



# BEZMÎÂLEM science

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**ORAL PRESENTATIONS**

**Guest Editor**

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OP-1

# Nuclear Magnetic Resonance-based Metabolomics in Patients with Rheumatoid Arthritis

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**Introduction:** Rheumatoid arthritis (RA); is an inflammatory autoimmune disease that is common in the population and is characterized by painful and swollen joints that seriously affect physical function and quality of life. Patients with RA are much more prone to serious infections, respiratory distress, and cardiovascular disease than the normal population. In the diagnosis of RA, with the clinic of the patients, X-ray and ultrasound images and laboratory results of the patients help diagnosis; however, further investigations are still needed since RA is thought to be a genetically based disease. Nuclear magnetic resonance (NMR) has been used for years to diagnose different amounts of diseases, including rheumatologic diseases. NMR is the best option to image the body metabolism, which is crucial to understand better the autoimmune diseases such as RA.

**Method:** For this study, 2 different groups were determined. For the patient group, 120 urine samples were taken from patients with RA. For the control group, 120 urine samples were taken from healthy people. These samples were studied in an NMR device and the results were compared through liquid chromatography-mass spectrometer (MS)/MS device.

**Results:** After examining urine samples through an NMR device, 7 metabolites's p values were found different. P values of threonine, histidine, N-Acetylglutamine, and lactate were higher in the patient group, in contrast; p values of acetate, methylmalonate, and asparagine were lower in the patient group compared the control group. Besides p values, correlations were detected between 20 different metabolites, which were studied in urine samples of both groups

**Conclusion:** It is clear that there are some metabolic differences between the urine of an patient with RA and a healthy person. To diagnose and treat RA, these differences may be assessed.

**Key words:** Rheumatoid arthritis, autoimmunity, nuclear magnetic resonance, metabolomics

## ***In vitro* Investigation of the Effects of *Hylotelephium* and *Typha* Plant Extracts on Anti-inflammatory and Wound Healing**

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**Introduction:** *Hylotelephium* and *Typha* species are known for their widespread use for treating wounds. This study investigated the total phenolic and flavonoid content, antioxidant capacities, wound healing-inducing and anti-inflammatory activity of these two species.

**Method:** After preparation of *Typha domingensis* (TDME) and *Hylotelephium spectabile* methanolic extracts (HSME), total phenol, flavonoid content and antioxidant activity were determined. Cell viability and the optimal doses for the scratch assay for HaCat and CCD-1072 cells were evaluated by MTT assay. The migration abilities of cells were evaluated using the scratch assay and analyzed by ImageJ software.

**Results:** The total phenolic content of HSME and TDME was found to be 9.8 and 36.8 mgGallicacid Eq/g, while the total flavonoid content of HSME and TDME were 6.3 and 13.8 mgQueEq/g, respectively. Also, the antioxidant capacities of HSME and TDME were found to be 312.9 and 521.4 mgascorbicacidEq/g, respectively. The maximum non-toxic dose for extracts was determined as 200 µg/mL, according to the MTT results. The maximum percentage of wound closure area with HaCaT cells after 16 h was 85.28% for 10 µg/mL TDME and 89.55% for 200 µg/mL HSME. However, after 24 h, 50 µg/mL TDME showed 91.45% area closure, while 200 µg/mL HSME showed 93.42%. Also, with the fibroblasts, it was observed that 43.27% and 61.37% of the wound area were closed after 16 and 24 h for 100 µg/mL TDME, while the control group exhibited 22.29% and 35.89% area closure, respectively.

**Conclusion:** TDME was found to have higher phenolic and flavonoid content and antioxidant capacity. In conclusion, the findings of this study provide significant evidence for the presence of wound-healing properties in both HSME and TDME.

**Key words:** *Hylotelephium*, *Typha*, wound healing, anti-inflammatory

## OP-3

## Investigation of *IFIT3* and *KCNS3* Gene Expression Patterns in the Peripheral Blood of Cryptogenic Epilepsy Patients

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**Introduction:** Epilepsy is a neurological disease, characterized by recurrent seizures. Cryptogenic epilepsies are defined as epilepsies with a lack of previous signs of brain damage and of obvious etiology. The absence of obvious causative pathology creates challenges in the clinical management of the disease. Gene expression studies aid in better clinical management, in terms of providing a better sight into etiology, or mechanisms leading to the disease. In this study, we studied the expression levels of *IFIT3* and *KCNS3* genes in blood samples to enlighten the molecular etiology of patients with cryptogenic epilepsy patients.

**Method:** Our study includes cryptogenic epilepsy patients admissioning at Bezmialem Vakıf University (n=20) and healthy controls (n=20). The participants were all over the age of 18. Females and males were equally distributed in both groups. The peripheral blood samples were collected into EDTA tubes, and total RNAs were isolated immediately. Complementary DNAs (cDNAs) were synthesized from RNA samples within the approved range of purity. *ACTB* was designated as the housekeeping gene. Primers were designed for *IFIT3*, *KCNS3*, and *ACTB*. qRT-PCR was performed on cDNA samples of both patients and healthy controls.

**Results:** Biostatistical analysis was conducted with Student t-test, using the delta delta Ct approach on Cycle of threshold (Ct) data. The expression levels of *KCNS3* were higher in the patient group ( $p<0.05$ ). *IFIT3* levels did not have a statistically significant difference between the two groups.

**Conclusion:** Patient samples showed a higher expression of *KCNS3*, a potassium channel-related gene. Non-etheless, the *IFIT3* gene, a gene functioning in immunity, didnot show any significant difference. Our findings suggest that a channelopathy is more likely to underly the disease, and that *KCNS3* might have a role in the pathogenesis.

**Key words:** Epilepsy, gene expression, RNA, peripheral blood

OP-4

## Attachment of the Oxadiazole Ring to Tetrazole-containing Proteasome Inhibitor Increases Cell Death in ER+ Breast Cancer Cells

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**Introduction:** Breast cancer is one of the most common malignancies in the world that leads to women's death. Proteasomes are large protein molecules that remove damaged or redundant proteins from the body through an enzyme called proteolysis. The 26S proteasome complex participates in the destruction of various proteins, regulates cellular mechanisms, and abnormalities in the regulation are linked to the development of cancer. Proteasome inhibitors block the activity of proteasomes, which are responsible for breaking down proteins and regulating gene expression through various signaling pathways. Many proteasome inhibitors have been developed by targeting the 26S proteasome complex for antitumor effects. These proteasome inhibitors have shown anticancer action by activating apoptosis in different tumor types. This study aimed to investigate the anticancer activity of a unique molecule with proteasome inhibitory properties that contains a tetrazole and oxadiazole ring on breast cancer cells.

**Method:** In this study, ER+ breast cancer cells (MCF-7) were used. Cell viability was analyzed by MTT assay, and the half-maximal inhibitory concentration (IC<sub>50</sub>) value with the most effective time was determined. The apoptosis was detected by flow cytometry using Annexin V/PI in addition to Acridine Orange/Ethidium Bromide (AO/EB) double staining.

**Results:** Cell viability results revealed that a new designed proteasome inhibitor induced cytotoxicity in ER+ breast cancers best at 72 hrs. However, the results showed that inhibitor containing tetrazole and oxadiazole rings has a lower IC<sub>50</sub> value (200 µM) compared to inhibitor with only tetrazole. Flow cytometry analysis and AO/EB staining results also supported the viability data.

**Conclusion:** The results of this study showed that proteasome inhibitor containing a tetrazole ring has some anti-cancer effect, but the addition of an oxadiazole ring to the molecule increases cell death caused by the inhibitor.

**Key words:** Cancer, proteasome inhibitor, apoptosis, cell death

## OP-5

## Comparing the Severity of Fetomaternal Hemorrhage in Dermatoses of Pregnancy Cases with Healthy Pregnants

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**Introduction:** Fetomaternal hemorrhage (FMH) is defined as the passage of fetal blood into the maternal circulation. Fetal erythrocytes can stimulate the maternal immune system and cause an autoimmune response. Meanwhile, dermatoses of pregnancy (DP) are defined as heterogeneous groups of skin diseases that occur in pregnancy and progress with itchy skin lesions. This study aims to determine the role of autoimmunity by measuring FMH in the etiology of DP.

**Methods:** In this study the case group consisted of patients with DP and the control group consisted of healthy pregnant who applied to our clinic between November 2021-2022. FMH was determined by the Kleihauer-Betke test in the blood sample taken from the mothers. Mollison's formula was used to estimate FMH severity.

**Results:** A total of 82, 30 (36.6%) case and 52 (63.4%) control, participants were included in this study. The mean maternal age was  $31\pm 4$  in case and  $31\pm 5$  in control group; revealed no statistical difference ( $p=0.445$ ). There was no statistical difference between the two groups regarding mean gestational week ( $27\pm 10$ ,  $30\pm 7$ ,  $p=0.350$ ). The rate of multiparity was 10% in the case group and 5.8% in the control group and there was no statistical difference ( $p=0.136$ ). There was no statistical difference in the terms of gravida, parity, abortus or BMI. The mean quantity of bleeding measured significantly higher in the case group ( $7\pm 5$ ,  $3\pm 3$ ,  $p<0.001$ ). The presence of DP history were significantly higher in the case group (16.7%, 1.9%,  $p=0.023$ ).

**Conclusion:** There is a significant, positive and very strong relationship between DP and FMH which provides evidence to the relationship between FMH and autoimmunity. These results also tie well with previous studies wherein DP history is a predisposing factor for dermatoses in further pregnancies.

**Key words:** Fetomaternal hemorrhage, dermatoses of pregnancy

OP-6

## Evaluation of the Effect of Glutathione, an Antioxidant, with Hormonal, Metabolic and Inflammation Markers in DHEA-induced PCOS Rat Model

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**Introduction:** Our study aims to investigate the possible positive effect of glutathione on the treatment of polycystic ovary syndrome (PCOS) and compare with Diane-35 and metformin.

**Method:** Twenty female Wistar albino rats, were randomly divided into 4 groups after generating a PCOS model with DHEA 6 mg/100 g/day subcutaneously for 34 days. After randomization, PCOS group (n=5); 0.2 mL 1% CMC/day orally for 28 days; Diane-35 group (n=5); Diane-35 4.5 mg/kg/day orally dissolved in 1% CMC for 28 days; group 3 (metformin group, n=5); metformin 300 mg/kg/day orally dissolved in 1 mL saline for 28 days; group 4 (glutathione group, n=5); glutathione 100 mg/kg intraperitoneally on days 35, 42, 49. PCOS was also confirmed. Unilateral oophorectomy on the 35<sup>th</sup> day to evaluate the follicles in the ovaries and vaginal smear for 10 days to confirm the absence of the regular estrus cycle. On the 56<sup>th</sup> day, rats were sacrificed by taking intracardiac blood to evaluate serum inflammation markers, testosterone, and insulin levels.

**Results:** There was a significant difference between the groups in terms of serum interleukin-6, insulin, testosterone, hs-CRP, SHBG, and MDA values. There were significantly lower in the metformin and glutathione groups compared with the PCOS group while there was no significant difference between the metformin and glutathione groups for all parameters. There was no difference between Diane-35 and the PCOS groups for all parameters. The primary, secondary, atretic, and cystic follicle numbers were lower in the glutathione group compared to the PCOS group, the number of antral follicles was higher in the glutathione group compared to the PCOS group ( $p=0.003$ ). The primary follicle number in the glutathione group was lower compared with the Diane-35 group; the number of antral follicles was higher ( $p<0.01$ ,  $p=0.001$ , respectively).

**Conclusion:** The results of the study may provide evidence for the possible positive effect of glutathione on improvement of increased inflammation, hyperandrogenemia, and insulin resistance in PCOS.

**Key words:** PCOS, glutathione, Diane-35, metformin, antioxidant, rat

## OP-7

## The Protective Effect of Glutathione on Ovarian Function in Female Rats with Cy-Induced Ovarian Damage

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**Introduction:** Premature ovarian failure (POF) is a condition that causes significant health problems and infertility due to loss of ovarian function, which is seen in approximately 1-3% of women younger than 40 years of age. POF decreases the number of oocytes in the ovaries, especially due to accelerated atresia. This study aims to evaluate the protective effect of glutathione on ovarian function in female rats with cyclophosphamide (Cy)-induced ovarian damage.

**Method:** Forty-two adult female Sprague-Dawley rats were randomly divided into six groups. Intraperitoneal injections were made to all groups on the 0<sup>th</sup>, 7<sup>th</sup>, and 14<sup>th</sup> days. Group 1; (sodium chloride 0.9%; 1 mL/kg), group 2; (Cy 75 mg/kg), group 3; (glutathione 100 mg/kg), group 4; (glutathione 200 mg/kg), group 5; (Cy and 100 mg/kg glutathione), group 6; (75 mg/kg Cy and 200 mg/kg glutathione). On the 21<sup>st</sup> day, the rats were sacrificed and the ovarian follicle count was evaluated by histopathological examination of the ovarian tissue. Anti-mullerion hormon (AMH) AMH-positive staining intensity of the follicles, and serum AMH levels were evaluated by immunohistochemistry.

**Results:** Serum AMH levels, AMH-positive staining, primary, secondary, and antral follicle count were statistically different between the groups ( $p < 0.01$ ). Primordial, primary, secondary, and antral follicle count, AMH-positive pre-antral and antral follicle count, percentage, and staining intensity were similar in groups 1 and 2; there was a statistically significant difference between group 2 and group 6 ( $p = 0.02$ ,  $p = 0.04$ ,  $p = 0.04$ ,  $p < 0.01$ ,  $p < 0.01$ ,  $p = 0.03$ ,  $p = 0.03$ ,  $p = 0.01$ ,  $p = 0.04$ ,  $p < 0.01$ ,  $p = 0.04$ , respectively).

**Conclusion:** The results of the study may provide evidence that glutathione at appropriate doses may have a protective effect against ovarian damage induced by the chemotherapeutic agent, Cy. It could lead to improved primordial, primary, secondary, and antral follicle numbers.

**Key words:** Glutathione, premature ovarian failure, infertility, cyclophosphamide



## Epileptic Seizure in Elderly People: Etiological Factors

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**Introduction:** The etiologies of epileptic seizures in the elderly are expected to differ from those in the young. In this study, the relationship between etiological factors with age and the presence of seizures associated with coronavirus disease-19 (COVID-19) was investigated in adult patients who presented to the emergency department with epileptic seizures.

**Method:** The study included 1,026 patients who were admitted to emergency and consulted the neurology department in Bezmialem Vakıf University Hospital between 01.01.2021-31.12.2021. According to consultation reports, 115 patients (mean age: 51.12±21.97; 52% male) were presented with epileptic seizures. Demographic data, seizure characteristics, and etiological factors of the patients were documented in detail. Patients were grouped as 18-60 years old (group <60; 75 of them) and 60 years and older (group ≥60; 40 of them).

**Results:** The number of patients without a previous diagnosis of epilepsy and without seizure history were significantly higher in group ≥60 (p values <0.001, =0.049, respectively). Seizure types observed: 22% focal and 78% generalized. Possible seizure triggering factors were examined; it was found that unknown causes (25%), infection (20%), malignancy (14%), drug disruption (11%), COVID-19 (11%), stress (7%), metabolic causes (5%), after Biotech vaccination (6%), and trauma (2%) were seen as a potential predictor. There was no significant difference between the groups in terms of seizure types and triggers. The presence of stroke, neurodegenerative disease, and diabetes, which are etiological factors, were significantly higher in group ≥60 (p values, <0.001, <0.001, and <0.001, respectively). 30% of the patients who came with the suspicion of COVID had their first seizure. There was no difference between the groups in terms of COVID-19 (p>0.05).

**Conclusion:** Elderly people presenting to the emergency department with seizures are more likely to have a first seizure and not be diagnosed with epilepsy. Therefore, etiological investigations should be done carefully.

**Key words:** Epileptic seizure, emergency, seizure types

OP-9

## Comparison of BCL2 Positivity and Ki67 Expression Rates and Clinical Prognostic Parameters in Diffuse Large B-cell Lymphoma with Germinal Center and Activated B-Cell Immunophenotype

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**Introduction:** Due to its heterogeneous characteristics in prognosis and response to treatment, there are many pathological and clinical methods to categorize diffuse large B-cell lymphoma (DLBCL). We compared the clinical prognostic parameters of these 2 groups calculated by immunohistochemical BCL2 positivity and Ki67 expression rates and NCCN-IPI (National Comprehensive Cancer Network) score.

**Method:** The study was performed with 102 patients diagnosed with DLBCL between January 2014-February 2022. Biopsy reports of the patients were reviewed, and BCL2 positivity, Ki67 expression rates, and immunophenotypes were noted. The clinical findings (age, stage, number of extranodal involvement, performance degree and LDH values) of the determined patient group were examined, and NCCN-IPI scores were calculated.

**Results:** Of the patients, 32.4% (n=33) were female, 67.6% (n=69) aging from 17 to 93. No significant difference in BCL2 or Ki67 expression, immunophenotype, or NCCN-IPI score was found between genders. It was observed that the Ki67 expression rate was higher in which the antiapoptotic protein Bcl2 was negative (p=0.001). Also, those with BCL2 negativity were more in the GCB group as an immunophenotype (p=0.009). The difference between NCCN-IPI score and Bcl2, Ki67 and phenotype was found to be statistically insignificant (p>0.05). There was no correlation with the NCCN-IPI score in those with a Ki67 expression rate above 90, but it was detected at a higher rate in the BCL2-negative group and with the GCB phenotype (p=0.001; p=0.021).

**Conclusion:** This study indicates that there is no co-relation between clinical prognostic parameters and BCL2, Ki67 and immunophenotype. In contrast pathological examination routines are statistically significant among themselves.

**Key words:** Diffuse large B-cell lymphoma, BCL2, Ki67, NCCN-IPI

OP-10

## Investigation of the Anti-cancer Effects of the Tyrosine Kinase Inhibitor-pexidartinib on the Lung Cancer Cell Line

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**Introduction:** Lung cancer is the type of cancer that causes the most deaths at all ages. Tyrosine kinase inhibitors targeting driver genes are effectively used for treating adenocarcinoma. Pexidartinib is a tyrosine kinase inhibitor approved by the Food and Drug Administration in 2019 and is the first systemic agent with proven efficacy for treating tenosynovial giant cell tumors. In this study, we investigated the anti-proliferative and anti-metastatic effects of a tyrosine kinase inhibitor pexidartinib on lung adenocarcinoma cell lines.

**Method:** In this study, BEAS-2B cell line as a healthy cell and A549 lung cancer cell line was cultured in standard conditions. The viability of cells was tested using MTT assay with pexidartinib in increasing concentrations for 24 h and 48 h. The percentages of A549 cells undergoing apoptosis were measured by AV-PI staining A-cell migration assay was performed to reveal the effect of pexidartinib on cancer cell motility. Western blotting was realized to understand the cell death mechanism with pexidartinib treatment in lung cancer cells.

**Results:** No significant effect of treatment of pexidartinib on Beas-2B cells was observed on cell viability. However, the cell viability of lung cancer cells, A549, was decreased with treatment with pexidartinib that is even 1  $\mu$ M ( $p < 0.005$ ). Necrotic cells stained with PI were increased to pexidartinib treatment on A549 cells. There was no apoptosis induced at any concentration of pexidartinib on A549 cells. On the other hand, it was shown that pexidartinib induced necroptosis with RIP1/RIP3 expression via western blot.

**Conclusion:** Our findings confirm the anti-cancer effects and safety of pexidartinib therapy on lung cells. However, it needs further studies to understand the related pathways of pexidartinib treatment in lung cancer.

**Keywords:** Lung cancer, tyrosine kinase inhibitor, pexidartinib