanan kuşlar, avlarına yaklaşırken kanatlarını kullanarak kendi seslerini boğabilirler (Stettenheim, 2006).

14. Yuva yapma: Kuşlar yuvalarını yaparken kendi tüylerini ve diğer kuşların tüylerini kullanırlar (Winkler, 1993). Pek çok kuş (özellikle su kuşları) yuvalarını kuş tüyleriyle kaplar. Bu, yumurtalarını sıcak tutmaya yardımcı olur ve ayrıca yumuşak bir dolgu sağlar. Muhabbet kuşu gibi bazı kuşlar, otları ve yaprakları yuvalarına taşımak için aslında altlarında ve alt sırtlarında bulunan tüyleri kullanırlar (Stettenheim, 2006).

15. Su taşıma: Çölde yaşayan bazı kuşların (kum tavuğu gibi) su tutmada çok iyi olan özel karın tüyleri vardır (Benitez-Lopez ve ark., 2014).Yumurtaları ve civcivleri büyütürken, birçok yetişkin kuş yuvaya dönmeden önce tüylerini karınlarına batırır. Daha sonra yumurtaların kurumasını önlemek ve civcivlerine su vermek için

suyu kullanabilirler. Bu adaptasyon, su birikintilerine yakın bölgelerde bulunan daha yüksek sayıda avcıdan kaçınmak için su deliklerinden daha uzakta yuva yapmalarını sağlar (Stettenheim, 2006).

16. Avcılardan kaçma: Kuşlar saldırıya uğradıklarında veya korktuklarında kuyruk tüylerinin bir kısmını düşürebilirler. Buna korku tüy dökümü denir. Bu bazen kuşun kaçmasına yardımcı olur ve saldırganın yalnızca ağzı veya ayağı tüylerle dolu kalır (Stettenheim, 2006).

17. Görsel sinyaller gönderme: Tüy renkleri ve desenleri, eşlere ve rakiplere sinyal göndermek için kullanılır. Bu muhtemelen tüylerin en büyük ve en çok kullanılan işlevidir (Stettenheim, 2006). Tüy fenotipleri cinsiyet hormonları tarafından değiştirilebilmektedir (Yu ve ark., 2004). Erkek kuşlar dişilere göre daha renkli olma eğilimindedir ve bu cinsel dimorfizmi tanımlamaktadır (McGraw ve ark, 2002). En iyi bilinen örneği erkekleri cinsel teşhirde kullanılmak üzere olağanüstü tüy renkleri ve şekilleri sergileyen cennet kuşlarıdır (Scholes, 2008).

Tüyler çeşitli renklerde evrimleşmiştir. Tüyler, çarpıcı desenlerde dağılmış olağanüstü bir renk çeşitliliği sergiler. Renk, kimyasal renk, fiziksel renk veya her ikisinin kombinasyonu ile oluşturulabilir. Pigmentler, ışığı belirli dalga boylarında emen kimyasal bileşiklerdir. Kuşlarda üç ana pigment türü vardır: melaninler, karotenoidler ve porfirinler (Lucas ve Stettenheim, 1972).

18. Kamuflaj: Bazen parlak renkler iyi değildir. Yırtıcı hayvanlar tarafından görülmemek için birçok kuşun ölü yapraklara veya yaşadıkları çevrenin diğer kısımlarına benzeyen tüyleri vardır, bu nedenle yırtıcılar onları göremez. Bazı yırtıcı hayvanlar da avlarının daha yakına gelebilmesi ve avın yakalanmasını kolaylaştırmak için karışmayı severler (Stettenheim, 2006).

19. Koku: Bazı tüyler kendine özgü kokular yayar. En çok çalışılan tüy kokusu, tepeli alklet'in tüylerinin mandalina kokusudur (Hagelin ve ark., 2003).

20. Çiçeklerin döllenmesine yardımcı olma: Sinek kuşlarının, başlarının etrafındaki tüyler bir çiçekten polen toplarken çiçeklerin tozlaşmasına yardımcı olur. Daha fazla nektar aramaya devam ettikçe, polen diğer çiçeklere aktarılır (Stettenheim, 2006).

21. Rüzgâr, nem ve güneşten koruma: Güçlü ve çıkıntılı kontur tüyleri, kuşları rüzgardan korur. Tüylerin yapı taşını oluşturan beta-keratin sert bir malzeme olup, suya ve aşınmaya dayanıklıdır. Daha koyu renkli tüyler de güneşten koruma sağlayabilir. Tüyler ayrıca kuşları yağmurdan korur. Birbirine geçen tüy dikenleri ve yağlı veya mumlu özel kaplama ile suyun aktığı bir kalkan oluşturur (Stettenheim, 2006).

SONUÇ

Birçok işleve sahip olan tüyler, kuşların karakterize özelliğini yansıtmaktadır. Güzel görünümünün yanında birçok hayati öneme sahiptir.

KAYNAKLAR

Benitez-Lopez A, Vinuela J, Suarez F, Hervas I, Garcia JT 2014. Niche-habitat mechanisms and biotic interactions explain the coexistence and abundance of congeneric sandgrouse species. Oecologia, 176(1), 193-206.

Bock WJ 2000. Explanatory History of the Origin of Feathers. American. Zoologist, 40:478-485.

D'Alba L, Carlsen TH, Asgeirsson A, Shawkey MD, Jonsson JE 2017. Contributions of feather microstructure to eider down insulation properties. Journal of Avian Biology, 48:1150-1157.

Darbro JM, Harrington LC 2007. Avian defensive behavior and bloodfeeding success of the West Nile vector mosquito, Culex pipiens. Behavioral Ecology, 18(4), 750-757.

Dove CJ, Agreda A 2007. Differences in plumulaceous feather characters of dabbling and diving ducks. The Condor, 109(1): 192-199.

Dumbacher JP, Pruett-Jones S 1996. Avian chemical defense. Current Ornithology, 13: 137-174.

Gremillet D, Chauvin C, Wilson RP, Le Maho Y, Wanless S 2005. Unusual feather structure allows partial plumage wettability in diving great cormorants Phalacrocorax carbo. Journal of Avian Biology, 36(1), 57-63.

Hagelin JC, Jones IL, Rasmussen LEL 2003. A tangerine-scented social odour in a monogamous seabird. Proceedings of the Royal Society of London B: Biological Sciences, 270 (1522), 1323-1329.

Hausmann, L,Campenhausen M, Endler F, Singheiser M, Wagner H 2009. Improvements of sound localization abilities by the facial ruff of the barn owl (*Tyto alba*) as demonstrated by virtual ruff removal. PLoS One, 4(11): e7721.

Jablonski PG 2001. Sensory exploitation of prey: manipulation of the initial direction of prey escapes by a conspicuous rare enemy. Proceedings of the Royal Society of London. Series B: Biological Sciences, 268(1471): 1017-1022.

Jehl JR 2017. Feather-eating in grebes: a 500-year conundrum. The Wilson Journal of Ornithology, 129(3): 446-458.

Johnson KE 2012. Feather biomechanics of penguins and other seabirds. Theses Digitization Project, 4166. Available at: https://scholarworks.lib.csusb.edu/etd-project/4166 [28.06.23]

Kazilek CJ 2009. Feather biology. Available at: https://askabiologist.asu.edu/explore/feather-biology [28.06.23]

Kulp FB, D'Alba L, Shawkey MD, Clarke JA 2018. Keratin nanofiber distribution and feather microstructure in penguins. The Auk, 135(3), 777-787.

Kuru M 1999. Omurgalı Hayvanlar. Palme Yayıncılık. Ankara, pp, 437.

Lovvorn JR, Jones DR 1991. Body mass, volume, and buoyancy of some aquatic birds, and their relation to locomotor strategies. Canadian Journal of Zoology, 69(11): 2888-2892.

Lucas AM, Stettenheim PR 1972. Avian Anatomy – Integument Agricultural Handbook 362: Agricultural Research Services. US Department of Agriculture: Washington DC, pp. 272.

Manegold A 2008. Earliest fossil record of the Certhioidea (treecreepers and allies) from the early Miocene of Germany. Journal of Ornithology, 149(2): 223-228.

Maybury WJ, Rayner JMV, Couldrick LB 2001. Lift generation by the avian tail. Proceedings of the Royal Society of London. Series B: Biological Sciences, 268(1475): 1443-1448.

Mcgraw KJ, Hill GE, Stradi R, Parker RS 2002. The effect of dietary carotenoid access on sexual dichromatism and plumage pigment composition in the American goldfinch. Comparative Biochemistry and Physiology Part B Biochemical and Molecular Biology, 131:261-9.

McKeegan DE, Savory CJ 1999. Feather eating in layer pullets and its possible role in the aetiology of feather pecking damage. Applied Animal Behaviour Science, 65(1): 73-85.

Medina I, Newton E, Kearney MR, Mulder RA, Porter WP, Stuart-Fox D 2018. Reflection of near-infrared light confers thermal protection in birds. Nature Communications, 9(1): 1-7.

Murakami Y 2022. Basic Anatomy and physiology of birds. Entomol, Ornithol & Herpetol, 11: 1-1

Pap PL, Osvath G, Daubner T, Nord A, Vincze O 2020. Down feather morphology reflects adaptation to habitat and thermal conditions across the avian phylogeny. Evolution, 74(10): 2365-2376.

Pap PL, Vincze O, Wekerle B, Daubner T, Vagasi, CI, Nudds RL, Dyke GJ, Osvath, G 2017. A phylogenetic comparative analysis reveals correlations between body feather structure and habitat. Functional Ecology, 31(6): 1241-1251.

Prum RO 1999. Development and Evolutionary Origin of Feathers. Journal of Experımental Zoology, 285:291-306.

Ruiz-Rodriguez M, Valdivia E, Soler JJ, Martin-Vivaldi M, Martin- Platero AM, Martinez-Bueno M 2009. Symbiotic bacteria living in the hoopoe's uropygial gland prevent feather degradation. Journal of Experimental Biology, 212(22): 3621-3626.

Scholes E III 2008. Evolution of the courtship phenotype in the bird of paradise genus Parotia (Aves: Paradisaeidae): homology, phylogeny, and modularity. Biological Journal of the Linnean Society, 94(3), 491-504.

Spear LB, Ainley DG 1997. Flight behaviour of seabirds in relation to wind direction and wing morphology. Ibis, 139(2): 221-233.

Stettenheim P 2006. What Feathers Do. Birder's World, 25-34. Available at: https://askabiologist.asu.edu/content/23-functions-feathers [28.06.23]

Van Els P, Whitney BM 2011. Arboreal roosting as a possible explanation for tail stiffness in the genus Sclerurus. Ornitologia Neotropical, 22: 477-479.

Whitmer ER, Elias BA, Harvey DJ, Ziccardi MH 2018. An experimental study of the effects of chemically dispersed oil on feather structure and waterproofing in common murres (*Uria aalge*). Journal of Wildlife Diseases, 54(2), 315-328.

Winkler DW 1993. Use and importance of feathers as nest lining in tree swallows (*Tachycineta bicolor*). The Auk, 110(1): 29-36.

Yu M, Yue Z, Wu P, Wu DY, Mayer JA, Medina M, Widelitz RB., Jiang TX, Chuong CM 2004. The developmental biology of feather follicles. International Journal of Developmental Biology, 48(0): 181-191.

DRAFT

ORAL PRESENTATION

Effectiveness of tannic acid and ascorbic acid on the synthesis process of silver nanoparticles

Awaleh Daher Waberi[1], Mücahit Uğur[1] (*ORCID: https://orcid.org/0000-0002-3746-5683*), Özge Bildi Ceran[1] (*ORCID: https://orcid.org/0000-0002-3147-735X*), Barış Şimşek[1*] (*ORCID: https://orcid.org/0000-0002-0655-4368)*, Ömer Faruk Dilmaç[1] (*ORCID: https://orcid.org/0000-0002-9660-0638)*

[*]Çankırı Karatekin University, Faculty of Engineering, Department of Chemical Engineering, Çankırı, Turkey.

*Corresponding author e-mail: barissimsek@karatekin.edu.tr

Abstract

Silver nanoparticles have been one of the nanomaterials that have been widely studied in recent years due to their unique antibacterial and mechanical properties. Reducing agents play a key role in the synthesis of silver nanoparticles with desired features. Hydrazine and sodium boron hydride are used as reducing agents, however, the fact that these agents are carcinogenic and toxic materials. Green reducers such as glucose and ascorbic acid are important candidates instead of these toxic chemicals which enable the synthesis of silver nanoparticles at low nano sizes. In this study, the properties of tannic acid and ascorbic acid-synthesized silver nanoparticles were compared statistically. The thermal conductivity, absorbance @410-420 nm, zeta potential, and particle size distribution of the synthesized silver nanoparticles were determined. By proposing a systematic experimental design, quality criteria were characterized using Taguchi L_4 orthogonal arrays, and the results were evaluated with the help of main effect plots. Characterization of the synthesized nanoparticles was carried out with FTIR, Zetasizer, and Ultraviolet-Visible Spectroscopy (UV-Vis) devices. It has been shown that glucose/$AgNO_3$ and ascorbic/$AgNO_3$ molar ratios are the most important factors in the particle size distribution of AgNPs. The findings show that the lowest variance AgNPs were produced at the highest ascorbic/$AgNO_3$ molar ratios. As a result, it can be said that a better quality AgNPs production process is possible by adjusting the ascorbic acid/$AgNO_3$ molar ratio.

Keywords: Ascorbic acid, tannic acid, silver nanoparticles, statistics.

INTRODUCTION

Nanotechnology is a multidisciplinary branch of science that provides unique material production that has a wide range of usage in fields such as medicine, biology, and chemistry (Kakakhel et al., 2021). Nowadays, nanomaterials have become attractive for applications in biochemistry, the medicine industry, health monitoring, scientific diagnostics, water treatment, and ensuring food safety (Sharma et al., 2021).

Primary components of metal nanoparticles include gold, silver, copper, magnetic elements (cobalt, nickel), and semiconductors (Ramya et al., 2022). Among these, silver nanoparticles (AgNPs) come in front due to their low toxicity and ease of production (Reidy et al., 2013). Moreover, AgNPs exhibit extraordinary physicochemical properties such as conductivity, chemical stability, catalytic and antibacterial activity (Rudrappa et al., 2022). Owing to the mentioned features, AgNPs are used in a wide variety of fields such as food (additives, food packaging, refrigerator liners), textiles (clothing and bedding), cosmetics and hygiene products (toothbrushes, hair straighteners, disinfectant sprays, etc.), optics, electronics, magnetic, catalysis, and medicine (He et al., 2019).

Dubas and Pimpan synthesized silver nanoparticles by exposing silver nitrate solution to a low-power UV source in the presence of poly (methacrylic acid) (PMA), which acts as both a reducing and capping agent and used it as a colorimetric analysis to detect ammonia applications in water (Dubas and Pimpan 2008). Karhan et al. (2017) first applied the response surface methodology-based desirability function approach to obtain silver nanoparticles of the desired size using the green synthesis method. Using the proposed methodology, they achieved a 7.9% improvement in the average particle size distribution in the AgNP production process and showed that an 89.3% lower standard deviation was achieved compared to studies in the literature. Ponsanmti et al. (2020) examined the synthesis of silver nanoparticles by the green method using corn starch,

cassava starch, and sago starch. They found that the size of flower-shaped silver nanoparticles obtained from corn starch had a smaller particle distribution of 47.8 nm than cassava starch (107.5 nm) and sago starch (118.9 nm). Zamarchi et al. (2021) obtained pine nut extract and used it as a reductant and stabilizer in the synthesis of silver nanoparticles. To optimize the synthesis of silver nanoparticles, they investigated factors such as temperature, extract: water ratio, silver nitrate concentration and extract stability time. Kanniah et al. (2021) used the seed extract of *P. nigrum* as a reducing agent for the synthesis of silver nanoparticles. Silver nanoparticles exhibited effective antibacterial activity against *Bacillus subtilis* and *Escherichia coli*, while silver nanoparticles also showed effective antibacterial activity against compromised human cancer cells and cytotoxicity. Mahendran et al. (2022) biogenically synthesized silver nanoparticles using *Cissus quadarangularis* aqueous leaf extract and examined their antimicrobial activities. They found the average crystallite size of AgNPs as 35 nm and confirmed that it has unique antibacterial activity.

In light of the above mentioned information, tannic acid and ascorbic acid were selected as green reducing agents, and starch was used as the capping agent for environmental friendliness. The effects of solid/liquid ratio (reducing agent/$AgNO_3$ molar ratio), reducing agent type, and pH on the thermal conductivity, absorbance @410-420 cm^{-1}, zeta potential, and particle size properties of the synthesized AgNPs were examined. Experiments were performed using the Taguchi L_4 orthogonal array and the results were evaluated with main effect graphs.

MATERIALS AND METHODS

Materials

Silver nitrate ($AgNO_3$, >99.5%) and soluble starch from potatoes used in the experiments were purchased from Sigma Aldrich. D-(+)-Glucose (anhydrous) and sodium hydroxide pellets (NaOH, >99%) were from Merck Millipore, tannic acid was from Aromel Chemistry Medical, L (+) ascorbic acid was obtained from Carlo Erba company.

Synthesis of Silver Nanoparticles

In a 2000 mL beaker, 50 mL of 0.2% starch solution was taken and stirred for 10 minutes on a magnetic stirrer at 65ºC. Then, depending on the reducing agent/$AgNO_3$ mole ratio (S/L), a defined amount of 0.001 M $AgNO_3$ solution and reducing agent (RA), (0.1 M glucose, 0.1 M tannic acid, or 0.001 M ascorbic acid) solution was added. Then, the pH of the mixture was adjusted as specified in the experimental design using 0.002 M NaOH solution, and the color change was observed. Thus, glucose, tannic acid, and Ascorbic acid reduced silver ions (Ag^+) to metallic silver (Ag^0) in a sodium hydroxide alkaline solution (Karhan et al., 2017; Waberi, 2023). This reduction mechanism is given in equation (1) below, and the experimental system and the synthesized AgNPs are given in Figure 1.

$$2AgNO_3 + C_6H_{12}O_6 + 2NaOH \longrightarrow 2Ag^0 + C_6H_{12}O_7 + 2NaNO_3 + 2H_2O \quad (1)$$

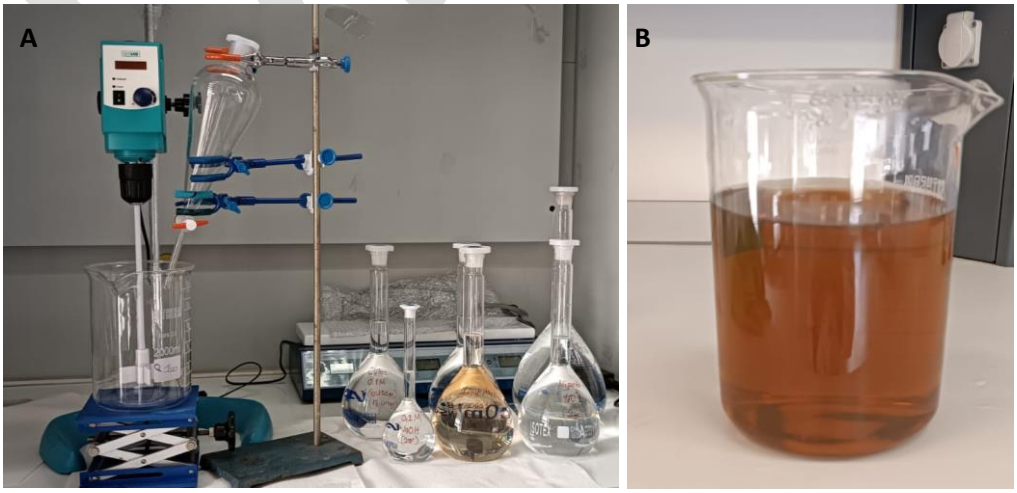


Figure 1. Synthesis of AgNPs a) experimental setup, b) synthesized AgNPs

DRAFT

Method

The Taguchi method uses orthogonal arrays to evaluate all possible combinations of parameters, thus reducing the number of experiments that are possible to solve a problem (Hasanzadeh et al., 2022). The selection of the appropriate orthogonal array is determined by the total degrees of freedom. The degrees of freedom of each factor are calculated by subtracting one from the number of levels of the factors (Şimşek, 2014). In this case, the total degree of freedom is 4 and therefore the L_4 orthogonal index was selected. By using the Taguchi method, the factor levels at which the best performance criterion value was obtained could be found, without performing all combinations. The results obtained were analysed with the help of the MINITAB software and main effect graphs were drawn. The symbols and levels of the factors determined for AgNPs synthesis are shown in Table 1.

Table 1. Factors and levels for AgNPs synthesis

Exp. No	Symbol	A (solid to liquid ratio)	B (Reducing agents)	C (pH)
0	REF	3.5	Glucose	9
1	15AA9	1.5	Ascorbic acid	9
2	15TA95	1.5	Tannic acid	9.5
3	35AA95	3.5	Ascorbic acid	9.5
4	35TA9	3.5	Tannic acid	9

Characterization of Silver Nanoparticles

The chemical structure of the AgNPs was investigated using FTIR spectroscopy. Bruker Tensor II device with a wavelength range of 400 to 4000 cm^{-1} was used to perform FTIR analysis. A PerkinElmer UV-visible spectrophotometer has been used to characterize AgNPs. The colloidal suspension of the AgNPs has been characterized by the UV-visible spectrophotometer within the range of 300−600 nm. The absorption peak is observed around 400 nm in the UV-visible spectrophotometric graphs, which have been confirmed in the presence of the silver nanoparticles for all experimental runs.

RESULTS and DISCUSSION

FTIR Characterization

FTIR spectra of the synthesized AgNPs are given in Figure 2. The apparent peak at 3401 cm^{-1} is related to O-H bond stretching vibrations due to ascorbic acid and tannic acid reduction reaction. The peak at 1720 cm^{-1} is attributed to the C=O bond stretching vibrations present in functionalities of ascorbic acid (ketones and aldehydes) and the carbonyl bond stretching vibration (C=O) in the carboxylic acid groups (COOH) of tannic acid. An absorption peak around 600 cm^{-1} is also observed, attributed to the stretching vibrations of carbon dioxide (CO_2).

DRAFT

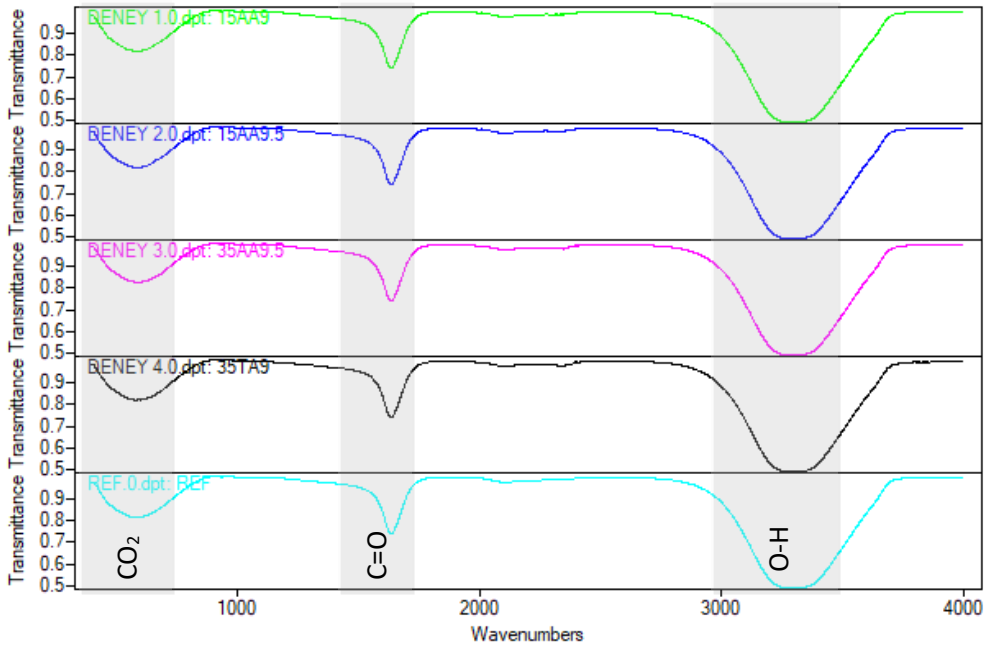


Figure 2. FTIR spectra of AgNPs

Experimental Results

Experimental results obtained for thermal conductivity, absorbance at the range of 410-420 nm, zeta potential, and particle size are given in Table 2, and the main effect graphs obtained with the help of the MINITAB program are given in Figure 3.

Table 1. Experimental results

Symbol	TC (W/m*K)	Absorbance @410-420 nm	Zeta potential (mV)	Particle size (nm)
REF	0.707±0.0389	4000	29.66±0.550	75.2±1.369
15AA9	0.771±0.0193	500	16.3±0.818	199.5±3.292
15TA95	0.648±0.0086	3900	19.3±2.551	92.2±5.830
35AA95	0.758±0.0394	1200	27.3±1.153	143.5±0.416
35TA9	0.767±0.0766	4000	26.1±1.877	108.3±1.021

When the main effect graphs were examined, it was seen that as the solid/liquid ratio increased, the thermal conductivity and particle size of AgNPs decreased, while the absorbance and zeta potential increased. Since AgNPs have a higher thermal conductivity, the presence of solid silver nanoparticles in solution can increase the overall thermal conductivity of the system. While AgNPs synthesized with ascorbic acid have the highest thermal conductivity, those synthesized with glucose and tannic acid have low thermal conductivity. Reducing the pH from 9 to 9.5 decreased the thermal conductivity of dispersed AgNPs.

While glucose-reduced AgNPs showed the highest absorbance value, increasing the pH to 9.5 decreased the absorbance value. The highest zeta potential of 30 mV was obtained by glucose-reduced AgNPs, while the lowest zeta potential of 22 mV was obtained with the use of ascorbic acid. Increasing the pH in the solution slightly reduced the zeta potential. While the highest particle size was obtained by ascorbic acid-reduced AgNPs, the particle size of those synthesized with glucose and tannic acid remains low. The concentration of silver ions affects the reduction kinetics and subsequent growth of nanoparticles. Higher concentrations of reducing agents lead to smaller particle sizes. However, above at a level of certain concentrations, particle size may begin to increase due to the agglomeration phenomenon. A slight decrease in particle size was observed

with an increase in pH value. An increase or decrease in pH can affect the surface charge of nanoparticles, affecting their stability and interaction with the surrounding environment.

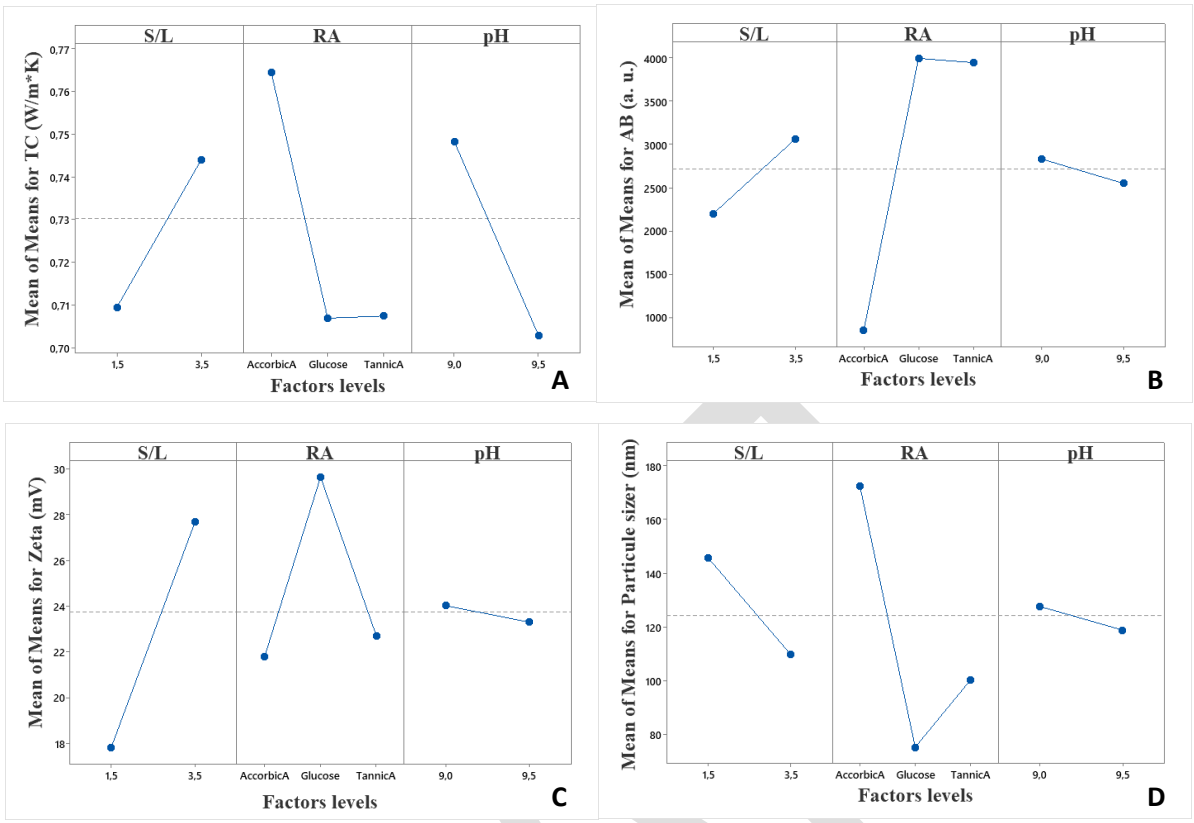


Figure 3. Main effect plots for a) thermal conductivity, b) absorbance @410-420 nm, c) zeta potential, and d) particle size of synthesized AgNPs

CONCLUSION

In this study, tannic acid and ascorbic acid-reduced AgNPs were prepared using Taguchi orthogonal arrays and compared with the glucose-reduced AgNPs using main effect plots. It was concluded that while the thermal conductivity of AgNPs decreases with increasing solid/liquid ratio, AgNPs obtained with ascorbic acid have the highest thermal conductivity. Moreover, the highest absorbance value at a range of 410-420 nm as an indicator of productivity was obtained by tannic acid at the ratio of 3.5 solid/liquid, and the level of pH=7. Furthermore; with the increase of the solid/liquid ratio, the zeta potential increased from 18 mV to 28 mV and the use of glucose showed the highest zeta-potential value for AgNPs. The use of ascorbic acid resulted in a high particle size for AgNPs.

REFERENCES

Dubas ST, Pimpan V 2008. Green synthesis of silver nanoparticles for ammonia sensing. Talanta, 76(1): 29-33.

Femi-Adepoju AG, Dada AO, Otun KO, Adepoju AO, Fatoba OP 2019. Green synthesis of silver nanoparticles using terrestrial fern (Gleichenia Pectinata (Willd.) C. Presl.): characterization and antimicrobial studies. Heliyon, 5(4): e01543.

Hasanzadeh R, Mojaver P, Chitsaz A, Mojaver M, Jalili M, Rosen MA 2022. Biomass and low-density polyethylene waste composites gasification: Orthogonal array design of Taguchi technique for analysis and optimization. International Journal of Hydrogen Energy, 47(67): 28819-28832.

He X, Deng H, Hwang H-m 2019. The current application of nanotechnology in food and agriculture. Journal of Food and Drug Analysis, 27(1): 1-21.

Kakakhel MA, Sajjad W, Wu F, Bibi N, Shah K, Yali Z, Wang W 2021. Green synthesis of silver nanoparticles and their shortcomings, animal blood a potential source for silver nanoparticles: A review. Journal of Hazardous Materials Advances, 1: 100005.

Kanniah P, Chelliah P, Thangapandi JR, Gnanadhas G, Mahendran V, Robert M 2021. Green synthesis of antibacterial and cytotoxic silver nanoparticles by Piper nigrum seed extract and development of antibacterial silver based chitosan nanocomposite. International Journal of Biological Macromolecules, 189: 18-33.

Karhan Ö, Ceran ÖB, Şara ON, Şimşek B 2017. Response Surface Methodology Based Desirability Function Approach To Investigate Optimal Mixture Ratio of Silver Nanoparticles Synthesis Process. Industrial & Engineering Chemistry Research, 56(28): 8180-8189.

Mahendran N, Anand B, Rajarajan M, Muthuvel A, Mohana V 2022. Green synthesis, characterization and antimicrobial activities of silver nanoparticles using Cissus quadarangularis leaf extract. Materials Today: Proceedings, 49: 2620-2623.

Ponsanti K, Tangnorawich B, Ngernyuang N, Pechyen C 2020. A flower shape-green synthesis and characterization of silver nanoparticles (AgNPs) with different starch as a reducing agent. Journal of Materials Research and Technology, 9(5): 11003-11012.

Ramya M, Senthil Kumar P, Rangasamy G, Uma shankar V, Rajesh G, Nirmala K, Saravanan A, Krishnapandi A 2022. A recent advancement on the applications of nanomaterials in electrochemical sensors and biosensors. Chemosphere, 308: 136416.

Reidy B, Haase A, Luch A, Dawson KA, Lynch I 2013. Mechanisms of Silver Nanoparticle Release, Transformation and Toxicity: A Critical Review of Current Knowledge and Recommendations for Future Studies and Applications. Materials (Basel), 6(6): 2295-2350.

Rudrappa M, Rudayni HA, Assiri RA, Bepari A, Basavarajappa DS, Nagaraja SK, Chakraborty B, Swamy PS, Agadi SN, Niazi SK, Nayaka S 2022. Plumeria alba-Mediated Green Synthesis of Silver Nanoparticles Exhibits Antimicrobial Effect and Anti-Oncogenic Activity against Glioblastoma U118 MG Cancer Cell Line. Nanomaterials, 12(3): 493.

Sharma S, Singh N, Ankalgi AD, Rana A, Ashawat MS 2021. Modern trends in analytical techniques for method development and validation of pharmaceuticals: A review. Journal of Drug Delivery and Therapeutics, 11(1-s): 121-130.

Some S, Bulut O, Biswas K, Kumar A, Roy A, Sen IK, Mandal A, Franco OL, Ince IA, Neog K, Das S, Pradhan S, Dutta S, Bhattacharjya D, Saha S, Mohapatra KD, Bhuimali A, Unni BG, Kati A, Mandal AK, Yilmaz MD, Ocsoy I 2019. Effect of feed supplementation with biosynthesized silver nanoparticles using leaf extract of Morus indica L. V1 on Bombyx mori L. (Lepidoptera: Bombycidae). Scientific Reports, 9(1): 14839.

Şimşek B 2014. A multi-response modeling and optimization application for determining the optimal mixing ratios of ready-mixed concrete: Response Surface Method with TOPSIS-based Taguchi Approach. Doctoral thesis, Ankara University, 207 pages, Ankara.

Zamarchi F, Vieira IC 2021. Determination of paracetamol using a sensor based on green synthesis of silver nanoparticles in plant extract. Journal of Pharmaceutical and Biomedical Analysis, 196: 113912.

Waberi AD 2023. Statistical Comparison of Silver Nanoparticle Properties Synthesized with Tannic Acid and Ascorbic Acid. Master thesis, Çankırı Karatekin University, 60 pages, Çankırı.

DRAFT

ORAL PRESENTATION

An evaluation to examine the change in average temperature and precipitation in the Mersin region

İlyas Bolat[1*] (ORCID: *https://orcid.org/0000-0002-5354-2968*), Akın Kamış[2] (ORCID: *https://orcid.org/0009-0002-5339-1706*), Hüseyin Şensoy[3] (ORCID: *https://orcid.org/0000–0001–6453–5723*), Kamil Çakıroğlu[4] (ORCID: *https://orcid.org/0000-0001-9057-1576*)

[*1]Bartın University, Bartın Faculty of Forestry, Department of Forest Engineering, Division of Soil Science and Ecology, Bartın, 74100, Türkiye

[2]Mersin Provincial Directorate of Environment, Urbanization and Climate Change, Natural Assets Protection Branch, New District 5307 St. No:8/A Akdeniz–Mersin, Türkiye

[3]Bartın University, Bartın Faculty of Forestry, Department of Forestry Engineering, Division of Watershed Management, Bartın, 74100, Türkiye

[4]Bartın University, Bartın Faculty of Forestry, Department of Forest Engineering, Division of Soil Science and Ecology, Bartın, 74100, Türkiye

*Corresponding author e-mail: bolat.ilyas@hotmail.com, ilyasbolat@bartin.edu.tr

Abstract

Climate change caused by global warming is seen as extreme changes, especially in temperature and precipitation. Data obtained from studies conducted by researchers and organizations in different parts of the world support that anthropogenic climate change is occurring. This study was carried out in Mersin, located in the Eastern Mediterranean part of the Mediterranean Region, in order to determine the changes in annual average temperature and total annual precipitation amounts on a regional scale in the 43–year period (1980–2022). Between 1980 and 2022, the annual average temperature in Anamur, Mersin, and Silifke varies between 17.90–21.30°C, 18.00–21.50°C, and 17.80–21.60°C, respectively. It was determined that the lowest and highest average temperatures in this period were in Silifke (17.80°C and 21.60°C, respectively). Simple linear regression model (SLRM) results revealed that the annual average temperature (Anamur; r = 0.8448, Mersin; r = 0.8865 and Silifke; r = 0.8329) had an increasing trend. However, according to the results of simple variance analysis, there is no statistical difference (P>0.05) between the average temperature values. During the 43–year period, the total annual precipitation in Anamur varied between 447.10 and 1755.10 mm, in Mersin between 301.60 and 1033.70 mm, and in Silifke between 300.50 and 1007.70 mm. During the study period, the lowest annual total precipitation was obtained in Silifke and the highest annual total precipitation was obtained in Anamur. In the regression analysis model made to determine the change between 1980 and 2022, the average total precipitation increased in Mersin (r = 0.0979) and decreased in Anamur (r = –0.05) and Silifke (r = –0.0436) appears to be trending. Simple variance analysis indicates that there is a statistical difference (P<0.05) between the annual total precipitation values of Anamur, Mersin, and Silifke. The results of the study reveal that temperatures tend to increase on a regional scale, as a sign of global warming, and that there are changes in the precipitation regime and distribution. If these trends continue, it is predicted that more severe floods or drought events may occur in this region and its surroundings.

Keywords: Anamur, global warming, regression model, temperature, Silifke, precipitation

INTRODUCTION

Temperature and precipitation, the main elements of climate, are of great importance in determining climate change trends in a region. Both climate elements can show large changes on both spatial and temporal scales. In particular, temporal changes in precipitation and temperature not only reveal spatial differences but also create oscillations in climate (Bahadır, 2011). Recent analyses of climate trends show that the global average surface temperature has increased by approximately 0.3 to 0.6°C since the late 19th century and by approximately 0.2 to 0.38°C in the last 40 years. It is also stated that daily minimum temperatures generally increase at a higher rate than maximum temperatures, causing a decrease in the daily temperature range in various parts of the world. While there was a slight increase in global precipitation by about 1% on land in the 20th century, there was a greater increase in the high latitudes of the Northern Hemisphere, especially in the cold season. Studies indicate that significant spatial and temporal changes have occurred in the past 100 years.

However, it is reported that the trends in increasing temperature (warming), precipitation, and decrease in daily temperature range are not the same globally (Nicholls et al., 1996; Zhang et al., 2000). On the other hand, it is reported that there are significant differences (increases–decreases) in the precipitation regime for various regions of the world (Popova et al., 2018). In a study examining temperature and precipitation trends in Bologna (Italy) from 1952 to 1999, it is emphasized that although the average annual precipitation was 750 mm, there was a lot of variability between years with a standard deviation of 159 mm. It is stated that this variability in the amount of precipitation is not statistically significant, but with a decrease of approximately 120 mm throughout the entire period, the region has evolved into a drier climate (Ventura et al., 2002).

Data from the Intergovernmental Panel on Climate Change (IPCC, 2001) report an increase in temperature of 0.6±0.2°C during the 20th century. It is noted that most of the warming has occurred in the last 20 years, with the nineties being the warmest decade since 1861. In the same report, it is predicted that the global average surface temperature will increase between 1.4–5.8°C in the period 1990–2100 (Apak and Ubay, 2007). However, according to the IPCC's latest report (6th Assessment Report – AR6) published on August 9, 2021, global temperature is increasing much faster than expected. Additionally, the report states that the temperature increase between 2010 and 2019 was more than 1°C (1.07°C) compared to the period 1850–1900 (IPCC, 2021).

Like the results obtained worldwide, a study conducted in our country reports that there is a significant change in the amount of precipitation and that this change is seen in both winter and autumn seasons. It is stated that winter precipitation in the western provinces of Turkey has decreased significantly in the last fifty years, while autumn precipitation has increased mostly in stations located in the northern parts of Central Anatolia. In general, precipitation decreases on the Aegean and Mediterranean coasts of Turkey, whereas it increases on the Black Sea coasts (Apak and Ubay, 2007). Another study reveals that there is a partial temperature increase in the Black Sea climate region (0.3°C), but the increase in temperature manifests itself excessively (0.5–0.6°C) in Continental and Mediterranean climate conditions. These data coincide with the predicted surface temperature increases according to climate regions around the world (Bahadır, 2011). Apart from these studies, according to the IPCC 3rd evaluation report published in 2001, it is predicted that annual average temperatures in Turkey will increase by 1–3°C by 2050. It is estimated that precipitation will show a decreasing trend in the Mediterranean climate area (IPCC, 2001). Global climate changes, which increased especially after the 1950s and became a problem for the whole world in the 1990s, have been the subject of many studies, directly or indirectly, within the framework of the developments explained above (Gao and Giorgi, 2008; Evans, 2009; Bolat et al., 2016; Öztürk et al., 2016; Bolat et al., 2017; Bolat et al., 2018, Şensoy and Ateşoğlu, 2018). In this study, data between 1980 and 2022 (43–year period) from some stations (Anamur, Mersin–Central, and Silifke), from which regular data has been received for many years in Mersin province, were used. By examining monthly–annual average temperatures and monthly–annual total precipitation amounts, it was aimed to determine the changes in these parameters on a regional scale.

MATERIALS AND METHODS

Location of the Study Area

Mersin, one of the 81 provinces of Turkey, is located between 36–37° northern latitudes and 33–35° eastern longitudes. Mersin province, formerly known as İçel, is surrounded by Adana in the east, Antalya in the west, Niğde, Konya, and Karaman provinces in the north, and the Mediterranean Sea in the south. Due to its geographical location, Mersin is located in the west of the Çukurova section of the Mediterranean Region and covers a significant part of the Eastern Mediterranean Basin. The surface area of the province is 15,853 km² (Eren and Sezgin, 2017; URL–1, 2023).

Study Data

In this study, periodic data of Anamur, Mersin (Central) and Silifke stations belonging to the Mersin Meteorology Station Directorate affiliated with the General Directorate of Meteorology for the years 1980–2022 (43 years) were used. Monthly and annual average temperature values, monthly average total precipitation, and annual total precipitation data of three stations with synoptic or automatic observation between 1980 and 2022 were evaluated. Annual averages are arranged as the arithmetic average of twelve months in the calendar year. Some information about the stations and their locations in Türkiye are given in Figure 1. The reason why this climate data is preferred is that changes in temperature and precipitation are the most important indicators of global climate change. While changes in temperature and precipitation affect the

distribution of vegetation around the world, these changes also cause more extreme weather and climate events (Çepel, 2003; Türkeş, 2010; Liu et al., 2017).

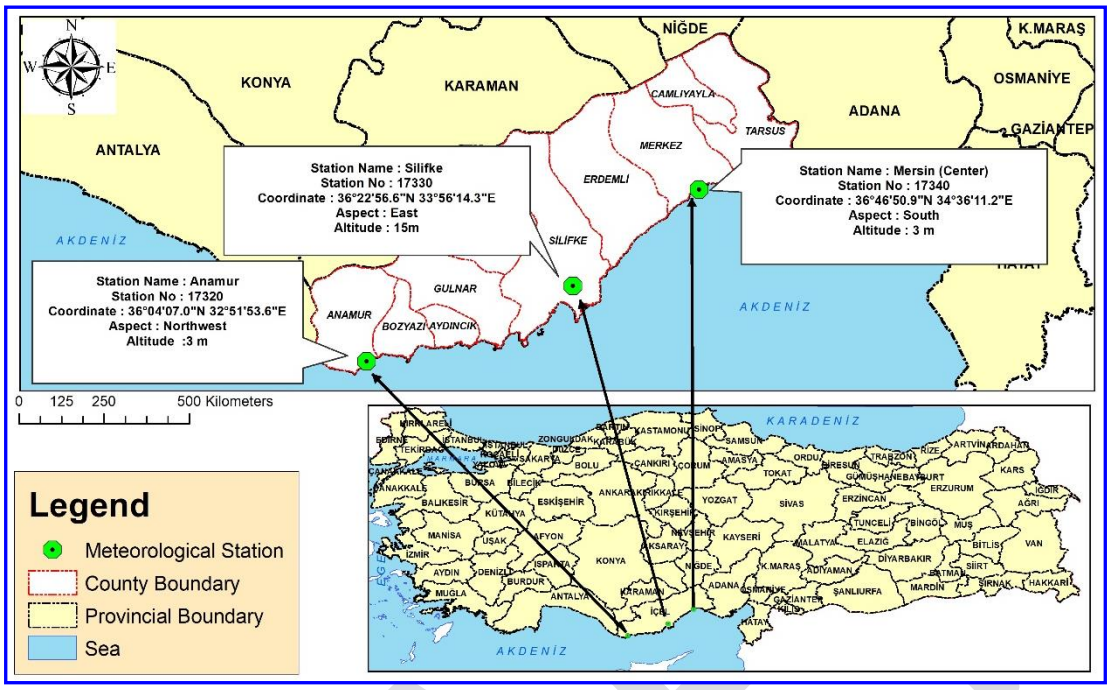


Figure 1. Mersin province and location of meteorological stations

Evaluation of Data and Statistical Analysis

Simple linear regression model (SLRM) was used to evaluate the average temperature and precipitation data used in the study. Whether there is a statistical difference ($P<0.05$) between the average temperature and total precipitation values of the meteorological stations was revealed by one–way analysis of variance (One–way ANOVA). S–N–K (Student–Newman–Keuls) test was applied to determine the different group or groups.

RESULTS AND DISCUSSION

Annual Average Temperature

The lowest average temperature in Mersin province Anamur meteorology station was 1992 (17.9°C) and the highest was 2018 (21.3°C) (Figure 2). According to SLRM (Figure 2), which shows the change over the years, the annual average temperature in Anamur tends to increase. The average temperature in Anamur between 1980 and 2022 is 19.6°C (Figure 5). According to this result, it is lower than the average temperature of Anamur compared to Mersin and Silifke. However, there is no statistical difference ($P>0.05$) between the average temperature values of Anamur, Mersin and Silifke (Figure 5).

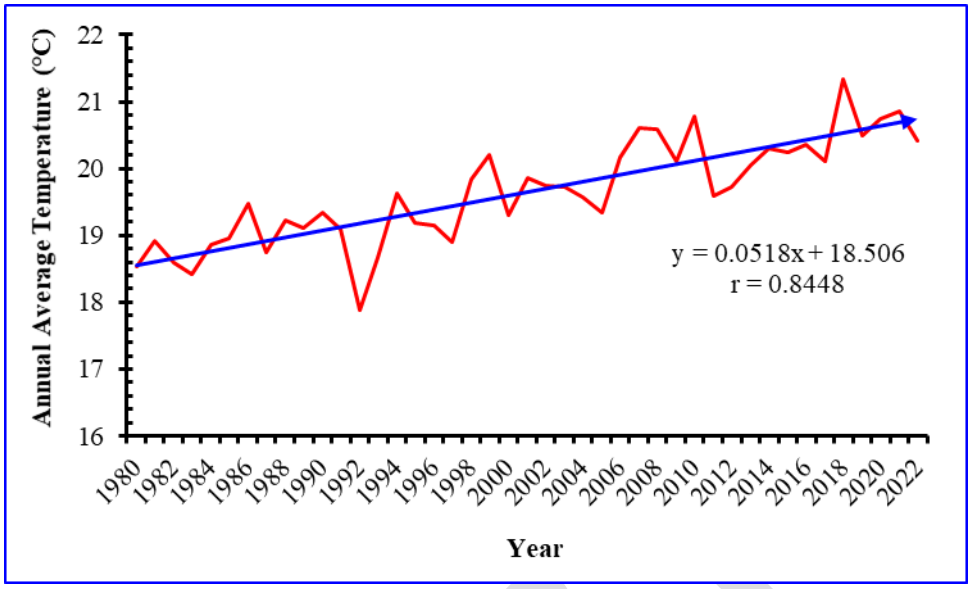


Figure 2. Change in monthly average temperature values of Anamur meteorological station for long terms (1980–2022)

The lowest annual average temperature at the Mersin meteorology station in Mersin province was 1992 with a value of 18.0°C, and the year with the highest was 2018 with a value of 21.5°C. (Figure 3). SLRM (Figure 3) reveals that the annual average temperature in Mersin has an increasing trend (r = 0.8865). The average temperature in Mersin between 1980 and 2022 is 19.9°C (Figure 5). This result shows that the average temperature of Mersin is higher than Anamur and Silifke. However, there is no statistical difference (P>0.05) between the average temperature values of Anamur, Mersin and Silifke (Figure 5).

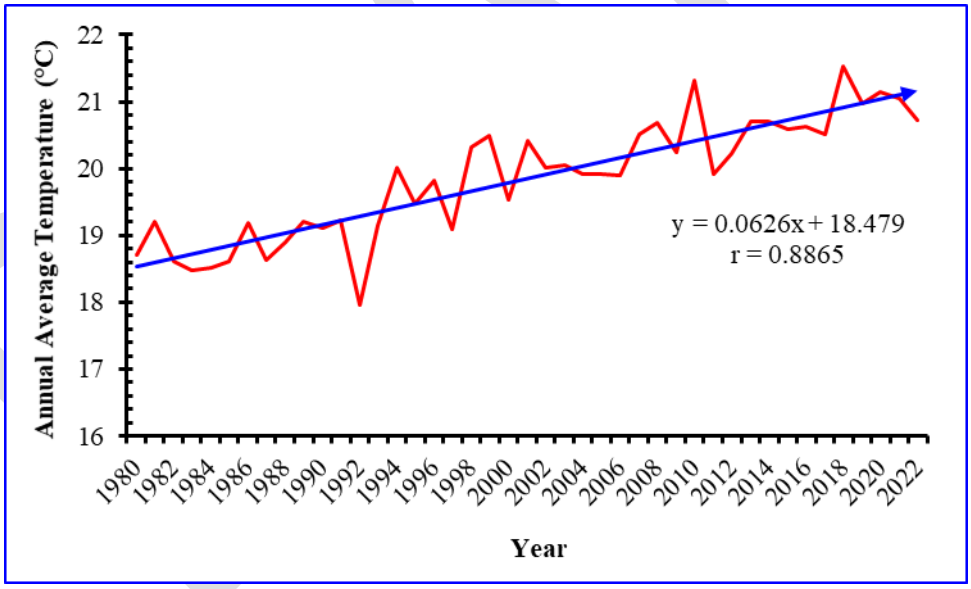


Figure 3. Change in monthly average temperature values of Mersin meteorological station for long terms (1980–2022)

The average annual temperature at Silifke meteorological station was the lowest in 1992 with 17.8°C, and the highest in 2018 with 21.6°C. According to SLRM (Figure 4), it was revealed that the annual average temperature in Silifke has an increasing trend (r = 0.8329). The average temperature in Mersin between 1980 and 2022 is 19.8°C (Figure 5). According to this result, the average temperature of Silifke is higher than Anamur and lower than Mersin. However, there is no statistical difference (P>0.05) between the average temperature values (Figure 5).

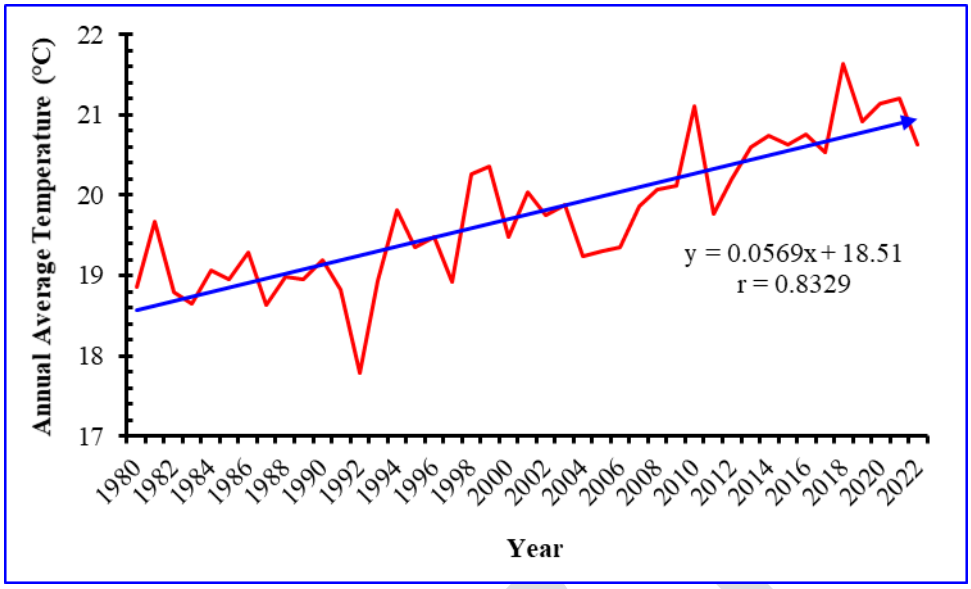


Figure 4. Change in monthly average temperature values of Silifke meteorological station for long terms (1980–2022)

As a result of the analysis of variance conducted to reveal whether there is a difference between the annual average temperature values of Anamur, Mersin, and Silifke, it was revealed that there is no difference (P>0.05). Between 1980 and 2022, Mersin has the highest (19.9°C) and Anamur the lowest (19.6°C) annual average temperature value (Figure 5).

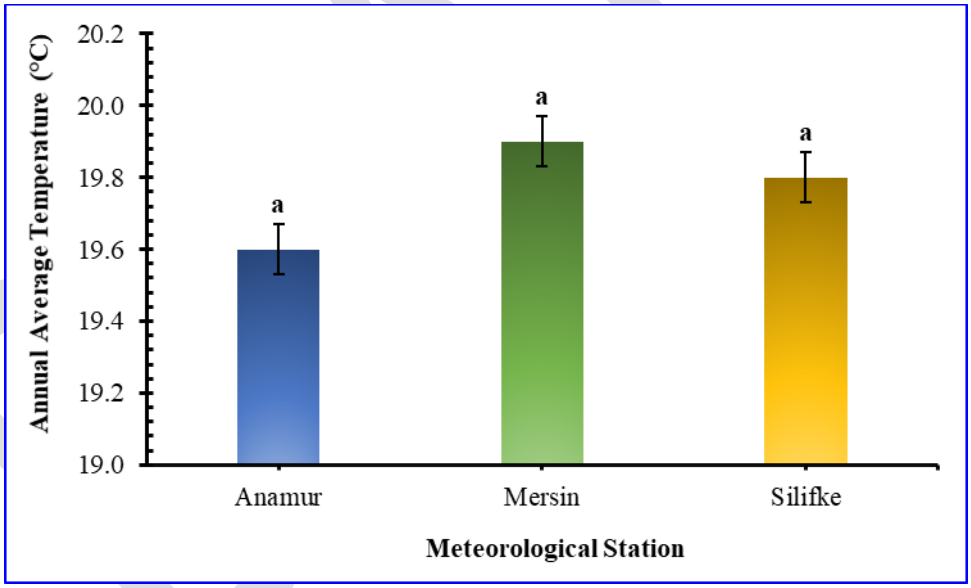


Figure 5. Average temperature values of Anamur, Mersin and Silifke meteorological stations between 1980 and 2022. Columns represent mean ± standard error. The same letters indicate that there is no difference between average temperatures (P<0.05)

The average temperature in Türkiye between 2010 and 2020 is 14.1°C. According to long–term data, it is reported that the average temperature in Turkey between 1970 and 2020 was 13.2°C (URL–2, 2022). Between 1971 and 2021, the highest annual average temperature in Turkey was 15.5°C in 2010, and the lowest annual average temperature was 11.7°C in 1992. There have been positive temperature anomalies in Turkey's average temperatures since 1994 (except for 1997 and 2011). This result means that the annual average temperature in Turkey has an increasing trend (URL–3, 2023). The average temperature values of Anamur, Mersin, and Silifke, which are the subject of this study, are above the general average of Turkey. Like the increasing trend of the annual average temperature in Turkey, it was determined that the annual average temperature of Anamur, Mersin, and Silifke also tended to increase between 1980 and 2022 (r = 0.8448; r = 0.8865; r = 0.8329, respectively). According to a recent study, it has been reported that Turkey's summer and autumn temperatures

have increasing trends and this warming is getting stronger. It is stated that this warming occurred relatively strongly in other regions except the Black Sea region (Türkeş, 2019). Based on the average temperature data from the study conducted at 27 stations in Turkey, it is reported that there are significant warming trends in the regions located in the south and southwest of Turkey, where a large amount of urbanization is experienced. However, it is emphasized that minimum temperatures show a general increasing trend and annual maximum temperature series tend to increase in the Mediterranean, Southeastern Anatolia, and the southern parts of Eastern Anatolia. Average temperatures in Turkey tend to increase, like global average surface temperatures. The rapid temperature increase, which has been continuing on a global scale since the 1980s, has been observed in Turkey since the 1990s (Demir et al., 2008). In a study conducted in the Western Black Sea region of Turkey, it was determined that the annual average temperature in Bartın, Zonguldak, and Düzce (Bartın; r = 0.568, Zonguldak; r = 0.653, Düzce; r = 0.584) had an increasing trend (Bolat and Şensoy, 2023a). A similar result is seen in the study conducted in Sinop and Kastamonu regions. According to the SLRM result in the study, it is stated that the annual average temperature in Sinop (r = 0.498) and Kastamonu (r = 0.225) has an increasing trend (Bolat and Şensoy, 2023b). Results similar to those showing that the temperature is on an increasing trend in Turkey are also seen on a world scale. One of the many studies was conducted in Southern Canada with data covering the years 1900–1998. The results report increasing trends in annual and seasonal daily average temperatures. It is stated that there was a statistically significant positive trend corresponding to an increase of 0.98 °C in the region, especially in the period 1961–1990 (Zhang et al., 2000). The results obtained in this study are compatible with the results of previous studies that draw attention to warming.

Total Annual Precipitation

The change in annual total precipitation (P_{TAn}) amount in Anamur, Mersin province is shown in Figure 6. P_{TA} was a minimum (447.10 mm) in 1990 and a maximum (1755.10 mm) in 2012. The average total precipitation (P_{TAv}) in Anamur between 1980 and 2022 was recorded as 904.0 mm. This result shows that Anamur receives more P_{TAn} compared to Mersin and Silifke (Figure 9). There is also a statistical difference ($P<0.05$) between P_{TAn} values. On the other hand, according to the SLRM created with the P_{TAn} amount distribution (Figure 6), it was revealed that the amount of precipitation in Anamur was in a decreasing trend (r = – 0.05).

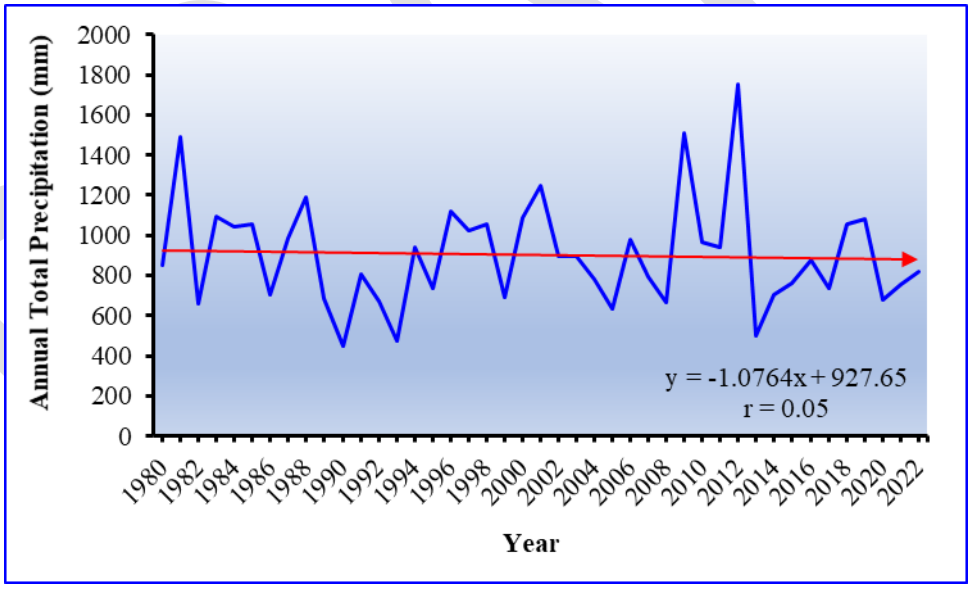


Figure 6. Change in annual total precipitation values of Anamur meteorological station between 1980 and 2022

The change in the annual total precipitation (P_{TAn}) amount in Mersin is shown in Figure 7. P_{TAn} was minimum (301.60 mm) in 1982 and maximum (1033.70 mm) in 2012. During the period 1980–2022, the average total precipitation (P_{TAv}) was recorded as 609.70 mm. This result reveals that Mersin receives less total annual precipitation than Anamur but more than Silifke (Figure 9). According to the SLRM created with P_{TAn} data (Figure 7), it was revealed that the amount of precipitation in Mersin tended to increase (r = 0.0979).

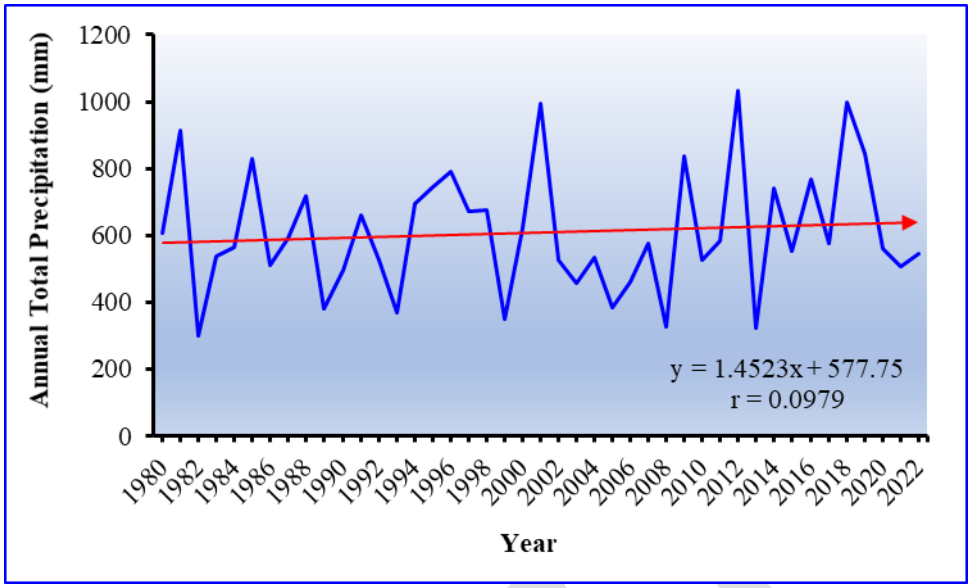


Figure 7. Change in annual total precipitation values of Mersin meteorological station between 1980 and 2022

P_{TAv} was minimum (300.50 mm) in 1999 and maximum (1007.70 mm) in 2001, and the change in the amount of P_{TAv} in the period 1980–2022 is shown in Figure 8. In Silifke, P_{TAn} was recorded as 561.0 mm in this period. According to this result, Silifke has less total annual rainfall than Anamur and Mersin (Figure 9). According to SLRM (Figure 8), it was revealed that the amount of precipitation in Silifke was in a decreasing trend (r = – 0.0436).

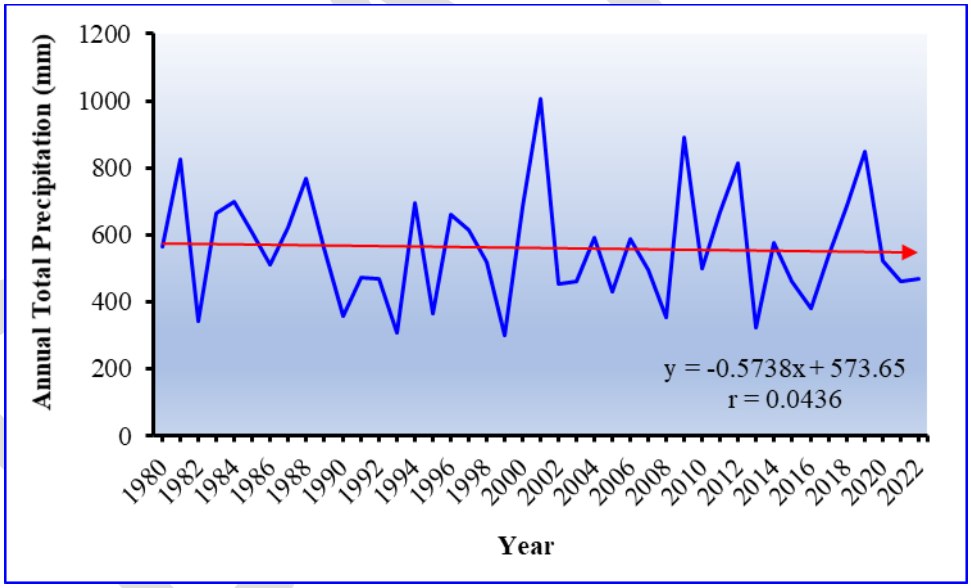


Figure 8. Change in annual total precipitation values of Silifke meteorological station between 1980 and 2022

As a result of the analysis of variance, it was revealed that there was a statistical difference ($P<0.05$) between the annual total precipitation of Anamur, Mersin and Silifke. According to the results of the S–N–K (Student–Newman–Keuls) test, Anamur was separated from Mersin and Silifke and placed in a different group, while Mersin and Silifke were included in the same group. Between 1980 and 2022, Anamur had the highest (904.0 mm) P_{TAv} value, while Silifke had the lowest (561.0 mm) P_{TAv} value (Figure 9).

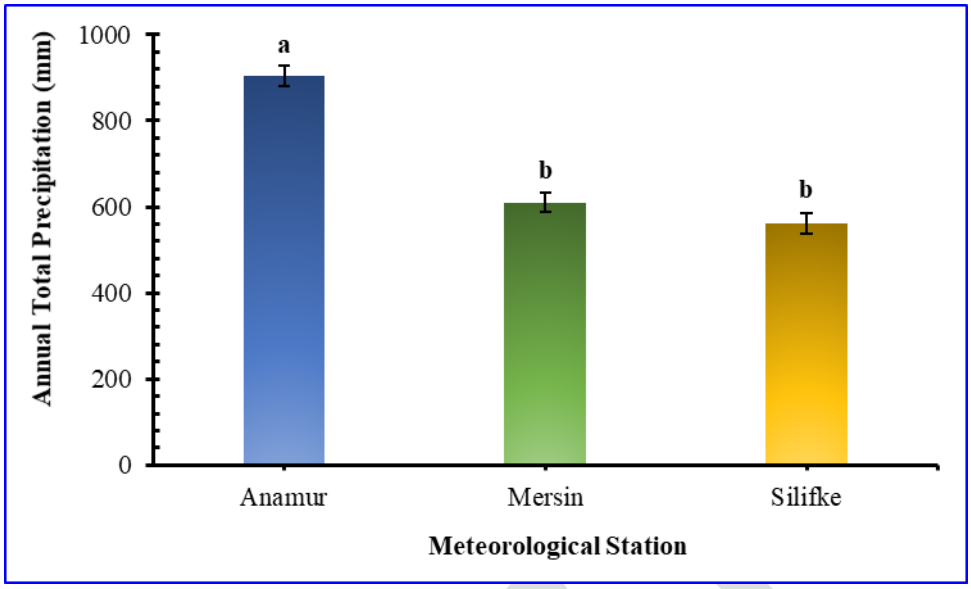


Figure 9. Total precipitation values of Anamur, Mersin and Silifke meteorological stations between 1980 and 2022. Columns represent mean ± standard error. Different letters indicate a difference between means at the P<0.05 significance level

It is reported that the average annual total precipitation in Turkey was 621.4 mm between 1970 and 2020, and 619.3 mm between 2010 and 2020 (URL–4, 2022). The average annual areal rainfall across Turkey between 1991 and 2020 is 573.4 mm. In terms of area, the average annual rainfall across Turkey between 1991 and 2020 is 573.4 mm. According to long–term averages, the highest precipitation in Turkey occurs above 1600 mm on the coasts of Rize and Artvin in the Eastern Black Sea Region, while the lowest precipitation is observed in the central parts of Central Anatolia and around Şanlıurfa and Iğdır. From 1930 to the present, 2008 is the year with the least precipitation (444.9 mm). In addition, precipitation in 2020 (500 mm) and 2021 (525 mm) was below normal. When evaluated spatially, the average precipitation in 2021 was 524.8 mm and there was a decrease of 8.5% compared to precipitation norms (URL–5, 2023). According to Turkey's long–term average rainfall values, Anamur and Mersin are above the average, while Silifke is below the average. In the regression analysis model made to determine the change in average total precipitation over the years, a relative increase in Mersin (r= 0.0979) and a decreasing trend in Anamur (r= – 0.05) and Silifke (r= –0.0436) were determined. Turkey's long–term trends and changes generally show that there is a significant decrease in winter and spring precipitation in the Marmara, Aegean, Mediterranean and Southeastern Anatolia regions, where the Mediterranean precipitation regime is dominant, and in the Central and Eastern Anatolia regions. In the cumulative rainfall calculated between 01 October 2013 and 17 January 2014 for Turkey in general, there was a decrease of 37% compared to the long–term average and 47.4% compared to 2013 (Türkeş, 2019). In a study conducted using data from 88 stations in Turkey for the period 1940–2006, although there was no significant trend in the annual standardized precipitation series, a slight increase was determined in the Black Sea and Continental Eastern Anatolia Regions and a slight decreasing trend in the Mediterranean, Mediterranean Transition, and continental Mediterranean regions (Demir et al., 2008). In a study conducted to determine the trend of average total precipitation by years, it was revealed that the average total precipitation tended to decrease in Bartın province (r = – 0.081) and increase in Zonguldak (r = 0.423) and Düzce (r = 0.306) provinces (Bolat and Şensoy, 2023a). On the other hand, in a study covering the years 1965–2015, no significant change trend was determined in monthly, seasonal, and annual precipitation in Bartın province (Yaman and Ertuğrul 2020). In a recent study in the Sinop and Kastamonu region, it was revealed that the average total precipitation in Sinop (r = – 0.094) and Kastamonu (r = – 0.276) provinces has a decreasing trend (Bolat and Şensoy, 2023b). Türkeş (2019) states that the variations in precipitation are not in the form of long–term trends but in the form of changes and fluctuations, but significant changes have been determined in the frequency and magnitude of dry–humid periods. It is noted that the spatial variation in precipitation variations is also strong. In terms of annual total precipitation trends, this study reveals similar results and is compatible with previous evaluations of precipitation trends and changes in Turkey.

CONCLUSIONS

The average temperature of Mersin is higher than Anamur and Silifke. However, there is no statistical difference between the average temperature values of Anamur, Mersin and Silifke. When the average temperature changes of Anamur, Mersin and Silifke are examined, it is seen that the temperatures tend to increase. Average total precipitation is higher in Anamur than in Mersin and Silifke. The trend of average total precipitation is relatively increasing in Mersin and decreasing in Anamur and Silifke. There is also a statistical difference between the annual total precipitation values of Anamur, Mersin and Silifke. As a result of all these, it can be said that annual average temperatures tend to increase, while total annual precipitation may become irregular, and the downward trend is more dominant. It is predicted that the Mersin region will evolve into a drier climate. Therefore, in the following years, undesirable meteorological events such as floods, drought, and increased irregular rainfall may occur frequently in the Mersin region and even in the entire southern parts of Turkey.

ACKNOWLEDGEMENTS

We would like to thank the Mersin Meteorology Station Directorate of the General Directorate of Meteorology of the Ministry of Environment, Urbanization, and Climate Change for the meteorological data used in the study.

REFERENCES

Apak G, Ubay, B 2007. First national communication of Turkey on climate change. Retrieved from: http://www.tr.undp.org/content/dam/turkey/docs/Publications/EnvSust/iklim1.bildirimi–27ocak2007.pdf

Bahadır M 2011. Türkiye'de iklim değişikliğinin iklim bölgelerine yansımasında kuzey–güney yönlü sıcaklık ve yağış değişim öngörüleri. Akademik Bakış Dergisi, 26: 1–18.

Bolat İ, Şensoy H 2023a. Analysis of some meteorological data and their variation trends in three provinces of the western black sea region between 2012 and 2021. Forestist, 73(3): 220–230.

Bolat İ, Şensoy H 2023b. Analysis and examination of change trends of some meteorological data between 2012–2021: The case study of Sinop and Kastamonu Provinces. Anatolian Journal of Forest Research, 9(2), DOI: https://doi.org/10.53516/ajfr.1308987

Bolat İ, Kara Ö, Tok E 2018. Global warming and climate change: a practical study on Bartın, Zonguldak and Düzce. Journal of Bartın Faculty of Forestry, 20(1): 116–127.

Bolat İ, Kara Ö, Tok E 2017. Kastamonu, Karabük ve Bolu'da 1980–1999 ile 2000–2015 yılları arasındaki sıcaklık ve yağışın değişimi. Bartın Orman Fakültesi Dergisi, 19(1): 276–289.

Bolat İ, Kara Ö, Öztürk M 2016. Short-term changes of leaf area index, light transmission, and gap in a temperate mixed deciduous forest ecosystem in Bartın, Turkey. Turkish Journal of Agriculture and Forestry, 40(4): 597–605.

Çepel N 2003. Ekolojik sorunlar ve çözümleri. TÜBİTAK, Popüler Bilim Kitapları, Aydoğdu Matbaası, 183 s., Ankara.

Demir İ, Kılıç G, Coşkun M, Sümer UM 2008. Türkiye'de maksimum, minimum ve ortalama hava sıcaklıkları ile yağış dizilerinde gözlenen değişiklikler ve eğilimler. TMMOB İklim Değişimi Sempozyumu, Bildiriler Kitabı, 69–84. TMMOB adına TMMOB Meteoroloji Mühendisleri Odası, 13–14 Mart, 2008, Ankara.

Eren AGFY, Sezgin AC 2017. Gastronomi turizmi açısından mersin yöresi mutfak kültürünün sürdürülebilirliği. 1st International Sustainable Tourism Congress / November 23–25, Proceedings Book, 161–170 pp, Kastamonu, Turkey.

Evans JP 2009. 21st century climate change in the Middle East. Climatic Change, 92: 417–432.

Gao X, Giorgi F 2008. Increased aridity in the Mediterranean region under greenhouse gas forcing estimated from high resolution simulations with regional climate model. Global and Planetary Change, 62: 195–209.

IPCC 2001. Intergovernmental panel on climate change, the scientific basis, contribution of working group ı to the third assessment report of the Intergovernmental Panel on Climate Change [Houghton, J.T., Y.

Ding, D.J. Griggs, M. Noguer, P.J. van der Linden, X. Dai, K. Maskell, and C.A. Johnson (eds.)], Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

IPCC 2021. Summary for policymakers, climate change 2021: the physical science basis, contribution of working group ı to the sixth assessment report of the Intergovernmental Panel on Climate Change, Valerie Masson–Delmotte vd., Cambridge University Press, 2021, s. 21.

Liu J, Du H, Wu Z, He HS, Wang L, Zong S 2017. Recent and future changes in the combination of annual temperature and precipitation throughout China. International Journal of Climatology, 37(2): 821–833.

Nicholls N, Gruza G, Jouzel J, Karl T, Ogallo L, Parker D 1996. Observed climate variability and change. In: Houghton JT, Meira Filho LG, Callander BA, Harris N, Kattenberg A, Maskell K, eds. Chapter 3, In: Climate Change 1995. The Science of Climate Change. Cambridge: Cambridge University Press, 132–192.

Öztürk M, Bolat İ, Gökyer E, Kara Ö 2016. Growth gradients of multi–aged pure oriental beech stands along the altitudinal gradients within a mesoscale watershed landscape. Applied Ecology and Environmental Research, 14(4): 101–119.

Popova EN, Popov IO, Semenov SM 2018. Assessment of variations in the annual sum of active temperatures and total precipitation during the vegetation period in Russia and neighboring countries. Russian Meteorology and Hydrology, 43: 412–417.

Şensoy H, Ateşoğlu A 2018. Bartın yöresinde iklim tipi değişikliğine yönelik bir değerlendirme. Bartın Orman Fakültesi Dergisi, 20(3): 576–582.

Türkeş M, 2010. Klimatoloji ve meteoroloji. 650 s., Kriter Yayınevi, İstanbul.

Türkeş M, 2019. Scientific basis of climate change and impacts on Turkey. Climate change training module series 1, the project co–funded by the European Union and the Republic of Turkey. http://www.iklimin.org/moduller/bilimmodulu.pdf [access: 24.12.2022].

URL–1 2023. https://mersin.ktb.gov.tr/#, Mersin İl Kültür ve Turizm Müdürlüğü web sayfası, (Erişim tarihi: 20.09.2023).

URL–2 2022. https://www.mgm.gov.tr/FILES/resmi–istatistikler/parametreAnalizi/Turkiye–Ortalama–Sicaklik–2020.pdf, Meteoroloji Genel Müdürlüğü web sayfası, (Erişim tarihi: 01.03.2022).

URL–3 2023. https://cevreselgostergeler.csb.gov.tr/sicaklik–i–85727#_edn1

URL–4 2022. https://www.mgm.gov.tr/FILES/resmi–istatistikler/parametreAnalizi/Turkiye–Yagis–2020.pdf, Meteoroloji Genel Müdürlüğü web sayfası, (Erişim tarihi: 01.03.2022).

URL–5 2023. https://cevreselgostergeler.csb.gov.tr/yagis–i–85728

Ventura F, Pisa PR, Ardizzoni E 2002. Temperature and precipitation trends in Bologna (Italy) from 1952 to 1999. Atmospheric Research, 61(3): 203–214.

Yaman B, Ertuğrul M 2020. Change–point detection and trend analysis in monthly, seasonal and annual air temperature and precipitation series in Bartın province in the western Black Sea region of Turkey. Geology, Geophysics and Environment, 46(3): 223–223.

Zhang X, Vincent LA, Hogg WD, Niitsoo A. 2000. Temperature and precipitation trends in Canada during the 20th century. Atmosphere–Ocean, 38(3): 395–429.

DRAFT

ORAL PRESENTATION

Thermal conductivity of sodium alginate cement pastes

Mücahit Uğur [1] (*ORCID: https://orcid.org/0000-0002-3746-5683*), Özge Bildi Ceran[1] (*ORCID: https://orcid.org/0000-0002-3147-735X*), Barış Şimşek[1*] (*ORCID: https://orcid.org/0000-0002-0655-4368)*, Tayfun Uygunoğlu[2] (*ORCID: https://orcid.org/0000-0003-4382-8257)*

[*1] Çankırı Karatekin University, Faculty of Engineering, Department of Chemical Engineering, Çankırı, Turkey.
[2] Afyon Kocatepe University, Faculty of Engineering, Department of Civil Engineering, Afyon, Turkey.

*Corresponding author e-mail: barissimsek@karatekin.edu.tr

Abstract

In order to reduce the amount of cement in concrete, researchers have turned to plant-derived additives in recent years. The addition of filler plays an important role in determining the mortar properties of cementitious composites. In this study, cement composites containing silver nanoparticles (AgNPs), multi-walled carbon nanotubes (MWCNTs), graphene oxide (GO) and sodium alginate (SOAL) were obtained. The thermal conductivity of these cement composites was measured and characterized using FTIR spectroscopy. The results showed that as the amount of sodium alginate increased in the cement mortar, the thermal conductivity decreased. The mechanical properties of sodium alginate added cement mortars could not be measured because the material dispersed during the curing process. It may be possible to determine its mechanical properties if mixed with different cement types instead of pozzolanic cement.

Keywords: Sodium alginate, vegetable cement paste, thermal conductivity, sustainability.

INTRODUCTION

The use of natural additives for sustainable concrete production has become extremely important with the increase in concrete production. Sustainability in concrete production is achieved by developing new concrete mixtures and improving performance (Greco et al., 2016). Natural additives and plant extracts have been shown to be valuable alternatives in the production of green and low-cost concrete, improving the mechanical properties of concrete while also reducing the production cost (Shanmugavel et al., 2020). Wang et al. (2023) used cactus sativus extract and mulberry leaf extract as additives in cement mortar making in their study and showed that these extracts increased the consistency of cement mortar and did not change the hydration reaction and hydration products of the mixture (Wang et al., 2023). Pan et al. (2023) reported that using carrot extract as a sustainable additive material significantly improved the mechanical properties and durability of concrete by increasing the compressive strength and splitting tensile strength of tunnel lining concrete (Pan et al., 2023).

Developments in nanotechnology in recent years affect many application areas such as chemical engineering, biology and construction (Dahlan et al., 2010). Nanomaterials provide extraordinary mechanical and thermal properties to cementitious composites thanks to their large surface areas. For this reason, the applications of these materials in the construction industry have attracted significant attention in recent years (Şimşek et al., 2021). When the literature is examined, it can be seen that carbon-based nanomaterials such as graphene oxide (Cheng et al., 2023), reduced graphene oxide (Wang R et al., 2023), and multi-walled carbon nanotubes (Mesquita et al., 2023) are being investigated to improve the thermal, optical, electrical, mechanical and other properties of cement composites (Bheel et al., 2023).

Sodium alginate ($(C_6H_7O_6Na)_n$) is a biodegradable and environmentally friendly bio-based polymeric additive consisting of the sodium salt of alginate, obtained by extraction from the cell walls of brown algae (Shang et al., 2022). It is seen as a low-cost alternative to synthetic additives and is reported to be renewable and abundant (Liu et al., 2023). Sodium alginate is rich in carboxyl and hydroxyl groups and has a circular long side chain structure (Yao et al., 2022). It also forms a viscous colloidal liquid when dissolved in water. Sodium alginate

DRAFT

is widely applied in the field of cell engineering such as cell immobilization technology due to its good biocompatibility (Zhang et al., 2023).

It is reported that alginate can form gel when in contact with cementitious materials. Liu et al. (2022) in their study, they investigated the effect and retardation mechanism of sodium alginate on the hydration properties of magnesium potassium phosphate cement (MKPC) suspension and reported that sodium alginate could prolong the hydration of MKPC with limited content and further increasing the sodium alginate content would not produce a better effect (Liu et al., 2022).

In this study, it was aimed to investigate the effect of silver nanoparticles (AgNPs), multi-walled carbon nanotubes (MWCNTs) and graphene oxide (GO) nanomaterials as well as sodium alginate on the thermal conductivity of cement composites. At the same time, the obtained cementitious composites were characterized by Fourier transform infrared spectroscopy (FTIR).

MATERIALS AND METHODS

Materials

Pozzolanic-type cement (grade 32.5R) equivalent to CEM IV/B (specific gravity of 2.78, Blaine fineness: 5105 cm^2/g) was used in this study. Polycarboxylate-ether-based type plasticizer (BASF Master Glenium 608) and distilled water were used for preparing cementitious composites. AgNPs (purity: >99.995%, size: 48-78 nm) and industrial grade MWCNT (purity: 92%, outer diameter: 48–78 nm, specific surface area: 50 m^2/g) were supplied from Nanografi Nanotechnology. GO particles (D-peak–to–G-peak intensity: 0.9, specific surface area: 180 m^2/g, thickness: 10–15 nm, average particle: 1.5–2 μm) were obtained by the improved Hummers method. Sodium alginate (molar mass: 216.121 g/mol) was supplied from Aromel Chemistry Medical.

Mixing and Casting Procedure

Table 1 presents the formulation detail of cementitious composites and the water-to-cement ratio was kept constant at 0.428. for all mixtures. Since the consistency of the cement mortar increases as the amount of sodium alginate increases, the amount of superplasticizer used was adjusted accordingly. Sodium alginate were added at a dosage of 0%, 0.3%, 0.6% and 1.0% by mass of cement. Three hybrid SOAL-cementitious composites were prepared with AgNPs, MWCNTs and GO at a dosage of 0.1% wt. Cement, water, and the super-plasticizer were premixed for 2 min., then nanofillers were put into the mixture gradually and the fresh mix was mixed for 2 more min. to obtain a compact design. After 24 h., all samples were de-molded.

Table 1. Mix proportions for cementitious composites

Exp. No.	Compounds						
	Cement (g)	DIW (ml)	SP (g)	SOAL (g)	AgNPs (g)	MWCNT (g)	GO (g)
SOAL0	525	225[1]	1.5	0	0	0	0
SOAL0.3	525	225	5.5	1.575	0	0	0
SOAL0.6	525	225	12	3.150	0	0	0
SOAL1	525	225	23	5.250	0	0	0
SOAL1AG	525	225	26	5.250	0.525	0	0
SOAL1CNT	525	225	26	5.250	0	0.525	0
SOAL1GO	525	225	26	5.250	0	0	0.525

Characterization of Cementitious Composites

Fourier Transform Infrared Spectroscopy (FTIR) analysis were carried out on powdered cementitious composites to evaluate the chemical changes in the cementitious composites. In the analysis, Bruker Tensor II device with a wavelength range of 400 to 4000 cm^{-1} was used.

Thermal Conductivity Measurements

A C-Therm thermal conductivity analyzer (Figure 1) was used to determine the 28-day thermal conductivity of the samples, according to ASTM D7984-16 (D-16, 2016).

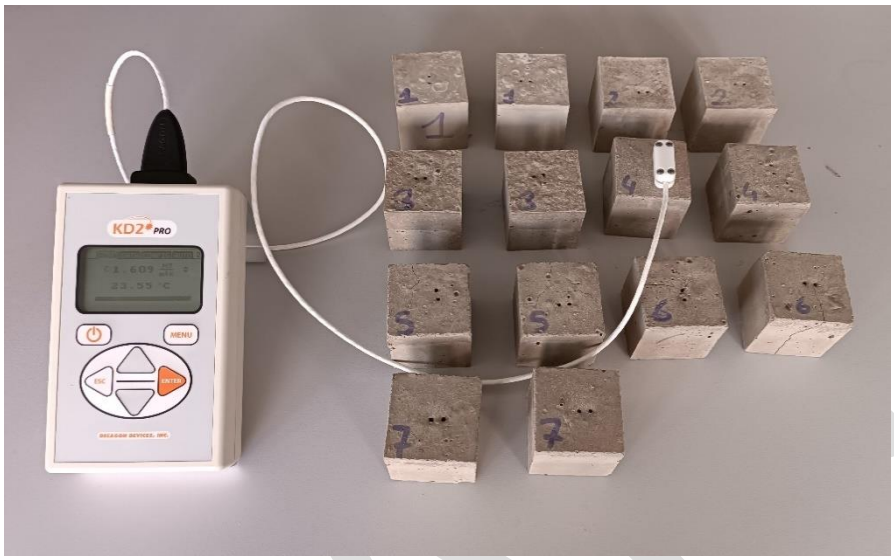


Figure 1. Thermal conductivity measurements of cementitious composites

RESULTS and DISCUSSION

FTIR Characterization

FTIR spectroscopy is frequently used to evaluate hydrated cement phases such as CSH (colloidal gel-like structures), portlandite, afwillite (crystalline microstructure) and calcite (Şimşek et al., 2022). FTIR spectra of the cementitious composites are given in Figure 2. The main characteristic peaks at 874, 1006 and 1100 cm^{-1}, indicating CSH content, correspond to Si-O-Si stretching vibrations. The intensity of the peaks between 874 and 1100 cm^{-1} decreased with the addition of sodium alginate. With the addition of 0.1 wt% MWCNT, the CSH peak intensity reached the highest level. The intensity of the peak observed around 1451 cm^{-1} due to C-O stretching vibrations and attributed to calcite increased with sodium alginate content. The peaks at 2322, 2900 and 3394 cm^{-1} correspond to the stretching vibrations of H-O-H and O-H can be attributed to H_2O molecules.

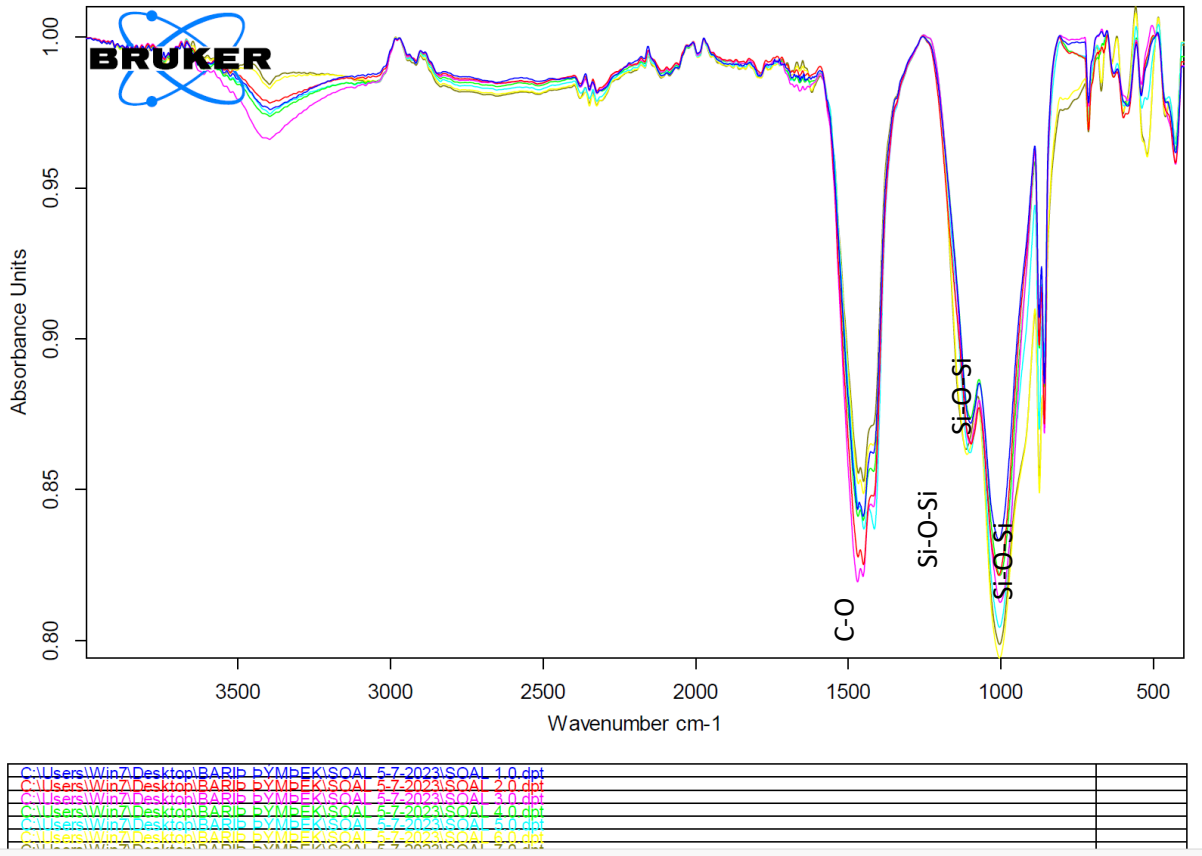


Figure 2. FTIR spectra of cementitious composites

Thermal Conductivity

Thermal conductivity results of the cementitious composites are given in Table 2. When the thermal conductivity results were examined, increasing the amount of sodium alginate in cementitious composites decreased the thermal conductivity values from 0.287 W/m.K to 0.194 W/m.K.

Table 2. Thermal conductivity results of the cementitious composites

	Temperature (ºC)	Thermal conductivity (W/m.K)
SOAL0	22.28	0.287±0.010
SOAL0.3	22.44	0.280±0.011
SOAL0.6	25.05	0.249±0.004
SOAL1	25.00	0.194±0.007
SOAL1AG	25.47	0.168±0.004
SOAL1CNT	25.61	0.164±0.004
SOAL1GO	25.09	0.153±0.003

CONCLUSION

In this study, it was aimed to analyse the properties of sodium alginate added cement mortars. Since it is a green material, it was preferred as an additive material and the following results were obtained:

As the amount of sodium alginate increased, the thermal conductivity of the cement mortar decreased and it was determined that it provided thermal insulation to the mortar.

In the mechanical tests performed on sodium alginate-added cementitious mortars, the 28-day compressive strength could not be determined due to the disintegration of the cement mortar. Even if graphene oxide and silver nanoparticle nanomaterials were added at 0.1% (by weight instead of cement), no significant improvement was observed in mechanical strength tests.

As a result of the study, it was concluded that different cement types should be used in order to use sodium alginate as a cement mortar substitute.

REFERENCES

Bheel N, Ali MOA, Kırgız MS, Shafiq N, Gobinath R 2023. Effect of graphene oxide particle as nanomaterial in the production of engineered cementitious composites including superplasticizer, fly ash, and polyvinyl alcohol fiber. Materials Today: Proceedings, In press.

Cheng Z, Liu Y, Wu J, Guo X, Chen W, Gao Y 2023. Graphene oxide-coated fly ash for high performance and low-carbon cementitious composites. Journal of Materials Research and Technology, 25: 6710-6724.

D-16 A 2016. Standard test method for measurement of thermal effusivity of fabrics using a modified transient plane source (MTPS) instrument. In: ASTM International West Conshohocken, PA.

Dahlan A S 2021. Impact of nanotechnology on high performance cement and concrete. Journal of Molecular Structure, 1223: 128896.

Greco E, Ciliberto E, Verdura PD, Lo Giudice E, Navarra G 2016. Nanoparticle-based concretes for the restoration of historical and contemporary buildings: a new way for CO_2 reduction in architecture. Applied Physics A, 122(5): 524.

Liu R, Fang B, Zhang G, Guo J, Yang Y 2022. Investigation of sodium alginate as a candidate retarder of magnesium phosphate cement: Hydration properties and its retarding mechanism. Ceramics International, 48(20): 30846-30852.

Mesquita E, Sousa I, Vieira M, Matos AM, Santos LPM., Silvestro L, Salvador R, D'Alessandro A, Ubertini F 2023. Investigation of the electrical sensing properties of cementitious composites produced with multi-wall carbon nanotubes dispersed in NaOH. Journal of Building Engineering, 77: 107496.

Pan J, Feng K, Chen W, Xing W, Wang Y 2023. Carrot extract as bio-admixture for performance enhancement of tunnel lining concrete. Journal of Building Engineering, 75: 107036.

Sanchez F, Sobolev K 2010. Nanotechnology in concrete – A review. Construction and Building Materials, 24(11): 2060-2071.

Shang X, Qu N 2023. Internal curing for high-performance concrete by a green composite capsule. Cement and Concrete Composites, 136: 104867.

Shanmugavel D, Selvaraj T, Ramadoss R, Raneri S 2020. Interaction of a viscous biopolymer from cactus extract with cement paste to produce sustainable concrete. Construction and Building Materials, 257: 119585.

Şimşek B, Doruk S, Ceran ÖB, Uygunoğlu T 2021. Principal component analysis approach to dispersed graphene oxide decorated with sodium dodecyl sulfate cement pastes. Journal of Building Engineering, 38: 102234.

Şimşek B, Uygunoğlu T, Dilmaç ÖF 2022. Optimization of Nanofiller-Blended Cementitious Composites Using Macrostructural and Microstructural Analyses. Journal of Materials in Civil Engineering, 34(10): 04022254.

Wang B, Lu K, Guangmin D, Wu Q 2023. Study on the effect of plant extracts as low carbon green admixtures on the performance of cement mortar. Case Studies in Construction Materials, 18: e02080.

Wang R, Sun R, Zhao L, Zhang T, Kong X, Fu Y 2023. Investigation of the dispersion of reduced graphene oxide in cementitious composites under different mixing strategies. Journal of Building Engineering, 77: 107447.

Yao F, Li M, Pan L, Li J, Xu N 2022. Synthesis of sodium alginate-polycarboxylate superplasticizer and its tolerance mechanism on montmorillonite. Cement and Concrete Composites, 133: 104638.

Zhang R, Xie D, Wu K, Wang J 2023. Optimization of sodium alginate aided bio-deposition treatment of recycled aggregates and its application in concrete. Cement and Concrete Composites, 139: 105031.

Synthetic food colorants and their health effects: A critical review

Nesibe Arslan Burnaz* (*ORCID: https://orcid.org/0000-0003-1163-4829*)

*Gümüşhane University, Faculty of Health Sciences, Department of Nutrition and Dietetics, Gümüşhane, Türkiye.

*Corresponding author e-mail: nsbburnaz@gmail.com

Abstract

The increase in diversity in food production, which is associated with the diversification of technological developments and changes in consumers' nutritional habits, has also increased the number of processed foods. In this context, additives applied to food products play an important role in consumers' preferences. Food additives are substances added to food to maintain, enhance, or improve its safety, freshness, taste, flavor, texture, or appearance (WHO, 2018). Colorants, which constitute an important class of food additives, are used to gain consumer appreciation, strengthen the natural color, restore the color lost during processing, or color a colorless product. Colorants may be derived naturally from plants and animals or may be synthetic. Synthetic colorants used in food industries are relatively cheaper, more durable, and brighter than natural ones. On the other hand, studies show that these additives pose a toxicological risk to health. The potential health risks associated with the use of these additives cause concern among consumers and spark controversy in society. Many synthetic food colorants have been banned in some countries on the grounds that they are carcinogenic due to experimental studies. Conflicting results in current studies evaluating its possible effects on health increase the controversy.

Keywords: Food additives, colorants, health

INTRODUCTION

With technological developments and changing consumer eating habits, the number of processed foods has also increased. Therefore, additives added to food products are important in consumers' preferences. (Silva vd., 2022).

Today, in the use of food additives in the food industry, many factors such as the sensory properties and nutritional value of the food, its ability to maintain its quality for a long time, and consumers' health and habits are taken into consideration. The diversification and widespread use of food additives also increases the risks to human health. The diversification and widespread use of food additives also increases the risks to human health (Bilişli, 2012).

In recent years, consumers' interest in sustainable foods, including compliance with environmental standards, food safety, product quality and effects on health, has increased. With the emergence of this awareness among consumers, food businesses have begun to consider the health risks of food products as well as their appeal (Gebhardt et al., 2020).

Despite using food additives in specified doses, some people who are sensitive to certain chemicals may experience many health problems such as allergies, hyperactivity, attention deficit, obesity, diabetes, gastrointestinal system, autoimmunity and neurodegenerative disorders (Körkoca and Bahşi, 2021; Sezgin and Ayyıldız, 2017). Therefore, before using food additives, their potentially harmful effects on human health should be evaluated. Recently, more and more attention has been paid to the toxicity of additives used in food. The Expert Committee on Food Additives (JECFA) is an international committee that conducts risk assessments of food additives (WHO, 2018). The European Parliament and the Council published a regulation stating that, as of 2009, the toxicity of food additives should be re-evaluated by the European Food Safety Authority (EFSA). EFSA is investigating available information on toxicity issues, particularly of synthetic food colourings, using official reports and current studies. The studies provide a summary of the evidence regarding the possible harmful effects of synthetic colorants and provide direction for future research (Amchova et al., 2015).

Food colorants constitute an important functional class of food additives. Recently, increasing attention has been paid to the toxicity of color additives used in foods, namely azo dyes. This group of colorants generally consists of bright colors. However, the main concern that often limits their use is their potential carcinogenic effect following reaction with metabolites by the gut microbiota (Feng et al., 2012).

Food Additives and Toxicological Assessments

Food additives can ensure the preservation of the quality and nutritional value of foods and increase their appeal, but their use in excessive amounts or for adulteration may cause potential risks to human health. Therefore, a risk assessment must be carried out before these additives are allowed to be used in foods (Bilişli, 2012).

Before starting to use food additives, ADI (Acceptable Daily Intake) and NOEAL (No Observable Adverse Effect Level) doses are calculated experimentally. “ADI” is the amount of additive that an adult can safely consume without experiencing any harmful effects, that is, the acceptable daily intake value. The ADI value is expressed as amount (mg) per body weight (kg). “NOEAL” is defined as the dose that does not cause any observable side effects for an additive. The ADI value is found by dividing the NOEAL value by the safety factor (100). When determining the maximum amount of use of a food additive in a food, the daily consumption amount and ADI value of foods containing this additive are considered (Amchova et al., 2015; Savaş, 2022). This value indicates the amount of food additive that can be consumed daily without posing a significant risk to consumer health (Amchova et al., 2015).

After food additives are approved by international organizations such as WHO, FAO, JECFA, a decision is made about which foods they can be added to and in what proportions. In the European Union (EU), safety assessments of food additives are carried out by a panel established within EFSA. The panel evaluates the safety of new food additives and the permitted uses of food additives. (EFSA, 2023). In our country, the competent authority for regulations regarding the use of food additives is the Ministry of Agriculture and Forestry.

Food Colorants

Color is considered one of the most important features that directly affects the consumer's food choice and eating desire (Sezgin and Ayyıldız, 2017). Colourants, which are indispensable components to attract the attention of consumers and determine their preferences, constitute an important class of food additives (Ukwo et al., 2022).

Colorants are used to restore the natural color of foods lost during production and storage or to strengthen the weak color and present it to the consumer. Colorants are also called food dyes. Today, colorants are added to products that are considered snacks, such as cakes, chips, biscuits, chocolate, chewing gum, pudding, flavored milk, fruit yoghurt, candies and jellybeans, which are especially consumed by children (Sezgin and Ayyıldız, 2017).

Colorants can be classified according to different criteria. The most used classification is that they are divided into natural or synthetic according to their source. Natural colorants can be obtained from plant tissue (carotenoids, anthocyanins, curcumin, chlorophylls etc.), animal cells (carminic acid), microorganism metabolism or mineral sources (calcium etc). Synthetic dyes are obtained by chemical synthesis and are not found in nature (Novais et al, 2022).

The first synthetic dye was a purple pigment called mauve, derived by William Henry Perkin in 1856 (Mota et al., 2023). Synthetic food dyes are azo dyes (Tartrazine, Sunset Yellow FCF, Azorubine, Amaranth, Ponceau 4 R, Allura Red, Brilliant Black BN, Brown FK, Brown HT, Red 2 G, Lithol rubine BK, Brilliant Blue, FCF, Patent Blue V and Fast Green FCF), Erythrosine, Indigotin and Quinoline Yellow) (Amchova et al., 2015).

In order to identify food additives, inform consumers and avoid any confusion, food additives approved for use by the EU are given the "E" code. It consists of the letter "E" and 3-digit numbers. All chemicals defined as additives and used in foods, whether natural or artificial, are included in this coding system (Sezgin and Ayyıldız, 2017). Food colorants contain E codes between E 100-180 (Karatepe and Ekerbiçer, 2017).

According to the Turkish Food Codex (TGK) Food Additives Regulation, the list of all colorants allowed to be used in foods in Türkiye is given in Table 1.

Table 1. Food Colorants (TGK, 2013)

E-code	Name	E-code	Name
E 100	Curcumin	E 150d	Ammonium sulfite caramel
E 101	Riboflavin	E 151	Brilliant Black BN, Black PN
E 102	Tartrazine	E 153	Vegetable carbon
E 104	Quinoline Yellow	E 155	Brown HT
E 110	Sunset Yellow FCF/Orange Yellow	E 160a	Carotenes
E 120	Cochineal, Carminic acid, Carmine	E 160b	Annatto, Bixin, Norbixin
E 122	Azorubine, Carmosine	E 160c	Paprika extract, Capsanthin
E 123	Amaranth	E 160d	Lycopene
E 124	Ponzo 4R, Cochineal Red A	E 160e	Beta-apo-8'-carotenal (C30)
E 127	Eritrosin	E 161b	Lutein
E 129	Allura Red AC	E 161g	Canthaxanthin
E 131	Patent Blue V	E 162	Beet root red, betanin
E 132	Indigotin, indigo carmine	E 163	Anthocyanins
E 133	Brilliant Blue FCF	E 170	Calcium carbonate
E 140	Chlorophylls and chlorophyllins	E 171	Titanium dioxide
E 141	Copper complexes of chlorophylls	E 172	Iron oxides, hydroxides
E 142	Green S	E 173	Aluminum
E 150a	Caramel	E 174	Silver
E 150b	Caustic sulphite caramel	E 175	Gold
E 150c	Ammoniac caramel	E 180	Litolrubin BK

Synthetic Food Colorants and Toxicity Assessment

Existing studies published since the 2009 EFSA official report have been reviewed and information on the toxicity of synthetic food colorants has been re-evaluated (Amchova et al., 2015).

Synthetic colorants are common ingredients added to food products, some of which can be metabolized by enzymes and absorbed or excreted after consumption. Metabolized dyes can become toxic and adversely affect health if consumed in large quantities (Gholami et al., 2021a, Rahnama et al., 2022). Studies provide evidence of the possible harmful effects of food coloring mixtures on children's behavior. Excessive consumption may aggravate attention deficit hyperactivity symptoms and asthma in sensitive children. Moreover, frequent headaches, cancer formation, neurological and genetic disorders, chromosomal damage, thyroid tumors, urticaria, allergies, respiratory problems, abdominal pain, weakening of the immune system, serious damage and disorders in fat metabolism, decrease in IQ and vitamin levels, etc. may cause (Gholami et al., 2021b).

Due to their negative effects on health, natural dyes extracted from plant materials of known origin are generally preferred by consumers in foods instead of synthetic dyes. However, instead of natural dyes, manufacturers have turned to synthetic dyes due to their low cost, stability and more attractive colors. (Shakoor et al., 2022). Today, numerous side effects (allergic reactions, behavioral and neurocognitive effects, and toxicity, etc.) are associated with both medium and long-term use of food colorants (Martins et al., 2016).

Tartrazine (E102) is a lemon yellow artificial azo dye used in products such as confectionery, sauces, chewing gum, puddings, beverages and ice cream. The FDA recommends an ADI of 3.75 mg/kg for tartrazine. Additionally, WHO recommends limiting ADI to 2.5 mg/kg (He et al., 2018). In a study, the neurobiochemical effects were investigated by giving ADI amount of tartrazine to rats, and it was found that tartrazine caused oxidative damage in brain tissue (Bhatt et al., 2018). For another azo dye, sunset yellow (E110), the ADI value allowed by JECFA and EFSA is 0-2.5 mg/kg body weight (EFSA, 2014). Certain studies have shown that dyes such as sunset yellow and tartrazine cause allergic reactions such as dermatitis, urticaria, exacerbation of asthma in sensitive individuals, and restlessness. Besides, it was reported that these colorants exacerbate sleep disorders and hyperactivity in children (Rahnama et al., 2022).

Quinoline yellow (E104) is also an artificial azo dye. It is used in products such as fruit drinks and chewing gum (25). A study not evaluated by JECFA and EFSA found increased hyperactivity in children aged 8-9 years exposed to quinoline yellow. EFSA Panel on Food Additives and Nutrient Sources added to Food (ANS)

evaluated the studies and concluded that quinoline yellow has negative long-term carcinogenicity as well as in vitro genotoxicity. The panel determined the ADI value as 0.5 mg/kg using the NOAEL obtained from the chronic toxicity and carcinogenicity study with reproductive toxicity stage in rats (EFSA, 2009a). A study has shown that quinoline yellow used in concentrations above the determined ADI value causes a mutagenic effect on human lymphocytes (Chequer et al., 2015).

Azorubine/Carmosine (E122) is a red-colored azo dye that is licensed as a food additive in the EU and is widely used in the food, pharmaceutical and cosmetic industries. In 2009, the EFSA ANS Panel re-evaluated the safety of the use of E122 as a food additive and the levels of use permitted in the legislation. The ADI value determined based on this evaluation is 0-4 mg/kg (EFSA, 2009a).

Amaranth (E123) is a synthetic dye with a red-brown appearance. It is used in cherry candies, some types of alcoholic beverages and fish roe (Gültekin, 2021). The ADI value, which was determined as 0-0.8 mg/kg for E123 based on the results of a 90-day animal trial study by JECFA and EFSA, was changed to 0-0.5 mg/kg after a long-term carcinogenicity study in animals. In 2010, the ADI of E123 was re-evaluated by the ANS Panel and determined as 0.15 mg/kg. Nowadays, allura red is preferred instead of amaranth due to its negative effects on health (EFSA, 2010a).

Allura Red (E129) is a colorant permitted for use in food and beverages, dietary supplements, pharmaceuticals and other consumer products worldwide (Gültekin, 2021). It is approved as a color additive by the "Food, Drug and Cosmetics" Committee in the USA. Research conducted since 1980 has shown that this colorant is genotoxic, etc. Although its effects are controversial, it was evaluated to be safe as a food additive at the last JECFA meeting in 2016 and its ADI value was determined as 0-7 mg/kg body weight/day (Joint, 2016). Consistent evidence has been found in many in vitro and in vivo studies that Allura red does not have carcinogenic and genotoxic potential (Bastaki et al., 2017).

Studies have shown that some food additives cause hyperactivity in children. A recent study in England found that children with no history of hyperactive disorder showed varying degrees of hyperactivity after consuming fruit drinks containing varying levels of additives. Tartrazine (E102), quinoline yellow (E104), Sunset yellow (E110), Carnosine (E122), Allura red (E129) were among the colorants examined (Kumar and Joshi, 2023).

Brilliant Blue (E133) is obtained from petroleum and is widely used all over the world. It is a food dye that is allowed to be used in many countries, including the EU, Japan, the USA and Turkey. It is used to color breakfast cereals, cakes, candies, chewing gum, dairy products, beverages, packaged soups, desserts, creams, flavored waters, frozen treats, etc. The ADI value for E133 by EFSA is 6.0 mg/kg body weight per day (Joint, 2017; EFSA, 2010b).

Indigotin (E132) is another food coloring of artificial origin. It is blue at pH 11.4 and yellow at pH 13.0. It is very sensitive to light and oxidizing substances. It is used in flavored and alcoholic beverages, sauces, confectionery, flavored fermented milk products, spices, desserts, edible ice and snacks. ADI value is 5 mg/kg. No changes or side effects were detected in chronic toxicity studies at doses lower than 500 mg/kg. However, some studies have reported the possible presence of potentially carcinogenic substance residues on human health and side effects such as allergy. In addition, simultaneous consumption with delicatessen products containing sodium nitrite (E250) is not recommended. Studies in animal models have reported adverse health effects such as nausea, vomiting, skin rashes, and brain tumors (Silva et al., 2022).

Iron oxides (E172) are widely used to color desserts, olives or the outer surface of cheese. E172 can be black, yellow, red or orange-brown. The ADI value has been determined by JECFA as 0.5 mg/kg body weight. Since there is no adequate biological and toxicological database, they are considered safe food pigments with low acute toxicity. E172 is permitted for use in cosmetics only in the United States. It is used as food coloring in many other countries, including our country (Sun et al., 2017; EFSA, 2015).

It is known that most approved food colorants are synthetic and have carcinogenic potential with side effects, toxicity and other adverse reactions when used incorrectly. In this regard, regulatory bodies around the world have banned some synthetic additives. However, a global consensus on food additives legislation has not yet been established. For example, Coloring agents Carmoisine (E122), amaranth (E123), and patent blue (E131) are allowed in the EU but banned in the USA. For all these reasons, especially in developed countries, curcumin (E100), carmines (E120), chlorophylls (E140, E141), carotenes (E160a), anatto extracts (E160b), paprika extract (E160c), lycopene (E160d), beet root (E162). anthocyanins (E163), such as there is a need to use natural additives with fewer side effects (Carocho et al., 2014; Novais et al., 2022).

DRAFT

CONCLUSION

The use of food colorants to improve and improve the sensory properties of foods is a technological necessity. The use of colorants contrary to legislation poses a risk to food safety. Additionally, most food colorants are synthetic, which raises concerns among consumers. Although research on food colorants and their effects on health is increasing day by day, health risks remain unclear, especially due to conflicting results obtained regarding synthetic colorants. This situation brings about discussions about the reliability of synthetic food colorants.

In order to prevent the use of food colorants contrary to the legislation, food producers should be made aware of this issue, inspected and control mechanisms should be constantly improved. Consumers should be made aware of synthetic food colorants and the health effects of foods containing them. In this context, it is important to avoid food items containing synthetic colorants that are considered dangerous to health or to reduce their use in the diet in order to avoid possible risks. Consumption of foods containing these additives should be limited, especially in children, pregnant women and sensitive people.

REFERENCES

Amchova P, Kotolova H, Ruda-Kucerova J 2015. Health safety issues of synthetic food colorants. Regulatory Toxicology Pharmacology, 73(3): 914-922.

Awuchi CG, Twinomuhwezi H, Igwe VS, Amagwula IO 2020. Food additives and food preservatives for domestic and industrial food applications. Journal of Animal Health, 2(1): 1-16.

Bastaki M, Farrell T, Bhusari S, Pant K, Kulkarni R 2017. Lack of genotoxicity in vivo for food color additive Allura Red AC. Food and Chemical Toxicology, 105: 308-314.

Bhatt D, Vyas K, Singh S, John PJ, Soni I 2018. Tartrazine induced neurobiochemical alterations in rat brain sub-regions. Food and Chemical Toxicology, 113: 322-327.

Bilişli A 2012. Gıda katkı maddeleri. In: Bilişli A, (edt). Gıda Kimyası. 2. Baskı. Çanakkale: Sidas Yayıncılık, pp. 271-308.

Carocho M, Barreiro MF, Morales P, Ferreira IC 2014. Adding molecules to food, pros and cons: A review on synthetic and natural food additives. Comprehensive Reviews in Food Science and Food Safety, 13(4): 377-399.

Chequer FMD, de Paula Venâncio V, de Souza Prado MR, da Silva LRC Junior C, Lizier TM et al. 2015. The cosmetic dye quinoline yellow causes DNA damage in vitro. Mutation Research/Genetic Toxicology and Environmental Mutagenesis, 777: 54-61.

EFSA 2014. Panel on Food Additives and Nutrient Sources Added to Food (ANS). Statement on a conceptual framework for the risk assessment of certain food additives re-evaluated under Commission Regulation (EU) No 257/2010. EFSA Journal, 12(6): 3697.

EFSA Panel on Food Additives and Nutrient Sources added to Food (ANS). Reconsideration of the temporary ADI and refined exposure assessment for Sunset Yellow FCF (E110). EFSA Journal. 2014;12(7): 3765.

EFSA 2010a. Panel on Food Additives and Nutrient Sources added to Food (ANS). Scientific Opinion on the re-evaluation of Amaranth (E123) as a food additive. EFSA Journal, 8(7): 1649.

EFSA 2010b. Panel on Food Additives and Nutrient Sources added to Food (ANS). Scientific Opinion on the re-evaluation of Brilliant Blue FCF (E133) as a food additive. EFSA Journal, 8(11): 1853.

EFSA 2015. Panel on Food Additives and Nutrient Sources added to Food (ANS). Scientific Opinion on the re-evaluation of iron oxides and hydroxides (E172) as food additives. EFSA Journal, 13(12): 4317.

EFSA 2009a. Panel on Food Additives and Nutrient Sources Added to Food. Scientific Opinion on the re-evaluation of Quinoline Yellow (E104) as a food additive. EFSA Journal, 7(11): 1329, 1-40.

EFSA 2009b. Panel on Food Additives and Nutrient Sources Added to Food. Scientific Opinion on the re-evaluation of Azorubine/Carmoisine (E122) as a food additive. EFSA Journal, 7(11): 1332, 1-40.

EFSA 2023. Food Additives, Available at: https://www.efsa.europa.eu/en/topics/topic/food-additives [21.08.23]

Feng J, Cerniglia CE, Chen, H 2012. Toxicological significance of azo dye metabolism by human intestinal microbiota. Frontiers in Bioscience (Elite edition), 4, 568.

Gebhardt B, Sperl R, Carle R, Müller-Maatsch J 2020. Assessing the sustainability of natural and artificial food colorants. Journal of Cleaner Production, 260, 120884.

Gholami Z, Marhamatizadeh MH, Yousefinejad S, Rashedinia M, Mazloomi SM 2021a. Vortex-assisted dispersive liquid-liquid microextraction based on hydrophobic deep eutectic solvent for the simultaneous identification of eight synthetic dyes in jellies and drinks using HPLC-PDA. Microchemical Journal, 170: 106671.

Gholami Z, Marhamatizadeh MH, Mazloomi SM, Rashedinia M, Yousefinejad S 2021b. Identification of Synthetic Dyes in Traditional Juices and Beverages in Shiraz, Iran. International Journal of Nutrition Sciences, 6(1): 39-44.

Gültekin F 2021. Gıda katkı maddeleri sözlüğü. In: Gültekin F (edt). Fark Etmeden Yediklerimiz: A'dan Z'ye Gıda Katkı Maddeleri. 1. Baskı. İstanbul: Server Yayınları, pp. 87-395.

He Q, Liu J, Liu X, Li G, Deng P, Liang J, Chen D 2018. Sensitive and selective detection of tartrazine based on TiO_2-electrochemically reduced graphene oxide composite-modified electrodes. Sensors, 18(6): 1911.

Joint, FAO 2016. World Health Organization, & WHO Expert Committee on Food Additives. Evaluation of certain food additives: eighty-second report of the Joint FAO. World Health Organization.

Joint, FAO 2017. World Health Organization, & WHO Expert Committee on Food Additives. Evaluation of certain food additives: eighty-fourth report of the Joint FAO. World Health Organization.

Karatepe TU, Ekerbiçer HÇ 2017. Gıda katkı maddeleri. Sakarya Tıp Dergisi, 7(4): 164-167.

Körkoca A, Bahşi Ş 2021. Gıda Katkı Maddeleri ve Sağlık. Muş Alparslan Üniversitesi Sağlık Bilimleri Dergisi, 1(1):26-32.

Kumar P, Joshi N 2023. Are the Food Additives, Safe or Harmful?-A Review. Journal of Ayurveda and Integrated Medical Sciences, 8(5): 140-144.

Martins N, Roriz CL, Morales P, Barros L, Ferreira IC 2016. Food colorants: Challenges, opportunities and current desires of agro-industries to ensure consumer expectations and regulatory practices. Trends in Food Science and Technology, 52:1-15.

Mota IGC, Neves RAMD, Nascimento SSDC., Maciel BLL, Morais AHDA, Passos TS 2023. Artificial dyes: Health risks and the need for revision of international regulations. Food Reviews International, 39(3): 1578-1593.

Novais C, Molina AK, Abreu RM, Santo-Buelga C, Ferreira IC, Pereira C, et al. 2022. Natural food colorants and preservatives: A review, a demand, and a challenge. Journal of Agricultural and Food Chemistry, 70(9): 2789-2805.

Rahnama H, Mazloomi SM, Berizi E, Abbasi A, Gholami, Z 2022. Identification of Tartrazine adulteration and evaluating exposure to synthetic dyes of sunset yellow and Quinoline yellow through consumption of food products among children. Food and Nutrition Sciences, 10(11): 3781-3788.

Savaş HB 2022. Gıda katkı maddeleri ve beslenme biyokimyası. In: Savaş HB (edt). Beslenme Biyokimyası. 1. Baskı. Ankara: Nobel Akademik Yayıncılık, pp. 511-518.

Sezgin AC, Ayyıldız S 2017. Food additives: colorants. In: Méndez-Vilas A (edt). Science within Food: Up-to-Date Advances on Research and Educational Ideas. Badajoz, Spain: Formatex Research Center, pp. 87-94.

Shakoor S, Ismail A, Sabran MR, Bekhit AEDA, Roohinejad S 2022. Impact of tartrazine and curcumin on mineral status, and thyroid and reproductive hormones disruption in vivo. International Food Research Journal, 29(1): 186-199.

Silva MM, Reboredo FH, Lidon FC 2022. Food colour additives: A synoptical overview on their chemical properties, applications in food products, and health side effects. Foods, 11(3): 379.

Sun B, Wang J 2017. Food additives. In: Joseph JJ, Chen J (eds). Food Safety in China: Science, Technology, Management and Regulation, Part3, Chapter 12; Wiley, pp. 186-200.

TGK 2013. Türk Gıda Kodeksi Gıda Katkı Maddeleri Yönetmeliği, 2013. Available at: https://www.mevzuat.gov.tr/mevzuat?MevzuatNo=18532&MevzuatTur=7&MevzuatTertip=5 [04.07.23]

Ukwo SP, Udo II, Ndaeyo, N 2022. Food additives: overview of related safety concerns. Journal of Food Science and Nutrition Research. 5: 1-10.

WHO, Food Additives, 2018. https://www.who.int/news-room/fact-sheets/detail/food-additives [04.07.23]

ORAL PRESENTATION

Biberiye (Rosemary) Yağının Diyabetik Sıçanlarda Beyin ve Testis Kolestrol ve Total Trigliserit Değerleri Üzerine Etkisi

Deniz Uluışık[1] *(ORCID: https://orcid.org/0000-0003-1462-0836)*, Ercan Keskin[1] *(ORCID: https://orcid.org/0000-0003-0598-8700)*, Durmuş Hatipoğlu[1*] *(ORCID: https://orcid.org/0000-0003-3790-7821)*

[*1] Selçuk Üniversitesi, Veteriner Fakültesi, Fizyoloji Anabilim Dalı, Konya, Türkiye

*Sorumlu yazar e-mail: drhatip@selcuk.edu.tr

Özet

Bu çalışmada, streptozotosin (40 mg/kg, s.c.) ile indüklenmiş diyabetik sıçanlarda beyin ve testis lipid değerleri üzerine Rosemary (biberiye) yağının etkinliği test edilmiştir. Otuz iki Wistar albino sıçan dört gruba ayrılmıştır: sağlıklı kontrol (K, n:6), Rosemarry (RSM, n:6), diyabetik kontrol (DM, n:10) ve diyabetik + Rosemarry (DM+RSM, n:10). Kontrol ve DM gruplarına herhangi bir tedavi uygulaması yapılmazken, RSM ve DM+RSM gruplarına Rosemary yağı (günde 200 mg/kg, oral) 21 gün boyunca uygulanmıştır. DM grubunda beyin ve testis total trigliserit (TG) ve kolestrol (TC) düzeyleri kontrol grubuna göre istatistiksel olarak önemle derecede artmıştır. Diyabet oluşturulan grupların Rosemary ile tedavi edilmesi (DM+RSM) ise bu etkileri tersine çevirerek bu lipid profilleri üzerine iyileşme sağlamıştır. Bu çalışmanın bulguları, biberiye uygulamasının diyabetik sıçanlarda daha iyi lipid profili sağladığını göstermektedir.

Anahtar Kelimeler: Diyabet, beyin, biberiye, kolesterol, testis, trigliserit.

The Effect of Rosemary Oil on Cholesterol and Total Triglyceride Levels in Diabetic Rats' Brain and Testis

Abstract

In this study, the effectiveness of Rosemary oil on brain and testis lipid levels was tested in diabetic rats induced with streptozotocin (40 mg/kg, s.c.). Thirty-two Wistar albino rats were divided into four groups: healthy control (K, n:6), Rosemary (RSM, n:6), diabetic control (DM, n:10), and diabetic + Rosemary (DM+RSM, n:10). While no treatment was applied to the control and DM groups, Rosemary oil was administered to the RSM and DM+RSM groups orally at a dose of 200 mg/kg for 21 days. In the DM group, total triglyceride (TG) and cholesterol (TC) levels in the brain and testis were significantly increased compared to the control group. Treatment with Rosemary in the diabetic groups (DM+RSM) reversed these effects, leading to an improvement in these lipid profiles. The findings of this study demonstrate that Rosemary application provides a better lipid profile in diabetic rats.

Keywords: Diabetes, brain, rosemary, cholesterol, testicles, triglycerides.

GİRİŞ

Diyabetes mellitus (DM), vücutta insülin aktivitesinin azalması veya insülin salınımının azalması sonucu meydana gelen bir metabolik bozukluktur. Hastalığın ilerlemesiyle birlikte, vücutta kaçınılmaz olarak nefropati, retinopati ve kardiyovasküler komplikasyonlar gibi patolojik değişiklikler meydana gelir (Padhi ve ark., 2020). Tip 2 Diyabetes Mellitus (T2DM), insüline duyarlı dokularda yetersiz insülin etkisi ve anormal insülin salınımı nedeniyle yüksek kan glukoz seviyeleri ile karakterize edilen en yaygın metabolik bozukluklardan biridir (Li ve ark., 2012, Roden ve Shulman 2019). Beta hücreleri, metabolik ihtiyaçları karşılamak için yeterli insülin üretme veya salgılama kapasitesini kaybettiğinde veya periferik dokular, insülin sinyaline yanıt verme yeteneklerini yitirerek insülin direnci geliştirdiğinde, bu durum açlık plazma glukoz seviyelerinin yükselmesine neden olur (Zaccardi ve ark., 2016). İnsülin direnci genellikle T2DM teşhisinden önce ortaya çıkar ve yüksek TAG ve NEFA, düşük HDL-C seviyeleri ve aterojenik küçük yoğun LDL partiküllerinin baskınlığı ile karakterize edilen belirgin bir dislipidemi ile birlikte görülür (Filippatos ve ark., 2017, Galicia-Garcia ve ark., 2020). T2DM'li hastalar arasında diyabetik dislipidemi (DD) çok yaygındır

(prevalans %72-85) (Turner ve ark., 1998, Vergès 2015). DD, aterosklerozun oluşumunda ve ilerlemesinde merkezi bir rol oynadığından, bu fenomen DM'si olmayan bireylere kıyasla önemli ölçüde artmış kardiyovasküler hastalıklar riski ile ilişkilidir (Battisti ve ark., 2003). DD'nin başlıca kantitatif lipoprotein anormallikleri artmış trigliseritler (TG'ler) ve total kolestrol (TC) ile ilişkilidir (Doucet ve ark., 2012). DD, temel olarak yağ dokusu ve karaciğerin bozulmuş insülin duyarlılığına bağlı olarak gelişir. Yağ dokusu insüline karşı dirençli hale geldiğinde, hormona duyarlı lipazın inhibisyonu ve glikoz emiliminin azalması nedeniyle kontrolsüz lipoliz gözlenir ve dolaşımdaki NEFA seviyelerinin artmasına neden olur (Filippatos ve ark 2017, Galicia-Garcia ve ark 2020). Karaciğerde, insülin TAG bakımından zengin lipoprotein üretimini düzenler; yani insülin direnci durumunda, hepatik aşırı üretim nedeniyle dolaşımdaki VLDL artar (Filippatos ve ark 2017, Galicia-Garcia ve ark 2020). T2DM ile ilişkilendirilen hiperglisemi ve hiperinsülinemi, HDL'nin yapısı ve işlevini etkileyebilir (Constantinou ve ark., 2016, Xepapadaki ve ark., 2021). T2DM'de dislipideminin patofizyolojisi tam olarak anlaşılmamış olsa da hiperglisemi, insülin direnci, hiperinsülinemi, adipokinler ve adipositokinler gibi çeşitli faktörlerin rol oynadığı düşünülmektedir (Taskinen 2003). Son gelişmeler, potansiyel olarak aterojenik lipid ve lipoprotein anormalliklerinin bir kümesi olan diyabetik dislipideminin karmaşık doğasını kabul etmiştir (Navab ve ark., 2011). DD'nin ana bileşenleri artmış plazma trigliseritleri ve düşük HDL kolesterol konsantrasyonlarıdır ve bu bileşenler, küçük yoğun LDL ve aşırı postprandiyal lipemi gibi daha yeni tanımlanan özelliklerle metabolik olarak sıkı bir ilişki içindedir (Taskinen 2003). Bu nedenle son yıllarda, DM ve DD gibi çeşitli metabolik sendromların tedavisinde yaygın olarak araştırılan, daha az yan etkiye sahip ve çok hedefli tedaviye yönelik bitkisel ilaçlara ilgi giderek artmaktadır (Ramadan ve ark., 2013, Bhatti ve ark., 2022).

Rosmarinus officinalis Linn. (Lamiaceae), biberiye (Rosemary) olarak bilinen, hipoglisemi tedavisinde kullanılan, çok çeşitli farmakolojik aktivitelere sahip, yenilebilir, tıbbi, süs ve esansiyel yağ içeren bir bitkidir (Kontogianni ve ark., 2013, Abdelrahman ve ark., 2020). Önemli bir şekilde, son klinik çalışma, T2DM hastalarının biyokimyasal parametrelerinin, antropometrik parametrelerinin ve lipid peroksid seviyelerinin, 3 ay boyunca biberiye çayı tüketiminden sonra önemli ölçüde azaldığını ve pankreatik β-hücre işlevinin ve insülin direncinin önemli ölçüde iyileştiğini bulmuş, bu da biberiyenin diyabet üzerindeki tedavi edici etkisini doğrulamıştır (Quirarte Báez ve ark., 2019). Ancak, biberiye uygulamasının diyabetle ilişkili olarak beyin ve testis lipid profilleri üzerindeki etkileri hala tam olarak anlaşılamamıştır. Bu yüzden mevcut araştırmada deneysel olarak (streptozotosin-STZ) DM oluşturulan ratlarda biberiyenin beyin ve testis dokuları trigliserid (TG) ve total kolesterol (TK) düzeyleri üzerine etkinliği araştırılmıştır.

MATERYAL VE METOT

Deney Hayvanı Temini ve Etik Kurul Kararı

Tüm deneysel prosedürler, hayvan refahına ilişkin Avrupa Ekonomik Topluluğu Direktiflerine (86/609/CEE ve 2010/63/EU) uygun olarak gerçekleştirildi. Çalışma sırasında sıçanlar, 12 saat gündüz ve 12 saat gece ışık döngüsüne sahip, oda sıcaklığı 22±2°C ve %50±10 nem olan kafeslerde barındırıldı. Selçuk Üniversitesi Veteriner Fakültesi Deney Hayvanı Üretim ve Araştırma Merkezi Etik Kurulu, çalışmanın araştırma etiğine uygunluğunu onayladı (Onay No: 2021-61).

Deneysel Grupların Oluşturulması ve Lipid Profilinin Belirlenmesi

Araştırmada 250-300 gr ağırlığında 32 adet sağlıklı yetişkin Wistar Albino sıçan kullanıldı. Ratlar şu şekilde dört gruba ayrıldı: Grup I (K,n:6): Herhangi bir uygulama yapılmadı. Grup II (RSM, n:6): 3 hafta boyunca her gün oral yolla 200 mg/kg biberiye yağı verildi (Yesilbag ve ark., 2011). Grup III (DM, n:10): Çalışmanın başlangıcında iki gün boyunca günde tek doz subkutan 40 mg/kg streptozotosin verilerek diyabet oluşturuldu. Diyabet oluşumu gerçekleşen ratlara herhangi bir uygulama yapılmadı. Grup IV (DM+RSM, n:10): Çalışmanın başlangıcında iki gün boyunca günde tek doz subkutan 40 mg/kg streptozotosin ile diyabet oluşturuldu. Diyabet oluşumu gerçekleşen ratlara 3 hafta boyunca her gün oral yolla 200 mg/kg biberiye yağı verildi.

Deneme gruplarında (DM ve DM+RSM), streptozotosin enjeksiyonundan bir hafta sonra ratların kuyruk venasından alınan kanlarını glikometre ile ölçerek diyabet oluşumunu kontrol ettik. Kan glikoz düzeyi 250 mg/dl'nin üzerinde olan ratlar, diyabet gruplarına dahil edildi. Deneysel çalışmanın ardından tüm hayvanlara sedasyon altında (10 mg/kg ksilazin ve 90 mg/kg ketamin) ötenazi ve nekropsi uygulandı. Servikal dislokasyon yöntemiyle ötenazi edilen hayvanların beyinleri ve testisleri hızlı bir şekilde çıkarılıp tuzlu su ile

yıkandıktan sonra sıvı nitrojenle dondurulduktan, TG ve TC düzeyleri belirlenene kadar -80°C'de saklandı. Beyin ve testis dokularında TG ve TC düzeyleri otoanalizörde belirlendi.

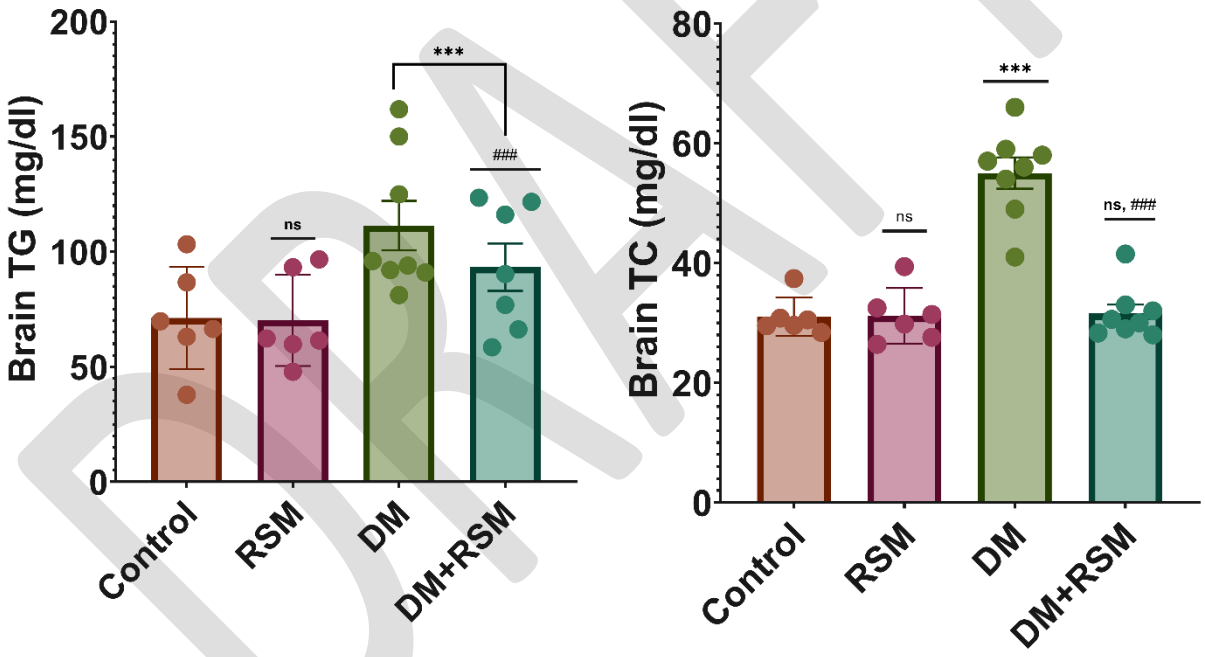
İstatiksel Analizler

Çalışmada elde edilen veriler SPSS istatistik programı yardımıyla One-Way ANOVA testi ve ardından çoklu karşılaştırma testlerinden Duncan testiyle analiz edilerek grupların ortalama değerleri arasındaki farkların önem dereceleri istatistiksel yöntemlerle belirlendi. Sonuçlar ortalama ± ortalamanın standart deviation şeklinde tablolandı. Tüm şekiller GraphPad Prism version 5.0 for Windows (GraphPad Software, San Diego, California USA) kullanılarak üretilmiştir.

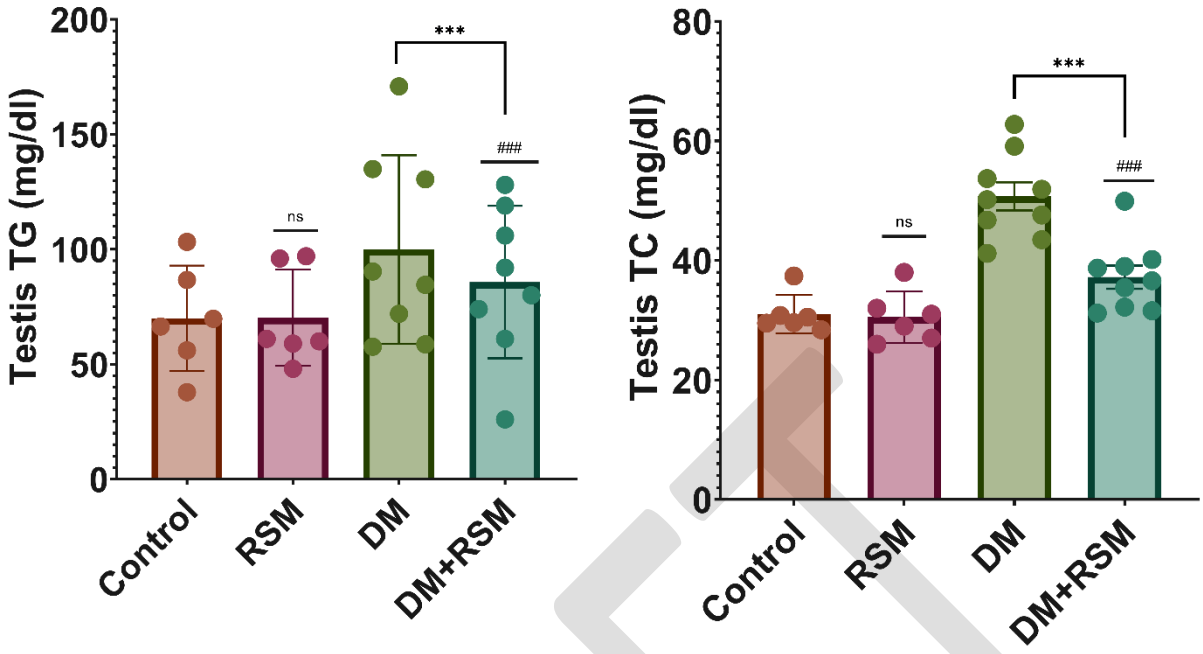
BULGULAR

Biberiye tüketimi DM'ye karşı beyin ve testis TG ve TC düzeyini iyileştirdi

Biberiye kullanımının, diyabetes mellitus'a karşı fitofarmakolojik tedavisinin potansiyeline dair veriler Tablo 1'de sunulmuştur. DM grubunun beyin ve testis dokularındaki total trigliserid (TG) ve kolesterol (TC) düzeyleri, kontrol grubuna kıyasla istatistiksel olarak anlamlı bir şekilde yükselmiştir ($p<0.05$). Ayrıca, biberiye takviyesinin özellikle beyinde TC düzeyini azalttığı ($p<0.05$) ve hatta kontrol grubuyla benzer seviyelere getirdiği gözlemlenmiştir. Buna ek olarak, bu fitoterapötik uygulamanın her iki dokuda da TG düzeylerini, DM grubuna göre anlamlı bir şekilde azalttığı tespit edilmiştir ($p<0.05$).



Şekil 1. Biberiyenin () STZ ile deneysel diyabet modeli oluşturulan ratlarda beyin total kolestrol (TC) ve beyin trigliserit (TG) düzeylerine etkisi. '***'; Kontrol grubuna göre istatistiksel olarak anlamlı farklılık göstermektedir ($p<0,05$), '###'; DM grubuna göre istatistiksel olarak anlamlı farklılık göstermektedir ($p<0,05$). 'ns'; Kontrol grubuna göre istatistiksel olarak anlamlı bir fark olmadığını göstermemektedir ($p>0,05$). RSM: Üç hafta boyunca günde 200 mg/kg vücut ağırlığında oral biberiye yağı uygulanan grup. DM: STZ ile diyabet oluşturulan grup. DM+RSM: STZ ile diyabet oluşturulduktan sonra üç hafta boyunca günde 200 mg/kg vücut ağırlığında oral biberiye yağı uygulanan grup.



Şekil 2. Rosemary'nin (RSM, 200 mg/kg vücut ağırlığı/gün) STZ ile deneysel diyabet modeli oluşturulan ratlarda (DM) testis total kolestrol (TC) ve testis trigliserit (TG) düzeylerine etkisi. '***'; Kontrol grubuna göre istatistiksel olarak anlamlı farklılık göstermektedir ($p<0,05$), '###'; DM grubuna göre istatistiksel olarak anlamlı farklılık göstermektedir ($p<0,05$). 'ns'; Kontrol grubuna göre istatistiksel olarak anlamlı bir fark olmadığını göstermemektedir ($p>0,05$). RSM: Üç hafta boyunca günde 200 mg/kg vücut ağırlığında oral biberiye yağı uygulanan grup. DM: STZ ile diyabet oluşturulan grup. DM+RSM: STZ ile diyabet oluşturulduktan sonra üç hafta boyunca günde 200 mg/kg vücut ağırlığında oral biberiye yağı uygulanan grup

TARTIŞMA

Diyabetin glukoz metabolizması üzerindeki etkilerini inceleyen çok sayıda çalışma mevcutken, diyabetin komplikasyonlara yatkın dokuların lipid içeriği ve metabolizmasını nasıl etkilediğini araştıran çalışmaların sayısı oldukça sınırlıdır. Bu bilgi eksikliğini ele almak amacıyla araştırma grubumuz, beyin ve testis dokularındaki trigliserit (TG) ve toplam kolesterol (TC) düzeylerini ve bu lipidler üzerine iyileştirici etkisi olabileceğini düşündüğümüz biberiye (rosemary) yağının etkinliğini incelemiştir. Bu çalışmada, oral yolla verilen biberiye yağının, STZ ile oluşturulan diyabet modeli üzerinde beyin ve testis dokularında TC ve TG düzeylerini nasıl etkilediğini değerlendirdik. Deneylerimiz sonucunda, diyabet oluşturulan ratların beyin ve testis dokularında TG ve TC düzeylerinin arttığını fakat üç hafta boyunca günde 200 mg/kg oral yolla uygulanan biberiye yağının bu profili iyileştirdiğini gösterdik.

T2DM'de glisemik kontrolün iyileşmesine rağmen diyabetik komplikasyon riskinin devam etmesi, metabolik sendromun diğer bileşenlerinin bu komplikasyonların başlangıcında ve ilerlemesinde rol oynayabileceğini düşündürmektedir (Eid ve ark., 2019). DM'nin makrovasküler komplikasyonlarıyla yakından ve nedensel olarak ilişkili olan en önemli bileşenlerinden biri diyabetik dislipidemidir (DD) (Turner ve ark 1998, Farmer 2008). T2DM hastalarında hiperinsülinemi, sıklıkla DD ile ilişkilidir (Isomaa ve ark., 2001). Yüksek kan glukozu ve insülin direncine yanıt olarak artan hepatik lipogenez ve yağ dokusu yağ asidi metabolizması hem tip 1 hem de tip 2 diyabette dislipidemiye katkıda bulunur (Vergès 2015). Araştırmamız sonuçlarına göre, DM'li sıçanlarda beyin ve testislerinde TG ve TC düzeylerinde kontrol grubuna kıyasla artışlar tespit edilmiştir. Birçok çalışma, plazma TG düzeylerindeki artıştan sorumlu patojenik mekanizmaların esas olarak TG'den zengin lipoprotein partiküllerindeki, özellikle de insülin direnciyle doğrudan bağlantılı olan büyük VLDL'deki artışın bir sonucu olduğunu belgelemiştir (Taskinen 2003, Selmi ve ark., 2017). Bunun yanı sıra beyinde TC'deki bu artış HMG-CoA redüktaz aktivitesinin artmasından kaynaklanmaktadır (Bopanna ve ark., 1997). Ayrıca diyabetik sıçanların beyninde gözlenen yüksek TG seviyeleri ise insülin eksikliği ile ilişkilidir, çünkü normal koşullar altında insülin TG'leri hidrolize eden lipoprotein lipaz enzimini aktive etmektedir (Mainzen Prince ve ark., 2011). DM'nin dünya çapında artan insidansı, kaçınılmaz olarak üreme çağındaki erkeklerde

bu patolojinin daha yüksek prevalansı ile sonuçlanacaktır ve DM ile ilişkili subfertilite veya infertilitenin önümüzdeki yıllarda dramatik bir şekilde artması beklenmektedir (Alves ve ark., 2013). Sonuçlarımızla benzer şekilde DM'nin rat testis dokularında kolesterol, esterleşmemiş yağ asitleri, trigliseritler ve fosfolipitleri artırdığı da bildirilmiştir (Ma ve ark., 2023). Her ne kadar DM'nin erkek üreme sağlığını reaktif oksijen türlerinin aşırı üretimi üzerinden bozsa da, testisi hangi mekanizmalarla etkilediği tam olarak bilinmemektedir (Atta ve ark., 2017). Ayrıca DM'nin erkek üreme fonksiyonu üzerinde farklı düzeylerde önemli bir etkisi olduğu görüşü de vardır (Alves ve ark., 2013).

Tip 2 diyabetin patofizyolojisinde obezite ve dislipidemi gibi faktörlerin rol oynadığı bilinmektedir (Kahn ve ark., 2006). Bu nedenle, bu değişikliklerin geri çevrilmesi, diyabetin ve onun komplikasyonlarının yönetimi ve tedavisinde etkili bir strateji olabilir (Sobczak ve ark., 2019). Mevcut araştırmamızda, biberiye yağının, diyabet grubuyla karşılaştırıldığında beyin ve testislerdeki TG ve TC düzeylerini azalttığı bulunmuştur. Çalışmamızda benzer şekilde STZ ile indüklenen diyabetik sıçan modelinde, biberiye sıcak su ekstresi günde 0,55 g/sıçan dozunda oral olarak verilmiş ve uygulamayı takiben kan şekeri, kolesterol, TG, LDL'de önemli bir düşüş ve HDL'de artış gözlenmiştir (Abu-Al-Basal 2010). Ayrıca STZ ile diyabet oluşturulan farelerde biberiye uygulamasının plazma glikoz, TG, TC ve LDL-C seviyelerini düşürdüğü, HDL-C'yi ise arttırdığı bildirilmiştir (Al-Jamal ve Alqadi 2011). Yukarıdaki bulgulara benzer şekilde, biberiyenin sulu ekstresi (200 mg/kg, STZ uygulamasından 2 hafta önce ve 3 hafta sonra) STZ-diyabetik sıçanlarda TC, LDL, TG ve HDL plazma seviyelerini iyileştirmiştir (Alnahdi 2012). Biberiyenin her iki dokuda da göstermiş olduğu hipolipidemik etki kısmen hem bağırsak emiliminin hem de lipid biyosentezinin baskılanmasıyla ilişkili olabileceği düşünülmüştür. Biberiye ve aktif bileşikleri lipid sentezinin yanı sıra TG sentezinden sorumlu ana enzimler olan DGAT1 ve DGAT2 üzerinden lipolizi de modüle edebilir (Cui ve ark., 2012). Biberiye ve aktif bileşikleri ayrıca Faz 2 enzimlerinin indüksiyonu yoluyla dislipidemiyi iyileştirebilir (Takahashi ve ark., 2009). Fakat hayvan ve *in vitro* deneylerden elde edilen birçok kanıta rağmen, biberiyenin insanlardaki etkinliğini ve güvenliğini doğrulamak için daha fazla klinik çalışmaya ihtiyaç vardır.

SONUÇ

Sonuç olarak, bu çalışma, biberiye yağının, streptozotosin ile oluşturulan diyabetli sıçan modelinde beyin ve testis dokularındaki trigliserit (TG) ve toplam kolesterol (TC) seviyelerini azalttığını göstermektedir. Diyabetin lipid profilleri üzerindeki olumsuz etkileri, biberiye yağı uygulaması ile tersine çevrilebilmektedir. Bu bulgular, biberiye yağının diyabetle ilişkili lipid değişiklikleri üzerinde olumlu etkilere sahip olabileceğini göstermektedir. Ancak, biberiye yağının benzer etkileri ve güvenlik profillerini insanlarda gösterip göstermediğini doğrulamak için daha fazla klinik araştırmaya ihtiyaç vardır. Yine de bu çalışma, diyabetin bu dokulardaki lipid metabolizmasını nasıl etkilediğini anlamamıza ve hastalığın potansiyel komplikasyonlarını hafifletmek için doğal bir yaklaşımın potansiyelini değerlendirmemize katkı sağlamaktadır.

KAYNAKLAR

Abdelrahman N, El-Banna R, Arafa MM, Hady MM 2020. Hypoglycemic efficacy of Rosmarinus officinalis and/or Ocimum basilicum leaves powder as a promising clinico-nutritional management tool for diabetes mellitus in Rottweiler dogs. Veterinary World, 13(1): 73.

Abu-Al-Basal MA 2010. Healing potential of Rosmarinus officinalis L. on full-thickness excision cutaneous wounds in alloxan-induced-diabetic BALB/c mice. Journal of ethnopharmacology, 131(2): 443-450.

Al-Jamal A-R, Alqadi T 2011. Effects of rosemary (Rosmarinus officinalis) on lipid profile of diabetic rats. Jordan J Biol Sci, 4(4): 199-204.

Alnahdi HS 2012. Effect of Rosmarinus officinalis extract on some cardiac enzymes of streptozotocin-induced diabetic rats. Journal of Health Sciences, 2(4): 33-37.

Alves MG, Martins AD, Cavaco JE, Socorro S, Oliveira PF 2013. Diabetes, insulin-mediated glucose metabolism and Sertoli/blood-testis barrier function. Tissue Barriers, 1(2): e23992.

Alves MG, Martins AD, Rato L, Moreira PI, Socorro S, Oliveira PF 2013. Molecular mechanisms beyond glucose transport in diabetes-related male infertility. Biochimica et Biophysica Acta (BBA) - Molecular Basis of Disease, 1832(5): 626-635.

Atta MS, Almadaly EA, El-Far AH, Saleh RM, Assar DH, Al Jaouni SK, Mousa SA 2017. Thymoquinone Defeats Diabetes-Induced Testicular Damage in Rats Targeting Antioxidant, Inflammatory and Aromatase Expression. International Journal of Molecular Sciences, 18(5): 919.

Battisti WP, Palmisano J, Keane WF 2003. Dyslipidemia in patients with type 2 diabetes. Relationships between lipids, kidney disease and cardiovascular disease.

Bhatti JS, Sehrawat A, Mishra J, Sidhu IS, Navik U, Khullar N, Kumar S, Bhatti GK, Reddy PH 2022. Oxidative stress in the pathophysiology of type 2 diabetes and related complications: Current therapeutics strategies and future perspectives. Free Radic Biol Med, 184(114-134.

Bopanna K, Kannan J, Sushma G, Balaraman R, Rathod S 1997. Antidiabetic and antihyperlipaemic effects of neem seed kernel powder on alloxan diabetic rabbits. Indian journal of Pharmacology, 29(3): 162.

Constantinou C, Karavia EA, Xepapadaki E, Petropoulou P-I, Papakosta E, Karavyraki M, Zvintzou E, Theodoropoulos V, Filou S, Hatziri A 2016. Advances in high-density lipoprotein physiology: surprises, overturns, and promises. American Journal of Physiology-Endocrinology and Metabolism.

Cui L, Kim MO, Seo JH, Kim IS, Kim NY, Lee SH, Park J, Kim J, Lee HS 2012. Abietane diterpenoids of Rosmarinus officinalis and their diacylglycerol acyltransferase-inhibitory activity. Food chemistry, 132(4): 1775-1780.

Doucet J, Le Floch J-P, Bauduceau B, Verny C 2012. GERODIAB: Glycaemic control and 5-year morbidity/mortality of type 2 diabetic patients aged 70 years and older: 1. Description of the population at inclusion. Diabetes & metabolism, 38(6): 523-530.

Eid S, Sas KM, Abcouwer SF, Feldman EL, Gardner TW, Pennathur S, Fort PE 2019. New insights into the mechanisms of diabetic complications: role of lipids and lipid metabolism. Diabetologia, 62(9): 1539-1549.

Farmer JA 2008. Diabetic dyslipidemia and atherosclerosis: evidence from clinical trials. Current diabetes reports, 8(1): 71-77.

Filippatos T, Tsimihodimos V, Pappa E, Elisaf M 2017. Pathophysiology of Diabetic Dyslipidaemia. Current vascular pharmacology, 15(6): 566-575.

Galicia-Garcia U, Benito-Vicente A, Jebari S, Larrea-Sebal A, Siddiqi H, Uribe KB, Ostolaza H, Martín C 2020. Pathophysiology of Type 2 Diabetes Mellitus. International Journal of Molecular Sciences, 21(17): 6275.

Isomaa B, Almgren P, Tuomi T, Forsen B, Lahti K, Nissen M, Taskinen M-R, Groop L 2001. Cardiovascular morbidity and mortality associated with the metabolic syndrome. Diabetes Care, 24(4): 683-689.

Kahn SE, Hull RL, Utzschneider KM 2006. Mechanisms linking obesity to insulin resistance and type 2 diabetes. Nature, 444(7121): 840-846.

Kontogianni VG, Tomic G, Nikolic I, Nerantzaki AA, Sayyad N, Stosic-Grujicic S, Stojanovic I, Gerothanassis IP, Tzakos AG 2013. Phytochemical profile of Rosmarinus officinalis and Salvia officinalis extracts and correlation to their antioxidant and anti-proliferative activity. Food chemistry, 136(1): 120-129.

Li Y, Chen M, Xuan H, Hu F 2012. Effects of Encapsulated Propolis on Blood Glycemic Control, Lipid Metabolism, and Insulin Resistance in Type 2 Diabetes Mellitus Rats. Evidence-Based Complementary and Alternative Medicine, 2012(981896.

Ma X, Ren X, Zhang X, Griffin N, Liu H, Wang L 2023. Rutin ameliorates perfluorooctanoic acid-induced testicular injury in mice by reducing oxidative stress and improving lipid metabolism. Drug and Chemical Toxicology, 46(6): 1223-1234.

Mainzen Prince PS, Kumar MR, Selvakumari CJ 2011. Effects of gallic acid on brain lipid peroxide and lipid metabolism in streptozotocin-induced diabetic Wistar rats. Journal of Biochemical and Molecular Toxicology, 25(2): 101-107.

Navab M, Reddy ST, Van Lenten BJ, Fogelman AM 2011. HDL and cardiovascular disease: atherogenic and atheroprotective mechanisms. Nature Reviews Cardiology, 8(4): 222-232.

Padhi S, Nayak AK, Behera A 2020. Type II diabetes mellitus: a review on recent drug based therapeutics. Biomedicine & Pharmacotherapy, 131(110708.

Quirarte Báez SM, Zamora Perez AL, Reyes Estrada CA, Gutiérrez Hernández R, Sosa Macías M, Galaviz Hernández C, Guerrero Manriquez GG, Lazaldo Ramos BP 2019. A shortened treatment with rosemary tea (Rosmarinus officinalis) instead of glucose in patients with diabetes mellitus type 2 (TSD). Journal of Population Therapeutics& Clinical Pharmacology, 26(4):e18–e28

Ramadan KS, Khalil OA, Danial EN, Alnahdi HS, Ayaz NO 2013. Hypoglycemic and hepatoprotective activity of Rosmarinus officinalis extract in diabetic rats. Journal of physiology and biochemistry, 69(779-783.

Roden M, Shulman GI 2019. The integrative biology of type 2 diabetes. Nature, 576(7785): 51-60.

Selmi S, Rtibi K, Grami D, Sebai H, Marzouki L 2017. Rosemary (Rosmarinus officinalis) essential oil components exhibit anti-hyperglycemic, anti-hyperlipidemic and antioxidant effects in experimental diabetes. Pathophysiology, 24(4): 297-303.

Sobczak A, A. Blindauer C, J. Stewart A 2019. Changes in plasma free fatty acids associated with type-2 diabetes. Nutrients, 11(9): 2022.

Takahashi T, Tabuchi T, Tamaki Y, Kosaka K, Takikawa Y, Satoh T 2009. Carnosic acid and carnosol inhibit adipocyte differentiation in mouse 3T3-L1 cells through induction of phase2 enzymes and activation of glutathione metabolism. Biochemical and biophysical research communications, 382(3): 549-554.

Taskinen M-R 2003. Diabetic dyslipidaemia: from basic research to clinical practice. Diabetologia, 46: 733-749.

Taskinen MR 2003. Diabetic dyslipidaemia: from basic research to clinical practice*. Diabetologia, 46(6): 733-749.

Turner R, Millns H, Neil H, Stratton I, Manley S, Matthews D, Holman R 1998. Risk factors for coronary artery disease in non-insulin dependent diabetes mellitus: United Kingdom Prospective Diabetes Study (UKPDS: 23). Bmj, 316(7134): 823-828.

Vergès B 2015. Pathophysiology of diabetic dyslipidaemia: where are we? Diabetologia, 58(5): 886-899.

Xepapadaki E, Nikdima I, Sagiadinou EC, Zvintzou E, Kypreos KE 2021. HDL and type 2 diabetes: the chicken or the egg? Diabetologia, 64(9): 1917-1926.

Yesilbag D, Eren M, Agel H, Kovanlikaya A, Balci F 2011. Effects of dietary rosemary, rosemary volatile oil and vitamin E on broiler performance, meat quality and serum SOD activity. British Poultry Science, 52(4): 472-482.

Zaccardi F, Webb DR, Yates T, Davies MJ 2016. Pathophysiology of type 1 and type 2 diabetes mellitus: a 90-year perspective. Postgraduate medical journal, 92(1084): 63-69.

DRAFT

ORAL PRESENTATION

Sağlıkta mesleki iyonlaştırıcı radyasyona maruz kalma ile tiroid bezi

Adem Keskin[1*] (*ORCID: https://orcid.org/0000-0003-1921-2583*)

[*1] Aydın Adnan Menderes Üniversitesi, Sağlık Bilimleri Enstitüsü, Biyokimya (Tıp) Bölümü, Aydın, Türkiye.

*Sorumlu yazar e-mail: ademkeskin78@gmail.com

Özet

Sağlık çalışanları hem teşhis hem de tedavi amacıyla iyonlaştırıcı radyasyona maruz kalabilmektedir. Ayrıca insan vücudundaki radyosensitif organlardan biri de tiroid bezidir. Bu çalışmada mesleki radyasyona maruz kalan sağlık çalışanlarında tiroid yapı ve fonksiyonlarında gözlenen değişikliklerin incelenmesi amaçlandı. Bu çalışmada öncelikle iyonlaştırıcı radyasyonun tıpta kullanıldığı alanlara değinilmiştir. Ayrıca iyonlaştırıcı radyasyon ile tiroid fonksiyonu ve yapısı arasındaki ilişki sağlık profesyonellerinin katıldığı çalışmalarda incelenmiştir. Radyolojik görüntüleme prosedürleri, iyonlaştırıcı radyasyona maruz kalmanın önde gelen uygulamaları arasındadır. Radyosensitif tümörlerin tedavi seçeneklerinden biri olan radyoterapi, tıpta iyonlaştırıcı radyasyonun kullanıldığı alanlardan biridir. İyonlaştırıcı radyasyonun kullanıldığı bir diğer tıp alanı olan nükleer tıp, radyoaktif maddelerin kullanıldığı en büyük teranostik alandır. İyonlaştırıcı radyasyona maruz kalan sağlık çalışanlarında hipotiroidizm riski artmaktadır. Ek olarak, bu maruziyet tiroid hormonları üzerindeki etkiyle ve tiroid bağışıklık hastalıklarının gelişme olasılığının artmasıyla ilişkilendirilmiştir. Ek olarak, kadın radyasyon sağlık çalışanları, erkek meslektaşlarına göre daha yüksek düzeyde anormal tiroid nodülleri, triiyodotironin ve tiroid uyarıcı hormon düzeylerine sahiptir. Ayrıca bu meslek grubundaki kişilerde yaşla birlikte anormal tiroid nodülleri, triiyodotironin ve tiroksin düzeyleri de artmaktadır. Öte yandan iyonlaştırıcı radyasyona maruz kalmanın bu sağlık çalışanlarında tiroid kanseri riski ile ilişkilendirilmemiştir. Sonuç olarak iyonlaştırıcı radyasyona mesleki maruz kalma, radyosensitif bir organ olan tiroid bezinde hasara neden olabilir. Tiroid organının fonksiyonlarında ve yapısında sağlık sorunlarının önlenmesi için mesleki olarak iyonlaştırıcı radyasyona maruz kalanların kişisel korunma yöntemlerini mümkün olduğunca titizlikle uygulamaları gerekmektedir.

Anahtar Kelimeler: Radyasyon, Tiroid, Triiyodotironin, Tiroksin, Sağlık çalışanı, Hipotiroidizm

Thyroid gland with occupational exposure to ionizing radiation in health

Abstract

Healthcare workers may be exposed to ionizing radiation for both diagnosis and treatment purposes. In addition, one of the radiosensitive organs in the human body is the thyroid gland. This study aimed to examine the changes observed in thyroid structure and functions in healthcare workers exposed to occupational radiation. In this study, firstly, the areas where ionizing radiation is used in medicine are mentioned. Additionally, the relationship between ionizing radiation and thyroid function and structure has been examined in studies involving health professionals. Radiological imaging procedures are among the leading applications of exposure to ionizing radiation. Radiotherapy, one of the treatment options for radiosensitive tumors, is one of the areas in medicine where ionizing radiation is used. Nuclear medicine, another field of medicine in which ionizing radiation is used, is the largest theranostic field in which radioactive substances are used. The risk of hypothyroidism increases in healthcare workers exposed to ionizing radiation. Additionally, this exposure has been associated with an effect on thyroid hormones and an increased likelihood of developing thyroid immune diseases. Additionally, female radiation healthcare workers have higher levels of abnormal thyroid nodules, triiodothyronine, and thyroid-stimulating hormone than their male counterparts. In addition, abnormal thyroid nodules, triiodothyronine and thyroxine levels increase with age in people

in this occupational group. On the other hand, exposure to ionizing radiation was not associated with the risk of thyroid cancer in these healthcare workers. As a result, occupational exposure to ionizing radiation can cause damage to the thyroid gland, which is a radiosensitive organ. In order to prevent health problems in the functions and structure of the thyroid organ, those who are occupationally exposed to ionizing radiation should apply personal protection methods as meticulously as possible.
Keywords: Radiation, Thyroid, Triiodothyronine, Thyroxine, Healthcare professional, Hypothyroidism

GİRİŞ

İyonlaştırıcı radyasyon, içinden geçtiği ortamın atomlarını iyonlaştırabilen malzeme parçacıklarının veya elektromanyetik radyasyonun fotonlarının akışını ifade eder. Hücresel düzeyde hasar, hücre ölümü veya sitogenetik bilgilerinin değişmesiyle ortaya çıkabilir. Bu olaylar, belirtilerin doz eşiğinin aşılmasına bağlı olduğu olumsuz doku reaksiyonlarına (deterministik etkiler) veya hasar olasılığının dozla birlikte arttığı sitokastik etkilere yol açabilir (Havránková 2020). İnsan vücudu üzerindeki en kötü sitolojik etkisi hücre ölümü olan iyonlaştırıcı radyasyon, hızlı teknik gelişmelerle birlikte sıradan yaşamda daha geniş bir uygulama alanında varolmuştur (Jiao ve ark., 2022). Tıpta hem tanı hem de tedavi açısından önemli bir uygulama alanı olan iyonize radyasyon, önemli ilerlemelere katkıda bulunmuş olsa da, risksiz de değildir. Bu risk, 1982'den bu yana hastaların tıbbi radyasyona maruz kalma oranındaki 3 kat artışı içermektedir; Bazı potansiyel riskler bilinmesine rağmen, bu risklerin nicelikselleştirilmesine yönelik zorluklar devam etmektedir (Chambers, 2017).

İyonlaştırıcı radyasyona maruz kalmanın deneysel modelleri ve bunların canlı organizmalar üzerinde hasar oluşturma mekanizmalarıyla ilgili tıbbi literatürdeki mevcut kanıtlar, deoksi ribonükleik asit (DNA) çift sarmalının bir veya her iki ipliğinin "sitokastik kırılması" teorisine dayanmaktadır. Bu teoriye göre, yüksek doz iyonlaştırıcı radyasyon, her iki DNA zincirine de zarar vererek hücre için potansiyel olarak ölümcül olan kırılmalara neden olabilirken, düşük doz iyonlaştırıcı radyasyon, esas olarak tek iplikçiklerde kolayca onarılabilen ve kalıcı bir hasara yol açmayan kırılmalara neden olabilir. Diğer bir taraftan düşük dozda iyonlaştırıcı radyasyon maruz kalmanın, hem maruz kalan bireylerde hem de sonraki nesillerde yıllar veya on yıllar sonra bile kanserojen etkilere sahip olduğu göstermektedir. Ek olarak, düşük dozlara maruz kaldıktan sonra hayatta kalan hücreler, görünüşte normal olmalarına rağmen, moleküler sinyaller ve komşu veya uzak hücreleri içeren karmaşık doku alışverişi nedeniyle doğrudan ışınlanmayan hücrelerde bile bulunabilen klonal olmayan kromozomal anormallikler gibi kendi nesillerinde belirgin hale gelen hasarları biriktirir (Burgio ve ark., 2018). Epidemiyolojik bir çalışmada radyasyonun neden olduğu genomik istikrarsızlık kanser insidansı ile ilişkilendirilmiştir (Eidemüller ve ark., 2009). Streffer (2010), karsinojenezde genomik istikrarsızlığın rolüne ilişkin kanıtları gözden geçirmekte ve artan radyosensitiviteye yönelik genetik yatkınlığın, artan genomik istikrarsızlık ve kanser yatkınlığı ile ilişkili olduğu sonucuna varmaktadır.

Tiroid bezi en büyük endokrin organdır ve boynun ön kısmında bulunur. Büyümeyi, metabolizmayı ve insan vücudunun diğer önemli fizyolojik süreçlerini düzenleyen tiroid hormonları, tiroksin (T4) ve triiyodotironin (T3)'i sentezlemek için iyotu içerir ve kullanır (Braverman ve Cooper, 2012). Tiroid bezi insan vücudunda radyasyona diğer organlara göre daha duyarlı (radyosensitif) olan organlardan biridir. Yüksek dozda ışınlamayı takiben, hipotiroidizm ile kendini gösteren önemli radyasyon etkileri gözlemlenebilmektedir. Hipotiroidizmden farklı olarak neoplazi, insanlarda tiroide radyasyonun neden olduğu iki önemli olumsuz sonuçtan biridir (Virgili, 2020). Diğer bir taraftan orta veya düşük dozda ışınlamanın hipotiroidizm ve otoimmün tiroidit üzerinde geçici bir etkisi olabildiği ve eğer varsa bu etkinin geri dönüşümlü olduğu ifade edilmektedir (Nagayama, 2018).

İyonlaştırıcı radyasyonun etkisi, hücre hasarı, hücre ölümü veya sitogenetik bilgilerinin değişmesiyle ortaya çıkabilir. Buna karşılık tıpta hem tanı hem de tedavi için gerek hastalar gerek sağlık çalışanları iyonize radyasyona maruz kalabilmektedir. En büyük endokrin bez olan tiroid bezi, vücudumuzda ki radyosensitif olan organlardan biridir. Bu çalışmada, düşük dozlarda iyonlaştırıcı radyasyona uzun süreli maruz kalan sağlık çalışanlarının tiroit bezi yapısında ve fonksiyonunda gözlenen değişikliklerin incelenmesi amaçlandı.

MATERYAL VE METOT

Düşük dozlarda iyonlaştırıcı radyasyona uzun süreli maruz kalan sağlık çalışanlarının tiroit bezi yapısında ve fonksiyonunda gözlenen değişikliklerin daha iyi anlaşılmasını sağlamak amacıyla PubMed, Scopus, Web of

Science ve Google Scholar veri tabanlarını kullanılarak literatür taraması yapıldı. Anahtar kelime olarak "radyasyon", "tiroid", "triiyodotironin", "tiroksin", "sağlık çalışanı" ve radyasyonun tiroit bezinde oluşturduğu en çok gözlenen sağlık sorunu "hipotiroidizm" kelimeleri kullanıldı. Alıntılama sayıları ve yayın tarihleri dikkate alınarak 30 makale seçildi.

Literatür taramasında öncelikle iyonlaştırıcı radyasyonun tıpta kullanıldıkları alanlara değinildi. Buna ek olarak iyonlaştırıcı radyasyon ve tiroid fonksiyon ve yapısı ile ilişkili bir başlık da oluşturuldu. Referans olarak değerlendirmeye alınan çalışmalarda özellikle sağlık çalışanları ile ilgili çalışmalara yer verildi.

İYONLAŞTIRICI RADYASYONUN TIPTA KULLANIM ALANLARI

İnsanların maruz kaldığı yapay radyasyon kaynaklarının çoğunluğunu tıbbi uygulamalar oluşturur (Tuncel, 2008). Bunların başında radyolojik görüntüleme prosedürleri gelmektedir. İyonlaştırıcı ışınları radyografi, floroskopi, anjiyografi ve bilgisayarlı tomografide (BT) kullanılır. İyonlaştırıcı radyasyon kullanan görüntüleme yöntemleri, radyoloji görüntüleme teknolojisindeki gelişmelerle birlikte çeşitli tıbbi alanlarda sıklıkla kullanılır (Divrik Gökçe ve ark., 2012). Son yıllarda çok dedektörlü BT gibi yüksek teknolojili cihazların geliştirilmesine paralel olarak BT anjiyografi, kardiyak BT ve sanal BT kolonoskopi gibi dinamik kontrastlı daha hızlı ve daha yüksek çözünürlüklü çalışmalar mümkün hale gelmiştir (Flohr ve ark., 2005). Bu durum, tanı yaklaşımlarını ve tedavi planlarını yeniden tasarlamak amacıyla klinisyenlerin radyolojik çalışmalara yönelik taleplerinin artmasına yol açmış ve bu da hastaların iyonlaştırıcı radyasyona maruz kalma oranında artışa yol açmıştır (Divrik Gökçe ve ark., 2012). İngiltere'de BT, 1990 yılında radyolojik uygulamaların %4'ünü oluştururken, 2000 yılında BT'nin radyolojik uygulamaların %10'unu oluşturduğu Amerikaya kıyasla, hastaların iyonlaştırıcı radyasyona maruz kalmasının büyük çoğunluğundan bu prosedürlerin sorumlu olduğu bulunmuştur. (UNSCEAR 2000). Kontrol edilebilir yapay radyasyon kaynaklarından alınan iyonize radyasyon miktarının giderek artması, yaşam boyu kansere yakalanma riskini doğurmakta ve dolayısıyla halk sağlığı açısından tehdit oluşturmaktadır (Hall ve Brenner 2008).

Tümörler normal dokulara göre radyasyon hasarına daha yatkındır (Minniti ve ark., 2012). Bundan dolayı radyoterapi en etkili kanser tedavilerinden biri olmaya devam etmektedir. Bununla birlikte, bireysel tümörlerin ve normal dokuların biyolojik özelliklerine göre tedaviyi mümkün kılan biyoloji odaklı kişiselleştirilmiş radyoterapi klinikte hala uygulanması gerekmektedir (Domina ve ark., 2018). İyonlaştırıcı radyasyonun sadece lokal etkisinin olmadığı, aynı zamanda birçok moleküler sinyal yolunu tetikleyerek sistemik etkilerinin de olduğu uzun yıllardan beri iyi bilinmektedir (Carvalho ve Villar, 2018). Kanser radyoterapisi sırasında her ne kadar lokal ışınlama uygulansa da, iyonize radyasyon, kan proteom ve metabolom düzeylerinde gözlemlenebilen tüm vücut tepkilerine neden olur. Radyaterapide ışınlanmış dokular radyasyonun dozundan ve hacminden etkilenirken kanın moleküler bileşimi, doz değerlendirmesi ve radyasyona sistemik tepkilerin tahmini ve izlenmesi için varsayımsal bir biyobelirteç kaynağıdır (Jelonek ve ark., 2017). Kanser hastaları için etkili bir geleneksel tedavi yöntemi olmaya devam eden radyoterapide klinik etkinlik, tedavi sırasında tümör hücrelerin radyorezistans gelişmesi nedeniyle tehlikeye girebilmektedir (Lu ve ark., 2022).

Nükleer tıp, biyolojik olayların görüntülenmesi için radyoaktif maddelere güvenen ve bu maddeleri alan dokulara iyonlaştırıcı radyasyon sağlayan en büyük teranostik kahramanıdır. Konsept, kişiselleştirilmiş tıbbın gelişmesiyle birlikte, hastalıkların kişiselleştirilmiş yönetimine izin verilmesi, hasta seçiminin iyileştirilmesi, yanıtların daha iyi tahmin edilmesi, toksisitenin azaltılması ve prognozun tahmin edilmesiyle önem kazanmıştır (Nunes ve ark., 2021). Standart radyografik incelemelerde, 0,01-10 mSv arasında değişen ortalama etkili dozlara sahip iken, çoğu nükleer tıp prosedürü için ortalama etkili doz 0,3 ile 20 mSv arasında değişmektedir (Mettler ve ark., 2008). İyonlaştırıcı radyasyonun tıpta kullanım alanlarından bir başkası olan girişimsel radyoloji çalışanlarının en yüksek dozlarda iyonlaştırıcı radyasyona maruz kalması ile birlikte nükleer tıp çalışanlarının iyonlaştırıcı radyasyona maruziyetinin izlenmesi göz ardı edilemez bir gerçektir (Liu ve ark. 2022).

İYONLAŞTIRICI RADYASYON VE TİROİD

Radyasyon esas olarak ışınlanmış dokulardaki molekülleri iyonize ederek DNA hasarına neden olur. Radyasyon enerjisi ya DNA, organeller ve membranlar gibi kritik hücresel yapılarda doğrudan emilir ya da sulu sitozolde oldukça reaktif serbest radikallerin indüksiyonu yoluyla dolaylı olarak emilir. Moleküler oksijenin varlığında geçici serbest radikallerin moleküler hasar oluşturma olasılığı artar, bu da biyokimyasal hasarın stabilizasyonuna yardımcı olur (Minniti ve ark., 2012). Radyasyonun neden olduğu hasar, apoptoz,

hücre döngüsünün durması, DNA onarımı ve kanserle sonuçlanabilecek, birbirine bağlı sinyal yollarından oluşan karmaşık bir ağdır. Nükleer felaketlerden kemoterapiye kadar radyasyona yanıt olarak tiroid kanserinin gelişimi uzun süredir bir araştırma konusu olmuştur. Tiroid bezinin duyarlılığının radyasyon ve kanser gelişimi üzerindeki iyonlaştırıcı ve iyonlaştırıcı olmayan radyasyonun etkilerine ilişkin temel bir genel bakış sağlanmıştır (Albi ve ark., 2017). İyonlaştırıcı radyasyon, papiller tiroid kanseri için iyi bilinen bir risk faktörüdür ve tiroid karsinogenezi için önemli olan mikroRNA ekspresyonunu serbest bıraktığı rapor edilmiştir. İyonlaştırıcı radyasyonun mikroRNA ekspresyonunu serbest bıraktığını, çift sarmallı DNA'yı etkileyerek ışınlanmış tiroid hücrelerinin onarım etkinliğini bozduğunu göstermektedir (Penha ve ark., 2018).

Cioffi ve arkadaşları (2020) sağlık çalışanlarında düşük doz iyonize radyasyona maruz kalma ile tiroid fonksiyonel değişiklik üzerine bir çalışma yapmışlardır. Cioffi ve arkadaşlarının (2020) yaptıkları çalışmada, düşük dozda iyonize radyasyona maruz kalmanın serbest triiyodotironin, serbest tiroksin ve tiroid uyarıcı hormon (TSH) düzeylerini önemli ölçüde etkilediği sonucuna ulaşmışlardır. Buna ek olarak iyonlaştırıcı radyasyona maruz kalma kategorisi ile herhangi bir ilişki belirlenemese de sağlık çalışanlarında hipotiroidizm riskinin olası bir artışını akla getirdiği belirtilmektedir (Cioffi ve ark., 2020). Luna-Sánchez ve arkadaşlarının 2019 yılında yaptıkları çalışmada da, benzer bir sonuç olarak, radyasyon dozlarına bağlı olarak subklinik hipotiroidizm riskinin arttığını gözlemişlerdir (Luna-Sánchez ve ark., 2019). Mesleki olarak iyonlaştırıcı radyasyona maruz kalan sağlık personelleri ile yapılan yakın tarihli bir çalışmada, düşük dozlarda mesleki iyonlaştırıcı radyasyona maruz kalmanın tiroid hormonlarını etkilediği ve tiroid immün hastalıklarının gelişme olasılığının daha yüksek olmasıyla bağlantılı olduğu saptanmıştır (El-Benhawy ve ark., 2022). 1039 sağlık çalışanı ile yapılan başka bir çalışmada, iyonlaştırıcı radyasyonun yıllık kümülatif dozu, total T4 ve serbest T4 düzeyleri ile pozitif, total T3 ve TSH düzeyleri ile negatif korelasyon gösterdiği sonucuna ulaşılmıştır. Buna ek olarak yaş ve yıllık kümülatif doz arttıkça tiroid fonksiyon bozukluğu riski arttığı sonucuna varılmıştır (Lu ve ark., 2022).

Düşük doz iyonize radyasyona uzun süreli maruz kalma, tıbbi meslek mensuplarının tiroid hasarına neden olabilir; bu daha geniş bir endişe kaynağı olmalıdır (Tu ve ark., 2018). Örneklem büyüklüğü 4308 radyasyon çalışanı olan bir çalışmada, radyasyon sağlık çalışanlarının %14,3'ünde anormal tiroid bezi saptanmıştır. Ayrıca tiroid bezi anormalliğinin ana semptomu olan anormal tiroid nodülü radyasyon sağlık çalışanlarının %5,1'inde ve anormal tiroid uyarıcı hormon düzeyi radyasyon sağlık çalışanlarının %7,1'inde saptanmıştır. Kadın radyasyon sağlık çalışanlarında anormal tiroid nodülleri, T3 ve TSH düzeyleri, erkek radyasyon sağlık çalışanlarına göre daha yüksek bulunmuştur. Ayrıca çalışma yaşı arttıkça anormal tiroid nodülleri, T3 ve T4 düzeylerinin arttığı belirtilmiştir. Radyasyon sağlık çalışanlarında iyonlaştırıcı radyasyonun tiroid hasarına neden olabileceği sonucuna varılmıştır. Bu nedenle radyasyon çalışanlarının sağlığını korumak için radyasyondan korunma yönetimine daha fazla önem verilmelidir (Yang ve ark., 2022). Sağlık çalışanlarının 37 yıllık kişisel dozimetrelerinin değerlendirildiği bir çalışmada tiroid bezine uzun süreli, düşük ila orta dozda iyonize radyasyona maruz kalma, tiroid kanseri riskinde artışla ilişkili olmadığı belirtilmiştir (Kitahara ve ark., 2018).

SONUÇ

Sonuç olarak iyonlaştırıcı radyasyonun düşük dozlarda uzun süreli maruz kalmanın radyosensitif bir organ olan tiroit bezinin fonksiyonlarında ve yapısında önemli bir değişikliğe neden olabilmektedir. İnsan vücudunda en büyük endokrin bez olan tiroit organının fonksiyonlarında ve yapısında sağlık açısından bir sorun oluşmaması amacıyla iyonlaştırıcı radyasyona maruz kalan sağlık çalışanlarının kişisel korunma yöntemlerini mümkün oldukça titizlikle uygulamalıdır. Buna ek olarak radyasyonlu ortamlarda az süre kalınmalıdır.

KAYNAKLAR

Albi E, Cataldi S, Lazzarini A, Codini M, Beccari T, Ambesi-Impiombato FS, Curcio F. 2017. Radiation and Thyroid Cancer. Int J Mol Sci. 18(5): 911.

Braverman LE, Cooper D. 2012. Werner & Ingbar's the thyroid: a fundamental and clinical text. Lippincott Williams & Wilkins.

Burgio E, Piscitelli P, Migliore L. 2018. Ionizing Radiation and Human Health: Reviewing Models of Exposure and Mechanisms of Cellular Damage. An Epigenetic Perspective. Int J Environ Res Public Health. 15(9): 1971.

Carvalho HA, Villar RC. 2018. Radiotherapy and immune response: the systemic effects of a local treatment. Clinics (Sao Paulo). 73(suppl 1): e557s.

Chambers CE. 2017. Health Risks of Ionizing Radiation: Dr Roentgen Today. Circulation. 136(25): 2417-2419.

Cioffi DL, Fontana L, Leso V, Dolce P, Vitale R, Vetrani I, Galdi A, Iavicoli I. 2020. Low dose ionizing radiation exposure and risk of thyroid functional alterations in healthcare workers. Eur J Radiol., 132: 109279.

Divrik Gökçe S, Gökçe E, Coşkun M. 2012 Radiology residents' awareness about ionizing radiation doses in imaging studies and their cancer risk during radiological examinations. Korean J Radiol. 13(2): 202-209.

Domina EA, Philchenkov A, Dubrovska A. 2018. Individual Response to Ionizing Radiation and Personalized Radiotherapy. Crit Rev Oncog., 23(1-2): 69-92.

Eidemüller M, Holmberg E, Jacob P, Lundell M, Karlsson P. 2009. Breast cancer risk among Swedish hemangioma patients and possible consequences of radiation-induced genomic instability. Mutat Res., 669: 48-55

El-Benhawy SA, Fahmy EI, Mahdy SM, Khedr GH, Sarhan AS, Nafady MH, Yousef Selim YA, Salem TM, Abu-Samra N, El Khadry HA. 2022. Assessment of thyroid gland hormones and ultrasonographic abnormalities in medical staff occupationally exposed to ionizing radiation. BMC Endocr Disord., 22(1): 287.

Flohr TG, Schaller S, Stierstorfer K, Bruder H, Ohnesorge BM, Schoepf UJ. 2005. Multi-Detector Row CT Systems and Image-Reconstruction Techniques. Radiology., 235: 756–773.

Hall EJ, Brenner DJ. 2008. Cancer risks from diagnostic radiology. Br J Radiol., 81(965): 362-378.

Havránková R. 2020. Biological effects of ionizing radiation. Cas Lek Cesk. 159(7-8): 258-260.

Jelonek K, Pietrowska M, Widlak P. 2017. Systemic effects of ionizing radiation at the proteome and metabolome levels in the blood of cancer patients treated with radiotherapy: the influence of inflammation and radiation toxicity. Int J Radiat Biol., 93(7): 683-696.

Jiao Y, Cao F, Liu H. 2022. Radiation-induced Cell Death and Its Mechanisms. Health Phys. 123(5): 376-386.

Kitahara CM, Preston DL, Neta G, Little MP, Doody MM, Simon SL, Sigurdson AJ, Alexander BH, Linet MS. 2018. Occupational radiation exposure and thyroid cancer incidence in a cohort of U.S. radiologic technologists, 1983-2013. Int J Cancer. 143(9): 2145-2149.

Liu G, Zhang R, Li Y, Wu XQ, Niu LM, Liu YY, Zhang X. 2022. Study of Low-Dose Radiation Workers Ionizing Radiation Sensitivity Index and Radiation Dose-Effect Relationship. Health Phys. 2022 123(4): 332-339.

Lu Z, Zheng X, Ding C, Zou Z, Liang Y, Zhou Y, Li X. 2022. Deciphering the Biological Effects of Radiotherapy in Cancer Cells. Biomolecules. 12(9): 1167.

Lu BF, Yin WJ, Xu T, Li NN, Yi GL. 2022. Correlation analysis of low-dose X-ray ionizing radiation and thyroid function in radiation workers. Zhonghua Lao Dong Wei Sheng Zhi Ye Bing Za Zhi. 40(10): 733-736.

Luna-Sánchez S, Del Campo MT, Morán JV, Fernández IM, Checa FJS, de la Hoz RE. 2019 Thyroid Function in Health Care Workers Exposed to Ionizing Radiation. Health Phys., 117(4): 403-407.

Mettler FA Jr, Huda W, Yoshizumi TT, Mahesh M. 2008. Effective doses in radiology and diagnostic nuclear medicine: a catalog. Radiology. 2008 248(1): 254-263.

Minniti G, Goldsmith C, Brada M. 2012. Radiotherapy. Handb Clin Neurol., 104: 215-228. Nagayama Y. 2018. Radiation-related thyroid autoimmunity and dysfunction. J Radiat Res., 59(suppl_2): ii98-ii107.

Nunes RF, Zuppani RMF, Coutinho AM, Barbosa FG, Sapienza MT, Marin JFG, Buchpiguel CA. 2021. General Concepts in Theranostics. PET Clin. 16(3): 313-326.

Penha RCC, Pellecchia S, Pacelli R, Pinto LFR, Fusco A. 2018. Ionizing Radiation Deregulates the MicroRNA Expression Profile in Differentiated Thyroid Cells. Thyroid. 28(3): 407-421.

Streffer C. 2010. Strong association between cancer and genomic instability. Radiat Environ Biophys., 49: 125–131.

Tu L, Wang SL, Dong Q, Song HY, Li XT, Tan CP, Dong X. 2018. Effect of low-dose ionizing radiation exposure on thyroid function in a medical occupational population. Zhonghua Lao Dong Wei Sheng Zhi Ye Bing Za Zhi. 36(2): 91-94. Chinese.

Tuncel E. 2008. Klinik Radyoloji. Genişletilmiş 2. Baskı. Nobel&Güneş Tıp Kitabevleri. s. 3–105.

UNSCEAR 2000. Sources and Effects of Ionizing Radiation, United Nations Scientific Committee on the Effects of Atomic Radiation. New York: United Nations; 2000. UNSCEAR 2000 Report to the General Assembly.

Virgili M. 2020. The management of benign thyroid pathologies in medical radiation protection. Giornale Italiano di Medicina del Lavoro ed Ergonomia, 42(4): 281-291.

Yang Y, Wang Q, Yang LT, Zhao ZX. 2022. Investigate the thyroid function of radiation workers and analysis of influence factors. Zhonghua Lao Dong Wei Sheng Zhi Ye Bing Za Zhi., 40(2): 113-116.

DRAFT

ORAL PRESENTATION

Lise 9. sınıf öğrencilerine verilen ilk yardım ve KBRN eğitiminin öğrencilerin bilgi düzeylerine etkisi

Gonca Kaleli[1*] (ORCID:0000-0003-3203-905X), Sefa Ertürk[2] (ORCID: 0000-0003-4051-9096),

[1*]Niğde Ömer Halisdemir. Üni, Fen Bilimleri Enstitüsü, Afet Yönetimi, Yüksek Lisans Öğrencisi, Niğde, Türkiye

[2]Niğde Ömer Halisdemir. Üni. Tıp Fak. Temel Tıp Bil. Böl. Biyofizik Anabilim Dalı, Niğde, Türkiye

* Sorumlu yazar e-mail:kaleligonca4@gmail.com

Özet

Bireylerin günlük hayatta hastalık, kaza, yaralanma ve afetlerle karşılaşmaları durumunda, sakatlıkların/ ölümlerin önlenebilmesi veya azaltılabilmesi için temel ilk yardım ve KBRN konusunda bilgi sahibi olmaları gerekmektedir. Her bireyin bu durumlarla karşılama riski bulunmasına rağmen özellikle lise öğrencileri hem yaş hem de uygulama alanları nedeniyle daha risklidirler. Ayrıca bu öğrenciler, aldıkları eğitimleri sayesinde çalışma alanları veya afet gibi durumlarda ilk yardım ve KBRN ile ilgili müdahalelerde bulunacak kişilerdir. Bu çalışma daha önce ilk yardım kursu almamış 9. Sınıf lise öğrencilerinin İlk Yardım ve KBRN bilgi düzeylerinin değerlendirilmesi, verilen eğitimin bilgi düzeylerine etkisinin belirlenmesi amacıyla yapılmıştır. Çalışma, Mesleki ve Teknik Anadolu Lisesi (MTAL) 9. sınıf öğrencilerine ön test- son test şeklinde yarı deneysel olarak uygulanmıştır. Çalışma, eğitim öğretime devam eden, eğitim programına gönüllü olarak katılan, verilen 16 saatlik ilkyardım ve 2 saatlik KBRN eğitimi tamamlayan ve ön test-son test anketlerini dolduran 109 öğrenci ile tamamlanmıştır. Veriler, "Tanıtıcı Bilgi Formu", "İlk Yardım Eğitimi ve KBRN Eğitimi Bilgi Değerlendirme Formu kullanılarak toplanmıştır. SPSS 22.0 analiz paket programı kullanılarak analiz edilen araştırma verilerinin değerlendirilmesinde tanımlayıcı istatistikler kullanılmıştır. Öğrencilerin %72.5'i kız olup, yaş ortalaması 14.46±0.58'dir. Öğrencilere verilen eğitimin etkisini göstermek adına eğitim öncesi ve sonrası bazı değerleri karşılaştırmak gerekirse; Öğrencilerin ilk yardımla ilgili Soluk ve göğüs basısı sayısı sorusuna verdikleri doğru cevaplar; ön test %8.3, son test %81.7 ve 2 ay sonrası için ise %55'dir. KBRN için ulusal zehir merkezi telefon numarasını doğru yanıtlama oranları ise; ön test %18.3, son test %87.2 ve 2 ay sonrası için ise %52.3 olduğu tespit edilmiştir. Öğrencilere verilen 16 saatlik ilk yardım ve 2 saatlik KBRN eğitiminin bu konudaki farkındalıkları ve bilgi düzeyleri üzerinde olumlu etkileri olduğu görülmüştür. Ön test ve son test verileri değerlendirildiğinde, uygulanan eğitimin öğrencilerin bilgi düzeylerini anlamlı olarak artırdığı gözlenmiştir. İlk yardım ve KBRN eğitimin bu yaş grubunda yaygınlaştırılması ve buna uygun eğitim programlarının düzenlenmesi önerilmektedir.

Anahtar Kelimeler: Öğrenciler, eğitim, ilk yardım, KBRN, bilgi düzeyi

The effect of first aid and CBRN training given to high school 9th grade students on students' knowledge levels

Abstract

In case individuals encounter diseases, accidents, injuries and disasters in daily life, they need to have knowledge about basic first aid and CBRN in order to prevent or reduce disabilities / deaths. Although every individual has the risk of encountering these situations, especially high school students constitute more risky groups due to both age and application areas. In addition, these students are the people who will intervene in first aid and CBRN-related interventions in situations where large masses are affected, such as work areas or disasters, thanks to the training they receive. This study was conducted to evaluate the level of First Aid and CBRN knowledge of 9th grade high school students and to determine the effect of the training given on their level of knowledge. The study was applied quasi-experimentally to 9th grade students of Vocational and Technical Anatolian High School (MTAL) in the form of pre-test and post-test. The study was completed with 109 students who were continuing their education, voluntarily participated in the training programme, completed 16 hours of first aid and 2 hours of CBRN training and completed the pre-test and post-test questionnaires. Data were collected using the "Introductory Information Form", "First Aid Training and CBRN

Training Knowledge Assessment Form". Descriptive statistics were used in the evaluation of the research data analysed using SPSS 22.0 analysis package programe. 72.5% of the students were female and the mean age was 14.46±0.58 years.. The correct answers given by the students to the question of number of breaths and chest compressions related to the first aid were 8.3% in the pre-test, 81.7% in the post-test and 89% 2 months later. The rate of correctly answering the phone number of the national poison centre for CBRN was found to be 18.3% in the pre-test, 87.2% in the post-test and 52.3% 2 months later. It was evaluated that the 16-hour first aid and 2-hour CBRN training given to the students had positive effects on their awareness and knowledge levels on this subject. When the pre-test and post-test data were evaluated, it is thought that the applied training significantly increased the knowledge levels of the students. It is recommended that first aid and CBRN training should be extended in this age group and appropriate training programmes should be organised.
Keywords: Students, education, first aid, CBRN, knowledge level

GİRİŞ

DSÖ, sağlığı yalnızca hastalık ve sakatlığın olmayışı değil fiziksel sosyal refah durumu olarak tanımlamıştır. (DSÖ, 1986). Sağlıklı toplumlar, sağlıklı bireyler hedefine ulaşabilmek için bireylerin bilinçlendirilmesi gerekmektedir. Bireylerin bilinçlendirilmesinde sağlık eğitimi anahtar rol oynamaktadır. DSÖ tanımına göre sağlık eğitimi bilginin artırılması, birey ve toplum sağlığına yardımcı yaşam becerilerinin geliştirilmesi ve sağlık okuryazarlığını iyileştirmek için tasarlanmış bir iletişim şeklini içermektedir (Sağlığın teşviki ve geliştirilmesi sözlüğü, 2011). Günlük hayatımızda her an hastalık, kaza, yaralanma ve afetlerle karşılaşma ihtimalimiz vardır. Bu nedenle sağlık eğitimi içinde yer alan ilk yardım ve KBRN eğitiminin önemi her geçen gün önem kazanmaktadır. İlk yardımı tanımlamak gerekirse; herhangi bir kaza durumunda hayatı tehlikeye düşüren bir durumda, profesyonel yardım sağlanıncaya kadar, hayatın kurtarılması veya durumun kötüleşmesini engelleyecek, tıbbı araç gereç aranmaksızın, mevcut araç ve gereçlerle yapılan ilaçsız uygulamalardır (İnan, 2011). Kazalara bağlı ölümlerin en aza indirilmesi, sakatlıkların azaltılması ve önlenmesi toplumun temel ilk yardım konusunda bilgi sahibi olması ile mümkündür. Toplumu ilk yardım konusunda bilgilendirmenin en etkin ve en önemli yollarından biri, temel ilk yardım eğitiminin yaygınlaştırılmasıdır. Kazalar ve ani yaralanmalar beklenmedik anlarda gerçekleştiği için, yerinde ve zamanında yapılan basit ama etkili bir ilkyardımla, yaralının hayatını kurtarmak, sakat kalmasını önlemek veya sakatlık derecesini azaltmak mümkündür. Kazalar ve kazalara bağlı ölümlerin oranı ülkeler arasında farklılık göstermekle birlikte, ülkemizde kazalara bağlı ölüm oranlarının yüksek olmasının temel nedeni, kaza sonrası bilinçli ilkyardım hizmetlerinin olmamasıdır (Aytaç, 2010). Temel ilkyardım eğitimi almış kişilerin zamanında, doğru ve etkin yapacağı uygulamalarla kaza, yaralanma ve afet gibi durumlarda ölüm ve sakatlanma sayıları azalacaktır.

Bu konuda gerçekleştirilen çalışmalarla ilgili literatür incelendiğinde gelişim evrelerinden kaynaklanan dikkat eksikliğinin adolesan döneminde, kaza ve yaralanma olasılığını artıran faktörler arasında, geldiği görülmektedir. (Aylaz ve ark. 2009). Adolesan dönem, genel olarak birey ve onun çevresi açısından karmaşık, fırtınalı ve zor bir dönemdir (Güler ve Akın,2006). Genel olarak, 13-17 yaşlar arası bireyler kendini ölümsüz, kudretli, karşı konulamaz gördükleri için riskli davranışlarda bulunma eğilimi fazladır (Ercan, 2005). Bu dönem yapılan riskli davranışlar hastalık, ölüm ve sakatlıkların önemli bir bölümüne neden olmaktadır. (Feldman, 2000). Adolesan dönemde meydana gelen ölümlerin en başta gelen nedenlerinden olan kazalar ve intiharlar da riskli davranışlarla yakından ilişkilidir (Tenore ve Lipsky,2001). Kara ve arkadaşları tarafından yapılan çalışmada şiddete bağlı davranışlar sergileme, silah taşıma, tehdit etme-edilme, kavgaya karışma, yaralanma ve intihar oranları oldukça yüksek bulunmuştur (Kara ve ark., 2003). Yine Aras ve arkadaşları tarafından yapılan bir çalışmada bahsedilen riskli davranış oranları oldukça yüksek bulunmuştur (Aras ve ark., 2007). Amerika Birleşik Devletleri 2000 yılı istatistiklerine göre 10-15 yaş arası çocuklarda ve 15-19 yaş arası ve ayrıca 11-18 yaş grubu için spor, Avrupa ve Kuzey Amerika'da önemli yaralanma nedenleri arasındadır. Yaralanmaların; % 62'si organize spor sırasında, % 20'si beden eğitimi sınıflarında, %18'i organize olmayan denetimsiz spor sonucu meydana gelmektedir. Raporlar okulda bütün yaralanmaların dörtte birinin (1/4) ciddi olduğunu göstermektedir. Spor aktivitelerine bağlı ölümler nadir ancak Amerika'da 6 yıllık bir periyodda 15-19 yaş grubunu içeren ölümle ilişkili 30 spor aktivitesi belirlenmiştir (Abernethy ve ark., 2008). Ayrıca ergenler arasında, önde gelen ölüm nedenlerini temsil eden bir halk sağlığı sorunudur (Reveruzzi, Buckley, ve Sheehan, 2016)

KBRN (CBRN), Kimyasal Biyolojik Radyasyon ve Nükleer kelimelerinin baş harflerinden oluşmaktadır. Günümüzde yaşanan savaş, sanayi ve endüstri sahasında yaşanan gelişmeler KBRN risklerini artırmıştır.

Bütün bu riskler, geniş halk kitlelerin yaşamını olumsuz yönde etkiler. Ayrıca çok sayıda insanın yaşamını yitirmesine ve canlı hayatın sona ermesine/değişimine neden olur (MEGEP 2011). Bu nedenlerden dolayı KBRN artık dünya için büyük bir tehlike unsuru olmaktadır.
Özellikle yukarıda değinilen adolesan dönemdeki öğrenciler için yapılan çalışmalar tarandığında yaygın olarak ilkyardım eğitimi almamış bireylerin bilgi, tutum düzeylerinin tarandığı, farklı okullardaki öğrencilerin ilkyardım bilgi düzeylerinin karşılaştırıldığı, 2 saatlik ilk yardım farkındalık eğitimlerinin bilgi düzeylerine etkisinin değerlendirildiği görülmüştür. Adolesan dönemindeki öğrenciler kaza ve yaralanmalar açısından riskli dönemde oldukları için bu yaş grubuna verilecek eğitim ve akran desteği ile ilkyardım ve KBRN'de erken müdahalelerin sağlanmasının, sakatlık ve komplikasyonların gelişmesini önemli oranda azaltılacağı düşünülmektedir. Bu araştırma, Mesleki ve Teknik Anadolu Lisesi 9. Sınıf öğrencilerine verilen İlkyardım ve KBRN eğitiminin öğrencilerin bilgi düzeylerine etkisinin belirlenmesi amacıyla planlanmıştır.

MATERYAL VE METOT

Evren ve Örneklem

Bu araştırma, Niğde ili merkezinde bulunan Mesleki ve Teknik Eğitim veren Atatürk Mesleki ve Teknik Anadolu Lisesinde gerçekleştirilmiştir. Niğde ili genelinde 15 tane Mesleki ve Teknik Anadolu Lisesi mevcut olup bu okullardan iki tanesi eski adıyla sağlık meslek lisesidir. Yapılan kura sonucunda Atatürk Mesleki ve Teknik Anadolu Lisesi bu çalışma için seçilmiştir. Bu lisede 2021-2022 eğitim öğretim döneminde öğrenim görmekte olan toplam 417 öğrenciden 100 tane 9. Sınıf öğrencisi ve 2022-2023 eğitim öğretim döneminde öğrenim görmesi planlanan toplam 420 öğrenciden 100 tane 9. sınıf öğrencisi araştırmanın evrenini oluşturmaktadır. Örnekleme alınma kriterleri olarak; 2021-2023 eğitim öğretim dönemlerinde eğitim öğretime devam eden, eğitim programına gönüllü olarak katılan, verilecek olan 16 saatlik ilkyardım +2 saatlik KBRN eğitimi tamamlayan ve ön test-son test ve 2 ay sonraki son testleri dolduran öğrenciler olarak belirlenmiştir.

Yapılan bu çalışmada kaza ve yaralanmalarda risk grubu oluşturan adolesan dönemdeki 9. Sınıf öğrencilerine 16 saatlik kapsamlı ilk yardım eğitimi +2 saatlik KBRN tehdit ve ilk yardım eğitimi verilmiştir.

Veri Toplama Araçları

Veriler, literatür bilgisi doğrultusunda araştırmacı tarafından hazırlanan "Tanıtıcı Bilgi Formu","İlk Yardım Eğitimi Bilgi Değerlendirme Formu" ve KBRN Eğitimi Bilgi Değerlendirme Formu kullanılarak toplanmıştır. Tanıtıcı Bilgi Formu öğrencinin demografik özelliklerini içeren 4 soru, İlk Yardım Eğitimi Bilgi Değerlendirme formu temel ilk yardım bilgilerini içeren 33 soru, KBRN Eğitimi Bilgi Değerlendirme Formu temel düzeyde KBRN bilgilerini içeren 13 soru olmak üzere toplam 50 sorudan oluşmaktadır.

Yöntemin Uygulanması

Eğitim Atatürk Mesleki ve Teknik Anadolu Lisesi toplantı salonu ve sınıflarında düzenlenmiş olup 4+4+4+6 saat teorik ve uygulamalı olarak tamamlanmıştır. Eğitim esnasında akıllı tahta, Kızılay eğitim sunumları, yetişkin, çocuk, bebek maketleri, sargı bezleri, tespit malzemeleri, OED cihazı kullanılmıştır. 16 saatlik Temel İlk Yardım Eğitimi İçeriğinde Türk Kızılay'ı tarafından hazırlanan sunumlardan faydalanılarak materyaller hazırlanmıştır. İçerik olarak genel ilk yardım bilgileri, hasta ve yaralı ve olay yerinin değerlendirilmesi, temel yaşam desteği ve hava yolu tıkanıklığı, kanamalar ve şok, yaralanmalar, yanık sıcak –soğuk çarpması ve donmalar, kırık-çıkık ve burkulmalar, bilinç bozuklukları, zehirlenmeler-böcek sokmaları –hayvan ısırmaları, göz–kulak ve buruna yabancı cisim kaçması, boğulmalar, acil taşıma teknikleri anlatılmıştır. Ayrıca 2 saatlik KBRN eğitiminde temel düzeyde kimyasal, biyolojik ve nükleer tehditler ve bu tehditlere maruz kalındığında yapılması gerekenlerin neler olduğu konusunda bilgilendirme yapılmıştır. Verilerin ön testi eğitim öncesi, son testi ise eğitimden hemen sonra ve iki ay sonra olacak şeklide iki kez toplanmıştır.

Verilerin Değerlendirilmesi

Bu araştırmada veriler, bilgisayar ortamında SPSS 22.0 analiz paket programı kullanılarak analiz edilmiştir. Verilerin değerlendirilmesinde sayı, yüzdelik, aritmetik ortalama ve standart sapma gibi tanımlayıcı istatistiklerden yararlanılmıştır. Öğrencilerin ön test -son test bilgi düzeylerinin değerlendirilmesinde ise elde edilen veriler bağımlı gruplara uygulanan uygun analiz yöntemleri kullanılmıştır. Grupların karşılaştırılmasında sonuçların istatistiksel anlamlılığı $p<0.05$ düzeyinde değerlendirilmiştir.

Etik İzinler

Araştırmaya başlamadan önce Niğde Ömer Halisdemir Üniversitesi Etik Kurulu'nda 28.04.2022 tarihte ve 2022/05-48 etik numarası ile etik izin ve Niğde İl Milli Eğitim Müdürlüğünden 26.05.2022 tarihinde 50436403

sayılı yazılı izin alınmıştır. Ayrıca öğrencilere ve velilerine araştırma hakkında bilgi verilerek yazılı ve sözlü izinleri alınmıştır.

BULGULAR ve TARTIŞMA

Araştırmaya 119 öğrenci katılmış olup, katılan öğrencilerin %72.5'i kız , %27.5i erkek olup yaş ortalaması 14.46±0.58'dir. Öğrencilerin ilk değerlendirmede kontrol edileni bilme oranları ön testte %40.4 son testte %83.5 2 ay sonrası için %71.6 dır. Temel yaşam desteği sırasında uygulanan soluk ve göğüs basısı sayısını ön testte %8.3, son testte 81.7 iki ay sonraki son testte ise %55 oranında öğrenci doğru bilmiştir. Öğrencilerin ilk yardımla ilgili solunumun nasıl değerlendirildiğine verdikleri doğru cevaplar; ön test %57.8, son test %91.7 ve 2 ay sonrası için ise %89'dur. Temel yaşam desteğine ne zaman son verileceği sorusunu ise ön testte %72.5, son testte %92.7oranında öğrenci doğru yanıtlamış olup iki ay sonrası son testte oran değişmeyerek %92.7 olarak kalmıştır (Tablo 1).

Değişkenler			
Yaş (**X±SD)**	**14.46±0.58**		
		n	**%**
Cinsiyet	**Kız**	79	72.5
	Erkek	30	27.5
İlk yardım ve KBRN öğrenmenin gerekliliği	**Evet**	106	97.2
	Hayır	3	2.8

Tablo1: Öğrencilerin sosyodemografik özellikleri

Hava yolunda tam tıkanıklık olması durumunda yapılması gereken müdahaleyi ön testte %27.5, son testte %78.9 oranında, iki ay sonraki son testte ise %%57.8 oranında öğrenci doğru bilmiştir. Şok pozisyonunun nasıl verileceği sorusuna verilen doğru yanıtlar değerlendirildiğinde ön testte %45.9,son testte 75.2 iki ay sonraki son testte ise 60.6 oranında olduğu görülmüştür. Sıcak çarpmasında doğru ilkyardım uygulamasını ön testte %37.6 son testte %27.5 iki ay sonraki son testte %28.4oranında öğrencinin doğru yanıtlamıştır. KBRN için ulusal zehir merkezi telefon numarasını doğru yanıtlama oranları ise; ön test %18.3, son test %87.2 ve 2 ay sonrası için ise %52.3 olduğu tespit edilmiştir. Son olarak KBRN ikaz ve alarm işaretlerinden olmayan ikazı doğru bilen öğrenci sayısı ön testte %30.3şson testte %56.iki ay sonraki son testte %46.8 oranında olduğu görülmüştür (Tablo 2).

DRAFT

Tablo 2. Öğrencilerin İlk Yardım ve KBRN sorularına verdikleri yanıtlarının dağılımı

	Ön Test		Son Test		Son Test (2 Ay Sonra)	
Yanıtlama Durumları (n=109)	**Doğru**	**Yanlış**	**Doğru**	**Yanlış**	**Doğru**	**Yanlış**
İlk Yardım	**n(%)**	**n (%)**	**n (%)**	**n (%)**	**n (%)**	**n (%)**
İlk değerlendirmede ilk kontrol edilen	44(40.4)	65 (59.6)	91(83.5)	18(16.5)	78(71.6)	31(28.4)
Soluk ve göğüs basısı sayısı	9(8.3)	100(91.7)	89(81.7)	20(18.3)	60(55)	49(45)
Solunumun değerlendirilmesi	63(57.8)	46(42.2)	100(91.7)	9(8.3)	97(89)	12(11)
Ne zaman sonlandırılır	79(72.5)	30 (27.5)	101(92.7)	8(7.3)	101(92.7)	8(7.3)
Tam tıkanıklıkta yapılan müdahale	30(27.5)	79(72.5)	86(78.9)	23(21.1)	63(57.8)	46(42.2)
Uygun şok pozisyonu	50(45.9)	59(54.1)	82(75.2)	27(24.8)	66(60.6)	43(39.4)
Sıcak çarpmasında ilk yardım	41(37.6)	68(62.4)	30(27.5)	79(72.5)	31(28.4)	78(71.6)
KBRN						
Ulusal zehir merkezi telefon numarası	20(18.3)	89(81.7)	95(87.2)	14(12.8)	57(52.3)	52(47.7)
KBRN ikaz ve alarm işaretleri	33(30.3)	76(69.7)	62(56.9)	47(43.1)	51(46.8)	58(53.2)

Araştırma bulgularımız değerlendirildiğinde ön test, son test ve 2 ay sonrası verileri arasında istatistiksel açıdan anlamlı farkın olduğu gözlenmiştir.

SONUÇ

Sağlık eğitimi yoluyla kalıcı sakatlık ve yaralanmaların önlenmesi için 9. Sınıf öğrencilerinin İlkyardım ve KBRN konusundaki bilgilerin belirlenmesi önem taşımaktadır. Standart ilk yardım eğitimine ek olarak KBRN tehditleri ve KBRN tehditlerinde ilkyardım eğitimi ve bu eğitim öncesi ve sonrası gerçekleştirilecek ön test-son test uygulaması ilk kez bu çalışma ile ortaya konulmuştur. İstatistiksel analizler ile ön test son test sonuçları değerlendirildiğinde öğrencilerin İlkyardım ve KBRN konusundaki bilgi düzeylerinin belirlenmesi konusunda verilen eğitim neticesinde eğitim programına katılan öğrencilerde eğitim öncesi ve eğitim sonrası istatistiksel açıdan anlamlı farklılığın olduğu tespit edilmiştir. Bu nedenle ilk yardım ve KBRN eğitimin bu yaş grubunda yaygınlaştırılması ve buna uygun eğitim programlarının düzenlenmesi önerilmektir.

TEŞEKKÜR

Çalışmamıza katılan öğrencilerimize teşekkür ederiz.

KAYNAKLAR

Abernethy, L. MacAuley, D. , McNally, O. , McCann,S . , (2008), İmmediate Care of School Sport İnjury , *İnjury Prevention* ; **9** :270-273

Aras Ş, Günay T, Özan S, Orçın E., 2007. İzmir İlinde Lise Öğrencilerinin Riskli Davranışları, *Anadolu Psikiyatri Dergisi* ;**8**:186-196

Aylaz R, Gözüm S, Yılmaz U, Bakış E, Güneş G, Aylaz A. The Efficacy of The First Aid Training Program for Apprenticeship Students. İnönü Üniversitesi Tıp Fakültesi Dergisi, 2009, 16:89-94.

Aytaç Ş, 2010. Ankara-Akyurt İlçesinde Bir Mobilya İmalat Fabrikasında Çalışanlarda İlk Yardım Gerektiren Durumların Sıklığı ile İlk Yardım Eğitimi Öncesi Ve Sonrası Bilgi Düzeyleri, Gazi Üniversitesi Sağlık Bilimleri Enstitüsü, Yüksek Lisans Tezi.

Ercan O, 2005. Adolesanın Psikososyal Gelişimi, *İ.Ü. Cerrahpaşa Tıp Fakültesi Sürekli Tıp Eğitimi Etkinlikleri Adolesan Sağlığı Sempozyum Dizisi* No: **43**

Feldman, E. , (2000), Adolescent Health : Risks , Resilience , Prevention . *Clin Fam Pract* **2; 767**-790

Güler , Ç.,Akın , L., (2006), *Halk Sağlığı Temel Bilgiler* , Hacettepe Üniversitesi Yayınları , Ankara

İnan HF, 2011. Temel İlkyardım Uygulamaları Eğitim Kitabı. H. F. İNAN içinde, *Temel İlkyardım Uygulamaları Eğitim Kitabı* (s. 1-7). Ankara: T.C. Sağlık Bakanlığı

Kara B, Hatun Ş, Aydoğan M, Babaoğlu K, Gökalp SA, 2003. Kocaeli İlindeki Lise Öğrencilerinde Sağlık Açısından Riskli Davranışların Değerlendirilmesi, *Çocuk Sağlığı ve Hastalıkları Dergisi* **46**: 30-37

MEGEP, 2011. *Acil Sağlık Hizmetleri. Kimyasal biyolojik radyasyon ve nükleer (KBRN)tehlikelerde acil yardım*

Reveruzzi, B., Buckley, L., & Sheehan, M. (2016). School-Based First Aid Training Programs: A Systematic Review. The Journal of School Health, 86(4), 266–272. https://doi. org/10.1111/josh.12373

Sağlığın teşviki ve Geliştirilmesine Yönelik Ottawa Sözleşmesi. DSÖ, Cenevre,1986

Tenore J. L. , Lipsky , M.S., (2001) Preventive Services For The Adolescent (13 -20 years) *Pediatr Clin North Am* ; **48** ; 289 – 311

DRAFT

ORAL PRESENTATION

Sensory analysis of hamburger patties enriched with psyllium

Ahmet Murat Günal[1*] (https://orcid.org/0000-0001-9109-1080), Hande Öngün Yılmaz[2] (https://orcid.org/0000-0002-3497-567X), Murat Baş[3] (https://orcid.org/0000-0002-0494-301X)

[*1]İstanbul Okan University, Faculty of Health Sciences, Department of Nutrition and Dietetics, İstanbul, Türkiye.
[2]Bandırma Onyedi Eylül University, Faculty of Health Sciences, Department of Nutrition and Dietetics, Balıkesir, Türkiye.
[3]Acıbadem University, Faculty of Health Sciences, Department of Nutrition and Dietetics, İstanbul, Türkiye.

*Corresponding author e-mail: ahmetmurat.gunal@okan.edu.tr

Abstract

This study explores the sensory effects of enriching hamburger patties with psyllium, intending to increase the dietary fiber content of this widely consumed fast-food item. Hamburger patties were enriched with psyllium at concentrations of 5% and 7.5%, and their sensory attributes, including taste, smell, appearance, mouthfeel, juiciness, and texture/consistency, were evaluated with comparison to control patties. Additionally, overall preference based on sensory factors and hedonic scale scores were assessed. A randomized controlled triple-blind sensory analysis panel comprising twelve dietitians, organized into two blocks respectively, was employed for the evaluation. Although the results revealed that in the first block, the control patty received higher scores for taste compared to the 7.5% psyllium-enriched patty, this difference was not observed in the second block. For other sensory factors, overall preference, and hedonic scale scores, the patties exhibited similarity. In summary, this study demonstrates that the sensory characteristics of hamburger patties remain largely unaffected when enriched with psyllium up to a concentration of 7.5%. These findings suggest that psyllium enrichment can be a viable strategy to increase the dietary fiber content of fast-food items without compromising their sensory appeal, potentially promoting healthier dietary choices among consumers.

Keywords: Psyllium, Dietary fiber, Hamburger, Sensory analysis, Hedonic scale, Fast food.

INTRODUCTION

Psyllium, also known as Plantago psyllium or ispaghula, and locally referred to as "karnıyarık otu" in Türkiye, belongs to the Plantago plant family. Due to specific climate requirements, it is primarily grown in India but is also found in regions such as Pakistan, Bangladesh, and Iran (Khaliq et al., 2015). Psyllium is primarily cultivated for its seed husk, which is rich in soluble fiber. Psyllium seed husk can absorb up to 80 times its weight in water, making it a valuable ingredient in the food industry, especially as a thickening and gelling agent (Phan et al., 2020). This mucilage does not ferment in the intestine and passes through the gastrointestinal system without alteration (Fischer et al., 2004).

The viscous gel formed by psyllium delays the interaction of complex carbohydrates with digestive enzymes by increasing the viscosity of chyme. This slows down digestion and glucose absorption, reducing postprandial blood glucose concentrations (Gibb et al., 2015). As a result of this delay, nutrients that reach the distal ileum stimulate the release of glucagon-like peptide-1 (GLP-1) into the bloodstream. GLP-1 has effects on metabolism, including reduced appetite and increased glucose-dependent insulin secretion (McRorie, 2015). Increased viscosity prevents effective bile absorption from the distal ileum, leading to a decrease in the bile acid pool, which stimulates hepatocytes to produce more bile. This, in turn, increases the number of LDL cholesterol receptors on hepatocytes, resulting in decreased blood LDL and total cholesterol concentrations, while HDL cholesterol levels remain unaffected (Narayanan et al., 2020). The U.S. Food and Drug Administration (FDA) has reported that adding 7 grams or more of psyllium to a diet low in saturated fat and cholesterol may reduce the risk of coronary heart disease (FDA, 1998).

When consumed with a meal or before a meal, psyllium expands in the stomach, helping to activate stretch receptors early. It delays gastric emptying and plays a role in allowing food to reach the distal regions of the small intestine, contributing to a prolonged feeling of fullness. Additionally, the reduction in postprandial glycemic peak levels and, consequently, insulin requirements suggest that psyllium may play a role in promoting prolonged satiety (Wanders et al., 2011). Due to these effects on digestion and appetite, psyllium consumption is thought to reduce daily energy intake, potentially contributing to weight loss (Howarth et al., 2001).

Because of its gelling properties, psyllium is commonly used in the food industry. Today, it is most commonly used in the production of baked goods, especially bread, as a gluten substitute for its binding and elasticity-enhancing properties (Roman et al., 2019). Psyllium can be an important source of dietary fiber for those who need to follow a gluten-free diet, such as individuals with celiac disease or other reasons (Fratelli et al., 2018). Examples of psyllium use as a fat replacer can also be found in foods like cakes and mayonnaise (Belorio et al., 2019; Amiri Aghdaei et al., 2014). In meat products, limited examples of psyllium usage include increasing water-holding capacity, improving product stability, reducing the fat content by using it as a fat replacer, enhancing production efficiency, improving texture, preventing volume loss during cooking, and extending shelf life (Zinina et al., 2019).

Considering its potential benefits in the food industry and metabolic effects, psyllium is an important source of soluble fiber. The Turkish Dietary Guidelines (TÜBER) classify dietary fiber as one of the "Foods and Nutrients That Should Be Consumed More" and recommend an intake of 25 grams per day for adult men and women. It is noted that 64.2% of the population does not meet this fiber intake requirement (TÜBER, 2022). The National Academies of Sciences, Engineering, and Medicine in the United States recommends daily fiber intakes of 25 grams for adult women and 38 grams for adult men, with a practical guideline of 14 grams of fiber per 1,000 calories (Trumbo et al., 2001).

Fast food products, which have seen a significant increase in consumption today, are generally characterized by high energy density, heterogeneous high-fat content favoring saturated and omega-6 fatty acids, high glycemic index, and significant carbohydrate content. Additionally, they tend to be rich in sodium while lacking in other micronutrients and dietary fiber (Varlamov, 2017). On average, fast food items have been found to have an energy density of 236 kcal/100g, with a typical meal containing around 1400 kcal (Jaworowska et al., 2013). A study analyzing menus available in 35 countries, observed that fast food meals typically contain 41-74 grams of total fat per meal (Stender et al., 2007). Another study found that per 100 grams, these foods contain 10-13 grams of total fat and 3.9-4.9 grams of saturated fat (Dunford et al., 2010). Fast food products often use refined flours separated from bran and germ. Additionally, side items like french fries and commonly consumed soft drinks are known for their high glycemic index, primarily due to the presence of sucrose or more frequently, high-fructose corn syrup (HFCS). The increased portion sizes in these menus also contribute to a higher glycemic load. Because these meals are typically low in vegetables and fruits and high in processed refined grains, they are also deficient in dietary fiber (Isganaitis and Lustig, 2005).

Globally, one of the most frequently consumed fast-food items is the hamburger. The objective of this research is to enrich hamburger patties with psyllium while preserving their sensory attributes.

MATERIALS AND METHODS

This research received ethical approval in accordance with the Helsinki Declaration, from the Acıbadem Mehmet Ali Aydınlar University Medical Research Evaluation Board (ATADEK), during its meeting held on December 17, 2021, under decision number 2021-24/31. Following the approval of the Ethics Committee, the study was conducted between December 2021 and January 2022 at the Istanbul Okan University Nutrition Principles Laboratory, involving participants who met the inclusion criteria and volunteered to participate. Participation in the research was based on a voluntary basis. The sensory analysis panel consisted of 12 trained panelists, including 2 expert dietitians and 10 dietitians, and was structured into two blocks to enhance the reliability of the panel. Besides, it was designed in a triple-blind fashion. Prior to their involvement in the study, panelists were informed and provided written informed consent.

The data collection form used in the sensory analysis panel consists of two sections. The first section utilizes a sensory analysis form for rating six factors on a scale from 1 to 9 (1: very poor, 5: neither good nor bad, 9: very good). These factors include taste, smell, appearance, mouthfeel, juiciness, and texture. The researcher calculated an overall liking score by averaging the ratings for each of these six factors. The second section employs a five-point Likert hedonic scale of subjective evaluation by panelists to assess overall liking (1: definitely disliked, 3: neither liked nor disliked, 5: definitely liked).

Product Enrichment

In the preparation of the meatball mixture, a composition consisting of 20% fatty ground beef, 0.8% salt, and 0.2% black pepper was utilized (Özvural, 2016). To produce hamburger patties, 3 kg of ground beef labeled with 20% fat content, available in 500-gram sealed packages at a retail chain, was purchased. The 2880 grams of ground beef were divided into three equal portions of 960 grams each. To each portion, 0.8% (7.68 grams) salt and 0.2% (1.92 grams) black pepper were added. Subsequently, to one portion, 5% (48 grams) of psyllium powder was added, while to the other, 7.5% (72 grams) psyllium powder was added. Each mixture was kneaded for 15 minutes until achieving homogeneity. Certified psyllium powder, produced through organic farming in India and imported by the Güzel Ada Gıda company, was employed as the source of psyllium powder. The weights of the ground beef and hamburger patties were measured using a Sinbo brand kitchen scale, model SKS-4524, with a sensitivity of 1 gram, available at the Istanbul Okan University Nutrition Principles Laboratory. Salt, black pepper, and psyllium were measured using a Kern brand precision scale, model PNS-600, with a sensitivity of 0.001 grams, located in the Biochemistry Laboratory. The exact amounts can be seen in Table 1.

Table 1. Content and Weights of Meatball Mixtures Prepared for Sensory Analysis Panel

	Control	**%5**	**%7.50**
Ground Beef (g)	960	960	960
Salt (g)	7.68	7.68	7.68
Black Pepper (g)	1.92	1.92	1.92
Psyllium (g)	0	48	72
Total (g)	969.6	1017.6	1041.6
g/Hamburger Patty	161.6	169.6	173.6

To ensure uniformity in shape, Plastlife brand KFTPRS model hamburger patty molds were employed while forming the patties from the prepared mixtures. After the hamburger patties were prepared, they were placed in three identical storage containers and then stored in the refrigerator. The following day, all patties were cooked uniformly and divided into quarters to prevent the analysis of satiety factors from affecting the panelists during the sensory evaluation.

Blinding of the Sensory Analysis Panel

To ensure the triple-blind nature of the panel, an independent academician was tasked with using the Google Random Number Generator tool to randomly select six numbers between 100 and 999 (156, 371, 527, 699, 852, and 897). Subsequently, the academician encoded these numbers in pairs, assigning two numbers to each storage container.

Sensory Analysis Panel

The sensory analysis panel consisted of 12 trained panelists, comprising 10 dietitians and 2 dietitians with master's degrees. To enhance the reliability of the panel, it was structured into two blocks. The panelists were selected from a pool of non-smoking females aged between 22 and 31 years, without any recent illnesses such as colds or congestion that could affect their taste and smell senses. They were instructed to arrive at the panel neither hungry nor full and to abstain from consuming any food or beverages (except water) one hour prior to the start of the session.

Before commencing the sensory analysis, the panelists were provided with information about the research, obtained written consent, and were reminded of specific guidelines for sensory evaluations, including rinsing their mouths with water before and after each taste and thoroughly chewing the samples.

The sensory analysis panel was conducted at the Istanbul Okan University Nutrition Principles Laboratory. The laboratory was designed with non-fatiguing grey color tones and intermittent blue accents. Both natural daylight and white artificial light were used for illumination, and the temperature and humidity of the environment were maintained stable throughout the panel session. After each taste, the area was ventilated to prevent the mingling of odors. Water was offered to panelists to rinse their mouths before each tasting, and they were advised not to consume anything except water during the breaks between blocks.

The sensory analysis panel was conducted in two parallel blocks with a one-hour interval between them. In each block, participants tasted samples labeled with three-digit random numbers. Although each of the six codes used was different, all panelists tasted the same three types of hamburgers: classic, 5% psyllium enriched, and 7.5% psyllium enriched. This was done to make panelists believe they were tasting six different products. Panelists were given products one by one and were asked to rate six factors: taste, smell, appearance, mouthfeel, juiciness, and texture on a scale from 1 to 9 (1: very poor, 5: neither good nor bad, 9: very good). The researcher calculated an overall liking score by averaging the ratings for each factor. Additionally, a five-point Likert scale (1: definitely disliked, 3: neither liked nor disliked, 5: definitely liked) hedonic scale assessment was used. The form also provided space for panelists to record notes for each code if desired. The panel was conducted in a triple-blind manner, and after the analysis results were documented, the independent academician revealed which pair of codes corresponded to each product.

Data Analysis

The analysis of the data was conducted using MS Excel 16 and SPSS 22.0 software packages. Descriptive statistics were employed, including frequency, percentage, standard deviation, standard error, minimum and maximum values, median, and quartiles. Cronbach's Alpha was utilized for internal consistency analysis. For inferential statistics, various tests were employed, including independent samples t-test, paired samples t-test, Wilcoxon signed-rank test, two-way analysis of variance (ANOVA), and the Kruskal-Wallis H test. Post-hoc analysis was performed using the Duncan test. Assumptions were checked using skewness and Mardia's multivariate normality analysis for normal distribution, Levene's test for homogeneity of variances, and Box's M test for covariance equality. All data were interpreted at a 95% confidence level.

RESULTS

In the sensory analysis panel, a distinction in taste among the patties was observed within the first block, indicating that non-fortified patties received higher scores compared to those fortified with 7.5% psyllium ($p=0.023$). However, no significant differences were detected among the average scores for taste, smell, appearance, mouthfeel, juiciness, texture/consistency, and overall liking of the patties ($p>0.05$). In the second block, all factors and overall liking averages were found to be similar ($p>0.05$) (Table 2).

Table 2. In-Block Comparison of Sensory Factors and Overall Preference of Patties

	1st Block (n=36)					2nd Block (n=36)				
	Control	**5%**	**7.50%**	**F/χ^2**	**p_1**	**Control**	**5%**	**7.50%**	**F/χ^2**	**p_2**
Taste**	6.50±1.38	5.58±1.16	5.17±0.83	**4.228^F**	**0.023***	6.83±1.11	5.67±1.50	5.83±1.27	4.228$^{\chi^2}$	0.071
Smell	7.17±1.40	6.58±1.44	7.00±1.60	0.967$^{\chi^2}$	0.615	6.33±1.67	6.67±0.98	6.25±1.22	0.334^F	0.718
Appearance	7.25±1.71	7.50±1.24	7.17±1.53	0.159^F	0.854	7.25±1.54	7.42±1.16	6.17±2.12	0.159$^{\chi^2}$	0.208
Mouthfeel	6.08±1.38	5.92±1.51	5.58±1.16	0.422^F	0.659	7.08±1.08	6.08±1.44	5.83±1.53	2.817^F	0.074
Juiciness	6.58±1.88	6.67±1.83	6.83±1.27	0.069^F	0.934	5.75±1.60	6.42±1.38	5.83±1.99	0.069$^{\chi^2}$	0.583
Texture	6.00±1.41	6.33±2.06	6.58±1.44	0.370^F	0.693	6.17±1.85	6.25±1.42	6.08±1.56	0.032^F	0.969
Overall Preference	6.60±0.99	6.43±1.16	6.39±0.77	0.150^F	0.861	6.57±1.10	6.42±0.98	6.00±1.25	0.838^F	0.442

Values are represented as Mean±SD; F: Two-Way Analysis of Variance; χ^2: Kruskal Wallis H Test Statistic; *: $p<0.05$; **: Duncan Post-Hoc, Control > %7.5 Enriched.

When participants conducted a re-evaluation without prior knowledge that they were consuming the same patty again, they awarded a higher score in terms of mouthfeel to the non-fortified patty during the second tasting (z=-2.360; p=0.018). However, no significant differences were observed among evaluations conducted for other patties and factors (p>0.05). The internal consistency of the data was examined using Cronbach Alpha values, revealing high reliability for both the non-fortified (Control) and 5% fortified patties (α>0.8), while the 7.5% fortified patty exhibited a considerably reliable level of consistency (α>0.6) (Table 3).

Table 3. Inter-Block Comparison of Sensory Factors and Overall Preference of Patties

	Control (n=24)				5% (n=24)				7.50% (n=24)			
	1st Block	2nd Block	t / z	p_1	1st Block	2nd Block	t / z	p_2	1st Block	2nd Block	t / z	p_3
Taste**	6.50±1.38	6.83±1.11	-0.886^t	0.394	5.58±1.16	5.67±1.50	-0.085^z	0.932	5.17±0.83	5.83±1.27	-1.526^z	0.127
Smell	7.17±1.40	6.33±1.67	-1.556^z	0.120	6.58±1.44	6.67±0.98	-0.192^t	0.851	7.00±1.60	6.25±1.22	-0.777^z	0.437
Appearance	7.25±1.71	7.25±1.54	-0.137^z	0.891	7.50±1.24	7.42±1.16	-0.333^z	0.739	7.17±1.53	6.17±2.12	-1.552^z	0.121
Mouthfeel	6.08±1.38	7.08±1.08	**-2.360^z**	**0.018***	5.92±1.51	6.08±1.44	-0.297^t	0.772	5.58±1.16	5.83±1.53	-0.442^t	0.667
Juiciness	6.58±1.88	5.75±1.60	-1.481^z	0.139	6.67±1.83	6.42±1.38	-0.142^z	0.887	6.83±1.27	5.83±1.99	-1.706^z	0.088
Texture	6.00±1.41	6.17±1.85	-0.632^z	0.527	6.33±2.06	6.25±1.42	-0.121^z	0.904	6.58±1.44	6.08±1.56	0.944^t	0.365
Overall Preference	6.60±0.99	6.57±1.10	-0.257^z	0.797	6.43±1.16	6.42±0.98	0.036^t	0.972	6.39±0.77	6.00±1.25	-0.393^z	0.694
Cronbach α	0.875				0.820				0.613			

Values are represented as Mean±SD; z: Wilcoxon signed-rank test; t: Dependent samples t-test value; *: p<0.05.

When comparing the hedonic liking scores given by the participants between the patties, it was found that they were similar in both the first and second blocks of evaluation (p>0.05). Furthermore, when the hedonic scale scores given by the participants for the same patties were compared between the blocks, it was statistically determined that there was no significant difference (p>0.05) (Table 4).

Table 4. In-Block and Inter-Block Comparison of Hedonic Appreciation of Patties

Variables	1st Block (n=36)	2nd Block (n=36)	t / z	p_1
Control (n=24)	3.67±.0.99	3.33±0.89	-0.518^t	0.615
5% (n=24)	3.17±1.27	3.83±1.19	-0.378^t	0.713
7.50% (n=24)	3.00±0.85	3.08±1.24	-0.171^z	0.864
F	1.312	1.400		
p_2	0.283	0.261		

Values are represented as Mean±SD; F: Two-Way Analysis of Variance; z: Wilcoxon signed-rank test; t: Dependent samples t-test value; p_1: Inter-block comparison; p_2: In-block comparison.

DISCUSSION

In the present study, an evaluation was conducted comparing 5% and 7.5% psyllium-enriched hamburger patties with control patties, focusing on sensory attributes, overall liking, and hedonic scale scores. It is noteworthy that previous research has explored the impact of psyllium enrichment on the sensory properties of various food products. For instance, Ferjančič et al. (2021) conducted a study involving low-fat chicken bologna sausages enriched with inulin, oat fiber, and psyllium at levels of 3% and 6%. Interestingly, their findings indicated a decrease in scores for both sensory attributes and overall appreciation when psyllium was added to the sausages. Mehta et al. (2013) investigated the enrichment of chicken nuggets with psyllium (2%, 4%, and 6%) in addition to 10% rice bran. They observed a decrease in sensory analysis scores as the fiber ratio increased, although the nuggets enriched with 10% rice bran and 4% psyllium were found to be organoleptically acceptable. Furthermore, Zhou et al. (2019) explored the sensory characteristics of pork sausages enriched with varying levels of psyllium (0.5-1%, 1%, and 2-3%) in comparison to control sausages without enrichment. Their study revealed that the desired sensory attributes increased in the sausages enriched with 0.5-1% and 1% psyllium when compared to control sausages. However, this trend reversed when the psyllium enrichment level reached 2-3%. Similarly, Danyliv et al. (2019) examined the impact of psyllium

addition to a pork and beef mixture sausage at levels of 2%, 4%, 6%, and 8%. Their findings suggested that the organoleptic properties of the final product could be enriched by 2% psyllium without any noticeable alteration. In another study, products resembling meat patties, produced using chicken meat paste, were evaluated for characteristics such as texture and appearance. These products were obtained by incorporating water in the range of 0% to 40% and psyllium in the range of 0% to 3%. It was noted that the addition of 40% water and 3% psyllium was suitable for increasing product volume and yield without compromising characteristic attributes. However, due to the absence of a sensory analysis panel in the study, it does not provide a clear insight into overall preference (Zając et al., 2020). In an effort to enhance the texture and consistency of a plant-based sausage product, psyllium enrichment was carried out at concentrations of 3%, 4%, 5%, and 6%. These enriched sausages were compared to non-enriched sausages in terms of sensory attributes, with a focus on evaluating their overall acceptability. Despite the observation of a darker color in the high-concentration control sausages, all concentrations were deemed suitable in terms of texture/consistency and overall acceptability. The authors have suggested that the utilization of psyllium in plant-based meat analog products can improve their texture and consistency, potentially making them palatable for omnivorous individuals as well (Noguerol et al., 2022).

In the context of the current study, it is worth noting that while the control hamburger patty initially received higher scores in terms of taste during the first block of the sensory analysis panel, no significant difference was observed in the second block. Furthermore, the comparison between the two blocks revealed that the hamburger patties exhibited similarity in terms of sensory attributes, overall preference, and hedonic appreciation. These findings contribute to the understanding of the sensory impact of psyllium enrichment in hamburger patties, aligning with previous research in the field.

CONCLUSION

This research investigates the effects of enriching hamburger patties with psyllium on sensory factors, aiming to increase the dietary fiber content of one of the most consumed fast-food items worldwide. In this study, hamburger patties were enriched with psyllium at concentrations of 5% and 7.5%, and compared with control patties that underwent no intervention in terms of sensory attributes, including taste, smell, appearance, mouthfeel, juiciness, and texture/consistency. Additionally, overall preference derived from these sensory factors and hedonic scale scores were compared.

A randomized controlled triple-blind sensory analysis panel consisting of twelve dietitians, organized in two blocks, was employed. According to the panel results, while the control patty received higher scores for the taste factor compared to the 7.5% psyllium-enriched patty in the first block, this difference was not observed in the second block. Regarding other factors, overall preference, and hedonic scale scores, the patties were found to be similar. The panel results indicate that the sensory characteristics of hamburger patties remain unaffected when enriched with psyllium up to a concentration of 7.5%.

ACKNOWLEDGEMENTS

This study is articulated from the corresponding author’s doctoral dissertation. Other authors were advisors of the corresponding author’s thesis. The study did not receive any external funding. The authors declare there are no conflicts of interest.

REFERENCES

Amiri Aghdaei, S. S., Aalami, M., Babaei Geefan, S., & Ranjbar, A. 2014. Application of Isfarzeh seed Plantago ovate L.) mucilage as a fat mimetic in mayonnaise. Journal of Food Science and Technology, 51(10), 2748–2754. https://doi.org/10.1007/s13197-012-0796-7

Belorio, M., Sahagún, M., & Gómez, M. 2019. Psyllium as a Fat Replacer in Layer Cakes: Batter Characteristics and Cake Quality. Food and Bioprocess Technology, 12(12), 2085–2092. https://doi.org/10.1007/s11947-019-02362-3

Danyliv, M. M., Vasilenko, O. A., Ozherelyeva, O. N., & Shestakova, Y. A. 2019. Improvement of sausage production technology. In IOP Conference Series: Earth and Environmental Science Vol. 341, No. 1, p. 012131. IOP Publishing.

Dunford, E., Webster, J., Barzi, F., & Neal, B. 2010. Nutrient content of products served by leading Australian fast food chains. Appetite, 55(3), 484–489. https://doi.org/10.1016/j.appet.2010.08.015

Ferjančič, B., Kugler, S., Korošec, M., Polak, T., & Bertoncelj, J. 2021. Development of low-fat chicken bologna sausages enriched with inulin, oat fibre or psyllium. International Journal of Food Science & Technology, 56(4), 1818-1828.

Fischer, M. H., Yu, N., Gray, G. R., Ralph, J., Anderson, L., & Marlett, J. A. 2004. The gel-forming polysaccharide of psyllium husk Plantago ovata Forsk. Carbohydrate Research, 339(11), 2009–2017. https://doi.org/10.1016/j.carres.2004.05.023

Food and Drug Administration. 1998. FDA allows foods containing psyllium to make health claim on reducing risk of heart disease. Rockville, MD: US Department of Health and Human Services.

Gibb, R. D., McRorie, J. W., Russell, D. A., Hasselblad, V., & D'Alessio, D. A. 2015. Psyllium fiber improves glycemic control proportional to loss of glycemic control: A meta-analysis of data in euglycemic subjects, patients at risk of type 2 diabetes mellitus, and patients being treated for type 2 diabetes mellitus. American Journal of Clinical Nutrition, 102(6), 1604–1614. https://doi.org/10.3945/ajcn.115.106989

Howarth, N. C., Saltzman, E., & Roberts, S. B. 2001. Dietary fiber and weight regulation. Nutrition reviews, 59(5), 129-139.

Isganaitis, E., & Lustig, R. H. 2005. Fast food, central nervous system insulin resistance, and obesity. In Arteriosclerosis, Thrombosis, and Vascular Biology Vol. 25, Issue 12, pp. 2451–2462. https://doi.org/10.1161/01.ATV.0000186208.06964.91

Jaworowska, A., Blackham, T., Davies, I. G., & Stevenson, L. 2013. Nutritional challenges and health implications of takeaway and fast food. Nutrition reviews, 71(5), 310-318.

Khaliq, R., Tita, O., Antofie, M. M., & Sava, C. 2015. Industrial Application of Psyllium: An Overview. ACTA Universitatis Cibiniensis, 67(1), 210–214. https://doi.org/10.1515/aucts-2015-0092

McRorie, J. W. 2015. Evidence-based approach to fiber supplements and clinically meaningful health benefits, part 1. Nutrition Today, 50(2), 82–89. https://doi.org/10.1097/NT.0000000000000082

Mehta, N., Ahlawat, S. S., Sharma, D. P., Yadav, S., & Arora, D. 2013. Sensory attributes of chicken meat rolls and patties incorporated with the combination levels of rice bran and psyllium husk. Journal of Animal Research, 3(2), 179.

Narayanan, S., & Pitchumoni, C. S. 2020. Dietary fiber. Geriatric Gastroenterology, 1-16.

Noguerol, A. T., Larrea, V., & Pagán, M. J. 2022. The effect of psyllium (Plantago ovata Forsk) fibres on the mechanical and physicochemical characteristics of plant-based sausages. European Food Research and Technology, 1-14.

Özvural, E. B., Huang, Q., & Chikindas, M. L. 2016. The comparison of quality and microbiological characteristic of hamburger patties enriched with green tea extract using three techniques: Direct addition, edible coating and encapsulation. LWT-Food Science and Technology, 68, 385-390.

Phan, J. L., Cowley, J. M., Neumann, K. A., Herliana, L., O'Donovan, L. A., & Burton, R. A. 2020. The novel features of Plantago ovata seed mucilage accumulation, storage and release. Scientific Reports, 10(1). https://doi.org/10.1038/s41598-020-68685-w

Roman, L., Belorio, M., & Gomez, M. 2019. Gluten-Free Breads: The Gap Between Research and Commercial Reality. In Comprehensive Reviews in Food Science and Food Safety, 18(3), 690–702. Blackwell Publishing Inc. https://doi.org/10.1111/1541-4337.12437

Stender, S., Dyerberg, J., & Astrup, A. 2007. Fast food: Unfriendly and unhealthy. In International Journal of Obesity, 31(6), 887–890. https://doi.org/10.1038/sj.ijo.0803616

Trumbo, P., Schlicker, S., Yates, A. A., & Poos, M. 2002. Food and Nutrition Board of the Institute of Medicine, The National Academies. Dietary reference intakes for energy, carbohydrate, fiber, fat, fatty acids, cholesterol, protein and amino acids. J Am Diet Assoc, 102(11), 1621-30.

Türkiye Beslenme Rehberi (TÜBER) 2022 Sağlık Bakanlığı, Halk Sağlığı Genel Müdürlüğü, Sağlık Bakanlığı Yayın No:1031, Ankara 2022

Varlamov, O. 2017. Western-style diet, sex steroids and metabolism. Biochimica et Biophysica Acta - Molecular Basis of Disease, 1863(5), 1147–1155. https://doi.org/10.1016/j.bbadis.2016.05.025

Wanders, A. J., van den Borne, J. J. G. C., de Graaf, C., Hulshof, T., Jonathan, M. C., Kristensen, M., Mars, M., Schols, H. A., & Feskens, E. J. M. 2011. Effects of dietary fibre on subjective appetite, energy intake and body weight: A systematic review of randomized controlled trials. Obesity Reviews, 12(9), 724–739. https://doi.org/10.1111/j.1467-789X.2011.00895.x

Zając, M. H. 2020. The properties of poultry batters depending on the amount of water and Plantago ovata husk. Acta Scientiarum Polonorum Technologia Alimentaria, 19(4), 475-482.

Zhou, Y., Ma, L., Yu, Y., Zhu, H., Wang, H., & Zhang, Y. 2019. Effect of psyllium husk powder addition on quality of meat patties. Journal of Food Science and Technology Beijing), 37(5), 42-49.

Zinina, O., Merenkova, S., Tazeddinova, D., Rebezov, M., Stuart, M., Okuskhanova, E., Yessimbekov, Z., & Baryshnikova, N. 2019. Enrichment of meat products with dietary fibers: A review. In Agronomy Research, 17(4), 1808–1822. Eesti Pollumajandusulikool. https://doi.org/10.15159/AR.19.163

DRAFT

ORAL PRESENTATION

Eremogone Fenzl (Caryophyllaceae) cinsi üzerine taksonomik incelemeler

Metin Armağan (*ORCID: https://orcid.org/0000-0002-3913-954X*)
Necmettin Erbakan Üniversitesi, Ereğli Ziraat Fakültesi, Tarla Bitkileri Bölümü, Konya, Türkiye
metinarmagan@erbakan.edu.tr

Özet

Türkiye florasında en fazla türe sahip familyalardan biri olan Caryophyllaceae (Karanfilgiller) dünya genelinde yaklaşık 100 cins ve 3000 tür içermektedir. Yapılan çalışmalarla, *Arenaria* Ruppius ex L.'nın *Eremogone* Fenzl alt cinsi (yaklaşık 70 tür) ve *Eremogoneastrum* Williams alt cinsi (22 tür) ile *Minuartia* Loefl.'nın *Spergella* (Fenzl) McNeill alt cinsi (3 tür) altında sınıflandırılan taksonlar *Eremogone* cinsi altına taşınmışlardır. *Eremogone* cinsi, genellikle yoğun şekilde yastık oluşturan, genellikle dallı, odunsu tabanlı çok yıllık bitkilerdir. Yapraklar genellikle çiçekli sapların tabanında veya yakınında kümelenmiştir. *Eremogone* cinsi dünyada 100 taksonla kuzey ılıman bölgelerde, özellikle Kuzeybatı Amerika'da, Avrasya Dağları'nda, Asya'da ve Küçük Asya'da yayılış göstermektedir. Türkiye'de ise 1 alt tür ile birlikte toplam 19 taksondan oluşur ve bunlardan 12 tanesi endemiktir. Bu çalışmada, Türkiye'de yayılış gösteren *Eremogone* taksonlarının morfolojiye dayalı revizyonu hedeflenmiştir. Yurtiçi ve yurtdışı herbaryum örnekleri stereo mikroskop altında incelenmiş, arazi çalışmaları ile toplanan canlı materyaller üzerinde çalışılmıştır. Taksonların teşhisinde kullanılabilecek karakterler ve türlerin ayrımı netleştirilmiştir. Çalışmalar sonucunda tür ayrımında glandular tüylerin ve çiçek durumundaki çiçek sayılarının değişkenlik gösterebildiği saptanmıştır. Kullanılacak tür teşhis anahtarlarında türlerin ayrımında sepallerin, petallerin, staminal glandların, gövdedeki yaprak çifti sayısının, yaprakların büyüklüğünün önemli değişmez karakterler olduğu tespit edilmiştir. Fakat özellikle sepallerin kenar kısımlarındaki içe katlanmalar tür ayrımında karışıklıklara neden olmaktadır. Bu çalışmada, taksonların teşhis karakterleri, birbiriyle morfolojik olarak benzer olan taksonların farklılıkları, habitat fotoğrafları ve yayılışları hakkında bilgiler verilmektedir.

Anahtar Kelimeler: *Arenaria, Eremogone, Minuartia*, Revizyon, Türkiye.

Taxonomic studies on the genus *Eremogone* Fenzl (Caryophyllaceae)

Abstract

One of the families with the most species in the flora of Turkey, Caryophyllaceae includes about 100 genera and 3000 species worldwide. As a result of the studies, taxa classified under the subgenus *Eremogone* Fenzl (about 70 species) and *Eremogoneastrum* Williams (22 species) of *Arenaria* Ruppius ex L. and subgenus *Spergella* (Fenzl) McNeill (3 species) of *Minuartia* Loefl. were moved to the genus *Eremogone*. The genus *Eremogone* is perennial plants with a woody base, usually densely cushion-forming and branched. Leaves are usually clustered at or near the base of flowering stems. The genus *Eremogone* is distributed in north temperate regions, especially in Northwest America, Eurasian Mountains, Asia and Asia Minor with 100 taxa worldwide. In Turkey, it consists of a total of 19 taxa including 1 subspecies and 12 of them are endemic. The aim of this study was to revise the Eremogone taxa distributed in Turkey based on morphology. National and international herbarium specimens were examined by stereo microscope and living material collected through field studies were examined. Characters that can be used in the identification of taxa and species distinguishing were clarified. As a result of the investigations, it was determined that glandular hairs and the number of flowers in the inflorescence can be variable in species classification. It was determined that sepals, petals, staminal glands, the number of leaf pairs on the stem and the size of the leaves are important stable characters in the species identification keys to be used. However, especially the inward folds on the margins of the sepals cause confusion in species determination. In this study, the diagnostic characters of the taxa, the differences of the morphologically related taxa, habitat photographs and information about their distribution are given.

Keywords: *Arenaria, Eremogone, Minuartia*, Revision, Türkiye

GİRİŞ

Eremogone cinsinin ait olduğu Caryophyllaceae (Karanfilgiller) familyası Dünya florasında en fazla türe sahip familyalarından biridir (Hernández-Ledesma ve ark., 2015; Govaerts, 2022). Bittrich (1993) tarafından familya Alsinoideae Burnett, Caryophylloideae Arn., ve Paronychioideae A.St. olmak üzere 3 alt familya ve 5 tribusa ayrılmıştır. Caryophyllaceae familyası taksonlarının farklı gen bölgeleri üzerinde yapılan moleküler çalışmalar ile familya monofiletik gruplar temelinde 11 tribusa ayrılmıştır (Harbaugh ve ark., 2010; Fior ve ark., 2006). *Arenaria* alt cinsi olarak sınıflandırılan *Eremogone* Fenzl cinsi moleküler analizler sonucunda ayrı bir cins olarak kabul edilmiş, *Arenaria* Ruppius ex L.'nın *Eremogone* Fenzl alt cinsi (yaklaşık 70 tür) ve *Eremogoneastrum* Williams alt cinsi (22 tür) ile *Minuartia* Loefl.'nın *Spergella* (Fenzl) McNeill alt cinsi (3 tür) altında sınıflandırılan taksonlar *Eremogone* cinsi altına taşınmışlardır (McNeill, 1962, 1967; Rabeler ve Wagner, 2015; Dillenberger ve Kadereit, 2014; Sadeghian ve ark., 2015).

Eremogone ilk kez Eduard Fenzl (1833) tarafından tanıtılmış. *Eremogone* Avrupa, Arktik, Küçük Asya, Kuzeybatı Amerika'da ve Kuzeydoğu Afrika'da bulunur. Başlıca çeşitlilik merkezleri, Orta ve Güney-Batı Asya ve Kuzeybatı Amerika'nın dağlarıdır (POWO, 2022). Ülkemizde yayılış gösteren 19 *Eremogone* taksonunun 12 tanesi endemiktir (Dinç, 2012).

Eremogone cinsi üyeleri, çok yıllık, tabanda odunsu, yastıksı formda bitkilerdir. Gövdeler yatık ya da yükselici, basit ya da dallanmıştır. Yapraklar genelde çiçekli gövdenin tabanında kümelenmiş, bir damarlı, ipliksi, iğnemsi, darca lineardır. Çiçek durumu bir veya daha fazla terminal talkımdan oluşur. Sepaller ince kenarlı genellikle geniştir. Çiçeksapları dik veya çiçekler sapsız. Çanak yapraklar 5, belirgindir. Taç yapraklar 5, beyaz, sarımsı beyaz veya bazen pembe veya kahverengimsi, pençeli veya değil, ayanın tepesi tam, aşınmış, girintili veya nadiren 2 parçalı; nektarlar genellikle 5 adet, çanak yaprakların karşısındaki filamentlerin tabanında belirgin. Stil 3, ipliksi. Tohumlar 1-10, koyu kırmızımsı veya yeşilimsi kahverengi, ten rengi, siyahımsı mor, siyah veya gri, tohum şekli oval, yanal olarak sıkıştırılmış, tohum yüzeyi pürüzsüz, ruguloz veya tuberkulat, kenarlarında kanatları yoktur (Harbaugh vd., 2010; Rabeler ve Hartman, 2005; Rechinger, 1964, 1988; Schischkin, 1995).

Bu çalışmasının amacı *Eremogone* cinsinin Türkiye taksonlarının morfolojilerinin incelenmesi, morfolojiye dayalı türler arasındaki farklılıkların ortaya konması, taksonların ayrımındaki kullanılacak varyasyon göstermeyen teşhis karakterlerinin belirlenmesidir.

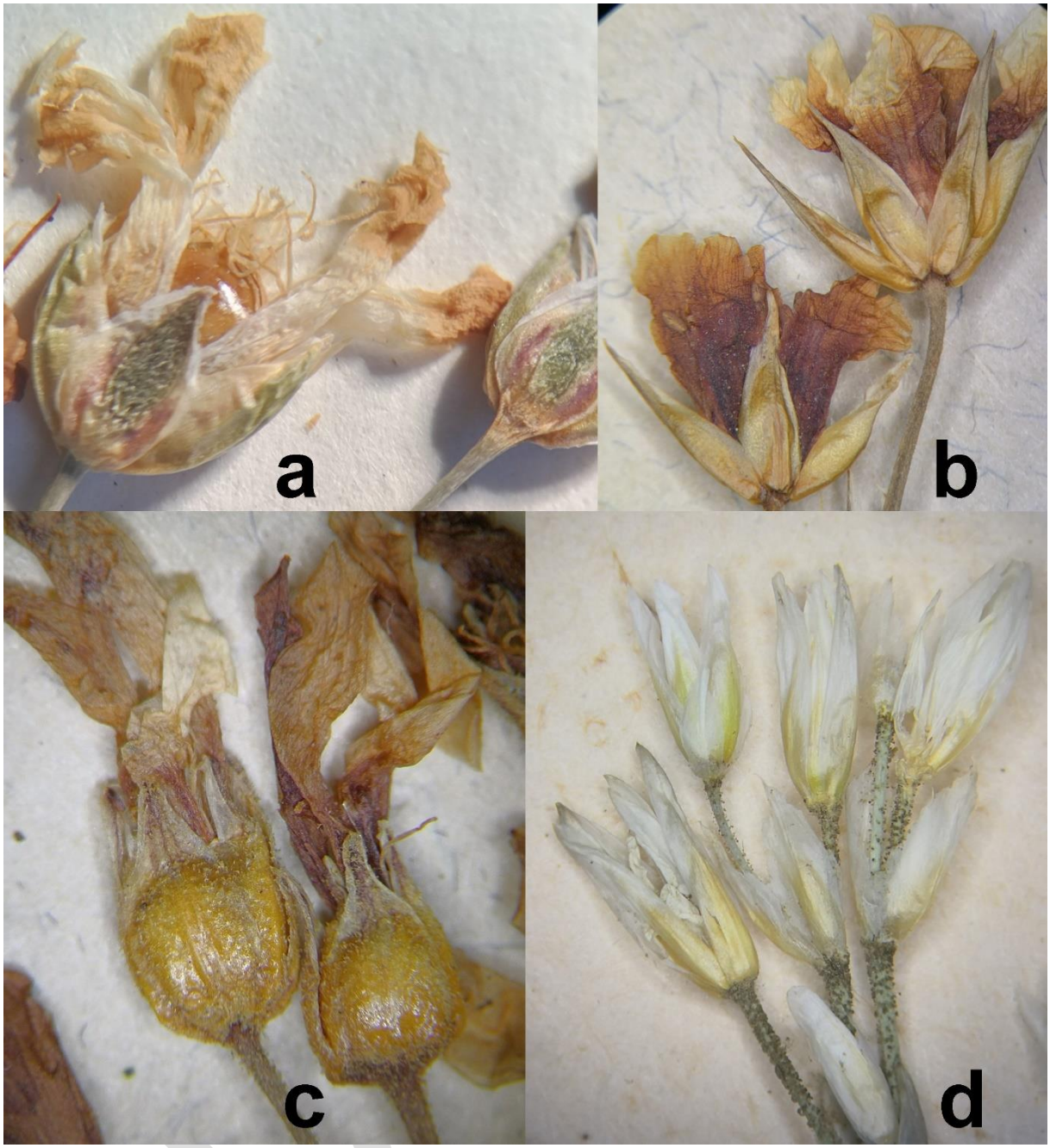
MATERYAL VE METOT

2018 – 2023 yılları arasında gerçekleştirilen arazi çalışmalarıyla toplanan canlı materyaller bu çalışmanın ana materyalini oluşturmaktadır. Ayrıca yurtiçi ANK, AEF, ESSE, GAZI, ISTO, KNYA, VANF ve yurtdışı E, G, K ve P herbaryumlarında (Herbaryum kodları Thiers (2023)'e göre yazılmıştır) bulunan örnekler stereo mikroskop altında incelenmiştir. Her taksonun morfolojisi mikroskop altında ölçülmüş, özellikle tayin karakterlerinin fotoğrafları çekilerek takson ayrımında kullanılmak üzere depolanmıştır.

BULGULAR ve TARTIŞMA

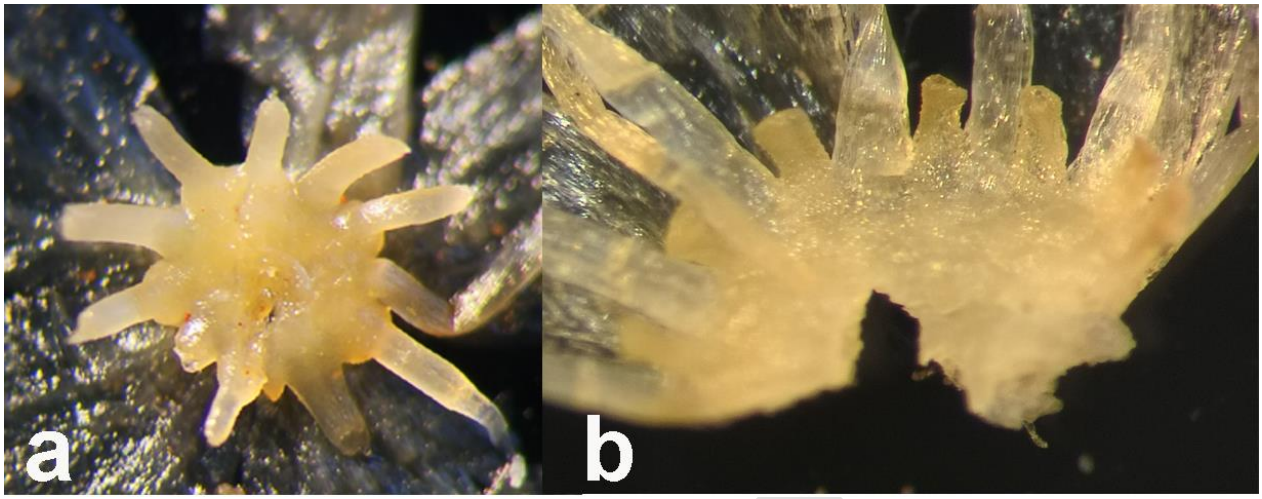
Örnekler üzerinde çiçek karakterleri, yaprak formları, tüylülük durumları, çiçekdurumları incelenmiştir. Flora çalışmalarında sepallerin şekilleri, sepallerin uç kısımları, petallerin sepallere oranı, petallerin uzunluğu ve şekli tür teşhislerinde belirleyici karakterler olarak kullanılmıştır. Sepaller genişçe akut ila aküminat veya kuspidat, uzun akut, oblong-lanseolat, hemen hemen tamamen zarsı, salgı tüylü veya tamamen tüysüz olabilir (şekil 1). Inflorescence kimöz panikula veya kümelenmiş salkımlardan oluşan korimboz veya tek olabilmektedir.

DRAFT

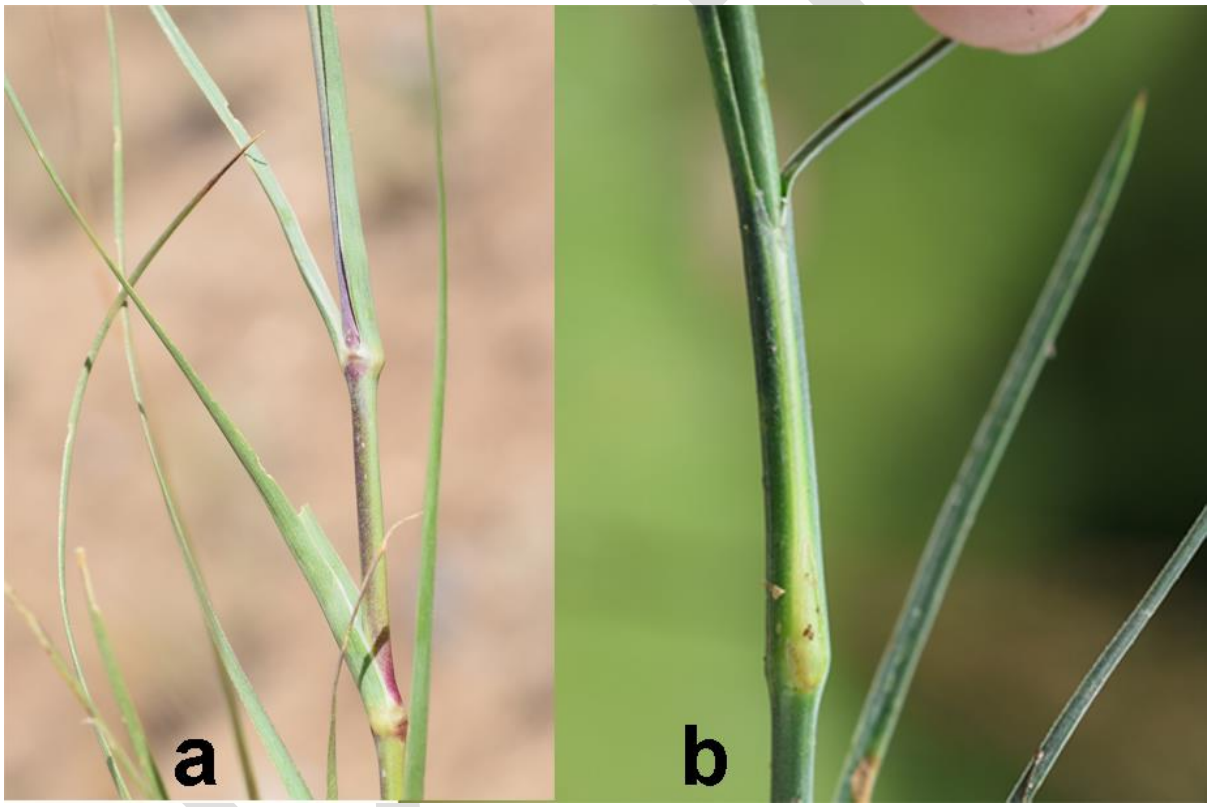


Şekil 1. *Eremogone*'de görülen bazı sepal şekilleri; **a)** geniş akut, **b)** uzun akut, **c)** akut-akuminat, **d)** zarsı.

Staminal glandlar (nektaryum) kanat benzeri (belirsiz) veya parmaksı olabilir (şekil 2). Yapraklar çimensi ya da batıcı olup dikensi, belirgin tek damarlı, gövde tabanında demetler halinde olur. Gövdede ki yaprak çift sayısı ise türler içinde sabittir. Yaprakların birleşme yerlerinde oluşturdukları kınlar (şekil 3) tür ayrımında önemlidir.



Şekil 2. *Eremogone* cinsinde görülen staminal glandlar: **a)** kanatsı, **b)** parmaksı.



Şekil 3. *Eremogone* gövde yapraklarının tabanı; **a)** serbest, **b)** kın oluşturmuş.

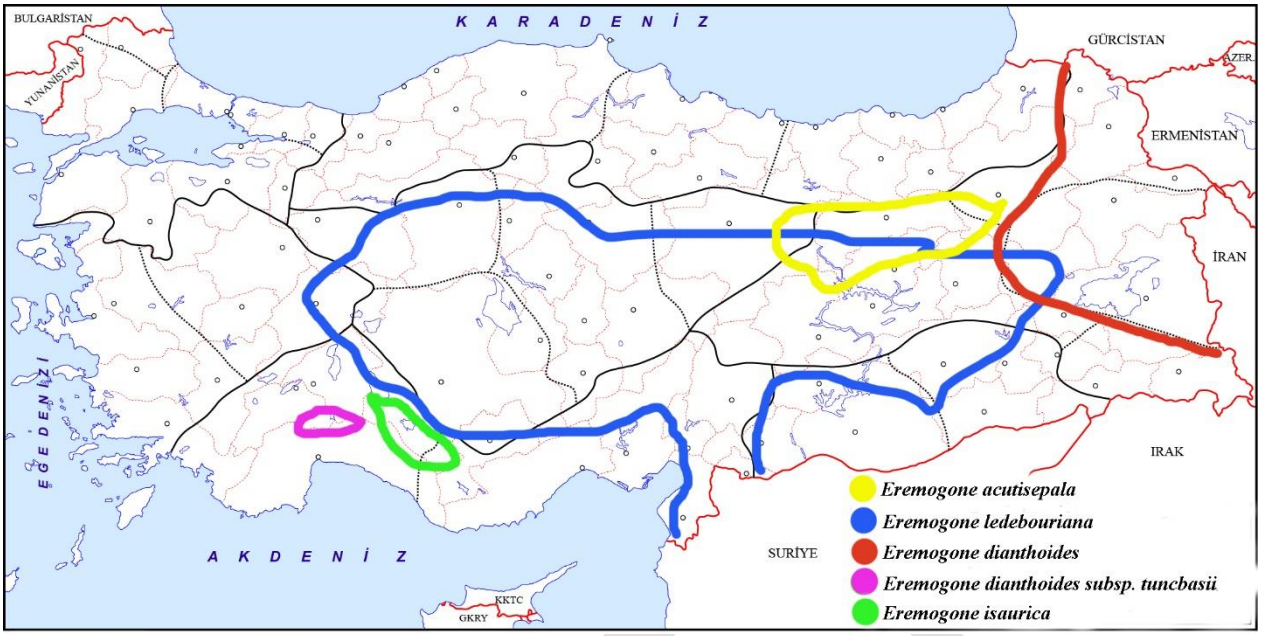
Türkiye'de 13'ü endemik toplam 19 takson yayılış göstermektedir (Tablo 1).

Tablo 1. Türkiye'de bulunan *Eremogone* taksonlarının yayılış bölgeleri ve endemizm durumları

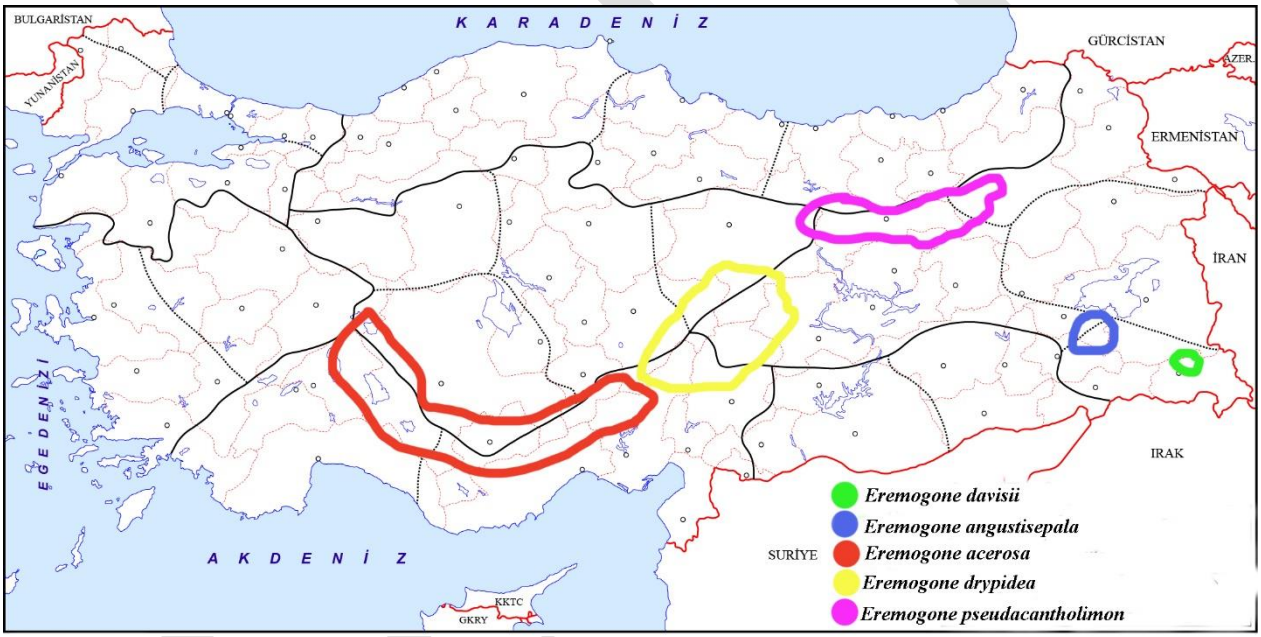
Takson adı	Bölge	Endemizm
E. acerosa (Boiss. & Heldr.) Ikonn.	İç Ege, Akdeniz	Endemik
E. acutisepala (Hausskn. Ex F.Williams) Ikonn.	İç Doğu Anadolu	Endemik
E. ali-gulii Koç & Hamzaoğlu	İç Doğu Anadolu	Endemik
E. angustisepala (McNeill) Ikonn.	Doğu Anadolu	Endemik
E. armeniaca (Boiss.) Holub	İç Doğu Anadolu	Endemik
E. blepharophylla (Boiss.) Ikonn.	Doğu Anadolu	değil
E. commagenae (Çeleb. & Favarger) Rabeler & W.L.Wagner		Endemik
E. cucubaloides (Sm.) Hohen.	Doğu Anadolu	değil
E. davisii (McNeill) Holub	Doğu Anadolu	Endemik
E. dianthoides (Sm.) Ikonn. subsp. *dianthoides*	Doğu Anadolu	değil
E. dianthoides (Sm.) Ikonn. subsp. *tuncbasii* (Eren & Parolly) M.Dinç	Akdeniz	Endemik
E. drypidea (Boiss.) Ikonn.	İç Anadolu'nun güneydoğusu	Endemik
E. gypsophiloides (L.) Fenzl	Doğu Anadolu	değil
E. isaurica (Boiss.) Ikonn.	Akdeniz	Endemik
E. ledebouriana (Fenzl) Ikonn.	Akdeniz, İç Anadolu, Doğu Anadolu, Güneydoğu Anadolu	değil
E. lychnidea (M.Bieb.) Rupr.	Karadeniz	değil
E. macrantha (Schischk.) Ikonn.	Doğu Anadolu	değil
E. pseudacantholimon (Bornm.) Holub	İç Doğu Anadolu	Endemik
E. scariosa (Boiss.) Holub	İç Doğu Anadolu	Endemik

Türkiye'deki 12 endemik *Eremogone* taksonundan 6 tanesi lokal endemiktir (*E. isaurica, E. dianthoides* ssp. *tuncbasii, E. ali-gulii, E. davisii, E. angustisepala, E. commagenae*). Batı Ege, Marmara ve Karadeniz'in büyük bir kesiminde *Eremogone* kaydı bulunmamaktadır. Türkiye'de en geniş yayılış alanına *E. ledebouriana* sahiptir. Genelde türlerin çoğunluğu doğu illerinde toplanmıştır (Şekil 4, 5, 6, 7).

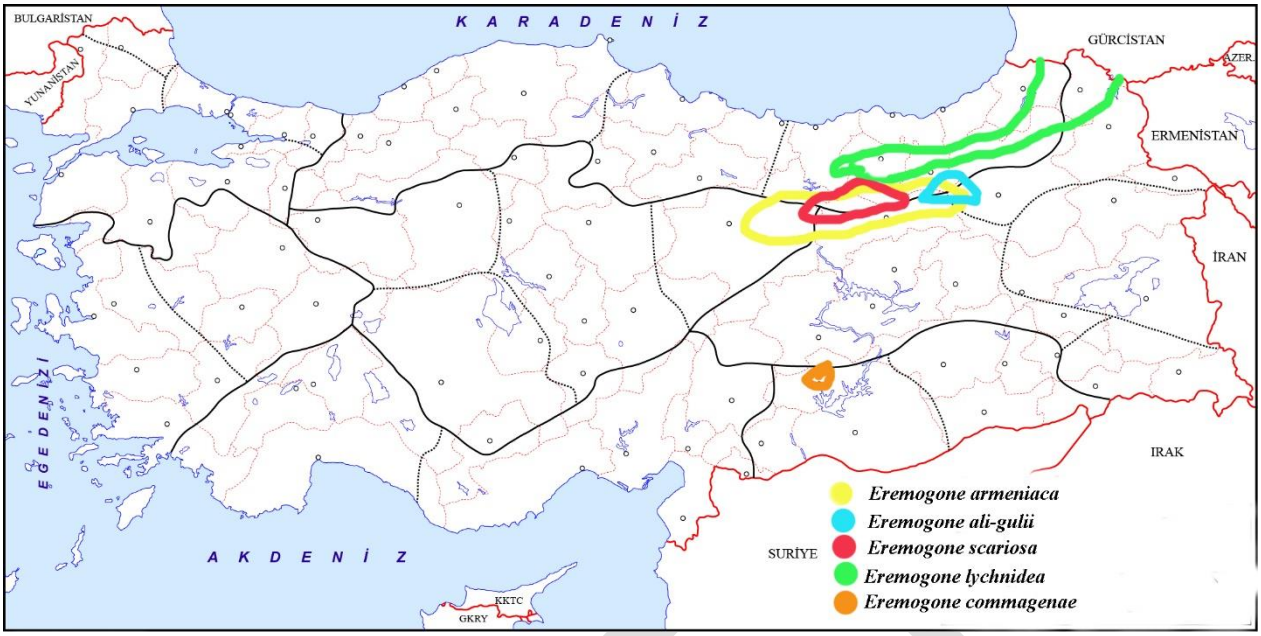
DRAFT



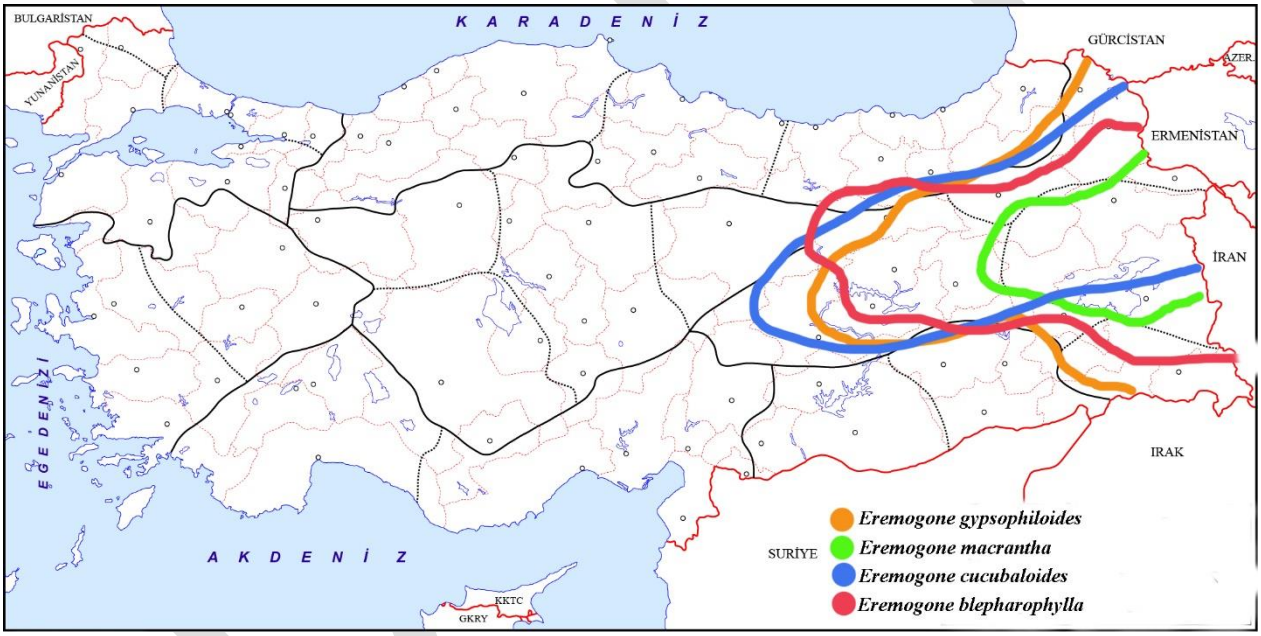
Şekil 4. *Eremogone* taksonlarının yayılışları (haritalar cografyaharita.com'dan alınarak düzenlenmiştir).



Şekil 5. *Eremogone* taksonlarının yayılışları (haritalar cografyaharita.com'dan alınarak düzenlenmiştir).

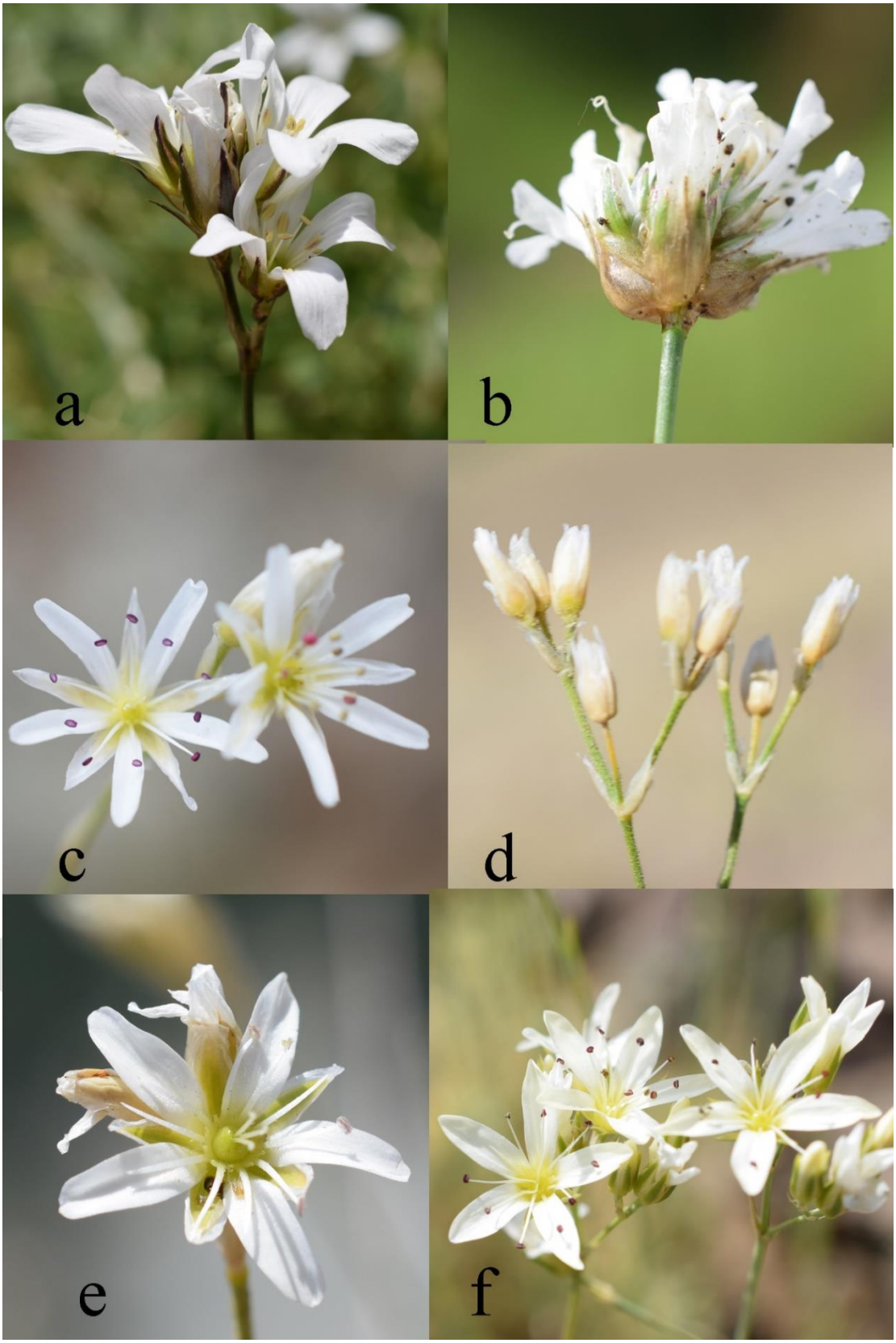


Şekil 6. *Eremogone* taksonlarının yayılışları (haritalar cografyaharita.com'dan alınarak düzenlenmiştir).



Şekil 7. *Eremogone* taksonlarının yayılışları (haritalar cografyaharita.com'dan alınarak düzenlenmiştir).

Türkiye'de bulunan bazı *Eremogone* taksonlarının teşhislerinde karışıklık yapılmaktadır. Genelde *E. ledebouriana*, *E. acerosa*, *E. drypidea* ve *E. commagenae* birbiriyle zaman zaman karıştırılabilmektedir. Bunun sebebi *E. ledebouriana*'nın çok fazla varyasyon aralığına sahip olmasıdır. Gövdede bulunan yaprak çift sayısı ve sepal şekilleri ile ayırt etmek daha kolaydır. *E. acutisepala*, *E. scariosa*, *E. ali-gulii* ve *E. armeniaca*'da teşhisinde karıştırılabilen bir gruptur (Şekil 8). Bu dört türün yayılış alanları da örtüşmektedir. Türkiye'deki tek alttür *E. dianthoides* subsp. *tuncbasii*'dir. Gövdenin tepesinde sıkı bir şekilde kümelenmiş çiçek durumu sadece *E. dianthoides* türünde görülmektedir (Şekil 8). Yaprak özellikleriyle *E. gypsophiloides*, *E. lychnidea*, *E. isaurica*, *E. macrantha* ve *E. cucubaloides* birbirine benzer yapıdadır. Taksonlar içinde *E. angustisepala*, *E. davisii ve E. pseudacantholimon* çok kolay ayrılabilen net türlerdir. Sadece *E. cucubaloides* ve *E. dianthoides* türlerinin gövde yaprak çiftlerinin tabanında 3 mm'den uzun kın vardır.



Şekil 8. Bazi *Eremogone* taksonlarının çiçekleri; a) *E. dianthoides* subsp. *tuncbsii*, b) *E. dianthoides*, c) *E. scariosa*, d) *E. ali-gulii*, e) *E. armeniaca*, f) *E. acutisepala*.

Sadece *Eremogone armeniaca*, *E. blepharophylla*, *E. scariosa*, *E. ali-gulii* türleri parmaksı staminal gland (nektaryum)lara sahiptirler. Bu özellikleri ile diğer türlerden rahatlıkla ayırt edilebilirler. Bu grupta *E. armeniaca*'da çiçek durumu kabuksu bir brakte ile çevrelenmiştir. Bu yönüyle kolayca ayırt edilebilmektedir.

Zaman zaman arazi çalışmalarında *E. gypsophiloides* ile *E. blepharophylla* karıştırılabilmektedir. Fakat bu iki türün arasında çiçeklenme zamanında farklılık mevcuttur. Önce *E. blepharophylla* çiçeklenir ve meyveye geçme zamanından sonra *E. gypsophiloides* çiçeklenir. Ayrıca staminal glandları ve sepal şekilleri ile de net ayrılabilmektedir.

SONUÇ

Çalışma sonunda Türkiye'de 12'si endemik toplam 19 takson değerlendirilmiştir (tablo 1). Yapılan morfolojik incelemelerde *E. scariosa*, *E. armeniaca* ve *E. ali-gulii* türlerinin birbiriyle çok yakın oldukları ve bunların evrimsel akrabalıklarının belirlenmesi için moleküler, polen, anatomi ve tohum mikromorfolojisi analizlerine ihtiyaç olduğu kanısına varılmıştır. Bu üç tür yayılış alanları iç içe olmakla birlikte, habitatları da benzerlik göstermektedir. Türkiye'de geniş bir bölgede bulunan *E. ledebouriana*'nın birbirinden farklı gövde yapıları ve çiçek durumları göze çarpmıştır. Bu türün deskripsiyonu daraltılarak sinonim olan bazı taksonların canlandırılması yapılabilir. *E. gypsophiloides* örnekleri üzerinde yaptığımız incelemelerde bir bireyin bir dalının salgı tüylü başka bir dalının ise tüysüz olduğu görülmüştür. Bu inceleme sonucunda çiçek durumundaki salgı tüylerinin tür teşhislerinde yanıltıcı olabileceği sonucuna varılmıştır.

Ayrıca yapılan herbaryum ve arazi çalışmaları sonucunda, Türkiye'de *E. szowitsii* (Boiss.) Ikonn.'nin bulunmadığı anlaşılmıştır. Herbaryumlarda *E. szowitsii* olarak teşhis edilen örneklerin *E. macrantha*'ya ait olduğu teşhisle kesinleştirilmiştir. Böylece Türkiye'de toplam takson sayısı 19 olarak belirlenmiştir.

TEŞEKKÜR

Resimli Türkiye Florası'ndaki *Gypsophila* ve *Eremogone* cinslerinin revize edilmesine maddi destek sağlayan Ali Nihat Gökyiğit (ANG) Vakfı'na teşekkür ederim. Ayrıca Türkiye Bilimsel ve Teknik Araştırma Kurumu (TÜBİTAK)'a (221Z314 nolu proje) teşekkür ederim.

KAYNAKLAR

Bittrich V 1993. Caryophyllaceae. In: Kubitzki K, Rohwer J, Bittrich V (Eds) The Families and Genera of Vascular Plants, Magnoliid, Hamamelid, and Caryophyllid Families, Vol. 2. Springer, Berlin, 206–236. doi: 10.1007/978-3-662-02899-5_21

Dillenberger MS, Kadereit JW 2014. Maximum polyphyly: Multiple origins and delimitation with plesiomorphic characters require a new circumscription of Minuartia (Caryophyllaceae). Taxon 63: 64–88. doi: 10.12705/631.5

Dinç M 2012. Eremogone Fenzl. In: Güner A, Aslan S, Ekim T, Vural M, Babaç MT (Eds) Türkiye Bitkileri Listesi (Damarlı Bitkiler). Nezahat Gökyiğit Botanik Bahçesi ve Flora Araştırmaları Derneği Yayını, İstanbul, 337–339.

Fenzl E 1833. Versuch einer Darstellung der Geographischen Verbreitungs- und Vertheilungs-Verhältnisse der natürlichen Familie der Alsineen 13, and unnumbered plate.

Fior S, Karis PO, Casazza G, Minuto L, Sala F 2006. Molecular phylogeny of the Caryophyllaceae (Caryophyllales) inferred from chloroplast matK and nuclear rDNA ITS sequences. American Journal of Botany 93: 399–411. doi: 10.3732/ajb.93.3.399

Govaerts R (2022). The World Checklist of Vascular Plants (WCVP). Royal Botanic Gardens, Kew. Checklist dataset https://doi.org/10.15468/6h8ucr [8. 3. 2023].

Harbaugh DT, Nepokroeff M, Rabeler RK, McNeill J, Zimmer EA, Wagner WL 2010. A new lineage-based tribal classification of the family Caryophyllaceae. International Journal of Plant Science 171: 185–198. doi: 10.1086/648993

Hernández-Ledesma P, Berendsohn,WG, Borsch T, Mering SV, Akhani H, Arias S, Castañeda-Noa I, Eggli U, Eriksson R, Flores-Olvera H, Fuentes-Bazán S, Kadereit G, Klak C, Korotkova N, Nyffeler R, Ocampo G, Ochoterena H, Oxelman B, Rabeler RK, Sanchez A, Schlumpberger BO, Uotila P (2015). A taxonomic backbone for the global synthesis of species diversity in the angiosperm order Caryophyllales. Willdenowia 45 (3): 281–383. https://doi.org/10.3372/wi.45.45301

McNeill J 1962. Taxonomic studies in the Alsinoideae. A revision of the species in the Orient. Notes from the Royal Botanical Garden Edinburgh 24(1): 102–140.

McNeill J 1967. Arenaria L. In: Davis PH (Ed.) Flora of Turkey and the East Aegean Islands, Vol. 2. Edinburgh University Press, Edinburgh, 17–38.

POWO (2023+). Plants of the World Online. Facilitated by the Royal Botanic Gardens, Kew. Available from: http://www.plantsoftheworldonline.org/ [6.4.2023].

Rabeler RK, Hartman RL 2005. Caryophyllaceae in Flora of North America North of Mexico, vol. 5. Oxford University Press, New York, 3–8.

Rabeler RK, Wagner WL 2015. Eremogone (Caryophyllaceae): new combinations for Old World species. Phytokeys 50: 35–42. doi: 10.3897/phytokeys.50.4736

Rechinger KH 1964. Arenaria L. In: Rechinger KH (Ed.) Flora of Lowland Iraq. Weinheim, Austria, 230–231.

Rechinger KH 1988. Arenaria L. In: Rechinger KH (Ed.) Flora Iranica (Caryophyllaceae II). Akademische Druck-u Verlagsanstalt, Austria, 6–28.

Sadeghian S, Zarre S, Rabeler RK, Heubl G 2015. Molecular phylogeny of Arenaria (Caryophyllaceae: tribe Arenarieae) and its allies inferred from nuclear DNA ITS and plastid DNA rps16 sequences. Botanical Journal of the Linnaean Society 178: 648–669. doi: 10.1111/boj.12293

Schischkin BK 1995. *Arenaria* L. In: Shishkin BK (Ed.) Flora of the U.S.R.R., Vol. 6. Bishen Singh Mahendra Pal Singh and Koeltz Scientific Books (English version), Moskva-Leningrad, 398–414.

Thiers B 2023 [continuously updated]). *Index herbariorum: A global directory of public herbaria and associated staff*. New York Botanical Garden, Bronx, NY. Available from: http://sweetgum.nybg.org/ih/ [20.09.2023].

DRAFT

ORAL PRESENTATION

Akdeniz midyesinden (*Mytilus galloprovincialis*) üretilen midye sosunun antioksidan aktivitesinin belirlenmesi

İbrahim Ender Künili[1*] (*ORCID: https://orcid.org/0000-0003-2830-6979*), Selin Özge Dinç[2] (*ORCID: https://orcid.org/0000-0003-1597-1929*)

[*1] Çanakkale Onsekiz Mart Üniversitesi, Deniz Bilimleri ve Teknolojisi Fakültesi, Su Ürünleri Avlama ve İşleme Teknolojisi Bölümü, Çanakkale, Türkiye
[2] Çanakkale Onsekiz Mart Üniversitesi, Uygulamalı Bilimler Fakültesi, Gıda Teknolojisi Bölümü, Çanakkale, Türkiye

*Sorumlu yazar e-mail: enderkunili@yahoo.com

Özet

Akdeniz midyesi (*Mytilus galloprovincialis*) besleyici değeri ve lezzeti nedeniyle ekonomik öneme sahip çift kabuklu yumuşakça türüdür. Midyelerin alternatif ürün yelpazesinin sınırlı olmasının yanında ürün üretimleri sırasında sıklıkla kayıplar yaşanabilmektedir. Diğer taraftan, tüketicilerin son yıllarda sağlıklı ve fonksiyonel özelliğe sahip gıda ürünlerini tüketme eğilimi artmış, bu özelliklere sahip ürünlerin iç ve dış pazarda yer edinmesine yönelik çalışmalar önem kazanmıştır. Yapılan bu çalışmada, mevcut midye ürünlerine alternatif olarak midye sosu üretimi gerçekleştirilmiş, elde edilen midye soslarının biyoaktif potansiyeli belirlenmiştir. Taze olarak toplanan midyelerden pilot çalışmalarda belirlenen formülasyonlara göre sos kurulumu yapıldıktan sonra fermantasyona tabi tutulmuştur. Üç ay süren fermantasyon işlemi sonrasında, soslar filtre edilmiş ve elde edilen filtratların DPPH radikallerini, referans antioksidan maddeler olan bütil hidroksi tolüen (BHT) ve Troloks (suda çözünebilen E vitamini analoğu) karşısında, süpürme etkisi belirlenmiştir. Elde edilen bulgulara göre, 2,5 – 40 mg/ml konsantrasyonları arasında BHT, trolox ve midye sosunun DPPH radikallerini süpürme kapasitesi sırasıyla, %19,5 – 94,2, % 18,1 - %93,1 ve %1,8 – 69,5 olarak tespit edilmiştir. Midye soslarının DPPH süpürme kapasitesinin 10 mg/ml konsantrasyondan sonra neredeyse sabit kaldığı belirlenmiştir. Sonuç olarak, alternatif ürün olarak midye sosunun biyoaktif özellikleri potansiyel olarak taşıyabileceği, DPPH radikallerini 10 mg/ml konsantrasyonlara kadar referans antioksidan maddelere benzer oranlarda süpürmesi özelliğinden anlaşılmıştır. Farklı fermantasyon koşullarının denenerek, midye soslarının biyoaktif özelliklerinin artırılabileceği bunun yanı sıra, iç ve dış pazarda yer edebilecek değerli ürünler olabileceği düşünülmektedir.

Anahtar Kelimeler: Akdeniz midyesi, *Mytilus galloprovincialis*, midye sosu, antioksidan aktivite, DPPH

Determination of microbiological quality of stews produced from Mediterranean mussels (*Mytilus galloprovincialis*)

Abstract

Mediterranean mussel (*Mytilus galloprovincialis*) is an economically important bivalve mollusc species due to its nutritive value and flavour. In addition to the limited range of alternative products of mussels, losses can often occur during product production. On the other hand, the tendency of consumers to consume food products with healthy and functional properties has increased in recent years, and studies to place products with these properties in the domestic and foreign markets have gained importance. In this study, mussel sauce production was carried out as an alternative to existing mussel products and the bioactive potential of the obtained mussel sauces was determined. Freshly collected mussels were subjected to fermentation after the sauce was made according to the formulations determined in pilot studies. After three months of fermentation, the sauces were filtered and the scavenging effect of the obtained filtrates on DPPH radicals against the reference antioxidants butyl hydroxy toluene (BHT) and Trolox (water-soluble vitamin E analogue) was determined. According to the results obtained, the DPPH radical scavenging capacity of BHT, trolox and mussel sauce at concentrations of 2.5 - 40 mg/ml was 19.5 - 94.2%, 18.1% - 93.1% and 1.8 - 69.5%, respectively. It was determined that the DPPH scavenging capacity of mussel sauces remained almost constant after 10 mg/ml concentration. As a

result, it was concluded that mussel sauces as an alternative product could potentially carry bioactive properties, as they scavenged DPPH radicals at similar rates to reference antioxidant substances up to 10 mg/ml concentrations. It is thought that by experimenting different fermentation conditions, bioactive properties of mussel sauces can be increased and they can be valuable products that can take place in domestic and foreign markets.

Keywords: Mediterranean mussel, *Mytilus galloprovincialis*, mussel stew, Sous-vide, shelf life

Giriş

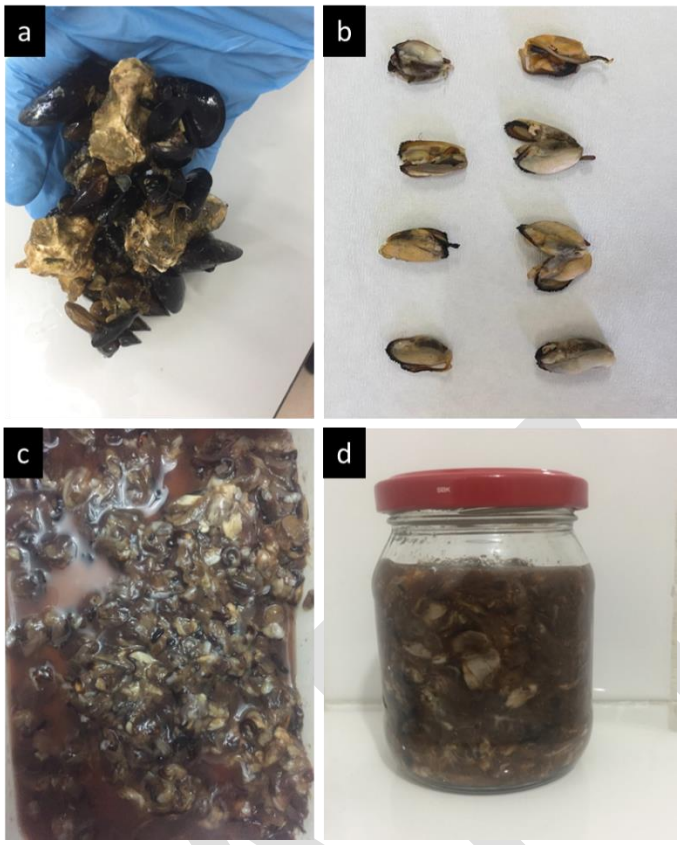
Akdeniz midyesi (*Mytilus galloprovincialis*), farklı mutfak kültürlerinde yer edinmiş, besleyici özellikleri ve lezzet bakımından rağbet gören ekonomik çift kabuklu yumuşakça türüdür. Üretimleri avcılık ve yetiştiricilik yoluyla gerçekleşmektedir. Pazarlık boy için kabul edilebilir kabuk uzunluğu olan 5 cm üzerinde midyeler toplandıktan sonra gerek görüldüğü durumlarda kendi kendilerini arındırmaları işlemi olan depurasyon uygulamasına tabi tutularak veya direkt olarak canlı ticaretleri yapılabilmektedir. Bunun yanında, midye eti çıkarma işlemleri de uygulanabilmekte ve koruyucu özelliklere sahip farklı aroma ve lezzet kazandıran işleme teknikleri ile işlenerek ticareti yapılmaktadır. Midye üretiminde, hasat, boylama, nakliye ve işleme aşamalarında çoğunlukla kabuk kırılmalarına dayanan kayıplar gerçekleşebilmekte yanı sıra, mikrobiyolojik açıdan da riskli üretimin olduğu durumlarda hem zaman hem de ekonomik açıdan kayıplar da görülebilmektedir. Bununla birlikte midye üretiminde alternatif ürün işlemeciliği genellikle tüketici talebi doğrultusunda şekillenmekte, iç ve dış pazarda bu türe ait fonksiyonel ürün çeşitliliği ve bulunabilirliği sınırlı kalmaktadır.

Son yıllarda tüketici gruplarının doğal gıda ürünlerine yönelmesi yanı sıra, güvenli ve biyolojik olarak fayda gösteren fonksiyonel ürünlere talepleri artmıştır. Bu talebe karşılık, gıda üretim firmaları ise, ürünlerinde fonksiyonel özellik gösteren bileşenlerin doğal formlarını veya gıda derecesinde güvenli olan sentetik formlarını ürettikleri ürünlere sonradan ilave ederek fonksiyonellik katmaya çalışmaktadır. Diğer taraftan, doğal gıda üretim metotlarından olan fermantasyon teknolojisi ile üretilen ürünlerin önemi arzu edilen doğallık ve fonksiyonelliğin aynı anda bulunması nedeniyle tüketici tarafından rağbet görmesine neden olmaktadır. Dolasıyla, midye gibi deniz ürünlerinin farklı üretim metotları kullanılarak doğal ve fonksiyonel özellik gösteren ürünlere dönüştürülmesi hem mevcut zaman için, hem de gelecekte önemli bir potansiyel taşımaktadır. Yapılan bu çalışma ile Akdeniz midyesi (*Mytilus galloprovincialis*) kullanılarak fermantasyon yöntemi ile midye sosu üretimi gerçekleştirilmiş, ürünlerin fonksiyonel özelliği ise potansiyel antioksidan aktivite varlığı araştırılarak tespit edilmiştir.

Materyal ve Metot

Araştırma materyali olarak Akdeniz midyesi (*Mytilus galloprovincialis*) kullanılmıştır (Şekil 1a-c). Toplam ağırlığı yaklaşık 3 kg olan ve her birinin boyu 4-6 cm aralığında bulunan örnekler bir yetiştiricilik çiftliğinden temin edilmiştir. Örnekler daha sonra Çanakkale Onsekiz Mart Üniversitesi laboratuvarlarına 4 saat içinde buz aküleri ile soğutulmuş yalıtımlı polietilen kutularda taşınmıştır. Laboratuvara getirilen örneklerin canlılık kontrolleri yapıldıktan sonra kabuklarından etleri ve kabuklar arası sıvısı alınarak fermantasyon için kullanılmıştır.

DRAFT



Şekil 1 – Akdeniz midyesi örnekleri **(a)**, midye etleri **(b)**, midye sosu üretimi için hazırlanan harç **(c)**, fermantasyon için kavanoza alınmış midye sosu harcı **(d)**

Midye sosu üretimi

Midye sosları için ayrılan et ve kabuklar arası sıvı, tuz, şeker, karabiber, kırmızı biber harmanlandıktan sonra el blenderı ile iyice homojenize edilmiştir. Homojenize edilen harç, 200 ml cam kavanozlara 150 olarak şekilde yerleştirilmiş ve fermantasyon işlemi için inkübatörde 30 °C'de üç ay bekletilmiştir. İkinci aydan sonra kavanozların renklerine göre her hafta bir kavanozdan ürün kontrolü yapılarak titrasyon asitliği, pH ölçümü, mikrobiyolojik analizler ve duyusal özelliklere göre kabul edilebilir duruma geldiğinde (üç ay) fermantasyon işlemi sonlandırılmıştır. Fermantasyon işlemi sonrasında olgunlaşmış ürünler nemlendirilmiş filtre kağıdı ile süzülmüş ve elde edilen süzüntü antioksidan analizlerde kullanılmıştır.

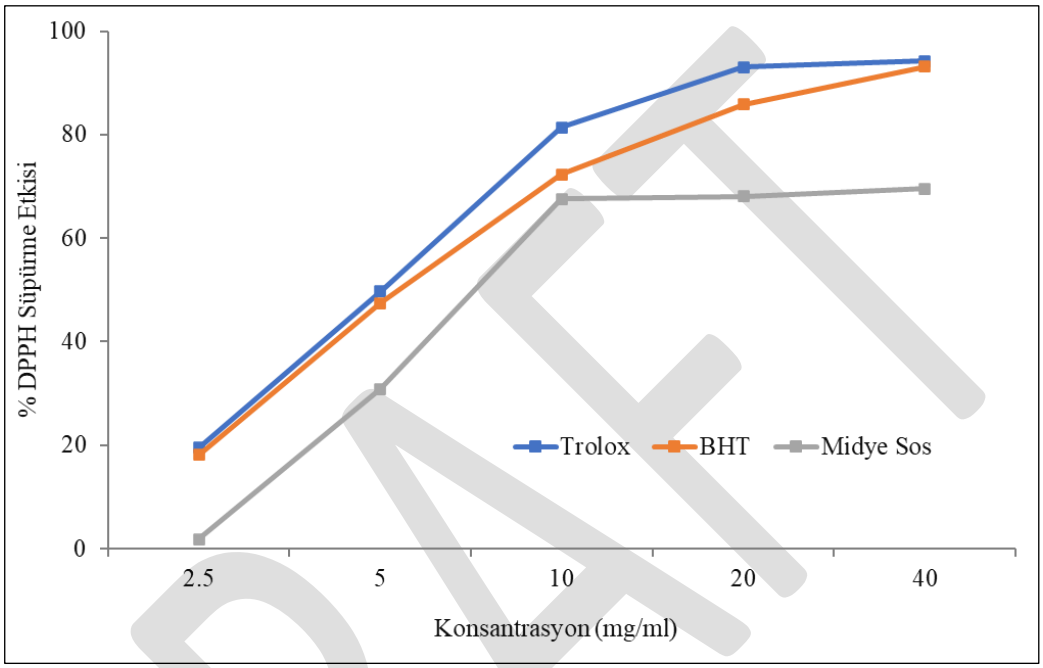
DPPH Süpürme Kapasitesi

Antioksidan aktivite için, bütil hidroksi tolüen (BHT), trolox ve midye sosu örneklerinden 2,5 mg/ml ila 40 mg/ml konsantrasyonlarında alt numuneleri hazırlanmıştır. Hazırlanan farklı konsantrasyonlardaki numunelerden 1ml alınarak, 3 ml hacmindeki 4 mg / 100 ml konsantrasyonunda hazırlanmış 2,2-diphenyl-1-picrylhydrazyl (DPPH) ile karıştırılmıştır. Reaksiyon tüpleri ise karanlık ortamda 30 dk bekletilmiş daha sonra santrifüj edilen örnekler 517 nm dalga boyunda absorbans değerleri ölçülmüştür. Ölçümleri yapılan absorbans değerlerinin yanında, metanol negatif kontrol, BHT ve Trolox pozitif kontrol olarak ölçülmüştür. Elde edilen sonuçlar aşağıdaki formül kullanılarak %DPPH süpürme etkisi olarak ifade edilmiştir.

%DPPH Süpürme Etkisi=$(A_0-(A-A_b)/A_0)*100$
A_0=DPPH Absorbansı
A=Örnek/Trolox/BHT Absorbansı
Ab=Kör (methanol) Absorbansı

Bulgular ve Tartışma

Midye dinamik deniz ortamlarında yaşayan ve oksidatif strese yol açan çeşitli serbest radikallere maruz kalan bir canlıdır. Dolayısıyla bu organizmalar kendi doğal ortamlarında antioksidan ve antioksidan enzim üretimi gibi savunma stratejileri geliştirme eğilimindedir. Bu çift kabuklular insan tüketimi ve biyoaktif bileşikler için mükemmel bir kaynak olduğundan son yıllarda üretim ve tüketimi üzerine yapılan araştırmaların çeşitliliği artmıştır. Bu çalışmada porsiyonluk boylarda bulunan midyelerden fermantasyon yöntemiyle midye sosu üretimi gerçekleştirilmiş, elde edilen ürünlerin ise antioksidan aktivitesi DPPH radikallerini süpürme yeteneği ile belirlenmeye çalışılmıştır. Çalışmada midye sosunun, referans antioksidanlar olan BHT ve troloks karşısındaki antioksidan aktivitesi Şekil 2'de özetlenmiştir.



BHT: Butylated Hydroxy Toluene, DPPH: 2,2-diphenyl-1-picrylhydrazyl.

Şekil 2: Akdeniz midyesinden üretilen sosların BHT ve trolox referans antioksidan maddelere göre DPPH radikallerini süpürme kapasitesi (%).

Elde edilen bulgulara göre, 2,5 – 40 mg/ml konsantrasyonları arasında BHT ve trolox referans bileşiklerinin DPPH radikallerini süpürme etkisi sırasıyla, %19,5 – 94,2 ve % 18,1 - %93,1 aralığında tespit edilmiştir. Midye sosunda ise bu aktivite %1,8 – 69,5 aralığında seyretmiştir. Ayrıca midye soslarının DPPH süpürme etkisinin 10 mg/ml konsantrasyondan sonra neredeyse sabit kaldığı belirlenmiştir. Dolayısıyla alternatif ürün olarak midye sosunun biyoaktif özellikleri potansiyel olarak taşıyabileceği, DPPH radikallerini 10 mg/ml konsantrasyonlara kadar referans antioksidan maddelere benzer oranlarda süpürmesi özelliğinden anlaşılmıştır.

Son yıllarda midye eti üzerine yapılan çalışmalarda tür, mevsim, çevresel yapı gibi faktörlere de bağlı olarak antioksidan, antimikrobiyal ve antiinflamatuar özellikler gibi birçok biyoaktivite belirlenmiştir (Coulson ve ark.,2015; Jayaprakash ve Perera 2020; Peralta ve ark., 2021). Chakraborty ve ark (2016) yeşil midyelerin (*Perna viridis L.*) DPPH radikallerini süpürme etkisini %24-32 aralığında belirlemiştir. Yapılan bir başka çalışmada ise yeşil midyelerin (*Perna viridis L.*) farklı ekstraktlarının DPPH radikalini süpürme aktiviteleri belirlenmiş ve %100 metanol ekstraktının %79,86'lık maksimum DPPH radikal süpürme aktivitesi gösterdiği bildirilmiştir (Shanmugam ve ark. 2020).

Bununla birlikte yapılan çalışmalar sonuçlarımıza benzer şekilde fermente edilen midyelerin biyoaktif özelliklerinin iyileştiğini bildirmiştir (Je ve ark. 2005; Jung ve ark. 2005; Rajapakse ve ark. 2005; Peralta ve ark., 2021). Je ve ark. (2005), mavi midyenin %25 tuzlu ortamda 12 ay fermente edilmesi ile elde edilen midye

sosundan saflaştırılan biyoaktif peptidlerin %89,5 oranında hidroksil radikalini temizleyebilme kapasitesine sahip olduğunu tespit etmiştir. 1:3,1:5, 1:7 ve 1:9 oranında farklı tuzlulukta 30 gün fermente edilen yeşil midyelerin ise farklı oranlarda DPPH süpürme aktivitesi gösterdiği belirlenmiş ve tuzluluğun 1:9 oranında kullanılması ile yaklaşık %80 oranında aktivite belirlenmiştir (Peralta ve ark., 2021).

Sonuç

Sonuç olarak bu çalışma ile doğası gereği hem besleyici ürün profile açısından hem de biyoaktif bileşikler açısından zengin olan midyenin fermente edilmesi ile midye sosu üretilmiştir. Fermantasyon ile üretimin sonucunda oluşan midye soslarında potansiyel antioksidan aktivite varlığı araştırılarak tespit edilmiştir. Farklı fermantasyon koşullarının denenerek, midye soslarının biyoaktif özelliklerinin artırılabileceği bunun yanı sıra, iç ve dış pazarda yer edebilecek değerli ürünler olabileceği düşünülmektedir.

Kaynaklar

Peralta, E. M., Endoma Jr, L. F., Sefil, A. S., & Palmos, G. N. (2021). Chemical and Antioxidant Activity Changes in Philippine Green Mussel (Perna viridis) during Fermentation at Varying Salt Concentrations. *Philippine Journal of Science*, *150*.

COULSON S, PALACIOS T, VITETTA L. 2015. Perna canaliculus (green-lipped mussel): bioactive components and therapeutic evaluation for chronic health conditions. In: Novel Natural Products: Therapeutic Effects in Pain, Arthritis and Gastrointestinal Diseases. Rainsford KD, Powanda MC, Whitehouse MW eds. Basel: Springer Basel. p. 91–132

CHAKRABORTY KSJ, CHAKKALAKAL D, JOSEPH D, ASOKAN PK, VIJAYAN KK. 2016. Nutritional and antioxidative attributes of green mussel (Perna viridis L.) from the Southwestern Coast of India. J Aquat Food Prod Tech 25(7): 968–985.

JAYAPRAKASH R, PERERA CO. 2020. Partial purification and characterization of bioactive peptides from cooked New Zealand green-lipped mussel (Perna canaliculus) protein hydrolyzates. Foods 9(7): 879

SHANMUGAM S, SHANKAR K, RAMACHANDIRAN S, NAIDU K, KALIMUTHU K, MUTHUVEL A. 2020. In vitro studies and characterization of tissue protein from green mussel, Perna viridis (Linnaeus, 1758) for antioxidant and antibacterial potential. Int J Pept Res Ther 26(1): 159–169

JE JY, PARK PJ, BYUN HG, JUNG WK, KIM SK. 2005. Angiotensin I converting enzyme (ACE) inhibitory peptide derived from the sauce of fermented blue mussel, Mytilus edulis. Bioresour. Technol. 96(14): 1624–1629.

JUNG WK, RAJAPAKSE N, KIM SK. 2005. Antioxidative activity of a low molecular weight peptide derived from the sauce of fermented blue mussel, Mytilus edulis. Eur Food Res Technol 220(5): 535–539.

RAJAPAKSE N, MENDIS E, JUNG WK, JE JY, KIM SK. 2005. Purification of a radical scavenging peptide from fermented mussel sauce and its antioxidant properties. Food Res Int 38(2): 175–182.

DRAFT

ORAL PRESENTATION

Hidrojellerin biyomedikal alanda kullanımları

Zeynep Güngör[1]* (ORCID: (0000-0002-2842-1638))

[1*] Çanakkale Onsekiz Mart Üniversitesi, Lisansüstü Eğitim Enstitüsü, Kimya Anabilim Dalı, Çanakkale, Türkiye.

* zeynpgungor@outlook.com, +90 538 470 07 98

Özet

Doğal veya sentetik kaynaklı monomerlerin, farklı çapraz bağlanma yöntemleri ve sentez metotlarıyla çapraz bağlanması sonucu elde edilen hidrojeller, biyomedikal alanda ilk kez Wichterle ve Lim'in poli(2-hidroksietil metakrilat) / p(HEMA) hidrojelleriyle gerçekleştirdikleri çalışmada kontak lens olarak kullanılmıştır. Biyomateryal üretiminde kontak lens kullanımıyla başlayan uygulamalar kontrollü ilaç salım ajanları, akıllı yara örtüleri, implantlar, diş dolguları, doku mühendisliğinde hücre ve doku iskeleleri, yapay organ üretimi gibi birçok alanda kullanılarak oldukça geniş bir kullanıma ulaşmıştır.
Hidrojellerin biyomedikal alandaki kullanım çeşitliliğinin artmasında en önemli faktör olarak hidrofilik içeriği ve yumuşak mekanik yapısıyla biyolojik dokulara benzer bir ortam sunması ve biyouyumluluğunun diğer sert ve sentetik materyallerden daha üstün özellikte olması gösterilebilir.

Anahtar Kelimeler: hidrojel, biyomedikal, biyouyumlu, doku mühendisliği, kontrollü ilaç salımı, biyomateryal

Uses of hydrogels in the biomedical field

Abstract

Hydrogels obtained as a result of cross-linking of natural or synthetic monomers with different cross-linking methods and synthesis methods were used as contact lenses for the first time in the biomedical field in the study carried out by Wichterle and Lim with poly(2-hydroxyethyl methacrylate) / p(HEMA) hydrogels. Applications that started with contact lens usage in biomaterial production have reached comprehensive uses in many areas, such as controlled drug release agents, smart wound dressings, implants, dental fillings, cell and tissue scaffolds in tissue engineering, and artificial organ production.
The most crucial factor in increasing the variety of uses of hydrogels in the biomedical field is that they provide an environment similar to biological tissues with their hydrophilic content and soft mechanical structure, and their biocompatibility is superior to other harsh and synthetic materials.

Keywords: hydrogel, biomedical, biocompatible, tissue engineering, controlled drug release, biomaterial

GİRİŞ

Biyomedikal Alanının Gelişim Süreci

Antik çağlardan bu yana ameliyat ve tedavilerde kullanılmak üzere birçok materyal ve yöntem örneğine rastlanmaktadır(Aljamali & Almuhana, 2021). Başta süngerler olmak üzere deniz hayvanlarından hazırlanan farmasötikler, ilk anestezi cihazı olarak tanımlanan uyku verici bitkilerin emdirilmesiyle kullanılan süngerler gibi uygulamalarla başlayan biyomateryal kullanımı, yapay kalp damarlarının üretildiği beyin ve kalp gibi hassas ve hayati organların takip ve tedavisinde kullanılacak ürünlerin üretimini sağlayan tıp ve mühendislik alanını bir araya getiren biyomedikal mühendisliğin bir çalışma alanı olarak varlığını sürdürmektedir.(Aljamali & Almuhana, 2021; Müller, Batel, Schröder, Müller, & Medicine, 2004)

DRAFT

Ağrılı artritik eklemlerin tedavisindeki eklemin hareketini ve işlevini yeniden sağlamak için korozyona dayanıklı metaller kullanılırken vücut ortamında daha stabil özellik sergileyen seramiklere yönelim gerçekleşmiştir. Biyouyumluluk, biyobozunurluk, mekanik dayanım, maaliyet gibi birçok faktörün gözönünde bulundurulması metal ve seramikler dışında daha kullanışlı materyal arayışlarını doğurmuştur. Kompozit malzemeler, polimerler, polimerik bir yapı olan hidrojeller bu arayışların sonucu olarak biyomateryal üretiminde kullanılmaya başlanmıştır.

Hidrojel Tarihine Genel Bir Bakış

‘’Hidrojel’’ terimi ilk kez 1894’te inorganik tuzlardan oluşan kolloidal jeli tanımlamak için kullanılmıştır. 1960’lardan günümüze ise fiziksel veya kimyasal çapraz bağlanma metotlarıyla elde edilen üç boyutlu polimerik yapıları tanımlamak için kullanılmaktadır. Keşfedildiği günden bu yana sağlıktan teknolojik aletlere, tekstilden tarıma birçok alanda kullanılabilirliği araştırma konusu olmuştur **(Şekil 1).**



Şekil 1. Hidrojellerin uygulama alanlarına genel bir bakış

Üç boyutlu yumuşak ve ağsı yapısı doku benzeri bir yapı sergilediğinden özellikle biyoloji ve tıp alanında dikkat çeken bir materyal olmuştur. Biyouyumlu ve biyobozunur özelliklere sahip örneklerinin de üretilebiliyor oluşu sağlık alanında sıkça araştırma konusu olmasını sağlayan bir diğer unsurdur.

Tablo 1. Hidrojellerin biyomateryal olarak kullanımında sahip olduğu avantaj ve dezavantajlar (Patel, A., & Mequanint, K. (2011), Rajen ve ark. (2022)).

Avantajları	Dezavantajları
Biyouyumluluk	Zayıf mekanik dayanım
Ayarlanabilir biyobozunurluk	Enfeksiyon riski
Su absorplayabilme ve tutabilme	Sık kullanımda maaliyet yüksekliği
Gözenekli yapı	Üretim zorluğu
Fonksiyonel kullanım	Toksisite varlığı (Sentetik kaynaklı hidrojellerde)

HIDROJELLERIN BIYOMEDIKAL ALANDA KULLANIMLARI

Hidrojellerin biyomedikal alanda başlıca kullanım alanlarını şu şekilde sınıflandırmak mümkündür.

1- Kontak lens kullanımı
2- Kontrollü salım ajanı
3- Tıbbi pansuman materyali
4- Doku mühendisliği
5- İmplantlar
6- Biyosensörler

1-Kontak Lens Kullanımı

Hidrojeller biyomedikal alanda ilk kez Wichterle ve Lim'in 1960 yılında gerçekleştirdikleri çalışmada poli(2-hidroksietil metakrilat) / p(HEMA) hidrojellerinin kontak lens olarak kullanılma potansiyeli araştırılmasıyla kullanılmıştır (https://www.nature.com/articles/185117a0) . İlk çalışmalarda poli(metil metakrilat) (PMMA) hidrofilik poli(2-hidroksietil metakrilat (HEMA)) (PHEMA) baz olarak kullanılırken daha sonra oksijen geçirgenliğini arttırmak için silikon hidrojeller geliştirilmiştir.(Shao, Kim, & Imran, 2016).

2- Kontrollü Salım Ajanı Olarak Kullanımı

Kontrollü ilaç salımı yapan sistemler geleneksel ilaç kullanımının aksine sadece hedef organda istenilen dozaj ve sürede ilacın salımını gerçekleştirerek oluşabilecek yan etkileri azaltacak ve gereksiz ilaç yükünü önleyecek bir imkan sunmaktadır. Antibiyotik, analjezik türde ilaçların yanı sıra hormon, ünsilin, kök hücre gibi biyoaktif moleküllerinde kontrollü salımını gerçekleştiren birçok hidrojel sistemin araştırıldığı çalışmalar mevcuttur.(Kaynak)

3- Tıbbi Pansuman Materyali

Hidrojeller, su ve vücut sıvısı gibi biyolojik sıvıları absorplayabilen gözenekli yapısıyla tıbbi uygulamalar için kullanışlı bir zemin sunmaktadır. Hidrojellerin tıbbi alanda kullanılacak materyallerden istenilen biyouyumluluk, oksijen geçirgenliği, antibakteriyel yüzey gibi özellikleri bünyesinde barındırabiliyor oluşu bu alanda birçok hidrojel sistemin üretilip araştırılmasını sağlamıştır. Yara iyleşme sürecini destekleyen akıllı yara örtüsü kullanımının dışında özelikle baskı uygulanamayacak organlardaki kanamalara müdahele de iyi bir hemostatik materyal olma potansiyeline sahip olduğu yapılan çalışmalarla kanıtlanmıştır(Cheng et al., 2021; Dong et al., 2022).

4- Doku Mühendisliği

Doku mühendisliği örneklerine 1933 yıllarında Bisceglie'nin fare tümör hücrelerini bir polimer membranla kaplayıp bunları bir domuzun karın boşluğuna yerleştirdiği çalışmasıyla rastlansa da multidisipliner bir mühendislik dalı olarak anılmaya 1980'lerin ortalarında Dr. Joseph Vacanti ve Dr. Robert Langer'ın dokuya benzer işlevler gösteren ağ yapılı sentetik biyouyumlu /biyobozunur polimer temelli iskeleler üzerine yaşayabilen hücreleri ekerek gerçekleştirdikleri çalışmaları ile başlamıştır(Bisceglie, 1934; C. Vacanti & Vacanti, 1991; C. A. J. J. o. c. Vacanti & medicine, 2006).

Hidrojellerin biyouyumlu ve biyobozunur yapıları, yüksek su içeriğine sahip oluşu ve ağsı yapısı hücrelerin çoğalması için uygun bir ortamın oluşmasının önünü açarak doku mühendisliğinde başarılı doku iskelelerinin üretilmesini sağlamaktadır. Hidrojellerin enjekte edilip yerinde jelleşebilen formları ise nakli hedeflenen hücreler için daha uygun bir ortam sağlayarak (kalp, kemik, kıkırdak, sinir gibi) doku onarımında önemli bir ajan olarak yer edinmeye başlamıştır. Ayrıca hücre dışı matris özelliklerini taklit eden hidrojellerle elde edilen 3boyutlu hücre kültürleri ileri düzey tıbbi ve biyolojik araştırmaları destekleyen verimli çalışma ortamları sunmaktadır (Ansar et al., 2022; Lee, 2018; C. A. J. J. o. c. Vacanti & medicine, 2006).

5- İmplantlar

İmplantlar kalp pilleri, eklem protezleri, göğüs implantları, ventriküler şantlar ve yapay kalp kapakçıkları gibi vücutta geçici veya kalıcı olarak kullanıldığında tedavi ve yaşam kalitesini artırmayı hedefleyen tıbbi cihazlardır (Ramon ve ark., 2021).
Hidrojeller metal, seramik, alaşım bazlı implantlara göre biyouyumluluğu yüksek bir materyaldir. Bu özelliğinden faydalanmak için implant yüzeyleri elektrokimyasal yöntem, sol-jel yöntemi gibi genel modifikasyon yöntemleri kullanılarak hidrojel filmlerle kaplanabilmektedir ((Alavi et al., 2022).Hidrojellerin yumuşak ve esnek yapıları sayesinde beyin ve kalp gibi hassas dokulara sahip organların tedavi ve takibinde kullanılabilecek soft hidrojel elektrotlar üretilmiştir(Tringides et al., 2021) Geliştirilmekte olan bir kullanım alanı da üç boyutlu yazdırılabilir hidrojeller ile insan dokusuna en yakın doku ve organların üretilmesidir ((Panja, Maji, Choudhuri, Ali, & Hossain, 2022) (Xie, Su, Zhou, Li, & Zhang, 2023).

6-Biyosensörler

İlk kez 1977 yılında Carl Kamman tarafından kullanılan "Biyosensör" terimi reaksiyondaki analitin konsantrasyonuyla orantılı olarak sinyaller üreterek biyolojik veya kimyasal reaksiyonları ölçen bir cihazı tanımlamaktadır. Hidrojeller biyosensör uygulamalarında kaplama malzemesi olarak, substratın olarak veya biyomoleküllerin ağ içinde hapsedileceği kapsülleme matrisi olarak kullanılabilmektedir. (Bhalla & Singh, 2016; Herrmann, Haag, & Schedler, 2021).

TARTIŞMA VE SONUÇ

Keşfedildiği günden günümüzde biyolojik sistemlere benzerliği ve uyumluluğuyla dikkatleri üzerine çeken hidrojel sınıfı yapılan çalışmalarla biyomedikal alandaki ihtiyaçlara cevap verebilecek potansiyelde biyouyumlu ürünlerin eldesini sağlayacağını gösterse de mekanik dayanım, toksisite ve sterilizasyon zorluğu gibi konularda aşılması gereken problemlere sahiptir.

KAYNAKLAR

Alavi, S. E., Panah, N., Page, F., Gholami, M., Dastfal, A., Sharma, L. A., & Shahmabadi, H. E. J. E. P. J. (2022). Hydrogel-based therapeutic coatings for dental implants. 111652.

Aljamali, N. M., & Almuhana, W. H. Y. J. J. o. A. i. E. D. (2021). Review on biomedical engineering and engineering technology in bio-medical devices. *6*(2), 18-24.

Ansar, R., Saqib, S., Mukhtar, A., Niazi, M. B. K., Shahid, M., Jahan, Z., . . . Ullah, S. J. C. (2022). Challenges and recent trends with the development of hydrogel fiber for biomedical applications. *287*, 131956.

Bhalla, P., & Singh, N. J. T. E. P. J. B. (2016). Generalized Drude scattering rate from the memory function formalism: an independent verification of the Sharapov-Carbotte result. *89*, 1-8.

Bisceglie, V. J. Z. f. K. (1934). Über die antineoplastische Immunität: I. Mitteilung. Heterologe Einpflanzung von Tumoren in Hühnerembryonen. *40*(1), 122-140.

Cheng, J., Liu, J., Li, M., Liu, Z., Wang, X., Zhang, L., . . . Biotechnology. (2021). Hydrogel-based biomaterials engineered from natural-derived polysaccharides and proteins for hemostasis and wound healing. *9*, 780187.

Dong, H., Wang, L., Du, L., Wang, X., Li, Q., Wang, X., . . . Ma, G. J. S. (2022). Smart polycationic hydrogel dressing for dynamic wound healing. *18*(25), 2201620.

Herrmann, A., Haag, R., & Schedler, U. J. A. h. m. (2021). Hydrogels and their role in biosensing applications. *10*(11), 2100062.

Lee, J. H. J. B. r. (2018). Injectable hydrogels delivering therapeutic agents for disease treatment and tissue engineering. *22*(1), 1-14.

Müller, W. E., Batel, R., Schröder, H. C., Müller, I. M. J. E.-B. C., & Medicine, A. (2004). Traditional and modern biomedical prospecting: Part I—the history. *1*, 71-82.

Panja, N., Maji, S., Choudhuri, S., Ali, K. A., & Hossain, C. M. J. A. P. (2022). 3D bioprinting of human hollow organs. *23*(5), 139.

Patel, A., & Mequanint, K. (2011). Hydrogel Biomaterials. InTech. doi: 10.5772/24856.

Ramon Z. Shaban, Brett G. Mitchell, Philip L. Russo, Deborough Macbeth. (2021) CHAPTER 8 - Infection associated with prosthetics and implantable devices, Editor(s): RAMON Z. SHABAN, BRETT G. MITCHELL, PHILIP L. RUSSO, DEBOROUGH MACBETH, Epidemiology of Healthcare-associated Infections in Australia, Elsevier, Pages 85-95.

Rajen Kundu, Pushpa Mahada, Bhawna Chhirang, Bappaditya Das. (2022). Cellulose hydrogels: Green and sustainable soft biomaterials, Current Research in Green and Sustainable Chemistry, (5), 100252.

Tringides, C. M., Vachicouras, N., de Lázaro, I., Wang, H., Trouillet, A., Seo, B. R., . . . Casiraghi, C. J. N. n. (2021). Viscoelastic surface electrode arrays to interface with viscoelastic tissues. *16*(9), 1019-1029.

Vacanti, C., & Vacanti, J. J. S. t. i. (1991). Functional Organ Replacement, The New Technology of Tissue Engineering. *1*, 43-49.

Vacanti, C. A. J. J. o. c., & medicine, m. (2006). The history of tissue engineering.

Xie, M., Su, J., Zhou, S., Li, J., & Zhang, K. J. G. (2023). Application of Hydrogels as Three-Dimensional Bioprinting Ink for Tissue Engineering. *9*(2), 88.

DRAFT

ORAL PRESENTATION

Yara teşhis ve bakımında hidrojeller

Zeynep Güngör[1*] (*ORCID: (0000-0002-2842-1638)*)

[1*] Çanakkale Onsekiz Mart Üniversitesi, Lisansüstü Eğitim Enstitüsü, Kimya Anabilim Dalı, Çanakkale, Türkiye.

* zeynpgungor@outlook.com, +90 538 470 07 98

Özet

Hidrojeller, yapısındaki hidrofilik gruplar sayesinde kuru kütlelerinin binlerce katı sıvıyı (su, vücut sıvıları vb.) absorplayabilen üç boyutlu polimerik yapılardır. Biyomedikal alanda kullanımı 1960'lı yıllarda HEMA hidrojellerinin kontak lens olarak kullanımının araştırılmasıyla başlamıştır. Kontak lens olarak kullanımıyla başlayan uygulamaları, kontrollü ilaç salımı, sensör kullanımı, akıllı tekstil ürünleri, su arıtım sistemleri, doku hücre iskelesi, akıllı yara örtüleri ve yapay damar üretimine kadar birçok alana yayılarak günümüzde en çok araştırılan malzemeler sınıfına eriştiği söylenebilir.
Biyolojik alanda kullanılan materyallerin antibakteriyel özellikte olması ve biyouyumlu olmaları istenilen en önemli özelliklerdendir. Hidrojellerin sentezinde kullanılan polimerik yapıların karakteristik özelliğiyle biyouyumluluk özelliği kazandırılabilirken hidrojel yapısına katılan veya yüklenen ilaçlarla ise tedavi edici materyal olarak kullanımı sağlanabilir.
Yara bakım süreci yaranın iyileşmesi için gerekli optimum şartların oluşmasında önemli bir faktördür. Bu şartların oluşmasında en önemli etkenlerden biri yara tedavi ve pansumanında kullanılacak materyal türüdür. Çalışmamız hidrojellerin yara teşhis ve bakımında ne gibi avantaj ve dezavantajlara sahip olduğuna dair bir bakış açısı kazandırmayı hedeflemektedir.

Anahtar Kelimeler: hidrojel, yara bakımı, biyomateryal, akıllı yara örtüsü

Hydrogels in wound diagnosis and care

Abstract

Hydrogels are three-dimensional polymeric structures that can absorb thousands of times more liquid (water, body fluids, etc.) than their dry mass, thanks to the hydrophilic groups in their structure. Its use in the biomedical field began in the 1960s with the investigation of the use of HEMA hydrogels as contact lenses. It can be said that its applications, which started with its use as contact lenses, have spread to many areas such as controlled drug release, sensor use, smart textile products, water purification systems, tissue cell scaffolds, smart wound dressings and artificial vein production, and have reached the most researched materials class today.
The most important desired features of the materials used in the biological field are that they have antibacterial properties and are biocompatible. While biocompatibility can be achieved with the characteristic feature of the polymeric structures used in the synthesis of hydrogels, it can be used as a therapeutic material with drugs added or loaded into the hydrogel structure.
The wound care process is an important factor in creating the optimum conditions for wound healing. One of the most important factors in the formation of these conditions is the type of material to be used in wound treatment and dressing. Our study aims to provide a perspective on what advantages and disadvantages hydrogels have in wound diagnosis and care.

Keywords: hydrogel, wound care, biomaterial, smart wound dressing

DRAFT

GİRİŞ

Yara Bakim Sürecine Genel Bakiş

Yaralanma, çeşitli fiziksel travmalar veya hastalıklar gibi birçok farklı nedenle bir hücrenin, dokunun veya organizmanın bütünlüğünün zarar görerek yaşayabilirliğinin veya temel fonksiyonlarının bozulmasına sebep olan değişiklik olarak tanımlanabilir (Eming ve ark., 2014, Baktır, 2020, Velnar, 2009)

Literatürde yara tedavisinde kullanılan yöntemlere ait ilk bilgilerin en eski tıbbi el yazmalarından M.Ö. 2200 yıllarına dayanan kil tabletlerdeki olduğu bildirilmiştir. Bu tabletlerde "üç iyileştirme hareketi" olarak ''yaraları yıkamak, alçıyı yapmak ve yarayı sarmak'' ifadeleri geçmektedir (Shah, J. B. (2011)). Bu ifadelerindende anlaşılacağı üzere eski çağlardan itibaren yarayı korumak ve yara sıvısını emmesi amacıyla çeşitli pansuman materyallerin kullanımı süregelmiştir (Sood, A. ve ark., 2014). Bu materyaller zamanla yarayı korumak dışında daha fonksiyonel, yara iyileşme sürecini de destekleyecek biyoaktif malzemelere dönüştürülmüştür (Ovington, L. G. (2007)).

Yaranın iyileşme süreci genellikle aşağıda maddeler halinde belirtilmiş olan dört aşamaya ayrılmıştır (Velnar ve ark., (2009), Raziyeva ark. (2021)).

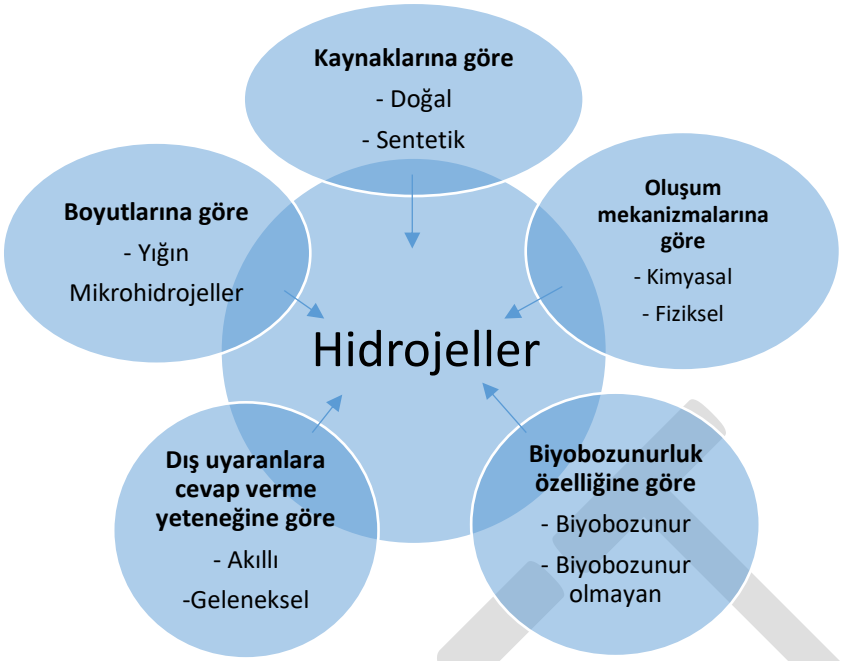
(i) Pıhtılaşma ve hemostaz
(ii) İltihaplanma
(iii)Çoğalma ve
(iv) Skar (yara izi) dokusu oluşumu ile yaranın yeniden şekillenmesi olarak

HIDROJELLER

Hidrojeller, tanımlarken incelendikleri alana göre çeşitli sınıflandırmaların kullanılabileceği üç boyutlu polimerik yapılardır **(Şekil 1)** (Peppas ve ark. (2012)). Birbirinden farklı birçok alanda kullanılabilecek potansiyel ve fonksiyonellikte olması popular bir çalışma alanı olmasını sağlamıştır (Ahmed, 2015).
Bünyesinde kütlesinin binlerce katı suyu tutabiliyor oluşu, doku benzeri yumuşak ve ağsı yapısının yanı sıra biyoyumlu ve biyobozunur özelliğe sahip örneklerinin üretilebilmesi, daha çok tıp ve biyoloji uygulamalarına yönelik araştırmaların gerçekleştirilmesine sebep olduğu söylenebilir (Lei ve ark. 2022).

Hidrojellerin tıbbi pansuman materyali olarak kullanılmasında da hidrofilik yapıları, hücre dışı matriks (ECM) gibi üç boyutlu (3D) gözenekli bir yapıyla dokulara benzer bir ortam oluşturması ve biyouyumlulukları etkin rol oynamaktadır (Mohammad ve ark., 2023, Abasalizadeh ve ark. 2020). Ayrıca kontrollü salım sistemlerinde de kullanılabiliyor oluşları, yaranın iyileşme sürecini destekleyen daha etkili yara örtülerinin üretilebilmesini sağlamaktadır (Fan ve ark. 2021)

DRAFT



Şekil 1. Hidrojellerin sınıflandırılması

1-Kontrollü Salim Yapan Hidrojellerin Yara Örtüsü Olarak Kullanimlari

Antibakteriyel hidrojel yara örtüsü üzerine yapılan çalışmalarda amoksisilin, ampisilin, tetrasiklin, gentamisin, siprofloksasin, moksifloksasin, kloramfenikol, sülfadiazin, linezolid gibi birçok antibakteriyel ilacın kullanımının yanı sıra gümüş, altın, bakır, çinko gibi inorganik metallerin antibakteriyel etkisi incelenmiştir (Liang ve ark. (2021), Shi ve ark. (2018)).
Hidrojeller sadece antibiyotik ilaç salımında değil, iyileşme sürecini olumlu yönde etkileyip hızlandıracak kök hücre, büyüme faktörü gibi biyoaktif moleküller, polipeptitler gibi moleküllerin salımında da kullanılarak doku onarımını teşvik eden yara örtüsü olarak birçok çalışmada incelenmiştir (Shin ve ark., (2019), Catoira ve ark., (2019)). Kronik yaralar ve yanıklar gibi ciddi enfeksiyon riski taşıyan yaralar için gümüş gibi antibakteriyel öğeler içeren hidrojellerin kullanımı dair çalışmalarda mevcuttur ((Rybka ve ark. (2022) Yao ve ark. (2021), Liang ve ark. (2021)).

2- Hemostatik malzeme olarak kullanimlari

Hidrojeller ayrıca iyi birer hemostatik malzeme olmakta da büyük bir potansiyele sahiptir. Açık yaralarda sıklıkla tercih edilen geleneksel malzemeler, iç kanamalı, baskı uygulanamayacak dokular için elverişsizdirler (Cheng ve ark., (2021)). Hidrojellerin enjekte edildiği yerde jelleşebilen örnekleri iç kanamalar ve baskı uygulanamayan yaralar için büyük bir avantaj sunmaktadır (Han ve ark., (2022), Bertsch ve ark. (2022)).

3- Yara takip süreci

Sağlıklı bir deri pH=4-6 aralığında bir pH değerine sahipken cilt hasar aldıktan sonra oluşan fizyolojik sıvıların ve enfeksiyon etkeniyle birlikte pH=9'a kadar yükselebilir (Arafa ve ark., (2021)). pH değişimine kolorimetrik olarak cevap verme yeteneğine sahip hidrojel sınıfıyla, yaralardaki bu pH değişimi göz önüne alındığında, yara takibini de sağlayabilecek akıllı bir yara örtüsü üretilebileceği düşünülmektedir (Arafa ve ark., (2021),Güngör, Z., & Ozay, H. (2022), Liu ve ark., (2017).

TARTIŞMA VE SONUÇ

Hidrojel pansuman materyaller, biyouyumlulukları, esnek yapıları, antibakteriyel özellikleri ve iyileştirici faktör olarakta kullanılabiliyor oluşlarının yanı sıra ıslak yüzeyde yapışkanlıklarının düşük oluşu, mekanik dayanımları, aşırı drenajlı yaralar için uygun olmamaları gibi zayıf yönlere de sahiptir. Bu alanda gerçekleştirilen birçok çalışma mevcut olsa da yara iyileşme süreci gibi karışık ve dinamik bir sürecin daha etkili yönetilmesini sağlamak için in vivo ortamda daha kapsamlı ve derinlemesine çalışmalara ihtiyaç olduğu söylenebilir. Yara takip sürecinde kullanılması hedeflenen kolorimetrik hidrojellerle ilgili ise henüz sınırlı sayıda araştırma mevcuttur.

KAYNAKLAR

Abasalizadeh, F., Moghaddam, S.V., Alizadeh, E. et al. Alginate-based hydrogels as drug delivery vehicles in cancer treatment and their applications in wound dressing and 3D bioprinting. J Biol Eng 14, 8 (2020).

Ahmed, E. M. (2015). Hydrogel: Preparation, characterization, and applications: A review. Journal of advanced research, 6(2), 105-121.

Arafa, A. A., Nada, A. A., Ibrahim, A. Y., Sajkiewicz, P., Zahran, M. K., & Hakeim, O. A. (2021). Preparation and characterization of smart therapeutic pH-sensitive wound dressing from red cabbage extract and chitosan hydrogel. International Journal of Biological Macromolecules, 182, 1820-1831.

Baktır, G. (2020). Wound repair and experimental wound models. Experimed, 9(3), 130-137.

Bertsch, P., Diba, M., Mooney, D. J., & Leeuwenburgh, S. C. (2022). Self-healing injectable hydrogels for tissue regeneration. Chemical Reviews, 123(2), 834-873.

Catoira, M. C., Fusaro, L., Di Francesco, D., Ramella, M., & Boccafoschi, F. (2019). Overview of natural hydrogels for regenerative medicine applications. Journal of Materials Science: Materials in Medicine, 30, 1-10.

Cheng, J., Liu, J., Li, M., Liu, Z., Wang, X., Zhang, L., & Wang, Z. (2021). Hydrogel-based biomaterials engineered from natural-derived polysaccharides and proteins for hemostasis and wound healing. Frontiers in Bioengineering and Biotechnology, 9, 780187.

Eming, S. A., Martin, P., & Tomic-Canic, M. (2014). Wound repair and regeneration: mechanisms, signaling, and translation. Science translational medicine, 6(265), 265sr6-265sr6.

Fan, F., Saha, S., & Hanjaya-Putra, D. (2021). Biomimetic hydrogels to promote wound healing. Frontiers in Bioengineering and Biotechnology, 9, 718377.

Güngör, Z., & Ozay, H. (2022). Ultra-fast pH determination with a new colorimetric pH-sensing hydrogel for biomedical and environmental applications. Reactive and Functional Polymers, 180, 105398.

Han, W., & Wang, S. (2022). Advances in hemostatic hydrogels that can adhere to wet surfaces. Gels, 9(1), 2.

Lei, L., Bai, Y., Qin, X., Liu, J., Huang, W., & Lv, Q. (2022). Current understanding of hydrogel for drug release and tissue engineering. Gels, 8(5), 301.

Liang, Y., He, J., & Guo, B. (2021). Functional hydrogels as wound dressing to enhance wound healing. ACS nano, 15(8), 12687-12722.

Liu, L., Li, X., Nagao, M., Elias, A. L., Narain, R., & Chung, H. J. (2017). A pH-Indicating colorimetric tough hydrogel patch towards applications in a substrate for smart wound dressings. Polymers, 9(11), 558.

Mohammad Hadi Norahan, Sara Cristina Pedroza-González, Mónica Gabriela Sánchez-Salazar, Mario Moisés Álvarez, Grissel Trujillo de Santiago, Structural and biological engineering of 3D hydrogels for wound healing, Bioactive Materials, Volume 24, 2023, Pages 197-235.

N.A. Peppas, B.V. Slaughter, M.A. Kanzelberger, 9.20 - Hydrogels, Editor(s): Krzysztof Matyjaszewski, Martin Möller, Polymer Science: A Comprehensive Reference, Elsevier, 2012, Pages 385-395.

Raziyeva, K., Kim, Y., Zharkinbekov, Z., Kassymbek, K., Jimi, S., & Saparov, A. (2021). Immunology of acute and chronic wound healing. Biomolecules, 11(5), 700.

Rybka, M., Mazurek, Ł., & Konop, M. (2022). Beneficial Effect of Wound Dressings Containing Silver and Silver Nanoparticles in Wound Healing—From Experimental Studies to Clinical Practice. Life, 13(1), 69.

Shah, J. B. (2011). The history of wound care. The Journal of the American College of Certified Wound Specialists, 3(3), 65-66.

Shi, G., Chen, W., Zhang, Y., Dai, X., Zhang, X., & Wu, Z. (2018). An antifouling hydrogel containing silver nanoparticles for modulating the therapeutic immune response in chronic wound healing. Langmuir, 35(5), 1837-1845.

Shin, J., Choi, S., Kim, J. H., Cho, J. H., Jin, Y., Kim, S., ... & Cho, S. W. (2019). Tissue Tapes—phenolic hyaluronic acid hydrogel patches for off-the-shelf therapy. Advanced Functional Materials, 29(49), 1903863..

Sood, A., Granick, M. S., & Tomaselli, N. L. (2014). Wound dressings and comparative effectiveness data. Advances in wound care, 3(8), 511-529.)

Ovington, L. G. (2007). Advances in wound dressings. Clinics in dermatology, 25(1), 33-38.)

Velnar, T., Bailey, T., & Smrkolj, V. (2009). The wound healing process: an overview of the cellular and molecular mechanisms. Journal of international medical research, 37(5), 1528-1542.

Yao, Y., Zhang, A., Yuan, C., Chen, X., & Liu, Y. (2021). Recent trends on burn wound care: Hydrogel dressings and scaffolds. Biomaterials science, 9(13), 4523-4540.

DRAFT

ORAL PRESENTATION

Kuşburnunun Biyoaktif Bileşenleri ve Sağlık Üzerine Etkileri

Tuba Eda Arpa Zemzemoğlu[1] (*ORCID: https://orcid.org/0000-0002-6836-4527*), Huri İlyasoğlu[1] (*ORCID: https://orcid.org/0000-0001-5710-2954*), Nesibe Arslan Burnaz[1*] (*ORCID: https://orcid.org/0000-0003-1163-4829*)

[*1]Gümüşhane Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Gümüşhane, Türkiye

*Sorumlu yazar e-mail: nsbburnaz@gmail.com

Özet

Günümüzde sağlık bilincine sahip tüketicilerin sayısının artmasıyla birlikte fonksiyonel ve sağlıklı gıda ürünlerine olan talep artmaktadır. Kuşburnu meyvesinin içermiş olduğu biyoaktif bileşenlerin fazla olması nedeniyle katma değeri yüksek ürünlere işlenme potansiyeli bulunmaktadır. Bu derlemenin amacı, kuşburnunun besin değeri, kimyasal bileşimi, gıda endüstrisindeki potansiyel uygulamaları ve sağlık açısından faydaları ile ilgili kapsamlı genel bir bakış sunmaktır. Kuşburnu, başta doğal antioksidanlar (C vitamini, polifenoller, karotenoidler ve tokoferoller) olmak üzere, benzersiz antioksidan, antiinflamatuar, antimikrobiyal, antidiyabetik ve osteoprotektif aktivitelere sahip değerli bir biyoaktif bileşik kaynağıdır. Kuşburnu ve ürünlerinin sağlık yararlarına ilişkin daha kapsamlı çalışmalar yapılmasına ihtiyaç vardır. Benzer şekilde biyoaktif bileşenler içeren ve sağlığa fayda sağlayacak doğal gıda kaynakları belirlenmeli ve elde edilecek ürün çeşitliliği artırılmalıdır.

Anahtar Kelimeler: Kuşburnu, *Rosa canina* L., sağlık

Bioactive Components of Rosehip and Their Effects on Health

Abstract

Today, with the increasing number of health-conscious consumers, the demand for functional and healthy food products has been increased. Due to the excess of bioactive components in rosehip fruit, there is a potential for processing into products with high added values. The purpose of this review is to provide a comprehensive overview of the nutritional value, chemical composition, potential applications in the food industry and health benefits of rosehip. Rosehip is a valuable source of bioactive compounds with unique antioxidant, antiinflammatory, antimicrobial and osteoprotective activities, mainly natural antioxidants (vitamin C, polyphenols, carotenoids, and tocopherols). More comprehensive studies are needed to reach the health benefits of rosehip and its products. Similarly, natural food sources with bioactive components providing health benefits should be identified and the variety of obtained products should be increased.

Keywords: Rosehip, *Rosa canina* L., health

GİRİŞ

Kuşburnu Avrupa, Batı Asya ve kuzeybatı Afrika'da yetişen çalı formunda yabani bir bitkidir. Kuşburnu meyveleri vitamin ve mineral bakımından oldukça zengin olup, C vitamini miktarı diğer meyve ve sebze türlerine göre nispeten daha yüksektir. Kuşburnu meyveleri karotenoidler ve fenolik bileşikler gibi antioksidan özellik gösteren bileşenleri de içermektedir. Kuşburnu meyveleri halk hekimliğinde uzun süreden beri kullanılmaktadır. Kuşburnu soğuk algınlığı, bulaşıcı hastalıklar, mide-bağırsak rahatsızlıkları, idrar yolu hastalıkları ve inflamatuar hastalıklara karşı profilaktik ve tedavi edici etkilere sahiptir. Kuşburnu yüksek miktarda askorbik asit içermesi nedeniyle Avrupa'da bitki çayı ve vitamin takviyesi olarak kullanılmaktadır. Kuşburnu reçel, marmelat, nektar ve sirke gibi ürünlere de işlenmektedir. Kuşburnu çekirdeği yağı, cilt üzerindeki tedavi edici etkisinden dolayı kozmetikte kullanılmaktadır (Ilyasoglu, 2014, Zhou 2023).

DRAFT

Kuşburnunun önemi dikkate alınarak bu derlemede kuşburnunun besin değeri, kimyasal bileşimi, gıda endüstrisindeki potansiyel uygulamaları ve sağlık açısından faydaları üzerinde durulacaktır.

KUŞBURNU

Kuşburnu *Rosaceae* familyasından *Rosa* cinsine ait, kışın yaprağını döken çalı formunda yabani bir bitkidir. Rosa cinsi 200'den fazla tür ve 40.000 çeşitten oluşur ve ılıman bölgelerden subtropikal bölgelere kadar geniş bir dağılım göstermektedir. Avrupa, Batı Asya ve Kuzeybatı Afrika'da yetişmektedir. Rosa cinsi bitkiler süs bitkisi olarak da yetiştirilmektedir (Zhou ve ark., 2023). Ülkemizde de pek çok yörede yetişen kuşburnu halk dilinde Yabangülü, Deligül, Gül burnu ve Gül elması olarak da bilinmektedir. *Rosa canina* L. ülkemizde yetişen kuşburnu türleri arasında meyve özellikleri bakımından işlenmeye en uygun türlerden birisidir. Orman açıklıklarında ve kayalık yamaçlarda yetişmektedir (Öz ve ark., 2018). Kuşburnu meyveleri; sağlık üzerine olumlu etkileri olduğu bilinen biyoaktif bileşenler içermesi nedeniyle büyük ilgi görmektedir. *R. canina* L.'nin meyveleri 2000 yılı aşkın bir süredir tıbbi bir bitki olarak kullanılmaktadır (Zhou ve ark., 2023).

Kuşburnu meyvesi makro besin öğesi olarak karbonhidrat açısından zengindir. Portekiz'de yapılan bir çalışmada *Rosa canina* L.'nin karbonhidrat içeriği % 93,16, protein içeriği % 2,72, yağ içeriği % 0,65 ve kül içeriği % 3,47 olarak bulunmuştur. Şeker olarak % 12,89 oranında fruktoz, % 12,17 oranında glukoz ve % 1,83 oranında sakaroz içerdiği belirlenmiştir (Barros ve ark., 2010). Türkiye'de yapılan bir çalışmada ise fruktoz miktarı % 18,84 ve glukoz miktarı % 17,11 olarak bulunmuştur (Demir ve ark., 2014). Kuşburnu meyveleri selüloz ve nişasta gibi polisakkaritleri de içermektedir (Baros ve ark., 2010).

Kuşburnu meyvesi mikro besin öğesi olarak askorbik asit açısından zengindir. Kuşburnunun askorbik asit içeriği meyve çeşidi, hasat yılı ve iklim koşullarına bağlı olarak değişkenlik göstermektedir. Türkiye'de yapılan bir çalışmada *R. canina* L.'nin askorbik asit içeriği 101 mg/100 g, Portekiz'de yapılan bir çalışmada ise 68 mg/100 g olarak bulunmuştur (Baros ve ark., 2010; Demir ve ark., 2014).

Kuşburnu A, C ve E vitaminlerinin yanısıra kalsiyum, potasyum, sodyum, fosfor, magnezyum, manganez, demir, çinko ve bakır gibi mineralleri içermektedir (Fan ve ark., 2014). Kuşburnu meyveleri potasyum ve kalsiyum açısından zengin bir meyvedir. *R. canina* L.'nin 4 genotipinin mineral madde içeriğinin belirlendiği bir çalışmada potasyum miktarı 8964-10450 mg/kg, kalsiyum miktarı 4988-17617 mg/kg, fosfor miktarı 1144-1769 mg/kg, magnezyum miktarı 1415-3135 mg/kg, demir miktarı 53-193 mg/kg, manganez miktarı 33-128 mg/kg, bakır miktarı 4,3-5,8 mg/kg ve çinko miktarı 5,1-12,5 mg/kg olarak bulunmuştur (Koç, 2020).

Meyvelerin tadını meyvelerdeki şeker ve organik asit oranı belirlemektedir. Kuşburnu meyvelerinde sitrik, malik, tartarik, süksinik, okzalik ve fumarik asit gibi organik asitler tespit edilmiştir (Murathan ve ark., 2016a**).**

Kuşburnu biyoaktif bileşenler içermektedir (Tablo 1). Kuşburnu meyvelerinin içerdiği biyoaktif bileşenlerden birisi karoteonoidlerdir. Barros ve ark. (2010) tarafından yapılan çalışmada β-karoten ve likopen tespit edilmiştir. Bir başka çalışmada ise β-karoten ve likopenin yanısıra lutein ve zeaksantin tespit edilmiştir (Fan ve ark., 2014).

Kuşburnu biyoaktif bileşikler açısından zengin olup biyoaktif bileşiklerin önemli bir bölümünü fenolik bileşikler oluşturmaktadır. Demir ve ark. (2014) tarafından yapılan çalışmada *Rosa canina* L.'de 12 tane bileşik tespit edilmiştir. Bu bileşikler: gallik asit, 4-hidroksi benzoik asit, ferulik asit, kafeik asit, p-kumarik asit, sinapik asit, kaftarik asit, klorojenik asit, kateşin, prosiyandinin-B2, 4-metil kateşol ve 2,5-dihidroksibenzoik asittir. Odriozola-Serrano ve ark. (2023) tarafından yapılan bir çalışmada ise fenolik ekstrakt üç fraksiyona ayrılıp analiz edilmiş ve fenolik bileşiklerin çoğunun esterleşmiş formda olduğu belirlenmiştir. Esterleşmiş ve konjuge formdaki bileşikler içinde başlıca fenolik bileşiklerin fenolik asitler olduğu tespit edilmiştir. Serbest formda ise flavonoller ve flavan-3-oller tespit edilmiştir. Fenolik asitlerin fenolik bileşiklerin % 92'lik kısmını oluşturduğu ve en yüksek miktarda bulunan fenolik asidin gallik asit olduğu belirlenmiştir. Çalışmada gallik asidin yanı sıra p-kumarik asit, protokateşik asit, p-salisilik asit, elajik asit, vanilik asit, kafeik asit ve ferulik asit de tespit edilmiştir. Ekstraktlarda tespit edilen başlıca flavonolün kuersetin ve türevleri olduğu ve fenolik bileşiklerin %5'lik kısmını oluşturduğu belirlenmiştir. Çalışmada flavonollerden izokuersetin, kuersetin, kaempferol, hiperoside ve rutin, flavan-3-ollerden kateşin ve prosiyanidin-B ve flavonlardan luteolin ve lutein-7-*O*-glukozit de tespit edilmiştir.

Tablo 1. *Rosa canina* L. : Biyoaktif bileşenler

Bileşen	Miktar	Kaynak
Askorbik asit (mg/100 g km)	68,04±1,11 101,38±0,91	Barros ve ark., 2010 Demir ve ark., 2014
Karotenoidler (mg/100 g km) β-Karoten Likopen	1,29±0,26 0,51±0,08	Barros ve ark., 2010
Tokoferoller (mg/100 g km) α-Tokoferol β-Tokoferol δ-Tokoferol	 7,05±0,28 0,19±0,01 1,09±0,06	Barros ve ark., 2010
Fenolik bileşikler (µg/100 g km) Gallik asit Ferulik asit Klorojenik asit Kafeik asit p-Kumarik asit 2,5-Dihidroksi benzoik asit Kateşin Prosiyanidin-B2	 12,67±4,35 10,55±3,18 12,11±5,53 9,38±3,56 9,00±4,09 10,40±4,47 19,96±28,22 7,54±10,48	Demir ve ark., 2014

Km: kuru maddede

Kuşburnu meyvelerinin çeşitli ürünleri işlenmesi sırasında atık ürün olarak kuşburnu çekirdeği elde edilmektedir. Kuşburnu çekirdeği yağının doymamış yağ asit içeriği yüksektir (>%80). Kuşburnu çekirdeği yağında bulunan başlıca tekli doymamış yağ asidi oleik asit, başlıca doymamış yağ asitleri ise linoleik ve linolenik asittir. Sırbistan'da yapılan bir çalışmada farklı bölgelerde yetişen *R. canina* L. çekirdeklerinden elde edilen yağların yağ asidi kompozisyonu belirlenmiş ve yağların oleik asit içeriği %3,89-13,82; linoleik asit içeriği % 24,53-46,68 ve linolenik asit içeriği %4,73-12,39 olarak bulunmuştur (Popovic-Djordjevic ve ark., 2023). Türkiye'de yapılan bir çalışmada ise oleik asit içeriği % 44,63, linoleik asit içeriği % 27,97 ve linolenik asit içeriği % 11,48 olarak bulunmuştur (Murathan ve ark., 2016b).

KUŞBURNU ÜRÜNLERİ

Ülkemizde kuşburnu meyvesi, taze meyvesinin hafif ekşi tadı nedeniyle genellikle kurutulmuş halde tüketilmektedir. Kuşburnu meyvesi, bitki çayı, reçel, marmelat, nektar, pestil gibi ürünlere işlenerek de tüketilmektedir (Ilyasoglu, 2014). Bunların dışında sirke, yağ ve toz içecek formu olarak da işlenmektedir.

Kuşburnu Sirkesi

Sirke, dünya genelinde baharat veya gıda koruyucusu olarak kullanılan, sos, mayonez ve salata sosu gibi gıdaların duyusal özelliklerine katkıda bulunmak için kullanılan fermente bir üründür (Ho ve ark., 2017). Sirke, çok sayıda meyve ve sebzeden üretilebildiğinden amino asitler, organik asitler, fenolik bileşikler, vitaminler, mineraller ve antioksidan maddeler bakımından zengindir. Son yıllarda geleneksel olarak üretilen bazı sirkelerinin ekonomiye kazandırılabilmesi için daha işlevsel ve duyusal özellikleri iyileştirilmiş olarak üretilmesi popülerlik kazanmıştır. Bu anlamda kuşburnu meyvesinden sirke üretimi ekonomik katma değeri ve işlevselliği yüksek bir ürün üretmek için iyi bir fırsat olarak görülmüştür. Özdemir ve ark. (2022), yapmış oldukları çalışmada kuşburnu sirkesinin toplam fenolik madde, toplam flavonoid ve antioksidan aktivitesini yüksek bulmuşlardır. Kuşburnu sirkesinin C vitamini miktarı fermentasyon boyunca artmış ve 60. günde en yüksek seviyeye ulaşmıştır (5,31-71,49 mg/ml). Aynı çalışmada kuşburnu sirkesinde 27 adet aroma bileşiği tespit etmiş ve bu sirkesinin sağlıklı ve aromatik bir alternatif olacağını bildirmişlerdir.

Kuşburnu Yağı

Kuşburnu, fonksiyonel gıda ve kozmo-nutrasötik ürünler için kullanılabilecek potansiyel bir meyve ürünüdür. Cilt iyileştirme potansiyeli nedeniyle çoğunlukla *R. canina* ve *R. moschata* gibi yabani gül çalı türlerinin tohumlarından elde edilen kuşburnu yağı, bitkisel kozmetiklerin, losyon ve krem gibi cilt bakım ürünlerinin geliştirilmesi için değerli bir malzemedir. Yağ içeriği bakımından en zengin (% 70-80) esansiyel yağ asitleri (EYA) kaynaklarından (linoleik asit ve linolenik asit) birisidir. Kuşburnu yağı, yara izleri, dermatit, akne, egzama ve yanıklar gibi cilt problemlerinin tedavi edilmesinde ve iyileştirilmesinde kullanılmaktadır. Cildi

gençleştirici özelliklere sahip EYA'leri, karotenoidler ve A vitamini (retinol) içeriği nedeniyle kuşburnu tohum yağı, doğal bir cilt bakım ürünü, nemlendirici ve yaşlanma karşıtı ajan görevi göstermektedir. Tohum yağı ayrıca yara izlerini iyileştirmek, yaşlanmayı azaltmak ve cilde pürüzsüzlük kazandırmak için de faydalıdır (Concha ve ark., 2006; Ahmad ve Anwar, 2016).

Kuşburnu Marmelatı

Marmelat, meyve pulpu, püre, meyve suyu ve sulu ekstraktlarının veya bitkilerin kök, yaprak ve çiçek gibi yenilebilen kısımlarına gerektiğinde şeker ve su ilave edilerek sürülme kıvamına getirilmesiyle elde edilen karışım olarak tanımlamıştır (Anonim, 2006). Kuşburnu marmelatı Türkiye'de kahvaltıda yaygın olarak tüketilen bir üründür. Daha bilinçli hale gelen tüketici bu tarz ürünlerde aroma ve lezzetin yanı sıra vitamin, mineral, fenolik bileşik ve antioksidan aktivitesi yüksek ürünleri tercih etmektedir. Kuşburnu meyvesi yüksek oranda içermiş olduğu vitamin, mineral ve fenolik bileşiklerle ön plana çıkmaktadır. Şengül ve ark., (2018) yılında yapmış oldukları çalışmada geleneksel olarak üretilen kuşburnu, kızılcık, çakal eriği ve ahlat armudu marmelatlarını karşılaştırmış ve en yüksek toplam fenolik madde içeren marmelatın kuşburnu marmelatı olduğunu (581,89 µg/GAE g) bildirmişlerdir.

Kuşburnu Çayı

Bitki çayları, pratik bir şekilde hazırlanan, hoş tada sahip, kalorisiz ve kafeinsiz içeceklerdir. İçermiş oldukları antioksidan bileşikler ile sağlık üzerine olumlu etkileri vardır ve tüm dünyada sıklıkla tercih edilmektedirler (Alarcòn ve ark., 2008; Yılmaz ve ark., 2021). Kuşburnu çayı, içermiş olduğu yüksek C vitamini sayesinde iyi bir antioksidan kaynağıdır (Nojavan ve ark., 2008). Taze ve kuru halde çay olarak tüketilmektedir (Wenzig ve ark., 2008). İlyasoğlu ve Arpa (2017), tarafından yapılan çalışmada kuşburnu çayı için 84–86°C derecede 6-8 dk demleme işleminin C vitamini ve toplam fenolik madde miktarı açısından en iyi koşullar olduğu belirlenmiştir.

KUŞBURNUNUN SAĞLIK ÜZERİNE ETKİLERİ

Genel olarak kuşburnunun beslenmedeki önemi, vitamin ve mineraller açısından zengin olmasına atfedilir. Aynı miktarda kuşburnu portakaldan 30-40 kat daha fazla C vitamini içerir (Yoruk ve ark., 2008). C vitamini kolajen sentezinde önemli olup soğuk algınlığı ve gribe karşı bağışıklık sistemini güçlendirir, polifenol içeriği ve antioksidan özellikleri sayesinde de potansiyel olarak kanser riskini azaltır. C vitamini demirin emilmesine yardımcı olur, kan basıncını ve kolesterolü düşürür (Çınar ve Çolakoğlu, 2004).

Kuşburnu içerdiği fenolik asitler, tanenler ve flavonoidlerle fitokimyasalların iyi bir kaynağıdır. Bu sayede kulak, burun ve boğaz sorunları dahil olmak üzere birçok hastalığın tedavisinde geleneksel olarak kullanılmaktadır. Avrupa halk hekimliğinde kuşburnu müshil, idrar söktürücü, gut önleyici ve romatizma önleyici ilaç olarak kullanılmaktadır (Ayati ve ark., 2018). Türk halk hekimliğinde ise meyve ve yaprakları soğuk algınlığı, öksürük, gastrit, ülser, anemi, anoreksi ve diyabet gibi hastalıkların geleneksel tedavisinde kullanılmaktadır. Ayrıca bitkinin kökleri ve yaprakları bronşite karşı da kullanılmaktadır (Sarikaya ve ark., 2010; Orhan ve ark., 2009).

Amerikan Botanik Konseyi tarafından yayımlanmış olan "Bitkisel İlaçlar Terapötik Rehberi"nde *R. canina* meyvelerinin, artrit, romatizma, gut ve siyatik gibi inflamatuar bozukluklarda; soğuk algınlığı ve grip gibi ateşli bulaşıcı hastalıklarda, mide-bağırsak rahatsızlıklarında, safra taşı şikayetlerinde, böbrek ve idrar yolu rahatsızlıkları dahil olmak üzere çeşitli hastalıklarda profilaktik ve tedavi edici aktivitelere sahip olduğu bildirilmiştir (Blumenthal, 1998). Ayrıca *R. canina*'nın antioksidan, antiinflamatuar, antiülser, antimikrobiyal, antimutagenik etkilerinin yanında anksiyete, osteoartrit, diyabet ve hipertansiyon gibi hastalıkların tedavisinde iyileştirici etkilerine ilişkin raporlar mevcuttur (Orhan ve ark., 2009; Öğüt, 2022).

Kuşburnu ve Osteoartrit İlişkisi

Osteoartrit, yaşlılarda önemli bir bozulma nedeni olan kronik ilerleyici bir hastalıktır. Ağrı, azalmış hareket açıklığı, fiziksel fonksiyonda azalma, eklem sertliği ve şişmesi, kas zayıflığı vb. ile karakterizedir. Tüm bu koşullar, bireylerde günlük aktivitelerin kötüleşmesine ve sakatlığa yol açarak yaşam kalitesinin bozulmasına yol açmaktadır. Osteoartritten yaygın olarak diz ve kalça etkilenir (Zampogna ve ark., 2020). Farmakolojik tedavisinde steroid olmayan anti-inflamatuar ilaçların (NSAID) kullanımı ciddi gastrointestinal komplikasyonlara neden olabilir (Lazzaroni ve Bianchi Porro, 2004). Ayrıca ağrıyı dindirmek için kullanılan parasetamolün de son zamanlarda gastrointestinal problem riskini arttırdığı rapor edilmiştir. Bu nedenlerle, bahsedilen yan etkiler olmadan ağrıyı ve sertliği en aza indirebilecek yeni bileşikler arayışına girilmiştir.

Çeşitli bitkisel ilaçlar denenmiş ve umut verici sonuçlara ulaşılmıştır. Bunlardan biri de kuşburnundan elde edilen özütler ve nutrasötiklerdir. Bir çalışma kapsamında *R. canina* (kuşburnu), *Boswellia serrata* ve *Harpagophytum procumbens* özütlerinin karışımından TriNyros kapsülü geliştirilmiştir. Bu karışımın antiinflamatuar, antioksidan ve kondroprotektif bir ajan olarak etkili olduğu bulunmuştur. Bu sayede eklem ağrılarını azaltarak osteoartrit hastalarının fiziksel fonksiyonel yeteneğini geliştirdiği rapor edilmiştir (Tapasvi, 2022). Yapılan bir başka çalışmada, kuşburnundan yapılan bitkisel bir ilaç, kalça ve diz osteoartriti olan hastalarda 3 aylık bir süre boyunca kullanılmış, ilacın osteoartrit semptomlarını hafifletebildiği görülmüştür (Winther ve ark., 2005).

Kuşburnu ve Kanser İlişkisi

Kanser, sinyalizasyon ve metabolizmanın değiştiği, kontrolsüz bölünmeye ve dönüştürülmüş hücrelerin hayatta kalmasına neden olan bir hastalıktır. Kanserler, her yıl milyonlarca ölüme yol açar (Upadhyay, 2021). 2020 yılında dünya çapında tahminen 19,3 milyon yeni kanser vakası ve neredeyse 10,0 milyon kanser ölümü meydana gelmiştir. Akciğer kanseri, tahmini 1,8 milyon ölüm (%18) ile kanserden ölümlerin önde gelen nedeni olmaya devam ederken, onu kolorektal, karaciğer, mide ve meme kanserleri takip etmektedir. Küresel kanser yükünün 2040 yılında %47'lik bir artışla 28,4 milyon vaka olması beklenmektedir (Sung ve ark., 2021). En acil patolojik durumlardan biri olan bu hastalık hakkında muazzam bir literatür bulunmasına rağmen, hastalığın başarılı bir şekilde tedavi edilmesi belirsiz görünmektedir (Upadhyay, 2021). Günümüzde kullanılan kemoterapötik ajanların neredeyse tamamı oldukça toksiktir ve normal hücrelerde de ciddi yan etkilere neden olur. Bu nedenle yüksek etkinliğe ve düşük toksisiteye sahip doğal kaynaklı yeni antikanser ilaçlarının geliştirilmesine yönelik çalışmalar yapılmaktadır. Son yıllarda bitki kaynaklı bileşiklerin kanser hücreleri üzerinde sentetik kemoterapi ilaçlarına göre nispeten daha etkili olduğu ve normal hücrelere çok daha az zarar verdiği gösterilmiştir (Shukla ve Mehta, 2015). Bu bağlamda literatürde kuşburnu ve özütlerine ilişkin de antikanser çalışmalar mevcuttur.

Prostat kanseri dünyada akciğer kanserinden sonra en sık görülen kanser türüdür. Her ne kadar hastalık kemoterapötiklerle tedavi edilse de ölüm oranı nispeten yüksektir (Russo ve ark., 2017; Özgönül ve ark., 2020). Son çalışmalar fenolik asit içeren diyetin daha düşük prostat kanseri riski ile ilişkili olduğunu göstermiştir. Buna yönelik yapılan bir çalışmada, kuşburnu çayında bulunan fenolik bileşiklerin prostat kanseri hücre hatlarındaki antiproliferatif etkisi araştırılmıştır. Çalışmada öncelikle kuşburnu çayının flavonol içeriği belirlenmiş ve daha sonra çay örneklerinin prostat kanseri hücre hattına (VCaP ve LNCaP) eklenmesiyle antiproliferatif etkisine bakılmıştır. Sonuç olarak, doğal bir flavonol kaynağı olan kuşburnu çayının androjenlere duyarlı prostat kanseri hücreleri hattında büyümeyi ve hücre çoğalmasını azalttığı görülmüştür (Özgönül ve ark., 2020). *R. canina*'nın sitotoksik etkisini belirlemek üzere yapılmış bir çalışmada, *R. canina*'nın insan akciğer (A549) ve prostat (PC-3) kanseri hücreleri üzerindeki sitotoksik etkileriyle birlikte ve antioksidan özellikleri ve ilgili olası mekanizmalar değerlendirilmiştir. Sonuçlar *R. canina* özütünün akciğer ve prostat kanseri hücrelerinin çoğalmasını engellemek için iyi bir aday olabileceğini göstermiştir (Kilinc ve ark., 2020). Literatürde mevcut bir başka çalışmada *R. canina* özütünün insan kolon kanseri (WiDr) hücrelerinde hücre canlılığı, hücre döngüsü, apoptoz ve telomeraz ekspresyonu üzerindeki potansiyel etkisini incelenmiştir. Özütün sitotoksik etkisinde yer alan mekanizma apoptoz ve hücre döngüsü açısından değerlendirilmiştir. Sonuç olarak *R. canina* özütünün WiDr hücreleri üzerinde seçici bir sitotoksik etki gösterdiği bulunmuştur. Özütün WiDr hücrelerinde apoptozu tetiklediği rapor edilmiştir. Böylece *R. canina*'nın kolon kanserinde de doğal ürün bazlı etkili bir antikanser ajan olabileceği görülmüştür (Turan ve ark., 2018).

Kuşburnu ve Diyabet İlişkisi

Diyabet dünya çapında yaygın bir metabolik hastalıktır. Hiperglisemi, lipid ve protein metabolizma anormallikleri ile karakterizedir. Retinopati, nöropati, nefropati vb. klinik komplikasyonlara eşlik eder. Diyabet tedavisinde birçok geleneksel bitki kullanılmaktadır. Bu bitkilerin biyoaktif bileşenleri antidiyabetik özelliklerine katkıda bulunur. Ayrıca, kullanılan kimyasal antidiyabetik ilaçlara doğal ve güvenli bir alternatif olarak düşünülebilir (Matalqah ve Al-Tawalbeh, 2019). Kuşburnu dünyanın birçok yerinde ve Anadolu'da geleneksel diyabet tedavisinde kullanılan bitkilerden biridir (Sarikaya ve ark., 2010). Yapılan bir çalışmada, *R. canina* meyvelerinin etanol özütlerinin antioksidan, hipoglisemik ve antidiyabetik aktiviteleri araştırılmıştır. 7 gün boyunca uygulanan etanol özütünün diyabetik sıçanlarda hipoglisemik etkisi olduğu saptanmıştır (Orhan ve ark., 2009). Obez farelerde yapılan bir çalışmada, kuşburnu tozunun yüksek yağlı diyetin neden olduğu obeziteyi ve glikoz intoleransını önlediği ve tersine çevirdiği, ayrıca plazma kolesterol seviyelerini düşürdüğü

gösterilmiştir. 6 hafta boyunca günlük kuşburnu tozu alan obez farelerde faydalı metabolik etkiler gözlenmiştir. Kuşburnu tozu içeceğinin tüketimi sistolik kan basıncında ve toplam plazma kolesterolünde önemli bir azalmayla sonuçlanmıştır (Andersson ve ark., 2012).

SONUÇ

Tüm dünyayı etkileyen COVID-19 salgını, sağlığa daha fazla vurgu yapılmasına ve biyoaktif bileşiklere sahip fonksiyonel gıdaların araştırılması, geliştirilmesi ve kullanılmasına hız kazandırmıştır. Kuşburnu meyvesi dünya genelinde büyük bir coğrafyada yetişebilmesi, askorbik asit, doymamış yağ asidi, polifenoller, karotenoidler ve polisakkaritler dahil olmak üzere insan sağlığına fayda sağlayan çeşitli fonksiyonel bileşikleri içermesi dolayısıyla dikkatleri üzerine çekmiştir. Doğal bir antioksidan kaynağı olan kuşburnu gıda, kozmetik, ilaç vekimya gibi çeşitli alanlarda ilgi uyandırmaktadır. İşlenmeyen birçok alt türü olan kuşburnunun kaynakları belirlenmeli, araştırmalar yapılmalı ve ekimi yapılan bir bitki haline gelmesi teşvik edilmelidir.

KAYNAKLAR

Ahmad N, Anwar F 2016. Rose hip (*Rosa canina* L.) oils. In Essential oils in food preservation, flavor and safety (pp. 667-675). Academic Press.

Anonim 2006. Türk Gıda Kodeksi Reçel, Jöle Marmelat ve Tatlandırılmış Kestane Püresi Tebliği, Resmi Gazete, Tarih: 30.12.2006, Sayı: 26392, Ankara.

Alarcón E, Campos AM, Edwards, AM, Lissi, E, López-Alarcón C 2008. Antioxidant capacity of herbal infusions and tea extracts: A comparison of ORAC-fluorescein and ORAC-pyrogallol red methodologies. Food Chemistry, 107(3): 1114-1119.

Andersson U, Berger K, Högberg A, Landin-Olsson M, Holm C 2012. Effects of rose hip intake on risk markers of type 2 diabetes and cardiovascular disease: a randomized, double-blind, cross-over investigation in obese persons. European Journal of Clinical Nutrition, 66(5): 585-590.

Ayati Z, Amiri MS, Ramezani M, Delshad E, Sahebkar A, Emami SA 2018). Phytochemistry, traditional uses and pharmacological profile of rose hip: A review. Current pharmaceutical design, 24(35): 4101-4124.

Barros L, Carvalho AM, Sa Morais J, Ferreira ICFR 2010. Strawberry-tree, blackthorn and rose fruits: Detailed characterisation in nutrients and phytochemicals with antioxidant properties. Food Chemistry 120: 247–254.

Blumenthal, M. (1998). The complete german commission e monographs: Therapeutic guide to herbal medicines. The American Botanical Council, Austin. First edition. 684 pp. ISBN: 978-0965555500.

Çınar İ, Çolakoğlu AS 2004, September. Potential health benefits of rose hip products. In: International Rose Hip Conference, pp. 253-258.

Concha J, Soto, C, Chamy R, Zuniga ME, 2006. Effect of rosehip extraction process on oil and defatted meal physicochemical properties. Journal of the American Oil Chemists' Society, 83 (9): 771-775.

Demir N, Yıldız O, Alpaslan A, Hayaloğlu AA 2014. Evaluation of volatiles, phenolic compounds and antioxidant activities of rose hip (Rosa L.) fruits in Turkey. Food Science and Technology, 57: 126e133.

Fan C, Pacier C, Martirosyan DM 2014. Rose hip (Rosa canina L): A functional food perspective Functional Foods in Health and Disease, 4(11):493-509.

Ho CW, Lazim AM, Fazry S, Umi Kalsum HZ, Lim SJ 2017. Varieties, production, composition and health benefits of vinegars: A review. Food Chemistry, 221: 1621-1630.

Ilyasoglu H 2014. Characterization of Rosehip (*Rosa canina* L.) seed and seed oil. International Journal of Food Properties, 17: 1591-1598.

Ilyasoglu H, Arpa TE 2017. Effect of brewing conditions on antioxidant properties of rosehip tea beverage: study by response surface methodology. Journal of Food Science and Technology, 54: 3737-3743

Kilinc K, Demir S, Turan I, Mentese A, Orem A, Sonmez M, Aliyazicioglu Y 2020. Rosa canina extract has antiproliferative and proapoptotic effects on human lung and prostate cancer cells. Nutrition and cancer, 72(2): 273-282.

Koç A 2020. Chemical changes in seeds and fruits of natural growing Rosehip (Rosa sp.) from Yozgat (Turkey). Acta Scientiarum Polonorum Hortorum Cultus, 19(2): 123–134

Lazzaroni M, Bianchi Porro G 2004. Gastrointestinal side-effects of traditional non-steroidal anti-inflammatory drugs and new formulations. Alimentary Pharmacology and Therapeutics, 20: 48-58.

Matalqah SM, Al-Tawalbeh DM 2019. Medicinal Plants Potential Against Diabetes Mellitus: Review Article. International Journal of Pharmacognosy, 6(2): 39-53.

Nojavan S, Khalilian F, Kiaie FM, Rahimi A, Arabanian A, Chalavi S 2008. Extraction and quantitative determination of ascorbic acid during different maturity stages of *Rosa canina* L. fruit. Journal of food composition and analysis, 21(4): 300-305.

Murathan T, Zarifikhosroshahi M, Kafkas E 2016a. Characterization of bioactive compounds in Rosehip spesies from East Anatolia Region of Turkey. Italian Journal of Food Science, 28: 314-325.

Murathan T, Zarifikhosroshahi M, Kafkas E 2016b. Determination of fatty acids and volatile compounds in fruits of rosehip(Rosa L.) species by HS-SPME/GC-MS and Im-SPME/GCMS techniquesTurkish Journal of Agriculture Forestry 40: 269-279

Odriozola-Serrano I , Nogueira DP , Esparza I , Vaz AA , Jiménez-Moreno N, Martín-Belloso O, Ancín-Azpilicueta C 2023. Stability and Bioaccessibility of Phenolic Compounds in Rosehip Extracts during In Vitro Digestion. Antioxidants, 12:1035.

Orhan N, Aslan M, Hosbas S, Deliorman OD 2009. Antidiabetic effect and antioxidant potential of *Rosa canina* fruits. Pharmacognosy Magazine, 5(20): 309-315.

Öğüt K 2022. *Rosa canina* L. In Novel Drug Targets With Traditional Herbal Medicines: Scientific and Clinical Evidence. Cham: Springer International Publishing, pp. 513-523.

Öz M, Baltacı C, Deniz İ 2018. Gümüşhane Yöresi Kuşburnu (*Rosa canina* L.) ve Siyah Kuşburnu (*Rosa pimpinellifolia* L.) Meyvelerinin C Vitamini ve Şeker Analizleri. Gümüşhane Üniversitesi Fen Bilimleri Dergisi, 8 (2): 284-292.

Özdemir N, Pashazadeh H, Zannou O, Koca I 2022. Phytochemical content, and antioxidant activity, and volatile compounds associated with the aromatic property, of the vinegar produced from rosehip fruit (*Rosa canina* L.). LWT - Food Science and Technology, 154: 112716.

Özgönül AM, Aşık A, Durmaz, B, Aslaminabad R, Gündüz C, Sozmen EY 2020. Antiproliferative effect of rosehip tea phenolics in prostate cancer cell lines. Turkish Journal of Biochemistry, 45(4): 423-428.

Popovic-Djordjevic J, Spirovic-Trifunovic B, Pecinar I, de Oliveira LFC, Krstic D, Mihajlovic D, Aksic MF, Simal-Gandara J 2023. Fatty acids in seed oil of wild and cultivated rosehip (*Rosa canina* L.) from different locations in Serbia Industrial Crops and Products, 191: 115797.

Russo GI, Campisi D, Di Mauro M, Regis F, Reale G, Marranzano M, ... Morgia G 2017. Dietary consumption of phenolic acids and prostate cancer: A case-control study in sicily, Southern Italy. Molecules, 22(12): 2159.

Sarikaya S, Öner H, Harput, ÜŞ 2010. Türkiye florasında diyabet tedavisinde kullanılan tıbbi bitkiler. Ankara Eczacılık Fakültesi Dergisi, 39(4): 317-342.

Shukla, S., & Mehta, A. (2015). Anticancer potential of medicinal plants and their phytochemicals: a review. Brazilian Journal of Botany, 38, 199-210.

Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, Bray F 2021. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. Cancer Journal for Clinicians, 71(3): 209-249.

Şengül M, Topdaş EF, Doğan H, Serencam H 2018. Artvin ilinde geleneksel olarak üretilen farklı marmelat çeşitlerinin bazı fiziksel ve kimyasal özellikleri, antioksidan aktiviteleri ve fenolik profilleri. Akademik Gıda, 16(1): 51-59.

Tapasvi S 2022. Clinical efficacy of nutraceuticals in the management of osteoarthritis. International Journal of Orthopaedics, 8(3): 17-23.

Turan I, Demir S, Kilinc K, Yaman SO, Misir S, Kara H, ... Deger O 2018. Cytotoxic effect of Rosa canina extract on human colon cancer cells through repression of telomerase expression. Journal of Pharmaceutical Analysis, 8(6): 394-399.

Upadhyay A 2021. Cancer: An unknown territory; rethinking before going ahead. Genes and Diseases, 8(5): 655-661.

Wenzig EM, Widowitz U, Kunert O, Chrubasik S, Bucar F, Knauder E, Bauer R 2008. Phytochemical composition and in vitro pharmacological activity of two rose hip (*Rosa canina* L.) preparations. Phytomedicine, 15(10): 826-835.

Winther K, Apel K, Thamsborg G 2005. A powder made from seeds and shells of a rose-hip subspecies (Rosa canina) reduces symptoms of knee and hip osteoarthritis: a randomized, double-blind, placebo-controlled clinical trial. Scandinavian Journal of Rheumatology, 34(4): 302-308.

Yılmaz C, Konuş M, Fidan C, Ergin D, Çetin D, Dilek Z, ... Elasan E 2021. Su kalitesi ve demleme şeklinin bitkisel çaylarda toplam antioksidan kapasite üzerine etkisi. Kahramanmaraş Sütçü İmam Üniversitesi Tarım ve Doğa Dergisi, 24(5): 921-929.

Yoruk IH, Turker M, Kazankaya A, Erez ME, Batta P, Celik F 2008. Fatty acid, sugar and vitamin contents in rose hip species. Asian Journal of Chemistry, 20(2): 1357-1364.

Zampogna B, Papalia R, Papalia GF, Campi S, Vasta S, Vorini F, ... Denaro V 2020. The role of physical activity as conservative treatment for hip and knee osteoarthritis in older people: a systematic review and meta-analysis. Journal of Clinical Medicine, 9(4): 1167.

Zhou M, Sun Y, Luo L, Pan H, Zhang Q, Yu C 2023. Road to a bite of rosehip: A comprehensive review of bioactive compounds, biological activities, and industrial applications of fruits. Trends in Food Science and Technology, 136: 76-91.

DRAFT

ORAL PRESENTATION

Elektroeğirme ve Solvent Döküm Yöntemleri ile Üretilen Polikaprolakton (PCL) Fiber ve Filmlerin Özellikleri

Büşra Boz[1](https://orcid.org/0009-0004-3923-7539), Ferda Mindivan[1*](https://orcid.org/0000-0002-6046-2456)

[*1] Bilecik Şeyh Edebali Üniversitesi, Mühendislik Fakültesi, Biyomühendislik Bölümü, Bilecik, Türkiye
*ferda.mindivan@bilecik.edu.tr

Özet

Bu çalışmada polikaprolakton (PCL) filmler ve fiberler sırasıyla solvent döküm (D-PCL) ve elektrospinning (E-PCL) yöntemleriyle üretilmiştir. E-PCL ve D-PCL örneklerinin morfolojisini ve moleküler etkileşimlerini karakterize etmek için X-ışını kırınımı (XRD), optik mikroskop (OM), Taramalı elektron mikroskobu (SEM), Fourier dönüşümü kızılötesi spektroskopisi (FTIR) ve temas açısı ölçümleri yapılmıştır. Solvent döküm yöntemiyle üretilen D-PCL'nin moleküller arası etkileşimleri elektroeğirme yöntemiyle üretilen E-PCL örneğinden daha fazla, kristal boyut değerleri ise daha büyük olduğu FTIR ve XRD analiz sonuçlarından tespit edilmiştir.D-PCL örneğinin OM yüzey görüntüsü PCL'nin karakteristik tane yapısını, E-PCL örneğinin SEM yüzey görüntüsü ise homojen boyutta üretilen nanofiberleri göstermiştir. D-PCL ve E-PCL örneklerinin temas açısı sonuçları PCL'nin hidrofobik yapısını yansıtmıştır. Bu çalışma PCL esaslı kompozit veya nanokompozit çalışmalarında solvent döküm yönteminin elektroeğirme yöntemine göre tercih edilebilir olduğunu önermektedir.

Anahtar Kelimeler: Polikaprolakton, Solvent Döküm, Elektroeğirme, yapısal karakterizasyon

Properties of Polycaprolactone (PCL) Fiber and Film Product by Electrospinning and Solvent Casting Process

Abstract

In this study, the polycaprolactone(PCL) films and fibers were fabricated by solvent casting (D-PCL) and electrospinning (E-PCL), respectively. X-ray difraction (XRD), optical microscope (OM), Scanning electron microscopy (SEM), Fourier transform infrared spectroscopy (FTIR) and water contact angle measurements were performed to characterize the morphology and molecular interactions of the E-PCL and D-PCL samples. It was determined from the results of XRD and FTIR analyses that the intermolecular interactions of the D-PCL sample produced by the solvent casting method were higher than the E-PCL sample produced by the electrospinning method, and the crystal size values were larger. The OM surface image of the D-PCL sample showed the characteristic grain structure of PCL, and the SEM surface image of the E-PCL sample showed the nanofibers produced in homogeneous size. The contact angle results of the D-PCL and E-PCL samples reflected the hydrophobic structure of PCL. This study suggests that the solvent casting method is preferable to the electrospinning method in PCL-based composite or nanocomposite studies.

Keywords: Polycaprolactone, Solvent Casting, Electrospinning, structural characterization

GİRİŞ

Nanomalzemelerin geniş bir uygulama alanına sahip olması son yıllarda kullanım alanlarını artırmıştır.Bunun bir sonucu olarak nanomalzemelerin üretilmesi için çeşitli teknikler ve yöntemler geliştirilmiştir. Bu teknikler arasında, basitliği ve çeşitli malzemelerden mikro/nano ölçekli fiberler üretebilme yeteneğine sahip elektroeğirme yöntemi kullanılmaktadır (Alharbi ve ark., 2023). Elektroeğirme, polimerik bir çözeltiye yüksek voltaj uygulanarak, hedef bir levha üzerinde toplanabilen nanolif üretim tekniğidir (Armentano ve ark., 2009). Nanolifler yüksek yüzey/hacim oranı, yüksek gözeneklilik ve ayarlanabilir mekanik özellikleri sayesinde çok sayıda biyomedikal uygulamalarda kullanılmaktadır (Alharbi ve ark., 2023). Aktif ve yenilebilir filmler için kullanılan basit ve özel ekipman gerektirmeyen bir diğer nanomalzeme üretim yöntemi de solvent döküm

yöntemidir. Aktif bileşiklerin film oluşturucu bir çözeltiye doğrudan dahil edilmesinin ardından çözeltinin düz bir plaka üzerine yayılması ve ardından kontrollü koşullar altında kurutularak, çözücünün uzaklaştırılıp film oluşumunun sağlanmasını içermektedir (Salević ve ark., 2022). Yeni araştırmaların bu tekniğe ilgi duyması zaman ve enerji tüketimi bakımından avantaj sunmasından kaynaklanmaktadır (Salević ve ark., 2022). Nanomalzeme olarak en çok tercih edilen Polikaprolakton (PCL); polyester grubundan, sentetik ve biyolojik olarak parçalanabilen bir polimerdir (Ghosal ve ark., 2021). Ayrıca PCL, yüksek mekanik özelliğinin yanında zayıf hücre yapışma özellikleri sergileyen, biyolojik olarak uyumlu ve hidrofobik bir polimerdir (Gautam ve ark., 2023). PCL' nin mekanik, fiziksel ve kimyasal özelliklerin değiştirilmesi, kontrol edilebilirliğinin de kolay olmasından dolayı fiber üretimimnde kullanılmaktadır (Alharbi ve ark., 2023). Literatürde PCL'ye farklı dolgu maddelerinin ilavesiyle hem solvent döküm hem de elektroeğirme yöntemiyle üretilen fiber ve filmler üzerine yapılan birkaç çalışma mevcuttur. Ghosal ve ark., (2021), hem elektroeğirme hem de solvent döküm yöntemiyle ürettikleri PCL bazlı, karbon kuantum nokta (hCQDs) katkılı hidrofobik nanokompozitin film ve fiberleri karakterize etmişlerdir. Filmlerin daha düşük temas açısına sahip oldukları ve ürettikleri bu filmlerin yara örtüsü olarak etkin bir şekilde kullanılabileceğini önermişlerdir. Del Gaudio ve ark., (2011) PCL ve poli(3-hidroksibutirat-ko-3-hidroksivalerat) (PHBV) fiber ve filmler solvent döküm ve elektroeğirme yöntemleriyle üretilmiştir. Yapılan karakterizasyon çalışmaları sonucunda elektroeğirme yöntemiyle üretilen fiberlerin daha homojen yapıda olduğu, solvent döküm yöntemiyle üretilen filmlerin ise mekanik dayanımının daha yüksek olduğu belirlenmiştir. Armentano ve ark., (2009) PCL matriksi içerisine karbon nanolif (CNF'ler) katkılı kompozit malzemeleri solvent döküm ve elektroeğirme yöntemleriyle üretmişlerdir. Kompozit filmler, saf PCL fiberine göre artan bir kristalizasyon sıcaklığı ortaya çıkarmıştır. Filmlerin ve fiberlerin mekanik özellikleri CNF içeriğine bağlı olarak iyileşirken, fiberlerin mekanik özellikleri CNF'nin eklenmesinden daha az etkilendiğini rapor etmişlerdir. Literatürde yer alan çalışmalar solvent döküm yöntemiyle elektroeğirme yönteminin üretilen örneklerin birçok özelliği üzerinde etkisi olduğunu göstermiştir. Bu çalışmada da elektroeğirme ve solvent döküm yöntemleriyle PCL fiber ve filmleri üretilmiştir. Üretilen PCL fiber ve filmlerin yapısal özellikleri karşılaştırılmıştır.

MATERYAL VE METOT

80.000 molekül ağırlıklı PCL kullanılarak, PCL filmleri solvent döküm, PCL fiberleri ise elektroeğirme yöntemleriyle üretilmiştir. Bu çalışmada elektroeğirme ve solvent döküm için kullanılacak polimer çözeltisi DMF/Kloroform (1/4) (v/v) çözücüleri içerisinde PCL konsantrasyonu %15 (w/v) olacak şekilde 3 saat karıştırılarak hazırlanmıştır. Çözünme sırasında sıcaklık 25-35°C arasında tutulmuştur. Üretilen polimer çözeltisi solvent döküm yöntemine göre üretilecek PCL film için bir kalıba dökülüp etüvde 35-40°C arasında 1 saat kurutulmuştur. Elektroeğirme işlemi ile polimer çözeltisinin 19,86 mm çapındaki iğne içerisine doldurularak, elekroeğirme cihazının pompa haznesine yerleştirilip, akış hızı 1,5 mL/saat, voltaj 10 kV, şırınga ucu ile toplayıcı levha arasındaki uzaklık 12 cm, oda sıcaklığı ve nem %34-47 arasında tutularak nanofiber üretimi gerçekleştirilmiştir. Solvent döküm yöntemiyle üretilen PCL film D-PCL ve elektroeğirme yöntemiyle üretilen PCL nanofiber E-PCL şeklinde kodlanmıştır. Şekil 1'de D-PCL ve E-PCL örneklerinin görüntüleri verilmiştir.



Şekil 1. E-PCL ve D-PCL örneklerinin görüntüleri

KARAKTERİZASYON

İki farklı yöntem ile üretilen PCL fiber ve filmlerin yapısal özelliklerini belirlemek için Fourier dönüşümlü infrared spektroskopisi (FTIR) analizleri Perkin Elmer spektrum 100 FTIR ile yapılmıştır. PCL fiberlerin görüntüleri taramalı elektron mikroskobu (SEM) ZEISS SUPRA 40 VP cihazı kullanılarak alınmıştır. Filmlerin ise optik mikroskopla (OM) yüzey görüntüsü alınmıştır. Kullanılan optik mikroskop Nikon marka ECLIPSE LV 150 modeldir. Örneklerin temas açısı ölçümleri Biolin Scientific, Theta Lite temas açısı ölçüm cihazı ile oda sıcaklığında gerçekleştirilmiştir. Fiber ve filmlerin XRD analizleri 2θ°= 5-50° açı aralığında ve 2º/dk.'lık tarama hızında yapılmıştır. Cu Kα (λ:1,5404) radyasyon kaynaklı PAN analitik, Empyrean marka XRD cihazı kullanılmıştır. Filmlerin ve fiberlerin kristalit boyut değerleri, Denklem 1 Scherrer Denklemi ile (Danilchenko ve ark., 2002), Mikro gerilme değerleri (ε) ise Denklem 2 (Monshi ve ark., 2012) kullanılarak hesaplanmıştır. Denklem 1'de yer alan L kristal boyut, k kristal şekille ilgili bir sabit, β analiz cihazı tarafından verilen FWHM değeri, λ dalga boyu ve θ Bragg açısıdır.

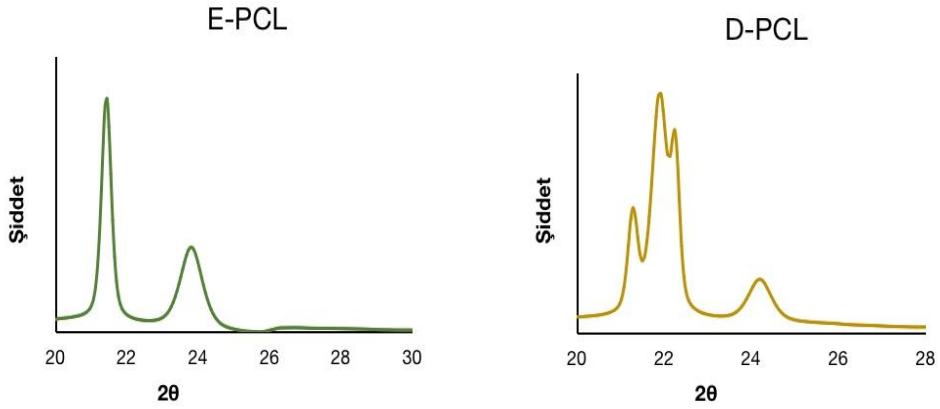
$$L = \frac{k.\lambda}{\beta.cos\theta} \quad (1)$$

$$\varepsilon = \frac{\beta}{4.tan} \quad (2)$$

BULGULAR VE TARTIŞMA

E-PCL ve D-PCL Örneklerinin XRD Analizi

PCL örneklerinin XRD analizinden elde edilen 2θ°, kristal boyut (A°), mikro gerilme (%) ve tabakalar arası mesafe (A°) değerleri Tablo 1'de, XRD difraktogramları ise Şekil 2'de verilmiştir. Şekil 2'de D-PCL'ye ait XRD difraktogramına bakıldığında 2θ°=21,27°, 21,88° ve 24,17° karşılık gelen PCL'nin karakteristik ortorombik kristal yapısını yansıtan (110), (111) ve (200) düzlemlerine ait pikler tespit edilmiştir. Bu sonuç literatürde daha önce yapılan PCL çalışma sonuçlarıyla uyumludur (Del Gaudio ve ark., 2011). Yine Şekil 2'den görüldüğü gibi E-PCL'nin yarı kristal yapısını yansıtan 2θº= 21.41° ve 23.79°'de (110) ve (200) düzlemlemlerine ait pikler tespit edilmiştir (Gautam ve ark., 2023). Tablo 1 incelendiğinde E-PCL ve D-PCL örneklerinin (110) düzlemine denk gelen değerleri karşılaştırıldığında D-PCL'nin tabakalar arası mesafe ve kristal boyut değerlerinin E-PCL'den daha yüksek olduğu görülmektedir. (200) düzlemine ait değerler karşılaştırıldığında ise tabakalar arası mesafe değerlerinin hemen hemen yakın ancak kristal boyut değerinin yine D-PCL'de yüksek olduğu, kristal boyut değerine bağlı olarak mikro gerilme değerinin de düşük olduğu belirlenmiştir. Tablo 1'den tespit edilen yüksek kristal boyut sonuçları solvent döküm yönteminin çekirdekleştirici etkisine atfedilmiştir (Uflyand ve ark., 2019).



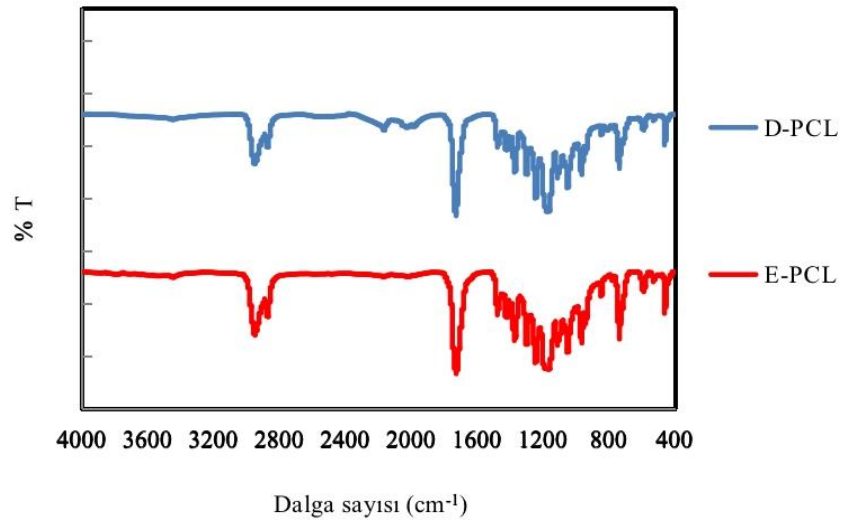
Şekil 2.E-PCL ve D-PCL örneklerinin XRD difraktogramları

Tablo 1. E-PCL ve D-PCL örneklerinin XRD analizinden elde edilen 2θ°, kristal boyut (A°), mikro gerilme (%) ve tabakalar arası mesafe (A°) değerleri.

ÖRNEKLER	2θ°	Tabakalar arası mesafe (Å)	Kristal boyut (Å)	Mikro gerilme (%)
E-PCL	21,4140	4,14958	297,6124	0,697145
	23,7906	3,74016	125,8231	1,486279
D-PCL	21,2701	4,17733	360,5816	0,579249
	21,8816	4,06195	221,2491	0,917958
	24,1790	3,68096	147,1053	1,251132

E-PCL ve D-PCL Örneklerinin FTIR Analizi

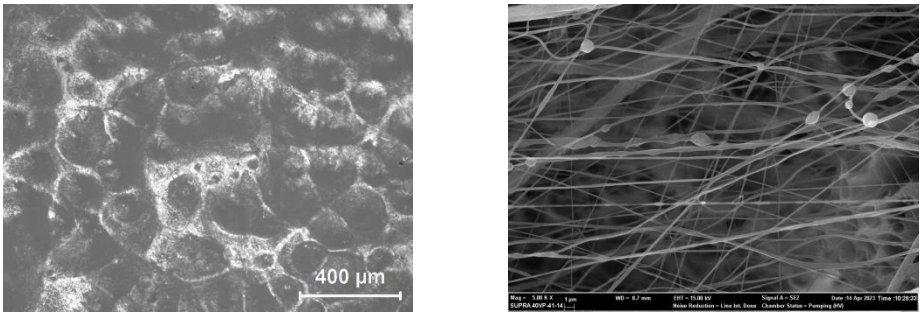
Şekil 3 E-PCL ve D-PCL örneklerinin FTIR spektrumunu göstermektedir. Şekil 3'ten görüldüğü gibi E-PCL ve D-PCL örneklerinin; 2934,41 cm^{-1} ve 2841,83 cm^{-1} dalga sayılarında sırasıyla -CH_2 grubunun asimetrik ve simetrik gerilme titreşimi, 1045,94 cm^{-1} 'de C-O-C epoksi, 731,83 cm^{-1} 'de CH_2 bükülme ve 711,23 cm^{-1} 'de ise C-C sallanma titreşimlerine ait bantlar tespit edilmiştir. Bu bantlar literatürde PCL'nin sahip olduğu fonksiyonel gruplara atfedilmektedir (Ishwarchand ve ark., 2021). Şekil 3 incelendiğinde D-PCL'nin fonksiyonel gruplarına denk gelen bantların E-PCL'nin sahip olduğu bant şiddetlerinden daha düşük olduğu görülmektedir. Literatürde bant şiddetinin düşüklüğü moleküler etkileşimin büyüklüğüne atfedilmiştir (Deng ve ark., 2018). Bu yüzden solvent döküm yöntemiyle üretilen D-PCL filminde tüm fonksiyonel gruplara ait moleküler etkileşimlerin E-PCL fiberine göre daha fazla olduğu anlaşılmıştır.



Şekil 3. E-PCL ve D-PCL örneklerinin FTIR spektrumları

E-PCL ve D-PCL Örneklerinin OM ve SEM Analizi

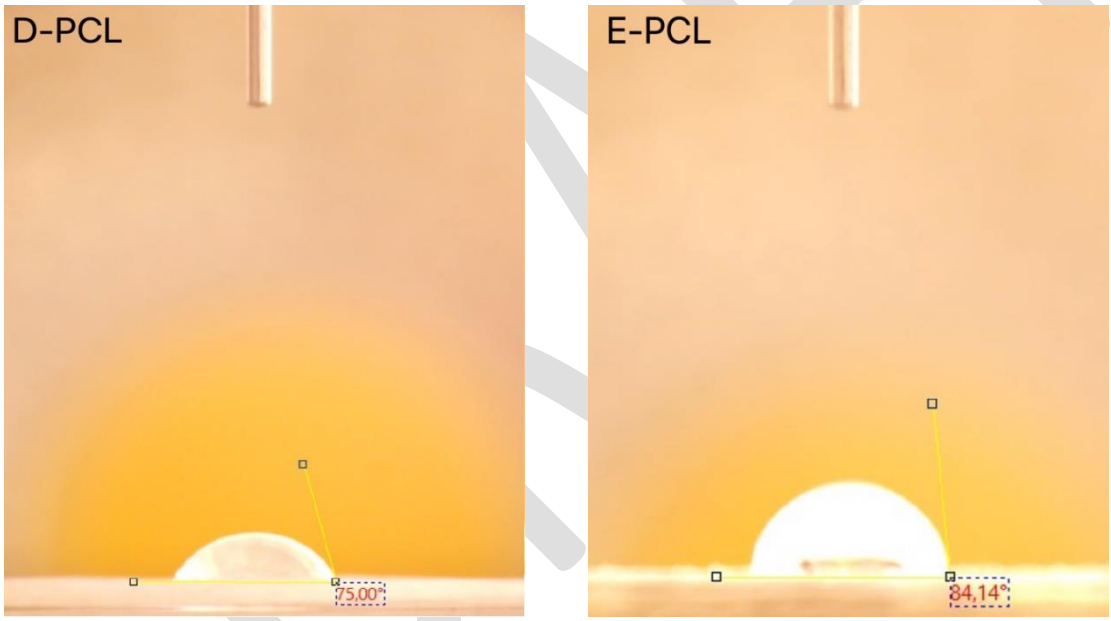
PCL örneğinin OM ve E-PCL örneğinin SEM görüntüsü Şekil 4'te verilmiştir. Şekil 4'te D-PCL örneğinin OM görüntüsünde çekirdekleşme noktaları ve PCL'nin karakteristik tane yapısı görülmektedir. Literatürde PCL'nin yüzey görüntüsü Şekil 4'te verilen görüntüyle benzer şekildedir (Castilla-Cortázar ve ark., 2019). E-PCL örneğinin SEM görüntüsünden ortalama fiber çapı 191.86 nm değerinde olduğu tespit edilmiştir. E-PCL örneğinin SEM görüntüsünde homojen boyutta nanofiberler ve bu nanofiberler üzerinde kısmen boncuk oluşumu görülmüştür. Fiberler üzerinde görülen boncuk oluşumu literatürde PCL'nin elektroeğirme çözeltisi içerisindeki Ağ.% miktarına atfedilmektedir (Fujihara ve ark., 2005).



Şekil 4. E-PCL ve D-PCL örneklerinin yüzey görüntüleri

E-PCL ve D-PCL Örneklerinin Temas Açısı Ölçüm Sonuçları

D-PCL ve E-PCL örneklerinin temas açısı ölçüm sonuçları Şekil 5'te verilmiştir. Şekil 5'te görüldüğü gibi D-PCL örneğinin temas açısı 75,00º ve E-PCL örneğinin temas açısı 84,14º olarak ölçülmüştür. Temas açısı ölçüm sonuçlarına göre solvent döküm ve elektroeğirme yöntemleriyle üretilen film ve nanofiber örneklerinin temas açıları birbirine yakın sonuçlar vermiştir. Literatürde elektroeğirme yöntemiyle üretilmiş PCL nanofiberin temas açısı sonucu 85,00º olarak ölçülmüştür (Joseph ve ark., 2023). PCL filmlerin literatürde hidrofobik özellikte olduğu, temas açısının ise 74-78º arasında tespit edildiği rapor edilmiştir (Gümüşderelioğlu ve ark., 2011). Bu çalışmada hem E-PCL hem de D-PCL için bulunan temas açısı sonuçları literatürle birebir benzer olduğu belirlenmiştir.



Şekil 5. D-PCL ve E-PCL örneklerinin temas açısı ölçümleri

SONUÇ

Bu çalışmada solvent döküm ve elektroeğirme yöntemleriyle üretilen PCL film ve nanofiberlerin yapısal özellikleri karşılaştırılmıştır. XRD analiz sonuçlarına göre her iki yöntemle de üretilen E-PCL ve D-PCL'nin kristal yapılarının benzer olduğu ancak D-PCL'nin kristal boyut değerlerinin daha yüksek olduğu belirlenmiştir. Bu sonuç solvent döküm yönteminin çekirdekleşme etkisi üzerinde daha etkin olduğunu ortaya çıkarmıştır. FTIR analiz sonuçları da D-PCL'nin moleküler arası etkileşimlerinin daha fazla olduğunu göstermiştir. D-PCL ve E-PCL örneklerinin yüzey görüntülerinde D-PCL örneğinin karakteristik tane yapısı, E-PCL örneğinin ise homojen boyutta nanofiberleri görüntülenmiştir.Temas açısı ölçüm sonuçları D-PCL ve E-PCL örneklerinin hidrofobik özellik sergilediklerini göstermiştir.

KAYNAKLAR

Alharbi N, Daraei A, Lee H, Guthold M 2023. The effect of molecular weight and fiber diameter on the mechanical properties of single, electrospun PCL nanofibers. Materials Today Communications, 35: 105773.

Armentano I, Del Gaudio C, Bianco A, Dottori M, Nanni F, Fortunati E, Kenny JM 2009. Processing and properties of poly (ε-caprolactone)/carbon nanofibre composite mats and films obtained by electrospinning and solvent casting. Journal of materials science, 44: 4789-4795.

Castilla-Cortázar I, Vidaurre A, Marí B, Campillo-Fernández AJ 2019. Morphology, crystallinity, and molecular weight of poly (ε-caprolactone)/graphene oxide hybrids. Polymers, 11(7): 1099.

Danilchenko SN, Kukharenko OG, Moseke C, Protsenko IY, Sukhodub LF, Sulkio-Cleff B 2002. Determination of the Bone Mineral Crystallite Size and Lattice Strain from Diffraction Line Broadening. Crystal Research Technology, 37(11): 1234-1240

Del Gaudio C, Ercolani E, Nanni F, Bianco A 2011. Assessment of poly (ε-caprolactone)/poly (3-hydroxybutyrate-co-3-hydroxyvalerate) blends processed by solvent casting and electrospinning. Materials Science and Engineering: A, 528(3):1764-1772.

Deng L, Kang X, Liu Y, Feng F, Zhang H 2018. Characterization of gelatin/zein films fabricated by electrospinning vs solvent casting. Food Hydrocolloids, 74: 324-332.

Fujihara K, Kotaki M, Ramakrishna S 2005. Guided bone regeneration membrane made of polycaprolactone/calcium carbonate composite nano-fibers. Biomaterials, 26(19): 4139-4147.

Gautam S, Purohit SD, Singh H, Dinda AK, Potdar PD, Sharma C, Mishra NC 2023. Surface modification of PCL-gelatin-chitosan electrospun scaffold by nano-hydroxyapatite for bone tissue engineering. Materials Today Communications, 34: 105237.

Ghosal K, Kováčová M, Humpolíček P, Vajďák J, Bodík M, Špitalský Z 2021. Antibacterial photodynamic activity of hydrophobic carbon quantum dots and polycaprolactone based nanocomposite processed via both electrospinning and solvent casting method. Photodiagnosis and Photodynamic Therapy, 35:102455.

Gümüşderelioğlu M, Kaya FB, Beşkardeş IG 2011. Comparison of epithelial and fibroblastic cell behavior on nano/micro-topographic PCL membranes produced by crystallinity control. Journal of colloid and interface science, 358(2): 444-453.

Ishwarchand W, Sarakar G, Swain BP 2021. Investigation of optical properties, chemical network and electronic environments of polycaprolactone/reduced graphene oxide fiber nanocomposites. Polymer Bulletin, 1:17.

Joseph B, John AJ, Glamočlija J, Stojković D, Soković M, Lazović S,Thomas S 2023. Processing and evaluation of the structure-properties of electrospun PCL/zirconium nanoparticle scaffolds. Materials Today Communications, 34: 104961.

Monshi A, Foroughi MR, Monshi MR 2012. Modified Scherrer Equation to Estimate More Accurately Nano-Crystallite Size Using XRD. World Journal of Nano Science and Engineering, 2: 154-160.

Salević A, Stojanović D, Lević S, Pantić M, Đorđević V, Pešić R, Nedović V 2022. The structuring of sage (Salvia officinalis L.) extract-incorporating edible zein-based materials with antioxidant and antibacterial functionality by solvent casting versus electrospinning. Foods, 11(3): 390.

Uflyand IE, Drogan EG, Burlakova VE, Kydralieva KA, Shershneva IN, Dzhardimalieva GI 2019. Testing the mechanical and tribological properties of new metal-polymer nanocomposite materials based on linear low-density polyethylene and Al65Cu22Fe13 quasicrystals. Polymer Testing, 74: 178-186.

DRAFT

ORAL PRESENTATION

Akdeniz midyesinden (*Mytilus galloprovincialis*) üretilen pilakilerin mikrobiyolojik kalitelerinin belirlenmesi

İbrahim Ender Künili[1*] (*ORCID: https://orcid.org/0000-0003-2830-6979*), Selin Özge Dinç[2] (*ORCID: https://orcid.org/0000-0003-1597-1929*)

[*1] Çanakkale Onsekiz Mart Üniversitesi, Deniz Bilimleri ve Teknolojisi Fakültesi, Su Ürünleri Avlama ve İşleme Teknolojisi Bölümü, Çanakkale, Türkiye
[2] Çanakkale Onsekiz Mart Üniversitesi, Uygulamalı Bilimler Fakültesi, Gıda Teknolojisi Bölümü, Çanakkale, Türkiye

*Sorumlu yazar e-mail: enderkunili@yahoo.com

Özet

Akdeniz midyesi (*Mytilus galloprovincialis*) ekonomik öneme sahip çift kabuklu türüdür. Dünyada birçok mutfakta yer edinmiş bu türün, ülkemizde tüketimi midye dolma ve midye tava şeklinde yaygındır. Ancak mikroorganizma bakımından risk içerebilen bu ürünlerin güvenli tüketimleri için alternatif ürün üretim modellerine ihtiyaç duyulmaktadır. Bu çalışmada, ülkemiz mutfağında yer edinmiş pilakinin Akdeniz midyesinden üretimi amaçlanmış, üretilen ürünlerin ise önemli bozulma parametrelerinden olan mikrobiyolojik kalitesinin değişimi incelenmiştir. Midye pilakiler sous-vide metodu kullanılarak, çiğ midye etlerinden sade ve sebzeli olarak üretilmiştir. Elde edilen bulgulara göre, çiğ midye etinde tespit edilen toplam aerobik mezofilik bakteri (TAMB) 4,47 log kob/g, maya ve küf (MK) 2,95 log kob/g, Bacillus sp. 1,65 log kob/g, Staphylococcus ve Micrococcus (S&M) 1,21 log kob/g seviyelerinde tespit edilmiş, diğer mikroorganizmalara ise rastlanamamıştır. Taze midye örneklerinde tespit edilen mikroorganizma seviyeleri ise ürünlerin ilk üretildiği gün de yaklaşık 2,0 – 3,0 log kob/g azalmış sırasıyla SM ve PM ürünleri için TAMB 2,37 – 1,34 log kob/g, MK 1,48 – 1,15 log kob/g olarak tespit edilmiş olup, diğer mikroorganizmalar belirlenememiştir. Midye pilaki ürünlerinin raf ömürleri ise mikrobiyolojik açıdan17 gün olarak tespit edilmiştir.
Sonuç olarak, Akdeniz midyesinden elde edilen pilaki ürünlerin hem görünüm hem de lezzet olarak tüketicinin ilgisini çekebileceği, ancak mikrobiyolojik açıdan ürün ticarileşebilmesi için, raf ömrünü uzatmak amacıyla ek yöntemlerin araştırılması ve uygulanmasının gerekebileceği düşünülmektedir.

Anahtar Kelimeler: Akdeniz midyesi, *Mytilus galloprovincialis*, midye pilaki, sous-vide, mikrobiyolojik kalite

Determination of microbiological quality of stews produced from Mediterranean mussels (*Mytilus galloprovincialis*)

Abstract

Mediterranean mussel (*Mytilus galloprovincialis*) is a bivalve species of economic importance. This species, which has gained a place in many cuisines in the world, is widely consumed in our country in the form of stuffed mussels and fried mussels. However, alternative product production models are needed for the safe consumption of these products, which may contain risks in terms of microorganisms. In this study, it was aimed to produce mussel stew, which has a place in the cuisine of our country, from Mediterranean mussels, and the change in microbiological quality of the produced products, which is one of the important spoilage parameters, was examined. Mussel stews were produced plain and with vegetables from raw mussel meat using sous-vide method. According to the results obtained, total aerobic mesophilic bacteria (TAMB) 4.47 log cfu/g, yeast and mould (MK) 2.95 log cfu/g, *Bacillus* sp. 1.65 log cfu/g, Staphylococcus and Micrococcus (S&M) 1.21 log cfu/g and other microorganisms were not detected in raw mussel meat. The levels of microorganisms detected in fresh mussel samples decreased approximately 2.0 - 3.0 log cfu/g on the first day of production and TAMB 2.37 - 1.34 log cfu/g, MK 1.48 - 1.15 log cfu/g for SM and PM products, respectively, and other microorganisms were not detected. The shelf life of mussel stewed products was determined as 17 days from microbiological point of view. In conclusion, it is thought that stewed products obtained from

Mediterranean mussels may attract the attention of consumers both in terms of appearance and taste, but additional methods may need to be investigated and applied to extend the shelf life to commercialise the product from a microbiologically.

Keywords: Mediterranean mussel, *Mytilus galloprovincialis*, mussel stew, Sous-vide, microbiological quality

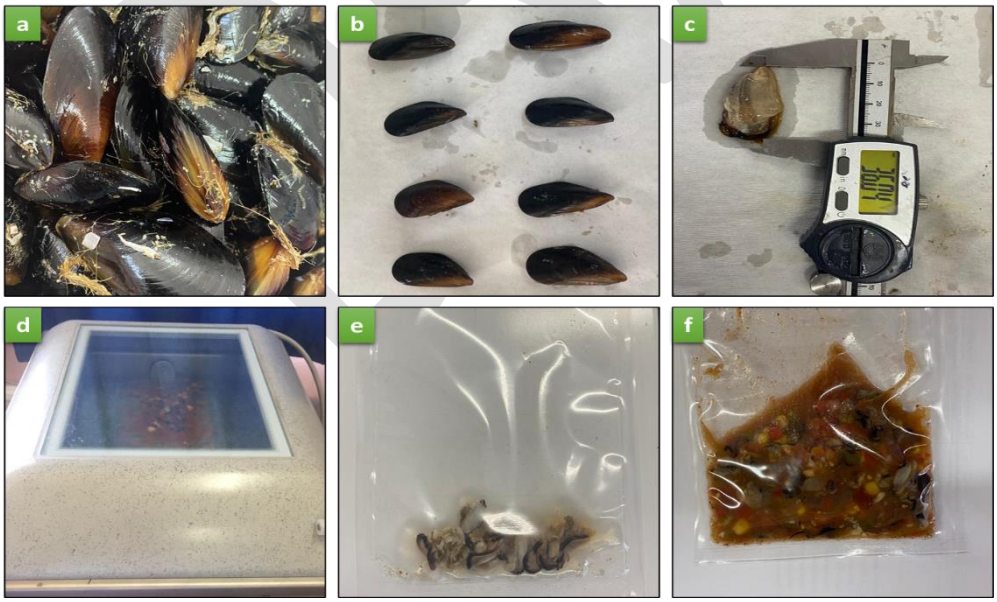
Giriş

Akdeniz midyesi (*Mytilus galloprovincialis*), dünyada hem lezzeti hem de besleyici özellikleri nedeniyle birçok geleneksel mutfakta yer edinmiş deniz ürünüdür. Ülkemizde yaygın olarak, midye dolma ve midye tava olarak tüketimi bulunan bu türün, alternatif ve güvenli ürünlere dönüştürülmesi, midyelerin yaşam şekillerinden ve üretim aşmalarında ekstra maaliyet ile zaman kaybından dolayı önem taşımaktadır. Bununla birlikte, tüketici kitlesine alternatif ürün sunulması ve ürün yelpazesinin genişletilmesi de bu noktada alternatif ürünlerin üretimi ile mümkün olabilmektedir.

Pilaki, genel olarak et ürünlerinin, çeşitli sebzeler ile hammadde türüne göre değişiklik gösteren süre ve sıcaklıklarda pişirilmesi işlemidir. Uygun süre ve sıcaklık uygulamaları ile hammadde içeriğinde yer alan mikroorganizma sayı ve türlerinin azaltılması da mümkün olacağından hem yeni ürünlerin üretimi hem de hammaddelerin bozulmasını geciktirerek tüketim süresinin uzatılması mümkün olabilmektedir. Ancak, midye gibi çift kabuklu yumuşakça türlerinde, yaşam şekillerinin aslını oluşturan bulundukları suyu süzerek beslenmelerinden dolayı başta mikroorganizmalar olmak üzere, birçok riski barındırabilmektedirler. Dolayısıyla, iç ve dış pazarda potansiyel sorun olabilecek bu durumlarda hem tüketiciye alternatif ürün sunumu hem de mikrobiyolojik açıdan standartlara uygun güvenli ürünlerin üretimine yönelik araştırmalar önem taşımaktadır. Yapılan bu çalışmada, Akdeniz midyesinden sous-vide metodu kullanılarak pilaki üretimi gerçekleştirilmiş, elde edilen ürünlerin çiğ ürünlerdeki mikrobiyolojik kaliteleri ile raf ömrü süresince değişimleri belirlenerek, potansiyel yeni ürün üretimi için bir ön çalışma gerçekleştirilmiştir.

Materyal ve Metot

Araştırma materyali olarak Akdeniz midyesi (*Mytilus galloprovincialis*) kullanılmıştır (Şekil 1a-c). Toplam ağırlığı yaklaşık 6 kg olan ve her birinin boyu 5 cm üzerinde olan porsiyonluk örnekler (Şekil 1b) Çanakkale Boğazı'ndan 2023 Temmuz ayında SCUBA donanımlı dalış sırasında el ile toplanmıştır. Toplanan örnekler Çanakkale Onsekiz Mart Üniversitesi laboratuvarlarına 2 saat içinde buz aküleri ile soğutulmuş yalıtımlı polietilen kutularda taşınmıştır. Laboratuvara getirilen örneklerin kırık kabuk ve canlılık kontrolleri yapıldıktan sonra ayrılan canlı ve sağlam örneklerden pilaki üretimi gerçekleştirilmiştir (Şekil 1d-f).



Şekil 1 – Akdeniz midyesi örnekleri **(a)**, örneklerin mikrobiyolojik ekim öncesi hazırlığı **(b)**, midye eti ölçümü **(c)**, sous-vide metodunda uygulanan vakum paketleme **(d)**, üretilen sade **(e)** ve sebzeli **(f)** pilakilerin görüntüsü

Midye Pilaki Üretimi

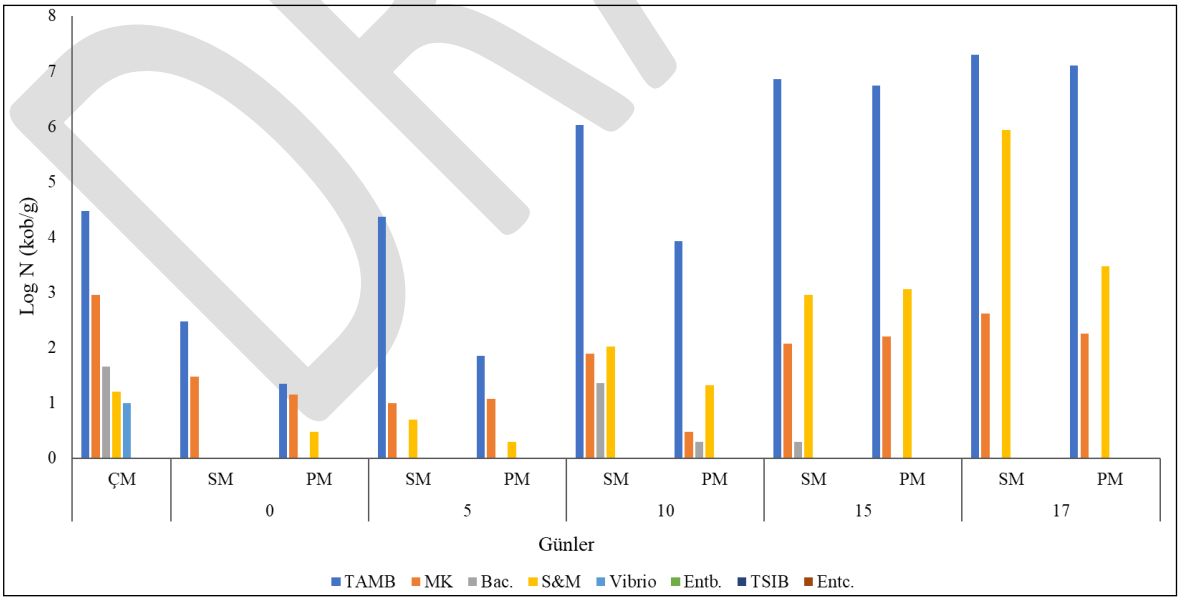
Pilaki üretimi için sous-vide metodu kullanılmış olup, haşlanmamış midye etleri sade (SM) ve sebzeli (PM) olarak vakum paketleme yapılmış ve 80ºC'de 15 dk vakum paket ile su banyosunda pişirilmiştir. Haşlama ile midye eti çıkarma işleminin sıcaklık ve süresi ise 95ºC'de 2 dk olarak uygulanmıştır. Sebzeli pilaki uygulaması için kullanılan sebzeler, domates, sarımsak, mısır, biber ve soğandan oluşmaktadır. Baharatlar ise yalnızca tuz ve karabiberdir. Üretilen her iki grup pilaki ürün mikrobiyolojik açıdan raf ömürleri süresince +4 ºC'de depolanmıştır.

Mikrobiyolojik Analizler

Midyelerin kabukları kum, çamur ve mikroorganizmalardan arındırıldıktansonra, alkollü pamuk ile temizlenmiştir. Steril bıçak yardımıyla açılan canlı midyelerin içinde bulunan kabuklar arası sıvı ve midye etlerinden toplam 10 g homojen örnek alınarak, 90 ml bakteriyolojik pepton (%0,1 Pepton + % 1NaCl) içerisine tartılmıştır. Tartım sonrasında örnekler Stomacher (Seward, 400) cihazında, 2 dk süre ile 2300 rpm'de homojen edilmiştir. Homojen edilen örneklerin yine peptonlu su kullanılarak 10^{-6}'ya kadar desimal seyreltmeleri yapılmıştır. Yapılan her seyreltmeden yayma plak yöntemi ile plate count agar (PCA-Merck), malt extract agar (MAE), mannitol egg yolk polymyxine (MYP-Merck), Baird-Parker agar (BP), thio citrate bile sucrose (TCBS-Merck) agar tryptose sulfite cyclocerine (TSC-Merck) agar, Endo Agar (Merck) ve D-coccosel (Biomeriux) besiyerlerine ekimleri yapılmıştır (FDA/BAM, 1998). Midye pilaki örneklerinden, ekim yapılan tüm petriler üreticilerin verdiği süre ve sıcaklıkta inkübasyona tabi tutulmuş, inkübasyon sonunda üreyen koloniler seçilerek biyokimyasal testler ile ön tanılama yapılmıştır.

Bulgular ve Tartışma

Bu çalışmada Çanakkale Boğazı'ndan toplanan porsiyonluk Akdeniz midyelerinden, mikrobiyolojik kontrolün sağlanması ve farklı tüketim şekillerinin aynı anda araştırılması amacıyla midye pilaki ürün üretimi gerçekleştirilmiştir. Sous-vide metodu esas alınarak üretilen ürünlerin görüntüleri Şekil 1e ve Şekil 1f'de gösterilmiş, elde edilen bu ürünlerin hammadde ve kontrol grubuna göre mikrobiyolojik kaliteleri ise Şekil 2'de özetlenmiştir.



ÇM: Çiğ midye eti, SM: Sade midye pilaki, PM: sebzeli midye pilaki, Kob: koloni oluşturan birim, TAMB: Toplam aerobik mezofilik bakteri, MK: maya ve küf, Bac.: Bacillus sp., S&M: Staphylococcus & Micrococcus, Entb.: Enterobacteriaceae, TSIB: Toplam sülfit indirgeyen bakteri, Entc.: Enterococcus sp.

Şekil 2: Taze midye eti, sade midye pilaki ve midye pilaki ürünlerinin raf ömrü süresince mikroorganizma seviyelerinin (log N kob/g) günlere göre değişimi.

Araştırma kapsamında kullanılan taze midye örneklerinin mikroorganizma seviyeleri sırasıyla toplam aerobik mezofilik bakteri (TAMB) 4,47 log kob/g, maya ve küf (MK) 2,95 log kob/g, *Bacillus* 1,65 log kob/g, *Staphylococcus* ve *Micrococcus* (S&M) 1,21 log kob/g tespit edilmiş, diğer mikroorganizmalara ise rastlanamamıştır. Taze midye örneklerinde tespit edilen mikroorganizma seviyeleri ise ürünlerin ilk üretildiği gün de yaklaşık 2,0 – 3,0 log kob/g azalmış sırasıyla SM ve PM ürünleri için TAMB 2,37 – 1,34 log kob/g, MK 1,48 – 1,15 log kob/g olarak tespit edilmiş olup, diğer mikroorganizmalar belirlenememiştir (Şekil 2). Ürünler depolandığı +4 ºC koşullarında 0, 5, 10, 15 ve 17 günlerde mikrobiyolojik analizleri yapılmış ve raf ömürleri benzer ürünler için belirtilen kriterlerde de geçerli olan log 6,0-7,0 kob/g (ICMSF, 1986; Anonim, 2011) seviyesine ulaşıncaya kadar bu analizler sürüdürülmüştür. TAMB sayısı log 7.0 sayısına ulaşan ilk mikroorganizma grubu olarak belirlenmiş, 17. Günde her SM ve PM gruplarında sırasıyla 7,3 ve 7,1 olarak tespit edilmiştir. Bununla birlikte, taze örneklerde ve depolamanın ilk günlerinde pilaki ürünlerde az sayılarda tespit edilen *Staphyloccocus – Micrococcus* (0 – 1,2 log kob/g) depolamanın ilerleyen günlerinde SM grubunda 5,93 log kob/g, PM grubunda ise 3,48 log kob/g seviyelerine kadar yükselmiştir. Diğer taraftan taze ürünlerde 1,65 log kob/g seviyelerinde olan *Bacillus* sp. pilaki ürünlerde tespit edilebilir değerlerin altında bulunmuştur. Genel olarak midye pilaki ürünlerde tespit edilen mikrofloranın TAMB, MK ve S&M mikroorganizma grupları ile şekillendiği belirlenmiştir. Midyelerin buzdolabı koşullarında raf ömürleri ancak haşlama sonrası elde edilen etler üzerinden yapılan analizler aracılığıyla belirlenebilmektedir. Midyeler öldükleri andan itibaren tüketim için uygunluğunu kaybetmektedirler. Dolayısıyla, ürünler ya canlı olarak tüketime kadar soğuk koşullarda bekletilir ya da pişirilerek veya etleri ayıklandıktan sonra işlenerek depolanabilmektedir.

Yapılan çalışmalar arasında ısıl işlem uygulanmış ve soğukta depolanmış midyelerin raf ömürleri paketleme türüne bağlı olarak oldukça değişken olabilmektedir. Turan ve ark., (2013) yaptıkları çalışmada çiğ midye etlerinde (*Mytilus galloprovincialis*) başlangıçta, 3,94 log kob/g tespit ettikleri TAMB sayısını raf ömrü sonu olarak belirledikleri dördüncü gün sonunda 7,54 log kob/g olarak tespit etmişlerdir. Kaba ve Erkoyuncu (2011) çiğ midye eti örneklerinde TAMB sayısını 5,28 log kob/g, raf ömrü sonu olan altıncı günde ise %0,2 potasyum sorbat katkılı çiğ etlerin 6,60 log kob/g ve haşlanmış katkılı etlerin ise 6,49 log kob/g olarak rapor etmişlerdir. Bildirilen bu iki çalışmada taze midye eti örnekleri için rapor edilen değerler benzer bulunmuş ancak katkı kullanarak haşlandıktan sonra paketlenmiş midye etleri için rapor edilen altı gün raf ömründen, çalışmamızdaki her iki üründe tespit edilen raf ömrü daha uzun bulunmuştur. Ayrıca, ilk uygulama sonrasında mikroorganizma yüklerindeki düşüş yine çalışmamızda daha yüksek seviyede bulunmuştur.

Sonuç

Sonuç olarak bu çalışma ile porsiyonluk Akdeniz midyelerinin geleneksel tüketim şekli olan midye dolma ve midye tava ürünlerine alternatif olarak midye pilaki ürün üretilmiştir. Sous-vide metoduna dayalı üretim metodu ile aynı zamanda başlangıç mikroorganizma yüklerinde önemli düşüşler gerçekleşerek raf ömürlerinin de uzatılması sağlanmıştır. Alternatif üretim metotlarının ayrı ayrı veya kombine olarak, bu şekilde denenmesi, ürün yelpazesinin artırılmasına olanak sağlarken, ürün zayiatı, ekstra iş gücü ve zaman kaybının önüne geçme potansiyeli taşıyacağı düşünülmektedir.

Kaynaklar

Anonim, 2011. Türk Gıda Kodeksi Mikrobiyolojik Kriterler Yönetmeliği, Resmi Gazete, Sayı: 28157.

FDA, Bacteriological Analytical Manual, 8th ed. Revision A, Food and Drug Administration, AOAC International, Washington, DC. (1998).

ICMSF. (1986). Microorganisms in foods. 2.sampling for microbiological analysis: principles and specific applications, 2nd ed. University of Toronto Press, Buffalo, NY.

Kaba, N. Ve Erkoyuncu, İ. (2011). Değişik İşlemler Uygulanan Midyelerde (Mytilus galloprovincialis Lamarck, 1819) Soğuk Depolama Sırasında Kalite Değişiminin Belirlenmesi. Akademik Gıda, 9 (6): 29-37.

Turan, H., Kocatepe, D., Altan, C.O., Erkoyuncu, İ. (2013). Soğukta Saklanan Midyelerin (Mytilus galloprovincialis L. 1819) Raf Ömrünün Belirlenmesi. Akademik Gıda 11 (3-4); 47-51.

ORAL PRESENTATION

Control of pre-slaughter and slaughter stress procedures that dromedaries undergo in Moroccan slaughterhouses

Abderrahim Moussahil[1]* (https://orcid.org/0000-0003-3321-8252) , Abdelilah Lemrhamed[1], Mohamed Farh[1], Abdelghani Iddar[2] and Mohammed El Khasmi[1]

[1] Laboratory of Physiopathology and Molecular Genetics, Faculty of Sciences Ben M'Sik, University Hassan II of Casablanca, PB. 7955 Sidi Othmane, Casablanca, Morocco.
[2] Unit of Radio-Immuno-Analysis/Division of Life Sciences, CNESTEN, PB. 1382 RP 10001, Rabat, Morocco.

*Correspondingauthor e-mail: moussahil.abderrahim@gmail.com

Abstract

Animals designated for slaughter undergo various stressors that can adversely impact their physiological functions and the quality of their meat. This study aimed to assess these stress factors during the pre-slaughter and slaughter processes in camels. A survey was conducted in Casablanca, Tit-Mellil Mediouna, Settat, and Mers Lkhir Skiratt-Temara slaughterhouses in Morocco, involving the evaluation of 246 camels from loading onto vehicles to the bleeding process. The findings revealed that a significant number of camels were transported in non-ventilated vehicles with covered roofs and dark floors. During transport, these camels were not mixed with other animal species, loaded using pushes and pulls with ropes due to the absence of ramps. They were transported in vehicles with slippery floors without any bedding and were led to the lairage area and slaughter room using ropes attached to their forelimbs. Many camels experienced longer transport distances and higher loading densities.
Furthermore, all camels were slaughtered according to halal practices without stunning. Most of the animals endured extended waiting times before loading and required a prolonged duration for the entire process, including loading, transport, unloading, lairage, water and food deprivation, escort to the slaughter room, slaughtering, and bleeding. During their journey to the lairage area and the slaughter room, the animals exhibited frequent falls, urination, defecation, and vocalizations. In our country, the animal welfare standards set by the World Organization for Animal Health are not consistently upheld. Often, the vehicles are multi-purpose and not specifically designed or equipped for camel transportation, lacking proper loading and unloading facilities and offering no protection from the sun. The camels were handled roughly by untrained and inexperienced handlers and were transported without any bedding in excessively high ambient temperatures.
In conclusion, in the municipal slaughterhouses examined, all the activities and procedures preceding slaughter could negatively impact the well-being of camels and compromise the quality of their carcasses and meat. Therefore, it is imperative for managers to adhere to internationally recognized standards for the appropriate welfare of camels in various slaughterhouses across Morocco.

Keywords: Preslaughter stress, Camels, Slaughterhousses, Survey, Morocco.

INTRODUCTION

The management of camels on farms, at markets, and in slaughterhouses, as well as their loading into trucks, transportation, unloading, and stabling at slaughterhouses, can potentially induce stress, leading to what is referred to as "pre-slaughter stress" (El Khasmi et al., 2015; Lemrhamed et al., 2018; Lemrhamed et al., 2019). Despite the longstanding attention of scientific researchers in developed countries to the welfare of farmed animals and its impact on product quality, this area of research remains relatively underexplored in underdeveloped regions, particularly concerning camels. Conversely, farm animals often encounter various

welfare challenges during and after transport, including prolonged periods of water and food deprivation, fatigue, injuries, diseases, handling, intermingling with different species, and exposure to diverse environmental conditions (Roadknight et al., 2021). Additionally, animals face additional potential stressors during transport, such as climatic conditions, accelerations, vibrations, noise, space limitations, road conditions, vehicle design, and air pollutants (Padalino and Raidal, 2020). The objective of this study was to conduct a survey on the pre-slaughter stress conditions experienced by camels, encompassing their loading into vehicles up to the slaughtering process, in Moroccan slaughterhouses.

MATERIALS AND METHODS

A survey was carried out on 246 male and female camels (Camelus dromedarius) to evaluate the stress conditions before slaughter and during the slaughtering process, spanning from loading to bleeding, in slaughterhouses located in Casablanca (63 camels), Tit-Mellil (59 camels), Settat (64 camels), and Mers Lkhir Skiratt-Temara (60 camels) in Morocco. This survey focused on various aspects, including animal and transport vehicle identification, camel positioning in the vehicles, loading density, co-mingling with different species, loading and unloading procedures, the process of guiding camels to the lairage area and slaughter room, and the slaughtering process. It also considered factors such as transport distance and duration, as well as loading, unloading, lairage, water and food deprivation, accompaniment to the slaughter room, slaughtering, and bleeding times. Additionally, the survey documented the frequencies of falls, urination, defecation, and vocalizations during the guidance of camels to the lairage area and slaughter room, as well as the presence of bruises on the carcasses. For each parameter, proportions were calculated by dividing the observed events by the total number of animals in the respective group. Descriptive analysis was performed using the statistical software SPSS version 19 to identify variables of interest.

The study examined variations among different groups of camels. The Chi-square test was employed to identify significant factors related to slaughterhouse conditions. Variations were deemed statistically noteworthy at a significance level of $P < 0.05$.

RESULTS

The majority of the camels included in the study were male (60.98%), aged between 3 and 9 years, weighing between 300 and 400 kg. They were predominantly intensively bred, sourced from markets, and individually valued between 15,500 and 24,000 DH. A significant portion of the camels (58.54%) were transported to the slaughterhouses in vans. For 72.36% of the animals, the transport vehicles lacked ventilation, had a covered roof, and had a dark floor. During transportation, 86.99% of the camels were not mixed with other species. All camels (100%) were loaded using pushes and pulls with ropes due to the absence of ramps. They were transported in vehicles with slippery floors without any bedding. Upon arrival at the lairage area and slaughter room, they were guided using pushing and pulling techniques with ropes while being restrained by a rope attached to the forelimb. Additionally, all camels were slaughtered in accordance with Islamic methods, without stunning.

The study's findings indicate that a significant number of camels waited between 25 and 36 hours before being loaded onto vehicles (78.86%). Loading required between 16 and 25 minutes for 57.52% of the camels. Transport and unloading times were, respectively, 10 to 11 hours for 68.29% of the camels and 11 to 20 minutes for 51.22% of the animals ($P < 0.05$). The durations of stay in the lairage and deprivation of water and food were between 8 and 12 hours for 71.54% of the camels and 25 to 72 hours for 69.11% ($P < 0.05$). The time taken for the animals to be guided to the slaughter room, slaughtered, and bled out were, respectively, between 11 and 20 minutes for 41.47%, 11 and 20 minutes for 45.53%, and 8 to 9 minutes for 54.47% of the camels ($P < 0.05$).

According to the same study, the transport distances and loading densities were, respectively, 120 to 350 km for 60.98% and less than 2.75 m2/animal for 51.22% of the camels. During the guidance of the animals to the lairage area and slaughter room, the frequencies of falls, urination, defecation, and vocalizations were, respectively, 5 to 10 times for 69.11%, 3 to 5 times for 65.85%, 2 to 4 times for 64.23%, and 15 to 40 times for 87.80% of the camels ($P < 0.05$).

DRAFT

DISCUSSION

The camels under examination experienced both psychological and physical stressors. Psychological stress arose from disruptions in social interactions, unforeseen events, and a lack of familiarity with various situations encountered by the animals. Physical stress resulted from factors such as water and food deprivation, exhaustion, collisions with enclosure elements or other animals, human interventions, and inter-animal conflicts during transportation and housing. The prolonged durations of loading, unloading, guiding to the slaughter room, slaughter, as well as increased instances of urination, defecation, and vocalizations, can be attributed, at least in part, to the animals' reluctance to move forward, attempts to walk backward, immobility, resistance despite operator interventions, escape efforts, slips, and/or falls. These observed reactions indicate a state of fear and a disruption in the well-being of the camels (Grandin and Shivley, 2015).

In this study, the camels were loaded and unloaded without the use of specialized facilities, transported in vehicles not designed for camel transport, and guided by ropes with forelimb restraint, potentially leading to depletion of muscle glycogen and elevated postmortem meat pH (Mounier et al., 2006). Furthermore, the slaughter process itself introduced a stress element, as it was performed in accordance with the halal procedure without prior stunning, rendering the animals conscious and sensitive. Consequently, prolonging this procedure could be significantly more stressful for camels that haven't undergone prior stunning. Specifically, the dromedaries were positioned in a squatting stance, with their front legs secured using a rope at the knees. The head was held in a caudal position (i.e., facing the tail). The animals were in visual and olfactory proximity to the operator responsible for the halal bleeding, the knife, and the dromedaries already slaughtered on the ground, potentially causing physical and psychological stress due to the release of volatile compounds from urine, blood, and feces (Terlouw and Bourguet, 2022).

CONCLUSION

In the four Moroccan municipal slaughterhouses examined in this study, camels face stressful conditions from the point of loading until slaughter. These conditions have the potential to disrupt the physiological balance of these animals and impact the quality of their meat. As a result, it is imperative for managers to take action and adhere to internationally recognized standards for the proper welfare of camels within these slaughterhouses.

REFERENCES

El Khasmi, M., Chakir, Y., Bargaâ, R., Barka, K., Lektib, I., El Abbadi, N., Belhouari, A., Faye, B.: Impact of transport distance on stress biomarkers levels in dromedary camel (*Camelus dromedarius*). Emir. J. Food Agric., 27(6), 507-512 (2015).

Grandin, T., Shivley, C.: How farm animals react and perceive stressful situations such as handling, restraint, and transport. Animals, 5(4), 1233-1251 (2015).

Lemrhamed, A., Farh, M., Riad, F., El Abbadi, N., Tahri, E.H., Belhouari, A., Faye, B., El Khasmi, M.: Evaluation of stress responses induced by the loading density in dromedary camel «*Camelus dromedarius*». Emir. J. Food Agric., 30(9), 803-808 (2018).

Lemrhamed, A., Tabite, R., Farh, M., Riad, F., El Abbadi, N., Tahri, E.H., Faye, B., El Khasmi, M.: Evaluation of preslaughter stress responses during waiting time at lairage in the dromedary camel (*Camelus dromedarius*). J Camel Pract Res 26,149-156 (2019).

Mounier, L., Dubroeucq, H., Andanson, S., Veissier, I., Etoile, M.L.: Variations in meat pH of beef bulls in relation to conditions of transfer to slaughter and previous history of the animals 1. Journal of Animal Science, 84, 1567-1576 (2006).

Padalino, B., Raidal, S.R.: Effects of Transport Conditions on Behavioural and Physiological Responses of Horses.Animals, 10,160 (2020).

Roadknight, N., Mansell, P., Jongman, E., Courtman, N., Fisher, A.: Invited review: The welfare of young calves transported by road. J. Dairy Sci., 104, 6343-6357 (2021).

Terlouw, E.M.C., Bourguet, C.: Quantifying animal welfare preslaughter using behavioural, physiological and carcass and meat quality measures. Preslaughter handling and slaughter of meat animals, pp.13-61(2022).

ORAL PRESENTATION

Vollastonitin doğal kauçuk karışımlarındaki yapışma ve mekanik özelliklere etkisi

Şehriban Öncel[1*] (*ORCID: https://orcid.org/0000-0001-8945-3054*), Tuba Ünügül[2] (*ORCID: https://orcid.org/0000-0002-2502-1129*), Rümeysa Kılıç[3], Ege Kaan Altınok[2], Bağdagül Karaağaç[2,3] (*ORCID: https://orcid.org/0000-0001-8747-8004*)

*[1]Kocaeli Üniversitesi, Ford Otosan İhsaniye Otomotiv MYO, Motorlu Araçlar ve Ulaştırma Teknolojileri Bölümü, Kocaeli, Türkiye

[2]Kocaeli Üniversitesi, Mühendislik Fakültesi, Kimya Mühendisliği Bölümü, Kocaeli, Türkiye

[3]Kocaeli Üniversitesi, Fen Bilimleri Enstitüsü, Polimer Bilim ve Teknolojisi, Kocaeli, Türkiye

*Sorumlu yazar e-mail: sehriban.oncel@kocaeli.edu.tr

Özet

Kauçuk esaslı malzemelerin özelliklerini iyileştirmek veya maliyeti ucuzlatmak gibi amaçlarla hamur bileşiminde çeşitli dolgu maddelerine yer verilmektedir. Karbon siyahı, kauçuk hamurlarında en yaygın olarak kullanılan dolgu maddesi olmasına rağmen üretimi sırasında yüksek enerji tüketimi ve CO_2 emisyonuna neden olması gibi dezavantajlara sahiptir. Bunun yanı sıra, petrol kökenli kaynaklardan üretilmesi dolayısıyla uzun vadede sürdürülebilir olmaması, statik elektriklenmeye yatkınlığı ve malzemelerde aglomere eğiliminde olmasından dolayı bu dolgu malzemesine alternatif bir malzeme arayışı devam etmektedir. Bu ihtiyaç doğrultusunda başlatılan bu çalışmada, alternatif dolgu maddesi olarak vollastonit kullanılmış ve karbon siyahı ile vollastonit dolgulu yedi farklı reçete oluşturulmuştur. Beyaz renkli, doğal bir mineral olan vollastonit, üstün fiziksel ve kimyasal özellikleri nedeniyle seramiklerde, plastiklerde, metalurjik uygulamalarda, boya ve kaplamalarda sıkça kullanılan bir maddedir. Bu çalışmada ham vollastonit (aktif olmayan, modifiye edilmemiş) ile epoksi vollastonit (aktif, modifiye edilmiş) doğal kauçuğa (NR) belirli miktarlarda eklenmiş ve karışımın özelliklerine etkisi araştırılmıştır. Hazırlanan kauçuk karışımlarında reolojik, fiziksel, mekanik, ısıl ve yapışma özellikleri açısından değerlendirmeler yapılmıştır. Ayrıca tüm vulkanizatlar, 70°C'de 72 saat boyunca etüvde yaşlandırılmış ve vollastonitin yaşlanma sürecindeki etkisi aydınlatılmaya çalışılmıştır. Yapılan analizler sonucunda, aktif vollastonit içeren karışımların yapışma ve mekanik özelliklerinde olumlu değişimler saptanmıştır.

Anahtar Kelimeler: Doğal Kauçuk, Dolgu Maddesi, Karbon Karası, Vollastonit, Doğal Dolgu Maddesi

The effect of wollastonite on adhesive and mechanical properties in natural rubber compounds

Abstract

In order to improve the properties of NR-based materials or to reduce the cost, various fillers are included in the compound composition. Although carbon black is the most commonly used filler in rubber compounds, it has disadvantages such as high energy consumption and causing CO_2 emissions during its production. In addition, the search for an alternative material to this filling material continues due to the fact that it is not sustainable in the long term due to its production from petroleum-based resources, its tendency to static electrification and its tendency to agglomerate in materials. In this study, which was initiated in line with this need, wollastonite was used as an alternative filler and seven different recipes with carbon black and wollastonite filling were created. Wollastonite, a natural mineral of white color, is a substance frequently used in ceramics, plastics, metallurgical applications, paints and coatings due to its superior physical and chemical properties. In this study, raw wollastonite (inactive, unmodified) and epoxy wollastonite (active, modified) were added to natural rubber (NR) in certain quantities and the effect of the mixture on its properties was

investigated. Evaluations were made in terms of rheological, physical, mechanical, thermal and adhesion properties in the prepared rubber mixtures. In addition, all vulcanizates were aged in the oven for 72 hours at 70°C and the effect of wollastonite on the aging process was tried to be elucidated. As a result of the analyzes, positive changes in the adhesion and mechanical properties of mixtures containing active wollastonite were detected.

Keywords: Natural Rubber, Filler, Carbon Black, Wollastonite, Natural Filler

GİRİŞ

Kauçuk esaslı malzemeler, Charles Goodyear'ın vulkanizasyonu keşfetmesinden bu yana hayatımızın birçok alanında yer almaktadır (De and Jim R. White 2001). Doğal kauçuğun yenilenebilir bitkisel kaynaklardan elde edilebilmesi ve görece üstün özelliklere sahip olması, sentetik kauçuklara göre daha fazla ilgi görmesini sağlamaktadır. NR, sahip olduğu iyi mekanik özellikleri, yüksek aşınma ve yırtılma direnci, dinamik çalışma koşullarına uygunluğu ve titreşim sönümleme özelliği sebebiyle tüm dünyada sayısız uygulama alanı bulmaktadır (Öncel and Karaağaç 2016). NR, yaygın olarak otomotiv, medikal gibi sektörlerin yanında; hortumlar, konveyör bantları, dayanıklı yük taşıyıcılar, titreşim sönümleyici parçalar, spor malzemeleri gibi birçok endüstriyel ürünün hammaddesi olarak kullanılmaktadır. NR esaslı malzemelerin özelliklerini iyileştirmek veya maliyeti ucuzlatmak amacıyla hamur bileşiminde çeşitli dolgu maddelerine yer verilmektedir. Kauçuk karışımların özellikleri; dolgunun dağılma derecesi, dolgu-polimer etkileşimi gibi özelliklere doğrudan bağlıdır. Kauçuk endüstrisinde en çok tercih edilen dolgu olan karbon siyahı (CB), doğalgaz veya ham petrolün kimyasal olarak bozundurulması yoluyla üretilmekte ve %90-95 oranında elementel karbondan (hidrojen ve oksijen ile birlikte) oluşmaktadır (Simpson 2002). En yaygın olarak kullanılan dolgu maddesi olmasına rağmen üretimi sırasında yüksek enerji tüketimi ve karbondioksit (CO_2) emisyonuna neden olması, petrol kökenli kaynaklardan üretilmesi sebebiyle uzun vadede sürdürülebilir olmaması, statik elektriklenmeye yatkınlığı ve karışım içinde aglomera eğiliminde olmasından dolayı karbon siyahına alternatif dolgu maddesi arayışı doğmuştur. Bu kapsamda alternatif dolgu malzemesi olarak doğal maddelerin kullanıldığı çalışmalar incelenmiş ve birçok doğal maddenin kauçuk hamur karışımı içinde kullanılabilirliğinin araştırıldığı görülmüştür; muz kabukları, mısır, şeker kamışı, pirinç çeltiği, nişasta, odun, bambu, palm yağı, hindistan cevizi kabuğu gibi selülozik (lignin, selüloz, hemiselüloz) içerikli kaynaklar ile mürekkep balığının iç kabuğu gibi doğal malzemeler (Akbay and Güngör 2018; Shebeer A. Rahim, G. Unnikrishnan, M. A. Joseph, Mubarak Ali 2021). Ayrıca doğal dolgu olarak; nano/mikro partiküllü haloysit, bentonit ve kalsiyum karbonatın da kullanıldığı çalışmalar da literatürde mevcuttur (Dintcheva et al. 2020; Rooj, Das, and Stöckelhuber 2013).

Doğal bir mineral olan vollastonit (Kalsiyum metasilikat, $CaSiO_3$), klivaj yapısı dolayısıyla öğütülmesi esnasında iğnemsi kırılım göstermektedir. Düşük su absorbsiyonu, aşınma ve sürtünme direnci, ısıl ve boyutsal kararlılık, ısıl iletkenlik, düşük elektrik iletkenliği, mukavemeti artırma gibi özellikleri sebebiyle plastik sektöründe fonksiyonel bir dolgu malzemesi olarak değerlendirilmiştir. İğnemsi tanecik morfolojisi sebebiyle bulunduğu ürünlere yüksek mukavemet sağlamakta olan vollastonit, kanserojen etkisinin olmaması sebebiyle 1980'li yıllardan itibaren asbestin yerini almıştır. Sonrasında ise beton boru dökümünde, yer karolarında, contalarda ve fren balatası gibi sürtünmeli ürünlerde alternatif bir malzeme olarak önem kazanmıştır. Ayrıca seramik, inşaat, plastik, kauçuk, boya, kaplama, sağlık gereçleri (kemik implantında), sanatsal ürünler gibi alanlarda kullanımı yaygınlaşmıştır (HANER and ÇUHADAROĞLU 2013). Vollastonit takviyeli polimer kompozitler, biyo uyumlu oluşu ve takviye edici özellikleri sebebiyle ilgi çekmektedir. Vollastonitin termoplastik kompozitlerde cam fiber gibi pahalı ürünlerin alternatifi olarak kullanılabilirliğinin araştırıldığı bir çalışmada, fonksiyonel bir dolgu potansiyeline sahip olduğu bulunmuştur. Ayrıca görece yüksek en-boy oranı ve sertliği sayesinde kompozitlerin kopma-eğilme dayanımını arttırabildiği saptanmıştır (Chan et al. 2020). 2018 yılında yapılan bir çalışmada, vollastonit içeren termoplastik elastomerde mineralin etkisi, uyumlaştırıcı ilavesi ile karşılaştırmalı olarak incelenmiştir (Abdul Karim and Ismail 2018). Uyumlaştırıcı olarak kullanılan maleik anhidrit (MAH) sayesinde dolgu ve matris arasındaki etkileşim artmış ve bunun sonucunda da kopma dayanımı artmıştır. Ayrıca MAH ilavesi, dolgunun aglomerasyonunu azalttığından kompozitin Young modülünde de pozitif bir etkiye yol açmıştır. İnorganik bir dolgu olan vollastonitin modifiye edilerek kauçuk karışımında kullanılması, dolgu etkileşimini artırmaktadır. Bu amaçla potasyum oleat (PO) ile de yüzey modifikasyonu gerçekleştirilebilmektedir. Modifikasyon sonucunda vollastonitin polarlığı azaldığı ve kauçuk-dolgu etkileşiminin arttığı tespit edilmiştir (Xiao et al. 2022). Modifikasyon yapılmadan 0, 10, 20, 30, 40 phr oranlarında karışıma eklenen vollastonitin etkilerinin araştırıldığı bir

çalışmada; vollastonit dolgulu NR karışımlarının pişme karakteristiği, mekanik dayanımı ve morfolojik özellikleri araştırılmıştır. Artan dolgu miktarı ile kopma dayanımı, kopmada uzama ve pişme sürelerinin düştüğü; fakat maksimum tork (MH) ve modül değerlerinin arttığı tespit edilmiştir (Diep et al. 2014).
Bu çalışmada, doğal bir mineral olan vollastonitin doğal kauçuk karışımdaki mekanik ve yapışma özelliklerine dayanımına etkisinin araştırılması hedeflenmiştir. Kauçuk esaslı malzemelerde kullanılan CB yerine vollastonit mineralinin alternatif olarak kullanımı ve ayrıca vollastonitin yapışma mekanizmasına etkisi araştırılmıştır. Reçeteler hazırlanırken hamurda kullanılan CB miktarı sabit tutulmuştur. Hamur içinde vollastonitin hiç olmadığı karışım (NR-REF), referans karışım olarak değerlendirilmiştir. İlk grupta (NR-W3, NR-W6, NR-W9) aktive edilmemiş vollastonit, ikinci grupta (NR-WA3, NR-WA6, NR-WA9) ise aktive edilmiş (aktif, modifiye) vollastonit alternatif dolgu malzemesi olarak kullanılmıştır. Reçete uyarınca hazırlanan örnekler, reolojik, fiziko-mekanik, ısıl analizlere tabi tutulmuştur. Ayrıca vollastonitin yapışma üzerine etkisini incelemek için H-yapışma testi uygulanmıştır.

MATERYAL VE METOT

Kauçuk hamurunun ana matrisi olarak seçilen NR (SIR10) Elkim Kauçuk firmasından satın alınmıştır. Takviye edici dolgu malzemesi olarak kullanılan karbon karası HAF N550, OMSK firmasından tedarik edilmiştir. Alternatif dolgu malzemesi olarak seçilen vollastonit hem aktive edilmiş hem de aktive edilmemiş durumda iken karışıma eklenmiştir. Vollastonit, Quarzwerke firmasından satın alınmıştır. Çinko oksit, stearik asit, TMQ (2,2,4-trimetil-1,2-dihidrokinolin), IPPD (N-izopropil-N'-fenyl-1,4-fenilendiamin), TMTD (tetrametiltiuram disulfit), CBS (N-siklohegzil-2-benzotiazol sulfenamid) ve kükürt Rubber Chem firmasından satın alınmıştır. Kauçuk hamurları 2 litrelik laboratuvar tipi dahili karıştırıcı (Banbury) ve çift silindirli mil peş peşe kullanılarak hazırlanmıştır. Karışımlara ait reçeteler, Tablo 1'de verilmiştir.

Tablo 1. Kauçuk hamur formülasyonları

	NR-REF	NR-W3	NR-W6	NR-W9	NR-WA3	NR-WA6	NR-WA9
	Bileşim (phr)						
NR	100	100	100	100	100	100	100
CB	50	50	50	50	50	50	50
Vollastonit	-	3	6	9	-	-	-
Vollastonit (Aktif)	-	-	-	-	3	6	9
Yağ	10	10	10	10	10	10	10
ZnO	5	5	5	5	5	5	5
Stearik asit	2	2	2	2	2	2	2
TMQ	1	1	1	1	1	1	1
IPPD	1	1	1	1	1	1	1
CBS	2	2	2	2	2	2	2
TMTD	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Kükürt	1.5	1.5	1.5	1.5	1.5	1.5	1.5

Hazırlanan reçetelerde, CB miktarı sabit tutulmuştur. Hamur içerinde vollastonitin hiç olmadığı referans örnekler NR-REF; aktive edilmemiş vollastonitin kullanıldığı karışımlar NR-W3, NR-W6, NR-W9 (sırasıyla 3, 6, 9 phr oranında), aktive edilmiş (modifiye) vollastonitin kullanıldığı karışımlar NR-WA3, NR-WA6, NR-WA9 (sırasıyla 3, 6, 9 phr oranında) kısaltmaları ile temsil edilmiştir. Tüm örneklere, reolojik, fiziko-mekanik, ısıl analizler uygulanmıştır. Ayrıca vollastonitin yapışma üzerine etkisini incelemek için H-yapışma testi uygulanmıştır. Endüstriyel bir mineral olan vollastonitin imalat sırasındaki birleştirici rolü hem literatürden, hem de daha önce çalışma grubumuzun yaptığı çalışmalardan bilinmektedir ve bu çalışma kapsamında da yapışmada olumlu etki yaratacağı düşünülmüştür. Bu amaçla reçetede alternatif bir dolgu maddesi olarak yüzeyi epoksi silan ile modifiye edilmiş ticari bir vollastonit kullanılmıştır.

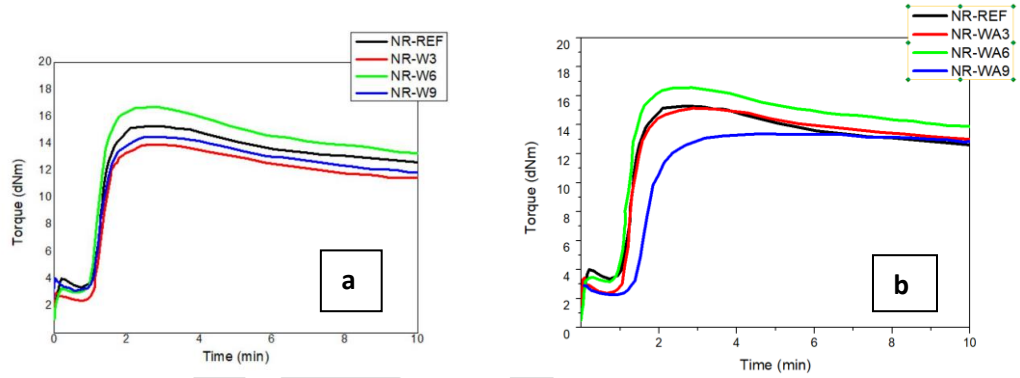
Reolojik özellikler, ASTM D5289 standardı uyarınca, döner kalıp reometresi (MDR, Alpha Technologies MDR 2000) kullanılarak 150°C'de incelenmiştir. Kauçuk hamurları, reometre eğrilerinden elde edilen optimum pişme süreleri esas alınarak, 150°C sıcaklık ve 150 bar basınç koşullarında, hidrolik sıcak pres kullanılarak vulkanize edilmiştir. Vulkanizatların mekanik özellikleri ASTM D412 standardı uyarınca universal test cihazı (Zwick/Roell 2.5 kN) kullanılarak, aşınma kayıpları ise ASTM D5963 standardına göre

aşındırıcı döner tambur kullanılarak ölçülmüştür. Sertlik değerleri, ASTM D2240 standartına göre Zwick Roell durometre ile Shore A cinsinden ölçülmüştür. ASTM D395 standardına göre, tüm vulkanizatlar iki plaka arasına yerleştirilmiş ve baskı uygulanarak kalıcı deformasyon testleri gerçekleştirilmiştir. Vulkanizatların yaşlandırma işlemi sonrası özellikleri de incelenmiştir. ASTM D573 standardına göre vulkanizatlara 70°C sıcaklıkta 72 saat boyunca, hava sirkülasyonlu etüvde ısıl yaşlandırma işlemi uygulanmıştır. Deneysel aşamada tüm karışımların termogravimetrik analizi (TGA, Mettler Toledo marka) ASTM E1131'e uygun olarak gerçekleştirilmiştir. Bu analizde malzeme, belirlenen sabit bir hızda ve seçilen atmosferik koşulda (azot), 25°C-600°C sıcaklıkları arasında ısıtılırken kütlesinde gerçekleşen değişim izlenmiştir. Son olarak, H-yapışma testi ile vollastonitin yapışma kuvvetine etkisi incelenmiştir. ASTM D4776 standardı uyarınca hazırlanan düzenekte özel bir kalıp kullanılmış ve kullanılan ağırlıklar ile ön germe uygulanmıştır. Belirlenen sürelerde karışımlar kalıp içinde pişirilmiş ve içinde elyaf bulunan karışımlar H şeklinde kesilerek çekme testine tabi tutulmuştur.

BULGULAR ve TARTIŞMA

Reolojik Özellikler

Şekil 1'de aktive edilmemiş (a) ve aktive edilmiş (b) vollastonit kullanılarak hazırlanan kauçuk hamurlarının reometre eğrileri sunulmuştur. Eğrilerden elde edilen minimum tork değeri (ML), maksimum tork değeri (MH), pişme genliği (CE) ve pişme hız indeksi (CRI) değerleri Tablo 2'de sunulmuştur.



Şekil 1. Reometre eğrileri (a) aktive edilmemiş vollastonit içeren karışımlara ait (b) aktive edilmiş vollastonit içeren karışımlara ait

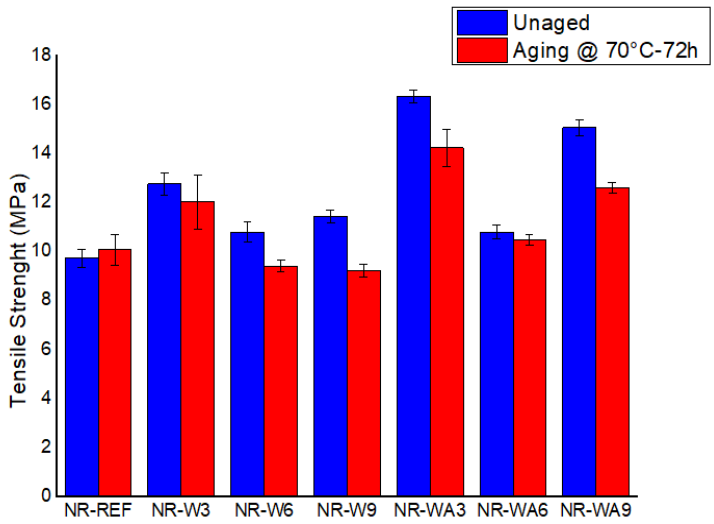
Tablo 2. Kauçuk hamurlarının önemli reolojik özellikleri

	ML (dNm)	MH (dNm)	CE (dNm)	CRI (min^{-1})
NR-REF	3.41	15.55	12.14	161
NR-W3	2.31	14.24	11.93	160
NR-W6	2.84	17.05	14.21	172
NR-W9	2.98	13.83	10.86	132
NR-WA3	2.33	15.24	12.91	152
NR-WA6	3.06	16.79	13.73	167
NR-WA9	2.30	13.84	11.54	116

Kauçuk hamurlarının önemli reolojik özelliklerini sunan Tablo 2'de sunulan pişme genliği (CE) değerleri ile çaprazbağ yoğunluğu ilişkilendirilmiştir. 6 phr vollastonit içeren karışımlar, kendi grupları içerisinde (aktif olmayan vollastonit içeren grup ve aktif vollastonit içeren grup) çapraz bağlanma düzeyinin en yüksek olduğu karışımlardır. ML değerleri incelendiğinde hamura vollastonit eklenmesinin işlenebilirliği kolaylaştırdığı söylenebilmektedir. Ayrıca vollastonit ilavesi ile viskozitenin referansa göre düştüğü de izlenmiştir.

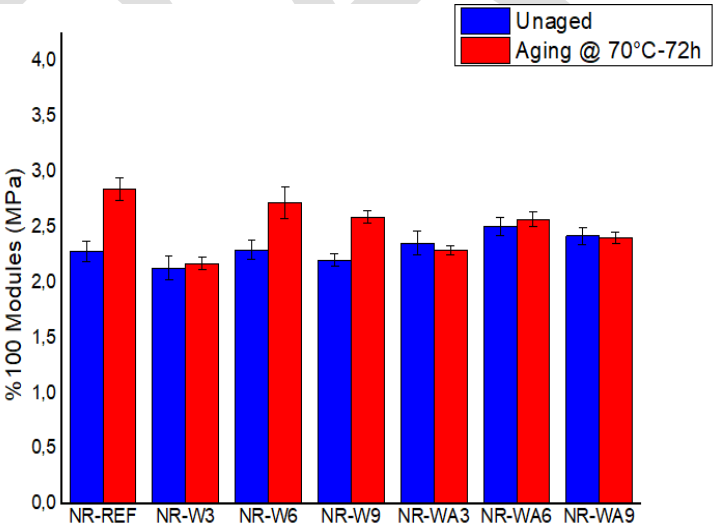
Mekanik Özellikler

Şekil 2'de vulkanizatların yaşlanma öncesi ve sonrasında kopma dayanımı değerlerindeki değişimler sunulmaktadır. Yaşlanma öncesindeki Referans ile karşılaştırıldığında vollastonit ilavesi ile tüm vulkanizatların kopma dayanımı değerlerinde bir iyileşme olmuş, özellikle aktive edilmiş vollastonit ilavesinin kopma dayanımında önemli bir artış sağladığı görülmektedir. Hem aktif hem de aktif olmayan vollastonit grubu incelendiğinde, 3 phr vollastonit ilavesinin en iyi kopma dayanımı sağlamaktadır. 70°C sıcaklıkta 72 saat boyunca yaşlandırma işlemine maruz bırakılan vulkanizatların kopma dayanımı değerlerinde bir düşüş gerçekleşmiştir.



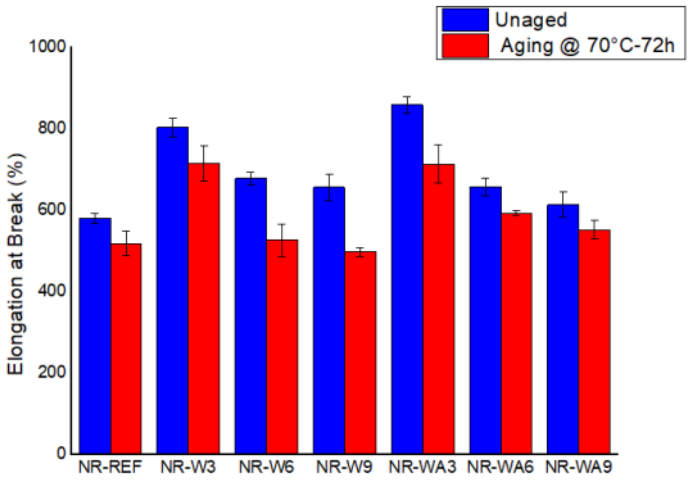
Şekil 2. Yaşlanma öncesi ve sonrasında vulkanizatların kopma dayanımı

Şekil 3'te vulkanizatların yaşlanma öncesi ve sonrasındaki %100 modül değerlerinin değişimi gösterilmektedir. Yaşlanma sonrasında, tüm vulkanizatların modül değerlerinde hafif bir artış izlenmiştir.



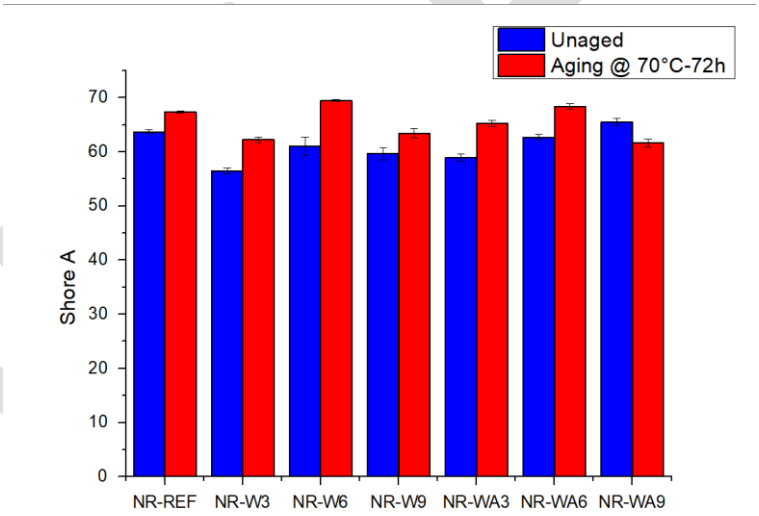
Şekil 3. Yaşlanma öncesi ve sonrasında vulkanizatların %100 modül değerleri

Şekil 4'te vulkanizatların yaşlanma öncesi ve sonrasındaki kopmada uzama değerlerinin değişimi gösterilmektedir. Karışımlara eklenen vollastonit, kopmada uzama değerlerinde bir artışa sebep olmuştur. Hem aktif hem de aktif olmayan vollastonit grubunu içeren sonuçlar incelendiğinde, 3 phr vollastonit ilavesinin en iyi kopmada uzama sağladığı anlaşılmaktadır. Yaşlandırma işlemi sonucunda tüm vulkanizatların kopmada uzama değerleri düşmüştür.



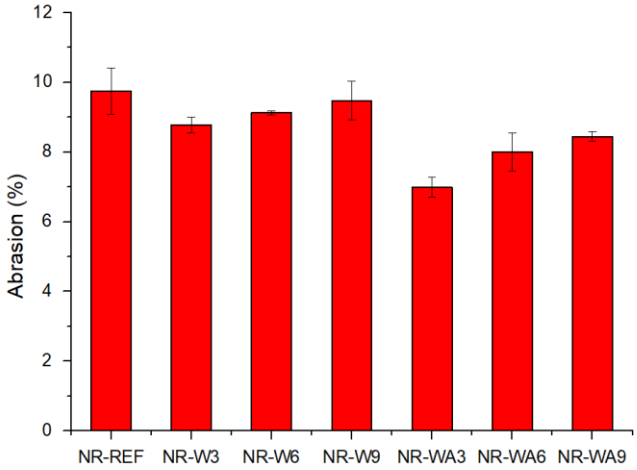
Şekil 4. Yaşlanma öncesi ve sonrasında vulkanizatların kopmada uzama değerleri

Şekil 5'te vulkanizatların yaşlanma öncesi ve sonrasındaki sertlik değerleri sunulmaktadır. Artan vollastonit miktarı ile sertlik değerlerinin arttığı görülmektedir. Vollastonitin tanecik yapısı sebebiyle oluşan bu durum doğaldır.



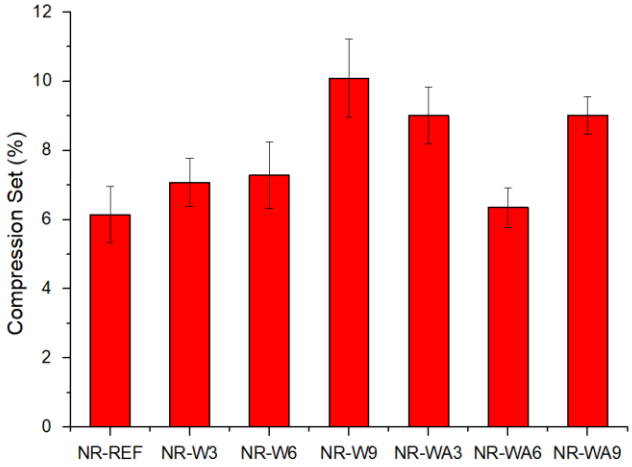
Şekil 5. Yaşlanma öncesi ve sonrasında vulkanizatların sertlik değerleri (Shore A)

Şekil 6'da vulkanizatlara 40 metre boyunca aşındırma işleminin uygulandığı teste ait sonuçlar sunulmuştur. Karışımlara uygulanan vollastonit ilavesinin aşınma kaybını azalttığı tespit edilmiş ve bu durum vollastonitin tanecik yapısı ile ilişkilendirilmiştir. Vollastonitin fren balataları gibi aşınma direncinin önemli olduğu alanlarda zaten kullanılıyor olması dolayısıyla, bu beklenen bir sonuçtur. Özellikle aktif vollastonitin matris ile yaptığı kuvvetli etkileşim, aşınma kaybı değerlerinde de izlenmiştir.



Şekil 6. Vulkanizatların aşınma kaybı (%)

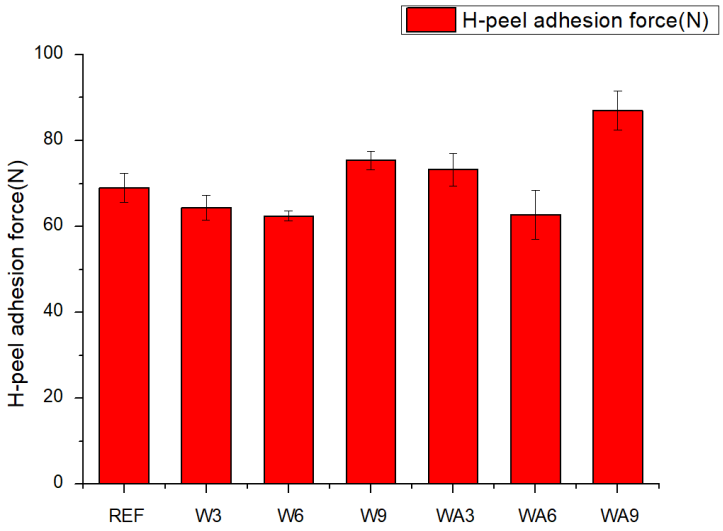
Şekil 7'de baskı altında kalıcı deformasyon testi sonuçları yer almaktadır. 6 phr aktif vollastonit ilavesinin en az kalıcı deformasyona yol açtığı, en fazla deformasyonun ise 9 phr oranında vollastonit içeren karışımlarda gerçekleştiği tespit edilmiştir. Bir başka deyişle, 6 phr vollastonit içeren hamurların çaprazbağ yoğunluğu daha yüksek olduğu görülmektedir.



Şekil 7. Baskı altında kalıcı deformasyon

H-yapışma Testi

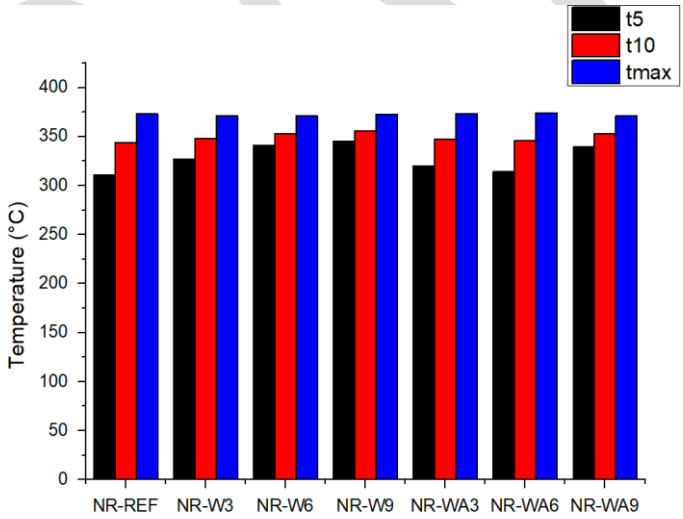
Takviye edici elyaf ile birlikte özel bir kalıpta pişirme ve ardından çekme testi uygulanması sonucunda elde edilen maksimum kuvvet, H-yapışma kuvveti Newton cinsinden kaydedilmiştir. Bu değerlerde meydana gelen değişimler, diğer bir deyişle vollastonitin aktif olduğu ve olmadığı durumlar arasındaki farklılık Şekil 8'de gösterilmektedir. Aktif yani yüzeyi epoksi silan ile modifiye edilmiş vollastonitin dolgu olarak kullanıldığı durumda yapışma kuvvetinin en yüksek olduğu ve bu durumun beklenildiği gibi artan ara yüzey etkileşiminden kaynaklandığı düşünülmektedir.



Şekil 8. H-yapışma kuvveti (N)

Termogravimetrik Analiz

Termogravimetrik analiz, 25°C-600°C sıcaklıkları arasında ve azot ortamında gerçekleştirilmiştir. Malzemelerin ısıl kararlılığının tespit edildiği bu teknikte; %5 ve %10 kütle kaybının gerçekleştiği sıcaklıklar T_5, T_{10} olarak Şekil 9'da sunulmuştur. Ayrıca bozunmanın en fazla olduğu sıcaklık da T_{max} ile ifade edilerek grafikte yer almıştır. T_5 değerleri incelendiğinde, vollastonit ilavesinin bozunmayı geciktirme etkisi olduğu söylenebilir. Aktif olmayan vollastonit içeren grubu kendi içinde değerlendirdiğimizde, dolgu miktarı artışı ile t_5 ve t_{10} sıcaklıklarındaki artışın doğru orantılı olduğu; Aktif vollastonit içeren grubu kendi içinde değerlendirdiğimizde ise en yüksek t_5 ve t_{10} sıcaklığının 9 phr ilave yapıldığı durumda olduğu tespit edilmiştir.



SONUÇ

Yapılan bu çalışmada, doğal kauçuk karışımlarında vollastonitin alternatif bir dolgu olarak kullanılabilirliği araştırılmıştır. Bu amaçla hazırlanan 7 karışımda hem aktif hem de aktif olmayan vollastonit kullanılmıştır. Referans amacıyla kullanılan karışımda ise hiç vollastonit kullanılmamıştır. Hamurdaki vollastonit miktarı ve dolgunun aktif olup olmaması test sonuçlarını değiştirmektedir. Özellikle aktif vollastonitin 9 phr kullanımı durumunda, kopma dayanımı ve yapışmanın olumlu anlamda değişmesi çalışmanın önemli bir sonucudur. Ayrıca vollastonit kullanımının aşınma direncine katkısı dolayısıyla, sürtünmenin önem kazandığı uygulamalarda kullanımı malzemelere katkı sağlamaktadır. Tüm sonuçlar değerlendirildiğinde, doğal dolgu maddesi olan vollastonitin karbon karasına sürdürülebilir bir alternatif dolgu maddesi olabileceği ve vollastonitin doğal kauçuk karışımlarındaki yapışma ve mekanik özellikleri olumlu yönde değiştirdiği tespit edilmiştir.

KAYNAKLAR

Abdul Karim, Ahmad Fikri, and Hanafi Ismail. 2018. "The Effects of a Compatibiliser on Processing, Tensile Properties and Morphology of Polystyrene (PS)/Styrene-Butadiene Rubber (SBR)/Wollastonite Composites." *Polymers and Polymer Composites* 26(8–9):454–60.

Akbay, İsmail Kutlugün, and Ahmet Güngör. 2018. "Investigation of Using Waste Banana Peels in EPDM as Bio-Based Filler." *International Advanced Researches and Engineering Journal* 2(2):132–36.

Chan, Jia X., Joon F. Wong, Azman Hassan, Zurina Mohamad, and Norhayani Othman. 2020. "Mechanical Properties of Wollastonite Reinforced Thermoplastic Composites: A Review." *Polymer Composites* 41(2):395–429.

De, Sadhan K., and Jim R. White. 2001. *Rubber Technologist 's Handbook*. Vol. 2. United Kingdom: Rapra Technology Ltd.

Diep, H. T. B., H. Ismail, A. R. Azura, Ng Van Tu, and T. Takeichi. 2014. "The Effect of Wollastonite on Curing Characteristics, Tensile and Morphology of Natural Rubber Compounds." *Advanced Materials Research* 858:199–204.

Dintcheva, Nadka Tzankova, Giulia Infurna, Marilena Baiamonte, and Francesca D'Anna. 2020. "Natural Compounds as Sustainable Additives for Biopolymers." *Polymers* 12(4).

HANER, Serhan, and Dilek ÇUHADAROĞLU. 2013. "Vollastonit: Bir Gözden Geçirme." *Jeoloji Mühendisliği Dergisi* 37(1):63–82.

Öncel, Şehriban, and Bağdagül Karaağaç. 2016. "Kauçuk Esaslı Malzemelerde Yaşlanma-Alternatif/Doğal Antioksidanlar." *Kauçuk Derneği* 50–52.

Rooj, Sandip, Amit Das, and Klaus Werner Stöckelhuber. 2013. "Understanding the Reinforcing Behavior of Expanded Clay Particles in Natural Rubber Compounds." *Soft Matter* 9:3798–3808.

Shebeer A. Rahim, G. Unnikrishnan, M. A. Joseph, Mubarak Ali, Asif Afzal. 2021. "Chitosan-Reinforced Nitrile Rubber – a Step towards Sustainable Development." *Plastics, Rubber and Composites*, 1–9.

Simpson, Richard B. 2002. *Rubber Basics*. United Kingdom: Rapra Technology Limited.

Xiao, Yao, Yingjie Hao, Lizhi Yan, Zhenchun Xu, Zhihua Sui, Yi Pan, Chuansheng Wang, Huiguang Bian, and Xiaoming Wang. 2022. "Mechanism on Surface Hydrophobically Modification of Fibrous Wollastonite and Its Reinforcement of Natural Rubber." *Journal of Polymer Research* 29(8):1–14.

DRAFT

ORAL PRESENTATION

ZnO-poli(indol-4aminokinaldin) elektrodun H_2O_2 tayininde sensör olarak performansının incelenmesi

Rukan SUNA KARATEKİN* (*ORCID: https://orcid.org/0000-0003-3052-1539*),

*Mersin Üniversitesi, Fen Fakültesi, Kimya Bölümü, Mersin, Türkiye

*rukansuna@mersin.edu.tr

Özet

Bu çalışmada hidrojen peroksitin elektrokimyasal tayini için ZnO ile modifiye edilmiş poli(indol-4aminokinaldin) iki aşamada sentezlenerek sensör performansı incelenmiştir. Bu çalışma ile ilk kez yeni sentelezlenip literatüre sunulmuş bir kopolimerin sensör alanındaki performansı incelenmiştir. Hazırlanan kopolimerin faklı pH'larda dahi aktif olması sensör uygulamaları için alışılagelmiş diğer iletken polimerlerden daha üstün bir özelliğe sahip olduğunu göstermektedir. Hazırlanan katalizörlerin karakterizasyonu alan emisyonlu taramalı elektron mikroskobu (FESEM), X-ışını kırınım spektroskopisi (XRD) ile gerçekleştirilmiştir. Diğer taraftan katalizörlerin elektrokimyasal özellikleri ve sensör performansı dönüşümlü voltametri (CV) ve amperometrik ölçüm ile incelenmiştir. Kopolimer ve kopolimersiz ZnO-ITO elektrotların H_2O_2 indirgenmesindeki performansları incelendiğinde kopolimerin varlığı ile birlikte H_2O_2'ye ait indirgenmenin daha yüksek akım ile gerçekleştiği gözlenmiştir. Amperometrik ölçüm ile birlikte hazırlanan elektrodun H_2O_2 algılamasında sensör olarak kullanılabileceği sonucuna varılmıştır.

Anahtar Kelimeler: poli(indol-4-aminokinaldin), sensör, ZnO

Investigation of the performance of ZnO@poly(indole-4aminoquianaldine) electrode as a sensor for the determination of H_2O_2

Abstract

In this study, for electrochemical determination of hydrogen peroxide and investigation of its sensor performance, ZnO doped poly(indole-4aminoquianldine) was synthesized in a two-step. In this study, the performance of a newly synthesized and presented copolymer in the field of sensors was examined for the first time. The fact that the prepared copolymer is active even at different pHs shows that it has superior properties than other conventional conductive polymers for sensor applications. To characterize synthesized catalysts field emission microscopy (FESEM), X-ray Powder Diffraction (XRD), measurements were performed. On the other hand, the electrochemical properties of catalysts were investigated by cyclic voltammetry (CV) and amperometric measurement. When the performances of copolymer and copolymer-free ZnO-ITO electrodes in H_2O_2 reduction were examined, it was observed that the reduction of H_2O_2 performed with a higher current with the presence of copolymer. According to the amperometric measurement, it was concluded that fabricated electrode can be used as a sensor to determine H_2O_2

Keywords: poly(indole-4aminoquinaldine), sensor, ZnO

GİRİŞ

Hidrojen peroksit çevre, endüstri ve biyomedikal alanlarında kullanılan önemli bir kimyasal maddedir (Stanković vd., 2020). Bununla birlikte birçok klinik, farmokoloji ve yiyecek endüstrisinde oksitleyici ajan olarak da kullanılmaktadır. Ancak hidrojen peroksitin fazlası merkezi sinir sistemine etki ederek Parkinson ve Alzeimer gibi hastalıklara neden olmaktadır (Yuvashree ve Balavijayalakshmi, 2019). Aynı zamanda kanser hücrelerinin normal hücrelere kıyasla daha fazla miktarda H_2O_2 ihtiva ettiği çeşitli çalışmalar ile kanıtlanmıştır. Bu durum kanser ile H_2O_2 miktarı arasında doğrudan bir ilişki olduğunu düşündürmektedir (Qian vd., 2021). Bu gibi ciddi ve yaygın hastalıklar ile H_2O_2 miktarı arasında doğrusal bir ilişki olması nedeniyle H2O2'nin doğru ve hızlı tespiti büyük önem taşımaktadır. H_2O_2'nin tayininde krotomotografik, spektroskopik ve titrimetri gibi yöntemler kullanılsa da bunlar karmaşık ve maliyeti yüksektir (Wang ve Wang, 2004). H_2O_2 miktar tayininde yaygın olarak kullanılan diğer bir yöntem ise elektrokimyasal yöntemdir. Bu yöntemin basit

ve maliyeti düşük olması sebebiyle günümüzde sıklıkla tercih edilmektedir. Bu yöntemde hidrojen peroksitin yükseltgenmesi veya indirgenmesine cevap veren ve sensör olarak seçiciliği ve hassasiyeti yüksek bir elektrot tasarımı büyük önem taşımaktadır. Sensör uygulamalarında kullanılmak üzere hazırlanan elektrotların seçiciliği ve hassasiyeti kullanılan elektrotun iletkenliğine ve hedef moleküle olan ilgisine bağlı olarak değişmektedir (Palanisamy vd., 2012). Stanković vd., arkadaşları (2020) hazırladıkları Ag nanopartikül kaplı polianilin grafen kompozit elektrotun askorbik asit ve glukoza karşı hassasiyetinin olmadığını ancak hidrojen peroksite karşı hassasiyeti olduğu belirtilmiştir. Hazırlanan elektrotun hedef moleküle hızlı cevap vermesi sensörün kullanılabilirliğini arttırmaktadır. Bunun için kullanılacak elektrotun hızlı ve tersinir adsorpsiyon/desorpsiyon kinetiğine sahip olması son derece önemlidir. Bahsedilen özelliğin iletken polimerlerde olmasından dolayı, sensör uygulamalarında kullanılan elektrotlara iletken polimerlerin modifiye edilmesi söz konusudur. Bunun dışında iletken polimerler iletkenlikleri, kararlılıkları ve modifiye edildikleri elektrotların aktif yüzey alanlarını arttırması sebebiyle de yaygın olarak kullanılmaktadır (Sudhakara vd., 2021).

Sensör uygulamalarında sensörün analite cevabının ortam şartlarından etkilenmemesi ve dolayısıyla ortamın pH değerinin değişmemesi için tampon çözeltiler (pH:7-7,5) kullanılmaktadır (Nor vd., 2019). Ancak tampon çözeltilerinin sahip olduğu pH aralığında kullanılan polimerin aktivitesini yitirmemesi, kararlılığını devam ettiriyor olabilmesi son derece önemlidir. Bilindiği üzere polianilin, polipirol gibi iletken polimerler düşük pH değerlerinde sentezlendiklerinden iletkenliklerini yine düşük pH ortamlarında koruyabilmektedir (Kim vd., 2020). Özellikle polianilinin yüksek pH'larda elektoaktivitesi azalmaktadır (Gicevicius, M. vd. 2018). Yapılan çalışmalarda bu problemi çözmek için polianilin yapısına sülfo veya karboksil grupları gibi asidik grupların eklendiği belirtilmiştir. Bu durum elektrot hazırlanmasında fazladan işlem uygulanmasını gerektirdiği için hem maliyet hem de zaman kaybı yaratmaktadır. Bu nedenle iletkenliğinin yüksek olması dışında iletkenlik değerini yüksek pH ortamlarında koruyan bir polimerin H_2O_2 tayininde sensör olarak hazırlanacak elektrotta kullanımı son derece önemlidir (Pundir vd., 2011). Özellikle metal ve metal oksitlerin iletken polimer yüzeyine depolanması ile partiküllerin kümeleşme sorunun ortadan kalktığını yüzeyde daha homojen bir yapı olduğu belirtilmektedir (Kim vd., 2020). Aynı zamanda polimerler inorganik metal oksit ile kaplandığında elektriksel ve mekaniksel özelliklerinin iyileştiği gözlenmiştir (Zhang vd., 2009). Literatürde bu metal oksitler arasında bulunan ZnO'nun sensör uygulamalarında yüksek katalitik etki göstermesi, biyouyumlu olması, kolay hazırlanması, toksik olmaması, yüksek termal ve kimyasal kararlılığının yüksek olması en çok tercih edilen elektrot malzemelerden biri olmasına neden olmuştur (Al-Hadran vd., 2016). ZnO hidrotermal, sol-jel, kimyasal depolama, termal bozunma ve elektrokimyasal yöntemler ile bir destek materyali üzerine sentezlenebilmektedir. Bu yöntemler arasında elektrokimyasal yöntem sentez kolaylığı, düşük sıcaklıklarda ve daha az toksik malzemeler ile çalışılabilme gibi avantajları ile ilgi görmektedir (Valatka ve Šulčiūtė, 2012).

Bu çalışmada tarafımızca sentezlenip literatüre kazandırılmış bir kopolimer olan poli(indol-4-aminokinaldin) kullanılmıştır. Yeni olan bu kopolimerlerin farklı metal/metal okistlerle modifiyesi ve bir alanda kullanılabilmesi son derece önemlidir. Bu çalışma ile birlikte ZnO ile modifiye edilmiş kopolimerin H_2O_2' nin dedeksiyonundaki performansı incelenmiştir.

MATERYAL VE METOT

ITO elektorlarının hazırlanması:

ITO camlar 2x1 cm^2 olacak şekilde kesilmiş ve sırası ile sonik banyado alkol, aseton ve su içerisinde 5 dakika bekletilerek yüzeyinin temizlenmesi sağlanmıştır.

Poli(indol-4aminokinaldin)' in elektrokimyasal sentezi:

Önceki çalışmamızda da belirttiğimiz gibi, 16 mM indol ve 16 mM 4-aminokinaldin (4-aq) 1:4 oranında karıştırılan asetonitril ve 1 M $HClO_4$ çözelti ortamına eklenmiş homojen bir karışım haline geldiğinde potansiyodinamik olarak indiyum kalay oksit (ITO) üzerinde sentezlenmiştir (Karatekin, 2022). Polimerizasyonda çalışma elektrotu olarak ITO, karşı elektrot olarak 2 cm^2 yüzey alanına sahip Pt levha, referans elektrot olarak ise Ag/AgCl (3,5 M KCl) kullanılmıştır.

ZnO' in elektrokimyasal olarak kaplanması

Bu çalışmada ZnO' nun kopolimer yüzeyine depolanması için öncelikle 0.1 M $Zn(NO_3)_2.4H_2O$ çözeltisi 20 dakika boyunca Ar gazından geçirilmiş ve sıcaklık 70 ^oC' de sabit tutulmuştur. Çözeltinin pH' sı NH_3 ile 9-10

arasına sabitlenmiştir. 0.8 mA sabit akımda yapılan deneylerde 25 saniyeden sonra yüzeyde ZnO tabakasının elde edildiği gözlenmiştir.

DRAFT

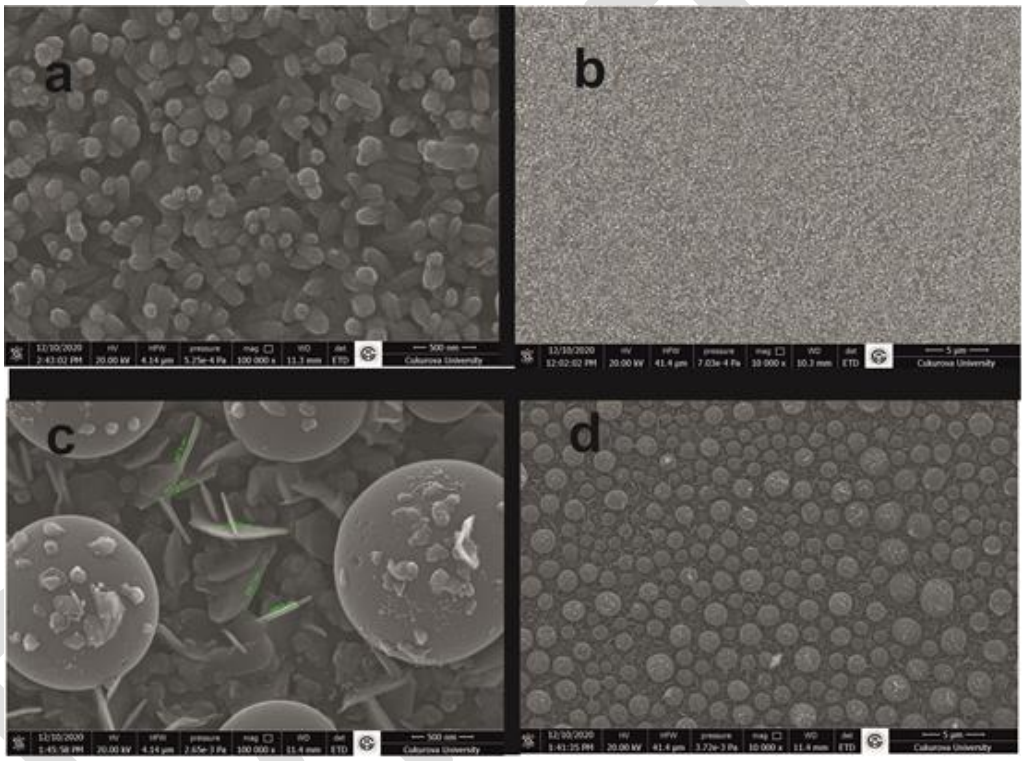
Kullanılan Cihazlar

Hazırlanan elektrotların morfolojik yapılarını incelemek üzere Zeıss marka SEM cihazı kullanıldı. XRD spektrumu Empyrean, Panalytical cihazı kullanılarak kaydedildi. Tüm elektokimyasal ölçümler için CHI-660C marka elektokimyasal unite tercih edildi.

BULGULAR ve TARTIŞMA

SEM sonuçları

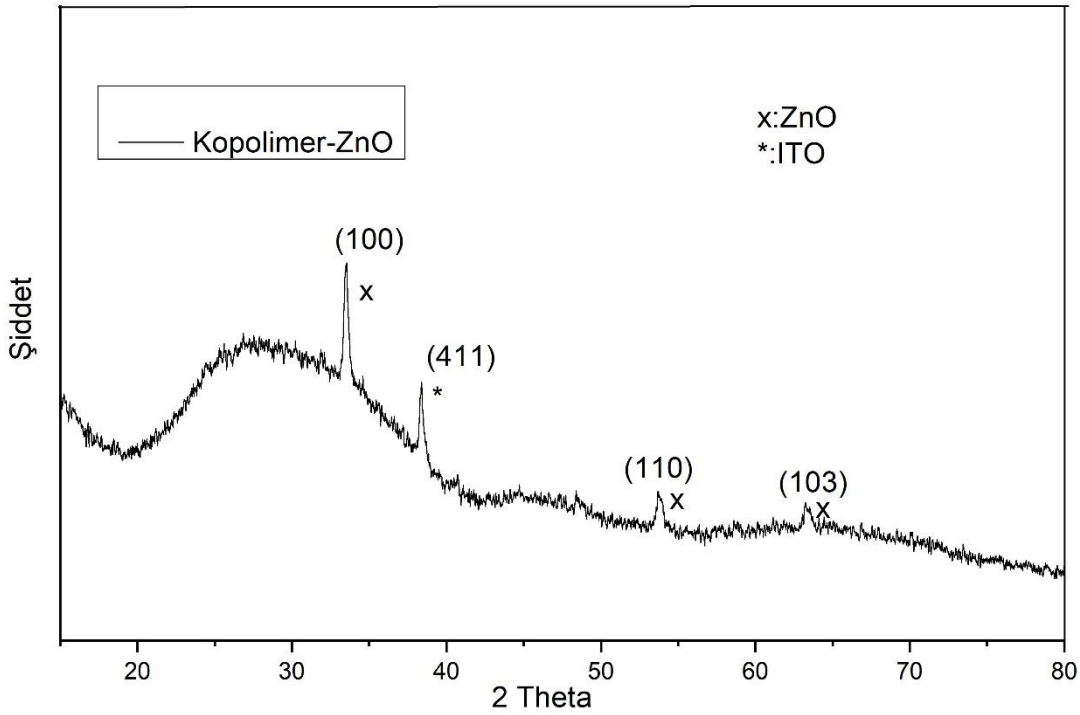
Şekil. 1 a ve b 'da verilen görüntüler ZnO' in ITO yüzeyindeki SEM görüntüsüne ait iken c ve d' de ki görüntüler kopolimer kaplı ITO yüzeyindeki ZnO' nun SEM görüntüsüne aittir. Şekilde görüldüğü üzere ZnO, ITO yüzeyine kaplandığında çubuk şeklini aldığını ancak kopolimer yüzeyine kaplandığında plate şeklini aldığı gözlenmiştir. Şekil 1c ve d' de oval şeklinde olan partiküllerin kopolimere ait olduğu ZnO partiküllerinin de bu oval şekillerin arasına depolandığı görülmüştür. Şekil 1c ve d'de elde edilen ZnO plate şeklinin yaklaşık 600 nm boyutunda olduğu SEM analizi sonucu ile belirlenmiştir.



Şekil 1. SEM görüntüleri a) - b) ZnO-ITO c) ve d) –e)Kopolimer-ZnO-ITO

XRD sonuçları

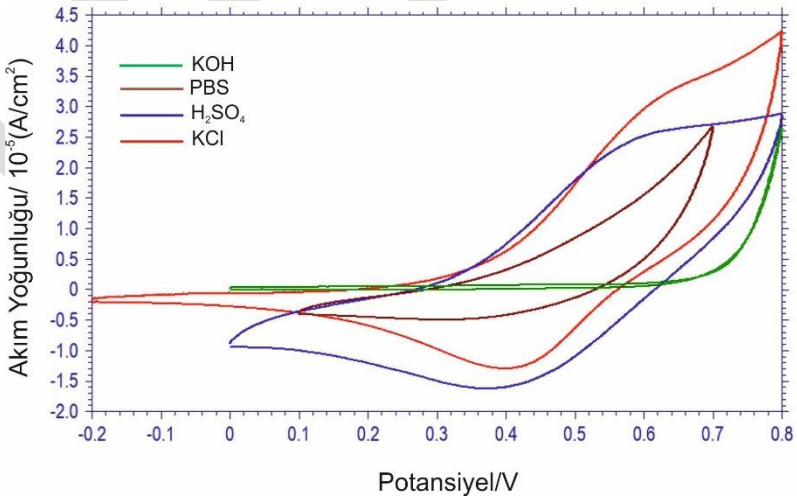
Şekil 2' de kopolimer-ZnO-ITO elektroduna ait XRD spektrumu verilmiştir. Yaklaşık $2\theta=30^{o}$' da görülen yayvan pikin kopolimere ait olduğu, diğer keskin piklerinde ITO ve ZnO' e ait olduğu belirlenmiştir (Karatekin, 2022). Buna göre hazırlanan elektrodun kopolimer, ZnO ve ITO dan oluştuğu bunların dışında herhangi bir pikin gözlenmemesi safsızlık içermediğinin göstergesidir.



Şekil 2. Kopolimer-ZnO-ITO elektroda ait XRD spektrumu

Elektrokimyasal Ölçümler

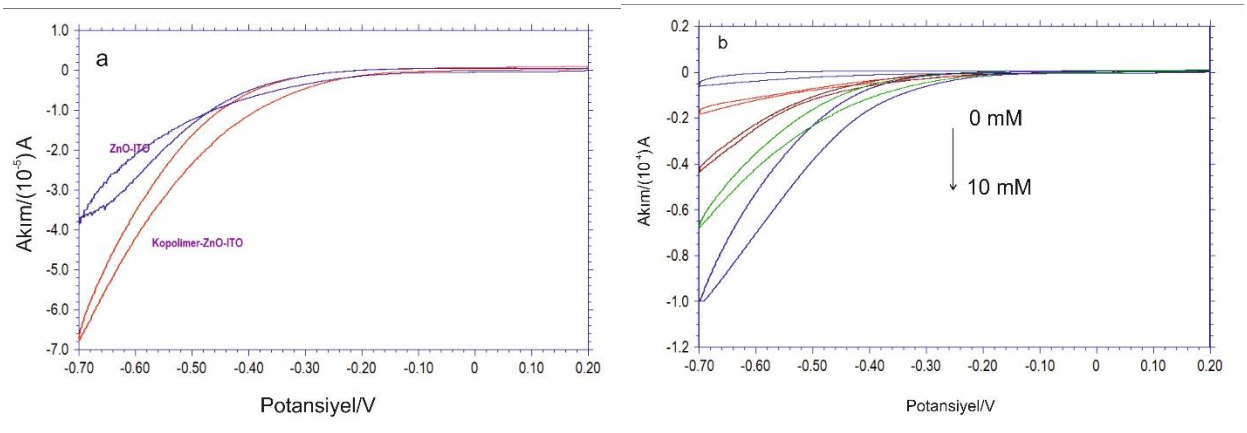
Poli(indol-4aq) kopolimerinin farklı pH ortamlarında alınan dönüşümlü voltamogramları Şekil 3' de görülmektedir. Yukarıda belirtildiği gibi kopolimerin iletkenliği sadece asidik ortamda değil nötr ortamda da korunmaktadır. Bu sonuçla birlikte aktivitesini çalışılan pH aralığında koruyan yeni bir kopolimer sentezi ve bir uygulamada kullanımı son derece önemlidir.



Şekil 3. Poli(indol-4aq)' ün farklı ortamlarda (KOH, PBS(fosfat tampounu), H2SO4 ve KCl) kaydedilmiş dönüşümlü voltamogramları (tarama hızı:10 mV/s

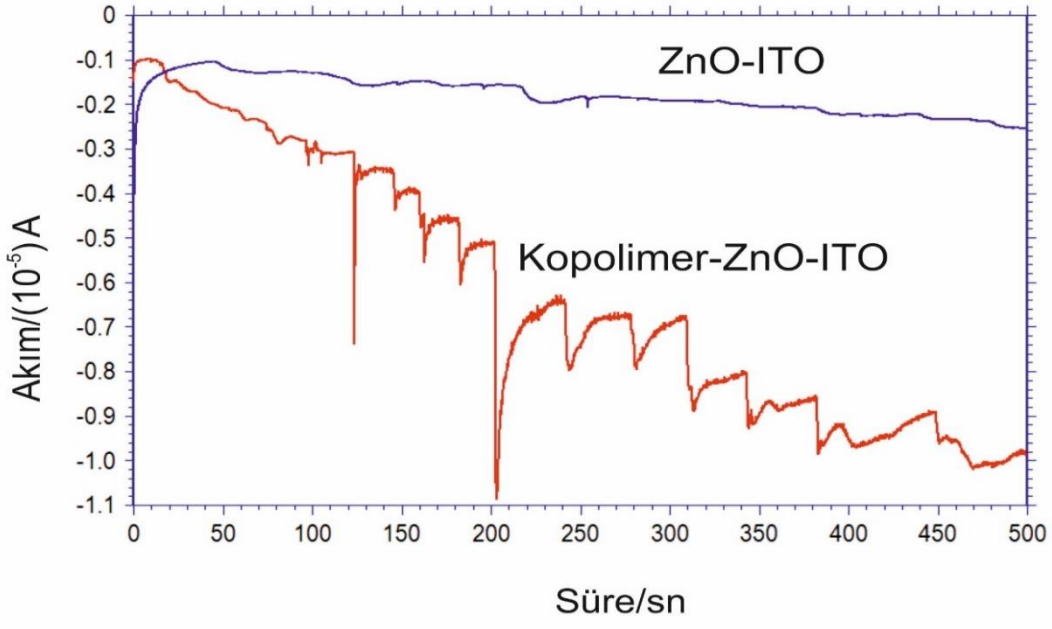
100 sn boyunca ITO ve kopolimer kaplı ITO elektroda ZnO kaplanması sonucu elde edilen ZnO-ITO ve kopolimer-ZnO-ITO elektrotların 5 mM hidrojen peroksit içeren çözelti ortamında (PBS, pH=7-7.5)

voltamogramları alınmıştır. Elde edilen voltamogram incelendiğinde katodik bölgede bir akım artışının olduğu, kopolimer kaplı ZnO modifiyeli elektrotun aynı derişimdeki hidrojen peroksite karşı ZnO-ITO elektrottan daha hassas olduğu gözlenmiştir. Bunun üzerine kopolimer-ZnO-ITO elektrotun farklı derişimlerde (0, 2.5, 5, 7.5, 10 mM) H_2O_2 içeren çözeltilerde de ölçümleri alınmış, hidrojen peroksit derişiminin artmasına bağlı olarak akımda artış olduğu gözlenmiştir (Şekil 4).



Şekil 4. a) ZnO-ITO ve kopolimer-ZnO-ITO elektrotlarının 5 mm H2O2 ihtiva eden 0.1 M PBS ortamında kaydedilen voltamograları b) Kopolimer-ZnO-ITO elektrodunun faklı derişimdeki H_2O_2 ihtiva eden 0.1 M PBS ortamında kaydedilen voltamograları (tarama hızı: 10 mV/s)

Şekil 5' de) ZnO-ITO ve kopolimer-ZnO-ITO elektrotlarının -0.5 V' da kaydedilen akım-zaman grafiği gösterilmişitir. Kopolimer varlığında elektrodun ortama eklenen H_2O_2' ye karşı hassasiyetinin arttığı gözlenmiştir. Bu durum kopolimer-ZnO-ITO elektrodunun H2O2 tayininde bir sensör olarak kullanılabileceğini göstermektedir. Diğer taraftan elektrodun cevap verme süreside sensör performansında önemli bir parametredir. Kopolimer-ZnO-ITO elektroduna ait amperometrik ölçüm incelendiğinde 5 saniyeden daha kısa bir sürede cevap verdiği anlaşılmıştır.



.

Şekil 5. ZnO-ITO ve kopolimer-ZnO-ITO elektrotlarının farklı sürelerde ard arda H_2O_2' nin ortama eklenmesi sonucu kaydedilen akım-zaman grafiği

SONUÇ

Bu çalışmada farklı pH'larda aktivitesini koruyan yüksek iletkenliğe sahip kopolimerin ZnO ile modifiyesi sonucu H_2O_2 indirgenmesindeki performansı incelenmiştir. Yapılan karakterizasyon ölçümleri ile birlikte ZnO' nun kopolimer yüzeyine tabaka şeklinde kaplandığı, kopolimer olmadığı durumda çubuk şeklinde depolandığı gözlenmiştir. Yapılan araştırmalar tabaka şeklindeki ZnO' in sensör performansının kristal yapısından dolayı daha iyi olduğu belirtilmiştir. Öyleki H_2O_2 içeren ortamda kopolimer-ZnO-ITO elektrodundan elde edilen akım ZnO-ITO elektrodundan elde edilen akımdan daha fazladır. Bu da kopolimerin etkisi ile birlikte ZnO' in H_2O_2' nin indirgenmesine daha fazla katalizlediği sonucuna varılmıştır.Bu sonuç H_2O_2 derişimin artması ile birlikte elde edilen katodik akım değerlerinin artması ile de anlaşılmıştır. Diğer taraftan yapılan amperometrik ölçüm ile elektrodun sensör olarak kullanılabileceği sonucuna varılmıştır.

KAYNAKLAR

Ahmed, N., A. vd. 2019. ¨Voltammetric determination of ascorbic acid with zinc oxide modified glassy carbon electrode˝, Journal of the Iranian Chemical Society, 16, 1957–1963.

Al-Hardan, N. H. vd. 2016. ¨Amperometric non-enzymatic hydrogen peroxide sensor based on aligned zinc oxide nanorods˝, Sensors, 16, 1004.

Gicevicius, M. vd. 2018. Analytical evaluation of optical ph-sensitivity of polyaniline layer electrochemically deposited on ıto electrode, Journal of The Electrochemical Society, 165 (14), 903-907.

Karatekin, R, 2022 Enhanced methanol oxidation on N-doped reduced graphene oxide/ZnO/nano-Pt catalyst, Diamond and Related Materials, 127; 109145.

Karatekin, R. 2022 Synthesis and characterization of highly conductive poly(indole-4-aminoquinaldine) copolymer. J Mater Sci: Mater Electron 33, 17923–17938 (2022).

Kim, W. vd. 2020. Comparative study on the effect of protonation control for resistive gas sensor based on close-packed polypyrrole nanoparticles, Applied Sciences, 10(5), 1850

Nor, N. M. vd. 2019. Effect of Concentration and pH of PBS to the electrocatalytic performance of enzymatic glucose biosensor, Solid State Phenomena, 290, 193-198

Qian, Z. vd. 2021. A nonenzymatic hydrogen peroxide electrochemical sensing and application in cancer diagnosis Small Methods.

Palanisamy, S. vd. 2012. z novel nonenzymatic hydrogen peroxide sensor based on reduced graphene oxide/ZnO composite modified electrode , Sensors and Actuators B, 166– 167, 372– 377

Pundir, C. S. vd. 2011. ¨A non-enzymatic sensor for hydrogen peroxide based on polyaniline, multiwalled carbon nanotubes and gold nanoparticles modified Au electrode , The Royal Society of Chemistry, 136, 4460–4466.

Stanković, V. vd. 2020. A novel nonenzymatic hydrogen peroxide amperometric sensor based on AgNp@GNR nanocomposites modified screen-printed carbon electrode , 876, 114487.

Sudhakara, S. M. vd. 2021. Phthalocyanine pendented polyaniline via amide linkage for an electrochemical sensing of H2O2 , Microchemical Journal, 16, 105781

Valatka, E. ve Šulčiūtė, A. 2012. Electrodeposition and Photoelectrocatalytic Activity of ZnO Films on AISI 304 Type Steel , Materials Science (Medžıagotyra), 18, 318-324

.Wang, L. ve Wang, E. 2004. A novel hydrogen peroxide sensor based on horseradish peroxidase immobilized on colloidal Au modified ITO electrode , Electrochemistry Communications, 6, 225–229.

Yuvashree, S. ve Balavijayalakshmi, J. 2019.¨Graphene based nanocomposites for electrochemical detection of H2O2 , Materials Today: Proceedings, 18, 1740–1745.

Zhang, Y. vd. 2009. An enzyme immobilization platform for biosensor designs of direct electrochemistry using flower-like ZnO crystals and nano-sized gold particles, Journal of Electroanalytical Chemistry, 627, 9–14

DRAFT

ORAL PRESENTATION

İmidaklopride maruz kalan *Melanopsis praemorsa (*Gastropoda:Prosobranchia)'nın ayak dokusundaki histopatolojik değişiklikler

Lokman DAMAR[1*] *(https://orcid.org/0009000945958371)*, Birgül OTLUDİL[2] (*https://orcid.org/0000-0002-6752-498X)*

[*1]DÜ Fen Bilimleri Enstitüsü, Biyoloji Anabilim dalı. Diyarbakır, TÜRKİYE
[2]DÜ Fen Fakültesi, Biyoloji Bölümü. Diyarbakır, TÜRKİYE

e-mail: birgulotl@dicle.edu.tr

Özet

Bu çalışmada tatlı su salyangozu *Melanopsis praemorsa*, İmidaklopridin sublethal konsantrasyonlarına (4.016µg /L^{-1}, 40.32 µg/L^{-1} ve 80.32 µg/L^{-1}) maruz bırakıldı. Deneyde 7, 14 ve 21 günlük periyotlar sonunda *M. praemorsa* ayak dokuları alınarak histolojik preparatları hazırlandı. İmidaklopridin farklı konsantrasyonlarında *M. praemorsa* ayaklarında meydana gelen histopatolojik değişiklikler ışık mikroskobunda incelendi. İmidakloprid intoksikasyonuna bağlı olarak ayak dokularında; mukus, pigment ve protein hücreleri ile lipid vakuollerinin sayısında artış, kas fibrillerinde atrofi ve nekroz, epitel tabakasında bozulma ve deskuamasyon gibi histopatolojik lezyonlar gözlendi. Ayaklarda lezyonların şiddeti, artan imidakloprid konsantrasyonuna ve imidaklopride maruz kalma süresine bağlı olarak paralel artış gösterdi. Çalışmamız için seçtiğimiz salyangoz bir kirleticinin etkisinin belirlenmesinde uygun biyobelirteçler olarak önerilir.

Anahtar Kelimeler: *Melanopsis praemorsa*, Histopatoloji, Neonikotinoid, İmidakloprid.

Histopathological changes in the foot tissue of *Melanopsis praemorsa* (Gastropoda: Prosobranchia) exposed to imidacloprid

Abstract

In this study, the freshwater snail *Melanopsis praemorsa* was exposed to sublethal concentrations of Imidacloprid (4.016 µg /L-1, 40.32 µg/L-1 and 80.32 µg/L-1). In the experiment, *M. praemorsa* foot tissues were taken at the end of 7, 14 and 21 day periods and histological preparations were prepared. Histopathological changes occurring in *M. praemorsa* feet at different concentrations of imidacloprid were examined under a light microscope. In foot tissues due to imidacloprid intoxication; Histopathological lesions such as an increase in the number of mucus, pigment and protein cells and lipid vacuoles, atrophy and necrosis in muscle fibers, deterioration and desquamation in the epithelial layer were observed. The severity of lesions on the feet increased in parallel with increasing imidacloprid concentration and duration of exposure to imidacloprid. The snails we chose for our study are recommended as suitable biomarkers to determine the impact of a pollutant.

Key Words: *Melanopsis praemorsa*, Histopathology, Neonicotinoid, Imidacloprid.

GİRİŞ

Günümüzde kentleşme, sanayileşme ve teknolojideki gelişmelerle birlikte hızla artan insan nüfusu beraberinde ihtiyaçları da arttırmakta ve bunun sonucu olarak da tarımsal verimi artıracak bir takım çalışmalar yapılmasını gerektirmektedir. Bu çalışmalardan en önemlisi zararlı organizmaların zararlarını azaltarak kontrol altına almak için kullanılan pestisitlerdir.

Pestisitler, gıda üretimi verimliliğini artırarak, gıda kaynaklı ve vektörle bulaşan hastalıkların azaltılmasıyla halk sağlığı yararına sahip olan geniş heterojen kimyasal grubudur (Blacquiere ve ark., 2012). Önemli bir pestisit olan neonikotinoit insektisitler, tahıl, sebze, çay ve pamuğun delici emici böceklerine ve pirelerin zararlı etkilerine karşı ekin koruma amacıyla bitkileri ve canlıları çok çeşitli zararlılara karşı tedavi etmek için

yaygın olarak kullanılan etkili bir böcek öldürücü sınıfıdır (Bolboaca ve Jaentschi, 2005). Birçok ürünün korunmasında yaygın olarak kullanılmaktadır (Van Duyn 2004).

Tarımda yaygın ve aşırı kullanımları sonucunda; bitki, toprak ve su ortamında yıllarca bozulmadan kalarak bir takım toksikolojik ve çevresel sorunlara yol açmaktadırlar (Li ve Randak, 2009; Qadir ve ark., 2014). Su kaynaklarına doğrudan uygulamalar veya yer altı sularına karışma yoluyla sucul ekosistemlere geçip ortamdaki canlı organizmada histolojik, fizyolojik ve davranışsal değişikliklere neden olmakta, aynı zamanda sucul ekosistemde yaşayan bütün organizmaların canlı yaşamını tehdit edebilmektedirler (Widdows ve Donkin, 1992; Walsh ve ark., 1994; Uno ve ark., 2001; Usheva ve ark., 2006). Çevrenin ve doğal kaynakların kirlenmesi sonucu biriktirilen insektisitler besin zinciri yoluyla biyolojik olarak biriktirilerek hayvan ve insan sağlığına olumsuz etkilere neden olmaktadırlar (Preston, 2002; Qadir ve ark., 2016).

Molluska grubundan *Melanopsis praemorsa,* toksik kimyasalların güvenilirliğini saptamak ve değerlendirmek için sucul ekosistemlerde yaygın bir şekilde kullanılan deneysel modellerden biridir (Melo ve ark., 2000; Mandauzio ve ark., 2004; Kılıçaslan ve Özbek, 2010). Tatlı su kirliliğinin potansiyel biyoindikatörlerinden biri olan *M. praemrsoa,* imidaklopridin sublethal etkilerini incelemek için umut verici bir türdür.

Histolojik inceleme, kirleticilere maruz kalmanın bir indikatörü olarak, özellikle subletal ve kronik etkiler için kirlilik derecesini değerlendirmede yararlı bir yöntemdir.

Bu çalışmanın amacı, tatlı su salyangozu *Melanopsis praemorsa*'nın neonikotinoit insektisit imidaklopridin sublethal konsantrasyonlarına maruziyeti sonucunda, ayak dokularında ortaya çıkabilecek değişiklikleri saptamaktır.

MATERYAL VE METOD

M. praemorsa örnekleri, iklimlendirme odasındaki akvaryumlara bırakılarak, 30 gün boyunca laboratuvar şartlarına aklimasyonları sağlanmıştır. Aydınlatma, 16 saat aydınlık ve 8 saat karanlık olacak şekilde ayarlanmıştır. Adaptasyon ve deneysel çalışmalarda laboratuvar 22±1 °C sabit sıcaklıkta tutulmuştur. 30 günlük adaptasyon süreci boyunca salyangozlar günde bir kez marul ile beslenmiştir (Otludil ve ark., 2004, Otludil ve Ayaz 2020, Bürçün Karakaş ve Otludil 2020).

M. praemorsa örneklerinin 96 saatlik LC_{50} değeri 401.6 µg/l olarak belirlenmiştir. Bu çalışma için seçilen subletal konsantrasyonlar 80.32 µg/l, 40.16 µg/l ve 4.016 µg/l (96 saatlik LC_{50} değerinin 1/5, 1/10 ve 1/100)'dir. Örnekler, biri kontrol grubu diğerleri de farklı konsantrasyonlardaki imidakloprid grupları olmak üzere 4 deney grubuna ayrılmış ve ayrı kavanozlara yerleştirilmiştir. Deneyin 7., 14. ve 21. günlerinde her bir maruz kalma ve kontrol grubundan rastgele 5 salyangoz alınmıştır. Salyangozlar dissekte edilerek ayak örnekleri alınmış, %10 formalin çözeltisi ile tespit edilmiştir. Formalinden alınan numuneler, musluk suyu altında 1 gece yıkanmıştır. Numuneler artan etanol serisi (%30, 50, 70, 80, 90, 96 ve 100) ile dehidre edilmiş, ksilen ile şeffaflaştırılmış ve parafin içine gömülerek 5µm kesitleri mikrotom (LEICA) ile alınmıştır. Kesitler, Hematoksilin-Eosin ile boyanmış ve ışık mikroskobu (Nikon NIS-Elements ECLIPS SE80i) ile incelenmiştir. Histopatolojik değişiklikler mikroskopta kamera (Nikon Digital SIGHT-DS2MV) ile fotoğraflanmıştır.

BULGULAR VE TARTIŞMA

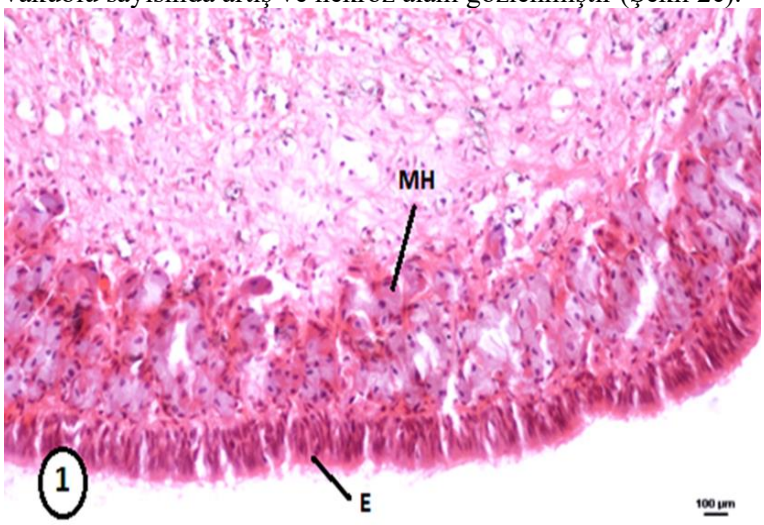
Kontrol Grubu

Ayak, vücudun ventralinde uzanan, geniş tabanlı, kaslı bir organdır. Tabanın kaslı ve geniş yüzeyinin yerle temas etmesi sonucu kolumnar kas fibrilleri nedeniyle hareket gerçekleştirilir. Suprapedal bez tarafından salgılanan mukus, ayağın zemine tutunmasını sağlar. Ayak dokusunda kolumnar kas fibrillerinin yanı sıra mukus hücreleri, protein hücreleri, pigment hücreleri, epitel hücreleri ve lipit vakuolleri de bulunur. 21 gün boyunca kontrol (Grup I) grubunda bulunan *M. praemorsa* bireylerinin ayak dokusunda herhangi bir histopatolojik değişikliğe rastlanmamıştır (Şekil 1).

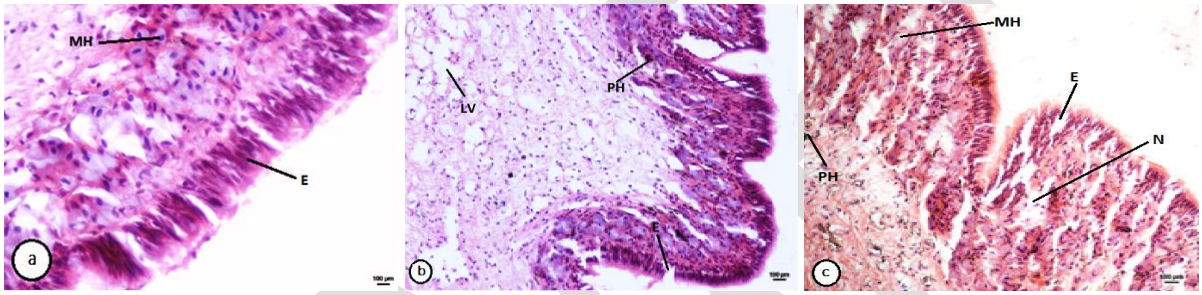
Deney Grupları

Deneyin 7 günlük tedavisi sonunda 4.016 µg/l İmidakloprid konsantrayonuna maruz bırakılan salyangoz grubunda (Grup II); ayak dokularında önemli bir değişiklik gözlenmemiştir (Şekil 2a). 40.16 µg/l İmidakloprid konsantrasyonuna maruz bırakılan salyangoz grubunda (Grup III); epitelde bozulmalar başlamış, mukus hücresi, pigment hücresi ve lipid vakuolü sayısında artış görülmüştür (Şekil 2b). 80.32 µg/l İmidakloprid konsantrasyonuna maruz bırakılan salyangoz grubunda (Grup IV); epitelde deskuamasyon, mukus hücresi,

pigment hücresi, lipid vakuolü sayısında artış ve nekroz alanı gözlenmiştir (Şekil 2c).

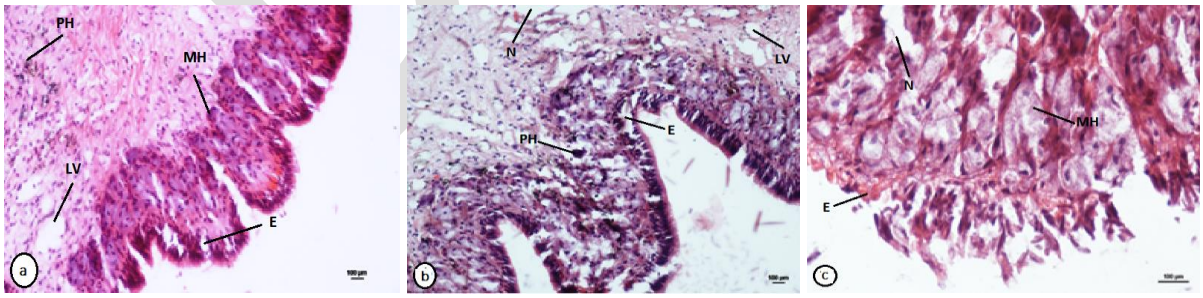


Şekil 1. *M. praemorsa*'nın 21. gün kontrol grubu (Grup I) ayak dokuları. E. Epitel, MH. Mukus hücresi. H&E.



Şekil 2. (a) 7. günde 4.016 µg/l İmidakloprid'e maruz kalan salyangozlar, (b) 7. günde 40.16 µg/l İmidakloprid'e maruz kalan salyangozlar, (c) 7. günde 80.32 µg/l İmidakloprid'e maruz kalan salyangozlar. (E. Epitel, MH. Mukus hücresi, LV. Lipid vakuolü, PH. Pigment hücresi, N. Nekroz). H&E.

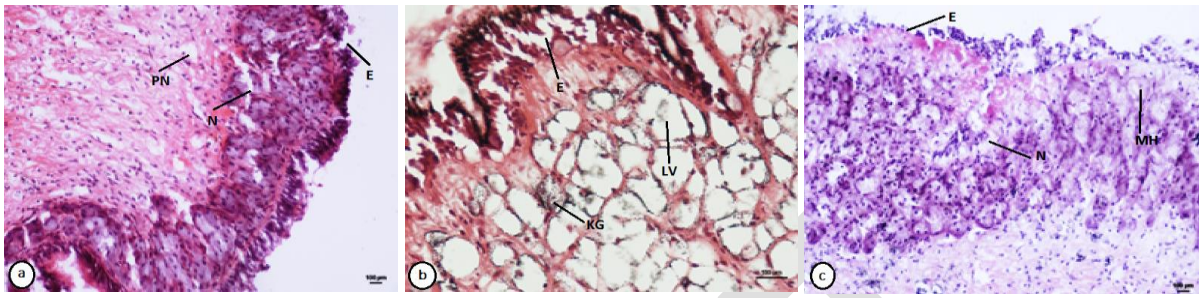
Deneyin 14 günlük tedavisi sonunda 4.016 µg/l İmidakloprid konsantrayonuna maruz bırakılan salyangoz grubunda (Grup II); ayak dokularında epitel tabakasında bozulmalar, mukus hücresi, pigment hücresi ve lipid vakuolü sayısında artış saptanmıştır (Şekil 3a). 40.16 µg/l İmidakloprid konsantrasyonuna maruz bırakılan salyangoz grubunda (Grup III); epitelde deskuamasyon ve kas fibrillerinde atrofi ilerlemiş, pigment hücresi, protein hücresi ve lipid vakuolü sayısı artış göstermiştir (Şekil 3b). 80.32 µg/l İmidakloprid konsantrasyonuna maruz bırakılan salyangoz grubunda (Grup IV); epitel deskuamasyonu aşırı ilerlemiş, mukus hücresi, protein hücresi ve lipid vakuollerinde aşırı artış görülmüştür (Şekil 3c).



Şekil 3. (a) 14. günde 4.016 µg/l İmidakloprid'e maruz kalan salyangozlar, (b) 14. günde 40.16 µg/l İmidakloprid'e maruz kalan salyangozlar, (c) 14. günde 80.32 µg/l İmidakloprid'e maruz kalan salyangozlar. (E. Epitel, MH. Mukus hücresi, LV. Lipid vakuolü, PH. Pigment hücresi, N. Nekroz). H&E.

Deneyin 21 günlük tedavisi sonunda 4.016 µg/l İmidakloprid konsantrayonuna maruz bırakılan salyangoz grubunda (Grup II); epitel tabakasında deskuamasyon, lipid vakuolü sayısında artış ve kas fibrillerinde atrofi

oluşmuştur (Şekil 4a). 40.16 µg/l İmidakloprid konsantrasyonuna maruz bırakılan salyangoz grubunda (Grup III); epitel deskuamasyonu ve kas fibrilleri atrofisi devam etmiştir, Lipid vakuolü sayısı aşırı derecede artmış ve içlerinde bol miktarda koyu granüller birikmiştir (Şekil 4b). 80.32 µg/l İmidakloprid konsantrasyonuna maruz bırakılan salyangoz grubunda (Grup IV); epitel tabakasında aşırı derecede deskuamasyon gözlenmiştir. Kas fibrillerinde aşırı derecede atrofi oluşmuş, kas doku yerini bağ dokuya bırakmıştır. Mukus hücresi ve lipid vakuolü sayısında aşırı artış gözlenmiş ayrıca nekroz tespit edilmiştir (Şekil 4c).



Şekil 4. (a) 21. günde 4.016 µg/l İmidakloprid'e maruz kalan salyangozlar, (b) 21. günde 40.16 µg/l İmidakloprid'e maruz kalan salyangozlar, (c) 21. günde 80.32 µg/l İmidakloprid'e maruz kalan salyangozlar. (E. Epitel, PN. Piknotik nukleus, N. Nekroz, LV. Lipid vakuolü, KG. Koyu granül, MH. Mukus hücresi) H&E.

M. praemorsa ayak dokularının histolojik incelemesi, İmidakloprid'in test edilen en düşük konsantrasyondan itibaren belirgin yapısal bozulmaya neden olduğunu gösterdi. Toksik etkilerin gözlemlenen dokularda belirgin olduğu ve bunların farklı salyangoz türleri üzerinde yapılan ağır metal ve pestisit çalışmaları ile uyumlu olduğu görülmüştür (Otludil ve ark., 2004; El-Feky ve ark., 2009; Otludil ve Ayaz, 2020; Bürçün Karakaş ve Otludil, 2020).
M. praemorsa örneklerinde mukus hücreleri, pigment hücreleri, protein hücreleri ve lipit vakuollerinde artış görüldü. Ayak dokusunda epitelde deskuamasyon ve kas fibrillerinde atrofi meydana geldiği gözlendi. Artan mukus üretiminin ardından artan mukus salgısı, gastropodların, mekanik uyaranlardan veya yumuşakça öldürücü kimyasallardan kaynaklanan tahriş dahil olmak üzere birçok stres türüne karşı ilk reaksiyonlarından biridir (El-Feky ve ark., 2009). Çalışmamızda ayak dokusunda görülen histopatolojik değişiklikler daha önce yapılan pestisit ve $CuSO_4$ ve Cd çalışmaları ile uyumludur.

SONUÇ

Bu çalışmanın histopatolojik gözlemleri İmidakloprid'in ölümcül olmayan konsantrasyonlarına maruz kalmanın tatlı su salyangozu *Melanopsis praemorsa*'nın vücut dokularında yıkıcı etkilere neden olduğunu göstermiştir. Bu değişiklikler, kirletici maddelere maruz kalmanın biyolojik son noktasını temsil etmektedir ve bu nedenle bu çalışma, sucul ortamların antropojenik kirlenmesini izlemek için histopatolojinin güçlü bir araç olarak uygulanmasını desteklemektedir. Bu çalışma, tatlı su salyangozu *Melanopsis praemorsa*'nın, dokularına ciddi zarar vermesi nedeniyle İmidakloprid gibi ağır metaller için bir biyoindikatör olarak hizmet edebildiğini ve su ortamında İmidakloprid kirliliğinin izlenmesi için bir biyobelirteç olabileceğini göstermiştir.

Salyangozlar üzerinde yapılan histopatolojik çalışma, çevresel kirleticilerin etkisinin değerlendirilmesinde oldukça hassas bir parametre olarak görülmektedir.

KAYNAKLAR

Blacquiere T, Smagghe G, Gestel C, Mommaerts V 2012. Neonicotinoids in bees: A review on concentrations, side-effects and risk assessment. Ecotoxicology, 21(4): 973-992.

Bolboaca SD, Jaentschi L 2005. Molecular descriptors family on structure activity relationships. 2. Insecticidal activity of neonicotinoid compounds. Journal of Pesticide Science, 4:78–85.

Bürçün Karakaş S, Otludil B 2020. Accumulation and histopathological effects of cadmium on the great pond snail *Lymnaea stagnalis* Linnaeus, 1758 (Gastropoda: Pulmonata). Environmental Toxicology and Pharmacology, 78:103403.

Downs CA, Dillon RT, Fauth JE, Woodley CM 2001. A molecular biomarker system for assessing the health of gastropods (*Ilyanassa obsoleta*) exposed to natural and anthropogenic stressors. J. Exp.

Mar. Biol. Ecol. 259: 189–214.
El-feky F, Raafat HA, Kamal H 2009. Physiological and Histopathological Effects of Tributyletin (TBT) on *Lymnaea natalensis* and *Physa acuta*. The Egyptian Journal of Hospital Medicine, 37: 610– 620.
Li ZH and Randak T 2009. Residual pharmaceutically active compounds (PhACs) in aquatic environment - status, toxicity and kinetics: A review. Vet. Med. 52: 295-314.
Mandauzio H, Monsinjon CG, Leboulonger F and Rocher B 2004. Seasonal variations in antioxidant defenses in blue mussel *Mytilus edulis* collected from a polluted area: major contributions in gills of an inducible isoform of Cu/Zn superoxide dismutase and of glutathione-S-transferase. Aqu. Toxicol., 70: 83-93.
Preston B. 2002. Indirect effects in aquatic ecotoxicology: Implications for ecological risk assessment. Environ. Manage, 29: 311-323.
Otludil B, Cengiz EI, Yıldırım MZ, Unver O and Unlu E 2004. The effect of endosulfan on the great rampshorn snail *Planorbarious corneus* (Gastropoda: Pulmonata): a histological study. Chemosphere 56:707-716.
Otludil B, Ayaz S 2020. Effect of Copper Sulphate (CuSO4) on Freshwater Snail, *Physa acuta* Draparnaud, 1805: A Histopathological Evaluation. Bulletin of Environmental Contamination and Toxicology. 104:738–747. Preston BL 2002. Indirect effects in aquatic ecotoxicology: Implications for ecological risk assessment. Environ. Manage, 29: 311-323.
Uno S, Shiraishi H, Hatakeyama S, Otsuki A. and Koyama J 2001. Accumulative characteristics of pesticide residue in organs of bivalves (*Anodontawoodiana* and *Corbicula leana*) under natural condition. Arch. Environ. Contam. Toxicol. 40: 35-47.
Usheva LN, Vaschenko MA, Durkina VB 2006. Histopathology of the digestive gland of bivalve mollusk *Crenomytilus grayanus* (Dunker 1853) from South Western Peter, The great Bay, Sea of Japan. Russian J. Mar. Biol. 32(3): 166-172.
Van Duyn J 2004. Neonicotinoid Insecticide Seed Coatings For Protection of Planted Corn Kernels and Seedlings, 78th Annual Meeting of the Southeastern Branch Entomological Society of America, February 15-18, USA.
Walsh K, Dunstan RH, Murdoch RN, Conroy BA 1994. Roberts TK and Lake P. Bioaccumulation of pollutants and changes in population parameters in the gastropod mollusc *Austrocochlea constricta*. Arch. Environ. Contom. Toxicol.26: 367-373.
Widdows J, Donkin P 1992. Mussels and environmental contaminants: bioaccumulation and physiological aspects. In: The Mussel *Mytilus*: Ecology, Physiology, Genetics and culture. (Ed. Gosling E) Amasterdam. Elsevier Press, pp. 383- 464.
Qadir S, Latif A, Ali M, Iqbal F 2014. Effects of Imidacloprid on the Hematological and Serum Biochemical Profile of *Labeo rohita*. Pak. J. Zool., 46(4): 1085-1090.
Qadir S, Bukhari R, Iqbal F 2015. Effect of Sub Lethal Concentration of Imidacloprid, on Proximate Body Composition of *Labeo rohita*. Iran J. Fish. Sci., In Press.
Qadir S, Iqbal F 2016. Effect of subleathal concentrtion of imidacloprid on the histology of heart, liver and kidney in Labeo rohitr. Pakistan journal of pharmaceutical sciences, 29 (6).

DRAFT

ORAL PRESENTATION

Preliminary Results on Pentose Phosphate Pathway Changes in Experimental Allergic Encephalomyelitis Mice

Ozden OZGUN ACAR* (*ORCID: https://orcid.org/0000-0002-2910-6349*)

*[1]Pamukkale University, Health Services Vocational School of Higher Education Denizli, Turkey.

*Corresponding author e-mail: ozdena@pau.edu.tr

Abstract

Metabolic disorders are thought to play a role in neurodegenerative disorders including Multiple Sclerosis (MS), Alzheimer's disease (AD), Huntington's disease (HD) and Parkinson's disease (PD). Research on the contribution of corrupted glucose metabolism to disease pathology in demyelinating diseases, particularly MS, is limited. The current study was designed to reveal changes in mRNA expression and activity levels of important pentose phosphate pathway energy metabolism enzymes in normal and diseased stated using an MS mouse model of experimental autoimmune encephalomyelitis (EAE). First, EAE was elicited in C57BL6 female mice by immunization with MOG_{35-55}/CFA Emulsion and PTX. Following immunization, mice were monitored daily for the development of EAE and clinically rated on a scale ranging from 0 to 5 for symptoms of the disease. The onset of EAE occurred on day 13.0±1.5, with a maximal clinical score of 3.8±0.2. Biochemical and molecular analyses were performed on the brain tissues of EAE and control mice to measure changes in energy metabolism. In the EAE group, the brain tissue transaldolase (TALDO) mRNA expression level and glucose 6-phosphate dehydrogenase (G6PD) enzyme activity decreased significantly compared to those in the control. These data revealed that there are impairments in brain energy metabolism in mice with experimental MS.

Keywords: Multiple sclerosis, experimental autoimmune encephalomyelitis, energy metabolism, glucose-6-phosphate dehydrogenase, transaldolase.

INTRODUCTION

Multiple sclerosis (MS) is a chronic, demyelinating neurological disease that affects over 2.8 million people worldwide (Raphael et. al., 2015, Wallin et. al., 2019). Most studies have focused on the abnormal functions of diverse immune cells, especially T cells, which play critical roles in MS pathogenesis (McFarland and Martin 2007; Compston and Coles 2008). Recent evidence indicates a new aspect of MS pathogenesis, namely, energy failure in the central nervous system (CNS) (Andersen et al. 2019; Podlecka-Pietowska et al. 2019). Metabolomic studies have demonstrated metabolic fluctuations in MS and have suggested that impaired metabolism may be associated with MS pathogenesis (Kim et al. 2017). In addition, the disruption of metabolism disrupts the brain nicotinamide adenine dinucleotide phosphate (NADPH) balance, which increases axonal oxidative damage. Protection against oxidative damage relies largely on the reducing power of NADPH, which is produced by glucose-6-phosphate dehydrogenase (G6PD) in the pentose phosphate pathway (Diaz-Vivancos et. al., 2015, Kim et. al., 2015, Nobrega-Pereira et. al., 2016).

Experimental autoimmune encephalomyelitis (EAE) is a commonly employed animal model for multiple sclerosis (MS). EAE is induced by myelin antigens, including myelin oligodendrocyte glycoprotein (MOG), myelin basic protein (MBP), or proteolipid protein (PLP) (Steinman et al., 2015).

The objective of this work was to collect initial data regarding the alterations in brain energy metabolism in mouse brain tissue that had developed experimental autoimmune encephalomyelitis (EAE), which serves as an animal model for multiple sclerosis (MS).

DRAFT

MATERIALS AND METHODS

Immunization of Experimental Autoimmune Encephalomyelitis (EAE)

Healthy 8- to 10-week-old female C57BL/6 mice were purchased from the The Bilkent University Animal House (Ankara, Turkey). They were housed in small cages at the animal care facility of the University of Pamukkale (Denizli, Turkey) at an ambient temperature of 22 ± 1 °C and a 12-h light/dark cycle. All experimental procedures and appropriate regimes for veterinary services within licensed projects were approved by the Institutional Experimental Animal Ethics Committee (PAUHADYEK-2021/51). After acclimatization for one-week, experimental allergic encephalomyelitis (EAE) was induced in mice by immunization with the Hooke Kit MOG_{35-55}/CFA Emulsion and PTX. Each mouse received 0.1 mL MOG35-55/CFA Emulsion subcutaneously at two sites. Following immunization, 400 ng of pertussis toxin in 100 µL of PBS was intraperitoneally injected into mice on days 0 and 1 (Senol at al., 2023). Followed by the development of EAE and graded clinically on a scale of 0 to 5 by a blinded observer for disease symptoms [0 = no symptoms; 1 = loss of tail tonicity; 2 = hind limb weakness; 3 = ataxia and/or paresis of hind limbs; 4 = complete paralysis of hind limbs; 5 = moribund or dead)] (Ozgun-Acar et. al., 2016). At the end of the study, which included 16 hours of fasting, the animals were euthanized. The brains were then extracted, frozen in liquid nitrogen, and stored at −80°C until use.

Reverse Transcription-Polymerase Chain Reaction (RT-PCR) and Real-Time Quantitative PCR for Gene Expression Analysis at mRNA Levels

Real-time PCR was performed as previously optimized (Senol et. al., 2023). Total RNA was extracted from mouse brains by using NucleoSpinTriPrep (Macherey-Nagel), Mini kit for RNA, DNA, and protein purification" according to the manufacturer's instruction with slight modifications. The RNA concentration was determined using a Nanodrop (MaestroNano micro-volume Spectrophotometer, USA) and the RNA was reverse transcribed using Easy Script cDNA Synthesis Kit (ABM). The reaction mixture was incubated for 45 min at 50ºC, followed by termination by heating at 5 min 85 ºC. Quantitative Real-Time PCR (qRT-PCR) analysis was performed using BlasTaq 2X qPCR Mastermix (GM, Taiwan) in an Bio-Rad CFX Connect Real Time System for G6PD, 6-phosphogluconate dehydrogenase (6-PGD), TALDO and transketolase (TKT) genes. Beta-actin (ACTB) gene was used as a control housekeeping gene.

Determination of the Glucose-6-Phosphate Dehydrogenase (G6PD) Activity

Brain G6PD activity was detected using the G6PD Colorimetric Assay Kit (Elabscience), according to the manufacturer’s instructions.

Statistical Analysis

Statistical analyses were performed using the Minitab 13 statistical software package (Minitab Inc. State College, PA, USA). Comparisons between groups were performed using Student's t-test, and $P< 0.05$ was selected as the level required for statistical significance.

RESULTS

Induction of Experimental Autoimmune Encephalomyelitis in C57BL6 Mice

EAE induction was performed in C57BL6 mice using the Hooke Kit MOG_{35-55}/CFA Emulsion and PTX. After the injection, the animals were checked daily, and the behavior of the mice was determined using a 5-step scale (Ozgun-Acar et. al., 2016) (Figure 1).

DRAFT

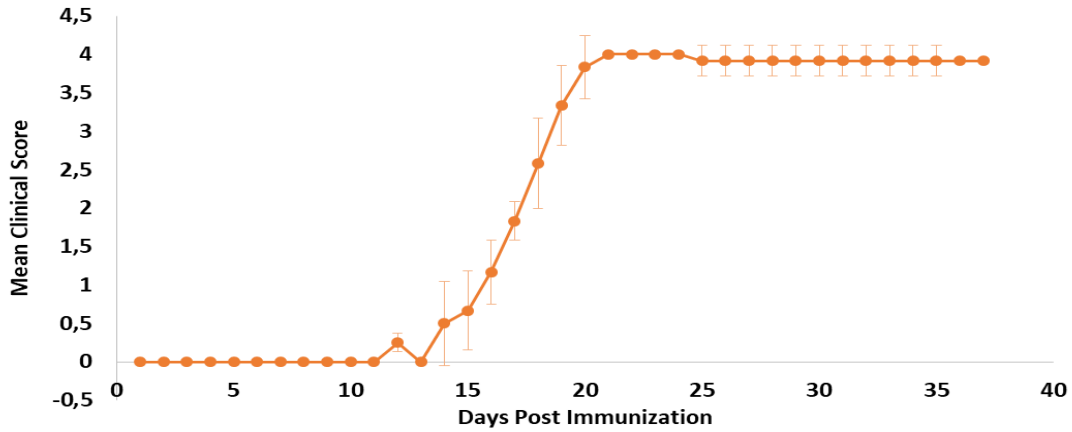


Figure 1. The graph indicates the changes in mice during the experimental period. Results are expressed as mean clinical scores ± standard deviation (SD).

The first disease symptoms appeared on the 13th day following immunization, and the disease score reached 3.8±0.2 on the 21st day.

mRNA Expression of Major Energy Metabolism Genes in EAE Mice

The mRNA expression levels of energy metabolism genes were determined in the brain tissues of the EAE and control groups mice. TALDO mRNA expression level was observed to decrease 2.7-fold in the EAE group compared to the control group. However, no significant difference was observed in the G6PD, 6-PGD TALDO and TKT mRNA expression levels between the EAE group and the control group (Figure 2).

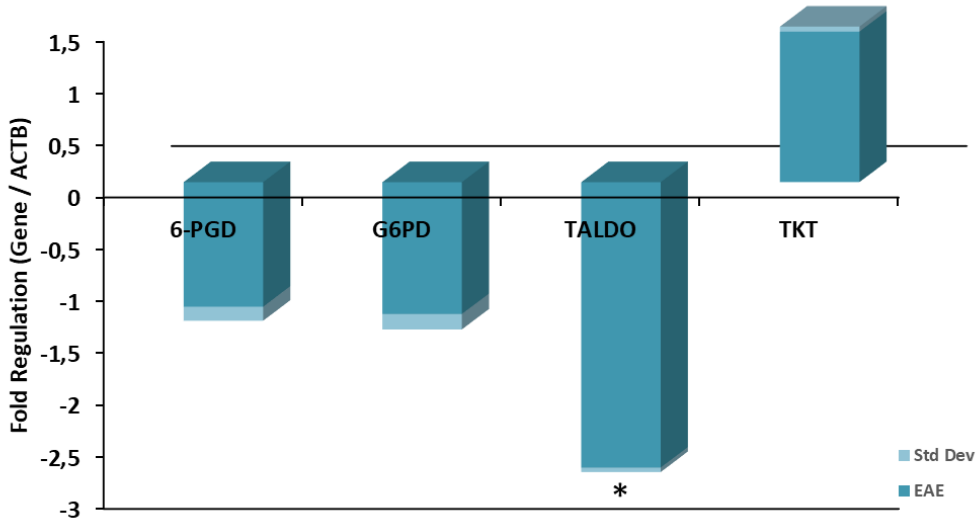


Figure 2. mRNA expression levels of selected genes (fold regulation) in the brain tissues of EAE mice. *Significantly different from the respective control value $p < 0.05$. Control values were taken as 1.

We found that the expression of TALDO mRNA level decreased by 2.7-fold in the EAE group, while the mRNA levels of G6PD, 6-PGD, and TKT were not significantly different from those in the control group.

Glucose-6-Phosphate Dehydrogenase (G6PD) Activity in EAE Mice

The G6PD activity was measured in the brain tissue of EAE-induced mice. The G6PD activity was determined in the control and EAE groups mice brain tissues. As shown in Figure 3, G6PD activity decreased by 51 % ($p<0.05$) in the EAE group compared to the control group.

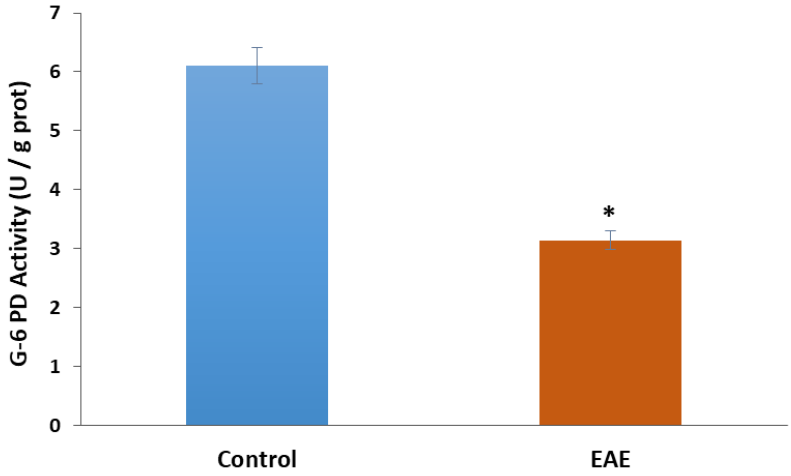


Figure 3. Changes in G6PD activity in the mouse brain control and EAE groups. *Significantly different from the respective control value $p < 0.05$. Control values were taken as 1.

DISCUSSION

In recent times, there has been a significant amount of research conducted on the association between diseases and energy metabolism, with a particular focus on glucose metabolism, as seen by the growing interest in this area (Vazquez et al., 2016). The pentose phosphate pathway (PPP) is a significant metabolic pathway that is closely linked to glucose metabolism. The PPP plays a significant role in various biological processes, including oxidative stress, immunity, embryogenesis, morphogenesis, myelination, inflammation, lymphocyte activation, radiation resistance, proliferation, and growth (Jacobson, 1963; Kelso et al., 1989). Furthermore, it has been observed that several metabolic, neurological, and neurodegenerative disorders are linked to abnormalities in the genes associated with the pentose phosphate pathway (Perl, 2011; Rahman and Hasan, 2014). Glucose-6-phosphate dehydrogenase (G6PD) is an enzymatic protein that is ubiquitously distributed throughout several tissues, with particularly elevated levels of activity inside the brain. The enzyme G6PD plays a crucial role in the regulation of inflammation through its production of nicotinamide adenine dinucleotide phosphate (NADPH) (Perl, 2017). While there was a considerable decrease in the activity of the G6PD enzyme, no changes were observed in the mRNA level. The findings of this study indicate a significant involvement of post-translational changes of G6PD in the regulation of its structure, enzyme activity, and function.

Another enzyme TALDO, which plays a significant role in the PPP, exhibits selective overexpression specifically in oligodendrocytes within the brain, as demonstrated by Banki et al. in 1994. The topic under consideration is of particular relevance due to the prevalence of demyelinating diseases in the central nervous system (CNS), with multiple sclerosis (MS) being the most frequently encountered. In MS, the primary focus of autoimmune activity is directed against the myelin layer, which is formed by oligodendrocytes. These cells are characterized by the loss of both cell integrity and myelin, as observed in previous studies (Martin et al., 1992). In the present investigation, it is probable that the observed reduction in TALDO mRNA expression might be attributed to the deleterious effects on the myelin sheath in mice with experimental autoimmune encephalomyelitis (EAE).

CONCLUSION

In summary, the available evidence clearly indicates that there is a notable impairment in the energy metabolism of PPP in mice with experimental autoimmune encephalomyelitis (EAE), therefore implying a potential involvement in the pathophysiology of the disease. Based on the available data, ongoing research is being conducted in our laboratory to identify metabolic variability resulting from energy metabolism abnormalities in multiple sclerosis (MS).

ACKNOWLEDGEMENTS

This work was supported by the Scientific and Technological Research Council of Turkey [TUBITAK-221Z131].

REFERENCES

Andersen SL, Briggs FBS, Winnike JH, Natanzon Y, Maichle S, Knagge KJ, Newby LK, Gregory SG 2019. Metabolome-based signature of disease pathology in MS. Mult Scler Relat Disord, 31: 12–21.

Banki K, Colombo E, Sia F, Halladay D, Mattson DH, Tatum AH, Massa PT, Phillips PE, Perl A 1994. Oligodendrocyte-specific expression and autoantigenicity of transaldolase in multiple sclerosis. Journal of Experimental Medicine, 180: 1649-1663.

Compston A, Coles A 2008. Multiple sclerosis. Lancet, 372: 1502– 1517.

Diaz-Vivancos P, de Simone A, Kiddle G, Foyer CH 2015. Glutathione-linking cell proliferation to oxidative stress. Free Radical Biology and Medicine, 89: 1154–1164.

Jacobson S 1963. Sequence of myelination in the brain of the albino rat. Journal of Comparative Neurology, 121: 5-29.

Kelso TB, Shear CR, Max SR 1989. Enzymes of glutamine metabolism in inflammation associated with skeletal muscle hypertrophy. American Journal of Physiology, 257: 885-894.

Kim GH, Kim JE, Rhie SJ, Yoon S. 2015. The role of oxidative stress in neurodegenerative diseases. Exp Neurobiol, 24(4): 325-40.

Kim HH, Jeong IH, Hyun JS, Kong BS, Kim HJ, Park SJ 2017. Metabolomic profling of CSF in multiple sclerosis and neuromyelitis optica spectrum disorder by nuclear magnetic resonance. PLoS One, 12: e0181758.

Martin R, McFarland HF, McFarlin DE 1992. Immunological aspects of demyelinating diseases. Annual Review of Immunology, 10: 153-187.

McFarland HF, Martin R 2007. Multiple sclerosis: a complicated picture of autoimmunity. Nat Immunol 8: 913–919.

Nobrega-Pereira S, Fernandez-Marcos PJ, Brioche T, Gomez-Cabrera MC, Salvador-Pascual A, Flores JM, et al. 2016. G6PD protects from oxidative damage and improves healthspan in mice. Nat Commun, 7: 10894.

Ozgun-Acar, O, Celik-Turgut G, Gazioglu I, Kolak U, Ozbal S, Ergur BU, Arslan S, Sen A, Topcu, G 2016. Capparis ovata treatment suppresses inflammatory cytokine expression and ameliorates experimental allergic encephalomyelitis model of multiple sclerosis in C57BL/6 mice. Journal of Neuroimmunol, 298: 106−116.

Perl A, Hanczko R, Telarico T, Oaks Z, Landas S 2011. Oxidative stress, inflammation and carcinogenesis are controlled through the pentose phosphate pathway by transaldolase. Trends in Molecular Medicine, 17: 395-403.

Perl A 2017. Metabolic control of immune system activation in rheumatic diseases. Arthritis Rheumatology, 69(12): 2259-2270.

Podlecka-Piętowska A, Kacka A, Zakrzewska-Pniewska B, Nojszewska M, Zieminska E, Chalimoniuk M, Toczylowska B 2019. Altered cerebrospinal fuid concentrations of hydrophobic and hydrophilic compounds in early stages of multiple sclerosis metabolic profle analyses. J Mol Neurosci, 69: 94–105.

Rahman M, Hasan MR 2014. Pentose phosphate pathway in disease and therapy. Advanced Materials Research, 995: 1-27.

Raphael I, Webb J, Stuve O, Haskins WE, Forsthuber TG 2015. Body fluid biomarkers in multiple sclerosis: how far we have come and how they could affect the clinic now and in the future. Exp. Rev. Clin. Immunol. 11: 69–91.

Senol H, Ozgun-Acar O, Dag A, Eken A, Guner H, Aykut ZG, Topcu G, Sen A 2023 Synthesis and Comprehensive in Vivo Activity Profiling of Olean-12- en-28-ol, 3β-Pentacosanoate in Experimental

Autoimmune Encephalomyelitis: A Natural Remyelinating and Anti-Inflammatory Agent. Journal of Natural Product, 86: 103−118.

Steinman, L. 2001. "Multiple sclerosis: a two-stage disease", Nature Immunology, 2, 762-764.

Vazquez A, Kamphorst JJ, Markert EK, Schug ZT, Tardito S, Gottlieb E 2016. Cancer metabolism at a glance. Journal of Cell Science, 129: 3367-3373.

Wallin MT, Culpepper WJ, Campbell J D, Nelson LM, Langer-Gould A, Marrie RA, Cutter GR, Kaye W E, Wagner L, Tremlett H, et al 2019. The prevalence of MS in the United States: A population-based estimate using health claims data. Neurology, 92:1029-1040.

DRAFT

ORAL PRESENTATION

Makromantarların kozmetik endüstrisinde kullanım olanakları

Funda ATİLA[1*] (*ORCID: https://orcid.org/ 0000-0003-1129-1045*)

[*1]Ege Üniversitesi, Bergama Meslek Yüksekokulu, Mantarcılık Programı, İzmir, Türkiye

*Sorumlu yazar e-mail:funda.atila@ege.edu.tr

Özet

Günümüzde, sentetik kimyasalların insan vücudu üzerindeki potansiyel zararlı etkilerine ilişkin kamuoyunun endişeleri ve doğal ürünlerin güvenli ve sağlıklı olduğuna dair genel bir inanç nedeniyle, kozmetik endüstrisi de sürekli olarak doğal kaynaklardan içerik arayışındadır. Mantarlar besin olarak ve tıbbi amaçlarla yüzlerce yıldan beri insanlar tarafından yaygın bir şekilde kullanılmaktadırlar. Mantarların sahip oldukları polisakkaritler, fenolik bileşikler, terpenoidler poliketidler, seramidler, vitaminler, selenyum ve L-ergotionin, gibi biyoaktif içerikler antioksidan, anti-tirozinaz, anti-inflamatuar, anti-hiyalüronidaz, anti-kollajenaz, anti-elastaz ve antimikrobiyal aktivitelere sahiptirler. Bu özellikler onları kozmetik endüstrisinde UV ışınlarına karşı koruyucu, yaşlanma karşıtı, nemlendirici ve cilt beyazlatıcı ürünlerin üretiminde kullanılmak üzere ideal adaylar haline getirir. Bu derlemede, doğadan gelen muhteşem hediyeler olarak potansiyel kozmetik uygulamalara sahip bazı mantar türlerinin içerdiği biyoaktif maddeleri ve kozmetik özellikleri gözden geçirilmiştir.

Anahtar Kelimeler: kozmesötikler, nutrikozmetikler, antiaging, cilt sağlığı

Possibilities of using macrofungus in the cosmetics industry

Abstract

Nowadays, due to public concerns about the potential harmful effects of synthetic chemicals on the human body and a general belief that natural products are safe and healthy, the cosmetics industry is constantly seeking ingredients from natural sources. Mushrooms have been widely used by humans for hundreds of years as food and for medicinal purposes. Mushrooms contain bioactive ingredients such as polysaccharides, phenolic compounds, terpenoids, polyketides, ceramides, vitamins, selenium and L-ergothionein, which have antioxidant, anti-tyrosinase, anti-inflammatory, anti-hyaluronidase, anti-collagenase, anti-elastase and antimicrobial activities. These properties make them ideal candidates for use in the cosmetics industry for the production of UV protection, anti-aging, moisturizing and skin whitening products. In this review, the bioactive substances and cosmetic properties of some mushroom species with potential cosmetic applications as wonderful gifts from nature are reviewed.

Keywords: cosmeceuticals, nutricosmetics, antiaging, skin health

GİRİŞ

Kozmetik, cildi temizlemek, güzelleştirmek, korumak, görünümünü değiştirmek veya durumunu iyileştirmek için kullanılan bir grup sağlık ve güzellik ürünü olarak tanımlanabilir (Luque de Castro, 2011). Kozmetiklerin "kozmesötikler" ve "nutrikozmetikler" olmak üzere iki ana kategorisi vardır: kozmesötikler, direkt olarak cilde uygulanan kremler, losyonlar, merhemleri kapsarken (Epstein, 2009; Taofiq ve ark., 2016), nutrikozmetikler ise görsel görünüm düzeltmek ve cilt sağlığını iyileştirmek için diyet ve besin takviyelerinin tüketimini içerir (Luque de Castro, 2011). Bu ürünlerin, cilt görünümünü, parlaklığını, dokusunu iyileştirip, yaşlanma karşıtı aktivite gösteren çeşitli bileşenleri içerdiği bilinmektedir (Hyde ve ark., 2010). Bu nedenle formülasyonları genellikle saf moleküller (karotenoidler, polifenolikler, vitaminler, amino asitler, poliglikanlar, çoklu doymamış yağ asitleri) içeren karışımlara veya bir bütün olarak alınan ekstraktlara dayanır (Barel ve ark., 2014).

Küresel kozmetik pazarı yüz milyarlarca ABD doları değerinde bir endüstridir ve ürün çeşitlendirmesine yönelik sürekli talebe yanıt vermek için küresel kozmetik pazarının sürekli büyümesi gerekmektedir. Günümüzde kimyasalların insan sağlığı üzerinde yarattıkları olumsuz etkiler artık toplumun tüm fertleri tarafından açık bir şekilde anlaşılmıştır. Bu durum tüketicilerin kozmetik ürün satın alırken de doğal kaynaklardan elde edilen ürünleri tercih etmesine sebep olmaktadır (Wang ve ark., 2015).

Birçok bitki özü (aloe vera, yaban mersini, hindistancevizi, salatalık, ginseng, limon, nar, pirinç) ve deniz kulağı, arı zehiri, koza, keçi sütü ve plasenta, bal, inci gibi hayvanların özleri, arı sütü ve salyangoz salgıları kozmetik mağazalarının raflarında sıradan hale gelmiştir (Taofiq ve ark., 2016; De Wet ve ark., 2013; Keller ve ark., 2014; Rodrigues ve ark., 2015). Yeni ortaya çıkan bir trend ise, mantarların sağlık ve güzellik ürünlerinde kullanılmasıdır. Gümümüzde mantarlar genellikle ciltle ilişkili çeşitli hastalıkların tedavisinde ham madde veya saf bileşikler olarak yaygın şekilde kullanılmaktadır (Hyde ve ark., 2010). Örneğin *Grifola frondosa* (maitake) mantar ekstraktları cilt hastalıklarına karşı bağışıklık sisteminin uyarımında ve *Sparassis crispa* (Wulfen) Fr. mantarından ise diyabet yaralarına karşı kollajen I sentezi ve epitelizasyon amaçlı faydalanılır (Xu ve ark., 2010; Thi Ngoc ve ark., 2018).

Mantarlar amino asitler, karbonhidratlar, yağlar, diyet lifleri, proteinler, fenolik bileşikler ve diğer temel mineraller ve vitaminlerden oluşan yüksek besin içeriğine sahiptir. Bu besleyici özelliklerin yanı sıra mantar türevi maddeler günümüzde birçok hastalığın tedavisinde kullanılmaktadır. Mantar bileşenleri ve bunların sekonder metabolitleri, antimikrobiyal, antiviral, anti-kanser, anti-inflamatuar, anti-aging, antioksidan, kırışıklık önleyici, nemlendirici, cilt beyazlatıcı ve immünomodülatör etkiler dahil olmak üzere çok sayıda biyolojik aktivite içerir (Taofiq ve ark., 2019; Taofiq ve ark., 2020; Nitthikan ve ark., 2022). Mantar kozmesötik içerikleri yavaş yavaş kozmetik pazarına girmektedir ve bu, mantar özleri ve/veya bunların biyoaktif bileşenlerini içeren, ince çizgilere, kırışıklıklara, yaşlanmaya, cilt dokusuna, ışıktan koruma ve pigmentasyona karşı faydalı etkiler iddia eden ticari kozmetik formülasyonlarının sayının arttığı açıkça görülmektedir (Taofiq et ark., 2016; Wu ve ark., 2016). Bu derlemede, mantarların yapısında yer alan ve cilde faydaları olduğu bildirilen bazı biyoaktif bileşikler özetlenmiş ve kozmetik endüstrisinde faydalanılabilecek bazı mantar türlerinin kozmetik özellikleri gözden geçirilmiştir.

MANTARLARIN YAPISINDA YER ALAN VE KOZMETİK ÖZELLİKLERİ OLDUĞU BELİRLENEN BAZI BİYOAKTİF BİLEŞENLER

Mantar ekstraktları ve misellerden, sporlardan ve mantar şapkalarından elde edilebilir. Bu nedenle bunlar, harici (kosmesötik) veya oral kullanım (nutrikozmetik) için toz, sıvı veya yarı katı olarak depolanan birincil ve ikincil metabolitlerin oldukça karmaşık karışımlarıdır (Badalyan, 2014). Mantarların en önemli avantajları, vegan doğaları nedeniyle kozmetik içerik maddeleri olarak popüler olmaları ve kimyasal maddelere göre uzun vadeli olumsuz etkilerinin daha az olmasıdır (Sujarit ve ark., 2021). Mantar günümüzde çeşitli kozmetik firmaları tarafından losyonlar, aydınlatıcı kremler, serumlar, nemlendiriciler, temizleme jelleri, astar kremler ve yüz maskeleri gibi cilt bakım ürünlerinde bileşen olarak kullanılmaktadır.

Mantar türleri yetiştirme şartlarına ve genetik özelliklerine göre büyük oranda değişiklik gösteren besin ve biyoaktif bileşik içeriklerine sahiptirler. Polisakkaritler mantarların yapısında yer alan önemli bir bileşendir ve bunu proteinler, antioksidanlar, mineraller ve vitaminler takip etmektedir (Rosdan Bushra, ve Nurul, 2022). Mantarlardaki biyoaktif bileşenler, bir besin kaynağı olmasının yanı sıra birçok hastalık için terapötik ve cilt yaşlanmasının belirtilerini geciktirmek için kozmetik ajanlar olarak birçok çalışmada yoğun bir şekilde incelenmiş ve bu bileşenlerin yeni doğal kozmesötik bileşenler olarak büyük bir potansiyele sahip olduklarına dair birçok kanıt elde edilmiştir.

Polisakkaritler

Polisakkaritler, mantar kuru ağırlığının büyük bir kısmını kapsayan yapısal birincil metabolitler oldukları için şu anda farmasötik pazarında yaygın şekilde faydalanılan bileşiklerdir. Lentinan, bağışıklık tepkisini güçlendirmesinin yanı sıra harici olarak kullanılan kremlerin bileşiminde de yer alabilen bir fungal polisakkarittir (Bisen ve ark. 2010). *Pleurotus ostreatus*'tan Pleuran, *Schizophyllum commune*'dan şizofillan, *Ganoderma lucidum*'dan Ganoderan, *Grifola frondosa*'dan grifolan ve galaktomannan, *Trametes versicolor*'dan krestin (polisakkarit-K) gibi birçok polisakkarit, mantar şapkalarından ve misel kültürlerinden

ekstrakte edilmiştir ve bu polisakkaritler yaşlanma karşıtı anti-oksidatif, antiterapötik ve immünomodülatör aktivitelere sahip oldukları belirlenmiştir (Zhao ve ark. 2020).

Mantar polisakkaritlerinin yüksek konsantrasyonu, epidermis ve stratum korneumun nem içeriğini artırarak, cildin nem tutma kapasitesini artırır ve cildin daha yumuşak olmasını sağlar (Yang ve ark., 2021). Bu polisakkaritler, topikal kozmetik ürünlerde kullanıldığında hyaluronik asit gibi güçlü nemlendiriciler olarak görev yapar. Mantar hücre duvarında, glukanlar sıklıkla kitine bağlanarak kitin-glukanları üretir. Bu ko-polimer, özellikle nemlendirici özelliklerinden dolayı yaşlanma karşıtı losyonlarda kullanılır (Gautier ve ark. 2008).

Mantarlar, kuruluk ve pul pul dökülme durumunda cildi nemlendirmek ve pürüzsüzleştirmek için kullanılan laktik asit ve seramidlerin üretimi için iyi bir biyofabrikadır. *Tremella fuciformis*'ten elde edilen ekstraktlar, nörodermatit ve sklerodermatite karşı kullanılan harici ürünlerin yapısında sıklıkla kullanılır (Wu ve ark. 2019).

Fenoller

Besin takviyeleri ve yaşlanma karşıtı kremler veya losyonların üretimi için aday olarak çeşitli mantar türlerinden fenolik bileşiklerin miktarını belirlemeye ve hatta saflaştırmaya yönelik birçok çalışma yapılmıştır.

Mantar özleri içeren kozmetik formülasyonlar, cilde zarar veren bakteri türlerinin büyümesini baskılar. Mantarlardan elde edilen baz kremdeki bazı biyoaktif bileşikler güzelliği arttırır ve iltihaplanma, cilt yaşlanması ve hiper-pigmentasyona karşı önemli bir rol oynar (Taofiq ve ark., 2016a)

Kafeik, ferulik, gallik, p-hidroksibenzoik, homojentisik, protokatekuik asit ve mirisetin, mantarlarda bulunan, melanin içeriğini azaltarak ve tirozinaz aktivitesini inhibe ederek cildin parlaklaşmasına yardımcı olduğu ve antoksidant potansiyele sahip olduğu bildirilen ticari fenolik bileşiklerdir (Abdelshafy, ve ark., 2022). Bu bileşikler ve bunların türevlerinin, *P. ostreatus, Pleurotus eryngii, A. bisporus* gibi bir çok mantar türünün içeriğinde yer aldığı rapor edilmiştir (Liu ve ark., 2013; Gąsecka ve ark., 2016). *A. bisporus* ekstraktından hazırlanan baz kremler, bazı bakteri türlerine karşı önemli bir antioksidan potansiyel ve antibakteriyel etki göstermiştir (Taofiq ve ark., 2016b). *Fomitopsis officinalis* mantarından elde edilen kumarinler, Herpes virüsünün kutanöz semptomlarına karşı bile uygulanabilen antimikrobiyal etkileri gösterirler (Girometta 2019).

Terpenoidler

Birçok terpenoid, kozmetik ve beslenme ürünlerinde önemli rol oynayan vitaminler, hormonlar ve ilaçları içeren biyolojik olarak aktif maddelerdir. Mantarlar birçok kozmetik amaç için topikal olarak uygulanabilen önemli bir terpenoid kaynağıdır.

Karotenoidler, serbest radikal temizleme ve UV koruma aktivitelerine sahip en yaygın tetraterpenoidlerdir. Karotenoidler uzun süredir cilt sağlığı ve kozmetik ürünlerin (Vahlquist ve Duvic 2007; Lademann ve ark., 2011), ve nutrikozmetik ve kozmesötik ürünlerin (Anunciato ve da Rocha Filho 2012) temel dayanak noktası olmuştur. Karotenoidler mantarlarda yaygındır. Yenilebilir mantar *Cantharellus cinnabarinus*'ta bulunan kırmızımsı-turuncu bir keto-karotenoid pigment olan Canthaxanthin, güçlü bir yaşlanma karşıtı gençleştiricidir ve cildi güçlendirir (Gharibzahedi ve ark., 2013). Sağlık ve renkle ilgili özellikleri nedeniyle karotenoidler geleneksel olarak doğal kozmetik bileşenler, gıda renklendiriciler, nutrasötikler ve hayvan yemi takviyeleri olarak kullanılır (Sujarid ve ark., 2021).

Mantar hücre zarındaki temel sterol olan ergosterol, kozmesötik bir bileşen olarak kullanılan lanostan tipi bir triterpenoiddir (Taofiq ve ark. 2019).

Makromantarların çoğunun neredeyse yalnızca β-karoten ürettiği görülmektedir (Ribeiro ve ark., 2011), oysa karotenoid sentezi mantar elisitörleri tarafından daha da artırılabilen *Cordyceps militaris* tarafından daha çeşitli bir spektrum sağlanmaktadır (Ribeiro ve ark. 2011; Tang ve ark., 2019).

DRAFT

Protein ve Aminoasitler

Kozmetikler, cilt kırışıklıklarına ve fotoyaşlanma dahil diğer yaşlanma semptomlarına karşı topikal kullanım için L-ergothioneine'i başarıyla denenmiştir. L-Ergotionin aslında cilt hücrelerini UV kaynaklı hasara karşı korur (Bazela ve ark. 2014) ve cilt hücreleri ve dokusu onu bir koruyucu madde olarak kullanabildiği için UVB'nin keratinositlere verdiği zararın neden olduğu fibroblastların yaşlanmasını hafifletir (Ko ve ark. 2021). antioksidan savunma sistemlerinin ayrılmaz bir bileşenidir (Markova ve ark. 2009). Daha yakın zamanlarda *Boletus edulis* Bull. (porcino mantarı) bu alanda en dikkate değer tür olarak vurgulanmıştır (Kalaras ve ark. 2017).

Kollajen ve elastin cilt sağlığı için gerekli olan ancak sağlıklı bir cilt matrisi için yeterli olmayan yapısal proteinlerdir. Cildin ayrıca dermal yenilenmeyi, çoğalmayı ve göçü desteklemek için uygun bileşenlere ihtiyacı vardır (Ito, 2014). Hyaluronik asit (HA), cilt gençleştirici olarak önemli bir rol oynayan, nemi tutan, viskoziteyi artıran ve hücre dışı sıvının geçirgenliğini azaltan, doğal olarak oluşan glikoz bazlı bir polimerdir (Saranraj ve Naidu, 2013). HA hem prokaryotik hem de ökaryotik hücrelerde eşit olarak dağılmıştır. İnsanlarda en çok deride bulunur, bunu göz vitreusu, göbek kordonu, sinoviyal sıvı, iskelet dokuları, kalp kapakçıkları, akciğer, aort ve penisin ereksiyon dokuları takip eder (Papakonstantinou ve ark., 2012).

Vitaminler ve mineraller

Mantarlar, B_1 vitamini (tiamin), B_2 vitamini (riboflavin), B_3 vitamini (niasin), B_7 vitamini (biyotin), B_9 vitamini (folik asit veya folat), B_{12} vitamini (kobalaminler) C (askorbik asit) ve D vitamini (kalsiferoller) de dahil olmak üzere birçok vitamin türünü içerir (Çağlarırmak ve ark., 2002). *A. bisporus, L. edodes* ve *P. ostreatus,* cildin bağışıklık sisteminin desteklenmesinde önemli bir rol oynayan D_2 vitamini (ergokalsiferol) açısından zengindir (Cardwell ve ark., 2018). Mantarlar güneş ışığına maruz kaldıklarında yüzeylerindeki ergosterolü D_2 vitaminine dönüştürebilmektedirler. D vitamini sağlıklı dişler ve kemiklerle ilişkilidir ve nem içeriğini ve cildin nemlenmesini etkili bir şekilde artırır (Moyad, 2009). B vitaminleri tüm mantarlarda bulunur ve nemlendirici ve yaşlanma karşıtı yararları nedeniyle en yaygın olarak cilt, saç ve tırnaklara yönelik güzellik ürünlerinde kullanılır. Siyah trompet (*Craterellus cornucopioides*), altın Chanterelle (*Cantharellus cibarius*) (Watanabe ve ark., 2012) ve *L. edodes* (Watanabe ve ark., 2014) şapkalarında yüksek düzeyde B_{12} vitamini tespit edilmiştir. *Coprinus comatus*'un tokoferoller (E vitamini) açısından zengin yenilebilir bir mantar olduğu, *Clitocybe odora*'nın ise askorbik asit (C vitamini) açısından zengin olduğu rapor edilmiştir (Vaz ve ark., 2011).

Hem üretimi yapılan mantar türleri hem de doğadan toplanan türler selenyum bakımından zengindir (Falandysz, 2008). Selenyum, birincil insan keratinositlerini UV radyasyonunun neden olduğu apoptozdan korur, dolayısıyla cilt, saç ve tırnak sağlığıyla ilişkilidir. Şampuanda bir bileşen olarak yaygın şekilde uygulanır (Rafferty ve ark. 2003).

MANTAR TÜRLERİNİN KOZMETİK ÖZELLİKLERİ

Mantar bazlı ürünlere olan ilgi, mevcut başarılı uygulama örneklerinin ışığında çok büyük ve henüz yeterince araştırılmamış bir potansiyele sahip olduğu varsayıldığından ilgi artmaktadır (Taofiq ve ark. 2016).

Lentinula edodes

L. edodes kayda değer bir antioksidan güç gösterir, yani serbest radikalleri temizler (Nam ve ark. 2021). Bu, bitkilerde de olduğu gibi, zengin fenolik içerikleriyle ilgilidir (Zhu ve ark., 2023). *L. edodes* antioksidan özellikleri sayesinde cilt beyazlatma ve canlanmayı iyileştirmek için kullanılır. Bu mantarlardan elde edilen ticari ürünler arasında sıkılaştırıcı krem, serum ve yüz kremi bulunmaktadır (Hyde ve ark., 2010).

Ayrıca *L. edodes*'in, oksidatif strese karşı yanıtta rol oynayan süperoksit dismutazları (SOD'ler) ve glutatyon peroksidazı (GPx) indüklediği rapor edilmiştir (Cheung ve ark. 2003).

L-ergotionin amino asidi bir histidin türevidir; mitokondriyal membranın oksidasyonunu azalttığı ve mitokondriyal fonksiyonları koruduğuna dair kanıtlar nedeniyle popülerlik kazanmaktadır (Paul ve Snyder

2010). Bugüne kadar *L. edodes, P. ostreatus, P. eryngii, G. frondosa* ve *A. bisporus* dahil olmak üzere birçok mantar türünde yüksek konsantrasyonlarda L-ergothioneine keşfedilmiştir (Dubost ve ark. 2006).

Ganoderma lucidum

Yaşlanma karşıtı aktiviteyi destekleyen önde gelen doğal içerik reishi mantarı (*G. lucidum*) ekstraktlarıdır. G. *lucidum* özleri, krem, serum ve misel temizleyici içeren yaşlanma karşıtı cilt ürünlerinde temel bir bileşen olarak yaygın şekilde kullanılmaktadır. Son farmakolojik çalışmalar, *G. lucidum* özlerinin antioksidan, antimikrobiyal, anti-tirozinaz ve antiinflamatuar özellikler için potansiyel gösterdiğini bildirmiştir (Chien ve ark., 2008; Joseph ve ark., 2009; Cör ve ark., 2018). Anti-aging, *G. lucidum*'un çok sayıda özelliğinin bir sonucu olarak kabul edilen ana etkisidir; bu türün hem topikal hem de oral kullanıma özellikle uygun olmasının nedeni budur (Wang ve ark. 2017). Diğer basidiomisetlerle karşılaştırıldığında, *G. lucidum* daha yüksek tirozinaz inhibisyonu göstermiştir ve şu anda birçok yüz maskesi kozmetiğinde bir bileşen olarak kullanılmaktadır (Chien ve ark. 2008).

Japon şirketi Menard, 1980'li yıllarda *G. lucidum*'u cildin yaşlanmasını önleyici ürünlerde kullanmıştır. Bu tür aynı zamanda batı pazarlarında da yaklaşık yirmi yıldır popülerlik kazanmış ve kapsül, ekstrakt ve yağ olarak kullanılmaktadır (Taofiq ve ark. 2016a, b).

G. lucidum, cilt bakım ürünlerinin yanı sıra, saçı nemlendirmek, saç kaybını önlemek ve güneşin zararlarına karşı koruma sağlamak için saç şekillendirici krem ve şampuanda da saç uyarıcısı olarak kullanılabilir (Hyde ve ark., 2010; Lourith ve ark., 2013; Wu ve ark., 2016)

Tremella fuciformis

Tremella fuciformis, cilt özü ve temizleme jeli gibi ticari ürünlerde cilt elastikiyetini arttırmak ve cilt sıkılığını arttırmak için kullanılır. *T. fuciformis*'in, antioksidan (Li ve ark., 2014), anti-elastisite ve anti-inflamatuar aktiviteye sahip polisakkaritler gibi biyoaktif bileşenler içerdiği rapor edilmiştir (Jo ve ark., 2021). *T. fuciformis* şapka ekstraktlarının, cilt dokusunu ve esnekliğini artırarak kırışıklık önleyici etkiler sergiler (Lai ve ark., 2010). *T. fuciformis* ekstraktından elde edilen suda çözünür bir polisakarit, melanin oluşumunu engelleyebilir ve böylece topikal uygulamayla melanin bazlı cilt lekelerinin temizlenmesine katkıda bulunabilir (Zhang ve ark. 2013). Ayrıca *T. fuciformis* polisakkariti içeren kozmetiklerin hidrasyon etkisinin hyaluronik asit içeren kozmetiklere göre daha yüksek olduğu ve *T. fuciformis* polisakkaritlerinin maliyetinin hyaluronik asite göre çok daha düşük olduğu bildirilmiştir (González-Laredo ve ark., 2021). *T. fuciformis*'den izole edilen bir polisakarit türevinin nem tutma özelliğinin çok yüksek olduğu ayrıca başka bir çalışmada da rapor edilmiştir (Wang ve ark. 2015b).

Cordyceps spp.

Cordyceps türleri, yaşlanma karşıtı özelliğe sahip benzersiz bir biyoaktif kordisepin kaynağıdır ve kozmesötik için potansiyel bir bileşeni temsil eder (Zhou ve ark. 2021). *Cordyceps sinensis*'in yaşlanma karşıtı etkisi özellikle ilgi çekicidir (Ji ve ark., 2009).

Tang ve ark. (2019) yenilebilir mantar *Cordyceps militaris*'ten izole edilen yeni bir doğal pigment olan kordisepini bildirmişlerdir. Bu pigment sarı renklidir ve karotenoid olarak nitelendirilmiştir. Cordycepene ayrıca yeni bir doğal kozmetik renk olma potansiyeline sahip antioksidan aktivite gösterir.

Diğer bir *Cordyceps* türü olan *Cordyceps bassiana*'nın ise deri kaynaklı dermatit üzerinde antiinflamatuar potansiyele sahip olduğu bildirilmiştir (Wu ve ark., 2011)

Grifolia frondosa

Grifola frondosa mantarından izole edilen bir polisakkaritin, NC/Nga farelerinde 2,4-dinitroklorobenzen kaynaklı atopik dermatit benzeri cilt lezyonunu önemli ölçüde baskıladığı bulunmuş ve antiinflamatuar bir ilaç olan deksametazon ile birlikte uygulandığında serum IgE ve sitokin ekspresyonunun azalmasına bağlı AD benzeri cilt lezyonunda sinerjistik bir etki gözlendiği rapor edilmiştir (Park ark ve ark., 2015),

Kim ve ark. (2007), *G. fondosa*'nın misel ekstraktının ve ayrıca bundan izole edilen bir ekzopolisakaritin (EPS) antioksidan, anti-tirozinaz ve anti-kollajenaz aktivitesini araştırdı. Elde edilen sonuçlar, izole edilmiş polisakkaritlerin yanı sıra mantarların da cilt yaşlanmasını ve hiperpigmentasyonu azaltan potansiyel bileşenler olduğunu göstermiştir.

Pleurotus spp.

Pleurotus türleri dünyada en çok yetiştirilen yenilebilir mantarlar arasındadır. Yüksek besin değeri sunarlar ve antiinflamatuar etki, antioksidan, immünomodülatör ve antimikrobiyal aktivite gibi tıbbi özellikler gösterdikleri rapor edilmiştir (Patel, 2012). Bu tıbbi özellikler bilim adamlarını olası kozmetik içeriklerin geleceği konusunda iyimser olmaya yöneltmiştir.

Meng ve ark.. (2011), *Pleurotus citrinopileatus* mantarının şapkalarından elde edilen ekstraktların, tirozinaz inhibisyon potansiyellerinin çok yüksek olduğunu ve melanin üretiminin önemli oranda inhibe ettttiğini rapor etmişlerdir. Anti-tirozinaz aktivitesi rapor edilen diğer Pleurotus türleri *Pleurotus eryngii* (Alam ve ark., 2011a); *Pleurotus nebrodensis* (Alam ve ark., 2011b); *Pleurotus ostreatus* (Banlangsawan ve ark.,2019)'tur.

P. eryngii mantarının etanol ekstraktları deri kaynaklı dermatit üzerinde antiinflamatuar potansiyele sahiptir (Choi ve ark., 2013). *P. citrinopileatus* mantarının, anti-atopik dermatit ve antioksidan aktivitesi birçok çalışmada rapor edilmiştir (Meng ve ark., 2011). *P. citrinopileatus*'un anti-hiyalüronidaz aktivitelsine sahip olması onu kozmetik ürünler için önemli bir malzeme haline getirmektedir (Abd Razak ve ark., 2020).

Agaricus bisporus

Tirozinaz inhibitörlerinin cilt beyazlatıcı ve önleyici etkileri nedeniyle kozmetik endüstrisinde kullanımı giderek önem kazanmaktadır. *A. bisporus* mantarında tirozinaz inhibisyonu rapor edilmiştir (Parvez ve ark., 2007; Pillaiyar ve ark., 2017). Mantar özlerinden hazırlanan baz kozmetik krem, tirozinaz aktivitesini baskılayarak nitrik oksit ve melanin üretimine karşı antiinflamatuar ve antioksidan potansiyel sergilemiştir (Taofiq ve ark., 2016b).

Mantar özleri içeren kozmetik formülasyonlar, cilde zarar veren bakteri türlerinin büyümesini baskılar. *A. bisporus*'tan elde edilen ekstraktlar, metisiline dirençli ve metisiline duyarlı *Staphylococcus aureus* dahil olmak üzere zararlı mikroorganizmalara karşı antibakteriyel aktiviteye sahiptir (Taofiq ve ark., 2016b). Bu bakterilerin yaralanma ve iltihaplanma sırasında deride kolonize olduğu bilinmektedir.

A. bisporus, meyve veren gövdelerinde çoğunlukla Se'yi eser mineral olarak biriktiren yenilebilir bir mantardır (Maseko ve ark., 2013). Kepek, yağlı saç, saç dökülmesi gibi saç sorunlarının çözümünde *A. bisporus* özütü kullanılarak şampuanlar formüle edilebilir. Saçların daha iyi büyümesi için saç ve saç derisini temizlemek için de kullanılabilir (Usman ve ark., 2021).

A. bisporus, saçların sağlıklı ve güçlü olmasında önemli rol oynayan, böylece saç dökülmesini ve kepeği önleyen D vitamini ve bakır, demir ve Se gibi çeşitli mineralleri içerir (Wu ve ark., 2016).

MANTAR BAZLI İÇERİKLİ TİCARİ OLARAK SATILAN KOZMETİKLER

Günümüzde, birçok mantar ticari kozmetik ürünlerinde kullanılmaktadır. OriginsTM Mega-Mushroom için Dr. Andrew Weil (https://www.origins.com/dr-weil-mega-mushroom), cilt bakımında mantarlardan yararlanan ilk premium Batılı markalar arasındaydı. 2006 yılında piyasaya sürülen ürün, cildi sakinleştirmek, yumuşatmak ve gözle görülür yaşlanma belirtilerine karşı korumak için kullanılan, cildi rahatlatan bir yüz maskesidir. Formülasyonunda *Hypsizygus ulmarius, Ganoderma lucidum* ve *Cordyceps sinensis* (Berk.) Sacc misel özleri içerir. Yakın zamanda Dr. Weil For Origins ürün kataloğuna eklenen Plantidote Mega-Mantar Vücut Kremi, üç mantar kompleksinin yanı sıra zencefil, zerdeçal ve kutsal fesleğen (*Ocimum sanctum* L.) gibi diğer bileşenleri içeren nemlendirici bir kremdir.

Menard, mantar içerikli bir kozmetik ürün markasıdır (www.menard-cosmetic.com). Toksinleri ortadan kaldırmak ve UV radyasyonuna ve serbest radikallere aşırı maruz kalma sonucu ortaya çıkan cilt hasarını onarmaya yardımcı olmak için Embellir serisindeki *Ganoderma lucidum* ekstraktını kullanmıştır.

Aveeno Positively Ageless, cildin doğal yenilenme süreciyle birlikte çalıştığı klinik olarak kanıtlanmış bir antioksidan olan doğal Shiitake Kompleksi içeren nemlendirici bir göz kremidir (http://www.aveeno.com). Bu formül, göz çevresindeki cildin görünümünü aydınlatmayı ve koyu halkaların görünümünü azaltmayı amaçlamaktadır.

Estée Lauder, yeni bir Re-Nutriv güneş bakım ürününde (www.esteelauder.com), kurt üzümü ve ginsenginin yanı sıra *G. lucidum* ekstraktı kullanmıştır. Bu ürünün, cilt parlaklığını koruduğunu, canlandırdığını ve yaşlılık lekeleri, düzensiz cilt tonu ve diğer kusurların oluşumu da dahil olmak üzere günlük UV radyasyonuna maruz kalmanın yaşlanma etkilerini azaltmaya yardımcı olduğu belirtilmiştir.

Dr. Patricia Wexler, New York City'de Wexler Dermatoloji'nin sahibidir (www.wexlerdermatoloji.com). Instant De-Puff Göz Jeli, Agaricus bisporus L. ve diğer önemli bileşenlerle yeniden formüle edildi. Bu yaşlanma karşıtı ürün, cilt yüzeyini iyileştiren ve kırışıklığın önlenmesi için gerekli olan ciltteki elastin seviyesini artıran bileşenler içerir. Murad's Age Diffusing Serum, hormonal yaşlanmanın belirti ve semptomlarını hedef alan yaşlanma karşıtı bir serumdur ve *L. edodes* içerir. Mushroom Wisdom Inc. Firmesı daha çok mantar içeren besin takviyeleri pazarlamaktadır. Firmanın ayrıca *T. fuciformis* özleri içeren cilt bakımına yönelik Aquamella Krem adlı ürünü de mevcuttur.

Formülasyonlarında mantar özleri ve mantar moleküllerini kullanan diğer kozmetik markaları arasında Bliss (Hut.com Ltd., Cheshire, İngiltere), La Roche (F. Hoffmann-La Roche Ltd., Basel, İsviçre), Nu-Derm (Obagi Medical Products Inc., Irvine, CA, ABD), SensiClear (Mission Scientific Skincare Inc., Gold River, CA, ABD) yer almaktadır (Choi ve Berson 2006).

SONUÇ VE ÖNERİLER

Mantarların muazzam bir biyoaktif molekül kaynağı olduğu iyi bilinmektedir ve günümüzde milyarlarca dolarlık kozmetik pazarında, çeşitli özelliklere sahip farklı mantar türevli kozmetikler ve nutrakozmetikler mevcuttur. Antioksidan, yaşlanma karşıtı, kırışıklık önleyici, temizleyici ve nemlendirici ajanlar olarak kozmetikte başarılı uygulamaları nedeniyle çeşitli mantar ürünleri ve ekstraktlarının patenti alınmıştır.

Mantar ekstraktlarının antioksidan, antiinflamatuar ve antimikrobiyal aktiviteleri birçok makalede rapor edilmiş olsada, bu aktivitelerden sorumlu biyoaktif metabolitler ve bunların etki mekanizmaları ile ilgili daha fazla çalışmanın yapılması gerekmektedir. Diğer taraftan mantarlar çok sayıda biyokimyasal madde içermelerine rağmen uygun olmayan çözücüler veya ekstraksiyon koşulları nedeniyle henüz karakterize edilememiş birçok içerik mevcuttur. Teknoloji ve bilimdeki ilerlemeler yeni biyoaktif maddelerin keşfine olanak sağlayacak, bu da kozmetik ürünlerin gelişimine önemli katkı sağlayacaktır. Ayrıca klinik çalışmalara ağırlık verilmesi, yüksek kaliteli ve kalitede standart sağlanmış ürünlerin üretilmesi bu tür kozmetiklerin sürdürülebilir üretiminde etkili olacaktır.

KAYNAKLAR

Abdelshafy AM, Belwal T, Liang Z, Wang L, Li D, Luo Z, Li L 2022. A comprehensive review on phenolic compounds from edible mushrooms: Occurrence, biological activity, application and future prospective. *Critical Reviews in Food Science and Nutrition, 62*(22): 6204-6224.

Abd Razak, DL, Jamaluddin A, Abd Rashid NY, Sani NA, Abdul Manan M 2020. Assessment of cosmeceutical potentials of selected mushroom fruitbody extracts through evaluation of antioxidant, anti-hyaluronidase and anti-tyrosinase activity. J- Multidisciplinary Scientific Journal *3*(3): 329-342.

Alam N, Yoon KN, Lee KR, Lee JS, Lee TS 2011a. Phenolic compounds concentration and appraisal of antioxidant and antityrosinase activities from the fruiting bodies of Pleurotus eryngii. Advances in Environmental Biology, *5*(6):1104-1113.

Alam N, Yoon KN, Lee TS 2011b. Evaluation of the antioxidant and antityrosinase activities of three extracts from Pleurotus nebrodensis fruiting bodies. African Journal of Biotechnology, 10(15):2978-2986.

Badalyan SM 2014. Potential of mushroom bioactive molecules to develop healthcare biotech products. In *Proceedings of the 8th International Conference on mushroom biology and mushroom products (ICMBMP8)* (pp. 373-378). New Delhi, India: Yugantar Prakashan Pvt. Ltd.

Badalyan SM, Barkhudaryan A, Rapior S 2022. Medicinal macrofungi as cosmeceuticals: A review. International Journal of Medicinal Mushrooms, *24*(4).

Banlangsawan N, Sripanidkulchai B, Sanoamuang N 2016. Investigation of antioxidative, antityrosinase and cytotoxic effects of extract of irradiated oyster mushroom. Songklanakarin Journal of Science and Technology, *38*(1).

Barel AO, Paye M, Maibach HI 2014. Handbook of Cosmetic Science and Technology. CRC press.

Bisen P S, Baghel RK, Sanodiya BS, Thakur GS, Prasad GBKS 2010. Lentinus edodes: a macrofungus with pharmacological activities. Current Medicinal Chemistry, 17(22): 2419-2430.

Cardwell G, Bornman JF, James AP, Black LJ 2018. A review of mushrooms as a potential source of dietary vitamin D. Nutrients, 10(10): 1498.

Çaglarlrmak N, Unal K, Otles S 2002. Nutritional value of edible wild mushrooms collected from the Black Sea region of Turkey. Micologia Aplicada International, 14(1): 1-5.

Cheung LM, Cheung PCK , Ooi VEC 2003. Antioxidant activity and total phenolics of edible mushroom extracts. Food Chemistry 81(2):249-255

Chien CC, Tsai ML, Chen CC, Chang SJ, Tseng CH 2008. Effects on tyrosinase activity by the extracts of Ganoderma lucidum and related mushrooms. Mycopathologia, 166: 117-120.

Choi JH, Kim HG, Jin SW, Han EH, Khanal T, Do MT, Jeong HG 2013. Topical application of Pleurotus eryngii extracts inhibits 2, 4-dinitrochlorobenzene-induced atopic dermatitis in NC/Nga mice by the regulation of Th1/Th2 balance. Food and Chemical Toxicology, 53:38-45.

Cör D, Knez Ž, Knez Hrnčič M 2018. Antitumour, antimicrobial, antioxidant and antiacetylcholinesterase effect of Ganoderma lucidum terpenoids and polysaccharides: A review. Molecules, 23(3): 649.

De Wet H, Nciki S, van Vuuren SF 2013. Medicinal plants used for the treatment of various skin disorders by a rural community in Northern Maputaland, South Africa. Journal of Ethnobiology and Ethnomedicine 9: 1-10

Dubost, N. J., Beelman, R. B., Peterson, D., & Royse, D. J. (2006). Identification and quantification of ergothioneine in cultivated mushrooms by liquid chromatography-mass spectroscopy. International Journal of Medicinal Mushrooms, 8(3).

Epstein SS 2009. *Toxic Beauty: How Cosmetics and Personal-Care Products Endanger Your Health... and What You Can Do About It*. BenBella Books, Inc..

Falandysz J 2008. Selenium in edible mushrooms. *Journal of Environmental Science and Health Part C*, *26*(3), 256-299.

Gąsecka M, Mleczek M, Siwulski M, Niedzielski P 2016. Phenolic composition and antioxidant properties of Pleurotus ostreatus and Pleurotus eryngii enriched with selenium and zinc. European Food Research and Technology, 242: 723-732.

Gautier S, Xhauflaire-Uhoda E, Gonry P, Piérard GE 2008. Chitin–glucan, a natural cell scaffold for skin moisturization and rejuvenation. International Journal of Cosmetic Science, 30(6): 459-469.

Gharibzahedi SMT, Razavi SH, Mousavi SM 2013. Microbial canthaxanthin: perspectives on biochemistry and biotechnological production. Engineering in Life Sciences, *13*(4): 408-417.

Girometta C 2019. Antimicrobial properties of Fomitopsis officinalis in the light of its bioactive metabolites: a review. Mycology, 10(1): 32-39.

González-Laredo RF, Valdez-Villarreal A, Rocha-Guzmán NE, Moreno-Jiménez MR, Gallegos-Infante JA 2021. Tremella fuciformis and Its Polysaccharides as an Alternative Therapy Against Oxidative Stress. Functional Foods and Nutraceuticals for Human Health: Advancements in Natural Wellness and Disease Prevention, 45.

Hyde KD, Bahkali AH, Moslem MA 2010. Fungi—an unusual source for cosmetics. Fungal Diversity, 43: 1-9.

Ji DB, Ye J, Li CL, Wang YH, Zhao J, Cai SQ 2009. Antiaging effect of Cordyceps sinensis extract. Phytotherapy Research: An International Journal Devoted to Pharmacological and Toxicological Evaluation of Natural Product Derivatives, 23(1): 116-122.

Jo MH, Kim B, Ju JH, Heo SY, Ahn KH, Lee HJ, Oh BR 2021. Tremella fuciformis TFCUV5 mycelial culture-derived exopolysaccharide production and its anti-aging effects on skin cells. Biotechnology and Bioprocess Engineering, 26: 738-748.

Joseph S, Sabulal B, George V, Smina TP, Janardhanan KK 2009. Antioxidative and antiinflammatory activities of the chloroform extract of Ganoderma lucidum found in South India. Scientia Pharmaceutica, *77*(1): 111-122.

Keller SH, Le Y, Rodiger C, Hipler UC, Kertscher R, Malarski A, Huntstock LM, Kiehntopf M, Kaatz M, Norgauer J, Jahreis G 2014. Supplementation of a dairy drink enriched with milk phospholipids in patients with atopic dermatitis—A doubleblind, placebo-controlled, randomized, cross-over study. Clinical Nutrition 33: 1010- 1016

Kim SW, Hwang,HJ, Lee BC, Yun JW 2007. Submerged production and characterization of Grifola frondosa polysaccharides–a new application to cosmeceuticals. Food Technology and Biotechnology, 45(3): 295-305.

Lai J, He C, Zhao J, Dong Y 2010. Optimization of extraction technology of polysaccharide from Tremella fuciformis on commercialized basis and its function in skin care cosmetics. China Surfactant Detergent and Cosmetic 40: 259-262.

Li H, Lee HS, Kim SH, MoonB, Lee C 2014. Antioxidant and anti-inflammatory activities of methanol extracts of Tremella fuciformis and its major phenolic acids. Journal of Food Science, *79*(4): C460-C468.

Liu, J., Jia, L., Kan, J., & Jin, C. H. (2013). In vitro and in vivo antioxidant activity of ethanolic extract of white button mushroom (Agaricus bisporus). Food and chemical toxicology, *51*, 310-316.

Lourith N, Kanlayavattanakul M 2013. Hair loss and herbs for treatment. Journal of Cosmetic Dermatology, 12(3):210-222.

Luque de Castro, M. D., 2011. Cosmetobolomics as an incipient -omics with high analytical involvement. Trends in Analytical Chemistry. 30:1365-1371.

Maseko T, Callahan DL, Dunshea FR, Doronila A, Kolev SD, Ng K 2013. Chemical characterisation and speciation of organic selenium in cultivated selenium-enriched Agaricus bisporus. Food chemistry, 141(4): 3681-3687.

Meng TX, Furuta S, Fukamizu S, Yamamoto R, Ishikawa H, Arung ET, Kondo R 2011. Evaluation of biological activities of extracts from the fruiting body of Pleurotus citrinopileatus for skin cosmetics. Journal of Wood Science, 57: 452-458.

Patel Y, Naraian R, Singh VK 2012. Medicinal properties of Pleurotus species (oyster mushroom): a review. *World Journal of Fungal and Plant Biology*, *3*(1), 1-12.Rodrigues, F., Pimentel, F. B., Oliveira, M. B. P. P., 2015. Olive by-products: challenge application in cosmetic industry. Industrial Crops and Products 70:116–124.

Nam M, Choi JY, Kim MS 2021. Metabolic profiles, bioactive compounds, and antioxidant capacity in Lentinula edodes cultivated on log versus sawdust substrates. Biomolecules, 11(11):1654.

Nitthikan N, Leelapornpisid P, Naksuriya O, Intasai N, Kiattisin K 2022. Potential and alternative bioactive compounds from brown Agaricus bisporus mushroom extracts for xerosis treatment. Scientia Pharmaceutica, *90*(4): 59.

Palacios I, Lozano M, Moro C, D'arrigo M, Rostagno MA, Martínez JA, Villares A 2011. Antioxidant properties of phenolic compounds occurring in edible mushrooms. Food Chemistry, 128(3):674-678.

Paul BD, Snyder SH 2010. The unusual amino acid L-ergothioneine is a physiologic cytoprotectant. Cell Death and Differentiation, 17(7), 1134-1140.

Park HS, Hwang YH, Kim MK, Hong GE, Lee HJ, Nagappan A, Kim, GS 2015. Functional polysaccharides from Grifola frondosa aqueous extract inhibit atopic dermatitis-like skin lesions in NC/Nga mice. Bioscience, Biotechnology, and Biochemistry, *79*(1): 147-154.

Parvez S, Kang M, Chung HS, Bae H 2007. Naturally occurring tyrosinase inhibitors: mechanism and applications in skin health, cosmetics and agriculture industries. Phytotherapy Research: An International Journal Devoted to Pharmacological and Toxicological Evaluation of Natural Product Derivatives, 21(9): 805-816.

Pillaiyar T, Manickam M, Namasivayam V 2017. Skin whitening agents: Medicinal chemistry perspective of tyrosinase inhibitors. Journal of Enzyme Inhibition and Medicinal Chemistry, *32*(1): 403-425.

Rafferty TS, Beckett GJ, Walker C, Bisset YC, McKenzie RC 2003. Selenium protects primary human keratinocytes from apoptosis induced by exposure to ultraviolet radiation. Clinical and Experimental Dermatology, *28*(3): 294-300.

Rosdan Bushra SM, Nurul AA 2022. Bioactive mushroom polysaccharides: The structure, characterization and biological functions. Journal of Liquid Chromatography and Related Technologies, *45*(13-16): 174-190.

Sujarit K, Suwannarach N, Kumla J, Lomthong T 2021. Mushrooms: Splendid gifts for the cosmetic industry. Chiang Mai Journal of Science 48: 699-725.

Tang H, Chen C, Zou Y, Lou H, Zheng Q, Guo L, Yun F 2019. Purification and structural characterization of a novel natural pigment: Cordycepene from edible and medicinal mushroom Cordyceps militaris. Applied Microbiology and Biotechnology, 103: 7943-7952.

Taofiq O, Barreiro MF, Ferreira IC 2020. The role of bioactive compounds and other metabolites from mushrooms against skin disorders-A systematic review assessing their cosmeceutical and nutricosmetic outcomes. Current Medicinal Chemistry, 27(41): 6926-6965.

Taofiq O, Rodrigues F, Barros L, Barreiro MF, Ferreira IC, Oliveira MBP 2019. Mushroom ethanolic extracts as cosmeceuticals ingredients: Safety and ex vivo skin permeation studies. Food and Chemical Toxicology, 127: 228-236.

Taofiq O, González-Paramás AM, Martins A, Barreiro MF, Ferreira IC 2016. Mushrooms extracts and compounds in cosmetics, cosmeceuticals and nutricosmetics—A review. Industrial Crops and Products, 90: 38-48.

Taofiq O, Heleno SA, Calhelha RC, Alves MJ, Barros L, Barreiro MF, Gonzalez-Paramas AM, Ferreira IC 2016. Development of mushroom-based cosmeceutical formulations with anti-inflammatory, anti-tyrosinase, antioxidant, and antibacterial properties. Molecules *21:* 1372.

Thi Nhu Ngoc L, Oh YK, Lee YJ, Lee YC 2018. Effects of Sparassis crispa in medical therapeutics: a systematic review and meta-analysis of randomized controlled trials. International Journal of Molecular Sciences. 19(5): 1487.

Usman, M., Murtaza, G., & Ditta, A. (2021). Nutritional, medicinal, and cosmetic value of bioactive compounds in button mushroom (Agaricus bisporus): a review. Applied Sciences, 11(13): 5943.

Vaz JA, Barros L, Martins A, Santos-Buelga C, Vasconcelos MH, Ferreira IC 2011. Chemical composition of wild edible mushrooms and antioxidant properties of their water soluble polysaccharidic and ethanolic fractions. Food Chemistry, *126*(2): 610-616.

Wang HMD, Chen CC, Huynh P, Chang JS 2015. Exploring the potential of using algae in cosmetics. Bioresource Technology, 184: 355-362.

Wang X, Zhang Z, Zhao M 2015. Carboxymethylation of polysaccharides from Tremella fuciformis for antioxidant and moisture-preserving activities. International Journal of Biological Macromolecules, *72*: 526-530.

Wang J, Cao B, Zhao H, Feng J 2017. Emerging roles of Ganoderma lucidum in anti-aging. Aging and Disease, *8*(6): 691.

Watanabe F, Schwarz J, TAkenaka S, Miyamoto E, Ohishi N, Nelle E, Yabuta Y 2012. Characterization of vitamin B12 compounds in the wild edible mushrooms black trumpet (Craterellus cornucopioides) and golden chanterelle (Cantharellus cibarius). *Journal of Nutritional Science and Vitaminology*, *58*(6):438-441.

Watanabe F, Yabuta Y, Bito T, Teng F 2014. Vitamin B12-containing plant food sources for vegetarians. Nutrients, *6(5)*: *1861*-1873.

Wu Y, Choi M H, Li J, Yang H, Shin HJ 2016. Mushroom cosmetics: the present and future. Cosmetics, 3(3): 22.

Wu YJ, Wei ZX, Zhang FM, Linhardt RJ, Sun PL, Zhang AQ 2019. Structure, bioactivities and applications of the polysaccharides from Tremella fuciformis mushroom: A review. International Journal of Biological Macromolecules, *121*: 1005-1010.

Xu H, Liu JH, Shen ZY, Fei Y, Chen XD 2010. Analysis of chemical composition, structure of Grifola frondosa polysaccharides and its effect on skin TNF-α levels, lgG content, T lymphocytes rate and caspase-3 mRNA. Carbohydrate Polymers, *82*(3): 687-691.

Yang M, Zhang Z, He Y, Li C, Wang J, Ma X 2021. Study on the structure characterization and moisturizing effect of Tremella polysaccharide fermented from GCMCC5. 39. *Food Science and Human Wellness*, *10*(4), 471-479.

Zhang K, Meng XY, Sun Y 2013. Preparation of *Tremella*, *Speranskiae tuberculatae* and *Eriocaulon buergerianum* extracts and their performance in cosmetics. Detergent Cosmetics 36:28–32

Zhao S, Gao Q, Rong C, Wang S, Zhao Z, Liu Y, Xu J 2020. Immunomodulatory effects of edible and medicinal mushrooms and their bioactive immunoregulatory products. Journal of Fungi, *6*(4):269.

Zhu, Y, Yu X, Ge Q, Li J, Wang D, Wei Y, Ouyang Z 2020. Antioxidant and anti-aging activities of polysaccharides from Cordyceps cicadae. International Journal of Biological Macromolecules, 157: 394-400.

Zhu H, Chen Z, Li G, Yao X, Hu Y, Zhao W 2023. Physicochemical, sensory, and antioxidant characteristics of stirred-type yogurt enriched with Lentinula edodes stipe powder. Food Science and Nutrition. https://doi.org/10.1002/fsn3.3563

Zi Y, Zhang B, Jiang B, Yang X, Liang Z, Liu W, Liu, L 2018. Antioxidant action and protective and reparative effects of lentinan on oxidative damage in HaCaT cells. Journal of Cosmetic Dermatology, 17(6): 1108-1114.

DRAFT

ORAL PRESENTATION

Hypsizygus ulmarius mantarı üretiminde yerfıstığı kabuklarının substrat olarak kullanımının mantar verimi ve şapka boyutlarına etkisi

Burak SALMANOĞLU[1*] (*ORCID: https://orcid.org/0009-0005-2729-244X*), Funda ATİLA[2*] (*ORCID: https://orcid.org/ 0000-0003-1129-1045*)

[1] Kırşehir Ahi Evran Üniversitesi, Fen Bilimleri Enstitüsü, Bahçe Bitkileri Anabilimdalı, Kırşehir, Türkiye

[*2]Ege Üniversitesi, Bergama Meslek Yüksekokulu, Mantarcılık Programı, İzmir, Türkiye

*Sorumlu yazar e-mail:funda.atila@ege.edu.tr

Özet

Çalışmada, *Hypsizygus ulmarius* mantarı üretiminde yetiştirme ortamı olarak yerfıstığı kabuğu kullanımının mantarın üretim döngüsü, verim ve şapka boyutları üzerindeki etkisini karşılaştırmak için yerfıstığı kabuğu (FK) ve buğday samanının (S) farklı oranlarda karıştırılması ile hazırlanan beş farklı yetiştirme ortamı kullanılmıştır. En hızlı misel gelişimi (20 gün), taslak oluşumu (29.6 gün) ve hasat aşaması (37 gün) tamamen yerfıstığı kabuğu ile hazırlanan FK100 ortamında gerçekleşmiştir. Misel gelişim süresi ile N (r^2= - 0.953) ve C:N oranları (r^2= - 0.955) ile negatif, hemiselüloz içeriği (r^2=0.939) ile pozitif bir korelasyon olduğu belirlenmiştir. Farklı yetiştirme ortamlarında üretilen *H. ulmarius* için verim 207.45 g/kg -266.98 g/kg ve BE %59.27-%78.52 arasında değişmiştir. En yüksek verim S25:FK75 ortamında elde edilmiştir. Genel olarak, samana eklenen fıstık kabuğu miktarı arttıkça BE (%) yükselirken, yerfıstığı kabuğunun tek başına kullanıldığı yetiştirme ortamı FK100'de en düşük BE (%) elde edilmiştir. Mantarların şapka çapları, 78.1 mm-98.3 mm, sap uzunluğu 45.8 mm-87.0 mm, sap çapları ise 17.5 mm-23.5 mm arasında değişmiştir. En büyük şapka çapı ve sap uzunluğuna sahip mantarlar, en yüksek veriminde elde edildiği S25:FK75 ortamından hasat edilmiştir. En kalın saplara sahip olan şapkaların ise kontrol ortamında geliştiği gözlenmiştir. Çalışmada elde edilen veriler yerfıstığı kabuğunun ticari olarak *H. ulmarius* yetiştiriciliği için umut verici bir alternatif substrat olduğunu göstermektedir.

Anahtar Kelimeler: karaağaç istiridyesi mantarı, lignoselülozik materyaller, tarımsal atık

Effect of using groundnut shells as substrate on mushroom yield and fruitbody size in cultivation of *Hypsizygus ulmarius* mushroom

Abstract

In this study, five different growing media prepared by mixing different proportions of groundnut shell (FK) and wheat straw (S) were used to compare the effect of using groundnut shell as substrate on the cultivation cycle, yield and fruitbody size of *Hypsizygus ulmarius* mushroom. The fastest mycelial growth (20 days), pinhead formation (29.6 days) and time to first harvest (37 days) occurred on FK100 medium prepared entirely with groundnut shells. There was a negative correlation with N (r^2= - 0.953) and C:N ratios (r^2= - 0.955) and a positive correlation with hemicellulose content (r^2=0.939). The yields ranged between 207.45 g/kg-266.98 g/kg and BE between 59.27% and 78.52% For H. ulmarius produced in different growing media, The highest yield was obtained in S25:FK75 medium. In general, BE (%) increased as the amount of groundnut shell added to the straw increased, while the lowest BE (%) was obtained in the growing medium FK100 where groundnut shell was used alone. The fruitbody diameters of the mushrooms varied between 78.1 mm-98.3 mm, stem length 45.8 mm-87.0 mm and stem diameters 17.5 mm-23.5 mm. The mushrooms with the largest fruitbody diameter and stem length were harvested from the S25:FK75 medium, where the highest yield was obtained. It was observed that the fruitbody with the thickest stems developed in the control medium. The data obtained in this study indicate that groundnut shell is a promising alternative substrate for commercial cultivation of *H. ulmarius*.

DRAFT

Key words: elm oyster, lignocellulosic materials, agricultural waste

GİRİŞ

Dünya nüfusunun sürekli artması, doğal kaynakların bozulması ve iklim değişikliğinin etkileri nedeniyle insanoğlunun hâlâ karşı karşıya olduğu ve karşılaşmaya devam edeceği en büyük sorunlar gıda üretiminin azalması, insan sağlığının bozulması ve çevre kirliliğidir (Oseni ve ark. 2012).

Mantar yetiştiriciliği tüm bu sorunların çözümüne katkı sağlayabilecek bazı özelliklere sahiptir. Mantar üretimi, güçlü bir enzim sistemine sahip mantarların lignoselülozik materyali parçalama yeteneklerini kullanarak düşük değerli ve kullanım alanları sınırlı olan tarım ve orman atıklarından besin değeri yüksek ve tıbbi özelliklere sahip gıda üretimidir. Mantar yetiştiriciliği lignoselülozik atıkları gıda maddesine dönüştürmekle kalmaz, aynı zamanda sağlıklı ve besleyici bir ürün sunar. Mantardaki esansiyel amino asitlerin içeriği yüksektir, sindirimi kolaydır ve kolesterol içermez (Gonzalez ve ark., 2020). Yenilebilir mantarlar oldukça besleyicidir ve besin değerleri yumurta, süt ve etle karşılaştırılabilecek düzeydedir (Okoro ve Achuba, 2012). Tıbbi mantarlardan elde edilen ekstrakte edilebilir biyoaktif bileşikler, bağışıklık sistemini güçlendirecek ve yaşam kalitesini artırabilecek özelliklere sahiptirler (Waktola ve Temesgen, 2018). Mantar hasadından arda kalan ve yoğun miktarda mantar miseli ve mantar kalıntıları içeren atık mantar yetiştirme ortamları, aynı zamanda hayvan yemi olarak, toprak verimliliğinin zenginleştirilmesi için biyo-gübre olarak ve biyogaz üretiminde kullanılabilir (Leong ve ark., 2022). Mantar yetiştiriciliği, özellikle gelişmekte olan ülkelerdeki kadınlar ve gençler için gelir ve istihdam yaratılmasına yardımcı olabilir (Thakur, 2014).

Tarımsal atıkların çoğu, kimyasal yapıları ve ayrışma özellikleri nedeniyle işlenmesi ve bertaraf edilmesi genellikle sorunlu olan lignoselülozik bileşikler açısından zengindir. Türkiye önemli bir tarım ülkesidir. Buğday ülkemizin hemen her bölgesinde yaygın olarak üretilen en önemli tarımsal ürünlerden bir tanesidir ve bu üretim sonucunda bol miktarda buğday samanı ortaya çıkar. TÜİK (2022) verilerine göre 2022 yılında ülkemizde 19.8 milyon tonluk buğday üretimi gerçekleşmiş olup ortaya çıkan saman miktarı yaklaşık 25 milyon tondur. Yerfıstığı ise ülkemizde en fazla Akdeniz bölgesinde üretilmekle birlikte Ege, Marmara ve Güneydoğu Anadolu Bölgelerinde de yerfıstığı plantasyonlarına rastlanmaktadır (Şahin, 2014). Yerfıstığı dünyada yağlı tohumlar kategorisinden önemli bir yağ bitkisi olmasına karşın ülkemizde elde edilen mahsulün tamama yakını çerez olarak tüketilir ve buğday da olduğu gibi bu ürünün işlenmesi sonucunda da büyük miktarda yer fıstığı kabuğu ortaya çıkar.

Bazı araştırmacılar, yerfıstığı kabuklarının mahsul gübrelemesi için bir karbon kaynağı olarak, kirli sudaki süzülmesinde bir substrat olarak, kompozit materyali olarak ve biyoetanol üretiminde kullanımını araştırmışlardır (Çelik ve Gürdal, 2010; Taşar ve ark., 2014; Li ve ark., 2018; Ganguly ve ark., 2020). Yerfıstığı kabukları gibi lignoselülozik atıklar her ne bazı amaçlarla kullanılsa da büyük oranda yakılarak, toprağa gömülerek ya da farklı şekillerde bertaraf edilmektedirler. Kullanım alanları sınırlı olan bu atıkları yüksek pazar değeri olan gıdalara dönüştürmek ve mantar yetiştiricisi için ucuz bir substrat kaynağı olarak kullanmak bu atıkların ortadan kaldırılmasında çözüm yöntemlerinden biri olabilir (Sa'nchez, 2010).

Mantar üretimi ve tüketiminden elde edilen potansiyel faydalara rağmen Türkiye, mantar üretimi ve ticaretinde son derece geridedir. 2021 yılı Dünya mantar üretimi 44.207.117 ton olarak kaydedilmiştir (FAOSTAT, 2023) ve aynı dönem için Türkiye mantar üretimi ise 61.460 ton'dur (FAOSTAT, 2023). Zengin bir tarımsal çeşitliliğe sahip olan ve her yıl milyonlarca ton tarımsal atığın ortaya çıktığı Türkiye'nin dünya mantar üretimine katkısı %0.13 civarındadır. Ancak yine de 2000 yılında 7000 ton olan mantar üretimimizin 21 yılda gösterdiği artış oldukça umut verici boyutlardadır. Türkiye'de *Agaricus bisporus* türü hala en fazla üretilen mantar türü olduğu halde son yıllarda *Pleurus ostreatus* ve *Lentinula edodes* gibi türlerinde üretiminin yaygınlaştığı gözlenmektedir (Eren ve Pekşen, 2019). Ülkemizde son yıllarda mantar üretimi ve tüketimine artan ilgi, *Ganoderma lucidum, Hericium erinaceus, Hypsizygus ulmarius* gibi dünya'da ticari değere sahip egzotik mantar türleri için bir pazarın bulunduğunu göstermektedir.

H. ulmarius, düşük maliyetli üretim teknolojisi ve daha yüksek biyolojik verimliliği sebebi ile özellikle Asya ve Avrupa'da ticari olarak popüler bir türdür (Kumar ve ark., 2019). Bu mantar büyük ve açık beyaz renkli şapkaya, mükemmel bir tada ve çekici bir şekle sahiptir (Baghel ve ark., 2019). *H. ulmarius* görünüş olarak istiridye mantarına benzer, ancak yetiştirme döngüsü ve biyolojik verimlilik açısından farklılık gösterir. *H. ulmarus* mantarı protein (%13.5-18.7), yağ (%2.6-4.3), kül (%7.6-10.3), karbonhidrat (%69.6-73.5) açısından yüksek besin değerlerine sahip olup, enerji değeri ise 372.2–391.1 kcal/100 g km'dir (Atila, 2023). Bu tür aynı

zamanda niasin, riboflavin, D vitamini, C, B1, B5 ve B6 gibi vitaminler açısından da zengindir (Munna ve ark., 2018).

H. ulmarius mantarı talaş, buğday samanı, fasulye samanı, mısır silajı, atık mantar kompostu vb. dahil olmak üzere çeşitli substratlarda yetiştirilebilir (Ozturk ve Atila, 2019). Saman gibi substratların azot içeriği yüksek bir materyal ile desteklenmesinin verimi artırdığı rapor edilmiştir (Malayil ve ark., 2017).

Mantar yetiştiriciliği, Akdeniz ülkelerindeki tarımsal faaliyetlerin geniş bir yelpazesindeki ligno-selülozik kalıntıların ve tarımsal faaliyetlerin yan ürünlerinin değerlendirilmesi için mükemmel bir fırsat sunsa da, tarımsal atıkların mantar üretiminde kullanımları söz konusu olduğunda buğday samanı bazlı substratlar hakimdir. Bu nedenle, yerel olarak bol miktarda bulunan tarımsal atıkların alternatif substratlar olarak değerlendirilmesi ihtiyacı ortadadır.

Mevcut çalışma, başta Akdeniz bölgesi olmak üzere, ülkemizin diğer bazı bölgelerinde de bol miktarda bulunması mümkün olan yerfıstığı kabuklarının mantar üretiminde kullanılarak değerlendirilmesine ve ülkemizde henüz ticari olarak üretimi yapılmayan bir tür olan *H. ulmarius* mantarının tanıtılarak Türkiye mantar sektöründeki mantar çeşitliliğinin arttırılmasına katkıda bulunmayı amaçlamaktadır. Bu mantıkla, yerfıstığı kabuklarının *H. ulmarius* mantarı üretiminde substrat olarak kullanımının mantarının büyüme performansı ve verimine etkileri araştırılmıştır. Ayrıca yerfıstığı kabuklarının kullanımının *H. ulmarius* mantarının şapka boyutları üzerindeki etkisi değerlendirilmiştir.

MATERYAL VE METOT

Mantar ırkı

H. ulmarius suşu Hollanda'da faaliyet gösteren Homegreen Spawn Company'den temin edilmiştir. Saf kültürden çoğaltılan misel kültürleri Malt ekstrakt agarda (MAE) muhafaza edilmiş ve kullanılıncaya kadar 4°C'de sbuzdolabında saklanmıştır.

Substratlar

Çalışma, 2022 yılında Kırşehir Ahi Evran Üniversitesi Ziraat Fakültesi Mantar üretim laboratuvarında gerçekleştirilmiştir. Buğday samanı (BS) ve yerfıstığı kabukları (FK) farklı oranlarda karıştırılması ile yetiştirme ortamları hazırlanmıştır (Tablo 1).

Tablo 1. Çalışmada yetiştirme ortamı hazırlığında kullanılan materyaller ve oranları

Kod	Substrat	Oran (%)	Substrat	Oran (%)
Kontrol	Saman	100	-	
S75:FK25	Saman	75	Yerfıstığı Kabuğu	25
S50:FK50	Saman	50	Yerfıstığı Kabuğu	50
S25:FK75	Saman	25	Yerfıstığı Kabuğu	75
FK100	Saman	-	Yerfıstığı Kabuğu	100

Tohumluk misel hazırlığı

Tohumluk miseller buğday tanelerinden hazırlanmıştır. Buğday taneleri 15 dakika boyunca kaynatılmış, daha sonra suyu süzülmüş %2 alçı eklenerek karıştırılmış ve 500 ml'lik kavanozlara doldurularak otoklavda 121°C'de 90 dakika boyunca sterilize edilmiştir. Soğuyan kavanozlar laminar flow (steril kabinde) misel kültürleri ile aşılanmış ve 25°C'de inkübe edilmiştir.

Yetiştirme ortamı hazırlığı

BS ve FK substrat hazırlığı öncesinde ayrı ayrı küvetlerde bir gece suda bekletildi. Substralardaki fazla su drene edildikten sonra farklı oranlarda karıştırılarak Tablo 1'de verilmiş olan 5 farklı yetiştirme ortamı hazırlanmıştır. Yetiştirme ortamı hazırlığında kullanılan substratlar homojen bir şekilde karıştırılarak polipropilen torbalara 1 kg gelecek şekilde doldurulmuştur. Hazırlanan torbalar otoklavda 121°C'de 90 dakika boyunca sterilize edilmiş, soğuduktan sonra laminar flow (steril masa)'da her bir torbaya %3 (w/w) oranında tohumluk misel gelecek şekilde tohumluk misel aşılaması yapılmıştır.

İnkübasyon periyodunda üretim odasının sıcaklığı 25±2°C'ye, oransal nemi %70-80'e ayarlanmıştır. Misel gelişim periyodu tamamlandıktan sonra torbalar, taslak oluşumunun tetiklenmesi için 18±2°C sıcaklık ve %80-90 bağıl nemdeki üretim odasına aktarılmış ve polipropilen torbaların ağız kısmındaki pamuk tıkaçlar

çıkarılmıştır. Üretim odalarında florasan lambalar kullanılarak 12 saat boyunca ışık sağlanmış, havalandırma, CO2 konsantrasyonunu 1000 ppm'in altında tutacak şekilde yapılmıştır. Mantarlar tam olarak olgunlaştıklarında hasat edilerek tartım ve ölçüm işleri tamamlanmıştır.

Deneme, her yetiştirme ortamı için on tekrarlı olarak, tesadüf parselleri tasarımında gerçekleştirilmiştir.

Yetiştirme ortamı analizleri

Sterilizasyondan sonra test edilen substrat numuneleri, 60°C'de sabit bir ağırlığa kadar kurutulmuş, ardından bir değirmen kullanılarak kaba bir toz halinde öğütülmüştür. Substratların kül içeriği standart prosedür (AOAC, 2019) takip edilerek belirlenmiştir. Toplam karbon, Tiquia ve Tam (2000) formülüne göre kül içeriğinden tahmin edilmiş, toplam azot (N), Kjeldahl yöntemiyle belirlenmiş ve yetiştirme ortamlarının C:N oranları hesaplanmıştır. Selüloz, hemiselüloz ve lignin içerikleri Van Soest ve ark. (1991)'in yöntemi kullanılarak ölçülmüştür. Numunelerin selüloz, lignin ve hemiselüloz içerikleri denklemlerde gösterildiği gibi hesaplanmıştır;

(1) Selüloz = ADF-ADL;(2) Lignin = ADL; (3) Hemiselüloz=NDF-ADF (Zadrazil ve Brunnert, 1982).

Değerlendirilen üretim parametreleri

Çalışmada şu parametreler üzerinde gözlemler yapılmıştır: (I) misel gelişiminin tamamlanması için geçen gün sayısı, (II) misel ekiminden sonra primordia oluşumuna kadar geçen gün sayısı,(III) misel ekiminden sonra ilk hasada kadar geçen gün sayısı (IV) verim (g/kg) (V) Biyolojik etkinlik (%) (VI) şapka boyutları (şapka çağı, sap boyu ve sap çapı)

Biyolojik etkinliği hesaplamak için şu formül kullanılmıştır;

BE (%)= (taze mantar ağırlığı/ kuru substrat ağırlığı) X 100

İstatistiksel analiz

Sonuçlar varyans analizi (ANOVA) kullanılarak istatistiksel olarak analiz edildi. Ortalamalar arasındaki bireysel farklılıkları belirlemek için Tukey'in post-hoc karşılaştırması (%5 anlamlılık düzeyinde) uygulandı. Tüm analizler SPSS 16.0 versiyonu kullanılarak yapıldı.

BULGULAR ve TARTIŞMA

Yetiştirme ortamlarının kimyasal özellikleri

Yetiştirme ortamlarının kül, karbon, azot içerikleri, C:N oranları, selüloz, hemizelüloz ve lignin içerikleri bakımından istatistiksel olarak önemli farklar mevcut iken ($p<0.001$), yetiştirme ortamlarının pH değerleri arasında önemli bir fark bulunmamıştır ($p>0.05$).

Yetiştirme ortamı	pH	Kül (%)	C	N (%)	C:N	Selüloz (%)	Lignin (%)	Hemiselüloz (%)
Kontrol	6.93±0.23	7.83±0.21 a	53.46±0.12 d	0.59±0.01 d	90.13±1.69 a	36.66±1.38 b	14.50±0.51 e	23.64±1.31 a
S75:FK25	7.25±0.06	6.74±0.14 b	54.09±0.08 c	0.74±0.03 c	72.92±3.30 b	37.27±0.44 b	16.39±0.44 d	19.44±0.55 b
S50:FK50	7.06±0.18	6.24±0.05 b	54.38±0.03 c	0.78±0.02 bc	70.09±2.23 bc	40.57±0.50 a	20.71±0.45 c	17.29±0.13 c
S25:FK75	7.18±0.21	4.77±0.25 c	55.23±0.15 b	0.84±0.02 ab	66.06±1.65 c	41.33±0.33 a	22.75±0.37 b	14.96±0.19 d
FK100	7.26±0.04	4.13±0.17 d	55.60±0.10 a	0.91±0.02 a	61.36±1.36 c	42.10±0.28 a	25.33±0.73 a	10.17±0.22 e
P değeri	0.307	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

Tablo 2. Çalışmada kullanılan mantar yetiştirme ortamlarının bazı kimyasal özellikleri

Her değer ortalama ± standart sapma olarak ifade edilmiştir (n=3). Sütunlarında yer alan aynı harf ile adlandırılmış değerler arasında Tukey testine göre istatistiksel olarak fark yoktur

DRAFT

Yetiştirme ortamlarının pH değerleri 6.93 ile 7.26 arasında değişmiştir. $CaCO_3$ ya da $CaSO_4$ gibi alkali bir ortamın yetiştirme ortamı hazırlığında kullanılması kullanılması genellikle rakip mikroorganizmaları azaltmak için önerilir (Stölzer ve Grabbe 1991). Ancak bu çalışmada yetiştirme ortamlarının pH değerleri hafif alkali olduğundan dolayı yetiştirme ortamı hazırlığı sırasında $CaCO_3$ veya benzeri bileşik eklenmemiştir.

Çalışmada değerlendirilen yetiştirme ortamları ortalama %4.13-7.83 oranında kül içerirken, substratların C içeriği %53.46 ile %55.60 arasında değişmiştir (Tablo 2). Substratların kimyasal analizi, yetiştirme ortamlarının azot içeriğinin %0.59-%0.91 arasında değiştiğini göstermiştir. FK eklenen yetiştirme ortamlarının azot içeriğinin kontrol ortamına göre yüksektir. Ancak çalışmada incelenen tüm yetiştirme ortamlarının N içeriği Azizi ve ark. (1990) tarafından önerilen aralıkta veya çok yakındırlar. Kontrol ortamının C:N oranı 90.13 ile diğer substratlardan daha yüksekti. Hemiselüloz, selüloz ve lignin oraları da sırasıyla %10.17–23.64, %36.66–42.10 ve %14.50–25.33 gibi geniş bir değişkenlik gösterdi. Lignoselülozik bileşenler açısından kontrol ortamı hemiselüloz için en yüksek değerleri ve lignin için en düşük değerleri sergilerken, FK100 ortamı, selüloz ve lignin için en yüksek değerleri ve hemiselüloz içerikleri için en düşük değerleri sergiledi. Benzer şekilde yetiştirme ortamlarında FK içeriği arttıkça ortamdaki selüloz ve lignin içeriği artarken, hemiselüloz içerikleri ise düşmüştür.

Farklı yetiştirme ortamlarının Hypsizygus ulmarius mantarının üretim periyoduna etkileri

Farklı yetiştirme ortamlarının *H. ulmarius* mantarının misel gelişim süresi, taslak oluşumuna kadar geçen gün sayısı ve ilk hasada kadar geçen gün sayısı üzerindeki etkileri belirlemek üzere yapılan varyans analizi, yetiştirme ortamları arasında önemli farklılıklar olduğunu göstermiştir ($p<0.001$) (Tablo 3).

Yetiştirme ortamı	Misel Gelişim Dönemi (gün)	Taslak oluşumuna kadar geçen süre (gün)	İlk hasada kadar geçen süre (gün)
Kontrol	21,4±0.5 a	34,10±0.7 a	39,80±0.4 b
S75:FK25	20,6±0.5 b	33,90±0.7 a	41,80±0.8 a
S50:FK50	20,6±0.5 b	34,00±0.8 a	40,10±0.7 b
S25:FK75	20,6±0.5 b	33,80±0..8 a	41,70±0.8 a
FK100	20,0±0.0 c	29,60±1.0 b	37,00±0.6 c
P değeri	<0.001	<0.001	<0.001

Tablo 3. Farklı yetiştirme ortamlarının *Hypsizygus ulmarius* mantarının üretim sürecine etkileri

Her değer ortalama ± standart sapma olarak ifade edilmiştir (n=10). Sütunlarında yer alan aynı harf ile adlandırılmış değerler arasında Tukey testine göre istatistiksel olarak fark yoktur

H. ulmarius, incelenen yetiştirme ortamlarında misel gelişim sürecini 20.0–21.4 gün içinde tamamlamıştır. FK100 ortamı test edilen diğer ortamlara göre daha kısa bir misel gelişim süreci sergilemiştir. Yetiştirme sürecinin başlangıcında yüksek kontaminasyon riski bulunduğundan kolonizasyon aşamasının süresinin kısaltılması büyük önem taşımaktadır. Bu çalışmada elde edilen misel gelişim süreleri Munna ve ark. (2018) ve Eswaran ve ark. (2019) 'nın bulguları benzerdi.

Taslaklar, ortalama olarak 29.6 güne ihtiyaç duyan FK100 ortamı hariç, diğer tüm yetiştirme ortamlarında yaklaşık 34 günde meydana geldi. İlk hasada kadar geçen gün sayısı 37 gün ile FK100 ortamında en kısa iken, en uzun süre 41.7 ve 41.8 gün ile sırası ile S75:FK25 ve S25:FK75 ortamlarında belirlenmiştir. Bu çalışmada elde edilen taslak oluşumu için geçen gün sayısı ve ilk hasada kadar geçen gün sayısı ile ilgili veriler, *H. ulmarius* taslaklarının ortaya çıkmasının 21.33-27.33 gün sürdüğünü ve ilk hasatın 24.67-31.33 gün sonra başladığını bildiren Munna ve ark. (2019) çalışmasına göre çok uzun olsa da, bu sürelerin sırası ile 44.2 -55.3 gün ve 49.5-63.0 gün sürdüğünü bildiren Atila (2022)'nin verilerinden daha kısa zaman almıştır. Çalışmalar arasındaki bu farklılıklar farklı yetiştirme ortamları ve farklı ırkların kullanılmasından kaynaklanmaktadır.

Sadece yerfıstığı kabuğu kullanılarak hazırlanan yetiştirme ortamının *H. ulmarius*'un üretim periyotlarının kısaltılması üzerinde olumlu bir etkisinin olduğu belirlenmiştir. Genel olarak FK100 ortamı hızlı

kolonizasyonu desteklemiş ve mantar üretiminde erkencilik sağlamıştır. Mantar misellerinin büyümesi ve taslak gelişimi üzerinde yetiştirme ortamının lignoselülozik içeriği ve C:N oranının önemli etkisi vardır. Yang (2000), C:N oranının yaklaşık 22-30:1 oranında olmasının taslak oluşumunu ve şapka gelişimini desteklediğini, daha yüksek C:N oranının ise misel gelişimini desteklediğini öne sürmüşlerdir. Çalışmamıza en hızlı misel gelişinin, taslak oluşumunun ve şapka gelişiminin desteklendiği FK100 yetiştirme ortamının C:N oranı 61.36 olup, diğer yetiştirme ortamlarının C:N oranından düşüktür. Diğer taraftan, Öztürk ve Atila (2021), *H. ulmarius* mantarının hızlı misel gelişimi ile yetiştirme ortamının selüloz içeriği arasında bir ilişki bulunduğunu ve yüksek selüloz içeriğinin misel gelişim süresini kısalttığını bildirmişlerdir. Çalışmada yürütülen korelasyon çalışmaları, misel gelişim süresi ile N (r^2= - 0.953) ve C:N oranları (r^2= - 0.955) ile negatif, hemiselüloz içeriği (r^2=0.939) ile pozitif bir korelasyon olduğu ortaya çıkarmıştır. FK100 yetiştirme ortamının düşük hemiselüloz içeriği muhtemelen miselin hızlı gelişimine katkı sağlamış olabilir. Bu durum yerfıstığı kabuğunun kimyasal özelliklerinin yanı sıra fiziksel yapısı ile ilgili olabilir. Havalanma kapasitesi yüksek, kaba bir yapıya sahip ortamların misel gelişimini hızlandırdığı ve mantar üretim sürecini kısalttığı bazı çalışmalarda vurgulanmıştır (Wang ve ark., 2021; Hu ve ark. 2022).

Farklı yetiştirme ortamlarının Hypsizygus ulmarius mantarının verim ve biyolojik etkinliği üzerine etkileri

Çalışmamızda S25:FK75 ortamı, diğer ortamlara kıyasla önemli ölçüde daha yüksek verim ve BE(%) sergiledi ($p<0.001$) (Tablo 4).

Tablo 4. Farklı yetiştirme ortamlarının *Hypsizygus ulmarius* mantarının verim ve biyolojik etkinliğine etkisi

Yetiştirme ortamı	Flaş I (g)	Flaş II (g)	Flaş III (g)	Toplam verim (g/kg)	Biyolojik etkinlik (%)
Kontrol	130.83±13.9 b	81.94±13.0 b	-	210.36±4.0 d	63.74±1.1 d
S75:FK25	143.10±12.7 a	77.56±10.5 b	-	220.66±6.2 c	66.87±1.8 c
S50:FK50	136.22±10.2 ab	111.08±11.5 a	-	246.19±8.5 b	70.34±2.3 b
S25:FK75	147.68±11.1 a	83.40±12.8 b	37.53±8.6	266.98±8.4 a	78.52±2.3 a
FK100	121.07±7.3 c	50.55±6.0 c	35.83±6.5	207.45±5.8 d	59.27±1.6 e
P değeri	<0.001	<0.001	<0.001	<0.001	<0.001

Her değer ortalama ± standart sapma olarak ifade edilmiştir (n=10). Sütunlarında yer alan aynı harf ile adlandırılmış değerler arasında Tukey testine göre istatistiksel olarak fark yoktur

Farklı yetiştirme ortamlarında üretilen *H. ulmarius* için verim 207.45 g/kg -266.98 g/kg ve BE %59.27-%78.52 arasında değişmiştir. Genel olarak, samana eklenen fıstık kabuğu miktarı arttıkça BE (%) yükselirken, yefıstığı kabuğunun tek başına kullanıldığı yetiştirme ortamı FK100'de en düşük BE (%) elde edilmiştir. Çeşitli substratlar üzerinde yapılan diğer yetiştirme çalışmalarında BE değerleri %36 ile %93 arasında (Ozturk veAtila, 2021) veya %231.7 ve %437.5 arasında (Malayil ve ark., 2017) bulunmuştur. Belirli suşların genetik arka planı, substratların bileşimi ve hazırlanmasındaki farklılıkların yanı sıra yetiştirme metodolojileri ve çevre koşulları, bu verim farklılıkları için açıklama sağlayabilir.

S25:FK75 ortamından elde edilen verim kontrol ortamına göre %26.9 oranında daha yüksekti. FK100 yetiştirme ortamı ise istatistiksel olarak kontrol ortamı ile aynı grupta yer almıştır. *H. ulmarius* mantarının üretiminde kullanılan beş yetiştirme ortamı arasında, FK100 ortamı diğer ortamlara göre daha hızlı misel gelişim hızı, taslak ve şapka oluşum süresi bakımından diğer ortamlara göre daha iyi sonuçlar sergilese de, aynı performans mantar verimi konusunda gözlenmemiştir. Özellikle yerfıstığı kabuğu karıştırılarak hazırlanan yetiştirme ortamlarındaki hızlı misel gelişimi, yüksek verim ve BE değerleri, bu tarımsal atığın ticari *H. ulmarius* üretimi için umut verici bir alternatif substrat olduğunu göstermektedir.

90 gün boyunca sürdürülen bu çalışmada S75:FK25 ve FK100 ortamlarından 3 hasat, diğer 3 yetiştirme ortamından ise 2 hasat elde edilmiştir. Ancak FK100 ortamında 3 kez hasat yapıldığı halde her bir hasatta elde edilen verim düşük olduğundan toplam verim düşük olmuştur. Çalışmada kullanılan yetiştirme ortamlarının hepsinde ilk hasatta elde edilen verim, toplam verimin %55-65'ini oluşturmuştur. Birinci hasatta en yüksek

verim 143.10 g/kg ve 147.68 g/kg ile S25:FK75 ve S75:FK25 ortamlarından elde edilmiştir. İkinci hasatta en yüksek verim 111.08 g/kg ile S50:FK50 yetiştirme ortamında belirlenmiştir. Sadece S75:FK25 ve FK100 ortamlarından eldilen 3. hasatlar ise sırası ile 37.53 g/kg ve 35.83 g/kg'dır.

Farklı yetiştirme ortamlarının *Hypsizygus ulmarius* mantarının şapka boyutları etkinliği üzerine etkileri

Bu çalışmada kullanılan beş farklı yetiştirme ortamında gelişen mantarların şapka çapı, sap uzunluğu ve sap çapı bakımından önemli farklılıklar gözlenmiştir ($p<0.001$) (Tablo 5).

Tablo 5. Farklı yetiştirme ortamlarının *Hypsizygus ulmarius* mantarının şapka boyutlarına etkisi

Yetiştirme ortamı	Şapka çapı (mm)	Sap uzunluğu (mm)	Sap çapı (mm)
Kontrol	86,3±8.7 ab	69,9±7.5 b	23,5±0.7 a
S75:FK25	81,4±2.2 ab	74,7±8.0 b	17,5±3.5 c
S50:FK50	78,1±6.2 b	75,4±6.3 b	17,7±1.7 c
S25:FK75	98,3±4.0 a	87,0±5.6 a	19,6±1.8 b
FK100	84,4±7.5 ab	45,8±5.9 c	17,7±0.8 c
P values	<0.001	<0.001	<0.001

Her değer ortalama ± standart sapma olarak ifade edilmiştir (n=10). Sütunlarında yer alan aynı harf ile adlandırılmış değerler arasında Tukey testine göre istatistiksel olarak fark yoktur

Mantarların şapka çapları, 78.1 mm-98.3 mm, sap uzunluğu 45.8 mm-87.0 mm, sap çapları ise 17.5 mm-23.5 mm arasında değişmiştir. En büyük şapka çapı ve sap uzunluğuna sahip mantarlar, en yüksek veriminde elde edildiği S25:FK75 ortamından hasat edilmiştir. En kalın saplara sahip olan şapkaların ise kontrol ortamında geliştiği gözlenmiştir. Büyük boyutlu şapkaların iyi kalitede olduğu kabul edilir ve mantar üretiminde yüksek kaliteli olarak derecelendirilir (Li ve ark., 2019) ancak bu tür şapkalar paketleme sırasında kırılma eğiliminde olabilir ve kalite düşebilir.

SONUÇ VE ÖNERİLER

Yetiştirme ortamları bileşenlerinin analizi, bunların kolonizasyonu ve şapka oluşumuna cisimlere biyolojik dönüşüm verimlilikleri ile ilgili veriler, yerfıstığı kabuğunun ticari olarak *H. ulmarius* yetiştiriciliği için umut verici bir alternatif substrat olduğunu göstermiştir. Ülkemizde, özellikle Akdeniz bölgesinde bol miktarda ortaya çıkan ve kullanım alanı sınırlı olan yerfıstığı kabuklarının mantar üretiminde kullanılması, bu atıkların çok değerli bir protein kaynağı ve çeşitli tıbbi özelliklere sahip bir ürüne biyodönüşümünün sağlanmasının yanı sıra, bu atıkların etkin bir şekilde değerlendirilmesi ve atık yönetiminin kolaylaşmasına da katkı sağlayacaktır.

Referanslar

Atíla F 2022. Using phenol-rich agro-wastes as substrates for the cultivation of hypsizygus ulmarius mushroom with enhanced functional and nutritional potential. Brazilian Archives of Biology and Technology, *65*. https://doi.org/10.1590/1678-4324-2022210669

Atila F 2023. Possibility of safe, easy and lower cost substrate disinfection with chlorine dioxide in Hypsizygus ulmarius cultivation. Scientia Horticulturae, 318: 112139.

Baghel D, Singh V, Shukla CS, Singh HK 2019. Studies on nutritional and physiological requirement for growth and biomass of *Hypsizygus ulmarius*. International Journal of Current Microbiology and Applied Science. 8:169–176. https://doi.org/10.20546/ ijcmas.2019.812.025.

Çelik Ç, Gürdal E 2010. Yerfıstığı kabuğunun agrega olarak kullanım olanakları. İTÜ Dergisi 4(1):37-46

Eswaran A, Henry DC, Jaiganesh V 2019. Effect of different substrate alone and in combination on the sporophore production of elm oyster mushroom *Hypsizygus ulmarius*. Journal of Pharmacognosy and Phytochemistry, *8*(3): 3167-3171.

FAOSTAT. Food and Agriculture Organization of the United Nations Statistical Databases. Available: http://www.fao.org/faostat/en/#data/QC [17 Eylül 2023]

Ganguly P, Sengupta S, Das P, Bhowal A 2020. Synthesis of cellulose from peanut shell waste and its use in bioethanol production. In: Ghaosh SK, Sen, R, Chanakya, HN, Pariatamby, A (eds), Bioresource Utilization and Bioprocess, New York: Springer, pp. 81-91.

González, A, Cruz M, Losoya C, Nobre C, Loredo A, Rodríguez R, Contreras J, Belmares R 2020. Edible mushrooms as a novel protein source for functional foods. Food & Function, 11(9), 7400-7414.

Hu W, Di Q, Liang, T, Liu J, Zhang J 2022. Effects of spent mushroom substrate biochar on growth of oyster mushroom (Pleurotus ostreatus). Environmental Technology and Innovation, 28: 102729.

Kumar K, Lal, AA, Mohle KK 2019. Evaluation of different substrates for growth, yield and nutritive value of *Hypsizygus ulmarius* (Blue Oyster Mushroom). Journal of Pharmacognosy and Phytochemistry. 8:1634–1638

Leong YK, Ma TW, Chang JS, Yang FC 2022. Recent advances and future directions on the valorization of spent mushroom substrate (SMS): A review. Bioresource Technology, 344: 126157.

Li R, Zhang Y, Chu W, Chen Z, Wang J 2018. Adsorptive removal of antibiotics from water using peanut shells from agricultural waste. RSC Advances, 8(24): 13546-13555.

Li Z, Zhou J, Lin Z 2019. Development and innovation of Ganoderma industry and products in China. Ganoderma and Health: Biology, Chemistry and Industry, 187-204.

Malayil S, Chanakya HN, Ashwath H, Suresh H 2017. Biogas digester liquid as a supplement for higher yields of Hypsizgus ulmarius. Environment Technology and Innovation 8:269–281.

Munna J, Lal A A ,Singh PK 2019. Performance of different substrate on the production and nutritional composition of blue oyster mushroom (Hypsizygus ulmarius (Bull.: Fr.) Redhead). International Journal of Chemical Studies SP6: 366-368

Okoro IO, Achuba FI 2012. Proximate and mineral analysis of some wild edible mushrooms. African Journal of Biotechnology, 11(30):7720-7724.

Oseni TO, Dube SS, Wahome PK, Masarirambi MT, Earnshaw DM 2012. Effect of wheat bran supplement on growth and yield of oyster mushroom (Pleurotus ostreatus) on fermented pine sawdust substrate. Experimental Agriculture & Horticulture, *30*: 40.

Öztürk C, Atila F 2021. Changes in lignocellulosic fractions of growing substrates during the cultivation of Hypsizygus ulmarius mushroom and its effects on mushroom productivity. Scientia Horticulturae, 2*88*: 110403.

Şahin G 2014. Türkiye'de Yerfıstığı (Arachis hypogaea L.) Yetiştiriciliği ve Bir Coğrafi İşaret Olarak Osmaniye Yerfıstığı. *Gaziantep* University Journal of Social Sciences, *13*(3):619-644

Taşar Ş, Kaya F, Özer A 2014. Biosorption of lead (II) ions from aqueous solution by peanut shells: Equilibrium, thermodynamic and kinetic studies. Journal of Environmental Chemical Engineering, 2(2):1018-1026.

Thakur MP 2014. Present status and future prospects of tropical mushroom cultivation in India: a review. Indian Phytopathology, 67(2): 113-125.

Waktola G, Temesgen T 2018. Application of mushroom as food and medicine. Advances Biotechnology and Microbiology 11(3): 10-19080.

Wang Q, Juan J, Xiao T, Zhang J, Chen H, Song X, Huang J 2021 The physical structure of compost and C and N utilization during composting and mushroom growth in Agaricus bisporus cultivation with rice, wheat, and reed straw-based composts. Applied Microbiology and Biotechnology, 105: 3811-3823.

Yang, X.M., 2000. Cultivation of Edible Mushroom. China Agriculture Press, Beijing, p. 36.

Zadražil F, Brunnert H 1982. Solid state fermentation of lignocellulose containing plant residues with Sporotrichum pulverulentum Nov. and Dichomitus squalens (Karst.) Reid. European Journal of Applied Microbiology and Biotechnology, 16: 45-51.

Van Soest PV, Robertson JB, Lewis BA 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal Of Dairy Science*, *74*(10): 3583-3597.

DRAFT

ORAL PRESENTATION

Alpha-tocopherol ameliorates hydrogen peroxide-induced apoptosis in human dental pulp stem cells

Mukaddes Mergen Dalyanoğlu[1*] (*ORCID: https://orcid.org/0000-0002-5862-7792*), Mücahit Seçme[2] (*ORCID: https://orcid.org/0000-0002-2084-760X*), Yavuz Dodurga[3] (*ORCID: https://orcid.org/0000-0002-4936-5954*), Sebahat Turgut[4] (*ORCID: https://orcid.org/0000-0002-7884-5623*)

[*1]Pamukkale University, Denizli Healthcare Vocational School, Medical Services and Techniques Department, Denizli, Turkey.
[2]Ordu University, Faculty of Medicine, Department of Medical Biology, Ordu, Turkey.
[3]Pamukkale University, Faculty of Medicine, Department of Medical Biology, Denizli, Turkey.
[4]Ondokuz Mayıs University, Faculty of Medicine, Department of Physiology, Samsun, Turkey.

*Corresponding author e-mail: mmergendalyanolu@hotmail.com
mdalyanoglu@pau.edu.tr

Abstract

Tooth whitening has been found to have side effects on cell metabolism in dental pulp tissue because of peroxide content. In this work, we evaluated the effect of hydrogen peroxide (HP) on apoptosis in human dental pulp stem cells (hDPSCs) and whether alpha-tocopherol (AT) has protective effects against possible apoptosis in vitro. Without treatment (control group), the second group; 2 µg/mL hydrogen peroxide was applied for 30 minutes, the third group; before 100 µM alpha-tocopherol was applied to cells for 24 hours after then 2 µg/mL hydrogen peroxide was applied to the same cells during for 30 minutes. Cytotoxicity of alpha-tocopherol was determined using the XTT assay. For the determination of apoptosis, mRNA levels of Bcl-2 and Bax were determined by Real-Time PCR analysis. In the comparisons of groups with the control, evaluations were performed using the Student's t-test ($p<0.05$). According to the results of the XTT analysis, the alpha-tocopherol dosage that provided the most cell viability to be applied in the experiment was determined as 100 µM. Apoptosis was induced by hydrogen peroxide and alpha-tocopherol inhibited hydrogen peroxide-induced apoptosis in HDPCs by regulating the mRNA levels of Bcl-2 and Bax.

Keywords: Human dental pulp stem cell, hydrogen peroxide, alpha-tocopherol, apoptosis

INTRODUCTION

Dental aesthetics especially in terms of tooth color are widely needed in daily life from past to present. Dental bleaching is preferred in the treatment of tooth discoloration due to its low-cost aesthetic dentistry procedure and non-invasive approach (Fioresta et al. 2023).

Hydrogen peroxide (HP) is widely used as a dental bleaching agent. It acts through the releasing reactive oxygen species (ROS) that has the ability to reach the pulp through the tooth enamel and dentinal tubules causing decreased cellular metabolism causing a pathologic oxidative condition, lipid peroxidation, as a side effect (Benetti et al. 2017).

Apoptosis is mitochondrial-mediated cell death and is key in homeostatic tissue mechanisms (Benetti et al. 2017). The increase in the mitochondrial membrane permeability due to mitochondrial swelling may trigger the release of cytochrome c that later activates Bax, Bcl-2 and caspase-3 leading to apoptosis (Arrozi et al. 2020).

Vitamin E is a fat-soluble antioxidant molecule that prevents lipid peroxidation by scavenging reactive oxygen species, which is known to possess protective abilities against aging processes. Tocopherol is the predominant isoform of vitamin E and a natural agent with antioxidant and anti-inflammatory properties (Escobar et al. 2020).

Human dental pulp stem cells (hDPSCs) are mesenchymal stem cells (MSCs) exhibiting clonogenicity and capability of self-renewal and have high proliferation rate and multilineage differentiation potential such as

neuronal cells, chondroblasts, adipocytes, and osteoblasts. They are ideal for tissue engineering, osteogenesis, and regenerative medicine (Escobar et al. 2020).

Nevertheless, the apoptotic pathway induced by hydrogen peroxide and possible anti-apoptotic effects of alpha-tocopherol on human dental pulp stem cells has not been fully studied; therefore, in this work we evaluated the effect of hydrogen peroxide (HP) on apoptosis in human dental pulp stem cells (hDPSCs) and whether alpha-tocopherol (AT) has protective effects against possible apoptosis in vitro.

MATERIALS AND METHODS

Cell line, Cell Culture

Human dental pulp stem cells were used in the study. DPSCs were cultured and subcultured into T75 cell culture flasks in Minimum Essential Medium Alpha (MEM alpha) modifications with L-glutamine, D glucose (Biological Industries Co.) supplemented with 10% fetal bovine serum (Gibco Co.), 100 IU/mL penicillin, 10 mg/mL streptomycin (Biological Industries Co.) in a humidified incubator (Nuve) with 5% CO2 and 95% air at 37°C.

Minimum Essential Medium Alpha (MEM alpha) modifications with L-glutamine, D glucose (Biological Industries Co.); supplemented with 10% fetal bovine serum (Gibco Co.), 100 IU/mL penicillin, 10 mg/mL streptomycin (Biological Industries Co.) was used to culture and subculture hDPSCs. Cells were fed in cell culture flasks and placed in a humidified incubator with 5% CO_2, 37°C (Nuve). The cell medium was refreshed on average every 3-4 days and cell passages were performed when the cells reached 90% confluence.

Determination of Cell Viability

The 100 µM, 200 µM, 500 µM, 1 mM, 2 mM concentrations of alpha-tocopherol (Sigma-Aldrich Co.) in MEM alpha medium were prepared. The cytotoxic effect of alpha-tocopherol on hDPSCs was determined by XTT (sodium 3´-[1- (phenylaminocarbonyl)- 3,4- tetrazolium]-bis (4-methoxy6-nitro) benzene sulfonic acid hydrate) assay. Cells reaching 90% density were removed from the flasks by Trypsin-EDTA solution. Cells were seeded into 96-well microplates with 2.5×10^4 cells per well. The seeded cells were incubated in 100 µL of completed MEM-Alpha for 24 h in a CO_2 incubator at 37°C. After incubation, the media in the wells were refreshed, then different concentrations of alpha-tocopherol were added to the microplate wells and incubated for 24 hours. XTT analysis was performed to evaluate cell viability at the end of the period, with a commercial cell proliferation kit (Biological Industries Co.). The cell medium in the wells of microplate was removed and 50µL of the XTT reaction solution and 50µL of MEM-Alpha were added to each well and the plate was incubated for 4 h at 37°C, 5% CO_2. After the incubation, the optical densities of cells in microplate were measured with an ELISA microplate reader (Thermo Fisher Scientific) both at a wavelength of 450 nm and at a wavelength of 630 nm at the same time. To measure reference absorbance; a wavelength of 630 nm measurement was subtracted from the 450 nm measurement for all wells. The average of the absorbance values obtained from the control wells was accepted as 100% viable cells. The percentages of viability values in the experimental wells were calculated by comparing the absorbance values obtained from alpha-tocopherol applied wells to the control absorbance value.

Determination of the Dosages of alpha-tocopherol and hydrogen peroxide

According to the results of the XTT analysis, the alpha-tocopherol dosage that provided the highest cell viability to be applied in the experiment was determined as 100 µM.

Hydrogen peroxide dosage applied to hDPSC was determined based on the study of Soares et al. 2014. The reason we choosed this dosage was that they simulated office tooth bleaching in cell culture. They measured the quantity of HP diffused through the tooth enamel and dentin into the cell culture medium as 2 µg/mL.

Experimental design

The following three groups were formed for this study. The first group without treatment (control group); the second group; cells were exposed to 2 µg/mL HP for 30 min, the third group; before 100 µM alpha-tocopherol was applied to cells for 24 h after then 2 µg/mL HP was applied to the same cells during for 30 minutes.

Total RNA isolation, cDNA Synthesis and RT- PCR

Total RNA was isolated from study groups with TRIzol reagent (Invitrogen, USA) according to manufacturer instructions. Complementary DNA (cDNA) synthesis was performed using cDNA synthesis kit , according to the manufacturers' instructions. Quantitative expression analysis of Bax, Bcl-2 were determined by Real-Time

PCR (StepOne Plus, Applied Biosystem). The specific binding forward and reverse primer sequences of the associated gene and gene symbols are given in Table 1. Beta Aktin was used as hosukeeping gene for normalization of RT-PCR data.

Statistical analysis

The analysis of the findings was performed using the Delta-Delta Ct method and quantified using the Gene globe analysis platform. In the comparisons of groups with the control, evaluations were conducted using the Student's t-test, and $p<0.05$ was considered as the level of significance.

RESULTS and DISCUSSION

The viability of the human dental pulp stem cells exposed to alpha-tocopherol is shown in Fig. 1. According to the results of the XTT analysis, 100 µM concentrations of alpha-tocopherol provided the highest cell viability (132%) and therefore this concentration was applied in the experiment. 200 µM concentrations of alpha-tocopherol did not reveal a change in cell viability compared to the control group, while the 0.5, 1 and 2 mM concentrations of alpha-tocopherol decreased the cell viability.

The mRNA expression levels of bcl-2 and bax genes of hydrogen peroxide and both hydrogen peroxide and alpha-tocopherol applied to human dental pulp stem cells were determined. The results were normalized using the mRNA expression level of the β-actin reference gene. mRNA expression rates of bcl-2 and bax genes were determined. The mRNA expression levels of all genes were evaluated as 1.0 for the control group and the results were given as fold values compared to the control group. Fold changes levels of bax and bcl-2 are given respectively in Fig. 2.

Although it had been reported hydrogen peroxide-induced apoptosis in human gingival fibroblasts (Venegas et al. 2015) and human dental pulp stem cells (Llena et al. 2019) but molecular mechanism of the prevention of apoptosis with antioxidants such as alpha-tocopherol remains unclear.

It has been reported that Bcl-2, anti-apoptotic protein and Bax pro-apoptotic resided in the mitochondrial outer membrane and were involved in mitochondrial-dependent apoptosis pathways by regulating mitochondria membrane permeability. Proapototic protein Bax induces cell apoptosis via mitochondrial membrane permeabilization (Venegas et al. 2015). In the present study, hydrogen peroxide treatment resulted in significantly increased expression of Bax and Bcl-2 and both hydrogen peroxide and alpha-tocopherol treatment significantly decreased expression of bax and Bcl-2 which played a role via mitochondrial pathways in hydrogen peroxide-induced apoptosis in hDPSCs. Results revealed that hydrogen peroxide exhibited potential to trigger apoptosis in mitochondria pathways as previously reported and alpha-tocopherol also reduced hydrogen peroxide-induced apoptosis.

Table 1. Forward and reverse primer sequences designed for use in real time polymerase chain reaction.

Gene Name	Forward	Reverse
Beta-actin	TCCTCCTGAGCGCAAGTACTC	CTGCTTGCTGATCCACATCTG
BCL-2	TGCACCTGACGCCCTTCAC	AGACAGCCAGGAGAAATCAAACAG
BAX	TCAGGATGCGTCCACCAAGAAG	TGTGTCCACGGCGGCAATCATC

DRAFT

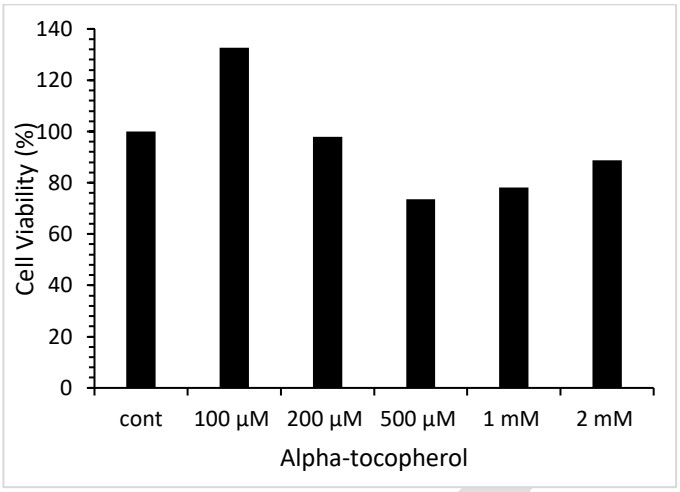


Figure 1. The % change in viability of hDPSCs human dental pulp stem cells treated with different concentrations of alpha-tocopherol for 24 hours.

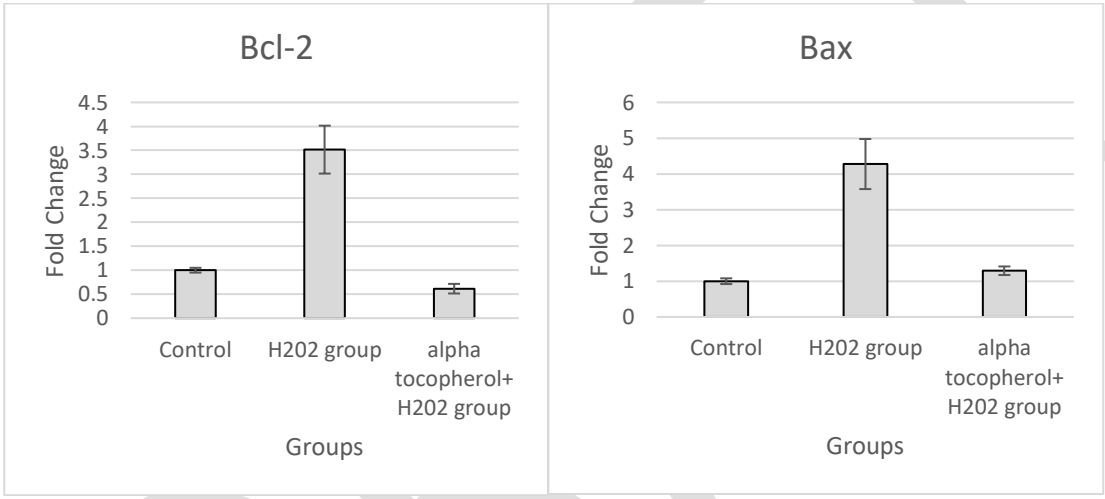


Figure 2. Fold changes levels of the control, hydrogen peroxide and both hydrogen peroxide and alpha-tocopherol groups in hDPSCs. Fold changes levels of bax and bcl-2 were examined by RT-PCR. β-Actin was used as a housekeeping gene. RT-PCR, real time polymerase chain reaction ($p<0.05$).

CONCLUSION

Hydrogen peroxide showed apoptotic effects by increased bax gene expression and alpha-tocopherol showed anti-apoptotic effects by decreased bax gene expression on human dental pulp stem cells. However, further studies are needed to better understand how alpha-tocopherol affects the molecular mechanisms in cell cycle.

REFERENCES

Aslina Pahrudin Arrozi, Siti Nur Syazwani Shukri, Wan Zurinah Wan Ngah, Yasmin Anum Mohd Yusof, Mohd Hanaf Ahmad Damanhuri, Faizul Jaafar & Suzana Makpo 2020. Comparative Efects of Alphaand Gamma-Tocopherol on Mitochondrial Functions in Alzheimer's Disease In Vitro Model.Scientific Reports, 10:8962.

Francine Benetti, João Eduardo Gomes-Filho, Luciana Louzada Ferreira, Edilson Ervolino, André Luiz Fraga Briso , Gustavo Sivieri Araújo, Eloi Dezan-Júnior, Luciano Tavares Angelo Cintra 2017. Hydrogen peroxide induces cell proliferation and apoptosis in pulp of rats after dental bleaching in vivo. Archives of Oral Biology. Reference: AOB 3857. Accepted date: 17-4-2017. DOI: http://dx.doi.org/doi:10.1016/j.archoralbio.2017.04.013.

Gloria Gutiérrez-Venegas, Adriana Guadarrama-Solís, Carmen Muñoz-Seca, Juan Antonio Arreguín-Cano 2015. Hydrogen peroxide-induced apoptosis in human gingival fibroblasts. Int J Clin Exp Pathol 8(12):15563-15572

Lina M. Escobar, Zita Bendahan, Andrea Bayona, Jaime E. Castellanos, and Marı´a-Clara Gonza´lez 2020. Effect of Vitamins D and E on the Proliferation, Viability, and Differentiation of Human Dental Pulp Stem Cells: An In Vitro Study. Hindawi International Journal of Dentistry Volume 2020, Article ID 8860840, 10 pages. Published 1 July 2020

Llena, M.Collado-González, D.García-Bernal, R. E.Oñate-Sánchez, C. M. Martínez, J. M. Moraleda, F. J. Rodríguez-Lozano, L. Forner 2019. Comparison of diffusion, cytotoxicity and tissue inflammatory reactions of four commercial bleaching products against human dental pulp stem cells. Sci Rep. May 23;9(1):7743.

Rossella Fioresta, María Melo, Leopoldo Forner, José Luis Sanz 2023. Prognosis in home dental bleaching: a systematic review. Clinical Oral Investigations 27:3347–3361

DRAFT

ORAL PRESENTATION

Integrating Carbon Dots into Upconverting Nanoparticles Using *In-Situ* Solvothermal Method

Rüveyda KÜÇÜK[1*] *(ORCID: 0009-0004-9365-4560)*, Melis Özge ALAŞ ÇOLAK[2] (*ORCID: 0000-0002-0546-087X)* Rükan GENÇ ALTÜRK[2] (*ORCID: 0000-0002-9569-8776)*

[*1]Department of Chemical Engineering, Faculty of Engineering Mersin University, Chemical Engineering, Mersin, Turkey
[2]Sabanci University,SUNUM Nanotechnology Research Centre, Istanbul, Turkey

*Corresponding author e-mail: rukan.genc@sabanciuniv.edu

Abstract

Nanoparticles exhibiting upconversion properties are preferred today due to many advantages such as sharp emission peaks, long fluorescence lifetime, low toxicity, and superior photostability. Upconverting nanoparticles (UCNPs) are often lanthanide-doped and are used in important areas such as bioimaging, biosensors, security systems, and drug delivery systems. UCNPs can absorb low-energy near-infrared (NIR) photons and emit higher-energy photons in the visible (VIS) or UV region. In contrast to UCNPs, luminescent materials exhibiting downconversion (DC) properties (e.g., organic fluorescence, quantum dots, MOFs, and carbon dots) are actively used in applications such as optics, energy, and bioimaging. Carbon dots (CDots, CDs), which exhibit DC properties and hold an important place among carbon-based nanomaterials, are known for their excellent optical properties. UC materials and carbon dots are garnering increasing interest because of their attractive features and wide-ranging applications. Integrating materials with DC and UC properties to create dual-modal luminescent materials is an emerging research topic. The aim of this study is to create a dual-modal luminescent material by integrating rare-earth-doped UC materials with carbon dots, which exhibit DC properties. Firstly, UCNPs were synthesized using a solvothermal method, and then UCNP@CD materials were obtained by integrating CDs into UCNPs using an *in-situ* method. The synthesized DC and UC fluorescent materials were subjected to physical and chemical characterization analyses using high-resolution transmission electron microscopy (HR-TEM), UV/Vis spectrophotometry, Zeta potential, Fourier-transform infrared spectroscopy (FT-IR), and X-ray diffraction (XRD) devices.

Keywords: Upconverting, carbon dot, downconverting nanoparticles, solvothermal syntheis

INTRODUCTION

Carbon dots, which exhibit characteristics such as low toxicity, biocompatibility, adjustable photoluminescence, and high quantum yield (QY), are prominent research subjects among materials capable of downconversion. Moreover, CDs demonstrate luminescence properties that adhere to Stokes' law (Liu, Li and Yang, 2020.). These properties make CDs valuable in various applications, including biomedical, bioimaging, biosensors, sensors, photocatalytic devices, and photocatalysis devices. These CDs are utilized in various fields such as tumor imaging, drug delivery systems, photoacoustic imaging, photosensitizers, and light-emitting diodes (Himaja, Karthik and Singh, 2015.).

Materials displaying upconversion (UC) characteristics possess several crucial attributes, including low toxicity, sharp emission peaks, and photostability. Rare-earth-doped upconverting materials exhibit enhanced fluorescence properties. UCNPs absorb low-energy near-infrared photons and emit photons in the high-energy visible and ultraviolet regions. Materials with upconversion capabilities are actively used in applications such as optics, bioimaging, biosensors, drug delivery systems, cancer treatment, security systems and pollution analysis (Chen *et al.*, 2014.; Mettenbrink, Yang and Wilhelm, 2022.).

Interest in materials capable of both downconversion and upconversion is steadily growing, expanding the scope of research. In recent years, there has been a focus on integrating two different dual-mode luminescent materials capable of both downconversion and upconversion. Fluorescent CDs and rare-earth-doped UCNP materials have been integrated into various studies. Wu and colleagues have introduced a simple solvothermal method for synthesizing dual-mode luminescent $NaYF_4$:Yb, Er(Tm)/CD composites for anti-counterfeiting

barcodes. In this study, CDs exhibited relatively weak adsorption on the surface of UC materials (Wu *et al.*, 2016.). Sun and colleagues have used CD-modified $NaYF_4$:Yb, Er nanoparticles (UCNPs@CDs) as fluorescent nanoprobes for the simultaneous detection of Fe^{2+} and Fe^{3+} ions (Sun *et al.*, 2021.). Wang and colleagues have modified the surface of UCNPs with ATP-sensitive CDs to develop an upconversion nanoprobe activated by lysosomal ATP for ratiometric imaging of hepatotoxicity (Wang *et al.*, 2022.). Research continues to improve the absorbance and dual-mode fluorescence properties of developed UCNP@CD materials.

In this current study, the aim is to synthesize dual-mode luminescent nanomaterials capable of downconversion and upconversion through the *in-situ* method using solvothermal synthesis and the combination of UCNP and CD. Thus, by integrating UCNPs with CDs, UCNPs@CDs are expected to possess the optical properties of both UCNPs and CDs, thanks to their excellent optical characteristics

MATERIALS AND METHODS

All chemicals used in the synthesis of the materials were used without purification. Yttrium (III) chloride hexahydrate ($YCl_3 \cdot 6H_2O$), Ytterbium (III) chloride hexahydrate ($YbCl_3 \cdot 6H_2O$), Erbium (III) chloride hexahydrate ($ErCl_3.6H_2O$), Ammonium fluoride (NH_4F) were purchase from Sigma-Aldrich. NaOH and urea were purchase from IsoLab. Ethanol was provide from Tekkim. Lemon salt was procured from local supermarket. Ultrapure water used throughout all the experiments was purified by the Milli-Q system (Millipore Inc., Ω=18 MΩ cm).

Synthesis of Upconverting Nanoparticles (UCNP)

$NaYF_4$:Yb, Er UCNPs were prepared according to a literature procedure (Tan *et al.*, 2019.; Sun *et al.*, 2021.). In brief, 0.15 mmol $YCl_3.6H_20$, 0.05 mmol $YbCl_3.6H_20$, and 0.0006 mmol $ErCl_3.6H_2O$ were first mixed in a beaker in ethanol/water (1:1 (v/v)) until a homogeneous solution was obtained. Oleic acid (OA) was added to the homogeneous solution. Then, in a separate beaker, a solution prepared in ethanol/water (1:1 (v/v)) containing NaOH (0.4 g) and NH_4F (0.05 g) was added drop by drop to this solution and stirred for 10 minutes on a magnetic stirrer. The final solution was transferred to a 100 mL Teflon-lined autoclave and subjected to heat treatment at 200°C for 7 hours. After the reaction, the obtained UCNP's were collected by centrifugation at 8000 rpm for 5 minutes, and the precipitate was washed three times with ethanol. Finally, the product was dried at 60°C for 12 hours.

Carbon Dot (CD) Synthesis

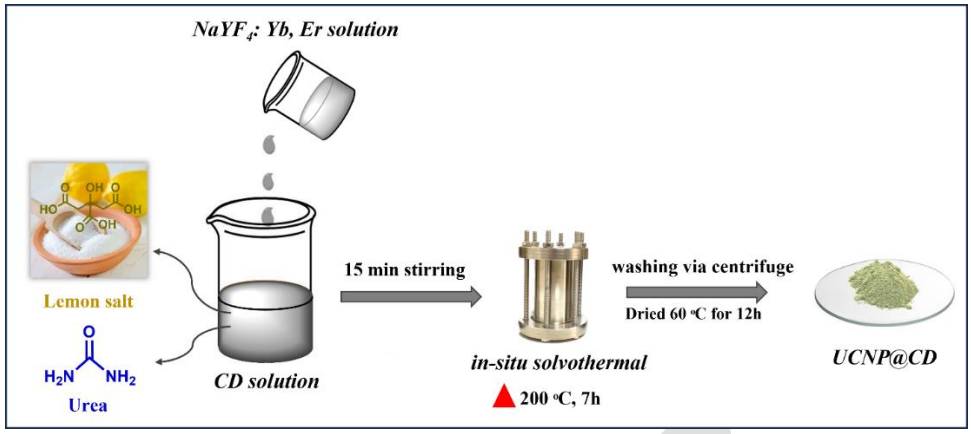
The synthesis of carbon dots was carried out by modifying a procedure from the literatüre (Alas *et al.*, 2019.). Initially, 0.12 g of citric acid and 0.12 g of urea were dissolved in EtOH:water (1:1 (v/v)). The resulting homogeneous solution was transferred to a 100 mL Teflon-lined autoclave and subjected to heat treatment at 200°C for 7 hours. After the reaction, the brown solution was centrifuged at 8000 rpm for 5 minutes, and the supernatant was collected. Finally, the product was dried at 60°C for 12 hours.

UCNP@CD Synthesis

In-situ integration of CDs into the obtained UCNP powder was achieved. In a beaker, citric acid and urea were dissolved in ethanol/water (1:1 (v/v)) until a homogeneous solution was obtained. This homogeneous solution was then added to a separate beaker containing a UCNP aqueous solution. The final mixture was transferred to a 100 mL Teflon-lined autoclave and subjected to heat treatment at 200°C for 7 hours. After the reaction, the UCNPs were collected by centrifugation at 8000 rpm for 5 minutes, and the precipitate was washed three times with ethanol. Finally, the product was dried at 60°C for 12 hours.

RESULT and DISCUSSION

As shown in Scheme 1, UCNP@CD were prepared by mixing UCNPs with CD precursors followed by an easy and simple in situ solvothermal process.



Scheme 1. Schematic illustration of synthesis of UCNP@CD by in-situ solvothermal.

Figure 1 shows digital images of the fabricated samples under 650 nm and 980 nm laser excitation. CDs exhibit faint radiation at 650 and 980 nm. However, when CDs are integrated into UCNPs, the radiation reflected by UCNP@CD under the laser is visibly more intense compared to UCNPs.

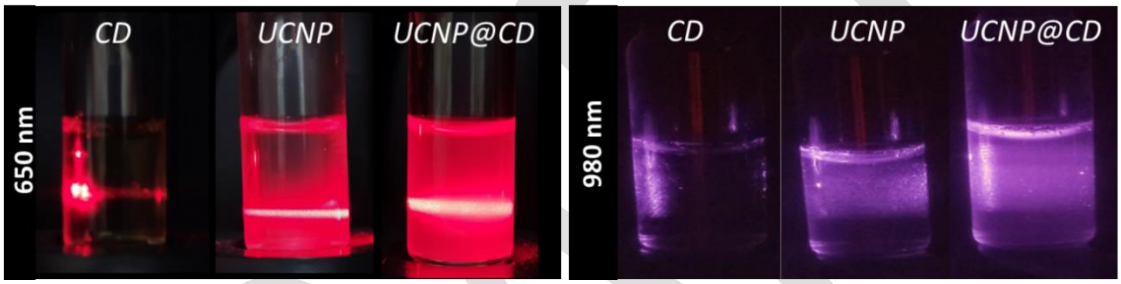


Figure 1. The digital images of CD, UCNP, and UCNP@CD under 650 nm and 980 nm laser excitation.

Under laser excitation at 650 nm and 980 nm, CD exhibits lower fluorescence compared to UCNP, but the fluorescence properties of UCNP@CD noticeably increase as a result of integrating the two materials.

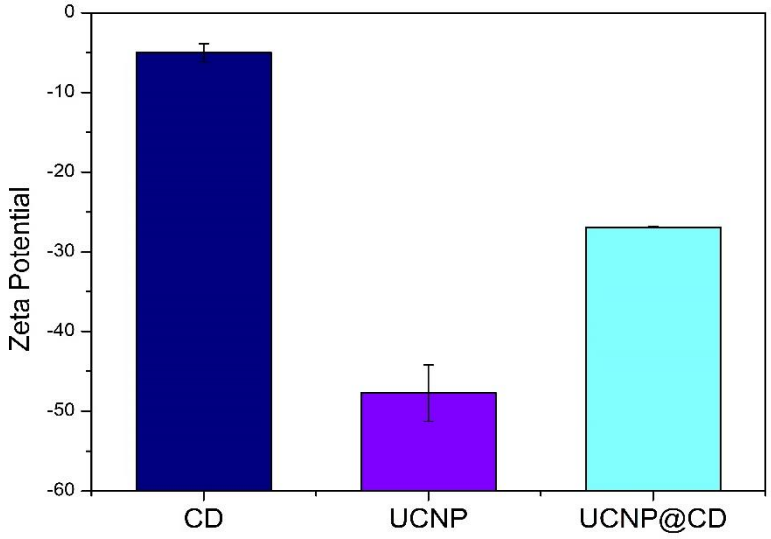


Figure 2. Zeta potentials of CD, UCNP, and UCNP@CD.

The zeta potentials of CD, UCNP, and UCNPs@CDs were measured in an aqueous environment and are shown in the Figüre 2. The zeta potentials of CD, UCNP, and UCNPs@CDs were -5 ± 1.12 mV, -47.73 ± 3.55 mV, and -26.9 ± 0.1 mV, respectively. It can be observed that the zeta potential of CD is opposite to that of UCNP, allowing the two particles to come together and form a compound due to the attraction of opposite poles. Additionally, the increase in zeta potential of UCNP@CDs compared to UCNP is attributed to the presence of numerous amine (-NH_2) groups on the surface of CD. This indicated that CDs successfully changed the surface charge of UCNPs and loaded onto the surface.

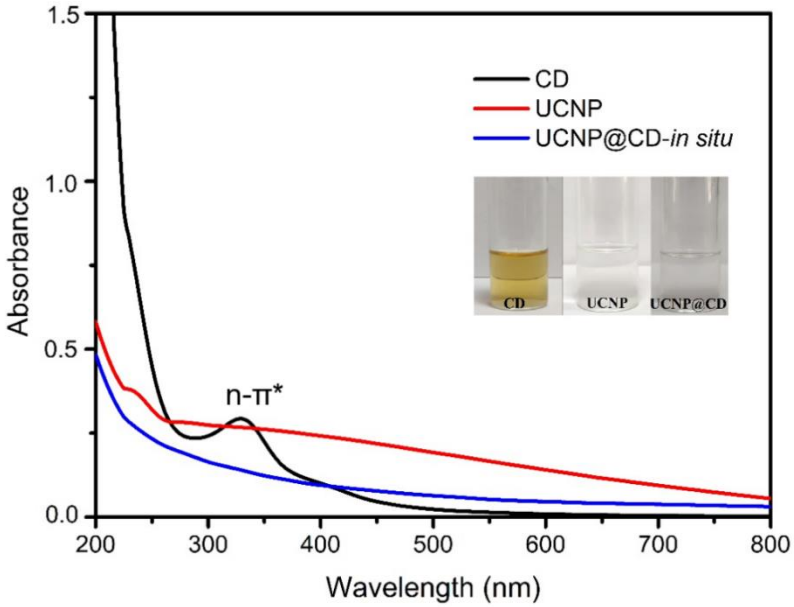


Figure 3. UV/Vis absorption spectra of CD, UCNP, and UCNP@CD

Figure 4 shows the optical UV/Vis spectrum of CD, UCNP, and UCNP@CDs. CDs exhibited a maximum absorption peak at around 330 nm, attributed to the n-π* transition of C=O or C=N bonds. UCNPs showed strong optical absorption in the UV region, along with a tail extending into the visible region. In the UV-Vis spectrum of UCNP@CDs, the peak points corresponding to CDs were not observed, likely due to the dominant optical contribution of UCNPs over CDs. UCNP@CDs, similar to UCNPs, exhibited strong optical absorption in the UV region with a tail extending into the visible region.

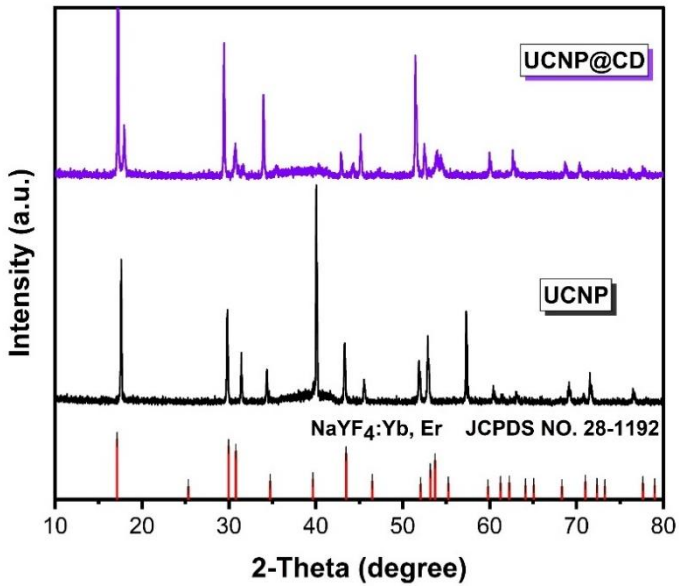


Figure 4. XRD spectra of UCNP and UCNP@CD.

The X-ray diffraction (XRD) patterns of the synthesized UCNP and UCNP@CD are presented in the Figure 4. The XRD profile of UCNPs clearly demonstrates the crystallization phase (JCPDS 28-1192) (Sun *et al.*, 2021.). The XRD pattern of UCNP@CDs shows well-crystallized structures identical to those of UCNPs. This indicates that the crystallographic structure of $NaYF_4$:Yb, Er was preserved during the modification process.

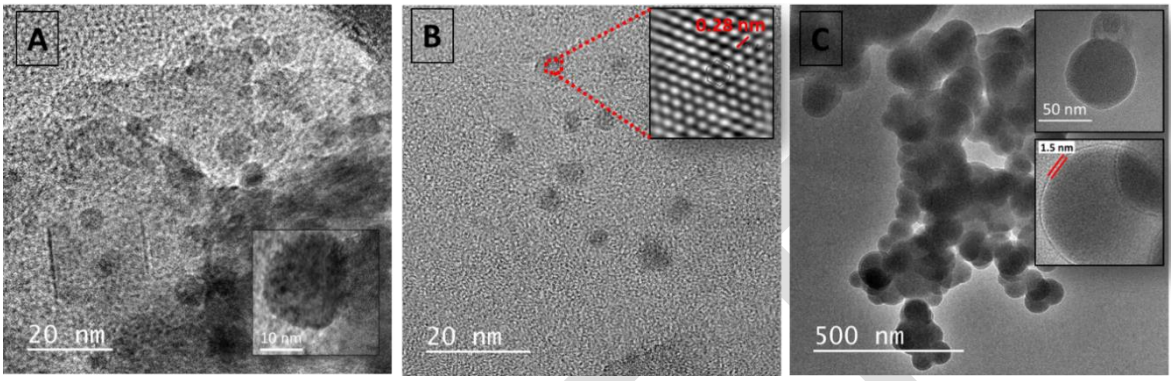


Figure 5. HR-TEM images of (A) CD, (B) UCNP, and (C) UCNP@CDs (Inset images: magnified views showing lattice spacing).

The morphological images of the synthesized materials were examined using HR-TEM analysis. CD nanoparticles appear to be uniformly spherical in shape, with an average particle size distribution of 5.49 ± 1.62 nm (Figure 5A). As shown in Figure 5B, UCNPs also exhibit a spherical shape with a homogeneous distribution and an average particle size of 4.06 ± 1.20 nm. All nanoparticles displayed similar shapes and uniform sizes. A lattice spacing of 0.28 nm was observed in UCNPs, corresponding to the (200) plane of the α-phase $NaYF_4$:Yb, Er (Janjua *et al.*, 2018.). UCNPs@CDs exhibited a core-shell structure, with the shell thickness measuring approximately 1.5 nm (Figure 5C). However, the absence of the crystal structure of CD within the UCNP structure indicates that the added CDs are amorphous.

CONCLUSION

As a result of the study, UCNP@CDs were successfully obtained by integrating CD and UCNP using the *in situ* solvothermal synthesis method. It was observed that under laser excitation at 650 nm and 980 nm, integrated UCNP@CD exhibited brighter fluorescence properties compared to CD and UCNP. Based on the zeta potential results, UCNP displayed a more negative value compared to CD. The integrated material obtained from the combination of these two materials exhibited intermediate properties between UCNP and CD. This indicates the successful synthesis of UCNP@CD. According to the UV-VIS analysis results, UCNP@CD did not show the characteristic 330 nm peak of CD. Instead, the integrated material exhibited strong optical absorption properties in the UV region, similar to UCNP. This dominance of UCNP over CD is reflected in the graphical results. XRD results indicated that the integrated material UCNP@CD retained the crystal characteristics of UCNP, confirming the successful integration of the two materials. Evaluation of HR-TEM analysis results revealed that CD, UCNP, and UCNP@CD were spherical in shape, homogeneously distributed, and that UCNP@CD had a core-shell structure. These materials can be used as promising candidates in applications such as anti-counterfeiting and excellent optical probes for multicolor cell imaging.

ACKNOWLEDGEMENTS

I would like to express my gratitude to my advisor, Associate Professor Dr. Rükan GENÇ ALTÜRK, and my co-advisor, Dr. Melis Özge ALAŞ ÇOLAK, for their guidance and support. I also extend my thanks to my colleagues in the FUNGROUP for their support throughout the study.

REFERENCES

Alas MO, Güngör A, Genc R, and Erdem E, 2019. Feeling the Power: Robust Supercapacitor from Nanostructured Conductive Polymer Fostered with Mn +2 and Carbon Dots. Nanoscale, 11: 12804–12816.

Chen G, Qiu H, Prasad PN, and Chen X, 2014. Upconversion nanoparticles: Design, nanochemistry, and applications in Theranostics. Chemical Reviews, 114(10): 5161–5214.

Himaja AL, Karthik PS, and Singh SP, 2015. Carbon Dots: The Newest Member of the Carbon Nanomaterials Family. Chemical Record, 15(3): 595–615.

Janjua RA, Gao C, Dai R, Sui Z, Ahmad Raja MA, Wang Z, Zhen X, and Zhang Z, 2018. Na+-Driven Nucleation of NaYF4:Yb,Er Nanocrystals and Effect of Temperature on Their Structural Transformations and Luminescent Properties. Journal of Physical Chemistry C, 122(40): 23242–23250.

Liu J, Li R, and Yang B, 2020. Carbon Dots: A New Type of Carbon-Based Nanomaterial with Wide Applications. ACS Central Science, 6(12): 2179–2195.

Mettenbrink EM, Yang W, and Wilhelm S, 2022. Bioimaging with Upconversion Nanoparticles. Advanced Photonics Research, 3(12): 2200098.

Sun YL, Zhang XP, Zhao CX, Liu X, Shu Y, Wang JH, and Liu N, 2021. Upconversion nanoparticles/carbon dots (UCNPs@CDs) composite for simultaneous detection and speciation of divalent and trivalent iron ions. Analytica Chimica Acta, 1183: 338973.

Tan H, Gong G, Xie S, Song Y, Zhang C, Li N, Zhang D, Xu L, Xu J, et al., 2019. Upconversion Nanoparticles@Carbon Dots@Meso-SiO2 Sandwiched Core-Shell Nanohybrids with Tunable Dual-Mode Luminescence for 3D Anti-Counterfeiting Barcodes. Langmuir, 35(35): 11503–11511.

Wang N, Yang Y, Zhang M, Zhu Q, and Li Z, 2022. Lysosomal Adenosine Triphosphate-Activated Upconversion Nanoparticle/Carbon Dot Composite for Ratiometric Imaging of Hepatotoxicity. Analytical Chemistry, 94(45): 15738–15745.

Wu F, Su H, Zhu X, Wang K, Zhang Z, and Wong WK, 2016. Near-infrared emissive lanthanide hybridized carbon quantum dots for bioimaging applications. Journal of Materials Chemistry B, 4(38): 6366–6372.

DRAFT

ORAL PRESENTATION

Comparison of two different doxorubicin dosages used for cardiotoxicity model

Meral Erdinç[1] *(ORCID ID: 0000-0001-5591-7607)*, Meryem Şeyda Kaya[2*] *(ORCID ID: 0000-0002-3800-8767)*, Zeynep Erdoğmuş Özgen[2] (*ORCID ID: 0000-0001-6607-836X)*, İlker Kelle[1] *(ORCID ID: 0000-0003-3232-7019)*, Fesih Aktar[3] *(ORCID:0000-0002-0760-568)*, Levent Erdinç[4] *(ORCID ID: 0000-0003-4661-9784)*,
Ayşe Nur Keleş[5] *(ORCID ID: 0000-0003-2617-159)*

[1]Dicle Üniversitesi, Tıp Fakültesi, Farmakoloji ABD, Diyarbakır, Türkiye.
[2]Dicle Üniversitesi, Eczacılık Fakültesi, Farmakoloji ABD, Diyarbakır, Türkiye.
[3]Dicle Üniversitesi, Tıp Fakültesi, Çocuk Sağlığı ve Hastalıkları ABD, Diyarbakır, Türkiye.
[4]Dicle Üniversitesi, Tıp Fakültesi, Biyokimya ABD, Diyarbakır, Türkiye.
[5]Dicle Üniversitesi, Tıp Fakültesi, Tıbbi Patoloji ABD, Diyarbakır, Türkiye.

*Corresponding author e-mail: meryemseydakaya@gmail.com

Abstract

Doxorubicin, an anthracycline antibiotic, is commonly used antineoplastic agent against different solid tumors and hematological malignancies. The most serious dose limiting side effect is cardiotoxicity which can occur in both acute and chronic form and characterized with loss of cardiac functions, arrythmia, and heart failure. Because of wide spectrum usage of doxorubicin, there are numerous studies in literature about molecular mechanisms of doxorubicin induced cardiotoxicity. In these studies, rat or mice models are commonly employed due to their cardiovascular system closely resembling to human. These studies utilize varying dosages and dosing schedules to assess the extent of doxorubicin's cardiotoxicity. However, in the current study, the aim is to focus on comparing two specific dosing schemes. Male Sprague-Dawley rats were grouped as; 1) Control group; 2) DoxA group (5 mg/kg i.p. injections on day 1, 3, 5), and 3) DoxB (10 mg/kg i.p. single injection on day 1). In all groups, firstly blood samples were taken intracardially and then isolated hearts were perfused by Langendorff system on day 7. Heart coronary perfusion pressure and heart rate per minute were measured. Moreover, the left ventricle of the heart tissue samples was fixed in formalin 10% for histopathological studies. Our findings indicate that a single 10 mg/kg doxorubicin injection increased coronary perfusion compared to DoxA group ($p<0.05$). Furthermore, creatine kinase levels in blood also increased significantly more in DoxB group ($p<0.05$). MDA analysis showed that both dosing schemes significantly increased MDA levels by comparing with control group ($p<0.05$), with no statistically significant difference between the DoxA and DoxB groups ($p>0.05$). The histopathological analysis further confirmed that DoxB grouped sustained more severe damage compared to DoxA group. In summary, the present study provides comprehensive evidence that a single 10 mg/kg doxorubicin dose induces more severe cardiotoxic effects than repeated doses.

Keywords: Doxorubicin, Cardiotoxicity, Perfusion Pressure

INTRODUCTION

Doxorubicin, an anthracycline antibiotic obtained from Streptomyces peucetius var. Caesius, is one of the commonly used antineoplastic agents against several solid tumors and hematologic malignancies in both adult and children (Sawicki et al.,2021). The major dose limiting side effect of doxorubicin is cardiotoxicity which clinically characterized with deterioration of cardiac functions, arrhythmia, cardiomyocyte function loss, heart failure. Besides these irreversible and deleterious side effects, doxorubicin also causes nephrotoxicity, neurotoxicity, and hepatotoxicity (Octavia et al., 2012). Due to the fact that doxorubicin is an important wide spectrum chemotherapeutic agent, there are several studies to understand the molecular mechanism of its cardiotoxicity and ameliorate cardiac functions such as designing of different analogues or co-usage of various antioxidants and iron chelators (Li et al., 2000, Cusack et al., 2006; Hole et al., 2014; Ozgen et al., 2016 & 2022; Yu et al., 2020). It has been proven that molecular mechanism of doxorubicin cardiotoxicity includes activation of programmed cell death pathways induced by elevated oxidative stress and reactive oxygen species

levels. However, the exact mechanism has not been solved yet and all the previous attempts only provided limited alleviation on cardiac functions. Therefore, the mechanism of doxorubicin cardiotoxicity remains still unclear and there are still numerous ongoing studies on preclinical phase (Rawat et al., 2021).

In the studies about doxorubicin cardiotoxicity, laboratory animal models are commonly employed due to possessing very similar and intact cardiovascular system to human and they could provide remarkably valuable information especially about the effects of experimental therapeutics. Therefore, establishment of high degree doxorubicin cardiotoxicity in laboratory animal models is highly essential (Hayward et al., 2007). In a literature survey, it can be easily found that there are several different dosages and dosing schedules to produce doxorubicin cardiotoxicity in laboratory animal models (Ozgen et al., 2016; Hole et al., 2014; Yu et al., 2020). The dosing schedules can be mainly divided into two groups: single dose injection and repeated injections. Both dosing schedules generated different levels of cardiotoxicity in animal models (Najafi et al., 2020). The aim of the current study is to investigate the difference between these two main groups about the establishment of doxorubicin cardiotoxicity by using the Langendorff model in regard to physiological, biochemical and histopathological manners.

MATERIALS AND METHODS

Doxorubicin Hydrochloride were purchased from Sigma Aldrich (Germany); Ketamin Hydrochloride from Pfizer (USA); Ksilazin from Bayer (Germany); and Heparin from Roche (Switzerland). Unless otherwise mentioned, all other reagents were purchased from Sigma Aldrich (Germany).

Pharmacological Experiments

Thirty-two adult male Sprague-Dawley rats, 250-300 g in weight were purchased from the Department of Medical Science Application and Research Centre of Dicle University (DUSAM), Diyarbakır. All rats were fed with standard diet and tap water, and all experiments were performed according to the "Guide for Care and Use of laboratory Animals"(National Institutes of Health publication 85-23, revised 1985). The experimental protocol was approved by the Dicle University Animal Research Ethics Committee (DUHADEK 2018/21).

The animals were divided into 3 groups (n=8);
Control Group: single intraperitoneal (i.p.) injection of 1 ml of sterile saline on day 1 and sacrificed on day 7.
Doxorubicin A (DoxA) Group: 5 mg/kg i.p. of doxorubicin injection respectively on day 1, 3, 5 and sacrificed on day 7.
Doxorubicin B (DoxB) Group: single i.p. injection of 10 mg/kg of doxorubicin on day 1 and sacrificed on day 7.

In all groups, seven days after first injection, rats were anesthetized with intramuscular injection of 100 mg/kg ketamine and 15 mg/kg xylazine. After heparinization with 500 IU/kg through femoral vein for purpose of preventing a coagulation during procedure, hearts were isolated and immediately cannulated through the aorta and placed to the Langendorff system. In this system, hearts were perfused with Krebs solution (aspirated with 5% CO2 and 95% O2 mix) at 11 ml/min constant flow rate. After 30-45 min stabilization period, perfusion pressure values were measured by pressure transducer located at infusion cannula and heart rates were recorded through electrodes placed on isolated heart. All the measured values were recorded by Biopac MP 30 software (Biopac Systems).

Biochemical Analysis

In all groups, the obtained heart tissue samples were stored at -80oC until further experiments. Tissue malondialdehyde (MDA) levels, an indicator of lipid peroxidation, were measured by the thiobarbutyric acid (TBA) method in non-perfused heart samples of all groups. In this method, firstly heart tissue samples were homogenized by adding cold TCA mixture (1 ml of 10% TCA and 8 ml of 5% TCA per 1 g tissue) and then centrifuged at 13000 rpm for 20 minutes. Next to collecting supernatant into a new eppendorf tube, an equal volume of 0,67 % TBA solution was added and heated to 100oC for 10 minutes. 1,1,3,3-tetraethoxypropane, standard for MDA analysis, was also studied under same conditions. Finally, UV absorptions of all samples were measured at a wavelength of 532 nm by using spectrophotometer. The absorption spectrum was recorded

in the range of 480-600 nm. The MDA concentrations were calculated using the molarity coefficient (1.56x105) reported by Jordan and Scenkman. The results were defined as nmol/gr of tissues.

Histopathological Studies

In all groups, the left ventricle of the heart samples was fixed in formalin 10% until histopathological studies. The tissue specimens were blocked by embedding in paraffin in the casting device (Thermo, Histostar) after histological tissue follow-up process with an automatic tissue tracking device (Thermo, Excelsior AS). 4-5 µm thick cross-section from the left ventricle of specimens were taken from the paraffined blocks with a microtome device (Leica, RM2245). Next to the deparaffinization, the sections were stained with Hematoxylin-Eosin (H&E) by standard methods in an automatic staining device (Shardon, Varistain Gemini). At the end, the heart samples were examined by a light microscope (Nikon, Eclipse 50i) for histopathological evaluation.

Statistical Analysis

In all experiments, the obtained data were expressed as arithmetic mean ± standard deviation. Statistical analyzes were performed using the SPSS program 11.0 (Chicago, ill., USA). Kruskal Wallis test was used to evaluate the difference between the groups. The difference between the two groups was calculated using the Mann-Whitney U test. Significance level was accepted as $p<0.05$ in all statistical analyzes.

RESULTS

Pharmacological Results

The obtained coronary perfusion pressure and heart rate data from the isolated hearts perfused by Langendorff system according to groups were shown in Figure 1 and Figure 2, respectively. It was found that there was a statistically significant increase of each parameter in both DoxA and DoxB groups compared to the control group ($p<0.05$). When the DoxA and DoxB groups were compared, it was seen that a single dose of 10 mg/kg doxorubicin statistically more increased the coronary perfusion pressure in DoxB group ($p<0.05$). Although there was a more increase in heart rate of DoxB group, it was not statistically meaningful due to $p>0.05$.

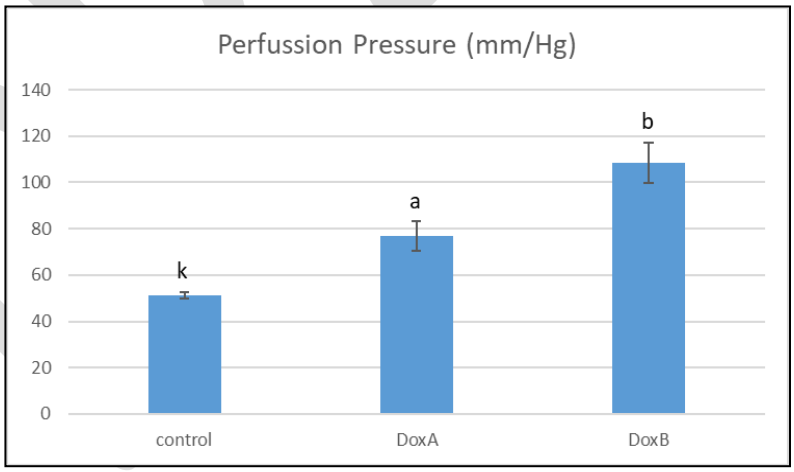


Figure1: Perfusion pressure and heart rate in all groups (a-k $p<0,05$; b-k $p<0,05$; b-a $p<0,05$)

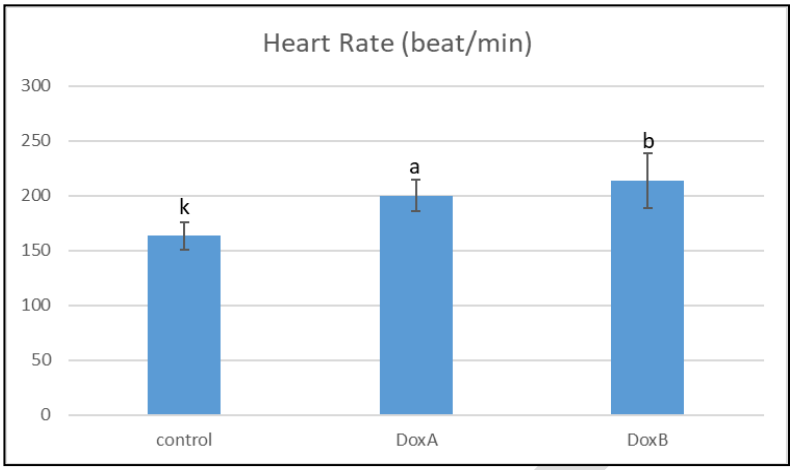


Figure 2: Heart rate (beat/min) in all groups (a-k p<0,05; b-k p<0,05; b-a p>0,05)

Biochemical Results

In Figure 3, the cardiac tissue MDA analysis results, the end product of lipid peroxidation, were shown and it can be inferred that doxorubicin injections significantly increased MDA levels in comparison to control group (p<0,05). When Dox groups were compared, there was not a statistically significant difference between DoxA and DoxB groups (p>0,05)

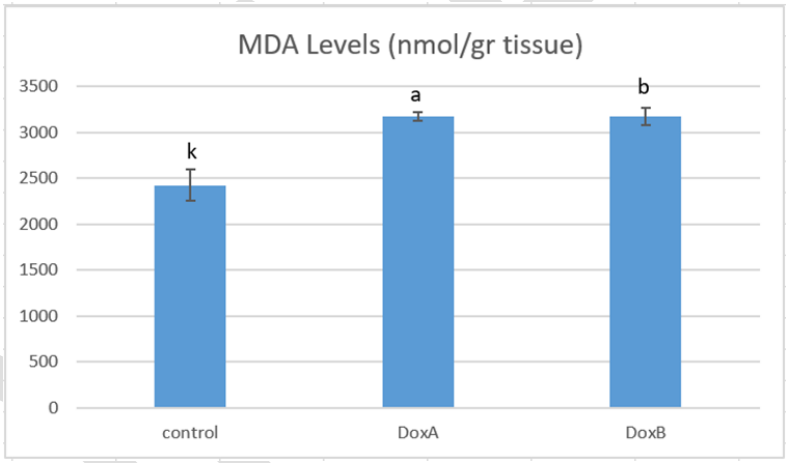


Figure 3: Cardiac tissue MDA levels in all groups (a-k p<0,005; b-k p<0,005; a-b p>0.05)

Histopathological Results

In histopathological examinations, cardiac tissue specimens were evaluated in terms of intracellular vacuolization, nuclear degeneration, cytoskeletal cardiomyocyte degeneration, intercellular edema, neutrophilic and lymphoplasmocytic infiltration, macrophage infiltration, necrosis, apoptosis, fibrosis. As seen in figure 4, doxorubicin injected groups showed significant cardiomyocyte degeneration and focal nuclear degeneration by comparing to control group. While significant cardiomyocyte degeneration characterized by a significant increase in the cytoplasm and a tendency to disperse, loss of cytoskeletal organization and striation, and indistinct cytoplasmic borders and focal nuclear degeneration was observed in DoxB group, only moderate significant cardiomyocyte degeneration was found in DoxA group. Besides that, heterogeneously distributed vesicular chromatin pattern, irregular nuclear membrane and markedly degenerated nuclei characterized by marked enlargement were also demonstrated in DoxB group.

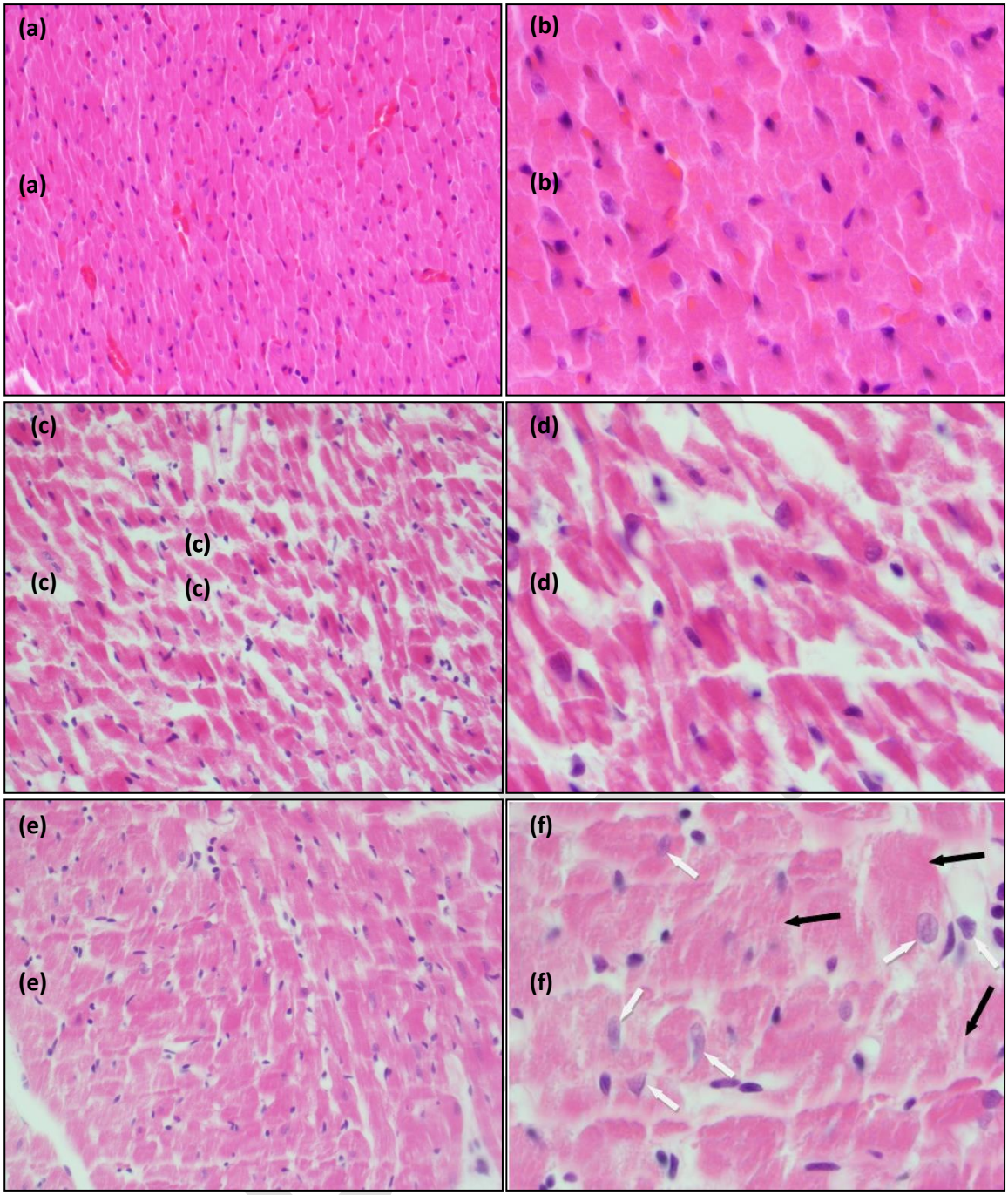


Figure 4: Photomicrographs of hematoxylin and eosin-stained sections of heart tissue. Control group (40x10 and 100x10; a and b, respectively); DoxA group (40x10 and 100x10; c and d, respectively); DoxB group (40x10 and 100x10; e and f, respectively).

DISCUSSION

Doxorubicin is one of the mostly used antineoplastic therapeutic agent against various cancer types. Cardiotoxicity is the most life threating side effect of Doxorubicin both in acute or chronic usage (Sawicki et al., 2021). There are several previous and ongoing studies aiming to understand and improve treatments for the mechanisms of doxorubicin cardiotoxicity (Octavia et al., 2012). Most of these studies usually have been performed by using laboratory animal models thanks to the similarity of their cardiovascular system and being reliable, well-accepted and cost-effective (Hayward et al., 2007).

In literature, different doses and dosing schemes are used for investigation of the mechanism of doxorubicin induced cardiotoxicity. There are two main groups about dosing scheme: single dose or repeated injections

(Najafi et al., 2020). For instance, while repeated injections of 10 mg/kg doxorubicin respectively on day 1, 3, and 5 were performed in the study of Yu et al. in 2020, a single dose of 10 mg/kg doxorubicin was injected on day 1 in the study of Ozgen group (Yu et al., 2020; Ozgen et al., 2022). There are several more examples of different dosing schedules that are recently analyzed in two different comprehensive review (Najafi et al., 2020; Sritharan and Sivalingam, 2021). In this study, the aim was to compare of these two main dosing schedules of doxorubicin by different analyses.

In the first dosing schedule, DoxA group, 5 mg/kg doxorubicin was injected on day 1, 3, and 5; and sacrification was performed on day 7. However, in the second dosing schedule, DoxB group, a single injection of 10 mg/kg doxorubicin was injected on day 1; and sacrification was performed on day 7. In all groups, hearts were isolated, cannulated through aorta and perfused with Krebs-Henseleit solution by Langendorff system. The coronary perfusion pressure values and heart rates were recorded. In accordance with literature, it was found that doxorubicin injection was significantly increased perfusion pressure and heart rate comparing to control group that indicates doxorubicin-induced cardiac dysfunction (Renu et al., 2018, Varga et al., 2015). Elevated tissue MDA levels pointed out that there was a generation of ROS and oxidative stress in induced cardiac damage as in previous studies (Sheibani et al., 2020). After being clear about production of cardiotoxicity in doxorubicin injected groups, two dosing schedules were compared. Firstly, coronary perfusion pressure was statistically significantly higher in DoxB group. When the heart rates were compared, although there was more increment in DoxB group, the statistical analysis revealed that the difference was not significant. Moreover, MDA analysis also indicated that there was not a statistically significant difference between DoxA and DoxB groups that can be concluded that both dosing schedules revealed similar degree of oxidative stress.

Our histopathological findings were also in accordance with both our other results and literature (El-Agamy et al., 2016). It is observed that there is a doxorubicin induced tissue damage such as cardiomyocyte degeneration and focal nuclear degeneration compared to control group. While, every other day injection of doxorubicin in DoxA group generated moderate significant cardiomyocyte degeneration, very significant cardiac damage was observed in DoxB group characterized by a significant increase in the cytoplasm and a tendency to disperse, loss of cytoskeletal organization and striation, and indistinct cytoplasmic borders and focal nuclear degeneration, heterogeneously distributed vesicular chromatin pattern, irregular nuclear membrane and markedly degenerated nuclei.

CONCLUSION

All in all, the present study revealed that a single dose of 10 mg/kg doxorubicin generated higher degree of cardiotoxicity than repeated doses based on perfusion pressure, heart rate, and histological anlysis results; while MDA experiment results indicate a similar level of toxicity.

REFERENCES

Cusack BJ, Gambliel H, Musser B, Hadjokas N, Shadle SE, Charlier H, Olson RD 2006. Prevention of chronic anthracycline cardiotoxicity in the adult Fischer 344 rat by dexrazoxane and effects on iron metabolism. Cancer Chemotherapy and Pharmacology, 58:517–526.

Erdogmus Ozgen Z, Erdinc M, Akkoc H, Kelle I 2016. Protective effects of melatonin on doxorubicin induced cardiotoxicity in isolated rat heart. Eastern Journal of Medicine, 21(3): 119–124.

Erdogmus Ozgen Z, Erdinc M, Kelle İ, Erdinc L, Nergiz, Y 2022. Protective effects of necrostatin-1 on doxorubicin-induced cardiotoxicity in rat heart. Human and Experimental Toxicology, 41:1–8.

El-Agamy DS, Abo-Haded HM and Elkablawy MA 2016. Cardioprotective effects of sitagliptin against doxorubicin- induced cardiotoxicity in rats. Experimental Biology and Medicine, 241(14): 1577–1587.

Hayward R, Hydock DS 2007. Doxorubicin cardiotoxicity in the rat: An in vivo characterization. Journal of the American Association for Laboratory Animal Science, 46(4): 20–32.

Hole LD, Larsen TH, Fossan KO, Limé F, Schjøtt J 2014. Diazoxide protects against doxorubicin-induced cardiotoxicity in the rat. BMC Pharmacology and Toxicology, 15-28.

Li T, Danelisen I, Bello-Klein A, Singal PK 2000. Effects of probucol on changes of antioxidant enzymes in adriamycin- induced cardiomyopathy in rats. Cardiovasc Res 46(3):523–530.

Najafi M, Hooshangi Shayesteh MR, Mortezaee K, Farhood B, Haghi-Aminjan H 2020. The role of melatonin on doxorubicin-induced cardiotoxicity: A systematic review. Life Sciences, 241, 117173.

Octavia Y, Tocchetti CG, Gabrielson KL, Janssens S, Crijns, HJ, Moens, AL 2012. Doxorubicin-induced cardiomyopathy: From molecular mechanisms to therapeutic strategies. Journal of Molecular and Cellular Cardiology, 52(6), 1213–1225.

Rawat PS, Jaiswal A, Khurana A, Bhatti JS, Navik U 2021. Doxorubicin-induced cardiotoxicity: An update on the molecular mechanism and novel therapeutic strategies for effective management. Biomedicine and Pharmacotherapy, 139, 111708.

Renu K, Abilash VG, Tirupathi Pichiah PB, Arunachalam S 2018. Molecular mechanism of doxorubicin-induced cardiomyopathy - An update. European Journal of Pharmacology, 818: 241–253.

Sawicki KT, Sala V, Prever L, Hirsch E, Ardehali H, Ghigo A 2021. Preventing and Treating Anthracycline Cardiotoxicity: New Insights. Annual Review of Pharmacology and Toxicology, 61: 309–332.

Sheibani M, Nezamoleslami S, Faghir-Ghanesefat H 2020. Cardioprotective effects of dapsone against doxorubicin- induced cardiotoxicity in rats. Cancer Chemotherapy and Pharmacology, 85(3): 563–571.

Sritharan S, Sivalingam N 2021. A comprehensive review on time-tested anticancer drug doxorubicin. Life Sciences, 278, 119527.

Varga ZV, Ferdinandy P, Liaudet L, Pacher P 2015. Drug-induced mitochondrial dysfunction and cardiotoxicity. American Journal of Physiology-Heart and Circulatory Physiology, 309(9): H1453–H1467.

Yu X, Ruan Y, Huang X, Dou L, Lan M, Cui J. Dexrazoxane ameliorates doxorubicin-induced cardiotoxicity by inhibiting both apoptosis and necroptosis in cardiomyocytes. Biochemical and Biophysical Research Communications, 523(1): 140–146.

DRAFT

ORAL PRESENTATION

Evaluation of Hydrothermal Production of Magnesium Borate Samples in the Treatment of Industrial Dyes from Wastewater

Busranur Ak[1,*] (*ORCID: https://orcid.org/0000-0002-6695-0477*), Enis Muhammet Gul[2] (*ORCID: https://orcid.org/0000-0001-5067-276X*), Fatma Tugce Senberber Dumanli[3] (*ORCID: https://orcid.org/0000-0002-3257-1524*), Azmi Seyhun Kipcak[4] (*ORCID: https://orcid.org/0000-0003-2068-6065*), Emek Moroydor Derun[5] (*ORCID: https://orcid.org/0000-0002-8587-2013)*

[1,2,4,5] Yildiz Technical University, Faculty of Chemical and Metallurgical Engineering, Department of Chemical Engineering, Istanbul, Türkiye
[3]Istanbul Nisantasi University, Faculty of Engineering and Architecture, Department of Civil Engineering, Istanbul, Türkiye

*Corresponding author e-mail: yaylabusranur@gmail.com

Abstract

A study was carried out to remove the Congo red dyestuff, which is extremely dangerous for human and environmental health, from wastewater by the photodegradation method using a boron mine, widely available in our country. The adsorbent used in the study is Magnesium Borate, which was synthesized by choosing the hydrothermal method from artificial methods. Prepared magnesium borate samples were characterized by methods such as X-ray diffraction (XRD), scanning electron microscopy (SEM) and fourier transform infrared spectroscopy (FT-IR). Dye solutions of the same concentration were prepared, and different masses of Magnesium Borate adsorbent were added to the dye solutions. The removal of Congo Red dye in the UV reactor by the photodegradation method was investigated. The obtained mass scanning results were examined. As a result of the research, it was seen that magnesium borate in different masses provided effective colour removal from wastewater. It has been found that the initial dye concentration and textural structure of the adsorbent play an important role in its adsorption capacity.

Keywords: Wastewater treatment, Congo Red, Magnesium Borate, Photodegradation.

INTRODUCTION

It is well known that organic dyes such as Congo red (CR) are highly toxic, potentially carcinogenic, and do not easily biodegrade (Guo et al., 2019). Excessive use of CR causes environmental pollution and has become a challenging task for environmental protection agencies (Rajesh et al., 2014). As a target in the United Nations World Water Development Report published in 2021; it is aimed to reduce wastewater pollution by 2030 (Nations, 2021).

Colour removal processes of industrial wastewater containing dyestuffs are gaining ecological importance (Kocaer et al., 2002). Before disposal, waste must be adequately treated using environmentally friendly technologies (Rashid et al., 2021). Researchers have put great effort into removing organic pollutants from the environment in order to maintain a clean environment. Various methods have been proposed for the removal of dye waste such as adsorption (Wang et al., 2022), photocatalytic treatment (Liu et al., 2022), electrochemical treatment (Di et al., 2020), and microbial treatment (Rizqi and Purnomo, 2017).

Borate is a kind of important functional material because of its unique and rich structure. For example, magnesium borates might be as antiwear and antifriction additives (Zeng et al., 2008), flame retardant (Zhang et al., 2017), and nanowhiskers (Zhu et al., 2009; Zhu et al., 2014). Magnesium borate compound can be synthesized by artificial methods as well as it can be found naturally. Magnesium borates can be produced artificially by hydrothermal (liquid-state) and thermal (solid-state) methods.

In this study, it is aimed to purify the non-biodegradable Congo Red dye, which is extremely dangerous for human and environmental health, from industrial wastewater by photocatalytic degradation by using magnesium borate synthesized by hydrothermal method. The synthesized Magnesium borate samples were characterized using X-ray diffraction (XRD), scanning electron microscopy (SEM) and fourier transform infrared spectroscopy (FTIR) techniques. CR dye solutions with the same concentration were prepared. Colour removal was investigated by photodegradation method in the UV reactor by adding different masses of Magnesium Borate to the dye solutions.

MATERIALS AND METHODS

The materials used in this study; Magnesium oxide (MgO) document source for magnesium borate production was Merck (99.9%) quality and boric acid (H_3BO_3) boron source was obtained from Etibank Bandırma Boron and Acid Factory. Congo Red dyestuff, which is a red crystalline powder with a molecular weight of 696.68 g/mol, was purchased from AFG Bioscience. Ethanol (C_2H_5OH) was purchased from Honeywell. The pure water used in the experiments was supplied from the "Human Power I+" brand water purification system. Raw materials were used without any pre-treatment.

Magnesium borate production was carried out using certain amounts of magnesium and boron sources with the help of FOUR E'S SCIENTIFIC magnetic hot plate stirrer. For all trials, it was decided to have a mole ratio of 1:6 (Mg:B) (Derun et al., 2015). During production, boric acid mineral was dissolved in 50 ml of distilled water and heated up to the determined temperature of 100°C. Then, magnesium oxide, which is a source of magnesium, was added and the reaction was carried out at 100°C, for 3 hours at a stirring speed of 500 rpm in a magnetic stirrer. While the experiments were carried out in a closed container, the reaction temperatures were kept constant with the temperature control unit.

After the reaction was complete, the products in the reaction vessel were allowed to cool. The samples were thoroughly washed with 96% ethanol and separated from unreacted boric acid by applying a vacuum filtration process. It was then dried in an oven at 40 °C for about 24 hours. The SEM image of the produced Magnesium borate was taken at 20 kV and analysed in the FTIR instrument for chemical bond analysis in the range of 4000-650 cm^{-1}. Experimental parameters for magnesium borate synthesis are given in Table 1.

Table 1. Experimental parameters for magnesium borates synthesis

Mg/B Molar Ratio	Temperature (°C)	Stirring Speed (rpm)	Reaction Time (h)	Dried Temperature (°C)
1:6	100	500	3	40

An example of the produced magnesium borate powders is shown in Figure 1.

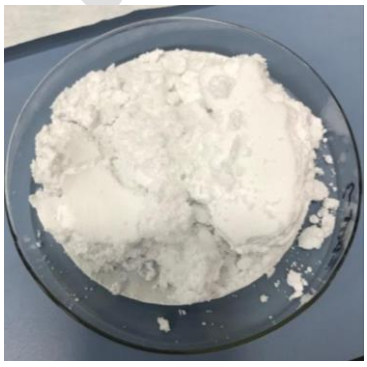


Figure 1. Sample of magnesium borate after drying

Congo Red aqueous dye solutions were prepared at a concentration of 50 mg/L for dye photodegradation in the laboratory. 5 ml of the dye solutions were taken and transferred to quartz tubes. 0.01, 0.03, 0.05, 0.07 and 0.09 g of magnesium borate were weighed into the quartz tube and kept in the Kerman UV 21/13 brand UV reactor for 15 minutes. The wavelength at which the CR dyestuff gave the highest absorbance in the UV vis spectrophotometer device was determined as 498 nm (λ_{max}) in the 400-800 nm range. The relationship between

absorbance and concentration was determined using calibration curves generated at the specified wavelength for Congo Red.

RESULTS and DISCUSSION

XRD Results

The XRD results of the products synthesized by the hydrothermal magnetic stirring method are given in Table-2. The synthesized products emerged as two different magnesium borate minerals. The products obtained are admontite, a magnesium borate mineral containing between seven and fifteen moles of crystalline water. XRD results is given in Table 3.

Table 3. Results of XRD

Identified Phases	Ref. Code	Chemical Formula	Score
Admontite	01-076-0540	$MgO(B_2O_3)_3(H_2O)_7$	54
Admontite	00-034-1438	$Mg_2B_{12}O_{20}15H_20$	29

The XRD pattern of the synthesized magnesium borate sample can be seen in Figure 2.

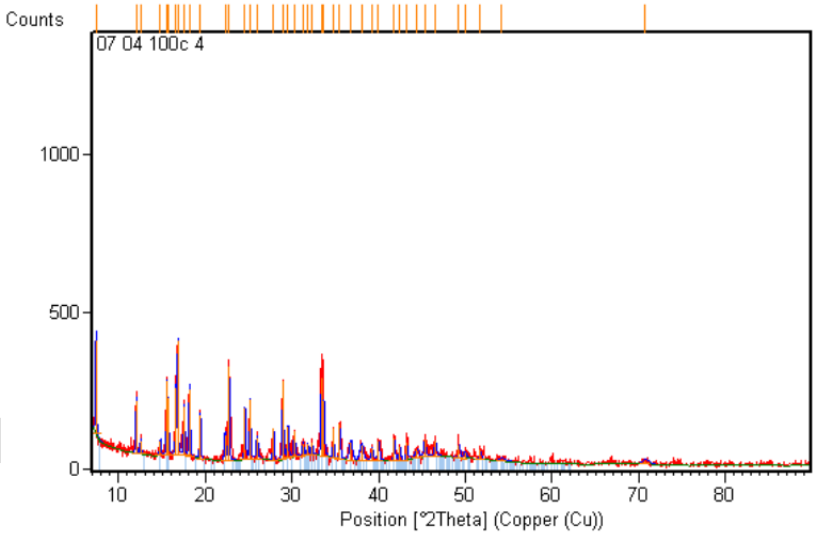
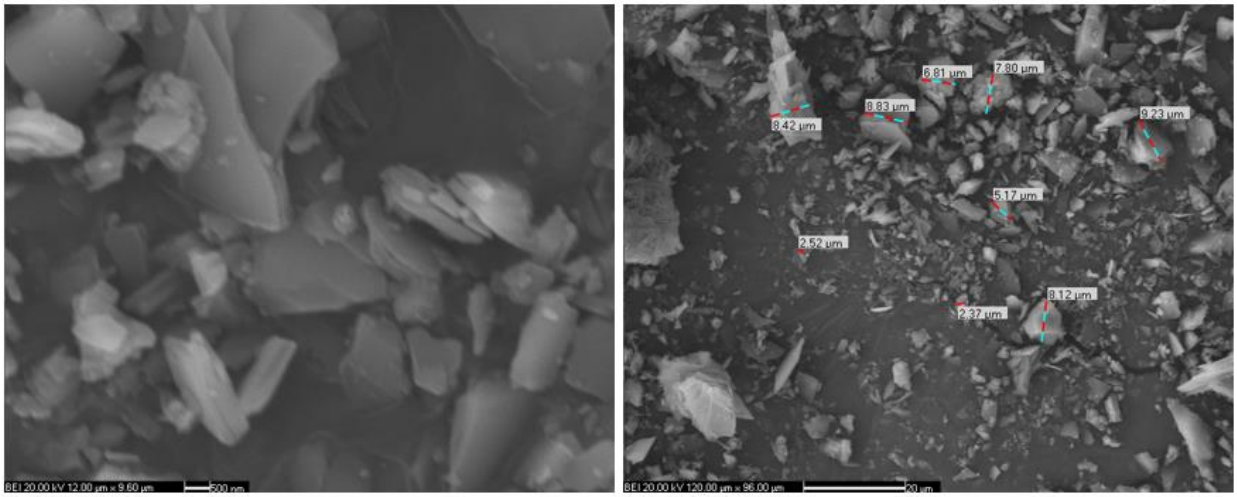


Figure 2. XRD pattern of prepared sample

SEM Results

SEM analysis was performed to determine the surface morphology of the produced Magnesium borate mineral and to perform grain size analysis. The surface morphologies of the produced magnesium borate are shown in Figure 3.



(a) (b)

Figure 3. The surface morphologies of the fabricated magnesium borate (a) 10000X (b) 1000X

When the SEM surface images are examined, the grain size of magnesium oxide varies between 2.52 µm and 9.23 µm. The hydrothermal method was preferred because it facilitates the adjustment of particle size and shape. Clumping was prevented by obtaining a homogeneous appearance.

FTIR Results

The synthesized Magnesium borate was characterized using FTIR. The FTIR spectrum of this synthesized magnesium borate is presented in Figure 4.

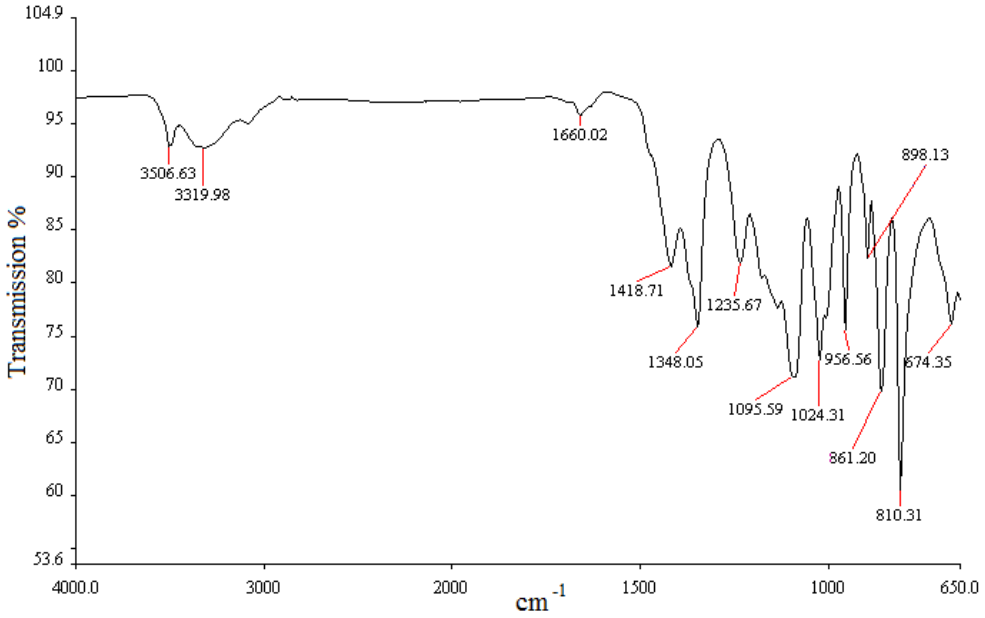


Figure 4. FT-IR Spectrum of the prepared sample

Variation of the Initial Dye Concentration of Congo Red

Variations in the initial concentration of the Congo red dye were performed to determine the optimum photodegradation concentration occurring using magnesium borate. The curve for determining the optimum conditions for photodegradation of Congo red dye on the effect of concentration is shown in Figure 5.

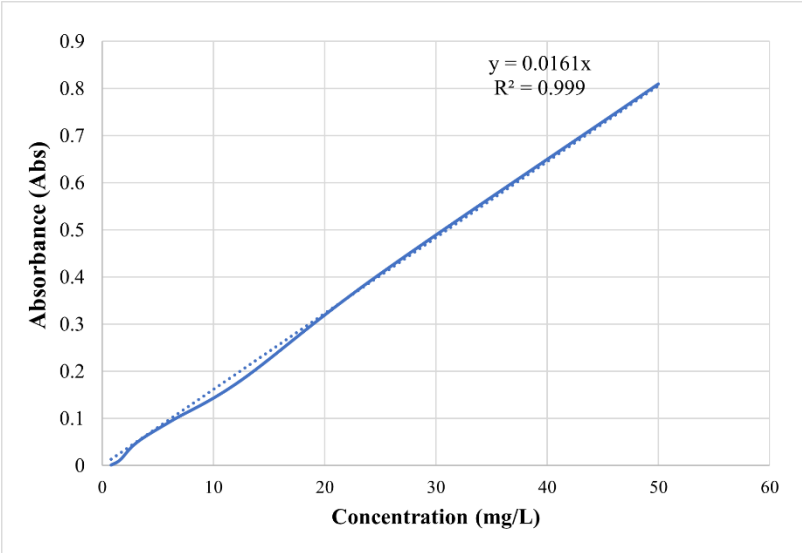


Figure 5. Concentration-Absorbance graph

Effect of Adsorbent Amount on Adsorption

The effect of the amount of adsorbent parameter on the removal of dyestuff with magnesium borate adsorbent was investigated. The working range in this parameter has been determined by means of literature information. When the magnesium borate mass absorbance graph was examined, the maximum removal of Congo Red dye was obtained at the optimum 0.0509 g mass. The results obtained as a result of the photooxidation reaction showed that the UV radiation intensity was effective in removing the colour from the wastewater. The analysis results of the amount of absorbent on the absorption are given in Table 2.

Table 2. Analysis results of the amount of absorbent on the absorption

Wavelength (nm)	Concentration (mg/L)	Weight (g)	Absorbance (Abs)
498 nm	**50 mg/L**	0.0118	0.651
		0.0303	0.609
		0.0509	0.564
		0.0705	0.567
		0.0901	0.596

CONCLUSION

Existing colour removal methods, physical or chemical treatments applied to wastewater containing dyestuffs are effective when the wastewater volume is small. Conventional treatment methods are not sufficient to treat high volumes of water. In this study, the applicability of the photodegradation method in the treatment of industrial wastewater was investigated. The functionality of the use of photochemical processes in the decolorization process of Congo red solutions and the effectiveness of industrially important magnesium borates were examined. It was produced by artificial hydrothermal synthesis method and its contribution to CR photodegradation was investigated. The obtained mass scanning results were examined.

1. As a result of the research, it was seen that magnesium borate in different masses provided effective color removal from wastewater. The impact on the UV Reactor environment allowed degradation of the CR dyestuff. Maximum removal of Congo Red dye at optimum weight was achieved.

2. Magnesium borate was successfully synthesized by hydrothermal method. This was demonstrated by the results of characterizations using XRD, SEM and FTIR.

3. It was found that the initial dye concentration and textural structure of the adsorbent play an important role in the adsorption capacity.

this study, magnesium borate, which is thought to be a potential adsorbent, will be a promising and innovative perspective in terms of wastewater treatment by affecting the photocatalytic degradation of Congo Red dyestuff.

ACKNOWLEDGEMENTS

This research was supported by the Coordination Unit for Scientific Research Projects, Istanbul Nisantasi University (project no. BAP2022/18).

REFERENCES

Derun EM, Kipcak AS, Senberber FT, Yilmaz MS 2015. Characterization and thermal dehydration kinetics of admontite mineral hydrothermally synthesized from magnesium oxide and boric acid precursor. Research on Chemical Intermediates, 41: 853-866.

Di J, Zhu M, Jamakanga R, Gai X, Li Y, Yang R 2020. Electrochemical activation combined with advanced oxidation on $NiCo_2O_4$ nanoarray electrode for decomposition of Rhodamine B. Journal of Water Process Engineering, 37: 1-9.

Guo RF, Ma YQ, Liu ZH 2019. Three hierarchical porous magnesium borate microspheres: A serial preparation strategy, growth mechanism and excellent adsorption behavior for Congo red. RSC Advances, 9: 20009-20018.

Kocaer FO, Alkan U 2002. Boyar madde içeren tekstil atıksularının arıtım alternatifleri. Uludağ Üniversitesi Mühendislik-Mimarlık Fakültesi Dergisi, 7: 47-55.

Liu K, Yang Y, Sun F, Liu Y, Tang M, Chen J 2022. Rapid degradation of Congo red wastewater by Rhodopseudomonas palustris intimately coupled carbon nanotube - Silver modified titanium dioxide photocatalytic composite with sodium alginate. Chemosphere, 299: 1-10.

Nations, U 2021. UN World Water Development Report 2021: Valuing Water. In The United Nations World Water Development Report 2021: Valuing Water.

Rajesh R, Kumar SS, Venkatesan R 2014. Efficient degradation of azo dyes using Ag and Au nanoparticles stabilized on graphene oxide functionalized with PAMAM dendrimers. New Journal of Chemistry, 4: 1551-1558.

Rashid R, Shafiq I, Akhter P, Iqbal MJ, Hussain M 2021. A state-of-the-art review on wastewater treatment techniques: the effectiveness of adsorption method. Environmental Science and Pollution Research, 28: 9050-9066.

Rizqi HD, Purnomo, AS 2017. The ability of brown-rot fungus Daedalea dickinsii to decolorize and transform methylene blue dye. World Journal of Microbiology and Biotechnology, 33: 1-9.

Wang Y, Gong Y, Lin N, Yu L, Du B, Zhang X 2022. Enhanced removal of Cr(VI) from aqueous solution by stabilized nanoscale zero valent iron and copper bimetal intercalated montmorillonite. Journal of Colloid and Interface Science, 606: 941-952.

Zeng Y, Yang H, Fu W, Qiao L, Chang L, Chen J, Zhu H, Li M, Zou G 2008. Synthesis of magnesium borate ($Mg_2B_2O_5$) nanowires, growth mechanism and their lubricating properties. Materials Research Bulletin, 43: 2239-2247.

Zhang L, Liu, ZH 2017. Preparation of $2MgO{\cdot}B_2O_3{\cdot}1.5H_2O$ nanomaterials and evaluation of their flame retardant properties by a thermal decomposition kinetic method. Thermal Analysis and Calorimetry, 129: 715-719.

Zhu W, Wang R, Zhu S, Zhang L, Cui X, Zhang H, Piao X, Zhang Q 2014. Green, noncorrosive, easy scale-up hydrothermal–thermal conversion: a feasible solution to mass production of magnesium borate nanowhiskers. ACS Sustainable Chemistry & Engineering, 2: 836-845.

Zhu W, Zhu S, Xiang L 2009. Successive effect of rolling up, oriented attachment and Ostwald ripening on the hydrothermal formation of szaibelyite $MgBO_2(OH)$ nanowhiskers. CrystEngComm, 11: 1910-1919.

ORAL PRESENTATION

Koyun ve keçilerde meme sağlığı ve hastalıkları

Ayşe Merve Köse[1*] *(ORCID: https://orcid.org/0000-0003-1863-5955)*, Sakine Ülküm Çizmeci[2] *(ORCID: https://orcid.org/0000-0003-2939-8019)*

[1] Hatay Mustafa Kemal Üniversitesi, Veteriner Fakültesi, Doğum ve Jinekoloji Anabilim Dalı, Hatay, Türkiye
[2]Selçuk Üniversitesi, Veteriner Fakültesi, Doğum ve Jinekoloji Anabilim Dalı, Konya, Türkiye

*Sorumlu yazar e-mail: aysemervekose@mku.edu.tr

Özet

Süt; memelilerin yaşamları boyunca en önemli biyolojik sıvılardan biridir ve canlıların yaşam döngülerinde, düzgün büyüme ve gelişmelerini sağlamak için gerekli olan ilk enerji ve besin kaynağı rolünü üstlenir. Sütte bulunan hem makro besinler (proteinler, laktoz ve lipitler) hem de aralarında belirli bir dengenin gözlendiği mikro besinler (vitaminler ve mineraller) yaşamın ilk aşamalarında vücuda fayda sağlamaktadır. Bunun dışında temel gıda maddesi olan süt ve süt ürünleri bu bileşimleri nedeniyle insan beslenmesinde önemli bir yer teşkil etmektedir. Enerji, protein, vitamin ve mineral maddeler içeren sağlık için önemi yüksek bir gıda kaynağı olan süt; doğrudan tüketilebildiği gibi peynir, yoğurt, tereyağı, kaymak gibi ürünlerin yapımında da kullanılmaktadır. Ülkemizde 2022 yılında TUİK verilerine göre çiğ süt üretiminin %92,3'ünü inek sütü, %4,9'unu koyun sütü, %2,5'ini keçi sütü ve %0,2'sini manda sütü oluşturmaktadır. Ülkemizdeki süt üretiminde koyun ve keçi sütünün payı düşük olarak gözükse de, tüketici açısından daha yüksek bir değere sahiptir. Koyun sütü, keçi ve inek sütünden daha yüksek toplam katı madde ve temel besin içeriği içerir. Keçi sütü ise daha iyi sindirilebilme özelliğe sahiptir, tıpta ve insan beslenmesinde bazı terapötik değerlere sahip olması açısından inek veya insan sütünden farklılık gösterir. Ayrıca, özellikle sert iklim koşullarına veya verimsiz topraklara sahip fakir bölgeler ve topluluklar için de önemli bir geçim kaynağıdır. Gıda kaynağı olarak değerlendirilmesi nedeniyle sütün hijyenik ve kaliteli olması gerekmektedir bu da ancak sağlıklı memeye sahip hayvanlardan elde edilebilir. Meme sağlığı, süt verimi ve kalitesini etkileyen en önemli faktör; ineklerde de olduğu gibi küçükbaş hayvanların da en önemli sorunlarından birisi olan mastitistir. Küçükbaş hayvanlarda mastitis; kontrol ve tedavisinin zorluğu, hayvan refahını bozması, ekonomik kayıplara yol açması (hayvanların ölümü, tedavi maliyetleri, sütün tüketim dışı kalması, sütün miktar ve kalitesinin azalması), hijyenik (enfekte sütü tüketerek tüketicilerin enfeksiyon kapması veya zehirlenmesi riski) ve yasal (bakteriyolojik süt kalitesinin tanımları) sebepler nedeniyle önemli bir hastalıktır. Bu çalışma koyun ve keçilerde meme sağlığı ve mastitisin özetlenerek aktarılması amacıyla ele alınmıştır. Bu bildiride küçük ruminantlarda memenin savunma mekanizmaları, mastitisin etiyolojsi, tanısı, tedavisi ve koruma ve kontrol stratejileri ile ilgili bilgiler verilecektir.

Anahtar Kelimeler: Koyun, Keçi, Meme sağlığı, Mastitis

Mammary gland health and diseases in sheep and goats

Abstract

Milk is one of the most important biological fluids throughout the lives of mammals and plays the role of the first source of energy and nutrients necessary to ensure proper growth and development in the life cycle of living things. Both macronutrients (proteins, lactose, and lipids) and micronutrients (vitamins and minerals), among which a certain balance is observed in milk, benefit the body in the first stages of life. Apart from this, milk and dairy products, which are basic foodstuffs, have an important place in human nutrition due to their composition. Milk, which is a food source of high importance for health, contains energy, protein, vitamins, and minerals. While it can be consumed directly, it is also used in the production of products such as cheese, yogurt, butter, and cream. According to TUIK data, in our country in 2022, 92.3% of raw milk production will be cow milk, 4.9% sheep milk, 2.5% goat milk, and 0.2% buffalo milk. Although the share of sheep and goat milk in milk production in our country seems low, it has a higher value for the consumer. Sheep milk contains higher total solids and essential nutrients than goat and cow milk. Goat milk, on the other hand, has better digestibility and differs from cow or human milk in that it has some therapeutic values in medicine and human

DRAFT

nutrition. It is also an important source of income for poor regions and communities, especially those with harsh climatic conditions or infertile soils. Since milk is considered a food source, it must be hygienic and of high quality, and this can only be obtained from animals with healthy mammary glands. The most important factor affecting mammary gland health, milk yield, and quality is mastitis, which is one of the most important problems for small ruminants as well as cows. Mastitis is an important disease in small ruminants due to many reasons, including difficulty in control and treatment, deterioration of animal welfare, causing economic losses (death of animals, treatment costs, milk being out of consumption, decrease in quantity and quality of milk), hygiene (risk of infection or poisoning of consumers by consuming infected milk), and legal requirements (bacteriological milk quality). This study was conducted to summarize mammary gland health and mastitis in sheep and goats. In this paper, information will be given about the defense mechanisms of the mammary gland in small ruminants and the etiology, diagnosis, treatment, protection, and control strategies of mastitis.
Key Words: Sheep, Goat, Mammary gland health, Mastitis

Giriş

Küçükbaş hayvanlarda mastitisin birçok açıdan oldukça önemli olduğu düşünülmektedir. Öncelikle meme bezi hastalıklarının süt veriminde niceliksel ve niteliksel düşüşe yol açması ve tedavi masrafları nedeniyle ekonomik açıdan önemlidir. Ayrıca, ciddi şekilde etkilenen hayvanlar çoğunlukla ölür ya da itlaf edilir. Süt ve süt ürünlerinde hijyen ve güvenlik kriterlerinin önemini de belirtmekte fayda vardır; kontamine olması muhtemel ürünlerin tüketimi nedeniyle tüketiciler arasında hastalık oluşturma riski vardır (Novac ve ark., 2019). Yani süt sığırlarında olduğu gibi, koyun ve keçilerde de meme içi enfeksiyon, hayvanların sağlığı, refahı, üretkenliği ve süt kalitesi açısından büyük bir sorundur. Klinik mastitisin sıklığı sığırlara göre önemli ölçüde düşüktür, ancak birçok işletmede ön sağımın olmaması, düşük dereceli mastitisin gözden kaçmasına neden olur. Koyun ve keçilerde klinik mastitis genellikle sığırlara göre daha şiddetlidir. Sığırlarda olduğu gibi koyun ve keçilerinde de subklinik mastitis meydana gelir ve süt üretiminin azalması ve süt kalitesinin düşmesi gibi benzer etkiler ortaya çıkar (Manning ve ark., 2021). Bu çalışma koyun ve keçilerde meme sağlığı ve mastitisin özetlenerek aktarılması amacıyla ele alınmıştır. Bu bildiride küçük ruminantlarda memenin savunma mekanizmaları, mastitisin etiyolojsi, tanısı, tedavisi ve koruma ve kontrol stratejileri ile ilgili bilgiler verilecektir.

Memenin Savunma Mekanizmaları

Meme dokusunun mastitisten korunması için yeterli bağışıklık fonksiyonları gereklidir. Meme bezi bağışıklığı, bezin anatomik özelliklerinin yanı sıra hücresel ve humoral savunma mekanizması da dahil olmak üzere spesifik olmayan ve spesifik koruma faktörlerinin koordinasyonuna bağlıdır. Meme bezinin savunma mekanizmalarından olan doğuştan gelen ve kazanılmış bağışıklık, meme bezinin enfeksiyonlardan korunmasında hayati bir rol oynar (Stoimenov, 2022). Doğal bağışıklık, genelde spesifik olmayan bağışıklık olarak tanımlanır ve meme başının anatomik bariyeri, makrofajlar, nötrofiller, Natural killer (NK) ve bazı salgısal maddeler tarafından yapılır (Rişvanlı ve ark., 2019).

Meme bezinin anatomik özellikleri

Meme bezinin derisi, meme başı ve meme başı kanalı mikroorganizmalara karşı ilk savunma hattıdır. Meme bezinin sağlıklı derisi; parankimi ve kanal sistemini çevresel faktörlerin etkisinden tamamen izole eder. Normalde sağlıklı bir bezin derisi bakteriyostatik ve bakterisidal etkiye sahip yağ asitleri ile kaplıdır (Alnakip ve ark., 2014). Meme bezinin ana savunma mekanizmalarından biri meme başı kanalıdır. Hem fiziksel bir bariyer hem de antimikrobiyal ajanların kaynağı olarak görev yapar. Meme başı kanal sfinkteri bakterilere karşı fiziksel bir bariyer sağlar. Kanalı çevreleyen düz kas sütün sızmasını önler ve tamamen kapanması sayesinde patojenlerin girişine karşı bir engel oluşturur. Ayrıca meme başı kanalının meme başı sinusuna açılan bölgedeki Fürstenberg rozeti de güçlü mukoza kıvrımlarından oluşmaktadır. Enfeksiyöz ajanların meme bezine nüfuzu çoğu zaman meme başı kanalı yoluyla gerçekleşir çünkü meme bezi ile çevre arasındaki tek doğrudan bağlantı yolu burasıdır (Zecconi ve ark., 2002; Paulrud, 2005; Sudhan ve Sharma, 2010; Kaçar ve Kırşan, 2016).

Meme başı kanalın antibakteriyel özellikleri; iç yüzeyinin, mikroorganizmaların tutunmasını ve tutulmasını ve ayrıca meme bezine daha fazla göçünü önleyen hidrofobik lipitlerle kaplı olmasıyla ifade edilir (Paulrud, 2005; Alnakip ve ark., 2014). Meme başı kanalı antimikrobiyal özelliği olan skuamöz epitel hücrelerinde üretilen serbest yağ asidi içeren balmumu benzeri bir madde olan laktosebum denen keratin tabakası ile kaplıdır (Kaçar ve Kırşan, 2016). Meme bezinin korunmasında bir diğer önemli faktör memenin yapısı ve meme başının konumudur. Örneğin küçük ve horizontal olarak yönlenmiş meme başlarında, ayrıca derin ve sarkık memelerde mastitis riski artmaktadır (Gelasakis ve ark., 2015). İlk savunma mekanizması olan meme başı kanalını geçen patojen bakteriler, ikinci savunma mekanizması olan hücresel savunma ile karşı karşıya gelirler. Doğal

savunmanın bir parçası olan hücresel savunmada nötrofiller, makrofajlar ve lenfositler görev yapar (Rişvanlı ve ark., 2019).

Hücresel savunma özellikleri

Sütteki somatik hücreler; makrofajlar, lenfositler ve polimorfonükleer nötrofiller gibi esas olarak bağışıklık sistemine ait hücrelerdir. Dökülen meme epitel hücreleri de aynı zamanda somatik hücreler olarak sınıflandırılır. Mastitis, meme bezinin esas olarak bulaşıcı mikroorganizmalara karşı inflamatuar tepkisidir. Somatik hücreler, meme içi enfeksiyona karşı inflamatuar yanıtta rol oynar. Somatik hücre sayısı hayvanların sağlık durumunu yansıtır ve sütün kalitesinin belirlenmesinde temel parametre olarak kabul edilir. Koyunların sağlıklı meme bezlerinden elde edilen sütteki bu hücrelerin sayısının, inek sütündeki somatik hücre sayısından daha fazla olduğu bilinmektedir. Somatik hücreleri oluşturan farklı hücre tiplerinin ayırt edilmesi ve tanımlanması da sağlıklı meme bezlerini mastitisli meme bezlerinden ayırmanın önemli bir parçasıdır. Lökositlerle karşılaştırıldığında meme epitel hücreleri somatik hücrelerin daha küçük bir kısmını oluşturur (Stoimenov, 2022). Boutinaud ve Jammes (2002), meme epitel hücrelerinin keçi sütündeki toplam hücre sayısının %26'sını oluşturduğu ifade etmektedir.

Lökositlerin sayısı ve türü, meme bezindeki patojenlerden korunma için önemlidir. Somatik hücrelerin bir parçası olan ve kan dolaşımı yoluyla beze ulaşan lökositlerin hemen her tipi meme bezinde bulunabilir. Lenfositler, makrofajlar ve polimorfonükleer nötrofiller (PMN), meme bezindeki inflamatuar yanıtlarda önemli bir rol oynar. Bu hücreler, mikroorganizmaları tanıma ve hızlı bir bağışıklık tepkisi oluşturma yeteneği sayesinde memenin patojenlere karşı hücresel korumasında rol alır. Sağlıklı bir meme bezindeki somatik hücre değerlerinin $2{,}5x10^5$ hücre/mL süt olmakla birlikte $6x10^5$ hücre/mL sütten daha fazla olabildiği de belirtilmiştir. Mastitis görülmeyen koyunlarda en fazla sayıda makrofajlar (%46-84) bulunur, laktasyonun başında ve ortasında seviyeleri daha yüksekken, sonuna doğru ise azalmaktadır. Somatik hücrelerin toplam sayısını oluşturan diğer hücreler PMN'ler (%2-28), lenfositler (%11-20) ve meme epitel hücreleridir (%1-2) (Katsafadou ve ark., 2019; Stoimenov, 2022).

Mastitis sırasında mukozayı oluşturan hücrelerin apikal yüzeyinde hücresel bağışıklık tepkisini tetikleyen reseptörlerinin varlığı tespit edilmiştir. Epitel hücreleri ayrıca patojeni kaplayan bir zarın (fagozom) oluşmasının neden olduğu fagositik aktivite de gösterir (Günther ve Seyfert, 2018). Meme epitel hücrelerinin meme bezinin korunmasındaki bir diğer önemli rolü de sitokinler, kemokinler gibi çeşitli inflamasyon aracılarının salgılanmasıdır. Meme epitel hücrelerinin koruyucu işlevi, iltihaplanma sürecine dahil olan serum amiloid ve haptoglobin üretimi ile tamamlanır. Ek olarak, meme bezinin bağışıklık savunmasında kritik bir rol oynayan antibakteriyel bir peptit olan katelisidin salgılayarak çeşitli patojenlere yanıt verirler. Bu peptid meme bezindeki meme epitel hücreleri ve nötrofiller tarafından sentezlenir (Cubeddu ve ark., 2017).

Mikroorganizmalara karşı ilk savunma hattını makrofajlar ve nötrofiller sağlar. Enfekte olmayan hayvanlarda somatik hücre sayısının (SHS) çoğunluğunu makrofajlar oluşturur. Makrofajlar meme bezine ulaştıklarında farklılaşan kan monositlerinden köken alır. En belirgin özellikleri fagositik aktiviteye sahip olmalarıdır. Makrofajlar nonspesifik olarak, bakterileri, hücresel döküntü ve birikmiş süt komponentlerini fagosite edebilen aktif hücrelerdir. Ayrıca damarlardan enfeksiyon bölgesine PMN lökositleri çeken bir takım kimyasal maddeler salgılar. Bunlar: sitokinler (tümör nekrozis faktör, interlökinler) ve kemokinler, antimikrobiyal peptitler (β-defensin ve katelisidin) dir (Rişvanlı ve ark., 2019; Stoimenov, 2022). Antijenlerin tanınabilmesi için antijen sunan moleküllere bağlanması gerekir ve ortaya çıkan bu kompleks hücre yüzeyinde yer alır. Antijen sunan moleküller ayrıca sınıf I ve sınıf II ana doku uyumluluk kompleksleri (MHC) olarak da bilinir. Makrofajlar, MHC sınıf II ile birlikte antijenleri işler ve lenfositlere sunarlar, böylece lokal bağışıklık tepkisinde rol oynarlar. Bu sayede kan damarlarının geçirgenliğinde artış sağlayarak kan hücresi elementlerinin ve moleküllerinin yangılı meme bezine nüfuz etmesini sağlarlar (Sordillo, 2005; Stoimenov, 2022).

Meme bezini enfeksiyona karşı koruyan ikinci savunma hattı niteliğindeki Polimorfnükleer lökositler, hücresel savunmanın en önemli hücreleridir. Lökositlerin ana işlevlerinden biri mikroorganizmaları yıkımlamak ve fagosite etmektir. Antikorlar ve komplement sistemi bakterileri opsonizasyon adı verilen bir işlemle bağlayarak nötrofillerin patojenleri sindirmesine aracılık eder. Nötrofiller, *E. Coli*'nin karbonhidrat açısından zengin fimbrialarını tanıyabilen lektin karbonhidrat reseptörleri sayesinde opsonize edici ajanların yokluğunda fagositik aktivitelerini ifade edebilirler. PMN'ler, süperoksit radikalleri ve hidrojen peroksit gibi reaktif oksijen bileşiklerini serbest bırakarak fagosite edilen bakterilerin yok edilmesini sağlar. Nötrofiller bu bileşiklere ek olarak peroksidazlar, lizozim, hidrolitik enzimler ve laktoferrin de salgılar. Bu hücrelerin hücre içi granülleri, defensinler, enzimler (miyeloperoksidaz) ve proteazlar (elastaz, katepsin tip B, D ve G) gibi bakterisidal peptitler içerir. Nötrofiller, mikroorganizmaların lipoprotein zarının yok edilmesinden sorumlu olan katelisidin

sentezleme yeteneğine sahiptir. PMN'ler meme bezinin korunmasında rol oynadıktan sonra apoptoza uğrarlar (Deligönül, 2011; Rişvanlı ve ark., 2019; Stoimenov, 2022).

Lenfositler hücresel savunmada görev alan diğer bir hücre grubudur. Ana temsilcileri T ve B lenfositleri ve doğal öldürücü hücrelerdir (NK hücreleri). Lenfositler, patojenik yapıları membran reseptörleri aracılığıyla tanır ve böylece bağışıklık tepkisine aracılık eder. Lenfositlerin ana işlevi bağışıklık hafızasıdır ve bu nedenle bir patojenle yeniden karşılaşıldığında hızlı bir bağışıklık tepkisi sağlarlar. B-lenfositlerin rolü, bulaşıcı ajanlara karşı antikor oluşumunu teşvik etmektir; bu özelliği patojenlere yönelik spesifik reseptörleri tarafından sağlanır. Hücre içi mikroorganizmalara karşı, enfekte hücreleri öldürerek ve makrofajları aktive eden NK'lar sitokin olan interferon-gamma'yı (IFN-γ) salgılatan özel hücrelerdir. Normal hücre moleküllerini tanıyan reseptörlere sahiptirler. MHC sınıf I molekülü olmayan bütün hücreleri öldürür (Sordillo, 2005; Rişvanlı ve ark., 2019; Stoimenov, 2022).

Hümoral savunma özellikleri

İmmünoglobulinler (Ig), süt ve kolostrumda bulunan en önemli spesifik humoral faktörlerdir. Bunlar meme bezine kan dolaşımı yoluyla girer veya doğrudan meme bezinde üretilebilirler. IgG1, IgG2 ve IgM, bakterilerin opsonizasyonunda rol oynar ve dolayısıyla bu patojenler, meme bezindeki nötrofiller ve makrofajlar tarafından tanınır. İmmünoglobulinler ayrıca kompleman fiksasyonunda da rol oynar, bakterilerin endotelyal hücrelere bağlanmasını önler, mikroorganizmaların aglütinasyonunu teşvik eder, virüsleri ve toksinleri nötralize eder (Sordillo ve Streicher, 2002; Stoimenov, 2022). Lemos ve ark., (2015) sağlıklı bir meme bezindeki immünoglobulin A konsantrasyonlarının 0,027 mg/mL olduğunu ve meme içi enfeksiyon varlığında konsantrasyonların neredeyse üç kat arttığını (0,06 mg/mL) ifade etmiştir.

Laktoferrin, esas olarak meme epitel hücreleri tarafından ve daha küçük miktarlarda nötrofiller tarafından üretilen, demir bağlayıcı bir glikoproteindir. Laktoferrin üretimi alveolar gelişimle ilişkilidir. Laktoferrin esas olarak mevcut demir için bakterilerle rekabet ederek veya bakteriyel yüzeylere bağlanarak bakteriyostatik etki gösterir; bu nedenle Gram-negatif bakterilere (örn., koyun mastitislerinin başlıca etkeni olan *M. haemolytica*) karşı özellikle önemlidir (Sordillo ve Streicher, 2002; Katsafadou ve ark., 2019). Enfekte koyun meme bezlerindeki ortalama laktoferrin konsantrasyonlarının, sağlıklı meme bezlerindeki ortalama laktoferrin konsantrasyonlardan 4,8 kat daha yüksek olduğu gösterilmiştir (Lemos ve ark., 2015).

Lizozim (N-asetilmuramil hidrolaz), meme bezinin antibakteriyel korumasının bileşenlerinden biridir. Lizozim kan dolaşımı yoluyla meme bezine ulaşır veya meme içi enfeksiyonlar sırasında meme bezinde lökositler tarafından sentezlenir. Esas olarak Gram-pozitif bakterilere karşı inhibitör veya litik etkiye sahiptir (Alnakip, 2014). Lizozim aynı zamanda düzenleyici T-lenfositleri aktive ederek epitelyal yüzeylerdeki immün homeostazdaki inflamatuar yanıtın düzenlenmesine de katkıda bulunur (Stoimenov, 2022).

Laktoperoksidaz, karaciğer kökenli tiyosiyanat ve bakteriyel veya endojen kökenli hidrojen peroksit varlığında meme bezinde lokal olarak sentezlenir. Antibakteriyel aktivitesini, lökositlerin bakterisidal aktivitesini artıran bir metabolit olan hipotiyosiyanat gibi aktif oksijen ürünlerinin oluşumu yoluyla gösterir (Katsafadou ve ark., 2019). *Staphylococcus aureus* gibi Gram pozitif bakterilere karşı bakteriostatik, *Escherichia coli* gibi Gram negatif bakterilere karşı ise bakterisidal etkiye sahiptir. Gram negatif bakteriler çok az H_2O_2 üretirler ve bu nedenle laktoperoksidaz sistemi, muhtemelen mastitis kontrolünde önemli değildir. *Streptococcus uberis* gibi Gram pozitif bakterilerin, laktoperoksidaz sistemi için yeterli H_2O_2 üretebildiği ve Gram pozitif bakterilerden kaynaklı mastitis kontrolu için bu enzimin yararlı olduğu bildirilmektedir (Rişvanlı ve ark., 2019). İn vitro koşullar altında sistemin, klinik durumlarda yalnızca kuru dönemde benzer etkinlik gösteren önemli antibakteriyel özelliklere sahip olduğu bulunmuştur; emzirme dönemindeki rolü, muhtemelen diğer süt proteinlerinin müdahalesi nedeniyle sınırlıdır (Katsafadou ve ark., 2019).

Meme savunmasını etkileyen diğer faktörler

Çeşitli faktörler meme bezinin savunma mekanizmalarının azalmasına ve dolayısıyla mastitis gelişmesine yol açmaktadır. Yetersiz beslenme mastitise zemin hazırlayan bir faktördür. A vitamini eksikliği, etkilenen hayvanların meme bezinin epitelyal bariyerinin bütünlüğünü ve işlevini bozarak klinik ve subklinik mastitis riskini arttırmaktadır (Koutsoumpas ve ark., 2013). Yetersiz selenyum konsantrasyonu koyunlarda meme bezinin hücresel korumasında azalmaya yol açar. Vasil ve ark., (2021) gebe koyunlarda oral selenyum ve E vitamini uyguladıkları koyunlarda mastitis vakalarında azalmaya neden olduğunu belirtmektedir. Çinko meme başı keratini ve derisinin bir bileşenidir; çinko eksiklikleri meme kanalının bütünlüğünü olumsuz yönde etkileyebilir ve dolayısıyla bakteri girişini kolaylaştırabilir. Ayrıca enerjinin fagositozu ve bakterilerin lökositler tarafından hücre içi öldürülmesini teşvik etmede önemli bir faktör olduğu kabul edilmiştir; bu bağlamda gebelik toksemisi olan koyunlarda doğum sonrası erken dönemde mastitis gelişme riskinin yüksek olduğu tespit edilmiştir (Barbagianni ve ark., 2015).

Eksik veya aşırı sağım, mikroorganizmaların gelişimi için uygun koşullar sağlaması nedeniyle meme içi enfeksiyon gelişme riskini artıtır. Makineli sağımda mastitis oluşumuna zemin hazırlayan vakum ve titreşim frekansındaki bozukluklar gibi faktörler, sağım sistemlerindeki önemli arızalardır. Sağım makinelerinin yanlış temizlenmesi ve dezenfekte edilmesinin yanı sıra aşırı kullanımı da patojenlerin birikmesine neden olur. Manuel sağımlarda ise meme içi enfeksiyonlara sağımcıların elleriyle bulaşan mikroorganizmalar neden olur, en yaygın olanı ise *Staphylococcus spp*. dir ki küçükbaş hayvanlarda mastitisin ana etiyolojik ajanıdır. Makineli sağım dokulara zarar vererek savunma mekanizmalarını ve bariyerlerini bozar. Emzirilen kuzular, *M. Haemolitica*'nın kuzu bademciklerinden koyunların papiller kanalına taşınmasına neden olmaktadır (Stoimenov, 2022). Küçük ruminantlarda mastitis oluşturan diğer riskler arasında çevresel, genetik ve hayvana bağlı faktörler yer almaktadır. Hayvanın yaşı, barınakların durumu ve temizliği, süt verim miktarı, hayvanın genel sağlık durumu, doğum sayısı, laktasyon dönemi ve laktasyon sayısı da bu faktörler içinde yer alır (Satılmış ve Yeşilkaya, 2023).

Mastitisin Etiyolojsi

Mastitis, sütte fiziksel, kimyasal ve genellikle bakteriyolojik değişiklikler ve glandüler dokularda patolojik değişikliklerle karakterize, meme bezinin iltihabıdır (Mbago, 2022). Mastitise, bakteri, virüs ve mantar gibi çeşitli mikroorganizmaların yanı sıra yaralanmanın da neden olabileceği belirtilmektedir (Mavrogianni ve ark., 2011; Satılmış ve Alkan, 2021). Koyun ve keçilerde, mastitis vakalarının büyük çoğunluğunun bakteriyel kaynaklı olduğu bilinmektedir (Satılmış ve Yeşilkaya 2023).

Mastitis genel olarak klinik ve subklinik olarak ayrılmaktadır (Olechnowicz ve Jaśkowski, 2014). Klinik mastit, belirgin semptomlarla ortaya çıkan meme bezinin bakteriyel enfeksiyonları için kullanılan terimdir. Klinik mastitisin gözle görülür belirtileri arasında bir veya daha fazla memede şişlik, kızarıklık, anormal süt (pıhtı veya serum varlığı) ve ayrıca anoreksi, ateş veya agalaksi gibi diğer semptomlar yer alır. Genellikle klinik mastitisin sonucu memede toksemi ve gangrenöz nekroz görülebilir. Klinik mastitis vakalarının oranı genellikle %5'in altındadır. Çeşitli patojenler mastitise neden olabilir ancak keçi ve koyunlarda meme içi enfeksiyonlarda en sık rastlanan mikroorganizmalar *Staphylococcus spp*. dir (Başatan ve Salar, 2016; Olechnowicz ve Jaśkowski, 2014).

Subklinik mastit, sütteki inflamatuar hücrelerin sayımı ile tespit edilen memenin inflamasyonu ile karakterizedir. Meme bezlerinde herhangi bir klinik anormallik olmayan ve görünüşte normal süt veren, bakteriyolojik olarak pozitif ve SHS'si $\geq 500 \times 10^3$ hücre/mL olan koyunlarda subklinik mastitis olduğu kabul edilir. Kronik mastitis klinik veya subklinik olabilir ancak memenin uzun süre devam eden inflamasyonu ile karakterizedir. Subklinik mastitisler genellikle sürünün genelini etkilemekte ve insidensi %5 ile %30 arasında değişmektedir (Manning ve ark., 2021).

Koyun ve keçilerde mastitise neden olan patojenlerin en önemlisi *Staphylococcus aureus* olmakla birlikte, *Streptococcus*, *Enterobacteriaceae*, *Corynebacteria* türleri, *Pseudomonas aeruginosa*, *Mannheimia haemolytica*, *Lentivirüs* (Visna-Maedi), *Aspergillus fumigatus*, *Serratia marcescens* ve *Burkholdelia cepacia* da mastitise neden olabilmektedir (Bergonier ve ark., 2003; Olechnowicz ve Jaśkowski, 2014; Baştan ve Salar, 2016; Satılmış ve Yeşilkaya, 2023).

Klinik mastitisin sık görülen iki etkeni *Staphylococcus aureus* ve *Mannheimia haemolytica* dır. Emziren koyunlardaki klinik mastitis vakalarının yaklaşık %40'ından *S. aureus* sorumlu iken sağılan koyunlarında yaklaşık %80'inden sorumludur. Aynı zamanda sağılan keçilerde de klinik mastitisin en yaygın nedenidir (Mavrogianni ve ark., 2011). *M haemolytica*, emzirilen koyunlarda klinik mastitis vakalarının yaklaşık %40'ından sorumludur. Emziren koyunlarda ve keçilerde klinik mastitise neden olan diğer bakteriyel ajanlar koagülaz negatif *Stafilokoklardır* (bunlar ayrıca subklinik mastitisin en sık görülen etkenleri), *Escherichia coli* ve *Streptococci*'dir. Mikoplazmanın çeşitli türleri de koyun ve keçilerde mastitise, özellikle de *Mycoplasma agalactia* bulaşıcı agalactia sendromuna (süt kesen) neden olur (Mavrogianni ve ark., 2011; Baştan ve Salar, 2016). Koyun ve keçilerin klinik mastitislerinin %20- %30'unda *Streptococcus spp.* gözlenmektedir. *Streptekoklar* sporadik patojenlerdir ve genellikle kötü barınma koşullarının olduğu ortamlarda daha çok izole edilmektedir (Satılmış ve Yeşilkaya, 2023).

Koyun ve keçilerde koagulaz negatif stafilokok türleri ile birlikte bazı streptekoklar, gram negatif bakteriler ve *Corynebacterialar* subklinik mastitise neden olmaktadır. Koyun ve keçilerde erken laktasyon döneminde Koagulaz negatif *Staphylococcus* (KNS) kaynaklı meme içi enfeksiyonlarda spontan iyileşme oranının %50 olduğu ve enfekte hayvanların yaklaşık %25'inin doğumdan sonraki 6. haftaya kadar enfekte kalabildiği belirtilmektedir. Enfekte meme loblarında SHS'nın sağlıklı meme loblarına göre daha yüksek olduğu, keçilerde subklinik mastitislerde sıklıkla *Corynebacteria spp*., *Streptococcus* spp. ve *S. aureus* gibi diğer patojenler izole edildiği bildirilmektedir (Baştan ve Salar, 2016).

Mastitisin Tanısı

Çoğu durumda, klinik muayene bulgularına dayanarak klinik mastit tanısı konulabilir. Sütçü sürülerde genellikle mastitisli hayvanlar ilk olarak sağım sırasında görülür. Etçi sürülerde koyunların memelerine bu kadar düzenli ve yakın ilgi gösterilmediği için hastalığın erken belirtilerinin gözden kaçma olasılığı yüksektir. Mastitisli hayvanlar, etkilenen meme bezinin olduğu tarafta topallık gösterebileceğinden, bu sürü içerisinde ayırt edilmesine yardımcı olabilir (Fragkou ve ark., 2014).

İnspeksiyon ve palpasyon ile kronik mastitislerin (memede asimetri, sklerozis, apse oluşumu gibi) teşhisi, tedavi uygulanacak veya sürüden çıkarılacak hayvanlar belirlenebilir (Baştan ve Salar, 2016). Sürü sağlığı yönetiminin bir parçası olarak da her laktasyon döneminin sonunda, sürüdeki tüm koyunların memeleri elle muayene edilmelidir. Hastalığın diğer patolojik durumlarla birlikte bulunabilmesi nedeniyle başlangıçta genel bir klinik muayene gereklidir. Hayvanın rektal ısısı ölçülmelidir. Daha sonra memenin detaylı klinik muayenesi yapılmalıdır (Fragkou ve ark., 2014).

Hastalığın etkili tedavisi için neden olan etkenin doğru tanımlanması önemlidir. Sütçü sürülerde mastitis tanısı için altın standart bakteri kültürüdür (Contreras ve ark., 2007; Toquet ve ark., 2021). Bakteriyolojik kültür mastitise neden olan patojenlerin izolasyonu için en uygun teşhis yöntemidir. Mastitis oranı yüksek sürülerde, fazla sayıda enfekte hayvandan çok sayıda süt örneğinde tekrarlı bakteriyolojik muayene yapılması yerine bazı hayvanlardan alınan örneklerde seçici bakteriyolojik testler, kapsamlı numune toplama maliyetini düşürmeye hizmet eder ve bu işletmelerde mastitis kontrol programlarını benimsemesine yardımcı olabilir (Contreras ve ark., 2007; Baştan ve Salar, 2016).

Subklinik mastit, yalnızca sütteki değişikliklerle karakterize edilir ve tespiti için özel testlere ihtiyaç vardır. Subklinik mastitten her zaman şüphelenilmelidir. Sütçü sürülerde süt üretiminin azalmasının başlıca nedenlerindendir (Fragkou ve ark., 2014). Subklinik mastit tanısı, enfeksiyonun (süt örneklerinden mikroorganizmaların izolasyonu) ve/veya meme bezindeki inflamatuar reaksiyonun saptanmasına dayanır. Sütün bakteriyolojik inceleme sonuçlarının doğru yorumlanması için aseptik numune alma çok önemlidir. Aslında subklinik mastitisin en yaygın etiyolojik ajanları olan koagülaz negatif stafilokoklar (Contreras ve ark., 2007; Fthenakis, 2023) aynı zamanda meme derisinde sık görülür. Bu nedenle, toplama işleminin uygun şekilde yapılmaması halinde süt numunesine kolaylıkla bulaşarak yanlış tanıya yol açabilirler (Fragkou ve ark., 2014).

Meme bezindeki inflamatuar reaksiyonun saptanması için en iyi yöntem, sütteki hücresel içeriğin arttığının gösterilmesidir (Olechnowicz ve Jaśkowski, 2014). California Mastitis Test (CMT), sütteki hücre sayısını indirekt saptayan süt içerisindeki somatik hücre sayısının yüksek veya düşük olduğu konusunda subjektif bir fikir veren, ucuz ve kolay bir tanı yöntemi olarak kabul edilen bir testtir. Somatik hücrelerin çekirdeklerindeki DNA ile reaksiyona giren testin içindeki deterjanlı ayıraç DNA'ları birbirlerine yapıştırıp, jel kıvamının oluşmasına yol açmaktadır. Koyunlarda CMT' nin sağım öncesi yapılması önerilmektedir, koyunlarda subklinik mastitis teşhisinde CMT +1 skorunun ideal eşik değer olduğu belirtilmiştir. Keçilerde de CMT'nin mastitis teşhisinde kullanılabilecek ucuz ve kolay bir tanı yöntemi olduğu, mastitis teşhisinde CMT' nin +2 ve üzeri olması gerektiğini belirtilmektedir (Baştan ve Salar, 2016; Manning ve ark., 2021).

Keçi ve koyunlar arasında mastitis teşhisini etkileyen en önemli farklılıklar sütteki SHS ile ilgilidir. Bu farklılıklar temel olarak enfekte olmamış keçi sütlerinde daha yüksek SHS' nin olması ile ilişkilidir. Keçi sütü salgısının daha yüksek apokrin salınımı nedeniyle lökositlerden daha fazla sayıda sitoplazmik partiküller ve epitel hücre içermektedir (Contreras ve ark., 2007). Keçi sütlerinde doğru SHS ölçümü için fluoro-optik elektronik hücre sayımı, Fossomatik gibi DNA spesifik metotlar ya da direkt mikroskobi ile ölçümde ise Pyronin Y-metil ile boyama kullanılmalıdır (Contreras ve ark., 2007; Baştan ve Salar, 2016). Enfekte olmayan sütçü koyunların SHS'si yaklaşık 200.000 ila 400.000 hücre/mL'lik bir değerdedir ya da daha azdır (Olechnowicz ve Jaśkowski, 2014). Normalde sağlıklı meme loblarında SHS'nin $<0,1x10^6$ olduğu görülmektedir. Somatik hücre sayısının $>0,1x10^6$ ve $<0,5x10^6$ bu değerler üzerindeki olması halinde durum mastitis olarak değerlendirilir (Souza ve ark., 2012; Fragkou ve ark., 2014; Satılmış ve Yeşilkaya, 2023).

Keçilerde kızgınlık, sağım mevsimi, süt verimi ve laktasyon aşaması gibi faktörler de SHS'nin artmasına neden olmaktadır (Olechnowicz ve Jaśkowski, 2014). Genellikle enfekte meme lobundan alınan sütteki SHS değerleri > 500.000 hücre/mL (laktasyonun ilk 90 günü) ve > 1.000.000 hücre/mL (laktasyonun ileri dönemleri)' dir. Geç laktasyon döneminde sağlıklı süt keçilerinde SHS 1 ml sütte 1.000.000'dan fazladır. Ticari standart olarak keçi sütlerindeki asgari SHS'yi Amerika Birleşik Devletleri, 1.000.000/mL olarak kabul etmektedir (Souza ve ark., 2012; Baştan ve Salar, 2016; Toquet ve ark., 2021).

Mastitisin Tedavisi ve Kontrolü

Saha koşullarında küçükbaş hayvanlarda, klinik ve subklinik mastit tedavileri birbirinden ayrılmalıdır. Çünkü hayvan başına ortalama tedavi maliyeti, seleksiyon değeri ve beklenen iyileşme karşılaştırıldığında farklılık söz konusudur (Bergonier ve ark., 2003). Klinik mastitis gelişen koyunlar genellikle ciddi şekilde hastadır ve

geliştirilen tedavi protokollerine göre hemen tedavi edilmelidir (Ruegg, 2011). Akut klinik vakalarda ilk amaç hayvanın hayatını, sonrasında ise memesini kurtarmaktır (Bergonier ve ark., 2003).
Koyunlarda *stafilokokal* mastit tedavisinde tilmikosinin tek dozundan (10 mg/kg) beş gün sonra semptomlarda tam bir azalma olduğunu bildirmektedir (Bergonier ve ark., 2003). Koyun ve keçilerde parenteral uygulamadan sonra çeşitli antibiyotiklerin farmakokinetiği incelenmiştir. İntravenöz yolla günde iki kez tobramisin (25 mg/kg) veya apramisin (20 mg/kg); kas içi yolla günde bir kez enrofloksasin (5 mg/kg) veya norfloksasin (10 mg/kg); kas içi yolla günde iki kez tiamulin (25 mg/kg); Florfenikol (20 ila 25 mg/kg) intravenöz veya intramüsküler yolla günde iki kez şeklinde terapötik kullanımlar önerilmektedir (Bergonier ve ark., 2003). Kuru dönem antibiyotik tedavisi koyunlar ve keçilerde mastit insidansını önemli ölçüde azaltmaktadır (Contreras ve ark., 2007; Olechnowicz ve Jaśkowski, 2014; Manning ve ark., 2021). Kuru dönemde uygulanan antibiyotik tedavisinin yüksek prevalans koşullarında genel sistemik uygulamadan ziyade seçici olarak meme içi uygulanması tercih edilebilir (Bergonier ve ark., 2003; Contreras ve ark., 2007; Olechnowicz ve Jaśkowski, 2014). Antibiyotik tedavisi, yeterli ve hijyenik uygulamayı sağlamak için veteriner hekim tarafından gerçekleştirilmelidir (Olechnowicz ve Jaśkowski., 2014). Çünkü, *P. aeruginosa* veya *A. fumigatus*'un şırınga ile kontaminasyonu yoluyla iatrojenik kaynaklı ciddi mastitis salgınları görülebilir (Bergonier ve ark., 2003; Contreras ve ark., 2007). Bu noktada antibiyotiklerin aşırı kullanımının antibiyotik direnci riskini arttıracağı ve bir halk sağlığı sorunu haline gelebileceği de göz önünde bulundurulmalıdır (Contreras ve ark., 2007).
Klinik gangrenöz mastitis insidansının yüksek olduğu durumlarda, piyasada küçükbaş hayvanlarda mevcut olan aşılar klinik grangrenöz mastitise karşı yaygın olarak kullanılmaktadır (Bergonier ve ark., 2003; Contreras ve ark., 2007). Ancak bu aşıların süt inekleri ve koyunlarda farklı koruyucu etkinliklerinin bildirilmesi ve yeni enfeksiyonları önlemedeki yetersizliklerine rağmen, *S. aureus* kaynaklı meme içi enfeksiyon prevalansının yüksek olduğu sütçü sürülerde mastitisten korumadan ziyade klinik semptomları azaltmak için aşıların kullanılması önerilmektedir (Contreras ve ark., 2007). Subklinik enfeksiyonların aksine aşılama ile klinik mastitis prevalansının azaldığı bildirilmektedir (Contreras ve ark., 2007; Olechnowicz ve Jaśkowski, 2014). Ancak mastitisten korunmada küçük ruminantlarda aşılamanın etkinliği konusunda henüz yeterli veriler ve tatmin edici bilgiler yoktur (Baştan ve Salar., 2016). Koyun ve keçilerde kullanım için lisanslı inaktif *S. aureus* içeren bir mastitis aşısı mevcuttur (koyun ve keçiler için VIMCO enjeksiyon emülsiyonu, Hipra Uk & Ireland Ltd) (Manning ve ark., 2021). Uygun şekilde hazırlanmış ölü (atenüe) aşı ile aşılama, *Pseudomonas aeruginosa*'nın neden olduğu yeni enfeksiyonların görülme sıklığını azaltabilir; ancak farklı virülansa sahip birden fazla suşa bağlı salgınlara da neden olabilir (Olechnowicz ve Jaśkowski, 2014). Her durumda aşılama, mastitisi kontrol altına almanın tek yolu olarak düşünülmemelidir. Enfeksiyonun kontrolünü arttırmak için diğer meme sağlığı yönetimi önlemleri de uygulanmalıdır (Vasileiou ve ark., 2019; Manning ve ark., 2021; Fthenakis, 2023).
Sürünün sağlık durumunu iyileştirmek için çiftliğin tamamı sıkı hijyen koşullarına tabi tutulmalıdır. Sağım makinesi standartlarının ve sağımhane sistemlerinin optimize edilmesiyle sütçü koyun sürülerinin meme sağlığının iyileştirildiği ifade edilmektedir (Contreras ve ark., 2007; Ruegg 2011; Fthenakis, 2023). Özellikle sürüde yüksek oranda meme içi enfeksiyon görülmesi durumunda, sağım sırası da dahil olmak üzere süt inekleri için uygulanan rutinlerin çoğu küçükbaş hayvanlar için de geçerlidir. Mekanik sağım sistemlerindeki veya sağım hijyenindeki eksiklikler nedeniyle fırsatçı mikroorganizma olan Koagulaz negatif *Staphylococcus*'un prevalansları artmaktadır (Contreras ve ark., 2007; Ruegg 2011). KNS kaynaklı meme içi enfeksiyonları kontrol etmek için, tüm sağım rutinleri gözden geçirilmeli ve vakum seviyesi, pulzasyon değeri ve oranı, sağım ünitesi başına vakum oranı vb. gibi sağım değişkenlerinin doğru olduğundan emin olmak için sağım ekipmanı periyodik olarak kontrol edilmelidir (Contreras ve ark., 2007; Ruegg, 2011; Manning ve ark., 2021). Küçükbaş hayvanlarda, sağım sonrası meme başı daldırma işlemi çoğunlukla yüksek oranda enfekte olan sürülerde kullanılmış ve yeni meme içi enfeksiyonların önlenmesinde çok etkili bir yöntem olduğu ortaya çıkmıştır (Bergonier ve ark., 2003; Contreras ve ark., 2007; Manning ve ark., 2021). Bununla birlikte, meme başı daldırma dezenfektanının kalite kontrolü çok önemlidir, çünkü bazı sporadik salgınlar, kuaterner amonyum bazlı meme başı daldırma maddesi kullanıldığında koyunlarda *S. marcescens* kaynaklı mastitise neden olabilmektedir (Contreras ve ark., 2007).
Modern süt işletmelerinde, üretim ve sağlık nedenleriyle pastörize kolostrum kullanımı artmaktadır. Bu prosedür, doğum sırasında yenidoğanların annelerinden ayrılmasına, akabinde kolostrum ve patojenlerden arındırılmış sütle beslenmelerine olanak tanıdığından hem oğlakların ya da kuzuların hem de annelerin memelerinin sağlık durumunu arttırır (Contreras ve ark., 2007).
Hayvan kaynaklarının kontrolünde, genel bir kural olarak, özellikle meme anomalileri bulunan, meme içi enfeksiyonu bulunan, uzun süredir devam eden veya tekrarlayan hastalıkları olan koyunların sürüden çıkarılması gerekir (Fthenakis, 2023). Bu, esas olarak subklinik ve hafif kronik mastitis'in kuru dönem

tedavisiyle ve akut veya şiddetli kronik masititis yaşayan hayvanların seleksiyonu ile gerçekleştirilir (Bergonier ve ark., 2003). Kuru dönem tedavisinden sonra, takip eden laktasyonun başlangıcında hala kronik belirtiler gösteren hayvanlar sürüden çıkartılmalıdır (Bergonier ve ark. 2003; Manning ve ark. 2021). Meme derisi, sağım öncesi meme ucunun daldırılmasıyla kontrol edilebilir; sürü büyüklükleri, sağım rutinleri ve muhtemelen toplu süt florası sorunlarının düşük görülme sıklığı nedeniyle bu önlem genellikle göz ardı edilmektedir. Stafilokokal dermatit veya bulaşıcı ektima vakalarında meme antisepsisi, etkilenen hayvanların ve kuzularının izolasyonu ve bazen de antibiyotik tedavisi uygulanmalıdır (Bergonier ve ark. 2003).
Özetle koyun ve keçilerde meme sağlığı koruma ve hastalık kontrolü prosedürü; 1. Hijyen (hem mera hem barınak hem de sağımda temizlik ve hijyen sağlanmalı), 2. Teat dipping, 3. Kuru dönem tedavisi, 4. Düzenli aralıklarla takip (günlük strip kap ile ön sütün muayenesi, aylık CMT testi ile sürü meme sağlığının kontrolü, 5. Seleksiyon (tedavi edilemeyen hayvanlar sürüden çıkarılmalı, gerekirse hasta memeler enfeksiyonun türüne göre köreltilmeli), 6. Sağımda memede süt kalmamasına önem verilmeli, 7. Aşılama (gerekli hallerde sürüye özel aşı hazırlatılmalı) basamaklarını içermektedir (Kocamüftüoglu, 2019; Fthenakis, 2023).

Kaynaklar

Alnakip, M.E., Quintela-Baluja, M., Böhme, K., Fernández-No, I., Caamaño-Antelo, S., Calo-Mata, Barros-Velázquez, J. (2014). The immunology of mammary gland of dairy ruminants between healthy and inflammatory conditions. J. Vet. Med., 31.

Barbagianni, M.S., Mavrogianni, V.S., Katsafadou, A.I., Spanos, S.A., Tsioli, V., Galatos, A.D., Nakou, M., Valasi, I., Gouletsou, P.G., Fthenakis, G.C. (2015). Pregnancy toxaemia as predisposing factor for development of mastitis in sheep during the immediately post-partum period. Small Ruminanat Research, 130, 246-251.

Baştan, A., Salar, S. (2016). Koyun ve Keçilerde Mastitis. Turkiye Klinikleri J Vet Sci Obstet Gynecol-Special Topics, 2(1), 9-17.

Bergonier, D., De Crémoux, R., Rupp, R., Lagriffoul, G., & Berthelot, X. (2003). Mastitis of dairy small ruminants. Veterinary research, 34(5), 689-716.

Boutinaud M., Jammes H., (2002). Potential uses of milk epithelial cells: a review. Reprod. Nutr. Dev. 42, 133-147.

Contreras, A., Sierra, D., Sánchez, A., Corrales, J. C., Marco, J. C., Paape, M. J., & Gonzalo, C. (2007). Mastitis in small ruminants. Small Ruminant Research, 68(1-2), 145-153.

Cubeddu T., Cacciotto C., Pisanu S., Tedde V., Alberti A., Pittau M., Dore S., Cannas A., Uzzau S., Rocca S., et al. (2017). Cathelicidin production and release by mammary epithelial cells during infectious mastitis. Vet. Immunol. Immunopathol. 89, 66-70.

Deligönül, E (2011). Laktasyon dönemdeki kıl keçisinde meme dokusunun hücresel ve sıvısal savunma sistemleri üzerinde histolojik ve immunohistokimyasal çalışmalar. Doktora Tezi, Mustafa Kemal Ünivesitesi, Sağlık Bilimleri Enstitüsü, Hatay.

Fragkou, I. A., Boscos, C. M., Fthenakis, G. C. (2014). Diagnosis of clinical or subclinical mastitis in ewes. Small Ruminant Research, 118(1-3), 86-92.

Fthenakis, G.C. (2023). Recent insights in ovine mastitis. 5. Ulusal-2. Uluslararası Koyun - Keçi Sağlığı ve Yönetimi Kongresi, 27-30 Nisan 2023, Marmaris/Türkiye, pp.61-64.

Gelasakis A.I., Mavrogianni V.S., Petridis I.G., Vasileiou N.G.C., Fthenakis G.C. (2015). Mastitis in sheep-The last 10 years and the future of research. Vet. Microbiol, 181, 136-146.

Günther J., Seyfert, H.M. (2018). The first line of defence: Insights into mechanisms and relevance of phagocytosis in epithelial cells. Semin. Immunopathol, 40, 555-565.

Kaçar, C., Kırşan, İ. (2016). Memenin Savunma Mekanizmaları, Kaymaz, M., Fındık, M., Rişvanlı, A., Köker, A. (eds). Evcil Hayvanlarda Meme Hastalıkları. 1.baskı, Malatya, Medipres, p.41-58.

Katsafadou, A. I., Politis, A. P., Mavrogianni, V. S., Barbagianni, M. S., Vasileiou, N. G., Fthenakis, G. C., & Fragkou, I. A. (2019). Mammary defences and immunity against mastitis in sheep. Animals, 9(10), 726.

Kocamüftüoglu, M. (2019). Koyun ve keçilerde meme sağlığı. Köker A, editör. Yetiştiricinin El Kitabı Çiftlik Hayvanlarında Meme Sağlığı ve Sağımı. 1. Baskı. Ankara, Desen Ofset; p.48-51.

Koutsoumpas A.T., Giadinis N.D., Petridou E.J., Konstantinou E., Brozos C., Lafi S. Q., Fthenakis G.C., Karatzias H. (2013). Consequences of reduced vitamin A administration on mammary health of dairy ewes. Small Ruminant Res., 110, 120-123.

Lemos V.F., Guaraná E.L.S., Afonso J.A.B., Fagliari J.J., Silva P.C., Soares, P.C. de Mendonça C.L. (2015). Effects of intramammary infection on whey proteinograms of sheep during lactation. Pesquisa Veterinaria Brasileira, 35 (3), 230-236.

Manning, A., Vasileiou, N., Crilly, J. P. (2021). Control of mastitis in dairy sheep and goats. Livestock, 26(3), 161-168.

Mavrogianni, V.S., Menzies, P.I., Fragkou, I.A., Fthenakis, G.C. (2011). Principles of mastitis treatment in sheep and goats. Vet Clin North Am Food Anim Pract, 27, 115-20.

Mbago, P. (2022). Bovine mastitis on selected farms in Kamwenge District: prevalence and antibiograms of the causative bacteria. Makerere university.

Novac, C. Ş., Andrei, S., Fit, N. I. (2019). An Overview of Specific Pathogens in Goat Mastitis. Bulletin of the University of Agricultural Sciences & Veterinary Medicine Cluj-Napoca. Veterinary Medicine, 76(2).

Olechnowicz, J. A. N., Jaśkowski, J. M. (2014). Mastitis in small ruminants. Med. Weter, 70(2), 67-72.

Paulrud, C.O. (2005). Basic concepts of the bovine teat canal. Veterinary Research Communications, 29 (3), 215-245.

Rişvanlı, A., Doğan, H., Şafak, T., Öcal, H. (2019). Memenin savunma sistemi: Meme savunmasında meme başı ve meme başı kanalının rolü. Öcal H, editör. İneklerde Mastitis Dışındaki Meme, Meme Başı ve Meme Derisinin Hastalıkları. 1. Baskı. Ankara: Türkiye Klinikleri; p.1-10.

Ruegg, P.L. (2011). Mastitis in small ruminants. In American Association of Bovine Practitioners Conference Proceedings, p. 111-119.

Satılmış, F,. Alkan, H. (2021). Koyun-Keçilerde Meme Sağlığı ve Mastitis. Erdem, H., Çiftci, E., Işık, M. K., ve Yorgancılar, M. Ü. (eds). Kuzu ve Oğlak Kayıplarının Önlenmesinde Koyun Keçi Sağlığı ve Yetiştiriciliği. 1. bs. Ankara, 225-231.

Satılmış, F., Yeşilkaya, Ö.F. (2023). Koyun ve keçilerde meme sağlığı. Alkan F, editör. Sağlıklı ve Sürdürülebilir Koyun ve Keçi Yetiştiriciliği. 1. Baskı. Ankara: Türkiye Klinikleri; p.62-66.

Sordillo, L. M., Streicher, K. L. (2002). Mammary gland immunity and mastitis susceptibility. Journal of mammary gland biology and neoplasia, 7, 135-146.

Sordillo, L.M. (2005). Factors affecting mammary gland immunity and mastitis susceptibility. Livestock Production Science, 98(1-2), 89-99.

Souza, F. N., Blagitz, M. G., Penna, C. F. A. M., Della Libera, A. M. M. P., Heinemann, M. B., Cerqueira, M. M.O.P. (2012). Somatic cell count in small ruminants: Friend or foe. Small Ruminant Research, 107(2-3), 65-75.,

Stoimenov, A. (2022). Defense Mechanisms of Mammary Gland in Sheep–A Review. Tradition & Modernity in Veterinary Medicine, 7(2).

Sudhan N., Sharma, N. (2010). Mastitis- an important production disease of dairy animals," in Gurgoan: Sarva Manav Vikash Samiti, 72-88.

Toquet, M., Gómez-Martín, Á., & Bataller, E. (2021). Review of the bacterial composition of healthy milk, mastitis milk and colostrum in small ruminants. Research in Veterinary Science, 140, 1-5.

Vasil', M., Zigo, F., Lacková, Z., Ondrašovičová, S., Kielb, E. P., & Bujok, J. (2021). Comparison of the effectiveness of selenium and vitamin E supplementation on the health of the mammary gland of sheep. Acta Fytotechnica et Zootechnica, 24, 75-79.

Vasileiou, N. G., Mavrogianni, V. S., Petinaki, E., Fthenakis, G. C. (2019). Predisposing factors for bacterial mastitis in ewes. Reproduction in domestic animals, 54(10), 1424-1431.

Zecconi, A., Hamano, J., Bronzo, V., Moroni, P., Giovannini, G., Piccinini, R. (2002). Relationship between teat tissue immune defences and intramammary infection. Biology of the Mammary Gland, 287-293.

DRAFT

ORAL PRESENTATION

Production of high-value biomaterials from *Chlorella vulgaris* utilizing airlift photobioreactor cultivation

Ugur Tepe[1] (https://orcid.org/0000-0003-0054-8941), Buse Dincoglu[1] (https://orcid.org/0000-0003-0054-8941), Bahar Aslanbay Guler[1] (https://orcid.org/0000-0002-0113-4823), Zeliha Demirel[1] (https://0000-0003-3675-7315), Esra Imamoglu[*1] (https://orcid.org/0000-0001-8759-7388)

[1]University of Ege, Faculty of Engineering, Department of Bioengineering, Izmir, Turkiye

*Corresponding author e-mail: esra.imamoglu@ege.edu.tr

Abstract

In recent years, there has been a lot of interest in high-value bioproducts obtained from microalgae, such as proteins, chlorophyll, carotenoids, and lipids. Microalgae bioproducts can be used in a variety of sectors, including animal feed, agricultural fertilization, natural components for cosmetics, bioactive chemicals in pharmaceuticals, renewable energy sources, and food supplements for nutrition. *Chlorella vulgaris* is one such microalgae that produces these valuable bioproducts on a commercial basis. Extensive studies on the production of high-value bioproducts from microalgae highlight the importance to optimize reactor types for specific microalgae. Among these reactor designs, airlift photobioreactors have various advantages, including the absence of extra components for mixing and the ability to interchange O_2 and CO_2 with aeration gas and medium. In this study, the cultivation of *C. vulgaris* microalgae was investigated in an airlift photobioreactor. The primary goal was to research biomass and high-value biomaterial production. *C. vulgaris* was cultured in an airlift photobioreactor for 7 days, and samples were obtained at regular intervals to assess turbidity, cell count, chlorophyll-a, chlorophyll-b, total carotenoid, and reactive oxygen species (ROS) content. Furthermore, the amount of lipid content was determined at the end of cultivation period. After the harvest, chlorophyll-a productivity, chlorophyll-b productivity, and carotenoid productivity of the *C. vulgaris* incubation were recorded 1.045±0.009, 1.322±0.048, and 0.892±0.005 mg/g dry weight/day, respectively while lipid content of incubated *C. vulgaris* was recorded 0.397±0.006 g/g dry weight. Considering the advantages of airlift photobioreactors, the cultivation of commercial microalgae *C. vulgaris* in an airlift photobioreactor presents great potential for biomass and bioactive material production.

Keywords: Microalgae, Airlift photobioreactor, *Chlorella vulgaris*, bioproduct

1. Introduction

Microalgae are known for producing high-value bioactive metabolites. *Chlorella vulgaris*, as one of those microalgae, has been studied intensively for its potential for producing chlorophyll-a, chlorophyll-b, carotenoids, and lipids (Jiang and Nakano, 2022; Li et al., 2023; Tunali et al., 2020). These metabolites are thought to be a prospective source for commercial product development in a variety of fields, including pharmaceuticals, cosmetics, food, and energy (Sun et al., 2023; Zhuang et al., 2022). Carotenoids could be synthesized from microalgae in various forms, which present different benefits such as anti-inflammatory, antioxidant, and immunomodulatory effects (Zhuang et al., 2022). Mostly due to those benefits, *C. vulgaris* itself has been reported as a fitting choice for human consumption along with its other beneficial applications (Y. Wong et al., 2017). Moreover, *C. vulgaris* also provides high lipid content for biofuel production (Ge et al., 2018). A wide range of applications of microalgae in general resulted in many ways of commercial scale production of microalgae, utilizing bioreactors (Fu et al., 2019). The use of airlift photobioreactors appears to

have certain advantages: it allows for the interchange of CO_2 and O_2 between the medium and the aeration gas; possible cell damage caused by mechanical pumping may be reduced; and circulation is performed without the use of moving components (Xu et al., 2009). Different types of bioreactors offer different advantages and disadvantages, which should be selected based on the target microalgae (Xu et al. 2009). In this study, the cultivation of *C. vulgaris* microalgae was investigated in an airlift photobioreactor. During the production, turbidity, cell count, chlorophyll-a, chlorophyll-b, total carotenoid, and reactive oxygen species (ROS) content of *C. vulgaris* were examined from periodic samples obtained from a 7-day long cultivation. Additionally, lipid content was determined after harvest. The main aim of this study is to investigate the biomass and biomaterial production of *C. vulgaris* in airlift photobioreactor.

2. Material and Methods

2.1. Algal strain and inoculum preparation

Stock culture of *C. vulgaris* was cultivated in the BG11 medium in a 2-L aerated sterile bottle (Demirel et al., 2015). For the preparation of the inoculum, cells from the stock culture were collected and concentrated by centrifugation (4100×g, 5 min), and the supernatant was discarded. The collected cells were transferred and incubated aseptically under continuous illumination (70 $\mu Em^{-2}s^{-1}$) with an aeration rate of 1 vvm in a 2-L airlift photobioreactor containing 1.6 L of BG11 medium.

2.2. Analytical methods

During the incubation period of 7 days, samples were collected at certain times in order to analyze turbidity (Huang et al. 2016), cell count (Cirik and Gökpınar, 1993), chlorophyll-a, chlorophyll-b, total carotenoid (Ajala and Alexander 2022), total protein (Lowry et al., 1951), ROS (Lu et al. 2020; Omidian, Rafiei, and Bandy, 2020), and lipid content (Isleten-Hosoglu, Gultepe, and Elibol 2012). All the experiments were done in triplicate and presented in figures and tables with average values. The specific growth rate and doubling time of the culture were also calculated (Eq. 1, 2) (Imamoglu, Sukan, and Conk Dalay 2007).

$$Specific\ growth\ rate\ (\mu) = \frac{\ln\left(\frac{C_n}{C_0}\right)}{dt} \quad (1)$$

$$Doubling\ time = \frac{\ln(2)}{\mu} \quad (2)$$

C_0 starting from the logarithmic phase, C_n and C_0 are the biomass concentrations (g/L) on the days t_n and t_0 (days), respectively.

3. Results and Discussion

3.1. Growth profiles of *C. vulgaris*

High biomass accumulation carries an important role in lipid production from microalgae (Demirel et al. 2015), which results in many other studies aiming to optimize both parameters in the same system (Liang, Sarkany, and Cui 2009; Ramanna et al. 2014). In this regard, turbidity, cell count, chlorophyll-a, and chlorophyll-b contents of *C. vulgaris* have been detected, along with the specific growth rate and doubling time. Turbidity and cell count of *C. vulgaris* have shown a correlation and steady increase, indicating suitable conditions for cell growth in airlift photobioreactor (Figure 1).

DRAFT

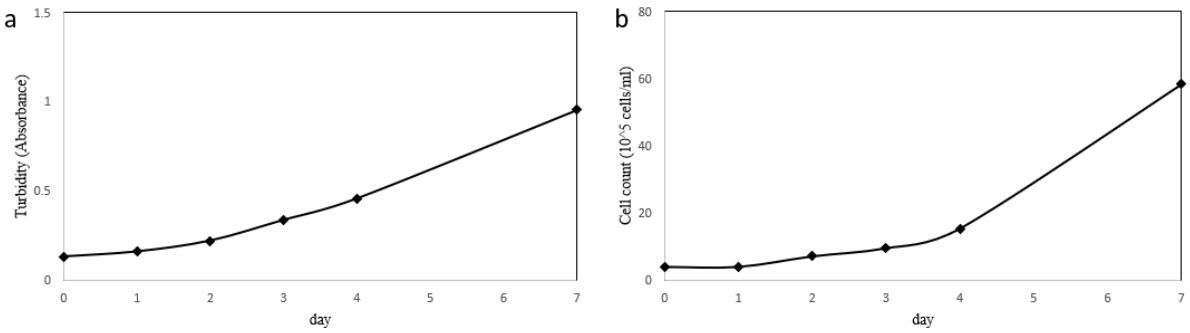


Figure 1. Growth profile of *C. vulgaris*. a: Turbidity (Absorbance); b: Cell count (10^5 cells/ml)

Aside from being a valuable component, chlorophyll contents are known indicators of photosynthesizing microorganism growth in any given medium (Doering, Chamberlain, and Haunert 2006), which is *C. vulgaris* in this study. Periodic examination of chlorophyll-a and chlorophyll-b content presents a constant rise, again indicating a successful incubation of *C. vulgaris* in airlift photobioreactor (Figure 2). It is also important to point out that chlorophyll-b synthesis includes chlorophyll-a (Eggink et al. 2004), which is possibly the reason for the stagnation period in chlorophyll-b accumulation during the first days of the incubation.

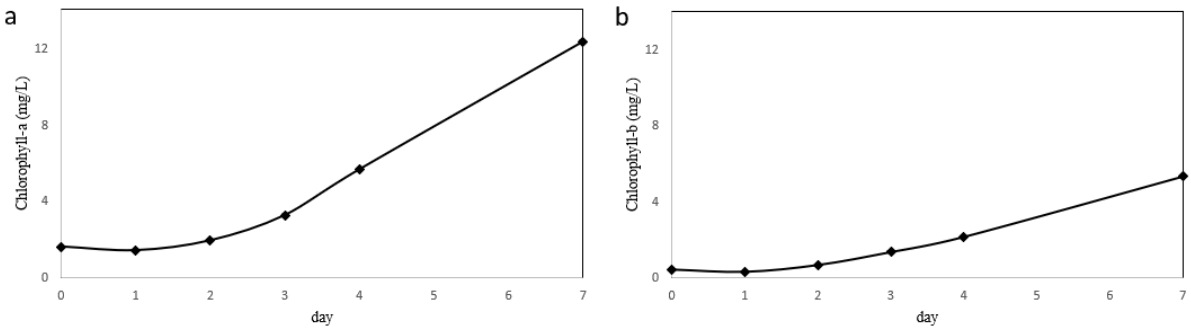


Figure 2. Chlorophyll content of *C. vulgaris*. a: Chlorophyll-a (mg/L); b: Chlorophyll-b (mg/L)

Moreover, the specific growth rate and doubling time of *C. vulgaris* incubation in airlift photobioreactor have been investigated and noted to be 0.516 ± 0.025 day^{-1}, and 1.345 ± 0.066 day, respectively (Table 1). In a study conducted by Balan and Suraishkumar in 2014, researchers cultivated *C. vulgaris* in various illumination conditions and reported specific growth rates of 0.551, 0.567, and 0.383 day^{-1} which correlates with the findings of our study.

3.2. Bioactive material production from *C. vulgaris*

Chlorophyll, carotenoids, and lipids are some of the valuable bioactive materials that can be obtained from *C. vulgaris* (Ge et al. 2018; Jiang and Nakano 2022; Levasseur et al. 2018). Periodic examination of the carotenoid and ROS content of *C. vulgaris* incubation has shown a gradual increase throughout the incubation (Figure 3). The increase in ROS content is an indicator of oxidative stress (Hazeem et al. 2019), which is expected to accumulate during the incubation due to nutrient starvation throughout the incubation (Elisabeth, Rayen, and Behnam 2021). On the other hand, stable ROS levels during the first days of incubation show that the airlift photobioreactor is a fitting tool for *C. vulgaris* incubation.

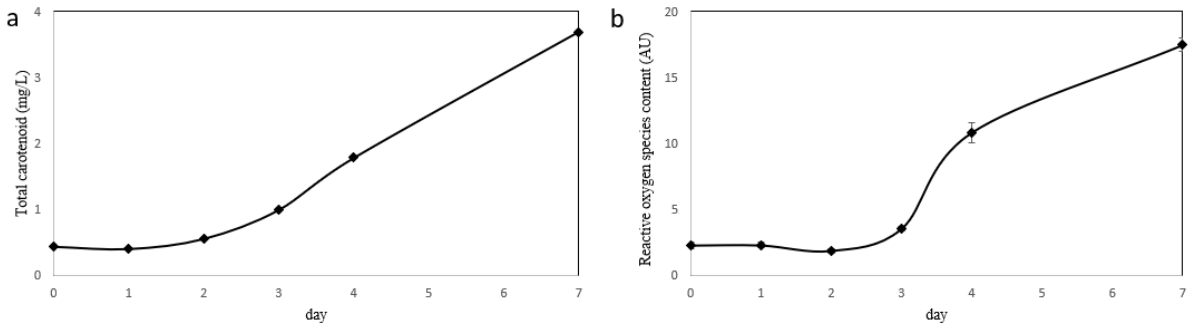


Figure 3. Total carotenoid and reactive oxygen species content of *C. vulgaris*. a: Total carotenoid (mg/L); b: Reactive oxygen species (Arbitrary Unit)

After the harvest, chlorophyll-a productivity, chlorophyll-b productivity, and carotenoid productivity of the *C. vulgaris* incubation were recorded 1.045±0.009, 1.322±0.048, and 0.892±0.005 mg/g dry weight/day, respectively while lipid content of incubated *C. vulgaris* was recorded 0.397±0.006 g/g dry weight (Table 1). In a different study conducted by Y. K. Wong et al. in 2017, researchers implemented varying nitrate/phosphate ratios on a *C. vulgaris* incubation medium in order to increase lipid accumulation. The study of the stated research presents a maximum of 53.202 g/g dry weight lipid content, which is higher than the results of this experiment mainly due to the optimization of the process. Similarly, in another study conducted by Ali et al. in 2021, researchers implemented increased salinity and light-intensity stress factors on *C. vulgaris* incubation in order to induce biomaterial production. Researchers have reported significant increases in carotenoid content up to 4.37±0.33 g/L, which is 1.18 times higher than our findings.

Table 1. Bioactive products and growth profiles of *C. vulgaris*

Microalgae	*C. vulgaris*
pH	7.65
Chlorophyll a productivity (mg/g/day)	1.045 ± 0.009
Chlorophyll b productivity (mg/g/day)	1.322 ± 0.048
Specific Growth Rate (day^{-1})	0.516 ± 0.025
Doubling Time (day)	1.345 ± 0.066
Carotenoid productivity (mg/g/day)	0.892 ± 0.005
Lipid content (g/g dw)	0.397 ± 0.006

4. Conclusion

C. vulgaris incubation in photobioreactors presents a vast potential for bioactive compound production. In this study, *C. vulgaris* was incubated in an airlift photobioreactor for 7 days in order to obtain valuable bioproducts namely chlorophyll-a, chlorophyll-b, carotenoids, and lipids. It has been observed that an airlift photobioreactor is a valid choice for the production of both *C. vulgaris* biomass and stated bioproducts. However, aeration speed and mixing rates are tied to each other in airlift bioreactor systems. The correlation of those parameters could limit the aeration rate due to a possible increase in shear stress during incubation, which makes incubation with airlift photobioreactor non-fitting for every microalgae. In other cases such as *C. vulgaris*, airlift photobioreactors offer less energy usage due to not requiring additional mixing equipment which makes them suitable for industrial-scale production. For this reason, further experiments for

optimization in aeration, light intensity, and medium modifications are required to increase the yield of targeted products in airlift photobioreactor incubation of *C. vulgaris*.

5. Acknowledgements

This study was a part of Cost Action CA20127 and the authors would like to thank the Scientific and Technological Research Council of Turkey (TUBITAK) with project number 121R100 for the financial support.

6. References

Ajala, Sheriff Olalekan, and Matthew L. Alexander. 2022. "Evaluating the Effects of Agitation by Shaking, Stirring and Air Sparging on Growth and Accumulation of Biochemical Compounds in Microalgae Cells." *Biofuels* 13(3):371–81. doi: 10.1080/17597269.2020.1714161.

Ali, Hamdy Elsayed Ahmed, Eman A. El-fayoumy, Wessam E. Rasmy, Ramadan M. Soliman, and Mohd Azmuddin Abdullah. 2021. "Two-Stage Cultivation of Chlorella Vulgaris Using Light and Salt Stress Conditions for Simultaneous Production of Lipid, Carotenoids, and Antioxidants." *Journal of Applied Phycology* 33(1):227–39. doi: 10.1007/s10811-020-02308-9.

Balan, Ranjini, and G. K. Suraishkumar. 2014. "Simultaneous Increases in Specific Growth Rate and Specific Lipid Content of Chlorella Vulgaris through UV-Induced Reactive Species." *Biotechnology Progress* 30(2):291–99. doi: 10.1002/btpr.1854.

Cirik, S., and S. Gökpınar. 1993. *Plankton Bilgisi ve Kültürü*. 47th ed. İzmir: University of Ege.

Demirel, Zeliha, Esra Imamoglu, and Meltem Conk Dalay. 2015. "Fatty Acid Profile and Lipid Content of Cylindrotheca Closterium Cultivated in Air-Lift Photobioreactor." *Journal of Chemical Technology and Biotechnology* 90(12):2290–96. doi: 10.1002/jctb.4687.

Doering, Peter H., Robert Chamberlain, and Kathleen M. Haunert. 2006. "Chlorophyll a and Its Use as an Indicator of Eutrophication in the Caloosahatchee Estuary, Florida." *Biological Sciences* 69.

Eggink, Laura L., Russell LoBrutto, Daniel C. Brune, Judy Brusslan, Akihiro Yamasato, Ayumi Tanaka, and J. Kenneth Hoober. 2004. "Synthesis of Chlorophyll b: Localization of Chlorophyllide a Oxygenase and Discovery of a Stable Radical in the Catalytic Subunit." *BMC Plant Biology* 4:1–16. doi: 10.1186/1471-2229-4-5.

Elisabeth, Bermejo, Filali Rayen, and Taidi Behnam. 2021. "Microalgae Culture Quality Indicators: A Review." *Critical Reviews in Biotechnology* 41(4):457–73. doi: 10.1080/07388551.2020.1854672.

Fu, Jingwei, Yun Huang, Qiang Liao, Ao Xia, Qian Fu, and Xun Zhu. 2019. "Photo-Bioreactor Design for Microalgae: A Review from the Aspect of CO2 Transfer and Conversion." *Bioresource Technology* 292:121947. doi: 10.1016/j.biortech.2019.121947.

Ge, Shijian, Shuang Qiu, Danielle Tremblay, Kelsey Viner, Pascale Champagne, and Philip G. Jessop. 2018. "Centrate Wastewater Treatment with Chlorella Vulgaris: Simultaneous Enhancement of Nutrient Removal, Biomass and Lipid Production." *Chemical Engineering Journal* 342:310–20. doi: 10.1016/j.cej.2018.02.058.

Hazeem, Layla J., Gamze Kuku, Etienne Dewailly, Christian Slomianny, Alexandre Barras, Abderrahmane Hamdi, Rabah Boukherroub, Mustafa Culha, and Mohamed Bououdina. 2019. "Toxicity Effect of Silver Nanoparticles on Photosynthetic Pigment Content, Growth, ROS Production and Ultrastructural Changes of Microalgae Chlorella Vulgaris." *Nanomaterials* 9(7). doi: 10.3390/nano9070914.

Huang, Yuanxing, Shengnan Qin, Daofang Zhang, Liang Li, and Yan Mu. 2016. "Evaluation of Cell Disruption of Chlorella Vulgaris by Pressure-Assisted Ozonation and Ultrasonication." *Energies* 9(3):1–11. doi: 10.3390/en9030173.

Imamoglu, Esra, Fazilet Vardar Sukan, and Meltem Conk Dalay. 2007. "Effect of Different Culture Media and Light Intensities on Growth of Haematococcus Pluvialis." *International Journal of Natural and Engineering Sciences* 1(3):5–9.

Isleten-Hosoglu, Muge, Idil Gultepe, and Murat Elibol. 2012. "Optimization of Carbon and Nitrogen Sources for Biomass and Lipid Production by Chlorella Saccharophila under Heterotrophic Conditions and Development of Nile Red Fluorescence Based Method for Quantification of Its Neutral Lipid Content." *Biochemical Engineering Journal* 61:11–19. doi: 10.1016/j.bej.2011.12.001.

Jiang, Mengqi, and Shin ichi Nakano. 2022. "New Insights into the Stoichiometric Regulation of Carotenoid Production in Chlorella Vulgaris." *Bioresource Technology Reports* 20:101227. doi: 10.1016/j.biteb.2022.101227.

Levasseur, Wendie, Behnam Taidi, Robin Lacombe, Patrick Perré, and Victor Pozzobon. 2018. "Impact of Seconds to Minutes Photoperiods on Chlorella Vulgaris Growth Rate and Chlorophyll a and b Content." *Algal Research* 36:10–16. doi: 10.1016/j.algal.2018.10.007.

Li, Suiyi, V. Meenakshi, S. Nithya, Sulaiman Ali Alharbi, Saleh H. Salmen, Rajasree Shanmuganathan, Li Zhang, and Changlei Xia. 2023. "Impact of the Combined Effect of Seawater Exposure with Wastewater and Fe2O3 Nanoparticles on Chlorella Vulgaris Microalgae Growth, Lipid Content, Biochar, and Bio-Oil Production." *Environmental Research* 232:116300. doi: 10.1016/j.envres.2023.116300.

Liang, Yanna, Nicolas Sarkany, and Yi Cui. 2009. "Biomass and Lipid Productivities of Chlorella Vulgaris under Autotrophic, Heterotrophic and Mixotrophic Growth Conditions." *Biotechnology Letters* 31(7):1043–49. doi: 10.1007/s10529-009-9975-7.

Lowry, O. H., N. J. Rosebrough, A. L. Farr, and R. J. Randall. 1951. "Protein Measurement with the Folin Phenol Reagent." *The Journal of Biological Chemistry* 193(1):265–75. doi: 10.1016/s0021-9258(19)52451-6.

Lu, Zhe, Jingcheng Dai, Ling Ling Zheng, Zhuoran Teng, Qi Zhang, Dongru Qiu, and Lirong Song. 2020. "Disodium 2-Oxoglutarate Promotes Carbon Flux into Astaxanthin and Fatty Acid Biosynthesis Pathways in Haematococcus." *Bioresource Technology* 299:122612. doi: 10.1016/j.biortech.2019.122612.

Omidian, Kosar, Hossein Rafiei, and Brian Bandy. 2020. "Increased Mitochondrial Content and Function by Resveratrol and Select Flavonoids Protects against Benzo[a]Pyrene-Induced Bioenergetic Dysfunction and ROS Generation in a Cell Model of Neoplastic Transformation." *Free Radical Biology and Medicine* 152:767–75. doi: 10.1016/j.freeradbiomed.2020.01.021.

Ramanna, Luveshan, Abhishek Guldhe, Ismail Rawat, and Faizal Bux. 2014. "The Optimization of Biomass and Lipid Yields of Chlorella Sorokiniana When Using Wastewater Supplemented with Different Nitrogen Sources." *Bioresource Technology* 168:127–35. doi: 10.1016/j.biortech.2014.03.064.

Sun, Han, Yuxin Wang, Yongjin He, Bin Liu, Haijin Mou, Feng Chen, and Shufang Yang. 2023. "Microalgae-Derived Pigments for the Food Industry." *Marine Drugs* 21(2). doi: 10.3390/md21020082.

Tunali, Merve, Edwin Nnaemeka Uzoefuna, Mehmet Meric Tunali, and Orhan Yenigun. 2020. "Effect of Microplastics and Microplastic-Metal Combinations on Growth and Chlorophyll a Concentration of Chlorella Vulgaris." *Science of the Total Environment* 743:140479. doi: 10.1016/j.scitotenv.2020.140479.

Wong, Y. K., Y. H. Ho, K. C. Ho, H. M. Leung, and K. K. L. Yung. 2017. "Maximization of Cell Growth and Lipid Production of Freshwater Microalga Chlorella Vulgaris by Enrichment Technique for Biodiesel Production." *Environmental Science and Pollution Research* 24(10):9089–9101. doi: 10.1007/s11356-016-7792-9.

Wong, YK, YH Ho, KC Ho, HM Leung, and KKL Yung. 2017. "Growth Medium Screening for Chlorella Vulgaris Growth and Lipid Production." *Journal of Aquaculture & Marine Biology* 6(1):1–10. doi: 10.15406/jamb.2017.06.00143.

Xu, Ling, Pamela J. Weathers, Xue Rong Xiong, and Chun Zhao Liu. 2009. "Microalgal Bioreactors: Challenges and Opportunities." *Engineering in Life Sciences* 9(3):178–89. doi: 10.1002/elsc.200800111.

Zhuang, Dingling, Ning He, Kuan Shiong Khoo, Eng Poh Ng, Kit Wayne Chew, and Tau Chuan Ling. 2022. "Application Progress of Bioactive Compounds in Microalgae on Pharmaceutical and Cosmetics." *Chemosphere* 291(P2):132932. doi: 10.1016/j.chemosphere.2021.132932.

DRAFT

ORAL PRESENTATION

Cinnamomum cassia kabuğundan hazırlanan farklı ekstrelerin antioksidan özellikleri ve fenolik/flavon içeriği üzerine bir araştırma

Sumru Sozer Karadaglı [1*] (0000-0001-8752-7355), Ferzan Lermioglu Erciyas[1]

[*1]Ege Üniversitesi, Eczacılık Fakültesi, Farmasötik Toksikoloji Anabilim Dalı, İzmir, Türkiye

*l.sumru.sozer@ege.edu.tr

Özet

Tarçın, tarçın ağaçlarının kabuğundan elde edilen eşsiz bir baharattır. Sadece lezzet için değil, aynı zamanda çeşitli farmakolojik etkileri ile de öne çıkmaktadır. Tarçın çeşitli türlerinin, özellikle kabuk ve yaprakların, çeşitli biyolojik işlevlere sahip olduğu gösterilmiştir. Antidiyabetik ve antitümör aktiviteleri, güçlü antioksidan aktivitesiyle ilişkilendirilmektedir. Bu nedenle son zamanlarda farklı tekniklerle hazırlanan tarçın ekstreleri üzerinde yoğun araştırmalar yapılmaktadır. *Cinnamomum cassia (C. cassia)* yüksek miktarda toplam fenol ve flavanoid içeren bir türdür.

Bu çalışmada *C. cassia* kabuğunun heksan, metanol ve kloroform ekstrelerinin antioksidan özelliklerini araştırılmıştır. Bu üç ekstre, toplam fenolik içerik, flavonoid içerik ve çeşitli konsantrasyonlarda DPPH radikal temizleme aktivitesi ile antioksidan potansiyelleri açısından taranmıştır. *C. cassia* ekstrelerinin hazırlanmasından sonra ekstrelerin bazı aktif maddeleri HPLC (Yüksek Performanslı Sıvı Kromatografisi) analizi ile değerlendirilmiştir. Quercetin, sinamaldehit, sinamilalkol ve sinnamik asit olmak üzere dört bileşiğin miktarı incelenmiştir. *C. cassia* ekstrelerinin lenfositler üzerindeki sitotoksik aktivitesi WST-1 yöntemi ile in vitro olarak araştırılmıştır.

Bulgularımıza göre, en yüksek sinnamaldehit ve polifenol düzeyleri metanol ekstresinde saptanmıştır. DPPH kullanılarak yapılan antioksidan aktivitenin heksan ve metanol ekstrelerinde kloroforma göre yüksek olduğu bulunmuştur. Üç organik çözücünün ekstraksiyon verimi araştırıldığında metanol çözücüsü ile (% 9) daha yüksek ekstraksiyon verimi elde edilmiştir. İnsan primer periferik kan lenfositlerinde yapılan sitotoksisite çalışmasında ekstrelerin lenfositlerle 24 saat inkübasyonu ile heksan, metanol ve kloroform için IC_{50} değerleri sırasıyla 91.74; 62,30 ve 66,42 µg/mL bulunmuştur. Sonuçlarımıza göre metanol ekstresi diğer ekstrelere göre daha yüksek düzeyde radikal temizleyici aktiviteye sahip ve en yüksek sinnamaldehit düzeyine sahip ekstre olarak saptanmıştır. Daha ileri araştırmalar, bu ekstrenin oksidatif stres altındaki hücreler için faydalı sonuçlar üretebileceğini düşündürmektedir.

Anahtar Kelimeler: *Cinnamomum cassia* kabuğu, heksan, kloroform, metanol, antioksidan aktivite

A research on the antioxidant properties and phenolic / flavone content of different extracts prepared from *Cinnamomum cassia* bark

Abstract

Cinnamon is a unique material obtained from the bark of cinnamon trees. Not only a flavor agent but also a variety of pharmacological applications. Cinnamon various types, especially bark and leaves, have been shown to have various biological functions. Its antidiabetic and antitumor activities were associated with its strong antioxidant activity. Therefore, intensive research has been carried out on the extracts of cinnamon prepared with different techniques recently. *Cinnamomum cassia(C. cassia)* contain higher amount of total phenols and flavanoids.

In the present study, we investigated the antioxidant properties of hexane, methanol and chloroform extracts of *C. cassia* bark. These three extracts were screened for their antioxidant potentials using total phenolic content, flavonoid content, DPPH radical scavenging activity at various concentrations. After preparation of *C. cassia* extracts, some active ingredients of the extracts were evaluated by HPLC (High Performance Liquid Chromatography) analysis. Four compounds namely quercetin, cinnamaldehyde, cinnamylalcohool and

cinnamic acide were quantified. The cytotoxic activity of the extracts of *C. cassia* on lymphocytes was investigated in vitro through WST-1 assay.
The findings indicated that the highest levels of cinnamaldehyde and polyphenols were detected in methanol extract. Antioxidant activity using DPPH were found to be high in hexane and methanol extracts. Extraction yield from *C. cassia* of three organic solvents were investigated. Higher extraction yields were obtained by methanol solvent(% 9).The cytotoxicity study in human primary peripheral blood lymphocytes, IC50 values in hexane, methanol and chloroform extracts were found to be 91.74; 62.30 and 66.42 µg/mL, respectively, by 24-hour treatment of extracts with lymphocytes. According to our results, methanol extract showed higher levels of radical scavenger activity and cinnamaldehyde than other extracts. Further research suggests that this extract may produce beneficial results for cells under oxidative stress.
Keywords: *Cinnamomum cassia* bark, hexane, chloroform, methanol, antioxidant activity

GİRİŞ

Bitkilerde bulunan antioksidan bileşikler, sağlığı koruyucu maddeler olarak hareket ederek insan sağlığına önemli katkıda bulunurlar. Antioksidanlar, metabolik ve yaşa bağlı hastalıklarda serbest radikallere karşı oluşan hasarı düzeltmede etkili olduklarından, insan sağlığındaki en önemli bileşikler olarak görülmektedirler. *C. cassia,* Çin ve diğer Güney ve Güneydoğu Asya ülkelerinde yetişen bir türdür. Ülkemizde de yaygın olarak kullanılmaktadır. Tarçın, sinnamaldehit, sinnamik asit, sinamil alkol gibi birçok esansiyel yağı ve çeşitli reçineli bileşikleri içerir. Singh, sinnamaldehitin varlığının tat ve kokuda önemli bir role sahip olduğunu bildirmiştir. (Singh ve ark., 2009)
Ancak bileşenlerin vücuttaki etkileşimi hakkında bilgi eksikliği tarçın kullanımının güvenliği konusunda tartışmalara yol açmaktadır. Son zamanlarda tarçın türlerinin antioksidan, antidiyabetik ve antitümör aktiviteleri çeşitli biyolojik çalışmalarda gösterilmiştir.(Gruenwald ve ark., 2009)
Ayrıca farklı ekstrelerin birçok aktif bileşen içerdiği gösterilmiştir. Diyabet ve çeşitli kanser türlerinin gelişimine karşı koruyucu etki, polifenoller içeren tarçın ekstrelerinin antioksidan aktivitesi ile ilişkilendirilmiştir. Tarçın kabuğunun aktif bileşenlerinden biri olan sinnamaldehitin bazı hücre kültürü çalışmalarında antikanser aktiviteye sahip olduğu gösterilmiştir.(Zaidi ve ark., 2015)
Metabolizmada reaktif oksijen türlerinin (ROS) oluşumu DNA'ya ve diğer hücresel moleküllere zarar verir. Bunun diyabet, kanser, kardiyovasküler ve nörodejeneratif hastalıklar dahil olmak üzere kronik hastalıkların başlangıcında ve gelişiminde önemli bir rol oynadığı ileri sürülmektedir. Canlı hücreler, kendilerini ROS'un neden olduğu hasardan koruyacak antioksidan savunma potansiyeline sahiptir. Serbest radikallere karşı hücresel antioksidan savunma yetersiz kaldığında oksidatif stres ortaya çıkar. Bunun sonucunda lipitlerde, proteinlerde ve DNA'da oksidatif hasar meydana gelebilmektedir (Halliwell, 1994, Hussain ve ark.,2020, Valko ve ark., 2004, Valko ve ark., 2007, Atanassova ve ark., 2011). Muhtemelen DNA, oksidatif saldırının biyolojik olarak önemli bir hedefidir. DNA'ya sürekli oksidatif saldırının kanser gelişiminde önemli bir rol oynadığı yaygın olarak kabul edilmektedir (Demirci-Çekiç ve ark., 2022, Ames ve ark., 1993). Diyetteki fenolik bileşikler; antioksidan, antikanser, antiviral ve antiinflamatuar aktiviteleri birçok hastalık için önemlidir. Bu konu çeşitli in vivo ve in vitro çalışmalarla ortaya konmuştur (Halliwell, 2012). *C. cassia* ekstrelerinin birçok aktif bileşen içerdiği gösterilmiştir. Diyabet ve çeşitli kanser türlerinin gelişimine karşı koruyucu etkisi gösterilmiştir. polifenoller içeren tarçın ekstrelrinin antioksidan aktivitesi ile ilişkilidir.
Ekibimiz daha önce *C cassia* kabuğundan farklı ekstreler ve fraksiyonlar hazırlamıştı. İnsan primer periferik kan lenfositleri, V79 Çin Hamster akciğer fibroblastları ve HL-60 insan promiyelositik lösemi hücreleri gibi farklı hücre tiplerinde, bu ekstrelerin endojen ve oksidatif DNA hasarı, sitotoksik ve apoptotik aktivite çalışmaları tarafımızdan yapılmıştır (Karadagli ve ark., 2014a, Lermioğlu Erciyas ve ark., 2015, Karadagli ve ark., 2014b). Bu çalışmada farklı polaritedeki organik çözeltiler ile doğrudan muamele ile hazırlanarak ekstrelerdeki bileşiklerin farkı çözücülere geçmesi sonucu antioksidan aktivite ve hücre canlılığına etkileri araştırıldı. Ayrıca HPLC ile ekstrelerdeki aktif bileşik miktarları da ölçülmüştür.

MATERYAL VE METOT

Bitki ekstrelerinin hazırlanması

Bu çalışmada ticari olarak temin edilebilen *C.cassia* kabuğu kullanıldı. Kurutulmuş ve toz haline getirilmiş malzeme (60g) 90 dakika süreyle ekstre edildi. +40°C'de heksan, metanol ve kloroform ile karıştırıldı ve süzüldü (Whatman filtre kağıdı No.4). Her bir organik çözücü ekstresi rotary evaporatör ile konsantre edildi. Kalıntı tersiyer bütanol ile alındı, daha sonra dondurulduktan ve liyofilize edildi. Yüzde verim, bitki materyalinin

kurutulmuş ağırlığı cinsinden ifade edildi. Tüm ekstreler +4°C'de karanlıkta tutuldu. Tarçın ekstreleri çalışma boyunca güneş ışığından korundu.

DRAFT

Lenfositlerin hazırlanması

Sağlıklı bir donörden alınan periferik venöz kan heparinize tüpe alınarak ışıktan korundu. Lenfositler, bir Histopaque(1:1) tabakası üzerine alındı ve oda sıcaklığında 2750 rpm'de 30 dakika süreyle santrifüjlendi. Daha sonra izole edilen lenfositler PBS ile yıkandı. Bu çalışma Ege Üniversitesi Tıp Fakültesi Klinik Araştırmalar Etik Kurulu tarafından İzmir, Türkiye (12.01.2012, 11.12.1/14) tarafından onaylanmıştır.

WST-1 analizi:

Sitotoksisite, WST-1 kullanılarak belirlendi. Lenfositler, %10 FBS ve %1 (h/h) penisilin streptomisin ile RPMI-1640 içerisinde süspanse edildi ve $1x10^6$/mL, 50 uL ortam konsantrasyonunda ekildi. Hücreler, CO_2 inkübatöründe (Nuaire 5510E, ABD) farklı konsantrasyonlarda (5-1000 µg/mL) *C. cassia* heksan, metanol ve kloroform ekstreleri ile 24 saat boyunca kültürlendi. 24 saatlik bekleme sonrası kuyucuk başına 10 uL WST-1 ilave edildi. 37°C'de 4 saatlik inkübasyonun ardından plaklar, 450 nm'de (Thermo Scientific, Varioskan® Flash) okundu. Tüm deneyler üç tekrarlı gerçekleştirildi. Ekstrelerin IC_{50}'si GraphPad Prism 5.0 yazılımı kullanılarak hesaplandı.

Toplam Fenolik İçerik

Ekstrelerin toplam fenolik içeriği, Folin-Ciocalteau kolorimetrik yöntemiyle analiz edildi. Metanol:su (1:10 h/h) içindeki ekstrenin/standart çözeltinin 0.2 mL'si, suyla 1:10 h/h seyreltilmiş 1 mL Folin-Ciocalteu reaktifine eklendi, 4 dakika sonra 800 uL doymuş sodyum karbonat çözeltisi (75 g/L) ilave edildi. Oda sıcaklığında 2 saatlik inkübasyon sonrası absorbanslar (Thermo Scientific, Varioskan® Flash) 765 nm'de ölçüldü. Gallik asit ile bir kalibrasyon eğrisi elde edildi ve toplam fenolik içerik, g GAE/g DW olarak ifade edildi. Tüm deneyler üç tekrarlı gerçekleştirildi. (Singleton and Rossi, 1965).

Toplam Flavonoid İçeriği

Toplam flavonoid içeriği Chang ve arkadaşlarının yöntemine göre tayin edildi. 0,1 ml %10 AlCl ve 1 M potasyum asetat (0,1 ml) eklendi ve 2 ml ekstre metanol içerisinde çözüldü. Toplam hacmi metanol ile 5 ml'ye tamamlandı. 40 dakika inkübasyonun ardından absorbans 415 nm'de ölçüldü. Standart kalibrasyon eğrisi kersetin kullanılarak elde edildi ve toplam flavonoid içeriği, g ağırlık başına ifade edildi. Tüm deneyler üç tekrarlı gerçekleştirildi. (Chang ve ark., 2002).

DPPH Testi

Ekstrelerin serbest radikal temizleme aktiviteleri DPPH yöntemi ile değerlendirildi. Numunenin farklı konsantrasyonlardaki 1,5 ml' si, metanol içindeki 0,5 ml taze hazırlanmış 0,5 mM DPPH 'a eklenerek, karanlıkta 30 dakika bekletildi. Absorbans, Thermo Scientific Varioskan® Flash mikroplaka okuyucu ile DPPH' a karşı 517 nm'de ölçüldü. Referans standardı olarak Trolox kullanıldı. Tüm örnekler üç tekrarlı çalışıldı. Bileşiklerin DPPH radikal temizleme aktivitesi aşağıdaki denkleme göre hesaplanmıştır (Yang ve ark., 2012, Kamleshiya ve ark., 2012):

Temizleme aktivitesi (%) = $(A_{DPPH}-A_{Örnek}) / (A_{DPPH}) \times 100$

HPLC analizi

HPLC analizi, fotodiyot dizisi detektörü ile donatılmış Thermo Scientific Accela Model 1250 sistemi (ABD) kullanılarak gerçekleştirildi Kersetin, sinamilalkol, sinamik asit ve sinnamaldehitten (Sigma Aldrich, ABD) oluşan stok çözelti, metanol (1 mg/mL) içerisinde hazırlandı. Ayırma için ACE-5 C_{18} kolonu (250x4,6 mm, 5 µm) kullanıldı. Kullanılan solventler; mobil faz A su içinde %0,25 trifloroasetik asit ve mobil faz B asetonitril içinde %0,25 trifloroasetik asit, gradyan koşulları Tablo 1'de verildi. Mobil faz, kullanımdan önce 0,45 um membran filtreden süzüldü. akış hızı 0,8 mL/dak olarak ayarlandı. Enjeksiyon hacmi 10 µL dir ve örnekler üç kez enjekte edildi. UV-VIS spektrumları 200 ila 600 nm arasında kaydedildi. Tüm kromatografik işlemler ortam sıcaklığında gerçekleştirildi. Hesaplamalar, external standart yöntemi kullanılarak pik entegrasyonu ile yapıldı. Çalışma standartları için 1-40 µg/mL konsantrasyonlar kullanıldı. Analitlerin kromatografik pikleri, alıkonma süreleri standardınkilerle karşılaştırılarak belirlendi.

Tablo 1. Gradient şartları

Zaman (dk)	Mobil faz A (%)	Mobil faz B (%)
0	90	10
15	30	70
17	90	10

İstatistiksel analiz

Veri analizi SPSS for Windows, sürüm 15.0 kullanılarak yapıldı. Sonuçların analizinde tek yönlü varyans analizi (ANOVA) testi kullanıldı. İstatistiksel olarak anlamlı fark $p<0,05$ düzeyinde kabul edildi.

BULGULAR

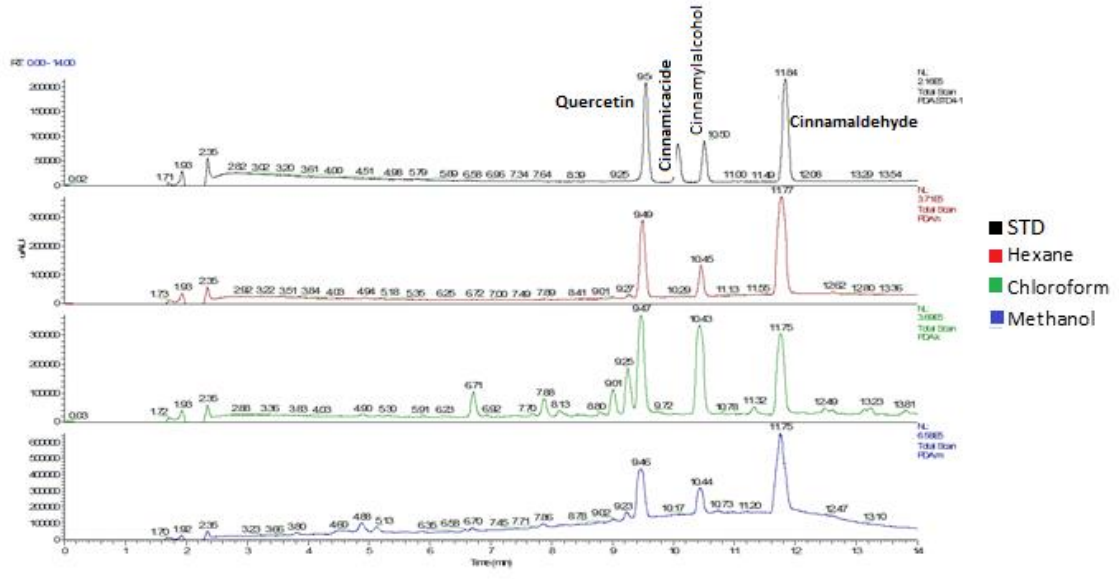
Çalışmada, ekstraksiyon verimi *C. cassia* hekzan, kloroform ve metanol ekstreleri için sırasıyla 1%, 1.2% ve 9% olarak hesaplandı (Tablo 2).

Ekstraksiyon verimi (%) = Dondurulmuş kuru ekstrenin ağırlığı/ örneğin ağırlığı x 100

Ekstrelerin bazı biyoaktif bileşiklerinin miktarını belirlemek için HPLC kullanıldı. *C. cassia* ekstrelerinde belirlenen sinnamaldehit ve sinamik asit miktarları Tablo 2'de verilmektedir. Ancak kersetin, sinnamil alkol tespit edilememiştir. *C. cassia* ekstrelerindeki toplam fenolik ve flavanoidlerin miktarları Tablo 3'te gösterilmektedir. Standart kersetin ve gallik asit kalibrasyon eğrileri, sırasıyla 0,992 ve 0,9929 R^2 değerlerine sahipti.

Çalışmamızda *C. cassia* 'dan elde edilen üç ekstrenin antioksidan aktivitesi DPPH yöntemi ile test edildi. Ekstrelerin serbest radikal temizleme aktiviteleri Şekil 3'te gösterilmektedir. *C. cassia* hekzan ve metanol ekstrelerinin kabukları dikkat çekici antioksidan aktiviteler sergilemiştir. Kabukların IC_{50}'si sırasıyla 22,55, 142,2, 25,24 µg/ml (hekzan, kloroform, metanol) bulunmuştur. Tüm ekstrelerin DPPH radikal temizleme aktivitesi, konsantrasyonun artmasıyla doğu orantılı artmıştır (Şekil 2). Kloroform ekstresi, hekzan ve metanol ekstrelerine göre düşük antioksidan aktiviteye sahiptir.

WST-1 yönteminde, lenfosit canlılığı, ekstrelerle 24 saat süre sonunda konsantrasyona bağlı olarak azalmaktadır. Kontrole göre hücre canlılığını %50 (IC_{50}) azaltmak için gereken konsantrasyon, hekzan, kloroform ve metanol ekstreleri için sırasıyla 91,74; 62,30 ve 66,42 µg/ml' dir.



Şekil 1: *C cassia* kabuğu ekstrelerinden elde edilen flavonoidlerin HPLC profili (1:kersetin; 2: sinnamilalkol; 3:sinnamik asit; ve 4:sinnamaldehit)

Tablo 2. Hekzan, kloroform ve metanol ekstrelerinin ekstraksiyon verimi ve *C. cassia* kabuğundan elde edilen aktif bileşikler

Ekstre	Ekstraksiyon verimi (%)	Sinnamaldehit (µg/ml)	Sinnamic Asit (µg/ml)
Hekzan	1 %	28,25	14,20
Kloroform	1.2 %	22,24	52,43
Metanol	9 %	40,13	24,84

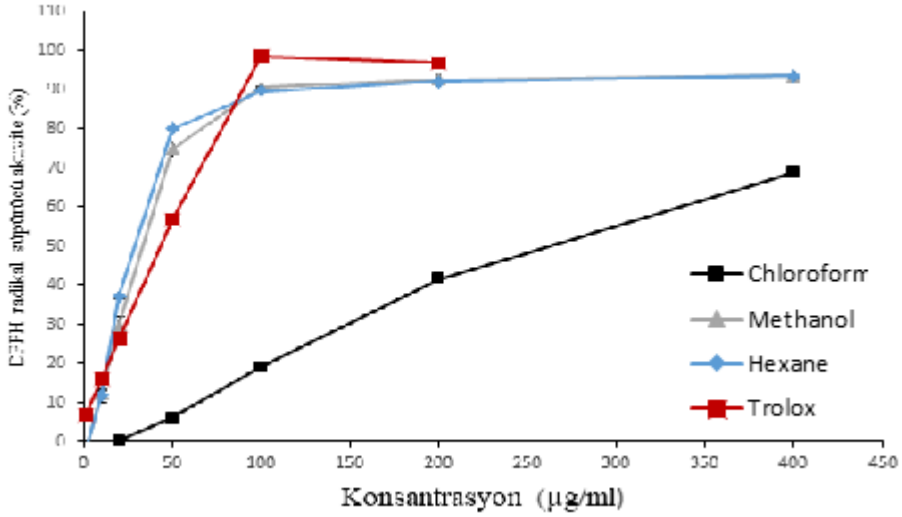
Tablo 3. *C. cassia* ekstrelerindeki toplam flavonoidler ve fenolik içerikler

	Hekzan	Kloroform	Metanol
Total flavanoid içerik (mg kersetin/g kuru ağırlık)	9,30±2,2	10,37±2,4	18,72±0,3
Total fenolik içerik (mg gallik asit/g kuru ağırlık)	21,28±0,3	27,85±0,2	57,91±0,6

Tüm analizler üç ölçüm ortalaması ± standart sapmadır.

TARTIŞMA

Kozmetikte, baharatlarda ve fitoterapi endüstrisinde kullanılan tarçın dünya çapında üretilmektedir. Bitkiden elde edilen ekstrelerin fiziksel ve kimyasal bileşimlerinin analizi, bileşenlerinin faydalı etkileri kadar toksik etkilerinin de olması, tüketici güvenliği açısından önemlidir. Ekstrelerin serbest radikal süpürme aktiviteleri, antioksidan bileşiklerin hidrojen kaybetme yeteneğine ve bu bileşenlerin yapısal konformasyonuna bağlıdır. DPPH serbest radikali, stabil bir molekül haline gelmek için antioksidan moleküllerden bir elektron veya hidrojen alır (Baliyan ve ark., 2022).



Şekil 2: *C. cassia* ekstrelerinin DPPH radikal temizleme aktivitesi.

Metanolde hazırlanan DPPH radikal çözeltisi, bir antioksidan madde varlığında DPPH-H (difenilhidrazin) moleküllerine dönüştürülür. Çalışmamızda tarçının hekzan ve metanol ekstrelerinin DPPH free radikal scavencing aktiviteleri 200 µg/ml nin üzerindeki dozlarda %90'nın üzerine çıkarken kloroform ekstreleri % 400µg/ml de % 68,84 olarak saptanarak daha zayıf bir aktivite göstermiştir. (Şekil 2) Referans standart olarak kullanılan trolox aktivitesi ise 100 µg/ml dozlarda %90 nın üzerine çıkmıştır.

Ekstreler HPLC ile analiz edildiğinde aktif bileşenleri olan sinnamaldehit hekzan kloroform ve metanol ekstrelerinde sırasıyla 28,25, 22,24, 40,13 µg/ml, ve sinnamik asit 14,20, 52,43 ve 24,84 µg/ml bulunmuştur. Sinnamaldehitin metanol ekstresinde sinnamik asitin ise kloroform ekstresinde daha yüksek düzeyde bulunduğu görülmektedir(Tablo 2).

Antioksidan bileşikler genellikle fenolik formdadır. Fenoller içerdikleri hidroksil grupları ile, radikalleri bağlayan bileşiklerdir. Bu önemli bitki bileşenleri, hidrojen atomlarını hidroksil gruplarından radikallere bırakır ve stabil fenoksil radikallerini oluştururlar. Bu nedenle, bitki ekstrelerinin antioksidan kapasitesini belirlemek için fenolik bileşiklerin miktarının belirlenmesi çok önemlidir (Shi ve ark., 2022). Tarçının fenolik bileşenlerinin de antioksidan ve serbest radikal temizleme aktivitelerinden sorumlu olduğu bildirilmektedir (Hamidpour ve ark., 2015). Fenolik bileşiklerin antioksidan özellikleri, proton kaybı, şelat oluşumu ve radikallerin dismutasyonundan kaynaklanmaktadır. Çalışmada tarçın ekstrelerinin total fenolik içerikleri hekzan kloroform ve metanol ekstreleri için sırasıyla 21,28, 27,85 ve 57,91 mg gallik asit/100g kuru ağırlık ve total flavon içerikleri 9,3, 10,37, 18,72 mg kersetin/100g kuru ağırlık olarak bulunmustur. Metanol ekstresinin diğer ekstrelere göre fenolik ve flavon içeriği açısından daha yüksek olduğu sonuçlarımızdan görülmektedir. Çalışma sonuçlarımız El-Baz ve ark. nın (2023) çalışmasıyla uygunluk göstermektedir

C. cassia kabuğunun temel bileşenlerinden olan sinnamaldehit ve diğer bileşenleri olan sinnamik asit, sinnamil alkol, kumarin ve metil öjenolün kantitatif analizine yönelik çalışmalarda *C. cassia'* nın buhar distilasyonu ya da metanol, aseton, kloroform, hekzan gibi çözücülerle ekstraksiyonu sonucunda bu bileşiklerin en yüksek düzeyde metanol çözücüsünde bulundukları gösterilmiştir (He ve ark., 2005)

Nile ve ark.ı (2023) *C. zeylanicum* kabuğunun aktif bileşeni sinnamaldehitin HCT 116 ve HT-29 kolon hücrelerinde serbest oksijen radikal oluşumunu artırdığını göstermişler, bileşiğin sitotoksik etkisini, ROS oluşumunun aracılık ettiği apoptozla ilişkilendirmişlerdir. *C. cassia* kabuğu metanol ekstresinin GS-MS analizinin yapıldığı bir çalışmada 35 fitokimyasal saptanmış ve majör bileşenlerin sinnamaldehit (%24.58), p-metoksi sinnamaldehit (%9.87), cis-2-metoksi sinnamik asit (%9.22), sinnamik asit (%7.39), kumarin (%5.31) ve 8,11-oktadekadienoik asit metil esteri (%4.27) olduğu bildirilmiştir (Ahmada ve ark., 2013)

Tarçın esasiyel yağının güçlü bir antiproliferatif etkiye sahip olduğu ve hücrelerde reaktif oksijen türlerinin oluşumunu indükleyebilen sinnamaldehiti yüksek oranda içerdiği bildirilmiştir (Cappelli ve ark., 2023). Sinamik asit, lignin, tanenler, flavonoidler, *pigmentler*, baharatların lezzet bileşenlerinin birçoğunun sentezlenmesinde öncüdür. Büyük moleküllü fenolik bileşiklerin oluşumunda kilit rol oynar. Çalışmamızda ekstrelerde elde edilen farklı antioksidan aktivite düzeylerinin sinnamaldehit ve sinnamik asit miktarlarından kaynaklandığı değerlendirilmiştir. Metanol ekstresinde yüksek fenol ve flavonoid içerik ve antioksidan aktivite saptanmıştır. Bu ekstrede sinnamaldehit düzeyi yüksektir. Kloroform ekstresinde ise DPPH aktivitesi düşük bulunmuştur ve daha yüksek sinnamik asit düzeyi saptanmıştır. Sonuçlarımıza bakıldığında kloroform ekstresi ile daha düşük antioksidan aktivite görülmesi(400 µg /ml da % 68) sinnamaldehit düzeyinin bu ekstrede daha düşük düzeyde olması ile ilişkili olabileceği değerlendirilmiştir.

SONUÇ

Sonuç olarak incelenen üç farklı ekstreden metanol ekstresi diğer ekstrelere göre daha yüksek düzeyde radikal temizleyici aktiviteye sahip ve en yüksek sinnamaldehit düzeyine sahip ekstre olarak saptanmıştır. Bu ekstrenin oksidatif stres altındaki hücreler için faydalı sonuçlar üretebileceği daha ileri çalışmalarla ortaya çıkarılabilir görüşündeyiz

TEŞEKKÜR

Yazarlar bu çalışmaya Ege Üniversitesi Eczacılık Fakültesi Farmasötik Bilimler Araştırma Merkezi'ne (FABAL, İzmir, TÜRKİYE) verdiği destek için teşekkür ederler.

KAYNAKLAR

Ahmada, SI, Capoorb M, Khatoona F 2013. Phytochemical analysis and growth inhibiting effects of Cinnamomum cassia bark on some pathogenic fungal isolates. Journal of Chemical and Pharmaceutical Research. 5 (3): 25-32.

Ames BN, Shigenaga MK, Hagen TM 1993. Oxidants, antioxidants and the degenerative diseases of aging. Proc Natl Acad Sci USA; 90:7915-22.

Atanassova M, Georgieva S, Ivancheva K 2011. Total phenolic and Total Flavonoid Contents, antioxidant capacity and biological components in medicinal herbs, J Univ Chem Tech and Metal; 46:81-8.

Baliyan S, Mukherjee R, Priyadarshini A, Vibhuti A, Gupta A, Pandey RP, Chang CM 2022. Determination of Antioxidants by DPPH Radical Scavenging Activity and Quantitative Phytochemical Analysis of Ficus religiosa. Molecules.16;27(4):1326.

Cappelli G, Giovannini D, Vilardo L, Basso A, Iannetti I, Massa M, Ruberto G, et al 2023. Cinnamomum zeylanicum Blume Essential Oil Inhibits Metastatic Melanoma Cell Proliferation by Triggering an Incomplete Tumour Cell Stress Response. Int J Mol Sci. 16;24(6):5698.

Chang C, Yang M, Wen H 2002. Estimation of total flavonoid content in propolis by two complementary colorimetric methods, J Food Drug Anal 10:178-82.

Demirci-Çekiç S, Özkan G, Avan AN, Uzunboy S, Çapanoğlu E, Apak R 2022. Biomarkers of Oxidative Stress and Antioxidant Defense. J Pharm Biomed Anal. 5;209:114477.

El-Baz YG, Moustafa A, Ali MA, El-Desoky GE, Wabaidur SM, Faisal MM 2023. An Analysis of the Toxicity, Antioxidant, and Anti-Cancer Activity of Cinnamon Silver Nanoparticles in Comparison with Extracts and Fractions of Cinnamomum Cassia at Normal and Cancer Cell Levels. Nanomaterials (Basel). 5;13(5):945.

Gruenwald J, Freder J, Armbruester N 2010. Cinnamon and health. Crit Rev Food Sci Nut. 50,9: 822-834.

Halliwell B 1994. Free radicals, antioxidants, and human disease: curiosity, cause, or consequence? Lancet 344:721-4.

Halliwell B 2012. Free radicals and antioxidants: updating a personal view. Nutr Rev. 70(5):257-65.

Hamidpour R, Hamidpour M, Hamidpour S, Shahlari M 2015. Cinnamon from the selection of traditional applications to its novel effects on the inhibition of angiogenesis in cancer cells and prevention of Alzheimer's disease, and a series of functions such as antioxidant, anticholesterol, antidiabetes, antibacterial, antifungal, nematicidal, acaracidal, and repellent activities. Journal of Traditional and Complementary Medicine. 5(2): 66-70.

He ZD, Qiao CF, Han QB, Cheng CL, Xu HX, Jiang RW, But PP, Shaw PC 2005. Authentication and quantitative analysis on the chemical profile of cassia bark (cortex cinnamomi) by high-pressure liquid chromatography. J Agric Food Chem. 6;53(7):2424-8.

Hussain S, Ashafaq M, Alshahrani S, Siddiqui R, Ahmed RA, Khuwaja G, Islam F 2020. Cinnamon oil against acetaminophen-induced acute liver toxicity by attenuating inflammation, oxidative stress and apoptosis. Toxicol Rep. 17;7:1296-1304.

Kamleshiya P, Meshram VG, Ansari AH 2012. Comparative evaluation of antioxidant and free-radical scavenging activity of aqueous and methanolic spice extracts. Int J Life Sci Pharm Res 2(3): 118-125.

Karadagli S, Agrap B, Erciyas FL 2014a. Investigation of Cytotoxic and Genotoxic Potential of Cinnamomum cassia Bark Water Extract. J Marmara Univ Inst Heal Sci. 4(1):17–23.

Karadagli SS, Agrap B, Erciyas FL 2014b. Investigation of the protective effect of C. cassia bark extract against H2O2-induced oxidative DNA damage in human peripheral blood lymphocytes and antioxidant activity. Marmara Pharm J.;18(1):43–8.

Lermioğlu Erciyas F, Ağrap B, Çanaklı G, Çeliksöz M, Sözer Karadağlı S, Gündüz C 2015. An investigation of the effects of C. cassia bark extracts on oxidative DNA damage and possible cytotoxic and apoptotic activities in transformed/untransformed cell lines from Type 1 diabetic patients, in vitro. Frontiers in Genetics. ICAW - 11th International Comet Assay Workshop.

Nile A, Shin J, Shin J, Park GS, Lee S, Lee JH, Lee KW, Kim BG, Han SG, Saini RK, Oh JW 2023. Cinnamaldehyde-Rich Cinnamon Extract Induces Cell Death in Colon Cancer Cell Lines HCT 116 and HT-29. Int J Mol Sci. May 3;24(9):8191

Shi L, Zhao W, Yang Z, Subbiah V, Suleria HAR 2022. Extraction and characterization of phenolic compounds and their potential antioxidant activities. Environ Sci Pollut Res Int. 29(54):81112-81129.

Singh G, Maurya S, deLAmpasona MP, Catalan CAN 2007. A comparison of chemical, antioxidant and antimicrobial studies of cinnamon leaf and bark volatile oils, oleoresins and their constituents. Food and Chemical Toxicology, 45, 9: 1650–1661.

Singleton VL, Rossi JA 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. Am J Enol Vitic 16:144–53.

Valko M, Izakovic M, Mazur M, Rhodes C. J, Telser J 2004. Role of oxygen radicals in DNA damage and cancer incidence. Mol Cell Biochem 266:37-56.

Valko M, Leibfritz D, Moncol J, Cronin M.T, Mazur M, Telser J 2007. Free radicals and antioxidants in normal physiological functions and human disease. Int J Biochem Cell Biol 39:44-84.

Yang CH, Li RX, Chuang LY 2012. Antioxidant activity of various parts of Cinnamomum cassia extracted with different extraction methods. Molecules 17(6): 7294-7304.

Zaidi SF, Aziz M, Muhammad JS, Kadowaki M 2015. Diverse pharmacological properties of Cinnamomum cassia: A review. Cinnamon: A Multifaceted Medicinal Plant Pasupuleti Visweswara Rao1,2 and Siew Hua Gan Pak J Pharm Sci. Jul;28(4):1433-8.

DRAFT

ORAL PRESENTATION

Preparation and *in vitro* evaluation of topical cream formulation containing Oleum rosmarini for use in the treatment of rheumatism

Tilbe Çevikelli[1*] (*ORCID: 0000-0002-0881-0644*), Umay Merve Güven[1] (*ORCID: 0000-0003-1547-0817*) Nurdan Tezcan[1] (*ORCID: 0009-0001-1442-5486)*, Serpil Demirci Kayıran[2] *(ORCID: 0000-0001-8340-3347)*

[*1]Cukurova University, Faculty of Pharmacy, Department of Pharmaceutical Technology, Adana, Turkey.
[2]Cukurova University, Faculty of Pharmacy, Department of Pharmaceutical Botany, Adana, Turkey.

*Corresponding author e-mail: tcevikelli@cu.edu.tr

Abstract

In this study, it was aimed to develop a topical cream formulation from y/s type emulsions containing Oleum Rosmarini so that the *Rosmarinus officinalis* plant can be used in the treatment of rheumatism. Triethanolamine was added for neutralization to some of the emulsion combinations containing oil, surfactant, copolymer and plasticizer at different rates over three different polymer concentrations in order to select the appropriate formulation with preformulation studies. Organoleptic controls of the selected formulations were made and characterizations were made in terms of pH, texture profile analysis, rheology and thermodynamics. The pH value of the optimized formulations was in the range of 5-7. The formulations were obtained homogeneously and no phase separation was observed. It has been observed that the creams provide proper flow and have high stability.

Keywords: Cream, Oleum rosmarini, *Rosmarinus officinalis*, Rheumatic Disease, Topical Application

INTRODUCTION

Rheumatoid arthritis is a multisystem disease of unknown cause, characterized by inflammation of the synovial membrane, leading to progressive destruction of joint cartilage, bone erosion, and chronic deformities that may also involve internal organs. Because chronic pain is a common symptom in most rheumatic diseases and the restrictive effect of pain, pain relief is one of the primary goals of antirheumatic treatment goals (Adami et al., 2019). *Rosmarinus officinalis* (rosemary) is a medicinal plant originating from the Mediterranean and grown worldwide. Several phytocompounds with pharmacological activities can be isolated from essential oils and extracts of *Rosmarinus officinalis*. Some characteristic chemical constituents of this oil include 1,8-cineole, α-pinene, camphor, bornyl acetate, borneol, camphene, α-terpineol, limonene and myrcene (Borges et al., 2019). Creams are emulsion-type semi-solid dosage forms with an opaque appearance containing a water phase, volatiles, hydrocarbons, waxes or polyols, dissolved or dispersed in a suitable base, for external application to the skin. These dosage forms are forms containing one or more drug substances dissolved or dispersed in a suitable base (Sahu et al., 2021). The aim of this study is to develop an alternative emulsion formulation prepared for topical use with the essential oil obtained from the Rosmarinus officinalis plant due to its anti-inflammatory and analgesic effects in the treatment of rheumatoid arthritis and to investigate its in vitro characterization.

MATERIALS AND METHODS

Materials

All materials used were in pharmaceutical grade.

Methods

Formulation Studies

Cream formulations were prepared by the emulsification technique. Carbopol 996 and HPMC were used as the water phase and Oleum rosmarini was added as the oil phase. Tween 20 was applied as surfactant while

propylen glycol was added as plastizer. Different ratios of the formulation ingredients were tried and charecterized *in vitro*, for determination of the optimum formulation and represented in Table 1.

Table 1. Formulation Ingredients of the Creams

	Water Phase				Oil Phase		
	Carbopol	**HPMC**	**TEA**	**Distilled water q.s. (g)**	**Oleum rosmarini(g)**	**Tween 20(g)**	**Propylen glycol (g)**
FF1	0.1	2	q.s.	100	30	1	5
FF2	0.1	2	q.s.	100	30	3	2.5
FF3	0.1	2	q.s.	100	30	3	10
FF4	0.1	2	-	100	30	1	5
FF5	0.1	2	-	100	30	3	2.5
FF6	0.1	2	-	100	30	3	10

Characterization of the creams

Organoleptic properties analyzed manually, pH was determined with the Mettler digital pHmeter, Texture Profile Analysis was done with the Texture Analyzer (TA.XT.PlusC, Stable Micro System, Haslemere, Surry, UK), rheological properties were analyzed by the Haake Rheometer I (Thermo Fisher Scientific Inc., Essen, Germany), and stability evaluation was done by centrufigation and freez-thaw cycles applied to formulations. All studies were conducted in triplicate.

RESULTS and DISCUSSION

Characterization of the creams

pH values of the all formulations were measured between 5- 7, and found to be applicable for skin. Results of the organoleptic evaluation was shown in Table 2. According to organoleptik evaluation F1,2,3 and F6 formulations were chosen for texture profile analysis and rheological analysis (n=3).

Table 2. Organoleptic Properties of the Formulations

Formulation	**Spreadability**	**Phase Separation**	**Homogenity**	**Appearance**
F1	+++	Stabile	Homogen	Convenient
F2	+++	Stabile	Homogen	Convenient
F3	+++	Stabile	Homogen	Convenient
F4	+	Occured	-	-
F5	+	Occured	-	-
F6	+++	Stabile	Homogen	Convenient

For rheological analysis, shear rate results and viscosity changes against shear stress were examined. As the shear rate increased, the viscosity decreased. Due to the increase in shear stress and shear rate, the flow characteristic showed results suitable for pseudoplastic flow (Namjoshi et al., 2020). It maintained its flow properties in both formulations and represented in Figure 1.

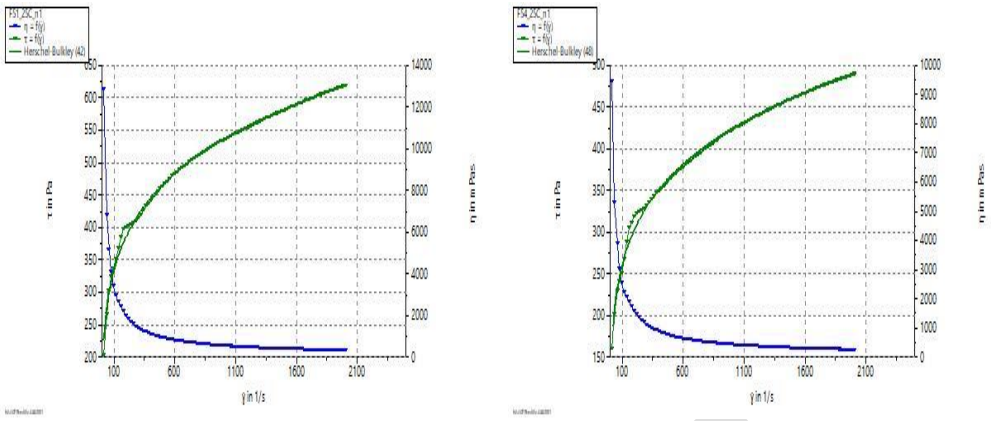


Figure 1. Rheograms of the F1 and F6 Formulations

It was stated that among the developed formulations, the formulation with high adhesion value and low cohesion value had low interaction with the skin and, accordingly, the cream showed high ease of application (Okur et al., 2020), and results are represented in Figure 2.

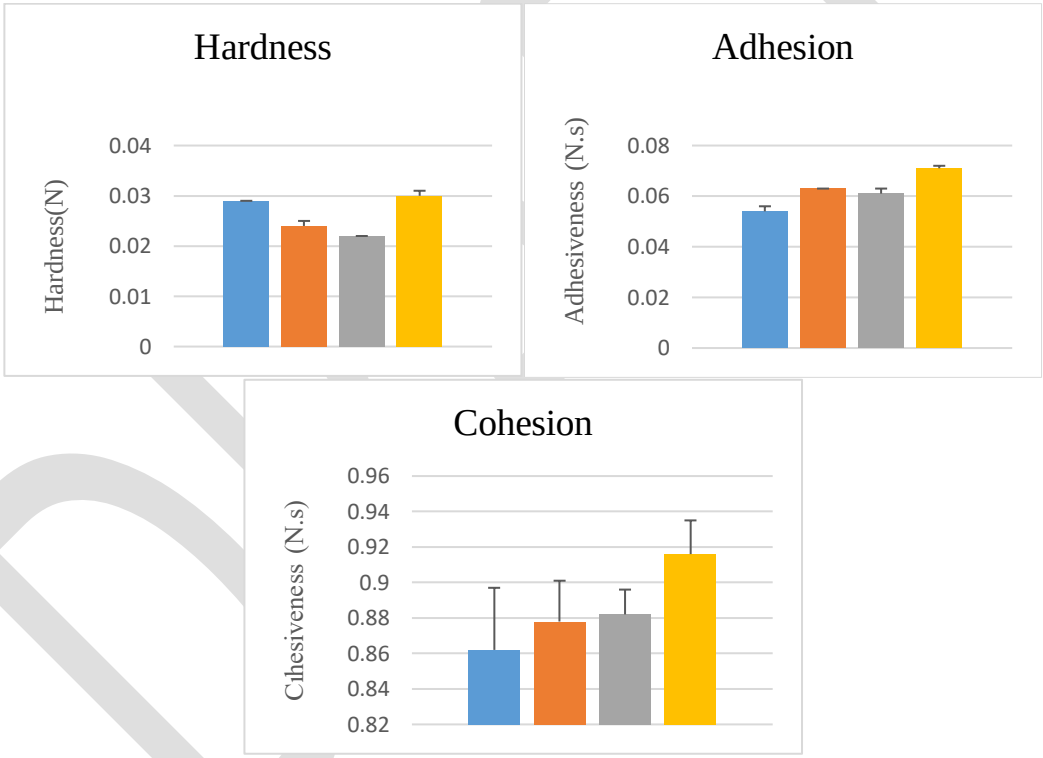


Figure 2. Texture Profile Analysis of the Creams

CONCLUSION

In the current study, a suitable formulation in terms of its physicochemical properties was developed by using rosemary oil, an active ingredient of natural origin. Considering the positive results, further research can be done with different studies. Additionally, further studies can be conducted on the effectiveness of the formulation through *ex vivo* and *in vivo* experiments.

REFERENCES

Adami G, Fassio A, Rossini M, Caimmi C, Giollo A, Orsolini G, Gatti D 2019. Osteoporosis in rheumatic diseases. International Journal of Molecular Sciences, 20(23): 5867.

Borges RS, Ortiz BLS, Pereira ACM, Keita H, Carvalho JCT 2019. *Rosmarinus officinalis* essential oil: A review of its phytochemistry, anti-inflammatory activity, and mechanisms of action involved. Journal of Ethnopharmacology, 229: 29-45.

Namjoshi S, Dabbaghi M, Roberts MS, Grice JE, Mohammed Y 2020. Quality by design: Development of the quality target product profile (QTPP) for semisolid topical products. Pharmaceutics, 12(3): 287.

Okur NU, Yozgatlı V, Senyigit Z 2020. Formulation and detailed characterization of voriconazole loaded *in situ* gels for ocular application. Journal of Faculty of Pharmacy of Ankara University, 44(1): 33-49.

Sahu T, Patel T, Sahu S, Gidwani B 2016. Skin cream as topical drug delivery system: a review. Journal of Pharmaceutical and Biological Sciences, 4(5): 149.

DRAFT

ORAL PRESENTATION

Mandalarda üreme ve üreme sorunları

Sakine Ülküm Çizmeci[1*] (*ORCID: 0000-0003-2939-8019*), Ayşe Merve Köse[2] (*ORCID: 0000-0003-1863-5955*)

[*1] Selçuk Üniversitesi, Veteriner Fakültesi, Doğum ve Jinekoloji Anabilim Dalı, Konya, Türkiye
[2] Hatay Mustafa Kemal Üniversitesi, Veteriner Fakültesi, Doğum ve Jinekoloji Anabilim Dalı, Hatay, Türkiye

*Sorumlu yazar e-mail: ulkum@selcuk.edu.tr,

Özet

Manda büyük oranda Asya kıtasında yaşayan, etinden, sütünden, iş gücünden, boynuzundan ve derisinden faydalanılan bir hayvandır. Mandalar yem seçmeden düşük kaliteli yemleri iyi değerlendirebilen ve farklı çevre koşullarına uyum sağlayabilen hayvanlardır. Türkiye'de manda yetiştiriciliği ağırlıklı olarak süt üretimi için yapılmakta olup, et üretimi sonraki amaç olarak gelmektedir. Diğer taraftan ülkemizde manda başına elde edilen et ve süt üretim değerleri manda yetiştiriciliği yüksek olan ülkelerin oldukça gerisindedir. Bu sebeple verimi arttıracak saf yetiştirme ile ırk geliştirme faaliyetlerinin yaygınlaştırılması gereklidir. Yurdumuzda 2021 yılı verilerine göre manda sayısı 185.574 ve çiğ süt üretimi ise 63.943 tona yükseldi. Türkiye'de yetiştirilen mandaların laktasyon süt verimleri 654-761 kg arasında değişmektedir. Mandalarda pubertasa ulaşma yaşı 13-22 ay ile 36-40 aylar arasındadır. Anadolu Mandaları 13-15 ay, bataklık mandaları 21-48 ay ve Nehir mandaları ise 15-18 ayda pubertasa ulaşır. Bir başka yaklaşımda ise erişkin vücut ağırlığının (250 ila 400) yaklaşık % 60'ına ulaştığında pubertasa erişmektedir. Östrus siklusu ortalama 21 (17-26) gündür. Siklus Nehir mandalarında 20-22 gün, Bataklık mandalarında ise 19-20 gün arasında değişmektedir. Mandalarda düzensiz östrus görülme insidansı oldukça yüksektir. Mandalarda da östrus siklusu proöstrus, östrus, metöstrus ve diöstrus olmak üzere 4 döneme ayrılır. Ortalama gebelik süresi Nehir mandalarında 310 gün (300-320) Bataklık mandalarında ise 330 gün (320-340) aralığındadır. Mandaların başlıca reprodüktif problemleri; repeat breder, postpartum sorunlar ve anötrüstür. Üreme performansının optimizasyonu ve verimin arttırılması gebelik, doğum ve postpartum dönemde takip edilerek yaşanacak aksaklıkların giderilmesi ile mümkün olacaktır.

Anahtar Kelimeler: Manda, Üreme, Gebelik, İnfertilite

Reproduction and reproductive problems in buffaloes

Abstract

Buffalo is an animal that lives largely in Asia and is used for its meat, milk, labor, horn and skin. Buffaloes are animals that can make good use of low-quality feed without choosing feed and can adapt to different environmental conditions. In Turkey, buffalo breeding is mainly done for milk production, with meat production being the next purpose. On the other hand, meat and milk production values per buffalo in our country are far behind countries with high buffalo breeding. For this reason, it is necessary to expand pure breeding and breed development activities that will increase productivity. According to 2021 data in our country, the number of buffaloes increased to 185,574 and raw milk production increased to 63,943 tons. Lactation milk yield of buffalos raised in Turkey varies between 654-761 kg. The age of reaching puberty in buffalos varies between 13-22 months and 36-40 months. Anatolian Buffaloes reach puberty in 13-15 months, swamp buffalos in 21-48 months and River buffalos in 15-18 months. In another approach, the adult reaches puberty when he reaches approximately 60% of his body weight (250 to 400). The estrous cycle is 21 (17-26) days on average. The cycle varies between 20-22 days in River buffaloes and 19-20 days in Swamp buffaloes. The incidence of irregular estrus in buffalos is quite high. The estrous cycle of buffalos is divided into 4 periods: proestrus, estrus, metestrus and diestrus. The average gestation period is 310 days (300-320) in river buffaloes and 330 days (320-340) in swamp buffaloes. The main reproductive problems of buffalos are; repeat breeder, postpartum problems and aneutrus. Optimizing reproductive performance and increasing productivity

will be possible by monitoring pregnancy, birth and postpartum periods and eliminating any disruptions that may occur.
Keywords: Buffalo, Reproduction, Pregnancy, Infertility

GİRİŞ

Manda daha çok Asya kıtasında yaşayan, etinden, sütünden, kaymağından, iş gücünden, boynuzundan ve derisinden faydalanılan bir hayvandır. Mandalar yem seçmeden düşük kaliteli yemleri iyi değerlendirebilen ve farklı çevre koşullarına uyum sağlayabilen hayvanlardır (Sarıözkan, 2011). Mandalar ineklere nazaran daha dirençli ve yetiştirme maliyetleri daha düşük hayvanlardır. Ancak, fertilite ve verim açısından ineklerden oldukça geride kalmaktadır ve yetiştirilmesi de karlı olmamaktadır. Fertilite problemlerinin en önemlileri ise; mevsime bağlı olarak üreme potansiyelindeki değişimler, pubertasın gecikmesi, düzensiz süklus, suböstrus, fertilizasyon oranının düşük olması, postpartum anöstrusun ve buzağılama aralığının uzun olmasıdır (Çizmeci, 2019).

Dünyada manda yetiştiriciliği genellikle aile tipi işletmelerde süt ve et üretimi amacıyla yapılmaktadır (Fao, 2022). Türkiye'de manda yetiştiriciliği; Ege Bölgesi'nde Afyon'da; Marmara Bölgesi'nde İstanbul'da; İç Anadolu Bölgesi'nde Sivas ve Yozgat'ta; Güneydoğu Anadolu Bölgesi'nde Diyarbakır'da; Doğu Anadolu Bölgesi'nde Muş'ta; Karadeniz Bölgesi'nde Samsun, Sinop, Çorum ve Amasya'da yoğunlaşmıştır (Özkan ve ark., 2017). Dünya genelinde 2020 yılında küçükbaş ve büyükbaş hayvan sayısı yaklaşık olarak 4.1 milyar olup, bunun %58.33'ü küçükbaş, %41.67'sini ise büyükbaş hayvan oluşturmaktadır. Yıllık toplam çiğ süt üretimi 900 milyon ton civarında olup 2020 yılı dünya süt üretiminin %81'i inek sütü, %15'i manda, %3,52'si küçükbaş ve %0,35'i ise deve sütüdür (Fao, 2022). Türkiye'de 2020 yılında manda varlığı 192.489 iken, 2021 yılında ise 185.574'e gerilemiştir (TÜİK 2022; HAYGEM, 2022). Türkiye'de 2022 yılında üretilen toplam sütün %92,3'ü inek sütü, %7,4'i küçükbaş (koyun ve keçi) sütü ve %0,3'ü ise manda sütüdür (TÜİK, 2022).

MANDALARDA ÜREME

Üreme Organlarının Anatomisi

Dişi üreme sistemi tübüler genital organlardan oluşur (Dyce ve ark., 2004, Hafez ve Hafez, 2004). Sığır ve manda, dişi üreme yolları anatomisi açısından benzerdir, ancak mandada serviksin daha dar olması, serviksin daha kalın olması, ovaryumların daha küçük ve uzun olması gibi bazı farklılıklar bulunmaktadır (Tonizza de Carvalho ve ark., 2014). Dişi genital sistem, gametler ve hormonlar üreten ovaryumları; uterusun bir uzantısı olan ve oosit, sperm ve salgıların taşınmasını sağlayan oviduktu; embriyonik ve fetal gelişimin devam ettiği uterusu; çiftleşme ve fetüsün geçiş kanalı olan vajina ve vulvayı içermektedir (Hafez ve Hafez, 2004, Tonizza de Carvalho ve ark., 2014). Mandalarda ovaryum folikülleri de ineklere nazaran küçüktür ve gelişmiş bir folikül 8 mm civarındadır ve rektal muayene sırasında Corpus Luteum (CL) ile karıştırılabilmektedir. CL küçüktür genelde ovaryuma gömülüdür ve belirgin bir taç yapmaz (Çizmeci, 2019).

Mandaların uterus ve serviksleri de ineklerden oldukça küçüktür ancak uterus duvarı daha kaslı ve kalındır. Kornuların arasındaki bağlar kuvvetli olduğu için uterus kornuları daha kıvrıktır. Gebe olmayan bir mandada uterus pelvik boşlukta yer almaktadır. Servikal kanal daha dardır ve halka sayısı da ineklerden daha azdır (Napolean ve Quayam, 1996). Servikal darlık özellikle düvelerde suni tohumlama uygulamalarını güçleştirmektedir (Carvalho ve ark., 2010). Mandalarda vulva daha gevşek, sarkık ve koyu pigmentlidir, ayrıca klitorisleri de daha büyüktür. Mandalarda malak vücutbüyüklüğünün anne vücut büyüklüğüne oranının küçük olması sayesinde doğum daha kısa sürede ve kolay olmaktadır (Tonizza de Carvalho ve ark., 2014).

Pubertas

Manda düveleri genellikle yetişkin vücut ağırlıklarının yaklaşık %55-60'ına ulaştıklarında pubertasa ulaşırlar. Pubertasa erişme yaşı ise 18 ila 46 ay arasında değişebilmektedir (Jainudeen ve Hafez, 1993). Mandalarda pubertasa ulaşma yaşı 13-22 ay ile 36-40 aylar arasındadır ve genellikle Anadolu Mandaları 13-15 ay, Nehir mandaları 15-18 ay, bataklık mandaları 21-48 ayda pubertasa ulaşmaktadır (Çizmeci, 2019). Pubertasa erişmeyi etkileyen faktörler genotip, beslenme, yönetim, sosyal çevre, iklim, doğum yılı, doğum mevsimi ve hastalıklardır. Uygun koşullar altında nehir tipi mandaların ilk östrusu 15-18 aylıkken, bataklık tipinin ise 21-24 aylıkken göstermektedir (Borghese, 2005). Ergenliğe ulaşılan vücut ağırlığı genotipten güçlü bir şekilde etkilenir ve bataklık tipi için 200-300 kg, nehir tipi için ise 250-400 kg civarındadır. Her ne kadar mandalar sığırlardan daha geç ergenliğe ulaşsa da, daha uzun bir üreme ömrüne sahiptirler ve bu da bu erken ekonomik dezavantajı telafi etmeye yardımcı olabilmektedir (Perera, 2011). Ancak pubertas yaşının gecikmesi, verimin düşmesine ve elde edilecek yavru sayısının azalmasına neden olmaktadır. Mandalarda östrus tespitinin güç olması nedeniyle pubertasın belirlenmesi de güçleşmektedir ve genellikle ilk buzağılama yaşı dikkate alınarak pubertas yaşı tahmin edilmektedir.

Üreme Özellikleri

Mandalar poliöstrik hayvanlardır ve yıl boyu östrus gösterirler. Fakat mevsimsel üreme özellikleri gösterdikleri de bildirilmektedir. Tropikal bölgelerde genelde sonbahar ve kış aylarında, ılıman iklim kuşağında ise ilkbahar sonu ve yaz aylarında üreme faaliyetleri sergilemektedirler (Perera, 2008). Mandalarda yaz aylarında sıcaklık stresine bağlı anöstrus görülebilmektedir (Roy ve Praksh, 2007). Fotoperiyodun sabit olduğu tropik bölgelerde yağışlardan etkilenirler ve sikluslarında değişimler yaşanabilir (D'Occhio ve ark., 2020). Ülkemizde bulunan Anadolu Mandalarının östrus ve doğumları genellikle Nisan-Ağustos ayları arasında görülmektedir (Çizmeci, 2019).

Mandalarda yetersiz bakım ve besleme sebebiyle reprodüktif sorunlarla karşılaşılmaktadır. Mandaların diğer reprodüktif problemleri ise, inaktif ovaryum, pubertasın gecikmesi, östrus semptomlarının zayıf olması, suböstrus ya da düzensiz siklus görülmesi ve mevsime bağlı şekillenebilen aöstrusun uzamasıdır. İlk buzağılama yaşı ortalama 3-4 yaş arasındadır. Ancak genellikle birçok manda ilk yavrusunu çok daha ileri yaşta doğurmaktadır (D'Occhio ve ark., 2020).

Seksüel Sikluslar

Mandaların östrus siklusu süresi ineklere benzer olup 17 ila 26 gün arasında değişmekle birlikteortalama 21-23 gün civarındadır (Jainudeen ve Hafez, 1993). Olumsuz çevre koşulları, beslenme, steroid hormonların salgılanmasındaki düzensizlikler gibi çeşitli faktörlere bağlı olarak sikluslar kısa (1–10 gün), normal (11–30 gün), uzun (31–60 gün) veya çok uzun (61–90 gün) olabilmektedir (Nanda ve ark., 2003). Siklusların uzamasında östrüs takibinin doğru yapılamaması, suböstrüs, ovulasyonun gecikmesi, implantasyonda yaşanan sorunlar, embriyonik ölümler etkili olmaktadır. Siklusların kısalması ise erken luteal regresyona bağlı şekillenmektedir (Perera, 2011). Östrus süresi nehir ve bataklık mandalarında benzerdir ve 5 ile 27 saat arasında değişmektedir ve ovulasyon östrusun başlangıcından yaklaşık 24-48 saat (ortalama 34 saat) sonra veya östrusun bitiminden 6-21 saat (ortalama 14 saat) önce gerçekleşir (Kanai ve ark, 1990; Perera, 1999). Sıcak iklimlerde östrus süresi genellikle daha kısadır ve östrus belirtileri daha çok gece veya sabahın erken saatlerinde ortaya çıkar (Perera, 2011).

Mandalarda östrus siklusu sırasında ovaryumun foliküler dinamikleri ineklere benzer. Östrus siklusu esnasında 1-3 foliküler dalga şekillenebilir, ancak yarısından fazlasında iki dalgalı siklus görülür (Baruselli ve ark., 1997; Presicce ve ark., 2005; Neglia ve ark., 2007). Mandalarda östrusun başlangıcı ile LH artışı arasındaki süre 1 ila 12 saat arasında değişebilir. Ovulasyon, LH dalgalanmasından 26 ila 33 saat sonra meydana gelir (Seren ve ark., 1995). Östrusun dış belirtileri (suböstrus) olmaksızın endokrin aktivitede önemli farklılıklar olduğu görülmektedir. Mandalarda östrus siklusu sırasında kan östradiol-17β, kan ve süt progesteron konsantrasyonlarındaki değişimler ineklerdekine benzerdir. Mandadaki CL sığırlara göre daha küçüktür ve dolaşımdaki progesteron (P4) konsantrasyonları da sığırlara göre daha düşük seyreder (Mondal ve Prakash, 2004). Siklik aktivitenin belirlenmesinde 1 ng/ml progesteron değeri eşik kabul edilmektedir. Süt sığırlarıyla karşılaştırıldığında mandalarda östrus belirtilerinin hafif olması, dolaşımdaki östradiol-17-β konsantrasyonunun düşük olmasından kaynaklanıyor olabilir (Seren ve ark., 1995; Singh ve ark., 2000; Roy ve Prakash, 2009).

Östrus Davranışları

Östrus davranışlarında manda ve sığır arasındaki temel fark, östrusun davranışsal belirtilerinin belirgin olmaması ve dişiler arasındaki eşcinsel davranışın daha nadir olmasıdır. Huzursuzluk, ayakta durma, hareketlerin artması, sütün azalması, heteroseksüel davranış, böğürme, vulvada ödem, çara akıntısı, sık idrar yapma ve erkek atladığında durma gibi belirtiler görülebilmektedir (Perera, 2011). Belirtilerin hepsi aynı anda görülmez ancak mandaların hemen hepsinde vajinal akıntı bulunur. Östrustaki hayvanların yaklaşık yarısında vulvada ödem ve hiperemi görülür (Warriach ve ark., 2015).

Gebelik ve Doğum

Mandalarda üreme, yıllardan bu yana dişi ve erkeğin yan yana olması ve doğal çiftleşmeye dayanmaktadır. Östrusların senkronize edilerek suni tohumlama ile elde edilen gebelik oranları oldukça düşüktür. Dişilerde gün uzunluğunun arttığı dönemde (ilkbahar-yaz) yapılan senkronizasyon denemelerinde inaktif ovaryum oranının artması gebelik oranını düşürmektedir ayrıca suni tohumlama uygulandığında gebelik oranları daha da düşmektedir (Çizmeci, 2019).

Mandada embriyonik gelişimin genel özellikleri diğer geviş getiren hayvanlarla benzer olmakla birlikte bazı önemli farklılıklar da bulunmaktadır. Akdeniz nehri mandasında, embriyolar tohumlamadan sonra 74 ila 100 saat oviductta kalır ve 4,5-5 günde uterusa erken morula aşamasında ulaşır (Anwar ve Ullah, 1998). Akdeniz ve Nili-Ravi mandalarında kompakt morulalar östrustan 125 ila 152 saat sonra, blastosist ise 141 saatten itibaren gözlenir (Mehmood ve ark., 1989).

Mandalarda gebelik ve doğum oranları da mevsime göre değişiklik göstermektedir. Doğumların çoğunluğu ülkemizde Nisan-Ağustos aylarında daha sıcak iklime sahip ülkelerde ise Ekim ve Şubatayları arasında olmaktadır (Çizmeci, 2019). Mandalarda gebelik süresi 300 ila 330 gün arasında değişmekte olup, nehir türleri için ortalama 310 gün, bataklık türleri için ise 320 gün civarındadır. Gebeliğin erken tanısı yaklaşık 20. günden itibaren ultrasonla (Presicce ve ark. 2004), 45. günden itibaren ise rektal palpasyonla yapılabilmektedir. Muayane teknikleri ve bulguları temel olarak ineklerdekine benzerdir. Ancak gebelik süresinin farklı olması nedeniyle fötusun gelişim dönemlerinde de bazı farklılıklar olacağı da göz ardı edilmemelidir. Laboratuvar tanı yöntemi, tohumlamadan 20-23 gün sonra progesteron analizine dayanmaktadır (Perera, 2008). Mandada doğum sürecindeki progesteron, östrojen ve prostaglandin $F_2\alpha$'da meydana gelen değişiklikler ineklerdekine benzerdir. Uterus involüsyonu genellikle doğumdan sonraki 25-35 günde tamamlanır (Lohan ve ark., 2004) ve laktasyon bu gelişme süresini kısaltabilir (Usmani ve ark., 1990)

Mandalarda doğumdan 1-2 hafta önce abdominal genişleme, memelerin gelişmesi, inmesi ve vulvada ödem gibi belirtiler görülmeye başlar. Doğum süresindeki hormonal değişimler ineklerdekine benzer seyretmektedir. Doğumu malak başlatır ve kortizol seviyesinin artmasıyla serum progesteron seviyesi hızla düşer. Doğumun birinci aşaması olan serviksin açılması ortalama olarak 1-2 saat kadar sürmektedir. Yavrunun kanaldan çıkmasına kadar geçen dönem ikinci dönemdir. Bu dönem genellikle 30-60 dakika kadar sürmektedir. Yavru zarlarının atılmasını içeren üçüncü dönem ise 4-6 saat kadar sürmektedir (Ahmad, 2003)

Doğum sürecinin ineklerden kısa olması güç doğum insidensinin de ineklere oranla düşük olmasının nedenleri arasındadır. Doğum kanalının malağın boyutuna oranla büyük olması güç doğum oranını azaltmaktadır. Ayrıca malakların %95'i normal prezentasyon ve pozisyonda doğum kanalına girer. Ancak doğumların az bir kısmında yavru kanala ters olarak girer ve güç doğum şekillenebilir (Purohit ve ark., 2011). Güç doğum olgularının çoğunluğunu (%59,16) uterus torsiyonu gibi (%83,33) anneye bağlı nedenlerden kaynaklanmaktadır (Srinivas ve ark., 2007). Anneye bağlı güç doğum sebepleri arasında; pelvik anormaliteler, vulvave vajinada görülen tıkanıklık, daralma yada yırtıklar, genital kanal tümörleri, torsiyo uteri,uterus inertiası vb. bulunmaktadır (Purohit ve ark., 2011). Yavruya bağlı nedenlerin en önemlisi ise geliş bozukluklarıdır (Srinivas ve ark., 2007).

Post-Partum Dönem ve Sorunları

İneklerde olduğu gibi, dişi mandalarda da yeni bir gebelik şekillenmesi için hem uterusun involе olması hem de östrus siklusunun yeniden başlaması gerekir.

Emzirilen bataklık mandasında uterusun involüsyonu 28 (16-60) günde tamamlanırken, elle sağım yapılan mandalarda bu süre 45 güne uzamaktadır. Uterus involüsyonu normal doğumlarda güç doğumlara göre daha erken, emzirilenlerde emzirmeyen veya sağılan mandalara göre daha erken ve düşük süt verenlerde yüksek süt verenlere göre daha erken meydana gelir (Jainudeen, 1986). Uterus involüsyonunu etkileyen faktörler doğum sezonu, doğum sayısı, güç doğum, beslenme ve enfeksiyonlardır. Postpartum anöstrus ya da asiklik dönem mandalarda oldukça değişkendir. Bu dönem genellikle ineklerden daha uzundur ve infertilitenin başlıca nedenlerindendir.

Doğumdan sonra östrus siklusunun yeniden başlaması, buzağılama aralığının uzamasına neden olan önemli bir sorundur. Mandalarda buzağılama aralığı mevsim, bakım ve besleme gibi faktörlere bağlı olarak 400 ile 600 gün arasında değişmektedir. Sığırlarda olduğu gibi hipofiz fonksiyon bozukluğu doğum sonrası inaktif ovaryumun önemli bir nedenidir. Foliküler aktivitede bir artış, doğumdan sonraki 30 ila 60. günler arasında, özellikle bir önceki gebeliğin şekillenmediği kornu tarafındaki ovaryumda meydana gelir, ancak ovulasyon oranı düşüktür. Sürekli emziren ve bakım beslemenin iyi olmadığı mandalarda 150-200 güne kadar ovaryumda aktivite görülmeyebilir (Jainudeen ve Hafez, 2016).

Bataklık mandalarında postpartum dönemde östrus ve ovulasyonlarda gecikme nehir mandalarından daha sık rastlanmaktadır. Ayrıca postpartum ilk 1-2 östrus siklusu normalden daha kısadır. İlk iki siklustan sonra ise luteal dönem uzayabilir ya da inaktif onaryuma bağlı siklik aktivite durabilir. Ancak sürüde boğa bulunması postpartum siklik düzensizlik oranını azaltabilir ve ovulasyon zamanını düzenlenmesine yardımcı olabilir (Jainudeen ve ark., 1983).

Kış veya ilkbaharda doğum yapan mandalarda yaz veya sonbaharda doğum yapanlara oranla siklik aktivite daha geç başlar. Postpartum ilk östrus mandalarda beslenme, gün ışığı süresi, yağış ve sıcaklıktan etkilenmektedir. Sıcaklıkların yükselmesi boğaların libidosunu düşürmekte ve dişilerde üreme fonksiyonlarını olumsuz etkilemektedir. Siklik aktivite genellikle gün uzunluğunun kısaldığı ve havaların serinlediği dönemde başlamaktadır (Gordon, 1996).

Mandaları etkileyen başlıca üreme problemleri anötrüs, repeat breder ve postpartum sorunlardır. Doğum esnasında yaşanan problemler uterus involusyonunda gecikme ve metritis gibi problemlere yol açmaktadır. Doğumla ilişkili sorunlar arasında distosi, ölü doğum, abortus, retensiyo sekundinarum gibi olgular yer alır.

Ölü doğum ve distosi, hayvanın gelecekteki üreme performansını düşüren en önemli faktörler olarak kabul edilmiştir. Metritisin şekillenme oranını arttırır ve plasentanın atılımını geciktirir. Bu tür durumlarda uterus involusyonu ve ovaryum aktivitesinin başlamasında gecikmeler yaşanır. Devamında sekonder enfeksiyonlar ve anormalliklerin yaşanma oranı da artmaktadır. Üreme performansının optimizasyonu, mandaların gebelik, doğum ve postpartum dönemde dikkatle takip edilmesi ve yaşanacak aksaklıkların giderilmesi ile mümkün olacaktır (Gordon, 1996).

Retensiyo, endometrit ve prolapsus uteri gibi postpartum problemler, doğum sonrası mandaların ovaryum fonksiyonu üzerinde olumsuz etkilere sahiptir. Ayrıca, metabolik bozukluklar, verim ve mastit gibi problemler ise ovaryum fonksiyonlarında azalmaya neden olmaktadır. Postpartum dönemde genital sistem komplikasyonları arasında kanama, vulval hematom, laserasyonlar, üreme organlarında (vajina, uterus) yırtılma, perineal yaralanmalar, pelvik organların distale yer değiştirmesi (vajina, uterus, mesane, perivajinal yağ) ve retensiyo yer almaktadır (Purohit, 2012). Metabolik problemler arasında ise süt humması, hipofosfatemi ve ketozis yer almaktadır (Purohit ve ark., 2014b). Meme bezinin komplikasyonları arasında meme ödemi, hemolaksi, hipogalaksi ve mastit gibi durumlar yer almaktadır. Mandalarda genital kanalın gevşemesi ve genişlemesi ile ilgili sorunlar nadiren görüldüğü için doğuma bağlı genital sistem yaralanmaları da nadir görülmektedir (Purohit ve Markandeya, 2015).

Ekzojen Hormonlarla Östrus Siklusunun Kontrolü

Geçmişte mandada östrus senkronizasyonu için kullanılan prosedürler ampirik olarak sığırlar için geliştirilen prosedürlere dayanıyordu. Senkronizasyon protokolleri, prostaglandinler kullanılarak luteal fazın kısaltılması veya progestagenler kullanarak luteal fazın uzatılmasını içermektedir. Çift doz prostaglandin uygulamaları senkronizasyon amacıyla başarıyla uygulanabilmektedir ancak daha iyi senkronizasyon, gebelik ve yavru elde etmek için foliküler gelişimin manipülasyonu gereklidir (De Rensis ve Lopez-Gatius 2007). Bu amaçla ovsynch protokolü kullanılabilmektedir (Paul ve Prakash 2005). Ovsynch, mandalarda östrus siklusunun senkronize edilmesini sağlar ve sabit zamanlı suni tohumlama ile kombine edildiğinde ovulasyon ve fertilizasyon oranlarını arttırabilir. Ötsrus siklusunun düzenlenmesi ve ovulasyonun uyarılması, östrus belirtilerinin takip edilmesi için ihtiyaç duyulan gözlem süresini ortadan kaldırarak mandanın üreme potansiyelinin arttırılmasına olanak sağlamaktadır (Perera, 2011). Mandalarda anöstrusların üstesinden gelmek için gonadotropin salgılatıcı hormon (GnRH) ve analoglarının enjeksiyonu veya progesteron içeren aparatların 10-12 gün boyunca intravajinal veya subkutan uygulanmasını içeren protokoller uygulanabilmektedir (Perera, 2008).

Mandalarda seçilen protokole ek olarak, başarıya ulaşmak için aşağıdaki faktörlerin de ele alınması gerekir:

(a) Bakım ve beslenme durumu iyi olan ve hastalıktan ari olan hayvanların seçimi,

(b) Hayvanların barındırıldığı yerlerin düzgün olması ve stres faktörlerinin minimize edilmesi (taşıma, kalabalık, iklim vb)

(c) Mevsimsel üreme dikkate alınarak siklusların başlayıp östrusların daha düzenli ve toplu olduğu dönemler tercih edilmelidir (Drost, 2007).

Başarılı suni tohumlama, spermanın kalitesine, hekimin tecrübesine, tohumlamanın doğru zamanlanmasına ve dişinin sağlık durumuna bağlıdır. Tohumlama için en uygun zaman östrus başlangıcından sonraki 8-12. saattir (Drost, 2007). Suni tohumlama; mandaların genetik özelliklerinin geliştirilmesine yardımcı olmaktadır, ancak mandalarda östrus davranışlarının zayıf olması ve gebelik oranlarının düşük olması nedeniyle tohumlama hala yaygın olarak kullanılamamaktadır. Tüm bu olumsuz faktörlere rağmen, ovsynch ve suni tohumlama kullanılarak uygulanan protokoller geliştirilmiştir (Perera, 2008).

MANDALARDA BİYOTEKNOLOJİK UYGULAMALAR

Günümüzde mandalarda üreme performansını arttırmak için suni tohumlama, multiple ovulasyon ve embriyo transferi (MOET), in vitro embriyo üretimi, kriyobiyoloji, cinsiyeti belirlenmiş malak üretimi ve klonlama gibi biyoteknolojik yöntemler kullanılmaya başlanmıştır (Çizmeci, 2019)

Mandada ilk başarılı embriyo transferi Amerika Birleşik Devletleri'nde gerçekleştirilmiştir. Ancak mandalarda bu biyoteknolojik uygulamanın başarı oranı reprodüktif verime paralel olarak oldukça düşüktür. Süperovule edilen hayvan başına düşen transfer edilebilir embriyo ortalaması 1'den azdır. Manda ovaryumunda ortalama 12.000 (ineklerde 133.000) primer folikül bulunmaktadır (Warriach ve ark., 2015).

Embriyo transferi, donör hayvanın seçimi, taşıyıcı hayvanın seçimi, donörün senkronizasyonu, donörün tohumlanması, embriyonun toplanması, embriyonun değerlendirilmesi, embriyo transferi ve taşıyıcının bakımı aşamalarını içermektedir. Yardımcı üreme teknikleri, konjenital üreme problemlerinin üstesinden gelmek, üreme performansını geliştirmek, süt üretimini arttırmak, yüksek verim ve genetik özelliklerin yaygınlaştırmak ve hastalıkların kontrol etmek amacıyla kullanılmaktadır. Bu teknolojiler, gelişim fizyolojisi, ıslah konularında

temel araştırmaların sürdürülmesi ve klonlama ve transgenesis gibi yeni ortaya çıkan biyoteknolojilerin ticari amaçla uygulanması için mükemmel bir embriyo kaynağı sağlar (Gasparrini, 2002; Manjunatha ve ark., 2009).

İNFERTİLİTE

Manda, postpartum uzun süreli anöstrus ve reprodüktif verimliliği etkileyen suböstrus göstermesi ile bilinmektedir. İnfertilitenin altında yatan nedenler pubertasa geç ulaşma, iklim, mevsim, yönetim, beslenme, düşük vücut kondüsyon skoru ve hastalıklardır (Dhami ve ark., 2020). Postpartum anöstrusun uzaması ve repeat breeder manda yetiştiriciliği ve süt endüstrisi için ekonomik kaybın ana nedenleri olarak kabul edilmektedir (El-Wishy, 2007). İnfertil mandaların tedavi edilmesi süt üretimlerinin arttırılmasında önemli bir faktördür. Asiklik ve repeat breeder mandaların normal siklus göstermesini sağlamak, üreme verimliliğini artırmak, böylece buzağılama aralığını ekonomik sınırlara getirmek amacıyla dolaşımdaki plazma progesteron seviyelerini modüle etmek için çeşitli hormonal preparatlar ve protokoller kullanılmaktadır (Mirmahmoudi., 2014; Mungad., 2017).

Anöstrus mandalarda infertilitenin en önemli nedenidir. Anöstrüs ise gerçek anöstrus, suböstrus, luteal aktivitenin uzaması, inakrtif ovaryum, ovaryum kist ve tümörleri olarak sınıflandırılabilmektedir. Mandalarda infertilitenin bir diğer önemli sebebi ise yangı oluşturarak anöstrus süresini uzatan ve fertilite oranını düşüren uterus enfeksiyonlarıdır.

Enfeksiyöz etkenler, üreme sistemini doğrudan etkileyerek veya genel sağlık durumunu dolaylı olarak etkileyerek üreme performansını olumsuz yönde etkilemektedir. Mandalar üzerinde yapılan çeşitli çalışmalarda birçok bakteri, virüs, mantar ve protozoa izole edilmiştir, ancak bunların manda türlerinde yaygın hastalık veya infertilitedeki önemi henüz tam olarak belirlenmemiştir (Purohit ve ark., 2014a).

Sürü yönetiminde metabolik profillerin değerlendirilmesi ve infertilite nedeni olarak enfeksiyöz ajanların belirlenmesi önemli etkenlerdir. Sürü infertilitesinin değerlendirilmesinde kan biyokimyasına bakılması faydalı minerallerin eksikliği hakkında bilgi vermektedir. Sürü infertilitesinin nedeni olabilecek muhtemel patojenlerin izolasyonu için uygun numuneler alınarak dikkatli ve uygun bir şekilde yetkili laboratuvarlara gönderilmelidir. İlk buzağılama yaşının uzaması (>42 ay), gebe kalma aralığının uzaması (24 ay), sürünün optimumun altında buzağılama oranına sahip olduğunu düşünmek için yeterli nedenlerdir. Bu problemlerin varlığında beslenme, yönetim ve sıcaklık stresine yönelik değerlendirmeler yapılmalıdır. Bir manda sürüsünde abortus vakalarının yüksek olması durumunda sürü acil olarak incelenmeli ve potansiyel bulaşıcı organizmaların izolasyonu için uygun örnekler yetkili teşhis laboratuvarlarına gönderilmelidir. Gebeliğin 6. ayından sonra abort yapan mandalar, *Brucella abortus* veya *Brucella melitensis* yönünden değerlendirilmeli, etken izolasyonu yapılmalı ve uygun karantina önlemleri alınmalıdır (Purohit ve ark., 20141).

Uterus enfeksiyonları içerisinde en önemli olan puerperal akut (toksik) septik metritistir. Pureperal akut septik metritli mandaların %14,3'ünde güç doğum, %52,4'ünde retensiyo sekundinarum %2,04'ünde prolapsus vagina, %11,9'unda prolapsus uteri, %19,1'inde ise çeşitli faktörlerin etkili olduğu belirtilmektedir. Puerperal akut septik metritisli mandalarda anoreksiya, depresyon, sallantılı yürüyüş, paraliz, ayağa kalkmakta zorlanma, siyah renkli dışkı ile kabızlık, sık ve tekrarlayan ağrılı ıkınma gibi belirtiler mevcuttur. Muayene sırasında rektal ısının 40.9±0.18°C, nabız sayısı ve solunum hızının arttığı görülebilir. Vajinal muayenede sulu, kahverengi ve pis kokulu akıntı tespit edilir (Purohit ve Markandeya, 2015).

KAYNAKLAR

Ahmad N 2003. Reproduction in the buffalo. In: Noakes DE, Parkinson TJ, England GCW (eds), Arthur's Veterinary Reproduction and Obstetrics. 8th edition China: Saunders, pp789-799.

Anwar M, Ullah N 1998. Early development and location of embryos in the reproductive tract of nili ravi buffalo [Bubalus bubalis]: a retrospective analysis. Theriogenology, 49:1187–93.

Baruselli PS, Mucciolo RG, Visintin JA, Viana WG, Arruda RP, Madureira EH, Oliveira CA, Molero-Filho JR 1997. Ovarian follicular dynamics during the estrus cycle in buffalo (Bubalus bubalis). Theriogenology, 47:1531–47.

Borghese 2005. Technical Series 67. Food and Agriculture Organization, Rome, Italy. https://www.fao.org/3/ah847e/ah847e00.htm [18.09.23]

Carvalho NAT, Soares JGS, Souza DC, Maio JRG, Sales JNS, Martins Júnior B, Macari RC, D'Occhio MJ, Baruselli PS 2010. Buffalo heifer's ovulation synchronization with GnRH or EB to FTAI during the off breeding season (partial results). Acta Sci Vet, 38:725.

Çizmeci SÜ 2019. Reprodüksiyon. In: Manda El Kitabı Dik B, Avcı O (eds), Güneş Tıp Kitabevi, İstanbul, Türkiye pp:147-155.

D'Occhio MJ, Ghuman SS, Neglia G, della Valle G , Baruselli PS, Zicarelli L , Visintin JA, Sarkar M, Campanile G 2020. Exogenous and endogenous factors in seasonality of reproduction in buffalo: A review Theriogenology, 150: 186-192.

De Rensis F, López-Gatius F 2007. Protocols for synchronizing estrus and ovulation in buffalo (Bubalus bubalis): a review. Theriogenology 67, 209–216.

Dhami AJ, Hadiya KK, Patel JA, Parmar SJ, Chaudhari DV 2020. Impact of different estrus induction and ovulation synchronization protocols in addressing infertility in buffaloes. Buffalo Bulletin.39:3

Drost M 2007. Advanced reproductive technology in the water buffalo. Theriogenology, 68(3):450-3

Dyce KM, Sack WO, Wensing CJG 1996. Textbook of veterinary anatomy. 2nd ed. Philadelphia: WB Saunders, pp:856.

El-Wishy AB 2007. Review The postpartum buffalo II. Acyclicity and anestrus. Animal Reproduction Science. 97:216–236.

FAO 2022 , Food and Agriculture Organization (FAO), http://www.fao.org/faostat/ [15.09.2023]

Gasparrini B 2002. In vitro embryo production in buffalo species: state of the art. Theriogenology 57:237-256

Gordon I 1996. Controlled reproduction in cattle and buffaloes. CABI, Wallingford, UK. vol. 1. pp. 452

Hafez ESE, Hafez B 20004. Reprodução animal. 7th ed. São Paulo: Manole, pp:513.

HAYGEM 2022. Hayvancılık Genel Müdürlüğü. "Hayvancılık Verileri, Ocak 2022", https://www.tarimorman.gov.tr/sgb/Belgeler/SagMenuVeriler/HAYGEM.pdf [05.09. 2023]

Jainudeen MR, Bongso TA, Tan HS 1983. Postpartum ovarian activity and uterine involution in the suckled swamp buffalo (Bubalus bubalis). Anim Reprod Sci, 5:181..

Jainudeen MR 1986. Reproduction in the water buffalo. In: Morrow DA (ed), Current Therapy in Theriogenology. Philadelphia: WB Saunders.

Jainudeen MR, Hafez ESE 2016. Cattle and Buffalo. Reproduction in Farm Animals, 157–171. doi:10.1002/9781119265306.ch11

Jainudeen MR, Hafez ESE 1993. Cattle and buffalo. In: Hafez ESE (ed), Reproduction in Farm Animals, 6th ed. Lea and Febiger, Philadelphia, USA, pp. 315–329.

Kanai Y, Abdul-Latief T, Ishikawa N, Shimizu H 1990. Behavioural and hormonal aspects of the oestrous cycle in swamp buffaloes reared under temperate conditions. In: Domestic Buffalo Production in Asia. International Atomic Energy Agency, Vienna, Austria, pp: 113–120.

Lohan IS, Malik RK, Kaker ML 2004. Uterine involutionand ovarian follicular growth during early postpartumperiod of Murrah buffaloes (Bubalus bubalis). Asian-austra-las J Anim Sci.17:313–316

Manjunatha BM, Ravindra JP, Gupta PSP, Devaraj M, Nandi S 2009. Effect of breeding season on in vivo oocyte recovery and embryo production in non-descriptive Indian river buffaloes (Bubalus bubalis). Anim Reprod Sci, 111: 376-383.

Mehmood A, Anwar M, Javed MH 1989. Superovulation with PMSG beginning on three different days of the cycle in Nili Ravi buffaloes [Bubalus bubalis]. Buffalo J, 5:79–84.

Mirmahmoudi R, Souri M, Prakash BS 2014. Endocrine changes, timing of ovulation, ovarian follicular growth and efficacy of a novel protocol (Estradoublesynch) for synchronization of ovulation and timed artificial insemination in Murrah buffaloes, 2:237-242. Doi: 10.1016/j.theriogenology.2013.09.016

Mondal M, Prakash BS 2004. Effects of long-term growth hormone releasing factor (GRF) administration on pattern of GH and LH secretion in growing female buffaloes (Bubalus bubalis). Reproduction,127:45–55.

Mungad KS, Patel JA, Dhami AJ, Hadiya KK, Borkhatariya DN, Sarvaiya NP 2017. Use of estrus induction strategy in true anestrus buffalo for reducing non-productive period. indian Journal of Animal Reproduction, 2:15-19.

Nanda AS, Brar PS, Prabhakar S 2003. Enhancing reproductive performance in dairy buffalo: major constraints and achievements. Reproduction, 61: 27–36.

Napolean RE, Quayam SA 1996. Biometrical studies of the female genitalia of non-descript buffalo (Bubalus bubalis). Indian J Anim Sci, 66:1269-1270.

Neglia G, Natale A, Esposito G, Salzillo F, Adinolfi L, Zicarelli L, Francillo M 2007. Follicular dynamics in synchronized Italian Mediterranean buffalo cows. Ital J Anim Sci, 6:611–4.

Özkan ZS, Arslan İ, Uçum F, Canik BU 2017. Tarımsal ekonomi ve politika geliştirme enstitüsü tepge. Samsun ilinde manda yetiştiriciliği faaliyetine yer veren işletmelerin mevcut durum analizi, Tepge Yayın No: 292. https://arastirma.tarimorman.gov.tr/tepge/Belgeler. [15.09.2023]

Paul V, Prakash BS 2005. Efficacy of the ovsynch protocol for synchronization of ovulation and fixed-time artificial insemination in Murrah buffaloes (Bubalus bubalis). Theriogenology 64:1049–1060.

Perera BMAO 1999. Reproduction in water buffalo: comparative aspects and implications formanagement. J Reprod Fertil, 54:157–168.

Perera BMAO 2008. Reproduction in Domestic Buffalo. Reprod Dom Anim, 43 (2):200–206.
Perera BMAO 2011. Reproductive cycles of buffalo. Animal Reproduction Science, 124(3-4):194-199.
Presicce GA, De Santis G, Stecco R, Senatore E, De MauroGJ, Terzano GM 2001. Foetal gender determination byultrasound in the Mediterranean Italian buffalo (Bubalusbubalis). Theriogenology, 55:532.
Presicce GA, Senatore EM, De Santis G, Bella A 2005. Follicle turnover and pregnancy rates following oestrus synchronization protocols in Mediterranean Italian buffaloes (Bubalus bubalis). Reprod Domest Anim, 40:443–7.
Presicce GA, Senatore Em, Bella A, De Santis G, Barile VL, De Mauro GJ, Terzano GM, Stecco R, Parmeggiani A 2004. Ovarian follicular dynamics and hormonal profiles in heifer and mixed-parity Mediterranean Italian buffaloes (Bubalus bubalis) following an estrus synchronization protocol. Theriogenology, 61:1343–1355
Purohit GN, Gaur M, Shekher C 2014a. Infertility Management in Female Buffaloes In:Bubaline Theriogenology, Purohit GN (ed), International Veterinary Information Service, Ithaca NY (www.ivis.org), pp:A5727.1114
Purohit GN, Ruhil S, Daga M, Gaur M, Bihani DK, Ahuja A 2014b. Parturition related disorders in the buffalo: A 10 year case analysis. Ruminant Sci, 3:241-244
Purohit GN 2012. Postpartum complications in large animals In: Purohit GN (ed), Domestic Animal Obstetrics Lambert Academic Publishers Germany, pp: 551-603.
Purohit GN, Markandeya NM 2015. Postpartum Compication in the Buffalo. Bubaline Theriogenology, A5718.0115.
Purohit GN, Barolia Y, Shekhar C, Kumar P 2011. Maternal dystocia in cows and buffaloes: a review. Open Journal of Animal Science. 1 (2):41-53
Roy KS, Prakas BS 2009. Plasma progesterone, oestradiol-17β and total oestrogen profiles in relation to oestrous behaviour during induced ovulation in Murrah buffalo heifers. J Anim Physiol Anim Nutr, 93:486-495.
Sarıözkan S 2011. Türkiye'de manda yetiştiriciliği'nin önemi. Kafkas Üniversitesi Veteriner Fakültesi Dergisi, 17 (1): 163-166.
Seren E. Parmeggiani A, Campanile G 1995. The control of ovulation in Italian buffalo. In: Proc. of the Symposium Reproduction and Animal Breeding: Advances and Strategy, Milan, Italy. pp. 265-275.
Singh J, Nanda AS, Adams GP 2000. The reproductive pattern and efficiency of female buffaloes. Anim Reprod Sci, 60-61:593-604.
Srinivas M, Sreenu M, Lakshmi RN, Subramanyam NK, Devi PV 2007. Studies on dystocia in graded Murrah buffaloes: A retrospective study. Buffalo Bulletin, 26: 40-45.
Tonizza de Carvalho NA, Soares JG, Kahwage PR, Garcia AR 2014. Anatomy of the Reproductive Tract of the Female and Male Buffaloes. In: Purohit GN (ed), Bubaline Theriogenology, International Veterinary Information Service, Ithaca NY (www.ivis.org), A5701.0714
TÜİK 2022. Türkiye İstatistik Kurumu. "Tarım ve Hayvancılık İstatistikleri", https://data.tuik.gov.tr/Kategori/GetKategori?p=tarim-111&dil=1 [15.09.2023]
Usmani RH, Dailey RA, Inskeep EK 1990. Effects of limitedsuckling and varying prepartum nutrition on postpartumreproductive traits of milked buffaloes. J Dairy Sci, 73:1564–1570
Warriach HM, McGill DM, Bush RD, Wynn PC, Chohan KR 2015. A Review of Recent Developments in Buffalo Reproduction — A Review. Asian Australas J Anim Sci, 28 (3): 451-455.

ORAL PRESENTATION

Türkiye Piyasasında Bulunan Chia (*Salvia Hispanica L.)* Tohumu Yağı İçeren Yumuşak Jelatin Kapsüller Üzerinde Kalite Kontrol Çalışmaları

Ebru Derici Eker[1] (0000-0002-7094-7625), Mohammed Yahia Asaad[1](0000-0002-0550-8415), Hatice Yıldırım Yaroğlu[2*](0000-0003-4866-313X)

[*1]Mersin Üniversitesi, Eczacılık Fakültesi, Farmasötik Teknoloji Bölümü, Mersin, Türkiye
[2]Mersin Üniversitesi, Sağlık Bilimleri Enstitüsü, Kök Hücre ve Rejeneratif Tıp Anabilim Dalı, Mersin, Türkiye

* e-mail: haticeyildirim@mersin.edu.tr

Özet

Kökeni tarih öncesi çağlara dayanan Chia tohumu yüksek oranda yağ, protein ve diyet lifi içermesiyle süper gıda olarak popülerleşen bir besindir. Chia tohumunda % 17-24 protein, %18-30 lif ve %25-40 oranında yağ mevcuttur. Tohumun yağ içeriğinin % 80'i α-linolenik asit (omega-3) ve linoleik asitten (omega-6) oluşmaktadır. Ülkemizdeki piyasada bulunan preparatları Tarım Bakanlığı izniyle piyasaya sürüldüğü için Sağlık Bakanlığı'ndan ruhsat alan beşeri ilaçlar gibi *in vitro* ve *in vivo* etkinlik, güvenlik ve kalite kontrol testlerinden geçmeleri gerekmemektedir. Çalışmamızda Türkiye piyasasında, değişik firmalarca pazarlanan farklı formülasyonlara sahip chia tohumu yağları ve yumuşak kapsül formülasyonlarının kalite kontrol açısından değerlendirilmesi amaçlanmıştır. Chia tohumu yağı içeren yumuşak jelatin kapsüller ve yağlar üzerinde organoleptik özelliklerin tayini, ağırlık sapması, dezentegrasyon testi, disolüsyon testi, DPPH antioksidan kapasitesi ölçümü ve fenolik madde tayini analizleri yapılmıştır. Elde edilen verilere göre kapsüller organoleptik özellikler açısından uygunluk göstermektedir. Yumuşak jelatin kapsüllerin ağırlık sapması ortalaması A numunesi için 0,7283±0.024g, B numunesi için 1,2947±0.015g olarak bulunmuştur. Yapılan dağılma testinden elde edilen sonuçlara göre kapsüller suni mide vasatında 5 dakika 14 saniye ile 6 dakika 32 saniye aralığında, suni barsak vasatında 5 dakika 14 saniye ile 6 dakika 32 saniye aralığında, suda ise 16 dakika 2 saniye ile 18 dakika 30 saniye aralığında dağılma göstermiştir. Chia tohumu yağı içeren yumuşak jelatin kapsüller ve yağlar üzerindeki kalite kontrol testlerindeki literatüre uygun sonuçlar nedeniyle bitkisel destek olarak kullanımının uygun olacağını düşünmekteyiz.

Anahtar Kelimeler: *Salvia hispanica l.* (chia tohumu yağı), *omega-3,* yumuşak jelatin kapsül, kalite kontrol

Quality Control Studies in The Turkey Market on Soft Gelatin Capsules Containing Chia (*Salvia Hispanica L.*) Seeds Oil

Abstract

Chia seed, which has its origins in prehistoric times, is a food that has become popular as a superfood with its high content of oil, protein and dietary fiber. Chia seeds contain 17-24% protein, 18-30% fiber and 25-40% fat. 80% of the oil content of the seed consists of α-linolenic acid (omega-3) and linoleic acid (omega-6). Since the preparations available in the market in our country are put on the market with the permission of the Ministry of Agriculture, they do not need to pass *in vitro* and *in vivo* efficacy, safety and quality control tests like the human drugs licensed by the Ministry of Health. In our study, it is aimed to evaluate the quality control of chia seed oils and soft capsule formulations with different formulations marketed by different companies in the Turkish market. Determination of organoleptic properties, weight deviation, disintegration test, dissolution test, DPPH antioxidant capacity measurement and phenolic substance determination analyzes were performed on soft gelatin capsules containing chia seed oil and oils. According to the data obtained, the capsules are suitable in terms of organoleptic properties. The mean weight deviation of soft gelatin capsules was 0.7283±0.024g for sample A and 1.2947±0.015g for sample B. According to the results obtained from the disintegration test, the capsules dispersed between 5 minutes 14 seconds and 6 minutes 32 seconds in the

artificial stomach medium, between 5 minutes 14 seconds and 6 minutes 32 seconds in the artificial gut medium, and between 16 minutes 2 seconds and 18 minutes 30 seconds in pure water. We think that its use as an herbal supplement would be appropriate due to the results in accordance with the literature in quality control tests on soft gelatin capsules and oils containing chia seed oil.

Keywords: *Salvia hispanica l.* (chia seeds oil), *omega-3*, soft gelatin capsule, quality control

GİRİŞ

Kökeni tarih öncesi çağlara dayanan Chia tohumu yüksek oranda yağ, protein ve diyet lifi içermesiyle "süper gıda" olarak popülerleşen bir besindir. Chia tohumu, yetiştirildiği iklim ve toprak şartlarına bağlı olarak değişmek üzere, %15-23 oranında protein içerir (Munoz ve ark.,2023). Chia tohumunun protein bileşimi dokuzu esansiyel aminoasit olmak üzere, on sekiz çeşit aminoasit içermektedir Chia tohumu gluten içermediği için çölyak hastalarının diyetleri için de uygun bir besindir. Chia tohumu, Avrupa Gıda Komisyonunun 2009/827/EC numaralı kararına göre "yeni gıda" olarak kabul edilmiştir. Amerikan Gıda ve İlaç Dairesi (FDA), chia tohumunu gıda katkı maddesi olarak değil, bir gıda olarak kabul ettiğini belirtmiştir. Chia tohumunun alerjik olmadığı, herhangi bir toksik etki göstermediği ve gıdalara katılarak yenilebilir olduğu yapılan bilimsel çalışmalarla kanıtlanmıştır (Erdoğdu ve ark.,2017)

Nutrasötikler, çoğunluğunu bitkisel kökenli farmasötik preparatların oluşturduğu diyet destek ürünleridir. Bu ürünler, çoğunlukla, Tarım Bakanlığı izniyle piyasaya sürülmektedir. Sağlık Bakanlığı'ndan ruhsat alan beşeri ilaçlar gibi *in vitro* ve *in vivo* etkinlik, güvenlik ve kalite kontrol testlerinden geçmeleri gerekmemektedir. Bu da pek çok sorunu beraberinde getirmektedir. Küresel anlamda bu konuda sorunlar mevcuttur. Bitkisel ürünlerin toplanma zamanı, toplanma, depolanma, kurutulma zamanları ve işlemleri, etken madde izolasyonu ve standardizasyonu, stabilite ve diğer kalite kontrol çalışmalarının, *in vivo* verilerin eksikliği bu ürünlerin alternatif tedavide ya da konvansiyonel tedaviye destek olarak kullanılmasında önemli faydalar sağlanmasına rağmen kullanımlarını kısıtlayan sorunlardır. Bu sorunların çözümlenmesi için piyasada bulunan preparatların etkinlik, güvenlik ve farmasötik açıdan değerlendirilmesi gerekir Başaran, 2008).

Bu çalışmada, Türkiye piyasasında, değişik firmalarca pazarlanan farklı formülasyonlara sahip chia tohumu yağları ve yumuşak kapsülleri temin edilerek üzerlerinde kalite kontrol testleri yapılmıştır ve böylece etkinlik ve güvenlikleri açısından karşılaştırılmaları amaçlanmıştır. Bu çalışmalara ek olarak chia yağlarının ve yumuşak kapsüllerinin, toplam fenolik madde içeriği ve 2,2-difenil-1pikrilhidrazil (DPPH) radikali giderme aktivitesi tayini yöntemleri kullanılarak antioksidan değerleri karşılaştırılmıştır. Çalışmamızın sonunda elde ettiğimiz verilerle, hemen hemen her yaş grubunda güvenli olduğu düşünülen hem besin desteği hem de şifa beklentisi olarak sıklıkla kullanılan chia tohumu kapsüllerinin ve yağlarının farklı firmalarca piyasaya sürülen ürünleri kalite, etkililik ve güvenlik açısından karşılaştırılmıştır.

MATERYAL VE METOT

Çeşitli firmalarca üretilmiş olan ve eczanelerde satışa sunulan 2 farklı markanın (Zadevital ve Desteque) chia tohumu yumuşak jelatin kapsül formu ve 2 farklı markanın chia tohumu yağı (Zadevital ve Naturoil) formu piyasadan temin edilmiştir. Testlerde gereken tüm hammaddeler ve materyaller Mersin Üniversitesi Eczacılık Fakültesi Farmasötik Teknoloji laboratuvarlarından kullanılmıştır. Farmasötik etkinlik ve kalite kontrol testleri kapsamında yumuşak jelatin kapsüllerde organoleptik özelliklerin tayini, ağırlık testi, dağılma testi (suda, suni mide vasatında ve suni bağırsak vasatında); DPPH antioksidan kapasitesi ölçümü ve fenolik madde tayini yapılmıştır.

Yumuşak jelatin kapsüllerde organoleptik özelliklerin tayininde en az 20 kapsülü ambalajından çıkarıldı ve incelendi. Pürüzsüz ve hasarsız olmalıdır. Fiziksel kararsızlığın kanıtı, sertleşme veya yumuşama, çatlama, şişme, beneklenme veya kabuğun renginin değişmesi gibi fiziksel görünümdeki büyük değişikliklerle gösterilir.

Ağırlık Sapması tayini doldurulan maddenin kötü niteliklerini, doldurma makinelerinin çalışmasındaki bozuklukları veya imalat safhasında kontrollerin iyi yapılıp yapılmadığını tespit eder. Chia tohumu yağı yumuşak jelatin kapsüllerinden her numuneden 10 kapsülün her biri tek tek tartılarak ağırlıkları tayin edildi. Uygun temiz ve kuru bir kesme materyali kullanılarak kapsüller kesildi ve içeriği uygun bir çözücü ile yıkanarak uzaklaştırıldı. Oda ısısında kapsülde kalan çözücünün buharlaşması için nemden koruyarak 30

dakika bekletildi. Her bir kapsül boşken tek tek tartıldı ve boş kapsülün kütlesi ilgili brüt kütleden çıkarılarak içeriğin net kütlesi her kapsül için hesaplandı.

Dağılma Testi kapsamında ilk olarak suda dağılma testi yapıldı. PharmaTest (Almanya) dezentegrasyon cihazında suda sürekli karıştırılarak (100 rpm) dağılma süresi tayin edildi. Suyun sıcaklığı 37°C'ye getirildi. Tüm numunelerden 6 kapsül kullanılarak ayrı ayrı su içinde dağılma süreleri hesaplandı. Suni mide vasatında dağılma testi için Türk Farmakopesi'ne (Avrupa Farmakope Adaptasyonu II-2017) göre 2L suni mide vasatı hazırlandı. Dezentegrasyon cihazına kondu. Her numunenin kapsüllerinden 6'şar adet dezentegrasyon cihazına yerleştirildi. Suni mide vasatının sıcaklığı 37°C'ye getirildi. Sonra her numune için ayrı ayrı kapsüllerin dağılma süresi hesaplandı. Suni bağırsak vasatında dağılma testi için Türk Farmakopesi'ne (Avrupa Farmakope Adaptasyonu II-2017) göre hazırlanan suni bağırsak vasatından 2L dezentegrasyon cihazına konuldu. Her numuneden üçer kapsül kullanılarak dağılma süreleri hesaplandı.

DPPH antioksidan kapasitesi ölçümü bir serbest radikal olan DPPH radikalinin antioksidan madde tarafından yakalanarak mor renginin açılmasının spektrofotometrede ölçülerek tayin edilmesine dayanır. Piyasadan temin edilen farklı chia kapsüllerinden elde edilen metanollu eksraktların her birinden 1'er ml alınarak, 4 ml 0,1 mM DPPH (metanolde) çözeltisi ilave edildi. Karışım karanlıkta ve oda koşullarında 30 dk bekletildi. Sonra spektrofotometrede 517 nm absorbans değerleri okundu. Deney 3 kez tekrarlandı ve aritmetik ortalamaları alındı. % DPPH radikali giderme aktivitesi hesaplandı.

Fenolik madde tayini için farklı chia kapsüllerinden elde edilen metanollu eksraktlardaki fenolik maddeler Folin-Ciocalteu reaktifi (FCR) ile saptandı. Piyasadan temin edilen farklı chia kapsüllerinden hazırlanan eksraktların her birinden 1ml alındı üzerine 45ml distile su ilave edildi. Sonra 1ml FCR eklendi. 3 dk sonra 3ml % 2 lik Na_2CO_3 çözeltisi ilave edilip oda koşullarında manyetik karıştırıcıda 2 saat bekletildi. Sonra spektrofotometrede 760nm'de absorbans değerleri okundu. Deney 3 defa tekrar edildi.

BULGULAR

Organoleptik Özelliklerin Tayini

Çalışmamızda kullandığımız çeşitli firmalarca üretilmiş olan ve eczanelerde satışa sunulan 2 farklı markanın chia tohumu yumuşak jelatin kapsüllerinin organoleptik özellikleri Tablo 1'de gösterilmiştir.

Tablo 1. Chia tohumu kapsülleri Organoleptik Özelliklerin Tayini

Karakteristik	**A Kapsül Numunesi**	**B Kapsül Numunesi**
Kapsüllerin şekilleri	Oval	Oval
Kapsüllerin kılıfının	Elastik	Elastik
	Normal yumuşaklık	Normal yumuşaklık
Rengi	Renksiz	Renksiz
Boyut	Orta	Büyük
Koku	Kokusuz	Kokusuz

Ağırlık Sapması

Her numuneden önce 10'ar tane dolu kapsül ayrı ayrı hassas terazi ile tartıldı, kapsüller boşaltılıp tartım işlemi her boş kapsül için tekrarlandı. Toplam 20 kapsül dolu ve boş olarak tartıldı. Sonuçlar tabloya yazıldı standart sapma değeri hesaplandı (Tablo 2, 3, değerler gram cinsinden verilmiştir).

Tablo 2. A Kapsül Numunesi için Standart Sapma Tablosu

Sıra No	Tanım	Dolu Kapsül	Boş Kapsül	İçerik (yağ) Ağırlığı	İçerik (yağ) Ortalama Ağırlık	İçerik (yağ) Standart Sapma	İçerik (yağ) Standart Sapma %
1	Tartım	0.744	0.268	0.476	0.4554	0.006866	1.44243
2	Tartım	0.747	0.312	0.435	0.4554	0.0068	1.56321
3	Tartım	0.729	0.250	0.479	0.4554	0.007866	1.642171
4	Tartım	0.727	0.263	0.464	0.4554	0.002866	0.617672
5	Tartım	0.748	0.282	0.466	0.4554	0.003533	0.758154
6	Tartım	0.737	0.270	0.467	0.4554	0.003866	0.827837
7	Tartım	0.736	0.278	0.458	0.4554	0.000866	0.189082
8	Tartım	0.681	0.265	0.416	0.4554	0.013133	3.156971
9	Tartım	0.747	0.276	0.471	0.4554	0.0052	1.104033
10	Tartım	0.687	0.265	0.422	0.4554	0.011133	2.638151
					Ortalama Standart Sapma	0.006219	

Tablo 3. B Kapsül Numunesi için Standart Sapma Tablosu

Sıra No	Tanım	Dolu Kapsül	Boş Kapsül	İçerik (yağ) Ağırlığı	İçerik (yağ) Ortalama Ağırlık	İçerik (yağ) Standart Sapma	İçerik (yağ) Standart Sapma %
1	Tartım	1.282	0.328	0.954	0.9373	0.0175052	1.83492
2	Tartım	1.295	0.351	0.944	0.9373	0.0070974	1.177032
3	Tartım	1.272	0.345	0.927	0.9373	0.0111111	1.111111
4	Tartım	1.313	0.376	0.937	0.9373	0.0003218	0.034343
5	Tartım	1.304	0.372	0.932	0.9373	0.0056866	0.610150
6	Tartım	1.305	0.366	0.939	0.9373	0.0018104	0.192800
7	Tartım	1.298	0.347	0.951	0.9373	0.0144058	1.514805
8	Tartım	1.284	0.336	0.948	0.9373	0.0112869	1.190601
9	Tartım	1.278	0.373	0.905	0.9373	0.0356906	3.943712
10	Tartım	1.316	0.380	0.936	0.9373	0.0013888	0.148376
					Ortalama Standart Sapma	0.0106304	

Dağılma Testi

Saf Suda Dağılma Testi: Numune kapsüllerimiz saf suda minimum 16 dk. 02 sn. ile maksimum 18 dk.30 sn. suresi arasında dağıldı.

Suni Mide Vasatında Dağılma Testi: Numune kapsüller minimum 5 dk 14 sn ile maksimum 6 dk 32 sn arasında dağılmıştır.

Suni Bağırsak Vasatında Dağılma Testi: Numune kapsüller minimum 4 dk 49 sn ile maksimum 5 dk 21 sn arasında tamamen dağılmıştır.

DPPH Antioksidan Kapasitesi Ölçümü

Bu çalışmada piyasadan temin edilen Chia tohumu yağı ve yağ kapsüllerinin metanollü ekstrelerinden hazırladığımız çalışma çözeltileri DPPH çözeltisi ile karıştırılıp 517nm'da spektrofofometrede absorbans değerleri ölçüldü. Numunenin çözüldüğü metanol ve DPPH çözeltisi kör olarak kullanılmıştır. Standart olarak

bütillendirilmiş hidroksitolun (BHT)'ye göre karşılaştırma yapıldı. DPPH radikalini giderme aktivitesi standart ile karşılaştırıldığında en yüksek A kapsül numunesinde (% 79,49), en düşük % DPPH giderme aktivitesi D yağ numunesinde (% 41,96) hesaplanmıştır (Tablo 4).

Tablo 4. Chia tohumu yağları ekstreleri ve kapsüllerinin % DPPH Giderme Aktivitesi

Numune	% DPPH giderme aktivitesi (inhibisyon)
A (Kapsül)	79,49±1,15
B (Kapsül)	74,50±4,04
C (Yağ)	48,51±2,15
D (Yağ)	41,96±3,87

Toplam Fenolik Madde Tayini

Piyasadan temin edilen Chia tohumu yağlarının ve kapsüllerinin metanollü ekstrelerindeki toplam fenolik madde miktarına ait veriler Tablo 5'de gösterilmiştir. En yüksek değer D yağ numunesinde 93,3 olarak, en düşük değer ise A kapsül numunesinde 28,67 olarak bulunmuştur.

Tablo 5. Chia tohumu yağları ekstreleri ve kapsüllerinin fenolik madde miktarları

Numune	Toplam fenolik madde miktarı (µg GAE/mg)
A (Kapsül)	27,69±2,11
B (Kapsül)	36,94±1,85
C (Yağ)	76,19±2,01
D (Yağ)	84,26±1,99

TARTIŞMA

Bu çalışmada, Türkiye piyasasında, farklı firmalarca pazarlanan chia tohumu (*Salvia hispanica L*), yağının ve bu yağı içeren yumuşak kapsül preparatlarının güvenli kullanımı açısından kalite kontrol testleri yapılarak etkinlik ve güvenliklerinin belirlenmesi amaçlanmıştır.

Yüksek miktarda omega-3 içeren chia tohumu tüketiminin kolesterol düşürme diyabetle mücadele, kanserle mücadele etkileri bilimsel araştırmalara konu olmuştur (Ayerza, ve Coates W,2007). Doymamış yağ asitlerinin zengin bir kaynağı olan chia tohumunda n-6/n-3 oranı 0,29 olarak bulunmuştur. Düşük n-6/n-3 yağ asidi oranı kardiyovasküler hastalıkların görülme riskinin azalması ile ilişkilendirilmiştir (Biçer ve ark. 2017) Chia tohumlarının alınmasının; serum trigliseridlerini ve düşük yoğunluklu lipoprotein (LDL) değerini önemli ölçüde azalttığı ve yüksek yoğunluklu lipoprotein (HDL) değerini arttırdığı bildirilmiştir. Ayrıca chia tohumunun doygunluk hissini ve bağışıklığı arttırdığı, kalp-damar ve diyabet gibi hastalıkların da riskini azalttığı bildirilmiştir(Ayerza, ve Coates W,2007). Benzer diğer bir çalışmada günlük 25 g chia tohumunu 7 hafta boyunca tüketen kadınlarda çoklu doymamış yağ asit değerlerinin (özellikle ALA ve EPA değerleri) yükseldiği belirlenmiştir (Jin ve ark. 2012). Nopal, chia tohumu, soya proteini ve yulaf içeren 235 kcal'lik içeceklerin 2 ay süresince belirli aralıklarla içilmesiyle vücut ağırlığının, trigliserid ve kan şekeri seviyelerinin azaldığı belirlenmiştir. Tavares ve ark. (2015) günlük 35 g chia ununun kişilerde, toplam kolesterolünde %13 azalma, HDL değerlerinde %25 artış sağladığını, ayrıca kişilerin kilo ve bel bölgelerinde de belirgin bir azalma olduğunu belirlemişlerdir. Azeem ve ark. (2015) chia tohum ekstraktının, pamuk tohumu yağının raf ömrünü arttırdığını ve lipid peroksidasyonunu önemli ölçüde inhibe ettiğini belirlemişlerdir.

Fareler ve insanlar üzerinde yapılan deneylerde, chia tohumunun Tip-2 diyabet hastalığının başlıca şikayetleri olan görme problemleri ve böbrek yetersizliğini azalttığı yönündedir (Chieeo ve ark., 2009). Chia tohumunun içerdiği diyet lifi, antioksidanlar, omega-3 yağ asidi gibi fonksiyonel bileşenlerin etkileri nedeniyle, düzenli tüketildiğinde, vücudu kansere karşı koruduğu ve kanserli bir bireyde toplam tümör ağırlığını azalttığı gözlenmiştir (Espad ve ark. ,2007).

Chia tohumu bazı fenolik bileşikler, tokoferol, karotenoid, vitaminler ve bazı peptitler gibi antioksidan bileşenleri içermektedir. Flavonoidler ve tokoferol tohumun antioksidan kapasitesinden sorumlu temel yapılardır. Chia tohumunun antioksidan kapasitesi 84/g'dır ve bu değer yaban mersininin antioksidan kapasitesine (96/g) yakındır. Antioksidan kapasitenin yüksek olması tohumun uzun süre saklanabilmesini sağlamaktadır. Tokoferoller, chia tohumu (238-427 mg/kg) ve yer fıstığı yağında (398.6 mg/kg) benzer miktarlarda olup chia tohumundaki en önemli antioksidan bileşiklerdir (Pellegrini ve ark. 2003). Chia tohumu aynı zamanda gallik asit, kafeik asit, klorojenik asit, rosmarinik asit, myristin, kuarsetin ve kampferol gibi fenolik bileşikleri de içermektedir (Munoz ve ark., 2013). Antioksidan etki gösterebilen bu fenolik bileşikler hücredeki oksidatif dengenin sağlanmasını destekleyerek kardiyovasküler hastalıklar, diyabet, kanser gibi kronik hastalıklardan koruyucu olabilmektedirler. Kafeik asit, klorojenik asit ve kuarsetin hücredeki yağlar, proteinler ve DNA'nın serbest radikaller ile okside olmasını engelleyerek antioksidan özelliği gösterebilmektedir (Munoz ve ark., 2013; Pellegrini ve ark. 2003).

Chia'nın in vivo olarak antioksidan kapasitesinin değerlendirildiği çalışmalar çok sınırlıdır. Obez ratlar üzerine yapılan bir çalışmada; ratlara 6 ve 12 hafta yüksek yağ ve yüksek fruktoz içeren diyet ile birlikte chia tohumu ve chia yağı verildiğinde chia tohumu ve yağının kanda katalaz (CAT), glutatyon peroksidaz (GPx) aktivitesini, karaciğerde glutatyon reduktaz (GRd) aktivitesini artırdığı bunun sonucunda kanda ve karaciğerdeki antioksidan kapasitenin sırasıyla %35 ve %47 oranında artış gösterdiği belirlenmiştir (da Silva ve ark., 2014).

Son yıllarda destekleyici tedavi seçeneklerinin tıp alanında, giderek artan bir önem kazandığı görülmektedir. Chia tohumu da bu doğal ürünlerin başında gelen ve biyolojik yönden aktif birçok bileşiği içeriğinde bulunduran bir üründür. Literatürde yer alan çalışmalar ışığında Chia tohumu doğal bir ürün olarak umut verici bir tedavi seçeneği olabileceği düşünülmüştür. Ancak standardizasyon ve yeterli farmakokinetik-farmakodinamik çalışmalar olmamasından dolayı kullanımı kısıtlıdır. Bir maddenin ilaç olabilmesi için gerekli üç temel özelliği olan hedefe yönelik olması, etkisinin doza bağlı olması ve etkisinin geçici olması gerekmektedir. Bundan dolayı chia tohumunun daha etkili ve güvenli kullanılabilmesi için, etki mekanizmalarını aydınlatabilecek moleküler düzeyde yeni çalışmaların yapılmasına ihtiyaç vardır. Modern tıbbın antibiyotik direnci, antineoplastik direnci, bozulmuş homeostazisin düzeltilmesi gibi patolojik durumlarda yetersiz kalması hali, yeni ilaç ve tedavi desteklerinin araştırılmasını önemli kılmaktadır.

Sağlık yönünden birçok faydası bilinen chia tohumunun *in vitro* ve *in vivo* klinik biyoaktivitesi ve güvenlik değerlendirmesiyle alakalı sınırlı sayıda çalışma bulunmaktadır. Buna karşın chia tohumunun iyi bir protein, diyet lif ve doymamış yağ asidi kaynağı olduğu yapılan çalışmalarda ortaya konmuştur. Chia tohumu gıdalarda jel oluşturucu, emülgatör ve stabilizatör gibi özelliklere sahip olup, yüksek omega-3 ve omega-6 içeriğiyle, besinlerin fonksiyonellik açısından zenginleştirilmesinde önemli bir kaynak konumundadır. Bu açıdan bakıldığında chia tohumunun ileri çalışmalar ve standardizasyon işlemlerinden sonra önemli bir yardımcı tedavi seçeneği olabileceği düşünülmektedir.

Chia tohumu *(Salvia hispanica L*) yağı içeren piyasa preparatları, ülkemizde Tarım Bakanlığı izni ile piyasada yer almaktadır. Dolayısıyla bu preparatlar, diğer beşeri ilaçlar gibi çeşitli *in vivo* ve *in vitro* kalite kontrol ve güvenlik testlerine tabi tutulmamaktadır. Bu durumun beraberinde getirdiği çeşitli problemler, preparattan beklenen etkinin yeterince gerçekleşmemesine neden olabilir. Yapılan bu çalışma ile piyasada yer alan bu ürünlerde kalite kontrol testleri yapılarak antioksidan aktivitesi saptandı.

Bu çalışmada farklı firmalardan sağlanan chia yağı ve kapsülleri üzerinde farmasötik etkinlik ve kalite kontrol testleri yapılmıştır. Chia'nın tıbbi kullanımını (oral takviyeleri) destekleyen çeşitli epidemiyolojik ve deneysel raporlara rağmen, ekstraksiyon ve etkili dozla ilgili protokoller, sağlam bilimsel verilerle desteklenen insan tüketimine geniş ölçekte uyacak şekilde standartlaştırılmalıdır.

SONUÇ

Diyette chia bulunması bir dizi farmakolojik özellik sağlar, ancak hücre ve memeli modellerinde mekanik yaklaşımlar kullanılarak biyolojik aktivitesinden sorumlu biyoaktif ve yağ asitlerinin bilgisi ve anlaşılması, daha geniş terapötik kullanımı için hala ana sınırlamalardır.

KAYNAKLAR

Ayerza, JR and CoatesW 2007. Effect of dietary α-linolenic fatty acid derived from chia when fed as ground seed, whole seed and oil on lipid content and fatty acid composition of rat plasma. Annals of Nutrition and Metabolism, 51(1): 27-34.

Azeem W, Nadeem M, and Ahmad S 2015. Stabilization of winterized cottonseed oil with chia (Salvia hispanica L.) seed extract at ambient temperature. Journal of Food Science and Technology, 52(11): 7191-7199.

Başaran AA 2008. Nutrasötikler. Turkiye Klinikleri J Med Sci, 28(6 Suppl 1): 146-9.

Biçer BN, Erdal B, Kılınç S, Çapaş M 2017. Chia Tohumu (Salvia hispanica L.). II. Ulusal Beslenme ve Diyetetik Öğrenci Kongresi, Kongre Kitabı, Kayseri, 23 – 26 Mart 2017, s.57.

Chieeo AG, D'Allessandro M.E, Hein GJ, Oliva M.E, Lombardo YB 2009. Dietary ehia seed rieh in alpha- 1 i no leni e aeid improves adiposity and normalise hypertriaeylglyeerolaemia and insulin resistanee in dyslipaemie rats. British Joumal of Nutrition, 101:41 -50

da Silva Marineli R, Moraes ÉA, Lenquiste SA, Godoy AT, Eberlin MN, Maróstica Jr MR 2014. Chemical characterization and antioxidant potential of Chilean chia seeds and oil (Salvia hispanica L.). LWT-Food Sci Technol. 59(2): 1304- 1310.

Espada CE, Berra MA, Martinez MJ, Eynard AR, Pasqualini ME, 2007. Effect of chia oil (Salvia hispanica) rich in ω-3 fatty acids on the eicosanoid release, apoptosis and t lymphocyte tumor infiltration in a murine mammary gland adenocarcinoma. Prostaglandins, leukotrienes and essential fatty acids, 77(1), 21-28.

Erdoğdu, M. & Geçgel, Ü. (2019). Chia Tohumu (Salvia hispanica L.) ve Yağının Fizikokimyasal Özellikleri ve Gıda Sektöründe Değerlendirilmesi. Gıda ve Yem Bilimi Teknolojisi Dergisi, 21: 9-17.

Jin F, Nieman DC, Sha W, Xie, G, Qiu, Y, Jia, W 2012. Supplementation of milledmchia seeds increases plasma ALAand EPAin postmenopausal women. Plant Foods For Human Nutrition, 67(2): 105-110.

Munoz LA, Cobos A, Daz O, Aguliera JM 2013. Chia seed (Salvia hispanica): An ancient grain and a new functional food. Food Rev Int29:394–408.

Tavares TL, Tavares TL, Leite Tavares R, Surama Oliveirada da Silva C. Silva, AS 2015. Chia induces clinically discrete weight loss and improves lipid profile only in altered previous values. Nutricion hospitalaria, 31(3): 1176-1182.

Pellegrini N, Serafini M, Colombi B, Del Rio D, Salvatore S, Bianchi M et al. 2003. Total antioxidant capacity of plant foods, beverages and oils consumed in Italy assessed by three different in vitro assays. J Nutr, 133(9): 2812-2819.

DRAFT

ORAL PRESENTATION

Examining the effectiveness of *tilia vulgaris* leaves in removing the cationic dye basic yellow 28 from aqueous environments

Beyza Moralı[1] (*https://orcid.org/ 0009-0009-7022-840X*)
Türkan Börklü Budak[2*] (*https://orcid.org/0000-0002-1294-2682*)

[1] Yıldız Technical University, Faculty of Art and Science, Department of Chemistry, İstanbul, Turkiye
[2*]Yıldız Technical University, Faculty of Art and Science, Department of Chemistry, İstanbul, Turkiye

*Corresponding author e-mail: turkanborklu@yahoo.com

Abstract

Considering the adverse effects of industrial wastes on the environment and water pollution problems, developing sustainable treatment methods is very important. This study was conducted to reduce the environmental impact of textile industry waste. The potential of *Tilia Vulgaris* tree leaves as an environmentally friendly and economical adsorbent and their potential to provide sustainable solutions for managing textile industry waste was investigated. This study mainly concerns the pollution caused by Basic Yellow 28 textile dye. Aqueous solutions of these dyes were purified by adsorption method using Tilia Vulgaris tree leaves. Within the scope of the research, batch adsorption experiments were carried out on aqueous solutions of shades, and the most suitable parameters were determined. These parameters include the amount of adsorbent, initial concentration, contact time, and ambient temperature. UV-VIS spectrophotometric method was used to determine the parameters. The results showed that the optimum conditions were 1.5 g, 50 mg/L, 60 min, and 30°C.

Keywords: Basic Yellow 28, Adsorption, Tilia Vulgaris Leaves

INTRODUCTION

With the world's population and rapid urbanization, textile products are needed (Hasani et al., 2022; Islam et al., 2021). Synthetic dyes are frequently employed in the dyeing process across various industries, including the textile industry, cosmetics, printing, food, and pharmaceuticals (Hisada et al., 2019). Synthetic dyes raise COD and BOD levels and are highly hazardous, carcinogenic, and non-biodegradable (Fu and Viraraghavan, 2022).

Since dyes cannot biodegrade, physical or chemical treatment techniques, such as adsorption, coagulation, precipitation, filtration, photocatalysis, and chemical oxidation, remove dye-containing effluents (Chaari et al., 2019; Aragaw and Alene, 2022). Compared to other techniques, adsorption has the most promising potential for treating dye effluents since it is simple to use, less expensive, highly effective, and can recycle dyes and regenerate adsorbents. (Janoš et al., 2003; Mutunga et al., 2020; Gad et al., 2023; Wang and Guo, 2020)

Tilia Vulgaris leaves, frequently used as traditional medicinal herbs, provide several health advantages. The flavonoids quercetin and kaempferol found in linden leaves have been associated with anti-inflammatory and anti-swelling activities (Toker et al., 2004). It has been investigated whether linden tree sawdust can act as a heavy metal ion adsorbent (Božić et al., 2009). Therefore, this research aims to examine the suitability and efficiency of linden leaves as an adsorbent for the filtration of synthetic dyes.

Basic yellow 28 dye, which is typically used to dye silk, textiles, paper, food, and cotton, was used in the experiment. This study used BY28 dye-containing aqueous solutions to conduct removal percentage experiments. As a result of the optimization studies, the best percentage removal values were tried to be obtained. The effects of the optimization parameters, such as the amount of adsorbent, the initial concentration of dyestuff, contact time, and temperature, were examined.

DRAFT

MATERIALS AND METHODS

Basic Yellow 28 (BY28) (MW = 433.5 g/mol, $C_{21}H_{27}N_3O_5S$, λ_{max} = 438 nm) is a cationic dye purchased from Sigma-Aldrich for the tests. Batch adsorption tests were conducted in a heated, shaking Julabo SW 22 water bath. During the adsorption studies, samples from the solution medium were obtained, and an Agilent 8453 device was utilized to analyze these samples in a UV-VIS spectrophotometer. The Mikrotest MSD-08 purification system provided the distilled water for the tests. Centrifugation was carried out using an Elektromag M615M centrifuge.

Preparation of Tilia Vulgaris Leaves

The leaf of the Tilia Vulgaris tree served as the adsorbent in this investigation. The adsorbent utilized was gathered from the İstanbul-Türkiye. The adsorbent was washed with tap water and then distilled to eliminate foreign particles. It was then dried in a Memmert UN55 oven at 80°C and ground to a size of 60 to 80 mesh. It was stored in a desiccator to protect it from moisture and other outside elements.

Adsorption Experiments

Using an adsorbent made from a Tilia Vulgaris tree leaf, the batch adsorption method was chosen to examine the cationic dye BY28's capacity for adsorption from aqueous solutions. The BY28 dye was diluted to varied quantities from stock solutions at 1000 ppm (mg/L). Standard keys were transferred into 100mL beakers at the starting concentration and set in a shaking water bath. The impact of several factors, including starting dye concentration (5-50 ppm), adsorbent amount (0.1-1.5 g), contact time (5-180 min), and temperature (25-40°C), were examined throughout these tests. The sample solution was spun at 5000 rpm for 3 minutes following adsorption, filtered through filter paper, and put in a test tube. The absorbance at maximum (λ_{max} = 438 nm) was examined using a UV-VIS spectrophotometer, and the results were noted. The removal percentage (Removal %) and adsorption capacity (q_e) of the dyes BY28 in solution were determined using the following equations:

$$Removal\,(\%) = \frac{C_0 - C_e}{C_0} x100 \qquad (1)$$

$$q_e = \frac{C_0 - C_e}{m} xV \qquad (2)$$

In the equations 1 and 2, m is the amount of adsorbent (g), V is the volume of dye solution (L), and C_0 is the initial dye concentration, C_e is the dye concentration at the end (mg/L).

RESULTS AND DISCUSSION

The effect of adsorbent quantity on BY28 adsorption

One of the most significant variables affecting the adsorption process and the removal % value is the fluctuation in adsorbent amount. Tilia Vulgaris tree leaf was used in the removal studies for the BY28 dye. The remaining variables remained at C_0 = 30 mg/L, V = 50 mL, pH = 6.5, 150 rpm, 25 °C, and 60 min. The amount of adsorbent used in the trials was changed from 0.1 g to 1.5 g. The removal percentage and q_e values of BY28 dye were calculated as shown in Figs. 1(a) and 1(b). Accordingly, it was found that employing 1.5 g of Tilia Vulgaris tree leaf as an adsorbent in the trial, the best removal percentage was attained at 99%.

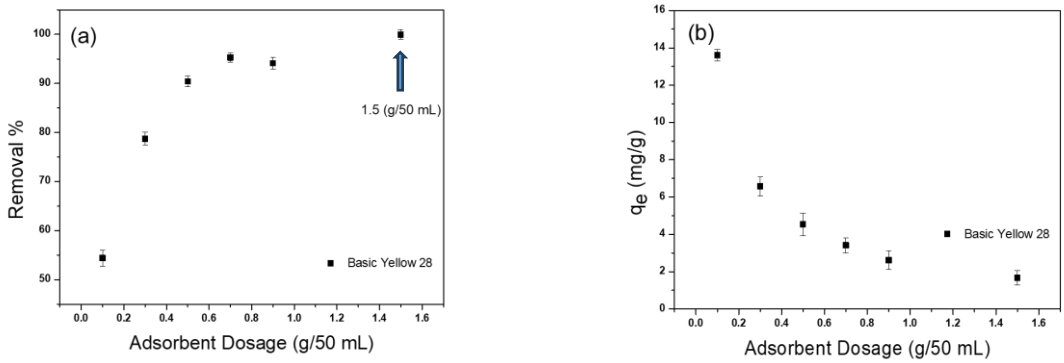
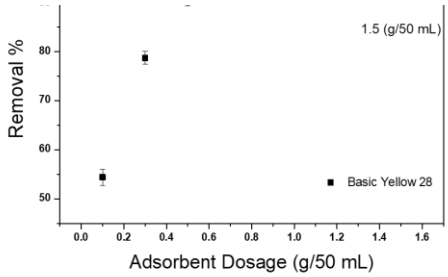


Figure 1. The impact of different adsorbent doses on the BY28 dye removal, Removal % (a), q_e (b).

Figure 1. The impact of different adsorbent doses on the BY28 dye removal, Removal % (a), q_e (b).



BY28 Adsorption and the Influence of Initial Solution Concentration

The initial concentration of the investigated dyestuff at various concentrations ranging from 5 to 50 mg/L is another parameter impacting the removal value. The adsorbent amount was kept at 1.5 g in the studies employing Tilia Vulgaris tree leaf as the adsorbent for BY28, while the other variables were held constant at V= 50 mL, pH 6.5, 150 rpm, 25°C, and 60 minutes. As shown in Figs. 2(a) and 2(b), the best removal was discovered at 94.93% for a 50 mg/L starting concentration.

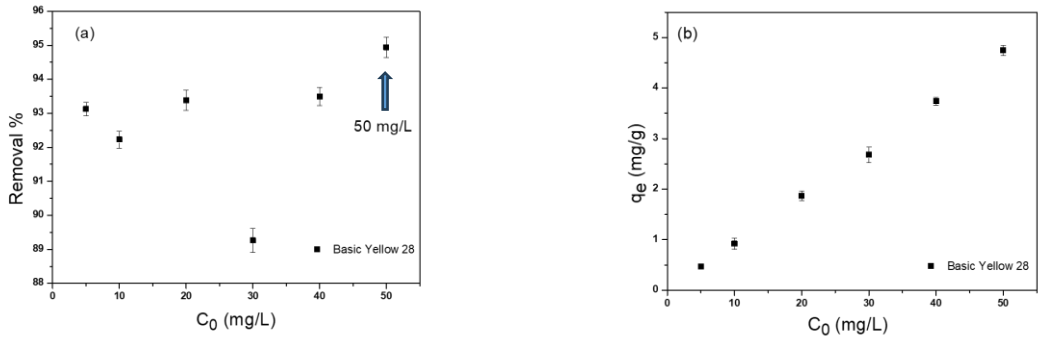
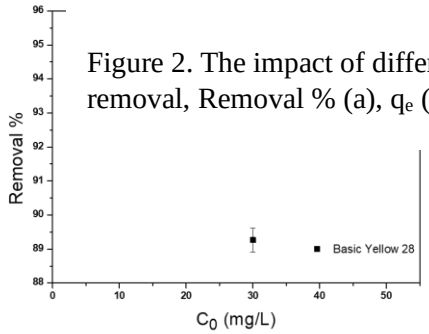


Figure 2. The impact of different initial concentrations on the BY28 dye removal, Removal % (a), q_e (b).

Figure 2. The impact of different initial concentrations on the BY28 dye removal, Removal % (a), q_e (b).



Adsorption of BY28 is affected by contact time.

Contact time is a critical element in determining the efficacy of adsorption. While other factors like V = 50 mL, pH 6.5, 25°C, and 150 rpm were held constant, contact times between 5 and 180 minutes were utilized to examine the effect of contact time on the adsorption process. The findings were reported to be 99.85% at 60 min when using 1.5 g of Tilia Vulgaris tree leaf and an initial concentration of 50 mg/L for BY28 dye, previously established specifications. In Fig. 3, all measured values are displayed.

DRAFT

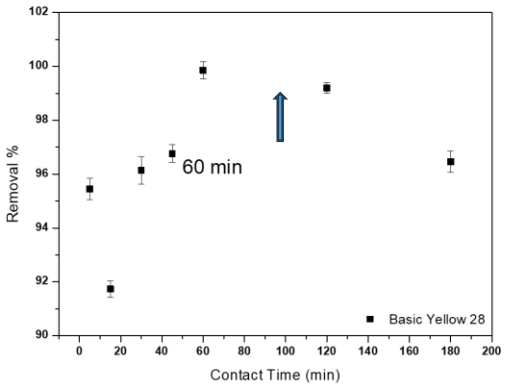


Figure 3. The impact of contact time on the BY28 dye removal %.

The Influen

Figure 3. The impact of contact time on the BY28 dye removal %.

Another elen ...perature. The impact of ambient temp ...atures ranging from 25 to 45°C while maintaining ... ...blished ideal values of 1.5 g adsorbent, 50 mg/L start ...alized when the Tilia Vulgaris tree leaf was utilized ... ...emoval was 94.94%, attained at 30°C.

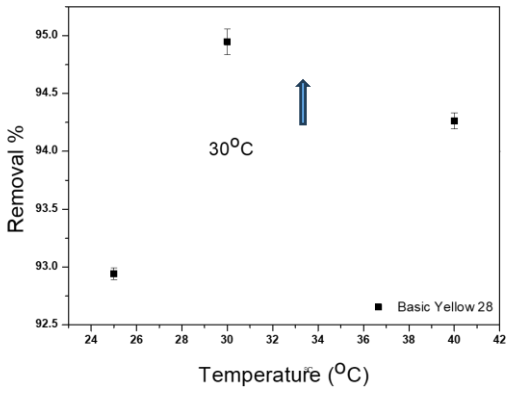
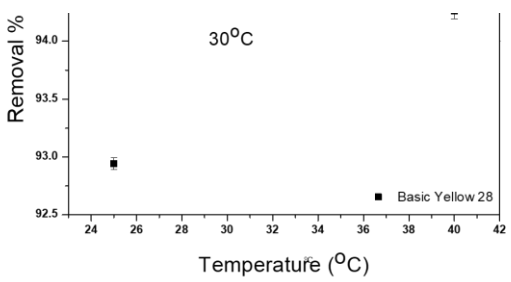


Figure 4. The impact of temperature on the BY28 dye removal %.

CONCLUS

Figure 4. The impact of temperature on the BY28 dye removal %.

This investig ...ployed in literature for therapeutic p ... the harmful dyes BY28 was tested, a ...adsorbent quantity, initial dye solution concentratio ... optimum values of 0.15 g, 50 mg/L, 60 min, and 30 ... Vulgaris tree leaves can remove BY28 hazardous dy ...e utilized to effectively, affordably, and environment



REFERENCES

Aragaw, T. A., & Alene, A. N. (2022). A comparative study of acidic, basic, and reactive dyes adsorption from aqueous solution onto kaolin adsorbent: Effect of operating parameters, isotherms, kinetics, and thermodynamics. *Emerging Contaminants*, *8*, 59–74. https://doi.org/10.1016/j.emcon.2022.01.002

Božić, D., Stanković, V., Gorgievski, M., Bogdanović, G., & Kovačević, R. (2009). Adsorption of heavy metal ions by sawdust of deciduous trees. *Journal of Hazardous Materials*, *171*(1–3), 684–692. https://doi.org/10.1016/j.jhazmat.2009.06.055

Chaari, I., Fakhfakh, E., Medhioub, M., & Jamoussi, F. (2019). Comparative study on adsorption of cationic and anionic dyes by smectite rich natural clays. *Journal of Molecular Structure*, *1179*, 672–677. https://doi.org/10.1016/j.molstruc.2018.11.039

Fu, Y., & Viraraghavan, T. (2002). Removal of Congo Red from an aqueous solution by fungus Aspergillus niger. In *Advances in Environmental Research* (Vol. 7)

Gad, Y. H., Helal, R. H., Radi, H., El-Nemr, K. F., & Khozemy, E. E. (2023). Preparation and application of irradiated polyvinyl alcohol/starch/pumice composites for adsorption of basic dye: Isotherm and kinetics study. *International Journal of Biological Macromolecules*, *249*. https://doi.org/10.1016/j.ijbiomac.2023.126106

Hasani, N., Selimi, T., Mele, A., Thaçi, V., Halili, J., Berisha, A., & Sadiku, M. (2022). Theoretical, Equilibrium, Kinetics and Thermodynamic Investigations of Methylene Blue Adsorption onto Lignite Coal. *Molecules*, *27*(6). https://doi.org/10.3390/molecules27061856

Hisada, M., Tomizawa, Y., & Kawase, Y. (2019). Removal kinetics of cationic azo-dye from aqueous solution by poly-γ-glutamic acid biosorbent: Contributions of adsorption and complexation/precipitation to Basic Orange 2 removal. *Journal of Environmental Chemical Engineering*, *7*(3). https://doi.org/10.1016/j.jece.2019.103157

Islam, A., Teo, S. H., Taufiq-Yap, Y. H., Ng, C. H., Vo, D. V. N., Ibrahim, M. L., Hasan, M. M., Khan, M. A. R., Nur, A. S. M., & Awual, M. R. (2021). Step towards the sustainable toxic dyes and heavy metals removal and recycling from aqueous solution- A comprehensive review. In *Resources, Conservation and Recycling* (Vol. 175). Elsevier B.V. https://doi.org/10.1016/j.resconrec.2021.105849

Janoš, P., Buchtová, H., & Rýznarová, M. (2003). Sorption of dyes from aqueous solutions onto fly ash. *Water Research*, *37*(20), 4938–4944. https://doi.org/10.1016/j.watres.2003.08.011

Mutunga, M. F., Wanyonyi, W. C., & Ongera, G. (2020). Utilization of Macadamia seed husks as a low-cost sorbent for removing cationic dye (basic blue 3 dye) from aqueous solution. *Environmental Chemistry and Ecotoxicology*, *2*, 194–200. https://doi.org/10.1016/j.enceco.2020.09.005

Toker, G., Küpeli, E., Memisoğlu, M., & Yesilada, E. (2004). Flavonoids with antinociceptive and anti-inflammatory activities from the leaves of Tilia argentea (silver linden). *Journal of Ethnopharmacology*, *95*(2–3), 393–397. https://doi.org/10.1016/j.jep.2004.08.008

Wang, J., & Guo, X. (2020). Adsorption isotherm models: Classification, physical meaning, application, and solving method. In *Chemosphere* (Vol. 258). Elsevier Ltd. https://doi.org/10.1016/j.chemosphere.2020.127279

DRAFT

ORAL PRESENTATION

Adsorption of basic blue 3 dye from colored effluents using *mulberry leaves* (Morus *nigra l.*) as a low-cost adsorbent

Adella Myori Hardieka[1] (*https://orcid.org/ 0009-0000-1136-2507*)
Türkan Börklü Budak[2*] (*https://orcid.org/0000-0002-1294-2682*)

[1]Yıldız Technical University, Faculty of Art and Science, Department of Chemistry, İstanbul, Türkiye.
[2*]Yıldız Technical University, Faculty of Art and Science, Department of Chemistry, İstanbul, Turkiye.

*Corresponding author e-mail: turkanborklu@yahoo.com

Abstract

Several industrial streams carry organic dyes that contribute extensively to environmental and water contamination. The adsorption method provides an efficient result and can be a long-term solution for these two primary concerns. In the present work, mulberry leaves (*Morus nigra L.*) were used as a low-cost adsorbent to remove basic blue 3 (BB3) dye from industrial wastewater. Batch adsorption experiments were performed under various conditions. The removal efficiency of the dye and the influence of different parameters, such as the initial concentration of dye, adsorbent mass, contact time, and temperature, were determined using a UV-Vis spectrophotometer. The optimal conditions found in this research were 30 mg/L, 1.5 g/50 mL, 45 min, and 40 °C, with a removal efficiency percentage of about than 99%.

Keywords: Adsorption, Basic Blue 3, Mulberry leaves, Wastewater treatment

Introduction

Water contamination is triggered mainly by industrialization, civilization, agricultural operations, and other environmental variables (Wu et al., 1999). A lot of organic pollutants have been discovered in water resources. Improperly treated wastewater containing toxic organic pollutants is extremely harmful because of the variety of adverse impacts and carcinogenic characteristics (Hanaa et al., 2000; Mehtab et al., 2017).

The textile sector has expanded significantly worldwide, and it has released dyeing wastewater containing hazardous chemical substances (Hasan and Jhung, 2015). Dyes are organic substances with a complicated aromatic molecular structure that may give any material an attractive and vivid color. On the other hand, dyes are more durable and difficult to biodegrade due to their complex aromatic molecular structures (Cardoso et al., 2011).

Currently, there are several ways to treat textile dye wastewater with varying degrees of effectiveness. Some techniques, such as adsorption, advanced oxidation, ion exchange, reverse osmosis, chemical precipitation, adsorption, coagulation, and flocculation, have been used for the removal of dyes from water (Yagub et al., 2014). Among them, the adsorption approach involving low-cost adsorbents obtained from different natural, agricultural, and industrial wastes is the most popular method in wastewater treatment (Khan et al., 2011).

Activated carbon is the most used adsorbent. The decolorization of dye effluent with activated carbon derived from agricultural waste has received a lot of interest (Peláez-Cid et al., 2020). The current study is focused on an alternative, low-cost, and effective adsorbent. Many studies have proven the potential of using adsorbents derived from natural, industrial, and agricultural waste as diverse, low-cost adsorbents. The mulberry leaves act as an effective adsorbent (Ahmad Khan and Farooqui, 2022; Poguberović et al., 2016).

DRAFT

The synthetic dye removal of basic blue 3 (BB3) (surfaced Khan et al., 2013; Taşar et al., 2017) was investigated in this study using adsorption techniques with mulberry leaves (*Morus nigra L.*) as an adsorbent and a variety of parameters, including initial dye concentration, amount of adsorbent, contact time, and temperature. The percentage of dye removal under optimum conditions was also examined.

MATERIALS AND METHODS

In this study, all the chemicals used were of analytical grade and were used without additional purification. Basic blue 3 (BB3) dyes were purchased from Sigma-Aldrich and used as received. The stock solution of 1000 mg/L of primary blue (BB3) dyes ($C_{20}H_{26}CN_3O$; molecular weight = 359.89 g/mol; λ_{max}= 654 nm) was prepared by using distilled water. The working solutions were prepared by diluting appropriate amounts of this stock solution.

Instrumentation

An analytical balance (Radwag), an oven (Memmert UN55), distilled water obtained using a purification device (Mikrotest MSD-08), a UV-Vis Spectrophotometer (Agilent 8453), a shaking water bath (Julabo SW22), an ultrasonic water bath (IsoLab) were used during the experiment.

Preparation of Crushed Mulberry Leaves

Mulberry leaves were collected from İstanbul-Türkiye, thoroughly washed with tap water, and then distilled to remove water-soluble impurities and other foreign particles. The mulberry leaves were dried at 80 °C for 24 hours in the oven. After the leaves are completely dry, they are crushed into a fine powder and then plant powders are sifted through a sieve (mesh size 24.0± 0.4 mm). The mulberry leaf adsorbent was stored in a desiccator.

The Adsorption Process of BB3

To investigate the adsorption of BB3 dyes, 50 mL BB3 with a specific concentration from stock solutions was taken into a 100 ml glass beaker. During these experiments, batch adsorption experiments were conducted under laboratory conditions at room temperature (25°C) on a water bath shaker for a specific time with a different amount of adsorbent. The effect of various parameters such as initial dye concentration (5-50 mg/L), amount of adsorbent (0.3-1.5 g), contact time (5-180 minutes), and temperature (25-40°C), have been investigated. By the end of the adsorption process, the sample solution had been filtered using filter paper (Whatman) and placed in a test tube. The absorbance value at λ_{max} = 654 nm for BB3 was analyzed using a UV-Vis spectrophotometer and record. To calculate the percentage removal (%R) and the adsorption capacity (q_e) of BB3 dye in solution, use the following equations 1 and 2:

$$\%R = \frac{(C_o - C_e)}{C_o} x\ 100 \quad (1)$$

$$q_e = \frac{(C_o - C_e)}{m} x\ v \quad (3)$$

Where m is the amount of adsorbent utilized (g), V stands for dye solution volume (L), and C_0 is the starting dye concentration. C_e is the dye concentration at time t (mg/L).

DRAFT

RESULTS AND DISCUSSION

Effect of Initial Dye Concentration on Adsorption of BB3

To study the influence of initial dye concentration on BB3, experiments were carried out in concentration ranges of 5, 10, 20, 30, 40, and 50 ppm (mg/L). In this experiment, 50 ml of BB3 dye solutions of each concentration were taken into a 100 ml glass beaker. 0.5 g of mulberry leaves as an adsorbent were weighed and added. The mixture was stirred using a water bath shaker at a speed of 150 rpm for 60 minutes under laboratory room temperature conditions. The mixed solution was filtered to separate the solid and liquid phases using filter paper (Whatman). Afterwards, the absorbance of each solution was determined using a UV-Vis spectrophotometer with a maximum wavelength (λ_{max} = 654 nm). The results of the optimum removal efficiency and adsorption capacity values are shown below in Figure 1. It shows that the optimum value for the percentage removal (%R) of BB3 dye is at a concentration of 30 mg/L.

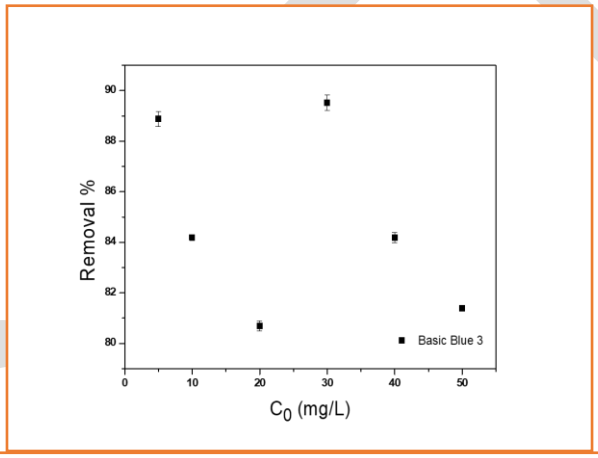


Figure 1. The effects of various adsorbent dosages on the R% of the BB3 dye.

Figure 1. The effects of various adsorbent dosages on the R% of the BB3 dye.

Effect of Adsorbent Amount on Adsorption of BB3

The effect of the amount of adsorbent was determined with a specific amount (0.1, 0.3, 0.7, 0.9, and 1.5 g). The adsorbent was weighed and added to each glass beaker (5). 50 ml of BB3 dye solution at 30 mg/L concentration was put into a 100 ml glass beaker. The solution was shaken at a constant speed of 150 rpm and a temperature of 26.9±0.3°C for 60 minutes. The filter paper was used to filter the solution. It was placed in a test tube. The absorbance of each solution was determined using a UV-Vis spectrophotometer with a maximum wavelength (λ_{max} = 654 nm). The results of the optimum removal efficiency and adsorption capacity values are shown below in Figures 2 (a) and (b). It is shown that the optimum values for the percentage removal (%R) of BB3 dye are found when the adsorbent dosage is 1.5 g per 50 ml. The percentage of removal increases with increasing mulberry leaf adsorbent dosage.

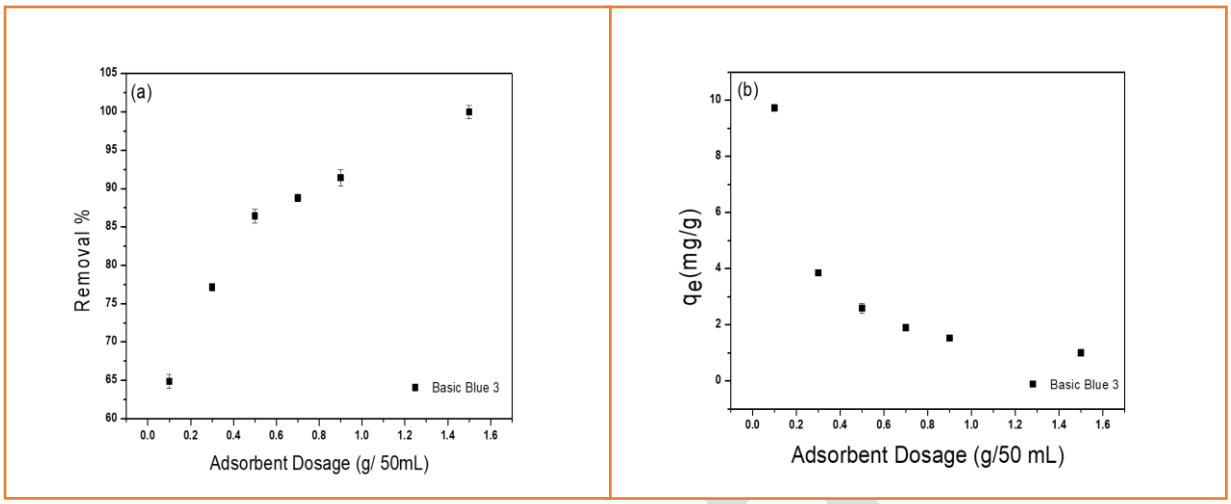


Figure 2. The effects of various adsorbent dosages on the Removal % (a), qe (b) of the BB3 dye.

Figure 2. The effects of various adsorbent dosages on the Removal % (a), qe (b) of the BB3 dye.

Effect of Contact

To determine the ...en and put into a 100 ml glass b... ...nd added. The solution mixture was shaken at 150 rpm at 25.7±0.2 °C within 5–180 minutes. The mixed solution was filtered using filter paper. Then, the absorbance of each solution was determined using a UV-Vis spectrophotometer with a maximum wavelength (λ_{max} = 654 nm). In a contact time experiment, the results of the optimum values of the removal efficiency are seen in Figure 3. This graph shows that the percentage removal (%R) of BB3 dyes is optimal for a contact time of 45 min.

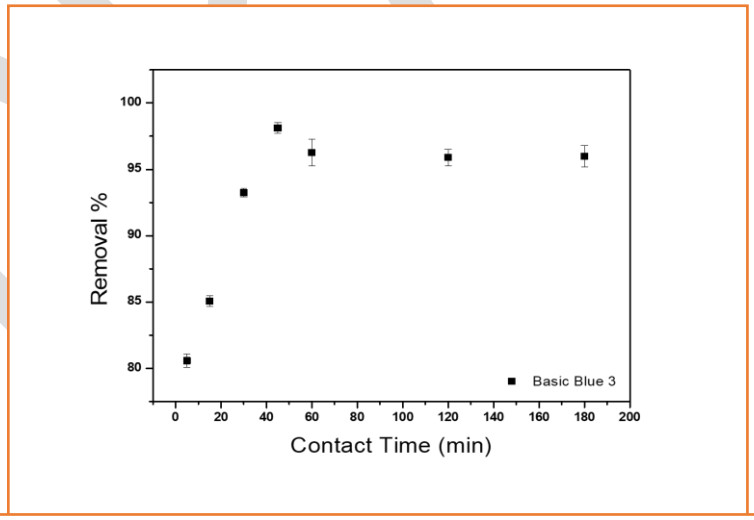
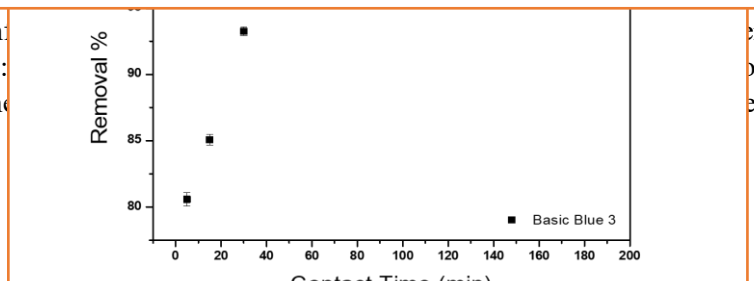


Figure 3. The effects of contact time on the Removal % BB3 dye.

Figure 3. The effects of contact time on the Removal % BB3 dye.

Effect of Tempera

To investigate the in... ...ere carried out under the following conditions: ...or BB3), stirring speed at 150 rpm, contact time ... ...e of 1.5 gram at different



temperatures ranging from 25 to 40°C. The mixed solution was filtered using filter paper. The absorbance of each solution was measured using a UV-Vis spectrophotometer with a maximum wavelength (λ_{max} = 654 nm). The results of the optimum values of the removal efficiency are illustrated in Figure 4. It indicates that the percentage removal (%R) of BB3 dye is optimal at temperatures of 40±0.2°C.

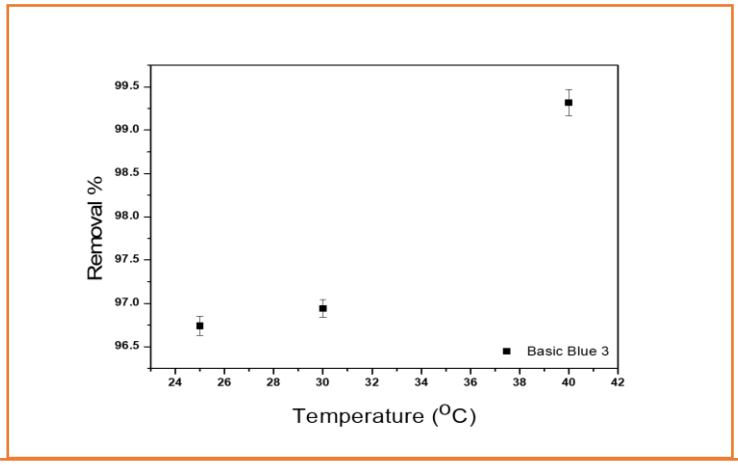


Figure 4. The effects of temperature on the Removal % BB3 dye.

CONCLUSION

Based on the present and previous research results, the adsorption method using mulberry leaves as an adsorbent has successfully been demonstrated to remove BB3 dye solution. The percentage of dye removal efficiency reached more than 99% under optimal conditions. According to the experiments that have been carried out, the optimum values of various parameters can be obtained as follows: 30 mg/L, 1.5 g/50 mL, 45 min, and 40°C for basic blue 3 (BB3). According to the data, C leaves can be successfully used as a natural, cheap, and environmentally friendly adsorbent to remove toxic BB dye.

References:

Ahmad KF, Farooqui M 2022. Removal of methylene blue dye from aqueous solutions onto Morus nigra L. (mulberry tree) leaves powder and its biochar equilibrium, kinetic and thermodynamic study. International Journal of Environmental Analytical Chemistry. https://doi.org/10.1080/03067319.2022.2103691

Cardoso NF, Pinto RB, Lima EC, Calvete T, Amavisca CV, Royer B, Cunha ML, Fernandes THM, Pinto IS 2011. Removal of remazol black B textile dye from aqueous solution by adsorption. Desalination, 269(1–3) : 92–103. https://doi.org/10.1016/j.desal.2010.10.047

Hanaa MS, Eweida A, Farag A 2000. Heavy metals in drinking water and their environmental impact on human health. ICEHM2000: 542-556.

Hasan Z, Jhung SH 2015. Removal of hazardous organics from water using metal-organic frameworks (MOFs): Plausible mechanisms for selective adsorptions. Journal of Hazardous Materials, 283: 329–339. https://doi.org/10.1016/j.jhazmat.2014.09.046

Khan TA, Sharma S, Ali I 2011. Adsorption of Rhodamine B dye from aqueous solution onto acid activated mango (Magnifera indica) leaf powder: Equilibrium, kinetic and thermodynamic studies. Journal of

Toxicology and Environmental Health Sciences, 3(10): 286–297. http://www.academicjournals.org/JTEHS

Mehtab H, Muhammad FM, Asma J, Sidra A, Nayab A, Sharon Z, Jaweria H 2017. Water pollution and human health. Environ Risk Assess Remediat. 1(3): 16-19. http://www.alliedacademies.org/environmental-risk-assessment-and-remediation/

Peláez-Cid A A, Romero-Hernández V, Herrera-González AM, Bautista-Hernández A, Coreño-Alonso O 2020. Synthesis of activated carbons from black sapote seeds, characterization, and application in the elimination of heavy metals and textile dyes. Chinese Journal of Chemical Engineering. *28*(2): 613–623. https://doi.org/10.1016/j.cjche.2019.04.021

Poguberović SS, Krčmar DM, Maletić SP, Kónya Z, Pilipović DDT, Kerkez DV, Rončević SD 2016. Removal of As(III) and Cr(VI) from aqueous solutions using "green" zero-valent iron nanoparticles produced by oak, mulberry, and cherry leaf extracts. Ecological Engineering. 90: 42–49. https://doi.org/10.1016/j.ecoleng.2016.01.083

sufaid Khan M, Ahmad A, Sikandar SS, Sufaid KM, Ullah KBF, Sikindar SS, Khan P 2013. Removal of Basic Dye from Aqueous Solutions Using Nano-Scale Zero Valent Iron (NZVI) as Adsorbent Synthesis of Organic Functional Materials as cathode part for Lithium-Ion Batteries View project Carbon-based Flexible energy storage electrodes for enhancing the performance of Lithium-ion batteries View project 744 Removal of Basic Dye from Aqueous Solutions Using Nano-Scale Zero Valent Iron (NZVI) as Adsorbent. J.Chem.Soc. Pak. 35(3). https://www.researchgate.net/publication/287482277

Taşar Ş, Kaya F, Özer A 2017. Adsorption of CI Basic Blue 3 Dye Molecules from Aqueous Media by Sulfuric Acid-Activated Montmorillonite Mineral. JOTCSB. 1(Sp. is. 1):1–16.

Wu C, Maurer C, Wang Y, Xue S, Davis DL 1999. Water Pollution and Human Health in China. Environmental Health Perspectives. 4 (107): 251-256.

Yagub MT, Sen TK, Afroze S, Ang HM 2014. Dye and its removal from aqueous solution by adsorption: A review. In Advances in Colloid and Interface Science. 209: 172–184). https://doi.org/10.1016/j.cis.2014.04.002

DRAFT

ORAL PRESENTATION

Cytotoxic activity of the root ethyl acetate extract of *Heptaptera cilicica* (Boiss. & Balansa) Tutin on renal cancer cell lines

Feyyaz Mıhoğlugil[1*] (*ORCID: https://orcid.org/0000-0003-0976-0244*)

[*1]Bahçeşehir Cyprus University, Faculty of Pharmacy, Nicosia, Northern Cyprus.

*Corresponding author e-mail: feyyaz.mihoglugil@baucyprus.edu.tr

Abstract

Heptaptera cilicica is one of the species of the genus *Heptaptera*. The root ethyl acetate extract of *H. cilicica* were fractionated column chromatography and twelve sesquiterpene coumarins; umbelliprenin (1), colladin (2), psoralen (3), bergapten (4), badrakemone (5), colladonin (6), badrakemin (7), marmesin (8), scopoletin (9), umbelliferon (10), 14'-acetoxybadrakemone (11) and 14'-acetoxybadrakemin (12) was isolated. Extract and isolated coumarins were performed against A498 and OU31 renal cancer cell lines. The root ethyl acetate extract of *H. cilicica* and colladin, colladonin, badrakemin, 14′-acetoxybadrakemin showed strong cytotoxic activity against UO31 renal cancer cell line.

Keywords: *Heptaptera cilicica*, cytotoxic activity, coumarin.

INTRODUCTION

The genus *Heptaptera* Marg. & Reuter (Apiaceae) is represented by 11 species worldwide, four of them; *H. cilicica* (Boiss. & Balansa) Tutin, *H. anisoptera* (DC.) Tutin, *H. anatolica* (Boiss.) Tutin and *H. triquetra* (Vent.) Tutin are growing in Turkey (The WHO Plant Lists, 2023, Herrnstadt et al.,1972).

H. cilicica is an endemic species and distributed in the eastern Taurus Mountains in Mersin province of the Mediterranean region of Turkey (Herrnstadt et al.,1972). *Heptaptera* species have been reported to contain sesquiterpene coumarin derivatives (Ban'kovskii et al.,1970, Borisov et al., 1975, Appendino et al., 1992, Appendino et al., 1993, Tosun et al., 2019, Tosun et al., 2021) to which various biological activities such as cytotoxicity, P-glycoprotein inhibitory, cancer chemopreventive, anti-inflammatory, antibacterial, antileishmanial, antiviral, antidiabetic and cholinesterase inhibitory are attributed (Tosun et al., 2021, Nazari et al., 2011, Gliszczynska and Brodelius, 2012, Dastan et al., 2016, Amin et al., 2016, Iranshahi et al., 2018, Erdogan Orhan et al., 2021).

In the present study, initially, the ethyl acetate extract of the roots of *H. cilicica* was tested on renal cancer cell lines and confirmed cytotoxic activity. Afterward, the extract was fractionated and twelve known coumarins were successfully isolated. Finally, isolated twelve sesquiterpene coumarins were performed on renal cancer cell lines (A498 and UO31).

MATERIALS AND METHODS

General experiments

NMR spectra were acquired on a Bruker Avance III spectrometer (Rheinstetten, Germany) operating at 500 MHz for 1H and equipped with a 5 mm indirect observation probe. Initial purification of the ethyl acetate extract was carried out on a Sephadex LH-20 (GE Healthcare, Chicago, IL, USA) column. Further purification of column fractions was performed using silica gel F254 PLC plates (1 mm thickness) (Merck KGaA, Darmstadt, Germany) or combination of PLC and silica gel column chromatography (Silica gel 60, 0.063-0.2 mm particle size, Merck KGaA, Darmstadt, Germany).

DRAFT

Plant material

The roots of *H. cilicica* were collected in the vicinity of Mersin-Tarsus in July 2013 and identified by Prof. A. Duran. A voucher specimen (A. Duran 9591) was deposited in the Herbarium of Selçuk University, Faculty of Sciences, Department of Biology (KONYA).

Extraction

Air-dried and coarsely powdered roots (100 g) of *H. cilicica* was extracted sequentially with dichloromethane (2 L) and methanol (3 L) at room temperature and concentrated, in vacuo, to dryness (yields; 4.99 g dichloromethane extract and 14.01 g methanol extract). The methanol extract was redissolved in a mixture of methanol/water (10:90) and then partitioned with ethyl acetate (EtOAc), the resulting extracts were separately concentrated in vacuo to dryness. Ethyl acetate and aqueous methanol extracts of the roots were 0.88 g and 12,95 g respectively.

Chromatography

Prepared EtOAc extract was separated on Sephadex LH-20 column using n-hexane:dichloromethane:methanol solvent mixture and fractions were collected. The fractions which had the same spots on TLC were combined. Combined fractions were separated, using silica gel with n-hexane:EtOAc to obtain subfractions. TLC was performed using silica gel 60 F254 plates. Subfractions were separately further purified by preparative TLC using n-hexane:ethyl acetate (90:10; 70:30; 50:50) as solvent system. Twelve coumarins were isolated from subfractions.

Cytotoxic assay on renal cancer cell lines

The assay used was a two-day, two cell line XTT bioassay (Skehan et al., 1990), an in vitro antitumor colorimetric assay developed by the MTP Assay Development and Screening Section. The renal cancer cell lines used were UO31 and A498. Cells were harvested and plated (45 µL) at a seeding density of 3.0 × 105 cells per well for the UO31 cell line, 2.5 × 105 cells per well for the A498 cell line. The respective cell lines were separately plated into 384-well assay plates and then incubated for 24 h. DMSO solutions of the test materials (8 µL) were diluted 1:25 with medium (192 µL) and then subjected to five 2:1 serial dilutions (100 µL each) on a 96-well plate. Duplicate 40 µL aliquots of each sample concentration were then transferred to a 384- well "dilution plate", which could accommodate the duplicate samples from two 96-well plates. A 5 µL portion of each solution in the dilution plate was transferred to the cell cultures in the 384-well assay plates to give a final volume of 50 µL and a DMSO concentration of 0.4%. Control wells included 8 wells with the positive control sanguinarine chloride at 20 µM, as well as DMSO only controls and no cell controls. The Z' factors for the individual plates were calculated and were >0.5 in all cases. The cells were incubated for 48 h at 37 °C in the presence of the test samples and then treated with the tetrazolium salt XTT (2,3-bis[2-methoxy-4-nitro-5-sulfophenyl]-2H-tetrazolium-5-carboxanilide). Viable cells reduced XTT to a colored formazan product, and after an additional 4 h incubation period the amount of formazan produced was quantified by absorption at 450 nm, using a 650 nm reference. Plates were read on a PerkinElmer EnVision (model # 2104) reader.

RESULTS and DISCUSSION

Twelve known sesquiterpene coumarins were obtained as a result of fractionation of the ethyl acetate extract of the roots of *H. cilicica*. Umbelliprenin (1), colladin (2), psoralen (3), bergapten (4), badrakemone (5), colladonin (6), badrakemin (7), marmesin (8), scopoletin (9), umbelliferon (10). 14'-acetoxybadrakemone (11), 14'-acetoxybadrakemin (12).

^{1}H NMR data of the known compounds agree with the data given in the literature for umbelliprenin (1) (Tian et al. 2013), colladin (2) (Ban'kovskii et al., 1970), psoralen (3), bergapten (4) (Tosun et al., 2020), badrakemone (5) (Appendino et al. 1992a), colladonin (6) (Perel'son, M et al., 1975), badrakemin (7) (Asghari et al. 2016), marmesin (8) (Abu-Mustafa and Fayez, 1961), scopoletin (9) (Tosun et al., 2020), umbelliferon (10) (Singh et al., 2010), 14'-acetoxybadrakemone (11) (Tosun et al., 2021), 14'-acetoxybadrakemin (12) (Tosun et., 2019).

The ethyl acetate extract of the roots of *H. cilicica* showed high cytotoxic activity against UO31 renal cancer cell line with IC50 value 0.08 µg/mL, in contrast extract didn't show cytotoxic activity on A498 renal cell line up to 50 µg/mL concentrations (Table 1.).

Umbelliprenin (1), colladin (2) and especially 14'-acetoxybadrakemin (12) showed strong cytotoxic activity against UO31 renal cell line. Colladonin (6) and badrakemin (7) showed strong cytotoxic activity against UO31 and mild cytotoxic activity against A498 renal cell lines. Other sesquiterpene coumarins which isolated from root ethyl acetate extract of *H. cilicica* didn't show cytotoxic activity (Table 1.).

Table 1. IC50 values of EtOAc extract and isolated sesquiterpene coumarins against renal cell lines.

	A498 (µg/mL)	UO31 (µg/mL)
EtOAc extract	>50	0.08
Umbelliprenin	>50	1.8
Colladin	>50	0.39
Psoralen	>50	>50
Bergapten	>50	>50
Badrakemone	>50	11
Colladonin	21	0.75
Badrakemin	20	0.38
Marmesin	>50	>50
Scopoletin	>50	>50
Umbelliferon	>50	>50
14'-acetoxybadrakemone	>50	>50
14'-acetoxybadrakemin	>50	0.017

CONCLUSION

Heptaptera species have been reported to contain sesquiterpene coumarins derivatives. Furthermore, these type of coumarins which isolated from *Heptaptera* species are reported as cytotoxic activity (Tosun, 2019-2020). Previously, six of these sesquiterpene coumarins (umbelliprenin, colladin, colladonin, badrakemin, 14'-acetoxybadrakemone, 14'-acetoxybadrakemin) were reported dichloromethane extract of the roots of *H. cilicica*. Tosun et al., reported that dichloromethane extract and colladin showed strong cytotoxic activity on colon cancer cell lines. (Tosun et al., 2021). In the present study, ethyl acetate extract of the roots of *H. cilicica* showed strong cytotoxic activity on UO31 renal cancer cell line. In conclusion, the ethyl acetate extract of the roots of *H. cilicica* has strong cytotoxic activity against UO31 renal cancer cell line and colladin, colladonin, badrakemin and especially 14'-acetoxybadrakemin may be responsible for the cytotoxic activity.

ACKNOWLEDGEMENTS

I would like to thank Dr. John A. Beutler, Molecular Targets Laboratory, CCR, NCI, Frederick, MD, U.S.A., for the cytotoxic activity testing and Prof. A. Duran for the collection and identification of plant material.

REFERENCES

Abu-Mustafa, E. A., & Fayez, M. B. E. (1961). Natural Coumarins. I. Marmesin and Marmesinin, Further Products from the Fruits of Ammi majus L. The Journal of Organic Chemistry, 26(1), 161-166.

Amin A, Tuenter E, Cos P, Maes L, Exarchou V, Apers S, Pieters L. Antiprotozoal and antiglycation activities of sesquiterpene coumarins from *Ferula narthex* exudate. *Molecules* 2016**;** 21: 1287-1296.

Appendino G, Özen HÇ, Jakupovic J. A sesquiterpene coumarin ether and a coniferyl ester from *Heptaptera anisoptera*. *Fitoterapia* 1993; 64: 505-506.

Appendino G, Özen HÇ, Nano GA, Cisero M. Sesquiterpene coumarin ethers from the genus *Heptaptera*. *Phytochemistry* 1992; 31: 4223-4226.

Appendino G, Özen HÇ, Tagliapietra S, Cisero M (1992a) Coumarins from Heptaptera anisoptera. Phytochemistry 31(9):3211–3213.

Appendino G, Özen HÇ, Tagliapietra S, Cisero M. Coumarins from *Heptaptera anisoptera*. *Phytochemistry* 1992; 31: 3211-3213.

Asghari J, Atabaki V, Baher E, Mazaheritehrani M (2016) Identification of sesquiterpene coumarins of oleo-gum resin of Ferula assa-foetida L. from the Yasuj region. Nat Prod Res 30(3):350–353.

Ban'kovskii, A. I., Ermatov, N. E., Perel'son, M. E., Bubeva-Ivanova, L., & Pavlova, N. S. (1970). Structure of the coumarins colladin and colladonin. II. Chemistry of Natural Compounds, 6, 170-176.

Ban'kovskii AI, Ermatov NE, Parel'son ME, Bubeva-Ivanova L, Pavlova NSt. Structure of the coumarins colladin and colladonin. *Khim. Prir. Soedin.* 1970; 6 (2): 173-180.

Borisov VN, Ban'kovskii AI, Pavlova NS, Bubeva-Ilanova, Sheichenko VI, Kabanov VS. Khim. Prir. Soedin. 1975; 247.

Dastan D, Salehi P, Aliahmadi A, Gohari AR, Maroofi H, Ardalan A. New coumarin derivatives from *Ferula pseudalliacea* with antibacterial activity. *Nat. Prod. Res***.** 2016**;** 30 (24): 2747-2753.

Erdogan Orhan İ, Tosun F, Senol Deniz F S, Eren G, Mıhoğlugil F, Akalgan D, Miski M. Butyrylcholinesterase-inhibiting natural coumarin molecules as potential leads. *Phytochemistry Letters* 2021; 44: 48-54.

Gliszczynska A, Brodelius PE. Sesquiterpene coumarins. *Phytochem. Rev.* 2012; 11: 77–96

Herrnstadt I, Heyn CC. Heptaptera Marg. & Reuter. In *Flora of Turkey and the East Aegean Islands*, Davis, P., Ed.; Edinburgh University Press, Edinburgh, 1972; Vol. 4: pp 388-390.

Iranshahi M, Rezaee R, Najafi MN, Haghbin A, Kasaian J. Cytotoxic activity of the genus *Ferula* (Apiaceae) and its bioactive constituents. *Avicenna J Phytomed*. 2018; 8(4): 296-312.

Nazari ZE, Iranshahi M. Biologically active sesquiterpene coumarins from *Ferula* species. *Phytother. Res*. 2011; 25: 315-323.

Perel'son, M. E., Kir'yalov, N. P., & Ban'kovskii, A. I. (1975). The question of the configurations of farnesiferol a, gummosin, badrakemin, and colladonin. Chemistry of Natural Compounds, 11(2), 246-247.

Singh, R., Singh, B., Singh, S., Kumar, N., Kumar, S., & Arora, S. (2010). Umbelliferone–An antioxidant isolated from Acacia nilotica (L.) Willd. ex. Del. Food Chemistry, 120(3), 825-830.

Skehan, P., Storeng, R., Scudiero, D., Monks, A., McMahon, J., Vistica, D., ... & Boyd, M. R. (1990). New colorimetric cytotoxicity assay for anticancer-drug screening. JNCI: Journal of the National Cancer Institute, 82(13), 1107-1112.

The WFO Plant List, A Working List of All Plant Species, Accessed September 6, 2023 at https://wfoplantlist.org/plant-list/taxon/wfo-4000017518-2023-06?page=1

Tian YO, Zhang ZX, Xu HH (2013) Laboratory and field evaluations on insecticidal activity of Cicuta virosa L. var. latisecta Celak. Ind Crop Prod 41:90–9.

Tosun F**,** Aytar EC, Beutler JA, Wilson JA, Miski M. Cytotoxic Sesquiterpene Coumarins from the Roots of *Heptaptera cilicica*. *Rec. Nat. Prod.* 2021; 15(6): 529-536.

Tosun F, Beutler JA, Ransom TT, Miski M. Anatolicin, a Highly Potent and Selective Cytotoxic Sesquiterpene Coumarin from the Root Extract of *Heptaptera anatolica*. *Molecules* 2019; 24: 1153-1160.

Tosun, F., Mıhoğlugil, F., Beutler, J. A., Eroğlu Özkan, E., & Miski, M. (2020). Neopapillarine, an unusual coumarino-alkaloid from the root extract of neocryptodiscus papillaris with cytotoxic activity on renal cancer cells. Molecules, 25(13), 3040.

DRAFT

ORAL PRESENTATION

Rüzgâr atlası analizi ve uygulama programı yardımıyla Sivas ili rüzgâr enerjisi potansiyelinin analizi

Ünal Yılmaz[1*] (*ORCID: https://orcid.org/0000-0003-0058-6323*),
Figen Balo[2] (*ORCID: https://orcid.org/0000-0001-5886-730X)*

[*1]Asri Mühendislik, Malatya, Türkiye
[2] Fırat Üniversitesi, Mühendislik Fakültesi, Met. Ve Malzeme Mühendisliği, Elazığ, Türkiye

*Sorumlu yazar e-mail:unalylmz4444@gmail.com

Özet

Fosil kaynakların daha az kullanılması ve çevreye salınan emisyonların dolayısıyla çevre kirliliğinin azaltılması amacıyla yenilenebilir enerji kaynaklarının değerlendirilmesi çevre ve insan sağlığının yanında ekonomik olarak ta önemlidir. Bir bölgenin mevcut yenilenebilir enerji potansiyelinin araştırılarak o bölge için yapılan fizibilite çalışması hem ekonomik açıdan hem de fosil kaynaklı olmayan bir enerji kaynağının kullanımıyla sağlanacak çevresel korumanın birçok avantajı için ilk adımdır.
Bu çalışmada, Danimarka Riso Ulusal Laboratuvarı olarak bilinen Danimarka Teknik Üniversitesi Rüzgâr Enerjisi Departmanı tarafından Weibull parametre tahmininden faydalanılarak geliştirilen WAsP Rüzgâr Atlası Analiz ve Uygulama Programı kullanılmıştır. Bu program vasıtasıyla Sivas ilinin 1 yıllık saatlik meteorolojik verilerinden faydalanılarak kurulması tasarlanan rüzgâr çiftliğinden elde edilebilecek rüzgâr enerjisi potansiyeli araştırılmıştır. Yapılan araştırmada bölgede hedeflenen enerji ihtiyacının karşılanması için 28 adet Vestas V52'ye (850 kW) türbin ile tasarlanan bir rüzgâr çiftliğinden elde edilebileceği belirlenmiştir.

Anahtar Kelimeler: Rüzgâr Enerjisi, WAsP, Yenilenebilir enerji, Sürdürülebilirlik, Rüzgâr Atlası

Analysis of the wind energy potential of Sivas province with the help of wind atlas analysis and application program

Abstract

Utilizing renewable energy resources in order to reduce the use of fossil resources and reduce emissions to the environment and therefore environmental pollution is important not only for the environment and human health, but also economically. A feasibility study conducted by investigating the existing renewable energy potential of a region is the first step towards many advantages of both economic and environmental protection that will be provided by the use of a non-fossil energy source.
In this study, the WAsP Wind Atlas Analysis and Application Program, developed by the Technical University of Denmark Wind Energy Department, known as the Danish Riso National Laboratory, using Weibull parameter estimation, was used. Through this program, the wind energy potential that can be obtained from the wind farm designed to be established by using the 1-year hourly meteorological data of Sivas province was investigated. In the research, it was determined that meeting the targeted energy need in the region could be obtained from a wind farm designed with 28 Vestas V52 (850 kW) turbines.
Keywords: Wind Energy, WAsP, Renewable energy, Sustainability, Wind Atlas

GİRİŞ

Enerji, bir ülkenin kalkınması bakımından ana unsurdur. Çevresel sürdürülebilirlik, modern çağın esas hedeflerinden biridir. Türkiye'nin enerji üretimi için kullandığı enerji kaynaklarının toprak, su ve hava gibi hayati elementler üzerinde önemli kirletici etkilere sebep olması, bu enerji kaynaklarının en kısa sürede kullanımını azaltmanın gerekliliğinin bir göstergesidir. Atmosferi ve olaylarını araştıran bilim olan meteoroloji, bu nedenle çevresel sürdürülebilirlik alanında kilit bir faktördür. Enerji üretimi ise çevreye salınan CO_2 emisyonlarının en büyük kaynaklarından biridir. Rüzgâr gibi yenilenebilir kaynaklardan enerji üretimi sırasında herhangi bir karbondioksit veya diğer hava kirleticileri üretilmemektedir(Abbasi ve Abbasi, 2010). Rüzgâr, incelenmesi çok karmaşık olan bir elementtir ve karakterize edilmesi, hızı, yoğunluğu, yönü, zeminin

pürüzlülüğünün etkisi, engellerin etkisi, rüzgârın stabilitesinin etkisi vb gibi çeşitli parametrelerin bir fonksiyonudur (IEA Statistics, 2021).
Rüzgâr çiftlikleri aracılığıyla yaygın olarak mekanik, elektrik ve termal olmak üzere üç tür enerji elde edilebilmektedir. Bu nedenle, rüzgâr çiftlikleri Dünya'da birçok ülkenin yayla bölgelerinde giderek daha fazla yer almaya başlamıştır (Barrios, ve Rodríguez, 2004). Rüzgar enerjisi sıfır emisyon eşliğinde bir elektrik akımı sağlayabilmektedir. Rüzgâr türbinleri bir rüzgâr çiftliğinin ana elemanlarından biridir. Son yıllarda sürekli desteklenen teknik gelişmelerle çıktısını doğrudan bir şebekeye sağlayan büyük rüzgâr türbinleri, elektrik santrallerindeki üretim tesisi ile rekabet edebilecek niteliklere sahiptir. Rüzgâr türbinlerinin kuşlarla veya habitatlarla olası etkileşimi ve özel korumaya veya diğer sosyal veya çevresel sınırlamalara ihtiyaç duyan toplulukların sağlığına ilişkin olası tehlikelerine rağmen, hızla fosil yakıtlarla rekabetçi bir enerji kaynağı olarak dikkat çekmektedir (Walker ve ark., 2015). Bir rüzgâr çiftliği tasarlanırken ekonomi için yeterli bir yıllık çıktı sağlamak esastır. Bu da, son derece rüzgârlı bir alan gerektirir. Bu nedenle, bir rüzgâr hızı araştırması, herhangi bir ülke veya bölgedeki büyük ölçekli rüzgâr enerjisi potansiyellerini araştırmanın ilk ve en belirgin adımıdır.
Türkiye'de rüzgâr enerjisinin potansiyelinin belirlenmesini içeren araştırmalar yapılmaktadır.
Bilir ve ark. Türkiye'nin başkenti Ankara'nın İncek ilçesi için rüzgâr enerjisi potansiyelini belirlemek amacıyla gerekli rüzgâr hızı ve yönü bilgilerini kullanmıştır. Yaptıkları analizlerin değerlendirilmesiyle araştırılan ilçenin büyük çaplı rüzgâr türbinlerinin kurulumu için uygun bir bölge olmadığını bununla birlikte küçük çaplı rüzgâr türbinlerini kurabilmek için değerlendirilebilir bir bölge olduğunu rapor etmişlerdir (Bilir ve ark., 2015). Ersöz ve Onat, rüzgâr enerjisi potansiyeli ve rüzgâr karakteristiklerinin analizini yapmak için yapay sinir ağları yöntemini kullanmışlardır (Onat ve Ersoz, 2011). Gökçek ve ark. Kırklareli şehrinin rüzgâr enerjisi potansiyelini ve rüzgâr karakteristiklerini araştırmışlardır. Rüzgâr hızı ve yönü bilgisi olarak, 2004 yılına ait saatlik verilerle hesaplamalarını yapmışlardır. Senelik ortalama güç yoğunluk değeri 139 W/m2 olarak rapor edilmiştir (Gökçek ve ark., 2007).
Bu araştırma çalışmasının temel amacı, aletli meteorolojik hava istasyonundan alınan yerel hava durumu verilerini kullanarak Sivas şehrinin bulunduğu alanda mevcut rüzgâr gücünü değerlendirmektir. Veriler WAsP yazılımı ile analiz edilerek rüzgâr yoğunluğu, rüzgâr yönü belirlenerek, rüzgâr çiftliği kurulumu için gerekli parametreler için elde edilen çıktılarla birlikte değerlendirilmiştir. Ayrıca WAsP simülasyonundan elde edilen veriler esas olarak rüzgâr türbininin çalışma alanları için en uygun rüzgâr türbinini ve hedeflenen enerjiyi sağlayabilecek türbin sayısını bulmak için kullanılmıştır.

Sivas için WAsP ile Rüzgâr Enerjisi Potansiyelinin Tespiti

Türkiye'nin büyük illerinden biri olan ve İç Anadolu bölgesinde yer alan Sivas ili 28 488 km2 yüzölçümüne sahip olup ve rakım yüksekliği ortalama 1285 m.'dir. Sivas ilinin ana geçim kaynağı tarım ve hayvancılıktır. Sanayi Sivas ilinde yeni yeni gelişmektedir. Küçük sanayinin daha etkin olduğu ana sanayi kuruluşları demir-çelik fabrikaları, süt fabrikaları, tuğla fabrikaları, dikimevi, yem ve un fabrikalarıdır. Fabrika sayısının giderek artması sebebiyle bu işletmelerin gereksinim duyduğu enerji miktarı da her geçen gün artmaktadır. Bu nedenle gerekli enerjinin karşılanması için bazı alternatif çözümler araştırılmaktadır.

Bu araştırmada Şekil 1'de Google üzerinden gösterildiği gibi 37,002 Doğu boylamı ve 39,7437 Kuzey enleminde yer alan Sivas ilinde araştırma yapılmıştır. Şekil 2'de ise araştırılan bölgenin izohipsli dem haritası gösterilmiştir.

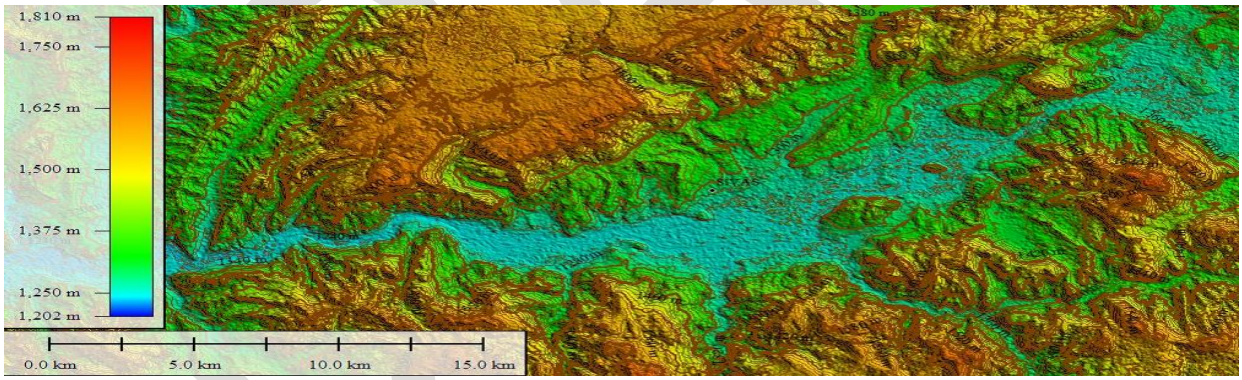
DRAFT



Şekil 1. 37,002 Doğu Boylamı ve 39,7437 Kuzey Enleminde yer alan Sivas ili Google earth görüntüsü

Sivas ilinde rüzgâr hızını ölçmek için kullanılan anemometrelerin yer aldığı alanda 100 metre aralıklarla izohipsli şekilde çizilmiştir.

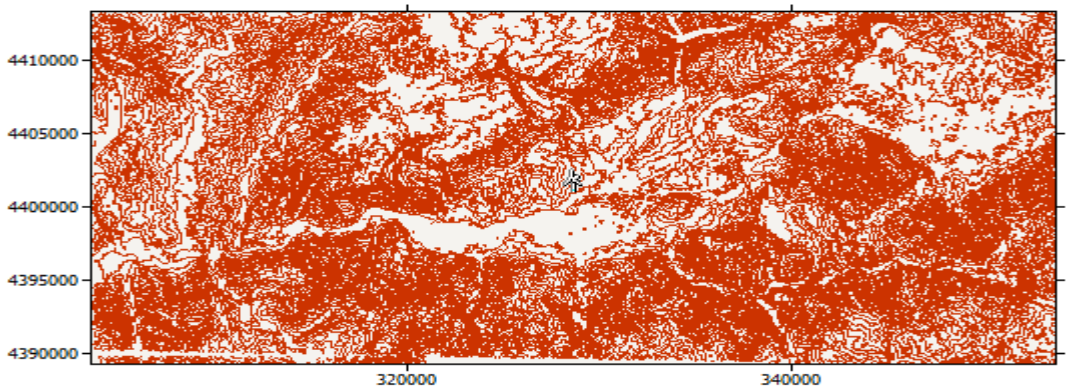
Sivas ili için yapılan bu araştırmada Meteoroloji Genel Müdürlüğü'nden alınan veriler yerden 10 m yükseklikte bir yıllık her güne ait 24 saatlik rüzgâr hızı ve yönü verileri söz konusu ile ait enlem ve boylamlar için datalar girilip WaSP simülasyon programına girdi olarak işlenmiştir. Daha sonra anemometre yeri 1,286.2 m. olarak belirtilmiştir. Sonuçta, kurulan sistem kuzey-doğu güney-batı genel rüzgâr yönlü olarak modellenmiş olup anemometreden elde edilen birim zaman aralığında birim alandan geçiş yapan rüzgâr gücü 4 W/m^2 ve ortalama rüzgâr hızı 1.12 m/s olarak elde edilmiştir. Tahmini olarak belirlenen güç yoğunluk değeri, 1.091 kg/m^3'lük hava yoğunluğu için bulunmuştur. Tablo 1'de Sivas için alan bilgisi verilmiştir.



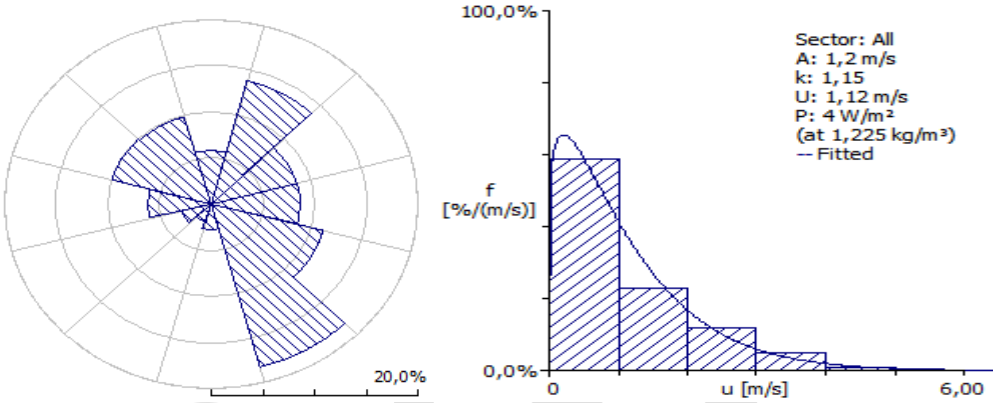
Şekil 2. Sivas İzohipsli Dem Haritası

Tablo 1. Sivas Alan Bilgisi

Anemometer height a.g.l	10m
Elevation a.s.l	1286,2 m
Net altitude a.s.l	1296,2 m
Mean air density	1,091 kg/m^3



Şekil 3. Sivas ilinde anemometrenin yerleştirildiği yer



Şekil 4. 10 m. Yüksekliğinde yerleştirilen anemometrenin rüzgâr yönlerine göre hızlarının gösterimi

Gözlemlenmiş rüzgâr enerjisi için anemometre verileri 39,44°D ve 37,00°K'de elde edilmiştir. Şekil 3'de Sivas ilinde anemometrenin yerleştirildiği yer gösterilmiştir. 10 m. Yüksekliğinde yerleştirilen anemometrenin rüzgâr yönlerine göre hızlarının gösterimi Şekil 4'de verilmiştir. Sivas ili için rüzgâr yönleri Tablo 1'de gösterilmiştir. Tablo 3'de Sivas ili için Histogram kutuları detaylandırılmıştır. Tablo 2'de Sivas ili için barometrik referans bilgileri verilmiştir.

Tablo 2. Sivas ili için rüzgâr yönleri

-	0°	30°	60°	90°	120°	150°	180°	210°	240°	270°	300°	330°
A [m/s]	1.5	1.4	0.9	1.0	0.8	0.8	0.7	0.6	1.0	2.0	1.8	2.2
k	1.33	1.26	1.47	1.40	1.25	0.93	1.52	2.04	1.14	1.57	1.79	2.18
U [m/s]	1.37	1.30	0.85	0.95	0.72	0.84	0.61	0.56	0.93	1.76	1.60	1.92
P [W/m²]	5	5	1	2	1	3	0	0	2	9	5	8
f [%]	5.8	13.9	8.5	8.5	11.3	18.3	2.8	1.9	3.0	6.2	10.0	9.8

Tabloya gore sıcaklık ortalama 13.60 derece civarındayken bağıl nem %77.55447 bulunmuştur. Yapılan hesaplamalar sonucunda; Sivas ili için ortalama hız Tablo 5'de verilmiştir.

Tablo 3. Sivas ili için Histogram kutuları

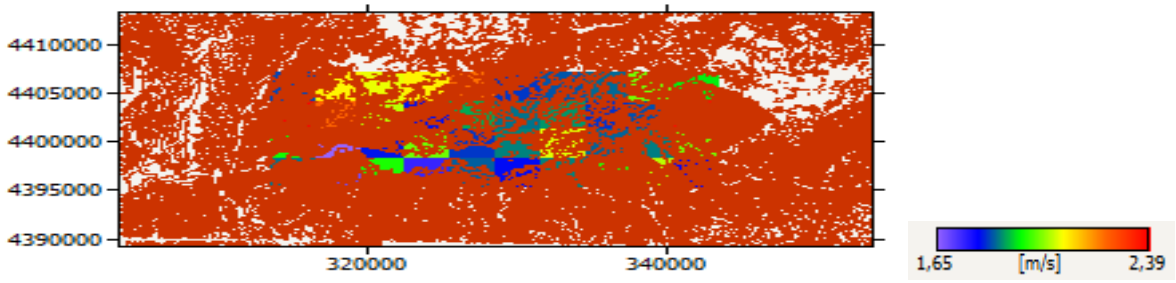
U [m/s]	0°	30°	60°	90°	120°	150°	180°	210°	240°	270°	300°	330°	All
1.0	514	537	680	615	838	732	936	959	643	333	335	295	590
2.0	207	213	282	313	128	180	45	35	265	309	364	243	229
3.0	169	154	35	61	26	41	16	6	54	205	224	321	118
4.0	101	82	3	11	7	29	2	0	35	104	66	129	51
5.0	9	12	0	0	1	16	0	0	4	40	8	12	10
6.0	0	2	0	0	0	2	0	0	0	9	3	0	2
7.0	0	0	0	0	0	0	0	0	0	0	0	0	0
8.0	0	0	0	0	0	0	0	0	0	0	0	0	0
9.0	0	0	0	0	0	0	0	0	0	0	0	0	0
10.0	0	0	0	0	0	0	0	0	0	0	0	0	0
11.0	0	0	0	0	0	0	0	0	0	0	0	0	0
12.0	0	0	0	0	0	0	0	0	0	0	0	0	0
13.0	0	0	0	0	0	0	0	0	0	0	0	0	0
14.0	0	0	0	0	0	0	0	0	0	0	0	0	0
15.0	0	0	0	0	0	0	0	0	0	0	0	0	0
16.0	0	0	0	0	0	0	0	0	0	0	0	0	0
17.0	0	0	0	0	0	0	0	0	0	0	0	0	0
18.0	0	0	0	0	0	0	0	0	0	0	0	0	0
19.0	0	0	0	0	0	0	0	0	0	0	0	0	0
20.0	0	0	0	0	0	0	0	0	0	0	0	0	0
21.0	0	0	0	0	0	0	0	0	0	0	0	0	0
22.0	0	0	0	0	0	0	0	0	0	0	0	0	0
23.0	0	0	0	0	0	0	0	0	0	0	0	0	0
24.0	0	0	0	0	0	0	0	0	0	0	0	0	0
25.0	0	0	0	0	0	0	0	0	0	0	0	0	0

Tablo 4. Sivas ili için barometrik referans bilgileri

Mean temperature	13,60 °
Ref. altitude for temperature a.s.l	2,5 m
Mean pressure	101.554 Pa
Ref. altitude for pressure a.s.l	,5 m
Relative humidity	77,55447 %

Tablo 5. Sivas ili için Ortalama Hız [m / s]

Maximum Value	2,39 m/s at (315255, 4402735)
Minimum Value	1,65 m/s at (318245, 4399745)
Mean Value	1,94 m/s

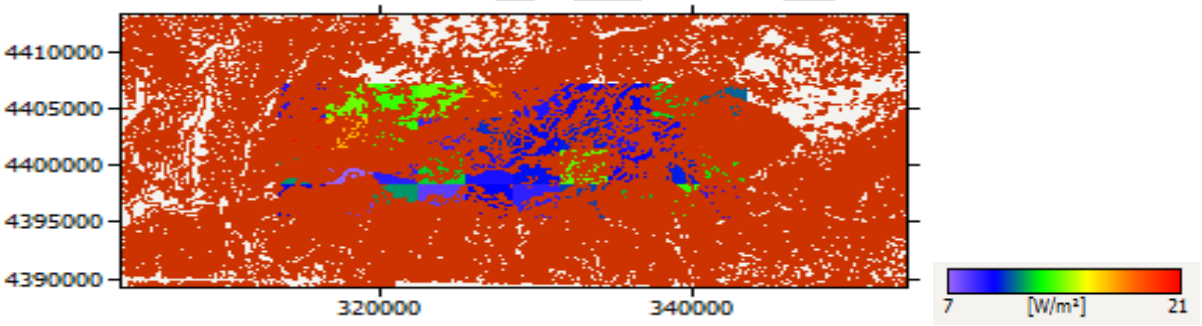


Şekil 5. Sivas ili için Maksimum ve Minimum Hız değerleri [m / s]

Şekil 5'de Sivas ili için maksimum ve minimum hız değerleri ve Tablo 6'da Sivas ili için güç yoğunluğu değerleri gösterilmiştir. Sivas Maksimum ve Minimum Güç Yoğunluğu Şekil 6'da ve Sivas ili için RIX değerleri Tablo 7'de verilmiştir. Sivas ili için Maksimum ve Minimum RIX değerleri Şekil 7'de mevcutlandırılmıştır.

Tablo 6. Sivas ili için Güç Yoğunluğu [W / m²]

Maximum Value	21 W/m² at (315255, 4402735)
Minimum Value	7 W/m² at (318245, 4399745)
Mean Value	11 W/m²



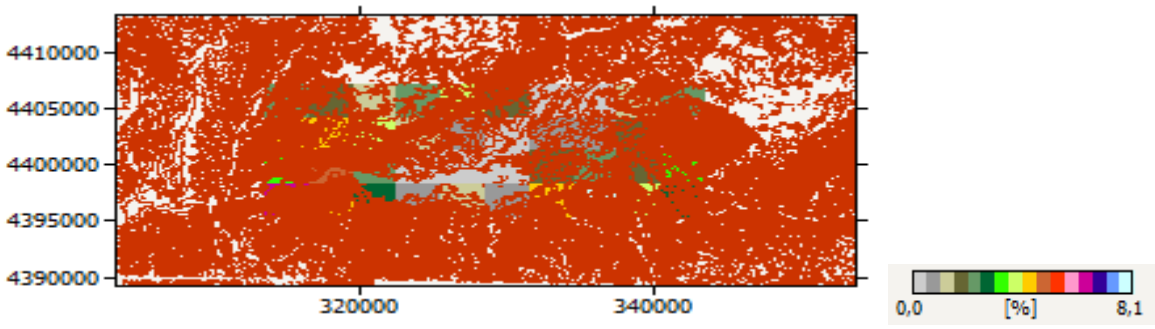
Şekil 6. Sivas Maksimum ve Minimum Güç Yoğunluğu [W / m²]

Sivas ili için gücün ve hızın minimum olduğu konum ise (318245,4399745) maximum olduğu konum (315255,4402735) olarak analiz edilmiştir.

Tablo 7. Sivas ili için RIX değerleri [%]

Maximum Value	8,1% at (336185, 4396755)
Minimum Value	0,0% at (327215, 4399745)
Mean Value	2,4%

Sivas ili için site rüzgâr iklimi ve site sonuçları sırasıyla Tablo 8 ve Tablo 9'de verilmiştir. Sivas ili için WAsP sonuç görüntüsü ise Şekil 8'de gösterilmiştir.



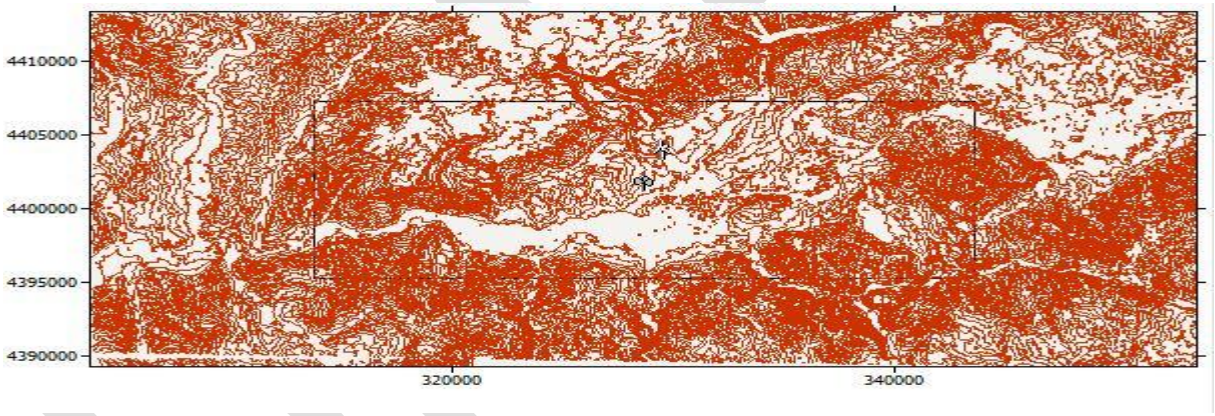
Şekil 7. Sivas ili için Maksimum ve Minimum RIX değerleri [%]

Tablo 8. Sivas ili için Site Rüzgâr İklimi

Site	Location [m]	H [m]	A [m/s]	k	U [m/s]	E [W/m²]	RIX [%]	dRIX [%]
Turbine site 001	(329628,3, 4403463,0)	55,0	1,8	1,44	1,66	7	0,6	0,5

Tablo 9. Sivas ili için Site Sonuçları

Site	Location [m]	Turbine	Elevation [m] a.s.l.	Height [m] a.g.l.	Air density [kg/m³]	Net AEP [MWh]	Wake loss [%]
Turbine site 001	(329628,3, 4403463,0)	Vestas V52-850 kW	1297,5	55,0	1,085	22,968	0,00



Şekil 8. Sivas ili için WAsP Sonuç Görüntüsü

WAsP simülasyonu aracılığıyla ile gözlemlenmiş ortalama rüzgâr iklimi analizini elde edebilmek için Meteoroloji istasyonları tarafından ölçülmüş veriler içe aktarılmıştır. Rüzgâr yönü ve hızını yerden 10 m yükseklikte ölçerek elde edilmiş Meteorolojik veriler, her 22.5^0 lik rüzgâr yönü için gözlemlenmiş ortalama rüzgâr iklimini yani rüzgâr frekansını, ortalama rüzgâr hızını göstermektedir. WAsP simülasyonu aracılığıyla rüzgâr enerjisi değerlendirmesi yapılırken, bu gözlemlenmiş ortalama rüzgâr iklimi içe aktarılmıştır. Gözlemlenmiş ortalama rüzgâr ikliminin, rüzgâr datalarının sektörler şeklinde sınıflandırılması ve Weibull dağılımı metodu ile hesaplanması sonucu oluşturulmuştur. Herbir sektör için k ve c karakteristikleri yardımıyla, istasyondan elde edilen rüzgâr verileri yerine ölçülen sonuçları temsil eden gözlemlenmiş ortalama rüzgâr iklimi değerleri kullanılır. Belli bir alanda rüzgâr yönü dağılımları rüzgârgülü ile gösterilir. Bu pusula şeklinde 12 sektöre bölünür ve ortalama rüzgâr hızı ile birlikte rüzgârın estiği yöne göre ortalama zamanın yüzdesel değerini gösterir. Güney Batı-Kuzey Doğu yönleri araştırılan lokasyonda hâkim rüzgâr yönüdür. Gözlemlenmiş ortalama rüzgâr iklimi ile girdi verisi ile WAsP yazılımı tarafından, türbin sahalarından gelen topografik etkileri türbin sahasında bölgesel rüzgâr iklimi ve göbek yüksekliğinde (55 m) rüzgâr türbini Vestas V52'ye (850 kW) ye göre uygulanmıştır. Sivas ili için planlanan hedef toplam gücü yani 22.968 MWh olduğu hesaba katılarak bu elektrik gücünü elde etmek için bölgede 28 adet rüzgâr türbini ile bir rüzgâr çiftliği kurulabildiği takdirde bu enerji ihtiyacının karşılanabileceği rapor edilmiştir.

SONUÇ

Fosil yakıtların tükenebilir doğası ve iklim değişikliğine katkıları, bugün toplumları kirletmeyen ve tükenmez yenilenebilir enerjilere doğru ilerlemeye itiyor. Sera gazları üreten birçok insan faaliyetine karşı, rüzgar enerjisi gibi bir yenilenebilir enerji kaynağının kullanımı açık ara en büyük emisyon azaltıcılardan biridir [37]. Rüzgâr enerjisi potansiyelinin değerlendirilmesi, rüzgâr türbinleri veya rüzgâr çiftlikleri ile ilgili kararları etkileyen önemli bir adımdır, ancak tüm yerler rüzgâr türbinlerinin kurulması için uygun değildir. Bu nedenle, güç üretiminin belirli bir yerini değerlendirmek için düşük rüzgâr potansiyelinin değerlendirilmesi gereklidir. Bu araştırmanın sonuçları, gelecekte Türkiye'nin Sivas ilinde veya benzer rüzgâr özelliklerine sahip başka bir yerde yerden 10 metre yükseklikte bir rüzgâr türbini geliştirmek için referans olarak uygulanabilir. Ek olarak, belirli sermaye harcamalarını tahmin etmek için kapsamlı ve dikkatli bir ekonomik analize ihtiyaç duyulacaktır.

TEŞEKKÜR

Bu çalışma, Fırat Üniversitesi MF.23.24 no'lu proje tarafından desteklenmiştir.

KAYNAKLAR

Abbasi, S.A., Abbasi, N., 2010. The likely adverse environmental impacts of renewable energy sources. Appl. Energy 65 (1-4), 121-144.

Barrios, L., Rodríguez, A., 2004. Behavioural and environmental correlates of soaring-bird mortality at on-shore wind turbines. J. Appl. Ecol. 41 (1), 72-81.

Bilir L, İmir M, Devrim Y, Albostan A. “An investigation on wind energy potential and small scale wind türbine performance at İncek region-Ankara, Turkey”. Energy Conversion and Management, 103, 910-923, 2015.

Gökçek M, Bayülken A, Bekdemir Ş. “Investigation of wind characteristics and wind energy potential in Kirklareli, Turkey”. Renewable Energy, 32(10), 1739-1752, 2007.

IEA Statistics, CO2 emissions from fuel combustion, highlights. International Energy Agency. http://www.iea.org/publications/freepublications/publication/CO_2 emıssıons from fuel combustıon Highlights (2021edition).

Onat N, Ersoz S. “Analysis of wind climate and wind energy potential of regions in Turkey”. Energy, 36(1), 148-156, 2011.

Walker, C., Baxter, J., Ouellette, D., 2015. Adding insult to injury: the development of psychosocial stress in Ontario wind turbine communities. Soc. Sci. Med. 133, 358-365.

DRAFT

ORAL PRESENTATION

Determination of electrokinetic properties of nitrite reductase (NirB) enzyme by protein film voltammetry (PFV)

Hilal Yılmaz[1*] (*ORCID: https://orcid.org/0000-0002-5782-0283*), Esra Meşe Erdoğan[1] (*ORCID: https://orcid.org/0000-0001-7425-2399*), Melek Özkan[1]*(*ORCID: https://orcid.org/0000-0001-9017-5389*),

[*1]Gebze Technical University, Faculty of Engineering, Department of Environmental Engineering Kocaeli, Türkiye

* Corresponding author e-mail:mozkan@gtu.edu.tr

Abstract

The protein film voltammetry method is used for understanding the electrokinetic behavior of an enzyme from the relation between chemical activity and electropotential. This method has some advantages such as fast and direct electron transfer between enzymes to the electrode surface, a small amount of enzyme is required for protein film voltammetry, and the absence of the complex coating on the electrode surface can increase the diffusion rates of the electrons. The electrokinetic properties of assimilatory nitrite reductase enzyme NirB from *Escherichia coli* were determined for the first time in the present study. The nitrite reductase enzyme was directly adsorbed on a glassy carbon electrode surface and this electrode was used for the characterization of the enzyme via cyclic voltammetry method. The electrokinetic properties of nitrite reductase NirB enzyme were analyzed by the Michaelis-Menten method, and the Vmax and Km values were determined to be 68.97 µA and 324.54 µM nitrite, respectively. The mass transfer mechanism of electrons to the electrode surface was determined from scan rate experiments between 25 to 500 $V.s^{-1}$ and the mass transfer was found to be diffusion controlled.

Keywords: Protein film voltammetry, Nitrite reductase, electrokinetic, Michaelis menten.

INTRODUCTION

The nitrogen cycle can be divided into seven enzymatic steps: Dinitrogen reduction to ammonia (ammonification), dissimilatory nitrite reduction to ammonia (also called ammonification), ammonia oxidation to nitrite (first step of nitrification), nitrite oxidation to nitrate (second step of nitrification), nitrate reduction to nitrite, nitrite reduction to dinitrogen by denitrification and anaerobic ammonium oxidation to dinitrogen (anammox) (Milton and Minteer, 2017). Nitrite reductase (NirB) is one of the important enzymes in the denitrification process of nitrite reduction to ammonia with the following reaction (Akhtar et al., 2013; Yılmaz et al., 2022):

$$NO_2^- + 3NADH + 5H^+ \rightarrow NH_4^+ + 3NAD^+ + 2H_2O$$

Enzyme kinetics of different types of nitrite reductases (NrfA, cytochrome c, and NirB) have been studied in some microorganisms, including *Bacillus firmus* (Gao et al., 2017), *Bacillus megaterium* (Chu et al., 2017), *Candida utilis* (Sengupta et al., 1996), *Escherichia coli* (Yılmaz et al., 2022), *Hydrogenobacter thermophilus* (Suzuki et al., 2006), *Mycobacterium tuberculosis* (Akhtar et al., 2013), and *Rhodobacter capsulatus* (Olmo-Mira et al., 2006). The electrokinetic properties of cytochrome c nitrite reductase were also studied by some researchers (Angove et al., 2002; Wijma et al., 2007). However, there is a lack of information on the electrokinetic characterization of nitrite reductase NirB of *E. coli*, which was cloned and characterized recently (Yılmaz et al 2022). The electrokinetic properties of enzymes can be determined by means of the protein film voltammetry (PFV) method.

The protein film voltammetry method is a relatively new electrochemical approach that is used for the determination of enzyme characteristics based on direct electron transfer between protein and electrode (Léger et al., 2003; Gulaboski et al., 2012). The current that results from the oxidation or reduction of the protein is proportional to the transported electrons per second. The protein is immobilized on an electrode surface by

adsorption to form an electroactive film then, by applying a potential, electrons are driven in and out of the active sites, resulting in diagnostically useful current signals. In this method, protein is configured as a film on an electrode. In the present study, the electrokinetic properties and mass transport mechanism of the nitrite reductase NirB enzyme were determined by the protein film voltammetry method.

MATERIALS AND METHODS

Reagents

All chemicals used in the present study were purchased from Sigma unless otherwise stated. Nitrite reductase NirB from *E. coli* was purified and quantitated as previously (Yılmaz et al., 2022). The purified enzyme was stored at +5 C° until used. The protein concentration in the purified enzyme mixture was measured according to the Bradford method using bovine serum albumin as a standard (Bradford, 1976). The protein concentration of the purified enzyme was calculated as 190 $\mu g.ml^{-1}$. The activity of the purified enzyme was determined by revising the protocol generated by Martínez-Espinosa. The amount of nitrite consumed in 5 mins in 50 mM PBS (phosphate buffered saline) using 3mM dithionite-reduced methyl viologen (MV) as the electron donor (Martínez-Espinosa et al., 2001) was calculated as enzyme activity. The remaining nitrite after reaction was determined colorimetrically by a diazo-coupling reaction, at 543 nm (Griess, 1879). The activity of the enzyme then the activity of ~521.1 µmol nitrite consumed min^{-1} mg^{-1} protein in 50 mM PBS, in the presence of 3 mM MV (as electron donor), and 16 mM sodium dithionite (reducing agent), pH 7.1. Phosphate buffered saline solution (50 mM, pH 7.1) was prepared by mixing K_2HPO_4, KH_2PO_4, and NaCl. Nitrite stock solutions (sodium nitrite, 100%; Isolab, Türkiye), were prepared in PBS pH 7.1.

Protein film voltammetry

Electrochemical measurements were carried out at potentiostat (VersaSTAT3). Glassy carbon working electrode (d=3mm, CHI104), platin wire counter electrode, and Ag/AgCl reference electrode (3 M KCl, BASI) were used in electrochemical measurements and electrolyte (PBS) was deoxygenated with N_2 for 5 mins before each test. Glassy carbon working electrodes were polished with 1, 0.3, and 0.05 micron alumina slurry respectively. Electrodes were checked by cyclic voltammetry in 0.5 M sulfuric acid and freshly cleaned electrodes were used for protein film voltammetry. Enzyme films on freshly prepared electrodes were prepared by covering the surface of the electrode with 20 µl of enzyme solution for 5 mins. After 5 minutes protein solution was gently removed from the electrode surface and the electrode was placed in the cell for electrochemical measurements. The cell containing 50 mM PBS was flushed with pure N_2 before each test for deoxygenation.

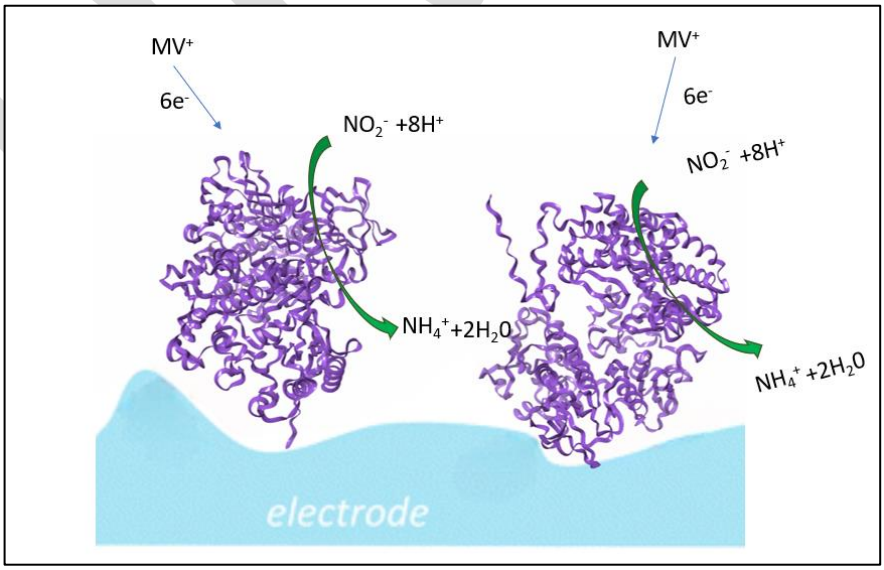


Figure 1. Schematic presentation of sub-monolayer film of nitrite reductase NirB on the glassy carbon electrode surface.

DRAFT

The Michaelis-Menten equation (Eq. (1)) was used to analyze the kinetic data. The values of Vmax and Km were determined by non-linear regression analysis of the corresponding Michaelis-Menten curves (v vs. [nitrite]) using the algorithm of the Lineweaver–Burk plot (Eq. (2)). Maximum catalytic current obtained at the addition of different concentrations of nitrite was accepted equal to Vmax (i_{max} =V_{max}).

The Michaelis Menten and Lineweaver Burk equations are as follows:

$$v = \frac{v_{max}*[S]}{[S]+K_m} \quad (1)$$

$$\frac{1}{V} = \frac{K_m}{V_{max}[S]} + \frac{1}{V_{max}} \quad (2)$$

[S]: Nitrite concentration at the beginning of the reaction.

V: Initial reaction rate
V_{max}: Maximum reaction rate, (i_{max} =V_{max})
K_m: Nitrite concentration when the reaction rate is $V_{max}/2$

The mass transport mechanism in the process was investigated by scan rate experiments. The cyclic voltammograms at the scan range between 25-500 mV.sec^{-1} were collected after the addition of 2 mM nitrite to the cell containing 1 mM MV.

RESULTS and DISCUSSION

Freshly polished glassy carbon electrodes were covered with 20 µl of NirB enzyme solutions for 5 min and then placed in an electrochemical cell containing PBS solution. To prevent the inactivation of nitrite reductase the electrolyte was flushed with pure N_2. Before the kinetic experiments were carried out, the effect of scan rates (between 25 to 500 V.s^{-1}) on catalytic currents was examined and the results are shown in Figure 2. The results show that catalytic currents were increased with increasing the scan rates as expected. As it is known the peak currents must be linearly proportional to the square root of the scan rate and for the diffusion-controlled processes, the slope of log (current) vs. log (scan rate) is 0.5. In the present study the mass transfer process was found to be diffusion-controlled one (slope is equal to 0.49).

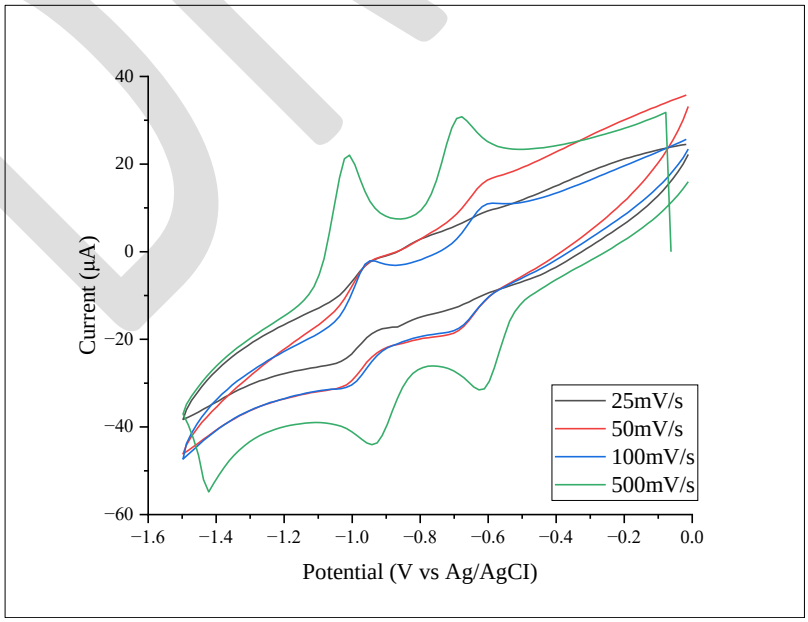


Figure 2. Effect of scan rates on the oxidation and reduction peaks of NirB immobilized GCE electrodes in the presence of 2 mM nitrite.

When the scan rates were set to 500 mV.sec^{-1}, separate peaks of oxidation and reduction were obtained. The reduction peaks seen near -0.6 V and -1V could belong to the reduction of methyl viologen (Ding et al., 2019). However, peak currents at these reduction potentials (-0.6 and -1 V) were not increased with increasing the nitrite concentration as seen in Figure 3. Only the reduction peak observed at -1.4 V was increased with nitrite addition and it can belong to nitrite reduction to ammonia. Electrokinetic nitrite reductase reaction rate and Km values were calculated from the currents at the potential of -1.4 V. The Vmax and Km values were determined by using the Lineweaver-Burk linearization method (Eq. (1)) and calculated to be 68.97 µA and 324.54 µM nitrite, respectively. This Km value of the enzyme is similar to the Km determined by *in vitro* enzyme assay (Yılmaz et al 2022).

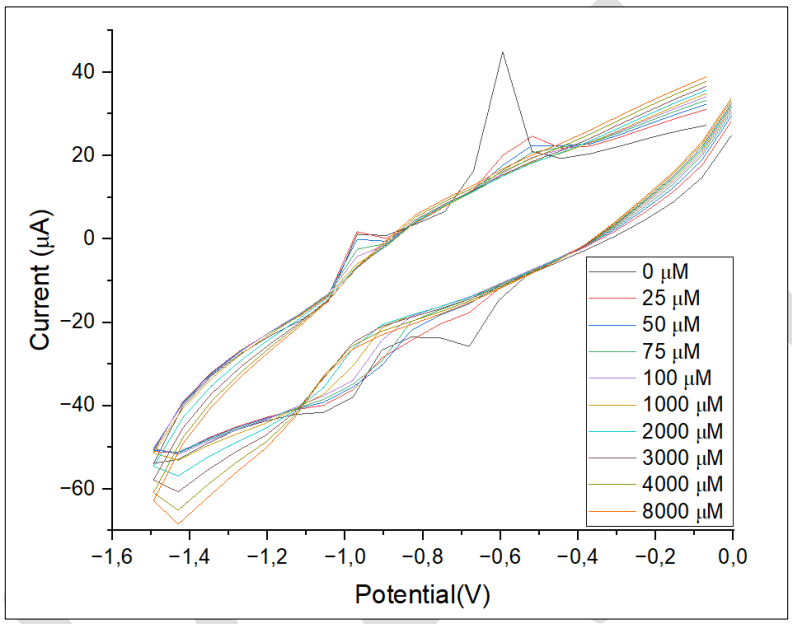
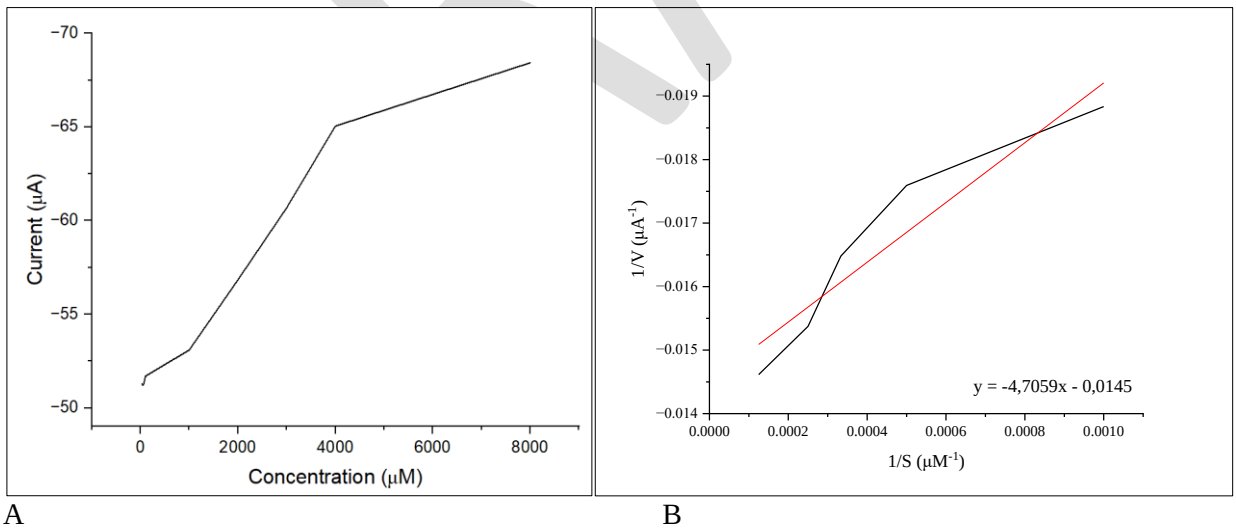


Figure 3. Cyclic voltammogram of protein film voltammetry *Scan rate:0.05V.



A B

Figure 4. A) Catalytic reduction currents vs. Nitrite concentration B)Lineweaver-Burk plot nitrite concentration range between 1-8 mM NO_2^- *Scan rate:0.05V.

CONCLUSION

Protein film voltammetry supplies much important information about the interaction of enzyme activity and electrokinetic properties of the enzyme. This method is a fast, easy, and economical alternative to the analytical methods for the characterization of the enzymes. In the present study, the mass transfer mechanism of NirB

nitrite reductase of *E. coli* was determined by scan rate experiments, and the process was found to be diffusion-controlled. The electrokinetics properties of the enzyme were determined by using Michaelis Menten's equation and the nitrite reduction peak potential was observed at a potential of -1.4 V. Km value of the NirB enzyme is found similar to its Km value obtained with in vitro enzyme assay. Protein film voltammetry was found to be a useful method for kinetic characterization of NirB nitrite reductase.

ACKNOWLEDGEMENTS
This work was supported by the Scientific and Technological Council of Turkey (TUBITAK) [grant number 120Z412].

REFERENCES

Akhtar S, Khan A, Sohaskey CD, Jagannath C, Sarkar D. 2013. Nitrite Reductase NirBD Is Induced and Plays an Important Role during In Vitro Dormancy of Mycobacterium tuberculosis. J Bacteriol 195:4592.

Angove HC, Cole JA, Richardson DJ, Butt JN. 2002. Protein film voltammetry reveals distinctive fingerprints of nitrite and hydroxylamine reduction by a cytochrome c nitrite reductase. Journal of Biological Chemistry 277:23374–23381.

Bradford MM. 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem 72:248–254.

Chu S, Zhang D, Wang D, Zhi Y, Zhou P. 2017. Heterologous expression and biochemical characterization of assimilatory nitrate and nitrite reductase reveals adaption and potential of Bacillus megaterium NCT-2 in secondary salinization soil. Int J Biol Macromol 101:1019–1028.

Ding J, Zheng C, Wang L, Lu C, Zhang B, Chen Y, Li M, Zhai G, Zhuang X. 2019. Viologen-inspired functional materials: synthetic strategies and applications. J Mater Chem A Mater 7:23337–23360.

Gao H, Li C, Ramesh B, Hu N. 2017. Cloning, purification and characterization of novel Cu-containing nitrite reductase from the Bacillus firmus GY-49. World J Microbiol Biotechnol 34.

Griess P. 1879. Bemerkungen zu der Abhandlung der HH. Weselsky und Benedikt „Ueber einige Azoverbindungen". Berichte der deutschen chemischen Gesellschaft 12:426–428.

Gulaboski R, Mirčeski V, Bogeski I, Hoth M. 2012. Protein-film voltammetry-electrochemical enzymatic spectroscopy: a review on recent progress. J Solid State Electrochem 16:2315–2328.

Léger C, Elliott SJ, Hoke KR, Jeuken LJC, Jones AK, Armstrong FA. 2003. Enzyme electrokinetics: using protein-film voltammetry to investigate redox enzymes and their mechanism. Biochem 42:8653–8662.

Martínez-Espinosa RM, Marhuenda-Egea FC, José Bonete M. 2001. Purification and characterisation of a possible assimilatory nitrite reductase from the halophile archaeon Haloferax mediterranei. FEMS Microbiol Lett 196:113–118.

Milton RD, Minteer SD. 2017. Enzymatic Bioelectrosynthetic Ammonia Production: Recent Electrochemistry of Nitrogenase, Nitrate Reductase, and Nitrite Reductase. Chempluschem 82:513–521.

Olmo-Mira MF, Cabello P, Pino C, Martínez-Luque M, Richardson DJ, Castillo F, Roldán MD, Moreno-Vivián C. 2006. Expression and characterization of the assimilatory NADH-nitrite reductase from the phototrophic bacterium Rhodobacter capsulatus E1F1. Arch Microbiol 186:339–344.

Sengupta S, Shaila MS, Rao GR. 1996. Purification and characterization of assimilatory nitrite reductase from Candida utilis. Biochemical Journal 317:147.

Suzuki M, Hirai T, Arai H, Ishii M, Igarashi Y. 2006. Purification, characterization, and gene cloning of thermophilic cytochrome cd1 nitrite reductase from Hydrogenobacter thermophilus TK-6. J Biosci Bioeng 101:391–397.

Wijma HJ, Jeuken LJC, Verbeet MP, Armstrong FA, Canters GW. 2007. Protein film voltammetry of copper-containing nitrite reductase reveals reversible inactivation. J Am Chem Soc 129:8557–8565.

Yılmaz H, İbici HN, Erdoğan EM, Türedi Z, Ergenekon P, Özkan M. 2022. Nitrite is reduced by nitrite reductase NirB without small subunit NirD in Escherichia coli. J Biosci Bioeng 134:393–398.

ORAL PRESENTATION

Nano Silisyum Uygulamasının Biberin (*Capsicum annuum* L.) Tuza Toleransı Üzerindeki Etkisi

Şebnem Kuşvuran[1] (*ORCID: https://orcid.org/0000-0002-1270-6962*), Ebru Derelli Tüfekçi[1*] (*ORCID: https://orcid.org/0000-0003-1097-8574*), Ferhat Mutlu[2] (*ORCID: https://orcid.org/0000-0002-9463-0171*)

[1] Çankırı Karatekin Üniversitesi, Gıda ve Tarım Meslek Yüksekokulu, Bitkisel ve Hayvansal Üretim Bölümü, Çankırı, Türkiye
[2]Çankırı Karatekin Üniversitesi, Fen Bilimleri Enstitüsü, Çankırı, Türkiye

*Sorumlu yazar e-mail: ebru.derelli@gmail.com

Özet

Biberde tuza toleransın artırılmasına yönelik olarak nano silisyum (Nano-Si) uygulamalarının etkinliğinin araştırıldığı çalışmada, materyal olarak Şölen biber çeşidi kullanılmıştır. Stres bitkileri için, 3 gerçek yapraklı aşamada 0 (kontrol), 50, 100 ve 150 mM olmak üzere 4 farklı tuz dozuna, Nano-Si uygulamalarında ise tuz stresi ile birlikte 0, 0,5, 1, 2 ve 3 mM dozlarına yer verilmiş, haftada bir kez olmak üzere yapraktan spreyleme şeklinde uygulanmıştır. Stres sonunda hasat edilen bitkiler, yeşil aksam yaş ve kuru ağırlıkları, gövde boyu ve çapı, yaprak sayısı ve alanı, yaprak su oransal içeriği (YOSİ), yeşil aksam Na, Cl, K ve Ca içeriği, malondialdehit (MDA), toplam klorofil ve karotenoid içeriği bakımından incelenmiştir. Tuz stresi biber çeşitlerinde bitki büyüme parametreleri ile YOSİ, K ve Ca iyon konsantrasyonu, toplam klorofil ve karotenoid içeriğinde azalmaya neden olmuş; Na ve Cl iyon konsantrasyonu ile MDA içeriğinde ise artış meydana gelmiştir. Buna karşın, tuz stresi ile birlikte nano-Si uygulamalarında tuz stresine oranla değişen oranlarda bitki büyüme parametreleri, YOSİ, K, Ca, fotosentetik pigmentlerde iyileşme sağlanmıştır. Ayrıca Na ve Cl iyon alımı ortalama olarak %6-23 düzeyinde sınırlandırılırken; MDA içeriğindeki artış %83-285 düzeyinde engellenebilmiştir. Çalışma sonucunda, nano-Si uygulamalarının tuz stresi ile ortaya çıkan zararlanmayı değişen oranlarda iyileştirdiği ve toleransı artırmada etkili olduğu, uygulamalar arasında bu olumlu etki açısından 0.50 mM ve 1 mM nano-Si uygulamalarının ön plan çıktığı belirlenmiştir.

Anahtar Kelimeler: MDA, Silisyum, Sodyum klorür, *Solanacae*, Tuz stresi

Effect of Nano Silicon Application on Salt Tolerance of Pepper (*Capsicum annuum* L.)

Abstract

In the study investigating the effectiveness of nano silicon (Nano-Si) applications to increase salt tolerance in pepper, Şölen pepper variety was used as material. For stress plants, 4 different salt doses of 0 (control), 50, 100 and 150 mM were applied at the stage of 3 true leaves, and Nano-Si applications included 0, 0.5, 1, 2 and 3 mM doses along with salt stress and were applied once a week by foliar spraying. Plants harvested at the end of the stress were analyzed in terms of fresh and dry weights, stem length and diameter, number and area of leaves, leaf water content (LWRC), Na, Cl, K and Ca ions content, malondialdehyde (MDA), total chlorophyll and carotenoid contents. Salt stress caused a decrease in plant growth parameters, RWC, K and Ca ion concentration, total chlorophyll and carotenoid content, and an increase in Na and Cl ion concentration and MDA content in plant. On the other hand, plant growth parameters, RWC, K, Ca, photosynthetic pigments were improved in nano-Si treatments with salt stress at varying rates compared to salt stress. In addition, Na and Cl ion uptake was limited by 6-23% on average, while the increase in MDA content was prevented by 83-285%. As a result of the study, it was determined that nano-Si applications improved the damage caused by salt stress at varying rates and were effective in increasing tolerance, and 0.50 mM and 1.00 mM nano-Si applications were prominent among the treatments in terms of this positive effect.

Keywords: MDA, Silicon, Sodium chloride, *Solanacae*, Salt stress

GİRİŞ

Artan dünya nüfusu ve buna bağlı olarak artan gıda ihtiyacı, tarımsal üretimde verimliliğin ön plana çıkmasına, mevcut üretim alanlarından en yüksek verimin alınmasına yönelik çalışmaların hız kazanmasına neden olmuştur. Özellikle abiyotik stres faktörleri, küresel gıda güvenliği, gıda kalitesi ve bitki verimliliği üzerinde sınırlayıcı etkilere sahiptir. Dünyadaki sulanabilir tarım arazilerinin yaklaşık %20'si toprak tuzluluğundan olumsuz etkilenmektedir. Bu sorun aşırı gübre kullanımı, uygun olmayan sulama sularının kullanımı, doğal çevre koşulları ve küresel iklim değişikliği ile birlikte artış göstermiştir (Zhao ve ark., 2021). Tuz stresi, çimlenme, büyüme ve gelişme, çiçeklenme ve meyve tutumunu engelleyerek bitkileri olumsuz etkilemektedir. Tuzlu topraktaki yüksek sodyum konsantrasyonları, bitkide su alımını ve besinlerin emilimini sınırlar. Su eksikliği ve beslenme dengesizliği, ozmotik stres ve iyonik stres dahil olmak üzere birincil stresleri ön plana çıkartmaktadır. Bu birincil stresler oksidatif stresle sonuçlanmakta, ikincil bir strese de neden olabilmektedir. Tuz stresi ile birlikte çeşitli fizyolojik ve moleküler değişiklikler meydana gelmekte ve fotosentez sınırlanarak bitki büyüme ve gelişmesinde sorunlar ortaya çıkmaktadır (Gong, 2021).

Silisyum (Si), oksijenden sonra yer kabuğunda (%28) en çok bulunan ikinci toprakta ise (%54) en bol bulunan elementtir. Stres altındaki bitkilerde önemli rollerinden dolayı Si, faydalı veya yarı gerekli bir element olarak bilinmesine karşın gereklilik kriterlerini karşılamadığı ve bitki metabolizmasında yer aldığına dair yeterli kanıt olmadığı için gerekli olmayan bir besin maddesi olarak sınıflandırılmıştır. Si, temel bir besin maddesi olarak geniş çapta tanınmamakla birlikte, genellikle bitki büyümesi, fizyolojik/metabolik yollar, hücre yapısı ve çok çeşitli abiyotik ya da biyotik çevresel streslerin hafifletilmesi için yararlı olan "değerli bir element" veya "yapısal element" olarak kabul edilir. Birçok çalışma farklı bitki türlerinde tuzluluk, ağır metal stresi, kuraklık gibi abiyotik stres koşullarında bitki büyüme ve gelişmesi ile verimi artırmadaki rolünü doğrulamıştır (El-Ramady ve ark., 2022).

Biyoteknoloji ve tarım sektöründe önemli bir yere sahip olan nanoteknoloji, gıda işleme endüstrileri, patojen tespiti ve teşhisi, gıda mühendisliği, ambalaj malzemeleri ve ekipmanları gibi farklı alanlarda yer almaktadır. Nanopartiküller boyutları 1-100 nm arasında değişen küçük moleküllü parçacıklar olup genellikle nano ölçekli parçacıklar (NSP'ler) olarak adlandırılmaktadır. Nanopartiküller, bitki büyüme ve gelişme destekleyicileri, herbisitler, nano-pestisit, nanogübreler gibi tarım sektöründe geniş kullanım alanına sahiptir. Nanogübrelerin az miktarda kullanımı, gübre tasarrufu sağlanmasına katkıda bulunmakta ayrıca toprakta kimyasal gübre ve pestisitlerin birikmesi nedeniyle oluşan artan toprak toksisitesini azaltmaya yardımcı olmaktadır. Bunun yanı sıra abiyotik veya biyotik stres koşullarında fizyolojik ve biyokimyasal düzeyde farklı mekanizmaları uyararak stres koşullarına toleransın sağlanmasında etkili olmaktadırlar (Saxena ve ark., 2016). Silisyum nanopartiküller geniş alanda etkili bir şekilde yayılım gösterebilmekte olup 7.0 nm çapa sahip 1.0 gram silika nanoparçacıklarının 400 m2'ye eşit geniş bir absorpsiyon yüzeyi gösterdiği tahmin edilmektedir. Ayrıca, Si nanopartiküller turgor basıncını artırarak yaprak nisbi su içeriği ve su kullanım etkinliğini olumlu etkilemektedir. Si partikülleri aynı zamanda antioksidan enzim aktivitelerinde artış sağlaması, iyon regülasyonunda etkili olması gibi özellikleri sayesinde tuzluluk, kuraklık, ağır metal toksisitesi gibi abiyotik stres koşullarına toleransın artırılmasında etkili olmaktadır (Liang ve ark., 2007; Saxena ve ark., 2016).

Dünyada ve ülkemizde önemli bir potansiyele sahip olan biber, geniş alanlarda yetiştirilmekte, taze- sofralık ve sanayilik olarak değerlendirilebilen önemli bir sebze grubunu oluşturmaktadır. Dünya biber üretimi 2021 yılında 2 milyon ha alanda 36.286.643 ton olup, Türkiye biber üretiminde Çin ve Meksika'dan sonra 802.617 da alanda 3.091.295 tonluk üretimle üçüncü sırada yer almaktadır (Anonim, 2022). Küresel iklim değişikliğinin en önemli sonuçlarından biri olan tuzluluk sonucu birçok tarımsal üründe verim ve verim parametrelerindeki olumsuzluklar karşısında farklı uygulamaların ele alınarak, tarımsal sürdürülebilirliğin sağlanması oldukça önemlidir. Bu doğrultuda nano-Si uygulamalarının bitki üzendeki pozitif etkileri değerlendirildiğinde, biberde tuz stresi karşısında bitki büyüme ve gelişme üzerinde nano-Si etkilerinin belirlenmesi bu çalışmanın en önemli çıkış noktasını oluşturmaktadır.

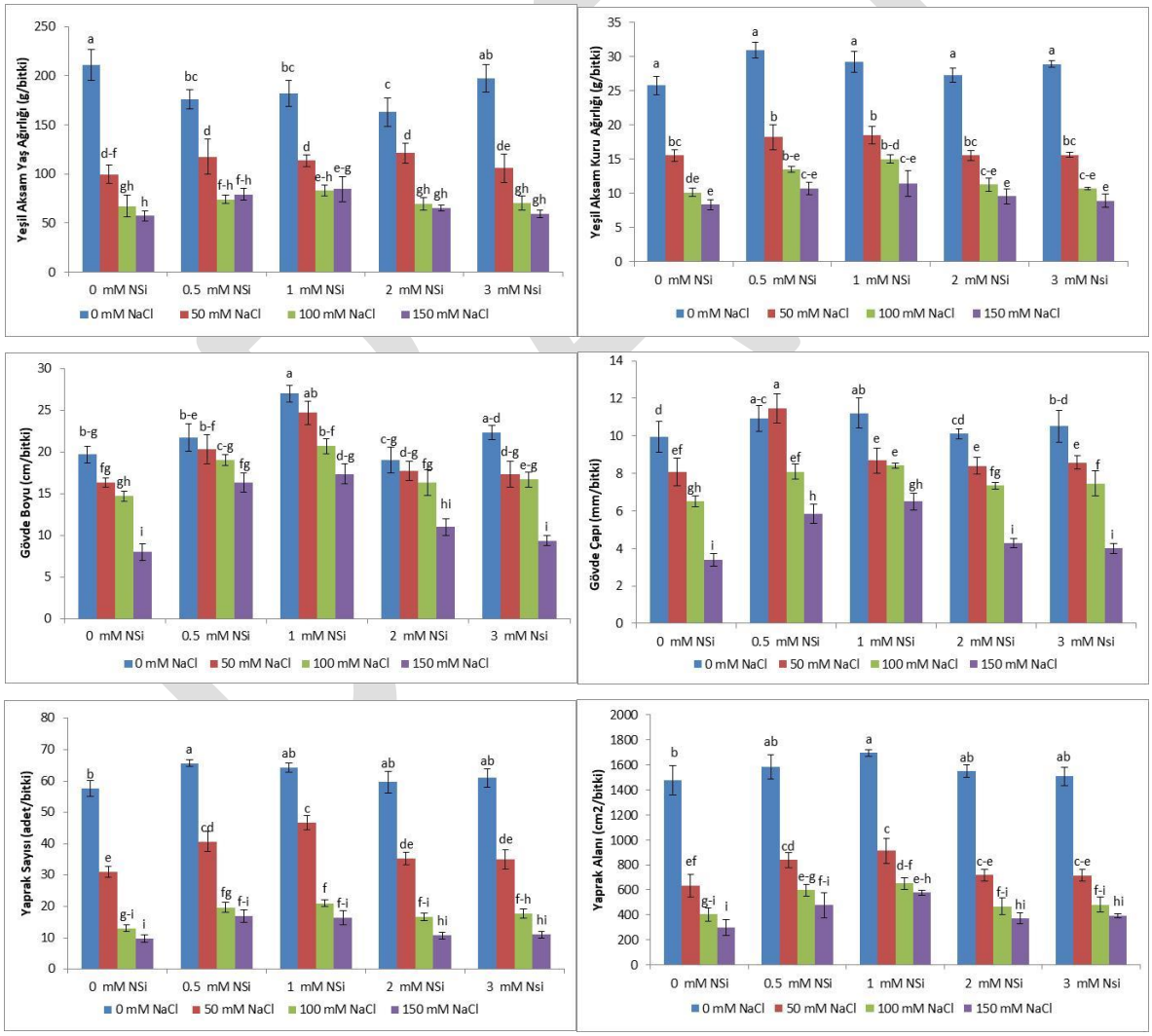
MATERYAL VE METOT

Çalışmada materyal olarak Şölen biber çeşidi kullanılmış, tohumlar 2:1 oranında karıştırılmış torf:perlit karışımı içeren viyollere ekilmiştir. Tohum ekiminden 55 gün sonra fideler 12 litre hacmindeki plastik saksılara (torf:perlit) her saksıda üç adet bitki olacak şekilde şaşırtılmıştır. Bitkiler saksılara şaşırtıldıktan 18 gün sonra tuz stresi uygulamalarına başlanmıştır. Bu amaçla çalışmada 4 farklı NaCl dozuna (0, 50, 100 ve 150 mM NaCl) yer verilmiştir. Nano-Si uygulaması için; 0, 0.5, 1, 2 ve 3 mM dozları kullanılmıştır. On altı gün stres uygulamalarına maruz bırakılan bitkiler hasat edilerek yeşil aksam yaş ve kuru ağılıkları, gövde boyu

ve çapı, yaprak sayısı ve alanı, yaprak oransal su içeriği, lipid peroksidasyon içeriği (MDA), toplam klorofil, yeşil aksam sodyum (Na), klor (Cl), potasyum (K) ve kalsiyum (Ca) içerikleri bakımından değerlendirilmiştir. Çalışmada, yaprak oransal su içeriği (YOSİ) Türkan ve ark. (2005), MDA içeriği Lutts ve ark. (1996) ve toplam klorofil Kusvuran ve ark. (2021) tarafından belirtilen yöntemler izlenerek gerçekleştirilmiştir. İyon içeriklerinin belirlenmesinde Dasgan ve Koc (2009) tarafından belirtilen yöntem izlenmiştir. Bitki yeşil aksam klor konsantrasyonun belirlenmesi Nielsen (2017)'e göre ve Mohr metodu ile yapılmıştır. Çalışma tesadüf parselleri deneme desenine göre 3 tekrarlamalı her tekrarda 3 bitki olacak biçimde yürütülmüştür. Verilerin istatistiksel analizinde JMP (versiyon 8.0) programı kullanılmıştır. Ortalamaları arasındaki farklılıklar LSD testine ($p \leq 0.05$) göre gruplandırılmıştır.

BULGULAR

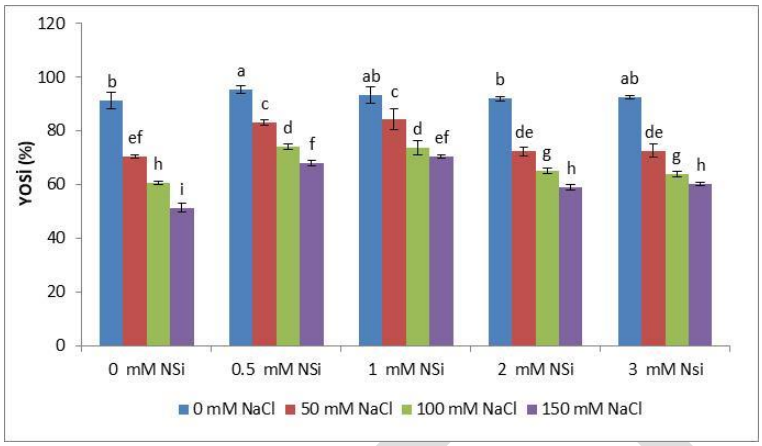
Nano-Si uygulamalarının tuza toleransın sağlanması yönündeki etkinliğinin incelendiği çalışmada, farklı tuz konstrasyonları ile oluşturulan tuz stresi koşullarında yaş ve kuru ağırlık, gövde boyu ve gövde çapı, yaprak sayısı ve yaprak alanı değerlerinde değişen oranlarda azalmaya neden olmuştur. Yaş ağırlık değerleri stres koşullarında %52-73, kuru ağırlık değerlerinde %28-65, gövde boyu ve çapında %16-59, yaprak sayısı ve yaprak alanı bakımından %19-83 oranında azalma göstermiş, bu değişim 150 mM NaCl uygulamasında ön plana çıkmıştır. Nano-Si uygulamaları ile genel olarak stresin olumsuz etkisi sınırlandırılmış ortalama olarak %17-39 oranında iyileşme sağlanmıştır. Nano-Si uygulamaları kendi içerisinde değerlendirildiğinde, en yüksek iyileşmenin sağlandığı dozlar 0,5 ve 1 mM dozları olmuştur (%17-116 artış) (Şekil 1).



Şekil 1. Nano-Si uygulamalarının tuz stresi koşullarında morfolojik parametreler üzerindeki etkisi

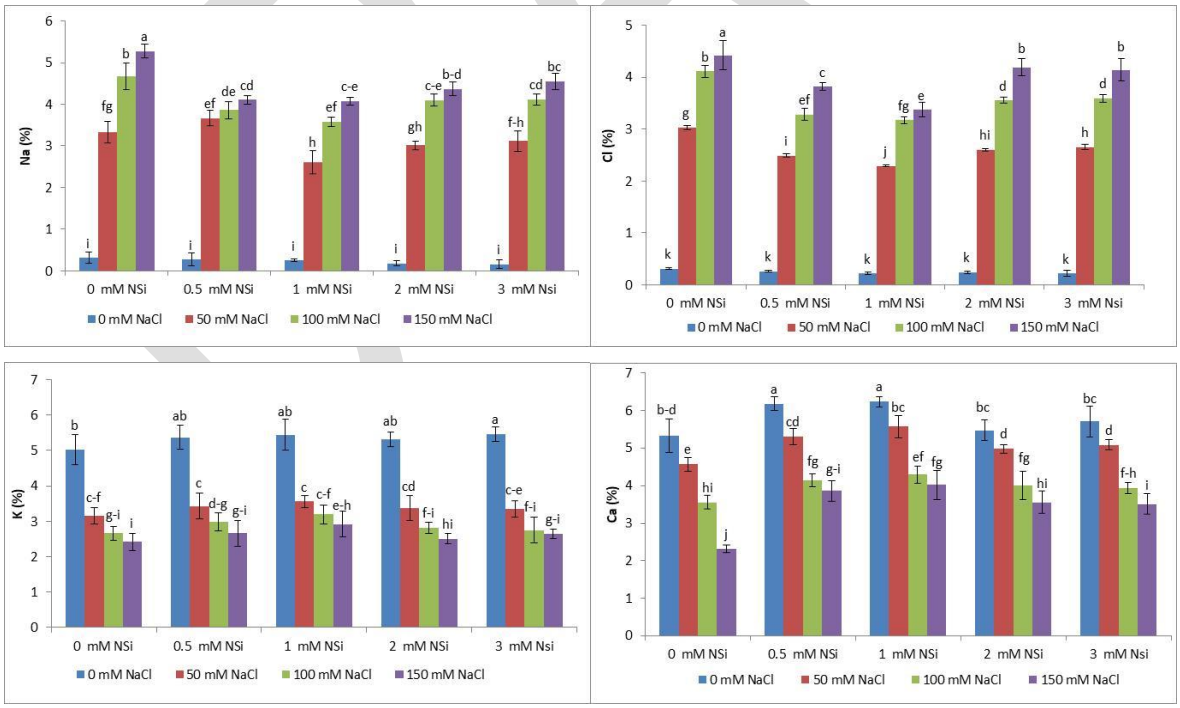
Yaprak oransal su içeriği (YOSİ), tuz stresine bağlı olarak kontrol bitkilerine oranla azalma göstermiştir. YOSİ %7-43 oranında azalmış, bu değişim 150 mM NaCl uygulamasında belirginleşmiştir. Nano-Si uygulamaları

ile birlikte YOSi değerlerindeki azalma sınırlandırılmış ve uygulama yapılmayan bitkilere oranla ortalama olarak %17 oranında iyileşme sağlanmıştır. Elde edilen bulgular çerçevesinde, en etkin nano-Si dozu 0.5 ve 1 mM konsantrasyonları olurken, bu uygulamalarda %18-37 oranında yaprak oransal su içeriği korunmuştur (Şekil 2).



Şekil 2. Nano-Si uygulamalarının tuz stresi koşullarında yaprak oransal su içeriği (YOSİ) üzerindeki etkisi

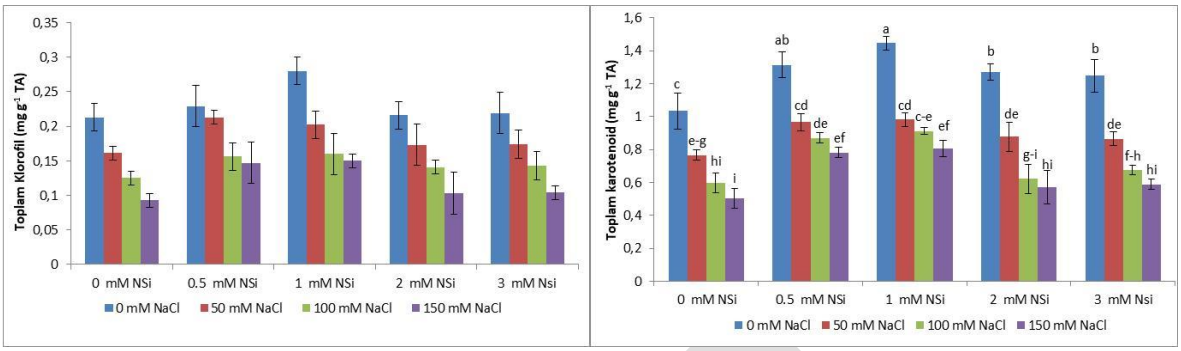
Yeşil aksam Na, Cl, K ve Ca iyon içeriklerinin de incelendiği çalışmada; Na ve Cl iyon içeriği tuz konsantrasyonundaki artışa bağlı olarak artış göstermiş, bu artış özellikle 150 mM NaCl uygulamasında en yüksek düzeye ulaşmıştır. Kontrol bitkilerine oranla 150 mM NaCl uygulamasında %1603 oranında Na ve %1133 oranında Cl iyon konsantrasyonunda artış tespit edilmiştir. Nano-Si uygulamaları ile birlikte Na iyon içeriğinde ortalama olarak %14 ve Cl iyon içeriğinde %16 oranında azalma meydana gelmiş ve toksik Na ve Cl iyon alımı sınırlandırılmıştır. Bu sınırlama 0,5 ve 1 mM nano-Si uygulamasında %17-24 düzeylerinde gerçekleşmiştir. K ve Ca iyon alımı tuz stresine bağlı olarak azalma göstermiş, bu azalma 150 mM NaCl uygulamasında %51 (K) ve %56 (Ca) oranlarında tespit edilmiştir. Nano-Si uygulaması ile birlikte K ve Ca iyon içerikleri korunmuş dolayısıyla %4-73 oranlarında iyileşme sağlanmıştır (Şekil 3).



Şekil 3. Nano-Si uygulamalarının tuz stresi koşullarında Na, Cl, K ve Ca içeriği üzerindeki etkisi

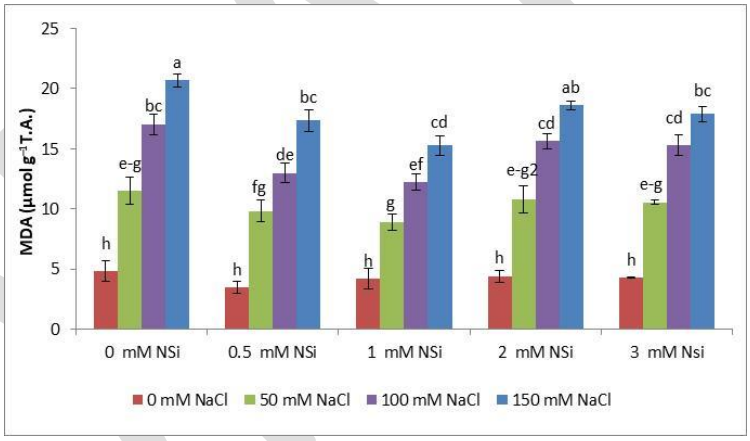
En yüksek fotosentetik pigment içerikleri kontrol bitkilerinin nano-Si uygulamalarında belirlenmiş, toplam klorofil ve karatenoid içeriği tuz stersi ile birlikte azalma göstermiştir (Şekil 4). Bu değişim

toplam klorofilde %5-56, karatenoid içeriğinde ise %6-51 oranında tespit edilmiştir. Nano-Si uygulamaları tuzun tüm düzeylerinde iyileştirici bir özellik göstermiş, ve toplam klorofil ve karatenoid içeriğinde ortalama olarak %7-61 oranında tuz stresinin olumsuz etkisi sınırlandırılmıştır.



Şekil 4. Nano-Si uygulamalarının tuz stresi koşullarında toplam klorofil ve karotenoid içeriği üzerindeki etkisi

Malondialdehid (MDA) içeriği stres koşullarında artmış (%138-328), en yüksek MDA içeriği 150 mM NaCl uygulamasında 20,68 µmol g–1T.A. (%328 artış) belirlenmiştir. Nano-Si uygulamalarında ise değişim %83-269 düzeyinde olup, ortalama olarak %16 düzeyinde MDA içeriğindeki artış sınırlandırılmıştır. Nano-Si uygulamaları içerisinde 0,5 ve 1 mM nano-Si uygulamalarında %14-28 oranında iyileşme sağlanmıştır (Şekil 5).



Şekil 5. Nano-Si uygulamalarının tuz stresi koşullarında MDA içeriği üzerindeki etkisi

TARTIŞMA

Artan dünya nüfusunun beslenme ihtiyaçlarının karşılanma gerekliliği, bitkisel üretim ve verimliliğin artırılmasına yönelik ilgiyi de artırmıştır. Bu nedenle, bitkisel üretimde verim ve kalitenin nasıl iyileştirileceği acil bir küresel tarım sorunu haline gelmiştir. Tuz stresi, dünya çapında tarımı tehdit eden önemli bir çevresel sorundur. Dünyadaki sulu tarım alanlarının yaklaşık %20'si toprak tuzlanmasından olumsuz etkilenmektedir. Doğal çevrenin bozulması, kötü sulama uygulamaları, gübreleme ve iklim değişiklikleri gibi faktörler sorunun giderek artmasına neden olmaktadır. Bu doğrultuda, verimin etkili bir şekilde iyileştirilmesine yönelik uygulamalar büyük önem taşımaktadır (Zhao ve ark., 2021). Tuzluluk, bitkilerde morfolojik ve biyokimyasal fonksiyonlar üzerinde olumsuz bir etkiye neden olmaktadır. Tohum çimlenmesini, bitki büyümesini, gelişmesini ve verimi olumsuz etkilemektedir (Zhang ve Dai 2019). Klorofil ve karotenoidlerin içeriğini azalmakta, kloroplast yapısını ve PSII sistemini bozarak fotosentetik sistemlerin etkinliği sınırlanmaktadır (Pan ve ark., 2020). Toprak tuzluluğu, toprak su potansiyelini ve yaprak su potansiyelini azalmasına; bitki su ilişkisinde dengesizlikler ve sonuçta ozmotik strese yol açmaktadır (Navada ve ark., 2020). Tuzluluk sodyum ve klor iyon içeriklerindeki artış ile ozmotik stresin meydana gelmesine sebep olmaktadır. Tarımsal üretimde

önemli bir yere sahip olan nano-Si partikülleri nano-herbisitler, nano-gübreler ve nano-pestisitler olarak kullanımları yoluyla çeşitli abiyotik stresleri hafifletmede etkinlik gösterebilmektedirler (Rastogi ve ark., 2019). Silisyum, bitkilerde fizyolojik fonksiyonları uyarmanın yanı sıra bitkinin stres koşullarında hayatta kalma potansiyelini geliştirmede önemli bir yer tutmaktadır. Bu doğrultuda silisyum, bitkinin abiyotik stres toleransını artırmakta, fotosentetik aktiviteyi iyileştirilmesinde, iyon regülasyonunun sağlanmasında ve elementlerin toksisite etkisinin azaltılmasında rol oynamaktadır (Merwad ve ark., 2018).

Farklı nano-Si dozlarının (0.5, 1, 2 ve 3 mM nano-Si) tuza toleransın sağlanması açısından etkilerinin incelendiği bu çalışmada, 2 farklı biber çeşidi, farklı düzeylerde (0, 50, 100 ve 150 mM NaCl) oluşturulan tuz stresine maruz bırakılmıştır. Tuz stresi bitki büyüme parametrelerinde azalmaya neden olmuş, bu azalma çeşitler ve tuz konsantrasyonlarına bağlı olarak farklılıklar göstermiştir. Özellikle 150 mM NaCl uygulamasında tuz stresi etkisi en belirgin biçimde görülmüştür. Buna göre kontrol bitkilerine oranla ortaya çıkan azalma büyüme parametreleri bakımından %15-83 düzeyinde gerçekleşmiştir. Bitkilerde büyüme ve gelişmeyi olumsuz etkileyen en önemli faktörlerin başında tuz stresi gelmektedir. Özellikle stres koşullarında ortaya çıkan ozmotik etki, iyon toksisitesi, fotosentez etkinliğinin azalması ve beslenme yetersizliği nedeniyle ortaya çıkan fizyolojik bozukluklardan kaynaklanmaktadır (Almodares ve ark., 2008). Biberde farklı NaCl dozlarının bitki büyüme ve gelişmesine etkisini inceleyen Kaouther ve ark. (2013), bitki yaş ve kuru ağırlıkları ile morfolojik parametreler ve yaprak oransal su içeriği gibi fizyolojik parametreler bakımından azalma meydana geldiğini ifade etmişlerdir. Benzer sonuçlar Afzal ve ark. (2014) tarafından gerçekleştirilen çalışmada da vurgulanmış, araştırıcılar tuz konsantrasyonundaki artışa bağlı olarak biber genotiplerin değişen oranlarda zararlanma ile karşı karşıya kaldıklarını ifade etmişlerdir. Nano-Si uygulamaları tuz stresinin ortaya koyduğu olumsuzluk ve zararlanmaları önemli düzeyde sınırlandırdığı, bu sınırlandırmanın uygulama dozuna göre değişim gösterdiği belirlenmiştir. Genel olarak bitki büyüme parametreleri bakımından nano-Si uygulamalarının %17-39 düzeyinde iyileşme sağladığı görülmüştür. Silisyumun bitki su dengesini sağlanması, bitki savunma sistemlerinin aktivasyonu, iyon regülasyonun sağlanması üzerindeki iyileştirici etkilerine bağlı olarak tuzluluğun bitkiler üzerindeki olumsuz etkilerini azalttığını, vurgulayan Haghighi ve Pessarakli (2013) domateste yaş ve kuru ağırlıklar, kök hacmi, yaprak su içeriği, fotosentetik oran, mezofil iletkenliğini ve bitki su kullanım verimliliği ve klorofil konsantrasyonu nano-Si uygulamasıyla artış göstermiştir.

Yüksek miktarda tuz konsantrasyonu bulunması, toprak ozmotik basıncının artmasına neden olarak, bitkinin su alım gücünü düşürmekte, hatta tamamen durdurmaktadır. Özellikle Na ve Cl iyonları ile ortaya çıkan iyon toksisitesi ise su taşınımında etkili olan hücrelerde zararlanmalara yol açmakta böylece bitki su ilişkisinde bozulmalara neden olmaktadır. Gerçekleştirilen bu çalışmada tuz uygulamalara NaCl oranına bağlı olarak YOSİ değerlerinde azalmalara neden olmuştur. Bu değişim 150 mM NaCl uygulamasında ön plana çıkmış ve ortalama olarak kontrol bitkilerine oranla %31 düzeyinde azalma meydana gelmiştir. Nano-Si uygulamaları stres koşullarında azalan yaprak oransal su içeriğini iyileştirmede (%3-37 artış) oldukça etkili olmuştur. Uygulamalar içerisinde 0.50 ve 1.00 mM nano-Si uygulamalarında bu etki ön plana çıkmıştır. Silisyum stres koşullarında transprasyon oranını düşürerek bitki bünyesindeki nemin korunmasında önemli bir rol oynamaktadır (Haghighi and Pessarakli 2013). Kalteh ve ark. (2014) silisyumun bir tabaka oluşturarak hücre duvarının dayanıklılığını arttırdığını, 7 nm çapındaki bir gram silisyum nanopartikülün 400 m^2'ye eşit bir soğurma yüzeyine sahip olduğunu, bu nedenle, Si nanoparçacıkları, ksilem nemini ve su translokasyonunu etkileyerek su kullanım etkinliğini artırdığı ifade etmiştir.

Kök bölgesinde tuz birikimi, ozmotik stresin yanı sıra K ve Ca gibi temel element ve minerallerin alımını kısıtlamakla beraber, Na ve Cl alımını indükleyerek hücre iyonu homeostazını (dengeleşimini) olumsuz etkilemektedir. Biberde gerçekleştirilen farklı çalışmalarda NaCl uygulamaları ile oluşturulan tuz stresi koşullarında bitki bünyesinde Na ve Cl iyon içeriklerinde artış meydana geldiği ifade edilmiştir. Stres karşısında büyümede azalmanın nedeni, stomaların kapanması ile yapraklarda toksik Na ve Cl birikimine bağlı olarak fotoasimilat üretiminin miktarının sınırlanmasıdır. Nano-Si, tuz stresi koşullarında terlemeyi kontrol altında tutarak, su kaybını önlemekte böylece bitki bünyesine Na ve Cl iyon girişi sınırlanmaktadır (Haghighi ve Pessarakli, 2013). Biberde gerçekleştirilen bu çalışmada, Nano-Si uygulamaları Na ve Cl iyon alımını %6-23 ve %5-23 oranında sınırlandırırken; K içeriğinde %4-21 ve Ca içeriğinde %9-73 oranında iyileşme sağlamıştır. Soyada gerçekleştirilen bir çalışmada, tuz stresi ile birlikte Na iyon içeriğinde artış meydana geldiğini, nano-Si uygulamasının K alımını teşvik ederken Na iyon alımının ise sınırlandığını ifade etmişlerdir (Farhangi-Abriz ve Torabian, 2018). Alsaeedi ve ark. (2019) hıyarda, nano-Si uygulamalarının tuz stresi koşullarında toksik Na alımını %38-77 oranında azalttığını, K içeriği korunarak K/Na oranı %127-735 oranında artış gösterdiğini bildirmişilerdir.

Stres sonucu oluşan serbest radikaller, membran lipid ve proteinlerin geri dönüşümsüz şekilde hasara uğramasından sorumludur. Lipid peroksidasyonun son ürünü, malondialdehid (MDA), eten ve pantendir. Oluşan MDA, hücre membranlarından iyon alışverişine etki ederek membrandaki bileşiklerin çapraz bağlanmasına yol açar ve iyon geçirgenliğinin ve enzim aktivitesinin değişimi gibi olumsuz sonuçlara neden olur. Lipid peroksidasyonu; membran bütünlüğünün yok olmasına, hücrenin elektrolitlere permeabilitesinin artmasına neden olur (Gözen ve Kuşvuran, 2021). Kavun, biber, domates, patlıcan ve brokoli gibi farklı bitki türlerinde gerçekleştirilen çalışmalarda tuz stresi koşullarında bitkilerde MDA içeriğinin arttığını göstermiştir. Nano-Si uygulamaları MDA içeriğindeki artışı %83-285 oranında engellemiştir. Farhangi-Abriz ve Torabian (2018) soyada ve Ismail ve ark. (2022) bezelyede gerçekleştirmiş oldukları çalışmalarında nano-Si uygulamaları ile MDA içeriğinde azalma meydana geldiğini ifade etmişlerdir.

Tuz stresi, kloroplastın parçalanmasına, pigment protein komplekslerinin kararsızlığına, klorofillerin yok olmasına ve karotenoidlerin miktar ve bileşiminde değişikliklere neden olmaktadır (Bayram ve ark., 2021). Nano-Si uygulamaları biberde toplam klorofil ve karotenoid içeriğinde kontrol bitkilerine oranla %5-56 düzeyinde azalmaya neden olmuştur. Nano-Si uygulamaları meydana gelen azalmayı belirli oranda sınırlandırmış fotosentetik pigmentlerde %9-87 iyileşme sağlanmıştır. Bu çalışmada, nano-Si uygulaması, tuzlu koşullar altında biberde daha yüksek fotosenteik pigment içeriğinin ortaya çıkmasına imkan sağlamış, NaCl'nin biberde klorofil içeriği üzerindeki olumsuz etkilerinin hafifletilmesine yardımcı olmuştur. . Önceki çalışmalar tuz stresinin börülce, fasulye, bakla ve soyada tuz stresinin bitki büyümesinde önemli azalmalara neden olduğunu, ancak Si uygulamalarının toplam fotosentetik pigment içeriği, fotosentetik hız, klorofil içeriği, stoma iletkenliği, terleme ve hücreler arası karbondioksit konsantrasyonu artırarak bitkilerin büyüme ve gelişmesini büyük ölçüde iyileştirdiğini göstermiştir (Avestan ve ark., 2019). Haghighi ve Pessarakli (2013) domateste, Avestan ve ark. (2019) çilekte gerçekleştirmiş oldukları çalışmalarında nano-Si uygulamalarının tuz stresi koşullarında klorofil içeriğinde meydana gelen kayıpları önemli ölçüde sınırlandırdığını vurgulamışlardır.

SONUÇ

Tuz stresi bitkisel üretimi ve verimliğiği etkileyen temel abiyotik stres faktörlerinin başında gelmektedir. NaCl ve nano-Si ile muamele edilen biber fideleri, büyüme ve fizyolojik özelliklerde önemli değişiklikler göstermiştir. Bu çalışmada fideler tuz stresine maruz bırakıldığında, bitki büyüme ve gelişmesinde, fotosentetik piment içeriği, YOSİ, K ve Ca içeriğinde azalma meydana gelirken MDA, Na ve Cl içeriğinde ise artış meydana gelmiştir. Bununla birlikte, nano-Si, hücresel oksidatif hasarı hafifleterek tuzluluğun olumsuz etkilerini önemli ölçüde azaltmıştır, bu değişim dozlara bağlı olarak değişim göstermiştir. Dozlar bakımından 0.5 ve 1 mM konsantrasyonları daha etkin bulunmuştur. Büyüme parametrelerinde, fotosentetik pigmentlerde, K ve Ca iyon içeriğindeki iyileştirmeler, nano-Si uygulamları sonrası bitkilerin tuzluluğa karşı toleransının artırılmasında etkili bulunmuştur.

TEŞEKKÜR

Bu çalışma Çankırı Karatekin Üniversitesi Bilimsel Araştırma Projeleri Koordinasyon Birimi tarafından KYO210621L09 numaralı proje ile desteklenmiştir.

KAYNAKLAR

Afzal M, Ahmad A, Alderfasi AA, Ghoneim A, Saqib M 2014. Physiological tolerance and cation accumulation of different genotypes of Capsicum annum under varying salinity stress. Proceedings of the International Academy of Ecology and Environmental Sciences, 4(1): 39-49.

Almodares A, Hadi MR, Dosti B 2008. The effects of salt stress on growth parameters and carbohydrates contents in sweet sorghum. Res. J. Environ. Sci., 2(4): 298-304.

Alsaeedi A, El-Ramady H, Alshaal T, El-Garawany M, Elhawat N, Al-Otaibi A 2019. Silica nanoparticles boost growth and productivity of cucumber under water deficit and salinity stresses by balancing nutrients uptake. Plant Physiology and Biochemistry, 139: 1-10.

Anonim 2022. FAOSTAT. https://www.fao.org/faostat/en/#data/QCL. Erişim Tarihi: 3.01.2022.

Avestan S, Ghasemnezhad M, Esfahani M,Byrt CS 2019. Application of nano-silicon dioxide improves salt stress tolerance in strawberry plants. Agronomy, 9(5): 246.

Bayram M, Dere S, Arpacı BB, Daşgan Y 2021. Sebzelerde çoklu stres faktörleri ve bitki reaksiyonları. In: Sebzelerde Stres Toleransı ve Islah Stratejileri. Ellialtıoğlu, Ş.Ş., Daşgan, H.Y., Kuşvuran, Ş. (eds.), Gece Kitaplığı, pp: 277-311, Ankara.

Daşgan HY, Koç S 2009. Evaluation of salt tolerance in common bean genotypes by ion regulation and searching for screening parameters. Journal of Food, Agriculture Environment, 7(2): 363-372.

El-Ramady H, Verma KK, Rajput VD, Minkina T, Elbehery F, Elbasiony H., Amer M 2022. Sources of silicon and nano-silicon in soils and plants. In: Silicon and Nano-silicon in Environmental Stress Management and Crop Quality Improvement. Etesamı, H., Al Saeedı, A., El-Ramady, H., Fujıta, M., Pessaraklı, M., Hossaın , M.A. (eds.), Academic Press, pp. 1-10, United Kingdom.

Farhangi-Abriz S, Torabian S 2018. Nano-silicon alters antioxidant activities of soybean seedlings under salt toxicity. Protoplasma, 255(3): 953-962.

Gong Z 2021. Plant abiotic stress: New insights into the factors that activate and modulate plant responses. J. Integr. Plant Biol., 63: 429.

Gözen V, Kusvuran S 2021. Düşük sıcaklık stresi. In: Sebzelerde Stres Toleransı ve Islah Stratejileri. Ellialtıoğlu, Ş.Ş., Daşgahn, H.Y., Kuşvuran, Ş. (eds.), Gece Kitaplığı, pp. 157-203, Ankara.

Haghighi M, Pessarakli M 2013. Influence of silicon and nano-silicon on salinity tolerance of cherry tomatoes (Solanum lycopersicum L.) at early growth stage. Scientia Horticulturae, 161: 111-117.

Ismail LM, Soliman MI, Abd El-Aziz MH, Abdel-Aziz HM 2022. Impact of silica ions and nano silica on growth and productivity of pea plants under salinity stress. Plants, 11(4): 494.

Kalteh M, Alipour ZT, Ashraf S, Marashi Aliabadi M,Falah Nosratabadi A 2018. Effect of silica nanoparticles on basil (Ocimum basilicum) under salinity stress. Journal of Chemical Health Risks, 4(3).

Kaouther Z, Nina H, Rezwan A, Cherif H 2013. Evaluation of salt tolerance (NaCl) in Tunisian chili pepper (Capsicum frutescens L.) on growth, mineral analysis and solutes synthesis. Journal of Stress Physiology and Biochemistry, 9(1): 209-228.

Kusvuran S, Kıran S, Altuntas O 2021. Influence of salt stress on different pepper genotypes: ıon homeostasis, antioxidant defense, and secondary metabolites. Global Journal of Botanical Science, 9: 14-20.

Liang Y, Sun W, Zhu YG, Christie P 2007. Mechanisms of silicon-mediated alleviation of abiotic stresses in higher plants: A review. Environmental Pollution, 147(2): 422-428.

Lutts S, Kinet JM,Bouharmont J 1996. NaCl-Induced senesence in leaves of rice (Oryza sativa L.) cultivars differing in salinity resistance. Ann. Bot., 78: 389-398.

Merwad ARM, Desoky ESM, Rady MM 2018. Response of water deficit-stressed Vigna unguiculata performances to silicon, proline or methionine foliar application. Scientia Horticulturae, 228: 132-144.

Navada S, Vadstein O, Gaumet F, Tveten AK, Spanu C, Mikkelsen O, Kolarevic J 2020. Biofilms remember: osmotic stress priming as a microbial management strategy for improving salinity acclimation in nitrifying biofilms. Water Research, 176: 115732.

Nielsen SS 2017. Sodium determination using ion-selective electrodes, Mohr titration, and test strips. In: Food analysis laboratory manual, Springer, pp. 161-170, Cham.

Pan T, Liu M, Kreslavski VD, Zharmukhamedov SK, Nie C, Yu M, Shabala S 2021. Non-stomatal limitation of photosynthesis by soil salinity. Critical Reviews in Environmental Science and Technology, 51(8): 791-825.

Rastogi A, Tripathi DK, Yadav S, Chauhan DK, Živčák M, Ghorbanpour M, Brestic M 2019. Application of silicon nanoparticles in agriculture. 3 Biotech, 9(3): 1-11.

Saxena R, Tomar RS,Kumar M 2016. Exploring nanobiotechnology to mitigate abiotic stress in crop plants. Journal of Pharmaceutical Sciences and Research, 8(9): 974.

Türkan İ, Bor M, Özdemir F, Koca H 2005. Differantial responses of lipid peroxidation and antioxidants in the leaves of droutght-tolerant P. Acutifolius gray and drought sensetive P. vulgaris L. subjected to polyethylene glycol mediates water stres. Plant Science, 168: 223-231.

Zhang Q, Dai W 2019. Plant response to salinity stress. In: Stress Physiology of Woody Plants, CRC Press, pp. 155-173.

Zhao S, Zhang Q, Liu M, Zhou H, Ma C, Wang P 2021. Regulation of plant responses to salt stress. International Journal of Molecular Sciences, 22(9): 4609.

DRAFT

ORAL PRESENTATION

Impedance Spectroscopy Analysis of an Electrochemical Cell Chamber for Diagnostics

Mehmet Yuksekkaya[1*] (*ORCID: https://orcid.org/0000-0002-2665-5799*), Utkan Demirci[2] (*ORCID: https://orcid.org/0000-0003-2784-1590*), Mustafa Kocakulak[3] (*ORCID: https://orcid.org/0000-0001-5029-0104)*

[*1]Ankara University, Faculty of Engineering, Department of Biomedical Engineering, Ankara, Türkiye.
[2]Stanford University, Canary Center for Cancer Early Detection, Department of Radiology, Bioacoustic MEMS in Medicine (BAMM) Labs, Stanford, USA.
[3]Izmir Democracy University, Faculty of Engineering, Department of Biomedical Engineering, Izmir, Türkiye.

*Corresponding author e-mail: myuksekkaya@ankara.edu.tr

Abstract
In biomedical diagnostics, electrochemical cells are important tools. This study explains the application of electrochemical cells in biomedical diagnosis, focusing on the modulation of impedance information at different frequencies as a function of cell fluid content. The relationship between the sample's composition and the chamber's reaction dynamics with impedance at distinct frequencies is examined. Also, a novel, practical and low-cost chamber design is presented.

To explain the underlying principles governing the electrochemical cell's behavior, an electrical equivalent circuit model for the chamber is proposed. The chamber's ionic fluid content for different concentrations is monitored through impedance spectroscopy. This study showed that electrochemical impedance spectroscopy can be used as economic and efficient tool for biomedical diagnostics with implications for a wide range of applications in the field of healthcare and life sciences.

Keywords: Impedance spectroscopy, microfluidics, biosensor, diagnosis, electrochemical chamber

INTRODUCTION

Electrochemical Cell

A basic Electrochemical impedance measurement setup consist of chamber with two electrodes in an electrolyte solution. The impedance for different alternating current (AC) signal with a constant frequency is dependent on several factor due to the solution dynamics and reactions. These factors include ion migration, the presence of free electrons in the solution, and the system's capacitive response. Electrical impedance spectroscopy is the impedance value for various frequencies. (Lasia, 2014) The resistivity of the bulk solution, permittivity of the solution, geometric electrode placement, electrode-solution interface, electrochemical processes occurring on the electrode and electrolyses have an effect on the impedance value in the two electrode systems. (Grimnes et al., 2005)

The data obtained from the EIS system is usually an impedance data for a specific frequency. An electrical equivalent model of an electrochemical system allows for more thorough data analysis, which may provide information about the structure of the materials in the electrochemical cell and the reactions formed in it.

Electrical Equivalent Model of a Cell

The design of the system, such as geometric placement of the electrodes and resistivity and permittivity features of the solution, makes bulk resistance and the capacitance of the model simply definable.

There is a relationship between resistivity and the quantity and mobility of the unbound electrons in a substance. Additionally, based on an electrical field applied to the substance, there is a relationship between permittivity and polarization. If the elements in the solution are changed, the resistivity and permittivity of the solution vary.

DRAFT

Different types of geometric configurations of two electrode systems exist, which are forward looking electrodes, co-planar electrodes and interdigitated electrodes. To reveal particular characteristics of electrical information, alternative electrode configurations may be utilized.

According to the description of the electrical equivalent circuit model, the experimental data and the equivalent circuit data must fit. The constituent parts of the electrical equivalent circuit model of the electrochemical cell should be evaluated based on an analysis and comprehension of the physical and chemical characteristics of the electrochemical system. (Krause, 2007)

In the electrical field, ion migration based on electrode charges leads to a double layer of charge on the electrode's surface, separated by a thick dielectric. This structure is called double layer capacitance (C_{DL}), which depends on frequency, and it is influenced by factors such as electrode potential, temperature, ion types, ionic concentrations, impurity adsorption, oxide layers, electrode roughness, electrode potential, and so on. (Luo, 2013 and Genry, 2017)

When surfaces are inhomogeneous, rough, or porous, which leads to observing frequency-dependent behaviour, since ideal capacitor model is suitable, constant phase element (*CPE*) is employed to model the actual double layer capacitance. α means phase coefficient, which represents the behaviour of CPE. (Hong et al., 2005 and Córdoba et al, 2015)

The ratio of the applied voltage to the current resulting from the charge transfer is defined as the charge transfer resistance (R_{CT}). The resulting current is influenced by the transferred ion concentration, potential formed during electrochemical reactions and reaction products. (Lvovich, 2012)

The impedance occurring because of the ion migration and diffusion in the diffusion layer stands for Warburg impedance (Z_W). It is strongly influenced by the frequency of the applied voltage. Higher frequencies result in lower Z_W because of faster ion migration or vice versa for lower frequencies. (Lvovich, 2012)

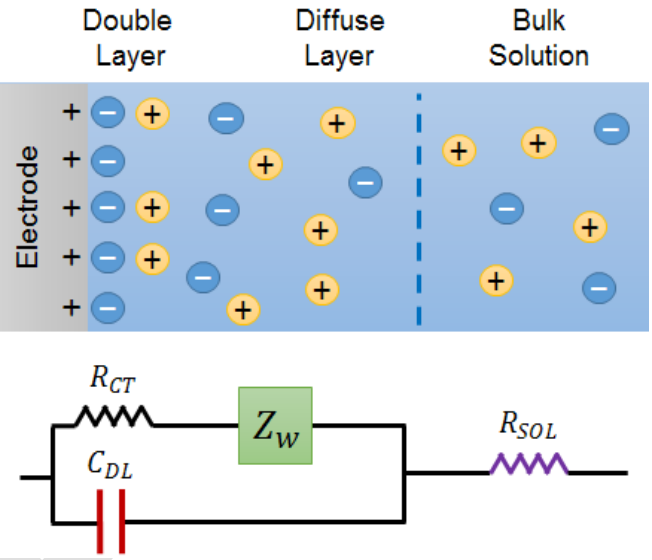


Figure 1. Randles electrical equivalent circuit model for an electrode. R_{CT} is the charge transfer resistor, Z_W is Warburg impedance, C_{DL} is the double layer capacitance, and R_{sol} is bulk resistance. R_{CT} and C_{DL} are related to the electrode-electrolyte interface and the reactions in that area that include the linear, semi-infinite diffusion of electroactive particles between electrode and electrolyte.

The resistance of the bulk solution can be easily clarified for higher frequencies in Bode plots of Randles electrical equivalent circuit model. In Nyquist plot, the imaginary versus real components of the impedance value graph can be plotted for the Randles equivalent circuit model, and the resulting graph includes information about both phase and magnitude. Nyquist and Bode plots provide the best visualization and description of the Randles model. (Randviir and Banks, 2013)

Lower frequencies have a linkage with the diffusion limited process, and high frequencies have a linkage with the electron transfer limited process. At high frequencies, R_{sol} value is obtained. Warburg impedance increases its visibility only at lower frequencies. On inhomogeneous, rough, or porous surfaces, CPE replaces C_{DL}. (Ahmed et al., 2014)

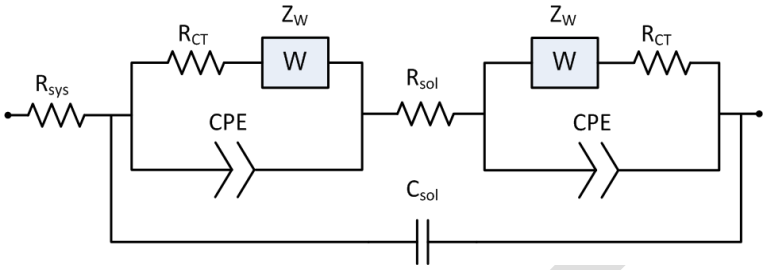


Figure 2. Randles electrical equivalent circuit model for full electrochemical system with two identical electrodes. R_{sys} is ohmic resistance of electrical wiring and C_{sol} is bulk capacitance.

In Figure 2 C_{sol} is significant. The impedance is determined predominantly by a co-planar electrode system's capacitance, which relies on material, geometry, and permittivity of solution. The capacitance value has a big effect on system's impedance, regardless of whether the capacitance value is high or low.

Impedance Spectroscopy

Electrochemical biosensors behave as a detector that can include EIS, and they have three different types according to their sensing method: impedimetric, amperometric, and potentiometric. Besides all these techniques that may contain EIS, impedimetric one mainly uses it. (Ahmed et al, 2014)

Biosensors based on EIS, which can be easily downsized, rapid, sensitive, and affordable, can be used in studies to determine the structural features of the solution/material that influence the permittivity and conductivity, or electrochemical reactions such as enzyme-catalysed reactions or specific binding reactions of some species on the surface of modified electrodes or in the solution. (Bahadır and Sezgintürk, 2016)

Pathogen and cell detection are also possible. Studies by Cheng et al. (2007), Shafiee et al. (2013), Toner et al. (2010), and Demirci et al. (2014) regarding impedance spectroscopy-based cell and virus detection and tracking with cell and virus lysate have been found in the literature.

MATERIALS AND METHODS

Chamber Design

Polymer-based microfluid chips are well suited to move the impedance measurement processes forward. The design of the chips is made up of three layers; the bottom polymer layer with electrodes on it, the medium layer with double sided adhesive and a patterned channel, and the top polymer layer with inlet and outlet patterns.

Electrode Layer

The screen-printing technique is used to obtain the electrode layer and a silver paste mixture, which has highly conductive silver ink composition and is silicon adhesive, is applied on the flexible polymer surface. With a flexible hydrophobic polymer film from the protective cover film of double-sided adhesive (DSA), a mask pattern is formed. The pattern is created with the help of computer-aided design software, and a laser cutter is used. The mask is first positioned on a flexible hydrophilic polymer. On one side of the mask, the silver paste mixture is dropped. The mixture is spread on the mask by using a microscope glass slide as a squeegee. The open spaces are filled with the mixture.

After the screen-printing process, the mask is carefully taken away continued in baking at 80° for sixty minutes. The electrodes are ready at the end of the baking process. It is possible that at the end of the process, errors are unavoidable since the laser cutter cannot cut the polymer in its actual dimensions, and baking has also an effect on the size of the electrodes.

Channel layer

The channel layer, which is patterned on DSA and created by a CAD software, is to form a cavity for solution to be filled, and using constant volume of solution is crucial in impedance measurements.

Inlet layer
The inlet layer, which is designed in CAD software, is formed with the flexible hydrophilic polymer cut by the laser cutter. The chamber is assembled by removing the protective film from one side of DSA so that it is laminated.

In order to minimize errors, this process is repeated numerous times. There is no requirement to use complex and expensive devices during the process. The steps may be done in any environment.

Experimental Setup

Probes of LCR meter, which is a device to measure the impedance, are attached to the electrodes of the chip, the sample solution is added into the channels, and magnitude and phase of impedance of the solution is recorded. The input AC potential is 1 peak-to-peak voltage (V_{pp}) and AC frequency is between 100 HZ and 1 MHz. In this paper, the maximum difference value for impedance change is approximately 1 kHz, which means that the magnitude of impedance is 1 kHz.

RESULTS

Even if the main purpose of this paper is to detect analytes, known conductance-valued solutions are measured firstly in order to obtain better knowledge about the system and impedance results. 1% Triton 100-X solution in DI pure water is used as control solution. Six test sample solutions, which are prepared at 0.05%, 0.1%, 0.25%, 0.50%, 1.0%, and 5.0% PBS in DI pure water, exposure to a conductivity test by using a conductivity meter.

Figure 3 (a) shows that different solutions with different concentrations have different conductivity levels. The determination of the resistivity for test samples and the control solution is done by impedance spectra measurements at a frequency range between 100 Hz and 1 MHz.

All the test samples and the control solutions are tested according to the procedure. For a solution 3 FFChips are used, and all the sample volumes are same with each other. The average of three impedance magnitude spectra measurements for a solution is calculated and this step is applied for all other solutions. According to the results, the Nyquist plot in Figure 4 is sketched.

DISCUSSION

The definition of one of the electrical models of the FFChip was made, along with the background of the electrical impedance spectroscopy analysis system. DI and some concentrations of PBS solutions filled channel measurements were done. The results of these known-concentrated solutions demonstrated that the system is capable of detect even $k\Omega$ of differences.

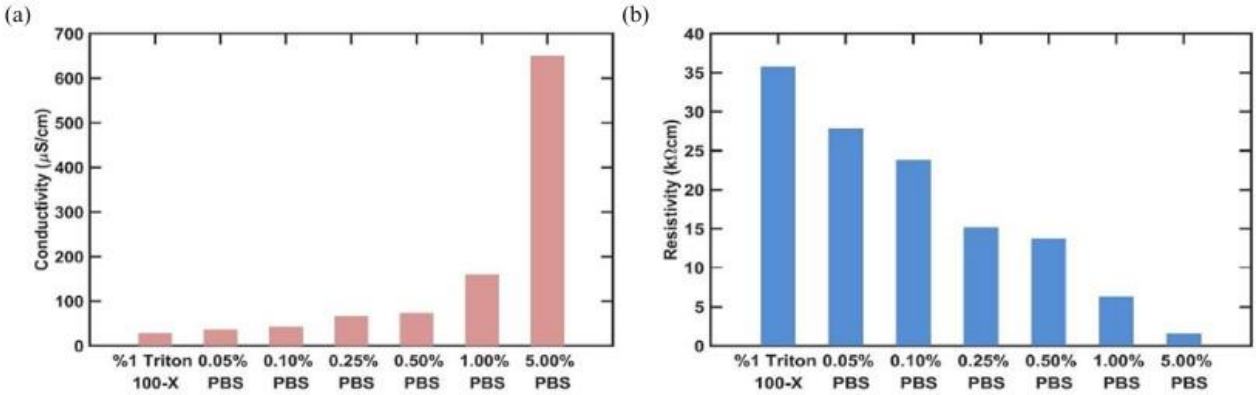


Figure 3. Test values a) Conductivity test values at various PBS concentrations b) Resistivity test values at various PBS concentrations.

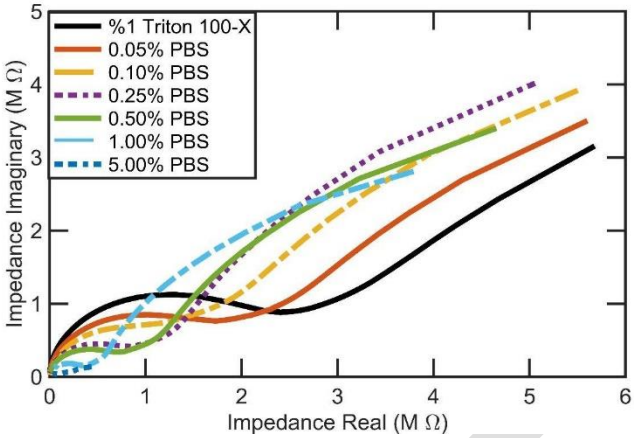


Figure 4. Nyquist plot of impedance values of control solution and different PBS concentration-solutions for a frequency range between 100 Hz and 1 MHz.

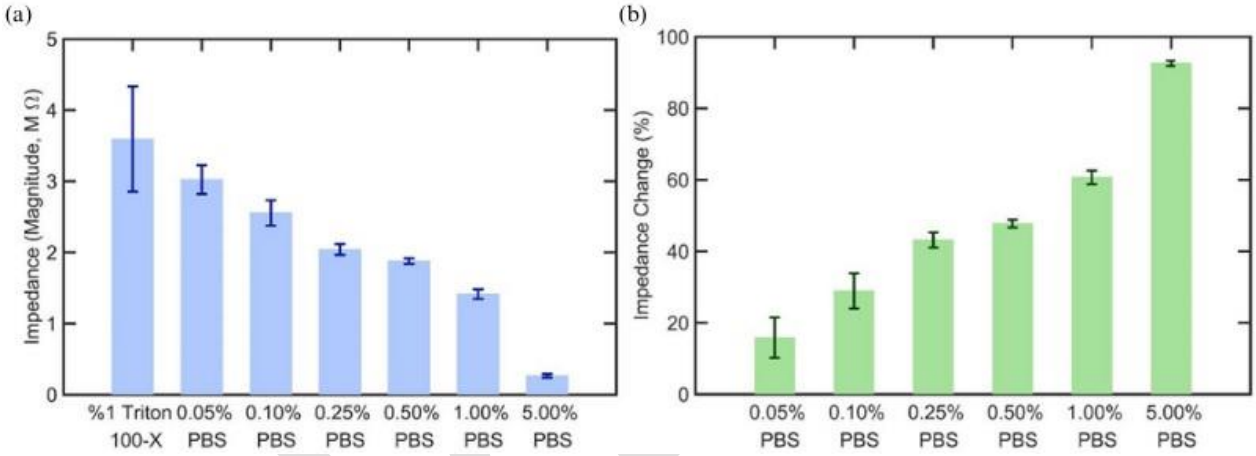


Figure 5. a) Impedance magnitude values of all the solutions at 1 kHz. Error bars represent standard deviations (STD) of the mean of impedance magnitude. (n=3), b) The impedance magnitude changes of all the sample test solutions with respect to average control value at 1 kHz. Error bars represent standard deviations (STD) of the mean of impedance magnitude. (n=3)

CONCLUSION

Lightweight, flexible, thin, affordable, easy to manufacture, disposable and mass-producible features make the flexible polymer film effective material for biosensors. With its portability, sensitivity, and rapid property, electrical impedance spectroscopy is efficacious in detection methodology.

ACKNOWLEDGMENTS

We would like to thank The Scientific and Technical Research Council of Turkey (TUBITAK) for providing financial support.

REFERENCES

Ahmed A., Rushworth J. V., Hirst N. A., Millner P. A. 2014. Biosensors for whole-cell bacterial detection. *Clinical microbiology reviews*, *27*(3), 631-646.

Bahadır E. B., Sezgintürk M. K. 2016. A review on impedimetric biosensors. *Artificial cells, nanomedicine, and biotechnology*, *44*(1), 248-262.

Cheng X., Liu Y. S., Irimia D., Demirci U., Yang L., Zamir,L., ..., Bashir R. 2007. Cell detection and counting through cell lysate impedance spectroscopy in microfluidic devices. *Lab on a Chip*, *7*(6), 746-755.

Córdoba-Torres P., Mesquita T. J., Nogueira R. P. 2015. Relationship between the origin of constant-phase element behavior in electrochemical impedance spectroscopy and electrode surface structure. *The Journal of Physical Chemistry C*, *119*(8), 4136-4147.

Demirci U., Shafiee H. 2014. *System and method for detecting pathogens* (U.S. Patent)

Gamry 2017. *Basics of EIS: Electrochemical Research-Impedance*. Available at: https://www.gamry.com/application-notes/EIS/basics-of-electrochemical-impedance-spectroscopy/ [17.09.23]

Grimnes S., Martinsen O. E. 2005. *Bioimpedance and Bioelectricity Basics*: Academic Press

Hong J., Yoon D. S., Kim S. K., Kim T. S., Kim S., Pak E. Y., No K. 2005. AC frequency characteristics of coplanar impedance sensors as design parameters. *Lab on a Chip*, *5*(3), 270-279.

Krause S. 2007. *Impedance Methods* in *Encyclopedia of Electrochemistry*, ed: Wiley-VCH Verlag GmbH & Co. KGaA

Lasia, A. 2014. Definition of impedance and impedance of electrical circuits. *Electrochemical Impedance Spectroscopy and its applications*, 7-66.

Luo X., Davis J. J. 2013. Electrical biosensors and the label free detection of protein disease biomarkers. *Chemical Society Reviews*, *42*(13), 5944-5962.

Lvovich V. F. 2012. *Impedance spectroscopy: applications to electrochemical and dielectric phenomena*. John Wiley & Sons.

Randviir E. P., Banks C. E. 2013. Electrochemical impedance spectroscopy: an overview of bioanalytical applications. *Analytical methods*, *5*(5), 1098-1115.

Shafiee H., Jahangir M., Inci F., Wang S., Willenbrecht R. B., Giguel F. F., ..., Demirci U. 2013. Acute on-chip hiv detection through label-free electrical sensing of viral nano-lysate. *Small*, *9*(15), 2553-2563.

Toner M., Bashir R., Cheng X, Demirci U., Irimia D., …, Rodriguez W. 2010. Methods for counting cells (USA Patent No: EP2156376 A1)

DRAFT

ORAL PRESENTATION

Doğal taşların bina enerji performansını destekleyebilirliği

Ünal Yılmaz[1*] (*ORCID: https://orcid.org/0000-0003-0058-6323*),
Figen Balo[2] (*ORCID: https://orcid.org/0000-0001-5886-730X)*

[*1]Asri Mühendislik, Malatya, Türkiye
[2] Fırat Üniversitesi, Mühendislik Fakültesi, Met. Ve Malzeme Mühendisliği, Elazığ, Türkiye

*Sorumlu yazar e-mail: unalylmz4444@gmail.com

Özet

Enerji darboğazı ve petrol sıkıntısı, araştırmacıları enerji kazancına katkı sağlayabilecek çalışmalara yöneltmiştir. Özellikle enerjinin büyük bölümünün kullanıldığı inşaat sektöründe enerjinin çok daha ekonomik kullanılması için çalışmalar artarak devam etmektedir. Binalarda daha az enerji kullanımı, daha az çevre kirletici emisyon yayılması ve daha düşük maliyet anlamına geldiği için bina kabuğunda doğru malzemelerin seçilmesiyle ekolojik, ekonomik ve enerji açısından oldukça önemli faydalar elde edilebilir.

Her yörenin jeolojik yapısında oluşmuş doğal taşları özellikle oluştuğu yörede kullanmak amacıyla geliştirilen bu çalışmada Şırnak iklim şartlarında Şırnak ilinde çıkarılan kayrak taşının bina kabuğunda duvar kaplama malzemesi olarak kullanılması durumunda enerji performansına etkileri ANSYS paket programı üzerinden dış duvarda sıcaklık dağılımı yönünden detaylandırılmıştır. Çalışmada farklı yapı malzemeleri ve taş yünü yalıtım malzemesi kullanılarak oluşturulan alternatif senaryolar karşılaştırılmıştır. Daha enerji performanslı malzeme ve daha az yakıt kullanılmasıyla çevresel etkilerin azaltılması amaçlanmıştır. Bu çalışma ile farklı yörelerdeki yapı ve yalıtıma destek sağlayabilecek doğal taşların; yapı, yalıtım veya duvar kaplama amaçlı kullanılarak yapılacak analizlerle enerji tasarrufuna destek olabilirliği hakkında karar vericilere, müteahhitlere ve mühendislere fikir verebilecek örnek bir çalışma hedeflenmiştir.

Anahtar Kelimeler: Doğal taş, kayrak taşı, ANSYS paket programı, bina kabuğu, sıcaklık dağılımı

Ability of natural stones to support building energy performance

Abstract

Energy bottleneck and oil shortage have led researchers to work that can contribute to energy gain. Efforts are increasingly continuing to use energy more economically, especially in the construction sector, where most of the energy is used. Since less energy use in buildings means less pollutant emissions and lower costs, significant ecological, economic and energy benefits can be achieved by choosing the right materials for the building envelope.

In this study, which was developed with the aim of using the natural stones formed in the geological structure of each region, especially in the region where it was formed, the effects of the slate stone extracted in Şırnak province, in Şırnak climate conditions, on the energy performance in case of use as a wall covering material in the building shell were detailed in terms of temperature distribution on the outer wall via the ANSYS package program. In the study, alternative scenarios created by using different building materials and rock wool insulation material were compared. With this study, natural stones that can support the structure and insulation in different regions; An exemplary study is aimed to give an idea to decision makers, contractors and engineers about its ability to support energy saving with analyzes to be made by using it for building, insulation or wall cladding purposes.

Keywords: Natural stone, slate, ANSYS package program, building envelope, temperature distribution

DRAFT

GİRİŞ

Kayrak taşı diğer adlarıyla dam taşı, arduvaz, kayağan taşı, ince yapraklar şeklinde yarılabilen ya da ayrılabilen, killi ve ince taneli, mikalı, kuvarlı, kloritli, siyah, kül ve gök renginde rezervinden çıkarılan bir çeşit başkalaşım doğal taşıdır. Alçak sıcaklık ve basınçta oluşmaktadır. Çekme mukavemeti fazladır.

Başkalaşım işlemine uğramış ana özdek, genellikle tortul kayaç şeklindeki volkanik tozlar ve kumuda ihtiva edebilen ince parçacıklı bir çeşit kildir (shale-şeyl, çamurtaşı). Esas doğal taş, bazı durumlarda bazı bölümleri başkalaşıma uğramış olduğundan, yapısında mevcut tortul katmanlaşmayı ve ilk mineralleri belli alanlarında barındırabilir. Tortul şeklindeki kayalar bazı durumlarda, yarılmış yüzeylerde izlendiği gibi, birbirini takip eden şerit yapısında katman katman bir yapıya bürünebilir. Yarılma olayı, yeryüzeyinin derinliklerinde uzun süre gömülü kalan taşın üzerine yapılan baskıdan dolayı ortaya çıkar. Bu sebeple, rezerv oluşumu sırasında sıkışık şekilde kıvrımlaşan daha yeni kısımlarda de yer yer gözükmekle beraber, kayrak taşı genellikle daha eski taşlar arasında yerleşir. Yarılmanın yönü, başkalaşım olayı esnasında kayrak taşına etki eden gerilimin yönüne göre değişir. Orijinal Kayrak taşı, katmanlaşmanın yönü boyunca ayrılmaz, yönlenmeyi olabildiğince fazla açıyla kesen yarılma yönünde ayrılır. Kayrak taşı, az-dereceli yerel meta-morfizma ile volkanik kül veya kil tarafınfan meydana çıkan orijinal bir şeyl taşı çeşidi olduğu için tortul taş şeklinde oluşan ince tanecikli, homojen, yapraklanmış meta-morfik taş yapısında iri tanecikli yapraklanan kısım, sadece meta-morfik sıkışmanın yönlenmesine dik olan düzlemlerde meydana gelir. Güçlü şekilde yapraklanma elde edilmişse "slaty bölünme" gerçekleşmiş olur. Ayrılmış tabakalar, pürüzlü veya düzgün yapıda olabilir (Çelik ve MY, 2003). Kayrak taşı geçirmiş oldukları meta-morfizma ile klivaj yapıda olurlarsa yapıları daha düzgün tabakalar şeklinde ayrışabilir. Kayrak taşı gözenekli bir yapıda bulunmaz. Aşınmalara ve atmosferik etkilere karşı oldukça dayanıklı olduğundan eğilme veya büzülme yapmaz (Çelik, ve ark., 2001). İşlenme sırasında yapraklanmış kısım yönünde vurmak suretiyle zemin karoları ve uzun çatı vb alanlar için kullanılabilir doğal taşın düz ve pürüzsüz levhalar elde edilecek şekilde biçimlendirilmesiyle yapı sektöründe kullanıma sunulur. Yuvarlak şekilde geniş yüzeyli yapıdada olabilir. Kayrak taşı çatılar için kaplama malzemesi olarak da yaygın şekilde kullanılmıştır. Çatı üzerinde kullanılan kayrak taşının birkaç yüzyıl boyumca bozulmadan kaldığı tarihi kayıtlarda bulunmuştur. Sadece kalsit ve pirit içerdiği durumlarda biraz daha çabuk bozulur ve ortaya çıkan H_2SO_4 çatı montajında kullanılan çivilerin paslanmasına sebep olduğundan çatı kaplamada kullanılan malzemeyi boşa çıkarabilir (Devlet Planlama Teşkilatı, 2001).

Kayrak taşının homojen yapıda, oldukça sert, fazla ısıya dayanımlı ve su geçirmeyen bir yapısı vardır. Kayrak taşının ısı yalıtım kabiliyeti yüksektir (Taşlıgil, ve ark., 2016).. Bu nedenle duvar kaplama malzemesi veya yapı malzemesi olarak kullanıldığında bina yalıtımına katkıda bulunur. Kayrak taşı yardımıyla binanın ısı yalıtımı ile sağlananı avantajlar bu taşla yapılan uygulamaların sürdürülebilirliği desteklemesi bakımından önemli bir referanstır. Aynı şekilde kayrak taşı hemen hemen her çeşit iklim şartlarının olumsuz etkilerine oldukça fazla direnç gösterebildiği için ve bina kabuğunun dış tabakasının yıpranmasını azalttığı içinde sürdürülebilirliğe önemli katkı sağlamaktadır (https://ardesiaslate.com.tr/bina-dis-cephe-kaplama).
Kayrak taşından ayrıca yalıtım özellikli mineral yünü de elde edilebilir. Rezervinden çıkarıldığı şekliyle doğal olarak kullanılabildiği gibi kesilip parlatarak da kullanılabilir. Arduvaz taşının artık parçaları katkı ve dolgu malzemesi olarak bazı endüstriyel alanlarında kullanılabilir. Genleştirilen sleyt düşük ağırlıklı yapı malzemesi olarak kullanılabilir toz ve pul sleyt taşından hem plastik-lastik hem de asfalt agregası ayrıca ilaç sektöründe dolgu malzemesi üretiminde faydalanılır (Aytekin, 2007).

Kayrak taşında gözlenen renk genellikle gri renktir. Bununla birlikte, kayrak taşı sadece bir bölgeden bile farklı renklerde çıkarılabilir. Mesela, Dünyaya baktığımızda Kuzey Galler'de çıkarılan kayrak taşı genellikle soluk ama koyuca bir griliktedir. Bununla birlikte içinde birçok farklı tonu barındırabildiğinden mavi, yeşil veya mor renklide çıkan bölümlerinde renk zenginliğini sağlayabilir. Yapısında organik madde fazla ise daha çok koyu gri veya gri renkte olabilir. Kayrak taşı, ince bir karbonat taş, ilksel taş tabaka ardalanması içeriyorsa mavimtırak bir gri renk alabilir. Günlenme maruziyeti varsa kahverengimsi, kızılımsı renklerin baskın olduğu bir renkte olabilir. Kayrağın bazen yüzey kısmında siyah renkli bulunan dentirik yapı taşa fosil görünüşü verir (Hudson, 1972).

Soykan andezit, mermer ve arduvaz taşları ile polimer beton üreterek ve yaptığı deneylerde arduvaz içerikli malzeme en yüksek basma mukavemetini en düşük aşınma kaybını veren malzeme olduğunu belirtmiştir (Soykan, ve ark., 2015).Yine Soykan ve ark. andezit ve arduvazın beton agregası olarak kullanıma uygunluğunu araştırmışlardır (Soykan, ve ark., 2015).. İnebolu'da tarihi yapıların çatı kaplama malzemeleri üzerine yaptığı çalışmada yalnızca kayrak taşının kullanıldığını belirtilmiştir (Halaç ve Dağlı 2022). Abay yine aynı bölgede çatı kaplama malzemesi olarak kayrak (yöresel adıyla marla) taşının kullanıldığını rapor etmiştir

(Abay, 2019). Yılmaz ve arkadaşları tüf taşlarının, Balo ve ark Limra taşının duvar kaplama malzemesi olarak kullanıldığında duvar enerji performansına etkilerini araştırmışlardır (Balo, ve Sua, 2018-Yılmaz ve ark., 2019).

Türkiye'de geniş bir yayılıma sahip olan Kayrak taşı önemli değerlendirilebilir nitelikte potansiyele sahiptir. Kayrak taşı Türkiye'de Eskişehir, İzmir, Denizli gibi şehirlerde ve en fazla İç Anadolu bölgesinde olmak üzere Akdeniz, Karadeniz ve Ege gibi farklı bölgelerde işlenmektedir. Kastamonu - Zonguldak arasında, Muğla, Bodrum ve Başkale-Hakkâri yolunda Türkiye'deki en çok rezerv alanı bulunmaktadır (Taşlıgil ve Şahin, 2016)

Kayrak taşı'nın üretimi Dünya'da birçok bölgede yapılmaktadır. Kanada, Hindistan, İngiltere, Amerika Birleşik Devletleri, Çin, Brezilya, Tayvan, kayrak üretimi yapan ülkelerin başında yer almaktadır. Dünya'nın en büyük rezervlerinden bir tanesi Rajasthan (Hindistan) bölgesindedir. Ayrıca İngiltere'de Galler, İtalya'da Lavanga, Almanya'da Bavyera'nın kuzeyi, Portekiz'de Valongo, Dünya'nın lider rezervlerine sahiptir. Amerika ve Avrupa ülkelerinde özellikle tarihi yapılarda çatı kaplama için eskiden beri kiremit yerine tercih edilmiştir. Türkiye'nin önemli firmalarından birinin yaptığı araştırmaya göre, her yıl Dünyada yaklaşık 30 milyon m²'lik çatı kaplama işleminin yapıldığı bve bunların. %5'i Amerika'da, %92'si Avrupa'da geri kalan kısmının ise başka bölgelerde yapıldığı belirtilmiştir. Kayrak taşını en fazla değerlendiren Avrupa ülkelerinin %43 oranla Fransa, %17 ile Almanya, %18 ile İspanya, %11 ile Büyük Britanya olduğu rapor edilmiştir. Avrupa'da 400 yıldır kullanılmakta olan Kayrak taşı günümüzde de çatı kaplama malzemesi olarak çatılarda yer almaktadır (Taşlıgil ve Şahin, 2016)

Kayrak taşının duvar kaplama malzemesi amaçlı kullanılabilmesi için Türk standartlarında belirtilen minimum yoğunluk değerinin 2,55 gr/cm³ olması gerekmektedir. Uludere bölgesindeki binalarda kullanılmak üzere araştırılan Şırnak yöresi Kayrak taşının yoğunluğu 2.75 gr/cm³ civarındadır. Bu değer standartı karşılamaktadır (Sert, 2010).Kayrak taşının bazı uygulama örnekleri Şekil 1'de gösterilmiştir.



Şekil 1. Kayrak taşının uygulam örnekleri

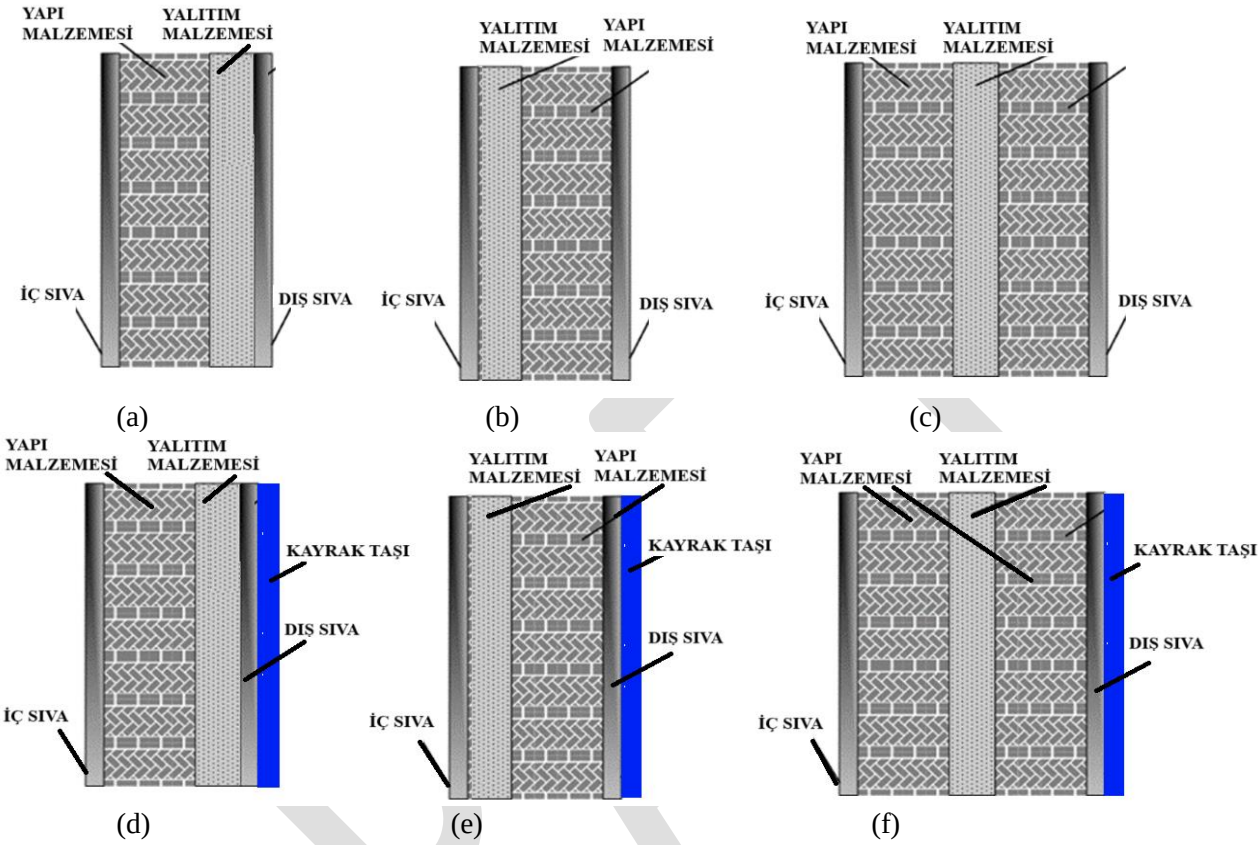
Bu araştırmada öncelikle Şırnak ilinde rezervi bulunan Kayrak taşına ait teknik özellikler ve kullanım alanları incelenmiştir. Daha sonra Uludere ilçesi iklim şartları için Kayrak taşının duvar kaplama malzemesi olarak kullanılması durumunda farklı duvar tipleri, farklı yapı malzemeleri ve taşyünü yalıtım ile tasarlanan bina kabukları için duvardaki sıcaklık dağılımı analizleri ANSYS yazılımı ile araştırılmıştır.

ANSYS yazılımı ile sıcaklık dağılımının analizi

Araştırma Türkiye ısı haritasında 1. Bölge üzerinde bulunan Şırnak Uludere ilçesi iklim koşulları altında yapılmıştır. Bu araştırmada öncelikle Şırnak ilinde rezervi bulunan Kayrak taşına ait teknik özellikler ve kullanım alanları incelenmiştir. Daha sonra Kayrak taşının, Uludere ilçesi iklim şartlarında bina tasarımlarında duvar kaplama malzemesi olarak kullanımı için farklı doğal içerikli yapı malzemeleri (ponza taşı içeren beton, gözeneksiz agrega içeren beton ve ahşap ürün içeren beton) ve piyasada en çok kullanılan yalıtım malzemelerinden biri olan taş yünü kullanılarak 18 farklı bina kabuğu tipi modellenmiştir. Modelleme için 3

faklı duvar yapısı (dıştan yalıtımlı, içten yalıtımlı ve sandviç) temel alınmıştır. Daha sonar ANSYS yazılımıyla sıcaklık dağılımlarına göre ısı geçişi analizleri ile sonuçlar değerlendirilmiştir.

Şekil 2'de dıştan-içten yalıtımlı duvar ve sanvdiç duvar önce duvar kaplama malzemesi kullanılmadan daha sonra dış duvarın kaplanmasında Kayrak taşının kullanılmasıyla oluşturulan duvar yapıları verilmiştir. Bina kabuğunu oluşturan bileşenler ve analizi yapılan alternatif tipler Tablo 1'de sunulmuştur. Dış duvar bileşenlerinin teknik özellikleri Tablo 2'de gösterilmiştir.



Şekil 2. Analizde faydalanılan duvar yapıları [(a) Duvar kaplama malzemesi kullanılmayan dıştan yalıtımlı duvar, (b) Duvar kaplama malzemesi kullanılmayan içten yalıtımlı duvar, (c) Duvar kaplama malzemesi kullanılmayan sanvdiç duvar, (d) Duvar kaplama malzemesi olarak Kayrak taşı kullanılan dıştan yalıtımlı duvar, (e) Duvar kaplama malzemesi olarak Kayrak taşı kullanılan içten yalıtımlı duvar, (f) Duvar kaplama malzemesi olarak Kayrak taşı kullanılan sandviç duvar]

Tüm duvar tipleri arasında en iyi performans, yapı malzemesi olarak pomza taşı içeren beton ve en dış katmanda Kayrak taşı kullanıldığı zaman tespit edilmiştir. En kötü performans ise yapı malzemesi olarak gözeneksiz agrega içeren beton en dış katmanda Kayrak taşı kullanılmadan oluşturulan alternatifle belirlenmiştir.

Tablo 1. Bina kabuğunu oluşturan bileşenler ve analizi yapılan alternatif tipler

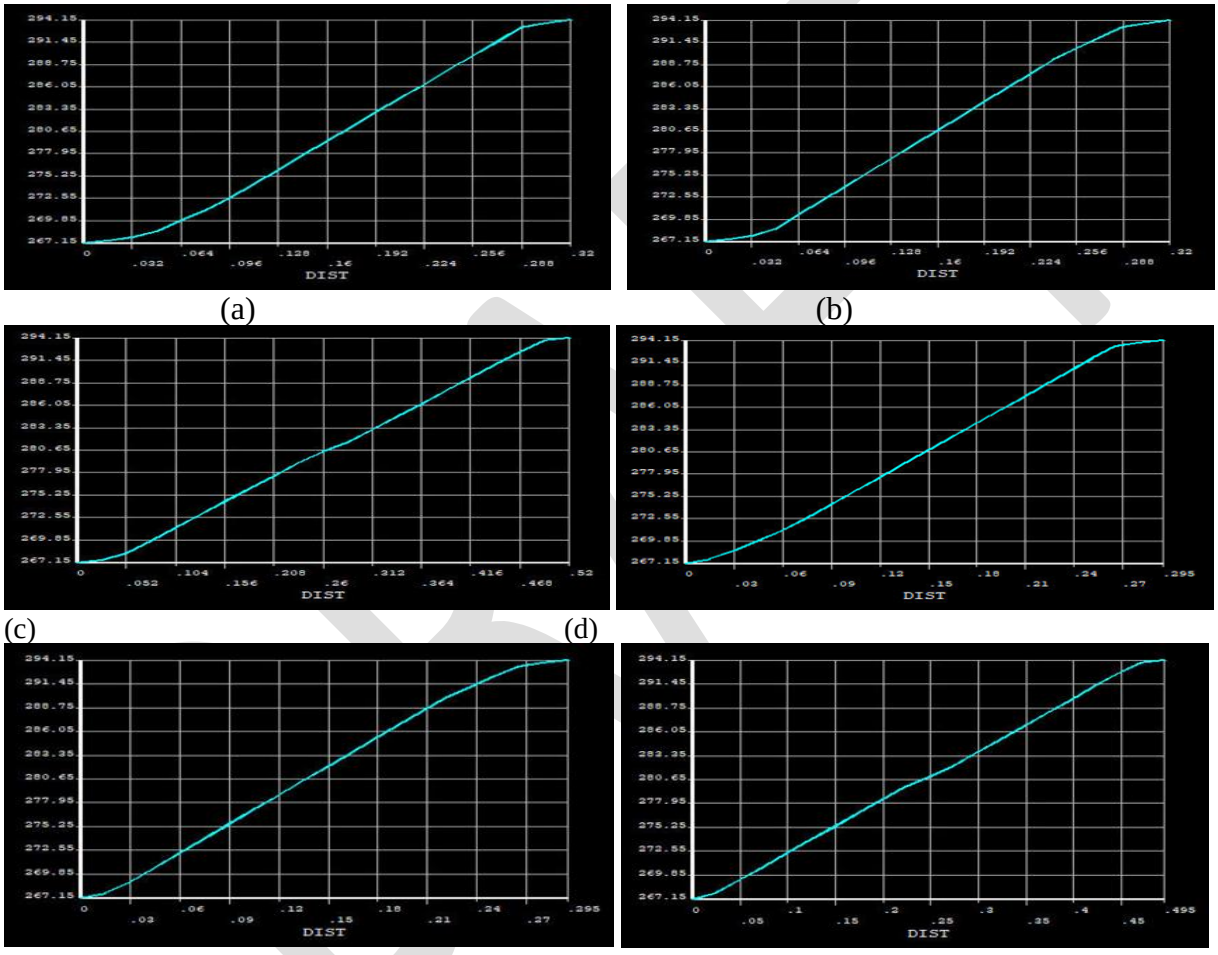
Tip	Duvar yapısı	Duvar Katmanları					
		Dış kaplama	Dış sıva	Yapı malzemesi	Yalıtım malzemesi	Yapı malzemesi	İç sıva
1	Dıştan yalıtımlı	yok	var	yok	Taş yünü	Ponza taşı içeren beton	var
2	Dıştan yalıtımlı	yok	var	yok	Taş yünü	Gözeneksiz agrega içeren beton	var
3	Dıştan yalıtımlı	yok	var	yok	Taş yünü	Ahşap ürün içeren beton	var
4	Dıştan yalıtımlı	Kayrak taşı	var	yok	Taş yünü	Ponza taşı içeren beton	var
5	Dıştan yalıtımlı	Kayrak taşı	var	yok	Taş yünü	Gözeneksiz agrega içeren beton	var
6	Dıştan yalıtımlı	Kayrak taşı	var	yok	Taş yünü	Ahşap ürün içeren beton	var
7	İçten yalıtımlı	yok	var	Ponza taşı içeren beton	Taş yünü	yok	var
8	İçten yalıtımlı	yok	var	Gözeneksiz agrega içeren beton	Taş yünü	yok	var
9	İçten yalıtımlı	yok	var	Ahşap ürün içeren beton	Taş yünü	yok	var
10	İçten yalıtımlı	Kayrak taşı	var	Ponza taşı içeren beton	Taş yünü	yok	var
11	İçten yalıtımlı	Kayrak taşı	var	Gözeneksiz agrega içeren beton	Taş yünü	yok	var
12	İçten yalıtımlı	Kayrak taşı	var	Ahşap ürün içeren beton	Taş yünü	yok	var
13	Sandviç duvar	yok	var	Ponza taşı içeren beton	Taş yünü	Ponza taşı içeren beton	var
14	Sandviç duvar	yok	var	Gözeneksiz agrega içeren beton	Taş yünü	Gözeneksiz agrega içeren beton	var
15	Sandviç duvar	yok	var	Ahşap ürün içeren beton	Taş yünü	Ahşap ürün içeren beton	var
16	Sandviç duvar	Kayrak taşı	var	Ponza taşı içeren beton	Taş yünü	Ponza taşı içeren beton	var
17	Sandviç duvar	Kayrak taşı	var	Gözeneksiz agrega içeren beton	Taş yünü	Gözeneksiz agrega içeren beton	var
18	Sandviç duvar	Kayrak taşı	var	Ahşap ürün içeren beton	Taş yünü	Ahşap ürün içeren beton	var

Şırnak Uludere ilçesimde bina dış duvarının sıcaklık dağılımlarina ait grafiksel görünümlerde en peformanslı duvar alternatifleri Şekil 3'de gösterilmiştir. Bina dış duvarında en kötü performansı veren duvar yapılarına göre ağ görünümleri Şekil 4'de gösterilmiştir. Yapı malzemesi olarak ahşap ürün içeren beton kullanıldığında en iyi ve en kötü performanslı duvarların vektörel görünümleri Şekil 5'da verilmiştir.

DRAFT

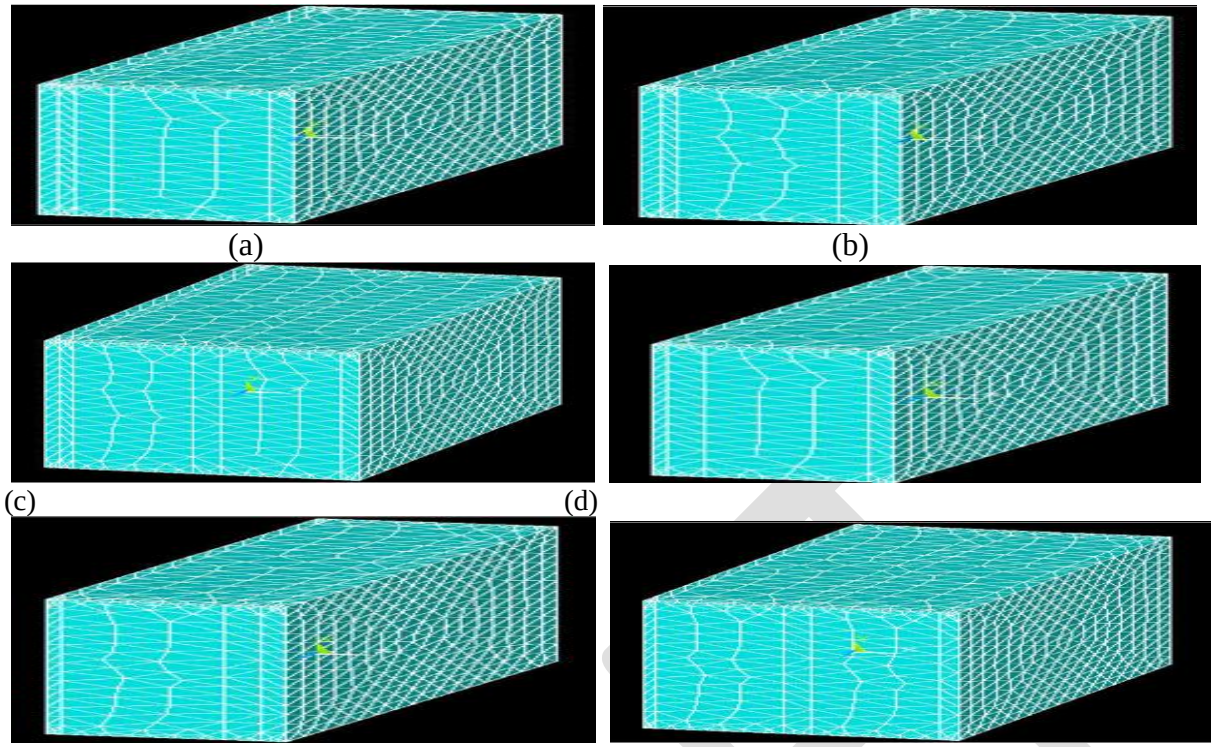
Tablo 2. Dış duvar bileşenlerinin teknik özellikleri [15]

Malzemeler	**Duvar kaplama**		**Yapı**			**Yalıtım**	
	Kayrak taşı	Dış sıva	Ponza taşı içeren beton	Gözeneksiz agrega içeren beton	Ahşap ürün içeren beton	Taşyünü	İç sıva
Kalınlık (m)	0,025	0,02	0,20	0,20	0,20	0,045	0,03
Isıl iletkenlik katsayısı (W/mK)	1. 69	0,87	0,29	0,64	0,41	0,39	1,40
Yoğunluk (g/cm^3)	2.750	1.400	0.800	1.500	0.800	0,034	2.000



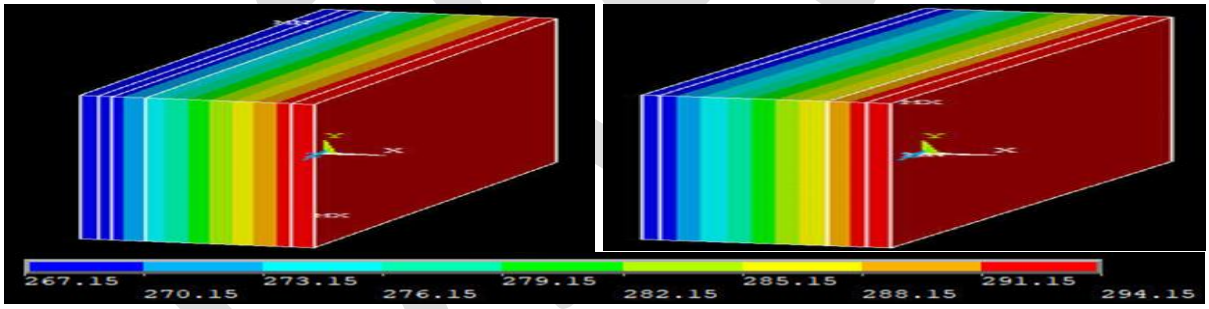
Şekil 3. Uludere ilçesinde grafiksel görünüm ile en peformanslı duvar alternatiflerin sıcaklık dağılımı [(a) En iyi performans veren Kayrak taşı kullanılan dıştan yalıtımlı duvar için (Tip 4), (b) En iyi performans veren Kayrak taşı kullanılan içten yalıtımlı duvar için (Tip 10), (c) En iyi performans veren Kayrak taşı kullanılan sandviç yalıtımlı duvar için (Tip 16), (d) En iyi performans veren Kayrak taşı kullanılmayan dıştan yalıtımlı duvar için (Tip 1), (e) En iyi performans veren Kayrak taşı kullanılmayan içten yalıtımlı duvar için (Tip 7), (f) En iyi performans veren Kayrak taşı kullanılmayan sandviç duvar için (Tip 13)]

Bu araştırmada, Türkiye'de birinci ısı bölgesinden Uludere ilçesi iklim şartları esas alınarak bina kabuğunda Şırnak Kayrak taşının kaplama elemanı olarak kullanılması halinde enerji geçişine etkileri sıcaklık dağılımları ile incelenmiştir. Analizlerde ANSYS yazılımından faydalanılmıştır. Analizlerin değerlendirilmesiyle ponza taşı agregalı beton ve Kayrak taşı kombinasyonun enerji geçişinde sıcaklık dağılımları açısından en iyi performansı gösterdiği gözlenmiştir.



(a) (b) (c) (d) (e) (f)

Şekil 4. Uludere ilçesinde ağ görünümlerle en kötü peformanslı duvar alternatiflerin sıcaklık dağılımı [(a) En kötü performans veren Kayrak taşı kullanılan dıştan yalıtımlı duvar için (Tip 5), (b) En kötü performans veren Kayrak taşı kullanılan içten yalıtımlı duvar için (Tip 11), (c) En kötü performans veren Kayrak taşı kullanılan sandviç yalıtımlı duvar için (Tip 17), (d) En kötü performans veren Kayrak taşı kullanılmayan dıştan yalıtımlı duvar için (Tip 2), (e) En kötü performans veren Kayrak taşı kullanılmayan içten yalıtımlı duvar için (Tip 8), (f) En kötü performans veren Kayrak taşı kullanılmayan sandviç duvar için (Tip 14)]



(a) (b)

Şekil 5. Uludere ilçesinde vektörel görünümlerle yapı malzemesi ahşap ürün içeren beton kullanılan du en iyi ve en kötü peformanslı duvar alternatiflerin sıcaklık dağılımı [(a) En iyi performans veren Kayrak taşı kullanılan dıştan yalıtımlı duvar için (Tip 6) (b) En kötü performans veren Kayrak taşı kullanılmayan içten yalıtımlı duvar için (Tip 9)]

Kayrak taşı dış kaplama bileşeni olarak kullanılmadan ve yapı malzemesi gözeneksiz agregalı beton kullanıldığında enerji etkinliğinde sıcaklık dağılımı açısından minimum katkı belirlenmiştir. Kayrak taşı ile en iyi performansı veren duvar yapısının dıştan yalıtımlı duvar olduğu görülmüştür. Tüm alternatifler değerlendirildiğinde Kayrak taşının bina kabuğunun enerji performansını arttırmaya destek olabileceği tespit edilmiştir. Duvar alternatiflerinde enerji performansını artması sebebiyle enerji kullanımı, salınan emisyon miktarı ve enerji maliyeti azaltacaktır. Petrol temelli yapı ve kaplama malzemelerinin yerine daha doğal içerikli malzemelerin kullanılması insan sağlığına, ekolojiye, ve hem ülkeye hem de araştırılan ilçeye enerji ve ekonomi açısından faydalar sağlayacaktır.

SONUÇ

Bu çalışma ile Uludere ilçesinde duvar kaplama malzemesi olarak Kayrak taşı kullanımının özellikle pomza taşı agregalı beton ve ilçe için tespit edilmiş optimum kalınlıkta kullanılan taş yünü kominasyonuyla en iyi performansın dıştan yalıtımlı duvarda elde edildiği görülmüştür. Bölgede yeni tasarlanacak binaların doğal malzeme seçimi ile yeşil bina konseptine yaklaştırma açısından örnek bir çalışma olarak tasarlanan bu çalışmann, müteaahhitlere, mühendislere ve karar vericilere yapılan ANSYS analizi desteğinde fikir vermesi amaçlanmıştır.

KAYNAKLAR

Abay A,. (2019). Kentsel Alanların Korunması Bağlamında İnebolu Geleneksel Konut Mimarisi Değerlendirmesi (Yüksek lisans tezi).

Aytekin, R. (2007). *Bodrum Yalıçiftlik Beldesi Alazeytin Köyü Kayrak Taşlarının Fiziko – Mekanik Özellikleri ve Değerlendirilebilirliği* (Yüksek lisans tezi).

Balo, F.Sua, L, Enerji-ekoloji-ekonomi açısından duvar kaplama malzemesi olarak limra taşı), 3rd Internatıonal Bozok Symposıum, 3-5 May 2018, Yozgat/Turkey

Construction Materials & Local Market Survey In Palestinian Territories, August 2022

Çelik, MY (2003a). Dekoratif Doğal Yapı Taşlarının Kullanım Alanları ve Çeşitleri. *Bilimsel Madencilik Dergisi, 42 (1),* 3-15.

Çelik, MY ve Kavuşan, G. (2001). Doğal taş ve mermerlere uygulanan yüzey şekillendirme teknikleri. *4. Endüstriyel Hammaddeler Sempozyumu*, 77-86.

Devlet Planlama Teşkilatı, Endüstriyel Hammaddeler Alt Komisyonu Çalışma Grubu. Sekizinci Beş Yıllık Kalkınma Planı Madencilik Özel İhtisas Komisyonu Raporu Yapı Malzemeleri II. (2001).

Halaç HH, Dağlı H, Marla Taşının Yapı Malzemesi Olarak Kullanımı; İnebolu'da Çatı Malzemesi Olarak Kullanım Örneği, Dokuz Eylül Üniversitesi Sosyal Bilimler Enstitüsü Dergisi, 24(1), 147-166, 2022

https://ardesiaslate.com.tr/bina-dis-cephe-kaplama

Hudson, K (1972). Yapı Malzemeleri; "Bölüm 2: Taş ve Kayrak". 14-27. ISBN 0-582-12791-2.

Sert M, Isparta ve Nevşehir Yöresi Volkanik Kökenli Taşların Fiziko Mekanik Özelliklerinin Belirlenerek Kullanım Alanlarının İrdelenmesi, Afyon Kocatepe Üniversitesi, Thesis · January 2010

Soykan, O, Özel, C., Öcal, C. (2015). Arduvaz ve Andezit'in Beton Agregası Olarak Kullanılabilirliğinin Araştırılması. *Süleyman Demirel Üniversitesi Fen Bilimleri Enstitüsü Dergisi*, 19(1), 69-74.

Yılmaz U, Sua L, Balo F, 3E methodology for tuff stones as wall coating material, 3rd World Conference on Technology Innovation and Entrepreneurship "Industry 4.0 Focused Innovation, Technology, Entrepreneurship and Manufacture" June 21-23, 2019, Istanbul

Taşlıgil, N. ve Şahin, G. (2016). Yapı Malzemesi Olarak Kullanılan Türkiye Doğal Taşlarının İktisadi Coğrafya Odağında Analizi. Marmara Coğrafya Dergisi, (33), 607-640.

DRAFT

ORAL PRESENTATION

Cryptodrassus ve *Drassyllus* (Araneae: Gnaphosidae) cinsi yer örümceklerinin seta morfolojilerinin karşılaştırılması

Durmuş OK[1,2] (https://orcid.org/0000-0001-8784-6356), Osman SEYYAR[1](https://orcid.org/0000-0002-0920-7943), Hakan DEMİR[1]

[1]Niğde Ömer Halisdemir Üniversitesi, Fen-Edebiyat Fakültesi, Biyoloji Bölümü, Niğde, Türkiye
[1,2]Nevşehir Hacı Bektaş Veli Üniversitesi, Avanos MYO, Bitkisel ve Hayvansal Üretim Bölümü, Nevşehir, Türkiye

Sorumlu Yazarı E-mail: durmusok@nevsehir.edu.tr

Özet

Örümceklerin vücutlarının dış kısmı seta adını verdiğimiz kıl benzeri yapılarla kaplıdır. Yer örümceklerinde bu yapıların cins düzeyinde farklılıklar gösterdiği bilinmektedir. Bu çalışmada, Erciyes Dağından toplanmış olan 2 cinse ait 2 türün seta morfolojileri çalışılmıştır *Cryptodrassus creticus* (Chatzaki, 2002) ve *Drassyllus praeficus* (L. Koch, 1866) türlerinin genelikle abdomen ve sefalotoraks gibi vücut kısımları üzerinde yer alan setaların morfolojileri Scanning Electron Microscopy (SEM) kullanılarak belirlenmeye çalışılmıştır.

Anahtar Kelimeler: Gnaphosidae, Seta, Erciyes Dağı, Örümcek, Araneae

Comparison of seta morphology of ground spiders of the genera *Cryptodrassus* and *Drassyllus* (Araneae: Gnaphosidae)

Abstract

Spiders cuticle is covered with hair-like structures that called setae. It is known that these structures differ at the genus level in ground spiders. In this study, the seta morphologies of 2 species belonging to 2 genera collected from Erciyes Mountain were studied. The morphologies of the setae of *Cryptodrassus creticus* Chatzaki, 2002 and *Drassyllus praeficus* (L. Koch, 1866), which are generally located on body parts such as abdomen and cephalothorax, were studied using Scanning Electron Microscopy (SEM) has been attempted to be determined.

Keywords: Gnaphosidae, Seta, Erciyes mountain, Spider, Araneae

Giriş

Gnaphosidae familyası hem cins hem de tür sayısı bakımından örümceklerin en zengin gruplarındandır. Örümcekler, Dünya'da 132 familya, 4313 cins, 51051 tür (World Spider Catalog, 2023), ülkemizde ise 55 familya, 370 cins, 1255 tür ile temsil edilmektedirler (Demir ve Seyyar, 2017; Danışman vd., 2023).

Gnafozidler genellikle 1–15 mm uzunluğunda, bacakları iki 2 tırnaklı, genellikle desenlenme göstermeyen örümceklerdir. Genellikle toprakta, taş altlarında, ağaç kabuklarında, kaya çatlaklarında veya kurumuş yaprak döküntüleri arasında yaşarlar ve buralarda tüp şeklinde ağlar örerler.

Tüm yer örümceklerinin vücutlarının üst kısmı seta adı verilen kıllarla kaplıdır. Yapılan çalışmalar incelendiğinde yer örümceklerinin vücut örtüsü üzerinde 10 farklı seta tipine rastlanılmıştır (Zakharov, Ovtsharenko, 2015). Setalar, yer örümceklerinin vücutlarının farklı kısımlarında değişik görevleri yerine getirmek üzere farklılaşmıştır. Örümcekler tarafından genellikle duyu organı ve mekanoreseptör olarak kullanılırlar. Vücut üzerinde abdomenin tüm kısımlarında, cephotrax üzerinde, bacaklar, pedipalp ve spineretler üzerinde yoğun bir şekilde bulunurlar. Örümceklerin bazı gruplarında vücut örtüsünü oluşturan bu

setaların farklı morfolojilerde olduğu ve örümcek sınıflandırmasında önemli olabileceği belirtilmiştir (Lehtinen 1967, 1975a,b).Gnaphozidler üzerine yapılan çalışmalar cins seviyesinde setaların morfolojik yapılarının fgarklı olduğunu göstermektedir. Salticidae ve Oxyopidae familyalarında da seta morfolojilerindeki farklılığın cins hatta tür ayrımında kullanılabilirliği ile ilgili birkaç çalışma yapılmıştır (Hill 1979; Townsend ve Felgenhauer, 2001).

Bu çalışmanın amacı, Gnaphosidae familyasına ait olan 2 cinse ait 2 türün seta morfolojilerini SEM ile belirlemek ve setaların bu cinslerin yer örümcekleri sınıflandırmasında kullanılabilirliğini ortaya koyabilmektir.

Materyal ve Metot

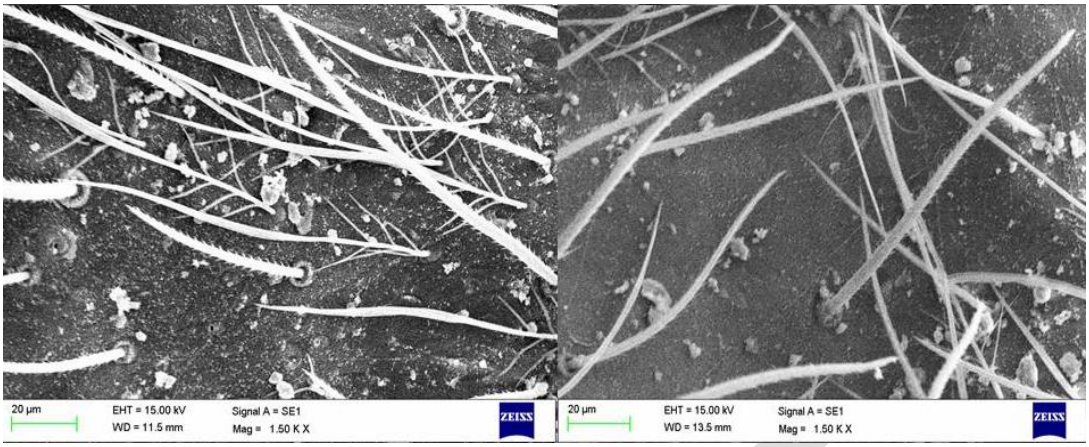
Yer örümceklerine ait örnekler Erciyes dağında (Kayseri) yapılan arazi çalışmaları sonucunda toplanmıştır. Teşhisi yapılan türlerin seta morfolojilerini belirlemek üzere Niğde Ömer Halisdemir Üniversitesi Merkezi Araştırma Laboratuvarında bulunan Elektron Mikroskobu kullanıldı. İki gnaphozid cinsine ait *Cryptodrassus creticus* (Chatzaki, 2002) ve *Drassyllus praeficus* (L. Koch, 1866) türlerinin prozoma, opistozoma ve bacakları ayıklandı. Her bir tür için bu vücut kısımları staplar üzerine düzgün konumda yerleştirildi ve daha net görüntü elde etmek için bu numunelerin yüzeyi Sputter Coater (Cressingto Auto 108) marka cihaz ile altınla kaplandı. Sonrasında yüzey morfolojisini incelemek için EVO LS 10 ZEISS marka cihaz kullanılarak fotoğrafları çekildi.



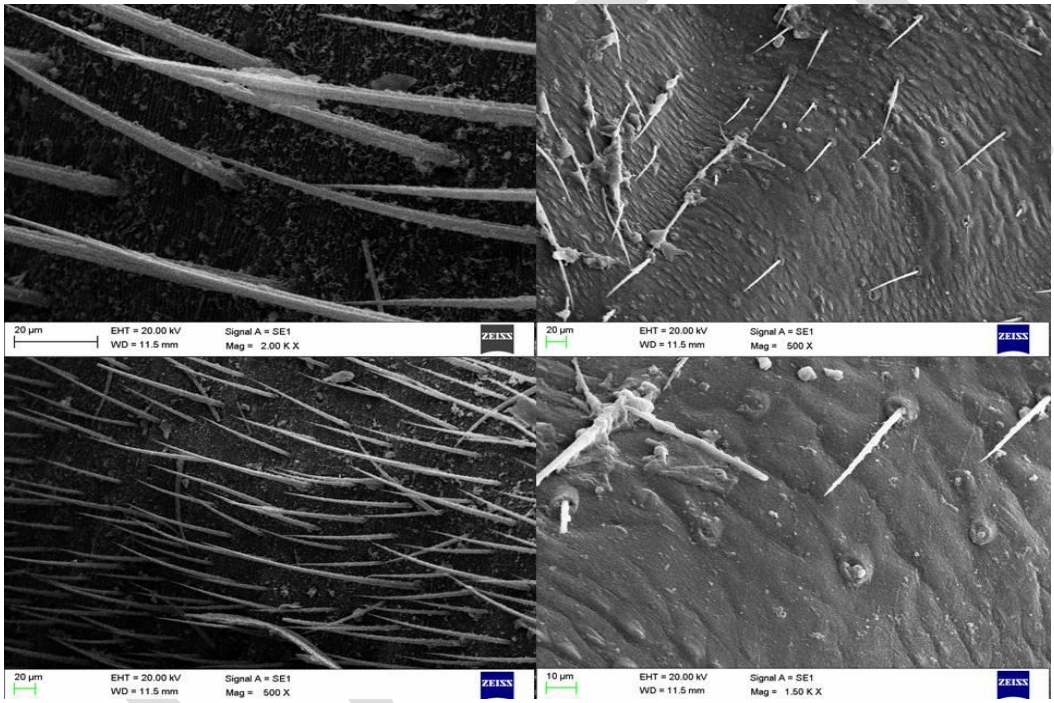
Şekil 1. Zeiss Marka evo 40 model SEM

Bulgular

Yer örümceklerinin iki cinsine ait iki türün, *Cryptodrassus creticus* (Chatzaki, 2002) ve *Drassyllus praeficus* (L. Koch, 1866) türlerinin prozoma ve opistozomaları üzerinde bulunan setaların morfolojileri SEM yardımıyla çekilmiş olup bu iki cinse ait setaların morfolojileri kıyaslanmıştır.



Şekil 2. *Cryptodrassus creticus* türüne ait plumose seta morfolojisi (abdomen)



Şekil 3. *Drassyllus praeficus* türüne ait setasız kütikula (abdomen)

Sonuç ve Tartışma

SEM yardımıyla fotoğraflanan Gnaphosidae familyasına ait 2 cinse ait 2 türü setalarının incelenmesi sonucunda *Cryptodrassus creticus* türünde tüysü (plumose) tip seta görülürken, *Drassyllus praeficus* türünde ise seta örneğine rastlanmamış olup Şekil 3.'de görülen yapıların sensilla olduğu tespit edilmiştir. Yapılan bu çalışma yer örümceklerinde bir ön çalışma niteliğinde olup diğer gnaphozid cinslerinin setalarının veya aynı cinse ait tüm türlerin seta morfolojileri incelenirse, yer örümceklerinin ayrımında seta morfolojisinin taksonomik olarak kullanılmasının ne derece de etkili olacağı görülebilir.

Teşekkür

Finansal desteğinden dolayı Niğde Ömer Halisdemir Üniversitesi Bilimsel Araştırmalar Birimine (Proje No: FEB 2018/09- TÜBİTAK C) teşekkür ederiz.

Kaynaklar

Chatzaki M., Thaler K., Mylonas M., Ground Spiders (Gnaphosidae: Araneae) from Crete and Adjacent Areas of Greece Taxonomy and Distrubution I, *Revue Suisse de Zoloogie*, 109: 559–601, 2002.

Danışman, T., Kunt, K.B., & Özkütük, R.S., The Checklist of the Spiders of Turkey. *http://www.spidersofturkey.info* (Erişim tarihi: 29.07.2023)

Demir, H., ve Seyyar, O, Annotated checklist of the spiders of Turkey. *Munis Entomology & Zoology*, 12(2): 433-469, 2017.

Hill D. (1979). The scales of salticid spiders, Zoological Journal of the Linnean Society 65: 193-218 – doi: 10.1111/j.1096-3642.1979.tb01091.x

Lehtinen PT., (1975) The significance of hair ultrastructure in phylogenetic classification of spiders, Journal of Ultrastructure Research 50: 362-395

Townsend VR Jr & Felgenhauer BE (2001) Phylogenetic significance of the morphology of the cuticular scales of the lynx spiders (Araneae: Oxyopidae), Journal of Zoology 253: 309-332 – doi: 10.1017/S0952836901000292

World Spider Catalog., World Spider Catalog. Version 24.5. Natural History Museum Bern, online at http://wsc.nmbe.ch (Erişim tarihi: 21.08.2023)

Zakharov, B., Ovtsharenko, V.,The covering setae of ground spiders (Araneae: Gnaphosidae) *Arachnologische Mitteilungen* 49: 34-46 Karlsruhe, Juni 2015

DRAFT

ORAL PRESENTATION

Falcarinol'ün Elektronik, Yapısal ve Farmakokinetik Karakterizasyonu: DFT, Moleküler Yerleştirme ve ADMET Çalışmaları

Özlem Akdere[1] (*ORCID: https://orcid.org/0009-0004-6988-9108*), Sevtap Çağlar Yavuz[2*] (*ORCID: https://orcid.org/0000-0001-6497-2907*)

[1]Erzincan Binali Yıldırım Üniversitesi, Fen Edebiyat Fakültesi, Kimya Bölümü, Erzincan, Türkiye
[*2]Erzincan Binali Yıldırım Üniversitesi, İliç Dursun Yıldırım Meslek Yüksekokulu, Tıbbi Hizmetler ve Teknikler Bölümü, Erzincan, Türkiye

*Sorumlu yazar e-mail: sevtap.yavuz@erzincan.edu.tr

Özet

Son yıllarda potansiyel sağlığı geliştirici bileşikler ile ilgili çalışmalara odaklanılmıştır. Bu bileşik gruplarından biri, havuç, maydanoz, kereviz, yabani havuç ve rezene gibi ilgili sebzelerde yaygın olan falcarinol tipi alifatik C17-poliasetilenlerdir. Falcarinol tipi poliasetilenler, anti-inflamatuar, antiplatelet, sitotoksik ve antitümör aktivitenin yanı sıra bakteri ve mikoplazmaya karşı birçok biyoaktivite göstermektedir. Bu çalışma kapsamında, farmakolojik olarak etkinliği yüksek olan falcarinolun yapısal ve elektronik özellikleri incelenmiştir. Bileşiğin moleküler optimizasyon hesapları Spartan 10 yazılımı kullanılarak Yoğunluk Fonksiyonel Teorisinin (DFT) B3LYP metodu ve standart 6-311+G** baz setleri kullanılarak yapılmıştır. Öncü moleküler orbitaller olan en yüksek dolu moleküler orbital (HOMO) ve en düşük boş moleküler orbital (LUMO) enerjileri kullanılarak hesaplanan sertlik ve yumuşaklık parametreleri ile global kimyasal aktivite verileri elde edilmiştir. Bileşiğin moleküler elektrostatik potansiyel (MEP) harita analizi gerçekleştirilmiştir. Ayrıca, moleküler yerleştirme, ADME ve toksisite özellikleri in silico olarak analiz edilmiştir. Asetilkolinesteraz (PDB kodu: 1EVE) ve pankreatik alfa-amilazın (PDB kodu: 1B2Y) falcarinol ile bağlanma enerjileri sırasıyla -7.0 kcal/mol ve -6.1 kcal/mol olarak hesaplanmıştır.

Anahtar Kelimeler: Falcarinol, Moleküler yerleştirme, Spartan 10

Electronic, Structural and Pharmacokinetic Characterization of Falcarinol: DFT, Molecular Docking and ADMET Studies

Abstract

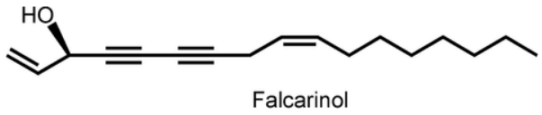
In recent years, it has been a focus on studies of potential health-promoting compounds. One of these groups of compounds is the aliphatic C17 polyacetylenes of the falcarinol type, which are common in related vegetables such as carrots, parsley, celery, parsnips, and fennel. Falcarinol-type polyacetylenes show anti-inflammatory, antiplatelet, cytotoxic, and antitumour activity as well as many bioactivities against bacteria and mycoplasma. Within the scope of this study, the structural and electronic properties of falcarinol, which has high pharmacological activity, were investigated. Molecular optimisation calculations of the compound were performed at B3LYP/standard 6-311+G** basis set of Density Functional Theory (DFT) using Spartan 10 software. Global chemical activity data were obtained with hardness and softness parameters calculated using the highest occupied molecular orbital (HOMO) and lowest unoccupied molecular orbital (LUMO) energies, which are the leading molecular orbitals. Molecular electrostatic potential (MEP) map analysis of the compound was performed. Additionally, molecular docking, ADME and toxicity properties were analyzed in silico. The binding energies of acetylcholinesterase (PDB code: 1EVE) and pancreatic alpha-amylase (PDB code: 1B2Y) with falcarinol were calculated as -7.0 kcal/mol and -6.1 kcal/mol, respectively.

Keywords: Falcarinol, Molecular docking, Spartan 10

DRAFT

GİRİŞ

Havuç, poliasetilenler ve karotenoidler olmak üzere iki grup fitokimyasal için özellikle iyi bir kaynaktır (Lund ve White, 1990; Bernart ve ark., 1996). Diğer sebzelerle karşılaştırıldığında havuç, insan diyetine daha fazla katkı sağlayabilir. Antioksidan, anti-kanserojen ve bağışıklık arttırıcı faydaları, yara iyileşmesi, kolesterolü düşürücü, anti-diyabetik, anti-enflamatuvar, antibakteriyel ve antifungal özellikleri araştırmacılar tarafından rapor edilmiştir (Pandey ve ark., 2020). Son zamanlarda yapılan bir dizi çalışma, Apiaceae familyasının üyelerinde bulunan bir grup C17 poliasetilenin kanser hücrelerine karşı sitotoksisiteye sahip olduğunu bildirmiştir (Kobæk-Larsen ve ark., 2005; Christensen ve Brandt, 2006. Havuçlar (Daucus carota L., Apiaceae) falcarinol tipinde 3 poliasetilen, yani falcarinol, falcarindiol ve falcarindiol-3-asetat içerir. Bu bileşikler bitkide (kök) mantar enfeksiyonlarını önler. Ayrıca insanlarda anti-bakteriyel, anti-inflamatuar ve anti-trombosit-agregasyon etkilerine sahiptirler (Rawson ve ark., 2011). İnsan tümör hücrelerine karşı önemli bir sitotoksik aktivite gösteren falcarinol, panaxynol olarak da adlandırılır (Zheng ve ark., 1999). Falcarinol havuçta bulunan en biyoaktif poliasetilen olarak bildirilmiştir. Falcarinol, iki karbon-karbon üçlü bağı ve iki çift bağı olan bir moleküldür (Larqué ve ark., 2022). Karbon 9 pozisyonundaki çift bağ, oksijen ve NADPH (veya NADH) kofaktörleri gerektiren desatürasyon ile ortaya konan *cis* stereokimyasına sahiptir, molekül, yağ asidinin hücre zarlarında katılaşmasını önleyen bir bükülme oluşturur (Yates ve England, 1982). Bu çalışma kapsamında, dikkat çekici biyaoaktif bir bileşik olan falcarinol bileşiğinin yapısal ve elektronik özellikleri incelendi ve alzheimer ile diyabet hastalıklarına karşı etkinliği moleküler yerleştime çalışmaları ile tahmin edildi.



Falcarinol

Şekil 1. Falcarinol bileşiğinin yapısı

MATERYAL VE METOT

Falcarinol bileşiğinin, Spartan 10 paket programı (Spartan'10, version 1.1.0.) içerisinde yer alan B3LYP/6-311+G** yoğunluk fonksiyonu yöntemi (DFT) kullanılarak kuantum kimyasal hesaplamaları yapıldı. Optimize yapının kimyasal kararlılığı için gaz fazında en yüksek dolu moleküler orbital enerjisi (E_{HOMO}), en düşük boş moleküler orbital enerjisi (E_{LUMO}), enerji farkı (ΔE), kimyasal sertlik (η), elektronegatiflik (χ) ve dipol moment (μ) değerleri gibi çeşitli parametreleri hesaplandı. Ayrıca moleküldeki elektrofilik ve nükleofilik tepkimelerin olabileceği bölgelerin belirlenmesinde ve molekül içi hidrojen bağı oluşumu hakkında önemli bilgiler veren Moleküler elektrostatik potansiyeli (MEP) haritası hesaplandı. Yerleştirme analizleri, AutoDock Vina yazılımı ve UCSF Chimera (Butt ve ark.,2020) kullanılarak gerçekleştirildi. Asetilkolinesterazin üç boyutlu yapısı (PDB kodu: 1EVE) ve pankreatik alfa-amilazın üç boyutlu yapısı (PDB kodu: 1B2Y) PDB formatlarında 3D yapısı RSCB PDB web sitesinden sağlandı. Ligand-reseptör 3D etkileşimlerini görselleştirmek için Biovia Discovery Studio Visualizer kullanıldı. Falcarinol bileşiğinin ADMET (absorpsiyon, dağılım, metabolizma, atılım ve toksisite) özellikleri tahmin etmek için SwissADME, SwissTargetPrediction, ADMETlab, Pro-Tox II ve admetSAR gibi çeşitli web araçları kullanıldı.

BULGULAR ve TARTIŞMA

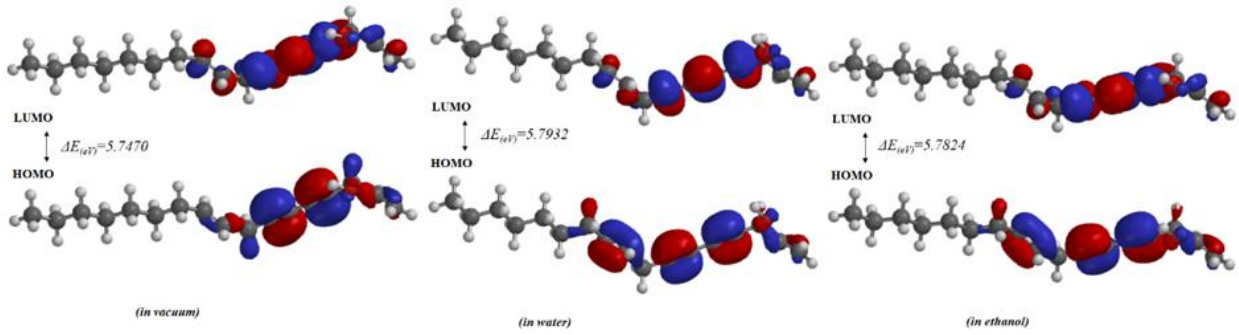
HOMO-LUMO enerji değerleri arasındaki fark önemli bir stabilite indeksidir. Büyük bir HOMO-LUMO enerji açığı molekülün daha kararlı ve daha az reaktif olduğunu, küçük bir HOMO-LUMO enerji açığı ise molekülün daha az kararlı ve daha reaktif olduğunu temsil eder (Kim ve ark., 2005). HOMO-LUMO enerji aralığına sahip HOMO ve LUMO enerji değerleri, elektronegatiflik, sertlik, yumuşaklık, elektrofiliklik indeksi ve kimyasal potansiyel gibi parametreleri tahmin etmek için kullanılır. Enerji açığındaki azalma genellikle molekülün sertliğinin azalmasına ve daha kolay polarizasyonuna yol açar. Molekül optimizasyonu, fonksiyonel B3LYP/6-311+G** temel setinde gaz, su ve etil alkol ortamında gerçekleştirildi ve HOMO-LUMO enerjileri hesaplandı. Falcarinol molekülü için hesaplanan HOMO-LUMO enerjisi (a.u.) ve bant aralığı değeri sırasıyla 0.2112 eV, 0.2129 eV ve 0.2124 eV'dir. Ayrıca falcarinol molekülünün elektronik özellikleri ve global reaktivite parametreleri hesaplandı ve hesaplanan sonuçlar Tablo 1'de verilmiştir.

Tablo 1'de verilen enerji bant aralığının 1.5 eV'den az olması molekülün termodinamik açıdan kararlı ve dayanıklı olmadığını göstermektedir. Bu aynı zamanda molekülün kendisiyle reaksiyona girebileceği,

dimerizasyon ve polimerizasyonun meydana gelebileceği anlamına da gelir. Elektrofilisiti indeksi (ω), özellikle organik moleküllerin güçlü veya zayıf elektrofilik özelliklerini anlamak için kullanılmaktadır. Buna göre moleküller; ω > 1.5 eV ise güçlü elektrofil, 0.8 eV ve 1.5 eV arasında ise orta dereceli elektrofil, ω < 0.8 eV ise zayıf elektrofilik karaktere sahip olarak değerlendirilmektedir (Domingo ve ark., 2008). Frontier moleküler orbitallerin (FMO) 3 boyutlu çizimleri ve FMO boşluklarının (ΔE) enerjileri Şekil 2'de gösterilmiştir.

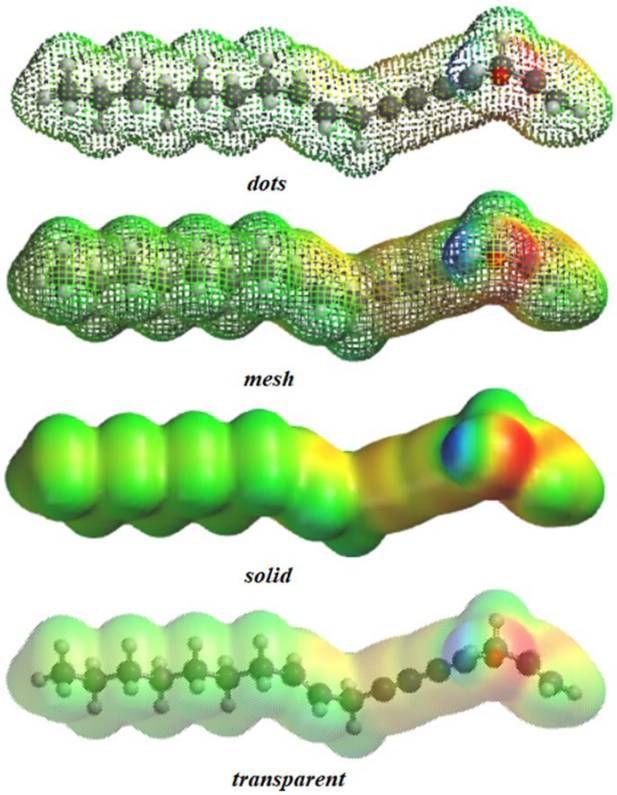
Tablo 1. Falcorinol molekülünün kimyasal reaktivite tanımlayıcıları

Parametreler	**(eV)**
E_{HOMO}	-0.240
E_{LUMO}	-0.027
Enerji bant aralığı ($\Delta E = E_{LUMO}-E_{HOMO}$)	0.212
İyonlaşma potansiyeli ($I = -E_{HOMO}$)	0.240
Elektron ilgisi ($A = -E_{LUMO}$)	0.027
Elektronegatiflik ($\chi = (I + A)/2$)	0.134
Kimyasal sertlik ($\eta = (I-A)/2$)	0.106
Kimyasal yumşaklık ($\sigma = 1/2\eta$)	4.697
Kimyasal potansiyel ($\mu = -(I + A)/2$)	-0.134
Elektrofiliklik indeksi ($\omega = \mu^2/2\eta$)	0.084
Maksimum yük aktarım indeksi ($\Delta N_{max} = -\mu/\eta$)	1.260



Şekil 2. Falcarinol için farklı ortamlarda HOMO-LUMO enerji farkları (ΔE)

Moleküler elektrostatik potansiyel (MEP), moleküler davranışın reaktivitesini, yapı-aktivitesini ve hidrojen bağını açıklamak için kullanılan yararlı bir yöntemdir (Murray ve Sen 1996). Yeşil renkli kısımlar elektrostatik potansiyelin sıfır olduğu bölgelerdir. MEP haritalarında elektrostatik potansiyel Kırmızı<Turuncu<Sarı<Yeşil<Mavi sırasıyla artar.

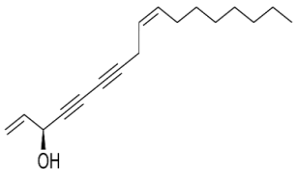


Şekil 3. Falcarinol molekülün MEP haritalarının gösterimi

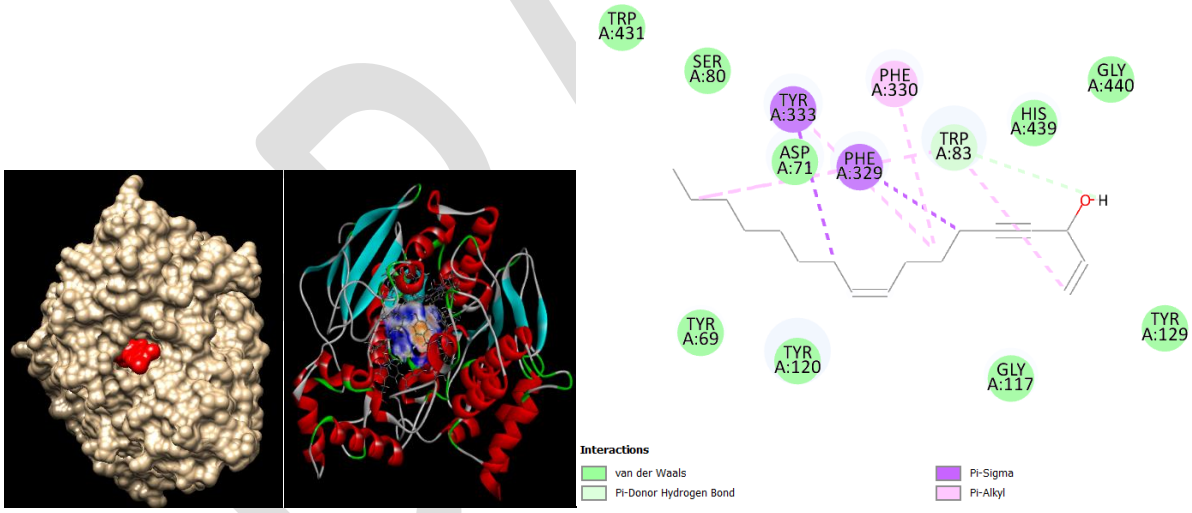
Şekil 3'te görüldüğü gibi haritadaki kırmızı renk hidroksil grubundaki oksijen atomunun elektronca en zengin bölge olduğunu göstermektedir. Bunun nedeni, oksijen atomu üzerindeki çiftleşmemiş ve proton çekici olan elektron çiftidir ve bu bölge yüksek elektrofilik saldırı etkinliği temsil etmektedir. Molekülün diğer kısımlarında görülen yeşil renk, elektron yoğunluğunun daha az olduğunu göstermektedir. Pozitif bölgeler hidrojen atomlarının üzerinde bulunur ve nükleofilik saldırılar için uygundur.

Moleküler yerleştirme analizleri, enzimler ve bileşikler arasındaki reaksiyonların moleküler etkileşimlerinin aydınlatılmasına yardımcı olur. Yerleştirme analizleri, hedef bileşiklerin etki mekanizmasının, ve uygun bağlanma bölgelerinin anlaşılmasını kolaylaştırır. Yerleştirme analizi sonucunda falcarinol molekülünün sırasıyla asetilkolinesteraz (AChE) (PDB kodu: 1EVE) ve pankreatik alfa-amilaz (PDB kodu: 1B2Y) ile etkileşimi gerçekleştirildi. Yerleştirme sonucu falcarinol molekülünün sırasıyla -7.0 kcal/mol ve -6.0 kcal/mol olmak üzere iyi bir bağlanma enerji değeri gösterdiğini destekledi.

Tablo 2. PDB ID: 1EVE ve PDB ID: 1B2Y ile falcarinol molekülünün yerleştirme sonuçları

Protein ID		**1EVE**		**1B2Y**	
Bileşik Adı	Chemical Structure	Docking score (kcal/mol)	Bağlanmada yer alan amino asitler	Docking score (kcal/mol)	Bağlanmada yer alan amino asitler
falcarinol		-7.0	ASP71, TRP83, PHE329, TYR333, PHE330	-6.1	TRP57, TYR61, HIS100, LEU161, ALA197, HIS200, ASP299, GLY305

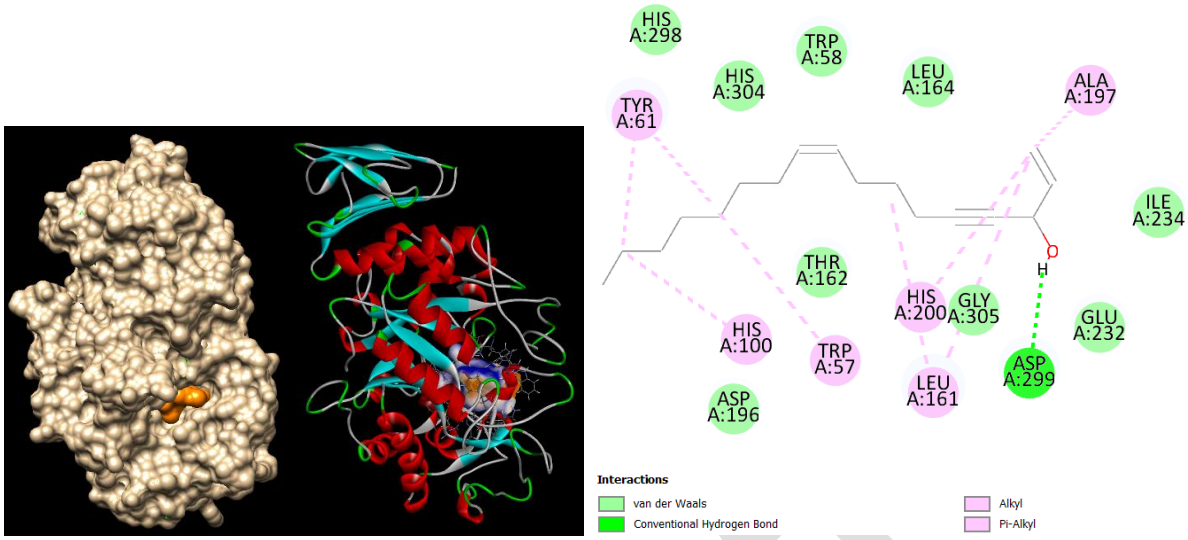
Ligand-reseptör 3D etkileşimleri, Biovia Discovery Studio Visualizer programı (D.S.J.S.D. Biovia, CA, ABD, Discovery Studio Visualizer) aracılığıyla görselleştirildi. AChE'nin (PDB kodu: 1EVE) 2.50 Å çözünürlükteki 3 boyutlu yapısı ile pankreatik alfa-amilazın 3.20 Å çözünürlükteki 3 boyutlu yapısı RSCB PDB web sitesi aracılığıyla elde edildi. Falcarinol molekülü, AChE kompleksi ile ikincil etkileşimler (van der Waals, pi-sigma, pi-donor hidrojen bağı ve pi-alkil) oluşturdu. Falcarinol-1EVE kompleksinde ASP71, TRP83, PHE329, PHE330 ve TYR333 kalıntıları arasında ikincil etkileşimler yer aldı. Bu etkileşimler Şekil 4'te gösterilmiştir.



Şekil 4. Falcarinol ve PDB ID: 1EVE'nin en kararlı kompleksi için protein-ligand etkileşim diyagramı

Falcarinol molekülü, pankreatik alfa-amilazı kompleksi ile ikincil etkileşimler (van der Waals, hidrojen bağı, alkil ve pi-alkil) oluşturdu. Etkileşimde yer alan amino asitler TRP57, TYR61, HIS100, LEU161, ALA197, HIS200, ASP299, GLY305'dir. Bu etkileşimler Şekil 5'te gösterilmiştir.

DRAFT



Şekil 5. Falcarinol ve PDB ID: 1B2Y'nin en kararlı kompleksi için protein-ligand etkileşim diyagramı

İlaç benzeri özellikler, molekülün fizikokimyasal ve ADMET özelliklerini ifade eder. In vivo ve in vitro analizlerin maliyetli ve zaman alıcı olması nedeniyle, in silico çalışmaları moleküllerin ADME özelliklerini tahmin etmek için yaygın olarak kullanılmaktadır (Pelkonen ve ark., 2011). Elde edilen sonuçlar Tablo 3'te verilmiştir.

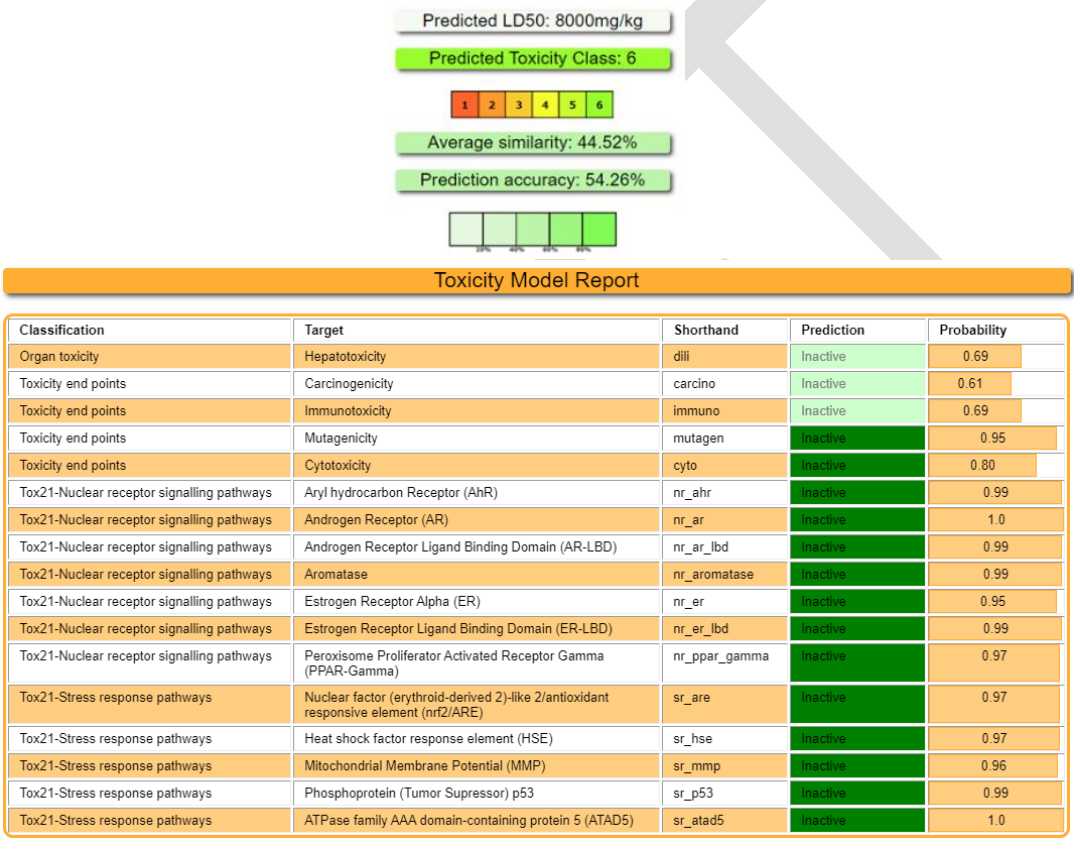
Tablo 3. SwissADME ve ADMETlab programlarına göre falcarinolun tahmini fizikokimyasal özellikleri

	SwissADME	**ADMETlab**
	Physicochemical Properties	
Formülü	$C_{17}H_{24}O$	$C_{17}H_{24}O$
Moleküler ağırlık	244.37g/mol	244.18g/mol
Ağır atomların sayısı	18	
Aromatik ağır atomların sayısı	0	
Fraction Csp^3	0.53	
Dönebilen bağ sayısı	8	
Hidrojen bağı alıcılarının sayısı	1	
Hidrojen bağı vericilerinin sayısı	1	
Molar kırılma	80.37	
TPSA	20.23Å^2	
Lipofiliklik		
$LogP_{o/w}$	3.86	
Su çözünürlüğü		
LogS	-4.49	
Çözünürlük	1.27e-02 mg/ml	
Emilim		
GI emilim		
Dağıtım		
BBB geçirgenlik		
P-gp substrat		
Metabolizma		
CYP1A2 inhibitör	Evet	
CYP2C19 inhibitör	No	
CYP2C9 inhibitör	Evet	
CYP2D6 inhibitör	No	
CYP3A4 inhibitör	No	
$LogK_p$ (cilt geçirgenliği)	-3.89cm/s	
İlaç-benzerliği		

DRAFT

Lipinski	Evet; 1 violation: MLOGP>4.15
Ghose	Evet
Veber	Evet
Tıbbi Kimya	
PAINS	0 uyarı
Brenk	2 uyarı
Sentetik Erişilebilirlik	4.33

Toksisite tahmini Pro-Tox II web sunucusu sayesinde gerçekleştirildi. Öngörülen sonuçlar Şekil 6'da verilmiştir. Toksisite sonuçlarına ilişkin olarak, falcorinol molekülü için bazı tahminler elde edilebilir: Molekül sitotoksik, kanserojen, immünotoksik veya mutajenik değildir. Pro Tox-II'ye göre molekül, toksisite sınıfı 6 olarak sınıflandırılmıştır.



Classification	Target	Shorthand	Prediction	Probability
Organ toxicity	Hepatotoxicity	dili	Inactive	0.69
Toxicity end points	Carcinogenicity	carcino	Inactive	0.61
Toxicity end points	Immunotoxicity	immuno	Inactive	0.69
Toxicity end points	Mutagenicity	mutagen	Inactive	0.95
Toxicity end points	Cytotoxicity	cyto	Inactive	0.80
Tox21-Nuclear receptor signalling pathways	Aryl hydrocarbon Receptor (AhR)	nr_ahr	Inactive	0.99
Tox21-Nuclear receptor signalling pathways	Androgen Receptor (AR)	nr_ar	Inactive	1.0
Tox21-Nuclear receptor signalling pathways	Androgen Receptor Ligand Binding Domain (AR-LBD)	nr_ar_lbd	Inactive	0.99
Tox21-Nuclear receptor signalling pathways	Aromatase	nr_aromatase	Inactive	0.99
Tox21-Nuclear receptor signalling pathways	Estrogen Receptor Alpha (ER)	nr_er	Inactive	0.95
Tox21-Nuclear receptor signalling pathways	Estrogen Receptor Ligand Binding Domain (ER-LBD)	nr_er_lbd	Inactive	0.99
Tox21-Nuclear receptor signalling pathways	Peroxisome Proliferator Activated Receptor Gamma (PPAR-Gamma)	nr_ppar_gamma	Inactive	0.97
Tox21-Stress response pathways	Nuclear factor (erythroid-derived 2)-like 2/antioxidant responsive element (nrf2/ARE)	sr_are	Inactive	0.97
Tox21-Stress response pathways	Heat shock factor response element (HSE)	sr_hse	Inactive	0.97
Tox21-Stress response pathways	Mitochondrial Membrane Potential (MMP)	sr_mmp	Inactive	0.96
Tox21-Stress response pathways	Phosphoprotein (Tumor Supressor) p53	sr_p53	Inactive	0.99
Tox21-Stress response pathways	ATPase family AAA domain-containing protein 5 (ATAD5)	sr_atad5	Inactive	1.0

Şekil 6. Falcarinol bileşiğinin Pro-Tox II ile toksisite tahmini

SONUÇ

Falcorinol molekülünün kuantum kimyasal hesaplamaları, B3LYP/6-311+G** temel seti kullanılarak DFT düzeyinde belirlenmiş ve analiz edilmiştir. Sonuçlardan görüldüğü gibi falcorinol molekülünün enerji aralığının (ΔE) küçük olması onun sert bir molekül olmadığını göstermektedir. Molekül çözünür. Yerleştirme sonuçları, bu molekülün her iki hedef protein ile etkileşimlerinde ikincil etkileşim türlerinin etkin olduğu gösterdi. Molekülün sitotoksik, kanserojenik, immünotoksik veya mutajenik olmadığı Pro-Tox II programı ile tahmin edildi.

TEŞEKKÜR

Bu çalışma Tübitak 2209-A Üniversite Öğrencileri Araştırma Projeleri Destekleme Programı (Proje Kodu: TBTK-0122-5631) tarafından desteklenmiştir.

KAYNAKLAR

Bernart MW, Cardellina JH, Balaschak MS, Alexander MR, Shoemaker RH, Boyd MR 1996. Cytotoxic falcarinol oxylipins from Dendropanax arboreus. Journal of Natural Products, 59(8): 748-753.

Christensen LP, Brandt K 2006. Bioactive polyacetylenes in food plants of the Apiaceae family: occurrence, bioactivity and analysis. Journal of pharmaceutical and biomedical analysis, 41(3): 683-693.

Domingo LR, Chamorro E, Pérez P 2008. Understanding the reactivity of captodative ethylenes in polar cycloaddition reactions. A theoretical study. Journal of Organic Chemistry, 73(12): 4615–4624.

Kim KH, Han YK, Jung J 2005. Basis set effects on relative energies and HOMO–LUMO energy gaps of fullerene C36. Theoretical Chemistry Accounts, 113: 233-237.

Kobæk-Larsen M, Christensen LP, Vach W, Ritskes-Hoitinga J, Brandt K 2005. Inhibitory effects of feeding with carrots or (−)-falcarinol on development of azoxymethane-induced preneoplastic lesions in the rat colon. Journal of Agricultural and Food Chemistry, 53(5): 1823-1827.

Larqué H, Carrillo-Aké AG, de la Vega J, Peraza-Sánchez SR, Carballo RM, Chávez-Montes A, del Olmo E 2022. Total synthesis of the falcarinol-type oxylipin (3S)-16, 17-didehydrofalcarinol and its activity against Leishmania mexicana. Tetrahedron, 132832.

Lund ED, White JM 1990. Polyacetylenes in normal and waterstressed 'Orlando Gold'carrots (Daucus carota). Journal of the Science of Food and Agriculture, 51(4): 507-516.

Murray JS, Sen K 1996. Molecular Electrostatic Potentials, Volume 3 1st Edition Concepts and Applications. Amsterdam: Elsevier.

Pandey N, Rijal S, Adhikari H, Bhantana B, Adhikhari M 2020. Production economics and determinants of carrot (Daucus carota L.) production in Chitwan, Nepal. International Journal of Social Sciences and Management, 7(4): 234-241.

Pelkonen O, Turpeinen M, Raunio H 2011. In vivo-in vitro-in silico pharmacokinetic modelling in drug development. Clinical pharmacokinetics, 50(8): 483-491.

Rawson A, Tiwari BK, Tuohy MG, O'donnell CP, Brunton N 2011. Effect of ultrasound and blanching pretreatments on polyacetylene and carotenoid content of hot air and freeze dried carrot discs. Ultrasonics Sonochemistry, 18(5): 1172-1179.

Spartan'10, version 1.1.0. Wavefunction, Inc. Irvine, CA. 2010.

Butt SS, Badshah Y, Shabbir M, Rafiq M. 2020. Molecular docking using chimera and autodock vina software for nonbioinformaticians. JMIR Bioinformatics and Biotechnology, 1(1): e14232.

Yates SG, England RE 1982. Isolation and analysis of carrot constituents: myristicin, falcarinol, and falcarindiol. Journal of Agricultural and Food Chemistry, 30(2): 317-320.

Zheng G, Lu W, Aisa HA, Cai J 1999. Absolute configuration of falcarinol, a potent antitumor agent commonly occurring in plants. Tetrahedron letters, 40(11): 2181-2182.

ORAL PRESENTATION

Ağır metaller ile kirletilmiş topraklarda yetişen semizotu bitkisinin fitoremediasyon potensiyeli üzerine bir ön çalışma

Elif Duyuşen Güven[1*] (*ORCID:h ttps://orcid.org/0000-0001-6046-7639),* Serpil Özmıhçı[2] (*ORCID: https://orcid.org/0000-0003-4817-7916*), Görkem Akıncı[3] *(ORCID: https://orcid.org/0000-0001-6288-3812),* Egesu Acar *(ORCID: https://orcid.org/ 0009-0005-7617-1898*), Erva Nur Poyraz (*ORCID: https://orcid.org/0009-0008-3931-5778*), Atakan Şatır (*ORCID: https://orcid.org/0009-0001-6511-4055)*

[*1,2,3] Dokuz Eyllül Üniversitesi Çevre Mühendisliği Bölümü, İzmir, TÜRKİYE

*Corresponding author e-mail: duyusen.kokulu@gmail.com

Özet

Çalışmada, Portulacaceae familyasına ait semizotu bitkisinin ağır metaller ile kirletilmiş toprakta fitoremediasyon denemesini içermektedir. Çalışma,sonbahar mevsiminde sera koşullarında ve toprak organik madde içeriği %5 olacak şekilde saksı deneyleri şeklinde gerçekleştirilmiş, orta seviyede ağır metallerle kirli toprakta yetiştirilen semizotu bitkilerinin ağırlıkları ve bitkilerin yenilebilir kısımlarına aldığı metal konsantrasyonları değerlendirilmiştir. Birbirine paralel olarak hazırlanan kirli toprak saksılarında süreç sonucunda ortalama %40 oranında çimlenme gözlenirken, herhangi bir kirletici müdahalesi olmamış toprak içeren saksılarda bu oran ortalama %82 oranında gerçekleşmiştir. Serada 55 günlük deney süreci sonucunda, topraktan bitkiye ağır metal geçişleri Cr, Cu, Ni ve Zn için transfer katsayısı 1 den büyük olup, anlamlı bir biçimde geçişi ifade etmektedir. Özellikle hareketliliğinin yüksek olduğu bilinen Zn için bu değer bitki gelişiminin daha kuvvetli olduğu kontrol saksılarında 2,88 ile yüksek bir geçişe işaret etmektedir.

Anahtar kelimeler: : Semizotu, ağır metal, fitoremediasyon, transfer katsayısı

Abstract

The study includes a phytoremediation trial of purslane plant belonging to the Portulacaceae family in soil contaminated with heavy metals. The study was carried out in the form of pot experiments in greenhouse conditions in the autumn season with a soil organic matter content of 5%, and the weights of purslane plants grown in soil contaminated with moderate levels of heavy metals as well as the metal concentrations in the edible parts of the plants were evaluated. While an average of 40% germination rate was observed as a result of the process in contaminated soil pots prepared in parallel with each other, this rate was 82% on average in pots containing soil without any pollutant intervention. As a result of the 55-day experiment period in the greenhouse, the transfer coefficients for heavy metal transfers from soil to plants for Cr, Cu, Ni and Zn were found to be greater than 1, indicating a significant transfer from soil to plants. Especially for Zn, which is known to have high mobility, this value indicates a high transition with 2.88 in control pots where plant growth is stronger.

Key words: Purslane, heavy metal, phytoremediation, transfer coefficient

GİRİŞ

Semizotu, *Portulacaceae* familyasına ait tek yıllık, çok yapraklı, sürünen otsu bir bitkidir. Semizotu diğer bitkilere göre yüksek oranda omega-3 yağ asitleri, B-kompleks vitaminleri içerir ve iyi bir C vitamini kaynağıdır. Semizotu demir, fosfor, bakır, çinko, magnezyum, kalsiyum, potasyum ve manganez gibi önemli mineraller açısından da önemli besin değeri olan bir sebzedir (Açak, 2017; Dweck 2001; Turan ve ark., 2003). Bitkinin yabani formları uzun yıllardan beri bilinmesine rağmen insan beslenmesindeki yeri ve önemi yeni yeni anlaşılmaya başlanmıştır. Türkiye'de semizotunun kültür formu ticari olarak Ege, Marmara ve Akdeniz bölgelerinde üretilmekte olup, yabani formları ise tüm bölgelerde çoğunlukla kullanılmaktadır (Eşiyok, 2012; Yurdagül, 2019). İlkbahar-yaz mevsiminde hızlı bir biçimde büyüdüğü bilinen bitkilerde kuru madde miktarı artmaktadır (Convington, 2004). Bitkinin en iyi gelişme gösterdiği mevsim erken ilkbahar olduğu için

yetiştiriciliği genellikle bu dönemde yapılmaktadır. Kullanılan toprak kaliteli ve organik olursa semizotu da bir o kadar besleyici olmaktadır.

Semizotu literatürde pek çok çalışmada fitoremediasyon bitkisi olarak tanımlanabilmektedir. Yedagari tarafından 2017'de yapılan bir çalışmada (Güneybatı İran) semizotu bitkisinin nikel ve kadmiyumla kirlenmiş toprakta ağır metal ayırıcı bir ürün olarak performansı incelenmiştir. Bu çalışmanın sonucunda, nikelin düşük kadmiyum konsantrasyonu ile daha fazla bitki dokusuna geçtiği, toprakta kadmiyumun 20 mg/kg'dan fazla olmasının nikelin bitkiye taşınmasını azalttığı görülmüştür. Sonuçlar ayrıca *Portulaca oleracea* L. ekstraksiyon içeriğinin toprağın ağır metal özelliğinin farklılaşmasından önemli ölçüde etkilendiğini göstermiştir. Büyüme mevsimlerinde semizotunda 29,6-30,2 mg/kg kadmiyum ve 85,4- 86,1 mg/kg nikel görülmüştür. Ayrıca mevcut çalışmada aynı zamanda *P. oleracea* 'nin ekstraksiyon içeriğinin, nikelin hareketliliğinden etkilendiği ve nikel ile iletilen kadmiyumun toksisitesinin bitki dokularında mevcut olduğu görülmüştür. Bu çalışma yararlı ağır metallerin etkisi hakkında bilgi vermiş olması ve semizotu bitkisinin fitoremediasyon potansiyelini göstermesi bakımından önem taşımaktadır. Deepa ve ark. (2006), büyük ölçüde, birçok ağır metalin neden olduğu stres koşulları altında bitkilerin büyüme kapasitesinin incelenmesine odaklanmıştır. Araştırmacılar bitki büyümesini etkileyen metallerin toksisite sırasını Cd>Cu>Al>Zn>Hg>Se>Pb olarak belirlemişlerdir. Eid ve Shaltout (2016), Mısır'ın kuzeyindeki Nil Deltası'ndaki kanalizasyon çamuru depolama alanında yetişen semizotu bitkisinin Zn ve Mn'yi aşırı biriktirebildiğini kanıtlamıştır.

Mısır'da gerçekleştirilen bir çalışmada, endüstriyel atıksuların toksik etkilerine maruz kalan tarım arazilerinde yetişen semizotu bitkisinin toprakta mevcut Mn(II), Cu(II), Zn(II), Fe(III) ve Pb(II) iyonlarını biriktirme kapasitesi incelenmiştir. Fitoremediyason verimi zenginleşme faktörü (BCF) olarak Cu (II) için 7.4, Fe(III) için 2.06; Zn(II) için 4.33, Mn(II) için 2.06 ve Pb(II) için 3.89 olarak tespit edilmiştir. Biyoakümülasyon faktörü (BF) ve translokasyon faktörleri denenen topraklar ve metal iyonları için 1'in altında bulunmuştur (El-Shami et.al. , 2019).

Prabha v.d. (2015), çeşitli Se konsantrasyonlarında alfisol içinde yetiştirilen semiz otunun gövde kesimleri tarafından selenyum (Se) alımını ve birikiminini araştırmışlardır. Hindistan'da gerçekleştirilen çalışmada, 42 günlük kısa bir büyüme döneminde maksimum 63.4 µg g^{-1} kuru ağırlık elde edilmiş ve bitki kısımları arasında Se birikme sırası yapraklar (31,5 µg g^{-1})> saplar (16,4 µg g^{-1})> kökler (15,5 µg g^{-1}) şeklinde olmuştur. Akümülasyon potansiyeli, bitkinin temin edilebildiği 15.2 µg g^{-1} Se g^{-1} toprak konsantrasyonundan dört kat daha yüksek olmuştur. Bitki dokularında Se biriktirebilmesine rağmen, topraktaki Se konsantrasyonlarının artırılması bitkilerin büyüme hızında artan konsantrasyona bağlı bir azalmaya neden olmuştur (yaprakların yenilenmesi, yaprak sayısı, kök sayısı, kök uzunluğu, gövde uzunluğu ve biyokütle).

Alyazour v.d. (2020) tarafından gerçekleştirilen bir çalışmada, kirli topraktan potansiyel bir Cr (VI) hiperakümülatör olarak görülen semziotu bitkisi yetiştirirken, toprak organik içeriği, pH ve sülfat konsantrasyonunun, Cr (VI) fito-ekstraksiyonu üzerindeki etkisi araştırılmıştır. Semizotu fideleri, (i) üç organik içerik bileşimi, (ii) altı pH seviyesi ve (iii) altı sülfat tuzu konsantrasyonunda 200 ppm Cr (VI) solüsyonları ile sulanmıştır. Farklı bitki dokularındaki krom konsantrasyonu, değişik koşullar altında izlenmiştir. Sonuçlar, Portulaca oleracea tarafından Cr (VI) alımının (i) düşük organik içerikli toprakta (% 0.42), (ii) hafif alkali pH aralığında (~8) ve (iii) 300–600 ppm arasında sülfat konsantrasyonunda tercih edildiğini göstermiştir.

Bir başka çalışma Hindistan Gujrat, Vadodra'daki tarla alanlarında yetişen iki semizotu türü bitkilerinin hem endüstriyel atık su ile hem de kuyu suyu ile sulanmasının etkilerini araştırmıştır. Sulanan endüstriyel atık sular ve bununla sulanan toprak, kuyu suyu ve bununla sulanan toprağa kıyasla çok yüksek düzeyde ağır metallere (Fe, Zn, Cd, Cr ve As) sahip olduğu tespit edilen çalışmada, sulanan atık topraklarda yetişen her iki türün bitkileri, maksimum köklerde ve en az çiçeklerde olmak üzere tüm bitki kısımlarında yüksek metal birikimi göstermiştir. Portulaca'nın her iki türü birden fazla ağır metal, yani Cd, Cr ve As. *P. tuberosa*'da Cd, Cr ve As'ın toplam sürgün konsantrasyonları (µg g^{-1} km) sırasıyla 1,571, 7,957 ve 3,118 iken, *P. oleracea*'da bunlar sırasıyla 1,128, 7,552 ve 2,476 olduğu tespit edilmiştir (Tiwari v.d., 2008).

Sivakumar v.d. (2020) *Portulaca oleracea* L.'nin gövde kesimlerinin fitoremediasyon verimliliğini araştırmıştır. Dietilen triamin penta asetik asit (DTPA) ile ekstrakte edilebilen Cu ve Cd seviyeleri tüm topraklarda test edilmiştir. Bitkinin sap kesimleri 30 gün büyütüldükten sonra bitki büyüme parametreleri ve Cd ve Cu birikimi test edilmiştir. DTPA ile ekstrakte edilebilen Cu (8.45 ila 35.34 µg / g) ve Cd (0.18 ila 3.10 µg / g) seviyeleri, bölgeler arasında çok farklılık göstermiş ve Cu seviyeleri tüm topraklarda Cd'den daha yüksek çıkmıştır (3,46 ila 135,92 kat daha yüksek). 30 günlük büyümenin ardından, bitkilerin yaprak sayısında (39 ± 2,0 ila 41 ± 1,5 yaprak), kök uzunluğunda (8,0 ± 0,7 ila 8,7 ± 0,7 cm) ve taze ağırlıkta (11.40 ± 0.59 ile 11.73 ± 0.36 g) Cu birikimi Cd'ninkinden daha yüksektir(2,45 ila 5,92 kat). Hesaplanan birikim faktörü (Cu ve Cd'nin topraktan bitkilere taşınması) Cd için (3.01 ila 39.22) Cu'dan (1.24 ila 4.59) daha yüksektir. Toprakta DTPA ile ekstrakte edilebilir Cu ve Cd, bitkilerde Cu ve Cd birikimi ve hesaplanan birikim faktörü arasında net bir eğilim bulunamamıştır.

Sunulan çalışma, ticari olarak satılan semizotu tohumlarıyla bir fitoremediasyon denemesini içermektedir. Çalışma, semizotunun iyi gelişme göstermediği sonbahar mevsiminde sera koşullarında ve toprak organik madde içeriği %5 olacak şekilde saksı deneyleri şeklinde gerçekleştirilmiş, orta seviyede ağır metallerle kirli toprakta yetiştirilen semizotu bitkilerinin ağırlıkları ve bitkilerin yenilebilir kısımlarına aldığı metal konsantrasyonları değerlendirilmiştir.

MATERYAL VE METOD

Çalışmada kullanılan tohum örnekleri yapılan ön çalışmalar ile çimlenme oranının %50 den yüksek olduğu belirlenen piyasadan temin edilen tohumlardır. Tohumların ekildiği toprak ve organik madde içeriğini yükseltmek amacıyla kullanılan keçi gübresi ve torf ise Ege Bölgesi'nde faaliyet gösteren yerel bir peyzaj firmasından temin edilmiştir.

Deneylerde kullanılan toprak karışımının %5 oranında organik madde içermesi için, başlangıç organik madde içeriği %1.8 olan toprağa sırasıyla %31 ve %74 organik madde içeren torf ve keçi gübresi karışımı ilave edilmiştir. Toprak karışımlarının tarla kapasitesi standart gravimetrik yöntemlere göre ölçülmüştür. (Güven vd., 2022). Deney düzeneği her biri 6 kg toprak karışımı içeren 8 L hacimli plastik deneme saksılarında tasarlanmıştır. Çalışmada kullanılan toprak karışımını ağır metaller açısından kirletmek amacıyla Güney Marmara bölgesi'nde terkedilmiş eski bir maden sahasından alınan pasa örneği kullanılmış, 6 kg lık toprak karışmına homojenize hale getirilmiş 350 gram pasa ilave edilerek yeni karışım sağlanmıştır. Saksılar kirletilmiş ve kirletilmemiş (kontrol) toprak karışımları ile 3 er tekrarlı olacak şekilde hazırlanmış ve Dokuz Eylül Üniversitesi Tınaztepe Yerleşkesi'nde bulunan seraya yerleştirilmiştir.

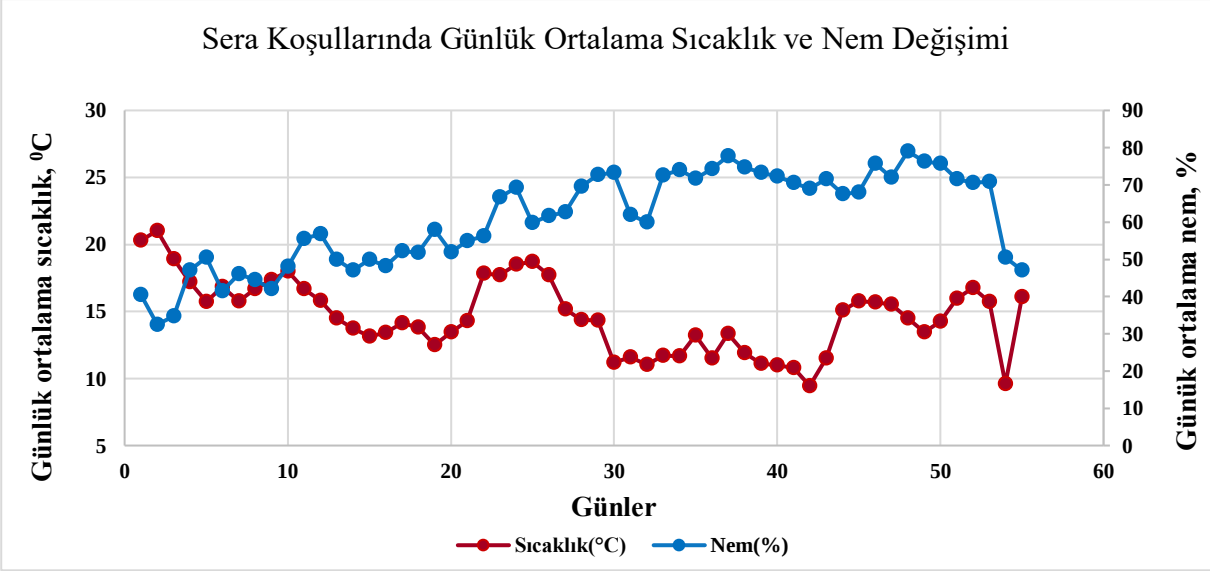
Çalışma Ekim-Aralık 2022 tarihinde gerçekleştirilmiştir. Saksılar seraya yerleştirdikten sonra her saksıya 20 adet tohum ekilmiştir. Bu tohumlar deneysel ekimde kullanılacağı için önce 1/5 oranında seyreltilmiş ağartıcı ve 1 ml Tween 20 eklenmiş su ile tohumların yüzey sterilizasyonu sağlanarak ekim gerçekleştirilmiştir. Ekim bittikten sonra saksılara can suyu verilerek her gün çimlenme takibi gerçekleştirilmiştir. Çimlenme tamamlandıktan sonra semizotu bitkisi ile kuraklık etkisini araştıran bir çalışmada verim üzerine en etkin sulama miktarı olarak belirlenen %75 tarla kapasitesi ile günaşırı sulamalar gerçekleştirilerek bitki büyümeleri değerlendirilmiştir (Güven vd, 2022).

Bitki büyümeleri, bitki sayısı ve bitki ağırlığı ile değerlendirilmiştir. Bitki yetiştirmeden önce toprak örneklerinde ağır metallerin enstrümental analizi için örnekleri asit ile parçalama esasına dayanan ön işlem gerçekleştirilmiştir. Modifiye edilmiş EPA 3050 B Yöntemi kullanılarak gerçekleştirilen parçalama işleminde Perkin Elmer Sample Preparation Block 50-48 kullanılmıştır. Bunun için 0.5 gram toprak örnekleri sırasıyla 5 ml %50 HNO_3, 2.5ml derişik HNO_3, 1.5 ml %30 H_2O_2 (oksidasyon sonuna dek 5 ml yi geçmeyecek şekilde ilaveler ile), ve nihayetinde 5 ml derişik HCl ile muamele edilmiştir. Farklı sürelerde 120 ^{0}C ve 95 ^{0}C sıcaklıklarda gerçekleştirilen bu parçalama sürecinin sonucunda ekstraktlar seyreltilerek okunmaya alınmıştır (USEPA, 1996). Bitki örneklerinde ağır metallerin enstrumental analizi için aynı blok parçalayışı kullanılmış, hava ile kurutularak öğütülen bitki organlarına dair örnekler 0.2 gram olacak şekilde tartılarak konulmuştur. Tüplere 5 ml konsantre HNO_3 eklenmiş ve önceden 110 ^{0}C ye ısıtılmış blok parçalayıcıda 2 saat süreyle ekstrakte edilmiştir. Süre sonunda soğumaya alınan tüplere, sonrasında 1ml %30 H_2O_2 eklenerek 1 saat daha parçalanmaya bırakılmıştır. İşlem sonunda soğuyan numuneler 50 ml'ye seyreltilerek 0.45 µm filtre kağıdından süzülmüş ve analizleri gerçekleşene kadar +4 ^{0}C de saklanmıştır (Ashrafi vd., 2015)

DRAFT

TARTIŞMA

Sera içerisindeki sıcaklık ve nem ölçümleri 27 Ekim – 21 Aralık 2023 tarihleri arasında 55 gün olmak üzere, 30 dakika aralıklarla günde 48 veri alınarak belirlenmiş ve günlük ortalama sıcaklıklar Şekil 1 ile ifade edilmiştir. Bu süre zarfında sera koşullarında en yüksek sıcaklık 36,3 ^{0}C ile 3 Kasım 2023 günü öğle saatleri, en düşük sıcaklık ise 4 ^{0}C ile 21 Aralık 2023 günü sabaha karşı kaydedilmiştir. Benzer şekilde ölçülen nem oranlarına göre, sera koşullarında en yüksek nem oranı %83 ile 14 Aralık 2023 günü belirlenirken, en düşük nem oranı ise %13 ile 28 Ekim 2023 günü kaydedilmiştir (Şekil 1).



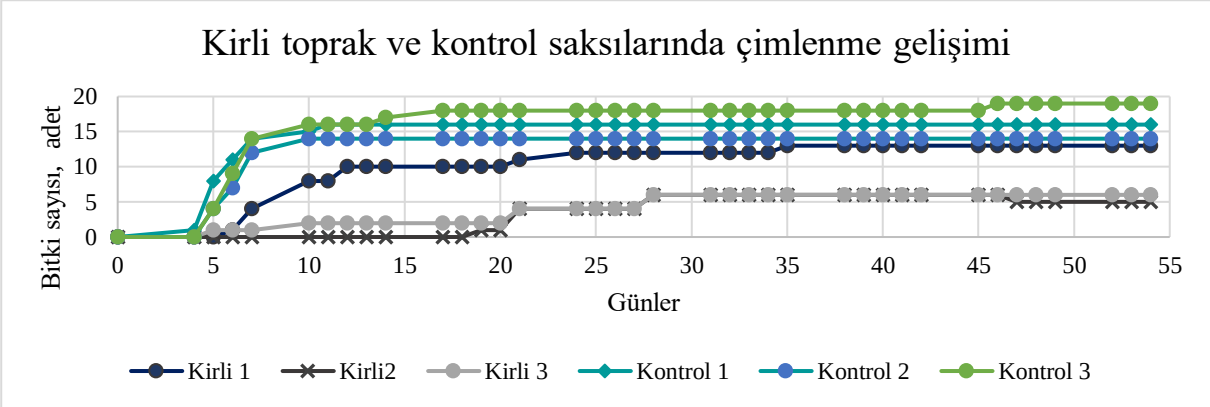
Şekil 1. Deney süreci byunca değişen sıcaklık ve nem salınımı

Saksılardaki ağır metal içeriği Tablo 1 ile ifade edilmektedir. Buna göre kirli saksılarda ortalama Al içeriği 8744 mg/kg iken, kontrol saksılarında bu değer ortalama 8922 mg/kg olarak görülmektedir. Bu durum, yerkabuğundaki doğal Al seviyesinin ortalama 82.300 mg/kg olması sebebiyle kirlilik olarak kabul edilmemektedir. Benzer şekilde, kontrol saksılarındaki Mn seviyesinin ortalama 446 mg/kg konsantrasyonunda olması, bu elementin doğal seviyesinin ortalama 950 mg/kg olması sebebiyle kontrol saksılarında kirliliği ifade etmemektedir (CRC, 2016). Diğer elementler dikkate alındığında, As seviyesi, kirli saksılarda ortalama 25 mg/kg olarak bulunurken kontrol saksılarında 12 mg/kg olarak belirlenmiştir. Bu durum kontrol saksılarında da As kontaminasyonu olduğunu göstermektedir. Cd elementi kirli saksıda 2.28 mg/kg ile kirliliği ifade ederken, kontrol saksısında 0.14 mg/kg ile kontaminasyon olmadığını göstermektedir. Cr değerleri saksılarda ortalama 44 mg/kg seviyesinde olup doğal konsantrasyon olan 102 mg/kg altındadır. Cu konsantrasyonları (kirli toprakta ortalama 69 mg/kg ve kontrol saksılarında ortalama 23.4 mg/kg) maden pasası ile müdahale edilmiş toprakta bu elemente dair önemli bir kontaminasyonu ifade etmemektedir. Benzer şekilde Ni açısından da kontaminasyon sözkonusu değildir. Pb konsantrasyonları, kontamine saksılarda ortalama 229 mg/kg ile ciddi bir kurşun kirliliğine işaret etmektedir. Zn ise ortalama 404.7 mg/kg lık seviyelerde kirliliği ifade etmektedir. Çalışmada kullanılan pasa örneklerinin terk edilmiş Pb-Zn maden sahasından geldiği dikkate alındığında, kontamine saksılarda yüksek Pb-Zn değerleri beklenen bir durumdur.

Tablo 1. Kontamine edilmiş (kirli= ve müdahale görmemiş (Kontrol) topraklardaki ağır metal değişimleri

	Kirli 1	Kirli 2	Kirli 3	Kirli Ortalama ± SS	Kontrol 1	Kontrol 2	Kontrol 3	Kontrol Ortalama ± SS
Al (mg/kg)	8331,67	9063,17	8838,53	8744.45±374.71	8685,18	8832,54	9249,07	8922,26±292,6
As (mg/kg)	25,07	26,72	23,29	25.028±1.72	11,65	11,68	12,59	11.97±0.53
Cd (mg/kg)	1,95	2,87	2,02	2.28±0.51	0,27	0,21	0,14	0.21±0.07
Cr (mg/kg)	42,97	45,19	43,53	43.90±1.16	44,34	43,73	46,21	44.76±1.30
Cu (mg/kg)	63,9	72,83	70,13	68.96±4.58	23,79	23,83	22,59	23.41±0.70
Mn (mg/kg)	1222,09	1597,98	1248,69	1356.26±209.77	439,96	440,65	458,78	446.47±10.67
Ni (mg/kg)	38,25	47,66	44,55	43.49±4.80	45,39	42,75	46,85	44.99±2.08
Pb (mg/kg)	201,73	276,44	208,37	228.85±41.35	9,97	9,61	11,02	10.20±0.73
Zn (mg/kg)	373,33	458,93	381,85	404.70±47.16	56,03	53,91	56,82	55.59±1.50

Saksılardaki çimlenmeler deney süreci boyunca düzenli olarak takip edilmiş olup Şekil 2 ile ifade edilmektedir. Buna göre birbirine paralel olarak hazırlanan kirli toprak saksılarında süreç sonucunda %25-65 (ortalama %40) oranında çimlenme gözlenirken, herhangi bir kirletici müdahalesi olmamış toprak içeren saksılarda bu oran %70-95 (ortalama %82) oranında gerçekleşmiştir. Bu durum, ağır metal varlığının bitki gelişimindeki olumsuz etkisini ifade etmektedir.



Şekil 2. Saksılardaki çimlenme sayıları

Çalışmada mevsime bağlı olarak bitki gelişimi zayıf gözlemlenmiş olup, kirletilmemiş topraktaki bitki gelişimi kirli toprağa göre daha belirgindir. Aynı zamanda mevsimin sonbahar-kış geçişine denk gelmiş olması ve iklim koşulları bitki gelişiminin olumsuzluğu üzerinde en önemli faktördür.

Bitkilerdeki ağır metal konsantrasyonları Tablo 3 ile ifade edilmektedir. Çizelge aynı zamanda, bitkilerin içerdiği ağır metal seviyeleri ile bu metallerin bitkinin yetiştiği topraktaki konsantrasyonları dikkate alınarak hesaplanan n toprak-bitki transfer katsayıları (TC) nı da içermektedir. Transfer katsayısı bir elementin toprak ortamındaki mobilitesini ve biyoelverişliliğini gösteren önemli bir indikatördür (Chen ve Ma, 2001) Bu değer TC= Bitki dokusundaki ağır metal konsantrasyonu / Topraktaki ağır metal konsantrasyonu ile hesaplanmaktadır (Pascual vd., 2004). Bu değerlere göre, As bitkilerde gözlemlenmemiş olup, topraktan bitkiye geçişi yoktur. Al elementinin kirli toprakta transfer katsayısı 0,21 iken yakın Al konsantrasyonuna sahip temiz saksılarda bu katsayı 494 mg/kg Al ile 0,06 olarak bulunmuştur. Cd elementinin kirletilmiş toprakta yetişen bitkide 2,1 ppm seviyesinde bulunması, 0,93 transfer katsayısına denk gelmektedir. Cr,

kirletilmiş toprakta 1,07 transfer katsayısı ile yüksek bir birikime işaret ederken kirletilmemiş toprakta bu değer 0,37 dir. Cu, her iki toprak türünde de 1,63 ve 1,92 TC ile bitkiye geçişi ifade eder. Mn geçişleri düşük oranda seyrederken (kirli toprakta 0,19 ve temiz toprakta 0,34), Ni kirli toprakta 56,6mg/kg ile 1,3 TC olarak bulunmuştur. Kirleticinin asıl bileşeni olan Pb ve Zn sonuçları değerlendirildiğinde ise, Pb kirli ve temiz topraklarda sırasıyla 0,19 ve 0,64 transfer katsayıları ile bitki bünyelerinde birikim göstermektedir. Diğer yandan kirli toprakta yetişen bitkide 528,1 mg/kg (1,3 TC) olarak ölçülürken, temiz toprakta bulunan Zn, bitkide 160,4 mg/kg birikerek 2,88 TC ye ulaşmıştır.

Tablo 2. Bitkilerde biriken ağır metal seviyeleri ve transfer katsayıları

	Kirletilmiş toprakta yetişen bitki (mg/kg)	Transfer katsayısı	Kontrol saksılarında yetişen bitki (mg/kg)	Transfer katsayısı
Al	1824.1	0.21	494.35	0.06
As	0.0	0.00	0	0
Cd	2.1	0.93	0	0
Cr	47.0	1.07	16.70	0.37
Cu	112.2	1.63	45.01	1.92
Mn	264.3	0.19	150.44	0.34
Ni	56.6	1.30	23.69542	0.53
Pb	43.4	0.19	6.525376	0.64
Zn	528.1	1.30	160.3968	2.88

SONUÇ

Çalışma, ülkemizde de yoğun olarak yetişen ve tüketilen semizotu bitkisinin madden pasa atığı ile kirletilmiş toprakta ağır metalleri bünyesine alabilme kapasitesi üzerine bir ön değerlendirme çalışmasıdır. 55 günlük çalışma sonucunda, kirli toprağın bitki büyümesini olumsuz etkilediği gözlemlenmiştir. Topraktan bitkiye ağır metal geçişleri Cr, Cu, Ni ve Zn için transfer katsayısı 1 den büyük olup, anlamlı bir biçimde geçişi ifade etmektedir. Özellikle hareketliliğinin yüksek olduğu bilinen Zn için bu değer bitki gelişiminin daha kuvvetli olduğu kontrol saksılarında 2,88 ile yüksek bir geçişe işaret etmektedir Çalışmanın sonbahar-kış döneminde gerçekleşmiş olması, ve buna bağlı bitki gelişiminin nispeten zayıf olmasına rağmen elde edilen bitkiye geçiş katsayıları, semizotu bitkisinin gelişimi daha yüksek olan bahar-yaz dönemlerinde kirletilmiş topraklardan fitoremediasyon yolu ile bitkiye ağır metal geçişinin geçerli bir arıtma yöntemi olarak kullanılabilirliğini belirtmektedir

TEŞEKKÜR

Çalışma, Dokuz Eylül Üniversitesi BAP Koordinasyon Birimi tarafından kabul edilen FBA-2022-2896 kodlu ve ve "İklim Değişikliğine Uyum Kapsamında Toprak Sağlığı Araştırmaları Canlı Laboratuvarı (LivingLab)" isimli proje ile Avrupa Birliği tarafından desteklenen Prima 2019-11, PRIMA/0009/2019, P2P/PRIMA/1218/0006, 01DH20006, Prima2019-12, STDF Valuefarm projesi tarafından mali olarak desteklenmektedir

KAYNAKLAR

Alam MS, Kaur G, Javed K, Athar M 2007. *Eruca sativa* seeds possess antioxidant activity and exert a protective effect on mercuric chloride induced renal toxicity. Food and Chemical Toxicology, 45: 910-920.

Açak, 2017. Gıda bilinci< https://gidabilinci.com/author/emir-kaboglu , Erişim Tarihi: 24.05.2022

Alyazouri A., Jewsbury R., Tayim H., Humphreys P., Al-Sayah M. H., 2020, Uptake of Chromium by *Portulaca Oleracea* from Soil: Effects of Organic Content, pH, and Sulphate Concentration, Applied and Environmental Soil Science, V:2020, Article ID 3620726 | https://doi.org/10.1155/2020/3620726

Ashrafi A, Zahedi M, Soleimani M 2015. Effect of Co-planted Purslane (Portulaca Oleracea L.) on Cd Accumulation by Sunflower in Different Levels of Cd Contamination and Salinity: A Pot Study. International Journal of Phytoremediation, 17:9, 853-860.

Chen M, Ma LQ 2001. Comparison of three aqua regia digestion methods for twenty Florida soils, Soil Science Society of America Journal, 65:491–499

Covington, MB 2004. Omega-3 Fatty Acids. American Family Physician Journal, 70(1): 133-140.

CRC, 2016, Abundance of elements in the earth's crust and in the sea, CRC Handbook of Chemistry and Physics, 97th edition (2016–2017), p. 14-17

Dweek A C 2001. Purslane (Portulaca oleracea). The Global Panacea, Personal Care Magazine, 4 (2): 7-15.

El-Shamy M , Heikal Y, Bonanomi G, 2019. Phytoremediation Efficiency of Portulaca oleracea L. Naturally Growing in some Industrial Sites, Dakahlia District, Egypt; Chemosphere 225: , DOI:10.1016/j.chemosphere.2019.03.099

Eşiyok D. 2012. Kışlık ve Yazlık Sebze Yetiştiriciliği, Meta Basım Matbaacılık Hizmetleri, s:408

Güven ED, Özmıhçı S, Akinci G, Tümer B, Uyar M 2022. Cultivation of Purslane (Portulaca oleracea) under Soil Stress Conditions. Turkish Journal of Agriculture - Food Science and Technology, 10(sp2), 3014–3021

Pascual I, Antolin MC, Garcia C, Polo A, Sanchez-Diaz M 2004. Plant availability of heavy metals in a soil amended with high dose of sewage sludge under drought conditions, Biology and Fertility of Soils, 40:291-299.

Prabha D, Sivakumar S, Subbhuraam CV, Son HK, 2015, Responses of Portulaca oleracea Linn. to selenium exposure, Toxicology and Industrial Health, 31:5; https://doi.org/10.1177/0748233713475502

Sivakumar S, Prabha D, Velmurugan P, Honga SC, Yi PI, Jang SH, Suh JM, 2020. Phytoremediation of Cu and Cd-contaminated roadside soils by using stem cuttings of Portulaca oleracea L, Environmental Chemistry and Ecotoxicology, 2:201-204.

Tiwari KK, Dwivedi S, Mishra S, Srivastava S, Tripathi RD, Singh NK, Chakraborty S. 2008, Phytoremediation efficiency of Portulaca tuberosa rox and Portulaca oleracea L. naturally growing in an industrial effluent irrigated area in Vadodra, Gujrat, India, Environmental Monitoring and Assessment 147:15–22.

U.S. EPA. 1996. "Method 3050B: Acid Digestion of Sediments, Sludges, and Soils," Revision 2. Washington, DC.

Yurdagül, 2019. Yabani Ve Kültür Semizotu (Portulaca Oleracea L.) Bitkisinin Antioksidan Özelliklerinin İncelenmesi. Aydın Adnan Menderes Üniversitesi, 90

DRAFT

ORAL PRESENTATION

Enfeksiyon hastalıklarında bakteriyofaj aracılı tedavilere güncel bir bakış

Tuğçe Soylamış [1*] (*ORCID: ttps://orcid.org/0000-0002-4663-897X),*
Gülseren Aktaş [2](ORCID: *https://orcid.org/0000-0002-1611-5289)*

[*1]İstanbul Üniversitesi, Sağlık Bilimleri Enstitüsü, Tıbbi Mikrobiyoloji, İstanbul, Türkiye
[2]İstanbul Üniversitesi, Tıp Fakültesi, Tıbbi Mikrobiyoloji, İstanbul, Türkiye

* tugce.soylamis@uskudar.edu.tr

Özet

Bakteriyofajların, antibakteriyel etkinlikleri, Sir Alexander Fleming'in penisilini keşfinden yaklaşık 10 yıl öncesinde ilk 'faj tedavisi' denemeleri ile başlamıştır. Antibiyotiklerin ortaya çıkmasıyla, fajların bakteriyel enfeksiyonların tedavisinde kullanımı azaltmıştır. Fakat antibiyotiklerin, tedavilerde uzun süreli, yaygın ve gelişi güzel kullanımlarından dolayı etkinliklerinin giderek azalması ve bunun sonucunda çoğul antibiyotik dirençli mikroorganizmaların giderek artması, etkili tedavi alternatiflerinin azalmasına yol açmıştır. Ayrıca, bazı bakterilerin oluşturduğu biyofilmler, antibiyotiklerin etkinliğini engelleyip tedavileri güçleştirmektedir. Son yıllarda, yeni antibiyotik geliştirilmediği için, dirençli mikroorganizmaların oluşturduğu enfeksiyonlarda tedavi olanakları azalmaktadır. Bu durum, uygulanabilir alternatif tedavi seçenekleri üzerinde yeni arayışlara zorlamaktadır. Faj tedavisi, şu anda karşı karşıya olunan bakteriyel direnç çağıyla mücadele etmek için önemli bir tedavi alternatifi olabilir. Fajların kısa replikasyon süresi ve çok sayıda yeni faj (virion) oluşturabilme yetenekleri, enfeksiyon etkeni tamamen öldürüldüğünde işlevini durdurması, ökaryot hücrelerde bilinen hiçbir yan etkilerinin olmaması, hızlı ve düşük maliyetli üretimleri enfeksiyon hastalıklarının tedavisinde önemli alternatifler olmasını sağlamıştır. Ayrıca tedavide fajların etkisini artırmak için faj-antibiyotik kombinasyonları veya faj kokteylleri kullanılabilir. İki veya daha fazla farklı fajın oluşturduğu faj kokteylleri, tedavide geniş etki spektrumu sağlar ve direncin ortaya çıkışını geciktirir/engeller. Böylece, tedavilerde daha uzun süre kullanımlarına imkân sağlanır. Biyofilm oluşturan patojen mikroorganizmaların neden olduğu enfeksiyonların güç olan tedavisinde antibakteriyel olarak kullanılabilirler. Bir diğer yaklaşım ise, fajların, özellikle tıbbi alanlarda veya klinik cihazlarda, kontaminasyonları ortadan kaldıran dezenfektan olarak kullanılmasıdır. Bu derleme, günümüzde enfeksiyon hastalıklarında bakteriyofaj aracılı tedavilere ışık tutmak ve önemini gündeme getirmek amacıyla yazılmıştır.

Anahtar Kelimeler: Antibiyotik direnci, bakteriyofajlar, bakteriyofaj tedavisi, kombinasyon tedavi, sinerjistik etki.

Abstract

The antibacterial activity of bacteriophages began with the first 'phage therapy' trials about 10 years before Sir Alexander Fleming's discovered penicillin. With the advent of antibiotics, the use of phages in the treatment of bacterial infections has decreased. However, the gradual decrease in the effectiveness of antibiotics due to their long-term, widespread and indiscriminate use in treatments and the consequent increase in multiple antibiotic resistant microorganisms has led to a decrease in effective treatment alternatives. In addition, biofilms formed by some bacteria hinder the effectiveness of antibiotics and complicate the treatment. In recent years, as no new antibiotics have been developed, treatment options for infections caused by resistant microorganisms are decreasing. This situation forces new searches for viable alternative treatment options. Phage therapy could be an important therapeutic alternative to combat the current era of bacterial resistance. The short replication time of phages, their ability to generate large numbers of new phages (virions), their ability to stop functioning when the infectious agent is completely killed, their lack of known side effects in eukaryotic cells, and their rapid and low-cost production have made them important alternatives in the treatment of infectious diseases. In addition, phage-antibiotic combinations or phage cocktails can be used to increase the effect of phages in treatment. Phage cocktails consisting of two or more different phages provide a broad spectrum of action in treatment and delay/prevent the emergence of resistance. Thus, they can be used in treatments for longer periods of time. They can be used as antibacterials in the difficult treatment of infections caused by biofilm-forming pathogenic microorganisms. Another approach is to use phages as

disinfectants to eliminate contamination, especially in medical areas or clinical devices. This review was written to shed light on bacteriophage-mediated therapies in infectious diseases today and to bring up their importance.

Key Words: Antibiotic resistance, bacteriophages, bacteriophage therapy, combination therapy, synergistic effect.

GİRİŞ

Yaklaşık 100 yıl önce keşfedilen bakteriyofaj (faj), kısa iplikli DNA ya da RNA içeren bakteri virüsleridir. Yirmi - otuz dakikalık yaşam siklusları ile basit bir virion yapısına sahiptirler (1). Bakteriyofaj, eski Yunan dilinde "bakteri yiyen" olarak adlandırılmıştır Bakterilerin geçemediği filtrelerden geçebilen, ultra-mikroskobik organizmalardır. Konak bakteri hücrelerinde çoğalırlar. Bakteriyofajın enfekte ettiği ve ürediği bakteri türleri kendisine spesifiktir (2). Dünya üzerinde yaklaşık 6000 farklı bakteriyofaj türü olduğu düşünülmektedir. Okyanusta en az 10^7 faj ml^{-1} olduğu ve toprak fajlarının sayısının 10^8 virion g^{-1} kadar olabileceği tahmin edilmektedir (3,4).

Bakteriyofajların, antibakteriyel etkinlikleri, Sir Alexander Fleming'in penisilini keşfinden yaklaşık 10 yıl öncesinde ilk 'faj tedavisi' denemeleri ile başlamıştır. Frederick William Twort (5) ve Felix d'Hérelle (6) , sırasıyla 1915 ve 1917 yıllarında birbirinden bağımsız olarak bakteriyofajları keşfetmişlerdir, d'Hérelle özellikle o zamana kadar başarılı bir tedavisi bulunmayan dizanteri gibi enfeksiyonları tedavi etmek için faj süspansiyonlarını kullanmıştır (7). Antibiyotiklerin ortaya çıkmasıyla, fajların önemi azalmasına rağmen, bakteriyel enfeksiyonların tedavisinde fajların kullanımı dünyanın çeşitli bölgelerinde, özellikle Doğu Avrupa ülkeleri ve Rusya'da devam etmiştir. Fajlar, en sıklıkla Gürcistan'da bakteriyel enfeksiyonların tedavisinde kullanılmıştır. d'Hérelle ve George Eliava 1923 yılında, Tiflis'te Eliava Enstitüsü'nü kurmuşlardır. Eliava Enstitüsü'nde, ikinci dünya savaşı sırasında yaralanan askerlerin tedavisinde faj kokteylleri kullanılmıştır. Günümüzde aynı enstitüde, bakteriyofaj uygulamaları devam etmektedir. Ayrıca, enfeksiyonu önleme ve tedavisi, veterinerlik, çevre kontrolü ve gıda güvenliği alanlarında da faj uygulama çalışmaları yapılmaktadır (8). Polonya'nın Wroclaw şehrinde, 2005 yılında, özellikle antibiyotik tedavisine dirençli kronik bakteriyel enfeksiyonları tedavi etmek için fajların kullanıldığı faj terapi merkezi kurulmuştur (9).

Çoğul antibiyotik dirençli mikroorganizmaların giderek artması, tedavide etkili antimikrobiyal alternatiflerinin azalmasına yol açmıştır. Dünya Sağlık Örgütü, 2017 yılında yeni antibiyotiğe ihtiyaç olan küresel dirençli patojenler listesi yayınlamıştır (10). Özellikle hastane kaynaklı ESKAPEE patojenleri olarak adlandırılan vankomisin dirençli *Enterococcus faecium*, metilisin ve vankomisin dirençli *Staphylococcus aureus,* karbapenem dirençli *Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa*, *Enterobacter spp.* ve karbapenem dirençli *Escherichia coli* etkenleri, yeni uygulanabilir alternatif tedavi seçenekleri için arayışları zorlamaktadır. Ayrıca, bazı bakterilerin oluşturduğu biyofilmler, antibiyotiklerin etkinliğini engelleyebilir. Biyofilm oluşumu, çoğu bakteri türleri tarafından hayatta kalmak için antibiyotiklere ve konak bağışıklık sistemine karşı direnci arttırmayı sağlayan önemli bir mekanizmadır (11,12). Fajlar, biyofilmle ilişkili enfeksiyonların tedavisinde geleneksel antimikrobiyallere göre çeşitli avantajlar sağlarlar (13,14). Fajların üretilmesi basittir ve antibiyotiklere göre daha ucuzdur. Mikrobiyotanın korunmasına yardımcı olur ve enfeksiyon etkeni tamamen öldürüldüğünde işlevini durdurur. Antibiyotik alerjisi olan hastalar için kullanılabilir ve ökaryot hücreleri üzerinde litik etkisi yoktur. Ayrıca fajların etkisini artırmak için faj-antibiyotik kombinasyonları veya faj kokteylleri kullanılabilir (15,16).

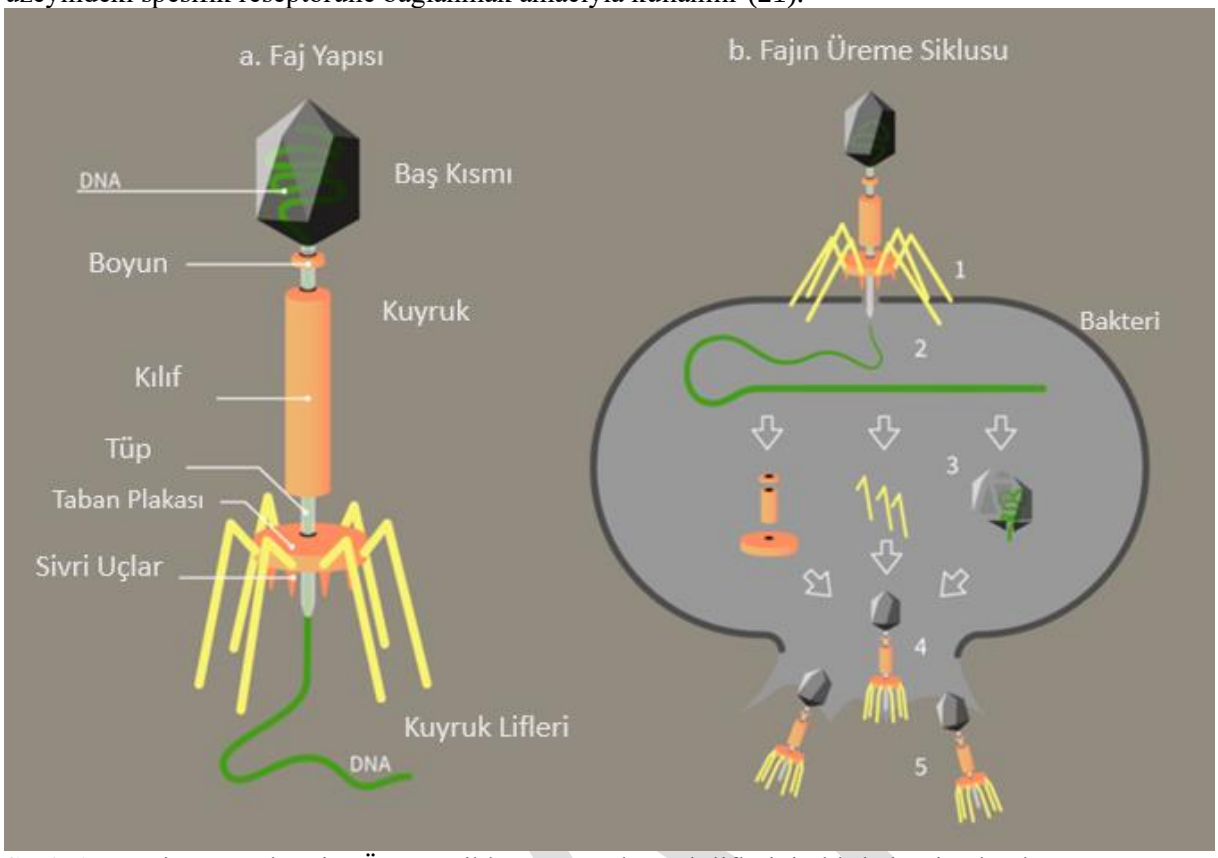
Antibiyotiklerin, bakteri enfeksiyonlarının tedavisinde uzun süreli, yaygın ve gelişi güzel kullanımlarından dolayı etkinlikleri azalmıştır (17). Son yıllarda, yeni antibiyotik geliştirilmediği için, antimikrobiyal dirençli mikroorganizmaların oluşturduğu enfeksiyonlarda tedavi olanakları azalmaktadır. Bundan dolayı farklı etki mekanizmalarına sahip, yeni terapötik ajanların araştırılması gerekmektedir (18). Faj tedavisi, şu anda karşı karşıya olunan bakteriyel direnç çağıyla mücadele etmek için önemli bir tedavi alternatifi olabilir. Bu derleme, günümüzde bakteriyofaj aracılı enfeksiyon hastalıklarının tedavilerine ışık tutmak ve önemini gündeme getirmek amacıyla yazılmıştır.

Bakteriyofaj Yapısı ve Mekanizması

Basit bir faj partikülü, içerisinde genetik materyali bulunan bir baş, boyun, kuyruk ve kuyruk liflerinden oluşmaktadır. Baş kısmı, tek tip bir nükleik asitten (DNA veya RNA) ve genetik materyali koruyan protein yapıda kasılabilen kapsid kılıftan oluşur (Şekil 1) (19). Baş bölgesini kuyruk kısmına bağlayan kısa bir boyun bulunur. Fajların büyük çoğunluğu, konak bakterinin yüzeyindeki spesifik bir reseptörün tanınmasını ve hücre yüzeyine bağlanmasını sağlayan taban plakasına ve kuyruk liflerine sahiptir. Kuyruk, enfeksiyon sırasında

DRAFT

nükleik asidin aktarıldığı bir tüptür (20). Faj kapsidine bağlı olan bu fibrin yapıdaki kuyruk, bakteri hücre yüzeyindeki spesifik reseptörüne bağlanmak amacıyla kullanılır (21).



Şekil 1. a. Faj Yapısı, b.Fajın Üreme Siklusu; 1: Faj kuyruk liflerinin bir bakteriye bağlanması, 2: DNA enjeksiyonu, 3: Faj yapı taşlarının oluşması, 4: Yeni virionların oluşması, 5:Bakteri lizisi ve virionların hücreden salınması (19).

Fajlar, litik ve lizojenik olmak üzere iki viral replikasyon siklusuna sahiptirler. Litik fajlar bakteri hücresini parçalayarak etki gösterirken, lizojenik fajlar hücre içinde latent kalır ve hücreyi hemen eritmezler (22). Litik bir faj enfeksiyonu, fajın kuyruk fibrillerini bakterilerin hücre yüzeyindeki spesifik reseptörlerine adsorbe etmesiyle başlar. Bakteri hücre yapısında çok çeşitli reseptör bulunmasına rağmen, fajlar esas olarak bakteri hücre duvarının bileşenleri üzerinde bulunan hücre yüzeyi yapıları, örneğin, porinler, efluks pompaları, lipopolisakkarit ve peptidoglikan gibi yapılara bağlanır (23). Fajlar, konak hücre reseptörüne bağlanarak genomunu bakteri sitoplazmasına aktarırlar. Transkripsiyon, translasyon ve replikasyon yoluyla kendi yapı taşlarının sitoplazma içerisinde oluşmasını sağlarlar. Bu yapı taşlarının bir araya gelmesiyle virionlar oluşur ve hücrenin lizisine yol açarlar. Serbest kalan yeni litik bakteriyofajlar, yeni bakteri hücrelerini enfekte ederler. Litik bakteriyofajlara benzer şekilde, lizojenik bakteriyofajlar da doğada oldukça yaygındır. Lizojenik bakteriyofaj genomu, konak bakteri tarafından alındıktan sonra konak hücre genomuna entegre olur. Faj genomu, konak hücre genomu ile birleştiği için yeni faj yapı taşları ve virionlar oluşturulmaz. Bundan dolayı faj tedavisinde lizojenik fajlar kullanılmaz sadece litik fajlar kullanılır (24).

Faj Tedavisi Uygulamaları

Faj tedavisi bir asır öncesinden beri kullanılmaktadır. Çoğul antibiyotik dirençli bakterilerin ortaya çıkışı, yeni tedavilere ihtiyaç duyulmasına yol açmıştır. Fajların, kısa replikasyon süresi ve dolayısıyla çok sayıda yeni fajı (virion) hızla oluşturabilme yetenekleri, ökaryot hücrelerde bilinen hiçbir yan etkilerinin olmaması, hızlı ve düşük maliyetli üretimleri enfeksiyon hastalıklarının tedavisinde önemli alternatifler olmasını sağlamıştır (25).

Fajların, patojen bakterilerin meydana getirdiği enfeksiyonları engellemelerinin yanı sıra bir diğer önemli özelliği, biyofilm oluşturan patojen mikroorganizmaların neden olduğu enfeksiyonların tedavisinde antibakteriyel olarak kullanılmasıdır. Bir diğer yaklaşım ise fajların, özellikle tıbbi alanlarda veya klinik cihazlarda kontaminasyonları ortadan kaldıran dezenfektan olarak kullanılmasıdır.

İki veya daha fazla farklı fajın oluşturduğu faj kokteylleri, tedavide geniş etki spektrumu sağlar ve direncin ortaya çıkışını geciktirir/engeller. Böylece tedavilerde daha uzun süre, etkili şekilde kullanımları mümkün olur.

Ayrıca bakteriyofaj kokteyllerinin, *E. coli* , *Salmonella enterica* veya *Listeria* cinsi bakterileri uzaklaştırarak gıdaların dekontamine edildiği gösterilmiştir (26,27).

DRAFT

Bakteriyofaj Kokteyl Tedavisi

Bakteriyofaj monoterapisinin dar spektrumlu olması sorunundan dolayı tek bir faj tedaviyi sağlayamayabilir. Bundan dolayı tedavilerde faj kokteylleri önerilmektedir. Faj kokteyllerinin, monofaj tedavisine kıyasla faja dirençli bakteriyel mutantların ortaya çıkışını azalttığı da gösterilmiştir. Mikroorganizma, tedavi kokteylindeki fajlardan birine karşı direnç geliştirirse, diğer faj antimikrobiyal etkiyi koruyarak bakteriyi enfekte etmeye devam eder. Bir faj kendi spesifik konak reseptörüne bağlandığında, bakteri hücresi bu reseptörün yapısını değiştirerek fajların bağlanma kapasitesini azaltabilir veya engelleyebilir (28). Ayrıca, direnç mekanizmaları faj replikasyon döngüsünün herhangi bir aşamasında da ortaya çıkabilir (29).

Bakteriyofaj kokteyl tedavisi kavramı genişletilebilir; yani her bakteriyofaj, bir kokteylde diğer bakteriyofajlarla birlikte değil sırayla kullanılabilir. Sürekli uygulama sayesinde direnç oluşsa bile direnç geliştirmemiş diğer bakteriyofajlar etki göstermeye devam edecektir. Hayvanlarla yapılan çalışmalarda, bakteriyofaj kokteyllerinin sıralı uygulanması ile, bakteri popülasyonlarını ve bakteriyofaj direncini azaltmada umut verici sonuçları elde edilmiştir (30). Esmael ve ark. (31), yaptıkları çalışmada çoklu ilaca dirençli *Proteus mirabilis* suşunu enfekte eden üç yeni bakteriyofajı izole etmiştir ve çoklu ilaca dirençli *P. mirabilis'in* biyofilmle ilişkili enfeksiyonlarını kontrol etmek ve ortadan kaldırmak için faj kokteylinin bireysel fajlardan daha geniş bir spektrum gösterdiğini bildirmişlerdir.

Bakteriyofaj ve Antibiyotik Kombinasyonu

Faj terapisi ilk olarak 1900'lü yılların başında uygulanmış olsa da fajların terapötik potansiyellerinin tam olarak ortaya çıkarılması zaman almıştır. Başlangıçta, batı dünyasında terk edilmesine rağmen, antibiyotik direncindeki artış, faj tedavisinin yeniden gündeme gelmesine yol açmıştır (32). Klinik olarak, tedavilerde bir antibiyotiğin kullanımı bazen yeterli olmayabilir. Sinerjist etkili antibiyotik kombinasyon tedavisi, monoterapiden daha güçlü tedavi etkisine sahiptir (33). Dirençli enfeksiyonları tedavi etmek için bakteriyofajlar ve antibiyotiklerin kombinasyonu umut vericidir. Faj-antibiyotik kombinasyon tedavisi, hastalığa neden olan bakterilerin antibiyotiklere dirençli olduğu veya antibiyotiklerin enfekte bölgeye yayılımının yeterli olmadığı durumlarda özellikle tercih edilebildiği bildirilmektedir. Fajların bakteriyle etkileşmesi sonucunda, bakterinin antibiyotikle etkileşimi azalır ve böylece antibiyotiğe dirençli bakterilerin ortaya çıkışı gecikir/engellenir. Ancak, çalışmalarda daha fazla klinik veri elde edilmedikçe, fajların antibiyotik yerine kullanılması mümkün olmadığı bildirilmektedir (18).

Comeau ve ark. (34), faj-antibiyotik sinerjistik etkileşimini (FAS), litik bakteriyofajların hızla çoğalmasına ve antibiyotiklerin öldürücü olmayan, düşük konsantrasyonuna (sub-lethal konsantrasyon) bağlamıştır. Yapılan bu çalışmada, antibiyotiğin sub-lethal konsantrosyonunun, bakteri hücrelerinin biyosentez kapasitesini arttırabildiği bildirilmiştir. Böylece litik fajların hızla çoğalması ve hücrelerin hızla parçalanarak fajların antibakteriyel etkinliklerinin artması sağlanır. Aynı çalışmada, bir sefalosporin olan sefotaksimin düşük dozda, üropatojenik bir *E.coli* suşunun wMEP faj üretimini 7 kattan daha fazla arttırdığı rapor edilmiştir. Buna benzer etkileşimin T4 benzeri fajların birlikte olduğu B-laktam, kinolon ve mitomisin C gibi antibiyotiklerin ölüm dozu altındaki konsantrasyonlarındaki çeşitli bakteri-faj sistemlerinde de gözlemlenmiştir. Bu durumun fajlara önemli bir avantaj sağladığı bildirilmiştir.

Yapılan bir çalışmada, nozokomiyal kaynaklı biyofilm oluşturmuş *Pseudomonas aeruginosa* suşu üzerinde faj-antibiyotik etkisi incelenmiştir. Kombine faj-streptomisin tedavisinin, tekli tedaviye kıyasla bakteri yoğunluğunda önemli ölçüde bir azalmaya neden olarak sinerjistik etkileşimi saptanmıştır (35).

Chhibber ve ark. (36), litik bakteriyofajlar ve linezolid kombinasyonunun, diyabetik ayak ülserlerinde antimikrobiyal dirençli enfeksiyonların tedavisinde monoterapiden daha etkili olduğunu bildirmiştir. Oechslin ve ark. (37), siprofloksasin ve bakteriyofaj kokteyllerinin bir kombinasyonunun sıçanlarda deneysel endokardit üzerinde sinerjistik bir etkiye sahip olduğunu bildirmişlerdir.

Bakteriyofajların Anti-biyofilm Etkileşimleri

Biyofilm oluşturabilen nozokomiyal patojenlerle oluşan enfeksiyonların sayısı giderek artmaktadır. Biyofilm içindeki bakteriler, genellikle biyosit ve antibiyotik gibi maddelere karşı yüksek düzeyde direnç gösterebilirler (12,38). Biyofilm matriksinin geçirimsiz olması nedeniyle, biyofilmlerin bakteriyofajlara direnç sağladığı sıklıkla varsayılsa da birçok bakteriyofajın, biyofilmler içindeki bakterileri enfekte edebildiği bildirilmiştir (39). Fajlar, biyofilmin parçalanmasını sağlayan depolimerazlar, endolizinler gibi enzimler üretirler (11). Ayrıca, fajlar tarafından üretilen enzimler biyofilmin dış kısmındaki bakterileri yok ettiğinde, biyofilmin iç kısmındaki mikroorganizmalar metabolik olarak daha aktif hale gelir (artan oksijenizasyon ve besin nedeniyle) ve böylece, bu mikroorganizmalar fajların ve antibiyotiklerin etkisine daha duyarlı hale gelirler (39,40). Ali ve ark. (41), yaptıkları bir çalışmada *Proteus spp.* suşlarına etki eden RP6 ve RP7 fajlarını izole ederek bu fajların özellikle *P.mirabilis* suşunun kristalin biyofilm üretimine etki ettiğini göstermişlerdir. Aynı çalışmada izole edilen fajların, geniş bir pH aralığında (4-10) hem asidik hem de alkali koşullarda iyi bir stabilite

gösterdiği bildirilmiştir. Ayrıca fajın pH değerlerinin dalgalanma yaptığı yerlerde etkili bir terapötik ajan olabilmesi için, geniş bir pH aralığını tolere etmesi gerektiği, böylece başarılı bir anti-biyofilm ve antibakteriyel etkili olabildiği de gösterilmiştir (41).

SONUÇ

Sonuç olarak, günümüzde bakteriyofaj araştırmalarında büyük ilerlemeler kaydedilmiştir. Bakteriyofaja dayanan tedavi seçeneklerinin, gelecekte antimikrobiyal ajanlara dirençli enfeksiyonların tedavilerinde önemli bir alternatif haline gelebileceği düşünülmektedir. Daha fazla araştırma yapılması gereğine rağmen, bakteriyofajların antibiyotiklerle kombinasyonu, sınırlı tedavi seçeneği olan veya antimikrobiyal tedavi imkânı olmayan mikroorganizmaların eradikasyonunda önemli bir tedavi seçeneği olabilir. Bu yaklaşım antibiyotik dozunun düşürülmesini sağlayabilir. Böylece tedavi sırasında antibiyotik direncinin ortaya çıkma potansiyelini de azaltabilir.

KAYNAKLAR

1. O'Flaherty S, Ross RP, Coffey A. Bacteriophage and their lysins for elimination of infectious bacteria. Vol. 33, FEMS Microbiology Reviews. 2009. p. 801–19.
2. Çetin ET. Genel ve Pratik Mikrobiyoloji. 3th ed. İstanbul: Sermet Matbaası; 1973. 671–687 p.
3. Ashelford KE, Day MJ, Fry JC. Elevated abundance of bacteriophage infecting bacteria in soil. Appl Environ Microbiol. 2003;69(1):285–9.
4. Ofir G, Sorek R. Contemporary phage biology: from classic models to new insights. Cell. 2018;172(6):1260–70.
5. Twort FW. An Investigation on The Nature Of Ultra-Microscopic Viruses. Lancet. 1915;186(4814):1241–3.
6. d'Herelle F. An invisible microbe that is antagonistic to the dysentery bacillus. CR Acad Sci. 1917;165:373–5.
7. Abedon ST, Kuhl SJ, Blasdel BG, Kutter EM. Phage treatment of human infections. Bacteriophage. 2011 Mar;1(2):66–85.
8. Kutateladze M. Experience of the Eliava Institute in bacteriophage therapy. Virol Sin. 2015;30(1):80–1.
9. Miedzybrodzki R, Borysowski J, Weber-Dąbrowska B, Fortuna W, Letkiewicz S, Szufnarowski K, et al. Clinical aspects of phage therapy. Adv Virus Res. 2012;83:73–121.
10. WHO. World Health Organzation. Global priority list of antibiotic-resistant bacteria to guide research, discovery, and development of new antibiotics. [Internet]. 2017. Available from: https://www.who.int/news/item/27-02-2017-who-publishes-list-of-bacteria-for-which-new-antibiotics-are-urgently-needed
11. Abedon ST. Ecology of anti-biofilm agents I: antibiotics versus bacteriophages. Pharmaceuticals. 2015;8(3):525–58.
12. Hoiby N, Bjarnsholt T, Givskov M, Molin S, Ciofu O. Antibiotic resistance of bacterial biofilms. Int J Antimicrob Agents. 2010;35(4):322–32.
13. Travis J, Potempa J. Bacterial proteinases as targets for the development of second-generation antibiotics. Biochim Biophys Acta (BBA)-Protein Struct Mol Enzymol. 2000;1477(1–2):35–50.
14. Rasmussen TB, Givskov M. Quorum sensing inhibitors: a bargain of effects. Microbiology. 2006;152(4):895–904.
15. Corban JE, Ramsey J. Characterization and complete genome sequence of Privateer, a highly prolate Proteus mirabilis podophage. PeerJ. 2021;9:e10645.
16. Maszewska A, Zygmunt M, Grzejdziak I, Różalski A. Use of polyvalent bacteriophages to combat biofilm of Proteus mirabilis causing catheter-associated urinary tract infections. J Appl Microbiol. 2018;125(5):1253–65.
17. Carson L, Gorman SP, Gilmore BF. The use of lytic bacteriophages in the prevention and eradication of biofilms of Proteus mirabilis and Escherichia coli. FEMS Immunol Med Microbiol. 2010;59(3):447–55.
18. Morrisette T, Kebriaei R, Lev KL, Morales S, Rybak MJ. Bacteriophage Therapeutics: A Primer for Clinicians on Phage-Antibiotic Combinations. Pharmacotherapy. 2020;40(2):153–68.
19. Guido. Hegasy Phage T4 [Internet]. Wikimedia Commons. 2017. Available from: https://commons.wikimedia.org/wiki/File:11_Hegasy_Phage_T4_Wiki_E_CCBYSA.png
20. Moineau S. Bacteriophage. In: Maloy S, Hughes K, editors. Brenner's Encyclopedia of Genetics (Second Edition). Second Edi. San Diego: Academic Press; 2013. p. 280–3.
21. Ackermann H-W. Tailed bacteriophages: the order Caudovirales. Adv Virus Res. 1998;51:135–201.
22. Wittebole X, De Roock S, Opal SM. A historical overview of bacteriophage therapy as an alternative to

antibiotics for the treatment of bacterial pathogens. Virulence. 2014;5(1):226–35.
23. Bertozzi Silva J, Storms Z, Sauvageau D. Host receptors for bacteriophage adsorption. FEMS Microbiol Lett. 2016;363(4):fnw002.
24. Ling H, Lou X, Luo Q, He Z, Sun M, Sun J. Recent advances in bacteriophage-based therapeutics: Insight into the post-antibiotic era. Acta Pharm Sin B. 2022;12(12):4348–64.
25. Gelman D, Eisenkraft A, Chanishvili N, Nachman D, Glazer SC, Hazan R. The history and promising future of phage therapy in the military service. J Trauma Acute Care Surg. 2018;85(1S):S18–26.
26. Knoll BM, Mylonakis E. Antibacterial bioagents based on principles of bacteriophage biology: an overview. Clin Infect Dis. 2014;58(4):528–34.
27. Pires DP, Melo LDR, Boas DV, Sillankorva S, Azeredo J. Phage therapy as an alternative or complementary strategy to prevent and control biofilm-related infections. Curr Opin Microbiol. 2017;39:48–56.
28. Oechslin F. Resistance development to bacteriophages occurring during bacteriophage therapy. Viruses. 2018;10(7):351.
29. Labrie SJ, Samson JE, Moineau S. Bacteriophage resistance mechanisms. Nat Rev Microbiol. 2010;8(5):317–27.
30. Hall AR, De Vos D, Friman V-P, Pirnay J-P, Buckling A. Effects of sequential and simultaneous applications of bacteriophages on populations of Pseudomonas aeruginosa in vitro and in wax moth larvae. Appl Environ Microbiol. 2012;78(16):5646–52.
31. Esmael A, Abo-Elmaaty SA, Khafaga EM, Abdelrahman S, Hassan MG. Efficacy of three lytic bacteriophages for eradicating biofilms of multidrug-resistant Proteus mirabilis. Arch Virol. 2021;166:3311–22.
32. Duckworth DH. Who discovered bacteriophage? Bacteriol Rev. 1976;40(4):793–802.
33. Worthington RJ, Melander C. Combination approaches to combat multidrug-resistant bacteria. Trends Biotechnol. 2013;31(3):177–84.
34. Comeau AM, Tétart F, Trojet SN, Prère M-F, Krisch HM. Phage-antibiotic synergy (PAS): β-lactam and quinolone antibiotics stimulate virulent phage growth. PLoS One. 2007;2(8):e799.
35. Torres-Barceló C, Arias-Sánchez FI, Vasse M, Ramsayer J, Kaltz O, Hochberg ME. A window of opportunity to control the bacterial pathogen Pseudomonas aeruginosa combining antibiotics and phages. PLoS One. 2014;9(9):e106628.
36. Chhibber S, Kaur T, Kaur S. Co-therapy using lytic bacteriophage and linezolid: effective treatment in eliminating methicillin resistant Staphylococcus aureus (MRSA) from diabetic foot infections. PLoS One. 2013;8(2):e56022.
37. Oechslin F, Piccardi P, Mancini S, Gabard J, Moreillon P, Entenza JM, et al. Synergistic interaction between phage therapy and antibiotics clears Pseudomonas aeruginosa infection in endocarditis and reduces virulence. J Infect Dis. 2017;215(5):703–12.
38. Pamp SJ, Gjermansen M, Johansen HK, Tolker-Nielsen T. Tolerance to the antimicrobial peptide colistin in Pseudomonas aeruginosa biofilms is linked to metabolically active cells, and depends on the pmr and mexAB-oprM genes. Mol Microbiol. 2008 Apr 1;68(1):223–40.
39. Harper DR, Parracho HMRT, Walker J, Sharp R, Hughes G, Werthén M, et al. Bacteriophages and biofilms. Antibiotics. 2014;3(3):270–84.
40. Chaudhry WN, Concepcion-Acevedo J, Park T, Andleeb S, Bull JJ, Levin BR. Synergy and order effects of antibiotics and phages in killing Pseudomonas aeruginosa biofilms. PLoS One. 2017;12(1):e0168615.
41. Ali S, Karaynir A, Salih H, Öncü S, Bozdoğan B. Characterization, genome analysis and antibiofilm efficacy of lytic Proteus phages RP6 and RP7 isolated from university hospital sewage. Virus Res. 2023;326:199049.

ORAL PRESENTATION

Analysis of *nDart1-0* DNA transposon by qRT-PCR in rice cultivars under drought stress

Ebru Derelli Tufekci[1*] (*ORCID: https://orcid.org/* 0000-0003-1097-8574)

[*1]Cankiri Karatekin University, Food and Agriculture Vocational School, Department of Field Crops, Cankiri, Turkey

Corresponding author e-mail: ebru.derelli@gmail.com

Abstract

Stress can be defined as factors that alter or disrupt metabolic homeostasis in plants, causing any change in the growth and development of the plant in its habitat. Stress conditions adversely affect the physiological functions of plants, leading to a decrease in their biosynthesis capacity, changes in their normal functions and causing great damage that can cause the death of the plant. Drought, which is one of the abiotic stress factors, is defined as "the lack of necessary and sufficient moisture for a plant to complete its normal growth and life cycle" and is considered to be the most destructive stress factor that reduces crop productivity in agricultural production compared to other environmental stresses. Transposons are sequences that can move to different locations in the genome of a cell through a process called transposition. Transposons are activated by various biotic and abiotic stresses and it has been determined that these activated transposons are associated with reactions that trigger the biosynthesis of secondary metabolites in the plant defence system, stimulation of signalling pathways. In this study, the gene expression level of *nDart1-0* DNA transposon, which is one of the transposons with important effects on gene function and genome, was analysed by qRT-PCR in Osmancık-97 and Halilbey wheat cultivars, which were drought tolerant and sensitive to drought stress, respectively, in order to understand the mechanism of drought stress. In the qRT-PCR study, it was observed that the gene level of *nDart1-0* DNA transposon increased and this transposon was active in Osmancık-97 and Halilbey rice varieties as a result of drought stress. In addition, the presence of this transposon in rice before is thought to be a transition between these species. In order to further understand the activity of these transposons, it will be more efficient to investigate them in different species.

Keywords: Drought, gene expression, *Oryza sativa*, transposon

INTRODUCTION

Since the optimum environmental requirements for the growth, development and survival of each plant species are different, the survival of plants under stressful conditions is possible by providing optimum conditions. Every abnormal change in the environmental conditions in which plants live adversely affects plant growth and development and gives rise to the concept of stress. Stress can be defined as factors that alter or disrupt metabolic homeostasis in plants and cause any change in the growth and development of the plant in its habitat (Shulaev et al., 2008). Stress conditions can negatively affect the physiological functions of plants, reducing their biosynthesis capacity, altering their normal functions and causing plant death. cause damages (Lichtenhaler, 1996). Drought, which is one of the abiotic stress factors, is defined as "the lack of necessary and sufficient moisture for a plant to complete its normal growth and life cycle" and is considered to be the most destructive stress factor that reduces crop productivity in agricultural production compared to other environmental stresses (Lambers et al., 2008). The negative impact of drought on the plant depends not only on the severity and duration of the drought, but also on the stage of development of the drought-exposed plant. While the growth and development of plants exposed to drought at the young stage is affected, crop yield is affected in plants exposed to drought during flowering or seed setting (Çırak and Esendal, 2006). Drought stimulates many adaptive physiological, molecular and biochemical responses in plants depending on its duration, its relationship with other types of stress, the genotype and developmental stage of the stressed plant. The elucidation of molecular control mechanisms related to drought stress is based on the expression of specific stress-related genes (Wang et al., 2003). These genes can be broadly classified as early and late

induced genes. Early genes are induced as soon as the stress signal is detected and are generally expressed for a short period of time. Many transcription factors, protein kinases, phospholipases and phospholipases are included in the group of early genes and the induction of these genes does not require new protein synthesis and signaling compounds are always readily available. Genes that are activated very slowly by stress belong to the group of late-induced genes and their expression is usually continuous. Water channel proteins, enzymes involved in the biosynthesis of osmotic protectors (sugars, proline, glycine betaine), LEA (late embryogenesis abundant) proteins, chaperones, proteins that protect macromolecules and membranes such as mRNA binding proteins, proteases, detoxification enzymes (Cramer et al., 2007, Verelst et al., 2013, Todaka et al., 2015).

Transposons are sequences that can move to different locations in a cell's genome through a process called transposition. Transposons change the expression of genes in their new locations on chromosomes. As a result, they affect existing genetic data and cause significant changes in gene expression. In addition, DNA transposons and retrotransposons, which proliferate by replication, lead to genome expansion. Transposons induce events such as insertions, inversions, deletions and duplications, leading to mutations and the formation of new alleles. These new formations are seen as an effective mechanism for the emergence of new species in the evolutionary process (Mao et al., 2015). Transposons regulate gene expression both through the influence of methyl groups on neighboring regions and by interfering with transcription through the RNAi (RNA interference) mechanism. Through this epigenetic gene regulation, transposons are involved in many mechanisms ranging from tissue differentiation during embryonic development to the maintenance of differentiation of different tissues in the adult (Jurka, 2008). In addition, some transposons were found to be activated by various biotic and abiotic stresses and it was revealed that these activated transposons were related to the plant defense system, stimulation of signaling pathways and reactions that trigger the biosynthesis of secondary metabolites.

Rice (*Oryza sativa* L.) is the plant from which rice, the most important staple food worldwide, is obtained. It is a member of the Wheat family (Poaceae). With an annual production of 400 million tons, it is the most widely cultivated irrigated cereal in the world. Rice provides 21% of global per capita human caloric needs and 15% of per capita protein. There is a rich genetic diversity in cultivated rice varieties, but this alone is not sufficient for yield increase. For this reason, it is important to study and understand DNA transposons that can alter the organization of the genome in order to increase productivity and to identify and develop rice varieties resistant to abiotic and biotic stresses (Baloch et al., 2015). Recently, a lot of focus has been placed on DNA transposons that result in changeable alleles in plants and how to use transposon tagging as a functional genomic tool to clarify the function of key genes (Kunze and Weil, 2002). A large part of the rice genome is composed of transposons. Since active excision/reintegration of these mobile elements may result in harmful genetic changes, many transposons are maintained in a genetically or epigenetically inactivated state. However, some non-autonomous DNA transposons of the *nDart1-3* subgroup, including *nDart1-0*, actively transpose in specific rice lines, such as pyl-v which carries an active autonomous element, aDart1-27, on chromosome 6.

The aim of this study was to investigate the expression level of *nDart1-0* DNA transposon in drought-tolerant Osmancık-97 and drought-sensitive Halilbey rice cultivars in leaf tissues under drought stress by qRT-PCR.

MATERIALS AND METHODS

Plant Material

Pots measuring 14x13 cm were cleaned and sanitized before being used to grow seeds of two distinct varieties of rice (Osmancık-97 and Halilbey). The seeds of the genotypes were sterilized three times with deionized water (dI-H2O) after being exposed to 5% sodium hypochlorite for 10 minutes. Three-wheeled plastic pots measuring 30 x 50 inches in diameter and filled with 1 kilogram of soil were used to plant the seeds of the different types. After all, until the period of 6-7 leaves, germination of the seeds was irrigated at the field's capacity. Irrigation was stopped so that the group may receive a therapy for drought stress. When the control group began to exhibit signs of stress after two weeks of watering at the field capacity, the irrigation group stopped. At the end of the 15th day of the stress application, leaf samples were taken randomly from 3 different pots containing both control and treatment plants and placed in separate transparent bags, quickly preserved in liquid nitrogen and the materials were placed in -80°C deep freezer for RNA isolation studies.

DRAFT

RNA Isolation Method

RNA isolation from leaf tissues was performed according to the TRIzol method. 1 ml TRIzol reagent was added to 2 ml ependorf tubes and kept ready at 4ºC. Approximately 100 mg of each root and leaf sample was taken and crushed in sterile mortars using liquid nitrogen and the powdered samples were quickly transferred to the tubes. The tubes were mixed by turning the tubes up and down rapidly and a homogeneous mixture was obtained. The tubes were kept at room temperature for 10 min. For 1 ml TRIzol, 0.2 ml chloroform was added to the tubes. The tubes were well capped and mixed by shaking vigorously by hand for 15 sec. After 5 min at room temperature, the mixture was centrifuged at 15,000 rpm for 17 min at 4°C. After transferring the upper liquid phase to new tubes, 500 µl of isopropyl alcohol (half the amount of TRIzol) was added to these tubes for RNA precipitation. After 10 min at room temperature, the tubes were centrifuged at 12,000 rpm for 10 min at 4°C and the supernatant was discarded. Precipitated RNA was washed with 1 ml of 75% ethanol. Samples were centrifuged at 10,000 rpm for 5 min at 4°C. The supernatant was discarded and the bottom RNA precipitate was allowed to dry. 30-70 µl ddH2O was added to the tubes and kept in the heat block at 57°C for 10 min to dissolve the RNAs. The quality and quantity of isolated RNAs were checked on 2% agarose gel and nano-drop spectrophotometer. The RNA samples were then removed to -80°C for use.

cDNA Synthesis and qRT-PCR Analysis

Fermentas Kit (Fermentas, Ontario, Canada) was used to obtain cDNA from RNA extracted from leaf tissue to determine the expression level of the *nDart1-0* transposon by simultaneous qRT-PCR. To start the study with equal amounts of RNA, the amount of RNA to be used was calculated as 1000 ng of total RNA from each sample. For 1X (1 sample), 1 µl Oligo dT, 1000 ng RNA and water were added to a total volume of 11 µL and the tubes were incubated in the PCR device at 65°C for 5 min. After this time, the tubes were immediately buried in ice and kept on ice for 2 min. Then, 4 µL 5X Buffer, 2 µL 10 mM dNTP, 1 µL RNase out, 2 µL Reverse Transcriptase enzyme were added to each tube and the total volume was completed to 20 µL. The PCR device was placed at 37°C for 60 min, 70°C for 5 min and the reaction was terminated at 4°C. Then, PCR optimization of all cDNAs generated with 18S rRNA primers was performed and the formation of cDNAs was checked by running on a 1.5% agarose gel. After controlling the cDNA reaction, the single-stranded cDNAs were amplified in the Thermo Scientific PikoReal Real-Time PCR System using primers, SYBR Green I Master Mix (Roche Applied Science, Penzberg, Germany) and after each cycle of PCR, the signals given by the fluorescent dye increasing in proportion to the amount of product formed were analyzed.Each sample was prepared in triplicate to minimize experimental errors.The 18S gene was used as a control for normalization of the expression levels of the samples.For qRT-PCR assay 1X, 18 µL of the mixture prepared using 0.1 µL forward primer, 0.1 µL reverse primer, 10 µL 2X Fast SYBR Green Master Mix (Roche, Cat no: 4385612) and 7.8 µL nuclease-free water was placed separately in each reaction well of the 96-well plate, 2 µL of the cDNA products to be measured were added and placed in the qRT-PCR device.

RESULTS and DISCUSSION

In general, transposons are divided into two groups, retrotransposons and DNA transposons, according to their mechanism of transposition in the genome. Retrotransposons are restricted to LTR ("Long Terminal Repeat") sequences, while DNA transposons are restricted to TIR ("Terminal Inverted Repeat") sequences and play an important role in transposition. Considerable attention has recently been given to DNA transposons causing mutable alleles in plants for utilizing transposon tagging as a functional genomic tool to elucidate the function of genes of interest (Johzuka-Hisatomi et al. 2008). While endogenous active DNA transposons, which are free from somaclonal variation because no tissue culture is involved in generating insertion mutants, have been extensively used for gene tagging in maize, snapdragon, petunia, and morning glories, only a few active endogenous DNA transposons, *mPing*, *nDart1*, *dTok*, and *nDaiZ*, have been identified in rice (Huang et al., 2009).

In this study, expression levels of *nDart1-0* DNA transposon in drought-tolerant Osmancık-97 and drought-sensitive Halilbey rice cultivars were analyzed under drought stress and its relationship with drought mechanism was examined. Drought stress conditions were applied and increases were observed in the expression level of *nDart1-0* transposon in the treatment group Osmancık-97 and Halilbey varieties due to drought stress compared to the control group (Figure 1).

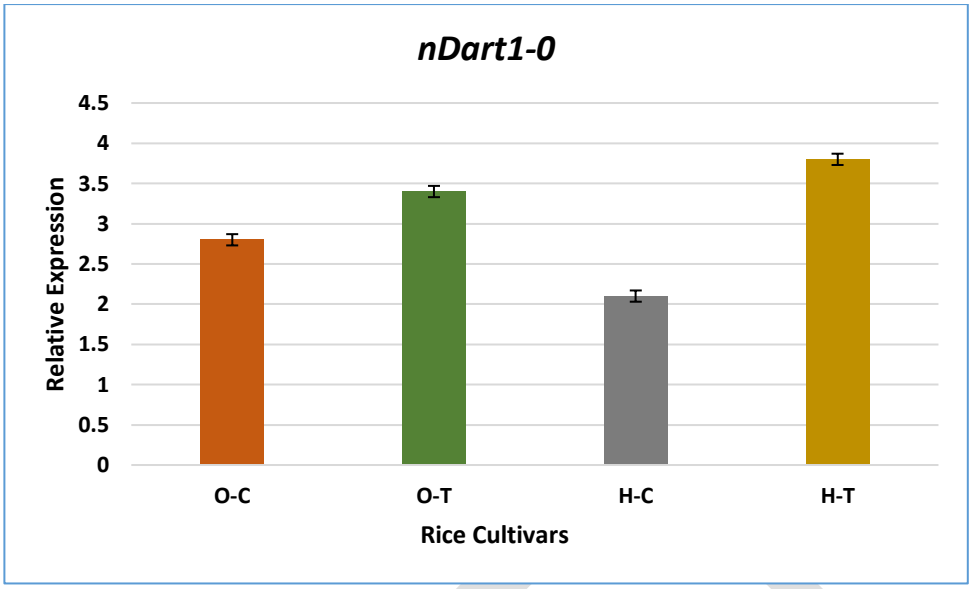


Figure 1. Expression level of *nDart1-0* DNA transposon in Osmancık 97 and Halilbey rice varieties (O=Osmancık 97, H=Halilbey, C=Control, T=Treated)

CONCLUSION

Understanding the molecular, cellular and physiological adaptation to drought and high temperature conditions is possible by revealing the signal transduction mechanisms in plants, the perception of stress signals, the activation of many transcription factors and other regulators, and the expression products of genes. Drought and high temperature stress tolerance is a complex event in which structural and functional changes in the cell membrane, tissue water content, composition of proteins, lipids, primary and secondary metabolites are controlled by the participation of many genes. Transposons are mobile elements that provide considerable dynamism to the genome and make up the bulk of the genome. In their new locations on the chromosomes they cause significant changes in the expression of genes. It is thought that the information obtained as a result of this study will make important contributions in terms of determining the molecular mechanism of abiotic stress tolerance in plants, identifying the related genes and molecular markers and using these markers in advanced plant breeding studies.

REFERENCES

Baloch FS, Derya M, Andeden EE, Alsaleh A, Cömertpay G, Kilian B, Özkan H 2015. Inter-primer binding site retrotransposon and inter-simple sequence repeat diversity among wild Lens species. *Biochemical Systematics and Ecology*, 58: 162-168.

Cheng J, Zhao Z, Li B, Qin C, Wu Z, Trejo-Saavedra DL, Luo X, Cui J, Rivera-Bustamante RF, Li S, Hu K 2016. A comprehensive characterization of simple sequence repeats in pepper genomes provides valuable resources for marker development in Capsicum. *Scientific Reports*, 6: 18919.

Çırak C, Esendal E 2006. Soyada kuraklık stresi. *Ondokuz Mayıs Üniversitesi Ziraat Fakültesi Dergisi*, 21(2): 231-237.

Cramer G, Ergül A, Grimplet J, Tillett R, Tattersall E, Bohlman M, Vincent D, Sonderegger J, Evans J, Osborne C, Quilici D, Schlauch KA, Schooley DA, Cushman JC 2007. Water and salinity stress in grapevines: early and late changes in transcript and metabolite profiles. Functional and Integrative *Genomics*, 7(2): 111-134.

Huang J, Zhang K, Shen Y, Huang Z, Li M, Tang D, Gu M, Cheng Z 2009. Identification of a high frequency transposon induced by tissue culture, nDaiZ, a member of the hAT family in rice. *Genomics* 93: 274-281

Johzuka-Hisatomi Y, Maekawa M, Takagi K, Eun C–H, Yamauchi T, Shimatani Z, Ahmed N, Urawa H, Tsugane K, Terada R, Iida S 2008. Homologous recombination-dependent gene targeting and an active

DNA transposon nDart-promoted gene tagging for rice functional genomics. In: Hirano H-Y, Hirai A, Sano Y, Sasaki T (eds) Rice Biology in the Genomics Era: Biotechnology in Agriculture and Forestry, vol 62. Springer, Berlin, pp 81-94.

Jurka J, Kapitonov VV, Pavlicek A, Klonowski P, Kohany O. Walichiewicz J 2005. Repbase update, a database of eukaryotic repetitive elements. *Cytogenetics and Genome Research*, 110: 462-467.

Kunze R, Weil CF 2002. The hAT and CACTA superfamilies of plant transposons. In: Craig NL, Craigie R, Gellert M, Lambowitz AM (eds) Mobile DNA II. American Society for Microbiology Press, Washington, DC, pp 565-610.

Lambers H, ChapinIII FS, Pons TL 2008. Plant water relations, plant physiological ecology (2nd edn). Springer, pp 163-223, New York.

Lichtenhaler, HK 1996. Vegetation stress: an introduction to the stress concept in plants. *Journal of Plant Physiology*, 148(1-2): 4-14.

Mao H, Wang H, Liu S, Li Z, Yang X, Yan J, Li J, Phan Tran, LS. Qin F 2015. A transposable element in a NAC gene is associated with drought tolerance in maize seedlings. *Nature Communications*, 6: 8326.

Shulaev V, Cortes D, Miller G, Mittler R 2008. Metabolomics for plant stress response. *Physiologia Plantarum*, 132: 199-208.

Todaka D, Shinozaki K, Yamaguchi-Shinozak K 2015. Recent advances in the dissection of drought-stress regulatory networks and strategies for development of drought-tolerant transgenic rice plants. *Frontiers in Plant Science*, 6, 84.

Verelst W, Bertolini E, De Bodt S, Vandepoele K, Demeulenaere M, Enrico Pè M, Inzé D 2013. Molecular and Physiological Analysis of Growth-Limiting Drought Stress in Brachypodium distachyon Leaves. *Molecular Plant*, 6(2): 311-322.

Wang W, Vinocur B, Altman A 2003. Plant responses to drought, salinity and extreme temperatures: towards genetic engineering for stress tolerance. *Planta*, 218(1): 1-14.

DRAFT

ORAL PRESENTATION

Decomposition of Jerusalem Artichoke using HNO_3 as catalyst

Nihal CENGİZ*[1] (*ORCID: https://orcid.org/*0000-0002-6572-7046)
Levent BALLİCE[1] (*ORCID: https://orcid.org/*0000-0002-3137-1352)

[1] Ege University, Faculty of Engineering, Department of Chemical Engineering, İzmir, Turkey.

*Corresponding author e-mail: nihal.cengiz@ege.edu.tr

Abstract

The production of bio-based chemicals based on renewable resources has become increasingly important in recent years. Chemicals synthesized based on raw materials produced from fossil resources necessitated the search for new resources and production technologies in terms of both the limitation of the reserve and the negative effects of the production process on
the environment. Lignocellulosic biomass is seen as an important raw material due to their abundance and economical nature. Hydrothermal decomposition of glucose and fructose which is one of the building blocks of cellulose and hemicellulos, which are basic components of biomass, under subcritical conditions of water into valuable chemicals is one of the important methods. In this study, Jerusalem artichoke, which is lignocellulosic biomass rich in inulin, was used as raw material. Hydrothermal decomposition experiments were carried out in batch- operated stirred reactors and in a temperature-programmed (heating speed adjustable PID unit) system. In experimental studies, the effect of reaction time, and catalyst on the conversion of the platform chemicals in the presence of mineral acid (HNO_3) was investigated. Liquid products determined in HPLC device; levulinic acid, hydroxymethyl furfural, acetic acid, and formic acid. The product yields were calculated by determining the amounts of these compounds and sugar monomers (glucose, fructose, arabinose, rhamnose, cellobiose, sucrose, mannose, xylose, galactose). It was determined that nitric acid was effective in hydrolyzing lignocellulosic biomass, but the organic acid yield was low.

Keywords: Jerusalem artichoke, levulinic acid, biomass decomposition, hydrolysis, HNO_3

INTRODUCTION

The rapid decrease in fossil fuel resources such as oil, natural gas and coal and the environmental pollution resulting from the use of these fuels have accelerated the search for renewable alternative energy sources and the production of valuable chemicals from these resources. Lignocellulosic biomass; It is one of the alternative energy sources as it is abundant, cheap and renewable. Only 5% of all chemicals produced today are obtained from renewable sources. Only 3-4% of the biomass produced in the world is consumed in food and non-food areas. The energy content of the unused part is 8 times the total energy consumed in the world. Intensive studies are being carried out on converting this great potential into liquid and gaseous fuels and various chemicals through chemical and biochemical processes. It is thought that biomass may be an alternative to fossil-based raw materials in the future.

Biomasses mainly contain cellulose, hemicellulose and lignin. With the hydrolysis, dehydration and rehydration of cellulose and hemicellulose in lignocellulosic biomass in acidic environment, many chemical substances are formed depending on the reaction conditions, especially 5-HMF, levulinic acid (LA), furfural and formic acid (D.W. and W.O. 2011; Signoretto et al. 2019; Werpy and Petersen 2004).

BASE. Department of Energy (DOE) studies have identified 300 candidate chemicals that can be produced from lignocellulosic biomass. By examining their production processes, the number of potentially important chemicals was reduced to 30, and a list of 12 leading chemicals (Building Blocks) was subsequently published in 2004. These chemicals will form the basis of biorefineries, which are planned to replace petrochemical refineries in the very near future. One of these 12 chemicals is stated as Levulinic Acid (LA).

Among some polysaccharides and monosaccharides, the highest efficiency in the conversion process to 5-HMF by acidic hydrolysis was achieved in studies conducted with fructose. Inulin, as an oligofructosaccharide

(polysaccharide), can be converted first into 5-HMF and then into Levulinic acid with an easier and higher conversion compared to cellulose. Inulin-rich biomass has a significant potential for levulinic acid production (Bedzo, Mandegari, and Görgens 2020; Fachri, Rasrendra, and Heeres 2015; Qiu et al. 2018; Sarchami and Rehmann 2015; Wang et al. 2015).

MATERIALS AND METHODS

Jerusalem artichoke was obtained and pre-dried for 3 days at 40°C in a Fischer Brand air circulating tray dryer in order to clean, roughen, dry and then make the samples suitable for grinding in the mill. Dried samples were ground in an MF-IKA Brand rotor grinder, and samples ground to <1 mm grain size by sieve analysis were reserved for use in subsequent studies. The samples, which were pre-dried and grinded, were first subjected to moisture analysis. In this context, the process was continued until a constant weight was reached in the oven kept at 105°C and the amount of moisture was determined. In addition, the samples were burned in a muffle furnace using the standard ash determination method and their ash content was determined. Lignocellulosic biomass has been used to obtain valuable water-soluble organic compounds under subcritical water conditions (below 374°C and 221 atm).

Hydrothermal cracking experiments were carried out in batch-operated stirred reactors. Feed solution of different concentrations was placed in reactors with an internal volume of 50 cm^3, tightly closed and purged with nitrogen gas. The reactors were heated to the desired reaction temperature in a temperature-programmed system (with a PID unit with adjustable heating rate), and were kept at this temperature for a 120 minutes' reaction time, and samples were taken to be analyzed at different time periods.



Figure 1. Batch, stirred and temperature-controlled reactor system

The purpose of selecting Jerusalem artichoke as a biomass source for valuable chemical production is the high inulin content taht belongs to extractive part as displayed in Table 1. Inulin is a naturally occurring polysaccharide found in many plants, particularly in the roots or tubers of certain species like chicory, Jerusalem artichoke, and dandelion. It is composed of linear chains of fructose molecules linked together by β(2→1) glycosidic bonds, with a terminal glucose unit. Inulin content can vary widely among plant species and even within different parts of the same plant. In general, plants that are known for their high inulin content, such as chicory and Jerusalem artichoke, can have inulin levels ranging from 15% to 20% or more in their dry biomass. However, this can vary significantly.

Inulin can be hydrolyzed and further processed to produce levulinic acid. Levulinic acid is a platform chemical that has applications in the production of biofuels, pharmaceuticals, and as a building block for various chemicals and plastics (Bhagia et al. 2017; Galletti et al. 2012; Gunnarsson et al. 2014; Sarchami and Rehmann 2015; Vassilev et al. 2010; Viet Bui et al. n.d.).

DRAFT

Table 1. Proximate, Elemental Analysis and Structural Constituents of Jerusalem Artichoke

Proximate Analysis (wt. %)	
Humidity	6.5
Ash	11.82
Structural Constituents (on dry and ashless basis %)	
Cellulose	7.82
Lignin	3.16
Hemicellulose	6.98
Extractives	
Elements (wt. %)	
C	42.3
H	6.4
N	1.76
S	-
O	49.2

Table 2 - Method properties in HPLC instruments for the analysis of organic acids and 5-HMF (Method I) and sugar compounds (Method II).

	Method I	**Method II**
Colon type	HPX-87H column	HPX-87H column
Mobile phase	5 mM H_2SO_4	5 mM H_3PO_4
flow rate of mobile phase	0.6 mL/minute	0.6 mL/minute
Detector type	Refractive Index (RID)	Refractive Index (RID)
Column Temperature	30°C	30°C
Detector Temperature	30°C	40°C
Injection Volume	20 µL	20 µL

RESULTS and DISCUSSION

Jerusalem artichoke decomposition in the presence of HNO_3 was investigated in this studty. Biomass/solvent ratio is selected as parameter and runs were carried out at three concentration levels of 1g/22 mL (0.0454 g/ml), 1.5 g/22 mL (0.068 g/mL) and 2 g/22mL (0.091 g/mL). The yields of the compounds in the aqueous product were determined by HPLC instrument. The methods conditions are given in Table 2. Samples were taken in during the reaction to evalute the effect of reaction time as 20, 40, 60 and 120 min. The variation of the yields of the compounds are represented in Figure 3 and Figure 4. The biomass/solvent ratio effects the organic acid and 5-HMF yields, the highest total organic acid and 5-HMF yield decreased as the concentration increases slighlty and the the highest yields are reached at different times. For the most diluted feed medium, 11.5 % total organic acid yields were obtained at 60 min while at moderate concentrated medium 11.0 % was achieved at 120 min of reaction time. The yields are lower in the high concentration run for organic acids (D.W. and W.O. 2011; Kang, Fu, and Zhang 2018; Mukherjee, Dumont, and Raghavan 2015) . Sugar compounds are the main products since they have 51.8 % total yield as highest in the most diluted feedstock run in the earlier times of reaction (20 min) and decreases while the reaction proceeds but still had high yields of sugar compunds. This shows a further decomposition of the sugar compunds exist in the reaction medium required to obtain organic acids in higher yields. It was determined that nitric acid was effective in hydrolyzing lignocellulosic biomass, but the organic acid yield was low. This decrease in the yield of target chemicals is due to the fact that the isomerization and conversion of sugars to fructose do not occur effectively in the presence of nitric acid.

DRAFT

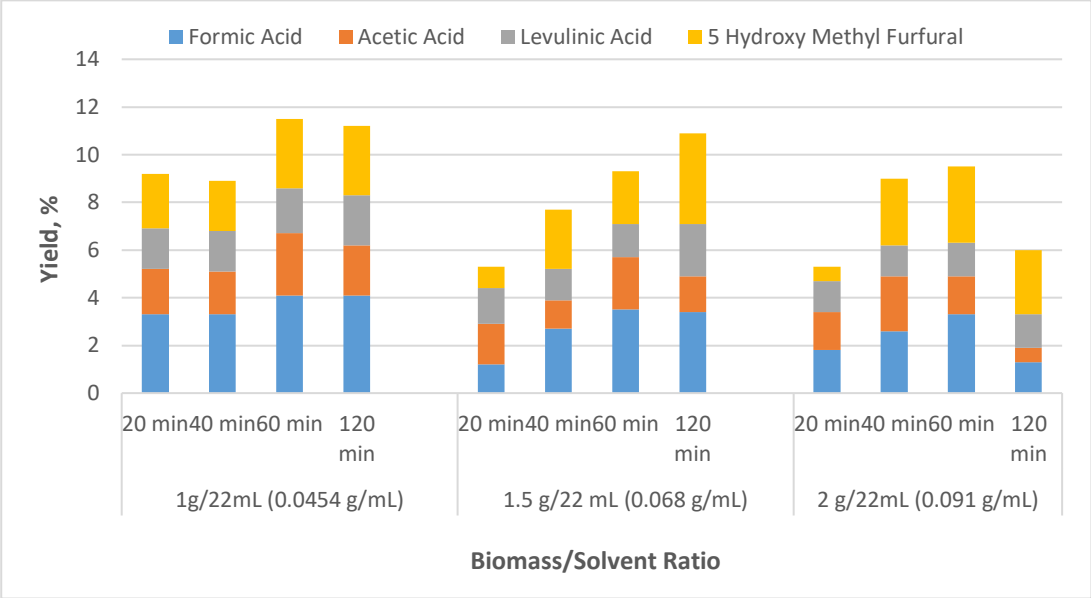


Figure 2. Variation of the yields sugar compounds in the liquid product as a result of the hydrothermal decomposition of Jerusalem artichoke at 180°C and pH=0.5 in the presence of HNO_3, according to the feed solution concentration and time.

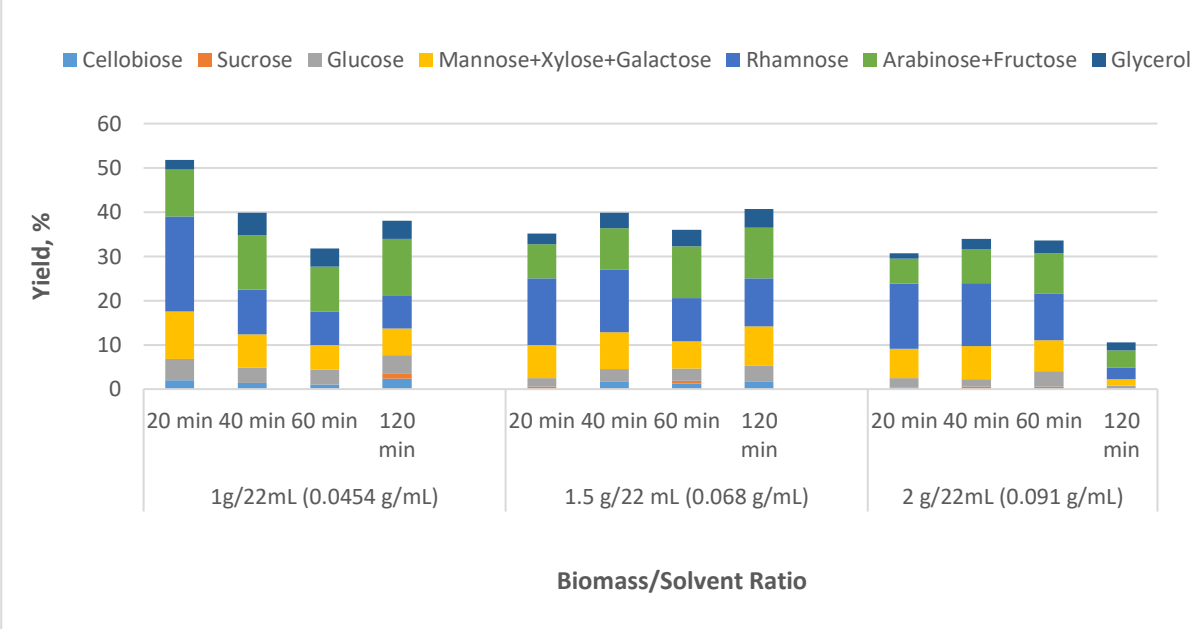


Figure 3. Variation of the yields of organic acid, furfural in the liquid product as a result of the hydrothermal decomposition of Jerusalem artichoke at 180°C and pH=0.5 in the presence of HNO_3, according to the feed solution concentration and time.

CONCLUSION

Jerusalem artichoke is decomposed to sugar compounds and organic acids, and 5-HMF in varying yields at 180°C and 0.5 pH level, various feedstock concentrations. To conclude, the levulinic acid was not effectively produced at this conditions as targeted while HNO_3 is successful in hydrolyzing the biomass into sugar compunds. It may be suggested that the study be carried out in two stages in order to benefit from the biomass solubilization effect of nitric acid but also to increase the yield of organic acids. In this sense, in the first stage, lignocellulosic biomass is first treated with nitric acid and solubilized, and sugar solution can be obtained. This

process can be carried out at low temperatures. Afterwards, nitric acid can be neutralized by adding base to the medium, and after the mixture is brought to the appropriate pH with Brönsted acids, the transformation of sugars into organic acids can be examined at higher temperatures.

ACKNOWLEDGEMENTS

We gratefully thank to Ege University and Office of Scientific Research Projects for their financial support during the project studies in project titled “From Lignocellulosıc Biomasses Investıgatıon Of Levulınıc Acid Production” and Number: FGA-2019-20183. We also thank to MSc. student Nazlıcan Atik for her contributions in the experimental studies.

REFERENCES

Bedzo, Oscar K. K., Mohsen Mandegari, and Johann F. Görgens. 2020. “Techno-Economic Analysis of Inulooligosaccharides, Protein, and Biofuel Co-Production from Jerusalem Artichoke Tubers: A Biorefinery Approach.” *Biofuels, Bioproducts and Biorefining* 14(4):776–93.

Bhagia, Samarthya, Hannah Akinosho, Jorge F. S. Ferreira, and Arthur J. Ragauskas. 2017. “Biofuel Production from Jerusalem Artichoke Tuber Inulins: A Review.” *Biofuel Research Journal* 4(2):587–99.

D.W., Rackemann, and Doherty W.O. 2011. “The Conversion of Lignocellulosics to Levulinic Acid.” *Biofuels, Bioproducts and Biorefining* 5(2):198–214.

Fachri, Boy Arief, Carolus Borromeus Rasrendra, and Hero Jan Heeres. 2015. “Experimental and Modeling Studies on the Conversion of Inulin to 5-Hydroxymethylfurfural Using Metal Salts in Water.” *Catalysts* 5(4):2287–2308.

Galletti, Anna Maria Raspolli, Claudia Antonetti, Valentina De Luise, Domenico Licursi, and Nicoletta Nassi O. Di Nasso. 2012. “Levulinic Acid Production from Waste Biomass.” *BioResources* 7(2):1824–34.

Gunnarsson, I. B., S. E. Svensson, E. Johansson, D. Karakashev, and I. Angelidaki. 2014. “Potential of Jerusalem Artichoke (Helianthus Tuberosus L.) as a Biorefinery Crop.” *Industrial Crops and Products* 56:231–40.

Kang, Shimin, Jinxia Fu, and Gang Zhang. 2018. “From Lignocellulosic Biomass to Levulinic Acid: A Review on Acid-Catalyzed Hydrolysis.” *Renewable and Sustainable Energy Reviews* 94:340–62.

Mukherjee, Agneev, Marie-Josée Dumont, and Vijaya Raghavan. 2015. “Review: Sustainable Production of Hydroxymethylfurfural and Levulinic Acid: Challenges and Opportunities.” *Biomass and Bioenergy* 72:143–83.

Qiu, Yibin, Peng Lei, Yatao Zhang, Yuanyuan Sha, Yijing Zhan, Zongqi Xu, Sha Li, Hong Xu, and Pingkai Ouyang. 2018. “Recent Advances in Bio-Based Multi-Products of Agricultural Jerusalem Artichoke Resources.” *Biotechnology for Biofuels* 11(1):1–15.

Sarchami, Tahereh, and Lars Rehmann. 2015. “Optimizing Acid Hydrolysis of Jerusalem Artichoke-Derived Inulin for Fermentative Butanol Production.” *BioEnergy Research* 8(3):1148–57.

Signoretto, Michela, Somayeh Taghavi, Elena Ghedini, and Federica Menegazzo. 2019. “Catalytic Production of Levulinic Acid (LA) from Actual Biomass.” *Molecules (Basel, Switzerland)* 24(15):1–20.

Vassilev, Stanislav V., David Baxter, Lars K. Andersen, and Christina G. Vassileva. 2010. “An Overview of the Chemical Composition of Biomass.” *Fuel*.

Viet Bui, Cuong, Wanwipa Siriwatwechakul, Wareepat Tiyabhorn, Thammanont Wattanasiritham, Natthaphat Limpraditthanont, and Siwarutt Boonyarattanakalin. n.d. “Conversion of Jerusalem Artichoke Tuber Powder into Fructooligosaccharides, Fructose, and Glucose by a Combination of Microwave Heating and HCl as a Catalyst.” *Thammasat International Journal of Science and Technology* 21(3).

Wang, Yingxiong, Christian Marcus Pedersen, Yan Qiao, Tiansheng Deng, Jing Shi, and Xianglin Hou. 2015. “In Situ NMR Spectroscopy: Inulin Biomass Conversion in ZnCl2 Molten Salt Hydrate Medium - SnCl4 Addition Controls Product Distribution.” *Carbohydrate Polymers* 115:439–43.

Werpy, T., and G. Petersen. 2004. “Top Value Added Chemicals from Biomass Volume I.” *Us Nrel* Medium: ED; Size: 76 pp. pages.

ORAL PRESENTATION

Innovative Functional Probiotic Food Products

Pınar ŞANLIBABA[2] *(https://orcid.org/0000-0003-4638-6765)* [1]*
Başar UYMAZ TEZEL *(https://orcid.org/ 0000-0002-4156-8861)*[2]

[*1] Ankara University, Department of Food Engineering, Faculty of Engineering, GÖLBAŞI/ANKARA/TÜRKİYE [2]Çanakkale Onsekiz Mart University, Bayramiç Vocational School, Laboratory Technology Program, BAYRAMİÇ/ÇANAKKALE/TÜRKİYE

*Corresponding author e-mail: sanlibab@ankara.edu.tr

Abstract

Over the past two decades, there has been a distinct and notable trend towards the widespread adoption of natural approaches for safeguarding human health. This trend has manifested through the utilization of various strategies, including probiotics, functional foods, biological stimulators, and regulators, among others. Each food categorized as a functional food undergoes a comprehensive evaluation that encompasses its nutritional content, caloric density, taste attributes, and its impact on human health. Specifically, these assessments consider the food's potential to support overall well-being and its capacity to contribute to the healing process in the event of illness. Among the emerging functional foods in the market, foods and beverages containing probiotics are widely recognized as future foods that enjoy substantial acceptance among consumers. This trend has led to a remarkable surge in the global market value, reaching 42.55 billion USD in 2017 and is anticipated to reach 94.48 billion USD by 2024. Within the wide array of probiotic foods available, there is a notable emphasis on producing and consuming fermented dairy products that contain live microorganisms. This category holds significant interest within the realm of probiotics. In recent years, non-dairy probiotic foods have garnered substantial attention from the research community, reflecting an evolving consumer demand and an expanding market for such alternatives. This review paper attempts to extensively provide insights into innovative probiotic-based foods and beverages.

Keywords: Innovative functional food, microorganisms, probiotic food, non-dairy food

1. INTRODUCTION

Functional foods are indeed a category that goes beyond providing essential nutrition. They are formulated to offer specific health benefits beyond simply meeting nutritional needs. These benefits can include promoting optimal health, reducing the risk of illness, and enhancing overall well-being. Functional foods often contain bioactive compounds or ingredients that have been demonstrated to have positive effects on health when consumed as part of a regular diet (El-Sohaimy et al., 2023). Microbial-derived functional foods include probiotics, prebiotics, symbiotic, and synbiotic. Functional foods of microbial origin operate by fostering the proliferation of probiotic bacteria, thereby constraining the growth of pathogenic bacteria (Koirala and Anal, 2021).

A probiotic product is expected to maintain a viable cell count of probiotic microorganisms exceeding 10^6 colony forming units (CFU) per mL or gram. Numerous factors can impact the viability of probiotics within food products during both processing and storage phases. These factors may stem from intrinsic characteristics of the product, encompassing aspects such as pH, titratable acidity, oxygen levels, water activity, the presence of substances like salt, sugar, hydrogen peroxide, bacteriocins, artificial flavorings, coloring agents, or treatments associated with food processing. Alternatively, these factors can be linked to external conditions, including fermentation parameters, incubation temperatures, heat treatments, cooling and storage conditions, the choice of packaging materials, the scale of production, or specific microbiological traits of the probiotics, such as the microbial strain or the rate and proportion of inoculation (Rodzi and Lee, 2021). Critical considerations in the selection of potential probiotics revolve around their viability during food processing and/or storage, and their survival post-transit through the gastrointestinal tract (GIT), and their capacity to

confer health benefits to the host. It is pertinent to note that a majority of bacteria struggle to endure the rigorous conditions encountered during their passage through the upper GIT.

Consequently, the judicious selection of the appropriate probiotic strain assumes pivotal importance in the formulation of products housing viable probiotic cells capable of bestowing health advantages upon consumers (Mojikon et al., 2022). Probiotics offer health advantages to the host when ingested in sufficient quantities. These benefits encompass: *i)* balancing intestinal homeostasis (Probiotics contribute to the equilibrium of the intestinal environment by either eliminating or inhibiting the growth of harmful microbial flora); *ii)* boosting the immune system (Probiotics have the potential to enhance the immune system's functionality, aiding in the body's defence against infections and illnesses); *iii)* reducing the risk of various cancers (There is evidence to suggest that probiotics may help mitigate the risk of certain types of cancer through their influence on the gut microbiota); *iv)* enhancing lactose digestion (Probiotics can improve the digestion of lactose, which can be particularly beneficial for individuals with lactose intolerance) and *v)* other host-microbe benefits (probiotics have been associated with various other advantages, including the prevention of conditions such as cardiovascular diseases, diabetes, and some allergic reactions). Additionally, the consumption of probiotics can lead to alterations in the composition of the intestinal tract's microbiome, further influencing overall health and well-being (Koirala and Anal, 2021). Various microorganisms, including *Lactobacillus rhamnosus* GG, *Lb. reuteri*, *Bifidobacteria*, specific strains of *Lb. casei*, and members of the *Lb. acidophilus* group, have been extensively studied and identified for their probiotic attributes. These microorganisms find prevalent utilization in probiotic food products, notably within the realm of fermented dairy products. Additionally, a subset of other microorganisms, such as *Escherichia coli* strain Nissle 1917, *Enterococcus faecium* SF68, and *Saccharomyces boulardii*, have exhibited therapeutic potential in various applications (El-Sohaimy et al., 2023).

The landscape of probiotic food products has witnessed a notable shift in recent times, transitioning from a predominance of dairy-based offerings to encompass a diverse array of plant-based alternatives, beverages, and snacks (Plessas, 2022). Traditionally, probiotic food products have centered on fermented dairy items, exemplified by kefir and yoghurt. Subsequently, a secondary category of probiotic food products has emerged, embracing fermented fruit and vegetable items, such as pickles or olives, along with select fermented meat products, including sausages. A compelling development in the realm of probiotic food products is the emergence of a new generation characterized by their intriguing non-dairy and non-fermented nature. Within this category, probiotics find incorporation into an assortment of items, spanning breakfast cereals, juices, fruit snacks, chocolates, and confections. Notably, in recent years, non-dairy probiotic foods have garnered substantial attention from the research community, reflecting an evolving consumer demand and an expanding market for such alternatives (Patel, 2017).

2. INNOVATIVE PROBIOTIC FOODS

The predominant probiotic products in the market are primarily derived from milk, including yoghurts, cheese, and fermented milk. Nevertheless, the utilization of milk-based probiotic products faces certain limitations, notably due to concerns related to allergies, cholesterol-related conditions, dyslipidemia, and the dietary preferences of vegans. Consequently, there has been a notable upsurge in research endeavours aimed to develop innovative and alternative mediums for delivering probiotics (Plessas, 2022).

2.1. FRUIT AND VEGETABLE-BASED PROBIOTIC FOODS

Fruit and vegetable juices have emerged as a promising alternative for probiotic delivery, particularly given the remarkable growth of their global market. Fermented fruits and vegetables are increasingly recognized as healthful food options suitable for individuals of all ages. They stand out as preferred dietary choices due to their exceptional functional and nutritional characteristics, which are enhanced through the fermentation process involving probiotic microorganisms. The health benefits associated with the consumption of fermented fruit and vegetable juices primarily stem from the presence of novel bioactive compounds. These bioactive compounds, such as phenolics and fatty acids, are augmented in quantity and efficacy during fermentation. Additionally, fermented juices contain elevated levels of minerals and vitamins compared to their unfermented counterparts, further contributing to their health-promoting properties (Mojikon et al., 2022). Numerous researchers have explored the suitability of various fruit and vegetable juices as potential raw materials for the production of probiotic juices and related beverages. These juices encompass a wide range of options,

including tomato, mango, orange, apple, grape, peach, omegranate, watermelon, carrot, beetroot, and cabbage juices. The probiotics most commonly employed in these formulations consist of different strains from a variety of microbial species, including but not limited to: *Lb. acidophilus, Lb. helveticus, Lb. casei, Lb. paracasei, Lb. johnsonii, Lb. plantarum, Lb. gasseri, Lb. reuteri, Lb. delbrueckii subsp. bulgaricus, Lb. crispatus, Lb. fermentum, Lb. rhamnosus, Bifidobacterium bifidum, B. longum, B. adolescentis, B. infantis, B. breve, B. lactis, B. laterosporus.* Additionally, other microbial species such as *E. coli* Nissle, *Streptococcus thermophilus, Weissella* spp., *Propionibacterium* spp., *Pediococcus* spp., *Enterococcus faecium, Leuconostoc* spp., and *Saccharomyces cerevisiae* var. *boulardii* have also been considered for their probiotic potential in these juice-based products (Patel, 2017). The criteria used for determining the suitability of a strain in fruit and vegetable juices are; *i)* ability of a substrate to acquire a starter culture; *ii)* the rate and total production of lactic acid, change in pH, loss of nutritionally important substances; *iii)* decrease in nitrate concentration and production of biogenic amines; *iv)* type of metabolism and ability of culture to produce desirable sensory properties of fermented products (Rodzi and Lee, 2021).

2.2. CEREAL-BASED PROBIOTIC FOODS

Cereals and their constituent components are poised to attain recognition as functional foods and nutraceuticals. This recognition is attributed to their rich content of antioxidants, vitamins, proteins, energy, and dietary fiber-nutritional elements of paramount importance for preserving human health (Tsafrakidou et al., 2022, Yang et al., 2022). Furthermore, cereals and their various constituents serve as integral substrates for the cultivation and development of probiotic bacteria through fermentation processes (Patil et al., 2022). Rice, wheat, corn, rye, barley, sorghum, millets (Pearl, Kodo, barnyard, foxtail, proso, little millet), pseudo-cereals (buckwheat, quinoa, and amaranth) are the most common cereals consumed by humans (Meena et al., 2023). It has been noted in empirical observations that cereals and their derived products are sometimes lesser or poorer than milk and dairy products. This disparity can be attributed to several factors, notably the cereals' relatively diminished protein content, inadequacy in specific essential amino acids, such as lysine, as well as challenges associated with their digestibility and nutrient bioavailability. These hindrances are compounded by the presence of various anti-nutritional factors, including polyphenols, tannins, and phytate, in conjunction with the coarse-grained nature of cereals. Numerous strategies have been recommended to enhance the nutritional profile of cereals. These encompass genetic enhancements, supplementation with amino acid protein concentrates, or the utilization of other protein-enriched resources like legumes and oilseeds. Furthermore, various processing techniques, such as cooking, sprouting, milling, and fermentation, have been recognized for their potential to enhance the nutritional attributes of cereals. Among these methodologies, fermentation has emerged as a particularly effective approach for augmenting the nutritional quality of cereals (Ganguly et al., 2021). The process of fermenting cereals has been demonstrated to exert positive effects on their bioavailability, digestibility, and organoleptic characteristics. Furthermore, the nutrient composition inherent to cereals has been observed to facilitate the proliferation of probiotic microorganisms. Notably, the presence of prebiotic compounds within cereals can further stimulate the growth and activity of probiotic microorganisms. To date, a variety of probiotics have been used in the development of functional cereal products, including *Lb. plantarum*, *Lb. acidophilus*, *Limosilactobacillus reuteri* and *Lb. casei* (Yang et al., 2022).

2.3. MEAT BASED PROBIOTIC FOODS

Meat and its derivative products constitute a fundamental component of human nutrition, notably serving as a rich source of high-quality proteins. Among the various techniques employed for food preservation and storage, fermentation and drying are venerable methods with a historical lineage. In particular, the manufacture of raw cured and ripened meat products, encompassing diverse varieties of sausages and smoke-cured meat items, prominently involves the utilization of fermentation processes. The traditional production of raw cured meat products hinges upon the fermentation of indigenous or supplemented carbohydrates by lactic acid bacteria, present either within the meat matrix or within its surrounding environment (Munekata et al., 2022). This intricate biochemical transformation gives rise to a myriad of compounds, including but not limited to lactic acid, pyruvic acid, alcohols, aldehydes, ketones, and carboxylic acids. The quantitative composition of these compounds profoundly influences the quality attributes and shelf-life stability of the end product. Predominantly, starter cultures comprising lactic acid bacteria (LAB), notably *Lb. sakei, Lb. curvatus, Lb. plantarum*, and *Lb. lactis*, assume a pivotal role in the fermentation of meat products. LAB hold a preeminent status as starter cultures, exercising paramount influence on the production of fermented meat items. The realm

of probiotic meat products represents a relatively nascent and not fully explored domain within the meat industry. Herein, paramount importance is accorded to striking a delicate balance between food safety, sensory attributes, and pro-health properties. The manufacturing of probiotic meat products proves considerably more challenging than conventional counterparts, primarily attributed to the distinctive characteristics of the raw materials. Probiotic bacterial strains suitable for incorporation into fermented meat products must exhibit the capacity to thrive in the distinctive milieu encountered in fermented goods. Additionally, they must assert dominance over other microbial entities present in the final product. Simultaneously, the sensory profile of the product must remain unaltered. Notably, probiotic bacteria need to be introduced during the preparation of the meat mixture and subsequently during the ripening phase. This strategic inclusion should facilitate the development of robust defensive mechanisms, ensuring the dominance of the probiotic strains over indigenous microflora. In light of these complexities, the production and introduction of probiotic meat products remain at an incipient stage, particularly when juxtaposed with their dairy-based counterparts (Kołożyn-Krajewska and Dolatowski, 2009; Sirini et al., 2022).

REFERENCES

El-Sohaimy, S.A., Hussain, M.A. (2023). Functional Probiotic Foods Development: Trends, Concepts, and Products. Fermentation, 9, 249. https://doi.org/10.3390/fermentation9030249

Ganguly, S., Sabikhi, L., Singh, A.K. (2021). Cereal-based Fermented Foods for Enhanced Nutritional Attributes and Better Gut Health. Intl. J. Ferment. Food, 10(1): 01-12. http://doi.org/10.30954/2321-712X.01.2021.1

Koirala, K., Anal, A.K. (2021). Probiotics-based foods and beverages as future foods and their overall safety and regulatory claims. Future Foods, volume 3, 100013, https://doi.org/10.1016/j.fufo.2021.100013.

Kołożyn-Krajewska, D., Dolatowski, Z.J. (2009). Probiotics in fermented meat products. Acta Sci. Pol., Technol. Aliment. 8(2), 61-74

Meena, K.K., Taneja, N.K., Jain, D., Ojha, A., Saravanan, C., Bunkar, D.S. (2023). Spontaneously Fermented Cereal Based Products: An Ancient Health Promoting Formulae for Consumption of Probiotic Lactic Acid Bacteria. Biointerface Research in Applied Chemistry, 13(5), 465. https://doi.org/10.33263/BRIAC135.465

Mojikon, F.D., Kasimin, M.E., Molujin, A.M., Gansau, J.A., Jawan, R. (2022). Probiotication of Nutritious Fruit and Vegetable Juices: An Alternative to Dairy-Based Probiotic Functional Products. Nutrients 14, no. 17: 3457. https://doi.org/10.3390/nu14173457

Munekata, P.E.S., Pateiro, M., Tomasevic, I., Domínguez, R., da Silva Barretto, A.C., Santos, E.M., Lorenzo, J.M. (2022). Functional fermented meat products with probiotics—A review, *Journal of Applied Microbiology*, 133(1), 91–103, https://doi.org/10.1111/jam.15337

Patel, A.R. (2017). Probiotic fruit and vegetable juices- recent advances and future perspective. International Food Research Journal 24(5): 1850-1857

Patil, A., Raktate, S., Sharma, B., Srivastava, E. (2022). Cereal based probiotic foods. Pharma Innovation Journal, 11(7): 4782-4788.

Plessas, S. (2022). Editorial: Innovations in Functional Food Production: Application of Fruit and Vegetable Juices as Vehicles for Probiotic Delivery. Frontiers in Microbiology, 13:854705. https://doi.org/10.3389/fmicb.2022.854705

Rodzi, N.A.R. M., Lee, L.K. (2021). Traditional fermented foods as vehicle of non-dairy probiotics: Perspectives in South East Asia countries. Food Research International, Volume 150, Part B, 110814, https://doi.org/10.1016/j.foodres.2021.110814

Sirini, N., Loyeau, P., Ruiz, M., Stegmayer, M., Soto, L., Werning, M., Frizzo, L., Ordoñez, V., Fernández-López, J., Rosmini, M. (2022). Development of Probiotic Fermented Sausages and Viability Monitoring of Supplemented Lactiplantibacillus plantarum BFL Strain. Fermentation, 8, 526. https://doi.org/10.3390/fermentation8100526

Tsafrakidou, P., Michaelidou, A.-M., G. Biliaderis, C. (2020). Fermented Cereal-based Products: Nutritional Aspects, Possible Impact on Gut Microbiota and Health Implications. Foods, 9, 734. https://doi.org/10.3390/foods9060734

Yang, Z., Zhu, X., Wen, A., Qin, L. (2022). Development of probiotics beverage using cereal enzymatic hydrolysate fermented with *Limosilactobacillus reuteri*. Food Science and Nutrition, 10(9), 3143-3153. https://doi.org/10.1002/fsn3.2913

DRAFT

ORAL PRESENTATION

Effect of drought stress on plant species in Karaman with terrestrial climate

Said Efe DOST[1]* *(0000-0003-4279-7292)*, Pınar ÖZTÜRK[2] (*0000-0002-1472-7812)*

[*1]Karamanoglu Mehmetbey University, Technical Science. Vocational School of Higher Education, Karaman, Turkey
[2]Karamanoglu Mehmetbey University, Department of Plant Production and Technologies, Karaman, Turkey

*saidefe.dost@gmail.com

Abstract

Water stress is an important abiotic stress affecting plant growth, yield, and quality decline. Sudden population growth in the world, global climate change, and many related factors cause an important drought problem in agriculture. To get over this problem, it is necessary to develop strategies for effectively using scarce water resources in plant production. The use of modern cultivation techniques in fruit growing has provided significant increases in the amount and quality of production. Irrigation and fertilization are the most important issues of this production model. Optimizing irrigation and fertilization significantly increases income per unit area while ensuring water consumption and sustainability. If the effect of drought damage on the plant is well-defined, the solution will be much easier. Water stress and drought cause oxidative stress in plants from the accumulation of free radicals. Plants try to resist oxidative stress with some enzymatic and non-enzymatic protection systems. However, the effectiveness of this defense system varies according to factors such as plant type, variety, stress density, and duration. Osmolites accumulate in plants under drought stress; It is divided into 3 parts carbohydrates (such as sucrose, and sorbitol), nitrogen compounds (proline and polyamines), and organic acids (oxalate and malate). Proline is a common osmolyte in plants exposed to water stress. Therefore, subsequent studies focused on elucidating the tolerance mechanisms of drought-resistant plants and on the conservation and transfer of plant genetic resources.

Keywords: Apple, Droughty stress, Proline,

INTRODUCTION

Stress is physically expressed as the force applied to an object per unit area. Plants encounter many stress factors throughout their lives. Stress is defined as abnormal changes that occur physiologically in plants under the influence of environmental or biological factors. The stress factors that plants encounter throughout their lives are examined under two main titles, and these factors are examined by researchers as abiotic (drought, salinity, radiation, high temperature, frost, etc.) and biotic (pathogen, competition with other organisms, etc.). These stress factors reduce the biosynthetic capacities of plants and change their normal functions, causing permanent damage and even death to the plant (Levitt, 1980; Hale & Orcutt, 1987; Mahajan & Tuteja, 2005; Kalefetoglu & Ekmekci, 2005).

Drought, which is one of the leading natural disasters, causes great losses by causing great damage to the environment and people. Forest fires have become an important problem threatening countries around the world due to global warming, decreasing precipitation, increasing greenhouse gases, insufficient water resources, and climate change (Hekimoglu & Altindeger, 2008). In addition, drought stress has the largest share with 26% among the stress factors observed in cultivated lands worldwide (Kalafatoglu & Ekmekci, 2005). In a general manner, drought is meteorological, it is defined as a period when there is no precipitation, the water content of the soil and plant development decreases significantly, and water scarcity reaches the amount it will suffer (Özcan et al., 2004). Drought is one of the most important environmental stress factors in the world affecting plant growth and yields within the abiotic stress group (Glombitza et al., 2004) and climate changes in recent years indicate that this situation is becoming more serious (Wang et al., 2003). Globally, drought stress, combined with high temperatures and radiation, significantly limits plant life and crop (Chaves et al., 2003; Kapluhan, 2013). Drought stress is common in areas where precipitation is not sufficient, as plants cannot find the moisture they need to sustain their lives (Sircelj et al., 2007).

Plants respond to the situation with morphological, biochemical, and metabolic changes that they show during the drought stress process. In plants exposed to drought stress, cell division, and differentiation come to a standstill, which negatively affects plant growth and development. The intake of mineral substances decreases in the plant and the yield is directly affected by slowing down the plant development with the decrease of turgor pressure in the plant under the influence of drought and the decrease of the amount of water lost by sweating. Stress has been determined to reduce trunk diameter and leaf area, as well as plant height and fresh root weight in pear trees exposed to drought. As a result, it has been found that plants exposed to water scarcity are more vulnerable to other biotic and abiotic stresses (Romo et al., 2001; Capell et al., 2004; Caruso et al., 2008; Farooq et al., 2009; Gür, 2018; Küçükyumuk, 2020; Faizi & Öztürk, 2022).

Even though drought in nature is caused by a combination of environmental factors, plants stimulate many physiological, biochemical, and molecular responses to water scarcity and, accordingly, they can develop tolerance mechanisms that adapt to limited environmental conditions (Arora et al., 2002). Stress tolerance is the ability of plants to survive against adverse environmental conditions. Plants form a two-way survival mechanism against drought stress, either by avoiding stress or by developing stress tolerance (Mundree et al., 2002). Plants draw water from the soil more strongly with the decrease in the amount of water lost by transpiration in drought stress and morphological changes in leaves and roots (Çırak & Esendal, 2006; Kutlu, 2010). When plants are under drought stress, they try to reduce water loss and transpiration by shedding their leaves and reducing leaf areas. As the leaf area decreases, the amount of CO2 per unit area also decreases. This decrease negatively affects the photosynthesis of the plant. While the plant increases the wax layer on the leaf surface to reduce water losses, the cuticle layer reflects the sun's rays, reducing the effect of heat and therefore reducing the rate of transpiration (Göksoy & Turan, 1991; Türkan et al., 2005). The relative moisture content is positively related to the photosynthesis rate. Leaf water content, relative moisture content, and transpiration rate decrease significantly in plants exposed to drought stress (Anjum et al, 2011). When the water decreases, the plant tries to preserve the available water by closing its stomata so as not to lose more water (Turkan et al., 2005). In addition, respiration, translocation, ion intake, carbohydrate, nutrient assimilation, and growth promoters are also damaged in combination with drought conditions (Siddiqui et al., 2015).

Dehydration causes a decrease in cell volume, an increase in cell cytoplasm concentration, and a constantly increasing dehydration of protoplasm, which in this case leads to an increase in reactive oxygen species (ROS). The activity of peroxidase or glutathione reductase decreases oxidative stress caused by drought. Adjustment in plants occurs through the deposit of low molecular weight organic solutions in water stress, osmotic. These osmolytes are found in plants as soluble carbohydrates and proline. One of the first reactions of the plant to reduce water stress damage is known as Proline deposition (Anjum et al., 2011; Marcińska et al., 2013; Adak et al., 2018). Drought resistance is behavior specific to species and even varieties, and it is of great importance to determine the method and ability of the plant to avoid stress in order to determine the tolerance of plants under stress conditions (Özcan et al., 2004). Various species-specific enzymatic defense systems have been developed against stress-induced oxidative stress in plant cells in addition to the tolerance mechanisms created against the mechanical damage caused by drought (Bian & Jiang, 2009). A high amount of antioxidant enzyme activity in plants significantly reduces the oxidative damage caused by the cell size in tolerance to drought stress. But the effectiveness and success of these antioxidant enzymes produced during drought vary depending on the type, variety, severity of stress, and duration of the stress of the plant (Sharma & Dubey, 2005; Türkan et al., 2005; Bian & Jiang, 2009).

WHAT IS STRESS

In our era, the word stress is included in our life in this way. The word stress is used in engineering fields such as physics and mechanics to fields related to human and animal health such as medicine and veterinary medicine. A kind of defense or even a psychological reaction occurs in the organism against the pressure or tension they encounter in creatures exposed to stress. Thus, ecological stress is defined as any factor that forces plant metabolism and the internal balance of the plant in its natural environment to change, as well as causing deficiencies in plant development and growth (Shulaeva et al., 2008; Nader et al., 2019). Physiological and biochemical events such as hormonal balance, respiration, photosynthesis, and nutrient transport are affected in plants exposed to stress, causing plant metabolism to slow down or stop. Plant development is negatively affected, resulting in a decrease in yield and quality as a result of all these reactions. Stress factors are studied under two groups, Biotic factors are stress factors caused by infection of microorganisms (fungi, bacteria, and

viruses) and attacks of harmful animals. Abiotic stress conditions involve many stresses caused by complex environmental conditions such as strong light, ultraviolet, high and low temperatures, frost, drought, salinity, heavy metals, and insufficient oxygen (Lichtenhaler et al., 1996; Hirayama & Shinoza, 2010). Naturally, biological tensions that occur as a result of stress in a living individual are also expected to be observed. What is expressed here as biological stress is the change in plant metabolism and morphology and the decrease in growth. According to this, if the plant is able to react to the environmental factors it lives in at the maximum level, it means that it has not started to be affected by environmental factors at the stress level. Contrary, if changes in factors other than optimal values cause changes in the response level of the plant as a result of tension, this condition is considered stress. Plants can naturally adapt to stressful conditions. Despite being exposed to stress conditions, these specialized plants are able to survive in the environment they are in and complete their life cycle (Boscaiu et al., 2008; Büyük et al., 2012; Noctor et al., 2018; Wang et al., 2019).

DROUGHT STRESS

Drought stress is one of the most important and common abiotic stress factors for plants in many parts of the world, especially in drought and semi-drought regions. Drought is used as a general term to describe atmospheric or weather events and is usually used to describe a period that will pass without precipitation (Jaleel et al., 2009; Dai, 2012).

Drought is divided into four main titles: meteorological, agricultural, hydrological, and socio-economic. Drought begins as a meteorological drought, develops as an agricultural and hydrological drought, and its effects become visible as a socio-economic drought. While meteorological drought is based on precipitation and is defined as a decrease in precipitation below normal values for a long time, the increase in air temperatures and atmospheric CO_2 together with global climate change affects the precipitation regime negatively in many regions and causes drought (Arbona et al., 2013). Agricultural drought is defined as the amount of water that plant roots can use in the soil that falls below a certain level or the inability of moisture in the soil to be taken up by the plant. In other words, it is seen that the amount of water lost by transpiration from the leaf surface is more than the amount of water taken up by the plant roots (Keyvan, 2010; Mengü et al., 2011; Dai, 2012; Örs & Ekinci, 2015). Imbalances in water consumption and water loss of the plant usually reveal themselves in cases where the water potential of the soil is lower than the water potential of the plant roots. Continuous water loss through transpiration and evaporation is positively or negatively affected by changing atmospheric conditions (Shao et al., 2008; Mafakheri et al., 2010; Dai, 2012; Trenberth et al., 2014). As a result, agricultural drought occurs immediately after meteorological drought depending on climatic conditions (Jaleel et al., 2009; Barriopedro et al., 2012; Dai, 2012). Hydrological drought, the development of large water losses in rivers, lakes, and underground water resources, the decrease in precipitation as a result of global warming and climate change, due to insufficient water reserves, the rapid increase in the world population and with this increase, the amount of water consumed per capita exceeds the standards, puts countries at risk of drought and desertification. Socioeconomic drought, on the other hand, occurs as a result of imbalances caused by other types of drought and is observed as a result of the deterioration of the economic balances of countries (Mishra & Singh, 2010). Climate change is a major factor inducing drought stress worldwide and disruption of the natural balance is a result of these changes, drought resulting from climate change is limiting crop yields by preventing many crops from being grown locally. Water losses in the plant and soil increase together with a combination of factors, and then it becomes easier for the plant to be exposed to water stress (Mafakheri et al., 2010; Dai 2012). As we have witnessed in many parts of the world, it is seen that precipitation decreases and is irregular in our country, especially in summer months due to climate change and global warming (Kaynaş & Kaynaş, 2003). As one of the effects of climate change, it is predicted that it will cause climate changes in our country, temperature values will increase with this change, and this rising temperature can seriously affect semi-drought and drought regions that do not have enough water. Irregularity in the amount of precipitation and its distribution over the months is an inevitable result that will negatively affect plant yield and agricultural production (Ozturk, 2002). Turkey will be among the countries in the risk group regarding climate change It is predicted that if measures are not taken, the consequences will be greater and our Central Anatolia and Mediterranean regions will be more affected by global warming in the future (Sircelj et al., 2007). Changes in production techniques have become necessary to ensure sustainability in agriculture. These changes in production techniques necessitate some practices that reduce water stress in order not to face the bigger problems of the future. Adjusting the planting time, mulch application, some microorganism applications, and fertilizer applications to reduce moisture losses in the growing environment

and plant can be given as examples of changes (Jaleel et al., 2009; Jing-min et al., 2010; Desoky et al., 2018). According to the United Nations World Agricultural Organization (FAO), drought damage is defined as the percentage of years in which crops are damaged due to reduced humidity (Hawkish & Cunning, 2014).

THE EFFECT OF DROUGHT STRESS ON THE PLANT

Water is one of the abiotic factors and is an important limiter affecting plant growth, development, and yield. Although drought stress is a multidimensional stress, the response of plants to water stress varies depending on the type of plant, the age of the plant, the growth and development period, the level and continuity of drought, and physical factors. Plants show resistance to the negative effects of stress with the changes they make in their physiological, morphological, ecological, biochemical, and molecular properties in order not to succumb to different stress factors (Kayabasi, 2011; Bhargava & Sawant, 2013; Marcińska et al., 2013). The stress factor also negatively affects plant growth, yield quantity, and quality. The decrease in plant growth due to stress develops due to the fact that shoot and root meristem tissues are affected and the cells in these tissues stop division and expansion The cease of cell division or expansion is directly related to a decrease in the rate of photosynthesis due to a lack of water (Jaleel et al., 2009; Anjum et al., 2011; Zlatev & Lidon, 2012; Nezhadahmadi et al., 2013).

The symptoms of drought stress vary depending on the plant type, growth stage, growing conditions, and other environmental factors (Arbona et al., 2013; Bhargava & Sawant, 2013; Nezhadahmadi et al., 2013). The severity of the drought, the duration of the drought, soil physicochemical conditions, and plant resistance are other effective factors that trigger the severity and effect of drought stress symptoms in plants. In general, the symptoms of drought stress are observed in the plant in the form of loss of leaf turgor pressure, sagging, fading, etiolation, yellowing, and finally early leaf fall, respectively (Bernacchia & Furini, 2004; Farooq et al., 2009; Jaleel et al., 2009; Zare et al., 2011; Bhargava & Sawant, 2013; Sapeta et al., 2013; Akhtar & Nazir, 2013). The effect of drought stress on plants can be summed up in two main titles fading and drying. The disappearance of leaf turgor, sagging, fading, etiolation, yellowing, and early leaf fall can be defined as the beginning and end stages of these two main headings (Smirnoff, 1993; Bhargava & Sawant, 2013; Sapeta et al., 2013; Akhtar & Nazir, 2013). If plants are exposed to drought conditions for a long time, the leaves wilt, especially in deciduous plants in winter, and the prolongation of the duration of arid conditions, this causes significant leaf losses in the plant, and these losses can lead to the weakening or even death of the plant. Plants exposed to a slight lack of water, which reaches about 70% of the water pressure in the leaves, carbon dioxide intake, and photosynthesis process are restricted due to the closure of the stomata, and the plant weakens and signs of pallor begin to appear. The first visible and known symptom of drought stress is wilting (Zeppel et al., 2015). Drying can be defined as excessive water loss, which can lead to complete disruption of metabolism and cell structure and stop enzyme-catalyzed reactions (Smirnoff, 1993; Kalefetoglu & Ekmekci, 2005). The plant loses turgor pressure in wilting, and the leaf water content decreases, and this decrease causes the collapse of cells. When the amount of water required for the plant is provided, it regulates the turgor pressure and reduces the severity of wilt, allowing the plant to return to its former state. However, since the wilting has reached a very advanced stage in the drying event, it is not possible for the plant to return to its former state (Akinci & Lösel, 2012; Kaçar, 2015). Most of the vegetative tissues of vascular plants that are sensitive to drying out are unable to enter the healing process after the proportional water content falls below 30% (Kalefetoglu & Ekmekci, 2005). When the plant cannot supply the water it needs from the root zone and this creates stress, the plant tries to get rid of this problem by reducing water losses or reducing its internal water potential below the soil water potential in order to maintain water intake (Bray, 1997; Rahdari & Hoseini, 2012), and the first effect that will occur as a result of the continuation of the problem is the loss of turgor pressure (Barlow et al., 1980).

Plant growth and development are significantly affected in drought conditions. The extent of damage and damage caused to the plant by these conditions depends on the duration of water scarcity. The fact that the water lost by transpiration from the leaves cannot be met by the plant roots and the turgor loss as a result of this water loss shakes the plant metabolism. Plasmolysis occurs in the leaf cells as a result of drought, which causes the leaves to wilt (Günay, 2005). The plant increases root development by slowing down trunk elongation and growth during the first periods of drought in order to defend itself and reach more water (Ozturk, 2015). On the other hand, if the drought lasts longer than the plant's endurance, the trunk and root development at this stage stops completely due to stress, the leaf area and the number of leaves decrease, even some leaves turn yellow and fall out. A decrease in the rate of photosynthesis and decreases in chlorophyll

content in dry conditions are considered typical signs of oxidative stress, which leads to pigment photooxidation and chlorophyll degradation (Anjum et al., 2011; Marcińska et al., 2013). The effects of drought on the plant not only affect plant water relations by reducing water content, turgor, and total water, but also affect the closure of stomata in plants under stress, reducing gas exchange and transpiration, and bringing the carbon assimilation (photosynthesis) rate to a standstill (Lisar et al., 2012). The decrease in plant growth and growth occurs due to the cessation of cell division in the shoot and root meristems and the expansion of cells. The cease of cell division or expansion is directly related to the decreased rate of photosynthesis due to water stress (Anjum et al., 2011). Photosynthesis is the most important process for plants. Growth and health are also affected due to any disruption in the photosynthesis process, and plant productivity (Zargar et al., 2017). Leaf water content, relative moisture content, and transpiration rate decrease significantly in plants exposed to drought stress. There is a positive relationship between the relative moisture content and the rate of photosynthesis (Anjum et al., 2011). Apart from that respiration, translocation, ion uptake, carbohydrate, nutrient assimilation, and growth promoters are damaged together with drought conditions (Siddiqui et al., 2015). One of the negative aspects of drought stress is that it leads to an increase in reactive oxygen species (ROS). The activity of peroxidase or glutathione reductase decreases oxidative stress caused by drought. Osmotic adjustment for water stress in plants takes place by deposition of low molecular weight organic solutions (Marcińska et al., 2013). These osmolytes exert their effects as soluble carbohydrates and proline in plants. The deposition of proline in the cytosol is known as one of the first plant responses to reduce damage under water stress conditions (Anjum et al., 2011).

PLANT REACTIONS TO DROUGHT STRESS

Plants are able to flex their growth and development mechanisms in such a way that they are minimally damaged by changes that may occur in environmental conditions, and even adapt to be minimally affected by environmental factors when exposed to certain climatic conditions for a long time. The distribution of plants belonging to the same species in regions of the world with different climatic characteristics is the best indicator of their ability to adapt to very different environmental conditions (Dolfeus, 2014). Drought stress, which has an important effect on plant growth and development, reveals many morphological, physiological, biochemical, and molecular responses, and thus the plant activates different tolerance mechanisms in order to adapt to adverse environmental conditions or survive (Pou et al., 2013; Nader, 2019). As an example of the tolerance effort of the plant, changes in the form of an increase or decrease in aquaporin gene expression are observed in plants under drought stress (Tyerman et al., 2002). The tolerance responses of the plant and the size of drought symptoms in the plant may vary depending on the type of plant, the stage of development, growing conditions, and other environmental factors (Arbona et al., 2013; Bhargava & Sawant, 2013; Nezhadahmadi et al., 2013). In other words, the factors that induce drought symptoms in plants have been determined as drought severity, drought duration, soil physicochemical conditions, and plant strength. and affected by drought stress, the effects, and symptoms of stress in general include loss of leaf turgor, drooping, wilting, etiolation, yellowing, and premature leaf fall (Bernacchia & Furini, 2004; Farooq et al., 2009; Jaleel et al., 2009; Zare et al., 2011; Bhargava & Sawant, 2013; Sapeta et al., 2013; Akhtar & Nazir, 2013). They developed three different strategies for surviving or recovering from the above-mentioned drought stress effects in plant physiology: adaptation, drought avoidance, and drought tolerance. Drought adaptation refers to the capacity to withstand drought processes, while avoidance is a method of strategies that will prevent the reduction of water potential in protoplasm from lasting long enough to be harmful, and tolerance is defined as showing more desiccation without harming the protoplasm (Gurel & Avcioglu, 2001; Mundree et al., 2002).

Leaf water content and turgor loss occur in the plant and the plant closes its stomata for defense and to prevent further water loss with drought stress or a decrease in soil water content in drought conditions (Jaleel et al., 2009; Akinci & Lösel, 2012; Küçükyumuk, 2020). One of the simplest responses of the plant under drought stress conditions is the closure of stomata and the reduction of CO_2 exchange. Photosynthesis is prevented depending on the severity of the stress and if this situation continues for a long time due to the behavior of the plant, it can lead to plant death (Jaleel et al., 2009). A plant needs water to control its internal temperature. Most of the water is lost through stomatal transpiration, more than the amount stored in plant cells. This loss of water allows the leaves and the plant to cool down. Plants cannot maintain their body temperature when faced with factors such as water scarcity and drought (Adams et al., 2009). The relative growth rate of the plant decreases and the plant height also decreases under water stress conditions (Lipiec et al., 2013). Lack of water reduces plant growth, leaf surface area, and dry matter; it also causes cell membrane degradation,

pigment damage, chlorophyll content decrease, and root growth slows down and stops. Plant tissues are also damaged during drought stress and deaths occur during long-term drought (Farooq et al., 2009). It is observed that while total phenolic substance, soluble sugar, proline, antioxidant, enzyme activities, Fe, Mg, and Mn values increase in the shade of damage to tissues and plant cells in plants under drought stress, N, Cu, B, Zn, P, Ca, and K values decrease (Gür, 2018). A high amount of sugar deposition occurs in plant tissues damaged by drought stress, which significantly hinders photosynthesis and causes stunted growth and necrotic leaves to form in the plant (Sami et al., 2016).

Drought conditions also change the plant cell turgor pressure, i.e. the amount of water potential. It is very important to maintain the osmotic balance in order for plant cells to be minimally affected by water stress. For this purpose, when plants perceive drought stress, they synthesize a group of soluble substances called "osmolytes", which play a role in maintaining cell turgor balance and accumulate in their cells. Examples of osmolyte substances can be given from different groups such as free amino acids such as asparagine, proline and glycine, betaine, organic acids, and carbohydrates. Osmolytes that accumulate in plant cells and play a role in maintaining water balance do not directly affect or increase the plant's resistance to drought stress. However, as they balance the intra-leaf water pressure, they increase the conductivity of the stomata, ensure the continuity of photosynthesis, and thus help growth. Osmotic regulation is one of the important components in adapting to drought stress. The leaf water potential helps to protect the cellular turgor and thus delays fading. The preservation of water balance and the ability to maintain cell metabolic activities through osmolytes, albeit limited, provide short-term resistance to plants in arid conditions. If drought and stress conditions persist for a long time, osmolyte deposition will not be enough to compensate for the loss of turgor caused by lack of water and will not prevent the plant from being affected by stress. The main intracellular function of proline is to protect membrane systems by preventing lipid oxidation and protecting protein structures through the compounds they form. It reveals that proline may also be involved in signal transduction and may be an important free amino acid that plays a role in regulating mitochondrial functions, cell division or death, and even gene expression levels (Anjum et al., 2011; Liang et al., 2018; Kishor & Sreenibasulu, 2014). As drought stress factors increase in drought-resistant plants, proline synthesis by the plant increases, providing resistance to structural denaturation caused by drought stress, and plant cell structures are protected by proline, which is responsible for protein synthesis (Xoconostle-Cazares et al., 2010). Osmolytes, which are protective molecules, are proteins that function in the cleaning of ROS caused by stress. They function as osmotic regulators and osmoprotectants. They retain water in the cytoplasm and protect the health of cellular structures by facilitating the retention of sodium in apoplasts and vacuoles (Beshir et al., 2016). If the stress conditions persist for a long time, the deposition of osmolytes will not be enough to compensate for the loss of turgor caused by lack of water (Ozturk, 2015). Some reactive oxygen species are formed in hydroxy (OH), superoxide (O_2^-), hydrogen peroxide (H_2O_2), and singlet oxygen ($^1O_2$) in drought conditions. Reactive oxygen compounds that deposit under stress conditions are actually a natural by-product of cell metabolism and are components that play an important role in the signal transduction mechanism (Anjum et al., 2011; Cabello et al., 2014). In case of excessive deposition of these components, they can lead to cell death by inducing lipid peroxidation, protein reduction, and DNA fragmentation. In addition to the negative effects of the components, reducing and preventing the deposition of these reactive oxygen compounds formed during drought stress is an important factor in the plants' struggle with stress conditions and even their survival. Plants struggling with oxidative stress caused by the deposition of reactive oxygen compounds use enzymatic (superoxide dismutase, peroxidase, catalase, ascorbate peroxidase) or non-enzymatic (glutathione, ascorbate, tocopherols, carotenoids) antioxidant molecules. While the main task of non-enzymatic antioxidant molecules is explained as the protection of photosynthetic membranes, enzymatic antioxidant molecules prevent their deposition by reducing reactive oxygen compounds (Farooq et al., 2009; Anjum et al., 2011; Dolferus, 2014; Osakabe et al., 2014). It can be determined whether oxidative signal or damage will occur in the plant thanks to the balance established between the amount of reactive oxygen ROS produced by the plant exposed to stress and the antioxidant enzyme activity to counteract it (Moller et al., 2007). The capture of reactive oxygen ROS by plant enzymes and their ability to reduce their harmful effects may be related to the drought tolerance of plants (Tsugane et al., 1999).

It has been determined that the artificial application of various plant growth regulators and osmotic preservatives such as cytokinin, abscisic acid, proline, glycine betaine, polyamine, and salicylic acid to plants has significant potential in increasing the plant's drought tolerance (Travaglia et al., 2007). It has been reported that ABA, known as the stress hormone in leaves, is synthesized in the roots during dry periods, causing stomata to close and changes in the expression of genes (Stellfeldt et al., 2018).

RESULT

In general, plants encounter many negative factors such as salinity, drought, pollution, heat, and cold throughout their lives, and growth and development events under normal conditions are damaged by these conditions. These factors that cause negative effects on the development and behavior of plants and the plant's different reactions to these conditions are generally defined as stress. When plants are exposed to these stress conditions, the faster they react to adverse conditions within their own bodies, the greater their chances of getting rid of adverse conditions. Of course, the reaction to stress conditions will be possible in drought-resistant or tolerant plants. Plants that are not drought tolerant should be excluded from this issue. It will be possible to develop plants with a high tolerance level that can provide better adaptation to environmental conditions in order not to disrupt production. It will be possible to produce these species in drought regions by transferring resistance genes to such plants by conducting breeding studies with hybridization or other gene transfer methods in plants that do not have tolerance and resistance to stress conditions. In addition, soil and water erosion will be prevented by using protective tillage methods instead of traditional tillage in semi-drought and drought regions, and it will be possible to reduce production costs by keeping soil moisture for a longer time. In our world where we feel the effects of global warming stronger day by day, and in our world where food shortages may occur due to the increase in population density, decrease in arable land, and destruction of water resources in the future, it has become very important to reduce product losses caused by stress. It is our duty to take action on a global scale and put a stop to this global climate change before it is too late.

REFERENCES

Vow, N., Gubbuk, H., & Trigger, N. (2018). Yield, Quality and Biochemical Properties of Various Strawberry Cultivars Under Water Stress. J Sci Food Agric., *98*(1), 304-311.

Adams, H. D., Guardiola-Clamonte, M., Barro Gafford, G. A., Villagas, J. C., Breshears, D. D., & Zou, C. B. (2009). Temperature sensitivity of droughts-induced tree mortality ports increased regional die-off under global-change-type droughts, Proceedings of the National Academy of Sciences, U.S.A. (106), 7063-7066. https://pubmed.ncbi.nlm.nih.gov/19365070/

Akhtar, I., & Nazir, N. (2013). Effect of waterlogging and drugged stress in plants. International Journal of Water Resources and Environmental Science, (2), 34-40. https://idosi.org/ijwres/2(2)13/3.pdf

Akıncı, S., & Lösel, D. M. (2012). Plant Water-Stress Response Mechanisms, Water Stress. Ismail Md. Mofizur Rahman (Ed.), Available from: http://www.intechopen.com/books /water-stress/plant-water-stress-response-mechanisms [Accessed: 11.11.2022].

Anjum, S. A., Xie, X., Wang, L., Saleem, M. F., Man, C., & Lei, W. (2011). African Journal of Agricultural Research, *6*(9), 2026-2032.

Arbona, V., Manzi, M., de Ollas, C., & Gómez-Cadenas, A. (2013). Metabolomics as a tool to investigate abiotic stress tolerance in plants. International Journal of Molecular Sciences, (14), 4885-911. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3634444/

Arora, A., Sairam, R. K., & Srivastava, G. C. (2002). Oxidative stress and antioxidant systems in plants, Curr. Sci., (82), 1227–1238. https://www.researchgate.net/publication/237817932_Oxidative_stress_and_antioxidative_system_in_plants#fullTextFileCont

Barriopedro, D., Gouveia, C., Trigo, R. M., & Wang, L. (2012). The 2009/10 droughts in China: possible causes and impacts on vegetation, American Meteorological Society, (13), 1251-67. https://journals.ametsoc.org/view/journals/hydr/13/4/jhm-d-11-074_1.xml

Bray, E. (1997). Plant responses to water deficit. Trends in Plant Science, (2), 48-54.

Bernacchia, G., & Furini, A. (2004). Biochemical and molecular responses to water stress in resurrection plants, Physiology Plant, (121), 175-81. https://onlinelibrary.wiley.com/doi/10.1111/j.1399-3054.2004.00321.x

Beshir, H. M., Bueckert, R.., & Scan, B. (2016). Effect of Temporary Drought at Different Growth Stages on Snap Bean Pod Quality and Yield. African Crop Science Journal, *24*(3), 317 – 330. https://www.researchgate.net/publication/308859654_Effect_of_temporary_drought_at_different_growth_stages_on_snap_bean_pod_quality_and_yield

Bian, S., & Jiang, Y. (2009). Reactive oxygen species, antioxidant enzyme activities and gene expression patterns in leaves and roots of Kentucky bluegrass in response to drugged stress and recovery. Scientia

Horticulturae, (120), 264-270. https://srv2.freepaper.me/n/WSz1mCIn3YKdBmK-AK6i9w/PDF/02/027b8b8fa38324acd61f45533bd3123a.pdf

Bhargava, S., & Sawant, K. (2013). Drought stress adaptation: metabolic adjustment and regulation of gene expression. Plant Breeding, (132), 21-32. https://onlinelibrary.wiley.com/doi/epdf/10.1111/pbr.12004

Boscaiu, M., Lull, C., Lydon, A., Bautista, I., Donat, P., & Mayoral, O. (2008). Plant responses to abiotic stress in their natural habitats. Bulletin UASVM, Horticulture, *65*(1), 53-58. https://www.researchgate.net/profile/Oscar-Vicente-2/publication/266881140_Plant_responses_to_abiotic_stress_in_their_natural_habitats/links/544521a00cf2dccf30b8fd2e/Plant-responses-to-abiotic-stress-in-their-natural-habitats.pdf

Barlow, E. W. R., Lee, J. W., Munns, R.., & Smart, M. G. (1980). Water relations of growing wheat grains. Australian Journal of Plant Physiology, (7), 519-525.

Büyük, İ., Soydam-Aydın, S., & Aras, S. (2012). Bitkilerin stres koşullarına verdiği moleküler cevaplar. Türk Hijyen ve Deneysel Biyoloji Dergisi, *69*(2), 97-110. https://www.researchgate.net/publication/271194163_Bitkilerin_Stres_Kosullarina_Verdigi_Molekuler_Cevaplar#fullTextFileContent

Cabello, J. V., Lodeyro, A. F., & Zurbriggen, M. (2014). Novel perspectives for the engineering of abiotic stress tolerance in plants. Current Opinion in Biotechnology, (26), 62-70. https://www.sciencedirect.com/science/article/pii/S0958166913006733?via%3Dihub

Capell, T., Bassie, L., & Christou, P. (2004). Modulation of the polyamine biosynthetic pathway in transgenic rice confers tolerance to droughted stress. Proceedings of the National Academy of Sciences, *101*(26), 9909-9914. https://www.pnas.org/doi/epdf/10.1073/pnas.0306974101

Caruso, A., Chefdor, F., Carpin, P., Depierreux, C., Delmotte, F. M., Kahlem, G., & Morabito D. (2008). Physiological characterization and identification of genes differently expressed in response to drug-induced by PEG 6000 in Populus canadensis leaves. Journal of Plant Physiology, (165), 932-941. https://www.sciencedirect.com/science/article/pii/S0176161707002192?via%3Dihub

Chaves, M. M., Maroco, J. P., & Pereira, J. S. (2003). Understanding plant responses to drought-from genes to the whole plant, Function. Plant Biol. (30), 239–264. https://pubmed.ncbi.nlm.nih.gov/32689007/

Çırak, C., & Esendal, E. (2006) Soyada kuraklık stresi, Anadolu Tarım Bilimleri Dergisi, *21*(2), 231-237. https://dergipark.org.tr/tr/download/article-file/187624

Dai, A. (2012). Drought under global warming: a review. Wires Climate Change, (2), 45-65.

Desoky, E. M., Rady, M. M., & Merwad, M. A. (2018). Response of Water Deficit-Stressed Vigna Unguiculata Performances to Silicon, Proline, or Methionine Foliar Application. Scientia Horticulturae, (228), 132-144. https://www.sciencedirect.com/science/article/pii/S030442381730612X?via%3Dihub

Dolferus, R. (2014). To grow or not to grow: A stressful decision for plants. Plant Sci., (2229), 247-261.

Faizi, Z. A., & Ozturk, A. (2022). Yumuşak Çekirdekli Meyve Türlerinde Kuraklığın Etkileri. Iğdır Üniversitesi Fen Bilimleri Enstitüsü Dergisi, *12*(3), 1224- 1237.

Farooq, M., Wahid, A., Kobayashi, N., Fujita, D., & Basra, S. M. A. (2009). Plant drought stress: effects, mechanisms, and management. Sustainable agriculture, 153-188.

Glombitza, P., Dubuis, P. H., Thulke, O., Welzl, G., Bovet, L., Gotz, M., Affenzeller, M., Geist, B., Hehn, A., Asnaghi, C., Ernst, D., Seidlitz, H. K., Gundlach, H., Mayer, K. F., Martinoia, E., Werck-Reichhart, D., Mauch F., & Schaffner A. R. (2004). Crosstalk and differential response to abiotic and biotic stressors reflect the transcriptional level of effector genes from secondary metabolism. Plant Molecular Biology, (54), 817-835. https://link.springer.com/article/10.1007/s11101-006-9011-7

Goksoy, A. T., & Turan, Z. M. (1991). Kuraklığın bitki fizyolojisi ve morfolojisi üzerine etkileri. Uludağ Üniversitesi Ziraat Fakültesi Dergisi, (8), 149-190. https://acikerisim.uludag.edu.tr/bitstream/11452/16614/1/8_1_20.pdf

Gür, İ. (2018). Effects of Water Stress Applied on Some Pear Rootstocks for Morphological and Biochemical Changes. Suleyman Demirel University, Ph.D. Thesis.

Günay, A. (2005). Sebze Yetiştiriciliği. Volume I, ISBN 975-00725-0-2, Izmir.

Gür, İ. (2018). Effects of Water Stress Applied on Some Pear Rootstocks for Morphological and Biochemical Changes. Suleyman Demirel University, Ph.D. Thesis (printed).

Gurel, A., & Avcioglu, R. (2001). Bitkilerde Abiyotik Stres Faktörlerine Dayanıklılık Mekanizmaları. Bitki Biyoteknolojisi, Genetik Mühendisliği, SÜ Vakfı Yayınları, Izmir, 288-326.

Hale, M. G., & Orcutt, D. M. (1987). The Physiology of Plants Under Stress, (206 pp).

Hekimoğlu, B., & Altındeğer, M. (2008). Küresel Isınma, Tarımsal Kuraklık ve Samsun Tarımına Etkileri. Küresel Isınma ve İklim Değişikliği. T.C. Samsun Valiliği ve İl Tarım Müdürlüğü. Samsun, (77pp).

Hirayama, T., & Shinozaki, K. (2010). Research on plant abiotic stress responses in the post-genome era: past, present and future, The Plant Journal, (61), 1041-1052. https://onlinelibrary.wiley.com/doi/10.1111/j.1365-313X.2010.04124.x

Jaleel, C. A., Manivannan, P., Wahid, A., Farooq, M., Somasundaram, R., & Panneerselvam R., (2009). Droughted stress in plants: a review on morphological characteristics and pigments composition, International Journal of Agricultural Biology, (11), 100-5. https://www.researchgate.net/publication/253008137_Drought_Stress_in_Plants_A_Review_on_Morphological_Characteristics_and_Pigments_Composition

Jing-Min, Z., Shang-Jun, X., Mao-Peng, P., Bingyao, M., Xiu-Mei, C., & Chunsheng, L. (2010). Effect of Humic Acid on Poplar Physiology and Biochemistry Properties and Growth Under Different Water Level. Soil Water Conserv, (6), 200-203.

Kaçar, B. (2015). Genel Bitki Fizyolojisi, Nobel Akademik Yayıncılık, Yayın No: 1243, Ankara.

Kalefetoğlu, T., & Ekmekçi, Y. (2005). Bitkilerde kuraklık stresinin etkileri ve dayanıklılık mekanizmaları. G.U. Fen Bilimleri Dergisi, *18*(4), 723-740.

Kapluhan, E. (2013). Türkiye'de kuraklık ve kuraklığın tarıma etkisi, Marmara Coğrafya Dergisi, (27), 487-510. https://dergipark.org.tr/tr/pub/marucog/issue/474/3913

Kayabaşı, S. (2011). Kuraklık stresinde yetiştirilen soyada (Glycine max L.) bazı fizyolojik parametreler ile prolin birikiminin araştırılması. Yüksek Lisans Tezi, Harran Üniversitesi, Şanlıurfa.

Kaynaş, N., & Kaynaş, K. (2003). Klon anaçları üzerine aşılı Angelona erik çeşitinin su stresi koşullarındaki fizyolojik değişimleri. Türkiye 4. Bahçe Bitkileri Kongresi, 08-12 Eylül, Antalya.

Kishor, P. B., & Sreenivasulu, N. (2014). Is proline accumulation per se correlated with stress tolerance or is proline homeost Thus a more critical issue? Plant Cell Environ., (37), 300-311. https://onlinelibrary.wiley.com/doi/epdf/10.1111/pce.12157

Kutlu, İ. (2010). Drought stress in cereals, Turkish Journal of Scientific Reviews, *3*(1), 35-41.

Küçükyumuk, C. (2020). Droughted Response of Young Pear Trees (Pyrus comminus). Applied Ecology and Environmental Research, 18(6): 7769-7781. https://www.aloki.hu/pdf/1806_77697781.pdf

Levitt, J. (1980). Responses of plants to environmental stresses. Academic Press, (497 pp).

Liang, B., Gao, T., Zhao, Q., Ma, C., Chen, Q., Wei, Z., & Ma, F., (2018). Effects of exogenous dopamine on the uptake, transport, and resorption of apple ionome under moderate droughts. Frontiers in plant science, (9), 755.

Lichtenhaler, H. K. (1996). Vegetation stress: An introduction to the stress concept in plants. J Plant Physiol, (148), 4-14.

Lipiec, J., Doussan, C., Nosalewicz, A., & Kondracka, K. (2013). Effect of drought and heat stresses on plant growth and yield: a review, International Agrophysics, *27*(4), 463- 477. https://ui.adsabs.harvard.edu/abs/2013InAgr..27..463L/abstract

Lisar, S. Y., Motafakkerazad, R., Hossain, M. M., & Rahman, I. M. (2012). Water stress in plants: causes, effects and responses, water stress, InTech, Croatia, 1-14.

Mafakheri, A., Siosemardeh, A., Bahramnejad, B., Struik, P. C., & Sohrabi, E. (2010). Effect of drug stress on yield, proline, and chlorophyll contents in three chickpea cultivars. Amer J Chin Study, (4), 580-585.

Mahajan, S., & Tuteja, N. (2005). Cold, salinity and droughted stresses: An overview. Archives of Biochemistry and Biophysics, (444), 139-158.

Marcińska, I., Czyczyło-Mysza, I., Skrzypek, E., Filek, M., Grzesiak, S., Grzesiak, M. T., Janowiak, F., Hura, T., Dziurka, M., Dziurka, K., Nowakowska, A., & Quarrie, S. A. (2013). Impact of osmotic stress on physiological and biochemical characteristics in drought-susceptible and drought-resistant wheat genotypes. Acta Physiol Plant, (35), 451-461.

Moller, I. M., Jensen, P. E., & Hansson, A. (2007). Oxidative modifications to cellular components in plants, Annual Review of Plant Biology, (58), 459-481.

Mundree, S. G., Baker, B., Mowla, S., Peters, S., Marais, S., Willigen, C. V., Govender, K., Maredza, A., Muyanga, S., Farrant, J. M., & Thomson, J. A. (2002). Physiological and molecular insights into drug tolerance. Afr. J. Biotechnol, (1), 23- 38.

Nader, K. B., Stoll, M., Rauhut, D., Patz, C. D., Jung, R., Loehnertz, O., & Gomès, E. (2019). Impact of Grapevine Age on Water Status and Productivity of Vitis vinifera L. cv. Riesling. European Journal of Agronomy, (104), 1-12. https://pubag.nal.usda.gov/catalog/6266126

Nezhadahmadi, A., Hossain Prodhan, Z., & Faruq, G., (2013). Drug tolerance in wheat, The Scientific World Journal, 1-12.

Noctor, G., Reichheld, J. P., & Foyer, C. H. (2018). ROS-Related Redox Regulation and Signaling in Plants. In Seminars in Cell & Developmental Biology, (80), 3-12. https://www.sciencedirect.com/science/article/pii/S108495211730246X?via%3Dihub

Osakabe, Y., Osakabe, K., Shinozaki, K., & Tran, L. P. (2014). Response of plants to water stress. Front Plant Science, (5), 86. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3952189/

Özcan, S., Babaoğlu, M., & Gürel, E. (2004). Bitki Biyoteknolojisi Genetik Mühendisliği ve Uygulamaları, S.Ü. Vakfı Yayınları, Konya.

Ozturk, K. (2002). Küresel İklim Değişikliği ve Türkiyeye Olası Etkileri. Gazi Üniversitesi Gazi Eğitim Fakültesi Dergisi, *22*(1).

Ozturk, N. Z. (2015). Bitkilerin kuraklık stresine tepkilerinde bilinenler ve yeni yaklaşımlar. Türk Tarım-Gıda Bilim ve Teknoloji Dergisi, *3*(5), 307-315. https://www.researchgate.net/publication/316444169_Bitkilerin_Kuraklik_Stresine_Tepkilerinde_Bilinenler_ve_Yeni_Yaklasimlar

Pou, A., Medrano, H., Flexas, J., & Tyerman, P. D. (2013). A Putative Role for MEDICINE and PIP Aquaporins in Dynamics of Leaf Hydraulic and Stomatal Conductance in Grapevine under Water Stress and Re-Watering. Plant, Cell & Environment, *36*(4), 828-843.

Rahdari, P., & Hoseini, S. M., (2012). Droughted stress: a review, International Journal of Plant Production, (3), 443-6.

Romo, S., Labrador, E., & Dopico, B. (2001). Water stress-regulated gene expression in Cicer arietinum seedlings and plants. Plant Physiol. Biochem., (39), 1017−1026.

Sami, F., Joseph, M., Faizan, M., Faraz, A., & Life, S. (2016). The Role of Sugars under Abiotic Stress. Plant Physiology and Biochemistry, (109), 54-61. https://www.researchgate.net/publication/307999199_Role_of_sugars_under_abiotic_stress

Sapeta, H., Costa, M., Lourenc, T., Marocod, J., Van der Linde, P., & Oliveira, M. M. (2013). Drug stress response in Jatropha curcas: growth and physiology, Environmental and Experimental Botany, (85), 76-84.

Sharma, S., & Dubey, R. S. (2005). Drought induces oxidative stress and enhances the activities of antioxidant enzymes in growing rice seedlings. Plant Growth Regulation, (46), 209-221.

Shulaeva, V., Cortesa, D., Miller, G., & Mittler, R. (2008). Metabolomics for plant stress response, Physiologia Plantarum, (132), 199-208.

Siddiqui, M. H., Al-Khaishany, M. Y., Al-Qutami, M. A., Al-Whaibi, M. H., Grover, A. M., Ali, H., Al-Wahibi, M. S., & Bukhari, N. A. (2015). Response of Different Genotypes of Faba Bean Plant to Drug Stress. Int. J. Mol. Sci., (16), 10214-10227. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4463642/pdf/ijms-16-10214.pdf

Sircelj, H., Tausz, M., Grill, D., & Batic, F. (2007). Detecting different levels of droughted stress in apple trees (Malus domestica Borkh.) with selected biochemical and physiological parameters. Scientia Horticulturae, *113*(4), 362-369.

Smirnoff, N. (1993). The role of active oxygen in the response of plants to water deficit and desiccation. New Phytol., (125), 27-58.

Stellfeldt, A., Maldonado, M. A., Hueso, J. J., & Cuevas, J. (2018). Gas Exchange and Water Relations of Young Potted Loquat cv. Algerie under Progressive Droughts Conditions. Journal of Integrative Agriculture, *17*(6), 1360-1368.

Travaglia, C., Cohen, A. C., Reinoso, H., Castillo, C., & Bottini, R. (2007). Exogenous abscisic acid increases carbohydrate accumulation and redistribution to the grains in wheat grown under field conditions of soil water restriction. J. Plant Growth Regulation., (26), 285–289.

Tsugane, K., Kobayashi, K., Niwa, Y., Ohba, Y., Wada K., & Kobayashi H. (1999). A recessive Arabidopsis mutant that grows photoautotrophically under salt stress shows enhanced active oxygen detoxification, Plant Cell, (11), 1195-1206.

Turkan, I., Bor, M., Ozdemir, F., & Koca, H. (2005). Differential Responses of Lipid Peroxidation and Antioxidants in the Leaves of Drug-Tolerant P.A cutifolius Gray and Droughty Sensuous P. vulgaris L. Subject to Polyethylene Glycol Mediates Water Stress. Plant Science, (168), 223-231.

Tyerman, S. D., Niemietz, C. M., & Bramley, H., (2002). Plant Aquaporins: Multifunctional Water and Solution Channels with Expanding Roles. Plant, Cell & Environment, *25*(2), 173-194.

Wang, W., Vinocur, B. & Altman, A. (2003). Plant responses to droughts, salinity and extreme temperatures: towards genetic engineering for stress tolerance. Planta, *218*(1), 1-14.

Wang, Y. T., Chen, Z. Y., Jiang, Y., Duan, B. B., & Xi, Z. M. (2019). Involvement of ABA and antioxidant system in brassinosteroid-induced water stress tolerance of grapevine (Vitis vinifera L.). Scientia Horticulturae, (256), 1-10.

Xoconostle- Cazares, B., Ramírez- Ortega, F. A., Flores-Elenes, L., & Ruiz- Medrano, R. (2010). Drug tolerance in crop plants, American Journal of Plant Sciences, (5), 241-256

Zare, M., Saint, M. H., & Bazrafshan, F. (2011). Effect of droughted stress on some agronomic traits in ten barley (Hordeum vulgare) cultivars, Technical Journal of Engineering and Applied Sciences, (1), 57-62. https://www.researchgate.net/publication/317358960_Effect_of_Pre-Anthesis_Water_Deficit_on_Plant_Height_Peduncle_Length_and_Spike_Length_in_13_Barely_Hordeum_vulgare_L_Genotypes#fullTextFileContent

Zargar, S. M., Gupta, N., Nazir, M., Mahajan, R., Malik, F. A., & Sofi, N. R. (2017). Impact of droughts on photosynthesis: molecular perspective, Plant Gene, (11), 154-159.

Zeppel, M. J. B., Harrison, P. P., Adams, H. D., Kelley, D. I., Li, G., & Tissue, D. T. (2015). Drought and resprouting plants, New Phytologist, (206), 583-589. https://pubmed.ncbi.nlm.nih.gov/27283977/

Zlatev, Z., & Lydon, F. C. (2012). An overview on droughted induced changes in plant growth, water relation, and photosynthesis, Emirates Journal of Food and Agriculture, (24), 57-72. https://www.researchgate.net/publication/266870948_An_overview_on_drought_induced_changes_in_plant_growth_water_relations_and_photosynthesis#fullTextFileContent

DRAFT

ORAL PRESENTATION

Comparative Biochemical Analysis of Brown And Green Pea Genotypes Grown Locally in Türkiye

Fatih HANCI [1]* (ORCID: https://orcid.org/0000-0002-2015-0351), Berk Can YILDIZ [1] (ORCID: https://orcid.org/0009-0004-6685-2094), Emir Furkan DEMİR [1] (ORCID: https://orcid.org/0009-0005-9019-3217)

*[1]Erciyes University, Faculty of Agriculture, Department of Horticulture, Kayseri, Türkiye.
[2] University, Faculty, Department, City, Country.

*Corresponding author e-mail: fatihhanci@erciyes.edu.tr

Abstract

In this study, two pea genotypes with different seed coat colors (green and brown) grown locally in Turkey were investigated based on biochemical components. The samples extracted from the grains of these two genotypes were evaluated. Total water-soluble protein, total flavonoids, total phenolics, H2O2 scavenging capacity, carotenoids, FRAP antioxidant capacity, CUPRAC antioxidant capacity, and total dry matter parameters were measured. For all the parameters examined, the difference between pea seeds with brown and green seed coat colors was statistically significant. In general, the studied characteristics of green peas were higher than brown peas. only for H2O2 scavenging capacity and dry matter content brown peas had higher results.

Key words: Brown pea, carotenoid, flavonoid, Pisum

INTRODUCTION

Edible grain legumes have many superiorities in terms of nutritional value compared to other plant-based foods. Their seeds contain 20-25% protein. There are some components in legume seeds that have positive or negative effects on their nutritional value. While anti-nutritional elements including enzyme inhibitors, lectins, gassing agents, polyphenols, tannins, phytic acid, and saponins can sometimes have detrimental impacts on nutrition and health, high protein, low fat content, vitamins, minerals, and dietary fibers often have good benefits (Pekşen and Artık, 2005).

Approximately 2/3 of the world's population lives on a grain-based diet that lacks sufficient protein. (Ekingen, 1992). Among the vegetable protein sources, the most The most protein is produced from edible grain legumes (Şehirali, 1988). Possessing due to its amino acid composition and acceptable functional properties peas are an alternative vegetable that can be used to enrich food products. protein source (Tömösközi, et al., 2001).

In 2021, the world dry pea cultivation area, which is the subject of field crops discipline, was 7.043.605 ha and the production amount was 12.403.521 tons (Anoymous, 2023). The annual production amount of green peas, which is the subject of the horticulture discipline, is 20.529.759 tons. This production was carried out on an area of 259.0367 ha. In 2020, the dry pea cultivation area in Turkey was only 679 ha and the production amount was 1805 tons. However, the production amount of green peas is 111625 tons on an area of 11.647 ha. Turkey ranks almost last in world dry pea production (87th among 95 countries). In fresh pea production, Turkey ranks 45th among 82 countries.

Turkey has a very important position in the world in terms of plant gene resources due to its geographical structure and different ecological conditions. Genetic material that fully adapts to the ecological conditions of the regions where they grow is the assurance of the future of agriculture and thus of humanity (Özgen, et al., 2000).

Antioxidants have long piqued the curiosity of the scientific community and the general public due to their ability to defend against free radicals (Bhoyar et al., 2018). Although there is information on the nutritional makeup of green peas, there is still a huge knowledge vacuum when it comes to the varietal assessment of antioxidant chemicals. There is a lack of research as well, much of which concentrates on methods for

extracting antioxidants (Xu and Chang, 2008; Oomah et al., 2011) and the antioxidant capabilities of green-pea seeds or seed coatings (Amarowicz et al., 2004; Petchiammal and Hopper, 2014).

The main hypothesis of this study was that there is a difference between some biochemical contents of pea grains with brownish seed coat color and pea grains with conventional green color.

MATERIALS AND METHODS

The experiment's agroclimatic site was in Turkey's Kayseri district in Middle Anatolia. The pH of the soil in the experimental plot was 7.1, making it neutral. Two different genotypes of brown and green peas were the plant material. Plants were grown in Erciyes University Cool Climate Vegetables Production and Research Area (ERUcsvra). Analyses were performed in the physiology laboratory of the same university. The procedure described by Danilcenko et al. (2017) was used to prepare data for analysis on pea seeds. The materials were completely dried at 60 degrees Celsius for 24 hours before being ground up in a plant grinder. Each clone's and organ's 3 g of powdered materials was homogenized for 2 minutes with 25 cc of pure methanol before being kept at +4°C for 16 hours. The supernatant was taken out and centrifuged for 20 minutes at 10,000 rpm for analysis (Thaipong et al. 2006). Both the copper (II) ion reducing antioxidant capacity determination (CUPRAC) method and the ferric reducing antioxidant power methodology (FRAP) (Zhang et al., 2013) were used to assess antioxidant capacity. The total quantity of phenolic compounds was determined using Singleton and Rossi's (1989) Folin-Ciocalteu method. Using the method outlined by Zhishen et al., the total quantity of flavonoids was determined. The technique suggested by Ruch et al. was used to calculate the hydrogen peroxide removing capacity (H2O2). The amount of soluble protein was determined using a method that Lowry et al. (1951) published.

To assess the significance of the data's variance, an ANOVA test was utilized. The least significant difference (LSD) test was used to compare results between groups when significant differences were discovered.

RESULTS and DISCUSSION

The differences between the results obtained for all measured parameters were statistically significant. In all parameters except H_2O_2 scavenging capacity and total dry matter percentage, pea seeds with green shell color had higher results. According to the results, green pea seeds had the highest total carotenoid concentration (0.785 g/g DW), followed by brown pea seeds (0.679 g/g DW) (Table 1).

Table 1. Some biochemical characteristics of brown and green peas

Seed Color	Chlorophyll a (mg/g)	Chlorophyll b (mg/g)	Carotenoid (mg/g)	Soluble Protein (mg/ml)	Dry weight (%)
Brown	1,921 b	0,998 b	0,679 b	12,103 b	25,957 a
Green	7,187 a*	4,880 a	0,785 a	21,563 a	15,802 b
LSD	*0,239*	*0,160*	*0,033*	*0,793*	*0,975*

*Significant ($p<0.01$), Means within a group that have a different small letter are significantly different from each other

In terms of H_2O_2 scavenging capacity, brown pea seeds had higher values than green seeds (32.609% and 8.633%, respectively) (Table 2). Regarding total phenolic matter concentration, green pea seeds ranked in the lead (11.529 GAE mg/g). Brown peas were found to have the lowest phenolic content (5.603 GAE mg/g). The ranking of the analyzed peas according to total flavonoid content was the same as for total phenolic matter. The total flavonoid content of green shelled seeds was 1.86 times higher than that of brown shelled seeds (84.238 and 45.190, respectively).

Table 2. Antioxidant capacities and some other biochemical properties peas

Seed Color	H_2O_2 Removal capacity (%)	Total Phenolic Content (GAE mg/g)	Total Flavonoid Content (mg Quercetin/g)	FRAP (mM Tloroks/g)	CUPRAC (mM Troloks /g)
Brown	32,609 a	5,603 b	45,190 b	7,698 b	5,074 b
Green	8,633 b	11,529 a	84,238 a	10,804 a	5,417 a
LSD	*1,081*	*0,411*	*3,065*	*0,426*	*0,238*

*Significant ($p<0.01$), Means within a group that have a different small letter are significantly different from each other

Various methods may be used to assess the antioxidant activity of plant extracts, but no single standard has been created because of how complex the extracts are. In the current study, the antioxidant activity of pea varieties with two distinct seed coatings was assessed using the CUPRAC radical scavenging test and the FRAP assay. The highest value was found using FRAP research in green pea seeds (10.804). Brown-shelled pea values (7.698) came after that. The CUPRAC results showed the same arrangement. Green-shelled pea seeds showed the highest scores in our investigation (5.417 and 5.074, respectively).

Evaluation and selection of variations with one or more desired traits are necessary for breeding and processing methods. The results of this study showed that two pea cultivars cultivated under same conditions but with varied seed coat colors showed substantial variance in their biochemical characteristics.

According to several studies (Leonardo and Doré, 2011; Vajdy, 2011), flavonoids have several significant biological actions, including impacts on photoaging, allergies, inflammation, and cardio protection. Additionally, according to Amarowicz et al. (2007) and Troszyska and Ciska (2002), flavonoids are the primary source of antioxidant activity in plants. In order to replace the usage of artificial antioxidants in meals, peas might be employed as a natural source of antioxidants. The total phenolic, total flavonoid, CUPRAC, and FRAP antioxidant capacities of green pea were substantially greater than those of the brown pea type, according to the results. Black soybeans, on the other hand, have a significantly higher phenolic content and antioxidant activity than light-colored beans, according to Xu and Chang (2007). Furthermore, Shem-Tov et al. (2012) discovered that FRAP, total flavonoid, and total phenolic content were greater in chickpea seeds with dark, yellow, and high color intensity compared to seeds with lighter colors. Red and black sword beans have higher phenolic contents and antioxidant capabilities than white sword beans (Gan et al., 2016). Except for hydrogen peroxide scavenging capabilities, these results go against everything we found. The total phenolic content of the green and brown pea genotypes evaluated in the current experiment ranged from 5,603 to 11,529 mg GAE/g and from 45,190 to 84,238 mg CE/g, respectively, with little variation between them. These findings have higher confidence than those of Klepacka et al. (2011), who found that the phenolic matter concentration of pea cultivars ranged from 28 to 42 mg/100 g. They agreed with the stated relatively little amount of variance, though.

CONCLUSION

The importance of pea, a substantial source of protein and minerals in the vegan diet, is increasing due to the various benefits associated with hunger control and intake. The study clearly demonstrated significant genetic diversity in the two pea genotypes examined for biochemical contents and their relationship to seed-coat color. The green pea variety had the highest total flavonoid content, total phenolic content, antioxidant activity and carotenoid values, brown pea seeds may have potential health benefits for capacity with more H2O2 scavenging capacity than light-colored seeds.

ACKNOWLEDGEMENTS

This study was supported within the scope of 2209-A- Research Project Support Programme for Undergraduate Students.

REFERENCES

Alam MS, Kaur G, Javed K, Athar M 2007. *Eruca sativa* seeds possess antioxidant activity and exert a protective effect on mercuric chloride induced renal toxicity. Food and Chemical Toxicology, 45: 910-920.

Dawson SS, Michler CH 2014. Afforestation, restoration and regeneration - Not all trees are created equal. Journal of Forestry Research, 25(1): 3-20.

Amarowicz R, Naczk M, Zadernowski R, Shahidi F 2000Antioxidant activity of condensed tannins of beach pea, canola hulls, evening primrose, and faba bean. Journal of Food Lipids, 7(3), 195-205,.

Amarowicz R, Troszynska A, Barylko-Pikielna N, Shahidi F 2004. Polyphenolics extracts from legume seeds: correlations between total antioxidant activity, total phenolics content, tannins content and astringency. J. Food Lipids 11, 278–286.

Anonymous 2023. Food and Agriculture Organization of the United Nations, Statistics Division, http://faostat3.fao.org/home/E, Accessed on 29.08.2023.

Bhoyar MS, Mishra GP, Naik PK, Singh SB 2018. Evaluation of antioxidant capacities and total polyphenols in various edible parts of Capparis spinosa L. Collected from Trans-Himalayas. Def. Life Sci. J. 3, 30–36.

Gan RY, Lui WY, Corke. 2016. Sword bean (*Canavalia gladiata*) as a source of antioxidant phenolics. International journal of food science & technology, 51(1), 156-162.

Klepacka J, Gujska E, Michalak J 2011. Phenolic compounds as cultivar- and variety distinguishing factors in some plant products. Plant Foods Hum. Nutr. 66, 64–69.

Leonardo CC, Doré S 2011. Dietary flavonoids are neuroprotective through Nrf2- oordinated induction of endogenous cytoprotective proteins. Nutritional Neuroscience, v.14, p.226- 236.

Oomah BD, Caspar F, Malcolmson LJ, Bellido AS 2011. Phenolics and antioxidant activity of lentil and pea hulls. Food Res. Int. 44, 436–441.

Pekşen E, Artık C 2005. Antinutritional Factors and Nutritive Values of Food Grain Legumes. Anadolu Journal of Agricultural Sciences, 20 (2), 110-120.

Petchiammal C, Hopper W 2014. Antioxidant activity of proteins from fifteen varieties of legume seeds commonly consumed in India. Int. J. Pharm. Pharm. Sci. 6, 476–479.

Shem-Tov Y, Badani H, Segev A, Hedvat I, Galili S, Hovav RAN 2012. Determination of total polyphenol, flavonoid and anthocyanin contents and antioxidant capacities of skins from peanut (*Arachis hypogaea*) lines with different skin colors. Journal of Food Biochemistry, 36(3), 301-308.

Troszynska A, Ciska E 2002. Phenolic compounds of seed coats of white and coloured varieties of pea (*Pisum sativum* L.) and their total antioxidant activity. Czech Journal of Food Sciences, 20(1), 15-22.

Vajdy M 2011. Immunomodulatory properties of vitamins, flavonoids and plant oils and their potential as vaccine adjuvants and delivery systems. Expert opinion on biological therapy, 11(11), 1501-1513.

Xu B, Chang SK 2008. Effect of soaking, boiling, and steaming on total phenolic content and antioxidant activities of cool season food legumes. Food Chem. 110, 1–13.

Xu BJ, Chang SKC 2009. Total phenolic, phenolic acid, anthocyanin, flavan-3-ol, and flavonol profiles and antioxidant properties of pinto and black beans (*Phaseolus vulgaris* L.) as affected by thermal processing. Journal of Agriculture and Food Chemistry, 57, 4754-4764.

DRAFT

ORAL PRESENTATION

Investigation of methods used in fruit thinning in apple species

Said Efe DOST[1]* *(0000-0003-4279-7292)*, Mehmet Emin DAĞ[2] *(0000-0003-3393-7855)*

[*1]Karamanoglu Mehmetbey University, Technical Science. Vocational School of Higher Education, Karaman, Turkey
[2]Karamanoglu Mehmetbey University, Department of Plant Production and Technologies, Karaman, Turkey

*saidefe.dost@gmail.com

Abstract

Apple, one of the temperate fruit types, is grown in Turkey in large quantities, especially in the Karaman region. Although Turkey is at the top of the world in apple production, it, unfortunately, lags behind many European countries in exports. One of the reasons for the low exports is mainly the low quality of production. The fact that the thinning of fruit trees in our country is not carried out correctly and to an appropriate extent is one of the main reasons for the low level of exports. Regardless of proper cultural measures such as pruning, fertilization, and irrigation at the right time and in the right way, thinking is a method that directly affects fruit quality. Thinning is done manually, mechanically, and chemically in three different ways. Because mechanical thinking damages the fruit, it is rarely used in commercial production. This method is preferred in organic farming, where the use of chemicals is limited. In our country, thinning is usually done manually, and chemical thinning is used as an alternative to save labor and production costs. Plant growth regulators such as NAA, 6-benzyl adenine, carbaryl, etephon, and NAD are used as chemical thinning agents. In this study, the factors affecting the thinning effect and success and the methods used by the producers were investigated.

Keywords: Apple, Chemical Thinning, Hormones, Thinning,

INTRODUCTION

The cultivated apple ″Malus x domestica Borkh″, which is an interspecies hybrid complex in the genus Malus, is an important fruit species recognized all over the world. The oldest archaeological remains of apples found in Anatolia, dating back to 6500 BC, show that this species has been known by humans for thousands of years (Luby 2003). Apple, a type of temperate climate fruit, is currently cultivated in a large number of countries (Özçağıran *et al.* 2011).

86 million tons of production were made according to the data of the world apple production year 2020. China ranked first with a production of 41.5 million tons, while the USA ranked and Turkey ranked second and third respectively with 4.84 million tons and 3.96 million tons (Anonymous 2022). When we look at the apple production areas of Turkey, Niğde ranks first with 235 thousand decares of orchards in 2019. When we look at the number of apple trees in the yield age, Karaman Province is one of the leading other provinces with 9.2 million trees. The provinces of Niğde, Isparta, Karaman, Antalya, and Konya constitute about 52% of the total apple production areas of Turkey with a production area of about 912 thousand decares. China and the USA, which are in the first places in terms of the amount of apple production, are in the top places as the largest exporters of the world in the apple export ranking. Although Turkey ranks 3rd in world apple production, it ranks 14th behind many countries in world apple exports (Anonymous 2022). Apple cultivation should be carried out with varieties that are accepted all over the world and modern practices and techniques that will increase quality production in these varieties should be given importance in order to improve the current situation in Turkey. The interest in different tastes and flavors in apples, as in other fruit types, especially in developed countries such as Russia, Germany, England, and the Netherlands, which are leading in apple imports with high-income levels, causes varieties with these characteristics to come to the fore in cultivation in countries with high export values (Dousti and Dumanoğlu 2011).

Fruit quality is affected by many factors such as genotype, climatic conditions, rootstock and variety characteristics, soil structure, location and direction of the garden, irrigation water and distribution, fertilizer

type and application time, dilution, and pruning. 'Thinning' is one of the main cultural processes that positively affects the quality of apple varieties, especially in many fruit types and varieties. Thinning is the process of removing flowers and small fruits on the plant from the plant, reducing the amount of fruit and lightening the product load (Karakuş and Kalyoncu 2011; Kaşka 2003).

Growers need to obtain proportionally more large and colorful fruits from their orchards in order to be able to produce in the competitive conditions required by domestic and foreign markets in apple cultivation. In other words, it has become important for producers to produce fruits of "marketable" quality (Çağlar and Balcı 2003). Thinning is especially important for apple species. If all cultivating processes are performed appropriately but without thinning, it may not be possible to obtain fruits of the desired quality due to excess fruit retention (Akgül 2004). The fruits become bigger, homogeneous and the shell color develops fully in the production of apples with the help of dilution. Branch breakage is prevented and the tendency of the periodical is reduced as a result of providing a balanced product distribution in the tree. In addition, struggling against diseases and pests is getting easier, and the costs of harvesting and post-harvest classification are decreasing (Butar *et al*. 2016; Kaçal 2009; Barritt 2000).

Manual thinning of fruits is a simple technique known since ancient times and used by manufacturers. Today, chemical thinning is indispensable for producers in order to save costs and labor in commercial apple cultivation. These chemicals used in thinning are used as substances produced by plant cells or synthetically produced substances. Plant growth regulators are substances that stimulate, inhibit, or otherwise change physiological functions in organisms (Morsünbül 2010; Güllüoğlu and Arioğlu 2005). Many plant growth regulators are used to improve the quality and yield in the production of agricultural products. Plant growth regulators are examined in five main groups according to their structures: auxins, gibberellins, cytokinins, dormins (abscisic acid), and ethylene (Yerlikaya 2008). In our country, which is an important apple producer, not much work has been done on fruit thinning and methods. Therefore, our article titled "Investigation of fruit thinning methods used in apple species" is important from this point of view.

Apple Flower Structure

Fruit quality in apple species arises as a result of the mutual relationship of grower practices such as pruning, other cultivating processes, and product load management with environmental factors other than plant genetic structure. In addition to product quality, it is important to be able to produce regularly every year in order to recover costs and maintain competitive power (Bound 2005). The apple plant forms flowers on short fruit branches (bouquets). There are about 5-6 flowers in each beam on the bouquet. The flower is located in the middle of this 5-point flower structure and the earliest blooming flower is called the “king flower”, and the others are called the “side flower”. In some cases, all flowers are pollinated, then fertilized, and fruit retention increases Depending on climatic conditions, pollination success, and the age of the tree. 8-16% of blooming flowers should be fruit for fruit set with high quality and market value. Fruit weight and fruit size are negatively affected in trees with a large amount of product, therefore fruit quality decreases (Mcafee 2006; Salvador *et al*. 2006; Çağlar and Balcı 2003).

Manual Thinning

Although manual thinning is the most reliable method, it is not economical in commercial production due to high labor costs (Janoudi and Flore 2005). In weather conditions where the risk of late spring frost is minimized, it is recommended to do manual fruit thinning after the product load is clearly determined. The distribution of fruits on the tree will also be monitored in this period and thinning will be performed accordingly, which will provide significant advantages to the producer. Manual thinning is a preferred method due to the fact that it is not a harmful application to the environment and the fruits are removed from the tree in a controlled manner. The most important disadvantage of manual thinning is that it increases labor costs (Demircan *et al*. 2005; Webster 2002). Some growers expect the thinning application to yield small fruits the size of walnuts. However, since the thinning process is so delayed, it will not be possible to reach the expected target as long as the fruits remain on the tree since they use the tree's food sources. In other words, thinning made at an early stage contributes more to the growth of fruits. Apple variety characteristics should be taken into account when making manual thinning. Varieties with strong tree growth could leave two fruits per beam, whereas varieties with weak growth should have left one fruit (Caglar and Balcı 2003). In general, in apple

cultivation, reducing the number of fruits to 4-6 fruits in the trunk cross-section area in order to achieve good fruit quality and size, and arranging this amount to 5 fruits in the trunk cross-sectional area in the varieties showing periodicity ensures the optimum success of dilution (Robinson 2008; Robinson and Watkins 2003). The thinning process can be done manually very simply and reliably by hand in apple species Considering that manual thinning requires a lot of labor and labor wages increase production costs in today's conditions, it is not a preferred method in commercial gardens. This method is mostly applied in small gardens or as a complement to chemical thinning. Therefore, thinning with chemicals is mainly preferred in today's commercial apple cultivation (Caglar and Balcı 2003).

Chemical Thinning

Thinning with chemical substances reduces the labor requirement and provides the possibility of use in large areas. Post-flowering thinners are used commercially in apples. Chemical thinners can be used at different phenological stages to dilute buds, flowers, or small fruits. However, reducing the buds before flowering is a risky practice due to the winter cold and late spring frosts, and its practical use is not widespread (Layne 2007; Janoudi and Flore 2005; Fallahi *et al*. 2004). Hormones are organic substances that can be synthesized and transported in plants, guide the growth and development events in plants, and can be effective even at very low changes. These substances are obtained from the organs of many plant species and some fungi. Hormones produced in some cells are transported to other cells and development is controlled. These natural and synthetic substances that have been determined to affect plant growth and development are called plant growth regulators. Plant growth regulators cause various physiological reactions in the plant (Kumlay and Eryiğit 2011; Aydoğdu and Boyraz 2005). Plant growth regulators are divided into two main groups due to their stimulating and retarding properties on plants; those that initiate and accelerate plant development are called (stimulants), and those that slow down and stop growth and development are called (inhibitors). Among them are those that have a stimulating effect on the plant; auxins, gibberellins, and cytokinins. The plant growth inhibitors are abscisic acid (ABA) and ethylene (Morsünbül 2010; Karakaya and Padem 2009). Synthetic hormones, which we call plant growth regulators, are widely used in garden plants for various purposes. For this purpose, Carbaryl (Sevin), a broad-spectrum insecticide, is often used alone or mixed with chemical substances (NAA, NAAm, BA) in small fruit thinning. In recent years, it has been determined that some other chemicals, such as Hydrogen Cyanamide, Etefon, and Thidiazuron, have thinning properties. Small fruit thinning is more common than flower thinning in modern apple cultivation (Karakaya and Padem 2009). The thinners used in the flower thinning process damage the floral organs, preventing pollination or fertilization and therefore reducing the fruit attitude. Small fruit thinners are taken up by the fruit, they inhibit the growth of the fruit, causing shedding. The level of thinning in apple species is greatly influenced by various factors such as the dose of the chemical used and the spraying time, the physiological state of the tree, the cultivation system, the variety, rootstock, and cultivating processes, as well as environmental conditions (Fallahi and Wilemsen 2002).

Auxins

Auxin-type plant growth regulators are hormones that have been used in agriculture for a long time. It is the most important group that affects the growth and development of plants. It performs the growth of the plant together with other plant growth regulators. It is naturally rare in the plant root. Auxins are synthesized in meristematic tissues such as leaves, apical buds, and flowers and are transported from top to bottom. It ensures the growth of the plant. It regulates cell division, and cell and tissue differentiation. It allows the plant to grow toward the sun (Algül *et al*. 2016). Auxins are studied under 4 groups and their important growth regulators are as follows. a) Indole group b) Naphthylene group c) Phenoxy group d) Benzole group (Baktır 2010). It stops growing when too much is secreted by the plant or when administered in high doses in artificial forms. When it reaches low levels in the plant, it causes the leaves to fall out. It is effective in fruit set and formation. Prevents fertilized flowers from falling. It ensures the development of the ovary and the formation of seedless fruit. Regulates the development of cambium in spring. Artificially obtained auxins are usually used in weed control. Some of these chemicals are known as 4-CPA (4-chlorophenoxyacetic acid), 2,4-D (2,4-dichloro phenoxy acetic acid), IAA (indol acetic acid), IBA (indolbutyric acid), and NAA (1- Naphthaleneacetic acid) (Karakuş and Koker 2007; Halloran and Kasım 2002).

DRAFT

NAA (1-Naphthaleneacetic acid) and NAAm (Napthalen Acid Amide):

NAA and NAAm, which are auxin-type thinners are widely used effectively in apples. These hormones are absorbed from the leaves and form the separation layer by promoting the formation of ethylene so that fruit falls are performed (Gadner 2003). The chemical structure of 1-Naphthaleneacetic acid (NAA) is $C_{12}H_{10}O_2$. Today, various formulations are available on the market in the form of ammonium, potassium, sodium salts, or ethyl esters. 1-Naphthaleneacetic acid, a plant growth regulator, is available on the market as a solid, white, odorless, tasteless substance with a molecular weight of 186.2 g/mol. May show chronic damage to the lungs when inhaled. It causes serious hazards in case of skin contact, inhalation, and ingestion. NAA promotes elongation and root formation in plants. However, its use in high concentrations has a growth-inhibiting effect. This chemical is in the toxicity level 3 class According to the data of the World Health Organization (WHO) and the Environmental Protection Organization (EPA) (Çetinkaya and Baydan 2006). 1-Naphthaleneacetic acid (NAA), especially for dilution in apples, pears, and olives, gives successful results when applied during the period when 70-80% of flowers are blooming. It is recommended to carry out the applications after the danger of late spring frost has passed, in order to guarantee the products. It is usually used for thinning in apples at concentrations of 2.5-20 ppm. As the dose of NAA used increases, the amount of thinning also increases. However high doses of NAA both damage the leaves and cause small fruit formation (the fruit remains on the tree without growing). Therefore it is emphasized that the use of high doses of NAA should be avoided. It will be better and faster for NAA prepared quite homogeneously in water to be taken up by the leaves. Emissive and adhesive substances should be added to the solution to increase the absorption effect from the leaf surface. Spraying is in the warm and windless morning hours when hormone uptake is highest and slow drying conditions are provided. NAA is not effectively applied in conditions where the air temperature is below 10oC. It is recommended to apply NAA 7-14 days before harvest to prevent pre-harvest fall in apples and pears. NAA applied to the plant shows its effect 2-3 days after application and its effect continues for 2 weeks (Baktır 2010; Halloran and Kasım 2002).

Ethylene

Ethylene, a simple compound produced by plants (C_2H_4), has been known for many years as a highly effective plant growth regulator in gaseous form (Westwood 1993). Ethylene synthesis may vary depending on environmental factors (Seçer 1989). Ethylene can be produced in all plant tissues. Ethylene is mostly synthesized from mature and aging tissues under stress. Leaves and flowers synthesize the highest amount of ethylene in the period before wilting and falling. The least amount of ethylene is synthesized by the roots of the plant. It is gaseous, volatile, and partially inactive under normal conditions. It is a hormone that can be produced by the plant at every stage of plant growth and development. Also known as maturation hormone (gas). Ripening, quality losses, aging, and deterioration of products occur faster due to ethylene secretion in fruits and vegetables in warehouses with insufficient ventilation (Baktır 2010; Kumlay and Eryiğit 2011). The effect of ethylene in these changes affects metabolic events such as the degradation of the chlorophyll structure and the synthesis of other pigments, the breakdown of the cell wall and the softening of the fruit, the synthesis of volatile compounds that make up the fruit smell, an increase in respiratory rate, and the conversion of starch into sugar (Öktüren and Sönmez 2005). In addition, the different effects of ethylene on plants are as follows; breaking dormancy, falling leaves and fruits, promoting flowering in some plants, stimulating adventitious root formation, encouraging female flower formation in monoic plants, and facilitating mechanical harvesting by encouraging abscission (Algül *et al.* 2016).

Cytokinins

Unlike other hormones that occur especially during cell division in plant tissues, quinine is an organic substance found in both plants and animals. The first plant-based cytokinin was zeatin, which was isolated from corn seeds. Zeatin, dihydroseatin, isopentenyl adenine (2IP), and dimethylallyl adenine are naturally synthesized cytokinins, kinetin (N6 furfuryl amino purine), benzyl adenine (BA) and tetrahydropyranylbenzyl adenine (PBA) are synthetic cytokinins (Üstün *et al.* 2010). The physiological roles of cytokinins in plant tissues; cell division and organ formation are their effects on cell growth. All tissues with active cell division contain a high amount of cytokinins. Cytokinins are effective in cell division and have a function in tissue and organ differentiation. They also play an active role in such formations as chlorophyll breakdown and aging

retardation, flower stimulation, frost resistance, seed germination, parthenocarpic fruit formation, gender differentiation, fruit attitude and fruit growth, and side bud development. Protein and RNA synthesis and enzyme activity are other main activities in which cytokinins are effective. Cytokinins are usually found in young tissues. It is synthesized especially in the root meristems and then transported to the green parts of the plant through the xylem. Auxins support root formation, while cytokinins support shoot formation. They contribute to the formation and development of organs in tissue-cultivating environments (Dodd 2003; Karakaya and Padem 2009; Baktır 2010; Kumlay and Eryiğit 2011).

Giberlin

Giberlic acid, which is one of the growth and development-promoting hormones, was first found in 1926 by Japanese researchers in the fungus Gibberella fujikuroi, which causes over-sizing in rice plants and takes its name from here. Later, this substance was isolated and named giberellic acid (GA) (Kılıç 2007; Morsünbül 2010). The most commonly used type among gibberellins is GA. It is known that there are at least 126 different species of gibberellin today. Gibberellin is found in large quantities in the buds, embryos, roots, young leaves, flowers, fruits, and cambium of plants. Only the GA type is commercially available and important Among the gibberellins (Baktır 2010). The most obvious effect of gibberellins is to increase the elongation of cells. Moreover, they are very effective in breaking seed and bud dormancy, eliminating stunting, meeting the need for cooling, parthenocarpic fruit set, and promoting germination. GA is mainly used in table and dried grapes to thin the bunches and increase the grain size (Tyler *et al*. 2004; Olszewski *et al*. 2002).

CONCLUSION

The thinning process has been recognized as one of the cultivating processes that should be paid attention to in many fruit types, especially apples, in recent years. When thinning is done correctly and on time, it causes an increase in fruit quality, especially in fruit size, color, taste, and many other parameters. In addition, thinning eliminates periodicity in fruit trees and ensures that a regular crop is obtained every year. Although manual thinning provides high success in apple species, this process is not preferred in large orchards and commercial production enterprises because it requires labor costs and a very long time. Chemical thinners that are more cost-effective and can be applied evenly in a short time are preferred in such large and high-production enterprises. Although chemical thinning is applied more easily and quickly than mechanical dilution, considerations such as the dose and time of administration of the drug, the age of the trees to be applied, and the suitability of the thinner for the flowering period or small fruit period should be considered. Any negligence or fault in chemical dilution can cause irreversible damage.

REFERENCE

Anonymous, 2022. Agricultural statistics, production. FAO, http://faostat.fao.org. Erisim Tarihi: 06.06.2022.

Akgül H, 2004. Ziraat Mühendisliği Dergisi 342: 42-44.

Algül BE, Tekintaş FE, Günver Dalkılıç G, 2016. Bitki büyüme düzenleyicilerinin kullanımı ve içsel ormonların biyosentezini arttırıcı uygulamalar. Journal of Adnan Menderes University Agricultural Faculty 13(2): 87-95.

Aydoğdu M and Boyraz N, 2005. Bitki Büyüme Düzenleyicileri (Hormon) ve Hastalıklara Dayanıklılık, Bitkisel Araştırma Dergisi 1: 35–40.

Baktır İ, 2010. Bitki Büyüme Düzenleyicileri Özellikleri ve Tarımda Kullanımları. Hasad Yayıncılık.

Barritt BH, 2000. Apple Quality for Consumers. The Compact Fruit Tree 34(2): 54-56.

Bound SA Tu, 2005. The impact of selected orchard management practices on apple. University of Tasmania, Ph.D. Thesis, 190p, Australia.

Butar S, Çetinbaş M, Atasay A, İşçi M, Koçal H. 2016. Elmada Meyve Seyreltmesi, Meyve Tutumu ve Meyve Kalitesi Üzerine Bitki Büyüme Düzenleyicisi Madde Uygulamalarının Etkisi. Journal of Agricultural Faculty of Gaziosmanpasa University 33(2): 9-18.

Çağlar S and Balcı S, 2003. Elma Yetiştiriciliğinde Uygulanan Meyve Seyreltme Yöntemleri Üzerine Bir İnceleme. KSÜ Fen ve Mühendislik Dergisi. 6(1): 117-128.

Çetinkaya MA and Baydan E, 2006. Bitki Gelişim Düzenleyicilerin Zehirliliğine Genel Bir Bakış. Veteriner Hekimler Derneği Dergisi 77(4): 26-31.

Demircan V, Yilmaz H, Binici T, 2005. Isparta ilinde elma üretim maliyeti ve gelirinin belirlenmesi. Tarım Ekonomisi Dergisi 11(2): 71-80.

Dodd IC, 2003. Hormonal interactions and stomatal responses. J. Plant Growth Regulation 22: 32-46.

Dousti S and Dumanoglu H, 2011. Braeburn, Fuji, Gala, Granny Smith, Jonagold ve Top Red Elma Çeşitlerinde M9 Anacı Üzerindeki Genç Ağaçların Verim ve Bazı Meyve Özelliklerinin Yaz Ayları Düşük Nemli Karasal İklim Koşullarında İncelenmesi. TÜRKİYE VI. ULUSAL BAHÇE BİTKİLERİ KONGRESİ. Şanlıurfa, 765-772.

Fallahi E and Willemsen KM, 2002. Blossom thinning of pome and stone fruit. HortScience 37(3): 474-477.

Fallahi E, Chun IJ, Mousavi Fallahi B, 2004. Influence of new blossom thinners on fruit set and fruit quality of apples. Proc. 9This on Plant Bioregulators. Acta Horticulturae 653:81-85.

Gadner J, Thininig and growth regulators notes for apples. www.gov.on.ca .

Gulluoglu L and Arioglu HH, 2005. Harran ovası koşullarında bazı bitki büyüme düzenleyici uygulamalarının ikinci ürün soyada (Glycine Max Merrill.) önemli tarımsal özellikler üzerine etkilerinin belirlenmesi, Harran Üni. Ziraat Fak. Dergisi 9(2): 37-43.

Halloran N and Kasım MU, 2002. Meyve ve Sebzelerde Büyüme Düzenleyici Madde Kullanımı ve Kalıntı Düzeyleri. Ankara Üniversitesi Ziraat Fakültesi.Gıda 27(5): 351-359.

Janoudi A and Flore JA, 2005. Application of ammonium thiosulfate for blossom thinning in apples. Scientia Horticulturae 104: 161-168.

Kaçal E, 2009. Elmalarda (Malus x domestica Borkh) meyve tutumu, meyve kalitesi ve çiçek tomurcuğu farklılaşması üzerine yeni çiçek seyrelticilerinin etkileri. SDU Fen Bilimleri Enstitüsü, Doktora Tezi, Isparta.

Karakaya D and Padem H, 2009. Sebzelerde Eşey Hücrelerinin Oluşumunda Büyümeyi Düzenleyici Maddelerin Etkileri, Süleyman Demirel Üniversitesi Ziraat Fakültesi Bahçe Bitkileri Bölümü, Isparta, Alaratarım 8(1): 43-50.

Karakuş A and Kalyoncu İH, 2010. Bazı Elma Çeşitlerinde Kimyasal ve Elle Seyreltme Uygulamalarının Meyve Kalitesi Üzerine Etkileri. Atatürk Üniversitesi Ziraat Fakültesi Dergisi, 41(2): 81-89.

Karakuş C and Köker R, 2007. "Tarımda Bitki Gelişim Düzenleyicilerin (BGD) Kullanımı ve Hormon Riski", Üniversite Öğrencileri 2. Çevre Sorunları Kongresi Kongre Kitabı 163-175.

Kaşka N, 2003. Türkiye'de Ilıman İklim Meyvelerinin Dünü, Bugünü ve Yarını. Türkiye IV. Ulusal Bahçe Bitkileri Kongresi 2003, 1-5, Antalya

Kilic Y, 2007. Fitohormonların Saplı Meşe (Quercus Robur L.) 1+0 Yaşlı Fidan Morfolojik Karakterleri Üzerine Etkisi. Gazi Üniversitesi Fen Bilimleri Enstitüsü. Yüksek Lisans Tezi, 74s, Ankara.

Kumlay MA and Eryiğit T, 2011. Bitkilerde Büyümeyi ve Gelişmeyi Düzenleyici Maddeler: Bitki Hormonları Iğdır Üni. Fen Bilimleri Enst. Dergisi 1(2): 47-56.

Layne DR, 2007. Layne DR, 2007. American/Western Fruit Grower, April.

Luby JJ, 2003. Taxonomic classification and brief history, In: Apples, Botany, Production and Uses. Ferree, D.C. and Warrington, I.J. (eds), CABI Publishing 1-14, Cambridge, USA.

Mcafee JD, 2006. Potential models for screening organic fruit thinners of apple. M.Sc Thesis, 104p, University of Arkansas, USA.

Morsunbul T, 2010. Bazı İleri Oksidasyon Teknikleri ile Bitki Gelişim Düzenleyici (BGD) Gideriminin Araştırılıması, (Yüksek Lisans Tezi). Uludağ Üniversitesi Çevre Mühendisliği Anabilim Dalı. Bursa.

Olszewski N, Sun T, Gubler F, 2002. Gibberellin signaling: biosynthesis, catabolism, and response pathways. Plant Cell 14: 61-80.

Öktüren F and Sönmez S, 2005. Bitki Besin Maddeleri ve Bazı Bitki Büyüme Düzenleyicileri (Hormonlar) Arasındaki İlişkiler. Derim. Batı Akdeniz tarımsal Araştırma Enstitüsü 22(2): 20-32.

Özçağıran R, Ünal A, Özeker E, Isfendiyaroğlu M, 2011. Ilıman İklim Meyve Türleri, Yumuşak Çekirdekli Meyveler. Cilt II, Ege Üniversitesi Ziraat Fakültesi Yayınları No: 556, İzmir.

Robinson TL, 2008. Crop load management of new high-density apple orchards New York Fruit Quarterly, 16(2).

Robinson, TL and Watkins CB, 2003. Crop load of Honeycrisp affects not only fruit size but many quality attributes. New York Fruit Quarterly, 11(3).

Salvador FR, Fisichella M, Fontanari M, 2006. Correlations between fruit size and fruit quality in apple trees with high and standard crop load levels. Journal of Fruit and Ornamental Plant Research. 14: 113-122.

Seçer M, 1989. Doğal Büyüme Düzenleyicilerin (Bitkisel Hormonların) Bitkilerdeki Fizyolojik Etkileri ve Bu Alanda Yapılan Araştırmalar. Derim 6(3): 109-124.

Tyler L, Thomas SG, Hu J, Dill A, Alonso JM, Ecker JR, Sun T, 2004. Della Proteins and Gibberellin-Regulated Seed Germination and Floral Development in Arabidopsis. Plant Physiol. 135: 1008–1019.

Üstün GE, Akal Solmaz SK, Morsünbül T, Azak HS, 2010. Advanced Oxidation and Mineralization of 3-Indolebutyric Acid (IBA) by Fenton and Fenton-like Processes Journal of Hazardous Materials 180(1-3): 508-513.

Webster T, 2002. Current approved thinking strategies for apples and pears and recent thinking research trials in Europe. The Compact Fruit Tree, 35(3).

Westwood MN, 1993. Hormones and growth regulators,Temp zone Pomology: Pysiology and Culture Timber Press Inc. 9999 Pp.W. Wilshire, Suite 124, Portland, Oregon.

Yerlikaya E, 2008. Endokrin Bozucular, Gazi Üniversitesi, Tıp Fakültesi. Ankara.

DRAFT

ORAL PRESENTATION

Investigation of the genetic defect of Cholesterol deficiency and Brachyspina syndrome in Holstein breed cattle breed in Eskisehir

Beste Goneci[1] (ORCID:0000-0002-7183-0170) and Muhammet Kaya[1*](ORCID:0000-0001-6474-121X)

[1] Department of Agricultural Biotechnology, Faculty of Agriculture, Eskisehir Osmangazi University, 26480, Eskisehir, Turkey

[1*] Department of Agricultural Biotechnology, Faculty of Agriculture, Eskisehir Osmangazi University, 26480, Eskisehir, Turkey

[*] muhammetkaya@ogu.edu.tr

Abstract

Hereditary diseases, which are likely to come from previous generations, can be transferred to the newly formed population and cause these diseases to spread from generation to generation. In this study, Holstein cattle population determined in Eskisehir was studied, DNA was isolated from these cattle and then examined in terms of Brachyspina syndrome and cholesterol deficiency diseases. Regarding these two hereditary diseases detected in the Holsteins in Turkey in previous studies, it was determined that 2 cattle for the BS genetic defect and 11 cattle for the CD genetic defect were heterozygous genotype, that is, carriers, in the population we determined in Eskisehir, in our study.

With regard to these two hereditary defects, in the study conducted to investigate samples were taken from the Holstein Friesian cattle reared in Eskisehir by using PCR teqnique. 2 and 11 cattle were found to be carriers of the BS and CD, respectively, among 112 Holstein Friesian cattle. The possibility of the spread of genetic defects and economic damage can be prevented using molecular techniques. The herds that are free of genetic defects be created by using some molecular methods detecting hereditary diseases, and by planning creation of new generations.

Keywords: ABOP genes, Brachyspina Syndrome, Cholesterol Deficiency, FANCI genes, Holstein

1. Introduction

Hereditary diseases in cattle can cause significant economic damage. These diseases are very difficult to detect. Today, with the development of technology, there are new hereditary diseases. One of them is "Cholesterol Deficiency (CD)", which is recessive and hereditary and was discovered in 2015, and the other is "Brachyspina Syndrome (BS)", which was first discovered in Denmark in 2006 (Özcan and Özbeyaz, 2017).

For autosomal recessive diseases to occur, functional changes must occur in both pairs of genes at the same location. This type of hereditary disease does not show up in every generation. Most known hereditary diseases in cattle are caused by autosomal recessive alleles. However, autosomal recessive alleles spread within the population because they are not seen in the external appearance and therefore cannot be identified if the animal shows heterozygous inheritance (Agerholm, 2007). According to OMIA, the total number of diseases found in cattle is 630 (OMIA, 2023). 73 of these diseases are found in the Holstein breed (OMIA, 2023).

Brachyspina syndrome first appeared in 2006 (Agerholm et al., 2006). To date, it has been found in the Netherlands (Agerholm and Peperkamp, 2007), Italy (Testoni et al., 2008), Germany (Buck et al., 2010) and Canada (Agerholm et al., 2010). Brachyspina syndrome is a rare hereditary defect in Holstein cattle. The mutation that causes Brachyspina Syndrome is located on the FANCI gene (OMIA, 2023). The disease is caused by a 3329 base pair deletion in the Fanconi anaemia complementation group 1 (FANCI) gene, which is located on the 21st chromosome of the bovine chromosomes (Charlier et al., 2012). While the FANCI gene normally contains 37 exons, the mutation results in a frameshift deletion from exon 25 to 27, creating a stop codon in exon 28 (Li et al., 2016). After the first two cases, two calves were stillborn in Italy and the cause was brachyspina syndrome. As in the first case, these two stillborn calves also had long legs and short bodies.

Various abnormalities were also observed in the autopsy findings examined radiographically (Testoni et al., 2008). Apart from these cases, dead Holstein Friesian calves with identical phenotype and morphological signs were reported in the Netherlands (Agerholm and Peperkamp, 2007), Germany (Buck et al., 2010) and Canada (Agerholm et al., 2010). In Poland, when 78 Holstein peregrine cattle were examined by Ruść and Kamiński (2015), 8 out of 78 animals were found to be carriers. In China, when 342 peregrine falcons born between 1996 and 2012 were screened by Li et al. (2016), 13 of them were found to be carriers. Chao et al. (2020) randomly identified 1688 cattle from herds in northern, central, southern and eastern Taiwan. In the first study conducted in Turkey, 250 Holstein Friesian cattle were tested for BS in 2021 and 1 cattle was found to be a carrier. These results of the study showed that this hereditary disease is also present in Holsteins reared in Turkey, even at low frequencies (Dibic, 2021).

The hereditary defect of cholesterol deficiency was first encountered in a bull named Mauglin Storm, born in 1991 (Menzi et al., 2016). As bulls descended from Mauglin Storm are used in artificial insemination, the frequency of the mutant allele causing the hereditary defect of cholesterol deficiency has increased in many countries. Calves with homozygous inheritance of the defect showed signs of hypolipidaemia and hypocholesterolaemia. It has been named cholesterol deficiency, which is also expressed as an autosomal recessive genetic defect and genetic lipid metabolism disorder in calves, resulting in death (Kipp et al., 2015). Cholesterol deficiency results in a 1.3 kb insertion in the 5th exon of the apolipoprotein B (APOB) gene on the 11th chromosome of BTA that affects lipid metabolism and results in early death (Schutz et al., 2016). APOB is the structural chylomicron protein of low (LDL) and very low (VLDL) density lipoproteins (Kane et al., 1980). The resulting 1.3 kb insertion creates a stop codon in the open reading frame (ORF) of APOB. As a result, the protein region is much shorter than its 140 amino acid length. In relation to this genetic disease, animals with heterozygous inheritance often have no symptoms, while this hereditary defect is encountered in some way at an advanced age.In animals with homozygous genotype in relation to cholesterol deficiency; It has been observed that excretion of chylomicron in the intestine cannot be made at an early age, but it has been shown that there is a problem in the cholesterol absorption stream (Schütz et al., 2016). Looking at the studies on CD, 27 Holstein Friesian cattle in Poland were selected by studying the Mauglin Storm pedigree, and 9 cattle were found to be carriers of the hereditary cholesterol deficiency disease. These results suggest that the mutation causing the genetic disease CD is also transmitted to Holstein cattle in Poland (Kamiński and Ruść 2016). In Switzerland, a study was conducted on the effect of the ABOP genotype on cholesterol metabolism in 254 Holstein cattle. As a result of the study, no homozygous alleles were found in 254 Holstein cattle, 36 Holstein cattle were carriers and 218 Holstein cattle had normal alleles. Of 1817 randomly selected Russian cattle born in Russia between 2010 and 2017, 147 were found to carry the defect. In order to carry out a study on this disease in domestic cattle and Holstein breeds bred in Turkey in 2021, 250 migratory animals were examined. As a result of the study, 4 cattle out of 250 were found to be carriers. These results of the study showed that this hereditary disease is also present in Holstein-Friesian cattle bred in Turkey, although at a low frequency (Dibic, 2021).

The genetic defects of Brachyspina Syndrome and Cholesterol Deficiency, which have been identified in cattle populations in different countries, were investigated using samples from the Holstein cattle population reared in Eskişehir. With this research, these two genetic defects in the Holstein breed, which is often preferred in dairy cattle breeding in Eskişehir, were investigated using the PCR method and it was aimed to benefit from healthy herds.

Material and Methods

This study was carried out at Eskişehir Osmangazi University, Faculty of Agriculture, Department of Agricultural Biotechnology, Molecular Genetics Laboratory. This research includes the steps of isolating the genomic DNA molecule from the blood samples, amplifying the regions related to hereditary defects by PCR, and interpreting the base pairs formed by the PCR products in terms of hereditary defects.

Material

DNA was purified from blood samples collected from 112 animals from 4 different Holstein farms in Eskişehir. Genomic DNA was isolated from the blood samples using the Thermofischer DNA isolation® kit and stored at 4°C.

Methods

In the study, blood samples from 112 animals were stored in 5ml EDTA tubes at -20°C until DNA isolation. DNA isolation was from blood using commecial kits (PureLink DNA isolation® genomic DNA isolation kit). After DNA isolation, DNA samples checked in agarose gel.

The genotypes for BS and CD were identified by using PCR method. The PCR reaction mixture (20 µl) contained 50–100 ng DNA template, 10X Taq polymerase buffer, 1.5 mM MgCl2, 2.5 mM dNTPs, 0.5 U Taq DNA polymerase and 5 pmoles of each primer (Table) per reaction. The PCR cycle profile was 94°C for 3 minutes; then 35 cycles of 94°C for 15 sec., 58°C (BS) and 65C (CD) for 20 sec., and 72°C for 30 sec; followed with 10 minutes at 72°C. PCR products were run by electrophoresis using 2% agarose gels. Primers for BS, ATP8 and CD related gene regions are shown in Table.

Table. Gene regions related to BS, ATP8 and CD

	Primer sequence	Fragment size	Literature
BS	F:5' GCTCAAGTAGTTAGTTGCTCCACTG3'	409 bp	Li, Y., et al., 2016
	R: 5' ATAAATAAATAAAGCAGGATGCTGAAA3'		
ATP8	F-W: 5' TAAGTTAGAGATTGAGAGCC3'	269 bp	
	R-W: 5' GATAAGGGTTACGAGAGGGA3'		
CD	Forward-W: 5'GGTGACCATCCTCTCTCTCTGC3'	436 bp	Menzi, F., et al., 2016
	Reverse: 5'AGTGGAACCCAGCTCCATTA3'		
	Forward-M: 5'CACCTTCCGCTATTCGAGAG3'	249 bp	

Results

DNA samples obtained from 112 Holstein cattle in the study population were run on a 1% agarose gel. The image of the DNA samples after electrophoresis is shown in Figure 1.

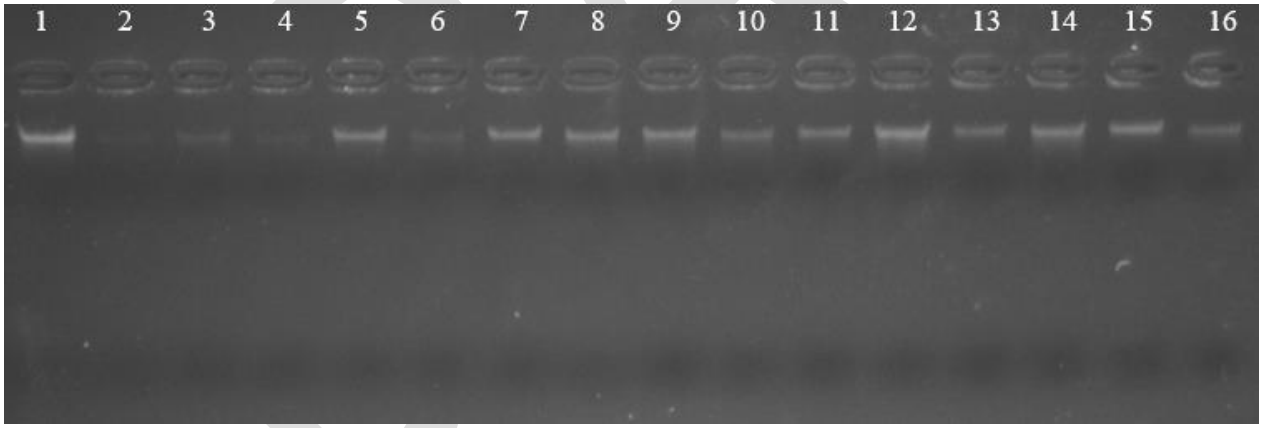


Figure 1. Image of some DNA samples to be used after electrophoresing using 1% agarose gel electrophoresis

PCR for Brachyspina Syndrome were visualised on a 2% agarose gel. As a result of the band pattern, 2 heterozygous individuals for the BS were detected among the cattle studied. According to Li et al. (2016), The ATP8 gene is used for PCR accuracy due to the positive control. Therefore, 269 base pairs were observed in the PCR results of all healthy and affected cattle in BS genetic defect.

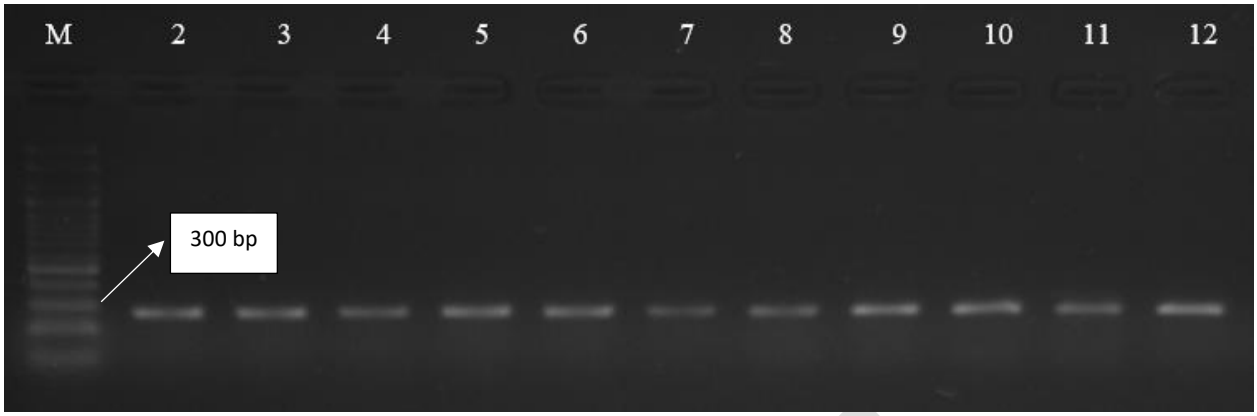


Figure 2. Detection of BS genotypes by 2% agarose gel electrophoresis of BS genotype expressions. 2. well and others; normal genotype (269 bp); M:100 bp ladder, Fermantas® GeneRuler SMO241

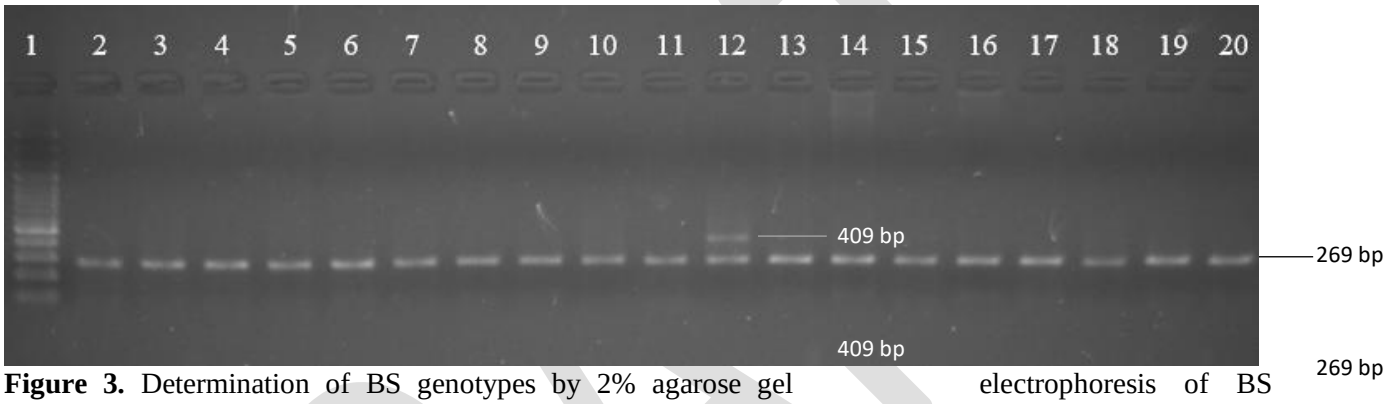


Figure 3. Determination of BS genotypes by 2% agarose gel electrophoresis of BS genotype expressions. 12. well ; Heterozygous genotype (269 and 409 bp) found in Holstein cattle; M:100 bp ladder, Fermantas® GeneRuler SMO241

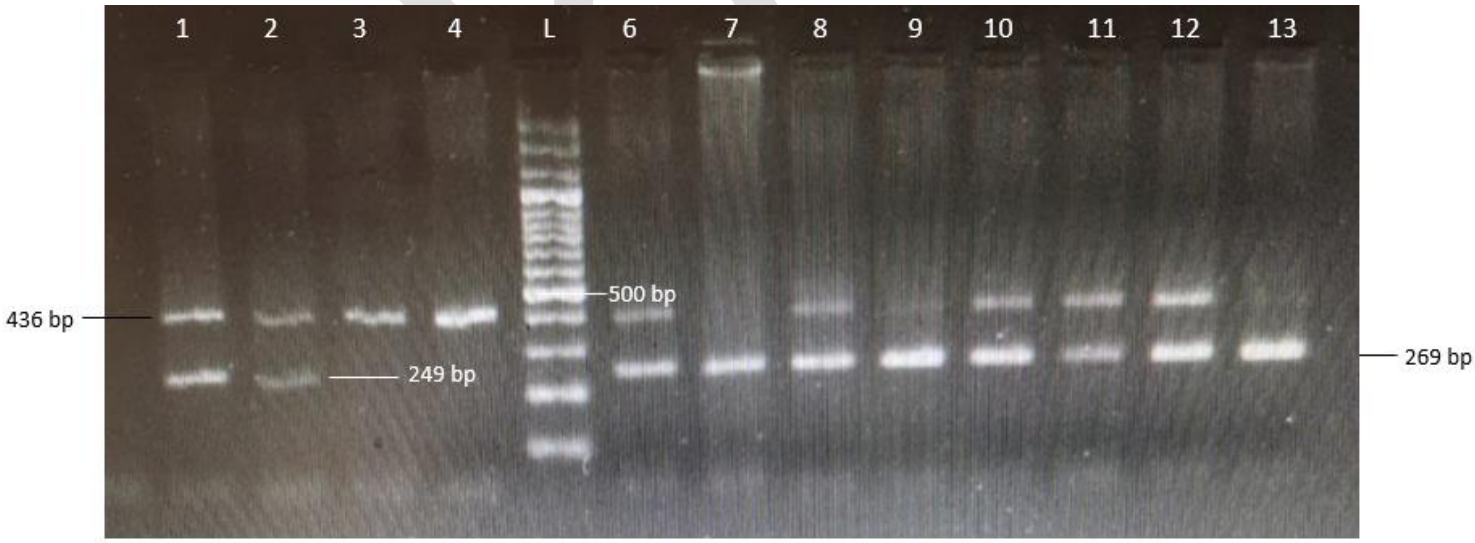


Figure 4. Detection of CD genotypes by 2% agarose gel electrophoresis method. wells 7 and 13 normal genotype (269 bp); wells 1 and 2 are heterozygous genotypes (249 and 436 bp) found in Holsteins; L:100 bp ladder, Fermantas® GeneRuler SMO241

In CD carrier and animals, 409 base pairs were also observed due to the mutation. Menzi et al, (2016) found that 249 base pairs were observed in healthy cattle in relation to the CD genetic disease. As a result of the visualisation process, 11 heterozygous individuals were found among the cattle studied in terms of cholesterol deficiency.

Following the PCR studies conducted in Eskişehir, it was found that two out of 112 Holstein Friesian cattle were affected by Brachyspina syndrome. Additionally, the deficiency allele for cholesterol was present in eleven of them. The mutant allele frequency of CD and BS was estimated 0.0491, and 0.0089 respectively.

Conclusions

Predicting genetic disorders in animal husbandry and ensuring population health through controlled breeding is a vital concern. It is crucial to detect heterozygous animals to manage genetic diseases in animal populations and mitigate risks by conducting a pre-breeding examination of the cattle intended for breeding. This approach will preserve the health of the future population and prevent any adverse impact on yield.

After the existence of the identified cases in Turkey (Dibic, 2021), the identified population in Eskişehir was also examined in a special framework with regard to BS and CD hereditary diseases and the existence of the diseases was determined.

By knowing the frequencies of these two hereditary diseases which directly affect the yield in the population, the cattle to be used as breeding stock in the future population can be known, which will prevent the direct decrease in yield by providing disease control.

As a result of this study, it was found that 2 cattle for BS genetic defect and 11 cattle for CD genetic defect were heterozygous genotype in Holstein cattle population in Eskişehir. Breeding selection in the formation of future populations by paying attention to these data will reduce the frequency of these hereditary diseases.

References

Agerholm, J.S., 2007, Inherited disorders in Danish cattle, APMIS- Acta Pathologica, Microbiologica, et Immunologica Scandinavica, 115, p.1-76.

Agerholm, J.S., McEvoy, F., Arnbjerg, J., 2006, Brachyspina syndrome in a Holstein calf, Journal of Veterinary Diagnostic Investigation, 18, p.418-422.

Agerholm, J.S., Peperkamp, K., 2007, Familial occurrence of Danish and Dutch cases of the bovine brachyspina syndrome, BMC Veterinary Research, 3, p.8.

Agerholm, J.S., DeLay, J., Hicks, B., Fredholm, M., 2010, First confirmed case of the bovine brachyspina syndrome in Canada, The Canadian Veterinary Journal, 51, p.1349-1350.

Buck, B.C., Ulrich, R., Wöhlke, A., Kuiper, H., Baumgärtner, W., 2010, Vertebral and multiple organ malformations in a black and white German Holstein calf, Berliner Und Münchener Tierärztliche Wochenschrift, 123, p.251-255.

Charlier, C., Agerholm, J.S.,Coppieters, W., et al., 2012, A deletion in bovine FANCI gene compromises fertility by causing fetal death and brachyspine, Public Library of Science ONE, 7(8).

Chao, C.H., Chen, Y.M., Lee, K.H., 2020, Genotype screening of bovine brachyspina in Taiwan Holstein cows, American Journal of Animal and Veterinary Sciences, 15(3), p.206-210.

Kaminski, S., Rusc, A., 2016, Cholesterol deficiency-new genetic defect transtmitted to Polish Holstein-Fresian cattle, Polish Journal of Veterinary Sciences, 19(4), p.885-887.

Kipp, S., et al., 2015, A new Holstein haplotype affecting calf survival, Interbull Bulletin, p.49.

Li, Y., et al., 2016, A novel multiplex polymerase chain reaction method for the identification of brachyspina syndrome carriers in Chinese Holstein cattle, Journal of Veterinary Science & Medical Diagnosis, 5(3).

Özcan, M., Özbeyaz, C., 2017, Holştayn ırkı sığırlarda otozomal resesif bir hastalık: kolesterol eksikliği, Lalahan Hayvancılık Araştırma Enstitüsü Dergisi, 57(1), s.61-67.

Menzi, F., et al., 2016, A transposable element insertion in ABOP causes cholesterol deficiency in Holstein cattle, Animal Genetics, 47(2), p.253-257.

OMIA, 2023, https://www.omia.org/OMIA001965/9913/ , Erişim Tarihi: 02/05/2023

Rusc, A., Kaminski, S., 2015, Detection of brachyspina carriers within Polish Holstein-Fresian bulls, Polish Journal of Veterinary Sciences, 18(2), p.453-454.

Schütz, E., et al., 2016, The Holstein Friesian lethal haplotype 5 (HH5) results from a complete deletion of TBF1M and cholesterol deficiency (CDH) from an ERV-(LTR) insertion into the coding region of APOB, Public Library of Science ONE, 11(4).

Testoni, S., Diana, A., Olzi, E., Gentile, A., 2008, Brachyspina syndrome in two Holstein calves, The Veterinary Journal, 177(1), p.144-146.

DRAFT

ORAL PRESENTATION

Indoor PM_{10} concentrations in a private gym

Özlem Özden Üzmez[1] (*ORCID: https://orcid.org/0000-0003-4310-788X)*, Gönenç Ozarlı[2*] (*ORCID: https://orcid.org/0009-0004-0689-3249*), Ozan Kaya[3] (*ORCID: https://orcid.org/0009-0007-3466-6774*)

[*1,2,3] Eskişehir Technical University, Engineering Faculty, Environmental Engineering Department, Eskişehir, Turkey

*Corresponding author e-mail: gonencozarli@gmail.com

Abstract

Nowadays, people spend most of their daily time (≈90%) indoors. Particulate matter is among the most important pollutants affecting indoor air quality. Indoor air quality is directly affected by indoor PM_{10}. Short and long-term exposure to PM_{10} can cause serious harm to human health. Most people dedicate a minimum of two days a week, with each day comprising at least one hour, to private gyms. In this study, measurements of indoor and outdoor PM_{10} concentrations were conducted at a private gym in Eskişehir. The measurements were taken during various training intensities, including light, medium, and heavy workouts, as well as during hours when the students were not engaged in training. When comparing the measurements to the indoor PM_{10} limit value set by the USEPA (American Environmental Protection Agency) at 150 µg/m^3, it was observed that PM_{10} levels exceeded this limit when the balcony door was opened to ventilate the indoor environment, both during heavy training sessions and during non-training hours. It was noted that PM_{10} levels did not exceed the limit value during medium and light training hours. On days characterized by heavy training sessions and poor outdoor air quality, indoor air quality poses a potential risk to both trainers and students. This underscores the significance of enhancing indoor air quality during these specific hours.

Keywords: Indoor Air Quality, PM_{10}, Private Gym

INTRODUCTION

In recent years, Indoor Air Quality (IAQ) has received increasing attention from the international scientific community, political institutions and environmental governments to improve the comfort, health and well-being of building occupants. Numerous studies on this topic have demonstrated both qualitative and quantitative shifts in indoor air quality (IAQ) over time, emphasizing the rise in pollutants and their concentrations. For this purpose, standards and regulations regarding IAQ, policies and monitoring plans for non-industrial buildings have been developed in various countries. It is known that people spend about 90% of their time in both private and public indoor environments such as homes, gyms, schools, workplaces, transportation vehicles. Therefore, IAQ has a significant impact on health and quality of life in general. For many individuals, the health hazards associated with indoor air pollution exposure may surpass those stemming from outdoor pollution. In particular, poor IAQ can be detrimental to vulnerable groups such as children, young adults, the elderly, or those suffering from chronic respiratory and/or cardiovascular diseases (Bralewska et al., 2022; Cincinelli and Martellini, 2017; Engel-Cox et al., 2013).

Particulate matter (PM) constitutes a primary focus in the study and control of air pollution (Dominici et al., 2014). Particulate matter includes both primary particles such as soot and dust from combustion sources and agricultural practices, and secondary particles such as sulfate and nitrate formed through chemical reactions from sulfur dioxide and nitrogen oxide in the atmosphere. The current focus is on PM_{10} and $PM_{2.5}$, which are particles less than 10 microns and 2.5 microns in diameter. Epidemiological studies of long-term exposure to ambient fine particulate air pollution have documented serious adverse health effects, including increased deaths from chronic cardiovascular and respiratory diseases, lung cancer, and adverse reproductive outcomes (Brunekreef and Holgate, 2002).

DRAFT

Indoor PM sources can be listed as smoking tobacco, burning wood and fossil fuels, burning candles and incense, cooking (especially frying and grilling), and cleaning (Zhang et al., 2021). Gyms typically do not contain most of these resources. Key sources of particulate matter (PM) within gym environments include settled dust that becomes airborne due to the activities of gym members and emissions from vehicles in nearby parking areas, serving as an external source. The first of these is specifically PM_{10} and the second is $PM_{2.5}$. In contrast to semi-urban and rural settings, urban gyms near heavy traffic, gyms with a high member density per unit area, and city gyms during winter months exhibit higher levels of indoor air pollution. Additionally, rural areas experience increased pollution during summer months, while sports halls with inadequate cleaning practices or excessive chemical use for cleaning also contribute to elevated pollution levels. Elevated levels of particulate matter have been detected in gyms, particularly in those where indoor air chemistry reactions, such as those involving ozone and terpenes, take place (Sofuoğlu, 2016).

The domains of sports and physical exercise encompass physical, psychological, and performance dimensions and are frequently conducted in indoor environments. The metabolic reaction to physical exercise places the body at a heightened risk of pollutant exposure, as the heightened respiratory ventilation rate during exercise leads to the inhalation of larger quantities of air and consequently, airborne pollutants. During exercise, individuals tend to inhale air through their mouth rather than allowing it to pass through the nasal filtering system. The amplified airflow rate carries pollutants deeper into the respiratory system, heightening the health risks to humans. This study assessed indoor air quality by quantifying particulate matter (PM_{10}) levels within a private gym located in Eskişehir, Turkey.

MATERIALS AND METHODS

Study area and measurement points

The research took place from January 4, 2023, to January 6, 2023, encompassing the training hall, locker room, and outdoor area (balcony) of a private gym, situated in the Yenibağlar neighborhood of Eskişehir, Turkey. Figure 1 displays schematics of the gym where measurements were conducted, along with the device placements. Measurements were conducted on January 4th during medium-intensity training, on January 5th for both light and heavy training sessions, and on January 6th during unstructured free training periods.

The gym has a total area of 85.215 m^2, including a locker room of 6.148 m^2 and a training area of 26.22 m^2. There is no mechanical ventilation in the studio, ventilation is provided by air conditioning and windows. The gym's sole entrance and exit door remains closed at all times.

Two DustTrak II Aerosol Monitors were used for PM_{10} measurements. One of the two devices employed was initiated in the locker room 15 minutes prior to the training session and relocated outdoors after 30 minutes, with measurements ongoing until 15 minutes post-training. The device positioned in the hall remained stationary throughout the study.

DRAFT

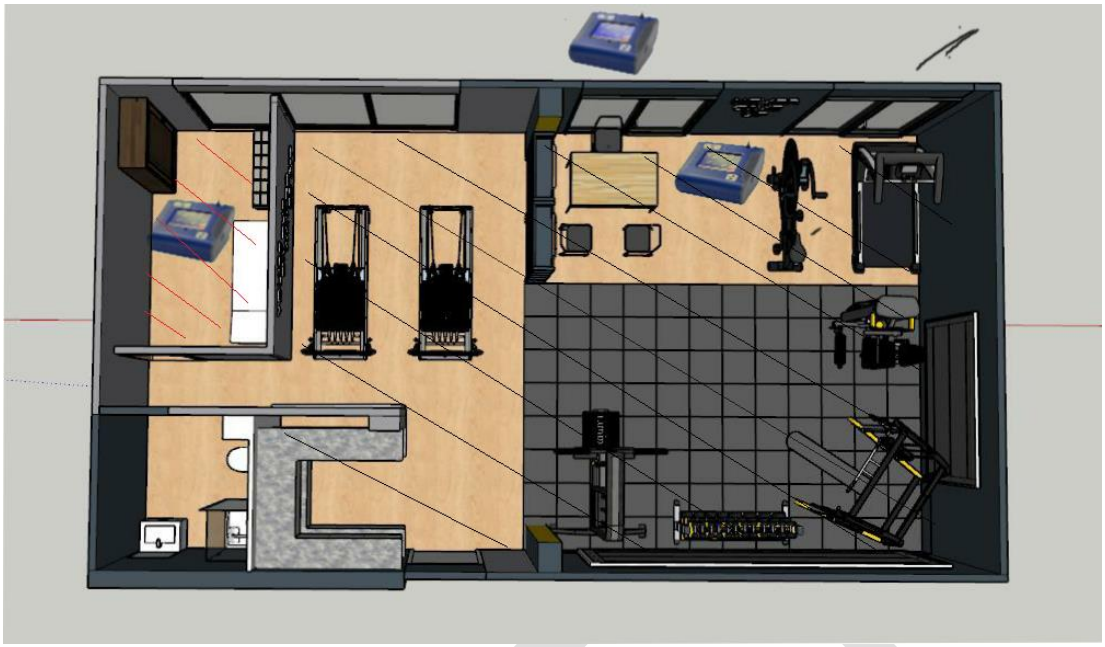


Figure 1. Drawing of the gym and locations of the devices

Study Program

Concurrent measurements were conducted in both indoor and outdoor environments, and the performance of the two devices employed was assessed through parallel laboratory testing. The comparison yielded an R^2 value of 0.97. This process affirmed the compatibility of the devices and validated their suitability for field measurements.

Measurements were conducted corresponding to the exercise intensity levels, encompassing inactive periods as well as light and heavy workouts, which took place in the locker room, training room, and outdoor areas. Typically, training sessions in the gym commence at the top of the hour and last for approximately 45 minutes. Upon arrival, students enter through the sole entrance and exit located in the hall and proceed to the locker room for changing into their workout attire. The duration of their stay in the locker room varies individually but averages around 5 minutes.

The initial measurement occurred during the 16:00-16:45 session, featuring intermediate-level training, with a focus solely on indoor environmental levels. The second measurement was conducted during the 08:00-08:45 session, with two individuals engaged in heavy training. The third measurement was carried out during the 13:00-13:45 session, with a single individual participating in light training. The fourth measurement occurred in the studio during the non-training session from 11:00 to 11:45. At that time, there were a total of three individuals present in the studio, and physical activity was minimized.

Also, I/O ratios were determined based on the results obtained from measurements conducted indoors and outdoors for all measurement points.

RESULTS and DISCUSSION

PM_{10} Concentrations

On January 4, 2023, measurements were conducted within both the locker room and the training hall. The training session commenced promptly at 16:05, at which point the student departed from the locker room. It is noteworthy that all windows and doors remained tightly shut throughout the duration of the training. The training session itself initiated with a series of warm-up exercises employing lightweight equipment, followed by the incorporation of squats and plank movements. The student arrived at the locker room at 16:43, where they engaged in a series of exercises, including lateral pull-downs, leg curls, and concluded with a running session. After their workout, the student left the locker room. Based on the collected data, it was found that

the impact of the student's mid-level training on the indoor air quality of the facility was minimal. Earlier in the day, the student had also visited the locker room briefly from 16:00 to 16:05 before heading to their class. Based on the collected data, it was observed that the concentration levels in the locker room were consistently higher than those in the training room. The highest concentration was recorded during the period when the student both entered and exited the locker room. Furthermore, an increase in concentration levels was noted after the student's entry into the locker room following their training session. The data obtained is given in Figure 2.

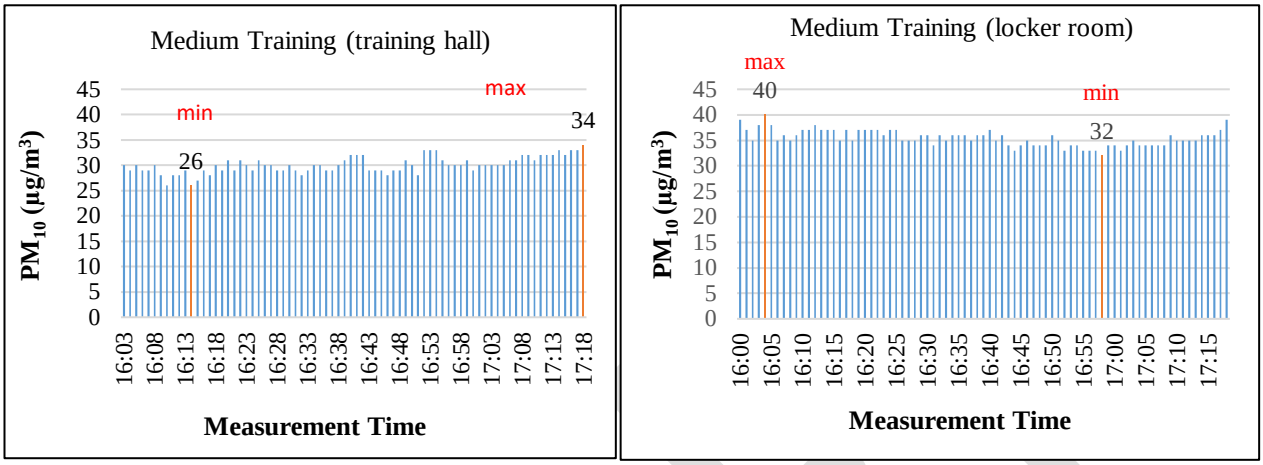


Figure 2. Medium training PM_{10} concentrations

On January 5, 2023, a series of measurements were conducted during a rigorous training session in various locations, namely the locker room, outdoor environment, and the training hall. The students commenced their training in the locker room at 07:57, beginning with a treadmill and spinning workout. Subsequently, both students alternated between intense rope exercises, which involved strenuous movements like weighted jumping and jumping planks. The training persisted until the conclusion of the session at 08:58. The outdoor measurements were initiated at 08:24, with the balcony door being partially ajar throughout this period. The data analysis revealed a significant spike in PM10 concentration, reaching its peak during the rope exercise session and the opening of the balcony door. Meanwhile, within the locker room, the highest concentration levels were recorded during the period when two students entered to use the facilities. Notably, concentrations outdoors consistently exceeded those indoors, indicating that external factors had the most pronounced impact on indoor air quality. The data obtained is given in Figure 3.

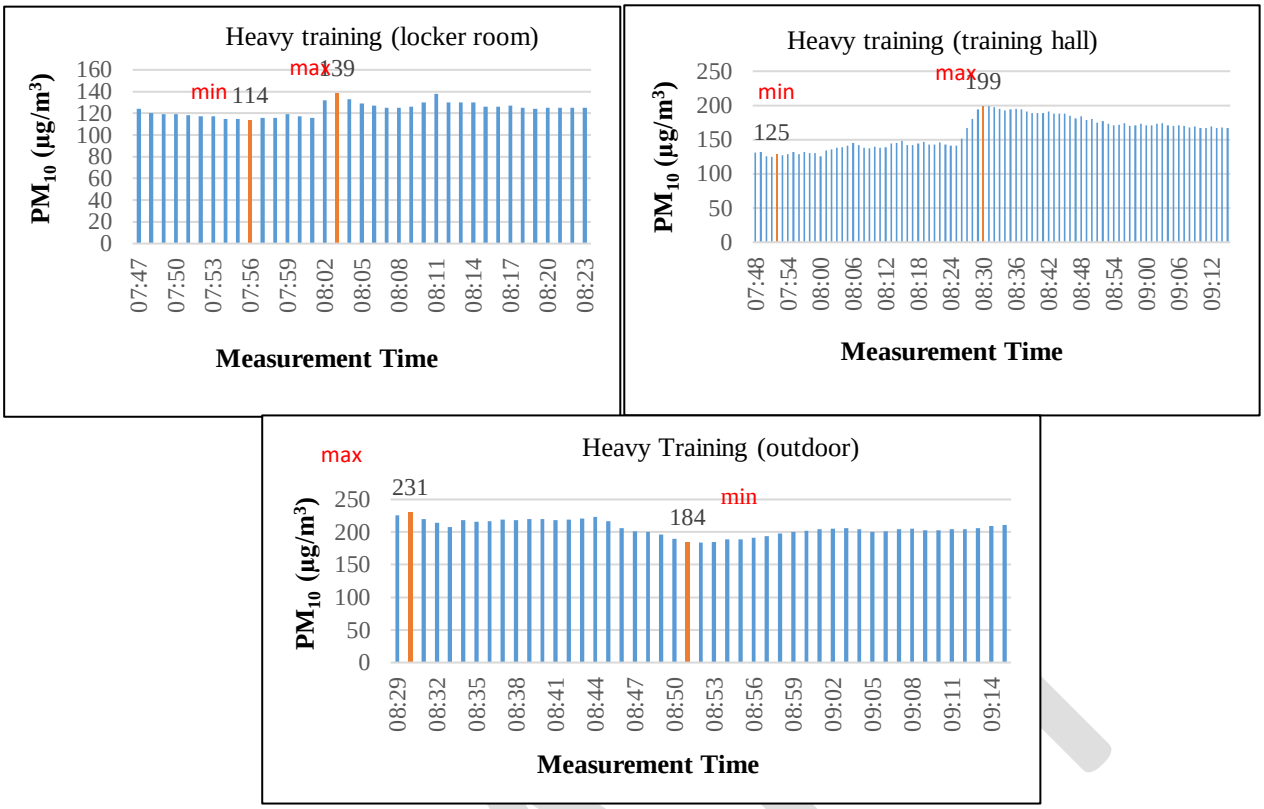


Figure 3. Heavy training PM_{10} concentrations

On January 5, 2023, a set of measurements was conducted during a light training session encompassing the locker room, outdoor environment, and training hall. The training regimen predominantly involved the utilization of a reformer pilates machine in the hall. The training session concluded at 13:38. Notably, at 13:16, Device 2 was relocated outdoors and activated for data collection. It is worth noting that the initial concentration levels were elevated before the commencement of the training. However, these concentrations decreased during the course of the training and subsequently exhibited a gradual increase as the balcony door was opened. The lowest recorded air quality levels were observed during the training session, indicating that the light training had minimal impact on indoor air quality. Conversely, the highest levels were registered after the training when the balcony door was opened. Inside the locker room, the peak concentration occurred when students entered after the 12:00 class, gradually declining to its minimum during moments of inactivity when no individuals were present. In the outdoor environment, the measurements began at their minimum recorded levels and displayed a consistent upward trend over time. The obtained data is given in Figure 4.

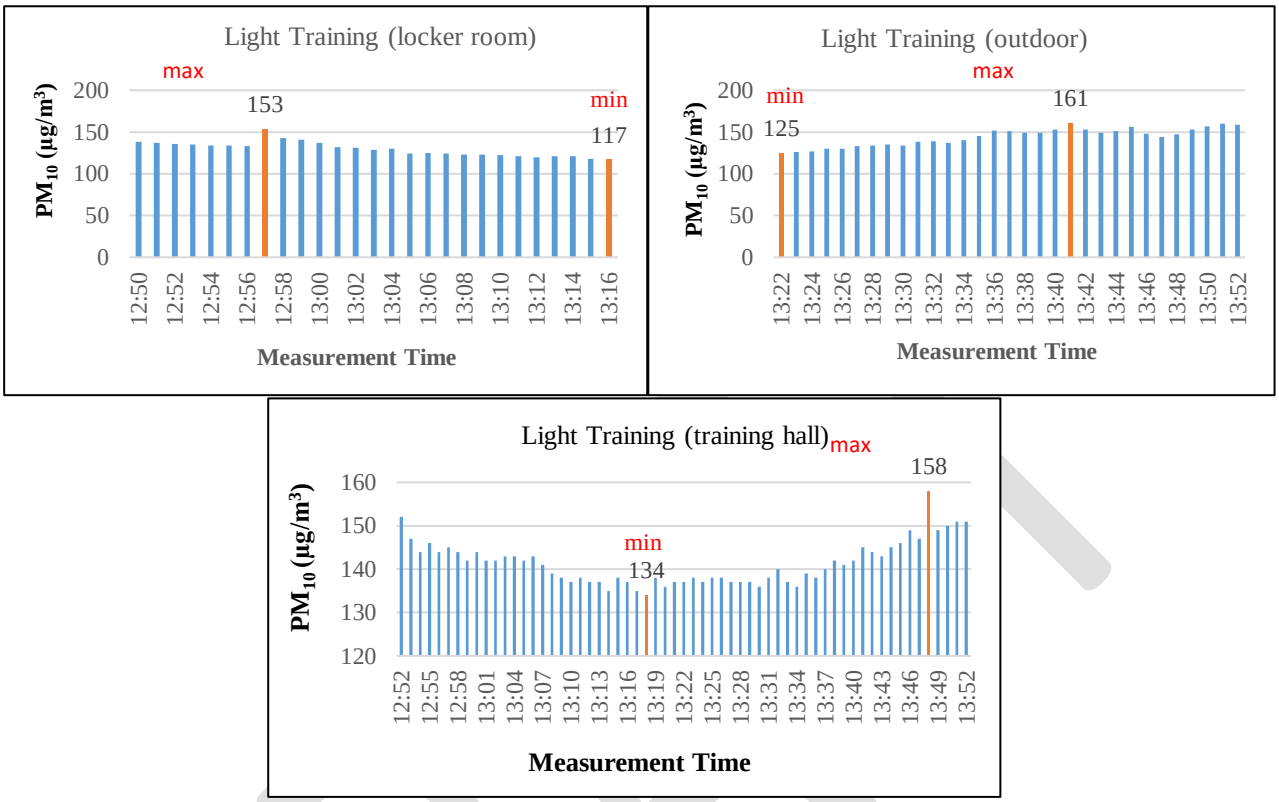


Figure 4. Light training PM_{10} concentrations

On January 6, 2023, a series of measurements were conducted in the locker room, outdoor environment, and training hall during a non-training period. The measurement session commenced at 11:10. At 11:29, Device number 2 was relocated from the locker room to the outdoors, where outdoor measurements were taken. Subsequently, the balcony door was left open. The measurement session concluded at 12:12. Initially, the air quality levels in the locker room were elevated, likely due to device preparation activities, but gradually decreased over time. In contrast, the changes in air quality levels within the training hall were minimal, but they reached their highest levels when the balcony door was open. This observed increase can be attributed to the outdoor environment being more polluted on the day of measurement compared to the indoor areas. The data obtained are given in Figure 5.

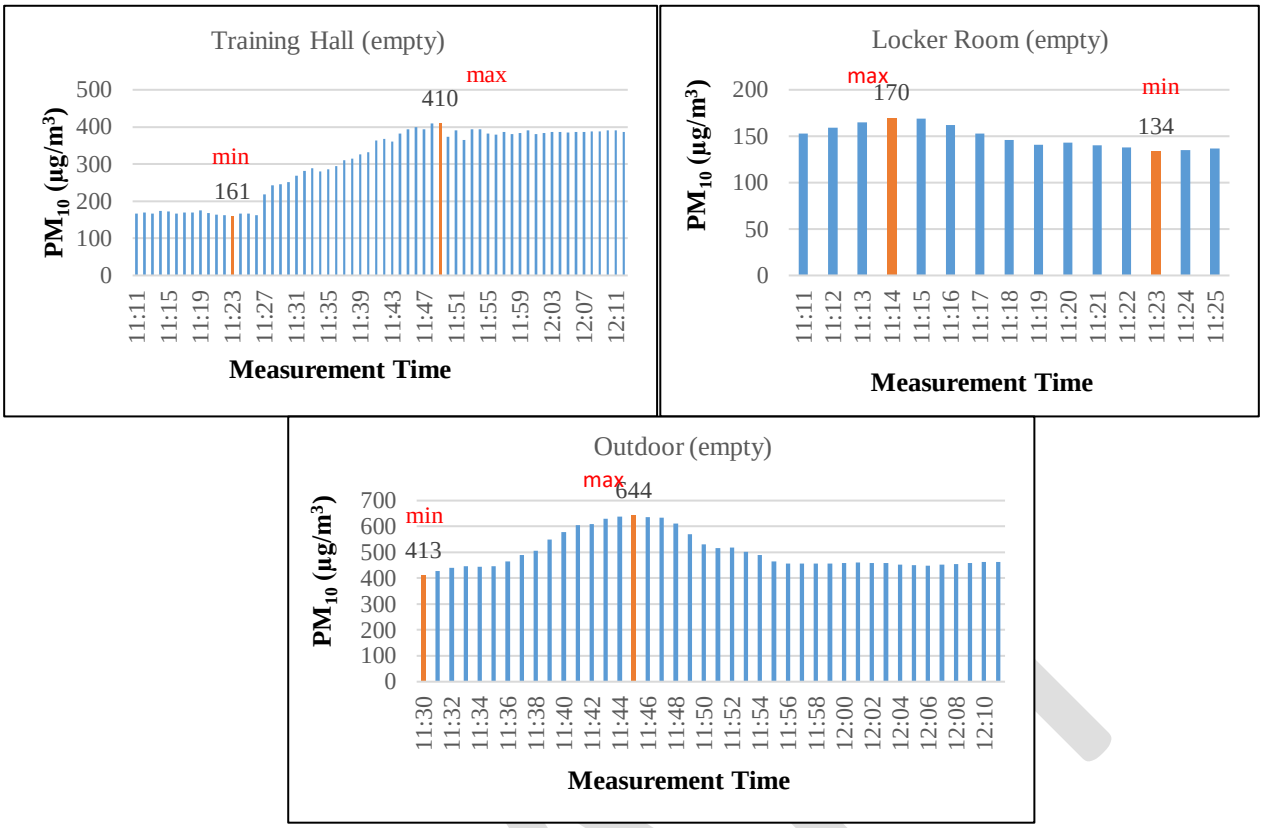


Figure 5. PM_{10} concentrations during non-training hours

I/O Concentration Ratios

Figure 6 presents the I/O ratio derived from concurrent measurements. When both the training hall and locker room are unoccupied, the I/O ratio is calculated at 0.72, which is below 1. A ratio below 1 signifies that indoor air quality levels remain lower than those of the outdoor environment when there is no activity within the indoor environment. In the context of light training, an examination of the I/O ratio reveals a value close to 0.98, approaching 1 but remaining slightly below it. This observation indicates that during light training, indoor PM_{10} concentrations experience an increase compared to an empty hall; however, they still remain somewhat lower than the levels outdoors. This underscores the fact that the influence of the outdoor environment on indoor air quality surpasses the impact of light training activities. Upon examining the I/O ratio during heavy training, it becomes evident that this value exceeds 1, indicating that the impact of heavy training on indoor air quality surpasses that of the outdoor environment.

CONCLUSION

Given the absence of nationally defined limit values for indoor air pollutants, a comparison is made between the average measured concentrations and the indoor PM_{10} limit set by the USEPA (United States Environmental Protection Agency), which stands at 150 $\mu g/m^3$. The data indicates that the measured values within the locker room consistently fall below this limit value across all training intensities and during unoccupied hours.

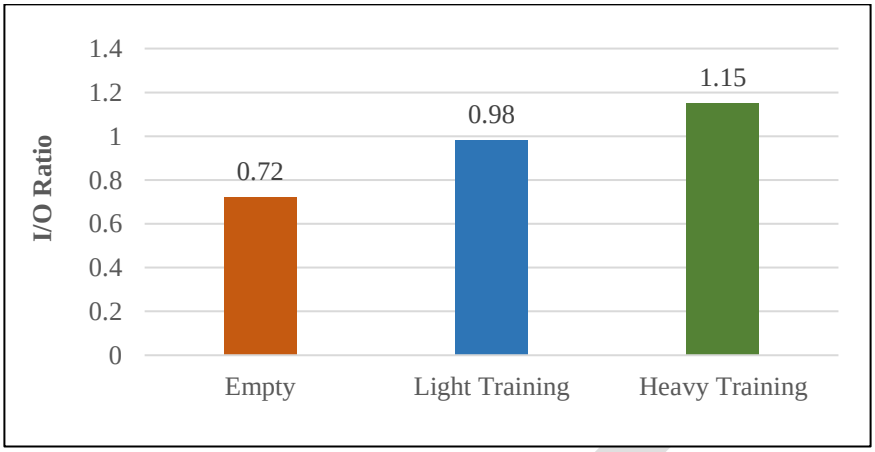


Figure 6. I/O Concentration Ratios

Upon examining the average values within the training hall, it becomes evident that the limit value was not exceeded during medium and light level training sessions. However, during heavy training and during unoccupied hours when the balcony door was open, PM_{10} concentrations exceeded the limit values, measuring at 160.94 µg/m^3 and 305.77 µg/m^3, respectively. This circumstance underscores the risk posed to coaches and students due to the influence of polluted air resulting from heavy training and external sources on indoor air quality. Clearly, measures must be taken to enhance the indoor air quality within this environment.

Short-term exposure to indoor air pollutants can lead to discomfort, including symptoms like headaches, throat irritation, and fatigue. Conversely, prolonged exposure to these pollutants poses more severe health risks, including the development of conditions like heart disease, respiratory ailments, and even cancer. Owners of private gyms should prioritize the provision of a healthier indoor air quality for all individuals frequenting their facilities. Achieving this goal can be accomplished through proper ventilation systems that consider the outdoor air quality, thereby ensuring gyms are comfortable and safe environments for everyone.

ACKNOWLEDGEMENTS
We would like to thank Yurdaer Songer for his valuable support for allowing us to make measurements in his private gym in our study.

References

Bralewska K, Rogula-Kozłowska W, Bralewski A 2022. Indoor air quality in sports center: Assessment of gaseous pollutants. *Building and Environment*.
Brunekreef B, Holgate ST 2002. Air pollution and health. *The Lancet*, 1233-1242.
Cincinelli A, Martellini T 2017. Indoor Air Quality and Health. *Int. J. Environ. Res. Public Health*, 14.
Dominici F, Greenstone M, Sunstein, CR 2014. Particulate Matter Matters. *Science*.
Engel-Cox J, Oanh NT, Donkelaar AV, Martin RV, Zell E 2013. Toward the next generation of air quality monitoring: Particulate Matter. *Atmospheric Environment*, 584-590.
Sofuoğlu SC 2016. İç Hava Kirleticileri ve İnsan Sağlığına Etkisi. *Tesisat Mühendisliği*, 33-44.
Zhang L, Ou C, Magana-Arachchi D, Vithanage M 2021. Indoor Particulate Matter in Urban Households: Sources, Pathways, Characteristics, Health Effects, and Exposure Mitigation. *Int J Environ Res Public Health*.

ORAL PRESENTATION

Lifestyle and risk for insulin resistance

Griselda Korçari , Jona Keri[2], Iris Plaku[3]

[1]Department of Technical Medical Laboratory and Imaging, Faculty of Medical Technical Sciences,

Aldent University, Tirana, Albania.

[2] Pharmacotherapeutic Research Center, Faculty of Medical Sciences, Aldent University, Tirana,

Albania

Email the corresponding author: Griselda KORÇARI (Griselda.zacaj@ual.edu.al1)

0355694443675

Abstract

Introduction: The increase in the number of patients with Diabetes Mellitus (type II) is closely related to the development of insulin resistance. This constitutes an international problem that requires special attention from the laboratory point of view and community awareness for recognizing risk factors that increase the risk of insulin resistance.

The purpose of this study is to identify whether lifestyle is a risk factor for possible insulin resistance and consequently for the development of type II diabetes mellitus after the Covid 19 pandemic.

Materials and methods: A random sample of forty-four students aged 18-22 living in the city of Tirana, who had different lifestyles, was selected to fill out a form with qualitative, evaluative data on lifestyle (physical activity, consumption of cigarettes, type of electronic or regular cigarette, family history of diabetes, concomitant pathology). For each member of the sample, the blood sample (serum) was examined for two parameters: fasting blood glucose and triglyceride. Based on these parameters, the TyG Index was calculated.

Results and discussions: 22 males and 22 females were included in the study.

• The ratio of cigarette users and non-users was 24/20 respectively.

• The ratio between normal body mass index (BMI) and abnormal BMI was 33/11 respectively.

Statistical processing with SPSS version 21 resulted:

• There is a statistically significant relationship between TyG and BMI *(p=0.000)*

• There is a statistically significant relationship between TyG and cigarette consumption *(p=0.000)*

• There is a statistically significant relationship between TyG and the daily amount of cigarettes smoked *(p=0.000)*

• There is a statistically significant relationship between TyG and alcohol *(p=0.000)*

• There is a statistically significant relationship between TyG and physical activity *(p=0.000)*

• There is a statistically significant relationship between TyG and medications *(p=0.000)*

Conclusions: Cigarette smoking, physical activity, alcohol and medications can be classified as independent risk factors for insulin resistance. Based on the results of the study, it is recommended to carry out other studies with a larger number of samples to better understand the role of each factor in increasing the risk of insulin resistance in order to make the population aware to avoid these factors. and to reduce the incidence of Diabetes in this age group.

Keywords: insulin resistance, BMI, cigarettes, alcohol, physical activity, Diabetes Type II.

Introduction

The increasing number of patients with diabetes mellitus (type II) is strictly correlated to the development of insulin resistance, which is the fundamental pathogenic component of many metabolic diseases, including type 2 mellitus diabetes, and is defined as a state of Reduced reactivity of insulin targeting fabrics to physiological insulin levels. (Lee et al., 2022). In recent years, it has been found that inflammation, the stress of the endoplasmic lattice and mitochondrial dysfunction, can promote insulin resistance (Gutiérrez-Rodelo et al., 2017) modifiable factors that are thought 'Insulin include diet, exercise, smoking, and stress. (Kelly, 2000) The

two basic forces that spread insulin resistance are the increase in the consumption of caloric low food fast food and the decrease in physical activity due to mechanized transport and the sedentary form of temporal activities. (Saklayen, 2018). A life factor that can have direct and indirect effects on insulin resistance is cigarette smoke (Artese et al., 2017). The prevalence of smoke of men and women aged 15 or over 15 in Albania, who currently smoke any tobacco product on a daily or non-daily basis for 2020 was 22.40% (Albania smoke rate 2000-2023, n.d.) except cigarette smoke, e-cigarettes are becoming very popular through young-adults. Electronic cigarettes (electronic cigarettes) are devices that can vaporize a nicotine solution combined with liquid flavours instead of burning tobacco leaves. (Rom et al., 2015) Electronic cigarettes are advertised aggressively as a help for the cessation of smoking; healthier, cheaper and more socially acceptable than conventional cigarettes (Rom et al., 2015). Therefore, the purpose of this study was to strongly refer to Jeong et al., 2021) should lay down the connection between lifestyle and insulin resistance in Aldent University students through the use of the Tyg index. The index of the triglyceride-glucose (Tyg index) is a marker of insulin resistance and metabolic dysfunction and has the advantage of being universally available (ZaIGham et al., 2022) and is greater than Homa-IR to predict type diabetes 2 (Park (Park. Et al., 2021). For these reasons, it is reasonable to evaluate the connection between lifestyle and insulin resistance using the Tyg index.

Materials and methods

The questionnaire

A random sample of 50 students from different departments of Aldent University has been selected to fill in a questionnaire with private data on lifestyle. The first part of this questionnaire suggested information on simple habits such as smoking, physical activity, alcohol consumption, personal and family history (including type II diabetes), age, height and weight and BMI, followed by the information part who requested the student's consent for performing phlebotomy.

Laboratory tests

Information from participants diagnosed with chronic illness was excluded leading to performing phlebotomy to 44 individuals. In order to receive fresh serum samples. Blood was collected from forearm veins using a 21G vacuum system and centrifuged at 3000 rpm. Serum samples were examined for fasting glucose and triglyceride levels. A total of 10ul serum samples was mixed with 1000 glucose reagents (product #:21-0672, GPSL-0507, ELITech Clinical system, France) Glucose was measured after 10 min of incubated mixture at 37 celcius degrees. All tubes were evaluated for hemolysis, icterus, and lipemia. The CYANSMART device (ELITEK) was used to analyze the samples. Triglyceride and glucose reagents were kept at a temperature of 18-25ºC for 15-20 minutes. After each test, a calibration and control procedure was performed.
The principle of the Glycemic measurement method is ***Glucose oxidase.***

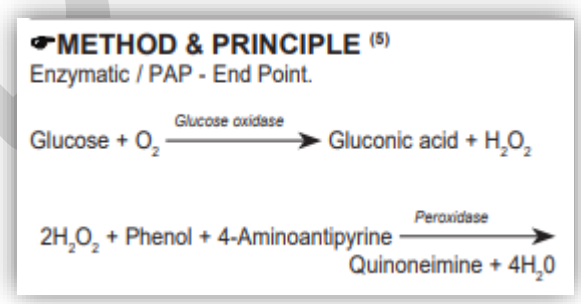


Figure 1. *The principle of the glucose method*

Manual procedure
Wavelength 505nm
Optical path: 1cm
Sample/ Reagent ratio: 1:100

Temperature: 37ºC

Read against reagent blank.

	Blank	Calibration	Test
Reagent R	1000 µl	1000 µl	1000µl
Distilled water	10 µl	-	
Standard/Calibrator	-	10 µl	-
Sample	-	-	10 µl

We mixed and read the absorbances (A) after 10 minutes of incubation.

Following the instructions triglyceride test was performed after the 10 minutes of incubated mixture of 10 ul serum sample and 1000 ul of triglyceride reagent (product #:22-0281, TGML-0427, ELITech Clinical system, France). The principle of the method of measuring triglycerides vs. Triglyceride Oxidase. The principle of the method of measuring triglycerides vs Triglyceride Oxidase. Both spectrophotometric methods are END points

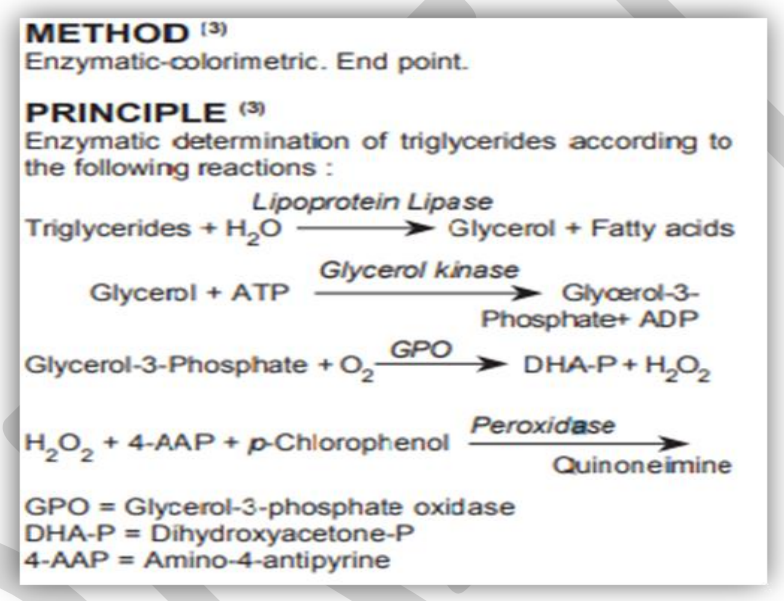


Figure 2. The principle of the Triglicerid method

Manual procedure

- Wavelength 505nm
- Optical path: 1cm
- Sample/ Reagent ratio: 1:100
- Temperature: 37ºC

Read against reagent blank.

	Blank	Calibration	Test
Reagent R	1000 µl	1000 µl	1000µl
Distilled water	10 µl	-	
Standard/Calibrator	-	10 µl	-
Sample	-	-	10 µl

We mixed and read the absorbances (A) after 10 minutes of incubation.



Figure 3. Semi-automatic biochemistry analyzer CYANSmart

All samples were measured at the beginning and their values were considered control values
2.3 TyG index Glucose and triglyceride results from each participant were necessary to calculate the TyG index. The TyG index is calculated using the formula ln [triglyceride (mg/dL) × fasting blood glucose (mg/dL)/2] ln [triglyceride (mg/dL)×fasting blood glucose (mg/dL)/2] and is expressed on a logarithmic scale (Jeong et al., 2021)

2.4 Statistical analysis

Before statistical analysis, participants were classified into 5 groups of smokers: only cigarette smokers, non-cigarette smokers, E-cigarette smokers, ex-cigarette smokers, and dual smokers (cigarette and E-cigarette smokers). Also participants were classified into the following insulin resistance groups according to the median TyG index (8.5): low insulin resistance group (< 8.5) and high insulin resistance group (≥ 8.5) (Jeong et al., 2021). IBM SPSS statistics version 21 was used to perform multinomial logistic regression and correlations between lifestyle habits and the TyG index. Two-sided $p < 0.05$ was considered statistically significant.

Results of the study

Table nr 1. Indicates the statistical relationship between the Body Mass Index and the TyG index. According to statistical processing, we have a statistically significant relationship *(P=0000).*

Paired Samples Test

		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Indeksi mases trupore - TyG Index	15.07935	2.94025	.44326	14.18543	15.97327	34.019	43	.000

Pair 2	TyG Index - Kosumues cigare	-6.89259	.76213	.11489	6.66088	7.12429	59.990	43	.000

Table nr 2. Indicates the statistical relationship between cigarette smoking, the amount of daily cigarettes consumed, alcohol consumption and the TyG index.
From the statistical processing, an important statistical relationship between them can be seen, *(P=0000).*

Paired Samples Test

		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	**Kosumues cigare - TyG Index**	-6.89259	.76213	.11489	-7.12429	-6.66088	-59.990	43	.000
Pair 2	**Sasia e cigareve qe tymosni ne dite - TyG Index**	-4.50622	1.59101	.23985	-4.98993	-4.02251	-18.787	43	.000
Pair 3	**TyG Index - Konsumimi i alkoolit te te intervistuareve**	5.09713	1.38767	.20920	4.67524	5.51902	24.365	43	.000

Table nr 3. Indicates the statistical relationship between the use of medications and the TyG index. From the statistical processing, an important statistical relationship between them can be seen, *(P=0000).*

Paired Samples Test

		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Sasia e cigareve qe tymosni ne dite - TyG Index	-4.50622	1.59101	.23985	-4.98993	-4.02251	-18.787	43	.000
Pair 2	Perdorimi i **medikamenteve - TyG Index**	-6.71077	.62086	.09360	-6.89953	-6.52201	-71.698	43	.000

Table nr 4. Indicates the statistical relationship between the TyG index and physical activity. From the statistical processing, an important statistical relationship between them can be seen, *(P=0000).*

Paired Samples Test

		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Sasia e cigareve qe tymosni ne dite - TyG Index	-4.50622	1.59101	.23985	-4.98993	-4.02251	-18.787	43	.000
Pair 2	TyG Index - Ushtrimi i aktivitetit fizik	5.84713	1.27738	.19257	5.45877	6.23549	30.363	43	.000

4. Discussions

Insulin resistance can be assessed by the Triglyceride-Glucose Index (TyG), a simple, low-cost and easy-to-apply method. With this index, the risk of patients developing pathologies can be calculated. cardiovascular disease, diabetes type II.
There are various studies that show the role of this index in predicting cardiovascular events with sensitivity and specificity values between 67 and 96% and 32.5-85%, respectively (Sánchez-García et al., 2020) as well as studies others that show the connection of TyG diabetes type II. (MAEDICA – a Journal of Clinical Medicine https://doi.org/10.26574/maedica.2021.16.3.375 2021; 16(3): 375-381)
Since the sedentary life is important for these pathologies, this study discussed the relationship between the TyG index and the sedentary life of students after the COVID-19 pandemic.
From our study, it was shown that there is a statistically significant relationship of sedentary factors (alcohol, physical activity, body weight, smoking) with TyG. These results are probably related to the lack of physical activity due to the isolation, perhaps due to the increased consumption of alcohol and cigarettes as an anti-anxiety tool to overcome the isolation, however, this should be verified in a wider group.
Also, it would be interesting if this study were carried out on the same group after a few years to see if people with high TyG would manifest Type II Diabetes, so TyG could be seen as an effective screening tool in predicting the control of diabetes. glycemia and insulin resistance in T2DM

5. Acknowledgement

This study was conducted at the Biochemical Laboratory at Aldent University. A special thanks goes out to Jona Keri, and Iris Plaku, for their help, support and review.

Reference

Albania Smoking Rate 2000-2023. (n.d.). Retrieved September 12, 2023, from https://www.macrotrends.net/countries/ALB/albania/smoking-rate-statistics

Artese, A., Stamford, B. A., & Moffatt, R. J. (2017). Cigarette Smoking: An Accessory to the Development of Insulin Resistance. *American Journal of Lifestyle Medicine*, *13*(6), 602–605. https://doi.org/10.1177/1559827617726516

Gutiérrez-Rodelo, C., Roura-Guiberna, A., & Olivares-Reyes, J. A. (2017). [Molecular Mechanisms of Insulin Resistance: An Update]. *Gaceta Medica De Mexico*, *153*(2), 214–228.

Jeong, S. H., Joo, H. J., Kwon, J., & Park, E.-C. (2021). Association Between Smoking Behavior and Insulin Resistance Using Triglyceride–Glucose Index Among South Korean Adults. *The Journal of Clinical Endocrinology & Metabolism*, *106*(11), e4531–e4541. https://doi.org/10.1210/clinem/dgab399

Kelly, G. S. (2000). Insulin resistance: Lifestyle and nutritional interventions. *Alternative Medicine Review: A Journal of Clinical Therapeutic*, *5*(2), 109–132.

Lee, S.-H., Park, S.-Y., & Choi, C. S. (2022). Insulin Resistance: From Mechanisms to Therapeutic Strategies. *Diabetes & Metabolism Journal*, *46*(1), 15–37. https://doi.org/10.4093/dmj.2021.0280

Park, H. M., Lee, H. S., Lee, Y.-J., & Lee, J.-H. (2021). The triglyceride–glucose index is a more powerful surrogate marker for predicting the prevalence and incidence of type 2 diabetes mellitus than the homeostatic model assessment of insulin resistance. *Diabetes Research and Clinical Practice*, *180*. https://doi.org/10.1016/j.diabres.2021.109042

Rom, O., Pecorelli, A., Valacchi, G., & Reznick, A. Z. (2015). Are E-cigarettes a safe and good alternative to cigarette smoking? *Annals of the New York Academy of Sciences*, *1340*, 65–74. https://doi.org/10.1111/nyas.12609

Saklayen, M. G. (2018). The Global Epidemic of the Metabolic Syndrome. *Current Hypertension Reports*, *20*(2), 12. https://doi.org/10.1007/s11906-018-0812-z

Zaigham, S., Tanash, H., Nilsson, P. M., & Muhammad, I. F. (2022). Triglyceride-Glucose Index is a Risk Marker of Incident COPD Events in Women. *International Journal of Chronic Obstructive Pulmonary Disease*, *17*, 1393–1401. https://doi.org/10.2147/COPD.S360793

DRAFT

ORAL PRESENTATION

Manyetik alan duyarlı veya çoklu uyaran duyarlı elektroeğirilmiş nanofiberlerin kanser tedavisinde kullanımı

Gökmen Arabacı[1*] (ORCID: 0000-0002-5848-6117), Yasemin İspirli Doğaç[2] (ORCID: 0000-0001-8616-0280)

[1*] Muğla Sıtkı Koçman Üniversitesi, Fen Bilimleri Enstitüsü, Moleküler Biyoloji ve Genetik Anabilim Dalı , Muğla, Türkiye
[2]Muğla Sıtkı Koçman Üniversitesi, Muğla Meslek Yüksekokulu, Kimya Ve Kimyasal İşleme Teknolojileri Bölümü, Muğla, Türkiye

* gokmen_arabaci@hotmail.com

Özet

Kanser, hücrelerin kontrolsüz bir şekilde artması ve büyümesiyle ilişkili en ölümcül olan hastalıklardan biridir. En yaygın kanser tipleri, meme, akciğer, mide, kolon, prostat ve cilt kanseridir. Ancak kanser her organda oluşabilir. Kanserin evresine, türüne, hastanın sağlık durumuna göre farklı kanser tedavi yöntemleri vardır. Bunlar arasında radyoterapi, kemoterapi, cerrahi yöntemler, hipertermi, biyolojik tedaviler, fotodinamik tedavi, kök hücre nakli, lazer tedavisi, kan ürünlerinin bağışı, hormon tedavileri veya immünoterapi gibi yöntemler sayılabilir. Klasik kanser tedavilerinin ağır yan etkileri, tümörlü hücrelerin yanında sağlıklı hücrelerinde zarar görmesi gibi olumsuzluklar yüzünde farklı tedavi yöntemleri araştırılmaya devam etmektedir. Nanofiberlerin yüksek yüzey alanı, farklı polimer çeşitleri ile kompozit olarak farklı özeliklere sahip malzeme üretme olanağı gibi avantajları, bu malzemelerin kanser tedavisinde kullanılabilirliğinin araştırılmasını çekici kılmaktadır. Bu çalışma, manyetik duyarlı elektroeğrilmiş nanoliflerin ve çoklu uyaranlara duyarlı elektroeğrilmiş nanoliflerin kanser tedavisinde kullanımına ilişkin çalışmalarla ilgili persfektif sunmaktadır.

Anahtar Kelimeler: Kanser, nanofiber, kanser tedavisi, manyetik duyarlı nanofiber, çoklu uyaranlara duyarlı nanofiber

Usage of magnetic sensitive or multi-sensitive electrospun nanofibers for cancer treatment

Abstract

Cancer is one of the most fatal diseases associated with the uncontrolled increase and growth of cells. The most common types of cancer are breast, lung, stomach, colon, prostate and skin cancer. However, cancer can occur in any organ. There are different cancer treatment methods depending on the stage and type of cancer and the health condition of the patient. These include methods such as radiotherapy, chemotherapy and surgical methods, hyperthermia, biological treatments, photodynamic therapy, stem cell transplantation, laser treatment, donation of blood products, hormone treatments or immunotherapy. Different treatment methods continue to be investigated due to the negative effects of classical cancer treatments such as severe side effects and damage to healthy cells as well as tumor cells. Advantages of nanofibers, such as high surface area and the possibility of producing materials with different properties as composites with various polymer types, make it attractive to investigate the usability of these materials in cancer treatment. This study focuses on studies on the use of magnetically responsive electrospun nanofibers and multi-stimulus-sensitive electrospun nanofibers in cancer treatment.

Keywords: Cancer, nanofiber, cancer treatment, magnetic sensitive nanofiber, multi-sensitive nanofiber

Kanser ve klasik tedavi yöntemleri

Kanser insan hastalıkları arasında kalp hastalıklarından sonra gelen en ölümcül hastalık olarak bilinmektedir. Yapılan çalışmalar önümüzdeki yıllarda kanser vakalarının artıcağını göstermektedir. 2025 yılına kadar yılda 20 milyondan fazla kanser vakası olacağı tahmin ediliyor (Zugazagoitia ve ark, 2016). Kanser hücre büyümesini ve çoğalmasını kontrol eden mekanizmanın doğru çalışmamasından kaynaklanır. Klinik belirtileri ağrı, ülserler, sıklıkla şişlikler, kanama gibi durumlarla kendini gösterir. Sistemik belirtiler kilo kaybı, yorgunluk ve iştah kaybıdır. Kanserin tedavisi üzerine tıp dünyası ve birçok çeşitli alandan araştırmacılar çalışmalar yapmaktadır (Wenhua ve ark, 2022). Kanser tedavileri genellikle radyoterapi, kemoterapi ve cerrahi yöntemler ile yapılırken, bazen hipertermi, biyolojik tedaviler, fotodinamik tedavi, kök hücre nakli, lazer tedavisi, kan ürünlerinin bağışı veya hormon tedavileri gibi yöntemler de kullanılabilir. Klasik kanser ilaçları kanser hücrelerinin DNA replikasyonunu engelleyerek etki gösterir, ancak araştırmalar seçici bir etki yaratmadığını göstermektedir. Bu tedavilerin yan etkileri arasında ağız yaraları, yorgunluk, ağrı ve enfeksiyonlar, saç dökülmesi, mide bulantısı bulunabilir. Alternatif tedavi yöntemleri araştırılmaktadır, ancak kanserin karmaşıklığı ve farklı türlerinin varyasyonları, tümör hücrelerinin yanında sağlıklı hücrelerede zarar vermeden etkili bir tedavi bulmayı zorlaştırmaktadır (Han ve Li, 2011; Maeng ve ark, 2010; American Cancer Society, 2023).

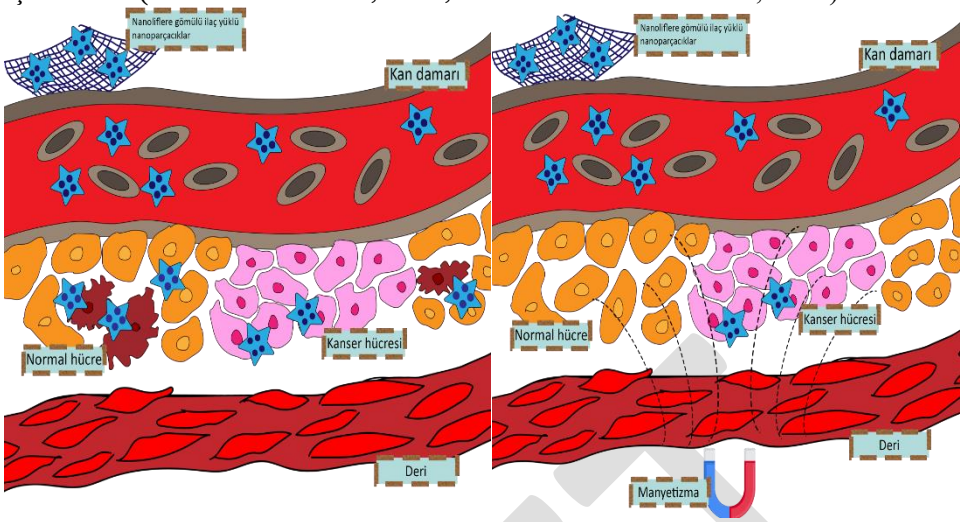
Elektroeğrilmiş (elektrospun) nanofiberler

Elektroeğirme yöntemi nanofiber üretim yöntemleri arasında en sık kullanılan yöntemlerden biridir. Temel fizik, elektrik fiziği, polimer kimyası, akışkanlar dinamiği gibi birçok alanı içinde barındıran multidispliner bir yöntemdir. Elektroeğirme cihazı yüksek voltaj güç kaynağı, besleme ünitesi (metal iğne, şırınga v.b.), Toplayıcı (iletken plaka, döner silindir v.b.) olmak üzere üç ana kısımdan oluşur. Yöntemin temel prensibi; polimer eriğinin besleme ünitesine alındıktan sonra güç kaynağı yardımı ile uygulanan voltaj sayesinde ilk önce damlacık halini alması ardından manyetik alan içinde çekilerek incelmesi ve lif yapısını alması şeklinde tanımlanabilir (Li ve ark, 2004; Xue ve ark, 2019).
Eleketroğirme yöntemiyle nanofiber üretiminde kullanılabilen polimer çeşidinin çok fazla olması istenilen özelliklere sahip nanofiber üretimini mümkün kılmaktadır. Elektroeğrilmiş (elektrospun) nanofiberler bu sebepten birçok farklı uygulma alanında kullanılmaktadırlar ancak bu yazıda konuyla bağlantılı olarak biyomedikal uygulamalardaki kullanımına yer verilecektir. Elektroeğrilmiş nanofiberler yüksek çözünürlük, yüksek yüzey-hacim oranı ve sağladığı geniş kullanım alanı gibi çeşitli avantajlara bağlı olarak özellikle farmasötik alanda ve biyomedikal uygulamalarda geniş kullanım alanı bulmaktadır. Nanofiber teknolojisi, yanık ve yara bakımı, organ onarımı, osteoporoz, doku rejenerasyonu ve ilaç dağıtımı gibi biyomedikal alanda yaşanan zorluklara potansiyel bir çözüm olarak birçok araştırmacının dikkatini çeken heyecan verici bir alandır. Güncel çalışmalar göstermiştir ki, biyomedikal nanoliflerin sağladığı geniş yüzey alanı, gelişmiş hücre aktiviteleri, ilaç kapsüllenmesi ve ilaç salınım hızının kontrolü için kullanılabilir. Malzeme seçimleri ve lif oryantasyonunun ayarlanmasıyla elektroeğirme prosesi optimize edildiğinde, hücre farklılaşması ve ilaç salınım kontrolü gibi alanlarda daha fazla gelişme sağlanabilir olması, araştırmacıların ilgisini giderek daha çok çekmektedir (Leung ve Ko, 2011; Yıldız ve ark, 2020; Kanmaz ve ark, 2018).

Manyetik duyarlı elektroeğrilmiş nanoliflerin kanser tedavisinde kullanımı

Kanser tedavi yöntemleri arasında en sık kullanılan kemoterapi ve radyoterapi olmasına rağmen sağlıklı dokularada zarar veren, toksisite ve olumsuz yan etkileri olan yöntemlerdir (Mokhosi ve ark, 2022). Lokal tümör bölgede tedavinin yoğunlaşması ve komşu sağlıklı hücrelerin en az düzeyde hasar görmesi hedefiyle manyetik elektroeğrilmiş nanoliflerin kanser tedavisi için kulanımı dikkatleri üstüne çekmektedir. Bu malzemeleri elektroeğirme yönteminde kulanılabilen birçok polimer ile manyetik nanoparçacıkları kompozit oluşturarak hazırlamak mümkündür (Huang ve ark, 2012). Böylece manyetik özellik kazanan nanolifler dış manyetik alan etkisiyle taşınabilir olması onları hedef bölgeye duyarlı kontrollü ilaç salınımı için mükemmel taşıyıcılar yapmaktadır. **Şekil 1**' de gösterildiği üzere klasik ilaç dağıtımı hasarlı hücre bölgesinin yanı sıra

sağlıklı hücreleride etkilemektedir. Manyetik ilaç yüklü nanolifler dış manyetik alan etkisi ile manipüle edilip hedef bölgeye çekilebilir (Patel ve Gundloori, 2023; Contreras-Cáceres ve ark, 2019).



Şekil 1. Manyetik özelikli ve klasik elektroeğrilmiş nanoliflerin ilaç salım farkı

Wang ve arkadaşları Fe_3O_4 manyetik parçacıklarını iki farklı selüloz türevi polimerle kompozit halinde elektroeğirme yöntemi kullanarak üretmişlerdir. Üretilen manyetik nanolifler hedefe duyarlı ilaç salım sisteminde kullanımış, model ilaç olarak indometasin (IDN) ve aspirin seçilmiştir. Özellikle sindirim sisteminde hedef bölgeye duyarlı ilaç salımı için olumlu sonuçlar alınmıştır (Wang ve ark, 2012). Başka bir çalışmada doksorubisin hidroklorür (DOX) yüklü elektroeğrilmiş kitosan/kobalt ferrit/titanyum oksit nano lifleri sentezlenmiştir. Bölgesel ilaç salım incelemeri ardından nanoliflerinin lokalize kanser tedavisinde kullanılabileceği sonucuna varılmıştır. (Radmansouri ve ark, 2018).

Manyetik nanolifler lokal bölge ilaç salım uygulamalarının yanı sıra hipertermi kanser tedavilerinde sağladığı pozitif etkiler nediyle dikkat çekmektedir. Hipertermi klasik kemoterapi ve radyoterapi yöntemi gibi etkili olan bir kanser tedavi yöntemidir (Hadi ve ark, 2019; Wei ve ark, 2022). Tümör hücreleri sağlıklı hücrelere göre 41° ila 45°C' arasındaki yüksek sıcaklıklara daha duyarlıdır. Bu tedavi yönteminde sağlıklı hücreler mümkün olduğu kadar korunarak tümör hücrelerin ısıtılarak apoptoza uğraması sağlanır (Moroz ve ark, 2002). Hipertermi etkisi yaratmak için manyetik nanopartiküller (MNP) dış bir manyetik alana maruz kaldığında manyetik enerji termal enerjiye dönüştülebilir, busayede tümör dokusuna nüfuz etmiş olan MNP hücrelerin ölümü için gereken ısıyı sağlanmış olur (Hervault ve Kim Thanh, 2014; Schneider-Futschik ve Reyes-Ortega, 2021; Mamun ve Sabantina, 2023).

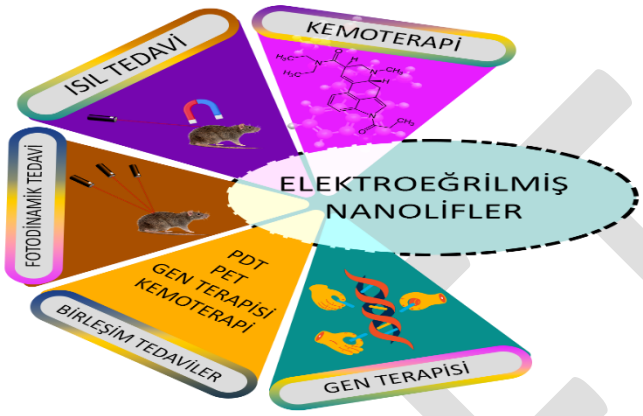
Polikaprolakton (PCL)/Fe_3O_4 kompoziti portatif bir elektro eğirme cihazı ile sentezlenmiş ısıtma verimi sonuçlarına göre hipertermi tevasi için kullanılabileciği öne sürülmüştür (Hu ve ark, 2020). Song ve arkadaşları Elektroeğirme yöntemiyle Fe_2O_3/Poliüretan manyetik nanolifleri sentezlemiş, manyetik nanolifler dış manyetik alan etkisiyle 70 saniyede 43°C' sıcaklığa ulaşmıştır (Song ve ark, 2018). Yine elektroeğirme yöntemiyle yağ asidi (Laurik asid) içine MNP kapsüllenmiş hipertermi tevasi için kullanılabilirliği araştırılmıştır. MNP 40-48°C'de 70-75 Jg-1 ısı emme ve salma kapasitelerine sahip olduğu bildirilmiştir (Sarier ve ark, 2018). Wei ve ark. önemli bir cilt kanser türü olan melenomun lokalize termo-kemoterapi tedavisi için ısıya duyarlı (açık-kapalı) ilaç yüklü manyetik nano elyaflar önerdi. Doğal antikanser ajanı olan kurkimin yüklü manyetik nano elyafların dış manyetik alan etkisiyle ısısının arttırıp kurkimin salımının kontrol edilebildiği gösterildi (Wei ve ark, 2022). Huang ve ark. polistiren/demir oksit nanoparçacıklarını elektroeğirme yöntemiyle sentezledi. Yüksek ısıtma kapasitesine sahip MNP lere bağlı olan kanser hücrelerinin manyetik alan etkisi altında ısınmaya bağlı olarak öldüğü gösterildi (Huang ve ark, 2012). 2021 yılında yapılan bir araştırmada poliakrilonitril/Fe304 manyetik özeliğe sahip nanoliflerin manyetik alan alan altında ısıtma ve hareket kabiliyetleri test edildi. Manyetik nanolifler 15.57 kA/m'lik bir manyetik alanda 49.9°C'ye kadar ısı üretti ve 2,15 mm/sn'lik bir hareket hızı sağladı (Lee ve Kim, 2021). Matos ve ark. manyetik hipertermi uygulamaları için selüloz asetat ve Fe304 MNP' leri birleştirip kompozit membranlar üretti. Manyetik özellikli membranlar iyi ısıtma değerleri gösterdi. Hipertermi tedavisinde komşu sağlıklıklı hücrelerin zarar görmemesi için ıstma değerlerinin nanopartikül konsantrasyonunun değiştirilerek düzenlenebileceği belirtildi (Matos ve ark, 2018). 2013 yıllında yapılan bir çalışmada Fe_2O_3/Poliüretan nanoliflerin yüksek frekans manyetik alan altında ısıtma değerleri incelendi. Yüksek frekanslı manyetik alan

altındaki kompozit nanolifler Fe_2O_3 miktarına ve manyetik alan kuvvetine bağlı olarak saf poliüretan nanoliflere göre %3-17'lik bir sıcaklık artışı göstermiştir (Park ve ark, 2013).

DRAFT

Çoklu uyaranlara duyarlı elektroeğrilmiş nanoliflerin kanser tedavisinde kullanımı

Uyaranlara yanıt veren akıllı nanoliflerin kanser tedavilerinde kullanımında, birden çok uyarana yanıt veren malzemeler geliştirerek sinerjistik olarak nanoliflerin faaliyetini arttırma yönünde çalışmalar yapılmaktadır. Elektroeğirme yönteminde kullanılabilecek olan polimer çeşitliğinin çok fazla olması bu anlayışı desteklemektedir. Akıllı ilaç tasarımlarında seçilen polimer dış ortamdan gelen çeşitli uyarılara karşı şişerek ya da büzüşerek cevap verebilmektedir. Bu kompozitlere MNP de dahil olunca ışık, sıcaklık, Manyetik alan, pH gibi manipülasyona neden olan özellikler aynı malzeme içinde birlikte kullanılabilmektedir (Zhao ve Cui 2020; Li ve ark, 2022; Kim ve ark, 2013; Samadzadeh ve ark, 2021). **Şekil 2.**' de kanser tedavisinde kullanılan nanoliflerin uygulamaları verilmiştir.



Şekil 2. Elektro eğrilmiş nanoliflerin kanser tedavisi uygulamaları

Melanom kanseri apoptozunu uyarmak için Kim ve ark. dış bir manyetik alandan yararlanarak eş zamanlı olarak ısı üreten ve buna bağlı olarak ilaç salımı gerçekleştiren bir malzeme tasarlamışlardır. Elektroeğirme yöntemiyle sentezlenen malzeme, sıcaklığa duyarlı bir polimer, manyetik partiküller ve bir antikanser ilacı (doksorubisin) içermektedir. Manyetik alan etkisiyle kendi kendine ısı üreten MNP' ler sayesinde sıcaklığa duyarlı polimer şişmekte ve ilaç salımı gerçekleşmektedir. Rapora göre insan melanom hücrelerinin %70'i ısı ve ilacın çifte etkisiyle 5 dakika içinde ölmüştür (Kim ve ark, 2013). Başka bir çalışmada hipertermi ve kemoterapinin birlikte uygulaması için dış bir manyetik alan yardımıyla kanser bölgesinde hem pH' a bağlı ilaç salımı hemde ısı ile kanser hücrelerini öldürcek bir yöntem önerilmiştir. Elektroeğirme yoluyla sentezlenen katekolik manyetik nanoliflere antikanser ilacı Bortezomib (BTZ) bağlandı. Nanoliflerin katekol kısımları ile BTZ'nin borat kısımları pH değişimleri ile bağlanma ve salınma gerçekleştirdi. Bununla birlikte hipertermi tedavileri için demir oksit nanopartiküller kullanıldı. Yapılan in vitro çalışmalar, katekolik manyetik nanoliflerin mükemmel bir antikanser etkisi sergilediğini göstermiştir (GhavamiNejad ve ark, 2015). Bortezomib (BTZ) antikanser ilacının kullanıldığı başka bir çalışmada elektroeğirme yöntemiyle sentezlenen ilaç yüklü manyetik nanoliflerin, manyetik alan ile hem hipertermi uygulaması hem de sinerjistik kanser tedavisini mümkün kılmak için kanser hücresine özgü ilaç salımında kullanılabilir olduğu bildirildi (Sasikala ve ark, 2016). Başka bir hipertermi uygulamıyla birlikte kontrollü ve sürekli ilaç salım uygulamısında denemek üzere; Akıllı nanolifli yapı iskeleleri, demir oksit (II, III) manyetik nanoparçacıklar, metformin ve gözenekli silika nanoparçacıklar ile kompozit oluşturulmuş, sıcaklığa duyarlı bir kopolimerin elektroeğrilmesiyle üretilmiştir. Sentezlenen ilaç yüklü manyetik nanoliflerin, cilt kanseri tedavileri için ısı ve ilaç salımının birlikte sinerjistik etkisi olduğu bildirilmiştir (Samadzadeh ve ark, 2021). Meme kanseri tedavisinde kullanılan Doksorubisinin elektroeğirme tekniği ile sentezlenen membranlarda pH ve redoks uyaranlarına göre salım profilleri incelendi. Elektroeğrilmiş membran asidik ve indirgeyici ortamda uyarılan ikili kontrollü salım göstermiş, meme kanseri hücre dizilerinde ciddi sitotoksik etki yaratmıştır (Federico ve ark, 2022).

Sonuç ve gelecek persfektifler

Nanoteknoloji, klinik bilimlere destek olan nanofiberlerle kontrollü ilaç salımı gibi konularda umut verici bir yaklaşım sunmaktadır. Nanofiberlerin, sürekli ilaç salımı göstererek ve hedef bölgeye yönlendirilebilir şekilde tasarlanabilmeleri saayesinde kanser hücrelerine karşı daha iyi performans göstermesi bu alandaki çalışmaların daha çok artmasını sağlacaktır. İlaç ve polimer seçimi, sürekli ilaç salımı için doğru kombinasyonun belirlenmesi açısından önemlidir. Manyetik özellikte nanolifler

üretilebilmesi, çeşitli uyaranlara tepki veren polimer seçme imkanı, bu alandaki çalışma pergeleni oldukça genişletmektedir. Nanofiberler, antikanser ilaçlarını ve doğal özleri kapsülleyebilir. Manyetik duyarlı nanofiberler ya da çoklu uyaranlara duyarlı nanofiberler tek tip ilaç yüklü nanofiberlere göre tercih edilebilir ve daha etkili bir kanser tedavisi sağlayabilir. Ancak, ilaç salımının hedeflenen şekilde sağlanması ve metastatik kanser hücrelerinin tedavisi için daha fazla çalışma gerekmektedir. Bu nedenle, bu nanofiberlerin ticarileştirilmesi için daha fazla araştırma yapılmalıdır.

KAYNAKLAR

American Cancer Society, "Types of Cancer Treatment." http://www.cancer.org/treatment/treatmentsandsideeffects/treatmenttypes/treatment-types-landing. [Erişim: 27 Eylül 2023].

Contreras-Cáceres R, Cabeza L, Perazzoli G, Díaz A, López-Romero JM, Melguizo C, Prados J 2019. Electrospun Nanofibers: Recent Applications in Drug Delivery and Cancer Therapy. Nanomaterials. 9, 656. https://doi.org/10.3390/nano9040656

Federico S, Martorana A, Pitarresi G, Palumbo FS, Fiorica C, Giammona G 2022. Development of stimulus-sensitive electrospun membranes based on novel biodegradable segmented polyurethane as triggered delivery system for doxorubicin. Biomater. Adv. Article 212769

GhavamiNejad A, Sasikala ARK, Unnithan AR, Thomas RG, Jeong YY, Vatankhah-Varnoosfaderani M, Stadler FJ, Park CH and Kim CS 2015. Mussel-Inspired Electrospun Smart Magnetic Nanofibers for Hyperthermic Chemotherapy. Adv. Funct. Mater., 25: 2867-2875.

Hadi F, Tavakkol S, Laurent S, Pirhajati V, Mahdavi SR, Neshastehriz A, Zadeh-Shakeri A 2019. Combinatorial effects of radiofrequency hyperthermia and radiotherapy in the presence of magneto-plasmonic nanoparticles on MCF-7 breast cancer cells. J Cell Physiol. 234: 20028– 20035. https://doi.org/10.1002/jcp.28599

Han H, Li XL 2011. "Multi-resolution independent component analysis for high-performance tumor classification and biomarker discovery," BMC bioinformatics, 12, 1, 1-14, doi:10.1186/1471-2105-12-S1-S7.

Hervault A, Kim Thanh, NT 2014. Magnetic nanoparticle-based therapeutic agents for thermo-chemotherapy treatment of cancer. Nanoscale. 6:11553.

Huang C, Soenen S.J Rejman J, Trekker J, Chengxun L, Lagae L, Ceelen W, Wilhelm C, Demeester J, and De Smedt SC 2012. Magnetic Electrospun Fibers for Cancer Therapy. Adv. Funct. Mater., 22: 2479-2486. https://doi.org/10.1002/adfm.201102171

Hu PY, Zhao YT, Zhang J, Yu SX, Yan JS, Wang XX, Hu MH, Xiang HF, Long YZ 2020. In situ melt electrospun polycaprolactone/Fe3O4 nanofibers for magnetic hyperthermia. Mater. Sci. Eng. C , 110, 110708.

Kanmaz D, Toprakci HAK, Olmez H, Toprakci O 2018. Electrospun Polylactic Acid Based Nanofibers for Biomedical Applications. Mat.Sci.Res.India;15(3).

Kim YJ, Ebara M, and Aoyagi T 2013. A Smart Hyperthermia Nanofiber with Switchable Drug Release for Inducing Cancer Apoptosis. Adv. Funct. Mater., 23: 5753-5761.

Lee JS and Kim SH 2021. Evaluation of Fe_3O_4/PAN Magnetic Nanofibrous Membrane for Heat Generation and Magnetic Actuation. in IEEE Access, vol. 9, pp. 77009-77016, , doi: 10.1109/ACCESS.2021.3081151.

Leung V, and Ko F 2011. Biomedical applications of nanofibers. Polym. Adv. Technol., 22: 350-365. https://doi.org/10.1002/pat.1813

Li D, Wang Y, Xia Y 2004. Elecrospinning Nanofibers as Uniaxially Aligned Arrays and Layer by Layer Stacked Films, Advenced Materials, 16, No.4.

Li L, Hao R, Qin Song, J Chen, X Rao, F Zhai, J Zhao, Y Zhang, L Xue J 2022. Electrospun Fibers Control Drug Delivery for Tissue Regeneration and Cancer Therapy. Adv. Fiber Mater. 4, 1375–1413 https://doi.org/10.1007/s42765-022-00198-9

Maeng JH, Lee DH, Jung KH, Bae YH, Park IS, Jeong S, Hong SS 2010. "Multifunctional doxorubicin loaded superparamagnetic iron oxide nanoparticles for chemotherapy and magnetic resonance imaging in liver cancer," Biomaterials, 31, 18, 4995- 5006. doi: 10.1016/j.biomaterials.2010.02.068.

Mamun A, Sabantina L 2023. Electrospun Magnetic Nanofiber Mats for Magnetic Hyperthermia in Cancer Treatment Applications—Technology, Mechanism, and Materials. Polymers 15, 1902.

Matos RJR, Chaparro CIP, Silva JC, Valente MA, Borges JP, Soares PIP 2018. Electrospun composite cellulose acetate/iron oxide nanoparticles non-woven membranes for magnetic hyperthermia applications. Carbohydr. Polym. 198, 9–16.

Mokhosi SR, Mdlalose W, Nhlapo A, Singh M 2022. Advances in the Synthesis and Application of Magnetic Ferrite Nanoparticles for Cancer Therapy. Pharmaceutics 14, 937.

Moroz P, Jones SK and Gray BN 2002. Tumor response to arterial embolization hyperthermia and direct injection hyperthermia in a rabbit liver tumor model. J. Surg. Oncol. 80: 149-156. https://doi.org/10.1002/jso.10118

Park CH, Kang SJ, Tijing LD, Pant HR, Kim JS 2013. Inductive heating of electrospun Fe2O3/polyurethane composite mat under high-frequency magnetic field. Ceram. Int. , 39, 9785–9790.

Patel PR, Gundloori RVN 2023. A review on electrospun nanofibers for multiple biomedical applications. Polym Adv Technol. 34(1): 44- 63. doi:10.1002/pat.5896

Radmansouri M, Bahmani E, Sarikhani E, Rahmani K, Sharifianjazi F, Irani M 2018. Doxorubicin hydrochloride-Loaded electrospun chitosan/cobalt ferrite/titanium oxide nanofibers for hyperthermic tumor cell treatment and controlled drug release, International Journal of Biological Macromolecules, 116, pp. 378-384.

Samadzadeh S, Babazadeh M, Zarghami N, Pilehvar-Soltanahmadi Y, Mousazadeh H 2021. An implantable smart hyperthermia nanofiber with switchable, controlled and sustained drug release: possible application in prevention of cancer local recurrence. Mat Sci Eng C-Mater. 118:111384.

Sarier N, Onder E, Carvalho MD, Ferreira LP, Cruz MM, Arat R 2018. Preparation of magnetite nanoparticle and fatty acid incorporated poly(methacrylic acid-ethyl acrylate) nanowebs via electrospinning for magnetic hyperthermia application. IOP Conf. Ser. Mater. Sci. Eng. 460, 012025.

Sasikala ARK, Unnithan AR, Yun YH, Park CH, Kim CS 2016. An implantable smart magnetic nanofiber device for endoscopic hyperthermia treatment and tumor-triggered controlled drug release. Acta Biomater. 31:122.

Schneider-Futschik E.K, Reyes-Ortega F 2021. Advantages and Disadvantages of Using Magnetic Nanoparticles for the Treatment of Complicated Ocular Disorders. Pharmaceutics , 13, 1157.

Song C, Wang XX, Zhang J, Nie GD, Luo WL, Fu J, Seeram R, Long YZ 2018. Electric Field-Assisted In Situ Precise Deposition of Electrospun γ-Fe2O3/Polyurethane Nanofibers for Magnetic Hyperthermia. Nanoscale Res Lett. 13, 273 https://doi.org/10.1186/s11671-018-2707-y

Wang L, Wang M, Topham PD, Huang Y 2012. Fabrication of magnetic drug-loaded polymeric composite nanofibres and their drug release characteristics. RSC Adv. 2(6): 2433- 2438.

Wei W, Zarghami N, Abasi M, Ertas YN, Pilehvar Y 2022. Implantable magnetic nanofibers with ON–OFF switchable release of curcumin for possible local hyperthermic chemotherapy of melanoma. J Biomed Mater Res. 110 (4): 851- 860. doi:10.1002/jbm.a.37333

Wenhua H, Qingxuan L, Yan L, Dingyue J, Yu G, Kai Z, Chuanming D 2022. Cancer Treatment Evolution from Traditional Methods to Stem Cells and Gene Therapy. Bentham Science Publishers. 22(5): s. 368-385(18), https://doi.org/10.2174/1566523221666211119110755

Xue JJ, Wu T, Dai YQ, Xia YN 2019. "Electrospinning and electrospun nanofibers: Methods, materials, and applications." Chemical reviews, 119, 8, 5298-5415, doi:10.1021/acs.chemrev.8b00593

Yıldız A, Kara AA, Acartürk F. 2020. Peptide-protein based nanofibers in pharmaceutical and biomedical applications. International Journal of Biological Macromolecules. 148, p. 1084-1097.

Zhao J, Cui W. 2020. Functional Electrospun Fibers for Local Therapy of Cancer. Adv. Fiber Mater. 2, 229–245, https://doi.org/10.1007/s42765-020-00053-9

Zugazagoitia J, Guedes C, Ponce S, Ferrer I, Molina-Pinelo S, Paz-Ares L 2016. Current challenges in cancer treatment. Clin. Ther., 38, pp. 1551-1566, 10.1016/j.clinthera.2016.03.026

ORAL PRESENTATION

Comparison of propofol alone and different doses of ketofol combinations on systolic, diastolic, and mean arterial pressures in dogs

Mümin Gökhan Şenocak[*] (*ORCID: https://orcid.org/0000-0002-8855-8847*, Latif Emrah Yanmaz[2] (*ORCID: https://orcid.org/0000-0001-5890-8271*)

[*1]Atatürk University, Faculty of Veterinary Medicine, Department of Surgery, Erzurum, Türkiye
[2] Burdur Mehmet Akif Ersoy University, Faculty of Veterinary Medicine, Department of Surgery, Burdur, Türkiye

*Corresponding author e-mail: mumingokhan@gmail.com

Abstract

This study aimed to compare the effects of propofol, either alone or in combination with varying doses of ketamine, on systolic, diastolic, and mean arterial blood pressure in unpremedicated dogs. Twenty-eight crossbred dogs were randomly divided into four groups: a Propofol-only group and three Ketofol groups with ketamine ratios of 1:1, 1:2, and 1:3, administered intravenously at a constant rate of 0.6 mg/kg/minute. Noninvasive blood pressure measurements were taken every five minutes for 60 minutes. The results indicated that the Propofol group showed no significant change in systolic pressure over time ($p > 0.05$). However, diastolic and mean arterial pressures decreased at various intervals. In contrast, the Ketofol 1:1 combination led to a consistent increase in systolic, diastolic, and mean arterial blood pressure over time ($p < 0.05$). The Ketofol 1:2 combination did not significantly affect systolic and mean blood pressure ($p > 0.05$) but increased diastolic pressure at various intervals. The Ketofol 1:3 combination did not significantly alter systolic and mean blood pressures but raised diastolic pressure; however, it proved inadequate in achieving an optimal plane of anesthesia, leading to the appearance of withdrawal reflex. Blood pressure changes were related to the varying ketamine amounts in the mixture. In conclusion, propofol reduced diastolic and mean arterial pressure, whereas its combination with ketamine resulted in a significant increase in systolic, diastolic, and mean arterial pressure in unpremedicated healthy dogs. Among the Ketofol combinations, the 1:2 ratio provided the closest approximation to initial blood pressure levels when compared to the 1:1 and 1:3 combinations. The Ketofol 1:3 combination could be misleading for blood pressure determination due to its inadequacy for achieving optimal anesthesia. The addition of ketamine to the propofol mixture can help balance blood pressure during propofol anesthesia.

Keywords: anesthesia, blood pressure, dogs, ketamine, ketofol, propofol.

INTRODUCTION

Anesthesia can influence blood vessel tone, potentially leading to fluctuations in blood pressure. Effective blood pressure management is crucial for maintaining the proper functioning of these organs. Anesthesia can induce a drop in blood pressure, a condition referred to as hypotension. Hypotension can diminish the blood flow to vital organs, possibly resulting in organ dysfunction or damage. Conversely, excessively high blood pressure, referred to as hypertension, during anesthesia can be problematic as well. It heightens the risk of bleeding, blood vessel damage, and other complications. (Den Boer et al. 1993; Akata 2007; Shi and Rodríguez-Contreras 2022)

Inadequate management of blood pressure during anesthesia in dogs can lead to a variety of complications, including cardiovascular issues, organ dysfunction, and an elevated risk of adverse events during surgery. During surgery, blood pressure serves as one of the continuously monitored vital signs. Surgery can elevate the body's demand for oxygen and nutrients due to factors such as pain, surgical trauma, and changes in body position. It acts as an indicator of the patient's response to anesthesia. Sudden shifts in blood pressure can signal potential issues such as anesthetic overdose, inadequate anesthesia, or other complications, enabling anesthetist to make timely adjustments.(Mazzaferro and Wagner 2001; Hughes 2008)

Different anesthesia drugs and techniques can impact blood pressure differently. Propofol, a common intravenous anesthetic in dogs, can cause a temporary drop in blood pressure due to its vasodilatory effect. Ketamine, often used in canine anesthesia, stimulates the sympathetic nervous system, generally increasing blood pressure. Ketamine is commonly used in combination with other agents for balanced anesthesia. When ketamine and propofol are combined for anesthesia in dogs, their effects on blood pressure are influenced by various factors, including dosage, rate of administration, individual patient characteristics, and the presence of other medications.(Haskins et al. 1986; Cullen 1996; Hughes 2008)

The objective of this study was to test the following null hypothesis: The administration of propofol alone or ketamine-propofol ratios at 1:1, 1:2, and 1:3 will not result in a change in systolic, diastolic and mean arterial pressure in unpremedicated dogs.

MATERIALS AND METHODS

The study was approved by the Atatürk University Ethics Committee for Animal Experiments (Approval No. 2015/6/119) and conducted at the Atatürk University Veterinary Teaching Hospital with privately owned male dogs brought in for castration surgery. The owners of the dogs provided written informed consent for their participation.

The animals were housed individually, and they were fasted for a minimum of 12 hours prior to anesthesia, whereas their water consumption was not restricted.

Animals

Twenty-eight male dogs of mixed breeds were enrolled in the investigation. The inclusion criteria included canines aged 1 to 6 years without a history of systemic disease and a clean clinical examination, complete blood profile, and serum chemistry analysis. The physical status score of the canines according to the American Society of Anesthesiologists was I, and the body condition score was 4 or 5 out of 9.

Drugs

Propofol (20 mg/mL, Fresenius Kabi, Graz, Austria) and ketamine (100 mg/mL, Ketasol, Interhas, Richter Pharma AG, Wels, Austria) were used in the current study. This mixture was manufactured daily to ensure that it was sterile and aseptic. Before administration, the admixture was combined into a single infusion container for each subject. Three distinct propofol and ketamine combinations were used for constant rate infusion (CRI): Ketofol 1:1, Ketofol 1:2, and Ketofol 1:3. The Ketofol 1:1 mixture contained 10 ml (1000 mg) of ketamine and 50 ml (1000 mg) of propofol, whereas the Ketofol 1:2 mixture contained 10 ml (1000 mg) of ketamine and 100 ml (2000 mg) of propofol. The Ketofol 1:3 mixture included 10 ml (1000 mg) of ketamine and 150 ml (3000 mg) of propofol.

Study design

The research was a longitudinal experimental clinical trial. Using a random drawing, dogs were assigned to anesthesia treatment groups.

The dogs were divided into four groups, each containing seven animals, and designated as group P, group Ketofol 1:1, group Ketofol 1:2, and group Ketofol 1:3. Each group received the assigned treatment for both induction and maintenance of anesthesia, with no pre-anesthetic medications being administered. The loading dose for induction of anesthesia was injected into the previously catheterized right cephalic vein at a rate of one milliliter every 15 seconds until jaw relaxation was observed, after which the dog was sternally positioned and intubated. Dogs breathed room air spontaneously.

Utilizing a volumetric infusion device (Birset IV Master, Birtan Medical, Ankara, Turkey), anesthesia was administered. Before the procedure, the IV infusion hose set was verified on the volumetric pump, and the same brand of IV set was used throughout the entire study. (Senocak et al. 2021) The total volume of substance in each group's mixture was administered as a CRI at a rate of 0.6 mg/kg/minute for sixty minutes. During the anesthesia, the pedal reflex was continuously evaluated by compressing the hindlimb pulvinus at one-minute intervals to determine depth of anesthesia.

Patient monitoring

Vital sign data was collected utilizing the Veterinary Patient Monitor (MVM Medical, GT9000F, Istanbul, Turkey) to track the animal's vital signs. To ascertain the systolic arterial pressure, diastolic arterial pressure, and mean arterial pressure, a blood pressure cuff with a circumference equivalent to 40% of the right pelvic limb circumference was placed proximal to the tarsus. (Şenocak 2023)

To obtain the respiratory frequency, and heart rate, electrocardiographic leads were attached to the extremities. A rectal probe was inserted into the rectum in order to measure rectal temperature. The end-tidal carbon dioxide tension adapter was also attached to the endotracheal tube connector for capnography. Manual ventilation was provided using an ambu bag when the patient's saturation level fell below 92%.

The systolic, diastolic, and mean arterial pressure data were recorded at prior to the study as baseline (BL), following intubation (T0), and every five minutes up to 60 minutes. When the swallowing reflex was observed, the dog was positioned on its left side and extubated after the final measurement at T60.

The surgical stimulus due to the open orchiectomy was started at T10 after induction of anesthesia.

Statistical analysis

The data's normality was assessed with the Shapiro-Wilk test, and variance homogeneity was checked using the Levene test. To assess the overall significance of treatment differences, a mixed ANOVA was conducted, as indicated by the omnibus p-value. For comparisons between the BL and subsequent time points (with time as the dependent variable), a repeated measures one-way ANOVA was performed, followed by post-hoc correction using the LSD method. In cases where Mauchly's Test of Sphericity was inconclusive, the degrees of freedom were adjusted using a Greenhouse-Geisser correction.

Blood pressure data from different groups underwent Univariate ANOVA for analysis, and the Post-Hoc LSD multiple comparison test was applied to assess pairwise differences.

In the study, parametric data were represented as mean values with standard deviations. The differences in means between different treatments were displayed as mean differences along with their corresponding 95% confidence intervals, showing the range from the lower to the upper bound of the CI.

We established a significance alpha level of 0.05. The statistical analyses were carried out using SPSS software, specifically Version 22.

RESULTS

In the investigation, a total of twenty-eight healthy dogs were included. Nonetheless, one animal from Group Ketofol 1:1 was excluded from the investigation due to apneic episodes. The animal recovered from anesthesia without any complications after receiving oxygen support via a positive-pressure ventilator. The study was conducted with six dogs in this group. During the investigation hypotension (mean arterial blood pressure < 60 mmHg) was not observed.

The dogs included in the study had an average age of 2.9 ± 1.1 years, and their average weight was 17.6 ± 6.4 kg.

Throughout the maintenance of anesthesia, two dogs in the Ketofol 1:3 treatment group exhibited a slight pedal reflex due to the light plane of anesthesia.

Propofol group had no significant change in systolic pressure compared to BL over time ($p > 0.05$). However, it was observed that diastolic pressure was decreased at T10 with a mean difference of 23.14 mmHg (95% CI 7.59-38.69, p=0.011), T15 with a mean difference of 26.29 mmHg (95% CI 9.56-43, p=0.008), T20 with a mean difference of 24.43 mmHg (95%CI 7-41.85, p=0.014), T25 with a mean difference of 19 mmHg (95% CI 4.25-33.75, p=0.02), and T30 with a mean difference of 19 mmHg (95% 4.55-33.45, p=0.018) compared to the BL. In addition, mean arterial pressure was decreased at T5 with a mean difference of 20 mmHg (95% CI 9.73-30.27, p=0.003), T10 with a mean difference of 16.7 mmHg (95% CI 2.48-30.95, p=0.028), T15 with a mean difference of 19.43 mmHg (95% CI 3.84-35, p=0.022), T20 with a mean difference of 18.29 mmHg (95% CI 1.28-35.29, p=0.039), T30 with a mean difference of 12.57 mmHg (95% CI 0.7-24.44, p=0.041) compared to the BL.

Ketofol 1:1 combination increased systolic blood pressure at T55 with a mean difference of 10 mmHg (95% CI 3.17-16.8, p=0.013), diastolic blood pressure at T50 with a mean difference of 21.83 mmHg (1.47-42.2, p=0.04), and mean arterial pressure at T25 with a mean difference of 19.83 mmHg (95% CI 2.8-36.8,p=0.03),

T30 with a mean difference of 20.3 (95%CI 3.2-37.4, p=0.028), T35 with a mean difference of 23 mmHg (95%CI 2.68-43.3, p=0.033), T40 with a mean difference of 24.33 mmHg (95% CI 3.36-45.31, p=0.031), T45 with a mean difference of 21.5 mmHg (95%CI 2.33-40.67, p=0.034), T50 with a mean difference of 24 mmHg (95% CI 5.76-42.28, p=0.02), T55 with a mean difference of 24 mmHg (95% CI 5.2-42.8, p=0.022), T60 with a mean difference of 22.5 mmHg (95%CI 2.8-42.16, p=0.032) compared to BL.

Ketofol 1:2 combination was not changed systolic and means blood pressure significantly ($p > 0.05$). However, it was increased diastolic pressure at T45 with a mean difference of 15.43 mmHg (95% CI 4.1-26.79, p=0.016), T50 with a mean difference of 16.57 mmHg (95% CI 5.77-27.34, p=0.009), T55 with a mean difference of 15.14 mmHg (95% CI 5.28-25.1, p=0.009), and T60 with a mean difference of 23.29 mmHg (95% CI 4.39-42.18, p=0.024) compared to the BL.

Ketofol 1:3 combination was not changed systolic and mean blood pressures significantly ($p > 0.05$) However, it was increased diastolic blood pressure T5 with a mean difference of 21.43 mmHg (95% CI 2.8-40.1, p=0.031), T10 with a mean difference of 24.57 mmHg (95% CI 4.9-44.2, p=0.022), T15 with a mean difference of 24.43 mmHg (95% CI 4.72-44.13, p=0.023), T20 with a mean difference of 27.43 mmHg (95% CI 4.15-50.7, p=0.028), T30 with a mean difference of 33.57 mmHg (95% CI 12.44-54.7, p=0.008), T35 with a mean difference of 33.43 mmHg (95% CI 13.2-53.67, p=0.007), T40 with a mean difference of 32.43 mmHg (95% CI 12.03-52.82, p=0.008), T45 with a mean difference of 33.86 mmHg (95% CI 15.84-51.87, p=0.004), T50 with a mean difference of 32.57 mmHg (95% CI 12.53-52.61, p=0.007), T55 with a mean difference of 32.57 mmHg (95% CI 11.89-53.25, p=0.008), and T60 with a mean difference of 33 mmHg (95% CI 10.66-55.34, p= 0.011) compared to the BL

In overall consideration, systolic blood pressure was significantly higher in Ketofol 1:3 group (146.15 ± 7.91 mmHg), Ketofol 1:1 (144.37 ± 5.57 mmHg), and Ketofol 1:2 (142.61 ± 7.24 mmHg) compared to Propofol group (136.59 ± 13.24 mmHg), ($p < 0.01$). In addition, systolic blood pressure was significantly ($p < 0.01$) higher in Ketofol 1:1 group compared to the Ketofol 1:3. It was also significantly (p=0.007) higher in Ketofol 1:3 group compared to Ketofol 1:2 group

Diastolic blood pressure was significantly higher in Ketofol 1:3 group (110.34 ± 13.68 mmHg), Ketofol 1:1 (106.41 ± 18.14 mmHg), and Ketofol 1:2 (103.49 ± 20.37 mmHg) compared to the Propofol group (95.77 ± 23.51 mmHg), ($p < 0.05$). In addition, diastolic pressure was significantly higher in Ketofol 1:3 group compared to the Ketofol 1:2 ($p < 0.01$).

Mean arterial blood pressure was significantly higher in Ketofol 1:3 (125.93 ± 12.23 mmHg), Ketofol 1:1 (125.91 ± 11.93 mmHg), and Ketofol 1:2 (121.96 ± 15.79 mmHg) compared to the Propofol group (113.01 ± 21.44 mmHg), ($p < 0.01$).

DISCUSSION

This study has provided an evidence that propofol administration is associated with a reduction in both diastolic and mean arterial pressure. Conversely, when propofol is administered in combination with ketamine, a significant improvement is observed in systolic, diastolic, and mean arterial pressure among unpremedicated, healthy dogs. It is noteworthy that variations in the ratios of propofol and ketamine in the mixture can lead to diverse magnitudes of blood pressure fluctuations. Specifically, the utilization of Ketofol at a 1:3 ratio may be insufficient for achieving the requisite level of anesthesia, potentially resulting in the acquisition of erroneous blood pressure data. The incorporation of ketamine into the admixture exerts a discernible influence on the elevation of blood pressure, and the administration of Ketofol at a 1:2 ratio appears to offer the most precise approximation to the baseline blood pressure values.

Multiple techniques exist for assessing arterial blood pressure in dogs, including invasive approaches like direct arterial puncture, as well as noninvasive methods such as oscillometry and Doppler ultrasonography, typically employed in clinical settings. It is noteworthy that both invasive and noninvasive means of measuring systolic, diastolic, and mean arterial pressure have demonstrated comparable results (Stepien and Rapoport 1999) In the current investigation, we opted for a noninvasive approach to blood pressure measurement utilizing oscillometry. This method involved the use of a blood pressure cuff with a width equivalent to 40% of the circumference of the right pelvic limb. The cuff was positioned proximal to the tarsus to facilitate the noninvasive assessment of blood pressure.

Certain premedications, particularly alpha2 agonists, can exhibit an initial surge in arterial pressure, succeeded by a more protracted but mild decrease in blood pressure. (Vainio 1989) This phenomenon associated with premedication has the potential to obscure the primary drug's primary impact on blood pressure. In the current research, we deliberately abstained from using any premedication agents to isolate and elucidate the inherent effects of the primary drugs on blood pressure. We selected propofol and ketamine for induction of anesthesia without the use of premedication. This combination is widely employed in anesthesia practices as a common choice. (Dogan et al. 2016; Yanmaz et al. 2020)

Blood pressure monitoring during the course of anesthesia serves as a valuable indicator of the anesthesia depth and provides insights into the potential presence of pain. (Saccò et al. 2013) Particularly, when confronted with surgical stimuli, deviations of 20% or more in blood pressure are conventionally regarded as indicative of a light plane of anesthesia or the potential existence of pain. (Şenocak 2023) In the context of our current investigation, it was observed that various combinations of propofol and ketamine at differing ratios elicited distinct alterations in systolic, diastolic, and mean arterial pressure. This variability may be attributed to the varying levels of ketamine within the administered mixtures (Smischney et al. 2012; Ghadami Yazdi et al. 2013)

Elevations in blood pressure can be attributed to various factors, including the white coat effect, anxiety, alertness, defensive responses, and the experience of pain. (Popilskis et al. 1993; Soares et al. 2012). Specifically, during surgical procedures, an increase in blood pressure is indicative of the dog perceiving a painful stimulus. Consequently, the monitoring of systolic blood pressure throughout surgery serves as a critical parameter for assessing the depth of anesthesia (Popilskis et al. 1993). However, it remains unclear whether the rise in blood pressure is primarily associated with the painful stimuli or is influenced by the pharmacological effects of administered drugs. In our study, surgical stimuli were introduced at the T10 level. Although systolic blood pressure did not exhibit significant changes, it did show a suspicious decrease. Notably, diastolic pressure and mean arterial pressure experienced a decrease within the initial thirty minutes, even at the T5 level of surgery. This observation suggests that this decrease may be attributed to both the chemical and physiological effects of propofol or the profound depression of the central nervous system when compared to the combination of propofol with ketamine. Alternatively, it could be related to the sympathomimetic effects of ketamine, stemming from increased monoamine release and the inhibition of presynaptic catecholamine reuptake. (Tso et al. 2004)

While propofol typically leads to a reduction in blood pressure, its combination with ketamine has been found to result in an elevation of blood pressure. This increase in blood pressure, particularly notable when using a 1:1 ratio of ketofol (ketamine combined with propofol), primarily manifested after the 25th minute and was particularly prominent in mean arterial pressure. The observation of withdrawal reflex in the group receiving propofol combined with ketamine at a 1:3 ratio led to the inference that this particular combination may not be adequate for achieving a sufficiently deep state of anesthesia. Consequently, the rise in blood pressure within this group could be attributed to the presence of consciousness. In contrast, when propofol was combined with ketamine at a 1:2 ratio, blood pressure remained relatively stable. Therefore, this particular combination warrants broader consideration and evaluation for its potential efficacy.

CONCLUSION

In conclusion, this study has provided the hemodynamic effects of propofol and its combination with ketamine in unpremedicated healthy dogs. Propofol alone is associated with a reduction in both diastolic and mean arterial pressure, whereas the combination with ketamine results in a significant increase in systolic, diastolic, and mean arterial pressure. Importantly, the observed alterations in blood pressure are contingent upon the specific ratios of propofol and ketamine administered. Notably, the use of Ketofol at a 1:3 ratio may prove insufficient for achieving adequate anesthesia, potentially leading to erroneous blood pressure measurements could be attributed to the presence of consciousness. Conversely, employing Ketofol at a 1:2 ratio appears to provide the closest approximation to the baseline blood pressure levels. These findings underscore the critical role of drug ratios in managing anesthesia and monitoring hemodynamic stability in dogs.

ACKNOWLEDGEMENTS

This research project was funded by the Scientific and Technological Research Council of Turkey (TÜBİTAK, Project No: 215S062). This study is based on the PhD thesis authored by Mümin Gökhan Şenocak.

REFERENCES

Akata T 2007. General anesthetics and vascular smooth muscle: Direct actions of general anesthetics on cellular mechanisms regulating vascular tone. Anesthesiology, 106(2): 365–391.

Den Boer MO, Van Woerkens LJ, Somers JAE, Duncker DJ, Lachmann B, Saxena PR, Verdouw PD 1993. On the preservation and regulation of vascular tone in arteriovenous anastomoses during anesthesia. Journal of Applied Physiology, 75(2): 782–789.

Cullen LK 1996. Medetomidine sedation in dogs and cats: A review of its pharmacology, antagonism and dose. British Veterinary Journal, 152(5): 519–535.

Dogan E, Yanmaz LE, Senocak MG, Okumus Z 2016. Comparison of propofol, ketamine and ketofol on intraocular pressure in New Zealand white rabbits. The Revue de Médecine Vétérinaire (Toulouse). 167(2), 18–21.

Ghadami Yazdi A, Ayatollahi V, Hashemi A, Behdad S, Ghadami Yazdi E 2013. Effect of two Different Concentrations of Propofol and Ketamine Combinations (Ketofol) in Pediatric Patients under Lumbar Puncture or Bone Marrow Aspiration. Iranian Journal of Pediatric Hematology and Oncology. 3(1): 187–92.

Haskins SC, Patz JD, Farver TB 1986. Xylazine and xylazine-ketamine in dogs. American Journal of Veterinary Research, 47(3): 636–641.

Hughes JML 2008. Anaesthesia for the geriatric dog and cat. Irish. Veterinary Journal, 61(1), 380–387.

Mazzaferro E, Wagner AE 2001. Hypotension during Anesthesia in Dogs and Cats: Recognition, Causes, and Treatment. Compendium on Continuing Education for the Practicing Veterinarian, 23(8): 728–736.

Popilskis S, Kohn DF, Laurent L, Danilo P 1993. Efficacy of Epidural Morphine Versus Intravenous Morphine for Post-Thoractotomy Pain in Dogs. Veterinary Anaesthesia and Analgesia, 20(1): 21–25.

Saccò M, Meschi M, Regolisti G, Detrenis S, Bianchi L, Bertorelli M, Pioli S, Magnano A, Spagnoli F, Giuri PG, Fiaccadori E, Caiazza A 2013. The relationship between blood pressure and pain. Journal of Clinical Hypertension, 15(8): 600–605.

Şenocak MG 2023. Comparison of spermatic cord ligation and the vas plexus ligation in canine orchiectomy: A prospective clinical study. Veterinary Medicine and Science 9(5): 2015–2021.

Şenocak MG, Yanmaz LE, Ersoz U, Okur S, Turgut F 2021. The effects of randomly selected IV serum sets on constant rate infusion: in-vitro demonstration. Atatürk University Journal of Veterinary Sciences 16(3): 330–335.

Shi L, Rodríguez-Contreras A 2022. The general anesthetic isoflurane inhibits calcium activity in cerebrovascular endothelial cells and disrupts vascular tone. bioRxiv 03(25): 485881.

Smischney NJ, Beach ML, Loftus RW, Dodds TM, Koff MD 2012. Ketamine/propofol admixture (ketofol) is associated with improved hemodynamics as an induction agent: A randomized, controlled trial. Journal of Trauma and Acute Care Surgery. 73(1): 94–101.

Soares FAC, Neuwald EB, Mombach VS, D'Avila AER, Conrado F de O, González FHD 2012. Systolic blood pressure of dogs at hospital and domestic environment. Ciencia Rural 42(7): 1243–1248.

Stepien RL, Rapoport GS 1999. Clinical comparison of three methods to measure blood pressure in nonsedated dogs. Journal of the American Veterinary Medical Association 215(11): 1623–1628.

Tso MM, Blatchford KL, Callado LF, McLaughlin DP, Stamford JA 2004. Stereoselective effects of ketamine on dopamine, serotonin and noradrenaline release and uptake in rat brain slices. Neurochemistry International 44(1): 1–7.

Vainio O 1989. Introduction to the clinical pharmacology of medetomidine. Acta veterinaria Scandinavica. Supplementum 85:85–88.

Yanmaz LE, Dogan E, Senocak MG, Ersoz U, Okur S 2020. Comparison the effect of different ratios of ketamine and propofol (ketofol) admixture on anesthesia induction in New Zealand White Rabbits. Ataturk Universitesi Veteriner Bilimleri Dergisi, 15(3): 251–256.

DRAFT

ORAL PRESENTATION

Spirulina platensis ilavesi ile üretilen klasik ve probiyotik yoğurdun duyusal nitelikleri

Çağrı Güler[1] (*ORCID: https://orcid.org/0000-0001-9512-9912* ,
Ayla Arslaner[1*] (*ORCID: https://orcid.org/0000-0002-2777-9697*)

[*1] Bayburt Üniversitesi, Mühendislik Fakültesi, Gıda Mühendisliği Bölümü, Bayburt, Türkiye
*e-mail: aylaarslaner@bayburt.edu.tr

Özet

İnsan beslenmesinin vazgeçilmez bir parçası olan ve günlük olarak sıklıkla tüketilen yoğurdun zengin besin içeriği ve insan sağlığına faydaları ile öne çıkan *Spirulina platensis* ile zenginleştirilmesiyle elde edilen fonksiyonel ürünün duyusal niteliklerinin belirlendiği bu araştırmada, 4 farklı oranda (%0, %0,3, %0,6, %0,9) *Spirulina platensis* içeren 4 klasik ve 4 probiyotik olmak üzere 8 çeşit yoğurt üretilmiştir. Üretilen yoğurtların duyusal nitelikleri 4 °C'de 1, 7, 14, 21 ve 28. günlerde analiz edilmiştir. Yoğurt üretiminde kullanılan *Spirulina platensis* tozunun, yoğurt örneklerinin duyusal özellikleri üzerine olan etkisi önemli bulunmuştur. Yapılan duyusal analizler sonucunda, yoğurt örneklerinin tümü duyusal değerlendirmeler açısından 5 puan üzerinden 4-5 arasında puanlara sahip olmuşlardır. Tüm duyusal değerlendirmeler gözönünde bulundurulduğunda; %0,3 oranında *Spirulina platensis* ihtiva eden (P%0,3), *Lactobacillus acidophilus*'lu, besin değeri yüksek yeni bir fonksiyonel yoğurt üretilebileceği kannatine varılmıştır. Ayrıca panelist değerlendirmelerine göre, %0,9 *Spirulina platensis* içeren örneklerin baharatlı yoğurt sosu gibi bir ürün olarak geliştirilebileceği belirtilmiştir.

Anahtar Kelimeler: Yoğurt, *Spirulina platensis, Lactobacillus acidophilus*

Sensory properties of classical and probiotic yogurt produced adding *Spirulina platensis*

Abstract

This study aims to determine the quality characteristics of the functional product obtained by enriching the yoghurt, an essential part of the human diet and frequently consumed daily, with *Spirulina platensis*, which stands out with its rich nutritional content and benefits for human health. For this purpose, 8 types of yoghurt, 4 classical and 4 probiotics, containing *Spirulina platensis* in 4 different ratios (0%, 0.3%, 0.6%, 0.9%) were produced. The sensory characteristics of the produced yoghurts were analyzed at 4 °C on the 1st, 7th, 14th, 21st and 28th days. The effect of *Spirulina platensis* powder in yogurt production on yogurt samples' sensory properties was significant. As a result of the sensory analysis, all of the yoghurt samples had scores between 4-5 out of 5 in terms of sensory evaluations ($p<0.05$). Considering all quality evaluations, a new enriched probiotic yoghurt with high nutritional value can be produced with *Lactobacillus acidophilus* and containing 0.3% Spirulina platensis (P0.3%). In addition, according to panellist evaluations, it was stated that the samples containing 0.9% *Spirulina platensis* could be developed as a product such as spicy yogurt sauce.

Keywords: Yogurt, *Spirulina platensis, Lactobacillus acidophilus*

GİRİŞ

"Fonksiyonel Gıda" terimi; insanın metabolik fonksiyonları ve fizyolojisi için ilâve yararlar sağlayan gıda anlamına gelir. İnsan beslenmesinde ihtiyaç duyulan, temel besin maddelerinin alınması yanında, sağlıklı yaşamı desteklemede de etkileri vardır (Berner ve O'Donnell, 1998; Çakıroğlu ve Uçar, 2018). Gıdanın, fonksiyonel olarak nitelendirilebilmesi için prebiyotikler, probiyotikler ve biyoaktif bileşenleri yeterli miktarda içermesi ve tüketildiğinde beklenen faydayı sağlayabilmesi gerekir (Erbaş, 2006). Fonksiyonel gıdalar, insanın temel fizyolojisi, bağışıklık, sinir, hormon, solunum, dolaşım ve sindirim sistemlerinde olumlu etkiler oluşturarak birçok hastalığın oluşma risklerini azaltmaktadırlar (Litwin ve ark., 2018). Beslenme alışkanlıklarımız içerisinde tüketim oranı yüksek süt ve süt ürünleri, fonksiyonel bileşenler için iyi birer

taşıyıcıdırlar. Bu nedenle, fonksiyonel süt ve süt ürünleri tüketimi, bireylerin yeterli ve dengeli beslenmesine hizmet etmek yanında, gıda takviyeleri olarak günlük diyetle alımı çok tercih edilmeyen fonksiyonel bileşenler için de uygun bir kaynak oluşturmaktadır.

Lb. acidophilus ürettiği antimikrobiyal metabolitlerle ve besin bileşenlerini kullanarak patojenlerle rekabet eder ve bağırsak florasında bozulan dengeyi yeniden sağlar. Bu nedenle *Lb. acidophilus* tıp alanında ilgi duyulan bir bakteridir. Bu bakteri fekal enteropatojenik E. coli suşları ile Salmonella ve Shigella türlerine, Proteus vulgaris gibi diğer bağırsak patojenlerine karşı kuvvetli antibiyotik etki göstermesi bakımından laktik asit bakterileri arasında ayrı bir öneme sahiptir. Gastrointestinal sistemimizin doğal mikroflorasının dengesi birçok faktör nedeniyle bozulabilir. Bunlar çevre kirliliği, iklim değişiklikleri, ilaç kullanımı, yanlış beslenme ve stres gibi olumsuz faktörden oluşur. Bu etkenlere maruz kalan bireylerin, barsak sistemlerindeki dengesizlik, patojenlerin gelişimi ve beraberinde çeşitli hastalıklar için uygun zemin oluşturmaktadır. Vücutta istenilen sağlık yararının sağlanabilmesi için probiyotiklerin düzenli olarak tüketilmesi zorunludur. Tüketim esnasında ürünün probiyotik mikroorganizma sayısının en az 10^6-10^7 kob/g seviyesinde olması gerekmektedir (Kailasapathy ve ark., 2008; Çakmakçı ve ark., 2012).

S. platensis en dikkat çekici yönü, yüksek protein içeriğidir. Ayrıca esansiyel yağ asidi, aminoasit, karotenoit, vitamin ve mineral bileşimi zengindir. Yüksek verimle elde edilebilmesi ticari üretimine başlanmasında önemli faktörlerden biridir. Ulusal Havacılık ve Uzay Dairesi ile Avrupa Uzay Ajansı uzay yolculuklarında ana yiyecek olarak belirlemiştir (Asghari, Fazilati, Latifi, Salavati, & Choopani, 2016). Birleşmiş Milletler ve Dünya Sağlık Örgütü (WHO) S. platensis'i çocuklar ve yetişkinler yüksek oranda yararlı gıda desteği olarak kabul etmektedir (Michaelsen vd., 2009). 2012 yılında *S. plantesis*'in güvenilirliği birçok toksikoloji araştırmaları ile ispatlanmıştır. Amerikan Gıda ve İlaç Dairesi (Food and Drug Administration, 2012) tarafından GRAS (Generally Recognised As Safe) listesine alınmıştır. Günlük 3-10 g tüketilmesi sağlık açısından önerilmiştir (Seyidoğlu ve ark., 2017).

Bu çalışmanın amacı, diyetimizin önemli bir parçası olup günlük hayatta sık sık tüketmiş olduğumuz yoğurdun, zengin besin içeriği ve insan sağlığına olan yararları bilinen *Spirulina platensis* ile zenginleştirilerek, kalite niteliklerinin arttırılıp her yaştan tüketiciye fonksiyonel bir gıda olarak sunulabilir duruma getirmektir. Bu amaçla klasik ve probiyotik olmak üzere 2 tip (klasik ve probiyotik) yoğurda, farklı oranlarda (%0, %0,3, %0,6, %0,9) *Spirulina platensis* ilave edilerek toplamda 8 çeşit yoğurt üretilmiş, duyusal kalite nitelikleri, 4 °C'de 1, 7, 14, 21 ve 28. günlerde analiz edilmiştir.

MATERYAL VE METOT

Deneme yoğurtlarının üretimi

Yoğurt örneklerinin üretiminde kullanılacak pastörize süt, 45-50 °C'ye arasında ön ısıtmaya tabi tutulmuştur. Yağsız kurumadde oranı en az %15 olacak şekilde önceden hesaplanan oranda yağsız süt tozu (YST) ve farklı oranlarda (%0, %0,3, %0,6, %0,9) *Spirulina platensis* ilave edilmiştir. 90 °C'de 5 dk süreyle ısıl işleme tâbi tutulduktan sonra 43±2 °C'ye soğutularak bir gün önceden liyofilize *Streptococcus thermophilus* ve *Lactobacillus delbrueckii subsp. bulgaricus* ve *Lactobacillus acidophilus* LA-5® ana kültüründen hazırlanan aktive edilmiş bulk kültürler ile %2 oranında inokule edilmiştir. Yoğurt örnekleri 42±1 °C'de pH 4,7±0,1'e ulaşıncaya kadar inkübe edilmiştir. İnkübasyon tamamlandıktan sonra 4±1 °C'de 24 saat tutulmuştur. Depolamanın 1, 7, 14, 21 ve 28. günlerinde gerekli analizler yapılmıştır.

Yoğurt örneklerinde yapılan duyusal analizler

Duyusal panellerde, Bodyfelt ve ark. (1988) ve TS 1330 (2009) tarafından verilen puantaj cetveli modifiye edilerek kullanılmıştır. Bu amaçla, yoğurt örneklerinin temel bazı organoleptik nitelikleri (dış görünüş, kıvam (kaşıkla), kıvam (ağızla), koku ve tat) depolamanın 1, 7, 14, 21 ve 28. günlerinde Gıda Mühendisliği Bölümü öğretim elemanlarından ve gıda mühendislerinden oluşan 10 kişilik gönüllü panelist grup tarafından her bir parametre için 5 puan, toplam 25 puan üzerinden değerlendirilmiştir.

İstatistiksel analizler

Araştırma sonucunda edilen değerler IBM SPSS Statistic 22 paket programında varyans analizine tabi tutulmuş, analiz sonucunda önemli çıkan faktörler Duncan çoklu karşılaştırma testi ile %99 ($p<0,05$) güven düzeyinde değerlendirilmiştir (SPSS, 2022).

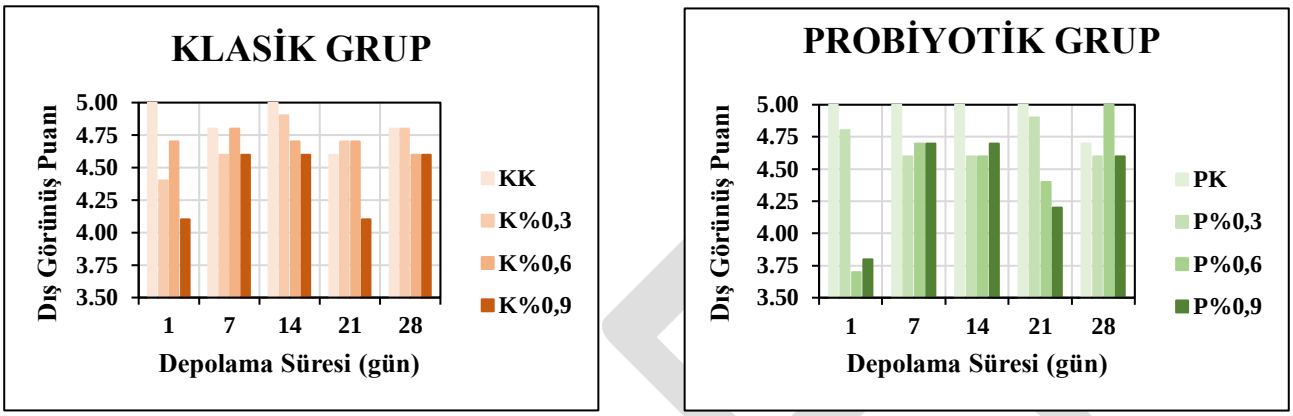
DRAFT

BULGULAR ve TARTIŞMA

Yoğurt Örneklerine Ait Duyusal Analiz Sonuçları

Dış görünüş.

Yoğurt örneklerine ait dış görünüş puanları 3,70 ile 5,00 arasındadır. Yoğurt örneklerinde en yüksek dış görünüş puanları 5,00 ile depolamanın 1, 7, 14 ve 28. günlerinde PK, 14. günde KK ve 28. günde K%0,6 örneğinde; en düşük dış görünüş puanı 3,70 ile depolamanın 1. gününde K%0,6 örneğinde tespit edilmiştir. Klasik ve probiyotik yoğurt örneklerinde depolama süresince dış görünüş değerlerinde meydana gelen değişim Şekil 1'de görülmektedir



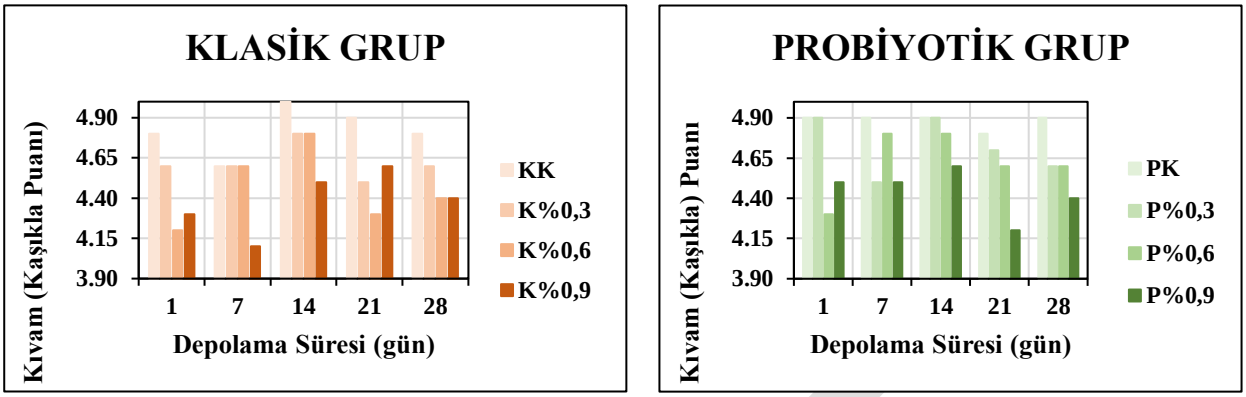
Şekil 1. Dış görünüş puanının depolama süresince değişimi

Dış görünüş değerlerine uygulanan varyans analizi sonucunda (Tablo 1), örnek değişkeninin dış görünüş puanı üzerine etkisi istatistiksel açıdan $p<0,01$ seviyesinde, depolama periyodunun etkisi ise $p<0,05$ seviyesinde önemli bulunmuştur. Örnek ve depolama değişkenlerinde, farklılığın kaynağını belirlemek amacıyla yapılan Duncan çoklu karşılaştırma test sonucu ise Tablo 2'de verilmiştir. Tablo incelendiğinde istatistiksel açıdan en yüksek ortalama değerin klasik grupta ve probiyotik grupta yer alan ve *Spirulina platensis* ihtiva etmeyen kontrol örneklerinde (KK & PK) olduğu görülmektedir. İstatistiksel olarak K%0,3, K%0,6, P%0,3 örnekleri benzerlik göstermiştir ($p>0,05$). K%0,9, P%0,6 ve P%0,9 örnekleri de istatistiksel olarak benzerlik göstermiştir ($p>0,05$).

En yüksek ortalama dış görünüş puanı, istatistiksel olarak aralarında fark olmayan ($p>0,05$) 1, 7, 14 ve 28. depolama günlerinde görülürken, en düşük ortalama dış görünüş puanı 21. günde belirlenmiştir ($p<0,05$).

Kıvam (kaşıkla).

Yoğurt örneklerine ait kıvam (kaşıkla) puanları 4,10 ile 5,00 arasındadır. Yoğurt örneklerinde en yüksek kıvam (kaşıkla) puanları 5,00 ile depolamanın 14. günde KK örneğinde; en düşük kıvam (kaşıkla) puanı 4,10 ile depolamanın 7. gününde K%0,9 örneğinde tespit edilmiştir. Klasik ve probiyotik yoğurt örneklerinde depolama süresince kıvam (kaşıkla) değerlerinde meydana gelen değişim Şekil 2'degörülmektedir.



Şekil 2. Kıvam (kaşıkla) puanının depolama süresince değişimi

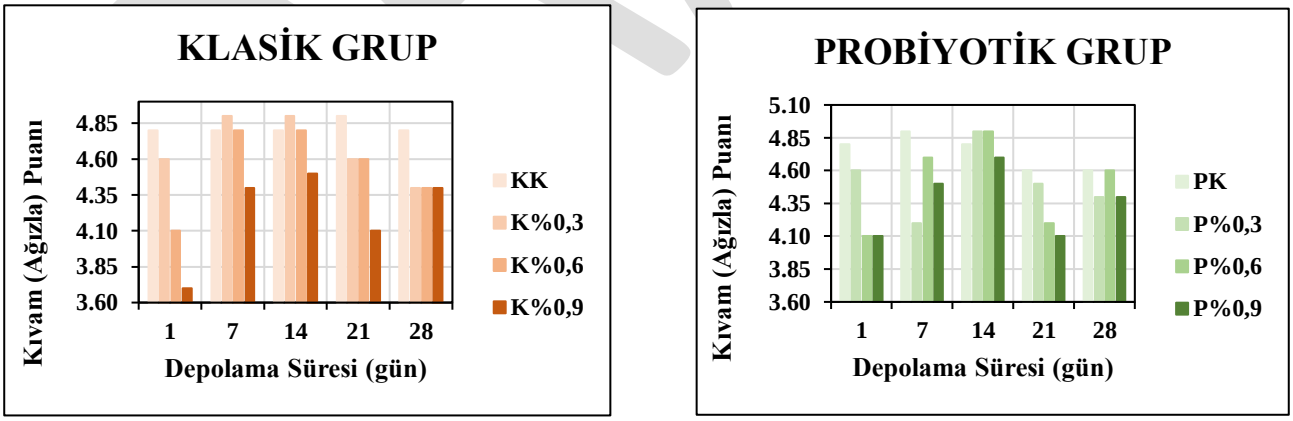
Kıvam (kaşıkla) değerlerine uygulanan varyans analizi sonucunda (Tablo 1), örnek değişkeninin kıvam (kaşıkla) puanı üzerine etkisi istatistiksel açıdan $p<0,01$ seviyesinde, depolama periyodunun etkisi ise $p<0,05$ seviyesinde önemli bulunmuştur. Örnek ve depolama değişkenlerinde, farklılığın kaynağını belirlemek amacıyla yapılan Duncan çoklu karşılaştırma test sonucu incelendiğinde (Tablo 2), istatistiksel açıdan en yüksek ortalama puanın probiyotik kontrol örneğinde (PK) olduğu görülmektedir. İstatistiksel olarak K%0,3 ve P%0,6 örnekleri benzerlik göstermiştir ($p>0,05$). En düşük ortalama puanın belirlendiği K%0,9 örneği ise istatistiksel olarak diğer örneklerden farklıdır ($p<0,01$).

En yüksek ortalama kıvam (kaşıkla) puanı depolamanın 14. günde görülürken, depolamanın diğer tüm günlerinde (1, 7, 21 ve 28. gün) benzerliğin olduğu ortaya çıkmıştır ($p>0,05$).

Kıvam (ağızla).

Yoğurt örneklerine ait kıvam (ağızla) puanları 3,70 ile 4,90 arasındadır. Yoğurt örneklerinde en yüksek kıvam (ağızla) puanları 4,90 ile depolamanın 7, 14 ve 21. günlerinde KK, K%0,3, PK, P%0,3 ve P%0,6 örneklerinde; en düşük kıvam (ağızla) puanı 3,70 ile depolamanın 1. gününde K%0,9 örneğinde tespit edilmiştir.

Klasik ve probiyotik yoğurt örneklerinde depolama süresince kıvam (ağızla) değerlerinde meydana gelen değişim Şekil 3'te görülmektedir.



Şekil 3. Kıvam (ağızla) puanının depolama süresince değişimi

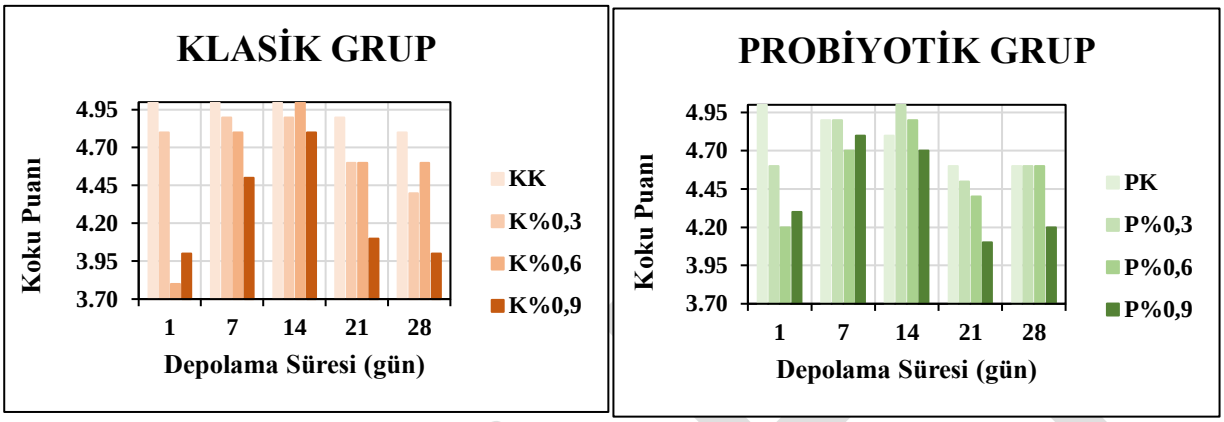
Yoğurt örneklerinin kıvam (ağızla) puanlarına ait varyans analiz sonuçları Tablo 1'de verilmiştir. Yapılan varyans analiz sonuçlarına göre, örnek değişkeninin ve depolama periyodunun etkisi kıvam (ağızla) puanı üzerine etkisi istatistiksel açıdan $p<0,01$ seviyesinde, önemli bulunmuştur. Duncan çoklu karşılaştırma test sonucu incelendiğinde (Tablo 2), istatistiksel açıdan en yüksek ortalama puanın klasik kontrol örneğinde (KK) olduğu görülmektedir. İstatistiksel olarak K%0,3, K%0,6 ve PK örnekleri; P%0,3 ve P%0,6 örnekleri kendi aralarında benzerlik göstermiştir ($p>0,05$). En düşük ortalama puan ise K%0,9 örneğinde çıktı ve istatistiksel olarak diğer örneklerden farklıdır ($p<0,01$).

En yüksek ortalama kıvam (ağızla) puanının depolamanın 14. gününde görülürken, depolamanın 21 ve 28. günlerinde benzerliğin olduğu ortaya çıkmıştır ($p>0,05$). En düşük ortalama kıvam (ağızla) puanının depolamanın 1. gününde olduğu ve istatistiksel olarak diğer günlerden farklı çıktığı görülmüştür.

Koku.

Yoğurt örneklerine ait koku puanları 4,00 ile 5,00 arasındadır. Yoğurt örneklerinde en yüksek koku puanları 5,00 ile depolamanın 1, 7 ve 14. günlerinde KK ve PK örneklerinde; en düşük koku puanı 4,00 ile depolamanın 1 ve 28. günlerinde K%0,9 örneğinde tespit edilmiştir. Klasik ve probiyotik yoğurt örneklerinde depolama süresince koku değerlerinde meydana gelen değişim Şekil 4'de görülmektedir

Şekil 4. Koku puanının depolama süresince değişimi

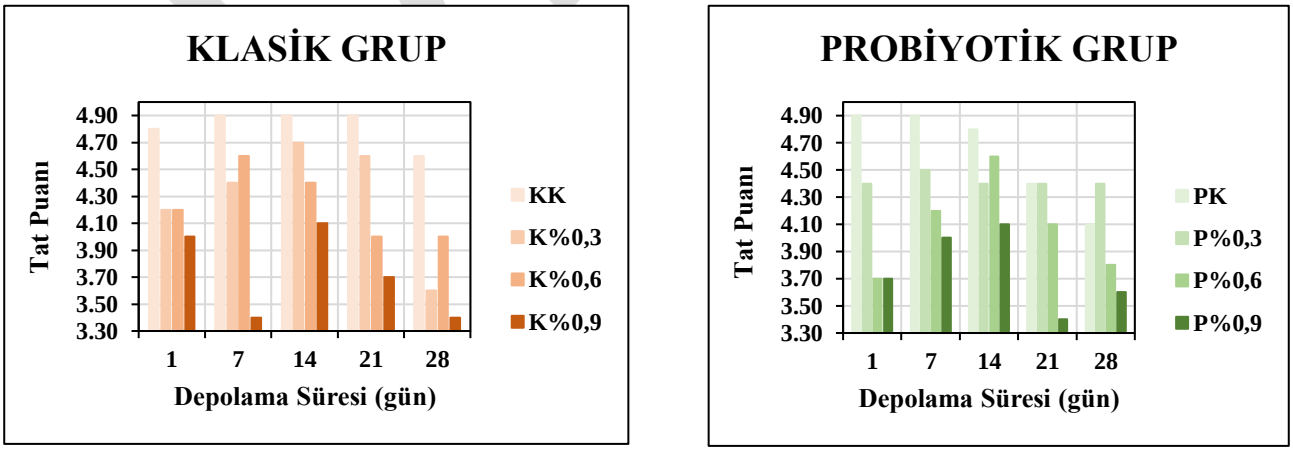


Koku puanlarına uygulanan varyans analizi sonucunda (Tablo 1), örnek değişkeninin ve depolama periyodunun koku puanı üzerine etkisi istatistiksel açıdan $p<0,01$ seviyesinde önemli bulunmuştur. Örnek ve depolama değişkenlerinde, farklılığın kaynağını belirlemek amacıyla yapılan Duncan çoklu karşılaştırma test sonucu incelendiğinde (Tablo 2), istatistiksel açıdan en yüksek ortalama koku puanının klasik kontrol örneğinde (KK) olduğu görülmektedir. İstatistiksel olarak K%0,3, K%0,6, PK ve P%0,3 örnekleri kendi aralarında benzerlik göstermiştir ($p>0,05$). En düşük ortalama değer ise K%0,9 örneğinde çıkmış ve istatistiksel olarak diğer örneklerden farklı bulunmuştur ($p<0,01$).

En yüksek ortalama koku puanları depolamanın 7 ve 14. gününde görülürken, depolamanın 1, 21 ve 28. günlerinde benzerliğin olduğu tespit edilmiştir ($p>0,05$) ve en düşük ortalama koku değerinin depolamanın 28. gününde olduğu tespit edilmiştir ($p<0,01$).

Tat.

Yoğurt örneklerine ait tat puanları 3,40 ile 4,90 arasındadır. Yoğurt örneklerinde en yüksek tat puanları 4,90 ile depolamanın 1, 7, 14 ve 21. günlerinde PK ve KK örneklerinde; en düşük tat puanı 3,40 ile depolamanın 7, 21 ve 28. günlerinde K%0,9 ve P%0,9 örneklerinde tespit edilmiştir. Klasik ve probiyotik yoğurt örneklerinde depolama süresince tat değerlerinde meydana gelen değişim Şekil 5'de görülmektedir



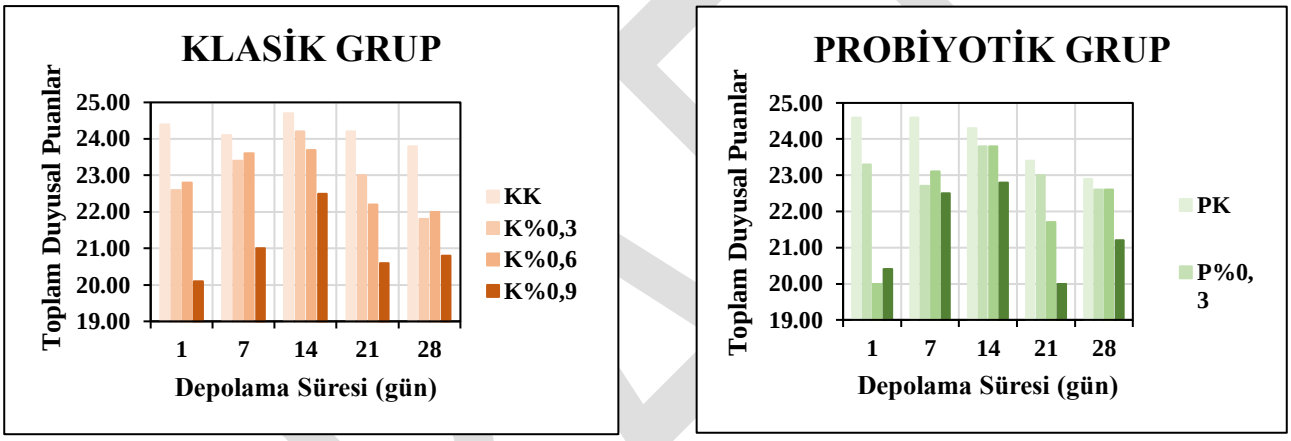
Şekil 5. Tat puanının depolama süresince değişimi

Tat puanlarına uygulanan varyans analizi sonucunda (Tablo 1), örnek değişkeninin ve depolama periyodunun tat puanı üzerine etkisi istatistiksel açıdan $p<0,01$ seviyesinde, önemli bulunmuştur. Örnek ve depolama değişkenlerinde, farklılığın kaynağını belirlemek amacıyla yapılan Duncan çoklu karşılaştırma test sonucu incelendiğinde (Tablo 2), istatistiksel açıdan en yüksek ortalama puanın 4,82 ile klasik kontrol (KK) örneğinde olduğu görülmektedir. İstatistiksel olarak K%0,3 ve K%0,6; K%0,9 ve P%0,9 örnekleri kendi aralarında benzerlik göstermiştir ($p>0,05$). En düşük ortalama değer ise 3,72 ile K%0,9 örneğinde çıkmıştır. Geriye kalan örnekler de kendi aralarında ve diğer tüm örneklerden istatistiksel olarak farklı çıkmıştır ($p<0,01$).

En yüksek ortalama tat puanının 4,50 ile depolamanın 14. gününde görüldüğü; depolamanın 1 ve 21. günlerinde benzerliğin olduğu ortaya çıkmıştır ($p>0,05$). En düşük ortalama tat değerinin 3,94 ile depolamanın 28. gününde olduğu tespit edilmiştir.

Toplam Duyusal Analiz Puanları.

Yoğurt örneklerine ait toplam duyusal analiz puanları 20,00 ile 24,70 arasındadır. Yoğurt örneklerinde en yüksek toplam duyusal analiz puanları 24,70 ile depolamanın 14. günü KK örneğinde; en düşük toplam duyusal analiz puanı 20,00 ile depolamanın 1 ve 21. günlerinde P%0,6 ve P%0,9 örneklerinde tespit edilmiştir. Klasik ve probiyotik yoğurt örneklerinde depolama toplam duyusal analiz puanlarında meydana gelen değişim Şekil 6'da görülmektedir.



Şekil 6. Toplam duyusal analiz puanının depolama süresince değişimi

Toplam duyusal analiz puanlarına uygulanan varyans analizi sonucunda (Tablo 36), örnek değişkeninin ve depolama periyodunun tat puanı üzerine etkisi istatistiksel açıdan $p<0,01$ seviyesinde, önemli bulunmuştur. Örnek ve depolama değişkenlerinde, farklılığın kaynağını belirlemek amacıyla yapılan Duncan çoklu karşılaştırma test sonucu incelendiğinde (Tablo 2), istatistiksel açıdan en yüksek ortalama puanın 24,24 ile klasik kontrol (KK) örneğinde olduğu görülmektedir. İstatistiksel olarak K%0,3 ve P%0,3 örnekleri kendi aralarında benzerlik göstermiştir ($p>0,05$). En düşük ortalama değer ise 21,00 ile K%0,9 örneğinde çıkmıştır. Geriye kalan örnekler de kendi aralarında ve diğer tüm örneklerden istatistiksel olarak farklı çıkmıştır ($p<0,01$). En yüksek ortalama toplam duyusal analiz puanlarının 23,73 ve 23,12 ile depolamanın 7 ve 14. günlerinde görüldüğü ve aralarında istatistiksel olarak benzerliğin ($p>0,05$) olduğu; en düşük ortalama toplam duyusal analiz puanının 22,21 ile depolamanın 28. gününde olduğu tespit edilmiştir. 1, 21 ve 28. günler de kendi aralarında benzerlik taşıdığı görülmektedir ($p>0,05$).

Tablo 1. Yoğurt örneklerinin duyusal niteliklerinin varyans analiz sonuçları

Varyasyon Kaynağı	SD	Dış görünüş	Kıvam (kaşık)	Kıvam (ağız)	Koku	Tat	Genel toplam
Örnek	7	4,018**	5,244**	5,469**	6,206**	13,212**	11,216**
Depolama	4	2,925*	2,258*	5,292**	8,015**	6,269**	6,374**
Örnek*Depolama	28	1,235	0,676	1,000	0,631	0,931	0,788

*: p<0,05, **: p<0,01

Tablo 2. Yoğurt örneklerinin duyusal niteliklerinin Duncan çoklu karşılaştırma test sonuçları

		Dış görünüş	Kıvam (kaşık)	Kıvam (ağız)	Koku	Tat	Genel toplam
ÖRNEK	**KK**	4,84±0,47^{a}	4,82±0,39^{ab}	4,82±0,39^{a}	4,94±0,24^{a}	4,82±0,39^{a}	24,24±1,44^{a}
	K%0,3	4,68±0,55^{ab}	4,62±0,49^{bcd}	4,68±0,47^{ab}	4,72±0,57^{ab}	4,30±0,71^{cd}	23,00±1,97^{bc}
	K%0,6	4,70±0,54^{ab}	4,52±0,58^{cde}	4,64±0,56^{ab}	4,76±0,52^{ab}	4,24±0,77^{cd}	22,86±2,44^{c}
	K%0,9	4,40±0,90^{b}	4,38±0,72^{e}	4,22±0,82^{d}	4,28±0,83^{d}	3,72±0,93^{e}	21,00±3,12^{e}
	PK	4,94±0,31^{a}	4,88±0,33^{a}	4,74±0,49^{ab}	4,78±0,54^{ab}	4,62±0,64^{ab}	23,96±1,83^{ab}
	P%0,3	4,70±0,65^{ab}	4,72±0,54^{abc}	4,52±0,68^{bc}	4,72±0,57^{ab}	4,42±0,67^{bc}	23,08±2,35^{bc}
	P%0,6	4,48±1,02^{b}	4,62±0,53^{bcd}	4,50±0,68^{bc}	4,56±0,64^{bc}	4,08±0,88^{d}	22,24±2,62^{cd}
	P%0,9	4,40±0,97^{b}	4,44±0,64^{de}	4,36±0,75^{cd}	4,42±0,84^{cd}	3,76±1,00^{e}	21,38±3,26^{de}
DEPOLAMA (GÜN)	**1**	4,44±0,95^{a}	4,60±0,56^{b}	4,41±0,76^{c}	4,59±0,67^{b}	4,24±1,06^{b}	22,27±2,84^{b}
	7	4,72±0,68^{a}	4,58±0,52^{b}	4,65±0,58^{ab}	4,81±0,45^{a}	4,36±1,16^{ab}	23,12±2,09^{a}
	14	4,76±0,53^{a}	4,79±0,44^{a}	4,79±0,41^{a}	4,89±0,31^{a}	4,50±0,74^{a}	23,73±1,43^{a}
	21	4,57±0,76^{ab}	4,57±0,57^{b}	4,45±0,65^{bc}	4,48±0,66^{b}	4,19±1,17^{b}	22,26±2,76^{b}
	28	4,71±0,66^{a}	4,59±0,67^{b}	4,50±0,69^{bc}	4,47±0,89^{b}	3,94±0,84^{c}	22,21±3,40^{b}

*Farklı harflerle gösterilen ortalamalar istatistiksel olarak birbirinden farklıdır.

SONUÇ

Yoğurt örneklerinin, depolama periyodu süresince 1, 7, 14, 21 ve 28. günlerde duyusal nitelikleri; dış görünüş, kıvam (kaşıkla), kıvam (ağızla), koku ve tat parametreleri ile toplam duyusal puanlar üzerinden değerlendirilmiştir. Spirulina platensis ilavesi ve depolama periyodunun tüm duyusal parametreler üzerine etkisi istatistiksel açıdan ($p<0,01$; $p<0,05$) önemli bulunmuştur. Spirulina platensis ilavesi tüm örneklerde duyusal puanları zayıflatsa da ortalama puanlar tüm gün ortalamalarında hiçbir örnekte 4 puanın altına düşmemiştir. Tüm parametrelerde en yüksek ortalama puanlar klasik grup örneklerde ve depolamanın 14. gününde belirlenmiştir. Duyusal analiz sonuçlarına göre kontrol örnekleri dışında en çok beğenilen ürün *Spirulina platensis* içeriği %0,3 olan probiyotik ve klasik yoğurt örnekleridir. *Spirulina platensis* miktarındaki artış, tat ve aromadaki artışa da doğrudan etki ettiğinden %0,6 ve %0,9 Spirulina platensis içeren örnekler %0,3 *Spirulina platensis* ihtiva eden örneklere göre daha düşük puan almışlardır. Fakat toplam skor 21 puanın (toplam duyusal analiz) altına düşmemiştir. Duyusal analiz sonuçları, depolamanın 21. gününe kadar tüm örneklerin duyusal genel kabul edilebilirlik açısından uygun olduğunu göstermektedir. Yüksek ihtimalle yosun tadından dolayı tercih edilmeyen bu zengin bileşimli ucuz besin ve enerji kaynağının ülkemiz insanının damak tadına hitap edecek şekilde formülasyonlarla gıda sektörüne kazandırılması önem arz etmektedir.

Teşekkür:

Bu çalışma, Doç. Dr. Ayla ARSLANER danışmanlığında Çağrı GÜLER tarafından tamamlanan "Spirulina platensis ilaveli probiyotik (Lactobacillus acidophilus) yoğurtlarin bazi kalite nitelikleri ve probiyotik raf ömrü" başlıklı Yüksek Lisans Tezi'nin bir bölümünden üretilmiştir.

KAYNAKLAR

Berner LA, O'Donnell JA 1998. Functional foods and health claims legislation: applications to dairy foods. International Dairy Journal, 8(5-6): 355-362.

Bodyfelt FW, Tobias J, Trout GM 1988. The Sensory Evaluation of Dairy Products. AVI Book, New York, 166-226.

Çakıroğlu FP, Uçar A 2018. Consumer attitudes towards purchasing functional products. Age, 18(25): 494.

Erbaş M 2006. Yeni bir gıda grubu olarak fonksiyonel gıdalar. Türkiye, 9: 24-26.

Litwin N, Clifford J, Johnson S 2018. Functional Foods for Health, Cobrado State University Extansion, 9: 391.

Çakmakçı S, Çetin B, Turgut T, Gürses M, Erdoğan A 2012. Probiotic Properties, Sensory Qualities, and Storage Stability of Probiotic Banana Yogurts. Turkish Journal Veterinary Animal Sciences, 36(3): 231-237.

Kailasapathy K, Harmstorf I, Phillips M 2008. Survival of Lactobacillus acidophilus and Bifidobacterium animalis ssp. lactis in stirred fruit yogurts, LWT - Food Science and Technology, 41(7):1317-1322,

Michaelsen KF, Hoppe C, Roos N, Kaestel P, Stougaard M, Lauritzen L, Molgaard C, Girma T, Friis H 2009. Choice of foods and ingredients for moderately malnourished children 6 months to 5 years of age. Food and Nutrition Bulletin, 30(3): 343-404.

SPSS 2022. SPSS 23.0 Statistical Package for the Social software. IBM Inc., 233 S. Wacker Dr., 11th Floor, Chicago, USA.

TSE 2009. TS 1330, Yoğurt Standardı. Türk Standartları Enstitüsü, Ankara

DRAFT

ORAL PRESENTATION

Salvia officinalis bitkisinin GC/MS yöntemi ile fitokimyasal olarak aydınlatılması ve *in silico* antioksidan aktivitesinin değerlendirilmesi

[1]Okan AYKAÇ*(ORCID: https://orcid.org/0000-0002-7363-8801)
[2]Ceylan HEPOKUR(ORCID: https://orcid.org/0000-0001-6397-1291)

[1] Sivas Cumhuriyet Üniversitesi Eczacılık Fakültesi Eczacılık Meslek Bilimleri Bölümü Farmasötik Kimya AbD, Sivas, Türkiye
[2]Sivas Cumhuriyet Üniversitesi Eczacılık Fakültesi Eczacılık Temel Bilimleri Bölümü Biyokimya AbD, Sivas, Türkiye

okanaykac@cumhuriyet.edu.tr

Özet

Türkiye'de tıbbi aromatik bitkiler denildiğinde aklımıza ilk gelen Ege Bölgesi ve Akdeniz Bölgesi kıyılarında yoğun olarak yetiştirilip (Bağdat, 2006), kültürü yapılan *Salvia off*. Geleneksel-tamamlayıcı tıp ve ticari açıdan kullanımları gün geçtikçe artmaktadır (Abreu ME, 2008). Bu özelliği ile bilim camiasının dikkatini yoğun bir şekilde çekmektedir. Geleneksel açıdan birçok terapötik etkisinden dolayı örneğin: Antioksidan, antikanser, asetil-kolinesteraz, bütil-kolinesteraz (Elmas S. ve Elmas O., 2021) tercih edilen Salvia off. cinsi bitkisinin literatürde fitokimyasal açıdan birçok araştırma konusu olduğu görülmüştür.

Bu çalışma kapsamında ticari adıyla adaçayı otunun biyolojik aktivite olarak antioksidan aktivitesine moleküler düzeyde anlam kazandırmak amaçlanmış ve GC/MS analizleri yapılarak ardından bu analiz sonuçlarında aydınlatılan bileşiklerin in silico olarak molekülar docking çalışmalarıyla desteklenebilirliği ortaya konuldu.

Anahtar Kelimeler: Antioksidan aktivite, *Salvia officinalis* , Moleküler modelleme

Phytochemical elucidation of *Salvia officinalis* plant by GC/MS method and evaluation of it is *in silico* antioxidant activity

Abstract

When we think of medicinal aromatic plants in Turkey, the first thing that comes to mind is Salvia off. Salvia, which is intensively cultivated and cultivated in the Aegean and Mediterranean coasts (Baghdad, 2006), and its traditional-complementary medicine and commercial uses are increasing day by day (Abreu ME, 2008). With this feature, it attracts the attention of the scientific community intensively. Due to its many traditional therapeutic effects, such as: Antioxidant, anticancer, acetyl-cholinesterase, butyl-cholinesterase (Elmas S. and Elmas O., 2021), Salvia off. has been the subject of many phytochemical studies in the literature.

Within the scope of this study, it was aimed to give meaning to the antioxidant activity of sage as a biological activity at the molecular level and GC / MS analyzes were performed and then the supportability of the compounds illuminated in these analysis results by molecular docking studies in silico was revealed.

Keywords: Antioxidant activity, Salvia officinalis , Molecular modeling

YÖNTEM

Ekstraksiyon Süreci:

Toplanan ve teşhis işlemi yapılan bitkinin toprak üstü kısımları gölgede kurutularak drog elde edildi. Bir öğütücüde uygun toz haline getirildi. Toz drog kalıntıları su, etanol, hekzan ve etil asetat çözücülerinde günde 8 saat olmak üzere 3 gün boyunca mukayese edildi. Ekstraksiyon işleminden sonra fraksiyonlar birleştirildi ve ekstrakt 40°C'de düşük basınç altında kurutuldu.

GC-MS Analizi:

DRAFT

Numune hazırlama aşamasında *Salvia off.* cinsinin ekstresinin tayini için analizden önce iki aşamalı türevlendirme yapıldı. Türevlendirmenin ilk aşaması için, analizden önce ekstrelerin sililenmiş türevi ve metoksiamin muamele edildi. Kromatogramda çoklu kromatografik heterozitlerin piklerini önlemek için metoksiamin kullanıldı. Metoksiaminleme işleminden önce, günlük olarak piridin (25 mg/ml) içinde metoksiamin hidroklorür (MOX) (Sigma-Aldrich, Almanya) çözeltisi hazırlandı. 100 µl numune hafif nitrojen akışı altında buharlaştırıldıktan sonra kurutulmuş numuneye 50 µl MOX solüsyonu ilave edilerek 30°C derecede 90 dakika bekletildi. Daha sonra 50 µl BSTFA + %1 TMCS (Sigma-Aldrich, Germany) eklendi ve aktif grupların korunmasını sağlamak amacıyla 70°C'de 45 dakika daha tutuldu.

Enstrümantal analiz için thermoscientific ISQ 7000 Single Quadrupole GC-MS System marka cihaz kullanıldı. Analitik kolon olarak teknokroma marka gaz kromatografisi kılcal kolonu kullanıldı. Taşıyıcı olarak distile su, etanol, etil asetat gazı kullanıldı. Kör numune olarak sadece türevlendirilmiş bir numune enjekte edildi. GC fırın sıcaklığı 50ºC'de başladı ve 2 dakika tutuldu, ardından 3°C/dk'da 280°C'ye yükseltildi ve 12 dakika tutuldu. Total sürüklenme zamanı 90 dk sürdü. Kütle spektrumu, 70 eV'de elektron iyonizasyonu ile elde edildi. Dedektör tarafından 50-500 atomik kütle birimi tarandı. Bileşikleri belirlemek ve tanımlamak için Mass Hunter yazılımı (Qualitative Analysis B.07.00) ve NIST Mass Spectral Library (2014) kullanıldı.

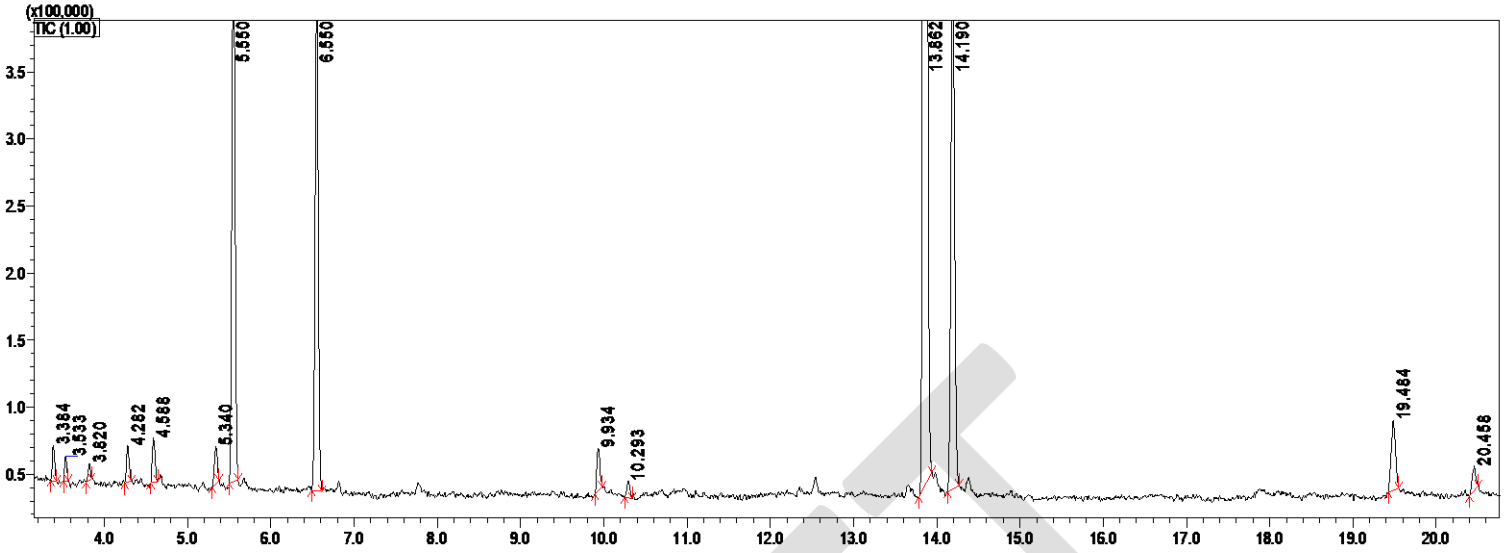
Moleküler Modelleme Çalışmaları:

İlk olarak *Salvia off*. cinsi bitkinin fitoterapideki kullanımına bağlı literatür taraması yapılarak hangi biyolojik aktivitenin daha çok bitkiyle ilişkilendirildiği sonucu araştırılarak, bu tarama sonucunda antioksidan aktivitenin çalışılmasına karar verildi ve uygun protein seçilimi yapıldı. Bu protein seçiminde PDB:6F7Q belirli saflaştırma özelliklerine göre elenerek çalışılacak protein olarak seçim yapıldı. PDB:6F7Q proteini daha önce Noshadi ve arkadaşlarının yapmış oldukları çalışmadaki hedef bölgesi baz alınarak çalışıldı (Noshadi et all., 2020). Hedef bölgedeki aminoasitler Tyr332, Phe329, Leu286, Trp231, Gly116, Glu197, Trp82 bilgisine Noshadi ve arkadaşının yapmış olduğu şekil 2'deki verilerden ulaşıldı (Noshadi et. all., 2020).

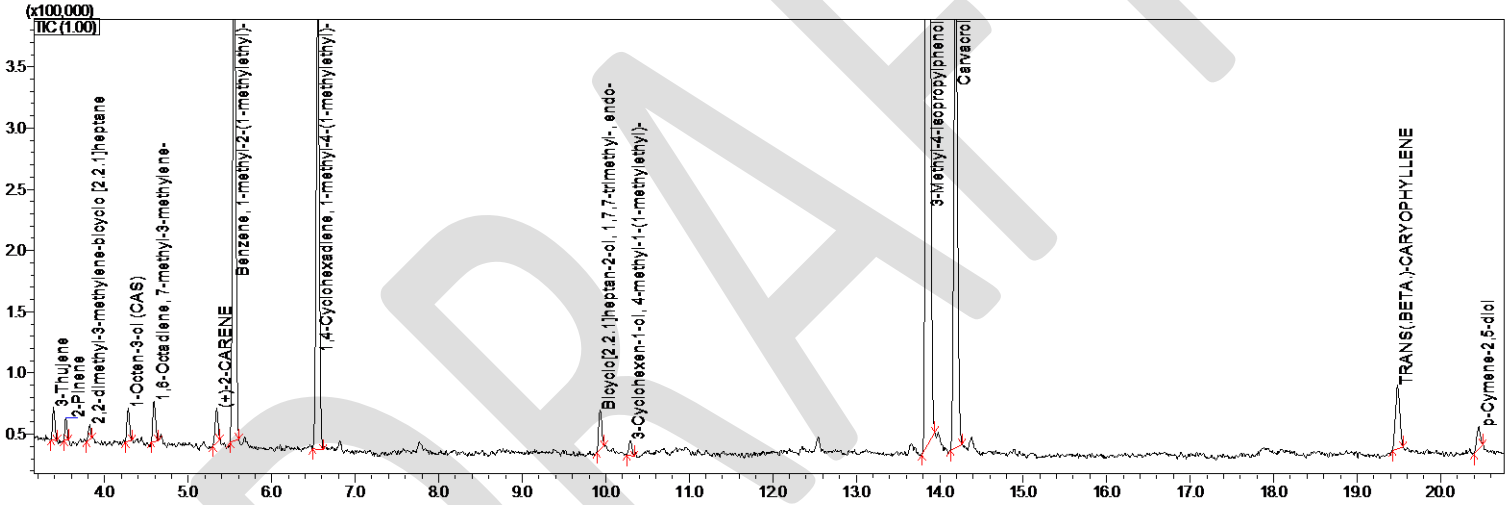
Salvia off. cinsinde GC-MS (gaz-kromatografisi) sonuçlarına göre farklı ve potansiyel olabilecek bileşikler 2D yapısı çizilerek ligandlar oluşturuldu. Daha sonra docking çalışması için Glide-2021(Schrödinger ©) arayüzü kullanıldı. Birinci adımda protein hazırlama yapıldı. A zinciri hariç diğer zincirler ve suların tümünün dışlanmasıyla OPLS_2005 işlemi yapıldı. Hedefin bağlanacağı ızgara alanı oluşturulurken spesifik aminoasitlerimizin bulunduğu yüzeyin olması sağlandı. İkinci aşama olarak LigPrep aşaması 2D olarak çizilen bileşiklerin seçilerek OPLS_2005 kuvvetine göre hazırlandı. Üçüncü aşama olarak docking aşaması için uygun koşullar seçilerek LigDock yapılarak görüntü ve sonuçlar elde edildi.

DRAFT

SONUÇLAR ve TARTIŞMA



Şekil 1. *Salvia off.*'nın GC/MS kromatogramı



Şekil 2. *Salvia off.*'nın GC/MS kromatogramı aydınlatılmış hali

DRAFT

Tablo 1. *Salvia off.*'nın GC/MS sonuçları

RT zamanı	m/z	Alan	Alan, %	Bileşikler	Benzerlik	Veribankası
3,384	TIC	51840	0,56	3-Thujene	91	W10N14
3,532	TIC	33183	0,36	2-Pinene	92	W10N14
3,82	TIC	23628	0,26	2,2-dimethyl-3-methylene-bicyclo [2.2.1]heptane	86	W10N14
4,281	TIC	58411	0,63	1-Octen-3-ol (CAS)	92	W10N14
4,588	TIC	75102	0,82	1,6-Octadiene, 7-methyl-3-methylene-	91	W10N14
5,34	TIC	67829	0,74	(+)-2-CARENE	92	W10N14
5,55	TIC	1104810	12,01	Benzene, 1-methyl-2-(1-methylethyl)-	96	W10N14
6,55	TIC	977489	10,62	1,4-Cyclohexadiene, 1-methyl-4-(1-methylethyl)-	96	W10N14
9,934	TIC	83350	0,91	Bicyclo[2.2.1]heptan-2-ol, 1,7,7-trimethyl-, endo-	94	W10N14
10,293	TIC	31563	0,34	3-Cyclohexen-1-ol, 4-methyl-1-(1-methylethyl)-	84	W10N14
13,862	TIC	5214670	56,68	3-Methyl-4-isopropylphenol	95	W10N14
14,19	TIC	1228515	13,35	Carvacrol	95	W10N14
19,484	TIC	191969	2,09	TRANS(.BETA.)-CARYOPHYLLENE	92	W10N14
20,458	TIC	57663	0,63	p-Cymene-2,5-diol	83	W10N14

GC/MS aydınlatma sonuçlarına göre şekil 1 ve şekil 2'deki kromatogram verilerinden tablo 1'de derlenen bileşiklerin varlığı saptanmış ve biyolojik aktivite aydınlatılması için hazır hale getirilmiştir.

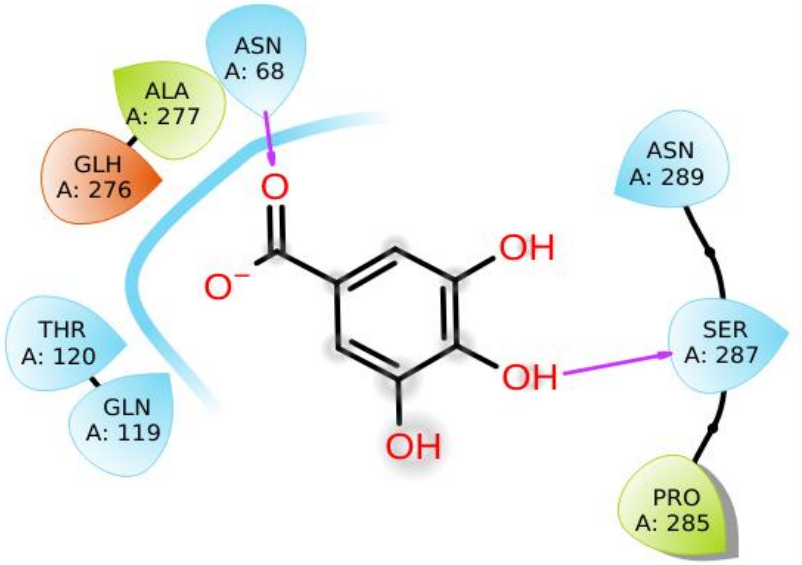
İn silico biyolojik aktivitede yorum yapılmasını kolaylaştırmak amacıyla gallik asit bileşiği (Sousa, J. N., Queiroz, L. D. R. P., vd,,2023) referans olarak belirlenip kullanılmıştır.

Tablo 2. Moleküler modelleme sonuçları

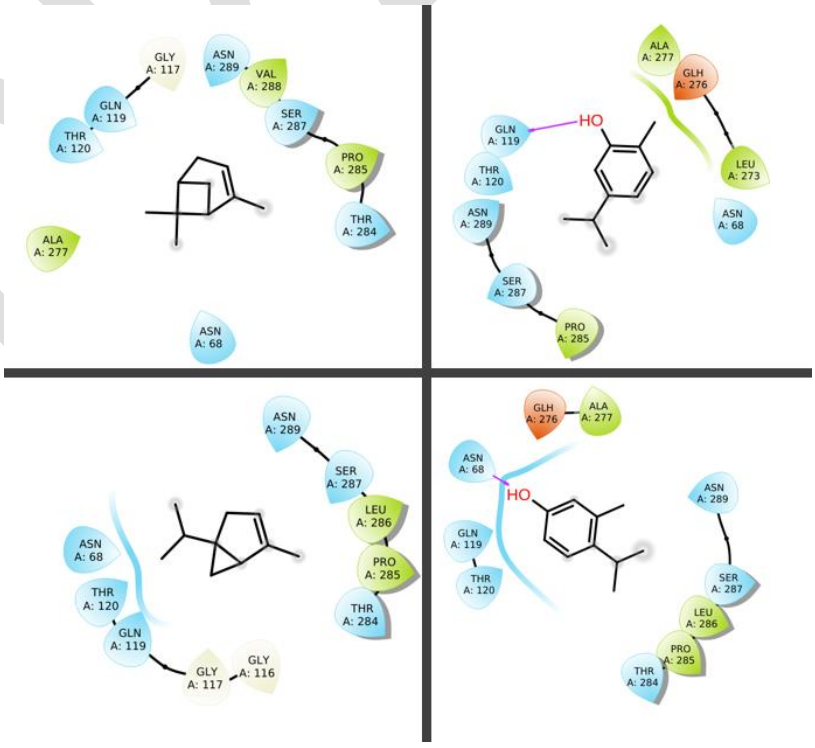
Bileşik Adı	Glide Energy	Docking Skore
Gallik asit	-25.269	-5.494
Karvakrol	-21.882	-4.672
2-isopropil-5-metil-hidrokinon	-19.742	-4.552
4-isopropil-3-metil-fenol	-21.566	-4.301
2,2-dimetil-3-metilen-norbornan-1,4-dikarboksilik asitin dimetil esteri	-24.047	-4.062
5-isopropill-2-metil-bisiklo[3.1.0]hekz-2-en	-15.458	-3.594
(1R,4E,9S)-4,11,11-trimetil-8-metilen-bisiklo[7.2.0]undek-4-en	-18.919	-3.496
2,6,6-trimetilbisiklo[3.1.1]hept-2-en	-14.598	-3.291
7-metil-3-metilen-okta-1,6-dien	-16.640	-2.315

DRAFT

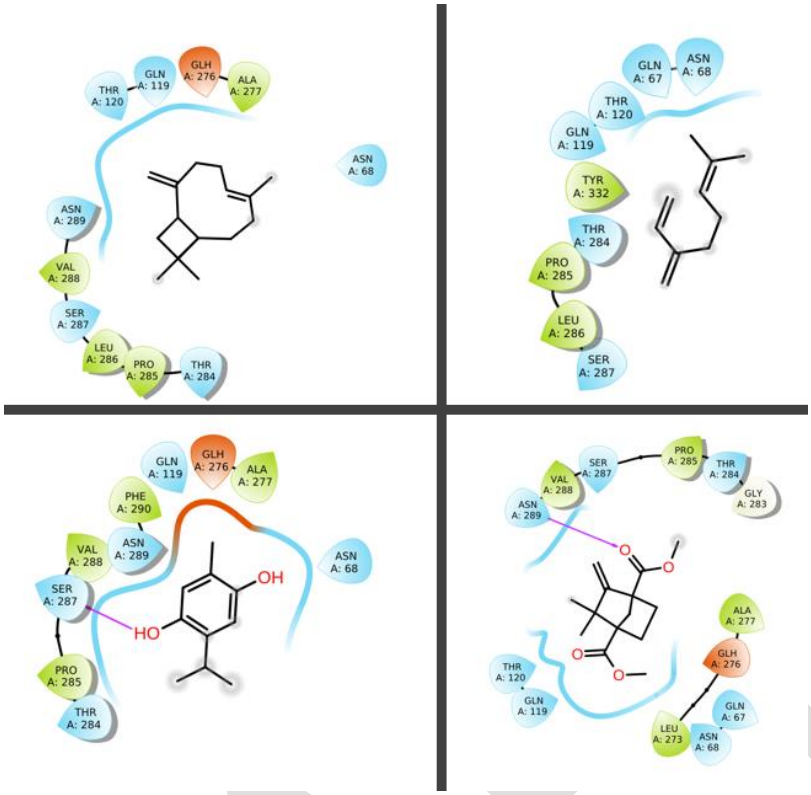
Tablo 2'de verilmiş olan verilere göre referans bileşik olan gallik asite oranla docking skoreu mutlak olarak en yakın olan karvakrol ve ardından gelen 2-isopropil-5-metil-hidrokinon bileşiğidir. Glide enerjileri tüm incelenen moleküllerde ortalama olarak aynı düzeydedir anlamlı bir fark gözlenmemiştir. Docking skoredan anlaşıldığı üzere aktivitede belirliyici faktör olma ihtimali yüksektir. Anlamlı korelasyon DPPH aktivite deneyi laboratuvar koşullarında gerçekleştiğinde yapılabilinmektedir.



Şekil 3. Referans bileşik olan gallik asidin ligand etkileşim diyagramı



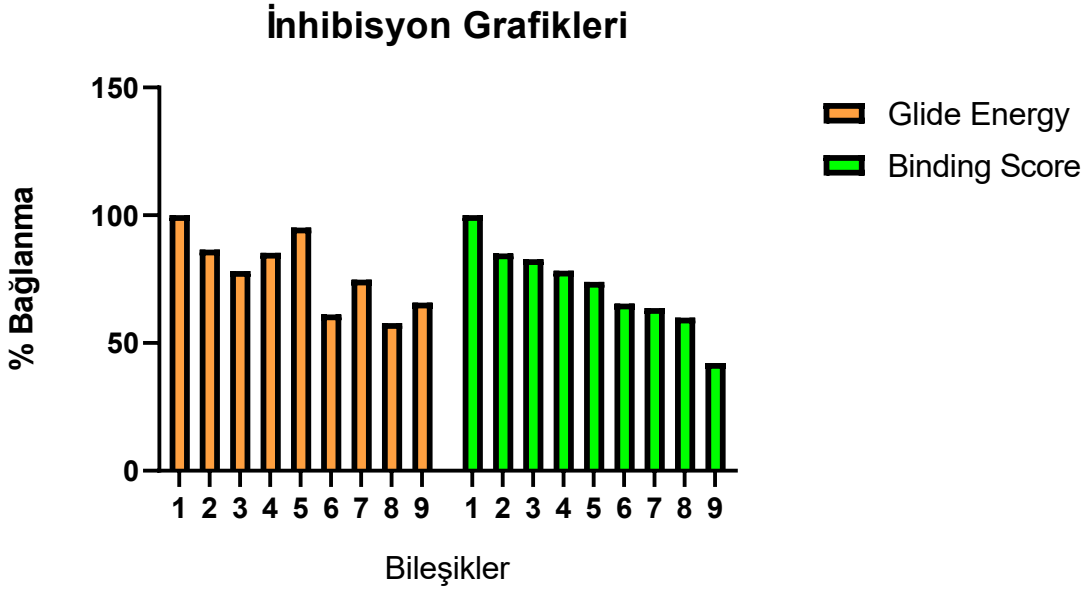
Şekil 4. Belirlenen bileşiklerin ligand etkileşimleri



Şekil 5. Belirlenen bileşiklerin ligand etkileşimleri

Şekil 4 ve 5 de verilen ligand interagtionlar için docking skore verisiyle hidrojen bağı yakalanan aminoasit sayısı ile orantı olabileceği sonucu ortaya çıkmıştır. Karvakrol gibi hidroksilce zengin bileşiklerde bağ yakalanma aktivite açısından önem teşkil ettiği gözlenmektedir.

Tüm bu veriler sonucunda *Salvia off.* tıbbi kültürü yapılarak antioksidan preparatı olarak farmakoekonomik açıdan uygun olmasa da başka etkinlikler için umut vaat edici özellikleri olabileceği tartışma konusu olmuş olup başka çalışmalar için esin kaynağı olabileceği ortaya çıkmıştır.



Şekil 6. Bağlanma enerjilerine göre in silico olarak inhibisyon değerlendirme oranları

KAYNAKLAR

Abreu ME, Munné-Bosch S, 2008. Salicylic acid may be involved in the regulation of drought-induced leaf senescence in perennials: a case study in field-grown Salvia officinalis L. plants. Environmental and Experimental Botany, 64 (2):105-112.

BAĞDAT, R. B. (2006). Tibbi ve aromatik bitkilerin kullanim alanlari, tibbi adaçayi (Salvia officinalis L.) ve ülkemizde kekik adiyla bilinen türlerin yetiştirme teknikleri. Tarla Bitkileri Merkez Araştırma Enstitüsü Dergisi, 15(1-2), 19-28.

Elmas, S., & Elmas, O. (2021). Salvia fruticosa'nın (Anadolu adaçayı) terapötik etkileri. *International Journal of Life Sciences and Biotechnology*, *4*(1), 114-137.

Noshadi, B., Ercetin, T., Luise, C., Yuksel, M. Y., Sippl, W., Sahin, M. F., ... & Gulcan, H. O. (2020). Synthesis, characterization, molecular docking, and biological activities of some natural and synthetic urolithin analogs. Chemistry & Biodiversity, 17(8), e2000197.

S. Schrödinger Release 2021-2: Glide, LLC, New York, NY, 2021.

Sousa, J. N., Queiroz, L. D. R. P., de Paula, A. M. B., Guimarães, A. L. S., Lescan, C. H., Aguilar, C. M., ... & Santos, S. H. S. (2023). Gallic Acid as a Sestrin (SESN2) activator and potential obesity therapeutic agent: a molecular docking study. Gene, 147683.

DRAFT

ORAL PRESENTATION

Erciyes Dağı (Kayseri) yer örümcekleri (Araneae: Gnaphosidae) faunası

Durmuş OK[1,2] (https://orcid.org/0000-0001-8784-6356), Osman SEYYAR[1](https://orcid.org/0000-0002-0920-7943), Hakan DEMİR[1](https://orcid.org/0000-0001-5456-4512)

[1]Niğde Ömer Halisdemir Üniversitesi, Fen-Edebiyat Fakültesi, Biyoloji Bölümü, Niğde, Türkiye

[1,2]Nevşehir Hacı Bektaş Veli Üniversitesi, Avanos MYO, Bitkisel ve Hayvansal Üretim Bölümü, Nevşehir, Türkiye

Sorumlu Yazar E-mail: durmusok@nevsehir.edu.tr

Özet

Kayseri ili sınırları içerisinde yer alan Erciyes Dağı'ndan 2018-2020 yıllarının Mayıs - Eylül ayları arasında yapılan arazi çalışmalarında Gnaphosidae familyasına ait örümcek örnekleri toplanmıştır. Toplanan örneklerin tür teşhisleri sonucunda 16 cinse ait 29 türün varlığı ortaya konmuştur. En çok tür tespit edilen cinsler sırasıyla *Drassodes, Micaria* ve *Haplodrassus* cinsleridir.

Anahtar Sözcükler: Gnaphosidae, fauna, Erciyes Dağı, örümcek, yer örümcekleri

Ground spiders (Araneae: Gnaphosidae) Fauna of Erciyes Mountain (Kayseri)

Abstract

Spider specimens belonging to the family Gnaphosidae were collected from Mount Erciyes in Kayseri province between May and September 2018-2020. Species identification of the collected specimens revealed the presence of 29 species belonging to 16 genera. The genera with the highest number of species were *Drassodes, Micaria* and *Haplodrassus,* respectively.

Keywords: Gnaphosidae, Fauna, Erciyes mountain, Spider, Araneae

Giriş

Arthropoda (Eklembacaklılar) şubesi içerisinde yer alan örümcekler, örümceğimsiler (Arachnida) sınıfına dâhildir ve biyolojik çeşitlilik bakımından oldukça zengin olan takımlarından birini oluşturur. Prosoma ile opistosoma'nın pedisel ile birleşik olması, erkeklerinde çiftleşme organının pedipalpler üzerinde bulunması ve opistosoma bölgesinde ağ bezi kabartılarının varlığı gibi özelliklere sahip olmalarıyla örümcekler diğer araknid gruplarından kolayca ayrılırlar (Foelix, 2011).

Örümcekler, Dünya'da 135 familya, 4335 cins, 51372 tür (World Spider Catalog, 2023), ülkemizde ise 55 familya, 370 cins, 1255 tür ile temsil edilmektedirler (Seyyar ve Demir, 2017; Danışman vd., 2023).

Ülkemizde yayılış gösteren örümceklerin büyük bir kısmını yer örümcekleri oluşturmaktadır. Gnaphosidae familyası 34 cinse ait 162 türle hem cins hem de tür sayısı bakımından ülkemizde ilk sırada yer alır (Danışman vd., 2023).

Gnaphosidae familyasına ait örümcekler siyah, koyu kahverengi veya griden yeşile kadar değişen renklerde, genellikle 1 ila 15 mm uzunluğunda ve iki tırnaklı bacaklara sahiptirler. Bazılarının sırt ve karın bölgelerinde desenler vardır. Ayrıca, familya üyelerinde gözlerin konumu ve büyüklüğü, labiyumun ve enditlerin şekli gibi önemli karakteristik özellikleri arasındadır. Bu yer örümceklerini diğer akraba familyalardan ayıran en önemli özelliği ise ön ağ bezi kabartılarının ayrık, silindirik şekilli ve uçlarının küt bir şekilde sonlanmasıdır (Platnick, 1978).

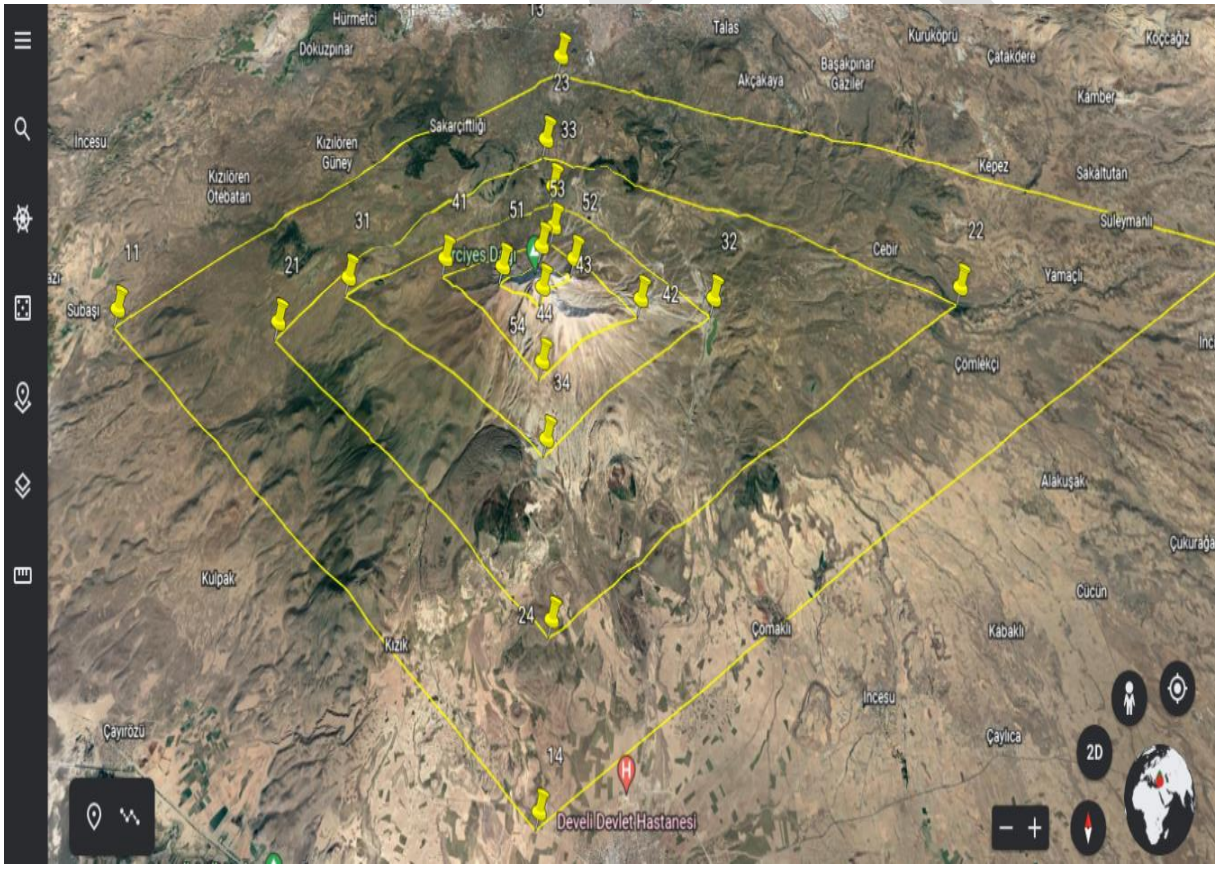
DRAFT

Gnafozidler genellikle 1–15 mm uzunluğunda, bacakları iki 2 tırnaklı, genellikle desenlenme göstermeyen örümceklerdir. Genellikle toprakta, taş altlarında, ağaç kabuklarında, kaya çatlaklarında veya kurumuş yaprak döküntüleri arasında yaşarlar ve buralarda tüp şeklinde ağlar örerler (Beccaloni, 2009; Foelix, 2011).

Bu çalışmanın amacı, Erciyes Dağı'nda yayılış gösteren yer örümcekleri faunasının ortaya çıkarılmasıdır.

Materyal ve Metot

Çalışma alanı olan Erciyes Dağı'nda 2018-2020 yılları arasında Mayıs-Eylül ayları aralığında arazi çalışmaları tamamlanmıştır. Şekil 1'de örnekleme yapılan alanlar verilmiştir. Örneklerin toplanacağı arazide önce lokalite tespitleri yapılmıştır. Bu toplama alanları belirlenirken, örümceklerin daha yoğun bulunacağı ağaçlık alanlar, kurumuş ağaç kabukları, bitki artıklarının olduğu döküntüler, kaya, taş, sulak alanlara yakın yerler tercih edilmiştir. Bu bölgelere sistematik olarak çukur tuzaklar kurulup, ağaçlık alanlarda da silkme şemsiye yöntemi ile örnek toplama tercih edilmiştir. Bu alanların dışında kalan bölgeler ise belirli çaplarda gezilerek atrap ve aspiratör yardımıyla da örnekler toplanmıştır (Şekil 2). Arazide çeşitli yöntemlerle toplanan örnekler, içerisinde % 70-75'lik etil alkol ve birkaç damla gliserin karışımı bulunan saklama şişelerine konularak muhafaza edilmiştir.



Şekil 1. Örnekleme yapılan lokaliteler (2018-2020)



Şekil 2. Arazi çalışmalarında kullanılan ekipmanlar (Pens, şemsiye ve aspiratör).

Muhafaza edilerek laboratuvara getirilen örnekler öncelikle Gnaphosidae familyasına ait olanları belirlemek amacıyla, familya düzeyinde teşhisleri yapılarak ayıklandı. Örneklerin cins ve tür teşhislerinde müze materyalleri ve literatür bilgilerinden faydalanıldı.

Bulgular

Araştırma alanından toplanan örneklerin faunistik değerlendirilmesi sonucunda Gnaphosidae familyasıdan 16 cinse ait 29 tür tespit edilmiştir. Türlerin eşey durumlarına göre dağılımları aşağıda Çizelge 1'de verilmiştir.

DRAFT

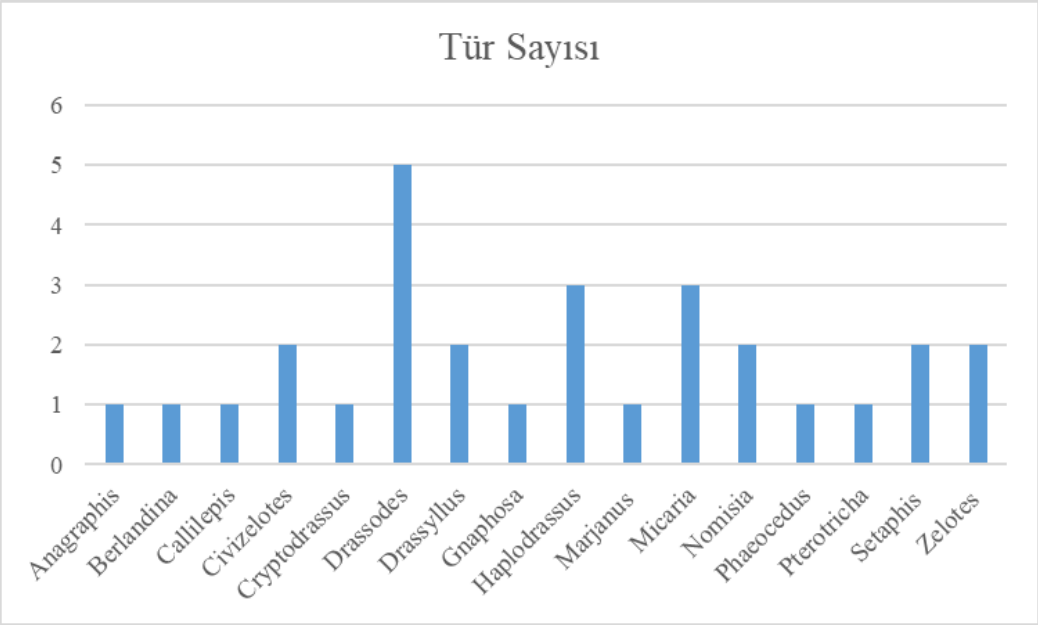
Çizelge 1. Çalışma alanından toplanın türler ve eşey durumları

TÜR LİSTESİ ve EŞEY DURUMLARI		
FAM. GNAPHOSIDAE		
Tür İsimleri	♀	♂
Anagraphis pallens	+	-
Berlandina plumalis	+	-
Callilepis cretica	+	+
Civizelotes caucasius	+	+
Civizelotes solstitialis	+	+
Cryptodrassus creticus	+	+
Drassodes bifidus	+	+
Drassodes lacertosus	+	+
Drassodes lapidosus	+	+
Drassodes lutescens	+	+
Drassodes pubescens	+	+
Drassyllus praeficus	+	+
Drassyllus pusillus	+	+
Gnaphosa lucifuga	+	+
Haplodrassus dalmatensis	+	-
Haplodrassus invalidus	+	-
Haplodrassus signifer	+	+
Marjanus platnicki	+	+
Micaria coarctata	+	+
Micaria formicaria	+	-
Micaria rossica	+	+
Nomisia aussereri	+	+
Nomisia exornata	+	+
Phaeocedus braccatus	+	-
Pterotricha kochi	+	-
Setaphis carmeli	+	+
Setaphis parvula	+	-
Zelotes longipes	+	+
Zelotes subterraneus	+	+

Sonuç ve Tartışma

Araştırma alanından toplanan örneklerin faunistik değerlendirilmesi sonucunda Gnaphosidae familyasıdan 16 cinse ait 29 tür tespit edilmiştir. Bu türlerden *Marjanus platnicki* (Zhang, Song & Zhu, 2001) türü son zamanlarda sadece ülkemizde Erciyes Dağı'nda tespit edilmiş bir türdür (Seyyar vd., 2019). Erciyes Dağı'ndan daha önce Nosek tarafından tespit edilmiş olan *Drassodes lutescens* (C. L. Koch, 1839), *D. similis*

Nosek, 1905, *Drassodes pubescens* (Thorell, 1856), *Haplodrassus signifer* (C. L. Koch, 1839), *Gnaphosa lucifuga minor* Nosek, 1905, *Zelotes longipes*, Z. *atrocaeruleus* türlerini tanımlanmıştır (Nosek, 1905). Yapılan arazi çalışmaları sonucunda bu türlerden 4 tanesi yeniden Erciyes Dağında rastlanılmıştır. En çok türü bulunduran cins 5 türle *Drassodes* cinsi olup bunu sırasıyla 3 türle *Micaria* ve *Haplodrassus* cinsleri takip etmektedir (Şekil 3).



Şekil 3. Erciyes Dağı'nda tespit edilen cinslerin türlere göre dağılımı

Teşekkür

Finansal desteğinden dolayı Niğde Ömer Halisdemir Üniversitesi Bilimsel Araştırmalar Birimine (Proje No: FEB 2018/09- TÜBİTAK C) teşekkür ederiz.

Kaynaklar

Beccaloni, J., Arachnids, University of California Press, Berkeley Los Angeles, 2009.

Danışman, T., Kunt, K.B., & Özkütük, R.S., The Checklist of the Spiders of Turkey. *http://www.spidersofturkey.info* (Erişim tarihi: 29.07.2023)

Demir, H., ve Seyyar, O, Annotated checklist of the spiders of Turkey. *Munis Entomology & Zoology*, 12(2): 433-469, 2017.

Foelix, R.F., Biology of Spiders, ***Oxford University Press,*** London, UK, 2011.

Platnick, N. I., Spinneret morphology and phylogeny of ground spiders (Araneae, Gnaphosoidea), ***Am. Mus. Nov.***, 2978, 1–42, 1978.

World Spider Catalog., World Spider Catalog. Version 24.5. Natural History Museum Bern, online at http://wsc.nmbe.ch (Erişim tarihi: 21.08.2023)

ORAL PRESENTATION

Investigation of the Effect of Temperature on pH in Kombucha Fermentation

Cihan DÜŞGÜN[1*] (*ORCID: https://orcid.org/0000-0003-2796-8356*)

[*1]Nigde Ömer Halisdemir University, Faculty of Arts and Sciences, Department of Biology, Niğde, Turkiye

*Corresponding author e-mail: cihandusgun@gmail.com

Abstract

Kombucha is a fermented beverage. Its consumption has significantly increased during the last decades due to its perceived beneficial effects. For this reason, it has become a highly commercialized drink that is produced industrially. During fermentation, pH, which determines acidity, is very important. The aim of this study was to determine the pH changes during fermentation at different temperatures and to investigate which temperature should be preferred for safe consumption. For this purpose, kombucha samples prepared with green tea were fermented at 20, 24, 28 and 32°C 16 days. pH measurements were made every 2 days. It was determined that the most suitable pH for optimum consumption was fermentation at 24 and 28°C.

Keywords: Kombucha, fermentation, pH, green tea, beverage

INTRODUCTION

Kombucha is a traditional fermented tea that has gained popularity due to its potential health benefits. It is rich in organic acids, minerals, vitamins, proteins, polyphenols, and other beneficial compounds. These compounds contribute to the various therapeutic properties of kombucha, including its antioxidant, antimicrobial, and antifungal activities (Mousavi et al., 2020). Scientific studies have shown that kombucha has potential anticancer effects and may enhance resistance to cancer (Watawana et al., 2015). The beverage has also been found to have hepatoprotective effects (Mousavi et al., 2020).

The fermentation process of kombucha involves a symbiotic culture of bacteria and yeast, known as SCOBY (Nizioł-Łukaszewska et al., 2020). This fermentation process leads to the production of bioactive compounds, such as antioxidants, which contribute to the health benefits of kombucha (de Miranda et al., 2022). Kombucha has been found to have high antioxidant activity, mainly attributed to its phenolic compounds. These phenolic compounds are responsible for the supposed health benefits of the beverage (Miranda et al., 2022)(de Miranda et al., 2022).

Furthermore, kombucha has been investigated for its potential as a probiotic. It has been found to have a broad microbial diversity, including probiotic bacteria and yeasts (Kaashyap et al., 2021). These microorganisms have been shown to have health benefits for the human gut, suggesting that kombucha can act as a powerful probiotic (Kaashyap et al., 2021). The presence of probiotics in kombucha contributes to its potential for improving gut health and overall well-being.

However, it is important to note that while kombucha has many potential health benefits, there are also risks associated with its consumption. Some health risks and side effects have been reported, including the presence of biological and chemical hazards (Kim and Adhikari, 2020). Therefore, it is important to ensure the safety and quality of kombucha products through proper manufacturing practices and food safety measures (Kim and Adhikari, 2020).

The aim of this study was to determine that the pH values of kombucha samples fermented with green tea at different temperatures at the end of the fermentation period are suitable for safe consumption.

DRAFT

MATERIALS AND METHODS

Green tea leaves and starter culture were collected from local shop in the Türkiye. The green tea broth was prepared by adding 7 g of green tea leaves to boiling water. After 30 min, the tea leaves were sieved and 70g of sucrose was added to prepare sugared green tea. Then, 1000 mL of sugared green tea was poured into a 2000 mL bottle, a 10% (v/v) starter culture was added. The bottle was covered with a cloth sheet and secured with rubber bands, then it was incubated for 16 days under aerobic conditions at temperatures ranging from 20°C to 32°C. pH values were measured and recorded every 4 days. The measurements were made in 3 parallels and averaged.

RESULTS AND DISCUSSION

In this study, it was aimed to determine the effect of temperature changes on pH changes during Kombucha fermentation. For this, a 14-day fermentation process was used. The pH values recorded as a result of the fermentation process are shown in Table 1.

Table 1. pH values measured during the fermentation period

Kombucha Fermentation	0th day	4th day	8th day	12th day	16th day
Kombucha at 20°C	6.74±0.12	6.35±0.08	6.01±0.04	5.57±0.04	5.31±0.08
Kombucha at 22°C	6.74±0.12	6.21±0.07	5.89±0.05	5.06±0.04	4.61±0.07
Kombucha at 24°C	6.74±0.12	5.99±0.08	5.10±0.05	4.23±0.05	3.74±0.06
Kombucha at 26°C	6.74±0.12	5.87±0.06	4.92±0.07	3.95±0.05	3.45±0.07
Kombucha at 28°C	6.74±0.12	5.80±0.08	4.90±0.06	3.85±0.03	3.30±0.07
Kombucha at 30°C	6.74±0.12	5.74±0.07	4.81±0.05	3.72±0.03	3.01±0.06
Kombucha at 32°C	6.74±0.12	5.70±0.05	4.69±0.04	3.48±0.04	2.87±0.05

Table 1 shows the pH change during fermentation depending on the temperature. At the end of the 14th day of fermentation at 20°C, the pH value was determined as 5.31±0.08. This value is the highest pH value obtained. this pH value is suitable for consumption. However, it is insufficient for the completion of kombucha fermentation. In addition, the pH value was measured as 2.87±0.05 as a result of fermentation at 32°C. This is the lowest pH value among all samples. this value is harmful for kombucha consumption. kombucha samples consumed at this pH are known to be harmful to the human stomach (Vitas et al., 2018).

Coskun and Kayisoglu, (2020) carried out kombucha fermentation using different plant samples. In their study, they reported that the optimum temperature for pH values was 24°C. Deghrigue et al., (2013) also carried out fermentation using a temperature of 25°C for kombucha fermentation. Fu et al., (2014) used a temperature of 30°C for kombucha fermentation of black tea. They ended the fermentation on the 8th day. In 8 days they reached the optimum pH level. The optimum fermentation time for traditionally prepared kombucha is 14 days. On the 8th day, they terminated the fermentation because they reached a low pH value that could be consumed. However, they did not reach the 14th day, which is the appropriate fermentation period for kombucha. This showed that the pH value decreased very quickly at 30°C. This study is in parallel with the results we obtained in this aspect. Gaggìa et al., (2019) used 28 degrees Celsius for fermentation temperature in their study. They measured pH on the 7th and 14th day. The result they obtained shows that kombucha fermentation takes place successfully. The pH measurements obtained with the temperatures used in the study are in parallel with our results.

As a result, different temperatures are used for kombucha fermentation. However, according to the results obtained, pH values suitable for human consumption differ. In this respect, it is suggested that the optimum pH range for kombucha consumption is between 3.74 and 3.30. In our results, it is seen that this pH range is reached at 24, 26 and 28°C.

DRAFT

CONCLUSION

In conclusion, kombucha is a fermented drink widely consumed among people. pH is important for the human stomach in the consumption of this fermented beverage. As a result of 14 days of fermentation, it was revealed with the results we obtained that the appropriate temperature for kombucha, which is ready for consumption, is between 24 and 28°C. The results we obtained were useful in obtaining preliminary information to reveal the importance of kombucha consumption in terms of pH for the human stomach. It has contributed to the literature in terms of other effects that kombucha samples consumed in this pH range may cause in terms of human stomach health. It is recommended that studies using human subjects be conducted to obtain more detailed information.

REFERENCES

Coskun F and Kayisoglu S 2020. Determination of Some Microbiological Properties of Kombucha Produced from Different Herbal Teas. Global Journal of Research In Engineering, 20(2):17-25.

de Miranda JF, Ruiz LF, Silva CB, Uekane TM, Silva KA, Gonzalez AGM, Fernandes FF and Lima AR 2022. Kombucha: A review of substrates, regulations, composition, and biological properties. Journal of Food Science, 87(2):503-527.

Deghrigue M, Chriaa J, Battikh H, Kawther A and Bakhrouf A 2013. Antiproliferative and antimicrobial activities of kombucha tea. African Journal of Microbiology Research, 7(27):3466-3470.

Fu C, Yan F, Cao Z, Xie F and Lin J 2014. Antioxidant activities of kombucha prepared from three different substrates and changes in content of probiotics during storage. Food Science Technology, 34(1):123-126.

Gaggìa F, Baffoni L, Galiano M, Nielsen D, Jakobsen R, Castro-Mejía J, Bosi S, Truzzi F, Musumeci F and Dinelli G 2019. Kombucha beverage from green, black and rooibos teas: a comparative study looking at microbiology, chemistry and antioxidant activity. Nutrients, 11(1):1-22.

Kaashyap M, Cohen M and Mantri N 2021. Microbial diversity and characteristics of kombucha as revealed by metagenomic and physicochemical analysis. Nutrients, 13(12):4446-4460.

Kim J and Adhikari K 2020. Current trends in kombucha: Marketing perspectives and the need for improved sensory research. Beverages, 6(1):15-34.

Mousavi SM, Hashemi SA, Zarei M, Gholami A, Lai CW, Chiang WH, Omidifar N, Bahrani S and Mazraedoost S 2020. Recent progress in chemical composition, production, and pharmaceutical effects of kombucha beverage: A complementary and alternative medicine. Evidence-Based Complementary and Alternative Medicine, 2020:1-14.

Nizioł-Łukaszewska Z, Ziemlewska A, Bujak T, Zagórska Dziok M, Zarębska M, Hordyjewicz Baran Z and Wasilewski T 2020. Effect of fermentation time on antioxidant and anti-ageing properties of green coffee Kombucha ferments. Molecules, 25(22):5394-5421.

Vitas JS, Cvetanović AD, Mašković PZ, Švarc-Gajić JV and Malbaša RV 2018. Chemical composition and biological activity of novel types of kombucha beverages with yarrow. Journal of Functional Foods, 44:95-102.

Watawana MI, Jayawardena N, Gunawardhana CB and Waisundara VY 2015. Health, wellness, and safety aspects of the consumption of kombucha. Journal of Chemistry, 2015:1-13.

ORAL PRESENTATION

Transcriptional and Translational Approaches in Spiders with Different Ecological Niche

Cihan DÜŞGÜN[1*] (*ORCID: https://orcid.org/0000-0003-2796-8356*), Simge ÖZARSLAN[1] (*ORCID: https://orcid.org/0000-0001-8920-4162*), Melek Handan ÖZPOLAT[1] (*ORCID: https://orcid.org/0000-0002-6849-5121*), Osman SEYYAR[1] (*ORCID: https://orcid.org/0000-0002-0920-7943*), Hakan DEMİR[1] (*ORCID: https://orcid.org/0000-0001-5456-4512*), Zeliha SELAMOĞLU2 (*ORCID: https://orcid.org/0009-0000-4132-4616*)

[*1]Nigde Ömer Halisdemir University, Faculty of Arts and Sciences, Department of Biology, Niğde, Turkiye

[2]Nigde Ömer Halisdemir University, Faculty of Medicine, Department of Medical Biology, Niğde, Türkiye

*Corresponding author e-mail: cihandusgun@gmail.com

Abstract

Spiders are the most successful of the arachnids (Arachnida) class of arthropods (Arthropoda), adapted to terrestrial ecosystems. Spiders have many enemies. Especially non-web-building spiders have many natural enemies during hunting. In this study, the similarities or differences between web-building and non-web-building spider species were examined by comparing total protein and total RNA levels and by performing biochemical analyses at the molecular level to obtain preliminary information on protein and gene structure of spider species with different hunting strategies through transcriptional and translational approaches. Total protein and total RNA measurements were analyzed for this study. As a result, it was determined that spiders hunted by making webs contained more total protein and total RNA than spiders hunted without making webs.

Keywords: Spider, total protein, total RNA

INTRODUCTION

Spiders are adapted to many of the world's ecosystems and therefore have a wide range of distribution across the globe. There have been many studies on spiders with different hunting strategies and ecological niches (Foelix, 2011). Studies on spiders, which have been the center of attention in the last 50 years, are mostly faunistic, systematic and ecological (Beccaloni, 2009). The number of molecular and biochemical studies on spiders is remarkably low, and these studies are mostly concerned with the detailed structure of spider webs and their silk. Spider silk produced by different spiders may have different composition and properties.

After protein synthesis, reactions such as phosphorylation and methylation play an important role in the functionalization of proteins and important biological molecules such as nucleic acids. Post-synthesis modifications of proteins make them important in signal transduction and enable a rapid and organized sequential collective response to extracellular events. The interaction of proteins plays an important role in many key cellular events including regulation of transcription (Clarke, 2003), cellular stress response, protein repair and aging (Kujubu et al., 1993), T cell activation (Cimato et al., 1997), nuclear transport (Vemuri and Philipson, 1988), neuronal differentiation ion channel function, cytokine signaling (Chen et al., 2004).

RNA analysis is important in determining the responses of organisms to different environmental conditions at the molecular level. RNA analysis provides preliminary information in determining the properties of proteins produced in organisms with different living conditions.

In this study, in order to obtain preliminary information on the transcriptional and translational expression and gene and protein size of spider species with different hunting strategies, total RNA and total protein levels were compared to determine the similarities or differences between different species at the molecular level.

DRAFT

MATERIALS AND METHODS

In this study, *Araneus quadratus* and *Araneus diedamatus* spider species, which are hunted by making webs, and *Alopecosa pulverulenta* and *Drassodes lapidosus* spider species, which are hunted without making webs, were obtained from nature and identified in the laboratory.

For homogenization of spiders, 0.1 g of sample was homogenized in 10 mL of 2 mM (pH: 7.4) phosphate buffer for 5 min. After homogenization, the samples were centrifuged at 10000 rpm for 5 min and 1 mL of the supernatant in the tubes was placed in sterile ependorf tubes and kept at +4°C.

The method proposed by Lowry was used to determine the amount of protein in 1 µL of supernatants from spider species (Lowry et al., 1951). Total RNA elution was performed according to the method of Chomczynski and Sacchi (Chomczynski and Sacchi, 1987).

RESULTS AND DISCUSSION

Lowry method was used to determine the protein content of the supernatants obtained from spider species. The total protein and RNA results we obtained are shown in Table 1. The protein content of *A. quadratus*, one of the spider species hunted by making webs, was 0.287 ± 0.023 ng/µL and the protein content of *A. diadematus*, another spider species hunted by making webs, was 0.251 ± 0.018 ng/µL. The protein content of *A. pulverulanta*, a spider species that hunts without making a web, was 0.122 ± 0.007 ng/µL and the protein content of *D. lapidosus*, another spider species that hunts without making a web, was 0.151 ± 0.008.

Table 1. Total protein and RNA results of the samples

Spider species	Total protein (ng/µL)	Total RNA (ng/µL)
Araneus quadratus	0,287 ± 0,023	0,280 ± 0,014
Araneus diadematus	0,251 ± 0,018	0,214 ± 0,023
Alopecosa pulverulenta	0,122 ± 0,007	0,084 ± 0,009
Drassodes lapidosus	0,151 ± 0,008	0,107 ± 0,008

The total RNA content of *A. quadratus*, one of the spider species that hunts by making webs, was 0.280 ± 0.014 ng/µL and the total RNA content of *A. diadematus*, another spider species that hunts by making webs, was 0.214 ± 0.023 ng/µL. The total RNA amount of *A. pulverulanta*, a spider species that hunts without making a web, was 0.084 ± 0.009 ng/µL and the total RNA amount of *D. lapidosus*, another spider species that hunts without making a web, was 0.107 ± 0.008.

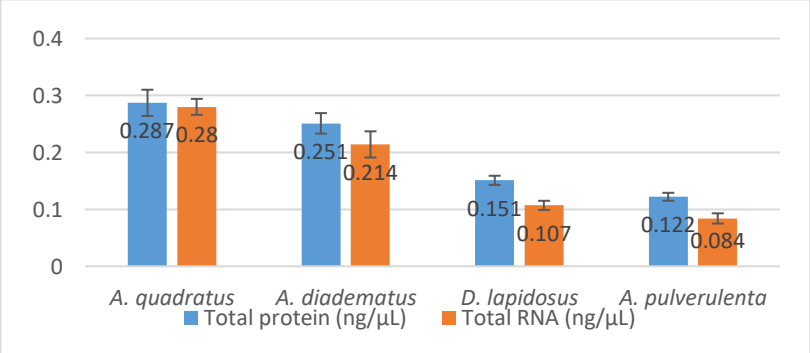


Figure 1. Total protein and RNA amounts of spider species

RNA analysis is important in determining the responses of organisms to different environmental conditions at the molecular level. RNA analysis provides preliminary information in determining the properties of proteins produced in organisms with different living conditions.

Saravanan, (2006) gave information about the structure and properties of spider web in his study. According to this information, spider web consists of 80% water, amino acids, glycoproteins, fats, salts and molecular weight compounds. The remaining 20% consists of filaments. The amount of glycoproteins in the filaments is determined by gene expression. This information shows that high amino acid and high protein synthesis is required to produce a network. The information obtained as a result of our study is in parallel with these data.

İde et al., (2011) determined the characteristic features of spider webs in their study. In this study, it was revealed that spider webs are of different types such as hiding, feeding, resting, protecting eggs and hunting. In this study, *A. diadematus* spider was used to make webs for hunting and total protein content was determined. The determination of total protein is in parallel with our study.

In this study, in order to obtain preliminary information on the transcriptional and translational expression and gene and protein size of web-building and non-web-building spider species with different ecological niches and different hunting strategies, total RNA and total protein levels were compared and similarities or differences between different species at the molecular level were determined by biochemical analysis. For this purpose, two spider species with ecological niches were selected.

As a result of the study, it was observed that the total protein ratio of web building spider species was higher than that of spider species that hunted without building webs. In addition, it was determined that the total mRNA ratio was higher in the web building spider species than the others. As a result, it was concluded that spider species that hunt by making webs do more transcription and translation than spider species that hunt without making webs. This result reveals that web-building spider species synthesize more proteins and express more genes than non-web-building spider species.

CONCLUSION

In conclusion, it was observed that spider species with different hunting strategies produced more RNA and protein transcriptionally and translationally than spider species with different hunting strategies. It is known that various factors have an effect on the amount of total RNA and therefore total protein. In this study, it has been demonstrated that when a spider has the capacity to make a web, it performs more transcription in order to produce the web, which has a protein structure, and as a result, it produces more protein.

ACKNOWLEDGEMENTS

This study was supported by the Niğde Ömer Halisdemir University Scientific Research Projects Coordination Unit (grantnumber: FEB 2017/33-HIDEP, managed by Hakan DEMİR).

REFERENCES

Beccaloni J 2009. Arachnids. University of California Press, Berkeley Los Angeles.

Chen YF, Zhang AY, Zou AP, Campbell WB and Li P-L 2004. Protein methylation activates reconstituted ryanodine receptor Ca^{2+} release channels from coronary artery myocytes. Journal of Vascular Research, 41(3):229-240.

Chomczynski P and Sacchi N 1987. Single-step method of RNA isolation by acid guanidinium thiocyanate-phenol-chloroform extraction. Analytical Biochemistry, 162(1):156-159.

Cimato TR, Ettinger MJ, Zhou X and Aletta JM 1997. Nerve Growth Factor–specific Regulation of Protein Methylation during Neuronal Differentiation of PC12 Cells. The Journal of Cell Biology, 138(5):1089-1103.

Clarke S 2003. Aging as war between chemical and biochemical processes: protein methylation and the recognition of age-damaged proteins for repair. Ageing Research Reviews, 2(3):263-285.

Foelix RF 2011. Biology of Spiders. Oxford University Press, London, UK.

İde S, Bayarı SH, Türkeş T, Mergen YO, Çelik Ö, Bütün V, Sargon MF, Kocatepe N and Kriechbaum M 2011. Structural characterization of a variety of spider silks from Turkey using different biophysical techniques. Journal of Spectroscopy, 25(3-4):155-167.

Kujubu DA, Stimmel JB, Law RE, Herschman HR and Clarke S 1993. Early responses of PC-12 cells to NGF and EGF: Effect of K252a and 5′-methylthioadenosine on gene expression and membrane protein methylation. Journal of Neuroscience Research, 36(1):58-65.

Lowry OH, Rosebrough NJ, Farr AL and Randall RJ 1951. Protein measurement with the Folin phenol reagent. Journal of Biological Chemistry, 193(1):265-275.

Saravanan D 2006. Spider silk-structure, properties and spinning. Journal of Textile and Apparel, Technology and Management, 5(1):1-20.

Vemuri R and Philipson KD 1988. Protein methylation inhibits Na+-Ca2+ exchange activity in cardiac sarcolemmal vesicles. Biochimica et Biophysica Acta (BBA)-Biomembranes, 939(3):503-508.

DRAFT

ORAL PRESENTATION

In vitro biological effects of some antagonist bacteria against potato common scab disease

Nida ÜNLÜ[1*] (*https://orcid.org/0000-0002-0740-4425),* Adem BOZKURT[2] (*https://orcid.org/0000-0002-4826-0317*), Eminur ELÇİ[1*] (https://orcid.org/0000-0002-6434-6321)

[*1]Niğde Ömer Halisdemir University, Faculty of Agricultural Sciences and Technologies, Department of Plant Production and Technologies, Niğde, TÜRKİYE
[2]Hatay Mustafa Kemal University, Faculty of Agriculture, Department of Plant Protection, Hatay, TÜRKİYE

*Corresponding author e-mail: eminur.elci@ohu.edu.tr

Abstract

Potato common scab is one of the most common diseases of potato, worldwide. There are some pesticides available but due to their side effects on human health and other living organisms, there is a need for more sustainable alternative ways. kUsing antagonistic bacteria may offer an alternative way to harmful pesticides. In this study, we aimed to determine the biological control activity of antagonistic bacteria isolates using the agar diffusion method against *Streptomyces scabiei* (N131). As a result of this study; Kn1c isolate was identified as a *Paenarthrobacter aurescens* by MALDI-TOF MS analysis. It was shown 29 mm inhibiton zone and 9.77 inhibition zone index. In future prospects, *Paenarthrobacter aurescens* may be proposed as a potential biological agent against potato common scab.

Keywords: Potato, common scab, antagonism, MALDI-TOF MS

INTRODUCTION

Potato is one of the most important food sources that fulfill the nutritional needs of the world's population. It is a rich source of protein, carbohydrates and various other vitamins and minerals. Pests and diseases are the key biotic factors that cause huge annual yield losses in potato crop. Among several diseases, Potato common scab, caused by *Streptomyces spp.* is indigenous to all potato growing areas in the world (Loria et al. 2006; Dees & Wanner 2012). A few species of *Streptomyces* can cause potato common scab but *Streptomyces scabiei* is considered to be predominant (Wanner, 2006). The disease has little impact on total potato yield but spoils the appearance, quality, and marketability of the potato tubers (Loria et al. 2006). For management of potato common scab, cultural practices (irrigation during tuber growing stage), crop rotation (Loria et al. 2006; Dees & Wanner 2012), physical, mechanical, organic and biological control methods have been used.
Nowadays, synthetic pesticides have been widely used for to management of pests and diseases. The heavy usage has damaged human health, ecological balance, and animals. For this reason, ecological and biological management methods have become important against chemical control. One of the most important methods is the use of biological control by antagonistic bacteria. Biological control including environmentally friendly applications is one of the most important disease control methods. The advantages of synthetic pesticides include a mix of active ingredients (Bedmutha et al. 2011), low mammalian toxicity and non-persistence in fresh water and soil (Isman 2000). Some studies have reported the antibacterial effect of nonpathogenic *Streptomyces* sp. or other nonpathogenic bacteria against potato common scab (Han *et al*., 2005; Hiltunen *et al*., 2009; Meng *et al*., 2013; Wanner *et al*., 2014; Arseneault *et al*., 2015; Chen et al., 2017; Lin et al., 2018; Karagöz et al., 2018; Shuang et al., 2020, Cui et al., 2022).

In this study; we isolated a biocontrol agent from soil. Biological control of obtained candidate isolates was detected by disc diffusion methods. The aim of this study was to evaluate the antibacterial activity of nonpathogenic bacteria obtained from soil against potato commons scab disease caused by *Streptomyces spp.*

DRAFT

MATERIALS AND METHODS

Pathogenic Bacterial Culture and Inoculum Preparation

Pathogenic S. scabies isolates N131 which were previously isolated from scab lesions on potato tubers grown in Niğde province. Pathogenicity of these isolates was confirmed using the pathogenicity test such as Potato slice test and raddish seedling. In previous work, pathogen isolate N131 was characterized by thaxomin production and potato tuber pathogenicity test.

Isolation of candidate antagonistic bacteria

Surveys for the isolation of antagonistic bacteria were carried out in different districts in Niğde province, Turkey. 200–300 g of rhizosphere soil from serious free PCS-infected area was collected at the potato planting period. All samples taken during the survey studies were individually labeled and placed in transparent polyethylene bags. The samples were kept in ice boxes until they were brought to the laboratory. Antagonistic bacteria were isolated from the healty potato tuber around. Suspensions with soil samples (10 g) and 90 mL of Nutrient Broth media (NB) were mixed in a shaker incubator at 27°C for 20 min, and diluted to 5 times. The diluted samples (100 µL) were then incubated on King B (KB medium) at 27°C for 48 h. After 48 h, bacteria with different sizes and morphologies appeared on the plates, and were individually isolated from single colonies and purified in KB medium.

Pathogenicity test of candidate antagonistic bacteria

Isolated antagonistic bacteria was tested by tobacco hypersensitivity (HR) test and potato pectolytic activity test. For the tobacco hypersensitivity (HR) pathogenicity test: the tobacco leaves with the help of a sterile needle, and bacterial solutions were injected between the veins of the tobacco leaf (*Nicotiana tabacum*) with a sterile injector. Sterile pure water was used as negative control. The isolates that produced necrosis were evaluated as HR positive (+), and those that did not form were evaluated as HR negative (–). For the potato pectolytic activity pathogenicity test; disease free potato tubers were sliced approximately 5 mm and placed in petri dishes. Then all isolates were inoculated on the potato slices. It was used the *Pectobacterium carotovorum* pv. *carotovorum* as a positive control and steril water used as negative control. Petri dishes was incubated at ±26°C. After 24-72 hours, they were checked whether there was softening and they were evaluated as positive in case of softening.

Evaluation *in vitro* antibacterial activity of antagonistic bacteria

In vitro antibacterial activity of the candidate antagonistic bacteria was evaluated by disc diffusion methods with the determination of inhibition zones. Pathogen strain was grown on Yeast Malt Extract Agar (YME) for one week at 28 ± 2 ºC. Then one loop spore was transferred to YME broth and incubated in same conditions at 200 rpm. After the incubation period, bacterial cells were harvested by centrifugation at 8000 x g for 10 min and were rinsed twice with sterile distilled water (sdH2O). Bacterial density was adjusted to ~ 1 X 106 cfu ml-1 with serial dilutions. Candidate antagonistic strains were grown on Nutrient Agar (NA) medium for 24 h, at 26 ± 2 ºC. Single colony was transferred to Nutrient Broth (NB) and incubated in same conditions at 200 rpm. Then bacteria were harvested by centrifugation at 5000 x g for 5 min and were rinsed twice with sdH_2O. Bacterial density was adjusted to OD600 = 0.5 with sdH2O. Inhibitory effects of the candidate antagonistic strains were defined with agar disc diffusion method (Kimura *et al*., 1998). Briefly, 50 µl of pathogen solution was pipetted on YME agar and was spread with sterile swab. Then 10 µl of candidate bacteria solution was pipetted on the 6 mm sterile disc, placed on center of the pathogen inoculated YME. The plates were incubated for one week at 28 ± 2 ºC. After the incubation period, inhibition zones around the discs were recorded. The antagonistic activity of the bacteria was calculated by subtracting the colony diameter from the inhibition zone diameter. The assays were independently repeated three times.

Identification of antagonistic bacteria

All candidate isolates were diagnosed firstly with MALDI-TOF MS according to the method described by Duman and Soylu (2019). The bacterial mass taken from the pure colonies of the isolates and obtained from the colonies developed for 24–36 h on Tryptic Soy Agar (TSA) medium was added to Eppendorf tubes with 300 µl of sterile distilled water. Tubes were agitated in the vortex, and the bacteria suspension was obtained. After adding 900 µl of pure ethanol (Merck, Darmstadt, Germany) into the tubes, the mixture was centrifuged at 13.000 rpm for 2 min. Supernatant discharged and the remaining pellet was centrifuged for 1 min at 13.000

rpm again. If ethanol residue was observed, the remaining ethanol was carefully removed with a pipette, and the resulting pellet was allowed to dry at room temperature (about 5 min). 30 µl of 70% formic acid (Merck, Darmstadt, Germany) was added into the tube containing dry pellet and vortexed again at 13.000 rpm for 1 min, then 30 µl of acetonitrile was added, and the mixture was vortexed at 13.000 rpm for 1 min. Finally, 1 µl of the süpernatant was added to the target plate in two replicates, followed by drying at room temperature (approximately 3–5 min), and addition of 1 µl HCCA Matrix on each sample point (α-Cyano-4-hydroxycinnamic acid) and drying. Brukers Bacterial Test Standard (Bruker Daltonics GmbH, Bremen, Germany) was used as Mass calibration standard. Samples were loaded onto the MALDI-TOF MS (Microflex LT; Bruker Daltonics GmbH, Bremen, Germany) with the Flex Control Software (Bruker Daltonics GmbH,Bremen, Germany). Each spectrum was obtained with 500 laser beams with the minimum laser power required for ionization of samples in automatic mode. The spectrum was analyzed at m/z (mass/charge) ratio in the range of 2–20 kDa. BIOTYPERTM 1.1 software (Bruker Daltonics GmbH, Bremen, Germany) was used as microorganism library. Four isolates, showing highly antagonistic effects in vitro, were selected for in vivo tests. Molecular identification of these four isolates were also performed using universal primers 27F and 1492R. Genomic DNA was extracted using the genomic DNA Purification Kit (GeneJET; Thermo Scientific Fermentas, Vilnius, Lithuania) according to the manufacturer's instructions. PCR assays were performed using universal primers 27F 5'-AGA GTT TGATCMTGG CTC AG-3' and 1492R 5' TAC GGY TAC CTT GTT ACG ACTT designed by Fredriksson et al. (2013) which produced a fragment of 1494 bp. The sequences were compared with those available in the NCBI Blast analysis (http:// www. ncbi. nlm. nih. gov/). In vitro, antibacterial activity was estimated using formula:

(ZD-DD)/ID×100

ZD: Inhibition zone diameter

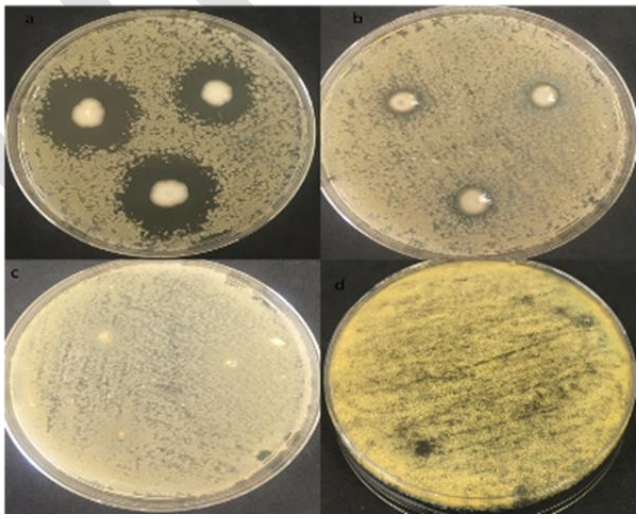
DD: Disc diameter

Statistical analysis

All data were replicated three times and analysis of variance (ANOVA) was done by using SPSS software. The mean values were compared with Duncan multiple range test at a probability of 5% ($P<0.05$).

RESULTS and DISCUSSION

A total of 50 bacterial isolates obtained from soil samples of potato rhizosphere soil showed antagonistic activity to S. scabies (Table 1). Among these isolates, 15 isolates showed pathogenic character according to tobacco hypersensitivity (HR) and potato pectolytic activity pathogenicity test. Totally 27 isolates showed antibacterial effect. Among these isolates, Kn1c isolate showed a higher antibacterial effect as compared with the other isolates. Kn1c isolate exhibited the strongest antagonistic activity towards the growth of S. scabies



and the growth inhibition zone 29 mm and 9,77 in inhibition zone index (Figure 1). According to the MALDI-TOF MS diagnostic results of Kn1c isolates, it was found *Paenarthrobacter aurescens* (Table 1).

Figure 1. Antibacterial effect of Kn1c against *Streptomyces scabiei*

Several studies have used biocontrol agents as; *Pseudomonas* spp. and *Bacillus* spp. to management of the potato common scab. Han et al. 2005, isolated the antagonistic bacteria and Streptomyces scabiei isolate from soil and identified the beneficial bacteria as a Bacillus sp. It was found decreasing the infection rate from 75 to 35% using by pot assay. Meng et al. 2012, isolated the antagonistic bacteria potato common scab suppressive soil, was identified as *Bacillus amyloliquefaciens* (BAC03). Antagonistic activity of BAC03 was displayed against *Streptomyces spp.* According to pot assay, BAC03 reduced common scab severity and potentially increased the growth of potato plants. Karagöz et al. 2018, detected the biological control effect of 1048 bacterial isolates from former studies were tested against *S. scabiei* in-vitro and 22 out of these strains were found to be effective. Among this isolates, four strains (KBA-10, K-19B, TV-91C and RK-92) showed highest antibacterial effect and those were chosen for field trials. It was identified as *Bacillus zhangzhouensis* by 16s general primers pairs by PCR analysis. According to field assay; KBA-10 was found most effective biocontrol agent for reducing potato scab disease and enhancing crop yield. Cui at al. 2022, were isolates 75 endophytic bacteria from the healthy potato tubers and strain 3-5 was selected as an optimal antagonistic bacterium against *Streptomyces griseoplanus* (*Streptacidiphilus griseoplanus*) using by the dual-culture method. Strain 3-5 was identified as *Bacillus amyloliquefaciens* based on its morphological characteristics and *16S rDNA* general genes. It was found successful in controlling potato scab with a 38.90 ± 3.2140% by field assay.

Isolate Number	Isolate Code	Result of the MALDİ-TOF analysis	MALDİ-TOF Similarity İndex	İnhibition Zone index
1	T1c****	Bacillus cereus	2,1	8,94 ^{b}
2	T3d	Bacillus cereus	1,89	5,55^{b}
3	T8ı	Bacillus cereus	1,76	2,26^{def}
4	T11e	Bacillus cereus	2,12	3,78^{c}
5	T12k	Bacillus cereus	1,93	2,33^{def}
6	T15o	Pseudomonas koreensis	2,21	6,19^{b}
7	T15ö	Pseudomonas koreensis	2,04	5,55^{b}
8	T17d	Leclercia adecarboxylata	1,74	2,55^{cdef}
9	T28a	Lysinibacillus pakistanensis	2,04	3,18^{cdef}
10	T36c	Bacillus cereus	2,03	3,18^{cdef}
11	T39d	Bacillus cereus	1,97	3,38^{cdef}
12	T46c	Bacillus cereus	1,86	3,48^{cde}
13	T47b	Pseudomonas aeruginosa	2,46	2,08^{f}
14	T51e	Bacillus cereus	1,72	2,58^{cdef}
15	T53b	Enterobacter cloacae	2,26	2,34^{def}
16	T53a	Bacillus cereus	2,02	3,04^{cdef}
17	T55c	Bacillus cereus	2,25	2,67^{cdef}
18	T55f	Bacillus cereus	2,12	5,23^{b}
19	T70c	Bacillus cereus	1,97	3,19^{cdef}
20	T76a	Paenarthrobacter aurescens	2,25	2,44^{cdef}
21	Kn1a	Bacillus mycoides	1,92	2,17^{d}
22	Kn1b	Paenarthrobacter aurescens	2,22	3,55^{cd}
23	Kn1c***	Paenarthrobacter aurescens	2,1	9,77^{a}
24	Kn1ı	Pseudomonas aeruginosa	2,38	2,83^{cdef}
25	Kn2c	Paenarthrobacter aurescens	2,05	5,93^{b}
26	Kn2e	Bacillus cereus	1,73	2,64^{cdef}
27	Kn2f	Siccibacter turicensis	1,44	2,83^{cdef}

Table 1. MALDI-TOF MS diagnostic test results of antagonistic bacteria isolates

CONCLUSION

Potato common scab is an important soil-borne disease, which occurs in most potato growing areas of the world. In this study, we evaluated the antibacterial activity of antagonist bacteria against potato common scab disease.

In this study, we used the Streptomyces scabiei as a pathogenic isolate. Then antagonistic bacterial samples were isolated from rhizosphere of healthy potato tubers. A totally 27 isolates showed antagonistic effect against the pathogen by disc diffusion test.
According to petri assay, Kn1c isolate showed highest inhibition zone and index then other isolates, 29 mm and 9,77 respectively.
Kn1c isolate was identified as *Paenarthrobacter aurescens* by MALDI-TOF MS analysis.

As a result of this study, *Paenarthrobacter aurescens* has highest antibacterial effect on potato common scab. In summary, the tested antagonistic bacteria isolate especially *Paenarthrobacter aurescens* seem to be promising tools for the control of potato common scab. Future works need to be confirmed these findings through the application of the studied isolates in field experiments. In addition, future experiments are needed to test genomic characterization and biological control mechanism as well.

ACKNOWLEDGEMENTS
This study was supported by TÜBİTAK 1002 project (121O224). The current research was produced by the Doctoral Thesis of Nida ÜNLÜ that was carried out at Niğde Ömer Halisdemir University Niğde, Turkey.

REFERENCES

Arseneault, TC Goyer and Filion M 2015. *Pseudomonas fluorescens* LBUM223 Increases Potato Yield and Reduces Common Scab Symptoms in the Field. Phytopathology. 105(10): 1311-1317.

Cui L, Yang C, Wang Y, Ma T, Cai F, Wei L and Tang M 2022. Potential of an endophytic bacteria Bacillus amyloliquefaciens 3–5 as biocontrol agent against potato scab. *Microbial pathogenesis*, *163*, 105382.

Chen S, Zhang M, Wang J, Lv D, Ma Y, Zhou B and Wang B 2017. Biocontrol effects of Brevibacillus laterosporus AMCC100017 on potato common scab and its impact on rhizosphere bacterial communities. *Biological Control*, *106*, 89-98.

Dees MW, A Sletten and A Hermansen 2013. Isolation and characterization of *Streptomyces* species from potato common scab lesions in Norway. Plant Pathology. 62(1): 217-225.

Han JS, JH Cheng, TM Yoon, J Song, A Rajkarnikar, WG Kim, ID Yoo, YY Yang and JW Suh 2005. Biological control agent of common scab disease by antagonistic strain *Bacillus* sp sunhua. J. Applied Microbiology. 99(1): 213-221.

Hiltunen LH, T Ojanpera, H Kortemaa, E Richter, MJ Lehtonen and JPT Valkonen 2009. Interactions and biocontrol of pathogenic *Streptomyces* strains co-occurring in potato scab lesions. J. Applied Microbiology. 106(1): 199- 212.

Karagoz K, Dadasoglu F, Mohammadi P and Kotan R 2018. Screening bacterial antagonists to common scab disease. *JAPS: Journal of Animal & Plant Sciences*, *28*(4).

Lin C, Tsai CH, Chen P Y, Wu CY, Chang YL, Yang YL and Chen Y L 2018. Biological control of potato common scab by Bacillus amyloliquefaciens Ba01. *PLoS One*, *13*(4), e0196520.

Loria R, Kers J, Joshi M 2006. Evolution of plant pathogenicity in Streptomyces. *Annual Review of Phytopathology*. 44, 469–487.

Meng Q, LE Hanson, D Douches and JJ Hao 2013. Managing scab diseases of potato and radish caused by *Streptomyces* spp. Using *Bacillus amyloliquefaciens* BAC03 and other biomaterials. Biological Control. 67(3): 373- 379.

Shuang M, Wang Y, Teng W and Jin G 2022. Isolation and identification of an endophytic bacteria Bacillus sp. K-9 exhibiting biocontrol activity against potato common scab. *Archives of Microbiology*, *204*(8), 483.

Wanner L A, WW Kirk and XS Qu 2014. Field efficacy of nonpathogenic *Streptomyces* species against potato common scab. J. Applied Microbiology. 116(1): 123-133.

DRAFT

ORAL PRESENTATION

Investigation of *Arenaria kotschyana*'s Biological Activity

Züleyha ASLAN ERGENEKON[1*] (*ORCID: https://orcid.org/0000-0002-5078-2317)*, Cihan DÜŞGÜN[1] (*ORCID: https://orcid.org/0000-0003-2796-8356*)

[*1]Nigde Ömer Halisdemir University, Faculty of Arts and Sciences, Department of Biology, Niğde, Türkiye

*Corresponding author e-mail: zuleyhaaslan@ohu.edu.tr

Abstract

The aim of this study is to investigate the biological activities of *Arenaria kotschyana* samples grown in middle Anatolia using different solvents. For this purpose, plant samples were extracted using ethanol, methanol and water. DPPH and CUPRAC methods were used to determine their antioxidant properties. Total phenolic content was also measured. According to the results obtained, the best antioxidant property was observed in the methanol extract with 69.54±0.14% for DPPH method. For CUPRAC method, 154.98±2.34 mg TE/g was observed in water extract. Furthermore, antioxidant properties were determined for each solvent. The highest total phenolic content was observed in methanol extract with 84±1.79 mg GAE/g. Our results suggested that the tested *A. kotschyana* could be valuable as source of natural agents in the development of health-promoting applications.

Keywords: Antioxidant, biological activity, total phenol, *Arenaria kotschyana*

INTRODUCTION

Plants remain significant sources of bioactive ingredients. Many secondary metabolites require extensive research to determine their potential as therapeutic agents. Recently, drug repositioning has emerged as an avenue for developing new therapeutic agents, and natural products have great potential in this field as they have been used for medical purposes for millennia (Huang and Zhang, 2022). Additionally, several natural compounds have been studied with restricted bioactivities, yet they possess the ability to evaluate new potential effects. Empirically, medicinal herbs have been used by humans with no rational knowledge of their pharmacological effects or active elements, learning by experience alone. In the early 19th century, plant-based rational drug discovery emerged, with an effort to combine the chemical composition with bioactivity (Atanasov et al., 2015). Although biotechnological drugs and monoclonal antibodies carry a significant value, medicinal plants present a distinct chance to explore new drugs through their extraordinary chemical diversity (Atanasov et al., 2021).

Plants are a plentiful source of phytochemicals such as flavonoids, anthocyanins, carotenoids, catechins, cinnamic acid derivatives, chalcones, stilbenes, and tocopherols, which have potential health benefits through their antioxidant properties (Rammohan et al., 2020). These phytochemicals have the ability to target multiple molecules and may serve as valuable agents for promoting health, especially considering that the pathophysiology of many illnesses involves multiple factors rather than a single cause (Efferth and Koch, 2011). Product safety necessitates careful consideration of pharmaceutical formulation, extraction, and manufacture, as well as the mode of action (Sut et al., 2016). To this end, medicinal plants should be scrutinised to discover potential antioxidants or enzyme inhibitors that could be employed as nutraceuticals, functional foods, or medications.

Arenaria genus belongs to the Caryophyllaceae family. The genus has 306 species that are distributed worldwide (Anonymous, 2023). *Arenaria* taxa are annual or perennial herbs, rarely spiny subshrubs, and often caespitose or mat-forming plants found in Eurasia, America, and North Africa. Their leaves lack stipules. The flower clusters, which can have as few as one or as many as fifty blooms, are found at the top or sometimes

also on the sides of the stem. The shape of the leaves can range from round to slender and pointed. The flower clusters, which can have as few as one or as many as fifty blooms, are found at the top or sometimes also on the sides of the stem. The clusters can be arranged in a branching panicle or a group of clusters. The flowers have five sepals. The petals are white and can be any of the following: free, herbaceous, scarious, or coriaceous. They may have a single midrib or one that is more prominent than the lateral veins, although sometimes they have an equally three-veined midrib. They may have a single midrib or one that is more prominent than the lateral veins, although sometimes they have an equally three-veined midrib. Entire or rarely subemarginate. They possess ten stamens, of which the outer five possess obsolete, single, or bipartite basal glands along with three styles. The capsule opening displays six teeth or occasionally six valves. The seeds are present inside. *Arenaria* can be either black or reddish, but the external morphological characteristics make distinguishing between the species a challenging task (Doğan and Çakır, 2019).

Hence, this study attempted to explore the phytochemical composition activity and investigate the antioxidant capabilities of *A. kotschyana*. Additionally, it also aimed to study the pharmacological potential of *A. kotschyana* against oxidative stres. It is also one of the aims of the study to determine the total phenolic content.

MATERIALS AND METHODS

The aerial parts of the *A. kotschyana* species were collected during Summer 2020 from Central Anatolia Region of Turkey. The plant specimens were dried under shaded conditions at room temperature for approximately one week. Subsequently, the pulverisation process was carried out employing a mill, and the specimens were stored out of the light.

Extracts were prepared using ethanol, methanol, and water. The plant material (10 g) with 100 mL of solvents (ethanol, methanol, and water) was extracted with the soxhlet extractor for 6 hours. Finally, solvents were evaporated from the mixtures. Plant materials (10 g) were extracted in 100 mL of boiling water for 15 min before being filtered. Water extracts were lyophilized. All extracts were stored at 4°C until further analysis.

The total phenolic contents were determined using the Folin–Ciocalteu. The results were presented as gallic acid equivalents (mg GAEs/g dry extract) for the assays (Selamoglu et al., 2017).

Antioxidant assays were performed using methods that have been previously reported (Selamoglu et al., 2017). The antioxidant potential was calculated as follows: 2,2-diphenyl-1-picrylhydrazyl (DPPH), cupric-reducing antioxidant capacity (CUPRAC) mmol TE/g extract in the.

The studies were conducted in 3 parallels and the results were averaged.

RESULTS AND DISCUSSION

The antioxidant activities of *A. kotschyana* extracts were evaluated using different assays. In our model, DPPH assays were used to assess radical quenching ability, while CUPRAC assays results indicated reducing power. This two test was useful to establish the total antioxidant capacity. The results are presented in Table 1.

Table 1. Results of biological activity

Species	Solvents	DPPH (%)	CUPRAC (mg TE/g)	Total Phenolic Content (mg GAE/g)
A. kotschyana	Ethanol	58.75±0.25	109.85±2.11	70.25±2.10
	Methanol	69.54±0.14	134.52±1.58	84±1.79
	Water	61.87±0.21	154.98±2.34	74±2.05

Values are reported as mean ± SD of three parallel experiments. TE: Trolox equivalent, GAE: Gallic acid equivalent

The findings showed that a correlation exist between DPPH and CUPRAC assays. Such a correlation was observed in another study (Bibi Sadeer et al., 2020). The methanolic and aqueous extracts exhibited the highest antioxidant activities in most of the assays, suggesting an important role of the phenolics of the extracts. The polarity-dependent increase in total antioxidant activity and reducing properties indicates that the extraction of strong antioxidant compounds is more favorable in polar solvents (Nawaz et al., 2020). Furthermore, in a

previous paper that highly polar solvents, such as methanol, can have a high effectiveness in the extraction of antioxidants (Altemimi et al., 2017), supporting our findings.

When the results were examined, the highest DPPH removal was observed in the extract in which methanol was used as solvent (69.54±0.14%). This result was followed by water extract (61.87±0.21%) and ethanol extract (58.75±0.25%). When the results of CUPRAC method were analyzed, the highest antioxidant property was observed in water extract (154.98±2.34 mg TE/g). In addition to these results, methanol extract (134.52±1.58 mg TE/g) and ethanol extract (109.85±2.11 mg TE/g) were found. It is known that antioxidant properties are related to phenolic substances in plant extracts. In this respect, the highest total phenolic content was obtained in methanol extract (84±1.79 mg GAE/g). This was followed by water extract (74±2.05 mg GAE/g) and ethanol extract (70.25±2.10 mg GAE/g).

As a result, differences in the biological activity of the extracts prepared with different solvents were observed. One of the reasons for these differences is the excess of biologically active compounds contained in the solvent. In addition, the ability of solvents to solubilize bioactive substances in plant samples varies with the solvent.

CONCLUSION

In this study, the biological activities of *A. kotschyana* extracts were compared using different solvents. This is the first study to investigate the biological activity of extracts prepared using this plant. As a result of this study, it was determined that the extracts prepared using ethanol, methanol and water have biological activities. In addition, it was observed that phenolic substance content was obtained from 3 different solvents. This research presents valuable preliminary data on a member of *Arenaria* genus. However, further investigations such as in vivo bioavailability and toxicity studies need to be performed in the future, before projecting the plant for possible nutraceutical/functional food and/or pharmaceutical applications.

REFERENCES

Altemimi A, Lakhssassi N, Baharlouei A, Watson DG and Lightfoot DA 2017. Phytochemicals: Extraction, isolation, and identification of bioactive compounds from plant extracts. Plants, 6(4):42-51.

Anonymous, *Arenaria* L., https://wfoplantlist.org/plant-list/taxon/wfo-4000002959-2023-06?page=1, 11 October 2023

Atanasov AG, Waltenberger B, Pferschy-Wenzig EM, Linder T, Wawrosch C, Uhrin P, Temml V, Wang L, Schwaiger S and Heiss EH 2015. Discovery and resupply of pharmacologically active plant-derived natural products: A review. Biotechnology Advances, 33(8):1582-1614.

Atanasov AG, Zotchev SB, Dirsch VM and Supuran CT 2021. Natural products in drug discovery: Advances and opportunities. Nature Reviews Drug Discovery, 20(3):200-216.

Bibi Sadeer N, Montesano D, Albrizio S, Zengin G and Mahomoodally MF 2020. The versatility of antioxidant assays in food science and safety—Chemistry, applications, strengths, and limitations. Antioxidants, 9(8):709-721.

Doğan C and Çakır N 2019. Pollen morphology of the genus *Arenaria* L. (Subgenus Arenaria)(Caryophyllaceae) in Turkey. Pakistan Journal of Botany, 51(6):2225-2235.

Efferth T and Koch E 2011. Complex interactions between phytochemicals. The multi-target therapeutic concept of phytotherapy. Current Drug Targets, 12(1):122-132.

Huang B and Zhang Y 2022. Teaching an old dog new tricks: Drug discovery by repositioning natural products and their derivatives. Drug Discovery Today, 27(7):1936-1944.

Nawaz H, Shad MA, Rehman N, Andaleeb H and Ullah N 2020. Effect of solvent polarity on extraction yield and antioxidant properties of phytochemicals from bean (*Phaseolus vulgaris*) seeds. Brazilian Journal of Pharmaceutical Sciences, 56:e17129.

Rammohan A, Bhaskar BV, Camilo Jr A, Gunasekar D, Gu W and Zyryanov GV 2020. In silico, in vitro antioxidant and density functional theory based structure activity relationship studies of plant polyphenolics as prominent natural antioxidants. Arabian Journal of Chemistry, 13(2):3690-3701.

Selamoglu Z, Dusgun C, Akgul H and Gulhan MF 2017. In-vitro antioxidant activities of the ethanolic extracts of some contained-allantoin plants. Iranian Journal of Pharmaceutical Research: IJPR, 16(Suppl):92-96.

Sut S, Baldan V, Faggian M, Peron G and DallAcqua S 2016. Nutraceuticals, a new challenge for medicinal chemistry. Current Medicinal Chemistry, 23(28):3198-3223.

DRAFT

ORAL PRESENTATION

Bilişsel gelişim ve laktasyon

Ahmet Murat Günal[1*] (https://orcid.org/0000-0001-9109-1080), Hande Öngün Yılmaz[2] (https://orcid.org/0000-0002-3497-567X)

[*1]İstanbul Okan Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, İstanbul, Türkiye.

[2]Bandırma Onyedi Eylül Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Balıkesir, Türkiye.

*Sorumlu yazar e-mail: ahmetmurat.gunal@okan.edu.tr

Özet

Bebek ve çocukların bilişsel gelişimi, genetik ve çevresel faktörlerden etkilenmektedir. Bilişsel gelişimin sağlanmasında gebelik dönemi dahil olmak üzere yaşamın ilk yıllarında çocuğun yeterli ve dengeli beslenmesi önemlidir. Doğumdan sonra çocuğun kendisi için en ideal besin olan anne sütü ile beslenmesinin bilişsel gelişimi olumlu yönde etkilediği, öğrenme, anlamlandırma, neden-sonuç ilişkisi kurma, görsel ve işitsel hafıza, okuma-yazma ve matematik yetenekleri gibi alanlarda akranlarına göre zeka testi sonuçlarının daha iyi çıktığı bilinmektedir. Anne sütü içeriğinde bulunan birçok madde yenidoğanın beyin gelişimi ile ilişkilendirilmiştir. Bilişsel gelişim potansiyelinde yeterli ve dengeli beslenmenin olumlu etkilere sahip olduğu bilinmektedir. Bilişsel gelişim üzerinde etkisi bilinen beslenme ile ilgili pek çok faktör vardır ve bu faktörler her zaman yeterince açık değildir. Özellikle anne sütü ve bilişsel gelişim üzerine yapılan araştırmalar maternal IQ, sosyokültürel çevre gibi birçok karıştırıcı faktör olduğunu belirtmiştir. Bununla beraber, yeterli süre ve miktarda anne sütü alımının bilişsel gelişim üzerine ılımlı bir pozitif etkisi olabileceğinin altı çizilmiştir.

Anahtar Kelimeler: Anne Sütü, Emzirme, Laktasyon, Bilişsel gelişim, Beslenme.

Cognitive development and lactation

Abstract

The cognitive development of infants and children is affected by genetic and environmental factors. Adequate and balanced nutrition in the first years of life, including the pregnancy period, is prominent in ensuring cognitive development. After birth, the ideal food for infants is breast milk. It has a positive effect on cognitive development. The intelligence test results are better than his peers in areas; such as learning, meaningfulness, cause-effect relationship, visual and auditory memory, reading-writing, and math skills known to come out. Many substances contained in breast milk are associated with the brain development of the newborn. Adequate and balanced nutrition has positive effects on cognitive development potential. There are many nutritional factors known to have an impact on cognitive development. These factors are not always clear enough. Studies on breast milk and cognitive development have stated many confounding factors such as maternal IQ and sociocultural environment. However, adequate time and amount of breast milk intake may have a moderately positive effect on cognitive development.

Key Words: Breast milk, Breastfeeding, Lactation, Cognitive development, Nutrition

GİRİŞ

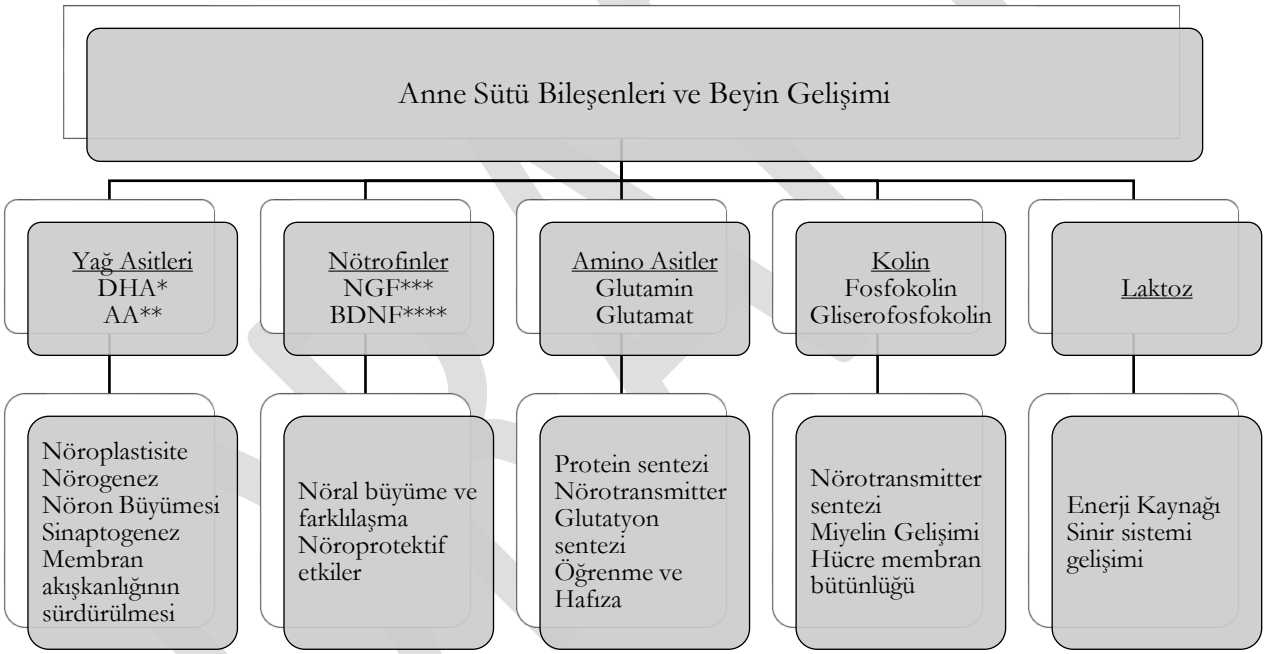
Anne sütü, bebekler için ideal besindir. Güvenli ve temizdir. Birçok yaygın çocukluk hastalığına karşı korunmaya yardımcı olan antikorlar içerir. Anne sütü, bebeğin yaşamının ilk aylarında ihtiyaç duyduğu tüm enerji ve besinleri sağlar. İlk yılın ikinci yarısında beslenme ihtiyacının yarısını veya daha fazlasını, yaşamın ikinci yılında ise üçte birine kadarını karşılamaya devam eder. Emzirilen çocuklar zeka testlerinde daha iyi performans gösterirler. Fazla kilolu veya obez olma olasılıkları daha düşüktür. Hayatlarının ilerleyen

dönemlerinde diyabete daha az meyillidirler. Emziren kadınlarda ayrıca meme ve yumurtalık kanseri riski daha düşüktür. Her beş bebekten üçü yaşamın ilk saatinde anne sütü alamıyor. 0-6 ay arası bebeklerin sadece %41'i sadece anne sütü alıyor. 0-23 ay arası optimal anne sütü alımı yapılırsa 820.000 çocuk kurtarılabilir. Anne sütü İkamelerinin uygunsuz pazarlanması, dünya çapında emzirme oranlarını ve süresini iyileştirme çabalarını baltalamaya devam ediyor. (Breastfeeding, WHO, 2020)

Dünya genelinde 12. ayda anne sütü alımı incelenmek için 1993-2013 yılları arasındaki raporlar araştırıldığında oranların gelişmiş ülkelere kıyasla gelişmekte olan ülkelerde daha yüksek olduğu görülüyor. Yine benzer şekilde düşük ve düşük-orta gelirli ailelere orta-yüksek ve yüksek gelirli ailelere göre, anne sütü alımı, ilk beş ay sadece anne sütü alımı, 12. ayda ve 23. ayda devam eden emzirme oranları daha yüksek bulunmuştur (Victora, ve ark., 2016).

BİLİŞSEL GELİŞİM

Anne sütünün bilişsel gelişim üzerine etkileri ilk kez 1929 yılında araştırılmış ve pozitif yönlü etkinin altı çizilmiştir (Hoefer ve Hardy, 1929). Sonrasında bu konu üzerine birçok araştırma yapılmıştır. Zeka katsayısı (IQ) testleri üzerinde yoğunlaşan çalışmalar emzirme süresi ile testlerden alınan skorların arttığını işaret etmektedir (Horta ve ark., 2015). Anne sütü içeriğinde bulunan birçok madde yenidoğanın beyin gelişimi ile ilişkilendirilmiştir. Bu maddeler ve etki mekanizmaları şekil 1'de özetlenmiştir (Kamini ve ark., 2020).



Şekil 1. Anne Sütü Bileşenleri ve Beyin Gelişimine Etkileri
(*Dokozahekzaenoik Asit, **Araşidonik Asit, ***Sinir Büyüme Faktörü, **** Beyin Kaynaklı Nörotrofik Faktör)

Bir kesitsel çalışma ve 17 kohortun dahil edildiği bir meta analiz sonuçlarına göre, emzirme, zeka testlerinde artmış performans ile ilişkili bulunmuştur (+3.44 puan). Bu ilişki, maternal IQ ayarlandıktan sonra da devam etmektedir. Uzun vadeli takip çalışmaları, emzirmenin okul başarısını ve yetişkinlikteki geliri de etkilediğini işaret etmektedir (Horta ve ark., 2015). 2008-2009 yıllarında doğmuş 1752 çocuk dahil edildiği bir kohort çalışmasında anne sütü alımı ile iletişim, problem çözme, ifade becerisi ve hesap becerisinde anlamlı sonuç çıksa da anne IQ'su üzerinden düzenleme yapılınca sadece problem çözme ve ifade becerisi anlamlı kalmıştır. Çalışma sonucunda bilişsel gelişim anlamında genetik ve sosyo-kültürel, sosyo-ekonomik durumun daha önemli olduğu, anne sütünün belirgin fark yaratmadığı gösterilmiştir (Kim, ve Choi, 2020). 2019'da yapılan anne sütü veya formülayla beslenen çocukların bilişsel gelişimlerinin değerlendirildiği sistematik derlemeye dahil edilen tüm çalışmalarda formülayla beslenen bebeklere göre anne sütü alanlar, anne sütü alanlar arasında ise daha uzun süre alanlar çeşitli testlerden daha yüksek bilişsel beceri skorları aldıkları tespit edilmiş ancak, düzeltici faktörler hesaba katıldıktan sonra sonuçlar anlamlı çıkmamıştır. Çalışma sonucu bilişsel gelişim

açısından anne sütü/formüla ile anne sütü alım periyotundan ziyade maternal IQ, maternal eğitim seviyesi ve sosyoekonomik çerçeve en önemli faktörler olarak belirtilmiştir (Kim, 2019). Girard ve arkadaşları tarafından yapılan bir popülasyon kohort çalışmasına 2007-08 yıllarında doğan 11.134 çocuk dahil edilmiş ve dokuzuncu ay ile üç ve beş yaşlarında değerlendirilmiş, veri kayıpları sonucu son sayı 7.478 kalmıştır. Çalışma diğer anne sütü – bilişsel gelişim çalışmalarında karıştırıcı faktörlerin göz ardı edildiğini öne sürmüştür. 13 alt ölçek açısından kohortu değerlendirdiklerinde tüm ölçeklerde anne sütünün pozitif etkisi gözlemlenmiş, karıştırıcı faktörlere göre düzenleme yapıldığında ise sadece 3. yaşta yapılan değerlendirmede bir faktörde (hiperaktivite) anlamlı sonuç kalmıştır (Girard ve ark., (2017). Couto ve arkadaşlarının 2020 yılında yayınladıkları ve 2015 – 2019 tarihleri arası yapılan sekiz çalışmanın analiz edildiği bütünleştirici derlemede öğrenme, anlamlandırma, neden-sonuç ilişkisi kurma, görsel ve işitsel hafıza, okuma-yazma ve matematik yetenekleri gibi alt ölçekler incelenmiştir. İlk altı ay sadece anne sütü alanlarda IQ, formüla alanlara göre bilişsel gelişim skorları anlamlı derecede daha yüksek (+8.9 puan) bulunmuştur (Couto ve ark., 2020).

Belarus'da yapılan Emzirmeyi Teşvik Müdahale Çalışması (PROBIT) adındaki randomize çalışmaya 31 doğum hastane ve polikliniği çalışmaya dahil edilmiş ve 96-97 yıllarında doğmuş totalde 17.046 anne-çocuk çiftinden 16.491'i 12 aylık süre ile takip edilmiştir. Hastanelerden 16'sı Dünya Sağlık Örgütü (DSÖ) ve Birleşmiş Milletler Çocuk Fonu girişimleriyle "Bebek Dostu Hastane" sıfatı ile müdahale, 15'i geleneksel hastane olarak kontrol grubu kabul edilmiştir. Müdahale grubunda ilk üç ay sadece anne sütü alanların oranı %43 iken kontrolde %6 bulunmuştur ($p<0.001$). İlk altı ay sadece anne sütü alanların oranı ise sırasıyla %7.9 ve %0.6 ($p<0.01$) olarak tespit edilmiştir (Kramer, ve ark., (2001). Probit çalışmasına katılanlardan 13.889'u 6.5 yaşında bilişsel gelişim bakımından değerlendirilmiş ve alt ölçekli IQ testleri ve öğretmen değerlendirmeleri incelenmiştir. Müdahale grubunda, kontrole kıyasla sözel IQ 7.5, performans IQ 2.9 ve total IQ 5.9 puan yüksek çıkmıştır. Müdahale grubunun öğretmen değerlendirmeleri de kontrole göre okuma-yazma becerilerinde anlamlı derecede yüksek bulunmuştur. Uzun süreli ve tek başına anne sütü verilmesi çocukların bilişsel gelişimini iyileştirir sonucuna varılmıştır (Kramer ve ark., 2008). Fakat uzun süreli ve tek başına anne sütü verilmesinin çocuk ve anne davranışları üzerinde risk veya faydasına dair kanıt bulunamamıştır. Bununla beraber müdahale grubunda olanların sonraki çocuklarını ilk üç ay sadece anne sütü ile beslemesi, kontrol grubuna göre iki kat daha olası bulunmuştur. Buradan da "Bebek Dostu Hastane" uygulamasının uzun dönemli pozitif etkilerinin de olduğu sonucuna varılmıştır (Kramer, ve ark., 2008). PROBIT çalışmaları term doğan sağlıklı bebekleri incelemiştir. 2017 yılında yayımlanan başka bir derleme ise PROBIT çalışmalarına ek olarak preterm doğumlarda anne sütünün etkisi ele alınmıştır. Hem term hem de preterm popülasyonlarda kanıtlar, emzirmenin veya anne sütü ile beslenmenin çocuk nörolojik gelişimine fayda sağladığına dair ikna edici bulunmuştur. Bu faydalar, formülaya kıyasla anne sütündeki besin öğeleri farklılıklarından kaynaklanabilir ve anne-bebek etkileşimlerindeki farklılıklar da işin içinde olabilir çıkarımı yapılmıştır. Genel olarak, nörolojik gelişimsel faydalara ilişkin mevcut kanıtlar, bebeklerin ilk altı ay boyunca sadece anne sütüyle beslenmesi ve hastaneye yatırılan preterm bebeklerin takviye edilmiş anne sütü alması gerektiğine dair mevcut önerileri desteklemektedir (Belfort, 2017).

Jing ve arkadaşları tarafından yapılan çalışmada 120 sağlıklı term çocuk, ilk altı ay anne sütü, süt bazlı formüla ve soya bazlı formüla ile beslenenler olarak üç gruba ayrılmış, her gruba 20 kız, 20 erkek çocuk dahil edilmiş ve üç, altı, dokuz ve 12. aylarda elektroensefalogram (EEG) ölçümlerine bakılmıştır. EEG aktivitesi ile miyelinizasyon hızlarına bakıldığında süt ve soya bazlı formüla ile beslenenler paralel özellik gösterirken anne sütü alanlar farklılık göstermiştir. Sonuçlara göre; formüla ile beslenenler anne sütü grubuna göre daha yüksek EEG aktivitesi görülmüş ve miyelinizasyon hızında sırasıyla altıncı ve dokuzuncu ay olmak üzere pik noktasına ulaşıldığı tespit edilmiştir. Yetişkinlik çağı nörolojik hastalıklar için geç pik noktasına ulaşmanın koruyucu olabileceği ifade edilmiştir (Jing ve ark., 2010).

Birleşik Krallıkta yapılan ve 2000 – 2001 yılları arasında, Birleşik Krallıkta doğan 11.544 çocuğun dahil edildiği "Millenium Cohort Study" adlı çalışma Koh tarafından analiz edilmiş, düşük test sonucuna sahip çocukların daha az emzirildiği ve bu test sonuçlarına göre 20. persentilden aşağı olanlar için anne sütünün etkisi, 80. persentilden yukarı olanlara göre iki kat fazla bulunmuştur. Emzirmeyi teşvik edici kamu politikalarının benimsenmesinin çocukların bilişsel gelişimindeki eşitsizliği azaltabileceği vurgulanmıştır (Koh, 2017).

Gibson ve Porter tarafından yapılan, emzirme döneminde maternal alkol ve sigara kullanımının çocuğun akademik başarısı ile ilişkisinin araştırıldığı çalışmaya 5.107 anne-çocuk çifti dahil edilmiş ve 2004'ten itibaren her iki yılda bir değerlendirmeye tabii tutulmuştur. Emzirme döneminde yüksek dozda alkol alan annelerin çocuklarında 7-10 yaş aralığında akademik başarıda düşüş gözlemlenmiştir. Sonuç olarak, laktasyon döneminde alkol tüketiminin doza bağlı olarak bilişsel gelişim ve akademik başarıyı olumsuz etkileyebileceği vurgulanmış, sigara tüketimi ile ilgili ise herhangi bir sonuca varılamamıştır (Gibson ve Porter, 2020).

DRAFT

SONUÇ

Anne sütü çocuk gelişimi için en önemli besindir. Birçok araştırma ilk altı ay tek başına ve ardından iki yaşa kadar ek besinlerle alınan anne sütünün, çeşitli kronik hastalıklara karşı koruyucu olduğunu göstermektedir. Bilişsel gelişim konusunda da anne sütünün önemi çeşitli boyutlarda araştırılmıştır. Fakat zeka ve bilişsellik konularında etkisi tam olarak hesaplanamayan karıştırıcı faktörler vardır. (Maternal IQ, maternal eğitim seviyesi, sosyokültürel ortam vb.) Bu konuda bilimsel açıdan kanıt niteliği yüksek randomize kontrollü çalışmalar yapmak çoğunlukla etik dışı kabul edilmiştir. Bu sebeple bilişsel gelişim üzerine anne sütünün doğrudan etkisini saptamak zor olsa da literatür ılımlı pozitif etki olabileceğini işaret etmektedir.

KAYNAKLAR

Belfort, M. B. 2017. The science of breastfeeding and brain development. Breastfeeding Medicine, 12(8), 459-461.

Breastfeeding, WHO, https://www.who.int/health-topics/breastfeeding, Erişim Tarihi: 03.01.2023

Couto G.R, Dias V, Oliveira I.J. 2020. Benefits of exclusive breastfeeding: An integrative review. Nursing Practice Today, 7(4):245-254

Gibson, L., & Porter, M. 2020. Drinking or smoking while breastfeeding and later academic outcomes in children. Nutrients, 12(3), 829.

Girard, L. C., Doyle, O., & Tremblay, R. E. 2017. Breastfeeding, cognitive and noncognitive development in early childhood: a population study. Pediatrics, 139(4), e20161848.

Hoefer C, Hardy MC. 1929. Later development of breast fed and artically fed infants: comparison of physical and mental growth. J Am Med Assoc., 92:615-619.

Horta, B. L., Loret de Mola, C., & Victora, C. G. 2015. Breastfeeding and intelligence: a systematic review and meta-analysis. Acta paediatrica, 104, 14-19.

Jing, H., Gilchrist, J. M., Badger, T. M., & Pivik, R. T. 2010. A longitudinal study of differences in electroencephalographic activity among breastfed, milk formula-fed, and soy formula-fed infants during the first year of life. Early human development, 86(2), 119-125.

Kamini, D., Amrita, K., Preeti, C. G., & Sadhana, J. 2020. Breast Milk Components and Neurodevelopment of Children. Indian Journal of Public Health Research & Development, 11(8).

Kim, K. M., & Choi, J. W. 2020. Associations between breastfeeding and cognitive function in children from early childhood to school age: a prospective birth cohort study. International breastfeeding journal, 15(1), 1-9.

Kim, S. 2019. A systematic review of the effects of b cognitive development am. Research Journal of Recent Sciences, 8(2), 60-64.

Koh, K. 2017. Maternal breastfeeding and children's cognitive development. Social Science & Medicine, 187, 101-108.

Kramer, M. S., Aboud, F., Mironova, E., Vanilovich, I., Platt, R. W., Matush, L., ... & Collet, J. P. 2008. Breastfeeding and child cognitive development: new evidence from a large randomized trial. Archives of general psychiatry, 65(5), 578-584.

Kramer, M. S., Chalmers, B., Hodnett, E. D., Sevkovskaya, Z., Dzikovich, I., Shapiro, S., ... & Shishko, G. 2001. Promotion of Breastfeeding Intervention Trial (PROBIT): a randomized trial in the Republic of Belarus. Jama, 285(4), 413-420.

Kramer, M. S., Fombonne, E., Igumnov, S., Vanilovich, I., Matush, L., Mironova, E., ... & Platt, R. W. 2008. Effects of prolonged and exclusive breastfeeding on child behavior and maternal adjustment: evidence from a large, randomized trial. Pediatrics, 121(3), e435-e440.

Victora, C. G., Bahl, R., Barros, A. J., França, G. V., Horton, S., Krasevec, J., ... & Group, T. L. B. S. 2016. Breastfeeding in the 21st century: epidemiology, mechanisms, and lifelong effect. The Lancet, 387(10017), 475-490.

ORAL PRESENTATION

Comparison of Various miRNA Expression Levels in the MJ Mycosis Fungoides Cell Line

Mücahit SEÇME[1*] (*ORCID: https://orcid.org/0000-0002-2084-760X*), Yavuz Dodurga[2] (*ORCID: https://orcid.org/0000-0002-4936-5954)*, Neşe Çallı Demirkan[3] (*ORCID: https://orcid.org/0000-0001-5860-100X*), Nida Kaçar[4] (*ORCID: https://orcid.org/0000-0001-8730-8056)*, Nur Selvi Günel[5] (*ORCID: https://orcid.org/0000-0003-0612-2263)*, İbrahim Açıkbaş[2] (*ORCID: https://orcid.org/0000-0001-7483-1147)*

[1]Department of Medical Biology, Faculty of Medicine, Ordu University, Ordu, Turkey.
[2]Department of Medical Biology, Faculty of Medicine, Pamukkale University, Denizli, Turkey.
[3]Department of Pathology, Faculty of Medicine, Pamukkale University, Denizli, Turkey.
[4]Department of Dermatology, Faculty of Medicine, Pamukkale University, Denizli, Turkey.
[5]Department of Medical Biology, Faculty of Medicine, Ege University, İzmir, Turkey.

*Corresponding author:mehtersecme@gmail.com

Abstract

Mycosis fungoides (MF) is the most common type of Cutaneous T-cell lymphomas (CTCLs). These lymphomas constitute a collection of disorders identified by the existence of malignant clonal CD4+ T lymphocytes within the skin. Although extensive research is underway, the exact pathogenesis of MF still remains partially understood. miRNAs play a vital role in modulating various biological mechanisms related to immune cell function, cell proliferation, and the cancer cell progression processes. The aim of this study is to determine the expression levels of various miRNAs in the MJ mycosis fungoides cell line and compare them with dermal fibroblasts. MJ and BJ cells were cultured under suitable conditions, and RNA isolation was performed using Trizol. Following cDNA synthesis, the expression levels of various microRNAs were determined using Real-Time PCR. When compared to BJ cells, statistically significant downregulation of miR-146a, let-7e, miR-210, miR-424, miR-130a, and miR-200a, as well as upregulation of miR-155 and miR-21, were observed in MJ cells. In conclusion, the differences in miRNA expression levels in this MJ mycosis fungoides cell line will contribute to our understanding of which signalling mechanisms are activated in mycosis fungoides cells and their roles in cell proliferation, progression, and cancer pathways.

Key Words: Mycosis fungoides, miRNA expression, MJ cells

INTRODUCTION

The skin is the most common site of extranodal involvement in lymphoma after the gastrointestinal system. Cutaneous lymphomas are a heterogeneous neoplastic disease group originating from T and B cells, characterized by distinct histopathological, immunological, and clinical features (Willemze et al., 2019). While being the most common lymphoma type when compared to other cutaneous lymphomas, primary cutaneous T-cell lymphomas represent a group of diseases characterized by malignant clonal CD4+ T lymphocytes present in the skin (Keehn et al., 2007).

MF encompasses 60% of cutaneous T-cell lymphomas and 50% of all primary cutaneous lymphomas (Willemze et al., 2019). Clinically, MF is a chronic and slowly progressing disease, typically originating from CD4 (+) helper T cells and presenting in four stages: patch, plaque, tumor, and erythroderma. The prognosis can vary from long-term survival to rapidly progressing fatal outcomes (Kim-James and Heffernan, 2001). The molecular pathogenesis and biological mechanisms of MF have not been fully elucidated yet. Various immunological, genetic, and environmental hypotheses have been proposed; however, recent research has suggested the accepted hypothesis that uncontrolled proliferation of T cells, resistance developed against the apoptosis mechanism known as programmed cell death, and as a result, chronic cutaneous inflammation trigger the development of lymphoma (Min, 2018).

MicroRNAs (miRNAs), approximately 19-24 nucleotides in length, are small non-coding RNAs that function by base-pairing with complementary regions in target mRNAs, thereby suppressing translation or directly leading to mRNA degradation (Bayraktar et al., 2019; Chen et al., 2013). They have been shown to play roles in many biological processes including development, differentiation, homeostasis, apoptosis, immune activation, signal transduction pathways, and cancer progression (Bartel, 2004). Recent research suggests that, in addition to the factors playing a role in the pathogenesis of CTCL, changes in miRNA expression may also be involved (Girardi et al., 2004; Ralfkiaer et al., 2014). Valencak and colleagues have reported that high DICER expression is a negative prognostic factor for CTCL (Valencak et al., 2011). Additionally, it has been noted that in CTCL, the expression of miR-155, miR-21, and miR-199/214 is upregulated, while miR-223 and miR-150 are downregulated. Some miRNAs may play roles in CTCL carcinogenesis by acting as oncogenic or tumor-suppressive factors, influencing cellular events such as proliferation, apoptosis, and invasion (Ralfkiaer et al., 2014; van Kester et al., 2011).

While there is a limited number of studies in the literature regarding miRNAs and their expression changes in CTCL, especially in the context of mycosis fungoides, determining miRNA changes between stages, and conducting comprehensive miRNA profiling studies involving both patients and MF cell lines can provide significant contributions to understanding the pathogenesis of MF (Ralfkiaer et al., 2014). The aim of the study is to determine the expression levels of certain miRNAs in a mycosis fungoides cell line, specifically the MJ cells, and to compare these levels with dermal fibroblast cells.

MATERIALS AND METHODS

Cell Culture

The study was conducted with the approval of the Pamukkale University Non-Interventional Clinical Research Ethics Committee, with the decision of the committee numbered 10 and dated 01.08.2017. In our study, the mycosis fungoides cell line "MJ (ATCC® CRL-8294™) ATCC® Number: CRL-8294" was commercially obtained. The BJ (ATCC® CRL-2522™) dermal fibroblast cell line was also used from our own stocks for the purpose of comparing the results obtained with MJ cells. The MJ cell line is a suspension-cultured T lymphocyte cell line characterized by CD2+; CD3+; CD4+ characteristics and is commonly used as a cell culture model in mycosis fungoides research. The cell culture medium used in our study to sustain the cells' viability and for experiment tracking consists of DMEM supplemented with 1% Sodium Pyruvate, 2mM L-glutamine, 10% Fetal Bovine Serum (FBS), 1% non-essential amino acids, and 100 IU/ml penicillin. The cells were monitored for their growth, confluency status, and characteristics using an inverted microscope (Nikon, Japan). The cultures were maintained under appropriate conditions at 37°C with 95% humidity and 5% CO2 in an incubator.

RNA Isolation, CDNA synthesis and Real-Time PCR

Total RNA isolation from MJ and BJ cells was carried out using the Trizol method. RNA purity and concentration values were determined using Nanodrop. Subsequently, cDNA synthesis was performed using the miRNA cDNA Synthesis Kit with Poly(A) Polymerase Tailing (Abm, Canada) following the protocol provided by the manufacturer. Next, miRNA expression levels were determined using SYBR Green master mix (Eva Green) by StepOne Plus Real-Time PCR (Applied Biosystems). In our study, we investigated 13 miRNAs, namely hsa-miR-146a-5p, hsa-miR-375, hsa-miR-200a-3p, hsa-miR-223-3p, hsa-miR-224-5p, hsa-miR-155-5p, hsa-miR-21-5p, hsa-miR-214-3p, hsa-miR-424-5p, hsa-let-7e-5p, hsa-miR-130a-3p, hsa-miR-210-3p, and hsa-miR-204-5p. U6 was used as a housekeeping miRNA for normalization in our study.

Statistical analysis

The ΔΔCT method was used for the analysis of miRNA expression levels detected by RT-PCR. Changes in expression were determined using the web-based "RT² Profiler™ PCR Array Data Analysis" platform (https://www.qiagen.com/tr/shop/genes-and-pathways/data-analysis-center-overview-page/). Statistical analysis of the expression changes between the two groups was performed within this platform and analyzed using the Student t-test. $p < 0.05$ was considered statistically significant.

DRAFT

RESULTS AND DISCUSSION

According to the obtained results, when comparing the MF cell line MJ with BJ dermal fibroblast cells, statistically significant changes in miRNA expression were observed. In MJ cells, miR-21 showed a 1.27-fold increase, miR-155 exhibited a 1.6-fold increase, while miR-146a had a significant 12.3-fold increase. Additionally, let-7e showed a 7.3-fold increase, miR-210 had a 2.1-fold increase, miR-130a exhibited a 2-fold increase, and miR-200a had a 6.2-fold decrease ($p < 0.05$). However, miR-204 expression did not show any significant change between the groups ($p > 0.05$). In the MJ cell line group, miR-214 demonstrated a 5.4-fold increase, miR-223 showed a 2.6-fold increase, miR-224 had a 3.1-fold increase, whereas miR-424 exhibited an 11.5-fold increase, and miR-375 showed a substantial 20-fold decrease; however, these changes were not statistically significant ($p > 0.05$). The results of the comparison for all miRNAs are demonstrated in Table 1.

Tale 1. Changes in miRNA expression levels when comparing MJ cell line with BJ

	miRNA	**Fold regulation**	**p value (*p<0,05)**
1	***miR-214***	5,4193	0,171957
2	***miR-223***	2,614	0,06719
3	***miR-21***	1,2763	0,000189*****
4	***miR-146a***	-12,3464	0,00025*****
5	***Let-7e***	-7,3415	0,0486*****
6	***miR-155***	1,6753	0,00745*****
7	***miR-210***	-2,1092	0,0473*****
8	***miR-424***	-11,5525	0,65
9	***miR-204***	1,1124	0,056
10	***miR-130a***	-2,0848	0,00025*****
11	***miR-224***	3,1171	0,32
12	***miR-375***	-20,6276	0,1
13	***miR-200a***	-6,2668	0,0036*****

Recent studies have revealed the involvement of not only genes in signalling pathways but also non-coding RNA types, such as miRNAs, in CTCL pathogenesis. It has been shown that miRNAs play a critical role in the progression and pathophysiology of malignancies, and they are abnormally expressed in various cancer types. It has been suggested that miRNAs play a role in the pathogenesis of mycosis fungoides and may serve as a potential marker in MF patients (Lindahl et al., 2018; Ralfkiaer et al., 2011). In our study with MJ cells, we also investigated miR-155 expression, which we found to have a significant increase. In a recent study, they explored the role of miR-155 in vitro and in vivo in MyLa and MJ MF cell lines, Hut78 SS cell lines, a primary cell line derived from human peripheral blood lymphocytes as a control, and a xenograft animal model. It was observed that miR-155 expression was high in MyLA and MJ cells, while it exhibited lower expression in HuT 78 cells compared to the other cells. In the animal model study, it was demonstrated that silencing miR-155 reduced tumor tissue and development in vivo. Thus, it has been suggested that miR-155 could be an effective strategic target in MF pathogenesis and may be associated with fundamental cellular mechanisms related to cell survival (Moyal et al., 2017). While miR-223 expression was found to be increased in our study with the MJ cell line, McGirt and colleagues reported lower miR-223 expression in HuT-78 and HH cell lines when compared with CD+ T cells. Additionally, they noted that miR-223 regulates cell development, inhibits clonogenic potential, and targets proto-oncogenes such as E2F1, MEF2C, and TOX in these cell lines (McGirt et al., 2014). The initial results regarding other miRNAs in MJ cells have been presented in our study and added to the literature.

CONCLUSION

In conclusion, considering the roles of miRNAs in cancer cells as both oncogenes and tumor suppressors, the detection of aberrantly expressed miRNAs can potentially play a role in the diagnosis and treatment of cancer. In dermatogenetic diseases such as mycosis fungoides, where the genetic and pathophysiological underlying causes are not fully elucidated, the expression levels of these non-coding RNAs are important. The determination of miRNA expression levels investigated in our study will guide research into their potential as markers for treatment strategies that can be developed by targeting MJ cells.

ACKNOWLEDGEMENTS

This study was produced from the PhD thesis of M.S. This study was supported by the Scientific Research Coordinatorship of Pamukkale University as the project numbered as 2018SABE004.

REFERENCES

Willemze, R., Cerroni, L., Kempf, W., Berti, E., Facchetti, F., Swerdlow, S. H., & Jaffe, E. S. (2019). The 2018 update of the WHO-EORTC classification for primary cutaneous lymphomas. Blood, 133(16), 1703–1714. https://doi.org/10.1182/blood-2018-11-881268

Keehn, C. A., Belongie, I. P., Shistik, G., Fenske, N. A., & Glass, L. F. (2007). The diagnosis, staging, and treatment options for mycosis fungoides. Cancer control : journal of the Moffitt Cancer Center, 14(2), 102–111. https://doi.org/10.1177/107327480701400203

Kim-James, H. Y., & Heffernan, M. P. (2001). The diagnosis, evaluation, and treatment of cutaneous T-cell Lymphoma. Current Problems in Dermatology, 13(6), 301–340. https://doi.org/10.1067/mdm.2001.119860

Min B. (2018). Spontaneous T Cell Proliferation: A Physiologic Process to Create and Maintain Homeostatic Balance and Diversity of the Immune System. Frontiers in immunology, 9, 547. https://doi.org/10.3389/fimmu.2018.00547

Bayraktar, R., Bertilaccio, M. T. S., & Calin, G. A. (2019). The Interaction Between Two Worlds: MicroRNAs and Toll-Like Receptors. Frontiers in immunology, 10, 1053. https://doi.org/10.3389/fimmu.2019.01053

Chen, X., Liang, H., Zhang, J., Zen, K., & Zhang, C. Y. (2013). microRNAs are ligands of Toll-like receptors. RNA (New York, N.Y.), 19(6), 737–739. https://doi.org/10.1261/rna.036319.112

Bartel D. P. (2004). MicroRNAs: genomics, biogenesis, mechanism, and function. Cell, 116(2), 281–297. https://doi.org/10.1016/s0092-8674(04)00045-5

Girardi, M., Heald, P. W., & Wilson, L. D. (2004). The pathogenesis of mycosis fungoides. The New England journal of medicine, 350(19), 1978–1988. https://doi.org/10.1056/NEJMra032810

Ralfkiaer, U., Lindahl, L. M., Litman, T., Gjerdrum, L. M., Ahler, C. B., Gniadecki, R., Marstrand, T., Fredholm, S., Iversen, L., Wasik, M. A., Bonefeld, C. M., Geisler, C., Krejsgaard, T., Glue, C., Røpke, M. A., Woetmann, A., Skov, L., Grønbæk, K., & Odum, N. (2014). MicroRNA expression in early mycosis fungoides is distinctly different from atopic dermatitis and advanced cutaneous T-cell lymphoma. Anticancer research, 34(12), 7207–7217.

Valencak, J., Schmid, K., Trautinger, F., Wallnöfer, W., Muellauer, L., Soleiman, A., Knobler, R., Haitel, A., Pehamberger, H., & Raderer, M. (2011). High expression of Dicer reveals a negative prognostic influence in certain subtypes of primary cutaneous T cell lymphomas. Journal of dermatological science, 64(3), 185–190. https://doi.org/10.1016/j.jdermsci.2011.08.011

van Kester, M. S., Ballabio, E., Benner, M. F., Chen, X. H., Saunders, N. J., van der Fits, L., van Doorn, R., Vermeer, M. H., Willemze, R., Tensen, C. P., & Lawrie, C. H. (2011). miRNA expression profiling of mycosis fungoides. Molecular oncology, 5(3), 273–280. https://doi.org/10.1016/j.molonc.2011.02.003

Lindahl, L. M., Besenbacher, S., Rittig, A. H., Celis, P., Willerslev-Olsen, A., Gjerdrum, L. M. R., Krejsgaard, T., Johansen, C., Litman, T., Woetmann, A., Odum, N., & Iversen, L. (2018). Prognostic miRNA classifier in early-stage mycosis fungoides: development and validation in a Danish nationwide study. Blood, 131(7), 759–770. https://doi.org/10.1182/blood-2017-06-788950

Ralfkiaer, U., Hagedorn, P. H., Bangsgaard, N., Løvendorf, M. B., Ahler, C. B., Svensson, L., Kopp, K. L., Vennegaard, M. T., Lauenborg, B., Zibert, J. R., Krejsgaard, T., Bonefeld, C. M., Søkilde, R., Gjerdrum, L. M., Labuda, T., Mathiesen, A. M., Grønbæk, K., Wasik, M. A., Sokolowska-Wojdylo, M., Queille-Roussel, C., … Odum, N. (2011). Diagnostic microRNA profiling in cutaneous T-cell lymphoma (CTCL). Blood, 118(22), 5891–5900. https://doi.org/10.1182/blood-2011-06-358382

Moyal, L., Yehezkel, S., Gorovitz, B., Keren, A., Gilhar, A., Lubin, I., Sherman, S., & Hodak, E. (2017). Oncogenic role of microRNA-155 in mycosis fungoides: an in vitro and xenograft mouse model study. The British journal of dermatology, 177(3), 791–800. https://doi.org/10.1111/bjd.15422

McGirt, L. Y., Adams, C. M., Baerenwald, D. A., Zwerner, J. P., Zic, J. A., & Eischen, C. M. (2014). miR-223 regulates cell growth and targets proto-oncogenes in mycosis fungoides/cutaneous T-cell lymphoma. The Journal of investigative dermatology, 134(4), 1101–1107. https://doi.org/10.1038/jid.2013.461

DRAFT

ORAL PRESENTATION

Utilization of Agrobacterium-Mediated Transformation Protocol for Enhanced Genetic Improvement of Tomato (*Solanum lycopersicum* L.)

Qurat ul ain Sajid[1] (*ORCID: http://orcid.org/0000-0002-6101-2975*), Eminur ELÇI[1*] (*ORCID: http://orcid.org/0000-0002-6434-6321*)

[1]Department of Plant Production and Technologies, Faculty of Agricultural Sciences and Technologies, Niğde Ömer Halisdemir University, Niğde, Turkey

*Corresponding author e-mail: eminur@gmail.com

Abstract

Agrobacterium mediated transformation protocol were employed to Rio Grande tomato cultivar with few modifications to develop the transformants. Transformation efficiencies were evaluated by checking different parameters including plant regeneration efficiency, percent transformation efficacy, co-cultivation, co-infection times, concentrations of 6-benzylaminopurine (BAP) in regeneration selection media, gibberellic acid (GA_3), and indole butyric acid (IBA) on shooting and rooting MS media. For tomato genetic transformation, *Agrobacterium tumefaciens* strain Eha-105 containing the desired gene was used. Eha-105 cells expressing the gene were streaked on LB plates with 50 gmL^{-1} kanamycin and incubated at 28°C for 48 hours. Three different explants of various ages were selected for transformation including cotyledons, cotyledon nodes, and hypocotyl. The highest percent regeneration efficiency was found in 14 days old cotyledonary nodes (74.68%). Kanamycin concentration was standardized, and 50 mgL^{-1} was shown to be optimal in terms of explant lethality. The highest percent regeneration efficiency was found for 48 hours of co-cultivation time (26.68%). The different concentrations of BAP in regeneration selection media significantly affected regeneration efficacy. Higher percent regeneration efficiency (15.82%) was observed at 1.5 mg L^{-1} of BAP. The highest healthy shoot frequency was obtained at 0.2 mg L^{-1} of GA_3. The highest frequency (47.60%) of rooting was observed for media having 0.3 mg L^{-1} of IBA concentration. Molecular analysis of T0 transgenic tomato plants revealed effective integration as well as increased relative expression of the gene. All these results indicated that an effective concentration of plant growth regulators is vital in the development of an efficient transformation Procedure for tomato transformation, ultimately leading to the establishment of improved tomato genotypes.

Keywords: Tomato, Rio Grande, IBA, BAP, Transformation.

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is an essential crop that is recognized for both its daily consumption and agricultural importance (Pozueta-Romero et al., 2001). The Rio Grande cultivar of tomato has received significant attention due to its distinctive traits and genetic potential. For improving the characteristics of this cultivar, such as resistance to biotic and abiotic stressors, gene characterization, and the production of useful proteins, genetic modification has emerged as an essential method (Goel et al., 2010; Khare et al., 2010). Utilizing the genetic potential of Rio Grande tomatoes requires the development of an effective and dependable transformation technique. Agrobacterium-mediated transformation stands out as a leading technique for establishing stable genetic variations because of its adaptability and applicability (McCormick et al., 1986). However, a robust methodology that can consistently produce transgenic plants with desirable features is necessary for a successful transformation.

In this work, we present a thorough investigation into the development and improvement of an Agrobacterium-mediated transformation strategy designed especially for the tomato cultivar Rio Grande. Our research focuses on the need for an approach that accelerates and simplifies the transformation process in order to overcome the difficulties experienced in genetic enhancement initiatives. We intend to build a methodology through a series of rigorous tests and studies that not only increases transformation efficiency but also decreases the time and resources needed for genetic modification (Velcheva et al., 2005).

The findings discussed in this study offer a promising tool for researchers and agriculturalists seeking to advance the genetics of Rio Grande tomatoes. By developing a consistent and robust transformation protocol,

we contribute to the wider aim of harnessing the genetic capability of this cultivar, ultimately promoting tomato production and crop improvement efforts.

MATERIALS AND METHODS

Growing of Tomato (cv. Rio Grande) Plants in Tissue Culture Conditions

To grow Tomatoes (cv. Rio Grande), MS-0 nutrient medium was used to plant. Magenta growth boxes are used to grow the plants. Seeds were used for propagation and 9 seeds were grown in each growth box. Plants were grown at 27 ± 2ºC with a day length of 16 h light, provided by fluorescent tubes in the growth chamber.

MSo Media Preparation

MSo was prepared by adding 2.16 g of MS salt with vitamins. 15 g of sucrose was also added along with 8 g of plant agar. PH should maintain between 5.7-5.8. Seeds have been surfaced sterilized and placed on MS media.

Seed Sterilization

Seeds have been surfaced sterilized to avoid contamination before placing on MSo media by following the protocol. Firstly, rinse the seeds with autoclaved distilled water almost three times. Washed it with 75% ethanol almost 2 times. Rinsed it again with water. 20-30ml water and 1 ml sulcid have been added. Mixed it well and washed it 2-3 times. After that fungicides have been added and washed for 1 min. More water has been added to it and rinsed the fungicide has. 1 ml Tween-20 has been added and washed 3-5 times.

Transfer of desired Gene to Tomato Plants

A modified protocol for tomato transformation have been used by Muneeb et al., 2022. Cotyledon, cotyledon nodes, and hypocotyl explants of tomato cultivar were used for gene transfer when plants were uniformly sized 3-4 nodes. First of all, explants were transferred to Petri dishes containing sterile MSo liquid medium, and A. tumefaciens culture containing pFGC 5941 vector was added and inoculated for 15 minutes with gentle shaking. The inoculated explants were put on co-cultivation media and kept in an incubator chamber at 28° C, for 48 hrs.

Co-Cultivation Media (CCM)

Explants were washed with sterile distilled water and sulcid (Antibiotic) for 20 min after 48 hrs. The explants were then dried on sterile filter paper. The washing process was performed to remove all agrobacterium remaining on the explants. Then the explants were placed on co-cultivation media. The explants were kept in a growth chamber at 24°C, 16 hours light and 8 hours dark.

Regeneration Selection Media (RSM)

To obtain transgenic plants, RSM media was used. Phosphinothricin (PPT) was used for plant selection in gene transfer. Ph should be set to 5.75-5.85 approximately.

Transfer and Selection on the Shoot Media (SEM)

Approximately 25 days later, a shoot formed on the explants. Developed candidate transgenic shoots were then grown in shoot-induction MS media. Ph should be set to 5.75-5.85 approximately.

Transfer and Selection Root Induction Media (RIM)

Approximately 25 days later, shoots formed on the explants. Callus and developed candidate transgenic shoots were then grown in root-induction MS media. Ph should be set to 5.75-5.85 approximately.

Confirmation of Transgenic Plants by Molecular Analyses

The DNA was extracted from the leaves by the Cetyl trimethylammonium bromide (CTAB) extraction method (Doyle and Doyle, 1987) with a few modifications. The extracted DNA was stored at –20 °C.

PCR Assays

According to DNA quantity and quality results, all samples were diluted to a final concentration of 50ng/µl. The PCRs were carried out with 16.8 µl sterilized water, 0.5 µl of 10 µM dNTP mix, 2 µl of 25 mM MgCl2, 2.5 µl of 10X Taq buffer, and 0.5 µl of 10 µM of each primer with 0.20 µl of 5 Unit/µl Taq DNA polymerase (Promega, Madison, WI, USA) and 2 µl of pure DNA. The total final reaction mixture of PCR was 25 µl.

Reactions were incubated at 94 °C for 2 min and following 40 amplification cycles (30 s at 95 °C, 30 s at 55 °C, and 30 s at 72 °C) were performed. The final PCR products were visualized under UV light after electrophoresis on ethidium bromide-stained 1.5% agarose gels.

RESULTS

Surface Sterilization of Seeds

Tomato (cv. Rio Grande) seeds have been arranged and used as the source for plants which would be the future source of explant for callus formation and development of transgenic lines. We have selected the Rio Grande because it is showing large plant produces high yields of large-size red tomatoes.



Figure 1. Different stages of seed germination

Placement of Explants on Co-Infection Media

Cotyledons, cotyledon nodes, and hypocotyl of tomato cultivar were used for gene transfer when plants were uniformly sized 3-4 nodes. First of all, explants were transferred to Petri dishes containing infection media, Acetosyringone and A. tumefaciens culture was added and inoculated for 15 minutes with gentle shaking.



Figure 2. Cutting of explants and Treatment of explants with grown inoculation media

A statistically significant interaction of the seedlings' age and explant type on plant regeneration efficiency was recorded as explained in table1.

Table 1. Effect of seedling age and explant type on percentage plant regeneration of *Solanum lycopersicon* L. cv. Rio Grande.

Seedlings age (d)	Plant regeneration efficiency (%)		
	Cotyledon leaf	Cotyledon nodes	Hypocotyl
7	32.14 bcd	42.30 bc	20.36 def
14	48.42 b	74.68 a	26.17 cde
21	22.24 def	31.43 cd	16.93 def
28	8.99 f	31.44 cd	11.83 ef
LSD $_{0.05}$		**16.76**	

The highest percent transformation efficacy was recorded in cotyledon nodes of 14 days old (74.68%) followed by 7 days old explants from cotyledonary leaves (32.14%**)**. Whereas the lowest percent transformation efficacy was recorded in the hypocotyl of 28 days old (11.83 %)(Table 2)

DRAFT

Table 2. Effect of seedling age and explant type on percentage transformation efficiency of Solanum lycopersicon L. cv. Rio Grande.

Seedlings age (d)	Transformation efficiency (%)		
	Cotyledon leaf	Cotyledon nodes	Hypocotyl
7	15.38	13.79	14.29
14	13.24	17.31	14.71
21	12.90	14.89	13.64
28	8.33	8.70	6.67

Placement of Explants on Co-Cultivation Media (CCM)

Later on, the inoculated explants were placed on co-cultivation media and kept in an incubator at 28°C, for 48 hours.

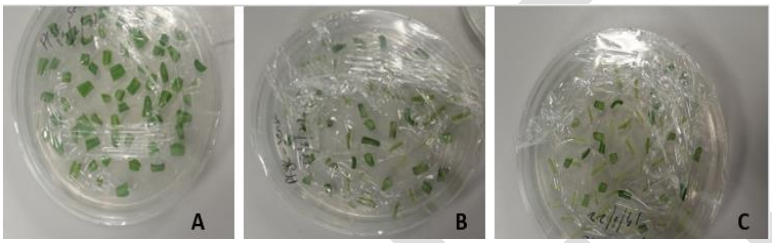


Figure 3. Placement of treated tomato explants on co-cultivation media (A, B, C)

The results indicated that there was a statistically significant co-cultivation time affecting the regeneration efficacy. The highest percent regeneration efficiency was found for 48 hours of co-cultivation time (26.68%) (Table 3).

Table 3. Effect of co-cultivation and co-infection times on percentage transformation efficiency of *Solanum lycopersicon* L. cv. Rio Grande.

Co-cultivation time (hr)	TE (%)	Co-infection time (min)	TE (%)
24	6.21 c	5	11.49 c
48	26.68 a	10	14.52 b
42	13.19 b	15	22.47 a
96	2.73 d	30	4.36 d
LSD $_{0.05}$	**0.96**	**LSD $_{0.05}$**	**1.69**

Regeneration Selection Media (RSM)

After almost 48 hours of infection, explants were washed with sterile distilled water and sulcid for 15-20 min. The explants were then dried on sterile filter paper.

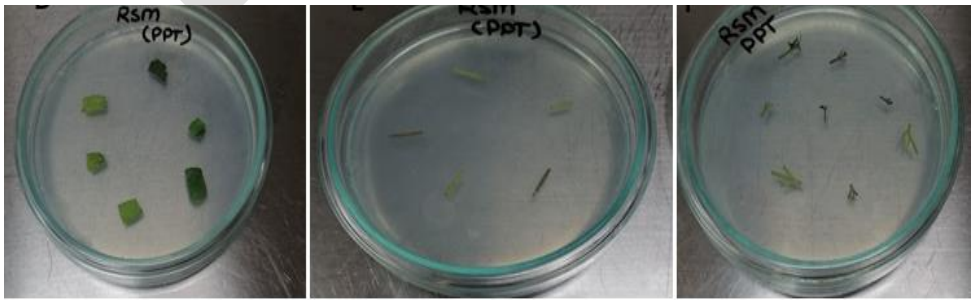


Figure 5. Placement of explants on RSM media containing PPT

Shoot Induction Media (SIM)

Approximately after six weeks, shoots formation started on media plates containing PPT as a selection antibiotic.

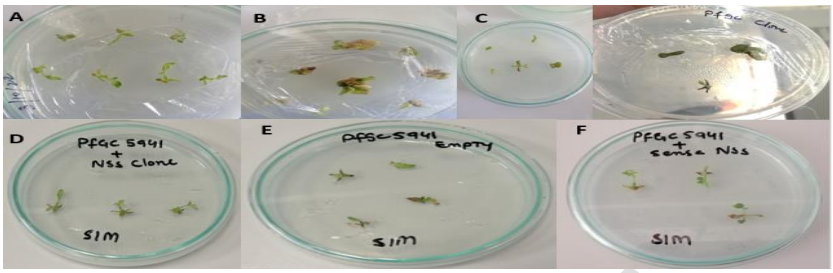


Figure 6. (A,B,C) Shoots formation from hypocotyls, Cotyledon leaves and stems, (D) Shoot formation of explants treated with pfgc+clone construct, (E) Shoot formation of explants treated with pfgc empty construct, (F) Shoot formation of explants treated with pfgc+sense construct.

Shoot Elongation Media (SEM)

Grown shoots were transferred to shoot elongation media containing ppt and sulcid as an antibacterial to avoid contamination.

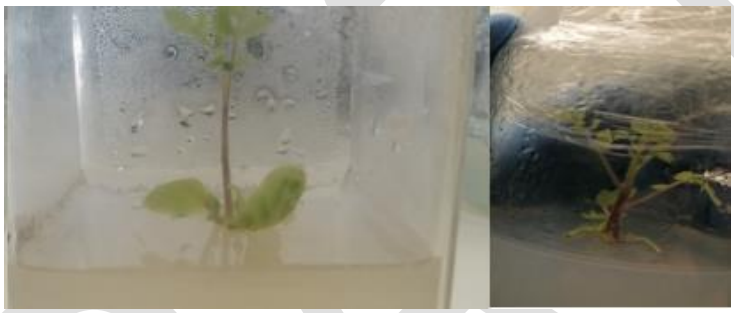


Figure 7. Transfer of grown shoots to shoot elongation media

Root Induction Media (RIM)

After 20-25 days on shoot induction media, plants were shifted to root induction media for root formation containing PPT and sulcid to avoid bacterial contamination.

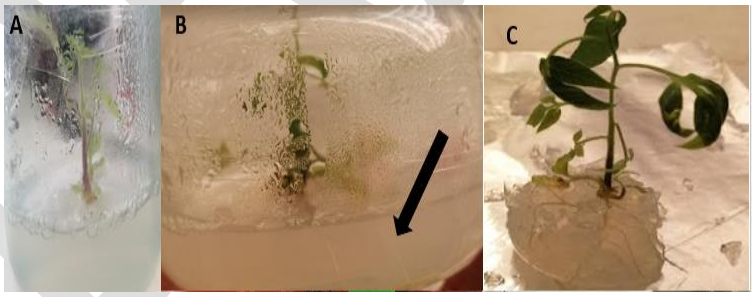


Figure 8. Shifting of growing shoots on rooting media (A,B,C)

A higher percent regeneration efficiency (15.82%) was observed at 1.5 mg L−1 of BAP. The highest healthy shoot frequency was obtained at 0.2 mg L−1 of GA3. The highest frequency (47.60%) of rooting was observed for media having 0.3 mg L−1 of IBA concentration (Table 4).

Table 4. Effect of different growth regulators used in plant regeneration media for calculating transformation efficacy

BAP (mg L^{-1})	Reg (%)	GA3 (mg L^{-1})	Shoot induction (%)	IBA (mg L^{-1})	Rooted shoots (%)
0.5	6.56 d	0.1	54.60 c	0.1	41.68 c
1.0	12.33 b	0.2	78.12 a	0.3	47.60a
1.5	15.82 a	0.4	75.47 b	0.5	43.70 b
2.0	8.65 c	0.6	38.72 d	0.7	29.80 d
LSD $_{0.05}$	**0.84**	**LSD $_{0.05}$**	**1.13**	**LSD $_{0.05}$**	**1.31**

Confirmation of Transgenic Plants by Molecular Analyses

Genomic DNA was taken from tomato plants by using the CTAB method with few modifications. DNA was quantified both with nanodrop and gel electrophoresis for quantity and quality.

PCR Assays

Pure extracted DNA was used and tested by PCR. PCR was performed by BAR primers to confirm transgenic plants.

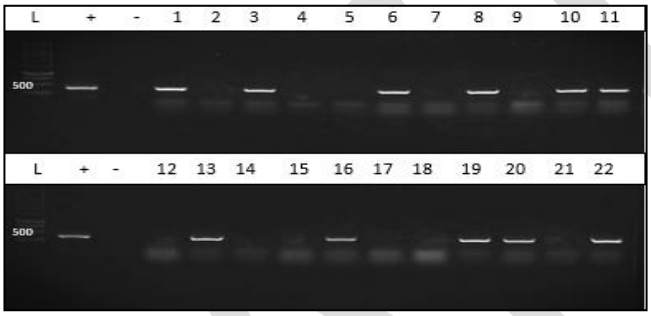


Figure 9. PCR assay for the confirmation of putative transgenic plants by BAR gene primers, Lane L: Gene Ruler 1kbp DNA Ladder (Thermo Scientific), + indicates positive control, - indicates negative control, Sample ranges from 1-22.

DISCUSSION

We have effectively enhanced a transformation technique using agrobacterium for the Rio Grande tomato cultivar, by miner modifications to previously employed methods. These strategies has greatly improved transformation efficiency by methodically assessing critical transformation characteristics, such as plant genotype, explant type, and agrobacterium strain selection (Bakhsh et al., 2014; Cortina and Culiáez-Macià, 2004). The achievement of this goal is vital for the robust development of transgenic tomato lines with desired features, such as viral diseases, insect resistance or tolerance to unfavorable environmental conditions (Madhulatha et al., 2007). Additionally, our streamlined approach minimizes resources while also enhancing efficiency, making it affordable for use in both agricultural and research applications (Davis et al., 1991). Furthermore, this methodology's applicability to additional tomato cultivars broadens its application, enhancing breeding initiatives and genetic research involving a variety of tomato cultivars (Velcheva et al., 2005). This protocol enables scientists and breeders to more effectively integrate beneficial features into the Rio Grande cultivar, resulting in the growth of healthier and more robust tomato crops (Liu, 1999).

Future studies should concentrate on improving the methodology and investigating cutting-edge genetic methods, such RNAi and CRISPR/Cas9-based genome editing, to increase the precision of genetic modification. As a result, our research represents an important step toward utilizing the genetic potential of the Rio Grande tomato cultivar, providing solutions to difficulties tomato growers encounter, and enhancing the security of the world's food supply.

CONCLUSION

Through our research, an effective agrobacterium-mediated transformation strategy was created specifically for the Rio Grande tomato genotype under in vitro conditions was highlighted by the optimization of key limiting parameters. These results provide useful information for enhancing transformation techniques in different tomato cultivars. Our work provides a useful and affordable method for developing agricultural traits

and enhancing tomato productivity and quality, which helps genetic improvement efforts in tomatoes overall for long term uses.

ACKNOWLEDGEMENTS

The current research was produced by the Doctoral Thesis of Qurat ul Ain SAJID that was carried out at Niğde Ömer Halisdemir University Niğde, Turkey.

REFERENCES

Bakhsh A, Anayol E, Ozcan SF 2014. Comparison of transformation efficiency of five Agrobacterium tumefaciens strains in Nicotiana tabacum L. Emirates Journal of Food and Agriculture, 26:259–264.

Cortina C, Culiáñez-Macià FA 2004. Tomato transformation and transgenic plant production. Plant Cell Tissue Organ Culture, 76:269–275.

Davis ME, Lineberger RD, Miller AR 1991. Effects of tomato cultivar, leaf age, and bacterial strain on transformation by Agrobacterium tumefaciens. Plant Cell Tissue Organ Culture, 24:115–121.

Goel D, Singh AK, Yadav V 2010. Overexpression of osmotin gene confers tolerance to salt and drought stresses in transgenic tomato (Solanum lycopersicum L.). Protoplasma, 245:133–141.

Hashmi MH, Saeed F, Demirel U, Bakhsh A 2022. Establishment of highly efficient and reproducible Agrobacterium-mediated transformation system for tomato (Solanum lycopersicum L.). In Vitro Cellular & Developmental Biology-Plant, 58:1066-1076.

Khare N, Goyary D, Singh NK 2010. Transgenic tomato cv. Pusa Uphar expressing a bacterial mannitol-1-phosphate dehydrogenase gene confers abiotic stress tolerance. Plant Cell Tissue Organ Culture, 103:267–277.

Liu LG 1999. The complementation of plant mutants with large genomic DNA fragments by a transformation-competent artificial chromosome vector accelerates positional cloning. Proceedings of the National Academy of Sciences of the United States of America 96:6535–6540.

Madhulatha P, Pandey R, Hazarika P, Rajam MV 2007. High transformation frequency in Agrobacterium-mediated genetic transformation of tomato by using polyamines and maltose in shoot regeneration medium. Physiology and Molecular Biology of Plants,13:191–198.

McCormick S, Niedermeyer J, Fry J 1986. Leaf disc transformation of cultivated tomato (L. esculentum) using Agrobacterium tumefaciens. Plant Cell Reports, 5:81–84.

Pozueta-Romero J, Houlné G, Cañas L 2001. Enhanced regeneration of tomato and pepper seedling explants for Agrobacterium-mediated transformation. Plant Cell Tissue Organ Culture, 67:173–180.

Velcheva M, Faltin Z, Flaishman M 2005. A liquid culture system for Agrobacterium-mediated transformation of tomato (Lycopersicon esculentum L. Mill.). Plant Science, 168:121–130.

ORAL PRESENTATION

Analysis of food preservatives in phyllo, bread, and lavash samples by a novel HS-GC-MS method

Orhan DESTANOĞLU* (*ORCID iD: 0000-0003-2477-0694*)

Istanbul University-Cerrahpasa, Institute of Forensic Sciences and Legal Medicine, Department of Science, 34500, Büyükçekmece, İstanbul, Türkiye

*Corresponding author e-mail: orhan.destanoglu@iuc.edu.tr

Abstract

The incredibly growing food consumption and demand in the world has confirmed the necessity of controlling food additives much more strictly. In Turkey, which is the country that consumes the most bread, lavash and phyllo per capita in the world, the analysis of the preservative content of these products is important for public health. Benzoic acid (BA), sorbic acid (SA), and their salts widely used preservatives for different types of foods inhibit proliferation of the microorganisms by interacting with the membranes, enzymes, proteins, and electron transport chain of the microorganisms. Although the use of BA and SA up to maximum limits do not affect health, excess consumption can cause some health problems such as metabolic acidosis, genotoxicity, convulsions, asthma, urticaria, and hyperpnoea. The main target of this study was to develope a sensitive, precise, and accurate headspace gas chromatography-mass spectrometry (HS-GC-MS) method with minimized chemical waste for determination of BA and sorbic acid SA in bread, lavash and phyllo samples. Esterification of the analytes and IS were conducted in HS vial during heating in HS oven. The validated method was successfully applied to the commercial samples. According to the results of the analyses, BA and SA were not detected except for one of the unbranded open products, while only SA was determined in the concentration range of 135 – 1412 mg/kg in all of the branded packaged products. This method, which was applied to real samples, is advantageous in terms of reducing toxic wastes to be used in routine analysis of BA and SA since it does not require large amounts of solvent as in the commonly used liquid chromatography methods.

Keywords: Preservative, food quality control, headspace gas chromatography, bread, lavash, phyllo.

INTRODUCTION

In every culture, people have tried to preserve foods in different ways since ancient times to prevent and delay changes that may occur in foods. Nowadays, with the increase in food production due to the increasing population in the world, the application of chemical preservatives has gained importance whereas people are correspondingly exposed to these chemicals. Benzoic acid (BA), sorbic acid (SA), and their salts widely used preservatives for different types of foods inhibit proliferation of the microorganisms by interacting with the membranes, enzymes, proteins, and electron transport chain of the microorganisms (Zengin et al. 2011; Hsu et al. 2014; Junqueira De Carvalho et al. 2016; Timofeeva et al. 2019; Bian et al. 2021; D'Amore et al. 2021; Feng et al. 2021; Pereira et al. 2021).

Although additions of BA and SA in the foods up to the maximum permitted concentration values of set by the regulatory agencies are accepted safe for health, excess consumption of the foods containing these preservatives can give rise to poisoning and health problems (Zengin et al. 2011; Hsu et al. 2014; Timofeeva et al. 2019; Bian et al. 2021; Feng et al. 2021; Pereira et al. 2021). Hence, proper controlling and monitoring of the preservatives in foods is crucial for food safety. In the rich cuisine of Türkiye, which ranks first in the world in per capita bread consumption, lavash and phyllo are also widely consumed. So, this work focused on measuring the concentration of BA and SA in bread, lavash, and phyllo samples.

Considering the importance of determination of BA and SA, it is a necessity to use a reliable analytical method. Many analytical methods have been reported for determination of BA and SA in food and beverage samples so far. Among them, high-performance liquid chromatography (HPLC) is the most common utilized technique (Techakriengkrai & Surakarnkul 2007; Zhao et al. 2012; Ozer et al. 2013; Ulca et al. 2013; Xu et al. 2013; Sugiura & Nakajima 2017; Iwakoshi et al. 2019; Timofeeva et al. 2019; Imanulkhan et al. 2020; Bian et al. 2021). Although HPLC is a widespread technique for simultaneous determination of BA and SA, it requires large amounts of toxic organic solvents in mobile phases and in sample pre-treatment processes. Also, some

extraction pretreatments such as LLE, SPE, QuEChERS, and DLLME should be employed prior to chromatographic analyses due to eliminating the matrix interferences (Dong & Wang 2006; Techakriengkrai & Surakarnkul 2007; Sugiura & Nakajima 2017; Iwakoshi et al. 2019; Bian et al. 2021; D'Amore et al. 2021). Furthermore, capillary zone electrophoresis (CZE) (Mato et al. 2006; Feng et al. 2021; Pereira et al. 2021), CE (Zhang et al. 2011; Hsu et al. 2014), capillary ion chromatography with conductivity detection (CIC-CD) (D'Amore et al. 2021), headspace solid-phase microextraction - gas chromatography - flame ionization detector (HS-SPME-GC-FID) (Dong & Wang 2006), thermal desorption gas chromatography mass spectrometry (TD-GC-MS) (Ochiai et al. 2002), GC-MS analysis after SPME (Sagandykova et al. 2017), and flow injection analysis – MS (FIA-MS) are the other developed methods published in the literature.

The main goal of this study was not only to adapt an HS-GC-MS method, which was previously developed for analysis of BA and SA in liquid beverages, to the solid bread, lavash and phyllo samples, but also to reduce amount of chemical waste. The static HS-GC systems are the gold standard for the analysis of volatile compounds since they commonly do not need complicated extraction procedures and provide highly reproducible and reliable results. Yet, BA and SA can not be analyzed directly by HS-GC-MS. Thus, BA, SA, and also salycilic acid (SalA) employed as internal standard (IS) were simultaneously derivatized to their volatile methyl esters with methanol in the vial under optimum HS conditions without applying any other pretreatment. This proposed method is a candidate to be used for routine analyzes in food control laboratories to protect public health.

MATERIALS AND METHODS

Chemicals

All reagents were of analytical reagent grade (≥ 99.0%, Sigma-Aldrich). A New Human Power I Scholar UV system (Human Corporation, Seoul, Korea) was used for obtaining deionized water (≥ 18.2 MΩ cm resistivity).

HS-GC-MS System

A Perkin Elmer Clarus 500 GC equipped with an HS40 autosampler and a Clarus 500 Mass Spectrometer was used for the analyses. Chromatographic separations were carried out by utilizing a Stabilwax-DA GC column (30 m length, 0.25 mm i.d., and 0.5 µm df.). The 22 mL HS vials containing the prepared solutions were kept in the HS oven at 140 °C for 15 min. The temperatures of the HS needle and the transfer line were 145 and 150 °C, respectively. The flow rate of the carrier gas was 1 mL/min. The temperature program of the GC oven was: I) 100 °C for 6 min; II) 20 °C/min to 220 °C; III) holding at 220 °C for 3 min. The temperature of the GC injection port was 250 °C. Total run time was 15 min. The SIR mode was utilized for quantification by selecting *m/z* 67 for SA between 5.5 – 6.5 min, *m/z* 105 for BA between 8.0 – 9.0 min, and *m/z* 120 for SalA 9.5 – 10.5 min. At same time, TIC chromatogram was obtained by performing the full scan mode in the range of 15-250 amu for identifying the analytes.

Sample Preparation

Firstly, 1-2 g of the representative pieces were collected from the samples purchased from the local bakeries, phyllo sellers, and supermarkets in Istanbul, Türkiye. After the samples were dried, they were finely grounded. 0.2 g of the grounded samples were transferred in to 10 mL of volumetric flasks. The samples were mixed and diluted with the solution of methanol:water 65:35 (v/v). It should be noted that the analytes and IS were derivatized to methyl esters since these organic acids could not be detected directly by HS-GC-MS. Optimal HS conditions for esterification reaction are mentioned above section. Therefore, 50 µL of the filtered solutions, 50 µL of the 10 mg/L of IS, and 200 µL of 4.5 M sulfuric acid were mixed in the 22 mL HS vials. After the vials were tightly sealed with PTFE-lined rubber septum caps, they were loaded to the HS autosampler. The shaking option of HS oven was activated.

RESULTS AND DISCUSSION

A static headspace GC-MS method of which parameters optimized in previous study (Destanoğlu 2023) for the analysis of BA and SA in non-alcoholic beverages was adapted to this work for determination of SA and BA in the different bread, lavash and phyllo samples. BA and SA were extracted from the samples using the solution containing 65:35 (v/v) methanol:water. Then, the esterification reaction took place in the presence of sulfuric acid/methanol in the 22 mL HS vial during heating in HS oven. GC oven temperature program and both MS modes were crucial for the selectivity and sensitivity of the method because the matrices of the

samples different and they were diluted 50 times. Although the dilution factor was not very high, sensitivity became important since 50 µL was taken for the vial.

Analytical Figures

Retention time, linearity, limit of detection (LOD), and limit of quantification (LOQ) are summarized in Table 1.

Table 1. Analytical figures of the HS-GC-MS method

Parameter	Analyte	
	BA	SA
Retention time	8.4	5.9
Linear range (mg/L)	2.5 – 5000	12.5 – 5000
Regression equation	y = 2.0602x - 0.4273	y = 0.2060x - 0.0258
R^2	0.9998	0.9998
LOD (mg/L)	1.5	4.5
LOQ (mg/L)	2.5	12.5

Analysis of the Samples

The proposed method was applied to the bread, lavash and phyllo samples for determination of BA and SA. The results are given in Table 2. According to the Turkish Food Codex Regulation on food additives, SA can be added to the samples investigated this study up to 2000 mg/kg while the use of BA is not permitted (Turkish Food Codex 2013). When looking at the Table 2, all samples were suitable in terms of SA amounts, even SA was not detected in 1 commercial branded phyllo (no 4), 3 unbranded open phyllos (no 1,2,3,5), 2 commercial branded breads (no 1,2) and 2 local bakery breads (no 1,2). A SIR chromatogram of the commercial branded phyllo (no 3) is presented in Figure 1.

Table 2. HS-GC-MS analysis results of the bread, lavash and phyllo samples

Sample	SA (mg/kg)	BA (mg/kg)
commercial branded phyllo 1	146 ± 7	N.D.
commercial branded phyllo 2	192 ± 8	N.D.
commercial branded phyllo 3	952 ± 38	N.D.
commercial branded phyllo 4	N.D.	N.D.
commercial branded phyllo 5	238 ± 11	N.D.
unbranded open phyllo 1	N.D.	N.D.
unbranded open phyllo 2	N.D.	N.D.
unbranded open phyllo 3	N.D.	N.D.
unbranded open phyllo 4	134 ± 8	N.D.
unbranded open phyllo 5	N.D.	N.D.
commercial branded bread 1	N.D.	N.D.
commercial branded bread 2	N.D.	N.D.
local bakery bread 1	N.D.	N.D.
local bakery bread 2	N.D.	N.D.
commercial branded lavash 1	1412 ± 49	N.D.
commercial branded lavash 2	1197 ± 56	N.D.

* N.D. : Not detected

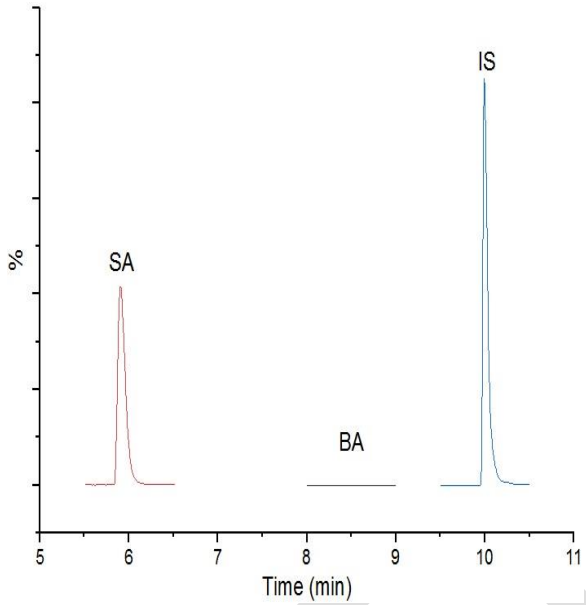


Figure 1. SIR chromatogram of the commercial branded phyllo (no 3)

CONCLUSION

In this paper, BA and SA were investigated in 16 samples consisting of the bread, lavash and phyllo samples by utilizing a validated static Headspace-GC-MS method of which optimal parameters were developed previous study for beverage samples. The baked products could be analyzed directly with the HS-GC-MS, but by 50-fold dilution with the solution containing methanol that was employed as the alcohol for the in-vial derivatization reaction, the measurements were successfully carried out in the linear ranges. It was aimed also to consume the toxic chemicals comparing to the most common HPLC method. Thus, sensitivity became important since 50 µL was taken for the 22 mL HS vial. The results showed that concentrations of BA and SA in all samples investigated in this study were suitable for the legal limits. Noteworthy, in terms of public health, it is an important issue that especially whether local producers use appropriate preservative content in a qualitative and quantitative manner must be strictly controlled. Therefore, it can be concluded that the proposed HS-GC-MS method is a candidate for routine analyses of the preservatives in the authorized laboratories.

REFERENCES

Bian Y, Wang Y, Yu J, Zheng S, Qin F, Zhao L. 2021. Analysis of six preservatives in beverages using hydrophilic deep eutectic solvent as disperser in dispersive liquid-liquid microextraction based on the solidification of floating organic droplet. J Pharm Biomed Anal [Internet]. 195:113889. https://doi.org/10.1016/j.jpba.2021.113889

D'Amore T, Di Taranto A, Berardi G, Vita V, Iammarino M. 2021. Going green in food analysis: A rapid and accurate method for the determination of sorbic acid and benzoic acid in foods by capillary ion chromatography with conductivity detection. Lwt [Internet]. 141(November 2020):110841. https://doi.org/10.1016/j.lwt.2020.110841

Destanoğlu O. 2023. Simultaneous determination of benzoic acid and sorbic acid in non-alcoholic beverages by a validated HS-GC-MS method with reduced waste. Food Addit Contam Part A. 40(7):812–823.

Dong C, Wang W. 2006. Headspace solid-phase microextraction applied to the simultaneous determination of sorbic and benzoic acids in beverages. Anal Chim Acta. 562(1):23–29.

Feng J, Li J, Huang W, Cheng H, Zhang Z, Li L. 2021. Capillary Zone Electrophoresis Determination of Five Trace Food Additives in Beverage Samples Using Counterflow Transient Isotachophoresis. Food Anal Methods. 14(2):380–388.

Hsu SH, Hu CC, Chiu TC. 2014. Online dynamic pH junction-sweeping for the determination of benzoic and sorbic acids in food products by capillary electrophoresis. Anal Bioanal Chem. 406(2):635–641.

Imanulkhan, Setyaningsih W, Rohman A, Palma M. 2020. Development and validation of hplc-dad method for simultaneous determination of seven food additives and caffeine in powdered drinks. Foods. 9(8):1–12.

Iwakoshi K, Shiozawa Y, Yamajima Y, Baba I, Monma K, Kobayashi C, Sasamoto T. 2019. Determination of nine preservatives in processed foods using a modified QuEChERS extraction and quantified by HPLC-PDA. Food Addit Contam - Part A Chem Anal Control Expo Risk Assess [Internet]. 36(7):1020–1031. https://doi.org/10.1080/19440049.2019.1615644

Junqueira De Carvalho L, Cristina Pires Do Rego E, Carius Garrido B. 2016. Quantification of benzoic acid in beverages: The evaluation and validation of direct measurement techniques using mass spectrometry. Anal Methods. 8(14):2955–2960.

Mato I, Huidobro JF, Simal-Lozano J, Sancho MT. 2006. Simultaneous determination of organic acids in beverages by capillary zone electrophoresis. Anal Chim Acta. 565(2):190–197.

Ochiai N, Sasamoto K, Takino M, Yamashita S, Daishima S, Heiden AC, Hoffmann A. 2002. Simultaneous determination of preservatives in beverages, vinegar, aqueous sauces, and quasi-drug drinks by stir-bar sorptive extraction (SBSE) and thermal desorption gc-MS. Anal Bioanal Chem. 373(1–2):56–63.

Ozer H, Psimouli V, Ozcan N, Ozer B, Papadaki I, Oreopoulou V. 2013. Ring trial for the simultaneous analysis of sweeteners and preservatives in soft drinks. Qual Assur Saf Crop Foods. 5(1):71–77.

Pereira LM, Della Betta F, Seraglio SKT, Schulz M, Nehring P, Gonzaga LV, Fett R, Costa ACO. 2021. Assessment of sorbate and benzoate content in mustard, ketchup and tomato sauce by sub-minute capillary electrophoresis. Food Technol Biotechnol. 59(3):376–384.

Sagandykova GN, Alimzhanova MB, Nurzhanova YT, Kenessov B. 2017. Determination of semi-volatile additives in wines using SPME and GC–MS. Food Chem [Internet]. 220:162–167. http://dx.doi.org/10.1016/j.foodchem.2016.09.164

Sugiura J, Nakajima M. 2017. Simultaneous determination of nine preservatives in food by liquid chromatography with the aid of coagulant in the clean-up process. Food Addit Contam - Part A Chem Anal Control Expo Risk Assess [Internet]. 34(5):695–704. https://doi.org/10.1080/19440049.2017.1293302

Techakriengkrai I, Surakarnkul R. 2007. Analysis of benzoic acid and sorbic acid in Thai rice wines and distillates by solid-phase sorbent extraction and high-performance liquid chromatography. J Food Compos Anal. 20(3–4):220–225.

Timofeeva I, Kanashina D, Stepanova K, Bulatov A. 2019. A simple and highly-available microextraction of benzoic and sorbic acids in beverages and soy sauce samples for high performance liquid chromatography with ultraviolet detection. J Chromatogr A [Internet]. 1588:1–7. https://doi.org/10.1016/j.chroma.2018.12.030

Turkish Food Codex. 2013. The Turkish Food Codex Regulation on food additives [Internet]. [place unknown]. https://kms.kaysis.gov.tr/Home/Goster/42410

Ulca P, Atamer B, Keskin M, Senyuva HZ. 2013. Sorbate and benzoate in Turkish retail foodstuffs. Food Addit Contam Part B Surveill. 6(3):209–213.

Xu J, Chen B, He M, Hu B. 2013. Analysis of preservatives with different polarities in beverage samples by dual-phase dual stir bar sorptive extraction combined with high-performance liquid chromatography. J Chromatogr A [Internet]. 1278:8–15. http://dx.doi.org/10.1016/j.chroma.2012.12.061

Zengin N, Yüzbaşioğlu D, Ünal F, Yilmaz S, Aksoy H. 2011. The evaluation of the genotoxicity of two food preservatives: Sodium benzoate and potassium benzoate. Food Chem Toxicol. 49(4):763–769.

Zhang X, Xu S, Sun Y, Wang Y, Wang C. 2011. Simultaneous determination of benzoic acid and sorbic acid in food products by CE after on-line preconcentration by dynamic pH junction. Chromatographia. 73(11–12):1217–1221.

Zhao YG, Chen XH, Yao SS, Pan SD, Li XP, Jin MC. 2012. Analysis of nine food additives in red wine by ion-suppression reversed-phase high-performance liquid chromatography using trifluoroacetic acid and

ammonium acetate as ion-suppressors. Anal Sci. 28(10):967–971.

ORAL PRESENTATION

Anti-inflammatory activity of some brown algae

Damla Kırcı[1*] *(ORCID: https://orcid.org/0000-0002-3479-3999)*, Ayşe Esra Karadağ[2] (*ORCID: https://orcid.org/0000-0002-3412-0807*)

[1]Selcuk University, Faculty of Pharmacy, Department of Pharmacognosy, Konya, Türkiye.
[2] İstanbul Medipol University, Faculty of Pharmacy, Department of Pharmacognosy, İstanbul, Türkiye.

*Corresponding author e-mail: damla.kirci@selcuk.edu.tr

Abstract

Algae is an important food source consumed by humans since ancient times. Macroalgae are classified into green, red, and brown algae. Brown algae, also known as Phaeophyta, comprises 20 classes; Phaeophyceae alone accounts for over 1800 species and 66% of the total algae consumption. Brown algae are a diverse group of marine organisms studied for their potential health benefits, including anti-inflammatory activities. These activities are often attributed to various bioactive compounds present in brown algae.
In this study, *Sargassum vulgare*, *Cystoseira tamariscifolia*, and *Padina pavonica* were collected in Mediterrenian Sea, Türkiye. *n*-Hexane and 70% ethanol extracts of the plants were prepared by maceration method. The *in vitro* anti-inflammatory activity was evaluated by 5-lipoxygenase (5-LOX) inhibitory effect of the *n*-hexane and 70% ethanol extracts of brown algaes spectrophotometrically. The anti-inflammatory activity of the *n*-hexane and ethanol extracts of *C. tamariscifolia* were determined as 60.33±4.4% and 67.58±1.5% in 130 µg/mL, respectively. The highest anti-inflammatory activity was observed in *C. tamariscifolia* extracts. The biological activity of *P. pavonica* extracts was also found to be more effective than *S. vulgare* extracts.

Keywords: Brown algae, anti-inflammatory activity, *Sargassum vulgare, Cystoseira tamariscifolia, Padina pavonica*

INTRODUCTION

Marine macroalgae, including seaweeds, have been recognized for centuries for their nutritional value and health benefits, particularly in Far Eastern countries such as Japan and Korea. Brown algae are the most consumed species, followed by red and green algae. Brown algae are rich in complex polysaccharides, minerals, proteins, vitamins, and phytochemicals, and have been associated with numerous health benefits, including cancer prevention, cholesterol reduction, and weight loss. Regular consumption of seafood, including seaweeds, has been linked to a longer life expectancy. As a result, there is growing interest in the production and consumption of high-value macroalgae-derived products in Western cultures (Lorenzo et al., 2017; Afonso et al., 2019).
Brown algae are also a rich source of bioactive compounds with excellent nutritional value, and are considered valuable food ingredients (Afonso et al., 2019; Li et al., 2021). Brown algae have been used for various purposes, including as a food source and traditional medicine. The brown algae are used in traditional practices for cancer, high cholesterol, thyroid disease and weight loss (Liu et al., 2012; André et al., 2021).
Sargassum vulgare is a brown algae species in temperate and tropical climates belonging to the Phaeophyceae family. These algae can be found primarily in shallow waters and coral reefs. Polyphenols, polysaccharides, phytohormones, carotenoids, vitamins, and unsaturated fatty acids are among the many important metabolites in *S. vulgare* (Marzban et al., 2022). The pharmacological activities of *S. vulgare* are found anticancer, anti-inflammatory, antibacterial and antiviral (Liu et al., 2012).
Cystoseira tamariscifolia has been shown to have antibacterial, antifungal, antiprotozoal, anti-inflammatory, antioxidant, and cytotoxic effects. These properties have been linked to the presence of many classes of compounds found in *C. tamariscifolia*, including phlorotannins (fucophloroethol, fucotriphloroethol, phlorofucofuroeckol, and bieckol) (Vizetto-Duarte et al., 2016; de Sousa et al., 2017).

DRAFT

Padina pavonica is a brown algae (Phaeophyceae) present worldwide and abundant along the Mediterranean and Atlantic coasts. *P. pavonica* extracts revealed many biological activities such as antimicrobial, anti-tumour, anti-osteoporotic, and anti-inflammatory (Abdelhamid et al., 2018; Drira et al., 2021).
In this research, *Sargassum vulgare*, *Cystoseira tamariscifolia*, and *Padina pavonica* were collected in Mediterrenian Sea, Türkiye. *n*-Hexane and 70% ethanol extracts of the plants were prepared by maceration method. The *in vitro* anti-inflammatory activity was evaluated by 5-lipoxygenase (5-LOX) inhibitory effect of the *n*-hexane and 70% ethanol extracts of brown algae spectrophotometrically. Anti-inflammatory activities of brown algae with different polarities were evaluated.

MATERIALS AND METHODS

Plant Material

The aerial parts of the brown algae were collected from Hatay, Türkiye, in 2020. The plant materials were diagnosed by Dr. Cemal Turan, and they were kept at the Herbarium of İstanbul Medipol University in İstanbul, Türkiye.

Extraction Method

The aerial parts of three brown algae were dried in the shade, and the dry stems and leaves were crushed in a blender. The powdered plant materials were then extracted n-hexane (Sigma Aldrich) and ethanol (70%, v/v) (Sigma Aldrich) at room temperature for 24 hours.

Extracts were eventually filtered using filter paper after the maceration step was repeated three times. The combined filtrates were concentrated with a rotary evaporator (Buchi R300) to dryness under reduced pressure at 40°C. The dried extracts were kept at -20°C until they were tested for enzyme inhibition activity. The yields of obtained extracts are given in Table 1.

Lipoxygenase (LOX) Enzyme Activity

Lipoxygenase (1.13.11.12, 7.9 Units/mg) enzyme activity inhibition levels were determined in a 96-well quartz plate, spectrophotometrically (Demirci et al., 2018). The experiments were carried out in 4 replicates, and the results are given in Table 2 as percent inhibition (%). Nordihydroguaiatic acid (NDGA) was used as a positive control.

% Inhibition = (E − S) / E x 100

E: the absorbance of the enzyme without sample.

S: the absorbance of the enzyme with the test sample

Statistical Analysis

The mean and standard deviation of data from *in vitro* enzyme inhibition was calculated (SD). The experiments were done in triplicates and all data were shown as mean ± standard deviation (SD). GraphPad Software Prism 8 (San Diego, CA, USA) was used to determine the % inhibitory impact of the extracts and standard compounds based on the extracts dose-response curve.

RESULTS and DISCUSSION

2.1. Yields of plant extracts

In the present work, the extracts were prepared from aerial parts of brown algae with maceration method. *n*-Hexane and ethanol were used for the extraction. The yields of the extracts were listed in Table 1. The yields of *n*-hexane extracts are lower than the ethanol extracts.

Table 1. Yields (%) of *n*-hexane and methanol extracts of brown algae

Plant	*n*-Hexane	70% Ethanol
Sargassum vulgare	0.54	12.35
Cystoseira tamariscifolia	0.87	8.94

Padina pavonica	0.63	12.12

DRAFT

2.2. Enzyme inhibition activity

The *in vitro* anti-inflammatory activity was evaluated by 5-lipoxygenase (5-LOX) inhibitory effect of the *n*-hexane and 70% ethanol extracts of brown algae spectrophotometrically. The anti-inflammatory activity of the *n*-hexane and ethanol extracts of *C. tamariscifolia* were determined as 60.33±4.4% and 67.58±1.5% in 130 µg/mL, respectively. The highest anti-inflammatory activity was observed in *C. tamariscifolia* extracts. The biological activity of *P. pavonica* extracts was also found to be more effective than *S. vulgare* extracts.

Table 2. Enzyme inhibition activities of brown algae (concentration: 130 µg/mL)

Plant	**Solvent**	**Inhibitory activity against 5-LOX (percentage ± S.D.[a])**
Sargassum vulgare	*n*-Hexane	40.84±4.8
	70% Ethanol	32.75±3.8
Cystoseira tamariscifolia	*n*-Hexane	60.33±4.4
	70% Ethanol	67.58±1.5
Padina pavonica	*n*-Hexane	49.42±1.5
	70% Ethanol	45.55±5.7
NDGA		100±0.0

[a]: Standard deviation

Previous research looked at the phytochemical and medicinal properties of *Sargassum vulgare* collected from the Suez Canal. The anti-inflammatory profile of *S. vulgare* methanol extract was found that COX-1, COX-2, IL6, and TNF activity were 77.39, 88.35, 75.38, and 71.24%, respectively. The study on *S. vulgare* from the Suez Canal found that it contains saponins, known to produce anti-inflammatory effects (Shreadah et al., 2018). Also, phlorotannins, a compound found in *S. vulgare*, have been shown to have anti-inflammatory activity (Chouh et al., 2022).

A study found that methanol extracts from *Cystoseira tamariscifolia* and *C. nodicaulis* were inhibited inflammation (Custodio et al., 2016). According to a review article, the anti-inflammatory properties of the genus *Cystoseira* were mostly attributed to polyphenols and sulfated polysaccharides (Almurshedi et al., 2023). There is no specific study on the anti-inflammatory activity of *C. tamariscifolia*, but a closer look at this species as a source of anti-inflammatory activity was suggested (De La Fuente et al., 2020).

These studies suggest that *C. tamariscifolia* and related species may have anti-inflammatory activity, possibly due to polyphenols and sulphated polysaccharides. However, more research is needed to understand these compounds' mechanisms and potential applications fully.

A study found that the polyphenol-rich fraction from *Padina pavonica* exhibits significant anti-inflammatory activity (Alshaikheid et al., 2019). Another study suggested that the terpenoids-rich dichloromethane extract of *P. pavonia* may exert free radical-scavenging and anti-inflammatory activities (Germoush et al., 2020). While there is no specific study on the anti-inflammatory activity of *P. pavonica*, it has been found to have antimicrobial, antioxidant, and anticancer activities (Al-Enazi et al., 2018; Rushdi et al., 2021). These studies suggest that *P. pavonica* may have anti-inflammatory activity, possibly due to polyphenols and terpenoids. However, more research is needed to understand these compounds' mechanisms and potential applications fully.

CONCLUSION

The brown algae consumption aligns with the increasing awareness of consumers' perceptions of organic and environmentally sustainable products. In this research, the brown algae were investigated anti-inflammatory activity. According to the results, the highest anti-inflammatory activity was observed in *C ystoseira tamariscifolia* ethanol extract. It is especially noteworthy that the efficacy in treating anti-inflammatories is exceptionally high. Therefore, *In vivo*, and clinical anti-inflammatory studies should proceed with the ethanol extract of *Cystoseira tamariscifolia* in the future.

REFERENCES

Abdelhamid A, Jouini M, Bel Haj Amor H, Mzoughi Z, Dridi M, Ben Said R, Bouraoui A 2018. Phytochemical analysis and evaluation of the antioxidant, anti-inflammatory, and antinociceptive potential of phlorotannin-rich fractions from three Mediterranean brown seaweeds. Marine biotechnology, 20: 60-74.

Afonso NC, Catarino MD, Silva AM, Cardoso SM 2019. Brown macroalgae as valuable food ingredients. Antioxidants, 8(9): 365.

Al-Enazi NM, Awaad AS, Zain ME, Alqasoumi SI 2018. Antimicrobial, antioxidant and anticancer activities of *Laurencia catarinensis, Laurencia majuscula* and *Padina pavonica* extracts. Saudi Pharmaceutical Journal, 26(1): 44-52.

Almurshedi AS, El-Masry TA, Selim H, El-Sheekh MM, Makhlof ME, Aldosari BN, Alfagih IM, AlQuadeib BT, Almarshidy SS, El-Bouseary MM 2023. New investigation of anti-inflammatory activity of *Polycladia crinita* and biosynthesized selenium nanoparticles: isolation and characterization. Microbial Cell Factories, 22(1): 173.

Alshaikheid M, Abdelhamid A, Bouraoui A 2019. Physicochemical characterization and pharmacological evaluation of marine polyphenols from the brown algae *Padina pavonica*. Journal of Advanced Research in Biotechnology, 4(1):1-6.

André R, Pacheco R, Bourbon M, Serralheiro ML 2021. Brown algae potential as a functional food against hypercholesterolemia. Foods, 10(2): 234.

Chouh A, Nouadri T, Catarino MD, Silva AM, Cardoso SM 2022. Phlorotannins of the Brown algae *Sargassum vulgare* from the Mediterranean Sea coast. Antioxidants, 11(6): 1055.

Custodio L, Silvestre L, Rocha MI, Rodrigues MJ, Vizetto-Duarte C, Pereira H, Barreira L, Varela J 2016. Methanol extracts from *Cystoseira tamariscifolia* and *Cystoseira nodicaulis* are able to inhibit cholinesterases and protect a human dopaminergic cell line from hydrogen peroxide-induced cytotoxicity. Pharmaceutical Biology, 54(9): 1687-1696.

De La Fuente G, Fontana M, Asnaghi V, Chiantore M, Mirata S, Salis A, Damonte G, Scarfì S 2020. The remarkable antioxidant and anti-inflammatory potential of the extracts of the brown alga *Cystoseira amentacea* var. *stricta*. Marine drugs, 19(1): 2.

de Sousa CB, Gangadhar KN, Macridachis J, Pavao M, Morais TR, Campino L, Varela J, Lago JHG 2017. *Cystoseira* algae (Fucaceae): Update on their chemical entities and biological activities. Tetrahedron: Asymmetry, 28(11): 1486-1505.

Demirci F, Karaca N, Tekin M, Demirci B 2018. Anti-inflammatory and antibacterial evaluation of *Thymus sipyleus* Boiss. subsp. *sipyleus* var. *sipyleus* essential oil against rhinosinusitis pathogens. Microbial pathogenesis, 122: 117-121.

Drira M, Mohamed JB, Hlima HB, Hentati F, Michaud P, Abdelkafi S, Fendri I 2021. Improvement of *Arabidopsis thaliana* salt tolerance using a polysaccharidic extract from the brown algae *Padina pavonica*. Algal Research, 56: 102324.

Germoush MO, Elgebaly HA, Hassan S, Kamel EM, Bin-Jumah M, Mahmoud AM 2019. Consumption of terpenoids-rich *Padina pavonia* extract attenuates hyperglycemia, insulin resistance and oxidative stress, and upregulates PPARγ in a rat model of type 2 diabetes. Antioxidants, 9(1): 22.

Li Y, Zheng Y, Zhang Y, Yang Y, Wang P, Imre B, Wong ACY, Hsieh YSY, Wang D 2021. Brown algae carbohydrates: Structures, pharmaceutical properties, and research challenges. Marine Drugs, 19(11): 620.

Liu L, Heinrich M, Myers S, Dworjanyn SA 2012. Towards a better understanding of medicinal uses of the brown seaweed *Sargassum* in Traditional Chinese Medicine: A phytochemical and pharmacological review. Journal of Ethnopharmacology, 142(3): 591-619.

Lorenzo JM, Agregán R, Munekata PE, Franco D, Carballo J, Şahin S, Lacomba L, Barba FJ 2017. Proximate composition and nutritional value of three macroalgae: *Ascophyllum nodosum, Fucus vesiculosus* and *Bifurcaria bifurcata*. Marine drugs, 15(11): 360.

Marzban A, Mirzaei SZ, Karkhane M, Ghotekar SK, Danesh A 2022. Biogenesis of copper nanoparticles assisted with seaweed polysaccharide with antibacterial and antibiofilm properties against methicillin-resistant *Staphylococcus aureus.* Journal of Drug Delivery Science and Technology, 74: 103499.

Rushdi MI, Abdel-Rahman IA, Saber H, Attia EZ, Madkour HA, Abdelmohsen UR 2021. A review on the pharmacological potential of the genus *Padina*. South African Journal of Botany, 141: 37-48.

Shreadah MA, El Moneam NMA, Al-Assar SA, Nabil-Adam A 2018. Phytochemical and pharmacological screening of *Sargassium vulgare* from Suez Canal, Egypt. Food Science and Biotechnology, 27: 963-979.

Vizetto-Duarte C, Custodio L, Gangadhar KN, Lago JHG, Dias C, Matos AM, Neng N, Nogueira JMF, Barraira L, Albericio F, Rauter AP Varela J 2016. Isololiolide, a carotenoid metabolite isolated from the brown alga *Cystoseira tamariscifolia*, is cytotoxic and able to induce apoptosis in hepatocarcinoma cells through caspase-3 activation, decreased Bcl-2 levels, increased p53 expression and PARP cleavage. Phytomedicine, 23(5): 550-557.

DRAFT

ORAL PRESENTATION

Retinal and Choroidal Mean Gray Value, Skewness and Kurtosis in Different Regions

Onur İNAM[1,2,*,**] (*ORCID: https://orcid.org/0000-0002-4726-1190*), Deniz SOMER[3,***] (*ORCID: https://orcid.org/0000-0003-1487-721X*), Güner Özkan ÜNEY[3] (*ORCID: https://orcid.org/0000-0001-8503-4258*), A. Ruhi Soylu[4,****] (*ORCID: https://orcid.org/0000-0001-8503-4258*)

[*1]Gazi University, Faculty of Medicine, Department of Biophysics, Ankara, Turkey.
[2]Columbia University, Irving Medical Center, Edward S. Harkness Eye Institute, Vagelos College of Physicians and Surgeons, Department of Ophthalmology, New York, NY, USA.
[3] University of Health Sciences, Ankara Training and Research Hospital, Clinic of Ophthalmology, Ankara, Turkey.
[****4]Hacettepe University, Faculty of Medicine, Department of Biophysics, Ankara, Turkey.

*Corresponding author e-mail: onurinam@gazi.edu.tr
****Double Corresponding author e-mail: arsoylu@hacettepe.edu.tr

***Onur İnam has performed this study while he is at Hacettepe University Graduate School of Health Sciences, Department of Biophysics, Ankara, Turkey and his current address is Gazi University, Department of Biophysics, Ankara, Turkey and he is now working as a researcher at the Department of Ophthalmology, Edward S. Harkness Eye Institute, Vagelos College of Physicians and Surgeons, Columbia University Irving Medical Center, New York, New York, USA*
****Deniz Somer has performed this study while she was at the Clinic of Ophthalmology, Ankara Training and Research Hospital, University of Health Sciences, Ankara, Turkey.*

Abstract

In this study we aimed to investigate the optical coherence tomography (OCT) images of healthy subjects for mean gray value (MGV), skewness and kurtosis, at retina and choroid in different regions. For this purpose, 200 OCT images of the 50 subjects (Female/Male is 1:1) retrospectively analyzed. Vertical and horizontal scans are processed for both retinal and choroidal region of interest (ROI) in a 6500 µm area centered to fovea. Images have been binarized using Fiji/ImageJ and Niblack auto local thresholding method. MGV, skewness and kurtosis are measured for entire ROI, 65 parts at 100 µm interval, and also for 3 vertical alignments as inner, middle and outer parts for these ROIs. For entire ROI retinal MGV is 117.2 ± 3.9 and 120.3±5.2 ($p<0.001$) and choroidal MGV is 76.3±6.8 and 78.5±6.7 ($p=0.019$) for vertical and horizontal scans, respectively. Retinal skewness value is 0.16±0.06 and 0.11±0.08 ($p<0.001$) and choroidal skewness value is 0.84±0.14 and 0.88±0.15 ($p=0.024$) for vertical and horizontal scans for entire ROI, respectively. Retinal kurtosis value in entire ROI is -1.97±0.02 and -1.98±0.02 ($p<0.001$) and choroidal kurtosis value is -1.28±0.26 and -1.20±0.29 ($p=0.037$) for vertical and horizontal scans, respectively. Image processing techniques are gaining importance for differentiating the healthy and diseased one, as well as fallowing up the disease progression. Thus, new methods, parameters and indexes could be a useful tool to investigate OCT images and define different properties of structures like retina and choroid. For retina and choroid; MGV, skewness and kurtosis were found to be significantly different in many vertical and horizontal scan regions and could be a candidate for image processing approaches.

Keywords: Biophysics, Image Processing, Retina, Choroid

DRAFT

INTRODUCTION

The retina possesses a fundamental plan that encompasses many neuronal cell classes and exhibits conserved features in the design of the retinal structure and this characteristic allows for the possibility of conducting detailed analysis and making comparisons across different species (Hoon et al., 2014). In order to fulfill specific visual function requirements, it is possible for structural and functional retinal specializations to manifest in various species (Hoon et al., 2014).

The retina, which is situated at the innermost layer of the eye, is a neurosensory tissue that plays a pivotal role in the process of transforming incoming light stimuli into electrical signals that are subsequently capable of being processed and comprehended by the visual system, thereby allowing for the perception and interpretation of visual information (Gupta et al., 2016). The retina, which is located at the optic nerve surrounding, has a thickness of approximately 400 µm; however, as we move towards the ora serrata surrounding, the thickness decreases significantly to around 140 µm (Gupta et al., 2016). These variations in thickness make the optic nerve surrounding the most thick part of the retina, while the ora serrata surrounding is the least thick part (Gupta et al., 2016). Furthermore, at the center of the retina lies the macular area, which is characterized by its circular shape with a radius of 1500 µm. The macular area also exhibits a range in thickness, with values ranging from 350 µm to 180 µm (Gupta et al., 2016). The vertebrate retina is organized into cell layers with microcircuits working together to encode visual information, and the cellular and molecular mechanisms can shape the structure and function of the retina during development (Gupta et al., 2016; Hoon et al., 2014). The structure can be rearranged and function can be disrupted in retinal diseases, and there are studies that try to find approaches to re-establish the functional architecture of the retina. (Gupta et al., 2016; Hoon et al., 2014).

The choroid, a highly specialized vascular tissue responsible for the supply of essential nutrients and oxygen to the outer retina, fulfills a pivotal role in the ocular system (Nickla & Wallman, 2010). Not limited solely to its blood supply function, the choroid exhibits a many other responsibilities, including the regulation of thermal dynamics within the eye, the precise positioning of the delicate retina, and the secretion of crucial growth factors that contribute to the overall health and functioning of the ocular structures (Nickla & Wallman, 2010). This nature of the choroid underscores its significance in maintaining the integrity and optimal performance of the visual system (Nickla & Wallman, 2010). Alongside this multifunctional nature, the choroid, an essential component of the eye, is comprised of a diverse array of anatomical layers, each possessing its own unique characteristics and functions (Ferrara et al., 2016; Nickla & Wallman, 2010). These layers include the Haller's layer, Sattler's layer, Bruch Membrane, choriocapillaris and additionally, the suprachoroidea, the outermost layer of the choroid (Ferrara et al., 2016; Nickla & Wallman, 2010). Each of these layers, in their own distinct way, contribute to the overall structure and function of the choroid, highlighting the remarkable complexity and interconnectedness of the ocular system (Ferrara et al., 2016; Nickla & Wallman, 2010).

The comprehension of the cellular and molecular mechanisms that contribute to the formation and functioning of the retina's structure has been greatly enhanced by advancements in molecular tools, imaging techniques, and electrophysiological approaches and the development of such tools has facilitated the understanding of how the retina's structure and function are shaped (Hoon et al., 2014).

The functional significance of the retina and choroid for the fundamental disciplines such as biophysics, physiology, and histology is progressively advancing and a remarkable instrument, known as optical coherence tomography (OCT), has emerged, revolutionizing the imaging resolution of these anatomical components to a level that is comparable to histological quality (Bajwa et al., 2015; Murthy et al., 2016).

First described in 1991 at the Massachusetts Institute of Technology, this innovative technique employs the principles of low coherence interferometry (Huang et al., 1991). This methodology, which bears some resemblance to the ultrasonic pulse-echo equivalent, deviates from the traditional approach by utilizing near-infrared light beams (NIR) rather than sound waves (Huang et al., 1991).

OCT, an advanced diagnostic imaging technology employed in the field of ophthalmology, serves the purpose of evaluating the structures of the retina and optic disk (Bajwa et al., 2015). Due to its high precision and accuracy, OCT has gained significant recognition as an effective modality in the aforementioned domain (Bajwa et al., 2015). By employing OCT, clinicians are able to acquire comprehensive topographic information

pertaining to the optic nerve head, the neuroretinal rim, and even the thickness of the nerve fiber layer (Bajwa et al., 2015). As a result of its remarkable capabilities, OCT has emerged as a particularly valuable tool for the early detection of glaucoma and the continuous monitoring of patients who may be at risk of experiencing vision loss associated with this ocular condition (Bajwa et al., 2015). It is noteworthy that OCT is also capable of assessing a diverse range of ocular diseases, including but not limited to choroidal tumors, diabetic macular edema, age-related macular degeneration, central serous chorioretinopathy, and epiretinal membranes (Bajwa et al., 2015). Remarkably, OCT has demonstrated its utility in facilitating the management of various ocular inflammatory disorders (Bajwa et al., 2015). To augment its diagnostic capabilities, OCT can be effectively employed in conjunction with other imaging techniques such as wide-field retinal angiography and fundus autofluorescence, thereby enabling a comprehensive assessment of diverse ocular pathologies (Bajwa et al., 2015).

By the time, OCT gain more advance technology and spectral domain OCT (SD-OCT) became available with a higher resolution (Nassif et al., 2004). Enhanced depth imaging (EDI-OCT) technique gave SD-OCT with a higher resolution which particularly is important the evaluation of the choroidal part and choroidal diseases (Margolis & Spaide, 2009).

This higher resolution allowed researchers to perform more deepened and focused studies especially at the fields of image processing (Agrawal, Gupta, et al., 2016). Especially Niblack binarization method have been used to assess choroidal vascularity index (CVI) (Agrawal, Gupta, et al., 2016). Thus, the main aim of this study is to define choroidal and retinal mean gray value (MGV), skewness and kurtosis after binarization in vertical and horizontal OCT scans.

MATERIALS AND METHODS

This study, which was conducted at the Department of Biophysics, Faculty of Medicine, Hacettepe University, aimed to provide a retrospective analysis of the optical coherence tomography image analysis findings and conducted in the context of a doctoral thesis. In order to achieve this, a total of 100 eyes from 50 subjects, comprising 25 females and 25 males, were included in the study. With vertical and horizontal scans there were a total of 200 OCT images. The data and information pertaining to each subject were collected from the Clinic of Ophthalmology, Ankara Training and Research Hospital, University of Health Sciences. Subjects who have applications to the Clinic of Ophthalmology, Ankara Training and Research Hospital, University of Health Sciences, in the three years prior to August 2017, and fall within the age range of 15 to 65, and have no previous record of ocular disease, systemic disease, or ocular surgery, and possess both conventional and computer-based archive data that can be accessed, and have SD-OCT and EDI-OCT images (Spectralis Heidelberg Engineering in Heidelberg, Germany), and have both vertical and horizontal images available, and possess SD-OCT images of sufficient quality for accurate interpretation, have been selected as meeting the inclusion criteria.

Both traditional archive systems within hospitals and digital archive systems were employed as tools for gathering data, in addition to the data collected from the SD-OCT device. In order to obtain a comprehensive understanding, various pieces of information were retrospectively collected and documented, including age, gender, demographic details, ocular disease history, systemic disease history, and history of ocular surgeries. All of the collected data were recorded within a sophisticated computerized database, ensuring accuracy and organization. To extract the SD-OCT images, the device's program was utilized. Through this program, the images were obtained and made accessible for further analysis and examination.

The images then were analyzed using the open-source image processing software Fiji/ImageJ (version 1.52d) (Schindelin et al., 2012; Schneider et al., 2012). The scale is set according to the image. Region of Interest (ROI) were delineated based on the horizontal scale, with a 6500 μm area centered around the fovea. The borders of the retina and choroid was designated as internal limiting membrane, retinal pigment epithelium (RPE) layer and the choroidoscleral junction, respectively. 6500 μm area is further divided to 65 horizontal and 3 vertical areas in order to compare the localized differences. Thus, calculations were performed for entire ROI, inner, middle and outer parts. In the binarization process images were converted to 8-bit and they were

binarized using Niblack auto local threshold method (Agrawal, Gupta, et al., 2016). After binarization process retinal and choroidal MGV, skewness and kurtosis have been measured for vertical and horizontal scans. All the measurements were performed using a semi-automated method utilizing Fiji/ImageJ program (İnam, 2018).

The data were imported into SPSS 21.0 (IBM SPSS Statistics V21.0) and Microsoft Excel (Microsoft Excel for Mac V16.11) for statistical analysis and graphical demonstrations. Descriptive statistics were used to summarize the data, with qualitative variables presented as numbers and percentages, and quantitative variables summarized using mean, standard deviation, minimum, and maximum values. The normality of variables was assessed using the Shapiro-Wilk test, and Student's t-test or Mann-Whitney U test was applied depending on normal distribution. Statistical significance was determined using a p-value of less than 0.05. This study has obtained approval from the Hacettepe University Medical Faculty Non-Invasive Clinical Research Ethics Committee (11.07.2017, GO 17-670), and conducted as a doctoral thesis study, with no external resources or support sought and no conflicts of interest.

RESULTS and DISCUSSION

There were 200 OCTs in this study with 100 horizontal and 100 vertical alignments for 2 eyes of the 50 cases (Female/Male ratio is 1:1). The age between female and male was not significantly different (40.71 ± 15.08, 39.86 ± 14.24, p=0.838).

In vertical scans mean retinal MGV value is 117.2 ± 3.9 for entire ROI and 120.3 ± 5.2 for horizontal scans (p<0.001). For the inner parts, retinal MGV value is 144.5 ± 4.0 for vertical scans and 147.1 ± 7.8 for horizontal scans (p=0.003). For the middle parts vertical retinal scans has a value of 84.8 ± 10.2 and horizontal retinal scans 82.0 ± 9.3 considering MGV (p=0.048). For the outer parts, retinal MGV value was found to be 124.8 ± 11.9 for vertical scans and 133.6 ± 11.5 for horizontal scans (p<0.001). In contrast, MGV in choroidal regions is more decreased when we look the general values. In vertical scans of entire ROI, choroidal MGV has a value of 76.3 ± 6.8 and horizontal scans has a value of 78.5 ± 6.7 (p=0.019). For the inner parts, vertical scans have a value of mean choroidal MGV 64.2 ± 12.7 and horizontal scans 65.0 ± 11.6 which is found insignificantly different from each other (p=0.678). For middle parts choroidal MGV in vertical scans has a value of 86.8 ± 10.4 and in horizontal scans 83.5 ± 9.9 (p=0.025). For outer parts in vertical scans choroidal MGV is 75.8 ± 12.0 and in horizontal scans 85.1 ± 10.9 (p<0.001). Thus, for retinal MGV there was a significant difference between vertical and horizontal scans for entire ROI, and inner, middle and outer parts as well. But for the choroidal MGV this difference is only significant for entire ROI, middle and outer parts. Inner regions of the choroid are seemed to be similar in terms of MGV between vertical and horizontal scan regions. All ROI is further investigated at 100 µm apart for MGV and plotted graph can be seen in Figure 1A-B for retinal and choroidal measurements.

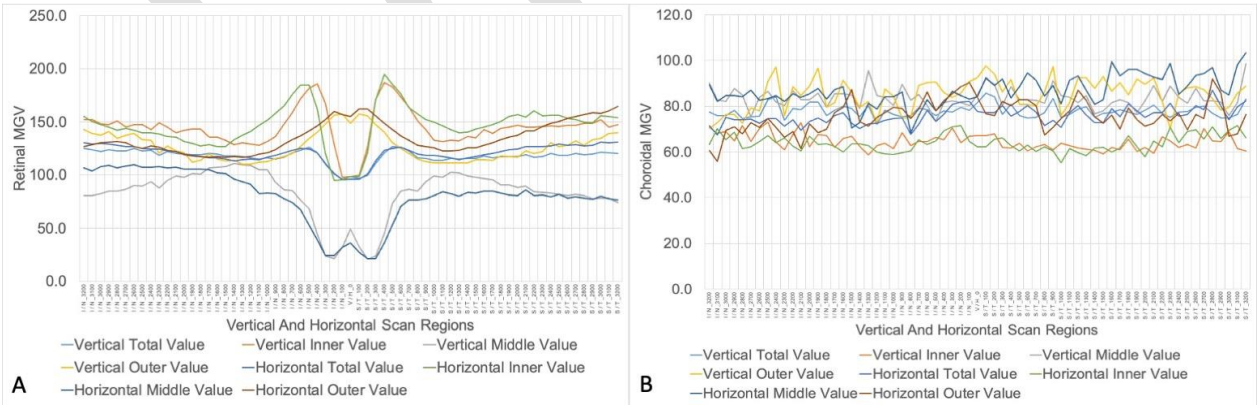


Figure 1. Retinal (A) and Choroidal (B) MGV for Vertical and Horizontal Scan Regions (MGV: Mean Gray Value)

Mean retinal skewness value for entire ROI in vertical scans is 0.16 ± 0.06 and in horizontal scans 0.11 ± 0.08 (p<0.001). In the inner parts retinal skewness is -0.27 ± 0.07 for vertical scans and -0.31 ± 0.13 for horizontal

scans (p=0.002). Middle parts have a retinal skewness value of 0.72 ± 0.19 for vertical scans and 0.77 ± 0.17 for horizontal scans (p=0.047) and outer parts have a retinal skewness value of 0.04 ± 0.19 in vertical scans in comparison horizontal scans have a value of -0.10 ± 0.18 (p<0.001). Mean choroidal skewness for entire ROI is 0.84 ± 0.14 for vertical scans and 0.88 ± 0.15 for horizontal scans (p=0.024). For the inner parts choroidal skewness is 1.15 ± 0.29 for vertical scans and 1.18 ± 0.37 for horizontal scans which is found insignificantly different from each other (p=0.530). In the middle parts, choroidal skewness in vertical scans was found to be 0.74 ± 0.19 and in horizontal scans 0.68 ± 0.20 (p=0.033). In the outer parts, choroidal skewness was 0.72 ± 0.21 for vertical scans and 0.90 ± 0.25 for horizontal scans (p<0.001). For retinal skewness, there was a significant difference between vertical and horizontal scans at the entire ROI, inner, middle and outer regions; in contrast choroidal skewness is only significantly different at the entire ROI, middle and outer regions, not in the inner regions. For skewness, all ROI is further investigated at 100 µm apart and plotted graphs for retinal and choroidal measurements which can be seen in Figure 2A-B.

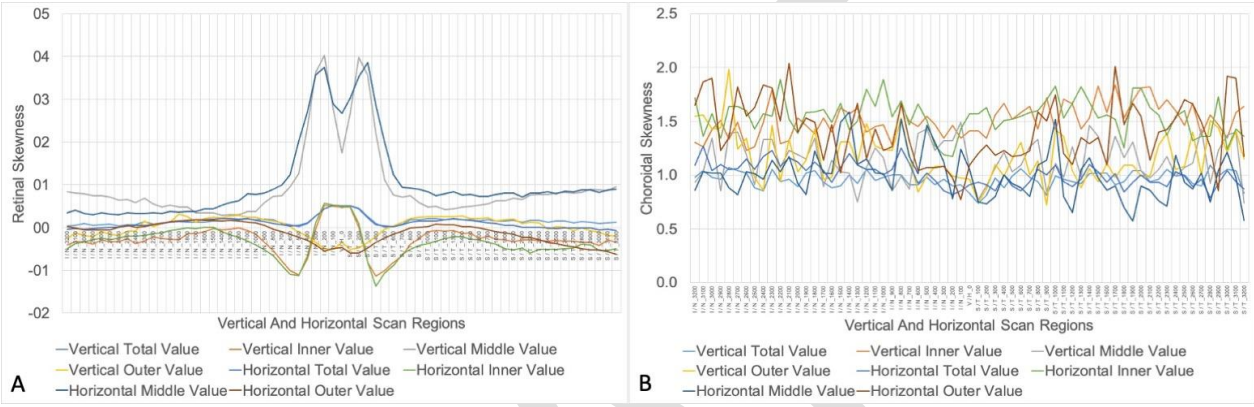


Figure 2. Retinal (A) and Choroidal (B) Skewness for Vertical and Horizontal Scan Regions

Mean retinal kurtosis was found to be -1.97 ± 0.02 in vertical scans and -1.98 ± 0.02 in horizontal scans for entire ROI (p<0.001). For the inner parts retinal kurtosis is -1.92 ± 0.04 for vertical scans and -1.89 ± 0.08 for horizontal scans (p<0.001). In the middle parts retinal kurtosis is found to be -1.45 ± 0.26 for vertical scans and -1.38 ± 0.25 for horizontal scans (p=0.05). Outer parts have a retinal kurtosis value of -1.96 ± 0.11 for vertical scans and horizontal scans have a retinal kurtosis value of -1.96 ± 0.06, which there was not a significant difference between groups (p=0.777). Mean choroidal kurtosis was found to be -1.28 ± 0.26 for the entire ROI in vertical scans and -1.20 ± 0.29 for horizontal scans (p=0.037). For the inner parts choroidal kurtosis is found to be -0.59 ± 0.73 in vertical scans and -0.47 ± 1.21 in horizontal scans (p=0.395). In the middle parts, vertical scans have a choroidal kurtosis value of -1.41 ± 0.32 and horizontal scans have a kurtosis value of -1.49 ± 0.33 (p=0.074). For the outer parts, choroidal kurtosis was found to be -1.44 ± 0.35 for vertical scans and -1.12 ± 0.49 for horizontal scans (p<0.001). When we compare retinal kurtosis for vertical and horizontal scans, only entire ROI and inner regions were found to be significantly different; in contrast choroidal kurtosis is different in entire ROI, middle and outer parts between vertical and horizontal scans. For kurtosis, all ROI is further investigated at 100 µm interval and resulting graph can be seen in Figure 3A-B.

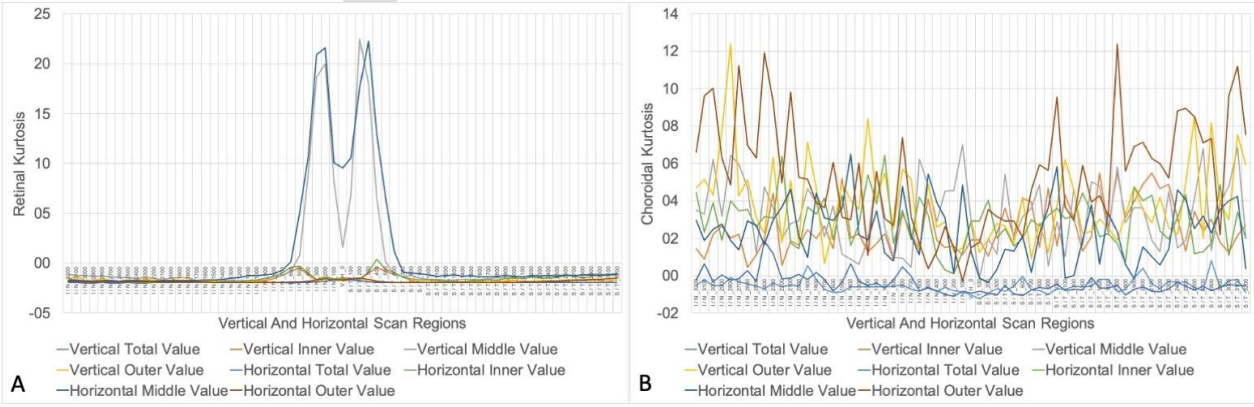


Figure 3. Retinal (A) and Choroidal (B) Kurtosis for Vertical and Horizontal Scan Regions

There are numerous studies in the literature aiming to find a better way to quantify OCT images and distinguish between healthy and diseased ones utilizing image processing techniques (Agrawal, Chhablani, et al., 2016; Agrawal, Li, et al., 2016; Agrawal, Salman, et al., 2016). Central serous chorioretinopathy have been investigated for CVI and found to have higher CVI values considering their fellow eyes (Agrawal, Chhablani, et al., 2016). In another study CVI is measured alongside choroidal thickness in Vogt-Koyanagi-Harada disease, they both found to be significantly higher compared to the control group (Agrawal, Li, et al., 2016). Interestingly it is found that both choroidal thickness and CVI values are decreased significantly at fallow up, compared to the baseline values (Agrawal, Li, et al., 2016). In a study conducted with panuveitis patients, at 3 months fallow up, CVI change is found to be considerably decreased in uveitic eyes, and this decrease is found to be statistically significant compared with the control group change (Agrawal, Salman, et al., 2016). Even systemic diseases that have effects on eyes were investigated in terms of CVI and although there was not found a significant difference for retinal thickness, choroidal thickness, and many other parameters; CVI of the diabetes patients have been found significantly lower than the control group (Tan et al., 2016). These findings can show that even retinal and choroidal thickness measurements could be nonsignificantly changed, many other parameters and indexes obtained via image processing techniques could be more useful for demonstrating the difference between diseased and healthy ones. Another study has investigated that CVI and structural choroidal changes in age related macular degeneration and they have found that exudative age-related macular degeneration have significantly lower CVI values compared to fellow eyes, despite insignificant change in choroidal thickness (Wei et al., 2017).
Thus, it is very important to investigate different parameters and indexes especially for healthy subjects. Different parameters and indexes could be useful for both differentiating the diseases and disease fallow up.

CONCLUSION

In conclusion, we investigated the retina and choroid, vertical and horizontal scans, in different regions for MGV, skewness and kurtosis at healthy subjects. We defined the mean values for different regions and found that there were localized differences between vertical and horizontal scans for these parameters. Further studies could be needed for investigating these parameters' role in different diseases.

REFERENCES

Agrawal R, Chhablani J, Tan KA, Shah S, Sarvaiya C, Banker A 2016. CHOROIDAL VASCULARITY INDEX IN CENTRAL SEROUS CHORIORETINOPATHY. Retina, 36(9): 1646-1651. https://doi.org/10.1097/iae.0000000000001040

Agrawal R, Gupta P, Tan KA, Cheung CM, Wong TY, Cheng CY 2016. Choroidal vascularity index as a measure of vascular status of the choroid: Measurements in healthy eyes from a population-based study. Sci Rep, 6: 21090. https://doi.org/10.1038/srep21090

Agrawal R, Li LK, Nakhate V, Khandelwal N, Mahendradas P 2016. Choroidal Vascularity Index in Vogt-Koyanagi-Harada Disease: An EDI-OCT Derived Tool for Monitoring Disease Progression. Transl Vis Sci Technol, 5(4): 7. https://doi.org/10.1167/tvst.5.4.7

Agrawal R, Salman M, Tan KA, Karampelas M, Sim DA, Keane PA, Pavesio C 2016. Choroidal Vascularity Index (CVI)--A Novel Optical Coherence Tomography Parameter for Monitoring Patients with Panuveitis? PLoS One, 11(1): e0146344. https://doi.org/10.1371/journal.pone.0146344

Bajwa A, Aman R, Reddy AK 2015. A comprehensive review of diagnostic imaging technologies to evaluate the retina and the optic disk. Int Ophthalmol, 35(5): 733-755. https://doi.org/10.1007/s10792-015-0087-1

Ferrara D, Waheed NK, Duker JS 2016. Investigating the choriocapillaris and choroidal vasculature with new optical coherence tomography technologies. Prog Retin Eye Res, 52: 130-155. https://doi.org/10.1016/j.preteyeres.2015.10.002

Gupta MP, Herzlich AA, Sauer T, Chan CC 2016. Retinal Anatomy and Pathology. Dev Ophthalmol, 55: 7-17. https://doi.org/10.1159/000431128

Hoon M, Okawa H, Della Santina L, Wong RO 2014. Functional architecture of the retina: development and disease. Prog Retin Eye Res, 42: 44-84. https://doi.org/10.1016/j.preteyeres.2014.06.003

Huang D, Swanson EA, Lin CP, Schuman JS, Stinson WG, Chang W, Hee MR, Flotte T, Gregory K, Puliafito CA, et al. 1991. Optical coherence tomography. Science, 254(5035): 1178-1181.

İnam O. (2018). Calculation of choroidal vascularity index and tissue distribution indexes by digital processing of retinal and choroidal images obtained via optical coherence tomography. Doctorate Thesis, Hacettepe University Graduate School of Health Sciences. Ankara.

Margolis R, Spaide RF 2009. A pilot study of enhanced depth imaging optical coherence tomography of the choroid in normal eyes. Am J Ophthalmol, 147(5): 811-815. https://doi.org/10.1016/j.ajo.2008.12.008

Murthy RK, Haji S, Sambhav K, Grover S, Chalam KV 2016. Clinical applications of spectral domain optical coherence tomography in retinal diseases. Biomed J, 39(2): 107-120. https://doi.org/10.1016/j.bj.2016.04.003

Nassif NA, Cense B, Park BH, Pierce MC, Yun SH, Bouma BE, Tearney GJ, Chen TC, Boer JFd 2004. In vivo high-resolution video-rate spectral-domain optical coherence tomography of the human retina and optic nerve. Optics Express, 12(3): 367-376. https://doi.org/10.1364/OPEX.12.000367

Nickla DL, Wallman J 2010. The multifunctional choroid. Prog Retin Eye Res, 29(2): 144-168. https://doi.org/10.1016/j.preteyeres.2009.12.002

Schindelin J, Arganda-Carreras I, Frise E, Kaynig V, Longair M, Pietzsch T, Preibisch S, Rueden C, Saalfeld S, Schmid B, Tinevez JY, White DJ, Hartenstein V, Eliceiri K, Tomancak P, Cardona A 2012. Fiji: an open-source platform for biological-image analysis. Nat Methods, 9(7): 676-682. https://doi.org/10.1038/nmeth.2019

Schneider CA, Rasband WS, Eliceiri KW 2012. NIH Image to ImageJ: 25 years of image analysis. Nat Methods, 9: 671. https://doi.org/10.1038/nmeth.2089

Tan KA, Laude A, Yip V, Loo E, Wong EP, Agrawal R 2016. Choroidal vascularity index - a novel optical coherence tomography parameter for disease monitoring in diabetes mellitus? Acta Ophthalmol, 94(7): e612-e616. https://doi.org/10.1111/aos.13044

Wei X, Ting DSW, Ng WY, Khandelwal N, Agrawal R, Cheung CMG 2017. CHOROIDAL VASCULARITY INDEX: A Novel Optical Coherence Tomography Based Parameter in Patients With Exudative Age-Related Macular Degeneration. Retina, 37(6): 1120-1125. https://doi.org/10.1097/iae.0000000000001312

DRAFT

ORAL PRESENTATION

Random Segment ve Blok Formda Poliimid Siloksan Kopolimer Sentezi

Türkan Doğan* (*https://orcid.org/0000-0003-1530-8940*)

*1 İstanbul Teknik Üniversitesi, Nanoteknoloji Uygulama ve Araştırma Merkezi, İstanbul, Türkiye

*tdogan@itu.edu.tr

Özet

Çeşitli mekanik özelliklere sahip olan polimid malzemeler havacılık ve uzay teknolojisinde geniş anlamda kullanım potansiyeline sahiptir. Bu tür poliimid malzemelerden olan poliimid siloksan kopolimerlerin, bu çalışmayla güneş enerjisi teknolojisinde altlık olarak kullanılabilecek esnek altlıklar olarak sentezlenmesi hedeflendi. Random segment ve blok formda poliimid siloksan kopolimer sentezi
, Benzofenon-3, 3', 4, 4'-tetrakarboksilik dianhidrit (BTDA), 4, 4'-oksidianilin (ODA) ve bis (3-aminopropyl) polidimetilsiloksan (APPS) kullanılarak sentezlendi. ODA ve BTDA, Blok kopolimer yapıdaki sert segmentleri, APPS ve BTDA ise yumuşak segmentleri oluşturmaktadır. Yumuşak ve sert segment uzunlukları ayarlanarak bu kopolimerlerin pek çok çeşidi üretilebilmektedir. Bu kopolimerler FT-IR analizi kullanılarak karakterize edildi. Random ve blok halde üretilen hibrit-nanokompozit poliimid siloksan kopolimer yapıların FT-IR sonuçlarında, Si-O-Si (asym Si-O-Si stretching) ve, Si-C (stretching) pikleri görüldü. FT-IR sonuçlarında tesbit edilen bu pikler, iki farklı biçimde üretilen her iki yapının da elastomer özelliğini doğrular yöndedir.

Keywords: Poliimid, poliimid siloksan, kopolimer.

GİRİŞ

Güneş pili teknolojisinde halen kullanılan ve araştırılan malzemeler üç gruba ayrılmaktadır:

• Kristal silisyum, çoğunlukla fotovoltaiklerde (PV) kullanılan ticari bir malzemedir.

• Fotovoltaik ince film hücreler geleneksel PV hücrelere göre daha ucuz olduğundan, üretim maliyetleri kristal silisyum güneş pillerine göre çok daha düşüktür.

• Üçüncü grupta çok eklemli yapılar yer almaktadır.

Bu çalışmayla, güneş pillerine esneklik fonksiyonu kazandırılarak kullanılabilirliğin arttırılması ve ayrıca alt tabakadan kaynaklanan verim düşüklüğünün ortadan kaldırılarak esnek güneş pillerinin performansının artırılmasına katkı sağlanması amaçlandı.

Güneş pillerinin sol-jel yöntemiyle incelenmesinde kullanılacak esnek altlıklar 3 farklı türdedir. Bu esnek altlıklar:

1) Polimid. Poliimid siloksanın özelliklerinde değişim yaratmak üzere, karşılaştırmalı olarak denenmiştir.

2) İnce esnek metalik yüzeyler. Bu çalışma kapsamında ince esnek metalik altlıklar ile diğer esnek altlıklar üzerinde üretilen filmlerin yapışma ve elektriksel özellikleri arasındaki farklar karşılaştırmalı olarak incelenebilmektedir.

3) Esnek poliimid siloksan hibrit-nanokompozit polimer yapılar üretildi. Bunun için, polidimetilsiloksan (PDMS) olarak kullanılan APPS ve poliimid sert ve yumuşak bloklar içeren blok kopolimerler oluşturulmuştur. Üretilen hibrit-nanokompozit kopolimer altlıklar, henüz yarıiletken ince filmlerle uyum açısından test edilmedi. Öte yandan, yarıiletken ince filmlerin ve esnek altlıkların uyumluluğu açısından karakterizasyon işlemleri yapılabilir.

Polimer malzemelerde karşılaşılabilecek bazı olumsuzluklar; aynı zamanda kısaca "Tg" denilen, camsı geçiş değerlerinin düşük olması, düşük sıcaklıkta kullanımda direncin düşük olması, sıcaklık genleşme katsayısının

DRAFT

(CTE) yüksek olması ve nem tutma oranlarının (WVTR) yüksek olması gibi durumlar, bu olumsuzluklar arasında sayılabilir.

Özellikle Tg oranlarının düşüklüğü en kritik olumsuzluktur. Çünkü Tg değeri yarı iletken ince filmlerle kaplama üretmek için en önemli kriteri oluşturmaktadır.

Genel olarak çoğu yarı iletken malzemenin hibrit-nanokompozit yapılarla birlikte üretimi sırasında 200 ^{o}C civarında bir sıcaklık değerine ihtiyaç duyulur. Bu nedenle, poliimid malzemeler yüksek Tg oranlarına sahip olmaları nedeniyle en çok tercih edilen malzemeler arasında yer almaktadır.

Poliimid malzemelerin olumlu yönleri:

• Yüksek Tg değerlerine sahip oldukları için yarı iletken ince film ile her türlü hibrit yapıyı oluşturabilirler.

• Mükemmel sıcaklık stabilitesine sahiptirler.

• Hibrit oluşturma açısından, yüksek mekanik dayanıma sahiptirler.

Poliimidler, organik diaminlerden ve organik tetrakarboksilik asitlerden (veya bunların türevlerinden) elde edilen yüksek molekül ağırlıklı polimerlerdir.

Esnek altlıklar üzerinde, yarıiletken ince film üretebilmek için polimidin, siloksan yapı ile hibritlenmesinin nedenleri şu şekilde sıralanabilir;

1) Poliimid yapıların termal genleşme katsayısını (CTE) ele aldığımızda, ısı yükselip camsı geçiş sıcaklığını (Tg) geçtiğinde sıcaklığın arttığı gözlemlenmektedir (Friedrich et al., 2004). Bu durumun, yarıiletken ince film yapılar üzerinde deformasyonlara neden olabileceği düşünülmektedir.

2) Polidimetilsiloksan (PDMS) kaynağı olarak bis(c-aminopropil) polidimetilsiloksan (APPS) yardımıyla üretilebilen poli(imid siloksan) kopolimerlerinin Tg değerindeki herhangi bir artış, CTE değerinin istenilen düzeye getirilmesine neden olur (Pei et al., 2013).

PDMS kaynağı olarak APPS kullanılması durumunda, PDMS'nin CTE değeri ile altlık üzerinde üretilen ince filmin CTE değeri birbirine yakınlaşabilmektedir.

Kopolimerizasyon, farklı kimyasal ve fiziksel özelliklere sahip iki yapının aynı polimer zincirine bağlanması sırasında amaçlanan özelliklere sahip yeni malzemelerin üretilmesini sağlayan en başarılı ve genel yöntemlerden biridir. Ayrıca, polisiloksan ve poliimid kombinasyonundan üretilen poliimid siloksan monomer blokları işlenebilirliği kolaylaştırmaktadır (Pei,X.; Chen,G.; Fang,X., Synthesis and Properties of Poly(imide siloxane) Block Copolymers with Different Block Lengths, J. APPL. POLYM. SCI. 2013).

Poliimid siloksan, yüksek esneklik ve yapışma özelliğine sahiptir ve her türlü agresif oksijen ortamında bozunmaya karşı dayanıklıdır. Bu nedenle bu çalışmanın içeriğinde esnek altlıkların incelenmesi için poliimid siloksan monomer blokları tercih edilmiştir.

Hibrit-nanokompozit polimer yapıların sentezinde kopolimerizasyon yöntemi kullanılmıştır. Kopolimerizasyon, farklı kimyasal ve fiziksel özelliklere sahip iki yapının aynı polimer zincirine bağlanması sırasında amaçlanan özelliklere sahip yeni malzemelerin üretilmesini sağlayan en başarılı yöntemlerden biridir. Ayrıca, polisiloksan ve poliimid kombinasyonundan üretilen poliimid siloksan monomer blokları işlenebilirliği kolaylaştırmaktadır (Pei,X.; Chen,G.; Fang,X., Synthesis and Properties of Poly(imide siloxane) Block Copolymers with Different Block Lengths, J. APPL. POLYM. SCI. 2013).

MALZEME VE METOD

Bu çalışma kapsamında poliamik asit PDMS (polidimetil siloksan) içerisine, yumuşak bloklar eklenerek esnek poliimid siloksan kopolimerlerinin üretilmesi planlanmaktadır. Kimyasal malzemeler yüksek saflıkta Sigma Aldrich'ten temin edilmiştir.

Yüksek direnç özellikli poliimid siloksan random segment ve blok kopolimer sentezi, NMP (Metil-2-pirolidinon ve ODCB (oksidiklorobenzen)'den oluşan ortak çözücü ortamda sentezlendi. Sentezlenen, sert ve yumuşak bloklar karıştırıldı. Bu amaçla ODA ve BTDA sert bloklarının karıştırılmasıyla, APPS ve BTDA yumuşak bloklarının karıştırılmasıyla blok kopolimer zincirlerinin üretilmesi planlandı. Poliimid siloksan bloklar yüksek esneklik ve yapışma özelliğine sahip, agresif oksijen ortamında ayrışmaya karşı dayanıklı ve

termal geçiş sıcaklığı yüksek olacak biçimde sentezlendi. Ayrıca, random segment poliimid siloksan görünür ışıkta optik olarak şeffaf olma özelliğine sahiptir.
Random segmentli poliimid siloksan hibrit-nanokompozit polimerik yapıların sentezi için, cam balon içerisine, azot ortamda, BTDA, NMP ve ODCB eklenerek, karıştırıldı. Dianhidrit tamamen çözüldükten sonra ODCB ile birlikte, APPS ve ODA sırayla ilave edilerek, 8 saat sürede 175°C'de karıştırıldı.
Poliimid siloksan hibrit-nanokompozit blok kopolimerlerin sentezlenmesi için, ODA ve BTDA poliimid sert bloğu oluştururken, APPS ve BTDA polisiloksan yumuşak bloğu oluşturacak biçimde sentez yapıldı. Cam flask içerisine, azot ortamda, BTDA, NMP ve ODCB eklendi. Dianhidrit tamamen çözündükten sonra ODCB ile birlikte, APPS yavaş yavaş cam flaska ilave edildi.

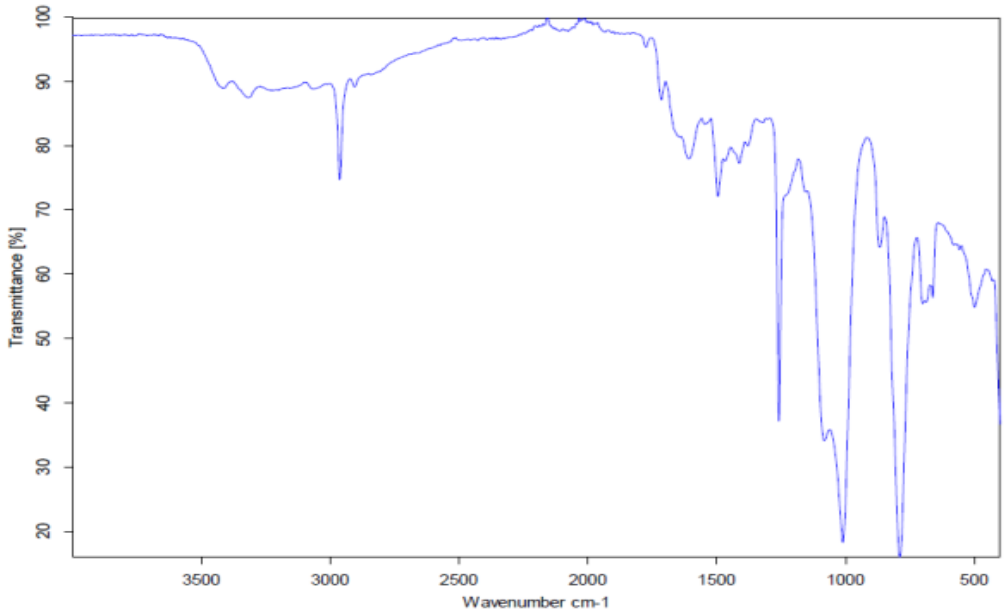
TARTIŞMA

Üretilen, esnek altlıkların fiziksel özellikleri incelendi. Özellikle, fiziksel değişiklikler bağlamında, üretilen hibrit-nanokompozit polimerik yapılar incelendi ve değerlendirildi. Böylece esnek yüzeyler üzerine daha önce literatürde incelenmemiş, uzay teknolojisine uygun özgün bir çalışma hedeflendi.
Bu çalışma, iki adımdan oluşmaktadır: İlk olarak random segmentlere ayrılan hibrit-nanokompozit poliimid siloksan ve ardından poliimid siloksan blok kopolimerler, BTDA, APPS ve ODA kullanılarak, oda sıcaklığında azot ortamda, kondensasyon polimerizasyonu tekniği kullanılarak sentez edildi.

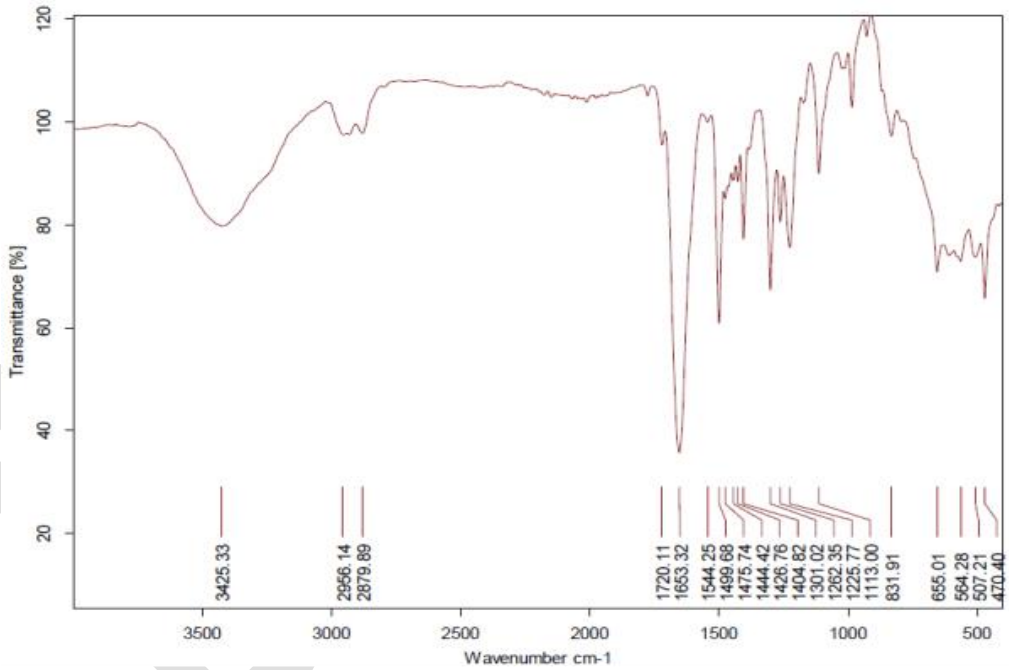
Random ve blok halde üretilen hibrit-nanokompozit poliimid siloksan kopolimer yapılar üzerinden gidilerek, yapının eldesinde kullanılan kimyasalların konsantrasyon oranlarında ki değişikliklerin, malzemenin yapısını nasıl etkilediği tesbit edilmeye çalışıldı. Konsantrasyon etkisini tesbiti amacıyla bu parametre üzerinde yapılan değişikliklerle, amid veya anhidrid kimyasal zincir halkalarının tamamlanarak kapandığını gösteren diamin-dianhidrid piklerinin tesbiti amacyla FT-IR analizi yapılmıştır. Yapılan FT-IR analizi sonucunda bu pikler tesbit edilmiştir. Random ve blok halde üretilen hibrit-nanokompozit poliimid siloksan kopolimer yapıların FT-IR analizi Şekil 1 ve 2'de gösterilmiştir.
Random biçimde üretilen poliimid siloksan kopolimer yapının FTIR spektroskopi analiz sonuçlarında yapı, random pikler sergilemektedir. 2900-3100 cm^{-1} arası alifatik C-H stretching , 1700-1800 cm^{-1} arası asym C=O stretching, 1710-1720 cm^{-1} arası sym C=O stretching, 1000-1100 cm^{-1} arası asym Si-O-Si stretching, 1000-1010 cm^{-1} arası sym Si-O-Si stretching, 750-790 cm^{-1} arası Si-C stretching pikleri Şekil 1'de görülmektedir.

FTIR spektroskopisinde 1700-1800 cm^{-1} arası ve 1710-1720 cm^{-1} arası karbonil amid gruplarını ve 1500 cm^{-1} lerde, amid yapının N-H bağları gözlenmektedir. Bu amid absorbsiyon pikleri 1700-1800 cm^{-1} arasında –COOH karboksil halka zincirlerini tamamlayarak kapatmak üzere yok olmuştur. Bu da kopolimer yapının aromatik halka zincirlerinin tamamlanması ile oluştuğunu göstermektedir. Tüm kopolimer yapılarda 1700-1800 cm^{-1} arası pikler imid grupları göstermektedir.

DRAFT



Şekil 1. Azot atmosfer ortamında random halde üretilen yüksek direnç özellikli hibrit-nanokompozit poliimid siloksan yapıların FTIR analiz sonuçları.



Şekil 2. Azot atmosfer ortamında blok halde üretilen yüksek direnç özellikli hibrit-nanokompozit poliimid siloksan yapıların FTIR analiz sonuçları.

Azot atmosfer ortamında blok halde üretilen hibrit-nanokompozit poliimid siloksan yapıların FTIR analiz sonuçları incelendiğinde ise, 3425 cm^{-1} de –OH (hidroksil) pikleri, 2956 ve 2879 cm^{-1} lerde alifatik C-H stretching düz zincir bağlar, 1720 ve 1653 cm^{-1} de asym C=O stretching diamin pikleri, 1544, 1499, 1475, 1444, 1426, 1404, 1301 ve 1225 cm^{-1} de aromatik halka zincirleri gösteren sym C=O stretching bağlar gözlenmektedir. Bu piklerden anhidrid yapının aromatik halka zincirin tamamlanması ile diamin piklerine dönüştüğünü anlıyoruz. 1113 ve 831 cm^{-1} de asym Si-O-Si stretching, silikon pikler görülmektedir. 655, 564, 507 ve 470 cm^{-1} de Si-C stretching pikleri görülmektedir.

Random biçimde üretilen poliimid siloksan kopolimer yapıda olduğu gibi, 1720-1650 cm^{-1} arası karbonil amid gruplarını ve 1544, 1499 cm^{-1} lerde, amid yapının N-H bağları gözlenmektedir. Bu amid absorbsiyon pikleri

1720-1650 cm-1 arasında –COOH karboksil halka zincirlerini tamamlayarak kapatmak üzere yok olmuştur. Kopolimer yapı, aromatik halka zincirlerinin tamamlanması ile oluştuğu görülmektedir.

SONUÇ

Poliimid blok kopolimer altlıklar, güneş pillerinde kullanılacak altlıklar olarak laboratuvar imkanları kullanılarak farklı esnek oranlarda üretildi. Böylece istenilen her formda, her konfigürasyonda, her ortamda kolaylıkla oluşturulabilen, erozyona dayanıklı alt tabakaların elde edilmesi mümkün olabilmektedir.
Bu çalışmanın sonuçları şu şekilde özetlenebilir:

- Esnek güneş pillerinde kullanılmak üzere poliimid siloksan polimer altlıkların üretilmesi ve yarı iletken kaplamalara uygunluğu incelenebilir.
- Literatür çalışmalarına göre poliimid siloksan hibrit polimerler, poliimidlere göre daha yüksek mekanik özelliklere ve cam geçiş sıcaklığı katsayısına sahiptir.
- Poliimid siloksanın bu özelliği sayesinde, güneş pillerinde kullanılan polimidin özelliklerinin arttırılarak esnek güneş pillerinin daha yüksek verimine katkıda bulunulması planlanmaktadır.
- Farklı kimyasal ve fiziksel özelliklere sahip iki yapının aynı polimer zinciri içerisinde birleştirilmesi olan kopolimerizasyon, amaçlanan özelliklere sahip yeni malzemeler üretmek için uygulanabilecek en başarılı ve genel yöntemlerden biridir.
- Ayrıca polisiloksan ve poliimid kombinasyonundan elde edilen poliimid siloksan monomer blokları, poliimidin işlenebilirliğini sağlar.
- Bu nedenle bu tez çalışması kapsamında poliimid siloksan esnek altlıkların incelenmesi tercih edilmiştir.

- Random biçimde üretilen poliimid siloksan kopolimer yapının FTIR spektroskopi analiz sonuçlarında yapı, random pikler sergilemektedir. 2900-3100 cm^{-1} arası alifatik C-H stretching , 1700-1800 cm^{-1} arası asym C=O stretching, 1710-1720 cm^{-1} arası sym C=O stretching, 1000-1100 cm^{-1} arası asym Si-O-Si stretching, 1000-1010 cm^{-1} arası sym Si-O-Si stretching, 750-790 cm^{-1} arası Si-C stretching pikleri gözlenmiştir. FTIR spektroskopisinde 1700-1800 cm^{-1} arası ve 1710-1720 cm^{-1} arası karbonil amid gruplarını ve 1500 cm^{-1} lerde, amid yapının N-H bağları gözlenmektedir. Bu amid absorbsiyon pikleri 1700-1800 cm^{-1} arasında –COOH karboksil halka zincirlerini tamamlayarak kapatmak üzere yok olmuştur. Bu da kopolimer yapının aromatik halka zincirlerinin tamamlanması ile oluştuğunu göstermektedir. Tüm kopolimer yapılarda 1700-1800 cm^{-1} arası pikler imid grupları göstermektedir.

- Blok halde üretilen poliimid siloksan kopolimer yapının FTIR spektroskopi analiz sonuçlarında ise yapı, 3425 cm^{-1} de –OH (hidroksil) pikleri, 2956 ve 2879 cm^{-1} lerde alifatik C-H stretching düz zincir bağlar , 1720 ve 1653 cm^{-1} de asym C=O stretching diamin pikleri, 1544, 1499, 1475, 1444, 1426, 1404, 1301 ve 1225 cm^{-1} de aromatik halka zincirleri gösteren sym C=O stretching bağlar gözlenmektedir. Bu piklerden anhidrid yapının aromatik halka zincirin tamamlanması ile diamin piklerine dönüştüğü anlaşılmaktadır. 1113 ve 831 cm^{-1} de asym Si-O-Si stretching, silikon pikler görülmektedir. 655, 564, 507 ve 470 cm^{-1} de Si-C stretching pikleri görülmektedir. Random biçimde üretilen poliimid siloksan kopolimer yapıda olduğu gibi, 1720-1650 cm^{-1} arası karbonil amid grupları ve 1544, 1499 cm^{-1} lerde, amid yapının N-H bağları görülmektedir. Bu amid absorbsiyon pikleri 1720-1650 cm^{-1} arasında –COOH karboksil halka zincirlerini tamamlayarak kapatmak üzere yok olmaktadır. Kopolimer yapının, aromatik halka zincirlerinin tamamlanması ile oluştuğu görülmektedir.

TEŞEKKÜRLER

Bu çalışma, İstanbul Teknik Üniversitesi, Bilimsel Araştırma Merkezi (İTÜ BAP) tarafından 37805 numaralı doktora tezi projesi kapsamında desteklenmiştir.

REFERANSLAR

Friedrich K, Dominik R 2004. Technological Aspects of Flexible CIGS Solar Cells and Modules. Solar Energy, 77: 685–695.

Gao C, Yan D. 2003. Hyperbranched Polymers: from Synthesis to Applications.
Jennifer B, Christen Andreas, Andreou G 2007. Student Member and fellow, IEEE, Design, Fabrication, and Testing of a Hybrid CMOS/PDMS Microsystem for Cell Culture and Incubation.
Meyer J-U 2001. Retina implant-a bioMEMS challenge.
Pei X., Chen G, Fang X 2013. Synthesis and Properties of Poly (imide siloxane) Block Copolymers with Different Block Lengths, J. APPL. POLYM. SCI. doi: 10.1002/app.38918, 3718-3727.
Wohl C J, Atkins BM, Belcher M A, Connell J W 2012. High. Perform. Polym. 24, 40.
Yoo H, Park S 2010. The fabrication of highly ordered blockcopolymer micellar arrays: control of the separation distances of silicon oxide dots.

DRAFT

ORAL PRESENTATION

Farelerde yaşlanmaya bağlı mesane fonksiyon bozukluğunda l-sistein/hidrojen sülfür yolağının rolü

Fatma Aydınoğlu[1*] (*ORCID: https://orcid.org/0000-0003-3691-208X.*), Nuran Öğülener[2]

[*1] Çukurova Üniversitesi, Eczacılık Fakültesi, Farmakoloji Abd, Adana, Türkiye
[2] Çukurova Üniversitesi, Tıp Fakültesi, Tıbbi Farmakoloji Abd, Adana, Türkiye

*faydinoglu@cu.edu.tr

Özet

Mesane fonksiyon bozukluğu, yaşlanmaya bağlı olarak çok sık görülen önemli sağlık sorunlarından birini oluşturmaktadır. Bu fonksiyon bozukluğu mesane detrusor kas kontraktilitesinin nöronal ve miyojenikkomponentlerinin modifikasyonu sonucu gelişmektedir. İnsan, sıçan ve fare mesane dokularında yapılan çalışmalarda yaşlanmaya bağlı detrusor kasında kolinerjik ve purinerjik yanıtların belirgin bir şekilde değiştiği ve mesane aktivitesindeki değişikliklerin bu farklılaşmadan kaynaklandığı ileri sürülmüştür. Hidrojen sülfür (H_2S) memeli dokularında L-sistein' den enzimatik olarak sentez edilen bir gaz nörotransmitterdir. H_2S'in sentezlendiği ve bu dokunun regülasyonunda rol oynadığı bildirilmiş ve ürogenital hastalıklar için önemli olabileceği ileri sürülmüştür. Ancak, mesane dokusunda yaşlanmaya bağlı L-sistein/H_2S yolağı aktivitesinde oluşabilecek değişiklik ile ilgili bir çalışma mevcut değildir. Çalışmamızda yaşlanmaya bağlı detrusor kasındaki fonksiyonel değişikliklerde L-sistein/H_2S yolağının olası rolünün araştırılması amaçlanmıştır. Çalışmamızda, izole fare mesane dokusunda kümülatif karbakol kasılma yanıtları değerlendirildi. Yaşlı ve genç kasılma yanıtlarında farklılık gözlemlendi. Kümülatif L-sistein gevşeme yanıtları genç ve yaşlı farelerden izole edilen dokularda karşılaştırıldı. Ayrıca, ekzojen H2S yanıtlarının araştırılması için kümülatif sodyum hidrojen sülfür gevşeme yanıtları da, genç ve yaşlı farelerden izole mesane prepatlarında araştırıldı. Endojen (L-sistein) ve ekzojen H_2S (NaHS) gevşeme yanıtlarının yaşlı farelerde genç farelere farklılık gözlemlendi. Bu bulgular, mesane dokusunda yaşa bağlı olarak H_2S yolağını etkilediğini düşündürmektedir.

Anahtar Kelimeler: fare, mesane, hidrojen sülfür

Role of the l-cysteine/hydrogen sulfide pathway in aging-related bladder dysfunction in mice

Bladder dysfunction is one of the most common health problems due to aging. This dysfunction develops as a result of modification of the neuronal and myogenic components of bladder detrusor muscle contractility. Studies conducted on human, rat, and mouse bladder tissues have suggested that cholinergic and purinergic responses in the detrusor muscle change significantly due to aging and that changes in bladder activity result from this differentiation. Hydrogen sulfide (H_2S) is a gaseous neurotransmitter synthesized enzymatically from L-cysteine in mammalian tissues. It has been reported that H_2S is synthesized and plays a role in the regulation of this tissue, and it has been suggested that it may be important for urogenital diseases. However, there is no study on the alterations of L-cysteine/H2S pathway activity due to aging in bladder tissue. In the present study we aimed to investigate the possible role of the L-cysteine/H_2S pathway in functional changes in the detrusor muscle due to aging. In the present study, the cumulative carbachol contractions were evaluated in bladder strips isolated from aged and young mice. The contractile responses to the cumulative carbachol were markedly different in aged mice compared tou young mice. Cumulative L-cysteine relaxation responses were evaluated in the bladder strips isolated from young and aged mice. Additionally, to investigate exogenous H_2S responses, the cumulative sodium hydrogen sulfide relaxation responses were performed in isolated bladder preparations from young and aged mice. Differences in endogenous (L-cysteine) and exogenous $_{H2S}$ (NaHS) relaxation responses were observed in aged mice compared to young mice. These findings suggest that the alterations of H_2S pathway may be occur in bladder tissue dependent to age.

Keywords: bladder, hydrogen sulfide, mice

GİRİŞ

İdrarın depolanması detrusor kasının gevşemesi ve üriner sifinkterin kasılmasıyla; idrarın boşaltılmı ise detrusor kasının kasılması ve üriner sfinkterin gevşemesi ile sağlanır (Coolsaet B, 1985; Geokas ve ark.,1985). Yaşlanma hemen hemen bütün fizyolojik sistemlerin aktivitesinde bir azalmaya neden olmaktadır (Diokno ve ark., 1992). Yaşlılarda, alt üriner sistem semptomlarının daha yaygın olduğu bilinmektedir (Brocklehurst ve Dillane, 1966). Mesanenin inervasyonunda kolinerjik, nitrerjik, peptiderjik sistemlerin rolü vardır. İnsan, sıçan ve fare mesane dokularında, yaşlanmaya bağlı innervasyondan sorumlu ilgili mekanizmalarda farklılaşmaların olduğu bildirilmiştir (Siroky MB, 2004). Yaşlı farelerden izole edilmiş detrusor kasında, mesane dolum, depolama ve boşaltım fonksiyonlarında hasar oluştuğu gözlemlenmiştir (Daly ve ark., 2014). Son çalışmalarda, yeni bir gaz nörotransmiter olan H_2S 'in de mesane düz kasının regülasyonunda rol oynadığı gösterilmiştir (Fernandes ve ark., 2013). Yapılan araştırmalarda, H_2S'in endojen olarak L-sistein 'den bu dokuda sentez edildiği ve L-sistein/H_2S yolağının ürogenital hastalıkların tedavisine katkı sağlayabileceği belirtilmiştir(Fernandes ve ark., 2013a; Fernandes ve ark., 2013b; Patacchini ve ark., 2005). Ancak, mesane dokusunda yaşlanmaya bağlı L-sistein/H_2S yolağı aktivitesinde oluşabilecek değişiklik ile ilgili bir çalışma mevcut değildir. Çalışmamızda yaşlanmaya bağlı detrusor kasındaki fonksiyonel değişikliklerde L-sistein/H_2S yolağının olası rolünün araştırılması amaçlanmıştır.

MATERYAL VE METOT

Çalışmalarda deney hayvanı olarak, Çukurova Üniversitesi Tıbbi Bilimler Deneysel Araştırma Merkezinden (DETAUM) temin edilen 25-35 gram ağırlıklarında swiss-albino genç (3-4 aylık) ve yaşlı (23-25 aylık) erkek fareler kullanıldı.

Çalışmada genç ve yaşlı fareler servikal dislokasyon ile öldürülerek, mesane detrusor dokuları izole edildi. İzole dokular, %95 O_2 ve %5 CO_2 ile gazlandırılan Krebs solüsyonu (mM: NaCI 118.1, KCI 4.7, CaCI2 2.5, MgCI2 6H2O 1.2, KH2PO4 1.2, NaHCO3 25, glucose 11.5) içeren 10 ml'lik organ banyolarına asıldı. Kas şeritleri 60 dakika boyunca inkübe edildi ve bu süre zarfında doku her 15 dakikada Krebs solüsyonu ile yıkandı. Kas uzunluğundaki değişiklikler izometrik bir dönüştürücü (MP35) aracılığıyla izometrik olarak kaydedildi.

Genç ve yaşlı farelerden izole edilen mesane şeritlerinin kasılma ve gevşeme yanıtları değerlendirildi. İlk deney grubunda, genç ve yaşlı fare gruplarından izole edilen idrar kesesi dokularında agonist kaynaklı kasılma yanıtları karşılaştırıldı. Şeritlerin kasılma kabiliyetini belirlemek için, izole edilmiş fare mesane şeritleri, 60 dakikalık dengeleme süresinden sonra 60 mM KCl ile önceden kastırıldı ve ardından dokular Krebs çözeltisiyle yıkandı ve 30 dakika boyunca yeniden dengelemeye bırakıldı. Bu süre sonunda kümülatif karbakol (10^{-4}-10^{-8} M) konsantrasyon-yanıt eğrisi elde edildi. Karbakol ile birinci seri kümülatif kasılma yanıtları elde edildikten sonra dokular 30 dakika dengelenmeye bırakıldı ve karbakol ile ikinci seri kümülatifkonsantrasyon-cevap eğrisi elde edildi.

Diğer deney grubunda ise, endojen H_2S'ye karşı gevşetici yanıtları değerlendirmek için mesane şeritleri karbakol ($3x10^{-6}$ M) ile önceden kastırıldı. Elde edilen kasılmadan sonra endojen H_2S substratı olarak kümülatif L-sistein (10^{-6}-10^{-2} M) uygulandı. Birinci seri gevşetici yanıtlar alındıktan sonra dokular 30 dakika inkübe edildi ve ikinci seri gevşemeler aynı şekilde kaydedildi.

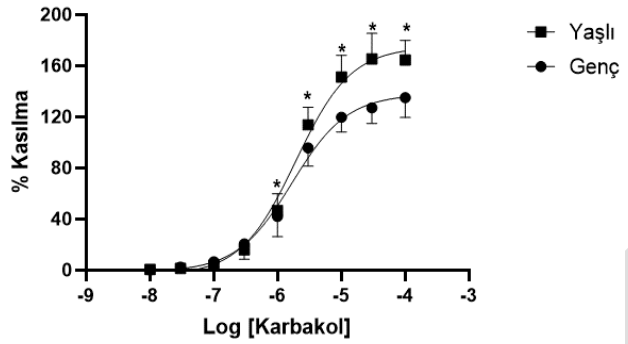
Ayrıca genç ve yaşlı farelerden izole edilmiş mesane dokularında ekzojen H_2S'ye verilen gevşetici tepkilerin değerlendirilmesi incelendi. Bu amaçla KCl (60 mM) sonrası kasılmalar elde edildi ve dokular yıkandı. 30 dakikalık inkübasyon süresinin ardından mesane şeritleri karbakol ($3x10^{-6}$ M) ile önceden kastırıldı. Kararlı kasılmalar elde edildikten sonra eksojen H_2S olarak kümülatif NaHS (10^{-6}-$3x^{10-3}$ M) uygulandı. Birinci seri gevşetici yanıtlar alındıktan sonra dokular 30 dakika inkübe edildi ve ikinci seri gevşemeler aynı şekilde kaydedildi.

İstatistiksel karşılaştırmalar için ikinci seri kasılmalar ve gevşemeler değerlendirildi. Tüm veriler ortalama ± S.E.M olarak sunulmuştur. Dokular arasındaki sonuçlardaki farklılıklar, tek yönlü varyans analizi ve çoklu karşılaştırmalar için düzeltilmiş eşleştirilmemiş t testi (Bonferroni düzeltmeleri) ile test edildi. 0,05'in altındaki *P* değerleri anlamlı kabul edildi.

BULGULAR ve TARTIŞMA

1.Yaşlanmanın karbakol kasılmaları üzerine etkisi

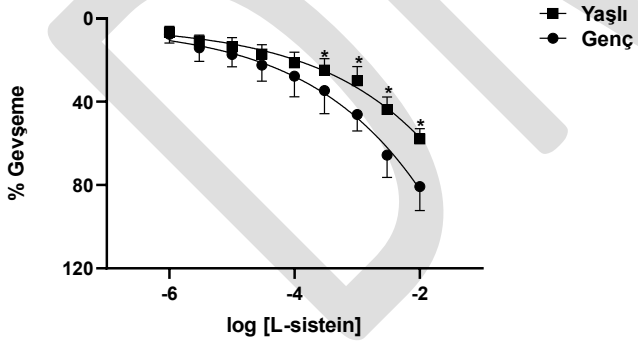
Yaşlanmanın karbakol kasılmaları üzerindeki etkisini araştırmak için genç ve yaşlı farelerde karbakolun kasılma yanıtları incelendi. Genç (3-4 aylık) ve yaşlı (23-25 aylık) farelerden izole edilen mesane şeritlerine spesifik M3-muskarinik reseptör agonisti karbakol kümülatif uygulanarak (10^{-8}-10^{-4} M) dokularda konsantrasyona bağlı kasılma yanıtları elde edildi. Karbakol' e verilen kasılma yanıtları, yaşlı (23-25 aylık) farelerde genç (3-4 aylık) farelere göre istatistiksel olarak anlamlı bir şekilde arttı ($P<0.05$; Şekil 1; n¬=5).



Şekil 1: Genç (3-4 aylık) ve yaşlı (23-25 aylık) farelerden izole edilen mesane şeritlende kümülatif karbakol (10^{-8}-10^{-4} M) kasılma yanıtları. Karbakol kasılma yanıtları 60 mM KCl'nin yüzdesi olarak ifade edildi. * genç fare grubuna göre anlamlılığı göstermektedir ($P<0.05$). Veriler ortalama ± SH olarak ifade edildi (n=5).

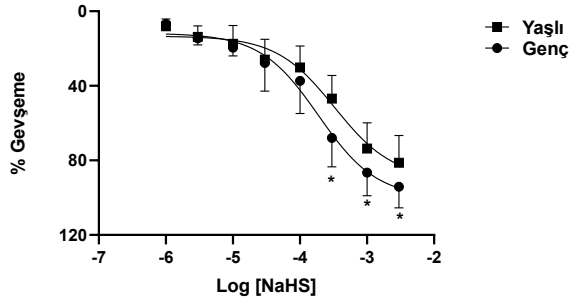
2. Yaşlanmanın H_2S sülfür gevşemeleri üzerine etkisi

Yaşlanmanın endojen H_2S gevşemeleri üzerindeki etkisini araştırmak için genç ve yaşlı farelerde endojen H_2S substratı L-sisteinin gevşeme yanıtları incelendi. Genç (3-4 aylık) ve yaşlı (23-25 aylık) farelerden izole edilen mesane şeritlerinde karbakol 10^{-6} M) ile kastırılmış dokulara kümülatif L-sistein (10^{-6}-10^{-2} M) uygulanması doza-bağlı gevşemeye neden oldu. L-sistein gevşeme yanıtları yaşlı (23-25 aylık) farelerde genç (3-4 aylık) farelere göre istatistiksel olarak anlamlı bir şekilde azaldı ($P<0.05$; Şekil 2; n¬=5).



Şekil 2: Genç (3-4 aylık) ve yaşlı (23-25 aylık) farelerden izole edilen mesane şeritlende kümülatif L-sistein (10^{-6}-10^{-2} M) yanıtlarının grafiği. * genç fare grubuna göre anlamlılığı göstermektedir ($P<0.05$). Veriler ortalama ± SH olarak ifade edildi (n=5).

Ayrıca, yaşlanmanın eksojen H2S gevşemeleri üzerindeki etkisini araştırmak için genç ve yaşlı farelerde eksojen H_2S prekürsörü NaHS gevşeme yanıtları incelendi. Genç (3-4 aylık) ve yaşlı (23-25 aylık) farelerden izole edilen mesane şeritlerinde karbakol 10^{-6} M) ile kastırılmış dokulara kümülatif NaHS (10^{-6}-10^{-3} M) uygulandı. Kümülatif NaHS (10^{-6}-10^{-3} M) uygulanması doza-bağlı gevşemeye neden oldu. NaHS gevşeme yanıtları yaşlı (23-25 aylık) farelerde genç (3-4 aylık) farelere göre istatistiksel olarak anlamlı bir şekilde azaldı ($P<0.05$; Şekil 3; n¬=5).



Şekil 3. Genç (3-4 aylık) ve yaşlı (23-25 aylık) farelerden izole edilen mesane şeritlende kümülatif NaHS (10-6-10-3 M) gevşeme yanıtlarının grafiği. * genç fare grubuna göre anlamlılığı göstermektedir (P<0.05). Veriler ortalama ± SH olarak ifade edildi (n=5).

SONUÇ

Sonuç olarak, yaşlı fare mesane düz kasında karbakol kasılmalarının genç fare grubuna göre artış göstermesi, mesaneden salınan endojen gevşetici maddelerin yaşlanmaya bağlı olarak azalmış olabileceğini düşündürmektedir. Ayrıca, yaşlanmaya bağlı olarak ürotelyumun hasar görmesi olasılığı da söz konusu olabilir. Endojen (L-sistein) ve ekzojen H_2S (NaHS) gevşeme yanıtlarının yaşlı farelerde genç farelere göre azalmış olması ise yaşlanmanın H_2S yolağını olumsuz olarak etkilediğini göstermektedir.

TEŞEKKÜR

Bu çalışma, "TAY-2019-11963" kodlu proje kapsamında Çukurova Üniversitesi Bilimsel Araştırma Proje Birimi tarafından desteklenmiştir.

REFERANSLAR

Brocklehurst JC, Dillane JB1966. *Studies of the female bladder in old age: II. Cystometrograms in 100 incontinent women*. Gerontol Clin (Basel), 8: 306–319.

Coolsaet B 1985. *Bladder compliance and detrusor activity during the collection phase*. Neuro Urodynamics, 4 (4): 263-273.

Daly DM, Nocchi L, Liaskos M, McKay NG, Chapple C, Grundy D 2014. *Age-related changes in afferent pathways and urothelial function in the male mouse bladder*. Send to J Physiol, 592(3): 537-49. doi: 10.1113/jphysiol.2013.262634.

Diokno AC, Brown MB, Goldstein N, Herzog AR 1992. *Epidemiology of bladder emptying symptoms in elderly men*.J Urol, 148(6): 1817-21.

Fernandes VS, Ribeiro AS, Barahona MV, Orensanz LM, Martínez-Sáenz A, Recio P, Martínez AC, Bustamante S, Carballido J, García-Sacristán A, Prieto D, Hernández M 2013a. *Hydrogen sulfide mediated inhibitory neurotransmission to the pig bladder neck: role of KATP channels, sensory nerves and calcium signaling*. J Urol.;190(2):746-56. doi: 10.1016/j.juro.2013.02.103.

Fernandes VS, Ribeiro AS, Martínez MP, Orensanz LM, Barahona MV, Martínez-Sáenz A, Recio P,

Benedito S, Bustamante S, Carballido J, García-Sacristán A, Prieto D, Hernández M 2013b. J Urol, *Endogenous hydrogen sulfide has a powerful role in inhibitory neurotransmission to the pig bladder neck.* 189(4): 1567-73.

Geokas MC, Conteas CN, Majumdar AP 1985. *The aging gastrointestinal tract, liver, and pancreas*. Clin Geriatr Med, 1: 177–205.

Patacchini R, Santicioli P, Giuliani S, Maggi CA 2005. *Pharmacological investigation of hydrogen sulfide (H2S) contractile activity in rat detrusor muscle*. Eur J Pharmacol., 21;509(2-3): 171-7.

Siroky MB 2004. *The Aging Bladder*. Rev Urol.; 6(Suppl 1): S3–S7.

ORAL PRESENTATION

Tortula lingulata Lindb. (Bryophyta)'nın Türkiye'den ikinci kaydı

Harun Çulha[1*](*ORCID: https://orcid.org/0000-0003-1537-1472*), Tülay Ezer[2] (*ORCID: https://orcid.org/0000-0002-6485-5505*)

[1*]Niğde Ömer Halisdemir Üniversitesi, Fen Edebiyat Fakültesi, Biyoloji Bölümü, Niğde, Türkiye
[2]Niğde Ömer Halisdemir Üniversitesi, Mimarlık Fakültesi, Peyzaj Mimarlığı Bölümü, Niğde, Türkiye

*Sorumlu yazar e-mail: culhaharun1@gmail.com

Özet

Bu çalışmada, ilk kaydı Erciyes Dağı'ndan verilen akrokarp karayosunu *Tortula lingulata* Lindb., Türkiye'den ikinci kez kaydedilmiştir. Avrupa'da nadir olan tür, bir deprem kırığı oluşumu olan Koramaz Vadisi'ndeki (Kayseri), Ağırnas yeraltı şehrinin girişindeki kumtaşı duvar üzerinden toplanmıştır. Avrupa IUCN Kırmızı Listesinde VU (hassas) kategorisinde değerlendirilen *Tortula lingulata*'nın morfolojik karakterleri fotoğraflarıyla birlikte verilmiş, ekolojisi ve Türkiye'deki yayılışı detaylı olarak anlatılmıştır.

Anahtar Kelimeler: Akrokarp, Briyofit, Koramaz Vadisi, Türkiye

The second record of _Tortula lingulata_ Lindb. (Bryophyta) from Türkiye

Abstract

In this study, the acrocarpous moss *Tortula lingulata* Lindb., first recorded from Erciyes Mountain, was recorded for the second time from Türkiye. The species, which is rare in Europe, was collected from the sandstone wall at the entrance of the Ağırnas underground city in the Koramaz Valley (Kayseri), which is an earthquake fracture formation. The morphological characters of *Tortula lingulata*, which is evaluated in the VU (vulnerable) category in the European IUCN Red List, are given with photographs, and its ecology and distribution in Türkiye are explained in detail.

Keywords: Acrocarpous, Bryophyte, Koramaz Valley, Türkiye

GİRİŞ

Briyofitler yaklaşık 15.000-25.000 tür ile dünyada Magnoliophyta'dan (~350 000 tür) sonra ikinci en büyük bitki grubudur. Yeryüzünde kutuplardan ekvatora suyun ve nemin var olduğu hemen her yerde yaşayabilirler (Glime, 2017).

Akrokarp karayosunu familyası olan Pottiaceae yaklaşık 80 cins ve 1.400'den fazla tür ile briyofitler içerisinde yer alan en büyük familyalardan birisidir (Zander, 1993). Pottiaceae üyelerinin çoğunluğu kurakçıl karakterli habitatlara uyum sağlamış olsa da yarı kurak ve sucul habitatlarda yayılış gösteren taksonları da içermektedir (Inoue ve Tsubota, 2016).

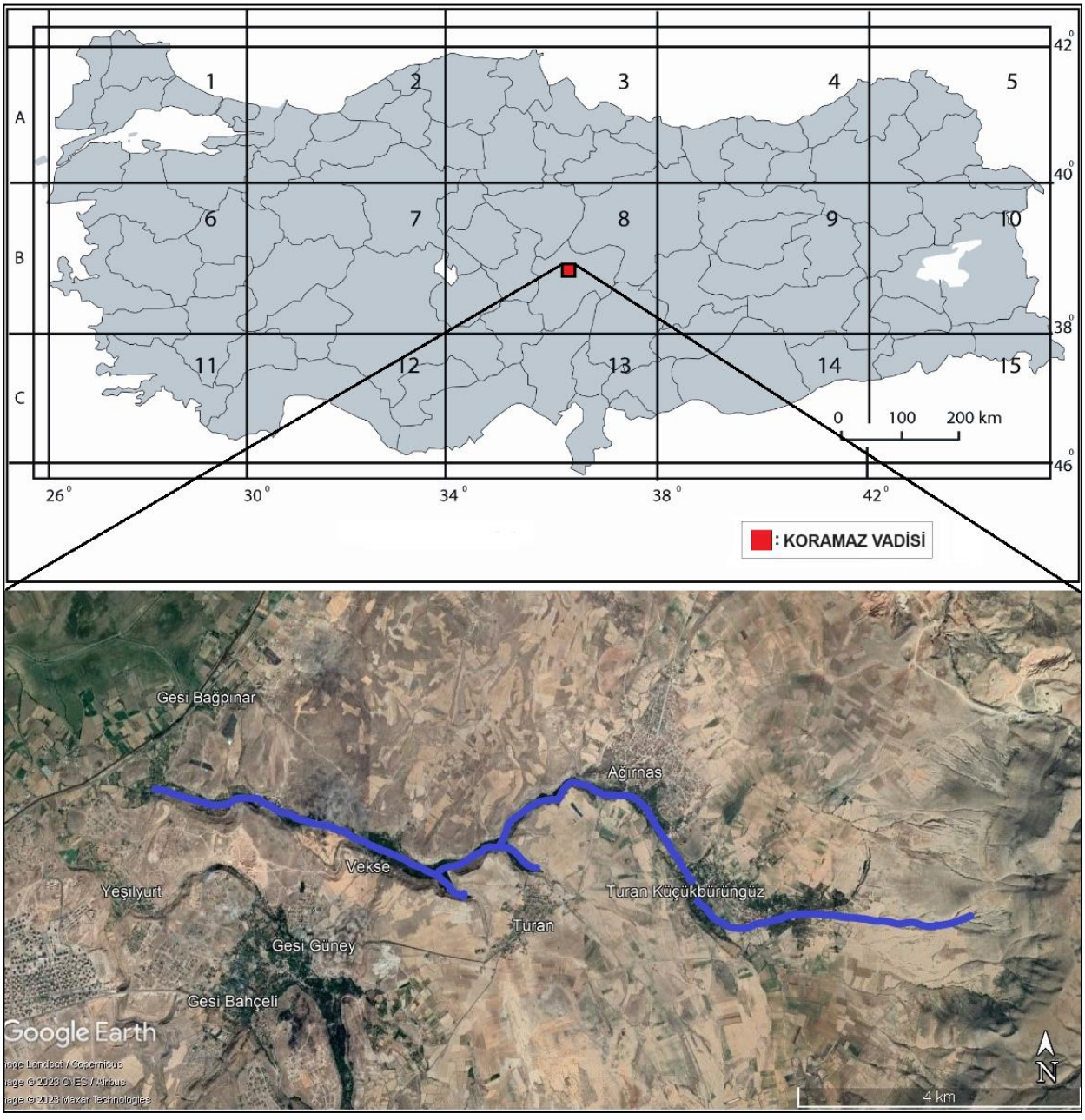
Pottiaceae familyası içerisinde *Tortula* Hedw. kozmopolit bir cins olup üyeleri morfolojik açıdan oldukça çeşitlilik gösteren bir gruptur. Cins dünya çapında yaklaşık 100 tür içermektedir (Košnar ve Kolář, 2009). Bu türlerden biri olan *Tortula lingulata* Lindb. Avrupa'da nadir bulunmakta ve özellikle Estonya'da koruma altındadır. Avrupa IUCN Kırmızı Listesinde VU (hassas) kategorisinde değerlendirilen *T. lingulata* Asya'da sadece Tacikistan'dan rapor edilmiştir (Ingerpuu ve ark., 2008). Türkiye'de ise tür ilk kez Erciyes Dağı'ndan Türkiye için yeni kayıt olarak saptanmıştır (Kara ve ark., 2014). Bu çalışmada, Koramaz Vadisi'nden toplanan *T. lingulata* Türkiye'den ikinci kez kaydedilmiş olup Asya için üçüncü lokalite kaydı verilmiştir.

DRAFT

MATERYAL VE METOT

Çalışma Alanı

Koramaz Vadisi, İç Anadolu Bölgesi'nin Kayseri ili, Melikgazi ilçesi sınırları içinde yer almakta olan 16 kilometrelik bir deprem kırığı oluşumudur. Henderson (1961) tarafından Türkiye briyofitleri için oluşturulmuş kareleme sistemine göre B8 karesinde yer almakta olan vadi, Kayır Hanı mevkiinden başlamakta ve Korumaz Dağı'nda son bulmaktadır (Şekil 1).



Şekil 1. Henderson (1961) kareleme sistemi ve Koramaz Vadisi (Google Earth'den değiştirilerek)

Orta Anadolu'daki Ihlara Vadisi'nden sonra en uzun vadi olma özelliğini taşıyan Koramaz Vadisi'nin batı kısmında yükseklik 1165 metre iken, doğu ucunda 1500 metreye kadar çıkmaktadır. Yerleşim alanlarının da yer aldığı Koramaz Vadisi üzerinde Isbıdın (Bağpınar), Vekse, Dimitre (Turan), Ağırnas, Kiçi Bürüngüz (Küçük Bürüngüz), Üskübü (Subaşı) ve Ulu Bürüngüz (Büyük Bürüngüz) olmak üzere 7 adet köy

bulunmaktadır. Vadinin en derin noktası Vekse ve Turan mahalleleri arasındaki Mançur mevkiidir (Cömert, 2008; Koramaz Vadisi, 2023; Yazlık, 2019; Allı, 2021).

Koramaz Vadisi UNESCO (Birleşmiş Milletler Eğitim, Bilim ve Kültür Örgütü) tarafından 2020 yılında Dünya Miras Listesi'ne dahil edilmesi üzere Dünya Miras Geçici Listesi'ne alınmıştır. Vadi sahip olduğu kültürel ve tarihi özelliklerinden dolayı uzun yıllardır kullanımda olduğu için bünyesinde tarihi taş evler, çeşmeler, su değirmenleri ve tümülüsler bulundurmaktadır (UNESCO, 2022). Ayrıca vadide 6 adet yeraltı şehri de bulunmaktadır. Bunlardan en büyüğü 1273 metre uzunluğuyla Bürüngüz yeraltı şehri olup, Türkiye'de tespit edilen en uzun yeraltı şehridir (Allı, 2021; Koramaz Vadisi, 2023).

Veri Kaynağı

Çalışmanın materyalini 19.11.2022 tarihinde Koramaz Vadisi'ne gerçekleştirilen arazi çalışmaları sırasında toplanan briyofit örnekleri oluşturmaktadır. Örnekler ilgili literatürler kullanılarak teşhis edilmiş olup Prof. Dr. Tülay Ezer'in kişisel koleksiyonunda ve Niğde Ömer Halisdemir Üniversitesi Herbaryumunda koruma altına alınmıştır (Zander, 1993; Cortini-Pedrotti 2001, Smith 2004; Košnar ve Kučera, 2010; Lüth, 2019; Dihoru, 2020).

BULGULAR ve TARTIŞMA

***Tortula lingulata* Lindb.** Revue Bryologique 7(3): 40–41. 1880. (Rev. Bryol.).

Sinonimleri: *Barbula montenegrina* Breidl. & Szyszył., *Tortula montenegrina* (Breidl. & Szyszył.) Broth.

İncelenen örnek: Türkiye, Orta Anadolu, Kayseri, Koramaz Vadisi, Ağırnas yeraltı şehri, giriş kapısı kumtaşı duvar üzeri (Şekil 2), 1301 m, 38°48'46"K 35°42'58"D, H.Ç.67, 19 Kasım 2022 (Niğde Ömer Halisdemir Üniversitesi Herbaryumu).

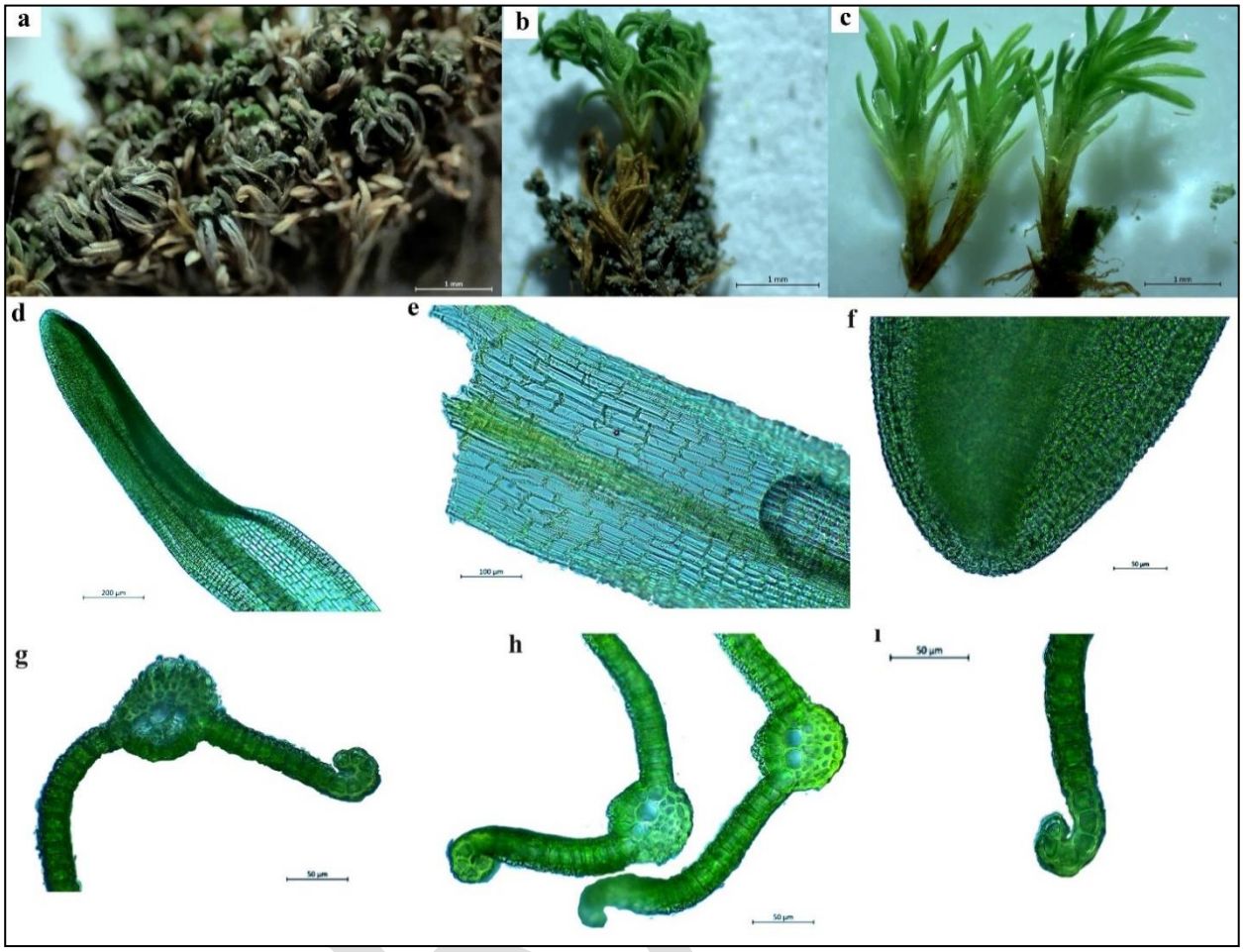


Şekil 2. *T. lingulata*'nın habitatı

Türün Morfolojik ve Anatomik Özellikleri

Tortula lingulata, küçük yoğun yeşil demetler halinde görülmekte olup kısa çim (short turf) hayat formu göstermektedir. Bitki dik gövdelidir ve 2,5 mm uzunluğundadır. Dilsi (lingulat) yapraklar kuruyken hafifçe kıvrık, nemliyken neredeyse dik bir şekilde durur. Yaprak ucu yuvarlak ve kukullattır. Yapraklar 700 x 250 µm boyutlarında olup yaprak kenarı kıvrıktır. Yaprak taban hücreleri dar dikdörtgen (35 x 17 µm), düz ve renksizken yaprağın orta ve üst hücreleri (19 x 12 µm) yuvarlak ve güçlü papillalıdır. Yaprak kenar hücreleri yaprağın ortasından itibaren birkaç sıralı olarak farklılaşmış olup bu hücreler kalın çeperli, hafif papillalı veya

papillasızdır. Ortalama 50 µm genişliğindeki kosta (orta damar) genellikle yaprağın ucunun hemen altında sonlanır (Şekil 3).



Şekil 3. *Tortula lingulata*. **a.** genel görünüm, **b.** kuru hali, **c.** nemli hali, **d.** yaprak, **e.** yaprak tabanı, **f.** yaprak ucu, **g, h.** yaprak enine kesitleri, **ı.** yaprak kenarı enine kesiti

Seta parlak kırmızı, ince, bükülebilir ve yaklaşık olarak 3 cm uzunluğundadır. Kapsüller yumurta biçiminde, geniş ağızlı, peristom dişleri kuru olduğunda dik, sporlar ise 20-30 µm'dir (Smith, 2004). İncelenen örnek steril olup sporofit görülmemiştir.

Türün Ekolojisi ve Dağılımı

Gölge habitatları tercih eden (sciofit) *Tortula lingulata* Avrupa'da (Estonya, Litvanya, Rusya, Ukrayna, Gürcistan, Çekya, Almanya, Romanya) kumtaşı anakaya üzerinde yayılış gösteren nadir bir türdür (Dierβen, 2001). Karadağ ve Fransa'daki yayılışı şüpheli olup *Tortula lingulata* var. *montenegrina* olarak rapor edilmiştir. *T. lingulata* Asya'da Tacikistan'dan ve Anadolu'dan kaydedilmiş olup dünyanın diğer kıtalarında rastlanılmamıştır (Ingerpuu ve ark., 2008; Kara ve ark., 2014; Dihoru, 2020). Bu çalışma ile Anadolu'dan ikinci kaydı ve Asya'dan da üçüncü lokalite kaydı verilmiştir. Avrupa Karayosunları, Ciğerotları ve Boynuzlu Ciğerotları Kırmızı Listesi'ne göre *Tortula lingulata* Avrupa ve AB (Avrupa Birliği) ülkeleri arasında VU (hassas) kategorisinde yer almaktadır (Hodgetts ve ark., 2019).

T. lingulata morfolojik olarak *T. obtusifolia* ile benzerdir. Ancak *T. lingulata*'nın yaprak ucunda sonlanan kostası ile geriye kıvrık yaprak kenarlarının birkaç sıra hücrelerinin kalın duvarlı ve hemen hemen papillasız olması *T. obtusifolia*'dan ayırdedici karakterleridir (Şekil 3, g-ı) (Košnar ve Kolář, 2009).

SONUÇ

Tortula lingulata Avrupa'da olduğu gibi Ülkemizde de nadir bir tür olup her iki kaydı da Orta Anadolu'da Kayseri ilinden verilmiştir. Türkiye'de briyofloristik çalışmalar hızla ilerlese de henüz çalışılmamış çok alan bulunmaktadır. Bu çalışma Türkiye'nin briyoflorasına katkı sağlayacaktır.

DRAFT

TEŞEKKÜR

Bu çalışma, FMT 2022/12-LÜTEP nolu proje ile desteklenmiş olup Niğde Ömer Halisdemir Üniversitesi, Bilimsel Araştırma Projeleri Koordinasyon Birimine teşekkür ederiz.

KAYNAKLAR

Allı H 2021. Koramaz Vadisi (Kayseri) ve Çevresi Makrofungusları. Türler ve Habitatlar. 2(2): 77-85.

Cortini Pedrotti C 2001. Flora dei muschi d'Italia, Sphagnopsida, Andreaopsida, Bryopsida (I parte). Medicina-Scienze. Roma.

Cömert H 2008. Koramaz Vadisi. Ağırnas Belediyesi Yayınları.

Dierssen K 2001. Distribution, ecological amplitude, and phytosociological characterization of European bryophytes. Bryophytorum Bibliotheca, Bd. 56, J. Cramer, Berlin Stuttgart.

Dihoru G 2020. *Tortula lingulata* in Romanian brioflora. Romanian Journal of Biology -Plant Biology 65(1-2): 43-47.

Glime JM 2017. Chapter 2. Life cycles and morphology, Bryophyte Ecology. Vol. 1. Physiological Ecology.

Henderson DM 1961. Contributions to the bryophyte flora of Turkey: V. Summary of present knowledge. Notes Roy Bot Gard Edinburgh 23: 279-301.

Hodgetts N, Cálix M, Englefield E, Fettes N, García Criado M, Patin L, Nieto A, Bergamini A, Bisang I, Baisheva ve ark. 2019. A miniature world in decline: European Red List of Mosses, Liverworts and Hornworts. Brussels, Belgium: IUCN.

Ingerpuu N, Maasikpalu K, Vellak K 2008. Morphology and habitat properties of *Tortula lingulata* in Estonia. Folia Cryptogamiva Estonica, Fasc. 44: 49-54.

Inoue Y, Tsubota H 2016. Systematics of the family Pottiaceae (Bryophyta) with special reference to the familial and subfamilial circumscriptions. Hikobia. 17: 117-129.

Kara R, Ezer T, Gözcü MC, Bozdoğan ŞG 2014. Bryophyte flora of Erciyes Mountain in Turkey, with 6 bryophyte records from the country. Turkish Journal of Botany. 38(4): 763-781.

Koramaz Vadisi 2023. Yerleşim Yerleri, http://www.koramazvadisi.com/yerlesim-yeleri/index.html, (05.05.2023)

Košnar J, Kolář F 2009. A taxonomic study of selected European taxa of the *Tortula muralis* (Pottiaceae, Musci) complex: variation in morphology and ploidy level. Preslia, 81: 399-421.

Košnar J, Kučera J 2010. The taxonomic identity and typification of *Barbula montenegrina* Breidl. & Szyszyl. (Bryopsida, Pottiaceae). Journal of Bryology. 32: 275-278.

Lüth M 2019. Mosses of Europe-A Photographic Flora Set of 3 Volumes, ISBN 978-3-00-062952-5.

Smith AJE. 2004. The moss flora of Britain and Ireland, Cambridge (GB): Cambridge University Press.

Yazlık B 2019. Koramaz Vadisi Columbarium Mezarları. Turkish Studies-Historical Analysis. 14(3): 669-733.

Zander RH 1993. Genera of the Pottiaceae: mosses of harsh environments. Bull Buffalo Soc Nat Sci., 32: 1-378.

DRAFT

ORAL PRESENTATION

The enzyme activity potentials of *Gladiolus halophilus* and *Gypsophila lepidioides* plant species growing in cypsiferous soils of Eastern Anatolia against some enzymes (tyrosinase and α-glycosidase).

Ali Rıza TÜFEKÇİ[1*] (*ORCID: 0000-0002-2951-3657*), Tuğçe VAROL[2] (*ORCID:0000-0001-7027-X*), Samed ŞİMŞEK[3] (*ORCID: 0000-0001-8451-3425*), Zeyad Adil HAMEED[4] (*ORCID: 0000-0001-5359-3194*), İbrahim FİLAZİ[5] (*ORCID: 0000-0002-8194-1421*), Şevki ADEM[6] (*ORCID: 0000-0003-2146-5870*), Hüseyin AKŞİT[7] (*ORCID: 0000-0002-1509-851X*), Etem OSMA[8] (*ORCID: 0000-0002-5250-8194*)

[*1]Çankırı Karatekin University, Faculty of Science, Department of Chemistry, Çankırı, Turkey.
[2]Erzincan Binali Yıldırım University, Faculty of Sciences and Arts, Department of Biology, Erzincan, Turkey
[3]Erzincan Binali Yıldırım University, Çayırlı Vocational School, Department of Medical Services and Techniques, Erzincan, Turkey.
[4]Çankırı Karatekin University, Faculty of Science, Department of Chemistry, Çankırı, Turkey.
[5]Çankırı Karatekin University, Central Laboratory Application and Research Center (ÇANKAM), Çankırı, Turkey.
[6]Çankırı Karatekin University, Faculty of Science, Department of Chemistry, Çankırı, Turkey.
[7]Erzincan Binali Yıldırım University, Faculty of Pharmacy, Department of Analytical Chemistry, Erzincan, Turkey.
[8]Erzincan Binali Yıldırım University, Faculty of Sciences and Arts, Department of Biology, Erzincan, Turkey.

*Corresponding author e-mail: areb@karatekin.edu.tr

Abstract

In this study, the inhibition effects of *Gladiolus halophilus* (GH) and *Gypsophila lepidioides* (GL) plants, which are endemic species growing in the chiselled soils of Erzincan province, Eastern Anatolia region, against tyrosinase and α-glycosidase enzymes were investigated. Methanol extracts of leaves, branches and roots of the plants were used in the study. GHY, GHD and GHK coded extracts with IC_{50} values of 75.17 µg/mL, 105.02 µg/mL and 83.92 µg/mL showed good activity against tyrosinase enzyme under *in vitro* conditions. The tyrosinase α-amylase inhibition effects of the extracts obtained from *G. lepidioides* were found to be lower. On the other hand, α-amylase enzyme inhibition effects of *G. halophilus* plant extracts were found to be very low.

Keywords: *Gladiolus* halophilus, Gypsophila *lepidioides,* enzyme activity, tyrosinase, α-glycosidase

INTRODUCTION

Many health problems have occurred in recent years with global warming, uncontrolled nutrition and increased genetic disturbances in the results affecting this. Diseases such as Alzheimer's diseases, skin cancer and diabetes are increasing very quickly (Yırtıcı, 2019). Treatment of these diseases with synthetic drugs is tesr in one place. Instead, studies on finding new therapeutic agents are increasing. It is hoped that the next decades, especially Alzheimer's disease and diabetes diseases, will affect approximately 81.1 million people by 2040 (Dhouafli et al., 2018). Diabetes's disease increases the risk of getting Alzheimer's disease. Therefore, studies on inhibition of α-glucosidase, which is important in controlling hyperglycemia, are important (Li et al., 2018). In addition, Type II diabetes can cause hyperpigmentation on the skin, and inhibition of tyrosinase can prevent hyperpigmentation (Mendes et al., 2017).

Enzymes have important tasks in various physiological processes, such as growth in plants, development, metabolic events and responses to environmental stresses. The tyrosinase enzyme is a very important enzyme found in most living groups, including plants, bacteria and mammals. Hydroxylation of monophenols to o-

DRAFT

diphenols using molecular oxygen as the mechanism of action; It is an enzyme containing copper that catalyzes the oxidation of o-quinones in o-diphenols. In the presence of molecular oxygen, they also play a role in the conversion of monophenols to those dihydroxyphenols in addition to the oxidation reactions of o- and vic-OH group (3,4,5-trihydroxy) phenolic compounds (Vamos-Vigyazo et al., 1981). Tyrosinase and polyphenols are commonly found in plants. It is an enzyme that was first determined to be abundant in edible mushrooms and then in most fruits and vegetables, and whose importance is understood afterwards (Keleş et al., 1987). The most pioneering benefit of tyrosinases in plants can be expressed as the creature's resistance to microbial or viral infections and resistance to unfavorable climatic conditions. The amount of tyrosinase in plants depends on its variety, age, cultural processes and maturity (Spille et al., 1997).

Tyrosinase industry, pulp and paper industry, textile industry, pharmaceutical and environmental technology are well suited for some biotechnological applications. Apart from that, it can be used as an antioxidant and nutrient colorant (Slap meat al., 2007). The high content of secondary metabolites gives the plant very strong biological activity such as antioxidant, antimicrobial, insectide, anticancer (Demirtaş et al., 2017). Tyrosinase enzyme inhibitors are substances that can inhibit the activity of the tyrosinase enzyme. Tyrosinase is an important enzyme involved in the biosynthesis of melanin, the pigment responsible for the color of our skin, hair and eyes. While melanin is necessary to protect the skin from harmful UV radiation, its overproduction can lead to a variety of skin conditions such as hyperpigmentation, melasma and age spots. Tyrosinase inhibitors are activated here. Tyrosinase inhibitors can help fade these pigmented areas and improve the overall appearance of the skin. In addition, the fact that this basic problem cannot be solved and another missing problem in the body causes the emergence of other problems. The lack of these tyrosinase inhibitors can cause skin cancer (Yırtıcı et al., 2022).

Diabetes mellitus is a multifactorial global health disorder that is rising at an alarming rate. Alpha-glucosidase inhibitors are used to treat type 2 diabetes. Plants contain many compounds that inhibit this enzyme. Plant extracts can have a strong inhibitory effect on the enzyme due to both the main component and the synergistic effect. In addition, they have the important advantage of being able to interact directly with the enzyme found in the small intestine. Therefore, it has an important place in research (Dirir et al., 2022).

This study describes the inhibition of endemic *Gypsophila lepidioides* and methanol extracts from the *Gladiolus halophilus* plant grown in the chips of the Eastern Anatolia region against some enzymes. Since *Gypsophila lepidioides* and *Gladiolus halophilus* plants are not particularly on enzyme activity studies in the literature, the results that occur here are noteworthy in terms of being an important scientific data.

MATERIALS AND METHOD

Plant materials and Extracts preparation

In the June-July, plant materials used in the study were collected during the flowering period from the serpentine soils in the serpentine areas around Yücebelen Village and 3. km away from the direction of Yahşiler Village on the Kemah-İliç highway in Erzincan province. *Gypsophila lepidioides* (37S 484606 E, 4381349 N, 1320 m) was recorded under herbarium number Kandemir 11534B and *Gladiolus halophilus* (37S 492090 E, 4372902 N, 1461 m) was recorded under herbarium number Kandemir 11340 in Erzincan University herbarium. The dried plants were divided into three parts: leaf, stem and root. The aerial parts were ground into a fine powder using a laboratory mill. In methanol, 100 grams of powdered materials were macerated. The extract was subsequently concentrated under a vacuum in a rotary evaporator at 40 °C. All extracts were stored in dark and 4°C until working time.

Chemicals

All materials including chemicals and solvents used for enzyme activity studies and extraction procedures in this study were obtained from Sigma Aldrich, Fluka and Biological Industries (BI, USA).

Enzyme activity tests

The tyrosinase inhibitory activity properties of extract were determined by making slight modifications in the dopachrome method used by Sarıkurkcu et al. (Sarıkurkcu et al. 2018, Tüfekçi et al., 2023). Accordingly, 25 mL of each extractant was taken from the stock extract solutions prepared at a ratio of 1:1 in DMSO and 40

mL of tyrosinase solution and 100 mL of sodium phosphate buffer (pH 6.8) were added and mixed. The mixture was incubated at 25°C for 15 minutes. After adding 40 mL L-DOPA, the mixture was incubated again at 25°C for 10 min. Absorbance was measured at 492 nm. Scutellarin compound was used as a positive control and the result was determined by comparing the inhibitory effect of this compound on tyrosinase enzyme with the effect of the compound. For this, 1 mg of scutellarin compound was weighed and dissolved in 1 mL DMSO, then diluted ten times with distilled water. Enzyme activity was tested at five different scutellarin concentrations.

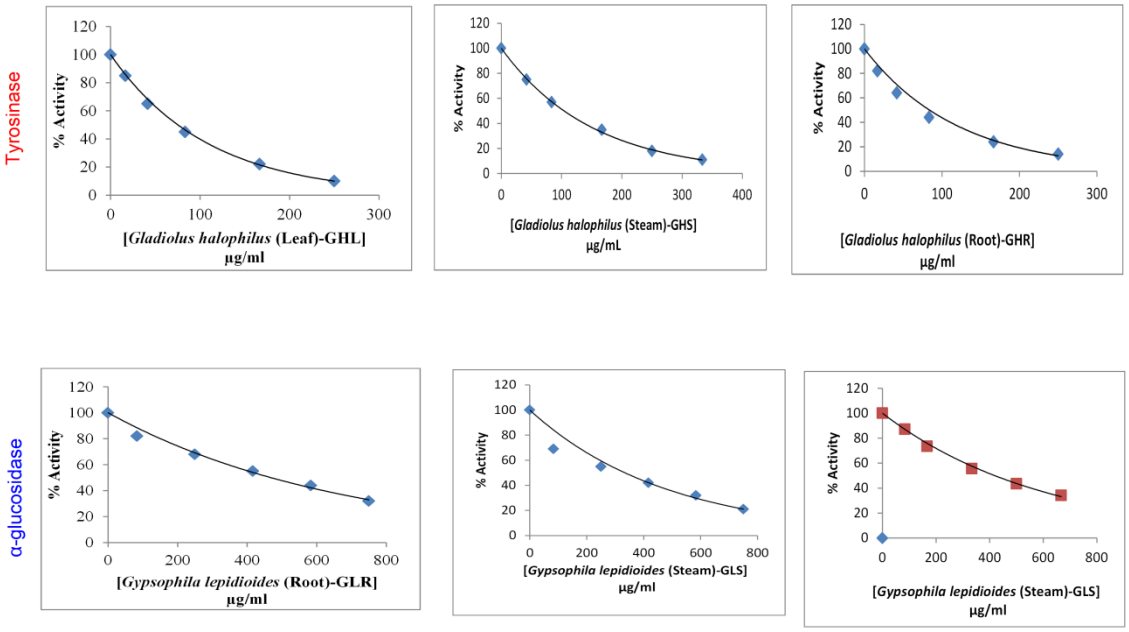


Figure 1. The inhibition effect of extracts on tyrosinase and a-glucosidase activity

The a-glucosidase inhibitory activity was determined using paranitrophenyl- a-D-glucopyranoside (pNPG) as substrate (Uysal et al., 2021). The extract (50 mL) was mixed with 50 mL a-glucosidase solution prepared in a buffer (sodium phosphate, pH 6.8), and 50 mL pNPG. The mix was incubated at 37°C for 15 min, and the reaction was followed with a spectrophotometer at 408 nm for 5 minutes.

The IC_{50} value was calculated by plotting the percentage inhibition of enzyme activity against the sample concentration. The concentration at which the enzyme activity was inhibited by 50% was determined as the IC_{50} value. The lower the IC_{50} value, the stronger the inhibitory activity of the extract against a-glucosidase. To compare the inhibitory activity of the extract with acarbose, the IC_{50} value of acarbose was also determined using the same method. The extract's inhibitory activity was then expressed in terms of mg standard equivalent per gram of extract, which represents the amount of extract required to achieve the same inhibitory activity as 1 gram of acarbose. This method allows for the quantification and comparison of the a-glucosidase inhibitory activity of the extract with a known standard (acarbose) and provides a measure of the extract's potential as an inhibitor of this enzyme.

RESULTS AND DISCUSSION

Table 1. Enzyme inhibitory activities of *G. halophilus* and *G. lepidioides* methanol extract.

Plant	IC_{50}=µg/mL	
	Tyrosinase	α-glucosidase
***Gladiolus halophilus* (Leaf)-GHL**	75.17	420.08
***Gladiolus halophilus* (Steam)-GHS**	105.02	-
***Gladiolus halophilus* (Root)-GHR**	83.91	-
***Gypsophila lepidioides* (Leaf)-GLL**	396.08	468.34
***Gypsophila lepidioides* (Steam)-GLS**	344.85	334.85
***Gypsophila lepidioides* (Root)-GLR**	465.19	266.59
Acarbose (Standart)	-	51.40
Scutellarin (Standart)	20.38	-

Thyrosinase and a-glucosidase inhibitory activity tests were performed to determine the anti-diabetic properties of methanol extracts from different branches of *G. halophilus* and *G. lepidioides* plants, as well as their protective properties for skin health. The results are presented as IC_{50} values (**Table 1**).

Tyrosinase inhibitory activity also differs between plant species and the plant's own branches. Enzyme activity results on this plant species have been reported for the first time. It has been observed that the Gladiolus halophilus plant has a high inhibition effect against the thyrosinase enzyme. The IC_{50} value of GHL extract was found as 75.17 µg/mL and was found to have the highest tyrosinase enzyme activity. GHR extract was determined to have the second highest inhibition with the IC_{50} value of 83.91 µg/mL and an inhibition property that came later than the other two branches with the GHS extract 105.02 µg/mL IC_{50} value.

The inhibitor effect of GL and GH extracts against the glucosidase enzyme has been found to be minimal. These differentials between the extras come from the adherence to the type and content of the chemical components it contains. The ability to inhibit this enzyme with its general reputation and any light exposed to the sun is generally seen in the aboveground parts. There is a mechanical synthesis event as a result of a kind of stress condition. The interesting thing is that this event fully supports the amount of content, and the extract belonging to the root part contains components that affect high inhibition (Figure 1).

CONCLUSION

In this study, tyrosinase and a-glucosidase inhibitory effects of *G. halophilus* and *G. lepidioides* were tested. *G. halophilus* extracts have been an effective tyrosinase inhibitor in studied plants. In addition, the a-glucosidase enzyme inhibition of *G. halophilus* and *G. lepidioides* extracts was found low. Although ghalophilus extracts generally show a high activity, it is necessary to first identify the compounds responsible for the activity and conduct additional in vivo, in vitro studies in order to use it as an inhibitor.

ACKNOWLEDGEMENTS
We thanks to Çankırı Karatekin University for their support in conducting this study.

REFERENCES

Demirtas I, Tufekci AR, Yaglioglu AS, Elmastas M 2017. Studies on the antioxidant and antiproliferative potentials of *Cirsium arvense* subsp. *vestitum*. Journal of Food Biochemistry, 41(1): e12299.

Dhouafli Z, Rigacci S, Leri M, Bucciantini M, Mahjoub B, Tounsi MS, Wannes WA, Stefani M, Hayouni EA 2018. Screening for amyloid-β aggregation inhibitor and neuronal toxicity of eight Tunisian medicinal plants. Industrial Crops and Products, 111: 823-833.

Dirir AM, Daou M, Yousef AF, Yousef LF 2022. A review of alpha-glucosidase inhibitors from plants as potential candidates for the treatment of type-2 diabetes. Phytochemistry Reviews, 21(4):1049-1079.

Keleş F. 1987. Gıdalarda Enzimatik Esmerleşme ve Kontrolü, Doğa Dergisi, 11:105-121.

Li W, Risacher SL, Gao S, Boehm SL, Elmendorf JS, Saykin AJ 2018. Type 2 diabetes mellitus and cerebrospinal fluid Alzheimer's disease biomarker amyloid beta1-42 in Alzheimer's Disease Neuroimaging Initiative participants. Alzheimers Dement (Amst), 10:94-98.

Mendes AL, Miot HA, Haddad VJ 2017. Diabetes mellitus and the skin. An Bras Dermatol, 92 (1):8-20.

Reddy DM, Reddy GVB, Mandal PK. 2018. Application of natural antioxidants in meat and meat products-A review. Food Nutrition Journal, 2018(03):1-12.

Sarikurkcu C, Kirkan B, Ozer MS, Ceylan O, Atilgan N, Cengiz M, Tepe B 2018. Chemical characterization and biological activity of *Onosma gigantea* extracts. Industrial Crops Products, 115:323-329

Spille GA. 1997. Caffeine, Chapter 3. Tea: The Plant and its manufacture; Chemistry and Consumption of the Beverage, CRC Press, 1-38.

Şimşek Ş, Yemenicioğlu A 2007. Partial Purification and kinetic characterization of Mushroom stem polyphenol oxidase and determination of its storage stability in different lyophilized forms, Process Biochemistry, 42:943-950.

Tüfekçi AR, Akşit H, Şimşek S, Karakoç ÖC, Adem Ş, Hameed ZA, Atalar MN, Topkara AR. 2023. Evaluation of insecticidal and enzyme activity potentials of essential oils and extracts of Chenopodium botrys against storage products pests. Bulletin of Biotechnology, 4(1):7-12.

Vamos-Vigyazo L. 1981. Polyphenol oxidase and peroxidase in fruits and vegetables, CRC Critical Reviews Food Science and Nutrition, 15:49-127.

Yirtici U. 2019. The determination of antioxidant properties and enzyme inhibition effect of *Centaurea fenzlii* Reichardt extract. Bitlis Eren Univ J Sci, 8(1):66-73.

Yırtıcı Ü, Ergene A, Atalar MN, Adem Ş 2022. Phytochemical composition, antioxidant, enzyme inhibition, antimicrobial effects, and molecular docking studies of *Centaurea sivasica*. South African Journal of Botany, 144: 58-71.

DRAFT

ORAL PRESENTATION

Molecular docking studies of Quercimetrin (Quercetin 7-*O*-D-glucoside) from *Centaurea paphlagonica* (Bornm.) Wagenitz

Ali Rıza TÜFEKÇİ[1*] (*ORCID: 0000-0002-2951-3657*), Emel EKİNCİ[2] (*ORCID:0000-0003-2323-2747*), Zeyad Adil HAMEED[3] (*ORCID: 0000-0001-5359-3194*), Şevki ADEM[4] (*ORCID: 0000-0003-2146-5870*

[*1]Çankırı Karatekin University, Faculty of Science, Department of Chemistry, Çankırı, Turkey.
[2]Çankırı Karatekin University, Central Laboratory Application and Research Center (ÇANKAM), Çankırı, Turkey.
[3]Çankırı Karatekin University, Faculty of Science, Department of Chemistry, Çankırı, Turkey.
[4]Çankırı Karatekin University, Faculty of Science, Department of Chemistry, Çankırı, Turkey

*Corresponding author e-mail: areb@karatekin.edu.tr

Abstract

In this study, Quercimetrin (Quercetin 7-*O*-D-glucoside) compound was isolated first time from *Centaurea paphlagonica*. The inhibitory effects of quercimetrin compound was tested towards tyrosinase enyzme. As a result of the enzyme inhibition experiment, *C. paphlagonic*a extract was found to be very active on the tyrosinase enzyme. The quercimetrin showed with IC_{50} value 14.47 µM against studied enzyme. The molecular docking study was performed in order to explain the interactions of the quercimetrin compound with target enzyme. The dock score was obtained as -131.986. As a result, isolated compound displayed a high inhibition effect against studied tyrosinase enzyme.

Keywords: Molecular docking, flavonoids, enzyme activity, tyrosinase, *Centaurea paphlagonica*

INTRODUCTION

Species of the genus *Centaurea* are generally distributed throughout Europe, Asia, North Africa and parts of America. This genus has a wide geographical distribution and the specific distribution of species may differ from species to species. These species are commonly found in alpine meadows, steppes, mountainous areas and agricultural land. On the other hand, the distribution of species within the genus Centaurea can vary depending on the specific species, and some species can only be found in certain geographical regions. The genus *Centaurea* is a plant genus with a fairly large number of species worldwide and in Turkey. The number of species of this genus worldwide is estimated to be around 700 to 800 (Kılıc et al., 2016). Due to the rich plant diversity of Turkey, the number of species of the genus *Centaurea* in Turkey is quite high. It is thought that there are more than 200 *Centaurea* species in Turkey (Tasar et al., 2018). These species are distributed in different geographical regions and habitats. Turkey's diverse climatic and topographic conditions increase the diversity of *Centaurea* species. The biological activities of plants belonging to the genus *Centaure*a may vary depending on the plant species and the components they contain. Since *Centaurea* species contain unique compounds, they offer a wide range of activity potential of the plant species. The high content of secondary metabolites gives the plant very strong biological activity such as antioxidant, antimicrobial, insecticide, anticancer (Demirtaş et al., 2017).

Enzymes play important roles in various physiological processes in plants, such as growth, development, metabolism and responses to environmental stresses. Centaurea species produce antioxidant enzymes such as superoxide dismutase (SOD), catalase and peroxidase. These enzymes help the plant fight oxidative stress by neutralising harmful reactive oxygen species (ROS) produced during various metabolic processes and in response to environmental stresses. Some Centaurea species increase the synthesis of phenolic compounds by activating enzymes such as phenylalanine ammonia lyase (PAL) and polyphenol oxidase (PPO) in defence against herbivores and pathogens. *Centaurea* species possess a number of metabolic enzymes involved in processes such as photosynthesis, respiration and synthesis of secondary metabolites such as alkaloids and

DRAFT

flavonoids. These enzymes play key roles in plant growth and response to environmental cues. In response to various environmental stresses such as drought, salinity and pathogen attack, Centaurea species can modulate the activity of stress-associated enzymes such as abscisic acid (ABA)-related enzymes and defence enzymes such as chitinases and glucanases. Some Centaurea species are used in traditional medicine and their enzyme activities may be related to their therapeutic properties. For example, some species may contain enzymes involved in the production of bioactive compounds with potential medicinal properties.

Research on *Centaurea* species shows that some species have tyrosinase enzyme activity. Tyrosinase is an enzyme responsible for the synthesis of phenolic compounds and plays a critical role in melanin production. Melanin is a pigment that provides pigmentation in the skin, hair and retina of the eye. Tyrosinase activity may be involved in various biological functions in plants, such as pigment synthesis, defence responses and antioxidant protection (Yırtıcı et al., 2022). Some *Centaurea* species may possess tyrosinase activity, and this activity may help the plant to regulate its phenolic components and melanin production. In particular, it is thought that some *Centaurea* species may have the potential to provide pigmentation regulation or antioxidant properties in medicinal or cosmetic uses.

Skin cancer, one of the most prevalent cancers in the world, continues to increase at alarming rates in both incidence and fatality (Ismaya et al., 2011). One of the most dangerous effects of skin cancer is melanoma, which is marked by active melanocyte proliferation and increased melanin pigment accumulation, resulting in skin pigmentation and discolouration. Tyrosinase enzyme levels that have been upregulated seem to have a key role in the increased production and storage of melanin in melanocytes (Rao et al., 2013).

Tyrosinase is a multifunctional and copper-containing enzyme and has very important functions in plants and animals. This enzyme is known to form dopaquinone via oxidation from L-3,4-dihydroxyphenylalanine (L-dopa) and then undergoes a series of reactions to form melanin (Sasaki et al., 2002). The suppression of melanin synthesis can be achieved by inhibiting tyrosinase activity.

In this study, quercimetrin compound was isolated from the *Centaurea paphlagonica* and its inhibition effect against the tyrosinase enzyme was studied *in silico*.

MATERIALS AND METHOD

Genaral

Molecular docking calculations were made using the Molegro Virtual Docker program. Interactions between quercimetrin and tyrosinase enzyme were visualized with the BIOVIA Discovery Studio program. The 3-D structure of the tyrosinase enzyme (PDB ID: 2Y9X)[4] was taken from the RCSB database (www.pdb.org). The 3D chemical structure of the isolated compound was taken as sdf format from the Chemdraw (ChemDraw 22.2) program. The active site of the tyrosinase enzyme was determined and the docking procedure was applied to analyze the interactions of the quercimetrin ligand in this active site. The coordinates of position of quercimetrin are X: -9.94907 Y: -28.2774 and Z:-44.4249 at 46.08 Å3 volume, 166.4 Å2 surface.

Pure compound

Quercimetrin (Quercetin 7-*O*-D-glucoside), a flavonoid glycoside, was isolated from *Centaurea paphlagonica* for the first time. The structure of the compound has been elucidated using NMR, HRMS-MS and a series of spectroscopic methods.

Enzyme activity tests

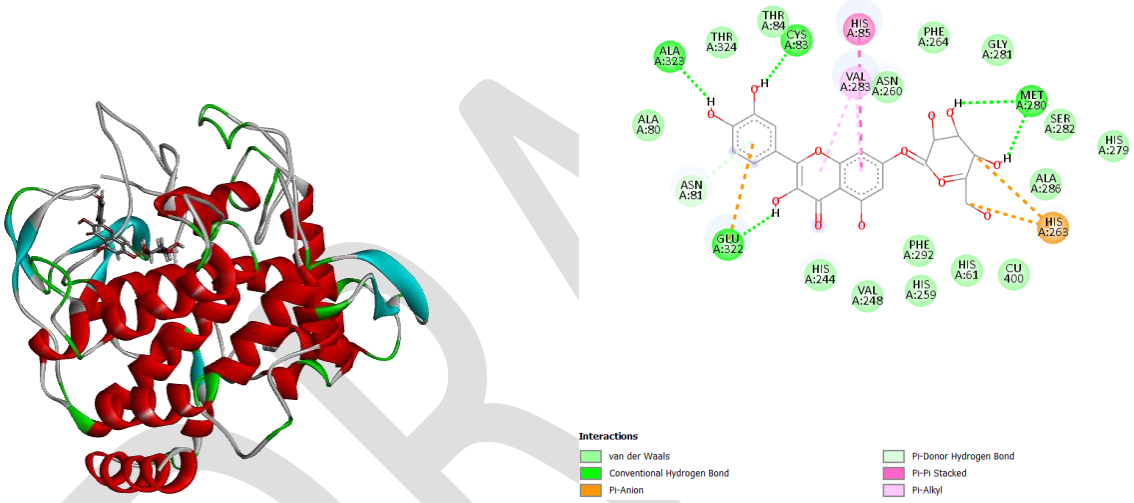
The tyrosinase inhibitory activity properties of quercimetrin compound were determined by making slight modifications in the dopachrome method used by Sarıkurkcu et al. (Sarıkurkcu et al. 2018, Tüfekçi et al., 2023).

Accordingly, 25 mL of each extractant was taken from the stock extract solutions prepared at a ratio of 1:1 in DMSO and 40 mL of tyrosinase solution and 100 mL of sodium phosphate buffer (pH 6.8) were added and mixed. The mixture was incubated at 25°C for 15 minutes. After adding 40 mL L-DOPA, the mixture was incubated again at 25°C for 10 min. Absorbance was measured at 492 nm. Scutellarin compound was used as

a positive control and the result was determined by comparing the inhibitory effect of this compound on tyrosinase enzyme with the effect of the compound. For this, 1 mg of scutellarin compound was weighed and dissolved in 1 mL DMSO, then diluted ten times with distilled water. Enzyme activity was tested at five different scutellarin concentrations.

RESULTS AND DISCUSSION

Molecular docking tests were conducted to identify the types of binding interactions and the amino acid residues relevant for the produced compound's biological activity. In this study, docking simulation was used to investigate the binding pocket of the tyrosinase protein receptors, and the outcomes were examined to identify the affinity scores and mode of contact (hydrophilic and hydrophobic contacts). 2D and 3D binding mode of quercimetrin in the binding pockets of the tyrosinase receptor is shown in Figure 1. Quercimetrin makes conventional hydrogen bonds with Glu 322, Ala 323, Cys 83 and Met 280 of tyrosinase. Also, it makes pi-donor hydrogen bond with Asn 81 of respective receptor. The residues Glu 322 and His 263 interact with quercimetrin by electrostatic interactions. In addition, quercimetrin makes pi-pi stacked interaction with His 85 and pi-alkyl interaction with Val 283 in the active site of the enzyme. The possible interaction types and energies obtained Molegro Virtual Docker (MVD) programme are given in Table 1. According to the results of docking calculation, the steric and hydrogen bond interactions played a major role in the binding of the said compound to the active site of the enzyme in protein–ligand interactions.



(b)

Figure 1. The docking conformation of quercimetrin-tyrosinase complex with lowest energy. (a) 3D view of the best selected conformation. (b) 2D view of the binding site interactions.

Table 1. MolDock score, electrostatic, steric interaction and the hydrogen bond energy of the docked quercimetrin.

Energy overview:Descriptors	Quercimetrin
Protein–ligand interactions	-167.814
Internal ligand interactions	33.617
MolDock Score	-131.986

CONCLUSION

Tyrosinase inhibitors uses as medicine in the prevention of Alzheimer, Parkinson and skin cancer diseases. The isolated compound is a biologically effective compound with therapeutic potential. The current in silico

study indicated that the quercimetrin may be a good agent in the development of selective tyrosinase inhibitor to prevent complications of this diseases in people with Alzheimer, Parkinson and skin cancer diseases.

DRAFT

ACKNOWLEDGEMENTS

We thanks to Çankırı Karatekin University for their physical and financial support in conducting this study.

REFERENCES

Demirtas I, Tufekci AR, Yaglioglu AS, Elmastas M 2017. Studies on the antioxidant and antiproliferative potentials of *Cirsium arvense* subsp. *vestitum*. Journal of Food Biochemistry, 41(1): e12299.

Ismaya WT, Rozeboom HJ, Weijn A, Mes JJ, Fusetti F, Wichers HJ, and Dijkstra BW 2011. Crystal structure of agaricus bisporus mushroom tyrosinase: ıdentity of the tetramer subunits and ınteraction with tropolone. Biological and Pharmaceutical Bulletin, 25:806–808.

Kilic O, and Bagci E 2016. Chemical composition of two endemic *Centaurea* L. taxa from Turkey, a chemotaxonomic approach. Journal of Essential Oil Bearing Plants, 19(1): 185-193.

Rao AR, Sindhuja HN, Dharmesh SM, Sankar KU, Sarada R, Ravishankar GA 2013. Effective inhibition of skin cancer, tyrosinase, and antioxidative properties by astaxanthin and astaxanthin esters from the green alga *Haematococcus pluvialis*. Journal of Agricultural and Food Chemistry, 61: 3842–3851.

Sarikurkcu C, Kirkan B, Ozer MS, Ceylan O, Atilgan N, Cengiz M, Tepe B 2018. Chemical characterization and biological activity of *Onosma gigantea* extracts. Industrial Crops Products, 115:323-329

Sasaki K and Yoshizaki F 2002. Nobiletin as a tyrosinase inhibitor from the peel of citrus fruit. Biological and Pharmaceutical Bulletin, 25:806–808.

Skin cancers. World Health Organization. https://www.who.int/uv/faq/skincancer/en/index1.html.

Taşar N, Doğan G, Kıran Y, Rahman MO, Çakılcıoğlu U 2018. Morphological, anatomical and cytological investigations on three taxa of *Centaurea* L.(Asteraceae) from Turkey. Bangladesh Journal of Plant Taxonomy, 25(2): 215-226.

Tüfekçi AR, Akşit H, Şimşek S, Karakoç ÖC, Adem Ş, Hameed ZA, Atalar MN, Topkara AR. 2023. Evaluation of insecticidal and enzyme activity potentials of essential oils and extracts of Chenopodium botrys against storage products pests. Bulletin of Biotechnology, 4(1), 7-12.

Yırtıcı Ü, Ergene A, Atalar MN, Adem Ş 2022. Phytochemical composition, antioxidant, enzyme inhibition, antimicrobial effects, and molecular docking studies of *Centaurea sivasica*. South African Journal of Botany, 144: 58-71.

DRAFT

DRAFT

DRAFT

DRAFT

DRAFT

2. POSTER PRESENTATIONS
2.1. ABSTRACTS

DRAFT

DRAFT

DRAFT

DRAFT

POSTER PRESENTATION

Covid-19 progression in north africa

Ilyes Zatla[1,*](ORCID: https://orcid.org/0000-0003-0476-8765), Lamia Boublenza [1], Amira Sebiane[1] & Ikram Salah[1]

1Laboratory of Microbiology applied to the Food industry, Biomedical and the Environment, Faculty of Natural and Life Sciences, Earth and Universe Sciences. Department of Biology. University of Tlemcen, Algeria.

*Author for correspondence, E-mail: ilyes.zatla@aol.com, Tel: +213-540-315-422, Address: Les Oliviers, Kiffane, Tlemcen.

Abstract

SARS-CoV-2 is the causative agent of the COVID-19 pandemic, causing a global health emergency, and affecting millions of people around the world. The objective of this work is to realize an epidemiological study about the evolution of COVID-19 in the five countries of North Africa (Morocco, Algeria, Tunisia, Libya, and Egypt) between January 2020 and May 2023. Methods applied were the observation and analysis of an international database using SPSS and Excel to compare the number of cases, number of deaths as well as the rate of vaccinations between the countries of this geographical area. Results obtained showed a total of 3,717,862 new cases and 83,757 deaths were recorded over the three years in these North African countries, where 2021 represented the most tragic year with the most COVID-19 cases and the most deaths (1767,410 and 50,341, respectively), and that the most affected countries were Morocco with 1,272,490 cases (34%) and Tunisia with 150,962 cases (31%). Similarly, the highest number of deaths was recorded in Tunisia with 29,331 deaths (35%), and in Egypt with 24,812 (30%). In addition, Morocco (47%) marked the highest rate of vaccinations out of a total of 384 851 69 injections. The COVID-19 pandemic has been considered a global health problem affecting each country in different ways depending on their people, control strategies, and vaccination campaigns.

Keywords: SARS-COV-2, COVID-19, North Africa, Cases, Deaths, Vaccination.

DRAFT

POSTER PRESENTATION

Impact of empirical antibiotic therapy on the management of native valve infective endocarditis with negative blood cultures : analysis of 45 cases

Harbi_Wafa[1*] , Manamani_Leila[2]

[*1]University Badji Mokhtar, Faculty of Medicine, ANNABA, ALGERIA.

*Corresponding author e-mail: harbi.wafa1@gmail.com

Abstract

Infectious endocarditis is a complex and heterogeneous pathology with significant morbidity and mortality. its management is essentially based on the eradication of microorganisms by bactericidal antibiotic therapy. We conducted a retrospective analysis of 45 patients hospitalized for infective endocarditis of native valve with negative blood culture from January 2021 to June 2023, the average age was 35 years with a male predominance. 60% of blood cultures were negative because of untimely prehospital antibiotic prescription.

Most of the patients had vegetation visible on transthoracic echocardiography, a minority had a mitro-aortic trigone abscess diagnosed on transesophageal echocardiography. All our patients were treated with a dual therapy combining vancomycin and gentamycin for two weeks followed by vancomycin monotherapy for 4 weeks, a favorable clinical and biological response was observed in 30 patients, no improvement with persistence of fever was observed in 10 patients, 5 patients underwent urgent cardiac surgery.

However, the main side effects observed with this dual therapy were renal failure (15) and allergic reactions such as urticaria (6) and Lyell syndrom (2)

This suggests that the use of a probabilistic antibiotic therapy combining vancomycin and gentamycin has been shown to be effective in the management of culture-negative infective endocarditis, due to its broad-spectrum, strong synergy and potentiation of bactericial activity, nevertheless, its tolerance and the side effects of each molecule still pose problems, including renal toxicity, often leading to premature discontinuation of gentamicin.

Keywords: endocarditis, antibiotic, empirical

DRAFT

POSTER PRESENTATION

Synthesis of iron copper phosphate catalyst for CC coupling reaction

Sarra Abbou[1*] , Amina Berrichi [1] , Drici Mohammed El Amine [1] ,Bachir Redouane [4]

[*1] University of Belhadj Bouchaib,Faculty of Science and Technology, Chemistry department, Ain Temouchent, Algeria.
[4] University of Tlemcen, Faculty of Science and Technology, Chemistry department, Tlemcen, Algeria.

*Corresponding author e-mail: sarra.abbou@univ-temouchent.edu.dz

Abstract

Almost all organic compounds that contain heterocyclic nitrogen are biologically active and have been used in the synthesis of several pharmaceutical molecules. Propargylamines are the most molecules which used to synthesis drugs molecules as rasagiline and selegiline for the Parkinson traitement. The C-C coupling reaction is a simple way to afford new structure of propargylamines in the presence of catalysts.

To ensure the synthesis of propargylamine with good yields, we used metal catalysts of binary copper iron phosphate catalyst to activate the reaction. The catalyst was characterized by several methods as XRD, MET and UV-Vis. Also, the catalyst presented a high activity and good stability for five runs.

Keywords: Catalyst, nanoparticle, syntheses .

DRAFT

POSTER PRESENTATION

Inclusion complexes study of a series of molecules with differents Cyclodextrins.

Sarra Abbou[1], Assia Keniche[2] , Berrichi Amina[1], Bachir Redouane[2]

[1]University of Belhadj Bouchaibof Ain Temouchent, Algeria.

[2]University Abou Bekr Belkaid of Tlemcen, Algeria.

[3]University of Belhadj Bouchaibof Ain Temouchent, Algeria.

Abstract

Cyclodextrins (CDs) are a group of cyclic oligosaccharides produced from starch or starch derivatives. They contain six (αCD), seven (βCD), eight (γCD), or more glucopyranose monomers linked via α-1,4-glycosidic bonds. CDs have a truncated cone shape with a hydrophilic outer wall and a less hydrophilic inner wall, the latter forming a more apolar internal cavity. Because of this special architecture, CDs are soluble in water and can simultaneously host lipophilic guest molecules. The major advantage of inclusion into CDs is increased aqueous solubility of such lipophilic substances. Accordingly, we present studies where the complexation of natural compounds. The different complex shows a higher antioxidant efficacy both in terms of capacity and rate of scavenging DPPH radical. The antioxidant activity of in free form has little difference with the differents guest in complexed form at the same concentration, this result indicates that the complexes formed maintained the Res antioxidant activity. This work shows the potential usage of Res/CD complexes. Also, we concluded that formulation of resveratrol and the other molecules with different cyclodextrin can improove the stability of resveratrol and incres the antioxidant activity.

Keywords: *Resveratrol*, Polyphenols, antioxidant activity, Cyclodextrin.

DRAFT

POSTER PRESENTATION

Valorization of urban wastewater treatment sludge in the construction field

Cheurfi Wassila[*1], Kebabi brahim[2]

[1,2]Constantine Mentouri University , Department of Chemistry, Faculty of Sciences, Pollution and Water Treatment Laboratory, Route Ain El Bey, Constantine 25000, Algeria

[*]Corresponding author: cheurfi_w@yahoo.fr

Abstract

The objective of this work is to identify alternatives that allow better management of urban wastewater treatment sludge. For this reason, a characterization of the annual sludge collected from the wastewater treatment plant in the city of Jijel Algeria is necessary. Physical, chemical and spectroscopic analyzes relating to the characterization of sludge. Based on a set of diverse and complementary techniques, such as XRF, DRX, FTIR, TG DSC. Thus the measurement of some physico-chemical parameters such as: density, porosity, pH, electrical conductivity, volatile matter, loss on ignition. This characterization aims to describe the composition of the sludge and ash of the sludge studied and distinguish the optimal conditions for obtaining residues with appropriate characteristics for better valorization.

The qualitative and quantitative analysis by different XRF, DRX, IR, TG-DSC techniques of the sludge collected during the years 2021-2022-2023 reveals that the main composition of the Jijel mud is stable and presents no danger during their reuse. The phases obtained by calcination of sludge (SiO_2, CaO, Al_2O_3, Fe_2O_3) are very essential in the field of manufacturing construction materials, which are the main minerals of clinker. Substituting calcined sludge in cement mortar does not present any risk to the resistance of the material but, on the contrary, allows it to be improved.

Keywords: sludge, wastewater, construction, clinker

DRAFT

POSTER PRESENTATION

Synthesis and assessement of antioxydant activity of spent coffee grounds chemically activated with potassium hydroxide

Naima Touafek[1,4*](*ORCID: https://orcid.org/0000-0002-6970-1169*),Nacera Baali[2] (*ORCID:https://orcid.org/0000-0001-9368-0241)*,Djanet Belkharchouche[3] (*ORCID:https://orcid.org/0009-0001-0912-2368*), Souad Belhour[4] (*ORCID: https://orcid.org/0000-0001-6408-0685*).

[1]Higher National School of Biotechnology "Toufik Khaznadar", Biotechnology Department. Constantine, Algeria.
[2]UFM-Constantine 1University, SNL Faculty, Animal Biology Department, Constantine, Algeria.
[3]Faculty of Process Engineering, Chemical Engineering Department,Constantine 3 University, Algeria.
[4]Mathematical and Subatomic Physics Laboratory, Exact Sciences Faculty, Physics Department, Constantine 1 University, Algeria

*Corresponding author e-mail: ntouafek@yahoo.fr

Abstract

The *Robusta* spent coffee grounds (SCG) were used as the basic material to produce activated carbon with potassium hydroxide (KOH). Initially, the SCG was washed with deionised water and dried at 70°Cfor 24h. After *pyrolysis* process at 700°C for 1h, the obtained SCG was mixed and stirred with KOH (1:1) at 80°Cfor 1h and subjected to calcination *under the same conditions*. The resulting material was mixed with H_2SO_4 (1N) and stirred at 90°C for 1h. After filtration and dryness, the chemical functional groups of SCG activated with KOH were checked using Fourier transform infrared spectroscopy analysis (FTIR) in the range of 4000-400 cm^{-1}. The spectrum exhibits a peak at 3650 cm^{-1} related to O-H stretching vibration. Also, a peak at 2987-2900 cm^{-1} indicated C-H stretching modes. The peaks found at about 1394 and 1066 cm^{-1} are assigned to the stretching vibration of the C-O bond. *Preliminary* results of *the antioxidant* assay *show* that the SCG had a weak DPPH radical scavenging ability *compared to standard.* The SCG *may serve* as *anew* basic material with *antioxidantpotentialand the enhancement of such effect needs the amelioration of the synthesis process.*

Keywords: SCG, KOH, Carbonization, FTIR, Antioxidant activity

DRAFT

POSTER PRESENTATION

Epidemiology and resistance to antibiotics of Enterobacteriaceae isolated from urinary tract infections in hospital

KHELILI Kaoutar* (*ORCID: https://orcid.org/0009-0001-5571-257X*)

*Frères Mentouri Constantine 1 University, Natural and Life Sciences Faculty, Applied Biology Department, Constantine City, Algeria Country.

*Corresponding author e-mail: kaoutar.khelili@gmail.com

Abstract

Enterobacteriaceae are the agents most frequently implicated in urinary tract infections. Their antibiotic resistance justifies epidemiological surveillance. The objective of our work is to carry out a statistical and epidemiological retrospective study over three years in order to calculate incidence and assess risk factors related to urinary *enterobacteriaceae* infections in hospitalized patients. Our results show that urinary infection is more common in women than in men. *Escherichia coli* dominates the epidemiological profile followed by *Klebsiella sp*. The study of the resistance profile of *enterobacteriaceae* has shown that most of them have a high resistance to penicillins, cephalosporins of first generation and trimethoprims. The epidemiological cohort investigation reveals that the female sex and the misuse of penicillins present a risk factor for urinary tract infection caused by *Escherichia coli*. The results of this study show a worrying increase in antibiotic resistance in uropathogenic *Enterobacteriaceae*. This requires rational prescription of antibiotics.

Keywords: urinary tract infections, *Enterobacteria*, antibiotic resistance, epidemiology, risk factors.

DRAFT

POSTER PRESENTATION

Case report of incidence correlation of aspergillosis and multiple antibiotic-resistant *Escherichia coli* in free-range poultry

Tana Kika[1*] (*https://orcid.org/0000-0002-4676-4108*), Jonida Boci[2] *(https://orcid.org/0009-0000-9480-6723),* Sonila Çoçoli[1] (*https://orcid.org/0009-0001-2822-1732*), Nikola Puvača[3] (*https://orcid.org/0000-0002-5500-7010),* Ilda Laci[4] (*https://orcid.org/0009-0007-5450-5177)* and Antonio Camarda[5] *(https://orcid.org/0000-0002-3961-585X),*

[*1]Agricultural University of Tirana, Faculty of Veterinary Medicine, Veterinary Public Health Department, Tirana, Albania.
[2]Food Safety and Veterinary Institute, Department of Animal Health, Tirana, Albania.
[3]University Business Academy, Faculty of Economics and Engineering Management, Department of Engineering Management in Biotechnology, Novi Sad, Serbia.
[4]Agricultural University of Tirana, Faculty of Veterinary Medicine, Preclinical Subjects Department, Tirana, Albania.
[5]University of Bari "Aldo Moro", Department of Veterinary Medicine, Bari, Italia

*Corresponding author e-mail: tana.shtylla@ubt.edu.al

Abstract

Forty one cases of sudden found dead free-range poultry were examined for the presence of *Aspergillus spp.* and avian *Escherichia coli*. The samples were collected from 10 different poultry flocks that had continuously been treated for respiratory clinical manifestations, low growth rates and poultry weakness. The post-mortem findings of the tested broiler chicks were mainly focused on air sacs, pericardium and liver. This case study was conducted in Elbasan district, during early spring months (March-April 2023).

Clinical and postmortem investigations; bacteriological and mycological examinations were carried out for each sample at the Veterinary Public Safety Laboratory, nearby the Faculty of Veterinary Medicine, Tirana. Simultaneous incidence of avian aspergillosis and collibacillosis resulted positive for 14 cases of the tested poultry (34, 14%), meanwhile 12 cases resulted positive only for *E.coli (*29, 26%). In the present study, the positive *E.coli* strains (n = 26) were evaluated against 6 different antimicrobial drug classes with the following resistance rates: Quinolones (46, 15%), Amphenicols (61, 53%), Tetracyclines (84, 61%), Penicillines-like (92, 3%), Sulfonamides (69, 23%), Aminoglicosides (73, 07%), by disk diffusion method. High levels of antibiotic-resistance were encountered in all of the 26 *E. coli* strains, where 14 *E. coli* strains (53, 84%) were identified as multi-resistant.

Previous national studies have been conducted for the presence of antibiotic-resistant avian *Escherichia coli* in poultry, but no data is yet reported on the correlation of collibacilosis infection incidence with respiratory mycosis. Continuous treatment of the poultry flock contributes to the development of antibiotic-resistant bacteria and in lack of poultry immunity towards other infectious agents. The preliminary results highlight that the incidence of multi antibiotic-resistant *E.coli* may be associated with other poultry respiratory diseases that should require further field investigations.

Keywords: *Escherichia coli*, *Aspergillus spp.*, poultry, Antibiotic-resistance, Albania.

POSTER PRESENTATION

Protective properties of some plant extracts against cellular damages induced by ionizing radiation

Saloua Mabsor Zgandaoui[1,4*] (ORCID: *https://orcid.org/0009-0006-4601-9059*), Nadia Errafiy[2] (ORCID: *https://orcid.org/0000-0003-1139-4466*), Laila Benbacer[3] (ORCID: *https://orcid.org/0000-0002-4200-6158*), Aboubaker El Hessni[4] (ORCID: *https://orcid.org/0000-0003-4890-7069*), Mohammed El Mzibri[3] (ORCID: *https://orcid.org/0000-0002-3148-1527*), Abdelghani Iddar[1] (ORCID: *https://orcid.org/0000-0001-6111-0607*), Adnane Moutaouakkil[1] (ORCID: *https://orcid.org/0000-0002-1762-881X*).

[1] National Center for Nuclear Energy, Science and Technology (CNESTEN), Biotechnology and Biomolecules Engineering Unit, Rabat, Morocco.
[2] Mohammed VI University of Health Sciences (UM6SS), Mohammed VI Center for Research and Innovation, Casablanca, Morocco.
[3] National Center for Nuclear Energy, Science and Technology (CNESTEN), Biomedical Research Unit, Rabat, Morocco.
[4] University Ibn Tofail, Faculty of Sciences, Department of Biology, Kenitra, Morocco.

[*] Corresponding author email: saloua.mabsor@uit.ac.ma

Abstract

The purpose of this study was to evaluate the protective effect of 4 plant ethanolic extracts with antioxidant properties: gum rockrose (*Cistus ladanifer*), false yellowhead (*Inula viscosa*), french lavender (*Lavandula stoechas*), and oleander (*Nerium oleander*). After evaluating the biological disturbances at low doses of ionizing radiation on *Tetrahymena pyriformis*, the protective effect of the 4 plant ethanolic extracts was evaluated by comparative studies of the effects on growth (generation time and number), on morphology and on physiology of the cell model by analyzing some metabolic enzymes and antioxidant biomarkers. The results obtained after exposure to ionizing radiation allowed us to highlight certain aspects of disturbances on the growth, morphology and physiology of the cell model, and to characterize the protective properties of the studied plant extracts. Some extracts are characterized by their important protective activities, which can be correlated to their antioxidant properties. These activities allowed the protozoan to escape the disruptive effects of ionizing radiation with a resumption of growth and a normal morphological form. The physiological parameters studied (GAPDH, SDH, Catalase, SOD, Peroxidases …) were also improved. These encouraging results shed light on the radioprotective effect of plant extracts, one of the most little-studied virtues.

Keywords: Protective properties, Plant extracts, Ionizing radiation, *Tetrahymena pyriformis*.

DRAFT

POSTER PRESENTATION

Development of rapid chemiluminescent enzyme immunoassay for determination of nonylphenol in drinking water

Anna N. Berlina* (*ORCID: https://orcid.org/0000-0002-3761-7472*), Maria Y. Ragozina, Nadezhda S. Komova (*https://orcid.org/0000-0002-3229-9073*), Kseniya V. Serebrennikova (*ORCID: https://orcid.org/0000-0001-9507-4085*), Anatoly V. Zherdev(*ORCID: https://orcid.org/0000-0003-3008-2839*), Boris B. Dzantiev (*ORCID: https://orcid.org/0000-0003-4008-4918*)

*[1]A.N. Bach Institute of Biochemistry, Research Center of Biotechnology of the Russian Academy of Sciences, Moscow 119071, Russia

*Corresponding author e-mail: anberlina@yandex.ru

Abstract

Nonylphenol is a more toxic decomposition product than the corresponding ethoxylate, which is used by humans in everyday life as part of various detergents. Having its effect on the reproductive, immune and nervous systems, this substance leads to disruption of the functioning of organ systems. Studies of toxic effects have led to the rationing of nonylphenol in drinking water. It is listed as priority pollutant in EPA's Contaminant Candidate List (US EPA, 2022). The main tools for determining nonylphenol are chromatographic methods. However, they cannot be used for screening purposes. In this regard, the development of relatively fast methods that allow the specific determination of the target analyte immediately in a series of water samples is an urgent task. The goal of this work was to develop a rapid chemiluminescent enzyme-linked immunoassay (ELISA) for nonylphenol. For this purpose, hapten-protein conjugates were synthesized and rabbit antibodies were obtained. The principle of the analysis is based on the competitive interaction between an immobilized analyte (hapten-protein conjugate) and a free one (nonylphenol) in the sample for the binding site of specific antibodies in the wells of a microplate. Goat anti-rabbit IgG antibodies labeled with horseradish peroxidase (HRP) were used as a developing immunoreagent. The HRP substrate of choice was luminol, combined with the enhancer p-iodophenol. By optimizing the composition and quantity of immunoreagents, the duration of the competitive stage, the detection time, it was possible to reduce the detection limit of nonylphenol to 7 ng/ml. The working range was 19-600 ng/ml. The achieved parameters are significantly better compared to the traditional approach with the use of colorimetric peroxidase substrate. Using colorimetric detection, the detection limit for nonylphenol was 55 ng/mL with the working range of 190-1300 ng/ml. The developed analysis was successfully applied to drinking water samples.

This work was supported by the Russian Science Foundation (grant no. 22-13-00293).

Keywords: nonylphenol, ELISA, chemiluminescense, screening, antibody, immunoassay

DRAFT

POSTER PRESENTATION

IgY antibodies targeting snake venom PLA_2 epitopes as potential Viper venom neutralizers

Vasiliki Moulasioti[1] (*ORCID: https://orcid.org/ 0000-0001-7075-1342)*, Evgenia Fotou[1] *(ORCID: https://orcid.org/ 0000-0003-0368-0526)*, Vassilios Moussis[1] (*ORCID: https://orcid.org/ 0000-0003-0658-8637)*, Dionyssios Sgouras[2] **(***ORCID: https://orcid.org/ 0000-0003-0975-2607)*, Vassilios Tsikaris[1] (*ORCID: https://orcid.org/ 0000-0002-2847-7340)*

[1]University of Ioannina, Faculty of Sciences, Department of Chemistry, Ioannina, Greece
[2]Hellenic Pasteur Institute, Laboratory of Medical Microbiology, Athens, Greece

*Corresponding author e-mail: moulasioti_v@hotmail.com

Abstract

Snakebites are a significant public health concern in many parts of the world, including Europe. Medically important cases of snakebite in the Old Continent are caused mainly by snakes belonging to the Viper family (*V. berus, V. aspis, and V. ammodytes*). It is well known that snake venom is a complex mixture, and not all the components contribute to its toxicity[1]. Snake venom phospholipases A_2 ($svPLA_2$), which are found in almost all venomous snake families, play a central role in snakebite toxicity and, particularly in Vipera species, possess the highest percentage among the venom components. Consequently, $svPLA_2$ may represent a promising candidate for achieving wide-spectrum antivenom activity[2]. Furthermore, currently available snake antivenoms produced in horses, immunized with whole venoms frequently cause adverse reactions in the patients administered[3]. On the contrary, IgY antibodies, which are analogous to mammalian IgGs, can be extracted from the egg yolk in large quantities with high yields, and do not activate the human complement system[4]. Thus, IgY technology has the potential to offer a sustainable, economical, and ethical alternative to traditional antivenom production methods. In this research, two peptide epitopes (10mers) from the C-terminal segment of PLA_2s of *V. berus* and *V. ammodytes* snake species were synthesized and conjugated via thioether bond to the CH_3CO-[K-Aib-C(1,3-CH_2CONH_2)]$_4$-NH_2 [CPSOC(3,9Acm)] peptide carrier. Hens were vaccinated with the immunogenic conjugates, and the specific IgY antibodies were isolated from the egg yolk. ELISA was employed to investigate the produced antibodies' potency to recognize both homologous and heterologous Viper venoms. Data shows that the produced antibodies have the ability to capture various Viper venoms from Greek territory.

Keywords: IgY antibodies, antivenom, CPSOC(3,9Acm), PLA_2, V. berus, V. Ammodytes.

Acknowledgements

The research work was supported by the Hellenic Foundation for Research and Innovation (HFRI) under the 3rd Call for HFRI PhD Fellowships (Fellowship Number:5428).
Also, we are grateful to Ilias Strachinis and Thomas Daftsios for their valuable contribution to this research by providing us with the appropriate viper venoms.

References

1. Z. Latinović et al., J. Proteomics, 146, 34–47, 2016.
2. H. Xiao et al., Biomed Res. Int., (2017).
3. G. León et al., Toxicon, 76, 63–76, (2013).
4. D. G. da Rocha et al., Eur. J. of Pharm. Sci., 106, 404-412, (2017)

DRAFT

POSTER PRESENTATION

Kinetic and thermodynamic study of paracetamol elimination by a food residu biosorbant

Didi Amel[1*] (*https://orcid.org/0000-0002-7019-319X)*, Bendallaa Ramila Chahinez [2], Amara Afaf [2].

[*1]University Abou Bakr Belkaid, Faculty of life and natural sciences, Department of biology, Tlemcen, Algeria.
[2] University Abou Bakr Belkaid, Faculty of sciences, Department of chemistry, Tlemcen, Algeria.

*Corresponding author e-mail: didiamel13@gmail.com

Abstract

Although pharmaceutical products play a major role in our lives and well-being, their contamination of our environment becomes a real public health problem. Among several known purification techniques, biosorption is one of the most economical and environmentally friendly method. This led us, among other things, to choose to use a residue from the food industry as biosorbent.

In our study, we are interested in eliminating by biosorption paracetamol from an aqueous solution by orange peel. For this, we studied the effect of some operating parameters on the adsorption efficiency (contact time, solution pH, temperature). Then we interested in studying some kinetic models and thermodynamic parameters to try to understand the mechanism that governed the biosorption process. The results showed that the kinetics were fast and well-adjusted with the pseudo-second order, and the maximum adsorption capacity was 9.94 mg/g. After the thermodynamic study, it was found that the process was favourable, spontaneous ($\Delta G0 < 0$) and endothermic ($\Delta H > 0$) and physico-chemical nature. The results obtained using this new green material as a biosorbent obtained from *Citrus sinensis* plant has proven to be an effective, economical and environmentally friendly alternative for the purification of water contaminated with paracetamol.

Keywords: biosorption, pharmaceutical wastes, orange peel, thermodynamics.

DRAFT

POSTER PRESENTATION

Validation of RP-HPLC method for simultaneous determination of Thiamin and Pyridoxin in pharmaceutical tablet

S. Batouche, M. Beldi

University Mohamed Cherif Messaadia, Faculty of Science and Technology, Department des Sciences de la Matière Souk Ahras , Algeria
Laboratoire des Sciences et Tehniques de l'eau et environnement

s.batouche@univ-soukahras.dz

Abstract

An isocratic RP-HPLC method has been validated for the dosage of Neurovit in pharmaceutical tablet form containing a binary mixture of Pyridoxin and Thiamin. The Chromatographic separation was performed on C 18 analytical column (4,6 mmx 250 mm, 5 µm particle size), using UV detection at 280 nm. With a mobile phase containing water with triethyilamine 2 % and sulfuric acid (pH = 2,5) at a flow rate of 1mL/min. The results of the study demonstrate that the method is specific, accurate, precise and reproducible, it could be applied for the quantitative analysis of thiamine and pyridoxin in Neurovit tablets.

Keywords: Neurovit-HPLC-Validation-statistics-Thiamin-Pyridoxin

DRAFT

POSTER PRESENTATION

Study, development and application of a new materials in A3 coupling synthesis

Zohra Hamiani [1,2] (*https://orcid.org/0000-0001-9014-0661*), Amina Berrichi [1,2] (*https://orcid.org/ 0000-0003-2586-6428)*, Redouane Bachir [1] (*https://orcid.org/0000-0001-6260-2179*)

[1] University of Tlemcen, BP 119, 13000, Laboratory of Catalysis and Synthesis in Organic Chemistry, Tlemcen, Algeria.
[2] University of Ain Temouchent, BP 284, 46000, Faculty of Sciences and technologies, Departement of Chemisty, Ain Temouchent, Algeria.

*Corresponding author e-mail: zohra.hamiani@univ-temouchent.edu.dz

Abstract

The three-component coupling of an amine, an alkyne and an aldehyde (A3) is a C-C coupling reaction that yields propargylamines, intermediates widely used in the preparation of biologically active and therapeutically useful products. These reactions involve the use of homogeneous and heterogeneous metal-based catalysts to activate the C-H bond and form a new C-C bond. In this work, we synthesize propargylamine in "one-pot" via A3 coupling reaction of an aldehyde, an alkyne and an amine in the presence of a heterogeneous iron-based catalyst in nanoparticles form. This material is prepared by hydrothermal method and characterized by different methods. The iron-catalysts present a good activity in the synthesis of propargylamine which good yield.

Keywords: A3 coupling, Heterogeneous catalyst, Hydrothermal method, Nanoparticles, Propoargylamine synthesis.

DRAFT

POSTER PRESENTATION

Root-Associated Endophytes in Five Co-Occurring Orchid Species: Isolation and Molecular Identification

Ines Harzli[1*] (*ORCID: https://orcid.org/0000- 0003-4009-2993*) and Yasemin Özdener Kömpe[1] (*ORCID:* https://orcid.org/0000-0003-1649-4298)

[*1]University of Ondokuz Mayis, Faculty of Science, Department of Biology, Samsun, Turkey.

*Corresponding author e-mail: inesharzli9@gmail.com

Abstract

The diversity of internal symbiotic fungi known as endophytes, which are associated with orchids, is vast. While previous research on orchid endophytic fungi primarily concentrated on endophytes linked to plant roots, there has been a growing inclination to investigate non-mycorrhizal endophytes from orchids due to their significant physiological functions and their potential as reservoirs of novel bioactive compounds. In this present study, we have successfully identified both morphologically and molecularly the fungal endophytes isolated from the roots of five terrestrial orchid species found in the same habitat: *Anacamptis papilionaceae, Orchis provincialis, Neotinea tridentata, Serapias orientalis,* and *Spiranthes spiralis* in the Samsun province (Turkey). To isolate and identify endophytic fungi associated with orchid species, root samples were collected during April 2022. The isolates underwent characterization through both morphological and molecular techniques. Molecular identification revealed that only 1 fungal strain was identified at family level, including Herpotrichiellaceae (isolated from *O.provincialis*). 2 fungal strains were identified at genus level, including *Trichoderma* (shared by *S.spiralis* and *O.provincialis*) and *Tomentella* (shared by *O.provincialis, A.papilionaceae,* and *S.orientalis*). 4 fungal strains were identified at species level, including *Rhizopus arrhizus* (shared by *S.spiralis,* and *N.tridentata*), *Ilyonectria robusta* (shared by *A.papilionaceae* and *N.tridentata*), *Fusarium oxysporum* (Shared by *S.spiralis, O.orovincialis,* and *N.tridentata*), and *Penicillium expansum* (*O.provincialis, S.orientalis,* and *A.papilionaceae*). In this research, we were able to identify culturable endophytic fungi of different orchid species found in the same habitat. These results may indicate that endophytic fungal partner(s) of orchids may vary and this could allow us to investigate their importance in the preservation and growth of orchids, as well as their possible use in uncovering bioactive compounds.

Keywords: Endophytes, Fungal diversity , Orchidaceae

DRAFT

POSTER PRESENTATION

Sustainable food alternatives

Özdegül Baştürk[1*] (*ORCID: https://orcid.org/0000-0003-2554-9588)*, Burak Demirhan[2] (*ORCID: https://orcid.org/0000-0001-8551-1472*), Buket Er Demirhan[2] (*ORCID: https://orcid.org/0000-0002-7938-6553*)

[*1] Gazi University, Institute of Health Sciences, Department of Food Analysis and Nutrition, Ankara, Turkey
[2] Gazi University, Faculty of Pharmacy, Department of Pharmaceutical Basic Sciences, Ankara, Turkey

*Corresponding author e-mail: ozdegul.basturk@gazi.edu.tr

Abstract

Food is one of the basic needs for sustaining life. As the population grows, hunger is a growing concern around the world. All foods, especially protein foods, are essential for global nutrition. However, rising food demands are increasingly damaging to the environment, as rising food needs increase the amount of land used and waste, resulting in greenhouse gases. For this reason, research has been started on what sustainable foods can be. The term sustainable food includes providing the food needs of today's people and subsequent generations with minor damage to the environment. This study aims to give examples of sustainable foods and compile studies related to sustainable foods with current data. Food alternatives produced or grown to ensure sustainable living are sustainable. Insects, algae, cultured meat, mycoprotein-Quorn, and more than 2,000 edible species are just a few of these sustainable food alternatives. Some species that pass as insect species used as food today are crickets and mealworms. According to some studies, algae may have antimicrobial, antiviral, and anticancer effects, but their total benefit and toxic properties to the human body are still issues that need to be investigated. Recently, algae are consumed both as food in Japan, Korea, and China and can also be used for other purposes. With cultured meat production, the skeleton of animals is not formed, so more edible parts and waste-free nutrients can be created. Also, cultured meats can eliminate the need for feed production. The main ingredient of Quorn Foods is mycoproteins, a type of fungus. Mycoproteins have been used in foods for over 50 years. Even on the mentioned alternatives, researches are still ongoing, and it is not known which of them is more sustainable. Additionally, studies are continuing how it can be more sustainable.

Key words: Sustainability, Food Alternatives, Sustainable Nutrition

DRAFT

POSTER PRESENTATION

BAP1 mutations in Glioblastoma

Ali Serçe[*] (0000-0002-8057-0411), Sabahattin Cömertpay (0000-0003-4850-6927)

[*1]Kahramanmaras Sutcu Imam University, Department of Bioengineering and Sciences, Kahramanmaras, Turkey.
[2]Kahramanmaras Sutcu Imam University, Faculty of Agricultural, Agricultural Biotechnology Department, Kahramanmaras, Turkey.

*Corresponding author e-mail: ali.serce.011998@gmail.com

Abstract

BAP1 (Brca-1 Associated Protein 1) gene is around 9 kilobases long and located on chromosome 3. The gene consists of 17 exons and codes the BAP1 protein(90kDa). BAP1 is a carboxy-terminal hydrolase(deubiquitinase) enzyme, and it can work both in the cytoplasm and nucleus. This tumor-suppressing protein is involved in important cellular pathways such as cell cycle, DNA repair, cellular differentiation, and cell death. Mutations of BAP1 were found in many cancers such as uveal malignant melanoma, malignant mesothelioma, renal cell carcinoma, and cutaneous melanoma. Most mutations were found in nuclear signal localization (NLS) regions of the gene, NLS-1 and NLS-2, which are responsible for the protein to translocate to nuclei. Glioblastoma also referred to as grade 4 glioma by World Health Organization is one of the most aggressive cancers in the central nervous system. Every year, around 240,000 cases of brain tumors diagnosed and most of them are glioblastoma. It is observed in middle. age people (around 64) but it could be also seen in any age, including children. Even though mutations in various tumors are investigated, very few studies included glioblastoma. In the present study, we examined 5 tumors and 4 blood samples of the glioblastoma patients and 2 non-patient blood sample. The participants went under surgery at Kahramanmaras Sutcu Imam University between 2022 June and January 2023. We sequenced and analyzed the NSL-1 and NSL-2 coding parts of the gene. Even though we have not found any mutation on NLS-1, we found point mutations on NLS-2 regions in two patients.

Keywords: Glial tumors, NLS, gene sequencing

DRAFT

POSTER PRESENTATION

Syntheses, Characterizations and Anticancer Studies of Ni(II) and Cd(II) Heterobimetallic Ferrocenyldithiophosphonato Complexes

Tuba ERKAN[1] (https://orcid.org/0009-0003-1288-1204) , Elif BULAT[2*] (https://orcid.org/0000-0002-2164-3641), Senem AKKOÇ[3,4] (https://orcid.org/0000-0002-1260-9425), Mustafa SAÇMACI[1] (https://orcid.org/0000-0001-8265-8233), Ertuğrul Gazi SAĞLAM[2] (https://orcid.org/0000-0002-7719-3934)

[1] Department of Chemistry, Yozgat Bozok University, Yozgat, Türkiye
[2] Department of Chemistry, Marmara University, _Istanbul, Türkiye
[3]Department of Basic Pharmaceutical Science, Süleyman Demirel University, Isparta, Türkiye
[4]Faculty of Engineering and Natural Sciences, Bahçesehir University, Istanbul, Türkiye

bulatelif55@gmail.com

Abstract

Ferrocenyl Lawesson Reagent (FcLR), an important member of perthiophosphonic acid anhydrides, has been researched in the last 30 years [1]. The reaction of FcLR with alcohols yields ferrocenyl dithiophosphonic acids (Fc-DTPOA). [2]. Fc-DTPOAs form coordination of four or six complexes with metal salts. [3].

In this study, ferrocenyl dithiophosphonic acids were obtained by the reaction of FcLR with aromatic or aliphatic alcohols. These acids that are known previously [2] were converted to ammonium ferrocenyl dithiophosphonates thereof. New heterobimetallic four-coordinated complexes of these salts with Ni(II) or Cd(II) ions were obtained. All of the compounds were characterized by some spectroscopic methods. The anticancer effects of the compounds were also studied in *vitro*.

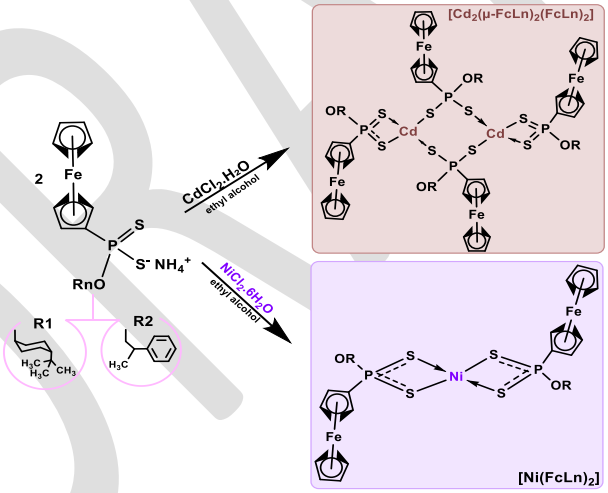


Figure 1. Synthesis reaction of ferrocenyl dithiophosphonato complexes.

Keywords: Ferrocenyl Lawesson Reagent, Fc-Dithiophosphonic acid, Fc-Dithiophosphonato complexes, Heterobimetallic complexes, Cytotoxic Activity.

Acknowledgments: Supported by Yozgat Bozok University Project Coordination Application and Research Center (BAP FYL-2022-1036).

References

[1] Sağlam E.G., Erden S., Tutsak Ö., Bayraktepe D. E., Durmuş Z.Y., Dal H., Ebinç A., 192 (2017) 322-329. https://doi.org/10.1080/10426507.2016.1238368
[2] Saglam E.G., Bulat E., Yıldırım A., 9 (2022) 183-191. https://doi.org/10.17350/HJSE19030000270
[3] Van Zyl, W. E.;Woollins, J. D. Coord. Chem. Rev., 257 (2013) 718-731. https://doi.org/10.1016/j.ccr.2012.10.010

DRAFT

POSTER PRESENTATION

Investigation of Sulfur-resistant Nickel-based catalysts in the dry reforming reaction of biogas: Effect of Yuttrium, Cerium, and Magnesium incorporation

Musab Esmail Khalil Doutoum (https://orcid.org/0009-0006-4857-559),
Hüseyin Arbağ (https://orcid.org/0000-0001-9301-7992)
Gazi University, Department of Chemical Engineering, 06570 Ankara, Turkey.
*Corresponding author e-mail: harbag@gazi.edu.tr

Abstract
Dry reforming of biogas is a crucial reaction that utilizes the biogas produced from biomass energy as a feedstock. The advantages of this reaction are mitigating the adverse effects of climate change by utilizing the two major greenhouse gases CH_4 and CO_2 as feedstock, production of hydrogen as a renewable energy carrier and sustainable alternative to fossil fuels, production of syngas with an H_2/CO ratio almost equal to 1 which is favorable for the production of some valuable chemicals via Fischer-Tropsch synthesis [1]. This reaction is highly endothermic and requires high temperatures to overcome the activation energy of the reaction. Therefore, a catalytic reaction system is required to increase conversion and selectivity, minimize waste, and reduce reaction times and energy demands. Nickel is frequently employed as active metal in catalysts due to it is cost-effectiveness and high activity in breaking C-C, C-H, and C-O bonds [2-5]. However, the Ni-based catalysts deactivate as a result of the coke deposition leading eventually to the block of the reactor, the reduction of the catalytic activity, selectivity, and product yield. The two side reactions methane cracking and/or Boudouard (CO disproportionation) reaction are thought to be the main contributors to the coke deposition due to the high C/H ratio of the feedstock in the CO_2 reforming of biogas [6]. As a result, Ni-based catalysts are promoted with other metals to overcome this limitation by the synergic effect between Ni and these metals. Depending on its source and the conditions of the anaerobic digestion process, biogas contains mainly CO_2 and CH_4, as well as trace amounts of H_2S, NH_3, and VOCs [7]. In the presence of H_2S even at ppm concentrations, besides carbon formation, another severe limitation is the loss of the activity of the catalyst due to sulfur poisoning. Thus, it is crucial to develop catalysts that can endure the harmful impacts of both coke deposition and sulfur poisoning. Despite the abundance of research efforts aimed at improving the stability of catalysts against coke deposition, the same cannot be said for research works focused on increasing the resistance of the catalysts against sulfur poisoning. In the present study, the influence of incorporating Ce, Y, and Mg elements on the activity and sulfur resistance of commercially available alumina-supported nickel catalysts during the dry reforming of the biogas reaction is investigated. Monometallic 8Ni@AL and bimetallic 8Ni-3X@AL catalysts (X= Ce, Y, and Mg) were prepared via wet impregnation route and the catalytic activity tests of the catalysts were carried out in a tubular fixed-bed reactor at 750°C and 1 atm with a total flow rate of 60 mL/min and an equivalent volumetric flow rates of CH_4, CO_2, and Ar under 50 ppm H_2S. The conversion rates of the X-doped catalysts were compared with that of the reference Ni@TAL catalyst. The Y and Ce-promoted catalysts exhibited enhanced activity and stability in the absence of H_2S. In terms of CH_4 conversion, the Y and Ce catalysts displayed higher initial conversion values compared to the unpromoted Ni@TAL catalyst. Notably, Ni-Y@TAL exhibited the highest initial conversion of 82%. Nevertheless, when H_2S was introduced at the 29th minute of the reaction, the conversion rates of the promoted Y and Ce catalysts experienced a sharp decline, ultimately leading to complete deactivation within just 15 minutes of Time on Stream (TOS). On the other hand, the Ni-Mg@TAL catalyst showed low initial CH_4 conversions compared to 8Ni@AL. The trends of the CO_2 conversion were similar to the CH_4 conversion. All catalysts exhibited CO_2 conversion rates higher than that of CH_4, with an H_2/CO ratio below 1. This can be attributed to the simultaneous occurrence of the reverse water-gas shift (RWGS) reaction as a side reaction. Despite the observed low stability of the Ni-Y@TAL and Ni-Ce@TAL catalysts in the presence of H_2S, these catalysts hold promise for future biogas valorization efforts.
Keywords: Dry reforming, Biogas, Syngas, Hydrogen sulfide, Sulfur poisoning, Promoter.

Aknowledgments: This work has been supported by the Scientific Research Projects Coordination Unit of Gazi University, Turkey, under grant number FYL-2022-8232.

References:
[1] Singh R, Dhir A, Mohapatra SK, Mahla SK. Dry reforming of methane using various catalysts in the process: review. Biomass Convers Biorefinery 2020; 10: 567-87.
[2] Wang HY, Ruckenstein E. Carbon dioxide reforming of methane to synthesis gas over supported rhodium catalysts: the effect of support. Appl Catal Gen 2000; 204: 143-52.
[3] Jeong H, Kim KI, Kim D, Song IK. Effect of promoters in the methane reforming with carbon dioxide to synthesis gas over Ni/HY catalysts. J Mol Catal Chem 2006; 246: 43-8.
[4] Arbag H, Yasyerli S, Yasyerli N, Dogu T, Dogu G. Coke minimization in dry reforming of methane by Ni based mesoporous alumina catalysts synthesized following different routes: effects of W and Mg. Top Catal 2013; 56: 1695-707.
[5] Akansu H, Arbag H, Tasdemir HM, Yasyerli S, Yasyerli N, Dogu G. Nickel-based alumina supported catalysts for dry reforming of biogas in the absence and the presence of H_2S: effect of manganese incorporation. Catal Today 2022; 397-399 : 37-49.
[6] Rostrup-Nielsen JR, Sehested J. Hydrogen and synthesis gas by steam- and CO_2 reforming. Adv Catal 2002; 47: 65–139.
[7] Iulianelli A, Manisco M, Bion N, Le Valant A, Epron F, Colpan C.O, Esposito E, Jansen J. C, Gensini M, Caravella A, Sustainable H_2 generation via steam reforming of biogas in membrane reactors: H_2S effects on membrane performance and catalytic activity, Int. J. Hydrog. Energy 2021; 46(57) : 29183–29197.

POSTER PRESENTATION

Components used as sugar substitute in cakes

Mustafa ÖZGÜR[1*] (https://orcid.org/0000-0002-7801-7932),

Aslı UÇAR[2] (https://orcid.org/0000-0001-9724-9571)

[1]Burdur Mehmet Akif Ersoy Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Burdur, Türkiye.

[2] Ankara Üniversitesi, Sağlık Bilimleri Fakültesi, Beslenme ve Diyetetik Bölümü, Ankara, Türkiye.

*Corresponding author e-mail: mozgur@mehmetakif.edu.tr

Abstract

It is thought that existence of supportive environments and communities that will direct people's preference to healthier foods are the main factors in prevention of obesity. According to Turkish Dietary Guidelines, it is reported that total amount of sugar consumed in daily diet should not exceed 10% of total energy, which is naturally present in foods and added during processing of foods. Therefore, products containing natural and artificial sweeteners have started to be used as sugar substitutes in food industry to limit total sugar. In our country and many parts of world, cakes are bakery products that contain added sugar, have high nutritive properties, and are produced in different mixtures and shapes, which are frequently consumed due to their hedonic effect. Cake production and consumption among bakery products ranks 3rd after bread and biscuits. Although cakes have high carbohydrate, protein and fat content, they are a poor product in terms of soluble and insoluble fiber, vitamins and minerals. Thus, there are studies to increase the nutritional aspect of cakes by adding dietary pulp and flour obtained from whole grain products in order to increase the quality parameters of cakes. Similarly, there are studies in which sugar and/or fat substitutes are used to reduce high sugar and fat content. Sugar substitutes are used to reduce sugar in cakes or to increase cake quality. Some studies, cakes were produced in which different molasses varieties, sugar grass, stevia extracts, honey dust, sugar alcohols, fruit purees, yacon (*Smallanthus sonchifolius*) syrup, and rare sugars (D-allulose) were used as sugar sources. Although the effects of these products, which are used as sugar substitutes, on physico-chemical, textural and sensory properties of cakes are different, they are generally preferred by consumers. For this reason, it is thought that use of sugar substitutes in food industry should increase.

Keywords: Cake, nutrititon, sugar substitutes

DRAFT

POSTER PRESENTATION

Multiple Sclerosis patients

[1]Emre Aktaş (https://orcid.org/:0000-0002-9422-3402) [2]Kübra Nur Şahin https://orcid.org/0009-0002-1806-9789), [3]Tuğba Sever (https://orcid.org/0009-0006-9766-5966) [4]Nehir Özdemir Özgentürk(https://orcid.org/0000-0003-3809-6303)

[1] Yıldız Technical University, Faculty of Arts and Science, Molecular Biology and Genetics, Istanbul, Turkey.
[2] Yıldız Technical University, Faculty of Arts and Science, Molecular Biology and Genetics, Istanbul, Turkey.
[3] Yıldız Technical University, Faculty of Arts and Science, Molecular Biology and Genetics, Istanbul, Turkey.
[4] Yıldız Technical University, Faculty of Arts and Science, Molecular Biology and Genetics, Istanbul, Turkey.

*Corresponding author e-mail: nehirozdemir@yahoo.com,

Abstract

Multiple sclerosis (MS) is the leading cause of neurological disability among adults and affects women three times more than men. Among the subtypes of MS, Remitting-Relapsing MS (RRMS) is the most common, characterized by disease states of remission and relapse, often transitioning to Secondary Progressive MS (SPMS). While the RRMS phase is marked by inflammatory processes, the SPMS phase is marked by neurodegeneration. Scientific evidence indicates that circulating microRNAs (miRNAs) play a significant role in various cellular and physiological functions. miRNAs circulate stably in human blood and are sensitive to processes occurring in the organism. miRNAs have been demonstrated to exert significant effects on MS pathology, with extensive investigations having been conducted and continuing inquiries pursued in this realm. Comprehensive studies on miRNAs exhibiting differing expression levels in male and female patients during the RRMS phase are lacking in the literature. In our study, we employed bioinformatics tools such as GEO2R, Cytoscape, and Morpheus to investigate the differences between remission and relapse disease states of RRMS in male and female subjects. MiRNAs displaying deviant expressions were analysed for both disease states and genders. The results of our investigation reveal that miRNAs with distinct expression levels during the remission and relapse phases of RRMS exhibit gender-specific variations. According to the results that will be obtained from this study, it is believed that specific miRNAs capable of distinguishing MS subtypes can be identified. Furthermore, gender-specific potential biomarkers can be discovered by assessing the shared and non-shared miRNAs between females and males. Additionally, there is potential for the identification of miRNAs that could be implicated in prospective treatment and drug development endeavours.

Keywords: Bioinformatics analysis, Multiple Sclerosis, Relapsing-Remitting Multiple Sclerosis, miRNA

DRAFT

POSTER PRESENTATION

Exploring Axolotl Regeneration: Bioinformatic Insights into the Wnt/ß-Catenin Pathway and Protein-Vitamin Interactions for Advancing Regenerative Medicine

Emre Aktaş[1] (https://orcid.org/0000-0002-9422-3402), Bengü Bena Koç[2] (https://orcid.org/0009-0007-1376-3727), Nehir Özdemir Özgentürk[3] (https://orcid.org/0000-0003-3809-6303)

Yıldız Technical University, Faculty of Arts and Sciences, Molecular Biology and Genetics, Istanbul, Turkey

*nehirozdemir@yahoo.com

Abstract

Regenerative medicine aims to address disorders caused by infections, inflammation, traumatic injury, and accidents by repairing tissue damage. The Mexican axolotl (Ambystoma mexicanum) serves as a main model organism for regenerative medicine. These species can regenerate all their organs and tissues in the case of injury or amputation without developing any sort of scar. In our study, we aim to unlock the mystery of this organism's extraordinary ability to divide by working at in-silico and molecular levels. We used preliminary bioassays to determine which of the proteins that constitute neural regeneration capabilities are more favorable. We identified that the Wnt/ß-catenin signaling pathway is one of the most prominent pathways regulating proliferation in Mexican axolotls and that the Wnt-1 is a major functional protein in this pathway. Bioinformatics tools such as Phyre2, HDOCK, and DisEMBL have been used to comprehensively investigate the components of the Wnt/ß-catenin signaling pathway synthesized from the apical epidermal cover and their relationship with neural regeneration. Analyses have shown that interactions of Wnt-1 proteins with various vitamins such as vitamin A, vitamin D, and vitamin E can affect this signaling pathway. In this respect, protein-vitamin binding complexes may be important not only in terms of docking scores but also in terms of their effects on signaling pathways. In conclusion, with the increasing relevance of bioinformatics studies, it is possible to understand the mechanism of regeneration more rapidly by developing studies at the molecular level. We believe that our study will contribute to regenerative medicine studies and may provide a different perspective on possible treatment methods.

Key Words: *Ambystoma mexicanum, regeneration, bioinformatics, molecular docking*

DRAFT

POSTER PRESENTATION

Yenilenebilir kaynaklı poliamid sentezi ve poliamid 6 ile karışımlarının karakterizasyonları

T. Sezgin (https://orcid.org/0000-0002-0279-6909), H. Esen (https://orcid.org/0000-0001-5649-6074)

Kocaeli Üniversitesi, Fen Bilimleri Enstitüsü, Polimer Bilimi ve Teknolojisi Programı, 41001, Kocaeli, Türkiye

Sorumlu yazar e-mail: huseyin.esen@kocaeli.edu.tr

Özet

Geleneksel plastiklerin çevreye verdiği zarar ve fosil yakıt tabanlı ürünlerde artan maliyetler sonucunda, yenilebilir kaynaklı polimerler, fosil yakıt bazlı polimerlerin yerine alternatif haline gelmektedir. Ayrıca petrolden elde edilen sentetik polimerler, plastik atık olarak doğaya bırakıldıklarında, toprakta uzun süre parçalanamadığından çevre kirliliğine ve toksik madde birikimine neden olmaktadırlar. Petrol kaynakları üzerindeki aşırı bağımlılığın, biyolojik kaynaklar ya da yenilenebilir kaynakları kullanarak geliştirilen biyoplastik üretimi ile kontrol altına alınabileceği öngörülmektedir. Biyobazlı polimerler ise yenilenebilir kaynaklardan üretilmektedir. Son yıllarda yapılan çalışmalar doğrultusunda, biyobazlı polimerlerin üretimi önem kazanmakta ve petrol bazlı polimerlerin yerini almalarına yönelik çalışmalar artmaktadır. [1-5] Bu çalışmada; ticari kullanımı olan PA6 içerisine kendi üretimimiz olan bio bazlı poliamid farklı oranlarda eklenmiştir. Bio bazlı poliamid izosiyanat ve dimer acid reaksiyonu ile elde edilmiştir. Elde edilen bio bazlı poliamid %5, 10 ve 15 oranlarında ticari PA6 içerisine katılarak ekstrüde edilmiştir. Yapılan analizler sonucunda eklenen bio bazı poliamidin termal özelliklerde herhangi olumsuz etki meydana getirmediği ve poliamid 6'nın darbe dayanımı arttırdığı gözlemlenmiştir.

Anahtar Kelimeler: Bio bazlı poliamid, dimer asit, PA6, mühendislik polimerleri, yenilebilir kaynaklar

Referanslar:

[1]: Kurt C. Frisch, JR., (2016), Foam products and methods of producing the same, United States, WO2016127016A1
[2]: P. Chaverot, S. Dreessen, L. Bouteiller, E. Bugnet, (2008), Thermoreversibly crosslinked elastic bituminous composition, France, US20110144242A1
[3]: D. Srinivas, A. Kumar, N. Devadutta, (2015), Process for Producing Amide Compounds, India, US2005090690A1
[4]: R. Nagelsdyk, S. Bühne, B. Jacobs, F. Calvert, D. Leutfeldt, D. Bernardo, (2018), Urea-group- and/or urethane-group-containig amides as and in rheology control agents, their preparation and their use, Japan, US2019390011A1
[5]: H. Zaigang, Z. Ke, Z. Xiaoyuchi, W. Chunqing, (2021), Phase-change microcapsule, preparation method and application thereof, heat-insulating coating, preparation method and application thereof, China, CN113214798A

DRAFT

POSTER PRESENTATION

Hasat Sonrası Yenilebilir Kaplama (Candelya) Uygulamasının 0900 Ziraat Kiraz Çeşidinin Muhafazasına Etkisi

Neslihan Ekinci[1*] (*ORCID: https://orcid.org/0000-0001-7022-5289.*), Serpil Varlı Yunusoğlu[2] (*ORCID: https://orcid.org/0000-0001-5699-0507.*), Neşe Yılmaz[3] (*ORCID: https://orcid.org/* **(**0000-0001-8720-2980)

[*1]Çanakkale Onsekiz Mart Üniversitesi, Ziraat Fakültesi, Bahçe Bitkileri Bölümü, Çanakkale, Türkiye
[2]Çanakkale Onsekiz Mart Üniversitesi, Lisansüstü Eğitim Enstitüsü, Bölüm, , Çanakkale, Türkiye
[3]Çanakkale Onsekiz Mart Üniversitesi, Lapseki Meslek Yüksekokulu, Bitkisel ve Hayvansal Üretim Bölümü, Çanakkale, Türkiye

*nekinci@comu.edu.tr

Özet

Bu çalışma ‘’0900 Ziraat’’ kiraz çeşidinde hasat sonrası yenilebilir kaplama (YK) uygulamasının depolama süresince meyve kalitesine olan etkisini belirlemek amacıyla yürütülmüştür. Hasat edilen meyveler, yenilebilir kaplama materyali uygulanmış meyveler ve hiç uygulama yapılmayan meyveler ile bunların ayrı ayrı modifiye atmosfer paketleme (MAP) ile kombinasyonu olmak üzere 4 gruba ayrılmıştır. Yenilebilir kaplama materyali meyvelere solüsyona daldırma şeklinde uygulanmıştır. Kontrol grubu meyveleri ise sadece saf suya daldırılmıştır. Daldırma işleminden sonra tüm meyveler, fazla suyun süzülmesi için 30 dk oda koşullarında bekletilmiştir. Tüm meyveler 1 gün süresince ön soğutma işlemine tabi tutulmuş ve sonrasında modifiye atmosfer uygulanacak gruba ait meyvelerde ambalajlama işlemi gerçekleştirilmiştir. Tüm uygulamalara ait meyveler 0±0,5 °C sıcaklıkta ve %90±5 oransal nem içeren normal atmosfer koşullarında 45 gün süresince soğuk hava deposunda muhafaza edilmiştir. Araştırmada muhafaza süresince 7 gün aralıklarla, meyve eti sertliği (N), ağırlık kaybı (% g), suda çözünür kuru madde miktarı (%SÇKM), meyve suyu pH’sı, titre edilebilir asitlik (% malik asit), meyve kabuk rengi (L*,C*, Hue^{o}), fenolik bileşen (GAE mg/100g), kiraz sapı toplam klorofil miktarı (mg 100g-1) gibi kalite özellikleri incelenmiştir. Çalışmada yenilebilir kaplama ve modifiye atmosfer paketleme kombineli uygulama, hasat sorası ‘’0900 Ziraat’’ kiraz çeşidinin muhafazasında, özellikle meyve eti sertliği, ağırlık kaybı, kiraz sapı klorofil miktarı, suda çözünür kuru madde miktarı ve meyve kabuk rengi üzerine olumlu sonuçlar vermiştir. Hasattan sonra başlangıç analizlerinde yapılan ölçümlerde 2.69 N olan meyve eti sertliği, 35 günlük muhafaza süresinin sonunda kontrol uygulamasında kontrol uygulamasında 0.97 N, YK+MAP uygulamasında 1.73 N değerinde tespit edilmiştir. Kiraz muhafazasında önemli bir kriter olan kiraz sapı klorofil miktarı hasattan sonra 46.39 mg 100g-1 değerinde, muhafaza süresinin sonunda ise kontrol uygulamasında 19.71 mg 100g-1, YK+MAP uygulamasında 24.61 mg 100g-1 değerinde belirlenmiştir.

Anahtar Kelimeler: *Prunus avium*, 0900 Ziraat, Yüzey Kaplama, Soğukta Muhafaza, Kalite

DRAFT

POSTER PRESENTATION

Immobilization of α-amylase on clear-epoxy-based photo-curable polymer for industrial use

Garen Karaburun[*1], Efe Baturhan Orman[1] (ORCID: https://orcid.org/0000-0002-8441-4061), Emrah Çakmakçı[1] (ORCID: https://orcid.org/0000-0002-2876-7460), Ayşe Ogan[1] (ORCID: https://orcid.org/0000-0002-8973-9762)

[*1]Marmara University, Science Faculty, Chemistry Department, Istanbul, Turkey.

*Corresponding author e-mail: garenkaraburun@gmail.com

Abstract

Enzyme immobilization is a process of physically or chemically attaching enzymes on a solid support or matrix via absorption, covalent binding, affinity immobilization, entrapment. These processes may modify the optimum working conditions of enzymes such as pH and temperature. Immobilized enzymes can be used several times unlike free enzymes in industrial processes. These properties make the immobilization of enzymes a favorable technique to the industry.

A-amylase is a highly specific hydrolytic enzyme that breaks down α-1,4-glycosidic linkage in starch molecules. As a cofactor α-amylase has a calcium ion (Ca^{2+}) which stabilizes the three-dimensional structure and works as an allosteric activator. They can be obtained from several sources, such as plants, animals and microorganisms. Amylases account for approximately 30% of the world's enzyme market. It is commonly used in various industries such as food, detergent, paper, textile, clinical and medical, production of fuel alcohol, treatment of waste water. Significantly in the food industry, amylases find numerous uses including; production of glucose, fructose and chocolate syrups, changing texture and volume when baking breads and cakes, production of fermented alcoholic beverages.

In our study, α-amylase was chemically immobilized to a photo-cured polymeric surface with the intent of creating a rapidly-producible material that is food grade. Photo-curing polymerization technique acquires a quick transformation of a liquid monomer into a solid film. Monomers of the studied polymer are biocompatible and have low toxicity levels. The CLEAR-epoxy-based photo-curable polymer was synthesized and characterized by FT-IR. Immobilization efficiency, effect of pH and temperature on activity, kinetics, reusability and storage stability were determined. According to our results, 27% of α-amylase was immobilized. Immobilized α-amylase showed better activity in acidic conditions than free enzymes, with maximum efficiency at pH 6.5. This indicates that α-amylase immobilized to CLEAR-epoxy-based photo-curable polymer can be utilized in industrial processes with acidic conditions.

Keywords: α-amylase, immobilization, photo-curable polymer, food industry

POSTER PRESENTATION

The bovine *MYOD1* p.Gly190Ser polymorphism (rs110708239) and its association with carcass traits in East Anatolian Red cattle and Holstein x Turkish native breeds

Nursen SENTURK[1] (0000-0003-1975-0103),
Sena ARDICLI[1*] (0000-0003-2758-5945)

[*1]Bursa Uludag University, Faculty of Veterinary Medicine, Department of Genetics, Bursa, Turkiye

*Corresponding author e-mail: sardicli@uludag.edu.tr

Abstract

The genes responsible for myogenic differentiation (MYOD) encode transcription factors specific to skeletal muscle development, and they are critical candidates for beef production. The collective understanding of the impacts of the *MYOD1* gene on carcass traits remains relatively restricted within native Turkish cattle breeds. Therefore, we evaluated the effects of p.Gly190Ser (c.568G>A) polymorphism in *MYOD1* gene on carcass traits in East Anatolian Red cattle and HolsteinxTurkish native breeds. The genotyping of the SNP within exon 1 of the gene was conducted through the PCR-RFLP technique. Population genetic parameters, encompassing adherence to Hardy–Weinberg equilibrium, heterozygosity, number of effective alleles, and polymorphism information content were computed. We employed information encompassing live weight, hot and chilled carcass weight, dressing percentage, chilling loss, back fat thickness, bone content, and valuable cuts yield. We conducted assessments for both the entire population and within each specific breed. The statistical analysis was performed utilizing the general linear model procedure employing the least squares methodology. The findings demonstrated a dominant prevalence of the BB genotype (~60%), resulting in a remarkably high frequency of the B allele (0.68). The $\chi 2$ test underscored a significant deviation from Hardy–Weinberg equilibrium for the respective locus ($P<0.001$). The ANOVA outcomes indicated that *MYOD1* rs110708239 marker does not serve as a robust marker within the examined breeds. We noted a significant association with dressing percentage exclusively within East Anatolian Red cattle ($P<0.05$). No significant alterations linked to genotype were observed in any of the other phenotypic measurements. This investigation centered on the variability exhibited by the bovine *MYOD1* p.Gly190Ser marker, as well as the potential relationships between the resultant genotypes and carcass characteristics. Although a possible association was identified with dressing percentage, the subsequent association analysis indicated that this marker did not exhibit significant genetic strength within the analyzed Turkish native breeds.

Keywords: East Anatolian Red breed, *MYOD1*, carcass characteristics, SNP

DRAFT

POSTER PRESENTATION

An Iminium Salt, The Berberine Molecule and Possible Aldehyde Form: A DFT Study

Fatma AYDIN[1] (https://orcid.org/0000-0002-7219-6407)*,
Aslı ÖZTÜRK KİRAZ[2] (https://orcid.org/0000-0001-9837-0779)

*[1]Çanakkale Onsekiz Mart Üniversitesi, Fen Fakültesi, Kimya Bölümü, Çanakkale, 17100, TÜRKİYE
[2]Pamukkale Üniversitesi, Fen Fakültesi, Fizik Bölümü, Denizli, 20070, TÜRKİYE

*faydin@comu.edu.tr

Abstract

Berberine (BBR) is named as 9,10-dimethoxy-5,6-dihydro-2H-[1,3]-dioxolo[4,5-g]-isoquinolino[3,2-a]-isoquinolin-7-ylium salt [1]. It is a natural molecule synthesized as cyclic quaternary ammonium salt and is known as a benzylisoquinoline alkaloid isolated from the plant Coptidis chinensis. The berberine molecule is found in the barks, leaves and twigs of the various plants such as barberry, Oregon grape and tree turmeric and widely used in traditional herbal medicine drug[2]. It is also as a natural dye and is used to dye wool, leather and wood due to its bright yellow color [3].

In this study, the optimized molecular geometries of the berberine and possible aldehyde form of it were calculated with DFT at the B3LYP/6-311++G(d,p) level. The results of theoretical calculations for two forms are compared in this study.

Key words: Berberine, Iminium salt, HOMO-LUMO, DFT calculations

References

[1] Nomenclature of Organic Chemistry: IUPAC Recommendations and Preferred Names, 2013.

[2] M.A. Neag, A. Mocan, J. Echeverría, R.M. Pop, C.I. Bocsan, G. Crişan, A.D. Buzoianu, Berberine: Botanical Occurrence, Traditional Uses, Extraction Methods, and Relevance in Cardiovascular, Metabolic, Hepatic, and Renal Disorders, Frontiers in Pharmacology. 9 (2018). https://doi.org/10.3389/fphar.2018.00557.

[3] ChemicalBook, in: CAS Database, 2086-83-1, n.d.

DRAFT

POSTER PRESENTATION

Osteosarkomada uzun kodlamayan RNA TUG1'in post-transkripsiyonel düzenlenmesinde rol oynayan miRNA'ların belirlenmesi

Beytullah Unat[*](*ORCID: https://orcid.org/0009-0006-1036-6710)*, Günhan Karakurum

[*1]Gaziantep Üniversitesi, Tıp Fakültesi, Ortopedi ve Tramvatoloji Bölümü, Gaziantep, Türkiye

*Sorumlu yazar e-mail: beytullahunat@hotmail.com

Özet

Uzun kodlamayan RNA'ların (lncRNA'lar) düzenlenmesinde meydana gelen bozukluklar, tümör oluşumunun önemli bir bileşenidir. Çeşitli tümörlerde lncRNA taurin yukarı regüle edilmiş gen 1'in (lncTUG1) anormal ekspresyonu rapor edilmiştir; ancak osteosarkomun (OS) ilerlemesinde kesin rolü ve temel hedefleri belirsizliğini korumaktadır. Bu çalışmanın amacı, bioinformatik analiz yöntemleri yardımıyla, osteosarkom patogenezinde muhtemel rolü olan ve TUG1 uzun kodlamayan RNA'sını hedefleyen miRNA'ların belirlenmesi amaçlanmıştır. TUG1 genin muhtemel miRNA düzenleyicilerinin belirlenmesi amacıyla mirtarbase (https://mirtarbase.cuhk.edu.cn/~miRTarBase/miRTarBase_2022/php/index.php, Erişim: 03.09.2023) çevrimiçi hedef tahmin aracı kullanılmıştır. Sonuç olarak yapılan analizler, miR-9-5p ve miR-1299 miRNA'ları tarafından düzenlenebileceğini göstermiştir. Önceki çalışmalar, miR-9-5p ve miR-1299 miRNA'larının osteosarkoma gelişimi ilerlemesinde önemli rolleri olduğunu göstermiştir. Bizde yaptığımız bu çalışmada, miR-9-5p ve miR-1299 miRNA'larının TUG1'in post-transkripsiyonel düzenlenmesi yolu ile osteosarkoma gelişimi ve ilerlemesinde önemli rollerinin olabileceğini gösterdik. Ancak bu sonuçları ileri analizler ve deneyler ile doğrulanması ve desteklenmesi gerekmektedir.

Anahtar Kelimeler: Osteosarkoma, TUG1, miRNA

DRAFT

POSTER PRESENTATION

Life cycle assessment (LCA) case study on cement-bonded particle board produced by using construction demolition waste wood

Ceren Serap Akın[1*] (*ORCID: https://orcid.org/0000-0001-5685-4556)*, İrfan Ar[2](*ORCID: https://orcid.org/0000-0002-6473-9205*), Sibel Hacıoğlu[3] (*ORCID: https://orcid.org/0009-0006-4731-2230*)

[1*] Gazi University, Graduate School of Natural and Applied Sciences, Ankara, Turkey
[2] Gazi University, Faculty of Engineering, Department of Chemical Engineering, Ankara, Turkey
[3] Tepe Betopan Yapı Malzemeleri San. ve Tic. A.Ş., Ankara, Turkey

*Corresponding author e-mail: cerenserap.akin@gazi.edu.tr

Abstract

Construction and Demolition Waste (CDW) is currently seen as one of the most important concerns that national authorities, particularly in Turkey, which is in the process of urban transformation and located in an earthquake zone. The search for alternative secondary raw materials for industries that use wood and its derivatives as raw materials is an important issue with the decrease on forest resources. CDW consists of bulky materials such as asphalt, bricks, wood and plastic. The main purpose of this study is use of waste wood those obtained by recycling the construction and demolition wood waste (CDWW) in production of cement-bonded particle board (CBPP). For this purpose, alkali treatment was carried out at four different concentrations as 2%, 3%, 5% and %8 NaOH alkaline solutions on the CDWW by using the dipping method. A new cement-bonded particle board design was made using these alkali treated CDWW. The mechanical and physical performance of the produced particle boards were carried out and a recipe that yield the particle board having the best results was determined and a life cycle assessment study was conducted. As the recipes were produced by helding the cement/wood (c/w) ratio stable at 1:2, the best results were obtained from the boards produced with 2% NaOH alkali treated CDWW. The modulus of rupture (MOR), modulus of elasticity (MOE) and density tests were applied to the produced boards in accordance with TS EN 310 and TS EN 323 standard, respectively. The vaules obtained as a results of MOR, MOE and density test are 10.37 N/mm^2, 6437.28 N/mm^2 and 1255.12 kg/m^3, respectively. The experimental outcomes showed acceptable mechanical and physical performance of the developed CBPP in compliance with the required standards. The feasibility of the study was evaluated by conducting LCA studies for the most effective recipe. The global warming potential (GWP) value of the recipe with the best result as a 677,11 kg CO2 equivalent was found. The results of this study can be considered as an effective roadmap for sustainability in all over the world and in appling secondy raw metarial CDW management.

Keywords: Construction and demolition waste, cement-bonded particle board, recycling, LCA.

DRAFT

POSTER PRESENTATION

Impact of incorporating waste glass in cement-based composite boards

Koray Kaysi[1] (ORCID: https://orcid.org/0009-0002-8252-4686), Birce Pekmezci Karaman[2] (ORCID: https://orcid.org/0000-0002-9051-2354), Nuray Oktar[2*] (ORCID: https://orcid.org/0000-0001-8980-7457)

[1]Gazi University, Graduate School of Natural and Applied Sciences, Ankara, Turkey.
[2]Gazi University, Faculty of Engineering, Department of Chemical Engineering, Ankara, Turkey.

*Corresponding author e-mail: oktarnuray@gmail.com

Abstract

In this study, the effect of using waste glass (WG) as aggregate on cement composite board properties was investigated. The large consumption of aggregates in the cement composite board production process and low quality requirements make the construction industry one of the most promising ways of solving the environmental burden of WG. One of the problems with glass is that the main use applications of it are disposable. WG is non-biodegradable and permanently fills landfills. In 2020, Europe's WG recycling rate was determined to be 79%. On the other hand, Turkey is quite behind in this regard with a recycling rate of 14%. Recycling WG will help conserve natural resources, minimize landfills and save energy. For this purpose, four different particle sizes (0-38, 38-63, 63-90 and 90-212 μm) of WG were obtained by grinding the WG in a ball mill and sieving it in the following process. In the cement composite board recipes, WG were substituted into the aggregate as 10%, 20% and 50% by weight, for all particle sizes. Modulus of rupture (MOR), water absorption (WA) and dry apparent density tests were carried out on the cement composite boards obtained in accordance with the TS EN 12467 standard. According to the test results, the best outcome was obtained from the recipe using 0-38 μm - 10% glass waste. MOR, density and WA values were found to be 6.26 N/mm^2, 1401 kg/m^3 and 15.51%, respectively. While MOR value increased by 2.3% compared to the control sample, density and WA values decreased by 1.72% and 9.57%, respectively. The use of WG in the construction industry can provide an environmentally friendly solution to these wastes, as well as providing a sustainable and alternative design opportunity for cement composite boards.

Keywords: Waste glass, cement composite, sustainability, recycling

DRAFT

POSTER PRESENTATION

Tip 2 diabetes mellitus hastalarında FTO ve PPARG genlerinin metilasyon profillerinin belirlenmesi

Menderes Yusuf TERZİ[1,2,*] (*https://orcid.org/0000-0001-8478-0451*), Meral URHAN-KÜÇÜK[1,2] (*https://orcid.org/0000-0003-1704-1370*), Müge ÖZSAN YILMAZ[3] (*https://orcid.org/0000-0001-8346-8941*), Zehra İLÇE KAYA[2] (*https://orcid.org/0009-0004-7070-4297*)

[1]Hatay Mustafa Kemal Üniversitesi, Tıp Fakültesi, Tıbbi Biyoloji Anablim Dalı, Hatay, Türkiye.
[2]Hatay Mustafa Kemal Üniversitesi, Sağlık Bilimleri Enstitütsü, Moleküler Biyokimya ve Genetik Anabilim Dalı, Hatay, Türkiye.
[3]Hatay Mustafa Kemal Üniversitesi, Tıp Fakültesi, İç Hastalıkları-Endokrinoloji Anabilim Dalı, Hatay, Türkiye.

*Sorumlu yazar e-mail: menderesyusufterzi@gmail.com / myterzi@mku.edu.tr

Özet

Tip 2 diyabet (T2DM), genetik duyarlılıklar ve çevresel etkileşimler ve çeşitli genler tarafından yönlendirilen kompleks, metabolik bir hastalıktır. Son zamanlarda sayısı artan çalışmalar diyabetes mellitus (DM) ile epigenetik, özellikle de DNA metilasyonu arasındaki ilişkiyi göstermektedir. Bu çalışmada, klinik olarak tip 2 diyabet (T2DM) tanısı konmuş hastaların periferik kan örneklerinde belirlediğimiz aday genlerden alpha-ketoglutarate dependent dioxygenase (FTO) ve peroxisome proliferator activated receptor gamma (PPARG)'ın metilasyon seviyesini ölçmeyi amaçladık. Çalışmamızda, Endkrinoloji Polikliniği'ne başvuran T2DM hastalardan (43) ve yaş ve cinsiyet uyumlu sağlıklı bireylerden (42) tam kanları alındı. Tam kandan izole edilen genomik DNA örnekleri bisülfit dönüşümünden sonra hedef genlerin metilasyon profili metil spesifik PCR ve jel elekroforez yöntemiyle analiz edildi. İstatistiksel analizler sonrası, FTO genindeki metilasyon durumu T2DM hasta ve kontrol grupları arasında farklılık göstermedi. Diyabetli hastalarda PPARG geninin metilasyon seviyesi kontrol grubuna göre anlamlı derecerede yüksek çıktı. PPARG'nin insülün duyarlılığını uyarıcı etkileri göz önüne alındığında, bulgularımız, metilasyon aracılı PPARG gen ekspresyonunun baskılanması diyabetli hastalarda insülin direncinin artmasına neden olabileceği ihtimalini güçlendirmektedir. PPARG genindeki metilasyonun T2DM hastalarındaki etkilerini ve hastalıkla ilişkisini daha iyi anlayabilmek için, daha büyük hasta popülasyonu ve kantitafi metodlar kullanarak ileri gen ekspresyon çalışmaları gerekli olacaktır.

Anahtar kelimeler: Tip 2 diyabetes mellitus, DNA metilasyonu, PPARG, FTO.

DRAFT

POSTER PRESENTATION

Silika esaslı ilaç taşıyıcı sistemlerin geliştirilmesi

Ecem Ünsal* *(ORCID: https://orcid.org/0000-0001-7534-3030),*
Müjgan Okur *(ORCID: https://orcid.org/0000-0002-1533-9408)*

Gazi Üniversitesi, Mühendislik Fakültesi, Kimya Mühendisliği Bölümü, 06570, Maltepe, Ankara, Türkiye

*Sorumlu yazar e-mail: ecemguder16@gmail.com

Özet

İlaç salım sistemleri kimyasal bir uyarıya cevap olarak ilacın serbest bırakıldığı sistemlerdir. Kontrollü ilaç salım sistemleri ise ilaç etken maddenin bölgesel veya sistematik olarak önceden belirlenmiş oranlarda ve spesifik zaman aralıklarında salım yapmasını sağlar. Kontrollü salım sistemlerinin temel hedefi, kan plazma konsantrasyonundaki ilaç etken madde miktarını sabitlemektir. Kontrolü ilaç salım sistemleri; pH, iyonik kuvvetler, enzim gibi fizyolojik koşulları oluşturan faktörlerden etkilenmektedir. Kontrollü ilaç salım sistemlerinin tedavi edici oranda ilaç düzeyinin sürekli korunması, hedeflenebilmesi nedeniyle zararlı etkilerin azaltılması, gerek duyulan ilaç miktarının azaltılabilmesi, önerilen ilaç rejimine hastanın uyumunu geliştirecek şekilde dozaj miktarının azaltılabilmesi, kısa yarılanma ömrüne sahip ilaçlar (örneğin proteinler ve peptid ilaçlar) için ilaç yönteminin kolaylaştırılması gibi avantajları bulunmaktadır. Kitosan, polietilen oksit, polikaprolakton, aljinat gibi polimerler, montmorillonit, haaloysite gibi killer, silika, selüloz gibi bazı malzemeler ilaç taşıyıcı destek malzeme olarak kullanılabilmektedir. Silika son zamanlarda kullanılan ve tıbbi uygulamalar için umut verici bir malzemelerden birisidir. Silika tetraetilortosilikat (TEOS) ve tetrametilortosilikat gibi alkoksisilanlar kullanılarak hazırlanabildiği gibi şeker kamışı küspesi, pirinç kabuğu, mısır koçanı, kahve kabuğu ve buğday kabuğu gibi biyokütlelerden de hazırlanabilmektedir. Doğal kaynaklardan elde edilen silika, düşük maliyetli olması, çevre dostu olması ve kolay elde edilebilirliği nedeniyle biyomedikal ve malzeme alanlarında kullanım alanı bulmaktadır. Bu çalışmada kullanılan silika, yaklaşık %90 oranında silika içeren çeltik artıkları külünden elde edilmiştir. Çeltik külü NaOH ile işleme tabi tutularak silika ekstraksiyonu gerçekleştirilmiş, daha sonra süzülerek sıvı kısım HCL ile nötralize edilmiştir. 1 gün yaşlandırma işlemi uygulanan silika süzülerek saf su ile iyice yıkanmış, çözücü değişimi basamağından sonra dondurarak kurutma yöntemiyle kurutulmuştur. Bu şekilde hazırlanan malzemeye emdirme yoluyla ilaç yüklenmiş ve bu şekilde hazırlanan ilaç yüklü malzemeden ilaç salım özellikleri incelenmiştir. Çalışmada kullanılmayı planlanan ilaç deksketoprofen trometamol olup, nonsteroid antiinflamatuar bir ilaçtır. İlaç salım deneyleri 37 ^{o}C sıcaklıkta, 100 rpm karıştırma hızında, pH 7.4 fosfat tamponu ortamında ve kesikli sistemde gerçekleştirilecektir. İlaç salım oranlarının belirlenebilmesi için deney ortamından belli aralıklarla örnekler alınmış, salınan ilaç miktarı UV spektroskopi cihazında 298 nm dalga boyunda absorbansları ölçülerek belirlenmiştir.

Anahtar Kelimeler: Çeltik, deksketoprofen, ilaç salımı, silika

Teşekkür: Bu çalışma Gazi Üniversitesi Bilimsel Araştırma Projeleri Koordinasyon Birimi (BAP) (FYL-2022-7760) tarafından maddi olarak desteklenmektedir.

DRAFT

POSTER PRESENTATION

Salçalarda Raf Ömrünün Arttırılması için Sinnamaldehit Enkapsülasyonu ve Antifungal Aktivitesi

Ayşegül AKI[1*] (ORCID: https://orcid.org/0000-0002-7017-2137), Şenol Alpat[2] (ORCID: https://orcid.org/0000-0001-5937-9949)

[*1]Dokuz Eylül Üniversitesi, Fen Bilimleri Enstitüsü, Biyoteknoloji, İzmir, Türkiye
[2]Dokuz Eylül Üniversitesi, Fen Bilimleri Enstitüsü, Biyoteknoloji, İzmir, Türkiye

*aysegul.aki@ogr.deu.edu.tr

Özet

Gıdaların mikroorganizmalar tarafından bozulması, henüz tam olarak çözülememiş dünya çapında bir sorundur. Gıdaların saklanması için, uygun bir çözüm hem ekonomik açıdan hem de insan sağlığı açısından faydalı olacaktır. Gıdayı güvenli hale getirmenin ve ona doğal veya 'çevre dostu' bir imaj vermenin yeni yollarına ilgi artmıştır. Gıdaların raf ömrünü uzatmak ve güvenli gıda sağlamak için gıda muhafazası, gıda endüstrisinin ve devlet kurumlarının en önemli endişelerinden biridir. Domates bazlı yemeklik ürünler için koruyucu olarak kullanılan sorbik asit-potasyum sorbat, benzoik asit-benzoatlar gibi koruyucuların yasaklanması planlanmaktadır. Son yıllarda, raf ömrünü uzatmak için gıdaların muhafazasında esansiyel yağların kullanımına ilişkin başarılı araştırmalar yapılmıştır.
Bu çalışmada, kimyasal koruyuculara alternatif olabilecek, %99 sinnamaldehit içeren tarçın yağının yeni nesil gıda koruyucususu olarak kullanımı hedeflenmektedir. İzole soy protein, kitosan ve gum arabik ile kompleks koaservasyon yöntemiyle enkapsüle edilmiştir. Enkapsüle edilen formülasyonların, enkapsülasyon verimi (EE), yükleme kapasitesi (LC) ve partikül boyutları karakterize edilmiştir. İzole soy protein / kitosan kompleks koaservasyonu %97,08 (EE) ve %45,30 (LC) ve izole soy protein / gum arabik kompleks koaservasyonu % 82,83 ve % 52.01 (LC) ölçülmüştür.

Bozulmuş salça örneklerinden izole edilen, *Aspergillus japonicus isolate D02* ve *Aspergillus flavus isolate 26* izolatlarına karşı antifungal etkinlikleri, spot kültür büyüme inhibisyonu (SPOTi) tahlili ve mikrotitre plakası (mikrodilüsyon) biyoanalizi iki yöntemle test edilmiştir. İzole soy protein / kitosan kompleks koaservasyonu MIC (minimal inhibisyon konsantrasyonu) ve MFC (minimal fungisidal konsantrasyonu) sırasıyla 187,5 ppm ve 62,5 ppm; izole soy protein / gum arabik kompleks koaservasyonu 125 ppm ve 250 ppm olarak tespit edilmiştir.

Anahtar Kelimeler: Enkapsülasyon, Kompleks koaservasyon, Antifungal aktivite

DRAFT

POSTER PRESENTATION

Cyclotetraphosphazene-based Hg(II) and Zn(II) coordination compounds

Afranur Pendar[1*] (https://orcid.org/0000-0003-1465-2811), Ceyda Duyar[2] (https://orcid.org/0000-0002-9012-2962), Yunus Zorlu[3] (https://orcid.org/0000-0003-2811-1872), Derya Davarcı[4] (https://orcid.org/my-orcid?orcid=0000-0002-0407-9604)

[*1,3,4]Gebze Technical University, Faculty of Science, Department of Chemistry, Kocaeli, Türkiye
[2]Afyon Kocatepe University, Faculty of Arts and Science, Department of Chemistry, Afyonkarahisar, Türkiye

*a.pendar2022@gtu.edu.tr

Abstract

Cyclophosphazenes are inorganic heterocyclic rings that contain alternating phosphorus and nitrogen atoms in the ring skeleton [1]. They show versatile coordination behaviour, particularly in the form of multidentate ligands carrying additional exocyclic donor functions [2]. Due to their easily functionalization, flexibility, and stability, cyclophosphazenes have significant interest to researchers [3]. Especially, trimer ($N_3P_3Cl_6$) and tetramer ($N_4P_4Cl_8$) have been the most striking structures [4]. Cyclophosphazenes are their versatility as ligands owing to ring flexibility and bonding groups, so they easily interact with transition metal ions [3,4].
Imidazole is a typical heterocyclic molecule which features a five-membered ring structure consisting of three carbon atoms and two nitrogen atoms. The nitrogen atoms are located at the first and third positions in the ring. Imidazole and its derivatives have been found to exhibit favourable ligand properties in the formation of metal complexes [5].
In this study, fully substituted cyclotetraphosphazene ligand with 2-isopropylimidazole reacted with $HgCl_2$ and $ZnCl_2$ salts, respectively. As a result, two new coordination compounds were obtained, and their structures were determined by single crystal X-ray diffraction method.

Keywords: Cyclotetraphosphazene, isopropyl imidazole, coordination compounds.

We would like to thank the project numbered TUBITAK-121Z236 for its financial support.

References

[1] Chandrasekhar, V., & Narayanan, R. S., *Chimia*, 67(1-2), 64-64, (2013).
[2] Richards, P. I., & Steiner, A., Inorganic chemistry, 43(9), 2810-2817, (2004).
[3] Jeevananthan, V., Thangavelu, S. A. G., Loganathan, P., & Shanmugan, S., ChemistrySelect, 6(7), 1478-1507, (2021).
[4] Chandrasekhar, V., Thilagar, P., & Pandian, B. M., Coordination Chemistry Reviews, 251(9-10), 1045-1074, (2007).
[5] Zhu, X. W., Luo, D., Zhou, X. P., & Li, D., Coordination Chemistry Reviews, 455, 214354, (2022).

DRAFT

POSTER PRESENTATION

Bioremediation of Toxic Textile Dyes with Mixed Microalgal and Cyanobacterial Culture

Seda ŞEN[1*] (*https://orcid.org/0000-0002-1187-2811)* Hatice Nur TOYCU[2] (https://orcid.org/0009-0009-5318-0539), Nur Koçberber KILIÇ[3] *(https://orcid.org/0000-0003-2668-3789)*

[*1] Faculty of Science, Department of Biology, Ankara University, Beşevler, Ankara, Turkey

*Corresponding author e-mail: nrkilic@ankara.edu.tr

Abstract

Various anthropological activities (agriculture, urbanization, industry, etc.) cause many harmful pollutants spreading into the environment. Among many toxic chemicals, dyes are the most dangerous organic pollutants. Biological treatment is based on the use of microorganisms with bioremediation capabilities in the treatment process. In this study, the mixed microalgal and cyanobacterial culture was obtained from hot spring of Denizli. The bioremediation capacity of mixed culture in media with Setazol Black B, Setazol Blue BRFX, and Setazol Navy Blue SBG textile dyes was determined. After selection of the pollutant, experiments were conducted in BG11 media at pH 8, with 8.7 mg/L Setazol Navy Blue SBG, and biomass concentrations [10% and 20% (v/v)]. The amount of dye remaining in the medium was determined spectrophotometrically. The highest bioremediation occurred in a medium containing 8.7 mg/L SNB dye with 10% (v/v) biomass, achieving 98.9% efficiency at the end of a 6-day incubation period.

Keywords: microalgae; cyanobacteria, textile dye; bioremediation

DRAFT

POSTER PRESENTATION

Scale-Up Approaches in Electrospinning Method

Mümine Tansu ÇELİK[1] (*ORCID: https://orcid.org/0000-0003-4194-0595*),
Serdar TORT[*1] (*ORCID: https://orcid.org/0000-0003-4945-5420*)

[1] Gazi University, Faculty of Pharmacy, Department of Pharmaceutical Technology, Ankara, Turkey.

*Corresponding author e-mail: serdartort@gazi.edu.tr

Abstract

Nanofibers are fine porous structures with average fiber diameters of one micron or less. As the fiber diameter decreases, the physical, biological and chemical properties of the fibers improve and properties such as large surface area, porosity and high permeability are obtained. In addition to tissue engineering, biomedical and regenerative medicine applications, in the field of pharmacy, nanofibers have found the opportunity to be used in many areas with the developing nanotechnology due to their features such as high surface/volume area, ease of application and low cost. Although there are many methods in nanofiber production, the most prominent method in recent years is the electrospinning method. It is based on the accumulation of the filaments coming out of the jets created by the electric field caused by the voltage applied to a polymer solution, on the collector plate. Although nanofibers produced by electrospinning technologies have gained a place in the market in many areas, there are many problems that need to be solved to ensure permanent stability in industrial production. In this study, the advantages and disadvantages of needle and needleless electrospinning methods in production will be examined and their suitability for scale-up will be investigated. Although the needle electrospinning method has advantages such as easy installation, low voltage requirement, relatively uniform fiber diameter, it has disadvantages such as clogging of the needles, inhomogeneous solution feeding rate and more irregular electric field. The needleless electrospinning method has advantages such as easy sustainability, no needle clogging problems, and higher efficiency. But, it has disadvantages such as the need for higher voltage and the difficulty of maintaining the viscosity and concentration of the solution. Although industrial devices are available for large-scale production of both methods, optimization of process parameters is very important for product optimization.

Keywords: Electrospinning, nanofiber, needle electrospinning, needleless electrospinning,

DRAFT

POSTER PRESENTATION

Zeytin yapraklarından ekstrakte edilen toplam fenolik bileşiklerin miktarına kurutma ve ekstraksiyon parametrelerinin etkisi

Melda Cebbar[1*] (*ORCID: 0009-0007-8696-3850)*, Özlem Doğan[2] (*ORCID: 0000-0002-1815-7239*)

[*1]Yıldız Teknik Üniversitesi, Kimya Metalurji Fakültesi, Kimya Mühendisliği Bölümü, Davutpaşa Kampüsü, 34220 İstanbul, Türkiye
[2]Yıldız Teknik Üniversitesi, Kimya Metalurji Fakültesi, Biyomühendislik Bölümü, Davutpaşa Kampüsü, 34220 İstanbul, Türkiye

*Sorumlu yazar e-mail: meldacebbar@gmail.com

Özet

Fenolik bileşikler açısından oldukça zengin olan zeytin yaprakları günümüzde pek değerlendirilmemektedir. Genellikle yakılmakta, toprağa gömülmekte ya da hayvan yemi olarak kullanılmaktadır. Yapraklar sürdürülebilirlik açısındanda, biyoaktif özellikleri nedeniyle katma değeri yüksek ürünlerin geliştirilmesinde kullanılabilecek ucuz, kolay erişilebilir bir hammaddedir. Bitkilerden ekstrakte edilen fenolik bileşiklerin miktarı kurutma, saklama ve ekstraksiyon yöntemlerine bağlıdır. Bu nedenle bu değerli bileşiklerin eldesinde optimum değerler birçok bağımsız değişkenin birbirleriyle olan ilişkilerinin de değerlendirilebileceği deneysel tasarım yöntemi ile belirlenmeli ve zamandan tasarruf sağlayarak endüstriye katkı sağlanmalıdır. Literatürde zeytin yapraklarına kurutma veya ekstraksiyon parametrelerinin toplam fenolik bileşiklerin miktarına etkisinin incelendiği çalışmalar olmasına rağmen, kurutma ve ekstraksiyon parametrelerinin birlikte değerlendirildiği ve kurutma kinetiğinin incelendiği çalışmalara pek rastlanmamaktadır. Bu nedenle zeytin yapraklarından ekstrakte edilen toplam fenolik bileşiklerin miktarına kurutma ve ekstraksiyon parametrelerinin etkileri, Box-Behnken deneysel tasarım yöntemi kullanılarak birlikte değerlendirilmiştir. Bu çalışmada, Balıkesir ili Edremit ilçesinden temin edilen zeytin yaprakları farklı çıkış güçlerinde mikrodalga fırında kurutulmuş ve yapılan matematiksel modelleme çalışmalarında kurutma kinetiği için Page modelin yüksek oranda uygunluk gösterdiği görülmüştür. Kurutma işlemleri sonrasında Box-Behnken deneysel tasarım yöntemi ile 3 faktörlü (mikrodalga çıkış gücü, etil alkol/su oranı, zaman), 3 seviyeli ve yanıt olan toplam fenolik bileşenlerin miktarını belirlemek için tasarım yapılmıştır. Merkez noktada 5 tekrarlı deney ve toplam 17 deney gerçekleştirilmiştir. Ekstraktların toplam fenolik bileşiklerin miktarı UV spektrofotometre ile belirlenmiştir. Deney sonuçlarını optimize edebilmek için ANOVA analizi gerçekleştirilmiş ve deney sonuçlarına kuadratik modelin uygunluğu görülmüştür. Analize göre deney koşullarının ekstrakte edilen toplam fenolik bileşenlerin miktarına etkisi; güç ve zaman lineer terimlerde anlamlı ($p<0,05$) iken etil alkol/su oranın tek başına önemli bir faktör olmadığı kuadratik ve interaksiyon terimlerinde etkili olduğu görülmektedir.

Anahtar Kelimeler: Zeytin yaprağı, fenol, kurutma kinetiği, ekstraksiyon, Box-Behnken

Teşekkür: Bu çalışma Yıldız Teknik Üniversitesi Bilimsel Araştırma Proje Koordinatörü (proje numarası FYL-2023-5679) tarafından desteklenmektedir.

DRAFT

DRAFT

POSTER PRESENTATION

Taze gül (Rosa damascena) ve posasının toplam fenolik bileşiklerinin miktarına hasat döneminin etkisi

Melda Cebbar[1*] (*ORCID: 0009-0007-8696-3850*), Özlem Doğan Aydeniz[2] (*ORCID: 0000-0002-1815-7239)*

[*1]Yıldız Teknik Üniversitesi, Kimya Metalurji Fakültesi, Kimya Mühendisliği Bölümü, Davutpaşa Kampüsü, 34220 İstanbul, Türkiye
[2]Yıldız Teknik Üniversitesi, Kimya Metalurji Fakültesi, Biyomühendislik Bölümü, Davutpaşa Kampüsü, 34220 İstanbul, Türkiye

*Sorumlu yazar e-mail: meldacebbar@gmail.com

Özet

Rosa damascena, Rosaceae familyasının Rosa cinsinde bulunan en önemli türlerinden birisidir. Güzel kokusu ile parfümeri sektöründe ve ayrıca yapısındaki fenol bileşiklerinin sayısının çokluğundan farmakolojik, kozmetik vb. alanlarda pozitif etkileri ile uzun yıllardır kullanılmaktadır. Dünyada öncelikle Türkiye, İran, Bulgaristan, İtalya, Rusya'nın güneyi, Hindistan, Çin ve Libya'da yetiştirilmektedir. Ülkemizde en fazla Isparta'da yetiştirilen Rosa damascena hasadı mayıs ortasından haziran ortasına kadar yaklaşık bir ay, senede bir kere gerçekleşmektedir. Türkiye'de yaklaşık 10000 ton Rosa damascena çiçeğinden gül yağı, gül suyu, gül absolütü ve gül konkreti üretilmektedir. Özellikle koku endüstrisinde yaygın olarak kullanılan gülün 4 tonundan 1 kilogram gül yağı elde edilmesinden dolayı oldukça değerlidir. Bu nedenle, gül yağının miktarını ve kalitesini arttırma, kokusunun hangi ekstraksiyon ve saklama koşullarında daha iyi olduğunu belirlemek adına yapılan çalışmalar literatürde yaygındır. Fakat bu gül ürünleri üretimi sonucunda açığa çıkan doğal fenol kaynakları olan posaların atık olmaktansa endüstriler açısından değerlendirilmesi için elde edilme sürecinin optimum koşullarını belirlemek literatür açısından eksik kalmıştır. Bu çalışmada, Ispartadan 2022 yılı hasat döneminin başında ve sonunda toplanmış taze gül ve bu güllerin hidrodistilasyon işlemiyle işlenerek açığa çıkan gül posasının yapısındaki toplam fenolik madde miktarına hasat döneminin etkisi incelenmiştir. En yüksek toplam fenolik madde miktarına ilk hasat taze gül ile ulaşılmıştır son hasatta ise toplam fenolik madde miktarında biraz düşüş görülmüştür. İşlenmiş gül distile posalarının ekstratlarındaki toplam fenolik madde miktarına bakıldığında, en yüksek değere yine ilk hasatta elde edilen posaların ektratlarında ulaşılmıştır. Deney sonuçları değerlendirildiğinde hasat döneminin az da olsa güldeki toplam fenolik madde miktarını etkilediği görülmüştür. İşlem görmüş gül posasının da hala yüksek oranda fenol içerdiği belirlenmiştir.

Anahtar Kelimeler: Rosa damascena, posa, fenol, ekstraksiyon

Teşekkür: Çalışmada kullanılan gülleri ve gül posalarını sağlayan Gülsha Kozmetik Sanayi ve Ticaret A.Ş'ye teşekkür ederiz.

DRAFT

POSTER PRESENTATION

Synthesis and Characterization of Tadpole-Like Single-Chain Nanoparticles as Polymer-Drug Conjugates

Hatice Kübra Batu[1](https://orcid.org/0000-0002-1287-7550), Binnur Aydogan Temel[1,2*](https://orcid.org/0000-0001-5252-6619)

[1]Bezmialem Vakif University, Institute of Health Sciences, Department of Biotechnology, Istanbul, Turkey

[2]Bezmialem Vakif University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, Istanbul, Turkey

*Corresponding author e-mail: baydogan@bezmialem.edu.tr

Abstract

Nanotechnology is focused on the development of synthetic techniques for producing nanoscale objects that enable precise size control and specific functionalization. For that purpose, single-chain polymer nanoparticles (SCNPs), created by the collapse or folding of polymer chains into structurally defined nanoparticles, are a dramaticly growing research topic in polymer science that has been developing over the last twenty-five years. SCNPs, which are ultra-small cross-linked nanoparticles, are obtained by individual folding or collapse of linear polymer chains through in-chain cross-linking under high dilution conditions. SCNPs allow the formation of polymer nanoparticles, which are much smaller than traditional polymer nanoparticles, 1-20 nm in size. Curcumin (Cur), a polyphenolic compound derived from *Curcuma longa* L., is known to have various therapeutic effects including anti-inflammatory, antiproliferative, antioxidant and antiangiogenic activities. Although many studies have showed the high anticancer potential of Cur, its bioavailability has been found to be low due to its poor absorption and rapid metabolism. These problems have been tried to be overcome with the use nanocarriers.

In this study, we aimed to design tadpole-like single-chain nanoparticles as polymer-drug conjugates. A poly(ethylene glycol) (PEG) based macroRAFT agent was synthesized and used for the reversible addition-fragmentation chain-transfer (RAFT) polymerization of methyl methacrylate (MMA) and methacrylic acid (MAA) monomers. MAA units of obtained amphiphilic copolymers were then reacted with Cur in a dilute solution to obtain tadpole-like single-chain nanoparticles. The polymers and SCNPs were characterized by FT-IR, GPC, DSC, UV-Vis, ^{1}H NMR and DLS analyses.

Keywords: Amphiphilic copolymers, reversible addition-fragmentation chain-transfer (RAFT) polymerization, single-chain polymeric nanoparticle (SCNP), curcumin.

Acknowledgements: This work was financially supported by Bezmialem Vakif University Scientific Research Projects Unit (Project No: 20210604). H.K.B. thanks to the Scientific and Technological Research Council of Turkey (TUBITAK) BIDEB 2211-C National PhD Scholarship Program in the Priority Fields in Science and Technology for financial support.

DRAFT

POSTER PRESENTATION

Fermentative melanin production with *Corynebacterium glutamicum*

Ceren Karcıoğlu (https://orcid.org/0009-0002-3681-8915), Eldin Kurpejovic (https://orcid.org/0000-0002-2743-1906), Dilek Kazan **(**https://orcid.org/0000-0002-0764-8876**),** Berna Sariyar Akbulut (https://orcid.org/0000-0002-4455-1192)

Marmara University, Engineering Faculty, Department of Bioengineering, Istanbul Kadıköy, 34722, Turkey.

*Corresponding author e-mail: berna.akbulut@marmara.edu.tr

Abstract

Melanin plays self-protective roles among bacteria, fungi, plants, animals, and humans. These roles include blocking of UV radiation, toxic iron chelation, and buffering against environmental conditions. Melanin can be synthesized from L-tyrosine through various enzymatic and non-enzymatic routes. A number of bacterial hosts, including *Streptomyces* and *Bacillus* species, have been used for the production of melanin, but the yield was not high enough for large-scale production. One route commonly employed by bacteria uses tyrosinase enzymes. This enzyme converts L-tyrosine to (3,4-dihidroksi-L-fenilalanin) L-DOPA. The same enzyme converts L-DOPA to L-dopaquinone, which is then non-enzymatically converted to L-dopachrome. Finally, L-dopachrome is polymerized to the black-brown pigment melanin. This work uses the Cu+2-dependent *Ralstonia solanacearum* tyrosinase enzyme for the efficient production of melanin in *Corynebacterium glutamicum* as the host. The industrial workhorse *C. glutamicum* has many advantages such as fast growth and genetic stability; it can reach high cell densities. To this end, *C. glutamicum* EKV-I has been used. This strain requires external L-tyrosine (1 g/L) addition which was converted to L-DOPA and finally polymerized to melanin. Here, melanin production was maximized by optimizing agitation speed, Cu^{+2} concentration, and production volume in a given flask. HPLC was used to confirm melanin synthesis. Then, melanin concentration was determined via measuring OD_{400} value. This value was converted to the dry cell weight using the correlation, 1 OD_{400} = 0.066 g/L. The optimum conditions for melanin production with EKV-I were in a 50 mL growth volume in a 500 ml flask agitated at 150 rpm for 48 hours. The concentration of Cu^{+2} was adjusted to 0.1 mM initially. The obtained yield was 0.75 g/L of melanin with 1 g/L of L-tyrosine. The authors acknowledge Tubitak BIDEB 2210D, Tubitak 120N728, and FYL-2023-10911 for support.

Keywords: Melanin, eumelanin, *C. glutamicum,* L-DOPA, L-tyrosine

DRAFT

POSTER PRESENTATION

Binary bioremediation of Toxic Textile Dyes and heavy metals with Mixed bacterial Cultures

A. İrem GÜNDÜZ[1] (https://orcid.org/0000-0003-6887-2660), Esra ERKOÇ[2*] (https://orcid.org/0000-0003-2457-2735), Nur Koçberber KILIÇ[3] (https://orcid.org/0000-0003-2668-3789)

1* Faculty of Science, Department of Biology, Ankara University, Beşevler, Ankara, Turkey

Corresponding author e-mail: nrkilic@ankara.edu.tr

Abstract

The industrial use of metals and organic dyes is widespread. Insufficient treatment of wastewaters can result in the release of dangerous materials, which can adversely affect the soil and aquatic life. Wastewaters including such pollutants have to be treated. In wastewater treatment, chemical and biological methods can be used. Chemical methods offer an alternative solution, they prove unfavourable due to their high cost and the secondary by-products they generate in large-scale treatment systems. Biological methods, on the other hand, are preferred in the first place because they require almost no chemicals and are easy to intervene in. In these methods, microorganisms capable of bioremediation are used. For this purpose, the mixed microbial culture was obtained from Çubuk stream (Ankara). Trials were done it media containing different heavy metals and Setazol Navy Blue SBG (SNB). Experiments were conducted with regards to pH levels, initial pollutant, and biomass concentrations. Mixed microbial culture had the maximum bioremediation capacity in media with Cr(VI)+SNB.

Keywords: bacteria, mixed culture, dye, heavy metal, bioremediation

DRAFT

POSTER PRESENTATION

Antimicrobial and Antifungal Borophene

Gamze Gürsu[1*] (*ORCID: https://orcid.org/0000-0003-3942-1198*), Aysu Şahin[2] (*ORCID: https://orcid.org/0009-0004-4397-0039)*, Nevin Taşaltın[3] *(ORCID: https://orcid.org/0000-0001-6788-1605)*

[*1]Maltepe University, Environment and Energy Technologies Research Center, Istanbul, Turkey.
[2]Maltepe University, Electrical and Electronics Engineering Faculty of Engineering and Natural Sciences, İstanbul, Turkey.
[3]Maltepe University, Environment and Energy Technologies Research Center, Istanbul, Turkey; Maltepe University, Department of Basic Sciences, Istanbul, Turkey, CONSENS Inc.; Maltepe University Research Center, Technopark Istanbul, Istanbul, Turkey.

*Corresponding author e-mail: gamzegursu@maltepe.edu.tr

Abstract

This paper addresses the critical problem of antibiotic resistance and the need for alternative antibacterial strategies. It focuses on the application of nanomaterials with different dimensionalities (0-D, 1-D, 2-D, and 3-D) in antibacterial contexts. The article explores the unique properties and mechanisms of low-dimensional nanomaterials like silver nanoparticles (AgNPs) and graphene quantum dots (GQDs) in fighting bacterial infections. These nanomaterials offer promise in wound dressings, medical implants, and food packaging, delivering clinical benefits while minimizing side effects.

This research explores the potential of β-rhombohedral crystalline structured borophene nanosheets as multifunctional antibacterial and antifungal agents. Prepared via physical exfoliation, these nanosheets exhibit significant inhibitory activity against pathogenic microorganisms, including *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Candida albicans*, and *Aspergillus brasiliensis*. The study suggests that borophene nanosheets, with their unique physicochemical properties, hold promise as effective antimicrobial coatings in biomedical and packaging applications, offering a novel solution to combat bacterial and fungal infections.

Keywords: antimicrobial, antifungal, borophene, microorganisms, nanosheets.

DRAFT

POSTER PRESENTATION

Akrilik / maleik kopolimer sentezi ve proses optimizasyonu

Gülşah Melikoğlu Konak[*1] *(https://orcid.org/0000-0001-8575-5557)*,
Nevzat Akyer[*2], Yasemin Ergin[*2] *(https://orcid.org/0009-0007-9567-0363)*

[*1]Yıldız Teknik Üniversitesi, Fen Edebiyat Fakültesi, Kimya Bölümü, İstanbul, Türkiye
[*2] Marlateks Teknoloji Tekstil Mak. ve Kimya San. TİC. A.Ş., İstanbul, Türkiye

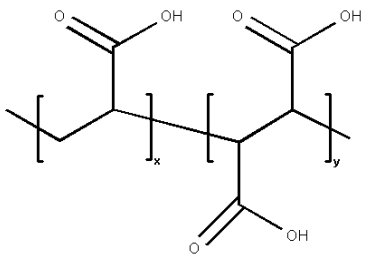
*gulsah.konak@marlateks.com

Özet

Suyun toprak, kaya gibi yer altı kaynaklarıyla temas etmesi sonucu Ca^{+2} ve Mg^{+2} iyonlarının çözünmesi su sertliğini oluşturur. Oluşan su sertliği ise endüstride kullanılmadan önce suyun işlem görmesini zorunlu hale getirmektedir. Her sanayi dalının kullandığı su miktarı ve sudan beklediği özellikler farklı olabilmektedir. Ancak su sertliği bütün endüstriyel uygulamaları etkileyen önemli bir parametredir. Su tüketim oranının yüksek olduğu tekstil sektöründe su içerisinde bulunan ve suya sertlik veren Ca^{+2} ve Mg^{+2} iyonları tutulmadıkları takdirde kumaş üzerinde lekelenmeye sebep olmaktadır. Bu lekelenme kumaşların renginde solma, dokusunda kalınlaşma gibi sorunlara yol açarak kumaşların estetik yapısını bozmaktadır. Bu sorunlar ancak kumaşların tekrar durulanmasıyla bertaraf edilebilir. Tekrar durulanma prosesi ise ekstra su sarfiyatına neden olmaktadır. Pek çok endüstriyel uygulamada kullanılan buhar kazanları, soğutma sistemleri gibi ekipmanlar sert suyun neden olduğu kireç birikintileriyle zarar görebilir. Ekipmanların bakım ve kullanım süresini uzatmak için su sertliği kontrol edilerek belirlenen seviyelerde tutulmalıdır. Çalışmamız kapsamında geliştirilen Akrilik / Maleik kopolimeri suya sertlik veren Ca^{+2} ve Mg^{+2} iyonlarıyla şelat oluşturarak bu iyonları ortamdan uzaklaştırabilmektedir. Su sertliği kontrol altında tutulduğunda veya önlendiğinde su kaynaklarının korunması, ekipmanların ömrünün uzatılması ve enerji tasarrufu konularında fayda sağlanacaktır. Geliştirmiş olduğumuz kopolimer aynı zamanda sahip olduğu dispersiyon kabiliyetiyle de yıkama sırasında tutunduğu yerden koparılan kirliliklerin dispersiyon halinde su ile uzaklaşmasını sağlamaktadır. Boya endüstrisinde ise pigmentlerin tanecik boyutlarının ve yapı sektöründe kalsit taneciklerinin küçültülmesinde katkı malzemesi olarak kullanılmaktadır.
Geliştirmiş olduğumuz ürünle benzer özelliklere sahip olan fosfat türevli ürünlerin çevreye çok ciddi boyutta zararları bulunmaktadır. Toplumun çevre konusunda duyarlılığının artması sonucunda geliştirmiş olduğumuz ürünün muadillerine göre çevreye daha az zarar veren çevre dostu bir yapıya sahip olması tercih sebebi olmaktadır ve muadillerine göre avantaj sağlamaktadır. Geliştirilen Akrilik / Maleik kopolimeri başta su sertliğinin önlenmesi ve azaltılması olmak üzere deterjan, tekstil, deri, yapı kimyasalları, su şartlandırma gibi birçok sektörde ve yaşamın farklı alanlarında yardımcı kimyasal olarak önemli bir yer edinmiştir.

Anahtar Kelimeler: Akrilik / Maleik kopolimeri, Ca^{+2} ve Mg^{+2} iyonları, çevre dostu, dispersiyon, su sertliği, şelat

Şekil 1: Akrilik/maleik kopolimer



DRAFT

POSTER PRESENTATION

Determination of some quality criteria in non-alcoholic beverages

Hanife Altınsoy[1*] (*ORCID: https://orcid.org/0000-0001-7038-0836*), İnci Barut[2] (*ORCID: https://orcid.org/0000-0002-0289-1447*), Burak Demirhan[2] (*ORCID: https://orcid.org/0000-0001-8551-1472*)

[*1]Gazi University, Institute of Health Sciences, Department of Food Analysis and Nutrition, Ankara, Turkey
[2] Gazi University, Faculty of Pharmacy, Department of Pharmaceutical Basic Sciences, Ankara, Turkey

*Corresponding author e-mail: dyt.hanifealtinsoy@gmail.com

Abstract

There are many factors that affect fruit juice quality. Considering the widespread consumption in our country and around the world, determining these quality criteria is of great importance in terms of food safety and human health. Hence, in this study, the aim was to determine quality criteria such as pH, Brix, electrical conductivity, total phenolic content, and the amount of furfural compounds in fruit juices. Total 100 samples, 4 different fruit juice samples (pineapple, apple, pomegranate, grape) and turnip juice sample from seven different brands (A, B, C, D, E, F, G) were analyzed. Furfural compounds in the fruit juices was analyzed using high-performance liquid chromatography with a diode array detector. In the analysis of 5-hydroxymethylfurfural (5-HMF) and furfural, linear results (5-HMF: R^2>0.999; furfural: R^2>0.999) were obtained over a wide concentration range. The average recoveries of 5-HMF and furfural were found as 109.1% and 98.5%, respectively. The limit of detection and the limit of quantification values for 5-HMF were determined as 0.0153 mg/L and 0.0463 mg/L, respectively. The limit of detection and the limit of quantification values were also determined as 0.0136 mg/L and 0.0413 mg/L for furfural. The mean 5-HMF values in pineapple, apple, pomegranate and grape juices were found as 0.054±0.034 mg/L, 0.052±0.056 mg/L, 0.661±0.451 mg/L, 0.092±0.047 mg/L, respectively. The amount of 5-HMF in turnip juice was determined as 0.003±0.003 mg/L. The mean furfural values in pineapple, apple, pomegranate and grape juices were found as 0.007±0.004 mg/L, 0,006±0,008 mg/L, 0.087±0.059 mg/L and 0.012±0.006 mg/L, respectively. Furfural could not be detected in turnip juice. The highest total phenolic content (TPC) were determined in pomegranate juice (399.867±149.795 mg/L), the lowest TPC values were determined in pineapple juice (49.307±16.536 mg/L). Other parameters such as pH, electrical conductivity, brix were generally determined in accordance with the standards.

Keywords: 5-HMF, Furfural, Total Phenolic Content, Non-Alcoholic Drink, HPLC

DRAFT

POSTER PRESENTATION

Utilization of agro-waste as a feedstock for L-Tyrosine production with *Corynebacterium glutamicum*

Emre Abdullahoğlu (https://orcid.org/0009-0006-6645-8737), Eldin Kurpejović (https://orcid.org/0000-0002-2743-1906), Nora Junker (https://orcid.org/0009-0004-6850-1167), Volker F. Wendisch (https://orcid.org/0000-0002-2743-1906), Berna Sariyar Akbulut (https://orcid.org/0000-0002-4455-1192)

Marmara University, Engineering Faculty, Department of Bioengineering, Istanbul Kadıköy, 34722, Turkey

Bielefeld University, Faculty of Biology and Center for Biotechnology (CeBiTec), Genetics of prokaryotes, Bielefeld 33615, Germany

*Corresponding author e-mail: berna.akbulut@marmara.edu.tr

Abstract

Lignocellulosic biomass is a potential source of sugars such as glucose and xylose which can be converted to value-added chemicals via microorganisms. Additionally, it also contain essential constituents for cell growth. In typical weather conditions, Turkey annually generates roughly 600,000 metric tons of hazelnuts, accounting for approximately 75% of the global production. This results in an equivalent amount of hazelnut husk waste which is often discarded and it is not harnessed for alternative purposes. In this work, the focus was to extract fermentable sugars and other essential components from hazelnut husk for L-tyrosine production in *Corynebacterium glutamicum*. *C. glutamicum* serves as the backbone of white biotechnology on an industrial scale, playing a pivotal role in the annual production of millions of tons of amino acids. L-tyrosine is among the three aromatic amino acids of protein biosynthesis. It is also the precursor of metabolites with defense, neurotransmitter, UV protectant, analgesic, and antioxidant functions that find applications in food, pharmaceutical, chemical and cosmetic industries. To release fermentable sugars from hazelnut husk, 50 g of dried hazelnut husk powder was suspended in 400 mL 3.8 % sulfuric acid and autoclaved for 50 minutes. Then, liquid fraction was collected by filtering and pH was adjusted to 7.0. The sugar content of the hydrolysate was 2±0.3 g/L of glucose and 8±0.6 g/L of xylose. Resulting hydrolysate was sterilized via autoclaving for growth and production. In order to determine the optimum growth conditions in 96-well plates, different amounts of hydrolysate were tested. Then, hydrolysate was tested for L-tyrosine production in 500-mL flasks for 72 hours. Results indicated that cells can efficiently grow and produce 277 mg/L L-tyrosine on pure hydrolysate without any supplementation. Although more biomass accumulated on higher hydrolysate amounts, this also prolonged the lag phase. Financial support by 120N728 and FYL-2023-10995 is gratefully acknowledged.

Keywords: L-tyrosine, *C. glutamicum,* Agro-waste, Bioproduction, Feedstock

DRAFT

POSTER PRESENTATION

Anti-microbial effects of lung microbiota-derived postbiotics against lung infections

Emine OMEROGLOU[1*] *(ORCID: 0000-0001-9345-7875)*, Fadime KIRAN[1] *(ORCID: 0000-0002-4536-2959)*

[1]Ankara University, Faculty of Science, Department of Biology, Ankara, Turkey.

*emineomeroglou@gmail.com

Abstract

Lung infections caused by various pathogens are defined as the damage in the lung. Although the severity of the damage may vary from mild to life threatening, burden of the disease is considered greater than other diseases which are well-recognized. Since pathogens causing lung infections, some of which are methicillin-resistant *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*, are becoming resistant to antibiotics worldwide, postbiotics as new and safe natural compounds that are beneficial to host, have gained attention as an alternative to antibiotics. The present study aimed to evaluate the anti-microbial and antioxidant activities of postbiotics derived from *Lactobacillus murinus* isolated from the lung microbiota of Spalax spp., which have unique lung microbiota due to their hypoxic and hypercapnic living conditions. For this purpose, lung tissue samples aseptically obtained from Spalax sp. were placed in MRS broth for pre-enrichment. Following pre-enrichment, bacteria were isolated using pour plate method. Postbiotics were extracted from the isolates by centrifugation and filtration, ant their anti-microbial activity was tested against MRSA, *P. aeruginosa* and *K. pneumoniae* by agar well diffusion method. Moreover, antioxidant activity of postbiotics were determined by investigating total phenolic and flavonoid content, as well as their effectiveness in eliminating DPPH radical. According to our results, postbiotics from *Lactobacillus murinus* displayed strong anti-microbial activity against all pathogens tested. Inhibition zones were measured as 13 ± 0.4 mm, 11 ± 0.5 mm, 13 ± 0.2 mm for *K. pneumonia*, MRSA and *P. auriginosa,* respectively. Total phenolic content was determined as 7 mg GAE/g while total flavonoid content was calculated as 6.52 mg/g QE. DPPH radical scavenging activity of postbiotics was found to be 100% efficient for all concentrations tested. These findings suggest that postbiotics derived from *L. murinis* isolated from lung microbiota of Spalax spp. may be used as an alternative to antibiotics.

Keywords: Lung microbiota, Postbiotics, Lung infections

DRAFT

POSTER PRESENTATION

Erken Gebelikte D vitamini Eksikliğinin Ovaryum Üzerindeki Etkileri

Deniz Küçükkaraca[1*] (ORCID: https://orcid.org/0009-0008-9506-9209), Çiğdem Elmas[1] (ORCID: https://orcid.org/0000-0002-8857-0918), Elif Seda Özdemir[2] (https://orcid.org/0000-0002-4628-9079) Deniz İrem Bulut[1], (ORCID: https://orcid.org/0000-0002-7820-0788), Gülnur Take Kaplanoğlu[1] (ORCID: https://orcid.org/0000-0002-3661-3488), Cemile Merve Seymen[1] (ORCID: https://orcid.org/0000-0002-8945-3801)

*[1]Gazi Üniversitesi, Tıp Fakültesi, Histoloji ve Embriyoloji ABD, Ankara, Türkiye

[2]Sivas Numune Hastanesi, Androloji laboratuvarı, Sivas, Türkiye

*deniz.zorba@gazi.edu.tr

Özet

D vitamini günümüzde gittikçe önem kazanan bir mikro besin olup, vücutta progesteron hormonuna benzer şekilde etki gösterdiği literatürde yer almaktadır. D vitamini eksikliğinin ovaryum dokusundaki etkileri üzerine çalışmalar daha çok poliskistik over sendromu ve preeklampsiyi kapsamaktadır. Gebeliğin erken döneminde ovaryum histolojisi üzerine yapılan çalışmalar oldukça sınırlıdır. Çalışmamızın amacı, D vitamini eksikliği modeli oluşturulmuş, gebeliğin erken dönemindeki dişi sıçan ovaryum dokularında oluşan hasarın, histomorfolojik düzeyde etkilerini inceleyerek ortaya koymaktır. Bu amaçla, 12 adet 4 haftalık, Sprague-Dawley albino cinsi dişi sıçan iki gruba ayrılmıştır. 6 haftalık deney boyunca kontrol grubu, D vitamini içeren standart yemle beslenen denekler, standart aydınlatma koşulları altında, UV-B yayan floresan aydınlatma altında, standart 12 saat aydınlık-12 saat karanlık döngüde tutulmuştur. D vitamini eksikliği modeli oluşturulan deney grubunda ise, denekler 6 hafta D vitamini içermeyen yemle beslenerek, vücutta D vitamini aktivasyonuna neden olan UV-B yaymayan ampüllerin kullanıldığı ortamda, standart 12 saat aydınlık-12 saat karanlık döngüde barındırılmıştır. Deney sonunda deneklerden alınan serum örneklerinde 25(OH)D düzeyleri HPLC yöntemiyle ölçülerek, 20 ng/mL ve altında olan deneklerde D vitamin eksikliği modelinin oluştuğu belirlenmiştir. Sonrasında kontrol ve D vitamini eksikliği gruplarındaki denekler çiftleşmenin tespit edilmesinden sonra gebeliğin ilk haftasında sakrifiye edilmiş, elde edilen ovaryum dokularının histolojik olarak incelenmesinde Hematoksilen-Eozin yöntemi kullanılmıştır. Histokimyasal incelemeler sonucunda kontrol gruplarında ovaryum dokusu ve ovaryum folliküllerinin normal histolojik yapılarında olduğu izlenmiştir. D vitamini eksikliği oluşturulan grupta, granüloza hücreleri ile teka yapılarının normal düzenlenimlerinin bozulduğu gözlenmiştir. Ayrıca gelişimlerinin farklı aşamalarında bulunan primordial, unilaminar ve multilaminar primer, sekonder, Graaf follikülleri, atretik foliküller ve korpus luteum yapıları ovaryum dokusundan alınan seri kesitlerde sayılmıştır. Sekonder ve Graaf folliküllerin D vitamini eksikliği grubunda kontrol grubuna kıyasla istatistiksel olarak anlamlı derecede az olduğu tespit edilmiştir. D vitamini eksikliğinin ovaryum dokusunda histokimyasal düzeyde folliküller hasara neden olduğu ve follikülogenezisin özellikle son basamaklarında matürasyonu bloke ederek olgun follikül gelişiminde azalmaya neden olduğu kanısına varılmıştır.

Anahtar Kelimeler: D vitamin eksikliği, Erken gebelik, Ovaryal folliküller, Histokimya

DRAFT

POSTER PRESENTATION

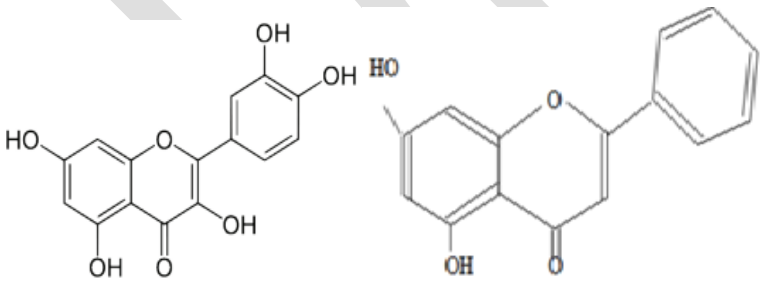
Kuarsetin ve krizinin paladyum(II) komplekslerinin sentezi ve karakterizasyonu, DPPH radikal süpürme aktivitelerinin incelenmesi

Murat Durmuş[1*] (*ORCID: https://orcid.org/0009-0009-9565-7470*),
Özlem Güngör[2] (*ORCID: https://orcid.org/0000-0003-2748-9179*)

[*1]Gazi Üniversite, Fen Fakültesi, Kimya, Ankara, Türkiye
[2]Gazi Üniversitesi, Fen Fakültesi, Kimya, Ankara, Türkiye
*e-mail:murat.md258@gmail.com

Özet

Flavonoidler,bitkilerde bulunan sarı, mavi ve kırmızı renkli pigmentlerin kaynağıdır. 2-fenil-benzo-a-piron yapısında bulunan meyvelerde, sebzelerde çay ve şarap gibi maddelerde bulunurlar. H ve OH gruplarının benzen ve heterosiklik yapılarına bağlanma şekilleri ile farklı isim alırlar. Antioksidan özellik göstermeleri flavonoidlerin en önemli biyolojik özelliklerinden biridir. Flavonoidler üzerinde yapılan araştırmalarda flavonoidlerin farklı türde kanser hücrelerine etki edip kanser oluşumunu azalttığı görülmüştür[1]. Kuersetin bitkilerde, sebze ve meyvelerde fazlaca bulunan flavonoid çeşididir. Turpgiller, üzüm, elma, domates ve yaban mersini gibi besin kaynaklarında farklı miktarlarda yer alırlar. Soğan yüksek miktarda kuersetin içeren bitkilerden biridir. Kuersetinin insanda papiller tiroid gibi kanser hücrelerinde apoptotik hücre ölümünü harekete geçirdiği söylenmiştir. Kuersetin ve kombonisyonlarıyla çalışmalar yapılmış olup antikanser madde şeklinde kullanılabilecek yapısının olduğu açıkça belirlenmiş, farklı olarak genotoksik ve toksik etki gösterebileceğini söyleyen araştırmalarda vardır[2]. Krizin (5,7-dihidroksiflavon) Passiflora gibi bitkilerden izole edilen flavonoid çeşididir. Doğal olan bu molekül antioksidan, anti-inflamatuar, anti-kanser, nöroprotektif ve anti-apoptotik gibi çeşitli etkiler gösteren farmakolojik özellik gösterir[3]. Flavonoidler endojen antioksidanların özelliklerini artırmanın yanı sıra doğrudan radikal süpürücü özellikleri oluşu ile reaktif oksijen türlerinin moleküler proteinlerdeki sülfidril bağları, çoklu doymamış yağ asitleri ve DNA'daki nükleotidler ile reaksiyona girerek doku hasarına neden olmalarını önlemektedirler. Çalışmalar neticesinde krizinin 5. Ve 7. Konumlarındaki hidroksil grupları ile serbest radikalleri süpürücü özelliğe sahip olduğu bildirilmiştir[4].Bu çalışmamızda ligant olarak kuersetin ve krizin (Şekil 1) kullanılarak, yeni paladyum(II) kompleksleri sentezlendi. Komplekslerin yapıları element analizi, FT-IR, NMR, kütle, UV-GB, TGA-DTA yöntemleri ve manyetik duyarlılık ile iletkenlik ölçümleri ile karakterize edildi. Kuarsetin ve krizin ile yeni paladyum(II) komplekslerinin antioksidan özellikleri 2,2-difenil-1-pikrilhidrazil (DPPH) radikal süpürme yöntemi ile incelendi. DPPH radikalinin stok çözeltisi DMSO ortamında 1 mM olarak hazırlandı ve 160 µM'a seyreltildi. Bileşiklerin stok çözeltisi DMSO ortamında 1mg/10 mL olarak hazırlandı.



Şekil 1. Kuersetin (a) ve krizinin (b) açık yapıları

Anahtar Kelimeler: flavonoid, kuarsetin, krizin, antioksidan, kompleks, radikal süpürme.

Kaynakça

[1] S. Karakaya, S. N. El, Beslenme ve Diyet Dergisi, 26(2) (1997) 54-60.
[2] A. S. Yalçın, A. M. Yılmaz, E. M. Altundağ, S. Koçtürk, Marmara Pharmaceutical Journal, 21 (2017) 19-29.
[3] J. F. Rodríguez-Landa, L. J. German-Ponciano, A. Puga-Olguín, O. J. Olmos-Vázquez, Molecules 27 (2022) 3551.

[4] Ö. Sedanur, Diklofenak İle İndüklenmiş Ht-29 Hücre Hattı Üzerinde Krisin'in Moleküler ve Biyokimyasal Etkileri, Yüksek Lisans Tezi, Bingöl Üniversitesi, 2020.

POSTER PRESENTATION

Electrochemical activity of mesoporous carbon supported and unsupported Fe-S and Ni-S catalysts for hydrogen evaluation reaction

Mukerrem Kavakli[1*] (*ORCID: https://orcid.org/0009-0000-3994-7355*), H. Mehmet Tasdemir[1] (*ORCID: https://orcid.org/0000-0001-9569-4161*), D.Dolunay Eslek Koyuncu[1] (*ORCID: https://orcid.org/0000-0001-8092-6740*), Alpay Sahin[1](*ORCID: https://orcid.org/0000-0002-1091-4979*)

[*1]Gazi University, Engineering Faculty, Chemical Engineering Department, Ankara, Turkey.

*Corresponding author e-mail: mukerremkavakl@gmail.com

Abstract

Hydrogen, which has a high gravimetric energy density, is a clean and sustainable energy carrier among alternative energy sources. Hydrogen can be produced cleanly by electrolysis of water. The electrolysis of water takes place through two half-reactions, the hydrogen evolution reaction (HER) at the cathode and the oxygen evolution reaction (OER) at the anode. Water is thus decomposed into oxygen and hydrogen. The hydrogen evolution reaction (HER) is an electrochemical process involving redox reactions at the electrode/electrolyte interface. One of the most important parameters for the industrial use of hydrogen production with HER is the development of active and economical electrocatalysis. In this study, the mesoporous carbon sample was prepared by the hard template method using the KIT-6 material prepared by the hydrothermal synthesis procedure. Ni-S and Fe-S active phases were synthesized by using the hydrothermal method. Wet impregnation was used for the preparation of the 10% by weight Ni-S@mezoC and Fe-S@mezoC catalysts. N_2 adsorption-desorption, XRD and SEM analyses were used to characterize the catalysts. Both the support material carbon and the catalysts exhibited Type-IV isotherm, which expresses mesoporosity. The surface area of the mesoporous carbon support was found as 1156 m^2/g. Due to the loading of metal sulfides on the support, the surface area of the Ni-S@mezoC and Fe-S@mezoC catalysts decreased due to clogging of the pores. Amorphous carbon phase was detected in the XRD patterns of the mesoporous carbon support. The Ni-S catalyst mainly exhibited the NiS_2 crystalline phase, while the crystal structure of the Fe-S catalyst was found to be FeS_2. Electrocatalytic activity of the mesoporous carbon and the catalysts were investigated in 0.5 M H_2SO_4 electrolyte medium by using linear sweep voltammetry (LSV) test. The overpotential of the mesoporous carbon support was 433 mV. The most active catalyst was Ni-S which showed the lowest overpotential value as 232 mV.

Keywords: HER, mesoporous carbon, KIT-6, metal sulfide, catalyst

DRAFT

POSTER PRESENTATION

Farklı çözücülerle ekstrakte edilen *Calliergonella cuspidata* (Hedw.) Loeske'nın biyokimyasal içeriğinin belirlenmesi

Dilay Turu[1*] (*ORCID: https://orcid.org/0000-0002-8485-0488*), Selime Deniz Bozkurt[2] (*ORCID: https://orcid.org/0009-0003-4782-8884*), Cenker Yaman[3] (*ORCID: https://orcid.org/0000-0002-3559-9098*), Gizem Gül[3] (*ORCID: https://orcid.org/0000-0003-3928-2917*), Atakan Benek[4] (*ORCID: https://orcid.org/0000-0001-6726-5968*), Kerem Canlı[3,5] (*ORCID: https://orcid.org/0000-0001-6061-6948*)

[*1]Dokuz Eylül Üniversitesi, Fen Bilimleri Enstitüsü, Biyoloji Bölümü, İzmir, Türkiye
[2]Ege Üniversitesi, Fen Fakültesi, Biyokimya Bölümü, İzmir, Türkiye
[3]Dokuz Eylül Üniversitesi, Fen Fakültesi, Biyoloji Bölümü, İzmir, Türkiye
[4]Kastamonu Üniversitesi, Fen Bilimleri Enstitüsü, Biyoloji Bölümü, İzmir, Türkiye
[5]Dokuz Eylül Üniversitesi, Fauna ve Flora Araştırma ve Uygulama Merkezi, İzmir, Türkiye

*Sorumlu yazar e-mail: dilayturu@gmail.com

Özet

Yaşamın birçok alanda kullanılan biyokimyasal bileşikler organizmaların hücrelerini ve diğer yapılarını oluşturmaktadır ve yaşam süreçlerini yürütmektedir. Proteinler, aminler, poliaminler, kompleks karbonhidratlar, organik asitler, lipitler, fenoller, flavonoidler, terpenoidler, aromatik bileşikler, hormonlar ve vitaminler gibi biyokimyasal bileşikler ve ikincil metabolitler, bitkilerinin fizyolojisi ve metabolizmasında çok önemli bir rol oynamaktadır. Hammadde olarak kullanılan doğal kaynaklı biyokimyasal bileşikler dünya üzerindeki birçok farklı bitki grubundan sağlanmıştır. Bu bitki gruplarından birisi de özellikleri bakımından birçok farklı alanda çok eski zamanlardan beri kullanılmakta olan karayosunlarıdır. Karayosunları çok uzun zamandan beri çeşitli hastalıkların tedavisinde kullanılmaktadır ve bu faydalarından dolayı son yıllarda yapılan çalışmalarda sıkça tercih edilmektedir. Bu çalışmada *Calliergonella cuspidata* (Hedw.) Loeske karayosunu türü etanol, metanol ve n-Hekzan olmak üzere üç farklı çözücüyle ekstrakte edilmiş ve Gaz kromatografisi ve kütle spektrometresi analizleri ile biyokimyasal içeriği belirlenmiştir. Elde edilen sonuçlar neticesinde, *C. cuspidata* karayosununun etanol ekstraktında 1-Allyl-4-(phenylthio)-6-(3-(trimethylsilyl)prop-2-ynyl)-5,6-dihydropyridin-2(1H)-one ve Phenol, 2,2'-methylenebis[6-(1,1-dimethylethyl)-4-(1-methylpropyl)-; metanol ekstraktında Phytol ve Neophytadiene; n-Hekzan ekstraktında ise 4-((E)-3-phenylallylideneamino)-5-(4-isopropylthiazol-2-yl)-4H-1,2,4-triazole-3-thiol bileşiklerinin varlığı tespit edilmiştir.

Anahtar Kelimeler: *Calliergonella cuspidata*, karayosunu, GC/MS, biyokimyasal içerik

DRAFT

POSTER PRESENTATION

Effects of ketamine treatment in an animal model of levodopa-induced dyskinesia: a pilot study

Nurdan Tekin[1*] (*ORCID: https://orcid.org/0000-0002-6310-7783*)
Tuğba Eryiğit Karamahmutoğlu[2] (*ORCID: https://orcid.org/0000-0002-0124-159X*)
Olca Kılınç[3] (*ORCID: https://orcid.org/0000-0001-9621-7543*)
Oya Orun[3] (*ORCID: https://orcid.org/0000-0003-1581-2207*)
Rezzan Gülhan[2] (*ORCID: https://orcid.org/0000-0002-1519-3170*)
[1]University of Health Sciences, Hamidiye School of Medicine, Departments of Medical Pharmacology, Istanbul, Turkiye
[2]Marmara University, School of Medicine, Departments of Medical Pharmacology, Istanbul, Turkiye
[3]Marmara University, School of Medicine, Departments of Biophysics, Istanbul, Turkiye

*Corresponding author e-mail: nrdntkn@gmail.com

Abstract

Levodopa-induced dyskinesia (LID) is a challenging motor complication associated with the long-term use of levodopa, which has remained the gold standard treatment for over 50 years in Parkinson's disease. Ketamine, an N-methyl-D-aspartate (NMDA) receptor antagonist, is a widely available drug developed as a general anesthetic. In recent years, evidence has emerged suggesting the potential utility of ketamine in various neuropsychiatric illnesses. This pilot study explores the effect of ketamine treatment on LID, aiming to provide insights into its therapeutic potential. Male Sprague-Dawley rats were unilaterally lesioned with 6-hydroxydopamine to induce the hemi-parkinsonism model and received levodopa + benserazide (12 mg/kg/day, 15 mg/kg) treatment for seven days. Dyskinesia was quantified using the Abnormal Involuntary Movements (AIMs) scores, and the rats developed stable dyskinetic movements subjected to ketamine treatment (5 x i.p. injections two hours apart). The AIMs scores were assessed before and after ketamine treatment. Our findings reveal a notable reduction in AIMs scores following ketamine treatment, underscoring the therapeutic potential of ketamine in managing LID. Notably, the Mann-Whitney U test demonstrated a statistically significant decrease in AIMs scores ($U=0$, $p=0.05$). This study provides preliminary evidence of the potential of ketamine to reduce AIMs scores in levodopa-induced dyskinesia. Although these findings are promising, further research in larger cohorts and in-depth exploration of the underlying mechanisms are needed. This pilot study lays the foundation for our further comprehensive research into the mechanisms of ketamine's effect on dyskinesia.

Keywords: Dyskinesia, Ketamine, AIM, Parkinson

DRAFT

POSTER PRESENTATION

Elektroaktif Hidrojellerin Sentezi, Karakterizasyonu ve Kontrollü İlaç Salım Davranışlarının İncelenmesi

Salih Muhcu[1*] (*ORCID: https://orcid.org/0009-0005-7132-6555)*, Melike Fırlak Demirkan[1] (*ORCID: https://orcid.org/0000-0003-1674-9086)*

[*1]Gebze Teknik Üniversitesi, Temel Bilimler Fakültesi, Kimya Bölümü, Kocaeli, Türkiye

*Sorumlu yazar e-mail: s.muhcu2021@gtu.edu.tr

Özet

Kontrollü salım teknolojisi, farmasötik ve tıbbi uygulamalarda büyük bir potansiyele sahiptir ve sağlık alanındaki etkisi nedeniyle hem akademi hem de endüstride yoğun bir araştırma faaliyeti konusudur. Teknoloji, tedavinin yüksek etkinliğini sağlar, tedavi maliyetini düşürür, toksisite risklerini ve ilacın yan etkilerini azaltır, ilaçların kanda ve plazmada kararlı emilim seviyeleri ve daha fazla hasta konforu sağlar. Ayrıca uyaranlara yanıt veren ilaç taşıyıcı sistemler, ilaçların belirli yerlerde ve zamanlarda kesin miktarlarda kontrollü olarak verilmesi potansiyeline sahip oldukları ve bu nedenle her kişinin ihtiyaçlarına göre uyarlanabildikleri için bu tür durumların tedavisi için daha fazla dikkat çekmiştir. Işık, pH, sıcaklık, elektrik, enzimler ve manyetik alanlar gibi halen araştırılmakta olan çeşitli dış uyaran türleri vardır. Ayrıca, bu tür cihazlar vücudun dışından tetiklenebilmektedirler. Her uyaran türünün kendine göre avantajları vardır, bu avantajların yanı sıra elektriksel uyarım kullanımının ON-OFF salım avantajları vardır, akımın büyüklüğü hassas bir şekilde kontrol edilebilir, bu şekilde salınan ilaç miktarı da kontrol edilir. İletken polimerler (CP'ler), yarı iletkenlere/metallere yakın elektriksel özellikler ve ek olarak elektrokimyasal ve optik özellikler kazandıran genişletilmiş π-bağlı bir polimer ailesidir. Bu benzersiz özellikler, onları çeşitli teknik endüstrilerde popüler hale getirdi.
Kontrollü salım teknolojisi birçok avantaj sağladığından, kontrollü ilaç salımını sağlayabilen biyouyumlu malzemelere yönelik bir pazar ihtiyacı vardır.
Bu çalışmada, yeni CP'ler hazırlanmış ve çeşitli tekniklerle (FTIR, TGA, CV) karakterize edilmiştir. Daha sonra, hazırlanan bu yeni CP'ler arasında en iyi iletkenlik ve mekanik özellik gösteren CP'nin elektrikle tetiklenen ilaç salım profili incelenmiştir.

Anahtar Kelimeler: Elektroaktif hidrojel, kontrollü ilaç salım sistemleri, elektriksel uyarımla ilaç salımı

DRAFT

POSTER PRESENTATION

Kumarin temelli kolorimetrik amonyak sensörü

Zeliha PERKTAS (0009-0000-9461-8941)

Gazi Üniversitesi, Fen Fakültesi, Kimya Bölümü, ANKARA, TÜRKİYE

zelihaperktas@gmail.com

Özet

Gıda israfı, dünya genelinde ciddi bir sorun olup sürdürülebilir gıda üretimi ve tüketimi için büyük bir engel teşkil etmektedir. Bu israf sadece gıda kıtlığına değil, aynı zamanda çevresel etkilere de yol açmaktadır. Özellikle sınırlı gıda kaynaklarına sahip bölgelerde gıda mevcudiyetini sınırlar ve küresel olarak su ve toprak kullanımını, gereksiz sera gazı salınımını ve verimsizliği içeren olumsuz çevresel etkilere neden olur. Özellikle artan kentsel nüfus, taze, sağlıklı, pratik ve hızlı gıda talebini artırmaktadır. Ancak bu talep, gıda kaybını ve israfını da artırmaktadır. Akıllı ambalaj, gıda kaybını ve israfını azaltmada önemli bir rol oynayabilir, çünkü gıda kalitesini koruyarak ve gıda güvenliğini sağlayarak etkili bir şekilde kullanılabilir. Gıda güvenliğini izlemek amacıyla çeşitli gıda sensörleri geliştirilmektedir. Akıllı ambalajlarda kullanılan bu sensörler, gıda kalitesi, menşei, kontaminasyon derecesi gibi bilgileri algılayabilir ve iletebilir. Bu sensörler aracılığıyla gıdadaki kimyasal, biyokimyasal, fiziksel ve mikrobiyolojik değişiklikler izlenebilir. Örneğin, tazelik sensörleri olan pH indikatörleri, gıdanın bozulma sürecini izlemek için kullanılabilir. Bozunma sonrasında ortaya çıkan biyojenik amin gazları, pH seviyesini değiştirir ve bu değişiklikler kolorimetrik veya florojenik indikatörlerle izlenebilir. Bu şekilde, tüketiciye gıdanın bozulma süreci hakkında bilgi verilebilir. Bu amaçla, gıda kaybını önlemek için kumarin temelli yeni bir tazelik sensörü geliştirmiştir. Bu bileşiğin fotofiziksel özellikleri çeşitli çözücülerde belirlenmiştir. Daha sonra bu bileşiğin gıda bozunmaları sonrasında ortaya çıkan amonyak ile etkileşimi çalışılmıştır. Hem çözücü ortamında hem de katı destek üzerinde sensörün etkisi inclenmiştir. Bu çalışmanın sonuçları, gıda kaybını ve israfını azaltmada akıllı ambalajların potansiyelini vurgulayabilir ve gelecekte daha sürdürülebilir bir gıda sistemi oluşturmak için önemli bir adım olabilir.

Anahtar Kelimeler: Kumarin, gıda israfı, akıllı ambalaj, biyojenik amin, sensör

DRAFT

POSTER PRESENTATION

Homalothecium philippeanum (Spruce) Schimp.'in farklı çözücülerle elde edilen ekstraktlarının biyokimyasal içeriğinin belirlenmesi

Selime Deniz Bozkurt[1*] (*ORCID: https://orcid.org/0009-0003-4782-8884*), Dilay Turu[2] (*ORCID: https://orcid.org/0000-0002-8485-0488*), Gizem Gül[3] *ORCID: https://orcid.org/0000-0003-3928-2917*), Cenker Yaman[3] *ORCID: https://orcid.org/0000-0002-3559-9098*), Atakan Benek[4] *ORCID: https://orcid.org/0000-0001-6726-5968*), Kerem Canlı[3,5] *ORCID: https://orcid.org/0000-0001-6061-6948*)

[*1]Ege Üniversitesi, Fen Fakültesi, Biyokimya Bölümü, İzmir, Türkiye
[2]Dokuz Eylül Üniversitesi, Fen Bilimleri Enstitüsü, Biyoloji Bölümü, İzmir, Türkiye
[3]Dokuz Eylül Üniversitesi, Fen Fakültesi, Biyoloji Bölümü, İzmir, Türkiye
[4]Kastamonu Üniversitesi, Fen Bilimleri Enstitüsü, Biyoloji Bölümü, Kastamonu, Türkiye
[5]Dokuz Eylül Üniversitesi, Fauna ve Flora Araştırma ve Uygulama Merkezi, İzmir, Türkiye

*Sorumlu yazar e-mail: ssdenizbozkurt@gmail.com

Özet

İnsanlık tarihinin başlangıcından bu günlere dek bitkiler, gıda, ilaç, inşaat, tekstil ve enerji gibi çeşitli sektörlerde kullanılmış ve kullanılmaya devam etmektedir. Özellikle ilaç sektöründe, bitkiler en az diğer sektörlerde oldukları kadar önemli olup ilaç arayışlarında ilk başvurulan kaynaklardan olmuşlardır. Bu kaynak bitki gruplarından birisi de karayosunlarıdır. Potansiyel olarak içerdikleri değerli bileşenler sayesinde ilaç araştırmalarında önemli roller üstlenmektedirler. Fakat karayosunlarının içerdikleri önemli bileşenlerin keşfi ve izolasyonu diğer bitkilere nazaran daha komplike olabilmektedir ve bu bileşenlerin düşük konsantrasyonlarda bulunabilme ihtimalleri kullanımlarını kısıtlamaktadır. Bu sebeple karayosunlarının potansiyel olarak içerdikleri bileşenlerin biyokimyasal analizlerinin yapılması önemlidir. *Homalothecium philippeanum*, Brachytheciaceae familyasına üye olan bir karayosunu olup ormanlar, sulak alanlar ve kayalıklar gibi çeşitli habitatlarda yaşayabilmeleri ile bilinmektedirler. Kuzey Yarımküre'nin ılıman bölgelerinde ve yüksek rakımındaki dağlık bölgelerinde yaygın olarak bulunmaktadırlar. Gerçekleştirilen bu çalışmada *H. phillippeanum*'un biyokimyasal içerik analizi için farklı çözücüler kullanılmıştır. Etanol, metanol ve n-hekzan çözücülerinde gerekli ekstraksiyon işlemleri yapıldıktan sonra gaz kromatografisi ve kütle spektrometresi cihazı ile *H. phillippeanum*'un içerik analizi yapılmıştır. Yapılan analiz sonucunda *H. phillippeanum*'un etanol ve metanol ekstraktlarında 1,2-Benzendikarboksilik asit, bis(2-metilpropil) ester, 2,2,4-Trimetil-1,3-pentandiol diizobütrat ve Propanoik asit, 2-metil-, 3-hidroksi-2,2,4-trimetilpentil ester bileşikleri; n-hekzan ekstraktında ise 2,6-Dibromo-3,5-difloro-4-piperidinilpiridin ve Oktasiloksan, 1,1,3,3,5,5,7,7,9,9, 11,11,13,13,15,15-hekzadekametil- bileşikleri başlıca bileşen olarak tanımlanmıştır.

Anahtar Kelimeler: Karayosunu, *Homalothecium philippeanum*, Biyokimyasal içerik, GC-MS

DRAFT

POSTER PRESENTATION

Cutting edge technology with mesenchymal stromal cells in 3D platform

Mesude Bicer[1*] (*ORCID: https://orcid.org/0000-0001-7089-5661*)

[*1]Abdullah Gul University, Faculty of Life and Natural Science, Bioengineering Department, Kayseri, Türkiye.

*Corresponding author e-mail: mesude.bicer@agu.edu.tr

Abstract

Adult stem cells, including mesenchymal stromal cells (MSCs), are easily isolated from various human tissues like adipose tissue and bone marrow. MSCs possess immunomodulatory abilities that affect numerous immune cell types and can differentiate into mesenchymal tissues such as bone, cartilage, and fat cells. They are predominantly well-tolerated and non-tumorigenic and have shown potential benefits in remedying various disorders. This has positioned MSCs as a promising source of stem cells for clinical applications. As of August 2023, there were over 1362 registered clinical trials, exploring their potential in treating conditions such as diabetes, cancer, cardiovascular diseases, and musculoskeletal disorders. MSCs were conventionally cultured and differentiated on flat two-dimensional (2D) surfaces composed of glass or polystyrene, which could cause abnormal cell polarity, chromosomal changes, reduced cell viability, and an altered differention potential. To overcome these issues, three-dimensional (3D) scaffolds have been engineered to mimic the natural MSC microenvironment, offering a more expansive culture surface. Recent research indicates that the therapeutic efficacy of MSCs, as observed in proof-of-concept and clinical studies, is not solely attributed to their integration into tissues but is remarkably influenced by the characteristics of 3D-scaffolds. However, visualizing cells within 3D frequently encounters obstacles, such as low signal-to-noise ratios, primarily due to the light-absorbing properties of hydrogels. In this study, adipose tissue-derived MSCs were cultured within a novel hydrogel that are comprised of plant-derived nanofibrillar cellulose. This 3D hydrogel formed nanofibers, consisting of a porous network suitable for MSC. Significantly, it exhibited minimal light absorption in the visible spectrum and displayed biocompatibility with MSCs, ensuring their viability. Moreover, the hydrogel facilitated uncomplicated light-based imaging techniques, including the utilization of conventional dyes and immunocytochemistry within 3D context. To conclude, this innovative 3D hydrogel holds promise as a material for expanding MSCs and studying cellular morphology and behavior within 3D environment.

Keywords: Mesenchymal stromal cells, regenerative medicine, 3D hydrogel

DRAFT

POSTER PRESENTATION

İlaca dirençli epilepsisi olan çocuklarda besin alımı ile yaşam kalitesi ve nöbet sayısı ilişkisi

Gamze Yurtdaş Depboylu[1*] (*ORCID: https://orcid.org/0000-0001-5410-7231*), Olgay Bildik[2] (*ORCID: https://orcid.org/ 0000-0002-0861-5267)*, Gülşah Kaner[1](*ORCID: https://orcid.org/0000-0001-5882-6049*), Pınar Gençpınar[3](*ORCID: https://orcid.org/0000-0002-3223-5408*), Nihal Olgaç Dündar[3](*ORCID: https://orcid.org/0000-0002-5902-3501*)

[*1] İzmir Katip Çelebi University, Faculty, Department of Nutrition and Dietetics, İzmir, Turkiye.
[2]Tepecik Training and Research Hospital, University of Health Sciences, Department of Pediatric Neurology, Izmir, Turkey.
[3]Izmir Katip Celebi University, Department of Pediatric Neurology, Izmir, Turkey

*Corresponding author e-mail: gmzyurtdas@hotmail.com

Abstract

Bu çalışmanın amacı ilaca dirençli epilepsisi olan çocuklarda besin alımı ile yaşam kalitesi ve nöbet sayısı arasındaki ilişkiyi değerlendirmektir. Bu çalışma, İzmir Tepecik Eğitim ve Araştırma Hastanesi Çocuk Nöroloji polikliniğinde takip edilen 2–18 yaş arası ilaca dirençli epilepsisi tanısı olan hastalarla yürütülmüştür. Çalışma verileri yüz yüze görüşme yöntemiyle anket kullanılarak elde edilmiştir. Besin alımının değerlendirilmesi için katılımcılardan üç günlük besin tüketim kaydı alınmıştır. Yaşam kalitesini değerlendirmek için çocuklar için yaşam kalitesi ölçeği (PedsQL) kullanılmıştır. Ölçekte yaşam kalitesi; fiziksel sağlık, duygusal işlevsellik, sosyal işlevsellik ve okul işlevselliği olmak üzere dört alanda puanlandırılmıştır. Katlımcıların %52,8'i kız olup tüm katılımcıların yaş ortalaması 10,6∓4,85 yıl'dır. Haftalık nöbet sayısı ortalama 15,2∓27,42 olarak belirlenmiştir. Toplam enerji alımları 1210,2∓390,75 kkal idi. Toplam enerjinin %46,7 karbonhidrat, %16,2'si protein ve %37'si yağlardan sağlanmaktaydı. PedsQL 'ye göre katılımcıların ortalama fiziksel sağlık puanı 72,4±31,16 puan, duygusal sağlık puanı 71,5±20,39, sosyal işlavsellik puanı 80,0±30,76, ve okul işlevselliği puanı 57,6±25,39 olarak tespit edildi. Yaşam kalitesi ölçek puanları her iki cinsiyette benzer dağılım göstermekteydi (p>0,05). Okul işlevselliği ile diyet yağ yüzdesi (r=-0,492, p=0,001) arasında negatif yönlü, karbonhidrat yüzdesi ile (r=0,523, p<0,001) pozitif yönlü anlamlı ilişki saptanmıştır. Sosyal işlevsellik ile diyet yağ yüzdesi (r=-0.349, p=0,010) arasında negatif yönlü diyet karbonhidrat yüzdesi (r=0,391, p=0,004) ile pozitif yönlü ilişki saptanmıştır. Haftalık nöbet sayısı ile duygusal işlevsellik (r=-0,600, p=<0.001) negatif yönlü ilişki iken enerji alımı (r= 0,312, p=0,023) ile pozitif yönlü ilişkili bulunmuştur. Bu çalışmada ilaca dirençli çocuklarda besin alımı ile yaşam kalitesi ve nöbet sayısı arasında ilişki olduğu belirlenmiştir. Bu çocuklarda nöbet sayısını kontrol altına almak ve yaşam kalitesi artırmaya yönelik bireysel beslenme müdahaleleri geliştirilmelidir.

Keywords: ilaca dirençli epilepsi, epileptik nöbet, besin alımı, yaşam kalitesi.

DRAFT

POSTER PRESENTATION

Improving the Sensitivity of Polymer Strain Sensors through Dielectric Enhancement

Mehmet Yuksekkaya[1*] (*ORCID: https://orcid.org/0000-0002-2665-5799)*

[*1]Ankara University, Faculty of Engineering, Department of Biomedical Engineering, Ankara, Türkiye.

*Corresponding author e-mail: myuksekkaya@ankara.edu.tr

Abstract

Flexible strain sensors have gained widespread popularity due to their diverse applications in wearable technology, health monitoring, and biomedical devices. Capacitive sensors are an important type of strain sensors. Their sensitivity, or gauge factor, closely tied to dielectric properties. One effective method to boost sensitivity is the incorporation of high dielectric constant materials with polymer base without decreasing elasticity. In this study, we present a practical silicon-based sensor enriched with barium titanate as an additive. The resulting sensor is being tested; significant improvements are obtained. Dielectric constant and gauge factor increased by almost 35% and the elasticity decreased by approximately by 15%. Barium titanate can be used to improve polymer based flexible strain sensors sensitivity.

Keywords: Flexible strain sensors, gauge factor, barium titanate, wearable sensors

DRAFT

POSTER PRESENTATION

Adli Bir Vakada Elde Edilen Liflerin FTIR ve SEM Yöntemleri Kullanılarak İncelenmesi

Beran_ÖNER[1], Belgin_İZGİ[2]

[1]Bursa Uludağ Üniversitesi, Fen Bilimleri Enstitüsü, Adli Bilimler Ana Bilim Dalı, Bursa
[2]Bursa Uludağ Üniversitesi, Fen-Edebiyat Fakültesi, Kimya Bölümü, Bursa

berannoner@gmail.com.tr

Özet

Bu çalışmada, adli bir vakada olay yerinde bulunan şüpheli veya şüphelilerden ele geçirilen ipliklerdeki lif yapılarının SEM (Taramalı Elektron Mikroskopisi) ve FTIR (Fourier Dönüşümlü Kızılötesi Spektroskopisi) kullanılarak mukayese yoluyla aydınlatılması amaçlanmaktadır. Çalışmada maktulün boynundan elde edilen ip ve olay yerinden elde edilen ip lifleri öncelikle fiziksel yöntem incelenmesine tabii tutulmuştur. Ardından bu liflerin mukayesesi SEM (Cihaz marka modeli) ve FTIR (Cihaz marka modeli) cihazlarından elde edilen sonuçlar ile gerçekleştirilmiştir. Farklı materyaller üzerinden elde edilmiş iki adet lif numunesine ait FTIR spektrumlarının birbirine benzer olup örtüştüğü gözlemlenmiştir. SEM görüntülerinde aynı büyütme oranlarında oluşan şekiller ve aynı hızlandırma voltajında meydana gelen görüntülere bakıldığında bu iki lif numunesinin benzer oldukları tespit edilmiştir. Bu çalışmanın sonucunda olay yerinden elde edilen lif numuneleri ile şüpheli veya şüphelilerden elde edilen lif numuneleri FTIR spektrumları ve SEM görüntülerinden söz konusu numunelerin fiziksel ve kimyasal olarak benzerlik gösterdiği görülmüştür. Bu bulgulardan yola çıkarak olay yerinden elde bulunan ve şüphelilerden elde edilen liflerin kaynağının aynı materyal olabileceği belirlenmiştir.

Anahtar Kelimeler: Lif Analizi, Taramalı Elektron Mikroskopisi (SEM), FTIR (Fourier Dönüşümlü Kızılötesi) Spektroskopisi

DRAFT

POSTER PRESENTATION

Inherently Antibacterial Polymeric Micelles for Antibacterial Agent Delivery

Hanan Paşaoğlu Belal, Serap Derman, Murat Topuzoğulları

Yıldız Technical University, Department of Bioengineering, Istanbul, Turkey.

hananvic95@gmail.com

Abstract

One of the major problems that threats the healthcare system worldwide is the multidrug- resistance of pathogenic bacteria against antibiotics. The advanced resistance of bacteria towards conventional antibiotics stimulates rapid intervention to find a new solution against this serious problem that can lead to several diseases and infections in human. Antibacterial polymers have been chosen as a promising alternative to traditional antibiotics, due to their unique mechanism action against bacteria, since antibacterial polymers having cationic amphiphilic structure show selectivity to bacteria via electrostatic interactions between the positive charges on the polymer and the negative charges on the surface of the bacterial cell membrane.

In this study antibacterial polymeric micelles carrying Juglone as antibacterial drug were produced using cationic co-polymers of 4-vinylpyridine (4VP) and oligo (ethylene glycol) methyl ether methacrylate (OEGMA) to obtain inherently antibacterial polymeric micelles as drug delivery system.

The produced copolymers were characterized by NMR, FT-IR, DLS spectroscopies and GPC. The hydrodynamic diameter of produced cationic micelles loaded with different Juglone concentrations ranged between 131-167 nm and the polydispersity indices were below 0.3.

Keywords: antibacterial cationic polymer, micelle, Juglone.

Acknowledgements: This study was supported by Yildiz Technical University Scientific Research Projects Coordination Unit [Project No: FCD-2022-4900].

DRAFT

POSTER PRESENTATION

Manufacture of an anti-ulcer food supplement in capsule form based on "cystoseira sensu lato" brown algae

Djahida Hadj Merabet [1*], Nour el Houda Brahimi [2] , Fayza Baghli [3]

[*1] University Abou Bekr BELKAID, Laboratory of Ppabionut (Physiology, physiopathology and Biochemistry of Nutrition Laboratory) , Faculty of Natural and Life Sciences and Earth and Universe , Department of biology , Tlemcen , Algeria
[2]University Abou Bekr BELKAID , Faculty of Natural sciences and life, earth and universe , Department of Biology , Tlemcen, Algeria
[2]University Abou Bekr BELKAID , Faculty of Medecine , Department of Pharmacology , Tlemcen, Algeria

*Correspondingauthor e-mail: Hadjmerabetd@gmail.com

Abstract

This study aimed to produce an anti-ulcer food supplement based on the brown seaweed "Cystoseira sensu lato", algae that has never been used as a food supplement. Our project is an innovative idea that falls within Decree 1275. A physicochemical study of the aqueous, methanolic and ethanolic extract and an in vivo study on wistar rats were carried out to determine the lethal dose (LD50), the effective dose (ED50) and to study the anti-ulcer activity of this algae. The data obtained showed a zero degree of toxicity with an LD50 equal to 10,000 mg/kg. A strong anti-ulcer effect was noted for the three extracts (aqueous, ethanolic and methanolic) as well as for C. sensu lato algal powder. The different doses of seaweed (50, 100 and 150 mg/kg) significantly reduced the percentage of ulceration with a high percentage of inhibition. It was found that the brown alga powder 'Cystoseira sensu lato' at 150 mg/kg gave a maximum effect in reducing gastric ulceration By following the dose-effect curve, we can estimate the ED50 of the brown alga 'Cystoseira sensu lato' at between 25 and 75 mg/kg. Our results also show that the brown alga C.sensu lato is rich in polyphenols and flavonoids and has significant antioxidant properties. It would therefore be interesting to carry out further studies to confirm and analyse other therapeutic activities of this miraculous algae

Keywords: Cystoseira sensu lato, Anti-ulcer food supplement, Wistar rats , LD50 , ED50

DRAFT

POSTER PRESENTATION

Nutritional Profile And Prevalence Of Polycystic Ovary Syndrome Among Female Students Of The University Of Tlemcen , Algeria

Djahida Hadj Merabet [1*], Meriem SAKER [2] , Imene Frid [3], Chaimaa Ferh [4]

[*1] University Abou Bekr BELKAID, Laboratory of Ppabionut (Physiology, physiopathology and Biochemistry of Nutrition Laboratory) , Faculty of Natural and Life Sciences and Earth and Universe, Department of Biology , Tlemcen , Algeria
[2] University Abou Bekr BELKAID, Laboratory of Ppabionut (Physiology, physiopathology and Biochemistry of Nutrition Laboratory) , Faculty of Natural and Life Sciences and Earth and Universe , Department of Biology , Tlemcen , Algeria
[3,4] University Abou Bekr BELKAID , Faculty of Natural sciences and life, earth and universe , Department of Biology , Tlemcen, Algeria

*Correspondingauthor e-mail: hadjmerabetd@gmail.com

Abstract

Polycystic ovary syndrome (PCOS) is the most common gynecological endocrinopathy in women of reproductive age . This study aims to estimate the prevalence of PCOS among female students at Tlemcen University-Algeria and to determine their nutritional profile. A prospective, cross-sectional study was conducted at the University of Tlemcen. 1124 female students aged 21.67 ± 3.21 years were recruited. Cases of PCOS were identified according to the Rotterdam criteria through a clinical interview, anthropometric measurements and an assessment of the participants' eating habits in the different faculties of the university. The data was analysed using Graph pad prism 9 software**.** The results of this study show that polycystic ovary syndrome affects around 12.81% of female students at the University of Tlemcen. The highest percentages were found in the faculties of natural and life sciences, earth and universe (25.34%), literature and languages (19.86%) and medicine (15.02%). Most subjects with PCOS are overweight (43.83%). It was also noted that most subjects with PCOS have irregular cycles and signs of hyperandrogenism. We also found that a large percentage of the PCOS population prefers fried foods (45%) and sweets (34.2%). Moreover, we noted that only 26% of PCOS patients engage in regular physical activity. The results revealed that the prevalence rate of PCOS is high among female students at the University of Tlemcen , Algeria. It is important for women to be aware of their lifestyle and nutritional profile in order to prevent this and other diseases.

Keywords: Polycystic ovary syndrome, Prevalence, Dietary habits, University of Tlemcen

DRAFT

POSTER PRESENTATION

Analysis of the chemical composition of *Leucojum aestivum* L. seed coats

Demet İncedere Uysal[1] (https://orcid.org/0000-0002-5923-6355) , Erdi Can Aytar[1,2] (https://orcid.org/0000-0001-6045-0183), Yasemin Ozdener Kömpe[1] (https://orcid.org/0000-0003-1649-4298)

[1]Ondokuz Mayıs University, Department of Biology
[2]Uşak University, Department of Horticulture

*sorumlu yazar e-mail: demetdere@gmail.com

Abstract

Alkaloids belonging to the Amaryllidaceae family are known for their antiviral and antitumor properties, with significant components such as galantamine widely used in the treatment of Alzheimer's disease. Additionally, an alkaloid named lycorine exhibits potent antiviral effects against poliovirus, measles, and Herpes simplex type 1 viruses. One of the important sources for the commercial production of galantamine is the *Leucojum aestivum* L. plant. *L.aestivum* is the sole native species of the Leucojum genus in Turkey and belongs to the Amaryllidaceae family. This plant thrives in damp and semi-shaded areas, ranging from sea level to high altitudes, and is locally known by names like 'yaz kar tanesi' (summer snowflake), 'loddon zambağı,' and 'kar tanesi zambağı' (snowflake lily). However, the collection of these plants from the wild is limited and poses a threat to their existence. Therefore, understanding the chemical composition of the seeds of this plant is of great importance. In this context, it has been determined that the seed husks of *L. aestivum* exhibit high DPPH free radical activity (IC_{50}: 214.97 mg/ml) and also possess important characteristics such as total flavonol content, total flavonoid content, total phenolic content, total proanthocyanidin content, and total tannin content.

Keywords: Antioxidant activity, *Leucojum aestivum*, Seed coat

DRAFT

POSTER PRESENTATION

Modelling Temperature and Moisture Profile of Carrot Under Different Storage Conditions

Rabitah Zakaria* (*ORCID: https://orcid.org/0000-0001-5098-3963*), Muhammad Syahmeer How (*ORCID: https://orcid.org/0000-0003-1087-0811*)

*University Putra Malaysia, Faculty of Engineering, Department of Process and Food Engineering, Serdang, Malaysia.

*Corresponding author e-mail: rabitah@upm.edu.my

Abstract

The loss of moisture during storage of fresh carrot can be predicted by developing a simultaneous heat and mass transfer model to predict the movement of water through diffusion, evaporation and convection and it can then be used to predict water loss at different storage conditions. Understanding the relationship between temperature and relative humidity is useful to minimize the water loss and to prolong the shelf life of vegetables. In this study a 3-dimension simultaneous heat and mass transfer mechanistic model was developed based on two primary mechanisms: heat transfer during the cooling of carrots and mass transfer due to evaporation of moisture from the carrots. The developed model was solved using the commercial software COMSOL Multiphysics. The simulated model was validated with experimental data at the storage conditions 8°C - 95% relative humidity (RH), 8°C - 60% RH and 30°C - 82% RH for the moisture profile and 8°C-80% RH for the temperature profile, respectively. The model was found to fit the experimental data well with a determination coefficient (R2) ranging from 0.80 to 0.99. The concentration of water inside carrot reduced from 51223 mol/m^3 to 15205 mol/m^3 when stored at 8°C - 60% RH for 240 hour and at 30°C - 82% RH the concentration of water decreased from 50130 mol/m^3 to 28285 mol/m^3. This indicates that relative humidity has a greater impact on moisture loss than temperature at the storage condition. The model predicted that the carrot has to be stored at higher than 90% RH and at temperature lower 5°C in order to maintain the water loss of lower than 8% for 240 day storage.

Keywords: mechanistic modelling, moiture loss

DRAFT

POSTER PRESENTATION

QSPR models for predicting the soil sorption partition coefficient (Koc) using machine learning algorithms

S. Batouche

University Mohamed Cherif Messaadia, Faculty of Science and Technology, Department des Sciences de la Matière Souk Ahras , Algeria
Laboratoire des Sciences et Tehniques de l'eau et environnement

s.batouche@univ-soukahras.dz

Abstract

A comparative study was undertaken to test the ability of different machine learning algorithms to predict the soil sorption partition coefficient (Koc) for organic compounds. In this work, three different quantitative structure-property relationship (QSPR) models were developed using four molecular descriptors. To develop the QSPR models, three different machine learning algorithms were applied and compared: random forest (RF), support vector machine (SVM), and deep neural network (DNN) algorithms. Based on the statistical parameters, the results showed that the DNN-QSPR model outperformed the RF-QSPR and SVR-QSPR models in terms of accuracy and prediction, (R^2 = 81,4 % mae = 0,3834, mse = 0,0604)

Keywords: QSPR-artificial intelligence,-machine learning, random forest-support vector machine-soil sorption coefficient.

DRAFT

DRAFT

DRAFT

2.2. FULL-TEXT PAPER

DRAFT

DRAFT

DRAFT

DRAFT

POSTER PRESENTATION

DRAFT

POSTER PRESENTATION

Ankara çayı su kalitesinin NSF-WQI indeksi kullanılarak değerlendirilmesi

Arzu ÇİÇEK[1] *(ORCID: https://orcid.org/ 0000-0002-7212-8084)*
Muhammed Tarık Durmuş[2*] (*ORCID: https://orcid.org/0000-0002-0362-0171*),

[*1] Eskişehir Teknik Üniversitesi, Mühendislik Fakültesi, Çevre Mühendisliği, Eskişehir, Türkiye
[2] Çevre, Şehircilik ve İklim Değişikliği Bakanlığı, Ankara, Türkiye

*Sorumlu yazar e-mail: mtarikdurmus@gmail.com

Özet

Ankara çayı, Türkiye'nin en büyük nehirlerinden birisi olan Sakarya Nehri'nin önemli bir kolu olmakla birlikte, yüksek kirlilik baskısı altında bulunan bölgenin kirlilik yükünü tüm havzaya taşıması sebebiyle izlenmesi ve takip edilmesi gereken önemli bir su kaynağıdır. Son yıllarda artan nüfus, kentleşme ve sanayileşmeyle birlikte, su tüketiminin kontrolsüz bir şekilde artması ve tarımsal faaliyetlerde bilinçsizce kullanılan gübre ve tarım ilaçlarının su kaynaklarına karışması sonucunda su kaynaklarındaki su kalitesinin izlenmesi oldukça önem kazanmıştır. Su kalite indeksi metodu ile su kalitesinin anlaşılması ve değerlendirilmesi oldukça pratik bir hal almıştır. Dünyada en çok kullanılan ve diğer su kalite indeks metotlarının dayanağını oluşturan su kalite indeksi Ulusal Sanitasyon Vakfı Su Kalitesi İndeksi (NSFWQI)'dir.

Bu çalışmada, Ankara Çayı'nda ilk kez kullanılan su kalite indeksi metodu çalışılmış ve su kalite indeksine bağlı su kalite sınıflarının tespit edilmesi hedeflenmiştir. Bu amaçla, Ankara Çayı'nın kaynağından başlayarak Sakarya Nehri ile birleştiği yere kadar akarsu boyunca seçilen on adet örnekleme istasyonundan 2019-2020 yıllarında mevsimsel olarak su örnekleri alınmıştır. Alınan numunelerde sıcaklık, pH, bulanıklık, fekal koliform, çözünmüş oksijen, derişimi ve doygunluğu, biyokimyasal oksijen ihtiyacı, toplam fosfat ve toplam nitrat analizleri yapılmış ve NSFWQI'ne göre su kalite sınıfları belirlenmiştir.
Anahtar Sözcükler: Su Kalite İndeksi, Ankara Çayı, NFS-WQI, Çubuk Çayı.

Abstract

While Ankara Stream is an important tributary of Sakarya River, one of the largest rivers in Turkey, it is an important water source that should be monitored and scrutinized since it carries the pollution load of the region that is under high pollution pressure to the entire basin. As a result of the uncontrolled increase in water consumption resulting from increasing population, urbanization and industrialization in recent years, and the careless uses of fertilizers and pesticides in agricultural activities that mix up with water resources, it has become very vital to monitor the water quality of water resources. The water quality index method has become very practical to understand and evaluate water quality. Specifically, the National Sanitation Foundation Water Quality Index, the basis of other water quality index methods, is the most widely used water quality index in the world.

In this study, the water quality index method, is used for the first time in Ankara Stream, with the aim of determining the water quality classification. For this purpose, in each season of 2019 and 2020, water samples were taken from ten sampling stations selected along the stream, starting from the source of the Ankara Stream to the point where it joins the Sakarya River. To be used in water quality index calculations, including taken sample's temperature, pH levels, turbidity values, fecal coliform, dissolved oxygen, concentration and saturation, biochemical oxygen demand, total phosphate and total nitrate analyzes, have been analyzed, and, consequently, Water quality classes are determined according to National Sanitation Foundation Water Quality Index.
Keywords: Water Quality Index, Ankara Stream, NSF-WQI, Sakarya River.

DRAFT

GİRİŞ

Sürdürülebilir kalkınmanın hayat bulmasındaki en önemli unsurlardan bir tanesi "su" dur; suyun yokluğunda kalkınma olmaz, kalkınma olmazsa hayal edilen refahı yüksek seviyelere ulaşılamaz ve yoksulluk engellenemez (Turan ve Eren, 2008). Tarih seyrinde uygarlıkların medeniyete doğru adımlarını hızlandıran en önemli neden su kaynakları olduğu gibi yaşadığımız zaman diliminde de en önemli stratejik unsurlar arasında yer almaktadır.

Dünyanın artan nüfusu ve büyüyen ekonomi hem dünyanın ekolojik dengesini bozmuş hem de su kaynaklarını tehdit edecek seviyeye gelmiştir. Sınırlı kaynaklarla sınırsız büyüme hırsına kapılan üretim sistemi, doğal kaynaklar üzerinde aşırı baskı oluşturmakta ve bunun neticesinde küresel ısınma, iklim değişikliği gibi tamiri mümkün olmayan arızalara sebebiyet vermektedir (Kılıç, 2008).

Su kalitesi, insan faaliyetlerinin neden olduğu genel kirlilik nedeniyle tüm dünyada büyük bir sorun haline gelmiştir. Su kalitesi indeksleri, su kalitesini değerlendirmek, su kirliliğini kontrol etmek, su kalitesini eski haline getirmek veya iyileştirmek ve havzadaki en iyi yönetim uygulamalarının etkilerini değerlendirmek için kolay bir sistemdir (Quilbe vd., 2006). Su kalitesi indeksi, sucul ekosistemdeki genel su kalitesi koşullarını yansıtmaktadır. Bu nedenle su kalitesi indeksi, sulak alanların kalitesi ve olası kullanımları konusunda yöneticiler ve karar vericiler için basit ve anlaşılır bir uygulama olmuştur (Bordalo vd., 2001; Maraşlıoğlu vd., 2018).

Su kalitesini değerlendirmek için birçok farklı yöntem kullanılmakla birlikte, en fazla kullanılan geleneksel yöntemlerde, analizleri yapılan parametrelerin standart sınır değerlerle karşılaştırılması yapılarak su kalitesi ortaya konulmaktadır. Ancak, geleneksel yöntemlerle su kalitesi parametrelerini incelemek hem zaman ve maliyet açısından dezavantajlıdır, hem de analizler sırasında çevre kirliliği oluşturduğu için olumsuz etkilere sahiptir (Noori vd., 2019).

Zaman, maliyet ve çevre kısıtlamaları nedeni ile geleneksel yöntemlerin yerini su kalite indeksi modeli almaktadır. Az sayıda su kalitesi parametreleri kullanılarak, belirli bir zamanda, bir su kütlesine ait genel su kalitesi durumunun, tek bir değer olarak ifade edilmesini sağlayan su kalite indeksinin amacı, farklı birimlere sahip farklı parametreleri içeren anlaşılması zor su kalitesi verilerinin, daha anlaşılabilir hale gelmesini sağlayarak, su kalitesi konusunda uzman olmayan bireylerin veya kurum yöneticilerinin, su kalitesinin durumu hakkında bilgi edinmesini ve bu bilgileri anlaşılır, rahat ve hızlı bir şekilde kullanabilmesini sağlamaktır (Kaçan, 2020).

Su kalitesini etkileyen parametrelerin her birinin, kullanılabilir su tüketimi için kabul edilen su kalitesi üzerindeki etkisini sağlayan derecelendirme tekniği, su kalite indeksi olarak tanımlanmakta olup, yüzeysel sularda olduğu gibi yeraltı sularının kalitesini belirlemek ve içme suyu olarak kullanımının uygunluğunu tespit etmek için önemli bir ölçüdür (Varol ve Şekerci, 2018).

Su kalite indekslerinin kullanılması, su kalitesindeki zaman ve mekandaki değişikliklerin incelenmesine izin veren su kütlelerindeki su kalitesi durumunu değerlendirmek için basit ve güçlü bir araçtır (Noori vd., 2019).

Dünyada pek çok su kalite indeksi formları mevcuttur ancak en çok bilinen su kalite indeksleri; Kanada Su Kalitesi İndeksi (CWQI), British Columbia İndeksi (BCWQI), Oregon Su Kalitesi İndeksi (OWQI), Ulusal Sanitasyon Vakfı Su Kalite İndeksi (NSF WQI)'dir. Bu çalışmada Ankara çayının su kalite indeksi Ulusal Sanitasyon Vakfı Su Kalite İndeksi (NSF WQI) ile değerlendirilmiştir.

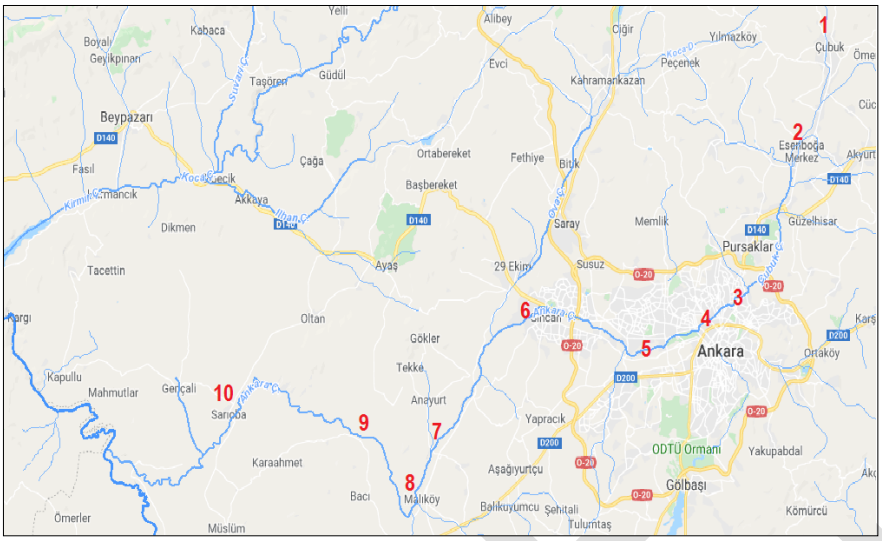
Bu çalışmada, Ankara Çayı'nın kaynağına yakın noktadan başlayarak Sakarya Nehri ile birleştiği yere kadar tespit edilen 10 istasyondan, 2019 yılında mevsimsel olarak su numuneleri alınmış, su kalite parametreleri ölçülerek NSF-WQI su kalite indeksi hesaplanmıştır.

DRAFT

MATERYAL METOT

2.1 Çalışma alanı

Çalışma alanı olarak seçilen Ankara Çayı, Sakarya Havzası'nın önemli akarsularından birisidir. Ankara İli içinden geçerek Sakarya Nehri'ne dökülen 140 km uzunluğundaki Ankara Çayı'nın iki önemli kolu Çubuk Çayı ve Hatip Çayı'dır (Eker ve Özkan, 2017).



Görsel 1. Çalışma alanı

Bu çalışma kapsamında, Ankara Çayı'nın üzerinde belirlenen 10 istasyondan su örnekleri alınmıştır. Çalışma alanının haritası Görsel 1'de ve örnekleme yapılan istasyonların bilgileri Tablo 1'de verilmektedir.

Tablo 1. Örnekleme yapılan istasyon bilgileri

İstasyon	Koordinat - Enlem / Boylam	Mevkii
1. İstasyon	(y) 40.264860 / (x) 33.025591	Çubuk
2. İstasyon	(y) 40.141782 / (x) 32.984644	Esenboğa
3. İstasyon	(y) 39.988622 / (x) 32.903226	Solfasol
4. İstasyon	(y) 39.955750 / (x) 32.837295	Akköprü
5. İstasyon	(y) 39.937872 / (x) 32.705455	Şaşmaz
6. İstasyon	(y) 39.973326 / (x) 32.570492	Sincan
7. İstasyon	(y) 39.834161 / (x) 32.401296	Polatlar Köyü
8. İstasyon	(y) 39.788582 / (x) 32.374978	Malıköy
9. İstasyon	(y) 39.845176 / (x) 32.298218	Girmeç Köyü
10. İstasyon	(y) 39.863670 / (x) 32.084781	Sarıoba Köyü

Deneysel Çalışmalar

2018-2019 tarihleri arasında, mevsimsel olarak Ankara Çayı'nda belirlenen 10 istasyondan su örnekleri alınmıştır. Numune alma, saklama ve taşıma işlemleri standartlar kapsamında gerçekleştirilmiştir. Ankara Çayı üzerinde belirlenen istasyonlardan alınan numuneler aynı gün Eskişehir Teknik Üniversitesi Çevre Uygulamaları Laboratuvarına getirilerek analizler yapılmıştır.

Alınan su örneklerinde pH, sıcaklık (°C), çözünmüş oksijen derişimi (mg/L) ve çözünmüş oksijen saturasyonu (%) seviyeleri arazide Thermo Orion Star A329 cihazı ile yerinde, toplam nitrat (mg/L), toplam fosfat (mg/L) ve BOİ (mg/L) değerleri uygun koşullarda muhafaza edilerek laboratuvara getirilen örneklerde Hach Lange marka (DR 2800) multi parametre ölçüm cihazı ile ölçülmüştür. Fekal koliform değerleri ise uygun koşullarda muhafaza edilerek laboratuvara getirilen örneklerde En Muhtemel Sayı (EMS) analizi ile tespit edilmiştir.

BULGULAR

Çalışmamız sonucunda elde edilen sıcaklık, pH, çözünmüş oksijen, nitrat, fosfat, BOİ, bulanıklık ve fekal koliform verilerinin mevsimsel değişimi Şekil 1'de verilmiştir. Elde edilen veriler Yerüstü Su Kalitesi Yönetmeliğine göre değerlendirildiğinde; sıcaklık, pH ve nitrat parametrelerinin I. sınıf, çözünmüş oksijen ve BOİ parametrelerinin III. sınıf ve fosfat değerlerinin ise IV. sınıf su kalitesinde olduğu tespit edilmiştir.

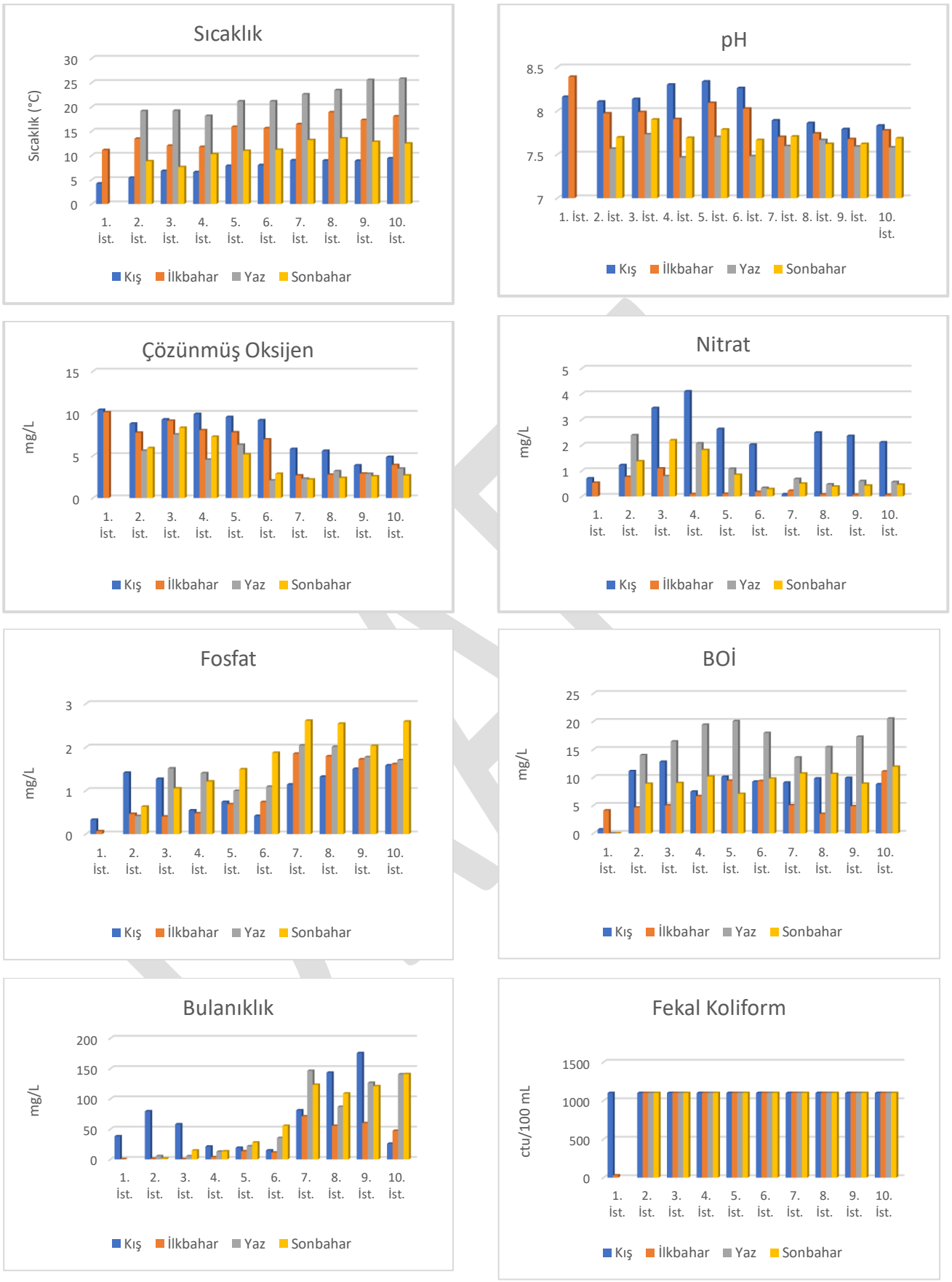
TARTIŞMA VE SONUÇ

Sucul sistemlerde su sıcaklığı birçok fiziksel, kimyasal ve biyolojik olayı etkilediğinden dolayı önemli bir faktördür (Çiçek vd., 2017). Ankara Çayı'nda en yüksek sıcaklık yaz mevsiminde 25,3°C olarak 9. ve 10. istasyonlarda, en düşük sıcaklık ise kış mevsiminde 4,7°C olarak 1. istasyonda ölçülmüştür. Tüm istasyonlarda sıcaklık değişim grafiği mevsimlerden bağımsız olarak aynı eğimde değişim göstermiştir. Ankara Çayı'nın kaynağına yakın olan 1. istasyonda en düşük sıcaklık değeri ölçülürken, şehir merkezine veya sanayi tesislerine doğru ilerledikçe su sıcaklığının arttığı tespit edilmiş, Sakarya Nehri ile birleşme noktasına en yakın konumdaki 10. istasyonda ise en yüksek su sıcaklığı ölçülmüştür. Ankara Çayı'nda yapılan sıcaklık ölçümleri literatürde yer alan çalışmalarla paralellik göstermektedir.

pH, sucul sistemdeki fizikokimyasal ve biyolojik faktörleri değiştirerek suyun asitliğini ve alkalinitesini yansıtan bir parametredir. Ankara Çayı'nda en yüksek pH değeri ilkbahar mevsiminde 8,39 olarak, en düşük pH değeri ise yaz mevsiminde 7,46 olarak ölçülmüştür. pH değerleri Yerüstü Su Kalitesi Yönetmeliği (YSKY)'ne göre değerlendirildiğinde, I. sınıf, yüksek kaliteli sular sınıfa girdiği ve literatürde yer alan çalışmalarla paralellik gösterdiği saptanmıştır.

Çözünmüş oksijen derişimi ve doygunluğu parametreleri, suda çözünen moleküler oksijendi ifade etmekte olup içeriği, su kütlesinin kendi kendini arıtma yeteneğini ölçmek için bir göstergedir. Bu parametre, en düşük yaz mevsiminde 0,82 mg/L olarak 6. istasyonda (III. sınıf) ve en yüksek ise kış mevsiminde 10,32 mg/L olarak 1. istasyonda (I. sınıf) ölçülmüş, mevsimlere bağlı değişiklik gösterdiği tespit edilmiştir. Çözünmüş oksijen derişimi, sıcaklık ve basıncın bir fonksiyonu olduğundan ve sudaki biyolojik aktiviteyi etkilediğinden önemli bir parametredir. Sıcaklıkla orantılı olarak yaz mevsiminde çözünmüş oksijen değerlerinin azaldığı ve en yüksek değerlerin kış mevsiminde ölçüldüğü görülmektedir. Genel olarak mevsimsel değişim grafiklerinin birbirine paralel olduğu ancak tüm mevsimlerde 6. istasyondan itibaren çözünmüş oksijen değerlerinde düşme olduğu görülmekte olup, bu düşüşün nedeni olarak, bu istasyondan itibaren sanayi tesislerinin ve kentsel yerleşimlerin sayısının ve bu tesislerden/yerleşimlerden kaynaklanan kirlilik yükünün artmış olması gösterilebilir. Çözünmüş oksijen derişimi ve doygunluğu değerlerinin, YSKY'ye göre değerlendirildiğinde III. sınıf, orta kaliteli sular sınıfa girdiği tespit edilmiştir.

Nitrat (NO_3^-), doğada var olan bitkiler için önemli bir besin kaynağıdır. Toprakla buluşarak sulara karışması; doğal veya suni gübre kullanımından, evsel veya endüstriyel atıksu deşarjlarından, septik tanklardan olabilmektedir. Nitrat parametresi, en düşük seviyede ilkbahar mevsiminde 0,065 mg/L olarak 10. istasyonda (I. sınıf) ve en yüksek seviyede ise kış mevsiminde 4,1 mg/L, olarak 1. istasyonda (II. sınıf) ölçülmüş, mevsimlere bağlı değişiklik gösterdiği tespit edilmiştir. Nitrat derişimi kış mevsiminde tüm istasyonlarda yüksek değerlerde seyrederken, diğer mevsimlerde 4. istasyondan itibaren düşüşe geçtiği görülmektedir. Nitrat derişiminin yüksek kaydedildiği istasyonların bulunduğu bölgeler, diğer istasyonlara nispeten sanayi tesislerinin daha az, tarım alanlarının daha çok olduğu bölgeler olması, nitrat kirliliğinin tarımsal faaliyetlerde kullanılan doğal veya suni gübrelerden veya evsel atıksu deşarjlarından kaynaklanabileceğini düşündürmektedir. Nitrat değerleri, YSKY'ye göre değerlendirildiğinde, kış mevsiminde 3. ve 4. istasyonlar (II. sınıf) hariç, diğer istasyonların tüm mevsimlerde I. sınıf yüksek kaliteli sular sınıfa girdiği tespit edilmiştir.



Şekil 1. Parametrelerin mevsimsel değişimi

Fosfor, sucul canlılar için mühim bir besleyici mineraldir ve doğal suların verimliliğini etkileyen önemli parametrelerden birisidir. Toplam fosfat parametresi, en düşük seviyede ilkbahar mevsiminde 0,066 mg/L olarak 1. istasyonda (I. sınıf) ve en yüksek seviyede ise sonbahar mevsiminde 2,59 mg/L olarak 10. istasyonda (III. sınıf) ölçülmüş, mevsimlere bağlı değişiklik gösterdiği tespit edilmiştir. Toplam fosfat analizlerine bakıldığında, yıllık ortalama ve mevsimlerdeki yükseliş trendinin 1. istasyonda en düşük seviyede iken 10. istasyona doğru arttığı görülmektedir. Her mevsimde özellikle 6. istasyondan sonra değerlerde ciddi bir artış görülmektedir. Toplam fosfat değerlerinin değişim grafiği, Ankara Çayı boyunca kirliliğin az olduğu yerden yoğun olduğu yere doğru besin girişi olduğunu göstermektedir. 6. istasyon yoğun sanayi tesislerinin ve evsel atıkların kaynaklandığı yerleşim bölgelerinin ortasında kalan bir konumdadır. Civarda Ankara Çayı'nın organik madde yükünü arttıran sanayi kuruluşlarının başında gelen süt ve süt mamulleri üreten tesisleri, entegre et ve tavuk işleme tesisi ve mezbahalar olması, aynı zamanda yakın bölgede hayvanat bahçesi ve evcil hayvan parkının varlığı, toplam fosfat kirliliğinin sanayi kaynaklı olmakla birlikte hayvansal atık kaynaklı olabileceğini de göstermektedir. Toplam fosfat değerleri, YSKY'ye göre IV. sınıf, zayıf kaliteli sular sınıfa girdiği tespit edilmiştir.

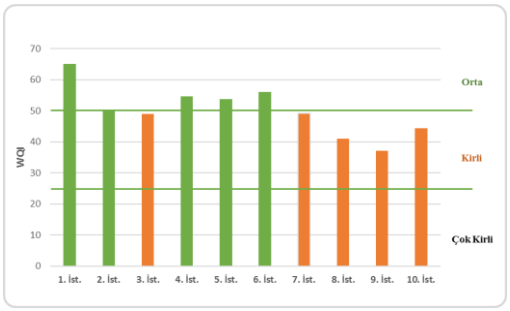
BOİ konsantrasyonu fazla olan bir çıkış suyunun oksijen tüketimi yeterince yüksekse yani anaerobik şartlara geçiyorsa, deşarj edildiği akarsuya zarar verebilmektedir. BOİ parametresi, en düşük seviyede kış mevsiminde 0,7 mg/L olarak 1. istasyonda (I. sınıf) ve en yüksek seviyede ise yaz mevsiminde 20,51 mg/L olarak 10. istasyonda (III. sınıf) ölçülmüş, mevsimlere bağlı değişiklik gösterdiği tespit edilmiştir. BOİ analizlerinde ölçülen değerler yaz mevsiminde yüksek seyrederken, diğer mevsiminde daha az ve aynı seviyede olduğu görülmüştür. BOİ değerleri YSKY'ye göre değerlendirildiğinde, III. sınıf, orta kaliteli sular sınıfa girdiği tespit edilmiştir. Ankara Çayı'ndaki organik kökenli kirliliğin yüksek olması sonucunda akarsuyun BOİ değeri de hatırı sayılır derecede yüksek olmaktadır.

Bulanıklık, suyun berraklık ve şeffaflığının bir ölçüsüdür. Bulanıklık parametresi, en düşük seviyede ilkbahar mevsiminde 3. istasyonda 0,42 NTU, en yüksek seviyede ise kış mevsiminde 9. istasyonda 175,5 NTU olarak ölçülmüş, mevsimlere bağlı değişiklik gösterdiği saptanmıştır. Kış mevsiminde yağışların etkisiyle her istasyonda bulanıklık değeri yüksek çıkarken, diğer mevsimlerde özellikle 7. istasyondan itibaren bulanıklık değerlerinde ciddi bir artış görülmektedir. Sanayi tesislerinin yoğun olduğu bu bölgedeki kirlilik durumu, her ne kadar diğer parametrelerdeki dalgalanmalar ile görülmekte ise de bulanıklık değerindeki artışın nedenlerinden birisinin de zemin farklılığından kaynaklandığı düşünülmektedir. Ankara Çayı ıslah çalışmaları kapsamında 7. istasyona kadar zeminin kaplamalı ve duvarların çift taraflı taş tahkimatlı, bu noktadan sonra zeminin toprak olduğu bilinmektedir.

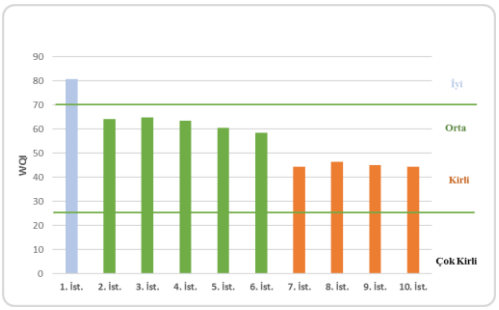
Potansiyel patojen olan fekal koliformun standartları aşan miktarda bulunması, suyun dışkı ile kontamine olduğu ve enfeksiyon taşıdığı anlamına gelmektedir (Çiçek vd., 2017). Fekal koliform değerleri tüm sene boyunca bir istasyon hariç aynı olmuştur. Sadece ilkbahar mevsiminde 1. istasyonda 29/100 ml olarak ölçülmüş, diğer tüm mevsimler ve tüm istasyonlarda ölçülen fekal koliform değeri 1100/100 ml'den fazla olarak tespit edilmiştir. Potansiyel patojen olan fekal koliformun standartları aşan miktarda bulunması, suyun insan sağlığına zararlı olduğu anlamına gelmektedir. Elde edilen bu sonuçlara göre, Ankara Çayı'na kanalizasyon sisteminden sızıntı olabileceği ya da bölgedeki hayvancılık faaliyetlerinden dolayı hayvan dışkılarının Ankara Çayı'na bulaşmış olabileceği tahmin edilmektedir.

Ankara Çayı'nda dört mevsim ölçülen parametrelere göre su kalite indeksi hesaplandığında; (Şekil 2) en düşük NSF-WQI indeks değeri kış mevsiminde 9. istasyon 37,07 (kirli), en yüksek NSF-WQI indeks değeri ise ilkbahar mevsiminde 80,86 (iyi) olarak 1. istasyonda ölçülmüştür.

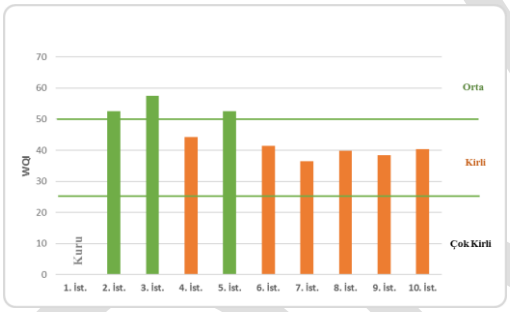
DRAFT



Kış mevsimi su kalite indeksi değerleri

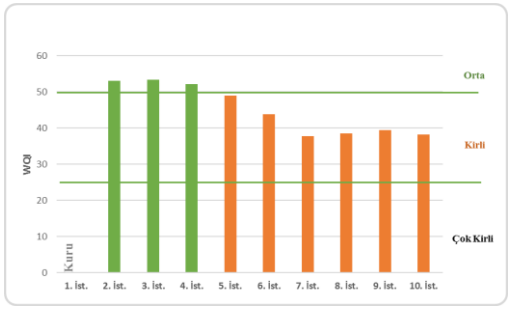


İlkbahar mevsimi su kalite indeksi değerleri

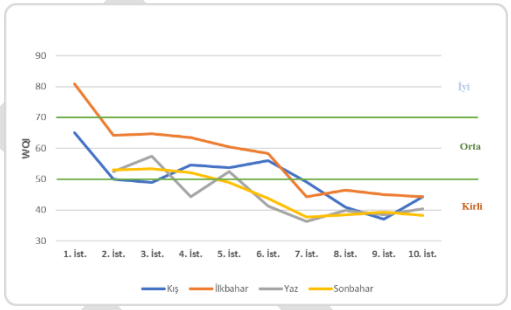


Yaz mevsimi su kalite indeksi değerleri

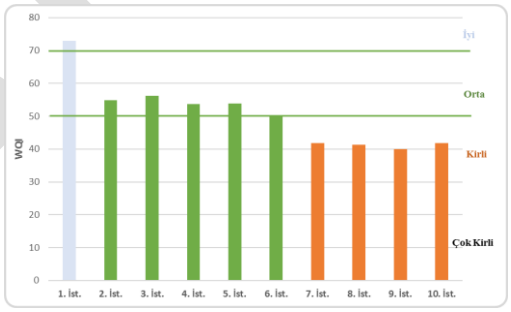
Şekil 2. Ankara Çayı Su Kalite İndeksi Mevsimsel değerleri



Sonbahar mevsimi su kalite indeksi değerleri



Su kalite indeksi değerlerinin mevsimlere değişimi



Ankara Çayı yıllık ortalama su kalite indeksi değerleri

2019 yılı kış, ilkbahar, yaz ve sonbahar mevsimleri su kalitesi sonuçları YSKY'ne göre değerlendirildiğinde; Ankara Çayı genel olarak "IV. sınıf" su kalitesinde, NSF-WQI su kalite indeksine göre değerlendirildiğinde ise su kalitesinin "kirli" olduğu tespit edilmiştir. NSF-WQI modeli uygulaması sonucu elde edilen veriler, her bir parametre için ulusal mevzuattaki su kalite standartlarıyla mukayese edildiğinde, sonuçların uyumlu ve gerçekçi olduğu görülmektedir. Bu da NSF-WQI modelinin Ankara Çayı üzerinde başarılı bir şekilde uygulandığını göstermektedir.

Türkiye'de pek çok akarsuda ciddi kirlilik sorunu yaşanmaktadır ve bu sorunun ortak nedeni arıtılmaksızın alıcı ortama verilen atık sulardır. Su Havzaları, Kullanımı ve Yönetimi Özel İhtisas Komisyonu Raporu'na (2001) göre, Türkiye'nin en fazla kirlenmiş akarsularından biri olan Sakarya Nehri, Ankara Çayı, Porsuk Çayı ve Çark Suyu tarafından taşınan atık sular ile kirletilmektedir. Raporda Ankara Çayı'nın, Ankara nüfusunun yaklaşık %90'ının evsel atık sularını Sakarya Nehri'ne taşıdığı ve IV. sınıf çok kirli sular kategorisinde olduğu ifade edilmektedir. Olumlu ve koruyucu önlemlerle su kütlelerinin potansiyel kirlilik riski etkili bir şekilde önlenebilmekte, yüksek kaliteli ve güvenli bir içme suyu kaynağı sağlanabilmekte ve su ortamının sürdürülebilir gelişimi korunabilmektedir (Zhan vd., 2021). Su kaynaklarının kirlenmesinin önlenmesi, kirletilmiş suların arıtılmasından daha ekonomik olduğundan hem ulusal hem de bölgesel anlamda kirliliğe neden olan hammadde, proses, tesis vs. tüm kaynakların belirlenerek engellenmesi ve su kaynaklarının kontrol altına alınması gerekmektedir (Turan ve Ülkü, 2013). Su kaynaklarını kirlenmeden önce gerekli koruyucu ve önleyici tedbirler almak gerekmektedir.

Ankara Çayı su kalitesinin iyileştirilmesi için öncelikle kirlilik kaynaklarının akarsuya ulaşmasının engellenmesi gerekmektedir. Tarım arazilerinde bilinçsiz ve denetimsiz kullanılan tarım ilaçlarından kaynaklanan kirliliğin yanı sıra bazı bölgelerde kanalizasyon atıklarının Ankara Çayı'na deşarj edildiği veya kanalizasyon sistemindeki kaçaklardan dolayı sızıntıların olduğu bilinmektedir. Bu kirlilik yükü Ankara Çayı su kalitesinin kötü olmasının en önemli sebeplerinden birisidir. Yapılması gereken ilk çalışma, Ankara Çayı üzerinde bulunan kaçak deşarj noktalarının tespit edilerek önlenmesi olacaktır.

Diğer yandan, Ankara Çayı'na deşarj eden atıksu arıtma tesislerinin deşarj kriterleri yeniden gözden geçirilmeli ve deşarj edilen arıtılmış su kalitesi denetlenmelidir. Bilindiği üzere, ASKİ'ye ait birçok atıksu arıtma tesisi ve organize sanayi bölgelerindeki arıtma tesisleri arıtılmış suları Ankara Çayı'na deşarj etmektedir.

Ankara'da faaliyetleri sonucu belli bir miktarın üzerinde atıksu elde eden sanayi tesisleri bir araya getirilip, Ergene Havzasında olduğu gibi ıslah OSB'ler kurulması ve sanayide daha az su, daha az enerji, daha az kirletici hammadde kullanılmasına yönelik proseslerin geliştirilmesi ve Ar-Ge çalışmalarının yapılması önerilmektedir. Atıksu Artıma Tesislerinin ve Organize Sanayi Bölgelerinin atıksu artıma sistemleri periyodik olarak denetlenmeli, Ankara Çayı'na deşarj edilen arıtılmış su kalitesinin yüksek seviyede olması sağlanmalıdır. Ayrıca OSB'lerde arıtılan suların tekrar OSB'deki tesislerde kullanılmasının sağlanabileceği ileri artıma sistemlerinin kurulması ve işletilmesi teşvik edilmelidir.

Sonuç olarak yapılan bu çalışmada, Ankara Çayı su kalite indeksi belirlenmiş, Ankara Çayı'nın su kalitesinin NSF-WQI kalite indeksine göre "kirli" olduğu tespit edilmiştir. NSF-WQI modeli uygulaması sonucu elde edilen veriler, her bir parametre için ulusal mevzuattaki su kalite standartlarıyla mukayese edildiğinde, sonuçların uyumlu ve gerçekçi olduğu görülmektedir. Ankara Çayı üzerinde kirlilik baskısı oluşturan; tarım arazilerinde bilinçsiz ve kontrolsüz kullanılan kimyasal gübrelerin kontrol altına alınması, şehrin kanalizasyon sistemindeki kaçakların tespit edilerek engellenmesi, atıksu arıtma tesislerinden deşarj edilen arıtılmış suların periyodik olarak denetlenmesi durumunda Ankara Çayı su kalitesinde iyileşmeler hemen görülecektir. Sakarya Nehri'nin önemli bir parçası olan Ankara Çayı, bölgedeki kirlilik yükünü tüm havzaya taşıdığından, ivedi bir şekilde uygulanabilir tedbirleri alınması hem Ankara Çayı hem Sakarya Havzası için önem arz etmektedir.

KAYNAKLAR

Bordalo, A. A., Nilsumranchit, W., & Chalermwat, K. (2001). Water quality and uses of the Bangpakong River (Eastern Thailand). Water Research,, 35, 3635–3642. doi:10.1016/ S0043-1354(01)00079-3.

Çiçek, A., Uysal, E., Köse, E., & Tokatlı, C. (2017). Eskişehir'de Yer Alan Bazı Sulama Göletlerinin Su Kalitesinin Değerlendirilmesi. Nevşehir Bilim ve Teknoloji Dergisi , Cilt 6(ICOCEE 2017 Özel Sayı) 440-446.

Eker, Ç. S., & Özkan, Ö. (2017). Ankara Çayı Dere Yatağı Sedimanlarının Zararlı Element İçeriği. Gümüşhane Fen Bilimleri Degisi (GÜFBED), 208-220.

Kaçan, M. (2020). Fethiye Körfezinde Su Kalitesi İndeksleri İle Değerlendirmenin Karşılaştırılması. Ankara: Ankara Üniversitesi Fen Bilimleri Enstitüsü Yüksek Lisans Tezi.

Kılıç, S. (2008). Küresel İklim Değişikliği Sürecinde Su Yönetimi. İ.Ü. Siyasal Bilgiler Fakültesi Dergisi, No:39 / 161-186.

Noori, R., Berndtsson, R., Hosseinzadeh, M., A. J., & Abyaneh, M. (2019). A critical review on the application of the National Sanitation Foundation Water Quality Index. Environmental Pollution, 244, 575-587.

Quilbe, R., Rousseau, A. N., Duchemin, M., Paulin, A., Gangbazo, G., & Villeneuve, J. P. (2006). Selecting a calculation method to estimate sediment and nutrient loads in streams: Application to the Beaurivage River (Quebec, Canada). Journal of Hydrology,, 326 (1-4), 295-310. doi: 10.1016/j.jhydrol.2005.11.008.

Turan, T., & Eren, Z. (2008). Türkiye'de Su Kaynakları Ve Su Politikası. TMMOB 2. Su Politikaları Kongresi (s. 25-31). Ankara: TMMOB.

Turan, F., & Ülkü, G. (2013). Gökpınar Ve Çürüksu Çaylarının Kirlilik Parametre Ve Yüklerinin İzlenmesi. Pamukkale Üniversitesi Mühendislik Bilimleri Dergisi, , Cilt 19, Sayı 3, Sayfalar 133-144.

Varol, S., & Şekerci, M. (2018). Korkuteli İlçe Merkezi (Antalya) Su Kaynaklarının Su Kalite İndeks Yöntemi İle Değerlendirmesi . Mühendislik Bilimleri ve Tasarım Dergisi, 6(1), 74-86,.

Zhan, S., Zhou, B., Li, Z., Li, Z., & Zhang, P. (2021). Evaluation of source water quality and the influencing factors: A case study of Macao . Physics and Chemistry of the Earth , 123, 103006.

DRAFT

POSTER PRESENTATION

Pharmaceutical aspects of drugs used in pathologies of the cardiovascular tract

Klodiola Dhamo* (ORCID: *https://orcid.org/0009-0004-6422-7471*

*1 Aldent University, Faculty of Medical Sciences, Department of Pharmacy, Tirana, Albania.
*Corresponding author e-mail: klodiola.dhamo@ual.edu.al

Abstract

Cardiovascular diseases (CVD) are a medical, social and economic problem. Their treatment today accounts for nearly 80% of the world's health expenditures. The treatment regimens for these diseases often include several drugs that can affect each other's effect. Diseases of the cardiovascular tract are the most frequent and life-threatening pathologies.
The aim of this study is to gain an overview of the diseases and their pharmacological treatment, to determine the prescription patterns of antihypertensive drugs and time trends among diagnosed cases of hypertension, and to investigate the pharmaceutical costs associated with different antihypertensive agents.
The study was carried out in the city of Korça, (for a period of two months), included patients treated for the disease of arterial hypertension, angina pectoris, ischemic heart disease, etc. 348 recipes were analyzed for the period November, December 2022. A total of 348 patients diagnosed with cardiovascular tract diseases (SCV), aged ≥50 years, were identified from the study population of 4000 patients in the computerized reimbursement database of the Health Insurance in the city of Korça.
The data were processed statistically. Data on age, category, dose/prescription, dose/total, reimbursement per prescription were obtained. The frequency encountered of each drug used and the direct cost of treatment were calculated.
Patients who receive drug treatment with arterial hypertension diagnosis have a higher number of medications for the cardiovascular tract with 65.5%. The smallest number with only 9.78% was occupied by patients treated with drugs for the diagnosis of Angina pectoris. The most frequently prescribed antihypertensive drug is Hydrochlorothiazide 25mg, which has the highest prescription frequency with 32.58%, while the lowest prescription frequency is Furosemide 40mg with 9.67%.

Keywords: *Diseases of the cardiovascular tract, diagnosis, drugs, patients, costs*

INTRODUCTION

Treatment of patients with hypertension is carried out by means of drugs as well as following the advice of specialists on lifestyle, nutrition and physical activity.
Some drugs are more effective in treating hypertension and have different good or side effects in patients..
The mortality rate from ischemic heart disease attributable to hypertension has doubled in Albania over the past 20 years, while the death rate from cerebrovascular diseases due to arterial hypertension has increased by more than 70%.
Hypertension is the most common disease and costs the most in terms of medications, which are covered by both the Insurance and Health Care Institutes.
Pharmacists intervention can modify factors associated with compliance, improve compliance in achieving the goal of lowering blood pressure in patients treated with antihypertensive agents.

MATERIALS AND METHODS

The study was carried out in the city of Korça, (for a period of two months), included patients treated for the disease of arterial hypertension, angina pectoris, ischemic heart disease, etc. 348 recipes were analyzed as a representative sample for the period November-December. A total of 348 patients diagnosed with cardiovascular tract diseases (SCV), aged ≥ 50 years, were identified from the study population of 4000 patients in the computerized database of Health Insurance reimbursements in the city of Korça.

DRAFT

The data were processed statistically. Data on age, category, dose/prescription, dose/total, reimbursement per prescription were obtained. The frequency encountered of each drug used and the direct cost of treatment were calculated

To prevent possible confounding by comorbidities in prescription patterns of antihypertensive agents, patients diagnosed with ischemic heart disease, pulmonary circulation disease, other forms of heart disease (including dysrhythmia and heart failure), stroke or kidney, were excluded from the study sample.

Six major classes of antihypertensive drugs are generally available, including angiotensin-converting enzyme (ACE) inhibitors, angiotensin receptor blockers (ARBs), beta-blockers, calcium channel blockers (CCBs), diuretics, and others (all other antihypertensive classes including alpha-blockers).

Prescriptions for a chronic disease in Albania, such as hypertension, most often included the prescription of drugs for 28-180-day periods, which would allow the patient to see a doctor every one to six months. As each prescription could contain different drug combinations and duration of medication, data analysis was undertaken using the prescription rate, calculated as the number of prescriptions containing a specific antihypertensive agent divided by the total number of prescriptions.

RESULTS

Table 1. Distribution of patients by age group in number and percentage, who use reimbursed drugs for the treatment of cardiovascular tract pathology.

Age	**Nr of patients**	**Age in %**
51-60	15	4.40
61-70	81	23.30
71-80	137	40.50
81-90	113	31.20
91-100	2	0.60

Table 2 Distribution of patients by gender in number and percentage, who use drugs with reimbursement, for the treatment of cardiovascular tract pathology

Gender	**Nr of patients**	**%**
Female	219	63
Male	129	37

Table 3 Distribution of patients according to the type of diagnosis of the cardiovascular tract medication

Diagnostic	Nr	%
AHT	228	65.5
Arithmia	43	12.35
Arterosklerosis	43	12.35
Angina pectoris	34	9.78

Table 4 Distribution of patients according to concomitant diseases

Diagnostic	**Co-occurring diseases**	**%**
AHT	82	71.9
Arithmia	1	0.87
Arterosklerosis	7	6.14
Angina pectoris	3	2.63
Fat metabolism disorder	21	18.4

DRAFT

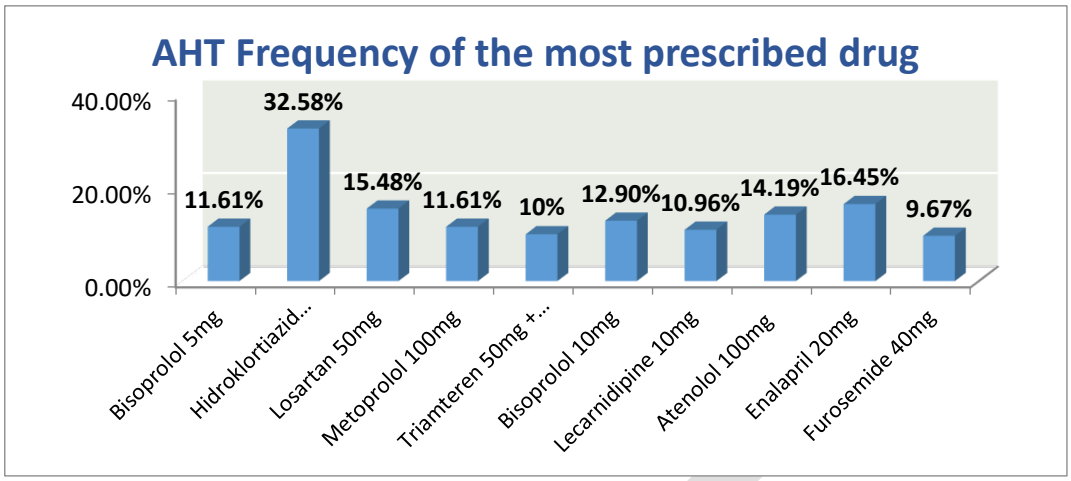


Figure 1 Distribution of the 10 most frequently prescribed antihypertensive drugs, ordered by frequency of prescription

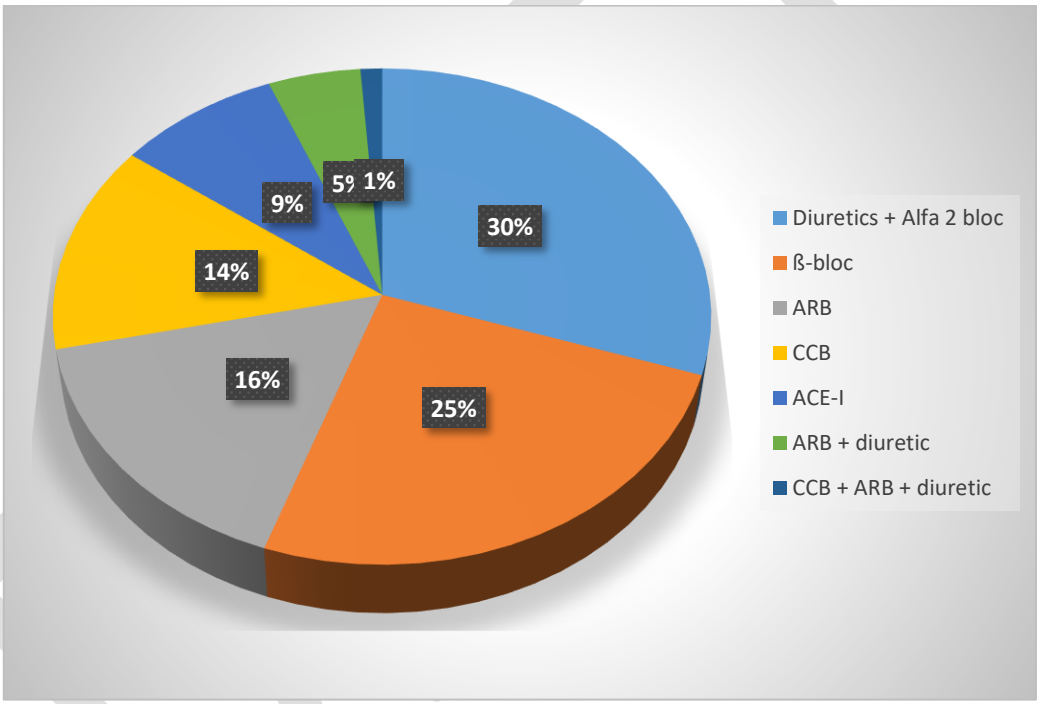


Figure 2 Distribution according to the 7 most frequently prescribed antihypertensive regimens, ordered by frequency of prescription

Table 5 Distribution of categories of pensioners by gender.

Gender/categoria	**Permanent pensioner**	**Totally disabled**
Female	204 (62.74%)	2 (50%)
Male	121 (37.26%)	2 (50%)

Table 6 Average cost distribution for each diagnosis

Diagnostic	**AHT**	**Arithmia**	**Angina pectoris**	**Arterosclerosis**
Average cost for prescriptions	487.34 Lek	911.95 Lek	435 Lek	263.61 Lek
Average cost in percentage	23.22%	43.46%	20.78%	12.56%

CONCLUSION

Patients who receive treatment for drugs with HTA diagnosis have a higher number of medication for the cardiovascular tract with 65.5%. The smallest number with only 9.78% was occupied by patients treated with drugs for the diagnosis of Angina pectoris.

The co-morbidity encountered most often in patients is HTA with 71.9%. While the less frequently encountered co-morbidity is heart disease with 0.87%.

The most frequently prescribed antihypertensive drug is Hydrochlorothiazide 25mg, which has the highest prescription frequency with 32.58%, while the lowest prescription frequency is Furosemide 40mg with 9.67%.

Females have the highest tendency for the category "Pensioner without term" with 62.74%. "Chronic patients" women occupy the largest part with 68.43%, while men make up the smallest number with 31.57%.

Diagnosis Heart rhythm disorder has the average cost of treatment of the highest diagnosis worth 911.95 Lek. The average cost per prescription is 650.94 Lek. The smallest reimbursement cost per prescription is 90 Lek for the drug Metaprolol 100mg.

The value of the prescription with the highest reimbursement cost belongs to the category of the disease Heart rhythm disorder with a value of 5800 Lek with the drugs Rivaroxaban Fenofibrate 200mg and Metaprolol 100mg.

REFERENCES

Albrecht, S. (2019). The Pharmacist's Role inMedication Adherence. U.S. Pharmacist, 36(5), 45-48.

Emberly, P., Davies, C., & Li, Y. (2017). Pharmacists' role in hypertension management. The Translator, 8, 1.

Carter BL, Bosworth HB, Green BB. (2021) The hypertension team: The role of the pharmacist, nurse, and teamwork in hypertension therapy. Journal of Clinical Hypertension.

Chobanian AV, Bakris GL, Black HR, et al.(2021) Seventhreport of the Joint National Committee on Prevention,Detection, Evaluation, and Treatment of High BloodPressure.

Christensen AJ, Howren MB, Hillis SL, et al.(2020) Patient andPhysician Beliefs About Control over Health:Association of Symmetrical Beliefs with MedicationRegimen Adherence. J Gen Intern Med.

http://FSDKSH.gov.al

DRAFT

POSTER PRESENTATION

Microbial activity and chemical composition of essential oil of myrtus communis

Fenghour Asma[1*], Harbi Wafa[2] ,Harzallah leila[3]

[*1]. Analytical Chemistry Laboratory, Department of Pharmacy, Faculty of Medicine, Badji Mokhtar University, BP 205, Annaba (23000), Algeria
[2]. Cardiology Service, Department of Medecine, Faculty of Medicine, Badji Mokhtar University, BP 205, Annaba (23000), Algeria

[*3]. Analytical Chemistry Laboratory, Department of Pharmacy, Faculty of Medicine, Badji Mokhtar University, BP 205, Annaba (23000), Algeria

*Corresponding author e-mail: asma.fenghour@gmail.com

Abstract

This study aims to determine the chemical composition of the essential oil of Myrtus Communis and the evaluation of its antibacterial activity. The extraction of essential oils was carried out by hydro distillation of the aerial parts. The chemical composition of the oils obtained was carried out using GC/MS chromatographic techniques, while the microbiological study was evaluated by the disk diffusion method on 13 pathogenic strains including gram (+) and gram (-).
The results obtained showed that the essential oil of Myrtus Communis is characterized by the presence of: alpha-pinene (49.93%), D-limonene (6.42%), Eucalyptol (34.46%), Linalool (2, 84%) , alpha-terpineol (2.33%) as main constituents. The antibacterial effect determined by the disc diffusion method showed that our oil proved to be inactive on all the strains tested except for two strains Staphyloccocus aureus and Proteus mirabilis with an inhibition diameter of 15 and 13mm respectively.

Keywords: communist; essential oil; CPG/MS-; antibacterial activity

Introduction : The myrtle (Myrtus communis) is a shrub of the Myrtaceae family of which it is the type. The name of the myrtaceae comes from the myrtle (genus Myrtus) the only European survivor of this vast family which, having been eliminated from Europe by the glaciations, found refuge in the tropical and subtropical regions of the globe.
Rich in essential oil, this species used in cooking has therapeutic virtues as an antiseptic, antispasmodic, anti-inflammatory...
The objective of this study is the valorization of Myrtle through the analysis of its essential oil and the evaluation of its antifungal and antioxidant activity.

MATERIALS AND METHODS:

1/ Plant material
The drug: dried leaves of Myrtus communis.
Harvest time: March (before flowering).
Provenance: Seraidi. wilaya of Annaba. Algeria.

2/ Extraction and identification of the essential oil
Extraction of essential oil from myrtle leaves was carried out by steam distillation for 3 hours. The average yield is 1.14ml per 100gr of dried leaves.
The identification of the chemical components of H.E. is carried out by GC/MS of the clarus 600 D Perkin Elmer type. The column used is a RESTEK Rtx®-5MS (30 meters in length, 0.25 mm internal diameter, film thickness is 0.25 µm).
A splitless injection mode is used. Helium was used as carrier gas at a flow rate of 1ml/min. The injector and transfer line temperatures were increased to 250°C. The initial temperature was maintained at 60° C. for 1 minute, then increased by 3° C. per minute to reach 200° C. and is maintained under isotherm for 13 minutes.

DRAFT

The volume injected was 1 µL. The standard solutions and the essential oil were diluted in absolute alcohol (1g/L).

Analyzes were performed in electron impact ionization (E-I) mode with an ionization energy of 70 eV using Scan mode acquisition (from 40 up to 600µ).

The identification of the compounds was made by comparison of the retention times, on the one hand, of the samples, and on the other hand, of the standard solutions, then by comparison of the mass spectra with those contained in the NIST® commercial library (National Institute of Standards and Technology).

The percentage of each compound is calculated by the internal normalization method.

3/ Evaluation of antifungal activity:

The antifungal activity is measured by the tube dilution technique on the reference strain Candida albicans ATCC 10231. The culture medium used is liquid Sabouraud Chloramphenicol. After incubation for 24 h at 35°C, the absorbance is measured at 560 nm.

The experimental data is subjected to statistical analysis. The graphical representation of the inhibition of fungal growth as a function of the concentration of essential oil presents a hyperbolic aspect with the equation $y = ax / (b+x)$. The use of an adjustment software makes it possible to determine the parameters a and b from which one will deduce the MIC 80% which characterizes the antifungal activity, and the constant of affinity Kaff (1/b) of the antifungal for yeast.

Parameter a: calculated maximum inhibition.

Parameter b: concentration of antifungal that induces a calculated inhibition equal to half of the calculated maximum inhibition.

4/ Evaluation of the antioxidant activity:

The antioxidant activity of the essential oil is measured by the ferric iron reduction method. The reducing power of ferric trichloride is determined by the following method reported by Zhao et al:

-100ul of essential oil is mixed with 2.5ml of phosphate buffer (pH 6.6) and 2.5ml of 1% ferricyanide.

-the mixture is incubated at 50° C. for 20 minutes. To the latter, 2.5 ml of 10% trichloroacetic acid is added followed by centrifugation at 3000 rpm for 10 minutes.

-5ml of supernatant is taken and then mixed with 5ml of distilled water and 1ml of 1% ferric trichloride

_The absorbance is then measured at a wavelength of 700nm. The final result of this measurement is expressed in ug of ascorbic acid equivalent / g of dry weight

RESULTATS :

1/ GC/MS analysis gave the following results:

The main constituents of H.E. are shown in the following table

N°	Denomination	Teneur en %
1	Alpha-pinene	49,93
2	D-limonene	6,42
3	Eucalyptol	34,46
4	Linalol	2,84
5	Alpha-terpineol	2,33
6	5-tetradecene	0,69
7	Geranyl-acetate	2,03
8	Eugeanol methyl ether	1,30

2/The antifungal action of H.E. myrtle on a strain of Candida albicans gave a low MIC value: 18.87µl/ml for a Kaff of around 0.11

3/the antioxidant activity measured by the iron reduction method (FRAP) revealed moderate activity with an EC50= 3.26mg/mL.

Conclusion:

Thanks to GC/MS applied to the analysis of H.E. of Myrtus communis, 8 major constituents have been identified, the most important being: alpha-pinene (49.93%), eucalyptol (34.46%), and D-limonene (6.42%). In parallel, this H.E. is endowed with a significant antifungal action against Candida albicans due to its richness in oxygenated compounds. This could justify its use for the treatment of dermatoses of fungal origin.

Références bibliographiques :

BABA AISSA, F. Encyclopedia of useful plants. Medicinal plants, aromatic plants, food plants. Editions elMaarifa, Algiers, 2011, 471 p.

2/ Encyclopedia of medicinal plants. Identification, preparations, care. 2nd edt. Editions Larousse, Paris, 2001, 335 p.

3/ European Pharmacopoeia, 6th edt. Edition Druckerei C. H. Beck, Nördlingen, 2007, volumes 1 & 2, 3538 p.

4/ GIORDANI, R., HADEF Y., KALOUSTIAN, J. "Compositions and antifungal activities of essential oils of some Algerian aromatic plants". Journal of Fitoterapia n°79, 2008, p 199-203.

DRAFT

POSTER PRESENTATION

Application of remote sensing for the study of the floristic composition at the level of the Middle Oued Righ *(Algeria)*

HADJAIDJI Sawsen[*1] (https://orcid.org/0009-0006-9055-3013) & HADJAIDJI-BENSEGHIER Fatiha[2]

[*1]Kasdi Merbah University, Faculty of Nature and Life Sciences, Department of Biological Sciences, Ouargla ,Algeria.
[2]Kasdi Merbah University, Faculty of Nature and Life Sciences, Department of Agricultural Sciences and Biology, Ouargla ,Algeria

**E-mail: shadjaidji@yahoo.com*

Abstract

The present study took place in the Middle Oued Righ, where we conducted an inventory of plants located in this Saharan area. For this, we used satellite imagery to choose study sites. Followed by floristic surveys ;according to subjective sampling. The identification of vegetation areas in the Middle Oued Righ was conducted through the visual interpretation of the color composition of the spectral bands (blue, green and red) and spectral analysis by N.D.V.I. The results identified 9 stations covering 6 geomorphological sites. Floristically, there are 91 registered plant species. These cover 28 families but the Asteraceae family is the most representative and the Saharo –Sindian element is the most remarkable. The plateaus, the ergs and the hills constitute the most floristically rich geomorphological sites compared to the sebkha, the lake and the wadi in the Middle Oued Righ area.

Keywords: Saharan species, remote sensing, vegetation zone, Middle Oued Righ, Central Northern Sahara.

INTRODUCTION

Algeria from West to East and from North to South is characterized by its great geographical, bioclimatic, geological, geomorphological and edaphic diversity at the origin of a considerable ecosystem and floristic richness. This diversity has aroused the interest of many researchers and work on the organization, dynamics and functioning of forest species that have emerged.

Indeed, in the Algerian South where the Sahara is precisely the northern Sahara, the Saharan vegetation carpet is discontinuous and very irregular; the plants especially use the locations where the water is a little more accessible than elsewhere. The vegetation of the arid zones, in particular those of the Sahara, is very sparse, with a generally bare and desolate appearance, the trees are as rare as scattered and the grasses appear there only for a very brief period of the year, when the climatic conditions become favorable (Quézel, 1965 and Ozenda, 1982)

Currently, the ecological balance that has lasted for centuries is broken due to desertification and climate change. Also, by browsing the ecosystems of the northern Sahara, their apparent homogeneity actually hides a great heterogeneity in detail (Hadjaidji-Benseghier et al., 2017).

The objective of this study is to identify vegetation zones using the remote sensing tool which allows us to acquire more information about a given area. As well as; the identification of the floristic composition of spontaneous plants; populating the ecosystems of the northern Sahara in Algeria (case of the Middle Oued Righ).

DRAFT

MATERIALS AND METHODS

1. Presentation the Middle Oued Righ

The Middle Oued Righ represents the central part of the Oued Righ Valley which consists of 3 zones: Upper, Middle and Lower Oued Righ. This valley is located in the northeast of the Algerian Sahara between the northern latitudes " 32°45'-34°30' " and the longitudes Are "5°45-6°15". It is an elongated depression (gutter-shaped) from south to north along the great Eastern Erg extending over a length of 150 Km and a width of 15 to 30 Km (Figure 1and 2).

The Middle Oued Righ corresponds to the area that extends from the south of Ain Shusha to the North of Tinedla (Figure 2). As, it is known by the palm groves of Sidi-Amrane, Skhouna, Ayata, Tamerna-Djedida, Tamerna-Guedima, Chmarra, Shusha, Djamaa, Ourlana, Tigdidine, Sidi-Yahya, Mazheur, Zaouiat-Riab, El-Arifiane and Tinedla .

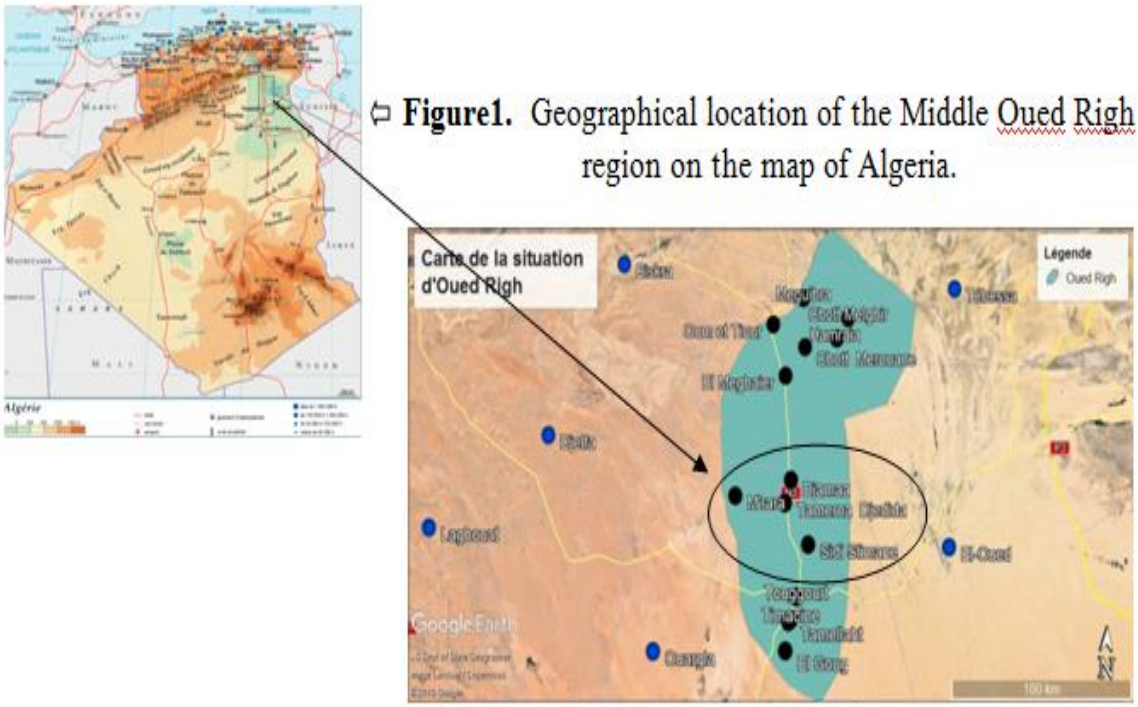


Figure1. Geographical location of the Middle Oued Righ region on the map of Algeria.

Figure 2. Satellite image of the Oued Righ and Middle Oued Righ region (1/1500); (Google Earth,2018)

2. Methodology

Our methodology consists of three stages; which are:

1. Remote sensing prospecting, which involves two phases:

a. Visual analysis: (colored composition)

b. Spectral analysis: (normalized Difference vegetation index: N D V I)

2 .Choice and establishment of the map of the study stations

3. The Floristic analysis (Sampling and Floristic Inventory)

Results

Image processing "SENTINEL-2" by ENVI 4.7 and geo-location of study stations

In order to properly explore the vegetation of the study area, we used satellite imagery. To this end, our investigations concerned (i) the colored composition, (ii) the standardized vegetation index "NDVI" and (iii) topographic maps for the geo-location of the study stations.

1. Interpretation of the colored composition of the "SENTINEL-2" image of the study area

The colored composition of the "SENTINEL-2" image of the Middle Wadi Righ was made from the Blue, Green and Red spectral bands (Figure 4a), because it is the most representative for the study area and the closest to the real colors. Indeed, we can clearly distinguish the different formations such as :

- the vegetation is dark green in color ;
- the dark brown to black depending on the degree of humidity represents the wetlands (wadi and lakes) as well as the canal ;
- the sands and the sandstones represent the colors ranging from beige to light brown respectively ;
- a large area colored in light to dark cyan also showing areas of efflorescence of salt crystals and saliferous crusts depending on the degree of humidity and the level of salinity (dry salty soil and wet salty soil).

2***. Spectral index: normalized Difference vegetation index (NDVI)***

Through this index, a good visualization is obtained for the vegetative zones; of which they appear in white for the strong chlorophyll activities.

The presentation of the NDVI image (Figure4 b) obtained after the analysis of the satellite image of the "Middle Wadi Righ" study area using the ENVI 4.7 software; and comparing with the map of the Middle Wadi Righ region (Fig. 9), shows the following :

- the white color represents the palm trees of Djamâa and M'rara where the vegetation cover is relatively dense ;
- the light gray color which means the dense vegetation cover; it represents the vegetation of the wetlands precisely of Wadi R'tem and lakes Ayata and Djamâa ;
- the less dense vegetation cover is indicated by the gray color. It represents the spontaneous plants that are found at the level of the plateaus and ergs ;
- the dark color represents the absence of vegetation in the depressions.

Figure 3. Map of the Middle Oued Righ region



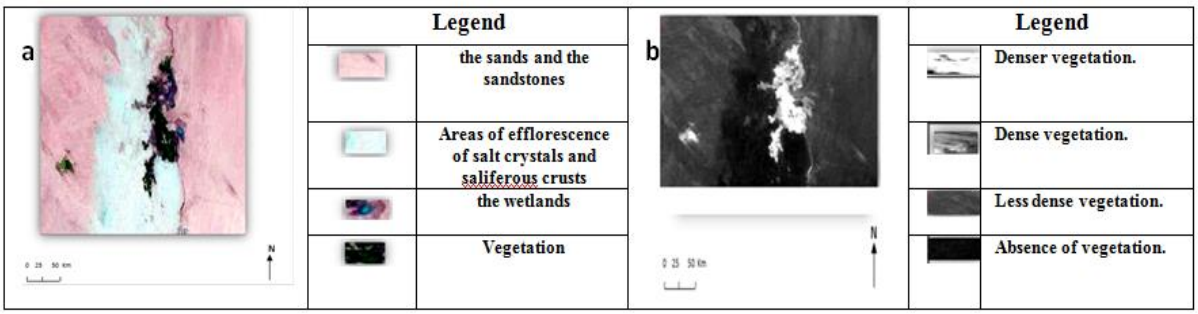


Figure 4.(**a**) Composition colorful from the spectral bands blue,Green and Red image “SANTINEL-2” of the 03/03/2018 and (**b**) the image of the index NDVI of the Middle Oued Righ region.

By using the topographic maps of Ghardaïa and Touggourt as well as ArcGIS version 10.1 software, we were able to establish the sampling map (Figure 5) At the level of the latter, we have geolocated the study stations, which are nine in number, by a field GPS and Google Earth pro.

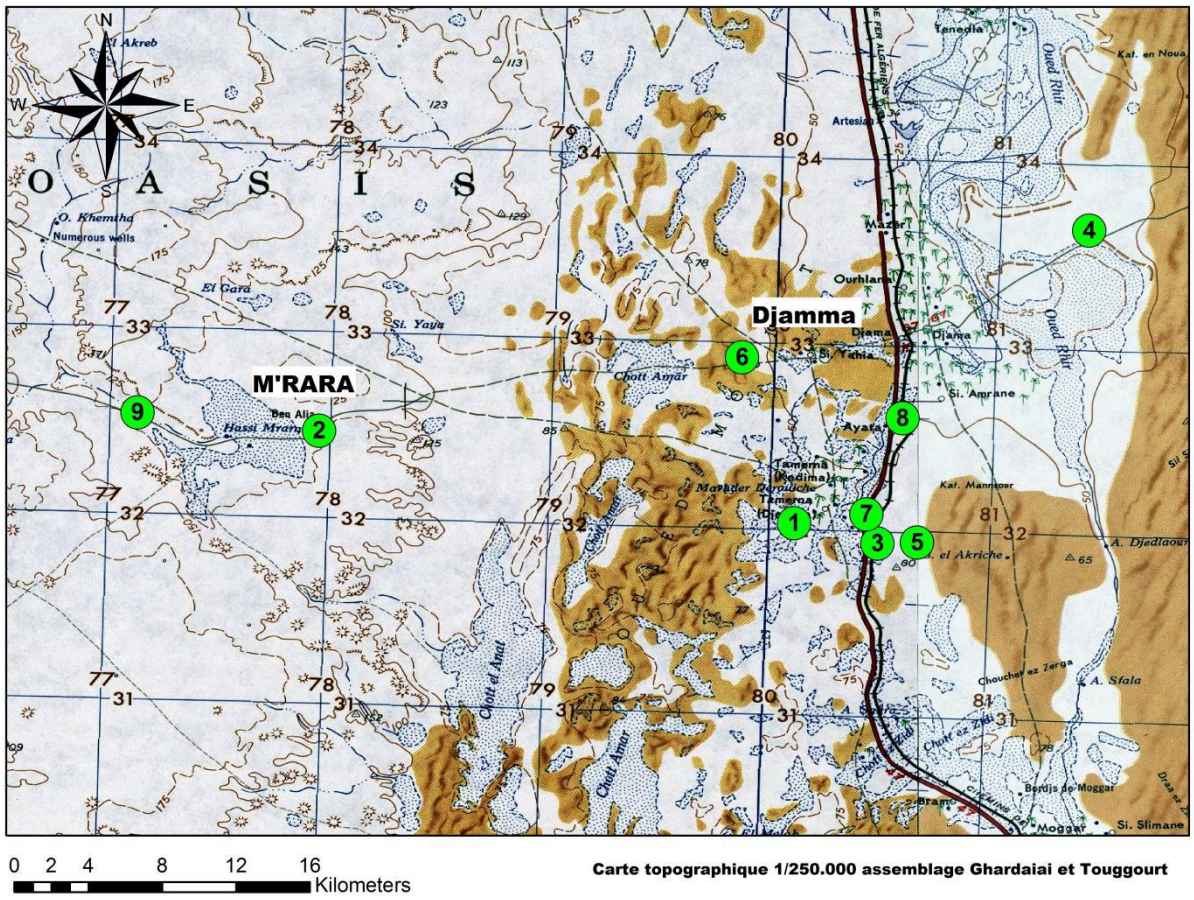


Figure 5. Location of study stations on the topographic map

On this map (Figure 5), we have represented the sites that are potentially floristically rich in the Middle Wadi Righ. This guided us in our approach to floristic study.

Floristic Inventory

The floristic inventory carried out at the level of the Middle Wadi Righ, allowed us to register 91 plant species. These cover 60 genera and 28 families **(**Table1**)**.

Table 1. Species inventoried according to the different families.

***Amaranthaceae* Juss., nom. cons.**	***Asteraceae***	***Boraginaceae***	***Caryophyllaceae***	***Euphorbiaceae***	***Plantaginaceae* Juss., nom. cons.**
Anabasis oropediorum *Maire* ***Cornulaca monacantha Delile*** ***Halocnemum strobilaceum (pall) M. Bied.*** ***Hammada scoparia (Pomel) Iljin*** ***Sarcocornia fruticosa (L.) A.J. Scott*** ***Salsola oppositifolia*** Desf. ***Suaeda vermiculata Forssk. ex J.F. Gmel.*** ***Traganum nudatum Del***	***Atractylis carduus (Forssk.) C. Chr.*** ***Brocchia cinerea (Delile) Vis.*** ***Cotula anthemoides L.*** ***Gymnarrhena micrantha Desf.*** ***Heteromera fuscata*** (Desf.) Pomel ***Ifloga spicata (Forssk.) Sch. Bip*** ***Launaea angustifolia (Desf.) Kuntze*** ***Launaea capitata (Spreng.) Dandy*** ***Launaea capitata Sp.*** ***Launaea mucronata (Forssk.) Muschl.*** ***Launaea nudicaulis*** (L.) Hook. f. ***Launaea quercifolia*** (Desf.) Pamp. ***Launaea fragilis (Asso) Pau*** ***Launaea fragilis sp*** ***Picris asplenioides L.*** ***Picris sp***	***Arnebia decumbens (Vent.) Coss. & Kralik*** ***Echium horridum Batt.*** ***Echium humile*** Desf. ***Echium humile subsp. pycnanthum (Pomel) Greuter & Burdet*** ***Echium trygorrhizum Pomel*** ***Moltkiopsis ciliata (Forssk.) Johnst*** ***Ogastemma pusillum (Bonnet & Barratte) Brummitt***	***Herniaria fontanesii J. Gay*** ***Paronychia arabica (L.) DC.*** ***Polycarpaea prostrata*** ***Polycarpaea repens (Del.) Asch et schw*** ***Pteranthus dichotomus Forssk***	***Euphorbia guyoniana Boiss. & Reut*** ***Euphorbia calyptrate Coss.& Kralik*** ***Euphorbia retusa*** Forssk.	***Plantago ciliata*** Desf.
					Plumbaginaceae
					Limoniastrum guyonianum Coss & Dur
					Poaceae
			***Cistaceae* Juss., nom. cons.**	***Fabaceae* Lindl., nom. cons.** *(= Leguminosae Juss., nom. cons.)*	***Centropodia forskalii (Vahl) Cope*** ***Phragmites communis Trin*** ***Panicum turgidum Forssk*** ***Stipagrostis plumosa (L.) Munro ex T. Anderson*** ***Stipagrostis obtusa (Del .) Nees .*** ***Stipagrostis pungens (Desf.) De Wint***
			Helianthemum getulum ***Helianthemum lippii (L.) Dum. Cours***	***Astragalus caprinus L*** ***Astragalus cruciatus Link.*** ***Astragalus gomboëformis Pomel*** ***Astragalus gyzensis*** *Bunge.* ***Astragalus mareoticus Delile*** ***Genista saharae Coss &Dur*** ***Retama raetam*** *(Forssk.) Webb*	
			Colchicaceae		***Rubiaceae***
			Colchicum wyssianum (Beauverd & Turrett.) J.C. Manning & Vinn.		***Crucianella hirta*** *Pomel*
		Brassicaceae	***Convolvulaceae***	***Geraniaceae* Juss., nom. cons.**	***Tamaricaceae***
					Tamarix aphylla ***Tamarix gallica sp***
		Eruca vesicaria (L.) Thell ***Eremobium aegyptiacum (Spreng.) Hochr.*** ***Eremobium aegyptiacum*** subsp. ***longisiliquum*** (Coss.) Maire ***Matthiola longipetala*** subsp. ***livida*** (Delile) Maire ***Henophyton deserti (Coss. & Durieu)*** ***Pseuderucaria clavata (Boiss. & Reut.) O.E. Schulz*** ***Pseuderucaria teretifolia (Desf.) O.E. Schulz*** ***Savignya parviflora subsp. longistyla (Boiss. & Reut.) Maire*** ***Savigna sp.*** ***Zilla spinosa subsp. macroptera (Coss.) Maire & Weiller*** ***Zilla sp.***	***Cressa cretica* L.**	***Erodium glaucophyllum*** *(L.) L'Her.* ***Monsonia heliotropioides*** *(Cav.) Boiss.* ***Monsonia nivea****(Decne.) Webb.*	***Thymelaeaceae***
			***Cucurbitaceae* Juss., nom. cons.**		***Thymelaea microphylla*** *Coss. & Durieu ex Meisn.*
Apiaceae	***Asparagaceae* Juss., nom. cons.**		***Colocynthis vulgaris*** Schrad.,	***Juncaceae***	***Zygophyllaceae*** R.Br., nom. Cons
Eryngium iliciflolium (Lam.)	***Drimia noctiflora*** *(Batt. & Trab.) Stearn*		***Cyperaceae* Juss., nom. Cons .**	***Juncus maritimus Lam.***	***Fagonia latifolia*** ***Fagonia microphylla Pomel*** ***Zygophyllum album L = Tetraena alba*** *(L. f.) Beier & Thulin* ***Zygophyllum album L = Tetraena gaetula*** *(L. f.) Beier & Thulin* ***Zygophyllum cornutum Coss.***
	Asphodelaceae		***Cyperus conglomeratus*** Rottb.	***Neuradaceae* Kostel., nom. Cons**	
Arecaceae	***Asphodelus refractus Boiss*** ***Asphodelus tenuifolius Cav***			***Neurada procumbens*** *L.*	
				***Nitrariaceae* Lindl .**	
Phoenix dactylifera L.				***Nitraria retusa*** (Forssk.) Asch.	
				Orobanchaceae	
				Cistanche phelypaea (L.) Cout.	

The rates of these families vary widely (Figure 6). Among the most abundant families, we note in descending order, the Asteraceae family with 17.58% (16 species), the Brassicaceae with 12.09% (11 species) and the Amaranthaceae with 8.79% (08 species).

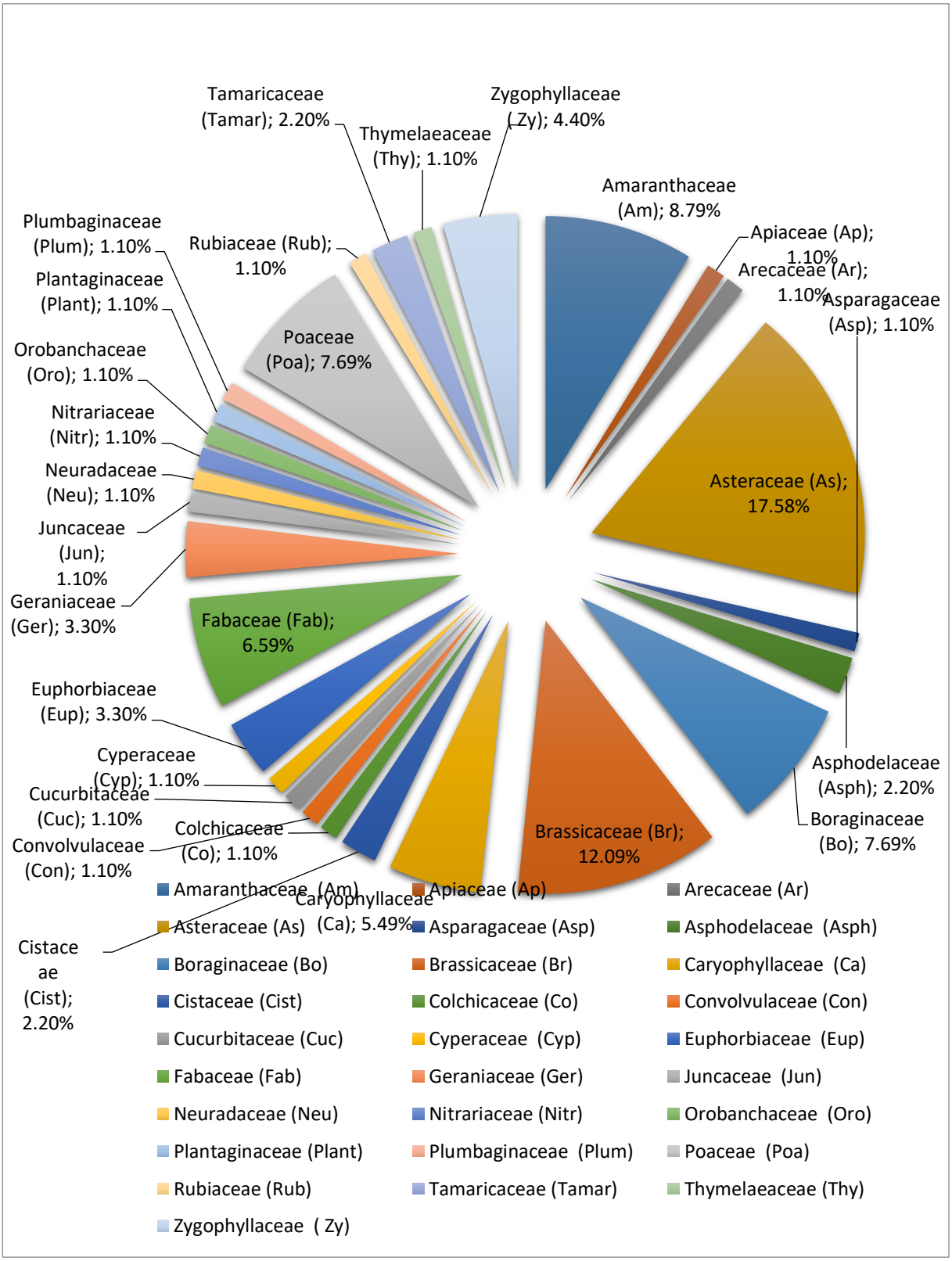


Figure6 . Proportion of families listed the Middle Oued Righ

The distribution of species according to the sites shows that it is variable. Indeed, we note in the most diversified site the plateau 21 families and 66 species; of which the dominant family is that of the Asteraceae with 10 species which are Brocchia cinerea (Delile) Vis. ,Cotula anthemoides L., Heteromera fuscata (Desf.) Pomel, Ifloga spicata (Forssk.) Sch. Bip, Launaea angustifolia (Desf.) Kuntze, Launaea capitata (Spreng.) Dandy, Launaea capitata Sp., Launaea mucronata (Forssk.) Muschl., Launaea quercifolia (Desf.) Pamp.,

Launaea fragilis (Asso) Pau ; followed by the Erg with 16 families and 53 species; of which the dominant family is that of the Asteraceae with 9 species which are Atractylis carduus (Forssk.) V. Chr., Brocchia cinerea (Delile) Vis. , Cotula anthemoides L., Launaea capitata (Spreng.) Dandy, Launaea nudicaulis (L.) Hook. F., Launaea fragilis (Asso) Pau , Launaea fragilis sp , Picris asplenioides L. and Picris sp;and the hills with 13 families and 25 species; of which the dominant family is that of the Boraginaceae with 6 species which are Echium horridum Batt. , Echium humile Desf. , Echium humile subsp. pycnanthum (Pomel) Greuter & Burdet, Echium trygorrhizum Pomel, Moltkiopsis ciliata (Forssk.) Johnst and Ogastemma pusillum (Bonnet & Barratte) Brummitt. . While the sites of Wadi, sebkha and the lake are the poorest in species. With 13 families and 16 species in the Wadi, of which the Amaranthaceae, Asteraceae and Tamaricaceae families are equal; 4 families and 4 species at the sebkha which are Amaranthaceae (Halocnemum strobilaceum (pall) M. Bied.); Juncaceae (Juncus maritimus Lam.); Poaceae (Phragmites communis Trin) and Tamaricaceae (Tamarix gallica sp) and the lake site with 6 families and 9 species of which the dominant family is that of the Amaranthaceae with 4 species which are Halocnemum strobilaceum (pall) M. Bied. , Sarcocornia fruticosa (L.) A.J. Scott , Suaeda vermiculata Forssk. ex J.F. Gmel. , Traganum nudatum Del.

The phytogeographic spectrum shows a very significant diversity of phytogeographic elements in the study region .Indeed, the Saharo -Sindian element dominates the Wadi Righ area with 34 species, and the endemic element comes in second position with 31 species ahead of the Mediterranean element which includes with 17 species.

DISCUSSION

The use of remote sensing through the "SENTINEL-2" image of the Middle Wadi Righ, led us to better explore the study area. Indeed, It appears that through the colored composition, we can observe that the cultivated plants (palm trees) in dark green color; whereas the spontaneous plants are not observed because of their low spectral reflectance, which poses a disadvantage. However, the spectral analysis of the satellite image by the normalized vegetation index (NDVI) makes it possible to obtain a good visualization for the vegetated areas of the study area. It appears in white for the strong chlorophyll activities.

However, it should be noted that the presentation of the NDVI image applied to the Wadi Righ Medium shows that there is a low spectral reflectance in the middle of the study area compared to these two East and West sides. This probably comes back to the high salinity levels characterizing the center of the study area. Mougenot B. (1993) demonstrated that when the salt level is high, it negatively affects the reflectance of vegetation.

The floristic inventory has allowed us to draw up a list of 91 plant species, covering 28 families where the Asteraceae family is predominant. The plateaus, the ergs and the hills constitute the most floristically rich geomorphological sites compared to the sebkha, the lake and the wadi in the Middle Wadi Righ area. Thus, our results are in accordance with those of Kherraze M.E., et al. (2010). However, in the Ouargla region (Chehma et al., 2005) as well as in the Central Sahara (Benhouhou and Benghanem, 2013), it is rather the wadis, lakes and dry valleys that are the most floristically rich. This seems obvious given the degree of aridity (Benseghier-Hadjaidji, 2018).

The phytogeographic spectrum shows a very significant diversity of phytogeographic elements in the study region. Indeed, the Saharo -Sindian element dominates the Wadi Righ area with 34 species, and the endemic element comes in second position with 31 species ahead of the Mediterranean element which includes with 17 species. It should be noted that most of the sampled species live in isolated areas. The distribution of spontaneous flora in this arid zone is usually due to chance it remains, however, rarely contagious. Indeed, no species adopts a regular distribution because of the heterogeneity of the environment (BAAMEUR, 2021).

Conclusion

The present study focused on the application of remote sensing for the study of the floristic composition at the level of the Middle Oued Righ which is located in the central northern Sahara. Indeed, this study made it possible to conclude the following:

The identification of the vegetation zones in the study area was carried out thanks to the visual interpretation of the colored composition of the spectral bands (blue, green and red) as well as to the spectral analysis by N.D.V.I. The results obtained highlight 6 geomorphological sites covering 9 stations.

The floristic analysis allowed us to identify 91 plant species, covering 28 families but the Asteraceae family is the most representative and the Saharo –Sindian element is the most remarkable. The plateaus, the ergs and the hills constitute the most floristically rich geomorphological sites compared to the sebkha, the lake and the wadi in the Middle Oued Righ area.

REFERENCES

BAAMEUR malika ,2021, Étude écologique de la flore spontanée du Sahara septentrional est Algérien , Résumé, THESE Pour l'obtention du diplôme de Doctorat En Sciences Agronomiques UNIVERSITE KASDI MERBAH – OUARGLA ,Algeria

Benseghier-Hadjaidji F., Talbi N., and Derridj A ., 2018. Contribution to the study of the zonal variation of the climate aridity in central northern Sahara (Algeria). AIP (AmericanInstitute of Physics) Conference Proceedings 1968, 030066 : 1-8 (2018) ; https://doi.org/10.1063/1.5039253

Chehma A., Djebar M.R., Hadjaidji F. et Rouabeh L., 2005. Étude floristique spatio-temporelle des parcours sahariens du Sud-Est algérien. Sécheresse ; 16 (4) : 275-85;

-Djellouli Y. et Daget Ph., 1987, Climat et flore dans les steppes du sud-ouestalgérien ,Bull. Soc. bot. Fr., 134, Lettres bot., 1987 (4/5), 375-384.

Hadjaidji-Benseghier Fatiha, Talbi Nadjib and Derridj Arezki, 2017. Did the global warming confirm in central northern Sahara (case of the region of Ouargla)? Elsevier. Energy Procedia, 119 : 852-862.

Kherraze M.E., Lakhdari K., Kherfi Y., Benzaoui T., Berroussi S., Bouhanna M., Sabaa A. 2010. Atlas floristique de la vallée de l'Oued Righ par écosystème. C.R.S.T.R.A. Guerfa. Biskra (Algérie). 91p.

Mougenot B., 1993. Effets des sels sur la réflectance et télédétection des sols salés, *Antenne de télédétection Orstom, Centre Agrymet, BP11416, Niamey, Niger*, Cah. *Orstom, sér, Pédol., vol. XXVIII, no 1, 45-54p,*

-S. BENHOUHOU et A. N BENGHANEM (2013), Flore et végétation du Sahara algérien : synthèse et perspectives de recherche, colloque scientifique :le Sahara impacts de changements environnementaux extrêmes sur la biodiversité ,p.23

POSTER PRESENTATION

Quality assessment of Shkumbin river water based on physico-chemical parameters.

Lirim Bekteshi[1*] (*ORCID: https://orcid.org/0009-0005-3280-1253*), Belinda Hoxha[2], Nikolin Gega[3], Piro Karamelo[4], Anxhela Dauti[5].

[*1]University of Elbasan, Faculty of Natural Sciences, Department of Chemistry, Elbasan, Albania.
[2,3,4,5] University of Elbasan, Faculty of Natural Sciences, Department of Chemistry, Ebasan, Albania.

*Corresponding author e-mail:lirimbekteshi@ymail.com

Abstract

The increase in demands for well-being, especially in recent decades, has led to an intensive use of land but also to an increase in industrial production. Both of these factors are related to the use of large amounts of water, therefore taking measures to have sufficient and quality water suitable for human and industrial consumption but also for agriculture (irrigation) is part of environmental policies. all over the world.
In this paper, a quality assessment of the water of the river Shkumbin, Albania, based on the physical and chemical parameters, has been carried out. In the study, the physico-chemical parameters of eight water samples, taken along the course of the river, were analyzed. The analyzed parameters are ten: pH. EC, T, DO, TDS, Nitrite (NO_2^-), Nitrate (NO_3^-), Ammonia (NH_4^+), salinity, and phosphates.PO_4^{-3}. The obtained data were subjected to statistical processing, with the aim of determining the polluted areas and sources of pollution. The watershed of the Shkumbin river is located in central Albania, and the sources of pollution are anthropogenic related to agricultural and industrial activity, but also natural related to the rock formations that lie in this basin and erosion.
The evaluation of the water quality of the river Shkumbin was carried out by comparing our parameters with those according to the WHO.

Keywords: pollution, water quality, physico-chemical parameters, irrigation, anthropogenic activity.

INTRODUCTION

Rivers and lakes are important sources of surface water. Albania is rich in many rivers and lakes which are used both for irrigation and fishing (Dreschel and Evans, 2010). The early urban centers were created by humans near the rivers, where river waters have been used for cleaning and other domestic purposes, as there were also the conditions for providing food (M.E.A. 2005). Rivers make up 1% of the planet's fresh water. In modern times, the industrial development and urbanization of society has led to the creation of large industrial and urban centers as well as the development of intensive agriculture. This has led to an increase in the need for fresh water, but at the same time also the pollution of river waters. The main sources of pollution are the anthropogenic ones related to industrial activity or urban discharges. Industrial waste and from excessive human activities that affect their physico-chemical characteristics which result in harmful effects on aquatic organisms.(Murhekar, 2011)(Vassilis et al., 2001). The discharge, in a continuous manner, into the waters of the rivers of the sewage of untreated cities, is the main problem of underdeveloped and developing countries (Singh, 2007). At present, more than 80% of the sewage generated in developing countries is discharged untreated into the environment, and about 50% of the population depends on polluted water sources for various uses, including irrigation (UNESCO 2003). Another potential polluter remains agriculture as a result of the use of fertilizers and pesticides, but also the discharges of livestock farms (Nahar et al., 2016). Water is the main component of living organisms, so their survival depends on water quality (Almeida et al., 2017). The assessment of water quality includes the assessment of its physio-chemical and biological parameters. Changes in parameters such as pH, salinity, DO, temperature, significantly affect the life of the water body (Kolawole et al., 2011)(Smitha et al., 2007). So the evaluation of the physico-chemical parameters gives us information about the pollution of the water body, the further study of this pollution is carried out with biological evaluations (Mali and Shumka 2012).
Natural pollution is related to the erosion of the soil on the banks of the rivers, but also from the rains, as well as the nature of the rock formations of the catchment area of the rivers. The nature of the mineralogical composition of the rocks of the watershed affects the values of parameters such as pH, EC or TDS. (Bucher et al, 2017).

DRAFT

In general, the parameters of surface bridges change according to the seasons, dry or wet, but also the change of agricultural activity in different seasons (Uddin et al., 2016)

MATERIALS AND METHODS

Study area:
Shkumbin River is one of the main rivers of Albania with a length of 181.4 km and a catchment area of 2445 km (fig 1) and an average height of the water flow of 753.2 m. This shows that the Shkumbin river is a fast-flowing river, as a result, in the rainy season, it causes high erosion, causing its pollution.

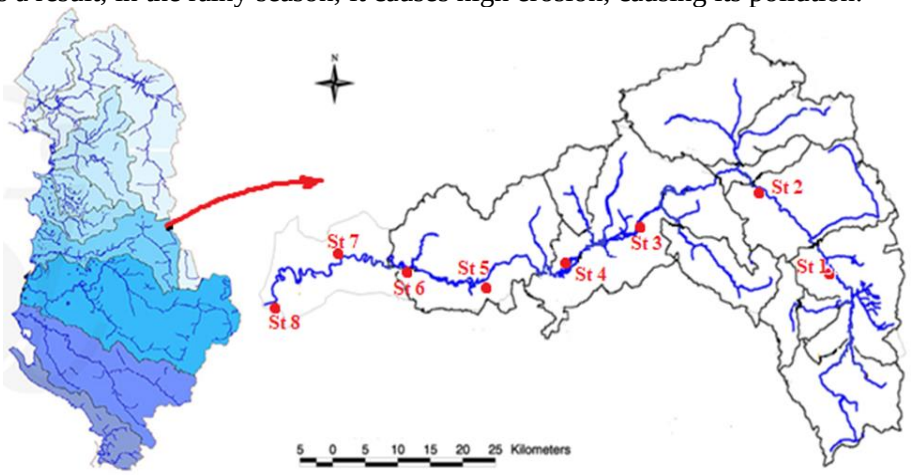


Figure 1. Shkumbin river catchment basin.

The river flows fluctuate a lot depending on the seasonality, the annual mean of the flows is 61.5 m/s and the total annual amount of sediments $7.2 \cdot 10^6$ tons/year (Pano et al.,1984)
Sampling: Samples were taken during the month of June 2023, almost along the entire length of the river at 8 sampling stations (fig1). Water sampling was done with 0.5 liter plastic bottles. The bottles were first washed with distilled water and then rinsed with 10 ml of 2% HCl. Before being filled, the bottles were again washed several times with river water and filled to the brim with the water sample, eliminating the existence of air bubbles in the bottles. and are closed with a safe cork, providing them with the appropriate label. Afterwards samples were transported to the lab within the same day using cooling boxes to keep the temperature of +4°C. The samples were stored under standard conditions till the analysis. Temperature, pH, conductivity (CE), dissolved solids (TDS), salinity and dissolved oxygen (DO) was measured directly with camera multimeter Hach (Sension 156). The concentration of nutrients (NO_2^-, NO_3^-, NH_4^+, PO_4^{3-}) was performed with UV– VIS spectrophotometer PYE UNICAM SP6 - 550, respectively at 543nm, 220 - 270nm, 630nm and 880nm.

RESULTS and DISCUSSION

The values of the ten parameters obtained from the analysis of 8 water samples of the Shkumbin River are summarized in a data matrix and descriptive statistics were used to interpret the results and to explain the data variations. The values of the statistical processing of the data are presented in table 1.
As can be seen from table 1, the median values for the physico-chemical parameters in the Shkumbin river are within the values recommended by WHO.

Table 1. Descriptive Statistics: pH, Temp, EC, DO, TDS, NO_2^-, NO_3^-, NH_4^-, PO_4^{-3}, Salinity.

Variable	Unit	Mean	StDev	CoefVar%	Minimum	Median	Maximum	WHO
pH		8.1675	0.2449	3.00	7.7400	8.1900	8.5500	6.5-8.5
Temp	^{0}C	21.362	2.356	11.03	18.500	20.550	24.700	<40
EC	uS/cm	351.13	26.05	7.42	329.00	341.00	398.00	750
DO	mg/L	9.238	1.060	11.47	7.900	9.100	10.800	> 5
TDS	mg/L	157.69	11.75	7.45	140.40	162.85	171.30	1000
NO_2^-	mg/L	0.03275	0.01597	48.77	0.00600	0.03550	0.05200	< 3
NO_3^-	mg/L	1.526	0.324	21.21	0.810	1.560	1.880	50
NH_4^+	mg/L	0.2437	0.1462	59.97	0.0500	0.2200	0.5400	0.5
PO_4^{-3}	mg/L	0.02500	0.01742	69.68	0.00500	0.02400	0.05700	0.3
Salinity	mg/L	0.06525	0.02307	35.36	0.04500	0.05700	0.11000	250

The coefficient of variation (CV) is the ratio of the standard deviation to the mean. The higher the coefficient of variation, the greater the level of dispersion around the mean. The high CV values of some parameters such as salinity, NH_4^+, PO_4^{-3}, NO_3^- show that we have an anthropogenic impact on the river water quality. The parameters for which the CV changes a little, that is, in the stations along the course of the river there are no significant differences, the anthropogenic influences are smaller.

Fig. 2 shows the probability of occurrence of the values of the studied parameters. The slope of the graphs for parameters such as NO_2^- NH_4^+ PO_4^{-3} shows the possibility of their occurrence in a wider range of concentrations. While this fact is less pronounced in parameters such as NO_3^-, salinity and very little in the parameters pH, DO, EC, T and TDS.

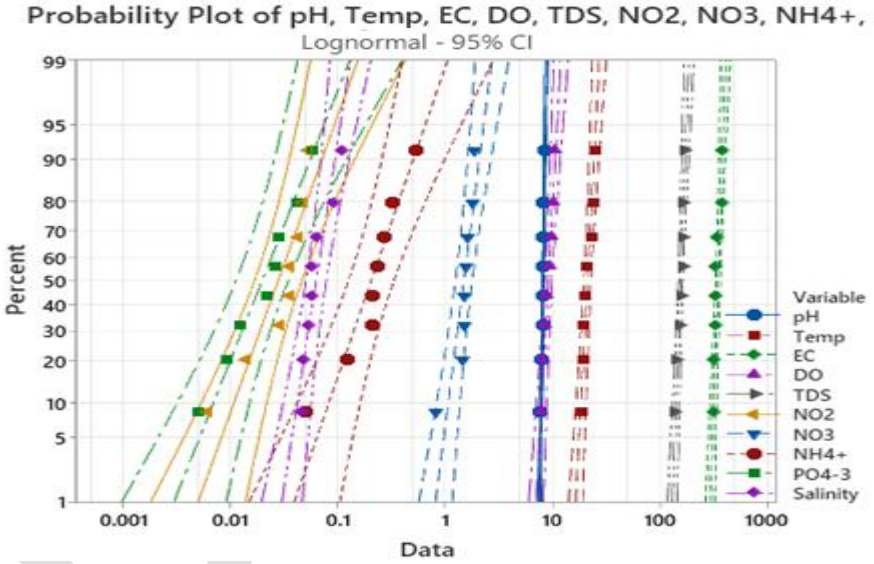


Figure 2. The probability of distribution of the values of the studied parameters along the stations.

Correlation analysis was carried out to distinguish the parameters with similar behaviors in the water body. The results of the correlation analysis are shown in Table 2.

As can be seen from the table, pH shows a high correlation with TDS, which is related to the presence of Ca^{+2} and Mg^{+2} metal ions as well as CO_3^- and HCO_3^- anions, which are the result of the action of river water with limestone rocks, which are the main part of the mineralogical composition of the basin watershed

Regarding the temperature parameter, a strong correlation can be seen with EC and salinity, but also with ion concentrations, since temperature affects their solubility.

Dissolved oxygen (DO) refers to the amount of oxygen gas present in water and is essential for aquatic life. All types of life in water, microorganisms, algae, fish, require oxygen for the development of their vital activity. There is a negative correlation with temperature, since the increase in temperature affects the amount of DO. Also, have a strong negative correlation with NO_2^- ions, which consume DO as a result of their oxidation to NO_3^-. There is also a negative correlation with NH_4^+ and PO_4^{-3} ions, which are related to the processes of decomposition of biological materials and sewage discharges.

The strong correlation of these four parameters NH_4^+ PO_4^{-3} NO_2^- NO_3^- shows that their source is the same and is related to urban discharges or livestock farms in the waters of the Shkumbin River.

Table 2. Pearson correlation coefficient between: pH, Temp, EC, DO, TDS, NO_2^-, NO_3^-, NH_4^+, PO_4^{-3}, Salinity.

	pH	**Temp**	**EC**	**DO**	**TDS**	**NO_2^-**	**NO_3^-**	**NH_4^+**	**PO_4^{-3}**
Temp	-0.301								
EC	-0.087	0.715							
DO	-0.238	-0.684	-0.415						
TDS	0.823	-0.109	-0.215	-0.493					
NO_2^-	0.044	0.538	0.504	-0.702	0.002				
NO_3^-	-0.107	0.505	0.417	-0.603	-0.156	0.910			
NH_4^+	-0.174	0.525	0.545	-0.382	-0.337	0.893	0.835		
PO_4^{-3}	0.081	0.486	0.513	-0.461	-0.122	0.906	0.803	0.963	
Salinity	-0.050	0.699	0.994	-0.450	-0.161	0.486	0.407	0.486	0.460

Some of the pairs of parameters with high positive or negative correlation are presented in table 3. (P-Value <0.05).

Table 3. Pairwise Pearson Correlations

Sample 1	**Sample 2**	**N**	**Correlation**	**95% CI for ρ**	**P-Value**
TDS	pH	8	0.823	(0.281, 0.967)	0.012
EC	Temp	8	0.715	(0.021, 0.944)	0.046
DO	Temp	8	-0.684	(-0.937, 0.040)	0.061
Salinity	Temp	8	0.699	(-0.010, 0.941)	0.053
Salinity	EC	8	0.994	(0.967, 0.999)	0.000
NO_2^-	DO	8	-0.702	(-0.941, 0.006)	0.052
NO_3^-	NO_2^-	8	0.910	(0.573, 0.984)	0.002
NH_4^+	NO_2^-	8	0.893	(0.509, 0.981)	0.003
PO_4^{-3}	NO_2^-	8	0.906	(0.556, 0.983)	0.002
NH_4^+	NO_3^-	8	0.835	(0.317, 0.969)	0.010
PO_4^{-3}	NO_3^-	8	0.803	(0.227, 0.963)	0.016
PO_4^{-3}	NH_4^+	8	0.963	(0.802, 0.993)	0.000

Fig. 3 shows the correlogram of the measured parameters, where it is clearly shown how the parameters correlate between them.

Table 3 shows the pairs of parameters that have a high negative or positive correlation (for values of P<0.05). This can be found in the correlogram, bearing in mind that pairs of parameters with high negative correlation are presented in strong blue color, while pairs of parameters with high positive correlation are presented without color (parameter pair Salinity, EC). Intermediate correlations based on level are shown in corresponding colors.

DRAFT

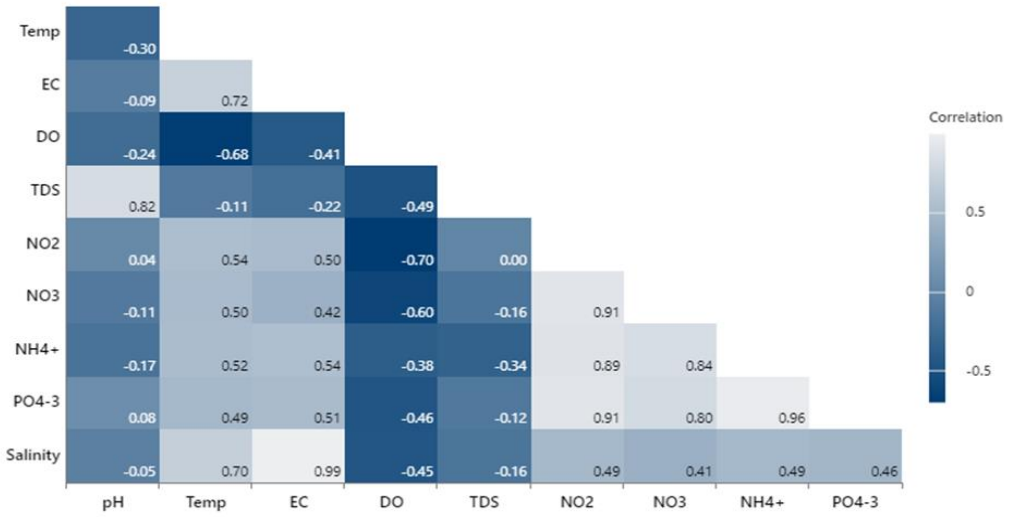


Figure 3. Correlogram of pH, Temp, EC, DO, TDS, NO_2^-, NO_3^-, NH_4^+, PO_4^{-3}, Salinity

Multivariable analysis (Cluster Analysis)
Through the computer program Minitab (Cluster method of data grouping) the dendograms of grouping of parameters based on the correlations between them were studied (Fig 4).

Cluster Analysis of Variables: pH, Temp, EC, DO, TDS, NO_2^-, NO_3^-, NH_4^+, PO_4^{-3}, Salinity
Correlation Coefficient Distance, Complete Linkage
Amalgamation Steps

Step	Number of clusters	Similarity level	Distance level	Clusters joined		New cluster	Number of obs. in new cluster
1	9	99.7110	0.00578	3	10	3	2
2	8	98.1352	0.03730	8	9	8	2
3	7	95.5114	0.08977	6	7	6	2
4	6	91.1440	0.17712	1	5	1	2
5	5	90.1664	0.19667	6	8	6	4
6	4	84.9743	0.30051	2	3	2	3
7	3	70.3599	0.59280	2	6	2	7
8	2	33.1733	1.33653	1	2	1	9
9	1	14.9131	1.70174	1	4	1	10

Final Partition
Cluster 1 pH TDS
Cluster 2 Temp EC NO_2^- NO_3^- NH_4^+ PO_4^{-3} Salinity
Cluster 3 DO

From the dendogram it can be seen that the parameters have similar values according to the stations forming the dendogram with 9 groups. In the first group, the parameters EC and salinity present high similarity of values between stations (similarity about 99%) which have 84% similarity with temperature. All three of these parameters influence each other, forming a set of parameters. EC is related to both temperature and salinity. High salinity values at station 8 have led to an increase in EC values. This happened because station 8 is located at the mouth of the river Shkumbin and the salinity of the sea water has an influence. Regarding the temperature, temperature fluctuations in the stations have led to EC fluctuations.
The second group of parameters consists of 2 subgroups and is related to the parameters NO_2^-, NO_3^-, which present 95% similarity and the parameters NH4, PO4, which have 98% similarity. These two subgroups have a distribution similarity according to the stations of about 91%, forming a special group.

These two groups with 70% similarity to each other, form the main part of the dendogram, which has a small similarity (about 33%) with the third group of parameters pH, TDS.
pH and TDS have similar values in stations around 84.9%, which means that TDS values have an impact on pH values. The first two groups and the third group of parameters form the large group where the similarity of the parameter values in the stations is about 33%
DO in the dendogram is presented as a single group since the values of DO are not similar to those of other parameters, the similarity in this case is 14.9%.

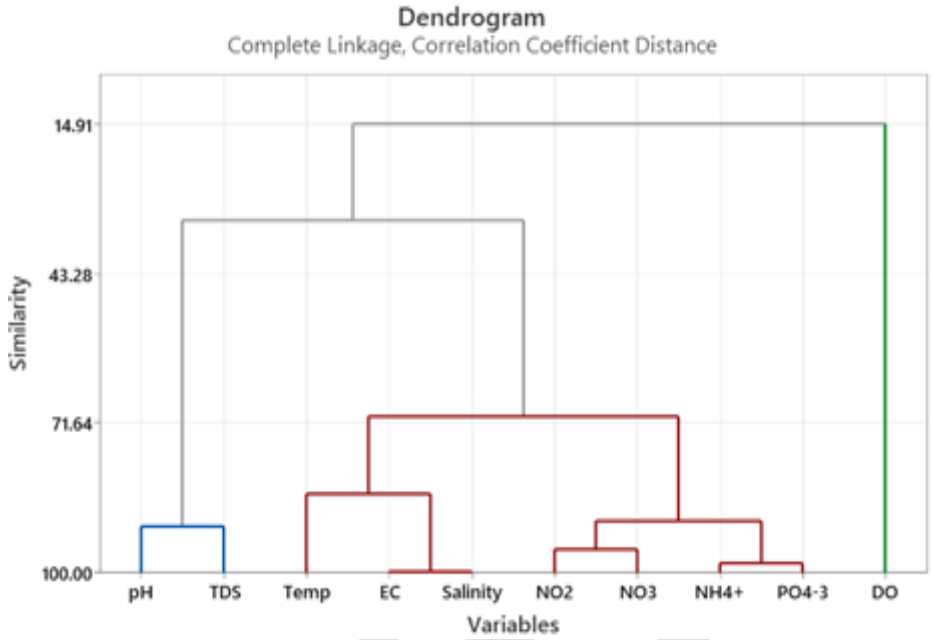


Figure 4. The dendrogram of cluster analysis of variables.

Cluster Analysis of Observations: pH, Temp, EC, DO, TDS, NO_2^-, NO_3^-, NH_4^+, PO_4^{-3}, Salinity
Standardized Variables, Euclidean Distance, Complete Linkage
Amalgamation Steps.

Step	Number of clusters	Similarity level	Distance level	Clusters joined		New cluster	Number of obs. in new cluster
1	7	78.1824	1.68921	5	6	5	2
2	6	69.8327	2.33569	2	3	2	2
3	5	59.7123	3.11925	5	7	5	3
4	4	44.7339	4.27894	5	8	5	4
5	3	44.4440	4.30140	1	2	1	3
6	2	36.0178	4.95379	4	5	4	5
7	1	0.0000	7.74245	1	4	1	8

Distances Between Cluster Centroids

	Cluster1	Cluster2	Cluster3	Cluster4	Cluster5
Cluster1	0.00000	3.28825	7.74245	4.81915	6.25929
Cluster2	3.28825	0.00000	5.32777	3.47958	4.95406
Cluster3	7.74245	5.32777	0.00000	4.64927	4.43376
Cluster4	4.81915	3.47958	4.64927	0.00000	3.78391
Cluster5	6.25929	4.95406	4.43376	3.78391	0.00000

In order to understand the relationship between the parameters in different stations and to evaluate the sources of change in these parameters, a Cluster Analysis of Observations was carried out. As can be seen from the dendogram in Fig. 5, the stations, based on the parameter values, are divided into seven groups, which consist of two large groups, one with 5 stations and the other with 3 stations.

In the large group, with 5 stations, we distinguish 4 sub-groups. Stations 5 and 6 form a group with a similarity level of 78%, which are connected to station 7, which represents a single group, with a similarity level of 59.7%. All these stations belong to the field area and the contributors to the parameter values are the anthropogenic ones related to urban and agricultural and livestock activity. The discharges of untreated water from the villages and the use of pesticides, chemical fertilizers and the discharges of livestock farms, make the parameters in stations 5 and 6 not differ much from each other, while the similarity is the lowest in station 7, expressing an impact with lower anthropogenic, as the area is hilly and less populated.

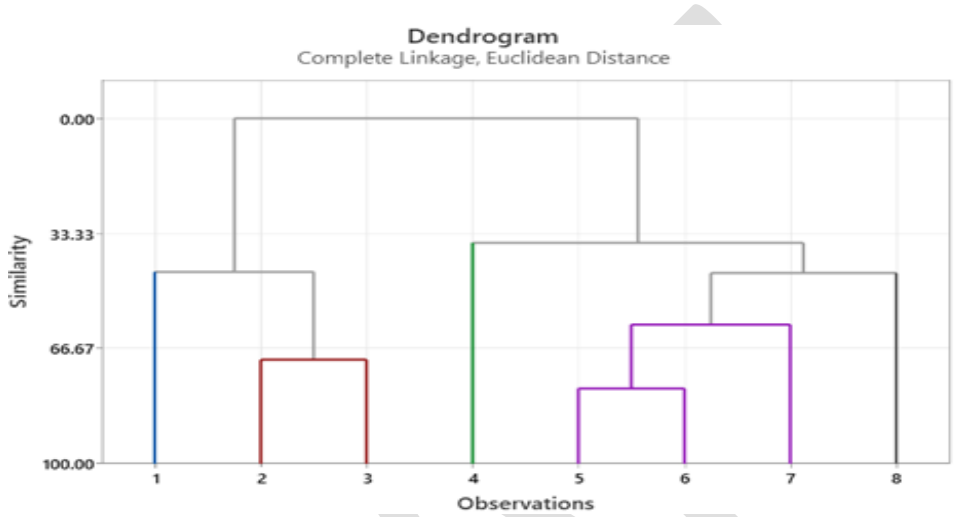


Figure 5. Dendogram of similarity between monitoring stations.

Station 8 is presented alone, and the similarity with the previous stations drops to 44.4%, due to the salinization of the river water from the sea, since this station is located in the Shkumbin river delta.

Station 4 is presented as a single group, and has about 36% similarity with stations 5,6,7,8. This station expresses the anthropogenic effect on the river water related to urban activity. The discharge of sewage from the city of Elbasan, untreated in advance, into the waters of the Shkumbin River presents increased values of nutrients and EC and a decrease in DO.

Regarding the other group of stations (stations 1, 2, 3.), we can say that station 1, being far from residential centers and in an area with traditional agriculture, appears clean, therefore it forms a special group in the dendogram. While stations 2 and 3, which have 69.8% similarity in the values of their parameters, are located after the town of Librazhdi, which is a small town and has a small effect on river pollution. But in these stations, erosion plays a role, since the river in these stations comes after a dislevelment of about 400 m with the first station and is erosive. These three stations form a common group with 45% similarity with the first station.

A presentation of the values of the physico-chemical parameters according to the stations is presented in figure 6

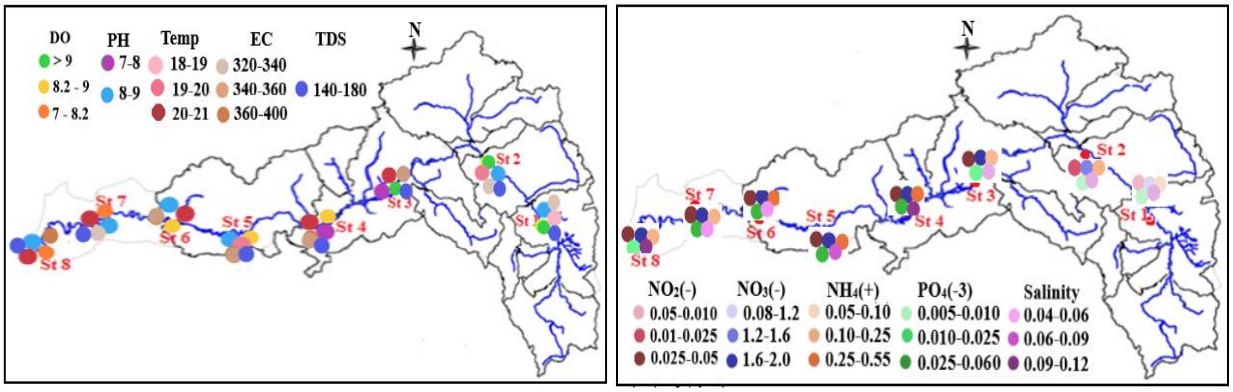


Figure 6. Values of physico-chemical parameters according to stations

CONCLUSION

From the evaluation of the physico-chemical parameters and their comparison with the values recommended by WHO, we see that the physico-chemical parameters of the Shkumbin river water are within the recommended values. Some parameters are within the limit values, especially in station 4, which is related to the urban discharges of the city of Elbasan or discharges from livestock farms. Therefore, the establishment of a plant for the treatment of the city's sewage, as well as the control of the waste discharge of livestock farms, would help to eliminate these potential pollutants of the river waters.

REFERENCES

Almeida C A, Quintar S, Gonzalez P, Mallea M A 2007. Influence of urbanization and tourist activities on the water quality of the Portero de los Funes River (San Luis – Argentina). Environment Monitoring and Assessment. 133: 459 – 465.

Bucher K, Zhou W, Stober I 2017. Rocks control the chemical composition of surface water from the high Alpine Zermatt area. Swiss Journal of Geosciences. vol 110, pp 811–831.

Dreschel P, Evans A E 2010. Irrigat. Drain. System. 24, 1–3.

Kolawole O M, Ajayi K T, Olayemi A B, Okoh A I 2011. Assessment of water quality in Asa River (Nigeria) and its indigenous Clarias gariepinus fish. International journal of environmental research and public health, 8(11): 4332-4352.

Mali S, Shumka S 2012. Possible management of fecal coliform pollution in Shkumbini basin following the WFD Guidelines. Journal of Environmental Protection and Ecology vol. 13(N: 3):p 1289 – 1296.

M.E.A (Millennium ecosystem assessment). 2005. Ecosystems and human well-being, 5, Washington, DC: Island press.

Murhekar G H 2011. Determination of Physico-Chemical parameters of Surface Water Samples in and around Akot City. International Research Journal of Chemistry and Environment, 1(2): 183-187.

Nahar, N., Lanon, M. A. H., Saha, B and Shaibur, M. R (2016). Assessment of physico-chemical properties of water of Gorai river at Kushtia town in 2014: a case study. Journal of Science, Technology and Environment Informatics, Vol. 2 No. 2, pp. 51–60.

Pano N, Selenica A, Puka V 1984. Hydrology of Albania., Tirana.

Singh L B, 2007. River Pollution. 1st Edn., APH Publishing,New Delhi, ISBN-10: 8131300854, pp: 192.

Smitha P G, Byrappa K, Ramaswamy S N 2007. Physico-chemical characteristics of water samples of Bantwal Taluk, South-Western Karnataka, India. J Environ Biol 28: 591-595.

Uddin G, Moniruzzaman M, Hoque M A, Khan M 2016 Seasonal Variation of Physicochemical Properties of Water in the Buriganga River, Bangladesh World Applied Sciences Journal 34 (1): p 24-34.

UNESCO (2003) Water for People, Water for Life. United Nations/World Water Assessment Programme,UNESCO, Paris and Berghahn Books, New York.

Vassilis Z. A, M. P. Dimitris and A.M. Konstantina, 2001.Statistical and trend analysis of water quality and quantity data for the Strymon River in Greece. Hydrology and Earth System Sciences, 5(4), 679–691.

WHO 2004. Guidelines for drinking-water quality, 3rd Edition, Volume 1: recommendations. Geneva: World Health Organization (www.who.int/water_sanitation_health).

DRAFT

POSTER PRESENTATION

A Non-Targeted Metabolomics Approach for Sex Identification from Human Dentine Samples Using ^{1}H-NMR-Based Metabolomics

Giatgong Konguthaithip[1], (*ORCID*: *https://orcid.org/ 0000-0002-0076-249X*), Karune Verochana[2], Tawachai Monum[3], (*ORCID*: *https://orcid.org/0000-0001-6621-9150*), Yutti Amornlertwatana[3], Churdsak Jaikang[3*], (*ORCID*: *https://orcid.org/0000-0002-9262-0404*),

[1] Chiang Mai University, Graduate School, Master's Degree Program in Forensic Science, Chiang Mai, 50200 Thailand.
[2] Chiang Mai University, Faculty of Dentistry, Division of Oral and Maxillofacial Radiology, Chiang Mai, 50200 Thailand
[3] Chiang Mai University, Faculty of Medicine, Department of Forensic Medicine, Chiang Mai, 50200 Thailand.

*Corresponding author E-mail: churdsak.j@cmu.ac.th;Tel.: +6653-934532

Abstract

After death, the tooth is the most remaining tissue and an important sample for forensic science investigation. It is covered by the enamel and dentine layers, which contain a variety of chemical compounds. Nuclear magnetic resonance spectrometry is an instrument used for determining targeted and non-targeted metabolomics profiles. The purpose of this study was to identify biomolecules contained in human dentine using 1H-NMR-based metabolomics. Dentine samples were collected from the second molar. The dentine powder was hydrolyzed at 100°C for 24 hours under acid condition. The solution was used for determining non-targeted metabolites by ^{1}H-NMR spectrophotometry. A total of one hundred and one chemical compounds were found in dentine, including organic acids, nucleic acids, fatty acyls, organoheterocyclic compounds, carbohydrates, organic nitrogen compounds, organic oxygen compounds, benzenoids, polyketides, alkaloids and lipids and lipid-like molecules. The levels of *L*-*N*-(3-carboxypropyl) glutamine, *N*-methyl-*L*-glutamic acid, citrulline, aminoacetone, 1-Methylguanine, glycine, dermatan sulfate, gamma-glutamylcysteine, *N*-acetylserine, ethyl glucuronide, creatinine, phosphohydroxypyruvic acid, phosphocreatine, mevalonic acid and L-pipecolic acid significantly decreased in males compared to females (p<0.01). Interestingly, this ^{1}H-NMR-based metabolomics technique utilizes a small amount of samples for chemical identification and could potentially be used for sex identification through machine learning models in the future.

Keyword: human dentine, ^{1}H-NMR, forensic science, biomolecule, Non-targeted Metabolomics

1. INTRODUCTION

Human dentine is a white layer and contained with solid substances as the main tooth. Teeth are the hardest organ in the human body and difficult to decompose from charred burning and bury a corpse. Chemical components in dentine both organic and inorganic compounds were elucidated. Enamel contains inorganic matrix (96%, w/w), the protein (90% amelogenin), collagen, lipids, and water (4%, w/w). Dentine is the next part of the tooth enamel. The composition of dentine is similar to the bone, combining with 70% (w/w) of inorganic matters, 20% of organic matters, and 10% of water.

Chemical compounds have been investigated and utilized for many forensic science studies. In forensic odontology, tooth have been investigated for specific and individual characteristics in each person including for age and gender. Chromatography techniques have been identified for D and L-aspartic acid. For chemical identification by chromatogram technique, there are many steps for sample preparation. Easy step for sample preparation, rapid identification and highly sensitivity are need available.

Nuclear magnetic resonance spectroscopy (NMR) was first discovered in 1940. NMR has been widely used in physics and chemistry identification. At present, NMR technique is identified for metabolomics in human

samples, including blood, serum, plasma, saliva, urine, and feces. The results have been used for prediction and diagnosis many diseases including heart diseases, diabetes and cancer.

The purpose of this study was to identify biomolecules contained in human dentine by ^{1}H-NMR 500 MHz. The chemical composition and the 1H-NMR technique can be applied in forensic science especially for gender and age identifications. Moreover, the cause of death, diseases, and toxic substances will be identified and might be linked to underlying disease in further.

2. MATERIALS AND METHODS

2.1 Chemicals

Hydrochloric acid (HCl) was obtained from Merck (Darmstadt, Germany). Deuterium oxide (D_2O), trimethylsilyl propanoic acid (TSP) were obtained from Sigma-Aldrich (Ontario, Canada).

2.2 Dentine sample collection

Two hundred samples of non-carious permanent second molar (100 males and 100 females) age ranged 21 to 85 years old. The teeth samples were obtained from Department of Forensic Medicine, Faculty of Medicine, Chiang Mai University, Thailand. This research was approved by the Ethics Committee of Faculty of Medicine, Chiang Mai University (FOR-2565-09049). The teeth samples were cleaned with 0.9% normal saline solution and distilled water to eliminate blood remaining according the method of Manuela et al. The dried samples were stored at -20 °C until analysis.

2.3 Dentine preparation

Dentine samples were isolated by transverse section (1 mm) by Isomet 1000 using a low-speed cross-sectional saw and the enamel surface was subsequently removed by Airotor handpiece, brand NSK model PANA-MAX. Both preparing steps were under cooling water condition to avoid the heat.

2.4 ^{1}H-NMR conditions

Dentine samples were firstly pulverized by a freezing mill (SPEX CertiPrep - 6750 Freezer/Mill) for 15 min. Fifty grams of the dentine powder was digested with 50 mg of 0.6 M HCl at 100 °C for 24 hrs. The digested solution was lyophilized. 0.1 M of TSP in D2O was added into the sample tube and then the mixed solution was transferred into NMR tube. Record the proton NMR spectrum was acquired at 27 ◦C on a Bruker AVANCE 500 MHz (Bruker, Bremen, Germany) with Carr-Purcell-Meiboom-Gill (CPMG, -RD-90◦-(t-180◦-t) n-acquire) using water-suppression pre-saturation pulse sequence. A 90◦ pulse with a 16 number of signal averaging (NSA) was applied. The baseline and phase correction were carefully adjusted by Bruker TopSpin versions 4.0.7 software.

2.5 Peak Assignment and Untargeted Metabolite Identification

Identification of each biomolecule was determined by using The Human Metabolome Data Base (HMDB) and previous published paper [David et al., 2013]. Acquisition of peak, *J* couplings joint value were analyzed by using Bruker TopSpin versions 4.0.7 software.

DRAFT

Table 1 shows the amounts of the top 101 chemicals and the variety of chemical compounds present in human dentin samples.

Position	Chemical compounds	HMDB ID	Amount (%)
1	5-hydroxyindoleacetic acid	0000763	0.04
2	Glycine	0000123	0.16
3	2,3,4,5-tetrahydro-2-pyridinecarboxylic acid	0012130	0.16
4	5-methylcytosine	0002894	0.17
5	L-alanine	0000161	0.17
6	Guanosine	0000133	0.18
7	inosine	0000195	0.18
8	3-Sulfinoalanine	0000996	0.19
Position	**Chemical compounds**	**HMDB ID**	**qNMR (%)**
9	L-allothreonine	0004041	0.19
10	Glycyl-aspartate	0028837	0.20
11	L-isoleucine	0000172	0.20
12	D-leucine	0013773	0.21
13	Oxoglutaric acid	0000208	0.22
14	Methylcysteine	0002108	0.22
15	Phosphocreatine	0001511	0.24
16	L-N-(3-carboxypropyl)glutamine	0029393	0.25
17	L-leucine	0000687	0.25
18	L-alpha-aminobutyric acid	0000452	0.25
19	Homocysteine thiolactone	0002287	0.26
20	4-hydroxy-L-glutamic acid	0002273	0.26
21	Ornithine	0000214	0.27
22	O-phospho-4-hydroxy-L-threonine	0006802	0.28
23	Malic acid	0000156	0.28
24	Serylvaline	0029052	0.31
25	2-Aminoadipic acid	0302754	0.32
26	5-Hydroxytryptophol	0001855	0.33
27	Pyruvic acid	0000243	0.33
28	Hydroxypropyl-asparagine	0028858	0.36
29	L-cysteine	0000574	0.39
30	Mevalonic acid	0000227	0.40
31	Quinolinic acid	0000232	0.41
32	5-Amino-2-oxopentanoic acid	0006272	0.42
33	Hydroxypyruvic acid	0001352	0.42
34	Aminoacetone	0002134	0.42
35	L-altruronic acid	0038491	0.44

DRAFT

36	1-Methylguanine	0003282	0.45
37	N-acetyltaurine	0240253	0.46
38	Aminomalonic acid	0001147	0.46
39	Hydroxypropyl-threonine	0028873	0.47
40	Cis-4-hydroxy-D-proline	0060460	0.48
41	Carnosine	0000033	0.48
42	Adenosine	0000050	0.49
43	Phosphoserine	0000272	0.51
44	N-methyl-L-glutamic acid	0062660	0.51
Position	**Chemical compounds**	**HMB ID**	**qNMR (%)**
45	L-phenylalanine	0000159	0.52
46	Dermatan sulfate	0000632	0.53
47	Isocitric acid	0000193	0.54
48	L-glutamine	0000641	0.54
49	3-methylguanine	0001566	0.54
50	Homocysteine	0000742	0.58
51	Spermidine	0001257	0.59
52	N-formyl-L-aspartate	0060495	0.59
53	Creatinine	0000562	0.61
54	D-glutamine	0003423	0.61
55	Norophthalmic acid	0005766	0.64
56	Gamma-glutamylcysteine	0001049	0.64
57	Betaine	0000043	0.66
58	Chondrotin sulfate	0000580	0.67
59	N-acetylserine	0002931	0.67
60	D-cysteine	0003417	0.67
61	Methionine sulfone	0062174	0.69
62	Glycolic acid	0000115	0.69
63	L-glutamic acid	0000148	0.69
64	L-3-phenyllactic acid	0000748	0.73
65	N-hydroxy-L-tyrosine	0038750	0.73
66	N-acetylglutamine	0006029	0.73
67	D-proline	0003411	0.73
68	Hydroxyprolyl-serine	0028872	0.75
69	L-homoserine	0000719	0.79
70	D-glutamic acid	0003339	0.79
71	N-carbamoylsarcosine	0012265	0.83
72	L-Norleucine	0001645	0.90

DRAFT

73	L-asparagine	0000168	0.90
74	Ethyl glucuronide	0010325	0.91
75	Cis-Aconitric acid	0000072	0.92
76	Citrulline	0000904	0.92
77	L-methionine	0000696	0.99
78	Succinic acid	0000254	1.00
79	3-Methylthiopropionic acid	0001527	1.01
Position	**Chemical compounds**	**HMB ID**	**qNMR (%)**
80	Cysteic acid	0002757	1.03
81	L-aspartyl-4-phosphate	0012250	1.05
82	L-2,4-diaminobutyric acid	0006284	1.05
83	L-proline	0000162	1.17
84	Methionine sulfoxide	0002005	1.18
85	Lanthionine	0240656	1.22
86	8-hydroxy-7-methylguanine	0006037	1.26
87	Creatine	0000064	1.29
88	Betaine aldehyde	0001252	1.29
89	L-alloisoleucine	0000557	1.39
90	L-2-hydroxyglutaric acid	0000694	1.49
91	L-2-Amino-3-oxobutanoic acid	0006454	1.60
92	2,3-diamonopropionic acid	0002006	1.76
93	2-Ketobutyric acid	0000005	1.85
94	2-oxo-3-hydroxy-4-phosphobutanoic acid	0006801	2.00
95	L-serine	0000187	2.19
96	7-methyladenine	0011614	2.27
97	D-asparagine	0033780	3.64
98	2-keto-6-aminocaproate	0012151	5.29
99	L-pipecolic acid	0000716	6.41
100	D-serine	0003406	6.57
101	Phosphohydroxypyruvic acid	0001024	8.22

RESULTS and DISCUSSION

In this study, human dentine samples obtained from Department of Forensic Medicine Chiang Mai University. The samples were hydrolyzed with hydrochloric acid to obtain free molecules. One hundred and one chemical compounds found in dentine samples and the results are shown in Table 1. The NMR chromatogram is shown in Figures 1. Chemical compounds including organic acids, nucleic acids, fatty acyls, organoheterocyclic compounds, carbohydrates, organic nitrogen compounds, organic oxygen compounds, benzenoids, polyketides, alkaloids and lipids and lipid-like molecules, and metabolites ethanol were found in human dentine. 5-hydroxy indole acetic acid (5-HIAA), 5-hydroxytryptopol (5-HTOL) and Ethyl glucuronide which are ethanol's metabolites could be detected and be indicate the deceased exposure to alcoholic beverages.

DRAFT

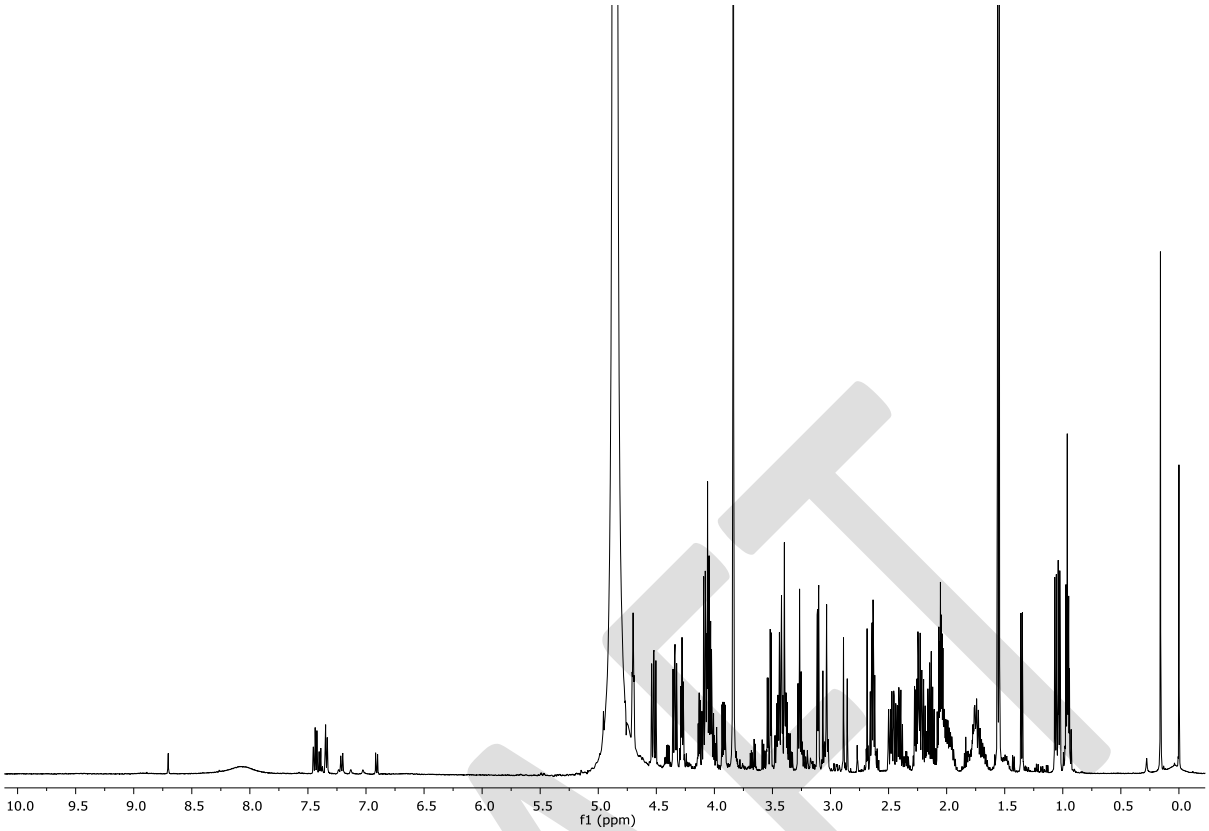


Fig.1 Chromatograms of a typical 500 MHz of ^{1}H-NMR spectra of hydrolyzed dentine at 0.00-7.44 ppm regions.

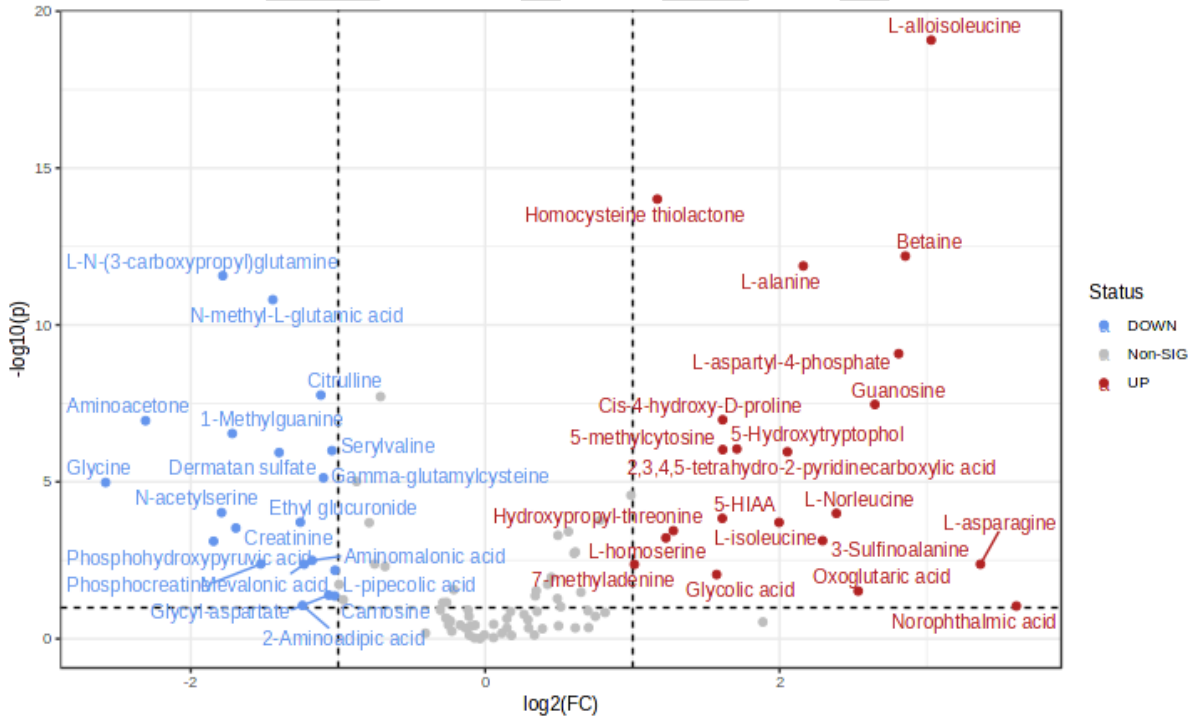


Fig.2 Volcano plot of between Male and Female when fold changes more than 1.2.

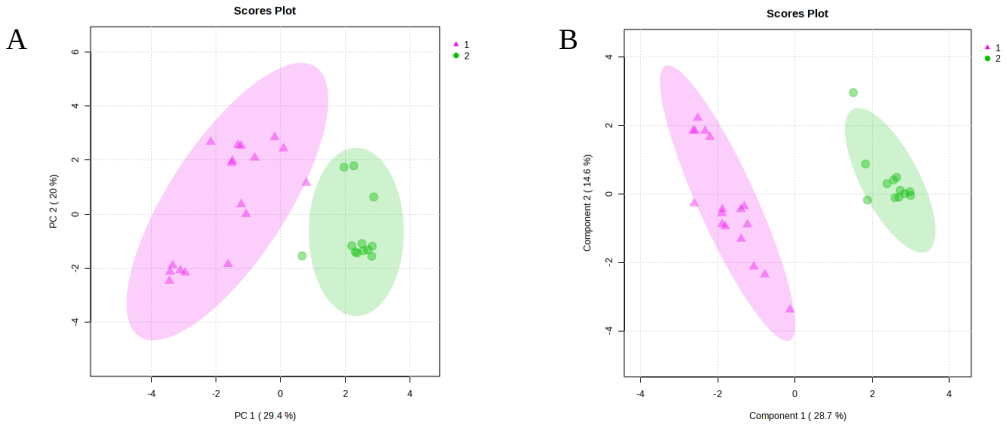


Fig.3 PCA and PLS-DA analysis of chemical in human dentine expression between the groups. PCA score plots (A) and PLS-DA score plots (B) of chemicals in male and female dentine samples data.

The ^{1}H-NMR is a popular technique for non-targeted metabolomic screening in various biological samples, including blood, urine, saliva and feces. This study represent the first research conducted in Thailand for non-targeted metabolites analysis in dentine performing by ^{1}H-NMR. Specific central peaks were identified and mapped for each chemical by Human Metabolome Data Base (HMDB).

In Figure 2, volcano plot are presented, depicting different chemical compounds compared between male and female samples which the x-axis being log fold change and the y being -log10(p-value) and logFC > 1.2, -log10(p-value) > 1.2 (< 0.05). A principal component analysis (PCA) and principle partial least square discrimination analysis (PLS-DA) plot demonstrated that certain chemical compounds could be exactly separated male and female.

Our results revealed that betaine , *L*-*N*-(3-carboxypropyl)glutamine, *N*-methyl-*L*-glutamic acid, Citrulline, Aminoacetone, 1-Methylguanine, Glycine, Dermatan sulfate, Gamma-glutamylcysteine, *N*-acetylserine, Ethyl glucuronide, Creatinine, Phosphohydroxypyruvic acid, Phosphocreatine, Mevalonic acid, *L*-pipecolic acid, Carnosine, Glycyl-aspartate, 2-Aminoadipic acid were significantly higher concentration in male more than female samples (p<0.01). But, Oxoglutaric acid, *L*-isoleucine, Glycolic acid, 7-methyladenine, 3-Sulfinoalanine, *L*-Norleucine, 5-hydroxyindoleacetic acid, 2,3,4,5-tetrahydro-2, pyridinecarboxylic acid, 5-Hydroxytryptophol, *L*-alloisoleucine, Homocysteine thiolactone, *L*-alanine, Betaine, *L*-aspartyl-4-phosphate, Guanosine, Cis-4-hydroxy-D-proline, 5-methylcytosine, *L*-asparagine, *L*-homoserine, Norophthalmic acid significantly increased in female more than male (p<0.01). These compounds could be applied for sex investigation in Forensic Science. Particularly in the identification of anonymous skull or tooth fragments by ^{1}H-NMR could be applied.

CONCLUSION

This experiment demonstrated that ^{1}H-NMR spectroscopy technique can identify 101 chemical compounds in human dentine samples. They categorized into organic acids, nucleic acids, fatty acyls, organoheterocyclic compounds, carbohydrates, organic nitrogen compounds, organic oxygen compounds, benzenoids, polyketides, alkaloids and lipids and lipid-like molecules. For application, *L*-*N*-(3-carboxypropyl)glutamine, *N*-methyl-*L*-glutamic acid, citrulline, aminoacetone, 1-methylguanine, glycine, dermatan sulfate, gamma-glutamylcysteine, *N*-acetylserine, ethyl glucuronide, creatinine, phosphohydroxypyruvic acid, phosphocreatine, mevalonic acid, *L*-pipecolic acid, carnosine, glycyl-aspartate, 2-aminoadipic acid categorized male and female. Another biomolecules would be more identified and applied for forensic cases further.

ACKNOWLEDGEMENTS

We greatful thanks Dr. Wirintra Watcharanone, Dr. Kanjana Inthongf, Dr. Atchara Pothikunapatg, Dr. Preeyajak Chinwaraphath for collcecting the teeth samples.

DRAFT

REFERENCES

Almhojd US, Lingstrom P, Nilsson A, Noren JG, Siljestrom S, Ostlund A, et al. Molecular Insights into Covalently Stained Carious Dentine Using Solid-State NMR and ToF-SIMS. Caries Res. 2017;51(3):255-63.

Andreev AS, Livadaris V. Characterization of Catalytic Materials through a Facile Approach to Probe OH Groups by Solid-State NMR. The Journal of Physical Chemistry C. 2017;121(26):14108-19.

Berglund B, Vaughan RW. Correlations between proton chemical shift tensors, deuterium quadrupole couplings, and bond distances for hydrogen bonds in solids. The Journal of Chemical Physics. 1980;73(5):2037-43.

Chan W, Banks R, Lynch E, Grootveld M. High-Resolution ¹H NMR Investigations of the Oxidative Consumption of Salivary Biomolecules by a Tooth-Whitening Formulation: Relevance to Safety Issues. Advances in Chemical Engineering and Science. 2012;02(01):62-73.

Coppel Y, Prigent Y, Gregoire G. Characterization of hydrogenated dentin components by advanced (1)H solid-state NMR experiments. Acta Biomater. 2021;120:156-66.

de Falco B, Incerti G, Pepe R, Amato M, Lanzotti V. Metabolomic Fingerprinting of Romaneschi Globe Artichokes by NMR Spectroscopy and Multivariate Data Analysis. Phytochem Anal. 2016;27(5):304-14.

Diez C, Rojo MÁ, Martín-Gil J, Martín-Ramos P, Garrosa M, Córdoba-Diaz D. Infrared Spectroscopic Analysis of the Inorganic Components from Teeth Exposed to Psychotherapeutic Drugs. Minerals. 2021;12(1).

Gardner A, Parkes HG, Carpenter GH, So PW. Developing and Standardizing a Protocol for Quantitative Proton Nuclear Magnetic Resonance ((1)H NMR) Spectroscopy of Saliva. J Proteome Res. 2018;17(4):1521-31.

Grootveld M, Page G, Bhogadia M, Edgar M. Updates and Original Case Studies Focused on the NMR-Linked Metabolomics Analysis of Human Oral Fluids Part I: Emerging Platforms and Perspectives. Applied Sciences. 2022;12(3).

Kim H-Y, Nam S-H, Han M-S. Hard Tissue Analysis of NMR after Fluoride Administration. Journal of Magnetics. 2016;21(4):599-602.

Letieri AdS, Freitas-Fernandes LB, Albarello LL, Fontes GP, de Souza IPR, Valente AP, et al. Analysis of Salivary Metabolites by Nuclear Magnetic Resonance Before and After Oral Mucosa Cleaning of Infants in the Pre-dental Period. Frontiers in Dental Medicine. 2021;2.

Nagana Gowda GA, Gowda YN, Raftery D. Expanding the limits of human blood metabolite quantitation using NMR spectroscopy. Anal Chem. 2015;87(1):706-15.

Ni Q, Chen S. Assessment of structural changes of human teeth by low-field nuclear magnetic resonance (NMR). Meas Sci Technol. 2010;21(1):15803.

Ni Q, Nicolella DP. The characterization of human cortical bone microdamage by nuclear magnetic resonance. Meas Sci Technol. 2005;16(3):659-68.

Pickard CJ, Mauri F. All-electron magnetic response with pseudopotentials: NMR chemical shifts. Physical Review B. 2001;63(24).

Qingwen N.; Shuo C. Assessment of structural changes of human teeth by low-field nuclear magnetic resonance (NMR). Molecular Biology. 2010 (27):1-15.

Tsai YL, Kao MW, Huang SJ, Lee YL, Lin CP, Chan JCC. Characterization of Phosphorus Species in Human Dentin by Solid-State NMR. Molecules. 2020;25(1).

Turunen S, Puurunen J, Auriola S, Kullaa AM, Karkkainen O, Lohi H, et al. Metabolome of canine and human saliva: a non-targeted metabolomics study. Metabolomics. 2020;16(9):90.

Verma M, Verma N, Sharma R, Sharma A. Dental age estimation methods in adult dentitions: An overview. J Forensic Dent Sci. 2019;11(2):57-63.

Vyalikh A, Mai R, Scheler U. OH(-) deficiency in dental enamel, crown and root dentine as studied by (1)H CRAMPS. Biomed Mater Eng. 2013;23(6):507-12.

Wishart DS, Jewison T, Guo AC, Wilson M, Knox C, Liu Y, et al. HMDB 3.0--The Human Metabolome Database in 2013. Nucleic Acids Res. 2013;41(Database issue):D801-7.

Zhou J, Ning K, Yang Y, Zou L, Xue J, Kong X, et al. 1H-NMR -based metabolic analysis on biocompatibility of dental biomaterials. Process Biochem. 2022;114:163-73.

Zia K, Siddiqui T, Ali S, Farooq I, Zafar MS, Khurshid Z. Nuclear Magnetic Resonance Spectroscopy for Medical and Dental Applications: A Comprehensive Review. Eur J Dent. 2019;13(1):124-8.

DRAFT

POSTER PRESENTATION

Pd (II)-phosphine-benzimidazole derivative complexes with *in vitro* antioxidant and *in vivo* antiangiogenic potentials

H. Tuba Kıyan[1*](*ORCID: https://orcid.org/0000-0001-5304-868X),*
Pervin Soyer[2] (*ORCID: https://orcid.org/0000-0002-6258-1993),*
Hakan Ünver[3] (*ORCID:* https://orcid.org/0000-0002-4858-4039),
Deniz Erol Kutucu[4] (*ORCID:* https://orcid.org/0000-0001-6867-2362)

[1*]Anadolu University, Faculty of Pharmacy, Department of Pharmacognosy, 26470 Eskişehir, Turkey
[2]Anadolu University, Faculty of Pharmacy, Department of Pharmaceutical Microbiology, 26470 Eskişehir, Turkey
[3]Eskişehir Technical University, Faculty of Science, Department of Chemistry, 26555 Eskişehir, Turkey
[4]Istanbul University, Faculty of Science, Department of Biology, 34134 Istanbul, Turkey.

*Corresponding author: htkiyan@anadolu.edu.tr

Abstract

Following the discovery of cis-platin, the field of medicinal chemistry experienced an explosion in interest to organometallic compounds. The researchers suggest that the discovery of the potential antiangiogenic effects associated with anticancer activities will provide significant advancements in cancer therapy. Palladium complexes have garnered significant attention in the field of cancer research due to their resemblance in chemical and electrical properties to platinum compounds. Moreover, the lipophilic nature of palladium complexes has been found to enhance their antiangiogenic and anticancer properties. Benzimidazole derivatives are a class of chemicals that has garnered significant attention within the field of medicinal chemistry. The bioactivity of benzimidazole-based compounds arises from their structural resemblance to DNA or enzymes. On the contrary, the presence of many donor atoms renders phosphine-type compounds more appealing in the field of organometallic chemistry. The efficacy of phosphines in inhibiting cancer growth has been extensively demonstrated in numerous academic investigations. The objective of this study is to synthesize and describe novel complexes consisting of palladium, phosphine, and benzimidazole. The antioxidant capabilities of these complexes will be evaluated by using *in vitro* ABTS cation decolorization assay, while their antiangiogenic activities will be assessed through *in vivo* chorioallantoic membrane (CAM) method. All of the complexes showed strong antioxidant and antiangiogenic activity.

Keywords: Antiangiogenic, Anticancer, Antioxidant, Benzimidazole, Phosphine, Palladium.

INTRODUCTION

The potential of palladium complexes as anticancer drugs is substantiated by the resemblances observed between the metal complexes of platinum (II) and palladium (II) molecules, as reported by Miklášová et al. (2009). Platinum-based chemotherapeutic agents have played a crucial role within the realm of metal-based anticancer treatments. According to several studies, it has been found that palladium derivatives possess a significant cytotoxic impact, which can be compared to that of well-established platinum-based drugs like cisplatin, carboplatin, and oxaliplatin. Furthermore, these derivatives have been observed to cause fewer adverse effects compared to other heavy metal compounds used in anticancer therapy (Divsalar et al., 2007; Miklášová et al., 2009). According to a study conducted by Saygideğer Demir et al. in 2014, it was shown that phosphine metal complexes have inhibitory effects on antioxidant enzymes and cytotoxic effects on lung and leukemia cell lines. Previous studies have reported the in vitro anticancer properties and in vivo angiogenic effects of several gold phosphine metal complexes (Bagowski et al., 2009; Reddy et al., 2018). The benzimidazole ring system is commonly found in numerous bioactive heterocyclic compounds, owing to its diverse range of biological and medicinal applications. Benzoimidazoles have garnered significant attention from medicinal and synthetic organic chemists due to their diverse range of biological actions. This group of heterocyclic molecules has been extensively studied and is known to possess various properties, including antidiabetic, anticancer, hormone antagonist, antiviral, anti-HIV, anthelmintic, antiprotozoal, antimycobacterial, antihypertensive, anti-inflammatory, analgesic, anxiolytic, coagulant, anticoagulant, antioxidant, antidiabetic, and antiallergic properties (Rashedy and Aboul-Enein, 2013; Shrivastava et al., 2017).

DRAFT

According of the molecules inherent biological characteristics, the present investigation involved the synthesis of two novel complexes including palladium, phosphine, and benzimidazole. The structural characteristics of these complexes were determined through the application of diverse spectroscopic methodologies. The objective of this study was to assess the antioxidant effects of these compounds *in vitro* using metal chelating, reducing power assessment, ABTS cation decolorization, and *in vivo* antiangiogenic activities utilizing the chorion allantoic membrane (CAM) method, which has not been previously explored.

MATERIALS AND METHODS

Synthesis of benzimidazole derivatives

During the synthesis step, the reaction was conducted by combining benzimidazole with benzyl bromide, and separately, 4-bromobenzyl bromide and 4-methylbenzyl bromide. This reaction took place in a dimethyl sulfoxide solvent under reflux conditions in a basic environment, utilizing potassium hydroxide. Following the completion of the reaction, a series of separation and purification techniques, including extraction, column chromatography, and thin layer chromatography, were employed to obtain the desired product. As a result, three distinct benzimidazole derivatives were obtained.

Synthesis of new palladium complexes

The experiment involved the addition of triphenylphosphine, tri-p-tolylphosphine, or tris (4-methoxyphenyl) phosphine chemical dissolved in acetonitrile dropwise to a solution of palladium chloride in acetonitrile. Following an hour-long stirring period at a temperature of 40ºC using a magnetic hot mixer, the mixture was dissolved in acetonitrile. Subsequently, the produced benzimidazole compounds were added dropwise. Following a 12-hour period of stirring, the entirety of the solution was subjected to evaporation through the process of filtration. Consequently, two distinct palladium complexes were obtained.

Characterization of synthesized molecules

X-Ray Single Crystal Spectroscopy, Nuclear Magnetic Resonance Spectroscopy (NMR), HR-Mass Spectroscopy, Infrared Spectroscopy or Fourier transform infrared spectroscopy (FT-IR) were performed using Elemental Analysis methods.

In vitro ABTS decolorization assay

The decolorization assay for ABTS and its reducing power were assessed using the methodology outlined by Re et al. (1999). ABTS was dissolved in water to achieve a concentration of 7 mM. The generation of ABTS radical cation was achieved by combining ABTS stock solution with a final concentration of 2.45 mM potassium persulfate. The resulting combination was then allowed to incubate in darkness at room temperature for a duration of 12-16 hours prior to utilization. The reagent utilized in this work necessitates fresh preparation before to each experiment. It was diluted with ethanol to achieve an absorbance reading of 0.700 (± 0.02) at a wavelength of 734 nm. This reagent was subsequently employed in the assessment of antioxidant activity. In the experimental procedure, a volume of 1 mL of reagent solution was introduced to a volume of 10 µL of sample solution, which had a concentration of 0.5 mg/mL. The resulting mixture was allowed to incubate for a duration of 5 minutes, after which the absorbance of the solution was determined at a wavelength of 734 nm. In the research conducted by Re et al. (1999), ethanol was employed as a blind, while a trolox solution with a concentration of 0.5 mg/mL was utilized as a positive control. The calculation of radical scavenging activity (RSA) involved determining the percentage inhibition of ABTS radical reduction using the following formula: "% ABTS scavenging = [(A control - A sample) / A control] × 100".

In vivo angiogenic activity (CAM assay)

Preparation of test samples

Benzimidazole derivatives and new palladium complexes, along with a positive control of (±)-Thalidomide and a negative control of SDS, were dissolved in a 2.5% (w/v) agarose solution. The chick chorioallantoic membrane was treated with 10 mL of these solutions, which were prepared and put onto circular stainless-steel supports with a diameter of 5 mm. The solutions were cooled to room temperature to solidify before being applied onto the CAM.

In vivo chick embryo chorioallantoic membrane (CAM) assay

The present study conducted an in vivo CAM analysis, using the methodology outlined in previous publications by Gurel-Gurevin et al. (2018), Ünver et al. (2022), and Yurtdaş-Kırımlıoğlu et al. (2022a, 2022b).

The fertilized hen eggs had a 72-hour incubation period at a temperature of 36.5°C, accompanied by a relative humidity of 80%. Subsequently, the eggs were placed in a horizontal orientation and subjected to many rotations. After the eggs were opened on the side opposite the snub, a volume of 10-15 mL of albumin was extracted. The eggs were incised using a scalpel at a position two-thirds of their height, measured from the pointed end. Subsequently, the shells were extracted using forceps. The cavity was sealed using stretch film, and the eggs were placed in an incubator set at a temperature of 36.5 ℃ and a relative humidity of 80% for an additional duration of 72 hours. The CAM was prepared by placing one pellet per egg, with each pellet having a diameter of roughly 2 cm. The hole in the egg shell was then sealed once more with stretch film. The eggs were seen after a duration of twenty-four hours utilizing a stereomicroscope. Each test condition employed a range of 10 to 15 eggs. The examination of each sample was conducted three times, with each examination being performed in duplicate. The anti-angiogenic effects were evaluated using a grading system, as shown in Table I. The subsequent equation was employed to get the anti-angiogenic scores: The mean score is determined by the formula [Number of eggs (score >1)2+Number of eggs (score >0.75-1)1]/ (total number of eggs scored) (Gurel-Gurevin et al., 2018; Ikitimur-Armutak et al., 2017; Kiyan et al., 2014). In order to provide positive and negative controls, the compounds Thalidomide and sodium dodecylsulfate (SDS) were used in the study. These controls were tested at a concentration of 50 mg/pellet. In order to conduct blank tests, solidified agarose-solution pellets (2.5%, w/v) were utilized for the treatment of CAMs. Each experiment was replicated three times.

Table 1. Score system of anti-angiogenic effect

Scale	Anti-angiogenic effect	Effects observed on CAM after treatment
<0.5	Inactive	No change in embryo
0.5 - 0.75	Weak	No capillary free area
0.75 - 1	Strong	Small capillary free area
>1	Very strong	Capillary free area around the pellet

RESULTS and DISCUSSION

Characterizations of complexes

Complex 1; Yellow solid (Yield=49%), FT-IR (KBr) cm-1: 3082-2837, 1592, 1498, 1290, 1251, 1178, 1099, 1021, 825, 799, 752, 727, 537. Calc.: $C_{32}H_{26}BrCl_2N_2PPd$: C, 52.88; H, 3.61; N, 3.85; Result: C, 52.75; H, 3.42; N, 3.83.
Complex 2; Yellow solid (Yield=49%), FT-IR (KBr) cm-1: 3104-2837, 1592, 1520, 1489, 1463, 1390, 1261, 1188, 752. Calc.: $C_{35}H_{32}BrCl_2N_2PPd$: C, 54.68; H, 4.20; N, 3.64; Found: C, 54.18; H, 4.35; N, 3.16.

In vitro ABTS decolorization assay

Antioxidant activity results of Pd complexes 1 and 2 by ABTS cation decolorization/inhibition assay are given in Table 2. The highest 68.56 % inhibition activity was determined with complex 2. Both of complexes showed strong antioxidant activity when compared with control groups.

Table 2. ABTS % inhibition rates

Pd complexes/Control groups	% Inhibition
Complex 1	49.79 ± 0.1
Complex 2	68.56 ± 5.01
BHT	43.55 ± 1.3
Ascorbic Acid	96.56 ± 0.5

***In vivo* angiogenic activity (CAM assay)**

Antiangiogenic effect scores regarding the results of antiangiogenic activity of Pd complexes 1 and 2 by *in vivo* CAM method are given in Table 3. When the results are evaluated according to the score table both of complexes showed strong antiangiogenic activity.

Table 3. Antiangiogenic activity scores

Pd complexes	Concentration (mg/pellet)	Average score	Anti-angiogenic effect	Irritation/Tox (%)
Complex 1	50	0.9 ± 0.2	Strong	-
Complex 2	50	0.85 ± 0.2	Strong	-
(±)-Thalidomide	50	0.9 ± 0.1	Strong	-
SDS	50	-	-	84.3 ± 0.02
Agar (blank)	% 2.5, w/v	-	Inactive	-

CONCLUSION

The current state of literature lacks research examining the antioxidant and antiangiogenic properties of Pd, phosphine, and benzimidazole complexes produced within the scope of this project. Hence, given that the investigation focused on evaluating the antioxidant and antiangiogenic properties of palladium complexes synthesized in this study, it is possible to consider that these synthesized complexes hold potential as a novel therapeutic agent in the development of drugs aimed at inhibiting angiogenesis induced by oxidative stress.

ACKNOWLEDGEMENTS

This study was supported by Anadolu University Scientific Research Projects (BAP) Commission. Project number is 1907S135. We greatly acknowledge the financial support.

REFERENCES

Bagowski CP, You Y, Scheffler H, Vlecken DH, Schmitz DJ, Ott I 2009. Naphthalimide gold(I) phosphine complexes as anticancer metallodrugs. Dalton Transactions 10799–10805.

Divsalar A, Saboury AA, Yousefi R, Moosavi-Movahedi AA, Mansoori-Torshizi H 2007. Spectroscopic and cytotoxic studies of the novel designed palladium(II) complexes: β-Lactoglobulin and K562 as the targets. Int J Biol Macromol 40, 381–386.

Gurel-Gurevin E, Tuba Kiyan H, Esener OBB, Aydinlik S, Uvez A, Ulukaya E, Dimas K, Armutak EI 2018. Chloroquine used in combination with chemotherapy synergistically suppresses growth and angiogenesis in vitro and in vivo. Anticancer Res 38, 4011–4020.

Ikitimur-Armutak EI, Gurel-Gurevin E, Kiyan HT, Aydinlik S, Yilmaz VT, Dimas K, Ulukaya E 2017. Anti-angiogenic effect of a Palladium (II)-Saccharinate Complex of Terpyridine in vitro and in vivo. Microvasc Res 109, 26–33.

Kiyan HT, Demirci B, Başer KHC, Demirci F 2014. The in vivo evaluation of anti-angiogenic effects of Hypericum essential oils using the chorioallantoic membrane assay. Pharm Biol 52, 44–50.

Miklášová N, Fischer-Fodor E, Lönnecke P, Schrepler MP, Virag P, Tatomir C, Cernea VI, Hey-Hawkins E, Silaghi-Dumitrescu L 2009. Antiproliferative effect and genotoxicity of novel synthesized palladium complexes with organoarsenic ligands. J Inorg Biochem 103, 1739–1747.

Rashedy A, Aboul-Enein H 2013. Benzimidazole Derivatives as Potential Anticancer Agents. Mini-Reviews in Medicinal Chemistry 13, 399–407.

Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, & Rice-Evans C 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free radical biology and medicine*, *26*(9-10), 1231-1237.

Reddy TS, Privér SH, Mirzadeh N, Bhargava SK 2018. Synthesis of gold(I) phosphine complexes containing the 2-BrC6F4PPh2 ligand: Evaluation of anticancer activity in 2D and 3D spheroidal models of HeLa cancer cells. Eur J Med Chem 145, 291–301.

Saygideğer Demir B, Keleş T, Serindağ O 2014. Antioxidant enzyme inhibitor role of phosphine metal complexes in lung and leukemia cell lines. Bioinorg Chem Appl 2014.

Shrivastava N, Naim MJ, Alam MJ, Nawaz F, Ahmed S, Alam O 2017. Benzimidazole Scaffold as Anticancer Agent: Synthetic Approaches and Structure–Activity Relationship. Arch Pharm (Weinheim) 350.

Ünver H, Dıkmen G, Kiyan HT 2022. Synthesis, X-ray characterization and evaluation of potent anti-angiogenic activity of a novel copper (II)-imidazole-bipyridyl complex. Inorganic and Nano-Metal Chemistry 52, 1153–1160.

Yurtdaş-Kırımlıoğlu G, Görgülü Ş, Güleç K, Kıyan HT 2022a. Nanoarchitectonics of PLGA based polymeric nanoparticles with oseltamivir phosphate for lung cancer therapy: In vitro-in vivo evaluation. J Drug Deliv Sci Technol 67.

Yurtdaş-Kırımlıoğlu G, Güleç K, Görgülü Ş, Kıyan HT 2022b. Oseltamivir phosphate loaded pegylated-Eudragit nanoparticles for lung cancer therapy: Characterization, prolonged release, cytotoxicity profile, apoptosis pathways and in vivo anti-angiogenic effect by using CAM assay. Microvasc Res 139.

DRAFT

POSTER PRESENTATION

High levels of antimicrobial resistance of *Pseudomona aeruginosa* and *Escherichia coli* against specific antibiotics in Albanian backyard poultry

Sonila Çoçoli[1*] (*https://orcid.org/0009-0001-2822-1732*), Tana Kika[1] (*https://orcid.org/0000-0002-4676-4108*), Jonida Boci[2] *(https://orcid.org/0009-0000-9480-6723)* Nikola Puvača[3] *(https://orcid.org/0000-0002-5500-7010), Ilda Laci[4] (https://orcid.org/0009-0007-5450-5177)*

[*1]Agricultural University of Tirana, Faculty of Veterinary Medicine, Veterinary Public Health Department, Tirana, Albania.
[2]Food Safety and Veterinary Institute, Department of Animal Health, Tirana, Albania.

[3]University Business Academy, Faculty of Economics and Engineering Management, Department of Engineering Management in Biotechnology, Novi Sad, Serbia.

[4]Agricultural University of Tirana, Faculty of Veterinary Medicine, Preclinical Subjects Department, Tirana, Albania.

*Corresponding author e-mail: scocoli@ubt.edu.al

Abstract

Antimicrobial agents are used to reduce clinical infections and production losses caused by microorganisms in poultry industry. Backyard poultry flocks have become increasingly popular and little is known about the main cause of bacterial infections in these populations. In this case study, were exanimated 30 different backyards poultry flocks in Elbasan district, Albania for the presence of *Pseudomonas aeruginosa* and *Escherichia coli* in sudden found dead poultry carcasses and their visceral organs respectively. These gram-negative agents were tested simultaneously against 5 antibiotics: Ampicillin AMP, Amoxicillin AMX, Ciprofloxacin CIP, Enrofloxacin ENR, Gentamicin GEN and Polymyxin B POLYB. In total, n=120 samples (carcasses and viscera) were tested, from which 32 strains of P. aeruginosa (26, 67%) and 44 of *E. coli* (36, 66%), were isolated.
The laboratory findings emphasised the presence of both bacteria in 30% of the assayed flocks. Moreover, all of the tested isolates were resistant towards up to three of the tested antibiotics. Relevant data of high resistance against Ciprofloxacin CIP (53, 94% of the n=76 tested isolates) and Polymyxin B POLYB (63, 15% of the n= 76 tested isolates) were reported. These data reveal relative risk of spread and development of antimicrobial resistance in zoonotic agents, which can lead to therapy failures, economic losses and concerns of veterinary public health.

Keywords: *Pseudomonas aeruginosa*, *Escherichia coli*, Poultry, Albania, Antimicrobial-resistance, Veterinary Public Health

INTRODUCTION

The inappropriate use of a wide range of antimicrobials for prophylactic, therapeutic, and growth-promotion purposes over a prolonged period of time contributes to the failure in poultry management and biosecurity, promoting the selective pressure of antimicrobial-resistant microorganisms and the emergence and spread of AMR strains. Antimicrobial resistance (AMR) is a growing concern in both commercial poultry farming and backyard poultry. The use of antimicrobials as growth promoters in small-scale chicken farming has been identified as a contributing factor to the development of antimicrobial resistance in poultry (Hedman et al., 2020). A pilot study (Alcaine et al., 2015) conducted in Albanian poultry farms found that bacterial isolates had much higher levels of antibiotic resistance compared to EU countries. Another study found that avian pathogenic Escherichia coli resistance patterns in Albania were extremely high against Neomycin, Doxycycline, Trimethoprim-Sulfamethoxazole, and other antibiotics (Shtylla et al., 2023). In backyard poultry, the use of antibiotics is generally lower than in commercial poultry farming, but AMR can still be a concern. Backyard poultry, which are generally raised without routine antimicrobial therapy, can function as

both a regular source of marginal income and a potential source of AMR (Hedman et al., 2020). Backyard poultry may be more susceptible to disease due to poor biosecurity measures, which can lead to the overuse of antibiotics (Nhung et al., 2017). Additionally, the close proximity of backyard poultry to humans can increase the risk of transmission of antibiotic-resistant bacteria (Hedman and Zhang, 2020). In Albania backyard poultry flocks have become increasingly popular and little is known about the main cause of bacterial infections in these populations or the antimicrobial resistance of these pathogens. This study is focused in 30 different backyards poultry flocks in Elbasan district, Albania for the presence of Pseudomonas aeruginosa and Escherichia coli. After the isolation of these two agents, the possible level of antimicrobial resistance was examined to better understand the present risk of AMR.

MATERIALS AND METHODS

In this case study, were exanimated 30 different backyards poultry flocks in Elbasan district, Albania for the presence of *Pseudomonas aeruginosa* and *Escherichia coli* in sudden found dead poultry carcasses and their visceral organs respectively. During the period of time March-April 2023, a total of 120 samples were collected and the laboratory analysis were carried out in the Faculty of Veterinary Medicine of Tirana.

The samples (visceral organs), were inoculated into the non-selective pre-enrichment medium, Buffered-Peptone Water and incubated at 37 °C for 24 h. Cultures obtained were plated onto MacConkey agar for the detection of *E. coli* and Cetrimide agar for the detection of *P. aeruginosa*, and incubated at 37 °C for 24 h. Suspected *E. coli* colonies were streaked onto Tryptone Bile X-Glucuronide and incubated at 42 °C for 24 h. Finally, all isolates were biochemically identified by using API 20E system, whereas potential *Pseudomonas* spp. colonies were submitted to oxidase test and processed by biochemical identification by API 20 NE system.

All isolates were submitted to antimicrobial susceptibility testing using the disc diffusion method (Kirby–Bauer test) against 5 different antibiotics: Ampicillin (AMP), Amoxicillin (AMX), Ciprofloxacin (CIP), Enrofloxacin (ENR), Gentamicin (GEN) and Polymyxin B (POLYB).

RESULTS and DISCUSSION

Overall 120 samples analysed, from 30 diffrent poultry flocks, 76 (53,95%) of them resulted positive for *Escherichia coli* and *Pseudomonas aeruginosa*, from which 44 (36,66%) strains of *E.coli* and 32 (26,67%) strains of *P.aeruginosa* were isolated.

Table 1. Isolated strains of *E.coli* and P.*aeruginosa* from backyard poultry

Total No.of sample analysed	*E.coli* strains	*P.aeruginosa* strains
120	44 (36,66%)	32 (26,67%)

DRAFT

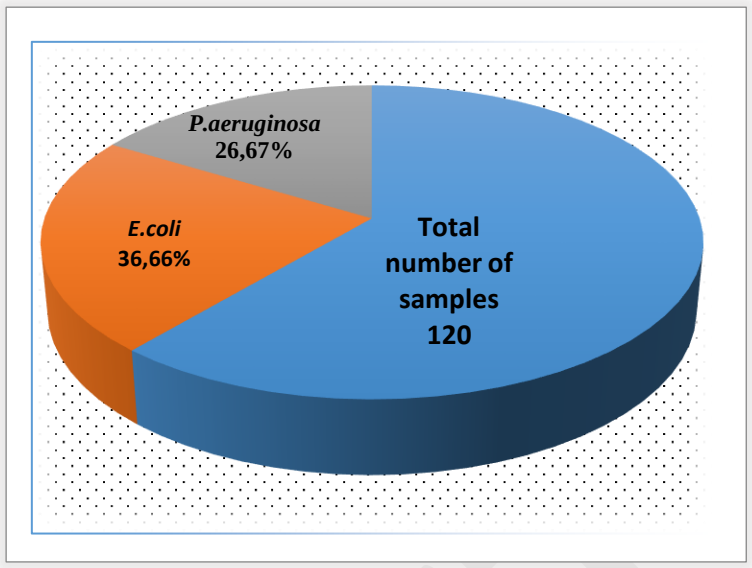


Figure 1. Isolated strains from 120 backyard poultry samples

Accordingly to the type of infection, all of the 44 *E.coli* strains were tested for their antibiotic-resistance performing antibiogram by disk diffusion method, Kirby Bauer 1966. The results presented in the table no. 2 evident high percentages of bacteria resistance against selected veterinary drug classes. Prominent results of antibiotic-resistance against Quinolones remain important in collibacillosis treatment.

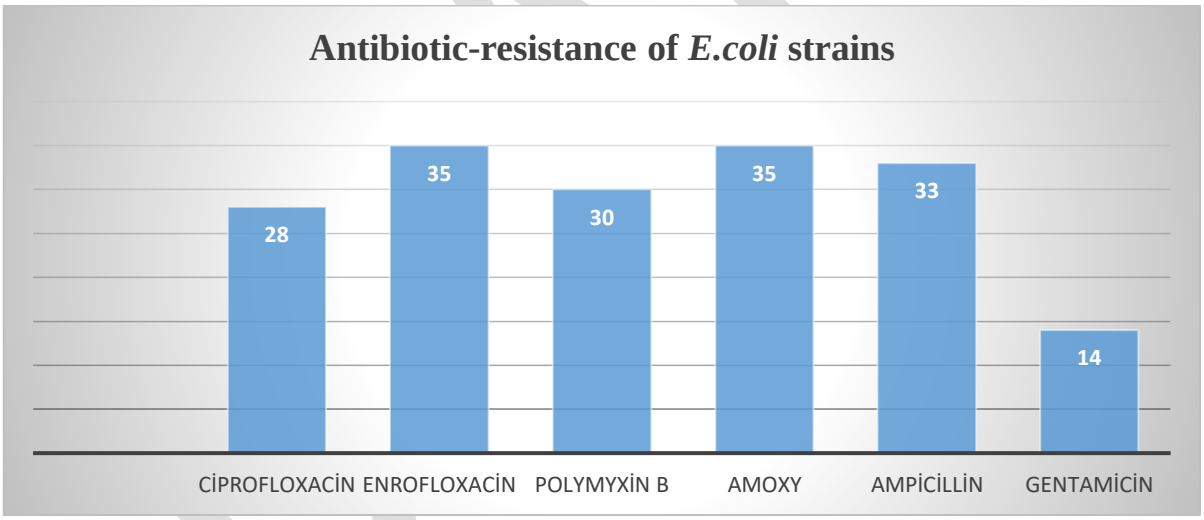


Figure 2. Antibiotic resistance for *E.coli* isolated strains

Pseudomona aeruginosa isolates were also investigated towards antimicrobial resistance. Important and elevated levels of resistance encountered in tested isolates highlighted the presence of 18 resistant strains against Polymyxin B or 56, 25% of all of the tested *P. areuginosa* isolates. Polymyxins are currently the last-resort antibiotics for the treatment of multidrug-resistant Gram-negative bacterial infections (Yang et al., 2023). Moreover, previous data for the human treatment of *P.aeruginosa* bacteremia have identified the efficasicity of Polymixin B in severe infections (Kvitko et al., 2011).

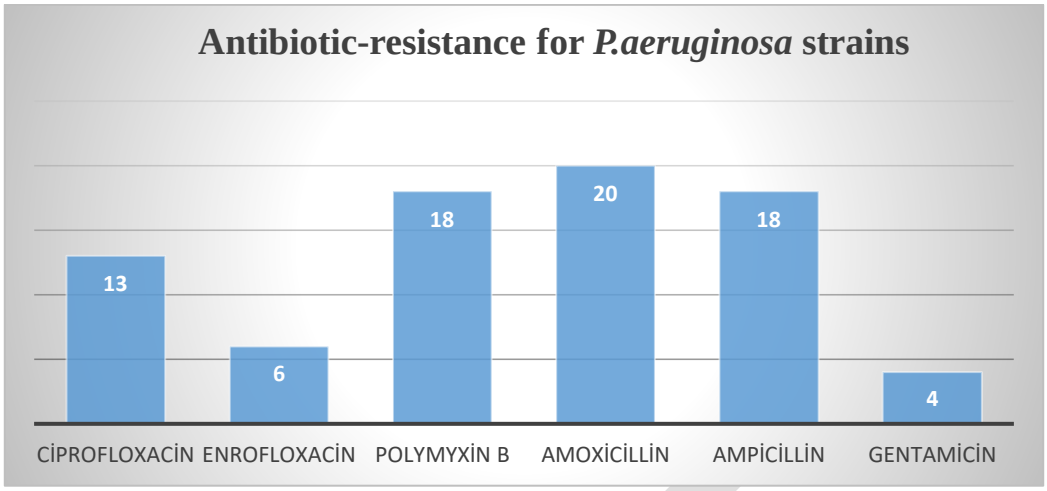


Figure 3. Antibiotic resistance of *P.aeruginosa* isolated strains

Nevertheless higher resistance percentages in *E.coli* isolated, sampled from clinically affected poultry and with evident colibacillosis lesions were presented, it is crucial to remark that similar antibiotic-resistance was detected in both bacteria, as shown in the figure no.4.

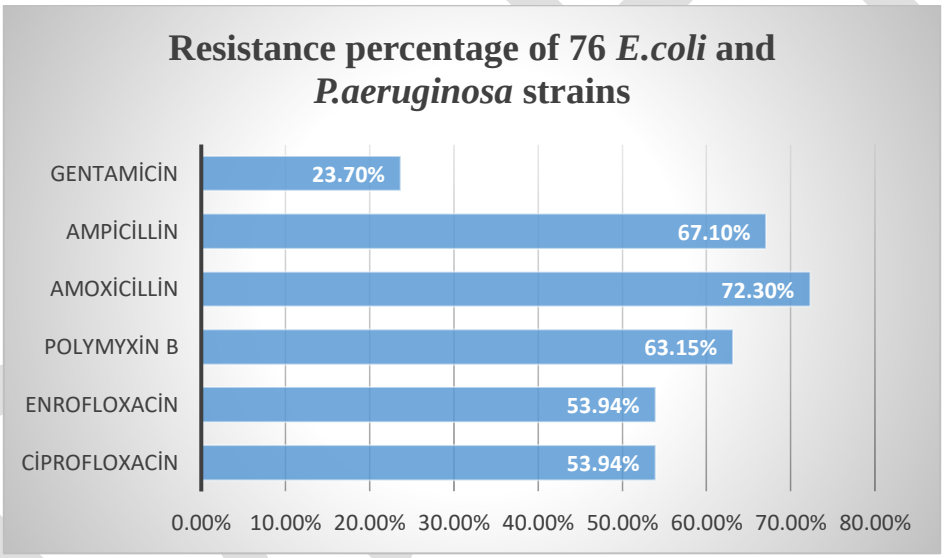


Figure 4. Resistance percentage of *E.coli* and *P. aeruginosa* strains

Disposal of animal wastes as a manure is another source of transmission of resistant pathogens from animal farms to the soil. This problem has been increased due to the concentrated animal feeding practices. As a result, resistant bacteria and antimicrobials residues accumulate in the soil and proliferate antibiotic resistance in soil inhabiting bacteria (Gosh and LaPara, 2007). It is important to note that both *Escherichia coli* and *Pseudomonas aeruginosa* can cause infections in poultry and can also be resistant to antibiotics (Varriale et al., 2020), (Nolan 2018). This study shows that backyard poultry are a potentional source of these pathogens and were both isolated in a high percentage which highlights the need for further investigations.

CONCLUSION

AMR in poultry is a complex issue that requires a multifaceted approach to address. Improved hygiene and sanitation, reduced use of antibiotics, and the use of non-conventional antimicrobial compounds are some strategies that can help reduce the prevalence of AMR in both commercial and backyard poultry farming. It is obvious that excessive use of antibiotics in poultry to increase the growth of broiler chicken is developing a serious sort of resistance in bacteria by production of antimicrobial resistance.

The conducted study in Elbasan district led to results similarities between different free-rage poultry farms. Thus it can be concluded that the pattern of resistance in the surrounding area of a poultry farm is highly influenced by mutual geographical area and choice of antibiotics used inside the farm.

In conclusion, high levels of antimicrobial resistance in *Pseudomonas aeruginosa* and *Escherichia coli* strains found in backyard poultry is a concern that requires immediate attention. The use of antimicrobials as growth promoters in small-scale chicken farming is a contributing factor to the development of AMR in poultry. To address this issue, it is important to promote judicious antimicrobial use in the poultry industry and to develop alternative strategies for disease prevention and control. Preventive measures have to be adopted in order to minimize the spread of antibiotic-resistant bacteria, especially *P. aeruginosa* in farm enviroinment and from poultry to humans.

REFERENCES

Alcaine SD, Molla L, Nugen SR, Kruse H. 2015 Results of a pilot antibiotic resistance survey of Albanian poultry farms. J Glob Antimicrob Resist. 2016 Mar;4:60-64. doi: 10.1016/j.jgar.2015.11.003. Epub 2015 Nov 30. PMID: 27436396

Ghosh, S., LaPara, T. The effects of subtherapeutic antibiotic use in farm animals on the proliferation and persistence of antibiotic resistance among soil bacteria. ISME J 1, 191–203 (2007). https://doi.org/10.1038/ismej.2007.31

Hedman, H.D.; Vasco, K.A.; Zhang, L. 2020. A Review of Antimicrobial Resistance in Poultry Farming within Low-Resource Settings. Animals 2020, 10, 1264. https://doi.org/10.3390/ani10081264

Hedman HD, Zhang L, Trueba G, Vinueza Rivera DL, Zurita Herrera RA, Villacis Barrazueta JJ, Gavilanes Rodriguez GI, Butt B, Foufopoulos J, Berrocal VJ, Eisenberg JNS. 2020. Spatial Exposure of Agricultural Antimicrobial Resistance in Relation to Free-Ranging Domestic Chicken Movement Patterns among Agricultural Communities in Ecuador. Am J Trop Med Hyg. 2020 Nov;103(5):1803-1809. doi: 10.4269/ajtmh.20-0076. PMID: 32876005; PMCID: PMC7646802.

Hedman HD, J.N.S. Eisenbergb, G. Truebac, D.L. Vinueza Riverac, R.A. Zurita Herrerac,J. Villacis Barrazuetac, G.I. Gavilanes Rodriguezc, E. Krawczykd, V.J. Berrocale, L. Zhangf,g 2019. Impacts of small-scale chicken farming activity on antimicrobial-resistant *Escherichia coli* carriage in backyard chickens and children in rural Ecuador One Health,Volume 8, 2019, 100112, https://doi.org/10.1016/j.onehlt.2019.100112

Kvitko CH, Rigatto MH, Moro AL, Zavascki AP. Polymyxin B versus other antimicrobials for the treatment of pseudomonas aeruginosa bacteraemia. J Antimicrob Chemother. 2011 Jan;66(1):175-9. doi: 10.1093/jac/dkq390. Epub 2010 Oct 20. PMID: 20961911

Nolan K. Lisa. 2019. Colibacillosis in Poultry, DVM, PhD, Department of Infectious Diseases, College of Veterinary Medicine, University of Georgia, Merck manual https://www.merckvetmanual.com/poultry/colibacillosis/colibacillosis-in-poultry

DRAFT

Nhung NT, Chansiripornchai N, Carrique-Mas JJ. Antimicrobial Resistance in Bacterial Poultry Pathogens: A Review. Front Vet Sci. 2017 Aug 10;4:126. doi: 10.3389/fvets.2017.00126. PMID: 28848739; PMCID: PMC5554362.

Shtylla Kika, Tana & Boci, Jonida & Cocoli, Sonila & Puvača, Nikola & Cabeli, Pranvera & Shoshi, Natalia & Camarda, Antonio. (2023). Avian Pathogenic Escherichia coli resistance patterns according to poultry species in Albania, a two years study. Albanian j. agric. sci. 2023; 22 (1): 1–6.

Varriale, L., Dipineto, L., Russo, T. P., Borrelli, L., Romano, V., D'Orazio, S., Pace, A., Menna, L. F., Fioretti, A., & Santaniello, A. (2020). Antimicrobial Resistance of Escherichia coli and Pseudomonas aeruginosa from Companion Birds. Antibiotics (Basel, Switzerland), 9(11), 780. https://doi.org/10.3390/antibiotics9110780

Yang, F., Zhou, Y., Bai, Y., Pan, X., Ha, U. H., Cheng, Z., Wu, W., Jin, Y., & Bai, F. (2023). MvfR Controls Tolerance to Polymyxin B by Regulating rfaD in Pseudomonas aeruginosa. Microbiology spectrum, 11(3), e0042623. https://doi.org/10.1128/spectrum.00426-23

DRAFT

POSTER PRESENTATION

In vitro biological evaluation on *Abies cilicica* (Ant. & Kotschy) Carr. subsp. *isaurica* Coode & Cullen methanolic extract

H. Tuba Kıyan[1] (*ORCID: https://orcid.org/0000-0001-5304-868X)*,
Melisa Manav[2] (*ORCID: https://orcid.org/0009-0000-6203-2102)*,
Melek Tekgöz[3] (*ORCID: https://orcid.org/0000-0003-2755-4655*),
Pervin Soyer[4] (*ORCID: https://orcid.org/0000-0002-6258-1993)*

[1]Anadolu University, Faculty of Pharmacy, Department of Pharmacognosy, 26470 Eskişehir, Turkey.
[2]Istanbul University, Faculty of Science, Department of Biology, 34134 Istanbul, Turkey.
[3]Eskişehir Technical University, Faculty of Science, Department of Biology, 26555 Eskişehir, Turkey.
[4]Anadolu University, Faculty of Pharmacy, Department of Pharmaceutical Microbiology, 26470 Eskişehir, Turkey.

*Corresponding author e-mail: pervinsoyer@anadolu.edu.tr

Abstract

Abies cilicica (Ant. & Kotschy) Carr. subsp. *isaurica* Coode & Cullen is an endemic species belonging to the Pinaceae family, commonly known as 'Bozkır Göknarı', distributed in the west of the Toros Mountains in Turkey. *A. cilicica* subsp. *isaurica* has antimicrobial, antiseptic, anti-inflammatory and antioxidant properties and is known to be used in the treatment of acne, ulcers, asthma and wound healing, bronchitis, stomach pain and digestive facilitator against various diseases. In this study, the phytochemical profile of *A. cilicica* subsp. *isaurica* leaf methanol extract was determined by HPLC system and within the scope of *in vitro* biological activities antimicrobial activity was investigated by microbroth dilution method, antioxidant activity by ABTS method and anti-inflammatory effect by COX-1, COX-2 enzyme inhibition method for the first time. The *A. cilicica* subsp. *isaurica* leaf methanol extract showed antimicrobial activity with the concentrations between 312,5-2500 µg/mL against Gram positive, Gram negative bacteria species and *Candida*. The leaf extract also exhibited high antioxidant and antinflamatory activity with 77.42% and 50.63% inhibition at the 200 µg/mL concentration.

Keywords: *Abies cilicica*, Phytochemical, Antimicrobial activity, Antioxidant activity, Anti-inflammatory activity.

INTRODUCTION

The genus *Abies*, belonging to the Pinaceae family, encompasses a total of 48 species and 78 taxa worldwide. In the specific context of Türkiye, there are 3 species and 5 taxa of *Abies* (Güner et al., 2012). Out of the taxa mentioned, two of them are considered endemic species. These species are classified as *Abies cilicica* subsp. *isaurica* Coode & Cullen lr (lc) and *Abies nordmanniana* subsp. *equi-trojani* (ASC. & Sint. ex Boiss.) Coode & Cullen, falling under the LR (nt) category (Pulatoğlu et al., 2023). *Abies cilicica* (Ant. & Kotschy) Carr. subsp. *isaurica* Coode & Cullen is an endemic species commonly known as 'Bozkır Göknarı', distributed in the west of the Toros Mountains in Türkiye (Yavaser et al., 2015). The resin derived from *Abies cilicica* has historically been employed for its medicinal properties, serving as an antiseptic, anti-inflammatory, antipyretic, antibacterial, and antiviral agent. In our country, it is known that the resin obtained from the cones of *A. cilicica* species is used externally as antiseptic and boil remedy as moxibustion or ointment (Baytop, 1999). In the treatment of gastrointestinal diseases, psoriasis, reflux, ulcers, asthma, bronchitis (Çakır, 2017) (Tumen et al., 2011) (Tumen et al., 2011), the resin contained in the cones of *A. cilicica* subsp. *isaurica* is used in the treatment of acne, ulcers, asthma and wound healing (Yeşilada et al., 1995) (Yavaser et al., 2015) (Çakır, 2017),

DRAFT

against psoriasis, bronchitis, stomach pain and as a digestive facilitator (Çakır, 2017). It is known that tar is used as antiseptic, in the treatment of abscess, abdominal pain and as a digestive facilitator (Yavaser et al., 2015). It has also been reported that its fruits (cones) are used as decoction in the treatment of vascular diseases (Yeşilada et al., 1995) (Tumen et al., 2011). Because of these properties, this study focused on determining the biochemical contents of *Abies cilicica* subsp. *isaurica* leaves methanol extract and examining the biological activities in the context of antimicrobial, antioxidant, anti-inflammatory properties.

MATERIALS AND METHODS

Preparation of Extract

The leaves of the endemic *A. cilicica* subsp. *isaurica* species were collected from Alanya (Antalya) and Fethiye (Muğla) in Antalya and Muğla regions in 2017 and identified by Forest Engineer Mahmut Tunahan. Methanolic extract of the leaves of *A. cilicica* subsp. *isaurica* were obtained by maceration with methanol at room temperature. The solvent was removed by using a rotary evaporator and dry extract were obtained.

Liquid-Chromatographic Analyses

The methanolic extract was analysed by LC-MS/MS (Liquid Chromatography-Mass Spectrometry/ Mass Spectrometry) system to assess its phytochemical composition.

In vitro Anti-inflammatory Activity Test by COX-1 and COX-2 Enzyme Inhibition Method

Purified COX-1 and COX-2 enzymes were used for *in vitro* determination of anti-inflammatory COX-1 and COX-2 inhibitory effect. The amount of PGE2 was measured by EIA (Vazquez et al., 1997). The activity results were determined colorimetrically in Elisa microplate reader and given as % inhibition.

Antimicrobial Activity Test

The Clinical and Laboratory Standards Institute's (CLSI) microbroth dilution susceptibility assay (CLSI 2012, 2002a) was used to test the antimicrobial activity of plant extract. *Escherichia coli (*ATCC 35218), *Bacillus subtilis (*NRRL B478), *Pseudomonas aeruginosa* (ATCC 27853) strains were subcultered overnight at 37 °C in Mueller Hinton Agar (MHA) and *Candida albicans* (ATCC 10231) in Sabouraud Dextrose Agar (SDA) at 30 °C. Overnight microorganism cultures were adjusted to 1×10^8 CFU/mL using McFarland 0.5 standard turbidity in sterile saline (0.85%) solution and homogenized by vortex. The plant extract was dissolved in methanol for the initial stock solution. The concentrations of plant extract was prepared by a two-fold serial dilution technique in 96-well microplate to 2500-4 µg/mL. After the dilution step, 100 µL of each microorganism culture were inoculated to the well plates and incubated at 37 °C, 24 h for bacteria and at 30 °C for 48 h for yeast. At the end of the incubation period, all wells were stained by adding 20 µL 0.01% resazurin solution to observe the color difference between dead and living cells (colors ranging from blue (green) to pink). Ketoconazole, ciprofloxacin and chloramphenicol were used as reference agents for antimicrobial activities against *C. albicans* and bacteria species. The experiment was repeated three times, and the results were averaged.

Antioxidant Activity Test

The antioxidant activity of plant extract was determined by using ABTS (2,2′-azinobis(3-ethylbenzothiazoline-6-sulfonic acid)) (Re et al., 1999) method. ABTS (7 mM) was dissolved in water with 2.45 mM potassium persulfate and the solution was kept in the dark room for 16 h. Standardized ABTS solution was prepared with ethanol to obtain 0.70±0.002 absorbances. Stock solutions of plant extract (200 µg/mL), ascorbic acid (100 µg/mL) and gallic acid (100 µg/mL) were prepared in ethanol. The ethanol was used as negative control and ascorbic acid (Vit C), gallic acid were used as a positive control. The radical scavenging activity (RSA) was calculated as the percentage inhibition of ABTS radical reduction as follows "%ABTSscavenging =[(*A*control−*A*sample)/*A*control]×100"

RESULTS and DISCUSSION

Liquid-Chromatographic Analyses

For LC-MS/MS analysis, Absciex 3200 MS/MS detector was used. Chromatographic separations were performed using Shimadzu 20A HPLC with an ODS 150 x 4.6 mm x 3 um column. The column oven temperature was set to 40 °C and the flow rate was set to 0.5 mL/min. Mobile phases were (A) Methanol: water: formic acid (10:89:1, v/v/v) and (B) Methanol: water: formic acid (89:10:1, v/v/v). The concentration of B was increased from 10% to 100% in 40 min. A mass range of 100-1000 amu was chosen for mass screening (EMS). The results were given in Table 1.

Table 1. Compounds identified in methanol extract

Rt	$[M-H]^-$	MS/MS	Identification
3.5	191	173, 127	Quinic acid
9.4	163	119	Coumaric acid
14.0	431	385, 223, 205,153	Rosaceous-like
16.2	315	299, 269, 255	Methylquercetin
17.0	433	343, 313	Naringenin-C-glucoside
18.0	577	456, 413, 311, 293	Apigenin-O-rhamnose-C-glucose
20.1	463	315, 300, 271	Methylquercetin rhamnoside
21.0	461	300, 271, 179, 151	Quercetin glucoside
22.8	345	301, 257, 179	Unknown
23.3	447	300	Quercetin rhamnoside
26.0	431	285, 255, 227	Luteolin/campferol rhamnoside
32.4	739	593, 453, 285	Kemferol/luteolin deoxyhexose derivative
37.0	723	577, 559, 437, 285	Kempferol/luteolin deoxyhexose derivative

COX-1 and COX-2 Enzyme Inhibition Test

The anti- inflammatory activity of *A. cilicica* subsp. *isaurica* leaf methanol extract was determined by COX-1 and COX-2 Enzyme Inhibition Test and % inhibition rates were determined (Table 2.). The 50.63% inhibition activity was determined at the 200 µg/mL concetration. When compared with the control group Selekoksib, the result was satisfactory.

DRAFT

Table 2. COX-1 and COX-2 Enzyme % inhibition rates

Concentration	**100 µg/mL**	**200 µg/mL**
	% Inhibition	
A. cilicica subsp. *isaurica* leaf methanol extract	15.00	50.63
Control group Selekoksib	**90.54**	**91.83**

Antimicrobial Activity Test

The MIC values of *A. cilicica* subsp. *isaurica* leaf methanol extract were given in Table 3. The minimum inhibitory concentrations of plant extract varied between 312,5-2500 µg/mL. According to the MIC values methanol extract of *A. cilicica* subsp. *isaurica* leaf exhibited poor antimicrobial activity against *Escherichia coli* and *Bacillus subtilis* with high concetrations. As a result, only the *Pseudomonas aeruginosa* is more sensitive than other bacteria species, similarly with *Candida albicans*.

Table 3. MIC values

	MIC µg/mL			
	Escherichia coli	*Bacillus subtilis*	*Pseudomonas aeruginosa*	*Candida albicans*
A. cilicica subsp. *isaurica* leaf methanol extract	2500	625	312,5	312,5
Ciprofloxacin	-	-	4	-
Ketoconazole	-	-	-	312,5
Chloramphenicol	8	4	-	

Antioxidant Activity Test

The methanol extract of *A. cilicica* subsp. *isaurica* leaf exhibited high antioxidant activity compared with control groups of gallic and ascorbic acid (Table 4). The 77,42% inhibition activity was determined at the 200 µg/mL concetration.

DRAFT

Table 4. ABTS % inhibition rates

	ABTS % inhibition	**Concentrations**
A. cilicica subsp. *isaurica* leaf methanol extract	77,42	200 µg/mL
Gallic Acid	88,28	100 µg/mL
Ascorbic Acid	84,23	100 µg/mL

CONCLUSION

In this study, the phytochemical composition of methanol extract prepared from the leaves of *A. cilicica* subsp. *isaurica* was investigated for the first time with HPLC system. In addition, the *in vitro* anti-inflammatory effect of the extract was examined for the first time using COX-1, COX-2 enzyme inhibition methods. The antimicrobial and antioxidant activities were also evaluated in this study. It is thought that the effects can be evaluated by *in vivo* animal experiments as a preliminary preparation for clinical studies.

ACKNOWLEDGEMENTS

This study was financed by Anadolu University Scientific Research Project Foundation (No: 1906S121).

REFERENCES

Baytop T 1999. Therapy with medicinal plants in Turkey (past and present). *Publication of the istanbul University*, *312*, 2-3.

Çakır EA 2017. A comprehensive review on Ethnomedicinal utilization of gymnosperms in Turkey. Eurasian J. For. Sci. 5, 35–47.

Clinical and Laboratory Standards Institute M07-A9 2012. Reference Methods for Dilution Antimicrobial Susceptibility Tests f or Bacteria That Grow Aerobically; Approved Standard- Ninth Edition, *CLSI document* M07-A9 *29(2).*

Clinical and Laboratory Standards Institute M27-A2 2002a. Reference Method for Broth Dilution Antifungal Susceptibility Testing of Yeasts; Approved Standard-Second Edition, *CLSI document 22(15).*

Güner A, Özhatay N, Ekim T, Başer KHC, & Hedge IC. (Eds.) 2000. *Flora of Turkey and the east Aegean Islands*. Edinburgh University Press.

Haliloglu Y, Ozek T, Tekin M, Goger F, Baser KHC, Ozek G 2017. Phytochemicals, antioxidant, and antityrosinase activities of Achillea sivasica Çelik and Akpulat. Int. J. food Prop. 20, S693–S706.

Pulatoğlu AÖ, Güney K, Çeter T, Yılmaz ES 2023. Chemical Composition of Essential Oils Obtained from Abies taxa in Türkiye and Investigation of Antimicrobial Activities. *Kastamonu University Journal of Forestry Faculty*, *23*(1), 31-46.

Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, & Rice-Evans C 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free radical biology and medicine*, *26*(9-10), 1231-1237.

Şeker Karatoprak G, Göger F, Yerer MB, Koşar M 2017. Chemical composition and biological investigation of Pelargonium endlicherianum root extracts. Pharm. Biol. 55, 1608–1618.

Tumen I, Akkol EK, Süntar I, Keleş H 2011. Wound repair and anti-inflammatory potential of essential oils from cones of Pinaceae: preclinical experimental research in animal models. J. Ethnopharmacol. 137, 1215–1220.

Vazquez MT, Rosell G, Pujol MD 1997. Synthesis and anti-inflammatory activity of rac-2-(2, 3-dihydro-1, 4-benzodioxin) propionic acid and its R and S enantiomers. Eur. J. Med. Chem. 32, 529–534.

Yavaser R, Erkus H, Sunna C, Karagozler AA 2015. Evaluation of antioxidant and antimicrobial activity of Abies Cilicica (Ant&Kotschy) subs: isaurica coodle&Cullen resi (). Eur. J. Biotechnol. Biosci. 3, 37–44.

Yeşilada E, Honda G, Sezik E, Tabata M, Fujita T, Tanaka T, Takeda Y, Takaishi Y 1995. Traditional medicine in Turkey. V. Folk medicine in the inner Taurus Mountains. J. Ethnopharmacol. 46, 133–152.

POSTER PRESENTATION

Impedance Spectroscopy Analysis of an Electrochemical Cell Chamber for Diagnostics

Mehmet Yuksekkaya[1*] (*ORCID: https://orcid.org/0000-0002-2665-5799*), Utkan Demirci[2] (*ORCID: https://orcid.org/0000-0003-2784-1590*), Mustafa Kocakulak[3] (*ORCID: https://orcid.org/0000-0001-5029-0104)*

[*1]Ankara University, Faculty of Engineering, Department of Biomedical Engineering, Ankara, Türkiye.
[2]Stanford University, Canary Center for Cancer Early Detection, Department of Radiology, Bioacoustic MEMS in Medicine (BAMM) Labs, Stanford, USA.
[3]Izmir Democracy University, Faculty of Engineering, Department of Biomedical Engineering, Izmir, Türkiye.

*Corresponding author e-mail: myuksekkaya@ankara.edu.tr

Abstract

In biomedical diagnostics, electrochemical cells are important tools. This study explains the application of electrochemical cells in biomedical diagnosis, focusing on the modulation of impedance information at different frequencies as a function of cell fluid content. The relationship between the sample's composition and the chamber's reaction dynamics with impedance at distinct frequencies is examined. Also, a novel, practical and low-cost chamber design is presented.

To explain the underlying principles governing the electrochemical cell's behavior, an electrical equivalent circuit model for the chamber is proposed. The chamber's ionic fluid content for different concentrations is monitored through impedance spectroscopy. This study showed that electrochemical impedance spectroscopy can be used as economic and efficient tool for biomedical diagnostics with implications for a wide range of applications in the field of healthcare and life sciences.

Keywords: Impedance spectroscopy, microfluidics, biosensor, diagnosis, electrochemical chamber

INTRODUCTION

Electrochemical Cell

A basic Electrochemical impedance measurement setup consist of chamber with two electrodes in an electrolyte solution. The impedance for different alternating current (AC) signal with a constant frequency is dependent on several factor due to the solution dynamics and reactions. These factors include ion migration, the presence of free electrons in the solution, and the system's capacitive response. Electrical impedance spectroscopy is the impedance value for various frequencies. (Lasia, 2014) The resistivity of the bulk solution, permittivity of the solution, geometric electrode placement, electrode-solution interface, electrochemical processes occurring on the electrode and electrolyses have an effect on the impedance value in the two electrode systems. (Grimnes et al., 2005)

The data obtained from the EIS system is usually an impedance data for a specific frequency. An electrical equivalent model of an electrochemical system allows for more thorough data analysis, which may provide information about the structure of the materials in the electrochemical cell and the reactions formed in it.

Electrical Equivalent Model of a Cell

The design of the system, such as geometric placement of the electrodes and resistivity and permittivity features of the solution, makes bulk resistance and the capacitance of the model simply definable.

There is a relationship between resistivity and the quantity and mobility of the unbound electrons in a substance. Additionally, based on an electrical field applied to the substance, there is a relationship between permittivity and polarization. If the elements in the solution are changed, the resistivity and permittivity of the solution vary.

DRAFT

Different types of geometric configurations of two electrode systems exist, which are forward looking electrodes, co-planar electrodes and interdigitated electrodes. To reveal particular characteristics of electrical information, alternative electrode configurations may be utilized.

According to the description of the electrical equivalent circuit model, the experimental data and the equivalent circuit data must fit. The constituent parts of the electrical equivalent circuit model of the electrochemical cell should be evaluated based on an analysis and comprehension of the physical and chemical characteristics of the electrochemical system. (Krause, 2007)

In the electrical field, ion migration based on electrode charges leads to a double layer of charge on the electrode's surface, separated by a thick dielectric. This structure is called double layer capacitance (C_{DL}), which depends on frequency, and it is influenced by factors such as electrode potential, temperature, ion types, ionic concentrations, impurity adsorption, oxide layers, electrode roughness, electrode potential, and so on. (Luo, 2013 and Genry, 2017)

When surfaces are inhomogeneous, rough, or porous, which leads to observing frequency-dependent behaviour, since ideal capacitor model is suitable, constant phase element (*CPE*) is employed to model the actual double layer capacitance. α means phase coefficient, which represents the behaviour of CPE. (Hong et al., 2005 and Córdoba et al, 2015)

The ratio of the applied voltage to the current resulting from the charge transfer is defined as the charge transfer resistance (R_{CT}). The resulting current is influenced by the transferred ion concentration, potential formed during electrochemical reactions and reaction products. (Lvovich, 2012)

The impedance occurring because of the ion migration and diffusion in the diffusion layer stands for Warburg impedance (Z_W). It is strongly influenced by the frequency of the applied voltage. Higher frequencies result in lower Z_W because of faster ion migration or vice versa for lower frequencies. (Lvovich, 2012)

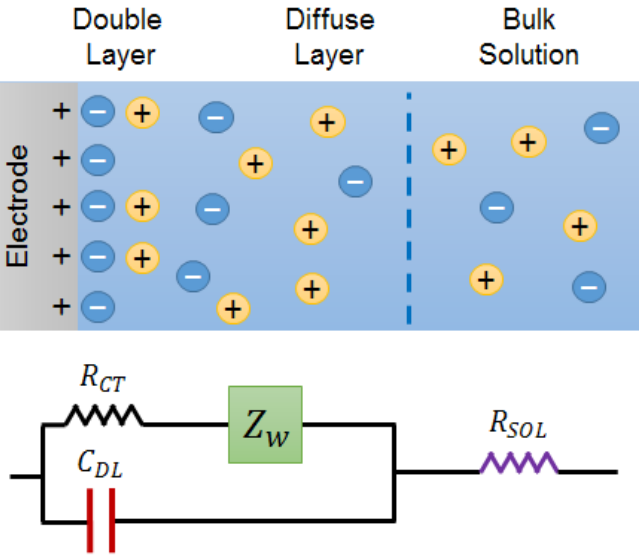


Figure 1. Randles electrical equivalent circuit model for an electrode. R_{CT} is the charge transfer resistor, Z_W is Warburg impedance, C_{DL} is the double layer capacitance, and R_{sol} is bulk resistance. R_{CT} and C_{DL} are related to the electrode-electrolyte interface and the reactions in that area that include the linear, semi-infinite diffusion of electroactive particles between electrode and electrolyte.

The resistance of the bulk solution can be easily clarified for higher frequencies in Bode plots of Randles electrical equivalent circuit model. In Nyquist plot, the imaginary versus real components of the impedance value graph can be plotted for the Randles equivalent circuit model, and the resulting graph includes information about both phase and magnitude. Nyquist and Bode plots provide the best visualization and description of the Randles model. (Randviir and Banks, 2013)

Lower frequencies have a linkage with the diffusion limited process, and high frequencies have a linkage with the electron transfer limited process. At high frequencies, R_{sol} value is obtained. Warburg impedance increases

its visibility only at lower frequencies. On inhomogeneous, rough, or porous surfaces, CPE replaces C_{DL}. (Ahmed et al., 2014)

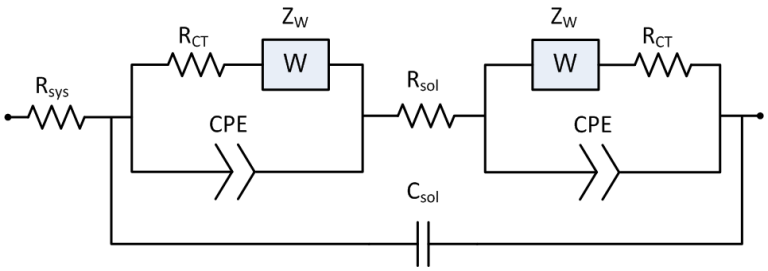


Figure 2. Randles electrical equivalent circuit model for full electrochemical system with two identical electrodes. R_{sys} is ohmic resistance of electrical wiring and C_{sol} is bulk capacitance.

In Figure 2 C_{sol} is significant. The impedance is determined predominantly by a co-planar electrode system's capacitance, which relies on material, geometry, and permittivity of solution. The capacitance value has a big effect on system's impedance, regardless of whether the capacitance value is high or low.

Impedance Spectroscopy

Electrochemical biosensors behave as a detector that can include EIS, and they have three different types according to their sensing method: impedimetric, amperometric, and potentiometric. Besides all these techniques that may contain EIS, impedimetric one mainly uses it. (Ahmed et al, 2014)

Biosensors based on EIS, which can be easily downsized, rapid, sensitive, and affordable, can be used in studies to determine the structural features of the solution/material that influence the permittivity and conductivity, or electrochemical reactions such as enzyme-catalysed reactions or specific binding reactions of some species on the surface of modified electrodes or in the solution. (Bahadır and Sezgintürk, 2016)

Pathogen and cell detection are also possible. Studies by Cheng et al. (2007), Shafiee et al. (2013), Toner et al. (2010), and Demirci et al. (2014) regarding impedance spectroscopy-based cell and virus detection and tracking with cell and virus lysate have been found in the literature.

MATERIALS AND METHODS

Chamber Design

Polymer-based microfluid chips are well suited to move the impedance measurement processes forward. The design of the chips is made up of three layers; the bottom polymer layer with electrodes on it, the medium layer with double sided adhesive and a patterned channel, and the top polymer layer with inlet and outlet patterns.

Electrode Layer

The screen-printing technique is used to obtain the electrode layer and a silver paste mixture, which has highly conductive silver ink composition and is silicon adhesive, is applied on the flexible polymer surface. With a flexible hydrophobic polymer film from the protective cover film of double-sided adhesive (DSA), a mask pattern is formed. The pattern is created with the help of computer-aided design software, and a laser cutter is used. The mask is first positioned on a flexible hydrophilic polymer. On one side of the mask, the silver paste mixture is dropped. The mixture is spread on the mask by using a microscope glass slide as a squeegee. The open spaces are filled with the mixture.

After the screen-printing process, the mask is carefully taken away continued in baking at 80° for sixty minutes. The electrodes are ready at the end of the baking process. It is possible that at the end of the process, errors are unavoidable since the laser cutter cannot cut the polymer in its actual dimensions, and baking has also an effect on the size of the electrodes.

Channel layer

The channel layer, which is patterned on DSA and created by a CAD software, is to form a cavity for solution to be filled, and using constant volume of solution is crucial in impedance measurements.

Inlet layer

The inlet layer, which is designed in CAD software, is formed with the flexible hydrophilic polymer cut by the laser cutter. The chamber is assembled by removing the protective film from one side of DSA so that it is laminated.

In order to minimize errors, this process is repeated numerous times. There is no requirement to use complex and expensive devices during the process. The steps may be done in any environment.

Experimental Setup

Probes of LCR meter, which is a device to measure the impedance, are attached to the electrodes of the chip, the sample solution is added into the channels, and magnitude and phase of impedance of the solution is recorded. The input AC potential is 1 peak-to-peak voltage (V_{pp}) and AC frequency is between 100 HZ and 1 MHz. In this paper, the maximum difference value for impedance change is approximately 1 kHz, which means that the magnitude of impedance is 1 kHz.

RESULTS

Even if the main purpose of this paper is to detect analytes, known conductance-valued solutions are measured firstly in order to obtain better knowledge about the system and impedance results. 1% Triton 100-X solution in DI pure water is used as control solution. Six test sample solutions, which are prepared at 0.05%, 0.1%, 0.25%, 0.50%, 1.0%, and 5.0% PBS in DI pure water, exposure to a conductivity test by using a conductivity meter.

Figure 3 (a) shows that different solutions with different concentrations have different conductivity levels. The determination of the resistivity for test samples and the control solution is done by impedance spectra measurements at a frequency range between 100 Hz and 1 MHz.

All the test samples and the control solutions are tested according to the procedure. For a solution 3 FFChips are used, and all the sample volumes are same with each other. The average of three impedance magnitude spectra measurements for a solution is calculated and this step is applied for all other solutions. According to the results, the Nyquist plot in Figure 4 is sketched.

DISCUSSION

The definition of one of the electrical models of the FFChip was made, along with the background of the electrical impedance spectroscopy analysis system. DI and some concentrations of PBS solutions filled channel measurements were done. The results of these known-concentrated solutions demonstrated that the system is capable of detect even $k\Omega$ of differences.

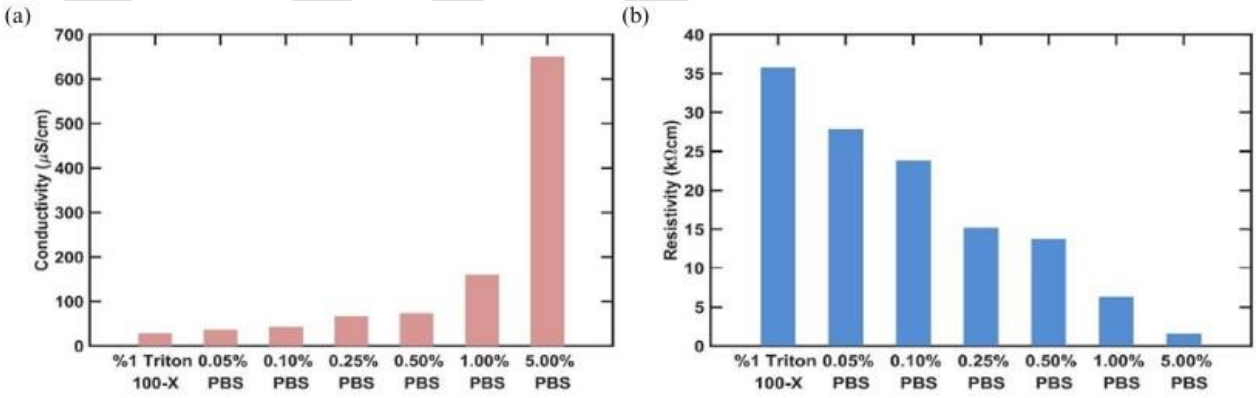


Figure 3. Test values a) Conductivity test values at various PBS concentrations b) Resistivity test values at various PBS concentrations.

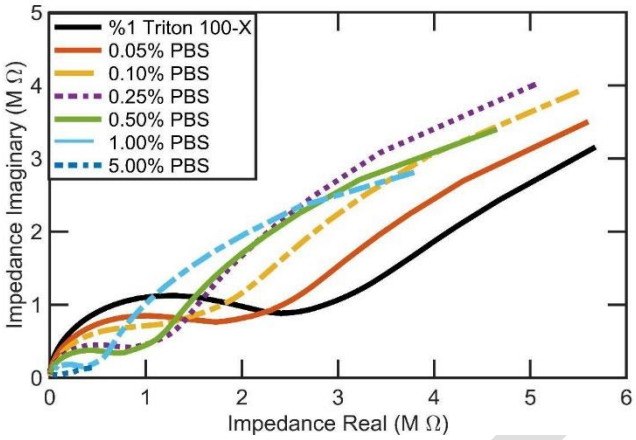


Figure 4. Nyquist plot of impedance values of control solution and different PBS concentration-solutions for a frequency range between 100 Hz and 1 MHz.

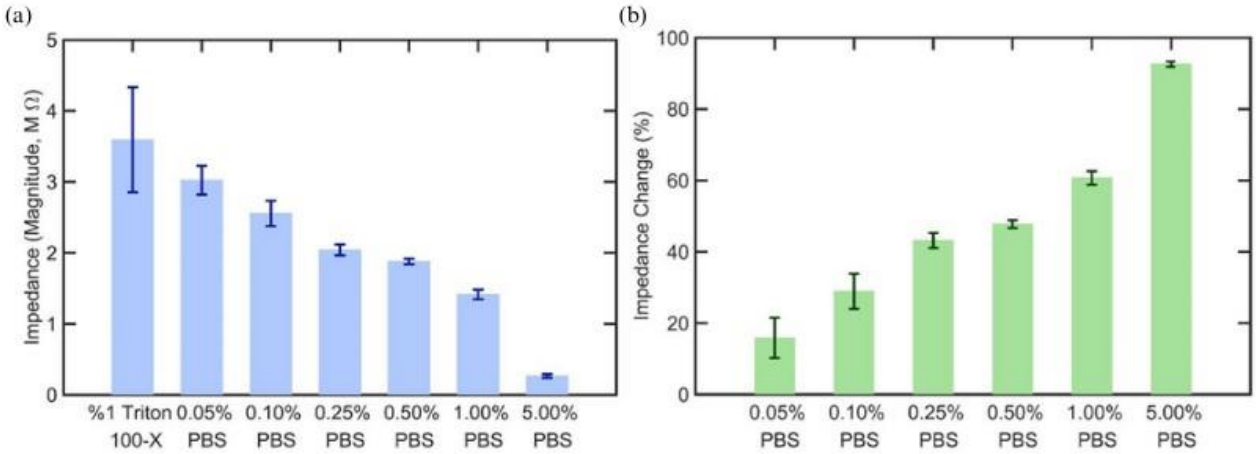


Figure 5. a) Impedance magnitude values of all the solutions at 1 kHz. Error bars represent standard deviations (STD) of the mean of impedance magnitude. (n=3), b) The impedance magnitude changes of all the sample test solutions with respect to average control value at 1 kHz. Error bars represent standard deviations (STD) of the mean of impedance magnitude. (n=3)

CONCLUSION

Lightweight, flexible, thin, affordable, easy to manufacture, disposable and mass-producible features make the flexible polymer film effective material for biosensors. With its portability, sensitivity, and rapid property, electrical impedance spectroscopy is efficacious in detection methodology.

ACKNOWLEDGMENTS

We would like to thank The Scientific and Technical Research Council of Turkey (TUBITAK) for providing financial support.

REFERENCES

Ahmed A., Rushworth J. V., Hirst N. A., Millner P. A. 2014. Biosensors for whole-cell bacterial detection. *Clinical microbiology reviews*, *27*(3), 631-646.

Bahadır E. B., Sezgintürk M. K. 2016. A review on impedimetric biosensors. *Artificial cells, nanomedicine, and biotechnology*, *44*(1), 248-262.

Cheng X., Liu Y. S., Irimia D., Demirci U., Yang L., Zamir,L., ..., Bashir R. 2007. Cell detection and counting through cell lysate impedance spectroscopy in microfluidic devices. *Lab on a Chip*, *7*(6), 746-755.

Córdoba-Torres P., Mesquita T. J., Nogueira R. P. 2015. Relationship between the origin of constant-phase element behavior in electrochemical impedance spectroscopy and electrode surface structure. *The Journal of Physical Chemistry C*, *119*(8), 4136-4147.

Demirci U., Shafiee H. 2014. *System and method for detecting pathogens* (U.S. Patent)

Gamry 2017. *Basics of EIS: Electrochemical Research-Impedance*. Available at: https://www.gamry.com/application-notes/EIS/basics-of-electrochemical-impedance-spectroscopy/ [17.09.23]

Grimnes S., Martinsen O. E. 2005. *Bioimpedance and Bioelectricity Basics*: Academic Press

Hong J., Yoon D. S., Kim S. K., Kim T. S., Kim S., Pak E. Y., No K. 2005. AC frequency characteristics of coplanar impedance sensors as design parameters. *Lab on a Chip*, *5*(3), 270-279.

Krause S. 2007. *Impedance Methods* in *Encyclopedia of Electrochemistry*, ed: Wiley-VCH Verlag GmbH & Co. KGaA

Lasia, A. 2014. Definition of impedance and impedance of electrical circuits. *Electrochemical Impedance Spectroscopy and its applications*, 7-66.

Luo X., Davis J. J. 2013. Electrical biosensors and the label free detection of protein disease biomarkers. *Chemical Society Reviews*, *42*(13), 5944-5962.

Lvovich V. F. 2012. *Impedance spectroscopy: applications to electrochemical and dielectric phenomena*. John Wiley & Sons.

Randviir E. P., Banks C. E. 2013. Electrochemical impedance spectroscopy: an overview of bioanalytical applications. *Analytical methods*, *5*(5), 1098-1115.

Shafiee H., Jahangir M., Inci F., Wang S., Willenbrecht R. B., Giguel F. F., ..., Demirci U. 2013. Acute on-chip hiv detection through label-free electrical sensing of viral nano-lysate. *Small*, *9*(15), 2553-2563.

Toner M., Bashir R., Cheng X, Demirci U., Irimia D., …, Rodriguez W. 2010. Methods for counting cells (USA Patent No: EP2156376 A1)

DRAFT

POSTER PRESENTATION

Comparative study of the diet of Oedipoda miniata in two regions of Tlemcen (Algeria)

Benmansour Bouchra Salima

University of Tlemcen

benmansoursalimab@yahoo.fr

Abstract

The present work, of economic importance, concerns the diet of an Orthoptera species, Oedipoda miniata, in the natural conditions of 02 stations, in the region of Tlemcen (Algeria): the first in Sebdou (south of the wilaya of Tlemcen), and the other located at Moutass (South West of Tlemcen). For this, a floristic inventory of the plant species of the 02 study stations was carried out. This allowed us to highlight the main families. The faunistic inventory of Orthoptera reveals the presence of species belonging to the Acrididae family and to the Caelifera Order. The diet of Oedipoda miniata was performed by microscopic analysis of the faeces content. This is a method that is so far the most objective. Moreover, it does not disturb the demographic balance of the populations. The study of the diet allowed to observe in faeces a limited number plant species, consisting mainly of poaceae and lamiaceae. The interest of the study of the locust diet is to understand the phenomenon of competition and the existing relationship between the plant and the insect. In the wild, it is the means of knowing if the locust chooses its food facing a diversity of plants, or is it a question of availability and quantity.

Key words: Diet, Orthoptera, Oedipoda miniata, Sebdou, Moutass, Tlemcen, Algeria.

Introduction

Food is one of the most important ecological factors in various biological parameters orthopteran populations namely, the fertility, longevity, speed of development and birth rate **(DAJOZ, 1982).**
DAJOZ (1971), considers food as a unique source of energy available to insects; it is
obviously a limiting factor when it is in insufficient quantity. In addition, he notes that the diet of a species is rarely constant throughout the year and in all places.
According to **GILLON(1983),** the use of food resources varies according to the environment in which the locust lives. The choice of the host plant is based not only on the insect-plant biochemical relationships but also on the structure of the environment. Similarly, **MOUMEN (1997)** states that the behavior of insects in the selection of the food substrate is a change in the opportunity to consume a plant rather than other. In fact, the choice of a plant by an insect depends on the presence of the substances stimulating or inhibiting food intake.
In the region of Tlemcen, more than 50 species have been described by **MESLI (2007)** and **DAMERDJI (2008)**. Most of these taxa belong to the family Acrididae including Oedipoda miniata. According to MESLI (2007), aromatic and medicinal plants form a major part of the locust diet. CHAPMAN (1982) noted that the majority of phytophagous insects have more than 50% of oligophagous species or monophages (feeding on a single family of plants), against against Orthoptera and in particular locusts, 60% of species are called polyphagous and 25% are called graminivores. PICAUD et al., (2003) mention that rare are specialist Orthoptera species and that graminivorous species make their food choices because of the high content of grasses in sugar.

The study of the diet allowed us to know the trophic behavior of Oedipoda miniata. In the wild, this tells us if a locust attacks the most abundant plants or it remains a matter of preference. Thus, we used the method of the examination of the contents of faeces which is to compare the fragments of epidermis of the plants ingested by the insect with those of a reference collection prepared from existing plant species in its biotope. The determination of the diet, by the microscopic analysis of the contents of the faeces remains until now the most objective. Moreover, this method does not disturb the demographic balance of populations. It is in this order of ideas that we chose to conduct this research.

Material and methods

Choice of stations

The choice of stations is based on the altitude parameter. Our study areas pass through the Tlemcen Mountains (Moutass) to the steppe region (Sebdou).

Station 01: Moutass (Tlemcen Mountains)

This station is located west of Tlemcen, with an exposure of 34.44'59.18"N and 1 ° 29'10.35"W, and an altitude of 1069 m. The recovery rate is 80%.

The climate of the region of Tlemcen among others Moutass, is of Mediterranean type, semi-arid in temperate winter, rainy in winter and dry in summer. Prolonged summer drought and erratic rainfall are ecological factors limiting and threatening the region's plant structures on a continuous basis.

Station 02: Sebdou

Sebdou is located south of Tlemcen wilaya, about 38 km. It is lies between latitude 34 ° 38 'N and longitude 1 ° 19' W. This station has an exposure of 34.36'49.16"N and 1 ° 20'04.24"W, with an elevation of 966 m and a recovery rate exceeding 75%.

The climate of Sebdou is arid marked by two seasons: a winter which is short characterized by the irregular rainfall with mild temperatures; the other summer, long, dry, characterized by high temperatures and a significant lack of precipitation.

Study technique

Samples are taken during the year 2015. The frequency of the exits is of once a month of two hours in each station, during the hot and sunny hours, a clear sky and a weak wind.

Two preliminary exits are carried out at least from December 2014 for vegetation sampling to establish a reference epidermotheca.

Methods of studying vegetation

For the study of floristic biodiversity, an exhaustive sampling was carried out in order to inventory all the plant species that exist in the 02 stations.

Thus, we have established the floristic characterization of the vegetation of the studied stations, from biological, morphological and geographical point of view. In addition, the development of an epidemotheque of references was duly realized.

Epidermothèque référence preparation

Plant samples (leaves and stems) were collected at the study stations and returned to the laboratory. The fresh cuticles are gently removed from the underlying tissues, under the binocular magnifying glass, using fine forceps, or by placing the cuticle to be studied in contact with a glass slide, then scratching the other face until internal tissue (Benhalima, 1983). Several methods of preparation of epidermis are indicated such as those of STEWART (1965) in BENHALIMA (1983), LAUNOIS (1976), BUTET (1985), CHARA (1987).

The protocol of preparation of the reference epidermothèque follows the following diagram:

The epidermis was photographed using a microscope connected to a digital camera.

Study method of Orthoptera

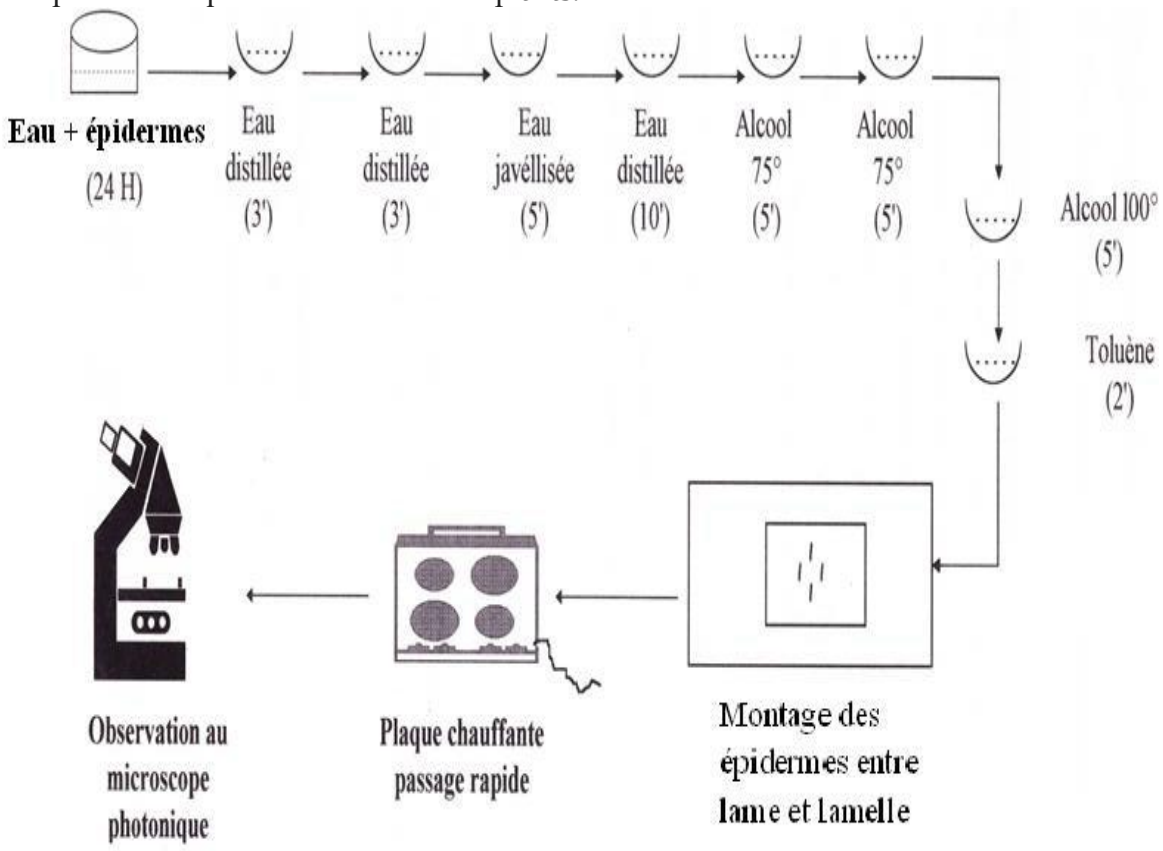
All the measurements were made in the form of a random walk within an approximate perimeter of 100m2. These samples are able to provide a good estimate of the faunistic richness.

The determination of Orthoptera necessitated key works such as that of CHAUPARD (1943), the key to Orthoptera of North Africa, FELLAOUINE (1989), LOUVEAUX and BENHALIMA (1987) catalog of orthopterans of Acrididae in North Africa.

Feces analysis

To determine the diet of Oedipoda miniata, we based on the composition of faeces. We therefore compared the plant fragments found in the feces of the species caught with the reference epidermotheca prepared from the plant species collected in the different study stations. This method has been widely used by many researchers including DOUMANDJI et al (1992), MESLI et al (2005), DAMERDJI (2008).

For this, individuals are harvested in the field and placed individually in plastic tubes and are put to rest for 8 hours (enough time to empty their digestive tract). The collected faeces are softened for 24 hours in water with wetting (polysorbate 80 or tween 80) and are then exposed to a series of baths. The experimental protocol of the preparation of the faeces is the same as that of the development of the epidermothèque of reference of the plants.



The fecal slide, once prepared, will be placed under a microscope, of which a 1mm2 window is glued under the objective (Method of windows adopted by Professor Doumandji).

III- Results and interpretations

Floristic composition

Table 1 illustrates the floristic composition of the two study areas.

Table 1: Floristic composition by families and species of the 2 study stations

Famillies	Species	Moutass	Sebdou
Anacardiaceae	*Pistacia lentiscus*	+	+
Apiaceae	*Ferula communis*	+	+
Astéraceae	*Scolymus sp.*	+	+
Borraginaceae	*Echium vulgare*	-	+
Caryophylaceae	*Paronychia argentea*	-	+
Lamiaceae	*Ballota hirsuta*	+	-
	Marrubium vulgare	-	+
	Phlomis herba-venti	-	+
	Rosmarinus officinalis	+	-
	Salvia officinalis	+	+
	Salvia verbanaca	-	+
	Thymus ciliatus	+	+
Liliaceae	*Asparagus stipularis*	+	-
Palmaceae	*Chamaerops humilis*	+	-
Poaceae	*Ampelodesma mauritanicum*	+	-
	Hordeum murinum	+	-
	Stipa tenacissima	-	+
Résédaceae	*Reseda luteola*	-	+
Rhamnaceae	*Ziziphus lotus*	+	-

The results obtained show that the floristic procession is very diversified. Indeed, we counted 8 families in the stations of Moutass and Sebdou. Regarding the species richness, the two stations are diversified with 12 species each. From the floristic surveys, there are 19 plant species, 7 of which are from the Lamiaceae family, 3 from the Poaceae family.

The remaining species each belong to the remaining families. In total, there are 11 families. Pistacia lentiscus, Ferula communis and Scolymus sp are species found in both stations.

Ecological indices

Global recovery

The total recovery rate of the environment is the sum of the recovery rates of all the plants present on 500 m2. It makes it possible to highlight the degree of occupation of the soil by the dominant species and to characterize the nature of the vegetal cover based on a scale proposed by DURANTON et al., (1982).

The results of calculations of plant species recovery rates evaluated at the two study stations are summarized in Table 2.

The formula considered most appropriate for estimating the degree of recovery is that of DURANTON et al (1982).

RG = Σ **Ss 100** / **s**

RG: overall recovery rate;

s: the surface of the plant transect;

Ss: the area occupied by a plant species projected orthogonally on the ground;

$Ss = 3.14\ r^2\ n$

r: mean radius of the tuft;

n: the number of tufts of the given species taken into consideration on the surface s.

Table 2: Overall overlap of the two stations

Station 1	RG%	Station 2	RG%
Ampelodesma mauritanica	22,68	***Stipa tenacissima***	33,21
Hordeum murinum	14,6	***Scolymus sp***	14,25
Ziziphus lotus	11,66	***Pile***	13,23
Bahi	10,52	***Thymus cilatus***	12,36
Pile	7,42	***Salof***	2,6
Scolymus sp	5,83	***Mavu***	1,92
Rosmarinus officinalis	3,52	***Phev***	1,79
Chamaerops humilis	3,1	***Ecvu***	1,83
Feco	2,18	***Relu***	1,6
Thymus ciliatus	1,92	***Feco***	1,2
Salof	1,15	***Salve***	0,88
Aspst	0,65	***Para***	0,4

In the first station, Ampelodesma mauritanicum, Hordeum murinum, and Ziziphus lotus are the most widespread, covering nearly 48.94% of the soil.
In the steppe region (station 2), Stipa tenacissima is the most representative species, covering 33.21%, followed by Scolymus sp with 14.25%.

Frequency of plant species in faeces

The principle is to note the presence or absence of the plant in the feces, according to BUTET (1985); it is expressed as follows:

$$\mathbf{F\ (i) = ni\ /\ N \times 100}$$

F (i): The relative frequency of the epidermis contained in the feces, expressed as a percentage.
ni: The number of times the plant fragments (i) are present.
N: Total number of individuals examined.

Attraction index

This method tells us about the relationship between the actual consumption of a given plant species and its recovery rate in the field.
The technique involves cutting on a graph paper a square of 1 millimeter
side and stick it on the light microscope tray so that the lens is facing; then place the end of the lamella at the level of the square, slide it vertically millimeter by millimeter and column by column thus sweeping the whole surface.
For the computation of the index of attraction we used the following formulas proposed by DOUMANDJI (1993).

$$\mathbf{Ss} = \Sigma \mathbf{xin}\ /\ '$$

$$\mathbf{S} = \Sigma \mathbf{Ss}\ /\ \mathbf{N}$$

$$\mathbf{T} = \mathbf{S}\ /\ \Sigma \mathbf{S} \times \mathbf{100}$$

$$\mathbf{IA} = \mathbf{T}\ /\ \mathbf{RG}$$

Ss: Ingested area of a given plant species calculated for an individual.
Xi: surface of plant fragments, representing a given plant species.
n ': scanned surface (sum of empty squares and solid squares).
n: surface of the lamella (400 mm2).
S: average total area of a given plant species calculated for all individuals.
N: number of individuals.
T: consumption rate of a given plant species.
IA: index of attraction.
RG: overall recovery for given plant species.

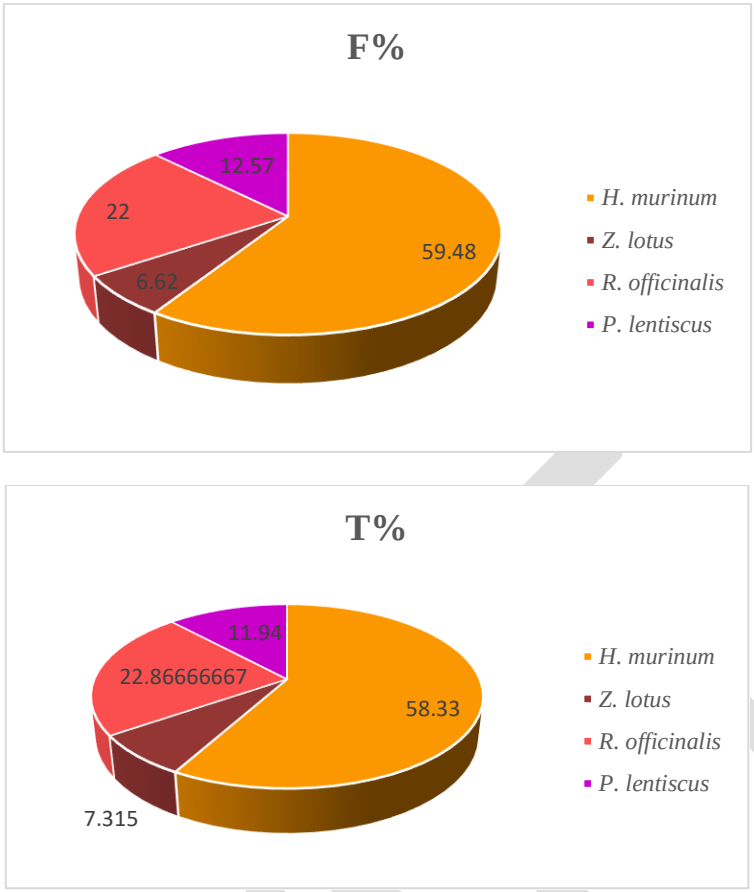
Table 3 and 4 summarizes the results of ecological indices in the two study stations.

Table 3: Areas (mm2), relative frequencies (F%), consumption rate (T%) and attraction indices (AI) of plant species found in Oedipoda miniata feces in Station 1.

Months	Indices	*H. murinum*	*Z. lotus*	*R. officinalis*	*P. lentiscus*
April 2015	S mm2	121,07	3,09	14,63	60,13
	F%	73,4	1,65	10,3	41,9
	T%	77,2	1,89	11,5	9,84
	IA	3,41	0,16	1,35	1,32
May 2015	S mm2	137,25	9,7	26,74	46,7
	F%	62,5	5,35	12,5	20,4
	T%	64,47	7,4	11,2	17,7
	IA	2,85	0,63	1,31	2,38
June 2015	S mm2	106,7	19,14	50,83	31,33
	F%	48,2	11,2	26,2	14,6
	T%	50,6	10,7	21,9	16,8
	IA	2,23	0,91	2,57	2,26
July 2015	S mm2	120,3	21,06	74,06	22,86
	F%	51,8	8,3	34,3	7,22
	T%	45,4	10,5	38,1	6,6
	IA	2	0,9	4,47	0,88
August 2015	S mm2	106,05	13,17	55,29	28,12
	F%	59,8	8,45	21,5	11,2
	T%	58,01	7,6	24,9	10,2
	IA	2,56	0,65	2,92	1,37
September 2015	S mm2	117,7	8,13	64,14	27,16
	F%	61,2	4,78	27,2	7,1
	T%	54,3	5,8	29,6	10,5
	IA	2,4	0,49	3,47	1,41

Table 4: Surfaces (mm2), relative frequencies (F%), consumption rate (T%) and attraction indices (AI) of plant species found in faeces of *O. miniata* in Station 2.

Mois	Indices	*S. tenacissima*	*P. lentiscus*	*T. ciliatus*	*M. vulgare*
March 2015	S mm2	97,19	9,13	52,27	75,1
	F%	38,1	2,72	20,68	38,5
	T%	38,2	5,3	25,2	31,3
	IA	1,15	0,4	2,03	16,3
April 2015	S mm2	98,36	12,02	43,63	57,06
	F%	34,2	7,8	17,9	41
	T%	34,6	5,9	17,5	42
	IA	1,02	0,44	1,41	21,87
May 2015	S mm2	117,56	9,7	42,11	38,7
	F%	58,9	3,23	19,95	18,3
	T%	64,5	4	20,31	11,19
	IA	1,94	0,3	1,64	5,82
June 2015	S mm2	104,26	11,14	39,6	44,17
	F%	61,7	5,3	21,2	11,8
	T%	60,6	4,6	15,5	19,4
	IA	1,82	0,34	1,25	10,1
July	S mm2	103,64	11,96	42,21	27,2
	F%	68	5,6	17,2	9,2
	T%	71,1	3,62	14,4	10,9
	IA	2,14	0,27	1,16	5,67
August 2015	S mm2	92,06	4,81	63,16	42,5
	F%	50,6	2,11	26	21,3
	T%	52,01	1,7	25,2	21,1
	IA	1,56	0,12	2,03	10,98
September 2015	S mm2	89,21	10,28	70,32	39,8
	F%	31,8	10,7	40,1	18,01
	T%	33,6	8,66	39, 5	18,6
	IA	1,01	0,65	3,19	9,68



Picture card 2: Relative Frequencies and Consumption Rates of Species Found in the feces of *O.miniata* in station 1

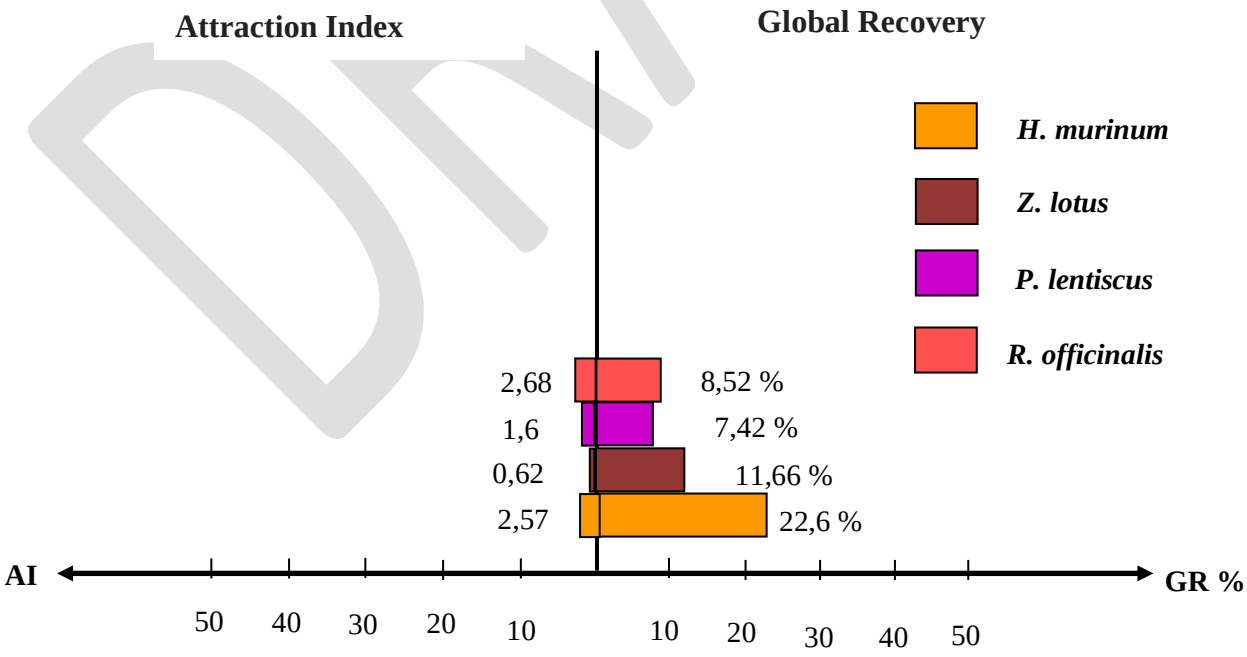


Figure 3: Comparison between the attraction indices of species consumed by O. miniata and their overall recovery rates at station 1

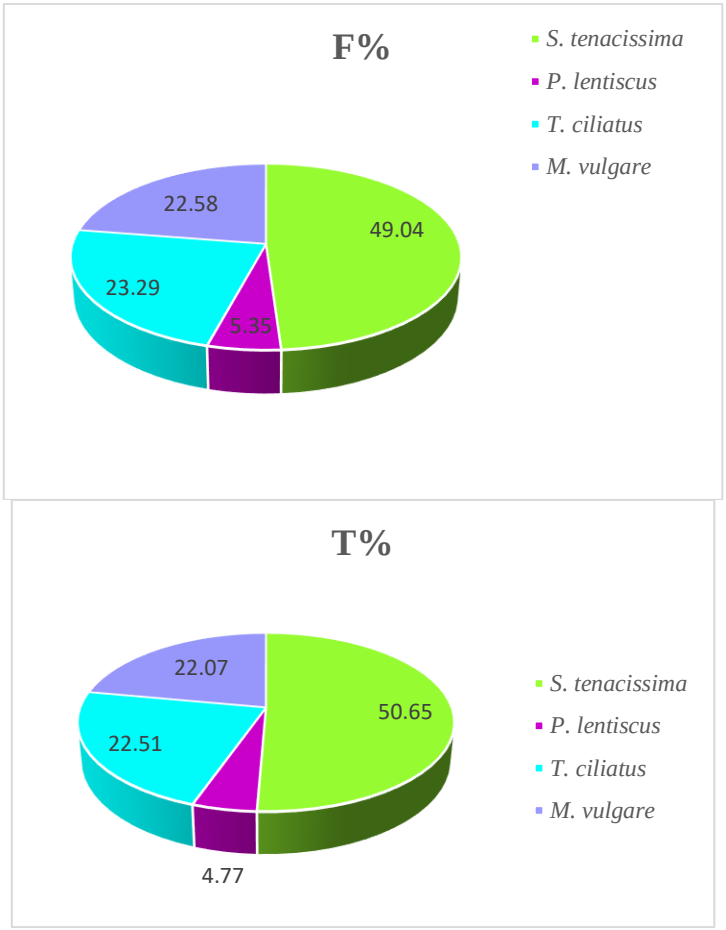


Figure 4: Relative Frequencies and Consumption Rates of Species Found in the feces of O. miniata station 2

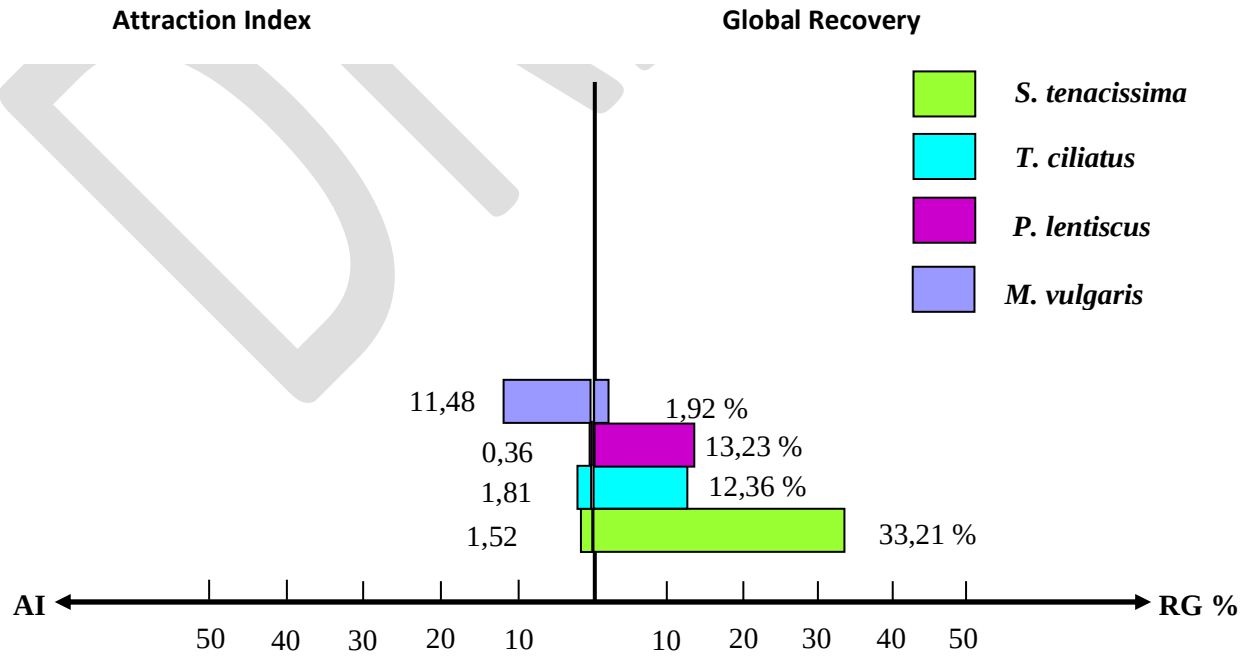


Figure 5: Relative Frequencies and Consumption Rates of Species Found in the feces of *O.miniata* in station 2

Discussion

At the Moutass station (Station 1), O. miniata consumes four plant species: H. murinum, Z. lotus, R. officinalis, and P. lentiscus. These species have a more or less low overall recovery; 22.6% for H. murinum, 11.66% for Z. lotus, 7.42% for P. lentiscuset 8.52
% for R. officinalis. The most consumed plant is H. murinum with a frequency of 59.48% and a consumption rate of 58.33%, followed by R. officinalis with F = 22% and
T = 22.86, P. lentiscus comes in third place with a frequency of 12.57% and a consumption rate of 11.94%, and finally, Z. lotus is placed fourth in the region of Moutasse with a frequency of 6.62% and a T equal to 7.31%.

We report that the consumption rate remains closely related to the relative frequency of plant fragments found in O faeces. miniata.

Comparing the global recovery of plant species consumed by O. miniata, with their indices of attraction and their consumption rates, it appears that H. murinum (Poaceae) is the plant species most consumed by this locust, while R officinalis (Lamiaceae) is the plant species that attracts it more with an index of attraction IA = 2.68; Z. lotus with an AI = 0.62 is the species that draws the least O. miniata. At the second station (Sebdou), 4 plant species form the floristic procession of O. miniata: Stipa tenacissima, Pistacia lentiscus, Thymus ciliatus and M. vulgare. The most consumed species is S. tenacissima with a consumption rate T = 50.65%. Then comes T.ciliatus with a T = 22.51; then M.vulgare with T = 22.07; P. lentiscus being the least consumed species with T = 4.77%. Although M. vulgare's rate of recovery is the lowest (1.92%), it attracts more O. miniata because it has the highest AI (11.81).

CONCLUSION

The study of the diet of the orthopter Oedipoda miniata in the region of Tlemcen realized in two stations Moutass and Sebdou, revealed to us information on the behavior of this species vis-à-vis the present vegetation.

It is through the examination of the contents of the feces that we were able to realize the study of the diet (method of the window). This method is well suited for field research. It is simple, fast and objective

Oedipoda miniata has a clear preference for 7 plant species. Therefore, it is said that it is polyphagous. In addition, it is a stenophagus indicating a medium that is poor in plant species and is appreciated in degraded environments. Comparing the consumption rates with the relative frequency obtained in the present work, we note that the level of consumption is not proportional to the recovery rates in the field. This is reflected by the fact that plants with low recovery rates are sometimes overexploited.

The results confirm what has been mentioned in several works, saying that the choice of the plant is not due to its nutritional value, nor its abundance in the field. This is the case of Rosmarinus officinalis in the first station and M. vulgare in the second station. The results also show that the species Oedipoda miniata has a high affinity for plant species of the family Haceae (Hordeum murinum, and Stipa tenacissima) which it consumes more than 50%, and lamiacees (Thymus ciliatus, Rosmarinus officinalis and Marrubium vulgare) with a consumption rate exceeding 22%.

In both stations studied, Oedipoda miniata shows a very wide diet with a minimum of 4 plant species ingested per station. We can therefore call it a euryphage species.

Bibliographical references

Allal-Benfekih L., 2006 - Recherches quantitatives sur le criquet migrateur *Locusta migratoria* (Orth. Oedipodinae) dans le Sahara Algérien. Perspective de lutte biologique à l'aide des microorganismes pathogènes et de peptides synthétiques. Thèse de doctorat, N° 17- 2006, Université de Limoges. 141pp.

Benhalima T., 1983 -Etude expérimentale de la niche trophique de *Dosiostaurus maroccanus* (Thunberg , 1815) en phase solitaire au Maroc . Thèse Doc. Ing Paris, 178 pp.

Chapman R.F., 1982 – Chemore reception: the signilcance of receptor numbers. Adv. InsectPhysiol. 16 : 247356.

Dajoz R., 1971 - Précis d'écologie. Ed. Dunod, paris, 434p.

Dajoz R., 1982- Précis d'écologie. Ed. Gautier-Villars, Paris, 503 p.

Dajoz R., 1985 - Précis d'écologie. Bordas, Paris. 505p.

Damerdji A., Mekkioui A., et Doumadji-Mitiche B., 2000 - Mise en évidence d'*Ampelodesma mauritanicum* (Diss) dans les fèces de différentes espèces de caelifères (Orthoptères) récoltées dans les monts de Tlemcen ; Etude qualitative. Revue INRAA. 1, 67-75.

Damerdji A., 2008 - Systématique et bio-écologie de différent groupes faunistiques notamment les Gastéropodes et les Orthoptères selon un transect nord-sud Ghazaouet, El Aricha. Thèse de Doct. Inst. Nat. Agro., El Harrach. 263p.

Doumandji –Mitiche B., Doumandji S et Benfkih L., 1992 – Données préliminaires sur la bioécologie de la sauterelle marocaine
Dociostaurus maroccanus(THUNBERG, 1815) à Ain Boucif (Médéa - Algérie) –
Med. Fac. Landbouw. Univ. Gent, 57/3 a, pp 659- 665.

El Ghadraoui L., 2002 - Etudes bioécologiques de criquet marocain
(*Dociostaurus maroccanus*) dans le site AL-Azghar du Moyen Atlas. Thèse de Doctorat d'Etat. Université de Fès, Maroc.

Fellaouine R., 1989 - Bioécologie des Orthoptères de la région de Sétif. Thèse de magister, Inst. Nat. Agro., El Harrach. 81p.

Louveaux A., Benhalima T., 1987- Catalogue des orthoptères Acridoidea d'Afrique du nord-ouest. Bull. Soc. Ent. France, pp 73-87.

Mesli L., Doumandji S., et Khelil M.A., 2005- Contribution à l'étude du
régime alimentaire de *Calliptamus barbarus* dans les monts de Tlemcen. Intergrated

protection in oakforests IOBC Wprs Bull. 28(8), pp 285-286.

Mesli L., 2007 - Contribution à l'étude bioécologique et régime alimentaire des principales espèces dans la wilaya de Tlemcen. Thèse Doc. Univ. Tlemcen. 102p.

Moumen K., 1997 - La transformation phasaire chez le criquet pèlerin *S. gregaria* (Forskål, 1775). Mécanisme et action de l'alimentation. Thèse de DEA, Fac., Scien., Biol., Univ., Tunis, 36 pp.

Picaud F., Bonnet E., Glouaguen V., and Petit D., 2003 – Decision Making for Food Choice by Grasshoppers (Orthoptera: Acrididae): Comparaison Between a Specialist Species on a Shurubby Legume and Three Graminivorous Species. Plant-Insect interactions. Faculté des Sciences, E.A. 3176, 123, av. A. Thomas, 87060 Limoges, France.

Zaim A., 2013 - Etude bioécologique des acridiens du Moyen Atlas, Perspectives de lutte biologique par les extraits des plantes. Thèse de Doctorat à l'Université de Fès Maroc.

.

POSTER PRESENTATION

Study of foodborne pathogen and hygienic status in albanian cheeses

Fatmira Shehu[1*] ORCID: 0009-0001-0854-3128 , Renis Maçi[2] ORCID: 0000-0003-3974-8588, Bizena Bijo[1] ORCID:0000-0003-4844-2003

[*1]Agricultural University of Tirana, Faculty of Veterinary Medicine,
Department of Veterinary Public Health, Tirana-Albania
[2]Food Safety and Veterinary Institute, Department of Food Microbiology, Tirana-Albania

*Corresponding author e-mail: s_fatmira@yahoo.it

Abstract

Cheese is considered a nutrient-rich food, important in the daily diet, but food-related diseases caused by cheese consumption have occurred in many countries Moreover, in recent years the consumption of cheese in Albania has increased and along with this, the necessity of microbial risk assessments on cheese also increased. The purpose of this study was to evaluate the microbiological quality of samples of different categories of cheese, originating from 19 different regions of Albania. During the period 2013-2019, 484 cheese samples, (n=150), (n=294), (n=40) of them were the soft, semi-hard and hard type of white cheeses respectively were tested for hygiene indicators *E.coli* and *coagulase-positive staphylococci*, as well as the parameters of the indicator of *Salmonella spp*., *Listeria monocytogenes.* The methods used were: ISO 11290: 2017 part one and two for *Listeria monocytogenes*, ISO 6579-1:2017 for *Salmonella spp.* and ISO 6888-1:2021 for *coagulase-positive staphylococci*. The results showed coagulase-positive staphylococci were found positive with a higher concentration above the legal criteria on 1.8 % (n=109), *E.coli* 13.2% (n=315). In addition, neither Salmonella nor Listeria monocytogenes were detected in 25 g to all types of cheeses (n=30 for both of them). This situation proves once again the importance and maximum care for ensuring a good hygienic quality of the milk used for cheese e production, as well as the need for rigorous implementation of the Hazard Analysis Critical Control Point (HACCP) system in dairy processing plants.

Keywords: Cheese, *E.coli, Staph. coagulase positive, Salmonella.spp, Listeria monocytogenes*

INTRODUCTION

Cheese is one of the favorite products of Albanian consumers and is a good source of protein, calcium, vitamins, and minerals. It also contains vitamin E, vitamin K, riboflavin, vitamin B6, folate, vitamin B12, pantothenic acid, and betaine. In Albania, the production and consumption of natural cheese are growing from time to time. Natural cheese is produced by curdling raw or pasteurized milk from healthy dairy animals including cows, sheep, and goats. Cows' milk is mostcommonly used for cheese making, and natural cheese is one of the most commonly consumed foods of animal origin consumption of cheese is becoming popular (INSTAT, 2019). Table 1 shows the production of cheese in Albania (in tons), in the years 2014-2019.

Table 1. Cheese production in Albania (in tons), in the years 2014-2019

Cheese texture	YEARS						
	2014	**2015**	**2016**	**2017**	**2018**	**2019**	**2020**
Soft	6.962	8.092	8.101	8.256	7.765	6.555	5.431
semi-hard	3.608	4.478	4.334	4.094	3.852	3.432	3.395
Hard	577	284	533	918	1.352	1.858	1.026

Cheese consumption has often been accompanied by reports of food outbreaks in various parts of the world, raising concerns about product safety (Sadik, C et al, 1986). Microbial populations present in raw milk can influence cheese quality dependent on the microbial profile and microbial load of the raw milk, ability of milk microflora to survive pasteurization, hygiene practices and build-up of microflora within the cheese manufacture plant, and additionally starter culture activity and acidity profiles, cheese manufacture

technology, cheese compositional parameters, and ripening temperature/environments (Sheehan J, 2013:; D'aoust, J et al 1985).

Recent studies from the year 2021 reported that O157 contaminationwas found in Gouda-like cheese (Hedberg C 1990). In recent years several outbreaks of human salmonella infection have been reported and cheese is a relatively uncommon wayof transmission (Felip, G and Toti L 1984, EFSA 2019, Jackson K et al 2011, Jackson K et al 2011). Ready to eat (RTE) food contaminated with *Salmonella* and *Listeria monocytogenes* is considered a risk to public health. Salmonellosis was the second most commonly reported foodborne gastrointestinal infection in humans after campylobacteriosis and was an important cause of foodborne outbreaks in EUMS and non-MS countries (EFSA 2021).

In 2020, *Listeria monocytogenes* was the causative agent of 16 foodborne outbreaksat the EU level, involving seven MS and 120 cases of illness, 83 hospitalizations, and 17 deaths Nine outbreaks were reported with strong evidence and 8 with weak evidence. The most commonly implicated food vehicles for the strong- evidence listeriosis foodborne outbreaks were fish and fish products, meat and cheese (Kousta, M et al 2010). Staphylococci are considered as indicator microorganisms for hygiene; therefore, their presence may be referred to neglected hygienic measures applied during production, processing and distribution of milk and dairy products.

The observed high levels of coliforms and Escherichia coli in dairy products are likely to indicate product contamination post pasteurization or inadequate pasteurization due to faulty equipment and lack of process monitoring. The presence of total coliforms and Escherichia coli in food of animal origin indicates environmental and feacal contamination, respectively.

Regulation (EC) No 2073/2005, and Albanian current Law on Food No.9863, 2008, lays down microbiological criteria, intended as food safety criteria (FSC) and process hygiene criteria (PHC), for *Salmonella* in specific food categories. According to current food law, the safety criterion for the products tested was the absence of *Listeria monocytogenes* in 25 g (Jackson, K., 2011). Cheeses should meet the safety criterion concerning *Listeria monocytogenes* and *Salmonella*; a product that does not com ply with these two criteria is considered unsafe and cannot be offered for sale (ISO 16649-2: 2001). Compliance with these criteria must be legally verified by the individual food business operator (FBO)as part of their HACCP program, through own-checks when implementing thegeneral and specific hygiene measures of Regulation (EC) No 852/2004 (Boyd E et al 2001).

MATERIALS AND METHODS

-Study area and collection of cheeses samples.

During seven years of surveillance (2013-2019), a total of 484 cheese samples were analyzed as they were considered to be at risk by the health authority. All samples were collected as official control collected from 19 different regions covering almost all the Albanian territory (Figure 1) and were categorized according to their texture and productive properties.

Samples were collected randomly at retails and processing dairy facilities. All samples were divided into five sub unit between 100and 250 g of each aliquot and analyzed separately for each parameter. Representative samples were transported under cooling conditions in a refrigeratedbox with thermometer 4-6 C within 24 h to the laboratories of Food Safety and Veterinary Institute and were tested for the presence of *Listeria monocytogenes*, *Salmonella spp, E. coli* and *coagulase positive staphylococci*. The microbiological survey involved the sampling of a variety of cheese. Cheeses matrices were categorized according to their texture and manufacturing properties.

Of the total samples examined (484 samples): (n=150) of them were soft of white cheeses, including mozzarella, ricotta: (n=294)semi-hard, and (n=40) hard type samples include caciocavallo, cheddar, Emmental, gouda gruyere. (Table 2). It allows comparison between cheese texture and years of surveillance. Semi-hard cheese is the most analyzed sample (n=294) or 61.0%. In the year 2018, we analyzed 103 samples which is by far the maximum observed number following the year 2015 (96 samples).

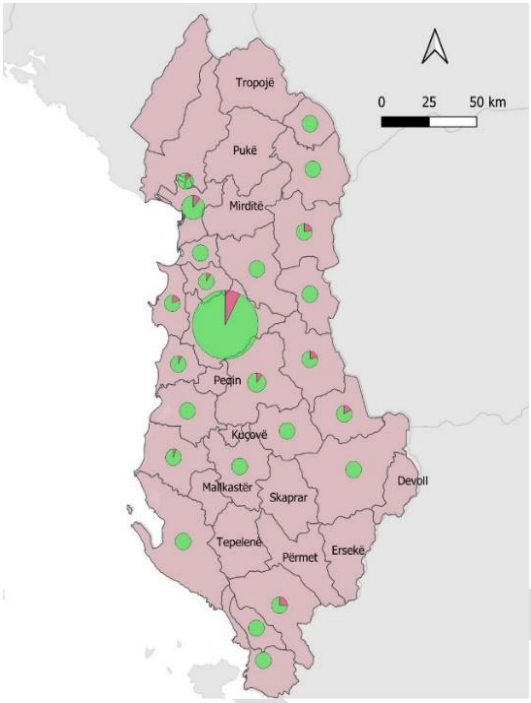


Figure 1. The geographical data pertaining to the various sampling locations.

Table 2. Distribution in numbers of all types of white cheeses between the period 2013 -2019

Reporting year	Cheese texture tested			Total samples
	Soft	semi-hard	Hard	
2013	15	18	3	36
2014	28	30	4	62
2015	32	49	15	96
2016	15	26	1	42
2017	24	54	10	88
2018	27	72	4	103
2019	9	45	3	57
Total	150	294	40	484

-Microbiological assays

Samples were analyzed to evaluate the parameters according to European Union requirements (Commission Regulation EC No 2073/2005). The cheese was analyzed to evaluate the presence of food pathogens such as *Salmonella spp* (ISO I. 6579-1:2017). *Listeria monocytogenes* (ISO 11290-1:2017), and *Coagulase-positive staphylococci.* Bacteria belonging to the indicator families *coagulase-positive staphylococci* (ISO 6888-2. 1999) and *Enterobacteriaceae*,including *E. coli b-D-glucuronidase-positive* (ISO 16649-2: 2001), were also detected andenumerated.

- Detection of Salmonella.spp

Salmonella analyses were performed according to ISO 6579-1 (Standardisation, I. ISO I. 6579-1: 2017), Microbiology of the food chain— a horizontal method for the detection, enumeration and serotyping of *Salmonella*—Part one: Isolation methods usually involve two steps pre-enrichment and enrichment steps. Shortly, each sub unit of sample was inoculated in Buffered Peptone Water (Biolife, Viale Monza, Italy) asa primary dilution of 1:9. After the incubation at 37 ^{0} C for 24 h, a 0.1 mL ofsample was transferred into Rappaport–Vassiliadis Broth (Biolife, Viale Monza, Italy) and incubated at 41.5^{0} C for 24 h and 1 ml from incubated BPW was transferred in parallel in MKTTN broth. Bacterial growth compatible with Salmonella spp. was plated into two selective media, Xylose Lysine Deoxycholate(XLD) agar (Biomaxima, Lublin, Poland) and XLT4 (Condalab, Madrid, Spain) and incubated at 37^{0} C for 24 h. All isolates were biochemically screened byusing TSI (triple sugar iron agar) and a urea agar (Christensen).

- Detection of Listeria monocytogenes

ISO 11290: 2017 part one, were used for detection of *Listeria monocytogenes*. The two steps that include pre-enrichment and enrichment processes in selective culture media followed by biochemical identification of suspected *L. monocytogenes* colonies. First, 25 g of each cheese sample was added to 225 mL half Fraser broth

(Condalab, Madrid, Spain) for initial selective enrichment and homogenized in a stomacher (Seward 400, Radnor, PA, USA). Incubation followed at $30^0 \pm 1^0$ C for 25 ± 1 h and after incubation 0.1 ml of the HF broth was transferred to tubes containing 10 ml of Fraser broth (Condalab, Madrid, Spain). Full-strength Fraser broth for the second enrichment, which was cultured at 37^0 C for 24 ± 2 h. Both enrichment cultures (HF and Fraser) were streaked onto the chromogenic Listeria agar Ottaviani and Agosti (ALOA agar) and Oxford medium. The plates were incubated for 48 h at 37^0 C and observed for the presence of typical *Listeria* colonies.

- *Enumeration of coagulase positive staphylococci*

The cheese samples were investigated for coagulase-positive staphylococci enumeration by the analytical reference method ISO 6888-1:2021. By means of a sterile pipette, 0,1 ml of the initial suspension (10^{-1} dilution) were transferred to a Baird-Parker agar (BPA) plate. For enumeration, the number of Petri dishes were used according to the dilutions was refer into ISO 7218:2007. The dishes prepared was Inverted above and placed for 24 h ± 2 h in the incubator set at 34^0 C to 38° C, and then re-incubated for a total of 48 h ± 4 h. For confirmation were used, 0.3 ml of rabbit plasma and 0.1 ml of each BHI (Brain Heart Infusion), incubated at 37^0 C. Calculation of the number of coagulase-positive staphylococci per gram of product was performed from the number of confirmed colonies found on plates chosen at dilution levels that were a significant result.

- *Enumeration of E.coli in TBX*.

In order to quantify *E. coli*, the aliquots of 10 g of cheese were homogenized with 90 mL of sterile BPW and decimal dilutionswere prepared (Biolife, Viale Monza, Italy). The homogenate was subjected to decimal serial dilutions using the same diluent in BPW and 1 ml of each dilution was plated on TBX agar medium (Biolife, Viale Monza, Italy). This medium included chromogenic substrates, X-glucuronide, for detecting dark blue colonies like *E. coli* that possess beta-glucuronidase activities. Expression of the results was done according to the ISO 7218. The mean of the values (cfu/g) of five subunits of each sample was calculated.

- *Statistical approach*

Descriptive analyses were used to summarize data such as frequencies and means, on types of cheese and hygienic conditions. IBM SPSS Version 20 (IBM Corporation, Armonk, NY) and Microsoft Excel 2013 (Microsoft Corporation, Redmond, WA) software packages were used in all analyses.

RESULTS AND DISCUSSION

Salmonella.spp. (n=30) and *L.monocytogenes* (n=30) were not detected while coagulase positive staphylococci was found positive with a higher concentration above the legal criteria on 2 % for (n=109) (Table 3). This information highlights the need to develop preventive measures by selecting good milk quality. These results showed that 2 % of all types of cheeses samples were found unsatisfactory legal criteria. In addition, neither *Salmonella* nor *L. monocytogenes* were detected in 25 g in all types of cheeses.

Table 3. Pathogenic bacteria detection from all cheese samples

Parameter	white cheese texture			Total tested	(+) ve samp. (%)
	Soft	semi-hard	Hard		
Salmonella spp.	12	14	4	30	0
L.monocytogenes	7	8	15	30	0
Coagulase pos staph.	18	86 (2.3%)	5	109	5 (1.8%)
E.coli beta-gluc(+)	102 (13.2%)	195 (13.4%)	18 (10.5%)	315	42 (13.2%)
Total	**139**	**303**	**42**	**484**	**47 (15 %)**

In regards to *E.coli*, a total of 315 samples were inspected and results indicated that 42 (13.2%) showed higher levels above 100 cfu/g with the lowest and highest counts of 10 cfu/g-10^5 cfu/g, respectively. (Table 4). Also, 277 (86.8%) of the samples including the negative ones had *E.coli* lower than 10^3 cfu/g while the counts of the remaining 42 exceed 10^3 cfu/g. This table shows that 20 samples of cheese analysed for *E.coli* (6.3%) had counts between 10^3 - 10^5 cfu/g.

Table 4. Safety status of traditional cheese in E.coli numbers

E.coli levels	Soft cheese	Semi-hard cheese	Hard cheese	Total
Less than LOD	82	161	17	260
[1-10]	6	5	0	11
10-100	4	2	0	6
100-1000	5	15	2	22
1000-10000	4	9	0	13
10000-100000	5	2	0	7
Occur. over 10^2	13.2%	13.4%	10.5%	13.2%
Occur. lower than 10^2	86.8%	86.6%	89.5%	86.8%
Occur. between 10^3 -10^4	8.5%	5.7%	0.0%	6.3%
Total	**106**	**194**	**19**	**319**

Coagulase positive staphylococci. A total of 110 samples were inspected for coagulase positive staphylococci. Among those with higher coagulase positive staphylococci growth, 3 samples out of 110 (2.7%) had the counts varying between10^1 and 10^4 cfu/g (Table 5). Moreover, among these, two samples of semi hard cheese (2.3%) exceeded 10^4 cfu/g.

Table 5. Safety status of traditional cheese in Staphylococcus numbers

Staph. Levels	Soft cheese	Semi-hard cheese	Hard cheese	Total
less than LOD	17	85	5	107
[1-10]	0	1	0	1
10-100	0	0	0	0
100-1000	0	0	0	0
1000-10000	0	2	0	2
10000-100000	0	0	0	0
Total	**17**	**88**	**5**	**110**
Occurence over 100	0.0%	2.3%	0.0%	1.8%
Occurence between 10^1 -10^4	0.0%	3.4%	0.0%	2.7%

There is undoubtedly a connection between the microbiology of milk, dairy products, and their processing environments. The dairy processing industry must take care of the hygienic quality of the processing facilities, by implementing strict controls that ensure and protect the hygienic quality of its products.

There are different factors in microbial diversity within a dairy processing plant, which depend directly on various factors such as raw material, technologicalprocess, products, workers' activities, infrastructure, and the implementation of environmental sanitation practices. If the above is not applied, the possible pathogens present in these environments, as a result of mismanagement, provoke the production of unsafe food, and consequently in causing the outbreak of foodborne illness and economic loss for Food Business Operators.

CONCLUSION

Raw milk cheese consumption is increasing in Albania also and worldwide with the growing demand for minimally processed, sustainable, healthy, and local foods. In this context, emerging and re-emerging pathogens once again represent a major food safety challenge. Therefore, good sanitation among plant workers, general sanitation in cheese plant, effective refrigeration, the use of active starters and good packaging are important in preventing related food poisoning from contaminated cheeses. Dairy processing plants should design and implement a food safety plan that carries out a risk analysis, preventive controls, monitoring procedures, and a corrective action plan. Also, training programs on good production practices for their employees are essential. Complete evaluation of the product chain of milk is also important for ensuring the hygienic quality of the cheese.

REFERENCES

Albanian Law on Food No.9863, dated 28.1.2008

Boyd E, Trmcic A, Taylor M, Shyng S, Hasselback et al 2021. Foodborne and Animal Contact Disease Outbreaks: Escherichia coli O121 outbreak associated with raw milk Gouda-like cheese in British Columbia, Canada, 2018. Canada Communicable Disease Report. 47; 11

Commission Regulation (EC) No 2073/2005 of 15 November 2005 on microbiological criteria for foodstuff.

D'aoust J, Warburton D. & Sewell A 1985. Salmonella typhimurium phage-type 10from cheddar cheese implicated in a major Canadian foodborne outbreak. Journal Of Food Protection. 48: 1062-1066

EFSA 2019.The European Union one health 2018 zoonoses report. EFSA J 17.

EFSA 2021, EU One Health Zoonoses Report 2020, EFSA, Journal; 19 (12): pg.39, 6971, www.efsa.europa.eu/efsajournal

Felip G. & Toti L 1984. Foodborne diseases caused by gram-negativemicroorganisms in milk products. Microbiol Aliments Nutr . 2 pp. 251-6

Hedberg C, Korlath J, D'Aoust J, White K, Schell 1992. A multistate outbreak of Salmonella javiana and Salmonella oranienburg infections due to consumption of contaminated cheese. Jama. 268: 3203-3207

INSTAT 2019. http://databaza.instat.gov.al/pxweb/sq/DST/ START_BU/media/ 7281/*vrojtimi-i-qumeshtit.* pdf

ISO 11290-1:2017. Microbiology of the food chain–Horizontal method for the detection and enumeration of Listeria monocytogenes and of Listeria spp.–Part 1: Detection method. (ISO Geneva, Switzerland.

ISO 16649-2:2001. Microbiology of food and animal feeding stuffs–Horizontal method for the enumeration of beta-glucuronidase-positive Escherichia coli–Part 2: Colony-count technique at 44 degrees C using 5-bromo-4-chloro-3-indolyl beta-D- glucuronide. (ISO Geneva,2001)

ISO I. 6579-1:2017. Microbiology of the Food Chain—Horizontal Method for the Detection, Enumeration and Serotyping of Salmonella—Part 1: Detection of Salmonella spp. International Organization For Standardization: Geneva, Switzerland.

ISO 6888-2:1999. Microbiology of food and animal feeding stuffs-Horizontal methodfor the enumeration of coagulase-positive staphylococci (Staphylococcus aureus and other species)-Part 2: Technique using rabbit plasma fibrinogen agar medium. (International Organization for Standardization)

ISO 7218: 2007. Microbiology of food and animal feeding stuffs — General requirements and guidance for microbiological examinations

Jackson K, Biggerstaff M, Tobin-D'Angelo M, Sweat D, Klos R 2011. Multistate outbreak of Listeria monocytogenes associated with Mexican-style cheese made from pasteurized milk among pregnant, Hispanic women. Journal Of Food Protection. 74: 949-953

Jackson K, Gould L, Hunter J, Kucerova Z. & Jackson B 2018. Listeriosis outbreaks associated with soft cheeses, United States, 1998–2014. Emerging Infectious Diseases. 24: 1116

Kousta M, Mataragas M, Skandamis P. & Drosinos 2010. E. Prevalence and sources of cheese contamination with pathogens at farm and processing levels. Food Control. 21: 805-815

Sadik C, Krending M, Mean F, Aubort J, Schneider P 1986. An epidemiological investigation following an infection by Salmonella typhimurium due to the ingestion of cheese made from raw milk. Proceedings Of The 2nd World Congress On Foodborne Infections And Intoxications 1 pp. 280-282.

Sheehan J 2013 Milk quality and cheese diversification. Irish Journal Of Agricultural AndFood Research. pp. 243-253

DRAFT

POSTER PRESENTATION

In vitro biological evaluation on *Pinus brutia* Ten. var. *pendulifolia* Frankis extracts

H. Tuba Kıyan[1] (*ORCID: https://orcid.org/0000-0001-5304-868X)*,

Melisa Manav[2] (*ORCID: https://orcid.org/0009-0000-6203-2102)*,

Melek Tekgöz[3] (*ORCID: https://orcid.org/0000-0003-2755-4655*),

Pervin Soyer[4] (*ORCID: https://orcid.org/0000-0002-6258-1993)*

[1]Anadolu University, Faculty of Pharmacy, Department of Pharmacognosy, 26470 Eskişehir, Turkey.
[2]Istanbul University, Faculty of Science, Department of Biology, 34134 Istanbul, Turkey.
[3]Eskişehir Technical University, Faculty of Science, Department of Biology, 26555 Eskişehir, Turkey.
[4]Anadolu University, Faculty of Pharmacy, Department of Pharmaceutical Microbiology, 26470 Eskişehir, Turkey.

*Corresponding author e-mail: pervinsoyer@anadolu.edu.tr

Abstract

Pinus brutia Ten. var. *pendulifolia* Frankis is a new variety of *Pinus brutia* Ten. commonly known as 'Kızılçam'. This species is an endemic species belonging to the Pinaceae family, which is mostly distributed in the Mediterranean and Aegean Regions. It has been reported that *Pinus* species are used in the treatment of respiratory and urinary system diseases, skin problems, diabetes, bronchitis, asthma, tuberculosis, cancer and gastrointestinal disorders such as reflux and ulcers. In this project, the phytochemical profile of *P. brutia* var. *pendulifolia* was determined by HPLC system and the antimicrobial activity of extracts prepared from the bark, leaves and green cones of *P. brutia* var. *pendulifolia* was investigated by microbroth dilution method, antioxidant activity by ABTS method and anti-inflammatory effect by COX-1, COX-2 enzyme inhibition method. The methanol extracts of *Pinus brutia* leaves, bark and green cone demonstrated antibacterial activity against Gram positive, Gram negative, and *Candida* species at concentrations between 312.5 and 2500 µg/mL. All of the extracts also showed strong antioxidant and anti-inflammatory activity, respectively with 57.14-72.14% and 50.63-88.13% inhibition at the 200 µg/mL concentration.

Keywords: *Pinus brutia*, Phytochemical, Antimicrobial activity, Antioxidant activity, Anti-inflammatory activity.

INTRODUCTION

Pinus brutia Ten. var. *pendulifolia* Frankis, a new variety of this species, is an endemic species belonging to the Pinaceae family (Güner et al., 2000) (Kızılarslan and Sevgi, 2013), which is mostly distributed in the Mediterranean and Aegean Regions (Güner et al., 2000) (Kızılarslan and Sevgi, 2013). It is known that Pinus species are used as antiseptic, tonic, expectorant and generally in respiratory and urinary system diseases, externally in the treatment of rheumatic pains and skin problems (Kızılarslan and Sevgi, 2013). In addition, ethnomedicinally, *P. brutia* bark in decoction against diabetes, bronchitis, asthma, tuberculosis, cancer, gastrointestinal disorders such as reflux, ulcers, pneumonia, abscess, intestinal spasm and dyspepsia, as carminative; buds as expectorant, diuretic and against cough; cones as decoction in the treatment of respiratory system diseases such as bronchitis, asthma, gastrointestinal diseases such as dyspepsia and ulcers, pneumonia, diabetes and cancer; resin against diabetes, asthma, tuberculosis, cancer, reflux, ulcer, bronchitis and low back pain; trunk against tuberculosis and respiratory system disorders; and tar against tuberculosis, cancer, asthma, diabetes and bronchitis (Çakır, 2017). In addition, it is known that red pine tar is used externally and its roots and shoots are used as decoction as a wound healer (Kızılarslan and Sevgi, 2013). This study was conducted to investigate the biochemical composition of *P. brutia* var. *pendulifolia* leaf, green cone and bark methanol extract and evaluate their biological activities, specifically in terms of antimicrobial, antioxidant, anti-inflammatory.

MATERIALS AND METHODS

Preparation of Plant Extracts

The leaves, green cone and bark of the *P. brutia* var. *pendulifolia* species, which is endemic to the Alanya (Antalya) and Fethiye (Muğla) regions in Turkey, were gathered in 2017, and identified by Forest Engineer Mahmut Tunahan. The extracts of *P. brutia* var. *pendulifolia* leaves, bark and green cone were acquired using the process of maceration using methanol at ambient temperature. The solvent was eliminated through the utilization of a rotary evaporator, resulting in the acquisition of desiccated extracts.

Liquid-Chromatographic Analyses

The extracts were analysed by LC-MS/MS (Liquid Chromatography-Mass Spectrometry/ Mass Spectrometry) system to assess their phytochemical compositions.

In vitro Anti-inflammatory Activity Test by COX-1 and COX-2 Enzyme Inhibition Method

The study was employed with COX-1 and COX-2 enzymes to assess the inhibitory effect of COX-1 and COX-2 *in vitro*. The quantification of prostaglandin E2 (PGE2) was conducted using the enzyme immunoassay (EIA) method, as described by Vazquez et al. (1997). The activity findings were evaluated using a colorimetric method in an ELISA microplate reader and expressed as a percentage of inhibition.

Antimicrobial Activity Test

The antimicrobial activity of plant extracts was assessed using the microbroth dilution susceptibility assay developed by the Clinical and Laboratory Standards Institute (CLSI) (CLSI 2012, 2002a). The strains *Escherichia coli* (ATCC 35218), *Bacillus subtilis* (NRRL B478), and *Pseudomonas aeruginosa* (ATCC 27853) were cultured overnight at 37 °C on Mueller Hinton Agar (MHA), while *Candida albicans* (ATCC 10231) was cultured on Sabouraud Dextrose Agar (SDA) at 30 °C. The microbe cultures were adjusted to a concentration of 1×10^8 colony-forming units per milliliter (CFU/mL) using McFarland 0.5 standard turbidity in a sterile saline (0.85%) solution. The cultures were then homogenized by vortexing. The plant extracts were dissolved in methanol to create the first stock solution. The plant extracts were made with varying concentrations using a two-fold serial dilution procedure in a 96-well microplate, ranging from 5000 to 4 µg/mL. Following the dilution procedure, 100 µL of each microbe culture were introduced into the well plates and subjected to incubation at 37 °C for 24 hours in the case of bacteria, and at 30 °C for 48 hours for yeast. Following the completion of the incubation period, the wells were subjected to staining by the addition of a 20 µL solution containing 0.01% resazurin. This staining process was employed to facilitate the visual differentiation between viable and non-viable cells, as indicated by the observed color variation, which ranged from blue (green) to pink. Ketoconazole, ciprofloxacin, and chloramphenicol were employed as standard agents to assess the antibacterial efficacy against *Candida albicans* and several bacterial species. The experiment was conducted on three separate occasions, and the outcomes were subsequently averaged.

Antioxidant Activity Test

The method was employed by Re et al. (1999) involved the utilization of ABTS (2,2′-azinobis(3-ethylbenzothiazoline-6-sulfonic acid)) to assess the antioxidant activity of plant extracts. A solution containing ABTS (7 mM) was prepared by dissolving it in water along with 2.45 mM of potassium persulfate. The resulting solution was then stored in a dark room for a duration of 16 hours. A standardized ABTS solution was made by diluting it with ethanol to achieve an absorbance value of 0.70 ± 0.002. Ethanol was utilized to generate stock solutions of plant extracts at a concentration of 200 µg/mL, ascorbic acid at a concentration of 100 µg/mL, and gallic acid at a concentration of 100 µg/mL. Ethanol was employed as the negative control, while ascorbic acid (Vitamin C) and gallic acid were utilized as the positive controls. The calculation of radical scavenging activity (RSA) involved determining the percentage inhibition of ABTS radical reduction using the following formula: "%ABTS scavenging = [(A control - A sample) / A control] × 100".

DRAFT

RESULTS and DISCUSSION

Liquid-Chromatographic Analyses

For LC-MS/MS analysis, Absciex 3200 MS/MS detector was used. The chromatographic separations were conducted using a Shimadzu 20A High Performance Liquid Chromatography (HPLC) system equipped with an ODS (octadecylsilane) column of 150 x 4.6 mm with a particle size of 3 um. The temperature of the column oven was adjusted to 40 °C, while the flow rate was set at 0.5 mL/min. The mobile phases utilized in the experiment consisted of two compositions: (A) a mixture of methanol, water, and formic acid in a volumetric ratio of 10:89:1, and (B) a mixture of methanol, water, and formic acid in a volumetric ratio of 89:10:1. The concentration of substance B underwent a significant increase, rising from 10% to 100% within a time frame of 40 minutes. The mass screening (EMS) was conducted within a selected mass range of 100-1000 amu. The findings were shown in Table 1.

Table 1. Compounds identified in methanol extracts

Rt	$[M-H]^-$	MS/MS	Identification	Statement
3.5	191	173, 127	Quinic acid	*P. brutia* var. *pendulifolia* leaf methanol extract, *P. brutia* var. *pendulifolia* green cone methanol extract
9.0	153	109	Hydroxybenzoic acid	*P. brutia* var. *pendulifolia* leaf methanol extract
9.7	289	245, 203	Catechin	*P. brutia* var. *pendulifolia* green cone methanol extract
14.0	179	135	Caffeic acid	*P. brutia* var. *pendulifolia* green cone methanol extract
16.2	491	327,315,300	Nepetin glucuronate-like	*P. brutia* var. *pendulifolia* leaf methanol extract
17.0	179	164, 145	Coniferyl alcohol	*P. brutia* var. *pendulifolia* leaf methanol extract
17.3	315	300, 283	Similar to Nepetin	*P. brutia* var. *pendulifolia* leaf methanol extract
17.7	285	241, 217, 175	Fisetin or similar	*P. brutia* var. *pendulifolia* leaf methanol extract *P. brutia* var. *pendulifolia* bark methanol extract
18.0	479	315, 287, 271	Mirsetine glucoside	*P. brutia* var. *pendulifolia* leaf methanol extract
18.4	325	307, 297,201,179, 151	Unknown	*P. brutia* var. *pendulifolia* bark methanol extract
20.1	463	315, 300, 271	Methylquercetin rhamnoside	*P. brutia* var. *pendulifolia* leaf methanol extract
21.0	461	300, 271, 179, 151	Quercetin glucoside	*P. brutia* var. *pendulifolia* leaf methanol extract
23.6	477	314, 299, 271	Isoramnetin glucoside	*P. brutia* var. *pendulifolia* leaf methanol extract
24.4	507	344, 329, 314, 301	346 MW Tetramethyl flavonoid + glucose	*P. brutia* var. *pendulifolia* leaf methanol extract
26.1	301	283, 273,257, 139	Herbasetin or similar	*P. brutia* var. *pendulifolia* leaf methanol extract
27.3	461	298, 269, 241	Apigenin-derived glucoside	*P. brutia* var. *pendulifolia* leaf methanol extract
27.3	301	273, 179, 151	Quercetin	*P. brutia* var. *pendulifolia* leaf methanol extract, *P. brutia* var. *pendulifolia* bark methanol extract

COX-1 and COX-2 Enzyme Inhibition Test

The *in vitro* anti-inflammatory activity of *P. brutia* var. *pendulifolia* leaves, green cone and bark methanol extracts was determined by COX-1 and COX-2 Enzyme Inhibition Test and % inhibition rates were determined (Table 2.). The 88.13% inhibition activity of green cone extract was determined at the 200 µg/mL concetration. The fact that the result was similar with control group indicates that it has high anti-inflammatory activity.

Table 2. COX-1 and COX-2 Enzyme % inhibition rates

Concentration	**100 µg/mL**	**200 µg/mL**
	% Inhibition	
P. brutia var. *pendulifolia* leaf methanol extract	28.13	56.88
P. brutia var. *pendulifolia* green cone methanol extract	81.25	88.13
P. brutia var. *pendulifolia* bark methanol extract	45.00	74.38
Selekoksib	**90.54**	**91.83**

Antimicrobial Activity Test

The MIC values were given in Table 3. The minimum inhibitory concentrations of plant extract varied between 312,5-2500 µg/mL. According to the MIC values methanol extracts of *P. brutia* var. *pendulifolia* exhibited poor antimicrobial activity against only *Bacillus subtilis* with high concetrations. As a result, *Pseudomonas aeruginosa* is more sensitive than other bacteria species, similarly with *Candida albicans*.

Table 3. MIC values

	MIC µg/mL			
	Escherichia coli	*Bacillus subtilis*	*Pseudomonas aeruginosa*	*Candida albicans*
P. brutia var. *pendulifolia* green cone methanol extract	625	2500	312,5	312,5
P. brutia var. *pendulifolia* leaf methanol extract	625	2500	312,5	312,5
P. brutia var. *pendulifolia* bark methanol extract	625	2500	312,5	312,5
Ciprofloxacin	-	-	4	-
Ketoconazole	-	-	-	312,5
Chloramphenicol	8	4	-	

Antioxidant Activity Test

All extracts of *P. brutia* var. *pendulifolia* exhibited high antioxidant activity compared with control groups of gallic and ascorbic acid (Table 4). The highest 72,14% inhibition activity was determined at the 200 µg/mL concetration with bark extracts.

DRAFT

Table 4. ABTS % inhibition rates

	ABTS % inhibition	**Concentrations**
P. brutia var. *pendulifolia* bark methanol extract	72,14	200 µg/mL
P. brutia var. *pendulifolia* green cone methanol extract	62,71	200 µg/mL
P. brutia var. *pendulifolia* leaf methanol extract	57,14	200 µg/mL
Gallic Acid	88,28	100 µg/mL
Ascorbic Acid	84,23	100 µg/mL

CONCLUSION

This study aimed to analyze the phytochemical compositions of methanol extracts derived from the leaves, green cone, and bark of *P. brutia* var. *pendulifolia* using a High-Performance Liquid Chromatography (HPLC) system. Notably, this investigation represents the first attempt to examine these extracts in terms of their phytochemical composition. Furthermore, the extract's *in vitro* anti-inflammatory efficacy was investigated for the first time through the utilization of COX-1 and COX-2 enzyme inhibition techniques. The present study additionally assessed the antibacterial and antioxidant activities. The subsequent phase of the study involved the investigation of *in vitro* anti-inflammatory antimicrobial and antioxidant effects. The evaluation of their effects can be conducted by *in vivo* animal tests, which serve as an initial step in preparing for clinical research. It is also envisaged that the results obtained by developing phytotherapeutic formulations that can be alternative to the examples in the literature within the scope of the project outputs can be published in reputable scientific journals and congresses.

ACKNOWLEDGEMENTS

This study was financed by Anadolu University Scientific Research Project Foundation (No: 1906S121).

REFERENCES

Clinical and Laboratory Standards Institute M07-A9 2012. Reference Methods for Dilution Antimicrobial Susceptibility Tests f or Bacteria That Grow Aerobically; Approved Standard- Ninth Edition, *CLSI document* M07-A9 *29(2).*

Clinical and Laboratory Standards Institute M27-A2 2002a. Reference Method for Broth Dilution Antifungal Susceptibility Testing of Yeasts; Approved Standard-Second Edition, *CLSI document 22(15).*

Çakır EA 2017. A comprehensive review on Ethnomedicinal utilization of gymnosperms in Turkey. Eurasian J. For. Sci. 5, 35–47.

Güner A, Özhatay N, Ekim T, Başer KHC, & Hedge IC. (Eds.) 2000. *Flora of Turkey and the east Aegean Islands*. Edinburgh University Press.

Haliloglu Y, Ozek T, Tekin M, Goger F, Baser KHC, Ozek G 2017. Phytochemicals, antioxidant, and antityrosinase activities of Achillea sivasica Çelik and Akpulat. Int. J. food Prop. 20, S693–S706.

Kızılarslan Ç, Sevgi E 2013. Ethnobotanical uses of genus Pinus L.(Pinaceae) in Turkey.

Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, & Rice-Evans C 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free radical biology and medicine*, *26*(9-10), 1231-1237.

Şeker Karatoprak G, Göger F, Yerer MB, Koşar M 2017. Chemical composition and biological investigation of Pelargonium endlicherianum root extracts. Pharm. Biol. 55, 1608–1618.

Vazquez MT, Rosell G, Pujol MD 1997. Synthesis and anti-inflammatory activity of rac-2-(2, 3-dihydro-1, 4-benzodioxin) propionic acid and its R and S enantiomers. Eur. J. Med. Chem. 32, 529–534.

DRAFT