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# **ORAL ABSTRACTS**

## OA-1

### Biological Variation for Thyroid Hormone Tests of Healthy Subject in Turkey

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**Aim:** Biological variation (BV) data is required to assist in interpretation of test results and to evaluate analytical performance. Our study aimed to calculate the BV and BV-based quality goals of thyroid stimulating hormone (TSH), free triiodothyronine (fT3), and free thyroxine (fT4) test.

**Methods:** A total of 21 Turkish healthy volunteers (12 males and 9 females) were included in the study. Blood samples were collected once a week for five weeks. The measurements of thyroid marker on each sample were performed in duplicate using the electrochemiluminescence assay method. Analytical variation (CVA), within-subject BV (CVI) and between-subject BV (CVG) were calculated. Quality goals, individuality index(II) and reference change value (RCV) were derived from these data. Ethics committee approval was obtained.

**Results:** For TSH, fT3 and fT4, CVA values were 3.3%(2.9%-3.8%), 1.7%(1.5%-1.9%) and 2.7%(2.4%-3.1%); CVI values were 22.3% (19.3%-26.3%), 4.4%(3.8%-5.3%) and 5.1%(4.3%-6.1%); CVG values were 26.6%(19.2%-39.8%), 9.2%(6.9%-13.6%) and 8.2% (6.1%-12.1%); allowable total errors were 27.1%, 6.2% and 6.6%; II values were calculated as 0.84, 0.48 and 0.61; and asymmetric RCV were (-40.3)-67.6, (-10.4)-11.6 and (-12.7)-14.5, respectively.

**Conclusion:** Our study provides updated BV data for thyroid function tests (TFTs) in healthy volunteers. TSH compared to fT3 and fT4 had higher BV. Since TFTs have shown a high degree of individuality, RCV should be preferred rather than population-based reference ranges in the assessment of serum concentrations.

**Keywords:** Within-subject variation, between-subject variation, thyroid stimulating hormone, triiodothyronine, thyroxine

## OA-2

### Biological Variation Estimates for Cancer Antigen 19-9, Carcinoembryonic Antigen and Prostate-Specific Antigen

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**Background:** In many malignancies, tumor markers have an important role in monitoring response to treatment and disease progression. Biological variation (BV) data are essential to accurately assess the clinical status of the patient and the analytical performance of the measurements. The aim of our study was to assess the BV estimates for carbohydrate antigen (CA) 19-9, Carcinoembryonic Antigen (CEA), and Prostate-Specific Antigen (PSA) in a group of healthy individuals.

**Material and method:** Weekly samples were collected a week for 5 consecutive weeks for CA 19-9 and CEA from 20 healthy individuals (12 male and 8 female) and PSA from 13 healthy males. The measurements on each sample were performed in duplicate using the electrochemiluminescence assay method. The data analysis was performed using BioVar, an online statistical tool. Analytical variation (CVA), within-subject BV (CVI), and between-subject BV (CVG) were calculated. Quality goals, individuality index(II), and reference change value (RCV) were derived from these data. Ethical approval was obtained from the Ethics Committee of Izmir Atatürk Training And Research Hospital.

**Result:** For CA 19-9, CEA, and PSA, CVA values were 11.9% (10.3%-14.0%), 9.7% (8.0%-12.1%), and 2.6% (2.2%-3.2%); CVI values were 9.4% (6.1%-12.7%), 10.4% (7.0%-14.7%) and 11.8% (9.7%-15.0%); CVG values were 43.2% (32.0%-66.1%), 36.1% (24.4%-66.4%) and 32.3% (22.2%-57.2%); allowable total errors were 18.8%, 17.8% and 18.3%; II values were calculated as 0.22, 0.28 and 0.37; and RCV were -29.6-42.0, -28.0-39.0 and -24.4-32.2, respectively.

**Conclusion:** Our study provides updated BV data for CA 19-9, CEA, and PSA in healthy volunteers. Findings show that there is marked individuality. Therefore, the use of RCV should be considered in patient follow-up for these markers.

**Keywords:** Within-subject variation, between-subject variation, carbohydrate antigen 19-9, carcinoembryonic antigen, prostate-specific antigen

## OA-3

### The Role of Presepsin in Sepsis: Diagnostic and Prognostic Advantages

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**Aim:** Sepsis is a condition caused by an infection that can progress to organ failure and death. There is a need for biomarkers that can be used for early diagnosis of sepsis, that are minimally affected by other conditions, and can also guide prognosis in sepsis. This study investigates the value of presepsin in diagnosing sepsis, assessing its severity, and predicting prognosis.

**Methods:** This study included patients hospitalized in intensive care units (n=100), and a healthy control group (n=30). Patients were divided into two groups according to sepsis-3 criteria: sepsis (n=57) and septic shock (n=43). Also, patients were divided into two groups, survivors (n=70) and non-survivors (n=30) according to 28-day mortality. We compared presepsin, C-reactive protein (CRP), procalcitonin (PCT), lactate, and SOFA scores between groups. Receiver operating characteristic (ROC) analyses were performed for parameters with significant differences between groups and area under the curve (AUC, 95% confidence interval), and cut-off values were determined.

**Results:** Presepsin, CRP and PCT values were significantly higher in the patient group compared to the control group (all  $p < 0.001$ ). In the diagnosis of sepsis, CRP showed maximum, presepsin and PCT showed excellent performance (AUCs were 1.000, 0.962, and 0.988, respectively). There was no significant difference in CRP values between sepsis and septic shock groups, while presepsin, PCT, lactate and SOFA scores were significantly higher in the septic shock group (all  $p < 0.001$ ). Lactate performed excellently in predicting disease severity, and presepsin, PCT, and SOFA scores performed moderately (AUCs were 0.920, 0.748, 0.711, and 0.781, respectively). The 28-day mortality was 17.5% in the sepsis group and 46.5% in the septic shock group. There was no significant difference in presepsin, CRP, and PCT values between survivors and non-survivors, while lactate and SOFA scores were significantly higher in non-survivors ( $p < 0.001$  and  $< 0.001$ , respectively). Lactate and SOFA scores performed moderately in determining prognosis (AUC 0.719 and 0.724, respectively).

**Conclusion:** Presepsin demonstrates excellent performance similar to CRP and PCT and surpasses them in determining sepsis severity. It may serve as an alternative to CRP and PCT, and its combined use with these biomarkers could be beneficial.

**Keywords:** presepsin, sepsis, septic shock

## OA-4

### Current Biomarkers on Sepsis

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**Objective:** Sepsis is a major cause of deaths in intensive care units. It is claimed that sepsis affects more than 30 million people annually and causes 5 million deaths. In 2011, it was found that sepsis and related complications accounted for over \$20 billion in expenditures in the United States. For these reasons, sepsis can be considered a public health issue. It has been determined that initiating appropriate antibiotic therapy within the first hour of sepsis treatment reduces mortality. This study aims to determine the diagnostic performance of neutrophil CD64, calprotectin, presepsin and sTREM1 parameters that we believe may have early diagnostic value in sepsis and to compare them with CRP and procalcitonin.

**Methods:** The research was initiated after the approval decision of Pamukkale University Faculty of Medicine Ethics Committee dated 23.05.2022 and numbered E.228284. Diagnostic performance values of sTREM1, presepsin, neutrophil CD64, and calprotectin were calculated. Sepsis patients were selected from those treated in the Intensive Care Unit of Pamukkale University between January 1, 2023, and January 31, 2024. After applying exclusion criteria, 76 sepsis patients and a control group consisting of 76 healthy volunteers were included in the study. Calprotectin, presepsin, and sTREM1 parameters were measured using a commercial ELISA kit. The neutrophil CD64 parameter was measured by flow cytometry. CRP was measured immunoturbidimetrically on the Roche c702 module, and procalcitonin was measured using electrochemiluminescence on the Roche e801 module. Cut-off values were determined by Youden Index. ROC curve analysis was performed for each parameter including CRP and procalcitonin.

**Results:** No significant differences were found in the demographic characteristics between the patient and control groups. Six parameters, including CRP and PCT, were found to be significantly higher in the patient group compared to the control group ( $p < 0.05$ ). The sensitivity, specificity, positive and negative predictive value of presepsin in the diagnosis of sepsis were 85,5%, 64,5%, 70,7%, 81,7%, respectively. The sensitivity, specificity, positive and negative predictive value of nCD64 in the diagnosis of sepsis were 93,4%, 80,3%, 82,6%, 92,4% respectively. The sensitivity, specificity, positive and negative predictive value of calprotectin in the diagnosis of sepsis were 36,8%, 100%, 100%, 60,8% respectively. The sensitivity, specificity, positive and negative predictive value of sTREM1 for the diagnosis of sepsis were 64,5%, 94,7%, 92,5%, 72,7% respectively. The sensitivity, specificity, positive and negative predictive value of CRP for the diagnosis of sepsis were 81.6%, 100%, 100%, 84.4% respectively. The sensitivity, specificity, positive and negative predictive value of procalcitonin for the diagnosis of sepsis were 63.2%, 100%, 100%, 73.1% respectively.

**Conclusion:** When the diagnostic performance data were evaluated, the highest performance was found in the Neutrophil CD 64 parameter. CRP shows very close values to the nCD64 parameter. Presepsin and Procalcitonin also showed comparable performance to CRP and NCD64. The diagnostic performance of calprotectin was found to be low compared to other parameters. In conclusion; presepsin and neutrophil CD64 are important current parameters in the diagnosis and clinical follow-up of sepsis patients.

**Keywords:** sepsis, biomarker, neutrophilCD64, nCD64, calprotectin, presepsin, sTREM1

## OA-5

### A Case Report on Polyethylene Glycol Interference in TSH Analysis

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**Abstract:** Thyroid stimulating hormone (TSH, thyrotropin) is secreted from the anterior pituitary and is the main regulator of thyroid function. TSH, stimulates the synthesis and release of the hormones thyroxine (T4) and triiodothyronine (T3) from the thyroid gland. TSH measurement is mainly performed to assess thyroid function. TSH analyses are performed for the diagnosis and treatment of hypo/hyperthyroidism and to monitor TSH suppression with treatment in some thyroid cancers. Measurements can be made by many immunoassay methods. The most frequently used method in routine analyses is chemiluminescence immunoassay. When unexpected results occur during measurements and cannot be explained clinically, this is called interference. Interference in chemiluminescence immunoassay can be caused by matrix effects, heterophilic antibodies, hook effect, as well as sample-related conditions such as the presence of rheumatoid factor and endogenous alkaline phosphatase. In case of unexpectedly high TSH levels, analyses can be repeated using polyethylene glycol and heterophilic antibodies to avoid possible interference.

**Aim:** In this case report, the effect of polyethylene glycol on the results of repeated analyses of a patient with isolated high TSH levels is presented.

**Method:** TSH measurements were performed on a Dxi 800 (Beckman Coulter, USA) using a double-sided chemiluminescence immunoenzymatic method. The analysis was repeated using polyethylene glycol from the same sample. The analysis was repeated using heterophilic blocking antibodies (Scantibodies, USA) from the same sample and the results were compared.

**Results:** In the routine controls of a 44-year-old male patient who underwent kidney transplantation at an external centre in 2019, TSH level was measured as 290  $\mu$ IU/ml, FT3 level as 3.69 pmol/L, and FT4 level as 4.43 pmol/L. Due to clinical mismatch; repeated results from the same sample were unchanged. The new sample was analysed and TSH was 294  $\mu$ IU/ml. Considering the possible presence of a macroprotein, the analysis was repeated after precipitation of the same sample with polyethylene glycol, but no measurement was possible. In the analysis using tubes containing heterophilic blocking antibody, the result was measured 285  $\mu$ IU/ml. The polyethylene glycol used was checked after reanalysis with polyethylene glycol failed again. Although the expiry period for polyethylene glycol is normally 6 months, it was found that the expiry period of the polyethylene glycol used had expired. The TSH result was measured as 258  $\mu$ IU/ml using freshly prepared polyethylene glycol.

**Conclusion:** In the detailed history, it was determined that the patient was started treatment for thyroid disease at the last follow-up visit at an external centre, but the patient did not use this treatment. As a result of the treatment and follow-up performed by the endocrine clinic, TSH result was measured as 5.31  $\mu$ IU/ml, FT3 3.15 pmol/L, FT4 8.74 pmol/L in the control analyses 3 months later. The use of polyethylene glycol in laboratory measurements is very important to prevent possible interferences, especially in the precipitation of macroproteins. Polyethylene glycol solution can be used up to 6 months after preparation and should be kept in accordance with the storage conditions during this period. Otherwise, it may cause erroneous results, especially in immunoassays.

**Keywords:** Immunoassay, TSH, Interference, Polyethylene glycol, Blocking antibodies.

## OA-6

### Comparison of Zybio Urine Analyzer with Urit Uc-1800 and Manual Urine Microscopy

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**Aim** Fully automatic urine devices are widely used in medical laboratories because they are at low cost and can be used quickly. There are many automated systems that uses different technologies. Before new systems are put into use, they should be compared both with existing systems and with manual urine microscopy. In our study, we aimed to investigate the clinical usability of the Zybio U2610 (Zybio Ins., China) device by comparing it with the currently used Urit Uc-1800 (URIT Medical Electronic Co., Ltd., China) device and manual urine microscopy. **Material and Method** In the comparison of Zybio with manual urine microscopy 100 pathologic urine samples were included in the study. In the Zybio and Urit device comparison, 206 pathologic urine samples were included in the study. Urine samples were analyzed within 2 hours using Zybio and Urit automatic urine devices and manual microscopic methods. The Zybio and Urit device used digital flow microscopy and automatic particle identification. The results were compared according to Choen's Kappa analysis and agreement rates. **Results** According to the Kappa values in the Zybio and Urit device comparison; epithelium, erythrocyte, leukocyte, pH, ketone and leukocyte esterase values are in good agreement; protein, glucose and hemoglobin moderately compatible; Nitrite, crystal, bacteria and casts were found to be poorly compatible. Urobilinogen and bilirubin were discordant. According to the Kappa values in the comparison of Zybio and manual microscopy; epithelium and erythrocytes were found to be well compatible, and leukocytes and crystals were found to be moderately compatible. Bacteria and cast were incompatible. **Conclusion** The compatibility of the two autoanalyzers, especially the cellular elements, erythrocyte, leukocyte, epithelium, pH, ketone, leukocyte esterase, protein, glucose and hemoglobin were acceptable. Apart from casts and bacteria, cellular elements were also confirmed by manual microscopic evaluation. The poor agreement for bacteria, casts, crystals, nitrite, urobilinogen and bilirubin was attributed to the lack of pathological samples. Based on the data of this study, it was thought that Zybio systems could be used safely in the laboratory.

**Keywords:** Urinary analysis, automated systems, microscopic evaluation

## OA-7

### Handicap in HbA1c Measurement in the Presence of Variant Hb: Ion Exchange HPLC

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**Aim:** To emphasize that the cation-exchange-HPLC method, which is frequently used in HbA1c measurement, may cause errors in the presence of many asymptomatic variant hemoglobin and that it is not unlikely to be overlooked.

**Methods:** The HbA1c test requested from a 52-year-old female patient who had no active complaints and applied to the internal medicine outpatient clinic for routine check-up was performed with the Lifotronic-H9 cation-exchange-HPLC method, and the result was 2.6% below the reference range. The concurrent serum glucose level was 82 mg/dL. With the suspicion of hemoglobinopathy, a peak coinciding with HbA2 was observed in the measurement made with the thalassemia column on the Tosoh-G8 cation-exchange-HPLC device (sum of HbA2 and HbX: 58.8%). As an alternative method, the HbA1c result repeated with the immune method on the Roche-Cobas-C501 biochemistry autoanalyzer was 4.8%. Written consent was obtained from the patient and the patient was referred to further genetic analysis.

**Results:** Hb G-Coushatta variant (Glutamate-Alanine change at the 23rd position) carrier was detected in DNA sequence analysis and beta chain scanning. The patient's hemogram, iron status, vitamin B12, folate levels, p50 value in blood gas, lipid panel, liver and kidney function tests were all within the normal reference range.

**Conclusion:** Considering the current literature information on Hb G-Coushatta and the absence of any anemia that could affect erythrocyte lifespan in our patient, we think that the erroneously observed decrease in HbA1c measurement is due to measurement error. In the presence of variant Hb; It has been reported in many publications that the highest rate of interference in HbA1c measurement methods is in the ion-exchange-HPLC method. It has been reported in the literature that the Hb G-Coushatta variant undergoes glycation just like the normal beta chain. Since the same retention time was not observed in the cation-exchange-HPLC method with Glike Hb G-Coushatta, it was reported that HbA1c was measured falsely low. This erroneous low HbA1c; It is more visible in homozygous variant forms and is easier to detect. It has been reported that HbA1c results can be obtained within the normal reference range in carriers, so it is important for the laboratory specialist to evaluate HbA1c results together with simultaneous glucose results. Preferring alternative methods (affinity HPLC, immune method, enzymatic method, etc.) in HbA1c measurement will reduce the risk of encountering erroneous results due to Hb variants and the loss of time and resources required for further analysis.

**Keywords:** HbA1c, interference, hemoglobinopathy, cation exchange HPLC



## OA-8

### A Rare Hemoglobin Variant First Identified in Turkey (Hb Iraq-Halabja): The Impact on HbA1c Measurement Methods

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**Aim:** The aim is to examine the Hb Iraq-Halabja variant, first reported in Turkey, the difficulties encountered in HbA1c measurement, and the use of alternative glycemic markers. Additionally, it aims to compare different methods in HbA1c analysis and evaluate the role of genetic analysis in identifying this rare Hb variant.

**Methods:** The HbA1c level of a 50-year-old male patient diagnosed with pre-diabetes and obesity who presented to the general surgery outpatient clinic for routine follow-up after obesity surgery could not be measured on the Adams HA-8180V analyzer (Arkray, Shiga, Japan - reverse-phase cation exchange chromatography), and a tail warning between stable HbA1c and HbA0 indicating abnormal peaks was observed (concurrent glucose level was 154 mg/dL). HbA1c results of %5.8 and %5.87 were obtained on Premier Hb9210 (Trinity Biotech, NY-USA - boronate affinity chromatography) and Cobas c501 (Roche Diagnostics, Germany - turbidimetric immuno-inhibition) analyzers, respectively (estimated average glucose equivalent 120 mg/dL). The patient was referred to the medical genetics outpatient clinic with suspicion of an Hb variant that could cause interference in HbA1c measurements based on literature review, and the rare Hb Iraq-Halabja variant was detected in the sequence analysis (ABI 3500 DNA Sequencer).

**Results:** After genetic verification and in light of literature information, an abnormal peak warning was observed with the HLC-723 G11 analyzer (Tosoh Bioscience, Tokyo, Japan - ion exchange HPLC), a variant Hb peak between stable HbA1c and HbA0 was detected on the graph, and the stable-HbA1c peak percentage was measured as %3.2 based on retention times, with a concurrent %5.4 HbA1c found on the Premier Hb9210 analyzer (concurrent glucose 94 mg/dL). Additionally, a fructosamine value contributing to the patient's clinic of 210 µmol/L was found (estimated average glucose equivalent for the last 2-3 weeks was 88 mg/dL). The patient's hemogram, reticulocyte, bilirubin parameters; haptoglobin, LDH, iron status; B12, folate levels were within normal reference ranges, and iron deficiency anemia, hemolytic anemia, microcytic anemia, B12-folate deficiency were ruled out.

**Conclusion:** It has been reported in the literature that the boronate affinity and immuno methods we used, which yielded HbA1c results consistent with the patient's fasting blood glucose and fructosamine results, are more resistant to interference by Hb variants. However, it was observed that the ion exchange HPLC method resulted in falsely low HbA1c measurements due to overlapping peaks of the Hb variant. This study demonstrates that hemoglobin variants can affect laboratory tests and highlights the need to consider such situations.

**Keywords:** Diabetes mellitus, HbA1c, Hb variant, HPLC, Hb Iraq-Halabja

## OA-9

### The Current Median Values of First Trimester Screening Test Parameters in Healthy Pregnant Women for Konya Province

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**Aim:** The prenatal risk calculation system produces detailed analysis reports and risk estimates for fetal genetic disorders using biochemical markers, ultrasound measurements and demographic data. The double screening test performed in the first trimester of pregnancy is used to detect diseases such as Down syndrome, Trisomy 13, Trisomy 18 and Neural Tube Defects. The median values used in such screening tests are derived from standardized data sets covering a wide range of populations. However, geographical, ethnic and other demographic differences between populations require the construction of regional median values. The aim of this study was to determine the new median values obtained from a population of healthy pregnant women in Konya province and to compare them with the median values from the current program used in screening tests. In this way, the aim was to optimize the performance of the screening tests and obtain more accurate results by taking into account regional characteristics.

**Materials and Methods:** This study was based on a retrospective evaluation of data from double screening tests performed on 736 healthy pregnant women at Konya City Hospital between 2021 and 2022. In this period, beta-human chorionic gonadotropin ( $\beta$ -hCG) and pregnancy-associated plasma protein-A (PAPP-A) test results were analyzed. Results: In the study, the median values for the free beta-hCG and PAPP-A values for the 11 to 13 weeks of pregnancy were recalculated for each week separately. The new median values obtained at 11 to 13 weeks gestation were compared with the reference median values recommended by the Prisca software. As a result of the analysis, the median values for free beta-hCG and PAPP-A were found to be statistically significantly different and lower than the current reference median values for each week ( $p < 0.001$  and  $p < 0.001$ , respectively).

**Conclusion:** The use of regional median values can contribute to more effective management of pregnant women by improving the accuracy and clinical effectiveness of prenatal screening tests. Re-evaluation of risk analyses using the new median values calculated in this study can help guide clinicians in the use of advanced diagnostic tests.

**Keywords:** Prenatal screening, First trimester screening, Median values , PAPP-A ,Free beta-hCG

## OA-10

### Immunophenotyping in Biphenotypic Cases; Case Report

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**Abstract:** Biphenotypic acute leukemia (BAL) is defined as the presence of multiple antigen co-expression on the same cell in leukemic blasts or the presence of two different types of blast populations. It constitutes 2-5% of all acute leukemias.

**Aim:** This study aimed to demonstrate the diagnostic role and importance of marker detection strategies used in flow cytometric (FC) immunophenotyping studies in patients with BAL.

**Methods:** Immunophenotypic analyzes from the bone marrow sample of the case diagnosed with Pre-B ALL in 2021 were performed with the FC method on the FACSCalibur (Becton Dickinson, USA) device, using markers of myeloid and lymphoid stem cells. In the analysis, CD45, CD19, CD7 CD3, CD4, CD117, CD34, HLA-DR, CD13 and CD33 markers were used in the immunophenotyping of the patient with a preliminary diagnosis of B-ALL. Subsequently, CD10, CD20, CD22, surface-IgM, c-CD79a, c-TdT, c-IgM markers were used. CD45, CD19, CD7, CD3, CD4, CD117, CD34, HLA-DR, CD38, CD13, CD33, surface-IgM, c-CD22, c-CD79a, c-IgM, TdT, CD20, CD10 markers were used in the immunophenotypic analysis performed on the Navios EX Flow Cytometer (Beckman Coulter, Ireland) device from the CSF sample of the patient, whose analysis was requested with the preliminary diagnosis of B-ALL in 2023. Following this, CD11b, CD14, CD15 and MPO markers were analyzed in accordance with the immunophenotypic character of the blastic area determined by the CD45/SSC graph.

**Results:** In the immunophenotyping study performed on the bone marrow in 2021, in the analysis performed in the blastic area determined by the CD45 / SSC graph, the patient was evaluated as Pre-B ALL, taking into account clinical consultation and other laboratory findings. In the immunophenotyping study performed on the CSF sample in 2023, CD19 %71, CD20 %42, CD4 %84, yüzey-IgM % 43, c-CD22 % 87, c-IgM %47, CD 38 % 44 was positive in the gated area; c-CD79, CD3, TdT, CD10 was found to be negative. The analysis result was compatible with Pre-B ALL, which was diagnosed in 2021, but due to the positivity of CD13, CD33, CD34, HLA-DR in the cells, it was necessary to evaluate the presence of an additional pathology of myeloid origin. Since CD3, CD11b, CD 14 positive, CD117, MPO negative were detected, it was understood that it was compatible with AML. According to these results, the patient was reported as biphenotypic Pre-B ALL / AML M0.

**Conclusion:** In immunophenotyping analyses, it is extremely important to determine the marker panels used in accordance with preliminary diagnosis and clinical evaluations. In order not to miss BAL cases, analyzes for markers of lymphoid and myeloid cell lines should be performed together in the first step in immunophenotyping analyses.

**Keywords:** Flow Cytometry, Biphenotypic Acute Leukemia, CD Markers

## OA-11

### Evaluation of Labsan Tricell-1000 Automated Urine Analyzer for the Erythrocyte and Leukocyte Count Performances

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**Aim:** Urine analysis is one of the most frequently studied tests in clinical laboratories. Automated urine analyzes are especially preferred for laboratories with intense workload. In our study, we aimed was to compare the results of erythrocyte and leukocyte counts on Labsan Tricell-1000 device with manual microscopy

**Methods:** A total of 100 first morning urine samples were studied by both Labsan Tricell-1000 and manual microscopy concurrently. The degree of concordance (Kappa coefficient) were evaluated. Analytical performance specifications for Kappa coefficients value of >0.6 as a minimum, and >0.8 as an optimal performance specifications were defined in European Urinalysis Guideline. The sensitivity and specificity value for the Labsan Tricell-1000 compared to manual microscopic examination were assessed.

**Results:** The degree of concordance of erythrocyte and leukocyte counts in microscopy of 100 urine specimens with manual microscopy was found to be 0.82 and 0.82 respectively (kappa coefficient). The sensitivity and specificity values of RBC and WBC were calculated for Labsan Tricell-1000 as 94%, 98%, 95.83%, 100%, respectively.

**Conclusions:** In our study, The sediment microscopy analysis of erythrocytes (cells/HPF) and leukocytes (cells/HPF) with Labsan Trion TriCell-1000 without any corrections demonstrated good with manual microscopy.

**Keywords:** Automated urine analysis, erythrocyte, leukocyte

## OA-12

### Calculated Low-Density Lipoprotein-Cholesterol: Comparability of the Friedewald, Martin/Hopkins, and Sampson/NIH Equations in the Turkish Population

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**Aims:** Ultracentrifugation, which is the reference method for low-density lipoprotein-cholesterol measurement, has been replaced by direct assays because it is not practical in standard clinical laboratories. However, due to the high cost and limited added value of direct assays, predictive equations are used. In this study, our aim was to compare the predictive performances of the Friedewald, Martin/Hopkins and Sampson/NIH to a direct assay when the triglyceride level is below 400 mg/dL. **Methods:** A total of 21,813 lipid profiles analyzed with the Roche Cobas e702 device (Roche Diagnostics, Mannheim, Germany) in the ISLAB-2 Core Laboratory were included in the study. Those that misclassified patients based on low-density lipoprotein-cholesterol thresholds of 70 mg/dL and 190 mg/dL were identified as clinically relevant errors. All statistical analysis was performed using R version 4.1.2. Statistical Language.

**Results:** The percentage of results meeting desirable bias specifications ( $\pm 5.4\%$ ) was 22.9%, 30.2%, and 32.2% in the Friedewald, Martin/Hopkins, and Sampson/NIH equations, respectively. The percentage of results meeting the total allowable error specifications ( $\pm 11.8\%$ ) was 62.8%, 76% and 75% in the Friedewald, Martin/Hopkins and Sampson/NIH equations, respectively. The Friedewald, Martin/Hopkins and Sampson/NIH equations showed negative proportional biases with a median bias of  $-9.6\%$ ,  $-7.8\%$  and  $-7.7\%$ , respectively. Despite this, Friedewald, Martin/Hopkins and Sampson/NIH equations had a good correlation with the Roche Cobas direct LDL-C assay (rs: 0.972; rs: 0.978; rs: 0.977, respectively). The rates of clinically relevant errors for the Friedewald, Martin/Hopkins, and Sampson/NIH equations were 8.3%, 6.7%, and 6.9%, respectively.

**Conclusions:** Our findings suggest that the Sampson/NIH and Martin/Hopkins equations performed better than the Friedewald equation in terms of desirable bias and total allowable error. In addition, all three equations tended to underestimate in our study group with negative proportional bias. In high-risk patients, current guidelines advocate for lowering LDL-C to under 70 mg/dL for secondary prevention, utilizing the Martin/Hopkins formula for more accurate prediction. The comparability of the Martin/Hopkins and Sampson/NIH equations was assessed on the Roche Cobas platform, revealing only a minimal overall difference between the two. We recommend monitoring patients using one of these methods, with each laboratory validating its chosen equation to ensure consistency and accuracy, thereby preventing variations between methods.

**Keywords:** Low-Density Lipoprotein Cholesterol; Cardiovascular Disease; Friedewald Equation; Bias

## OA-13

### Investigation of the Cause of High Glucosuria in a Euglycemic Patient, Case Report

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**Aim:** Glucose is filtered freely through the glomeruli. The glucose concentration in the glomerular ultrafiltrate is the same as in plasma. 90% of glucose is actively reabsorbed from the proximal tubule and 10% from the distal tubule. When a certain threshold is exceeded, glucose begins to be excreted in the urine, which is called glucosuria. Sometimes glucose can be seen in the urine without exceeding this threshold value in the blood; this is called renal glycosuria. Despite the normal glucose level in the serum sample studied simultaneously in our laboratory, a spot urine glucose result as high as 1762 mg/dL aroused our curiosity, so we aimed to investigate the cause of this result.

**Method:** Fasting blood glucose and Spot Urine Glucose were measured using the spectrophotometer method on the Abbott C.16000 autoanalyzer, urine glucose was measured using the photometer reflectance method on the Dirui H-800 automatic urine analyzer, and HbA1c was measured using the HPLC method on the Adams 8180-V analyzer.

**Results:** The results of a 47-year-old patient who applied to the internal medicine outpatient clinic, noticed during approval: Serum Glucose: 91mg/dL, Spot Urine Glucose: 1762 mg/dL, HbA1c%: 5.8, +++ Glucose in Complete Urine Analysis. The patient's medical history included diagnoses of Hypertension, Type-2 Diabetes, Asthma and Breast Fibrocyst. Tests at last year were Glucose: 91, 93, 95, 104 and HbA1c%: 5.8, 6.1, 6, 5.2. When the simultaneous analysis results were examined, it was seen that blood sugar regulation was achieved. Therefore, the possible causes of glucosuria were investigated. Consultation was made with the patient's physician for preanalytical error or source of interference, after ensuring that there was no analytical error. It has been learned that sodium-dependent-glucose-transporter-protein-2 inhibitor is used for the treatment of diabetes. Sodium-dependent-glucose-transporter-protein-2 inhibitors increase urinary glucose excretion by reducing glucose reabsorption in the kidney. As a result, it causes a decrease in plasma glucose and %HbA1c values. The raw material of sodium-dependent-glucose-transporter-protein-2 inhibitors is Phlorizin, a fermented poison obtained from the roots of the apple tree.

**Conclusion:** Other causes of glucosuria that are not accompanied by hyperglycemia include many reasons such as Renal Glucosuria, Fanconi Syndrome, Phlorizin, Heavy Metal Salts, Curare, Carbon Monoxide, Caffeine, Morphine, Strychnine, Chloroform, Glucoglycuria, Glucosuria of Pregnancy, Chronic Kidney Diseases. During patient results approved, we believe that not only the patient's previous diagnoses and previous results, but also the drug information used by the patient should be accessible when necessary.

**Keywords:** Glucosuria, drug effect

## OA-14

### Misleading High FSH Levels in an Asymptomatic Male Patient

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**Objective:** Laboratory data are critical for diagnosis and subsequent treatment strategies. However, various factors can influence laboratory data, leading to discordant interpretations of clinical outcomes. Macro-hormones primarily result from the binding of hormones to autoantibodies, leading to elevated serum concentrations due to slow clearance of hormone- autoantibody complexes. Hypophyseal gonadotropin adenomas are common, but secretion of biologically active luteinizing hormone and follicle-stimulating hormone is very rare. Follicle-stimulating hormone-secreting pituitary adenoma (FSHoma) is extremely rare and typically asymptomatic. It can lead to a presentation similar to ovarian hyper-stimulation syndrome in female patients. However, in male patients, there is usually no symptom or clinical indication. Additionally, macro forms of elevated FSH and LH are also very rare. The aim of this study is to present a rare clinical case related to the misinterpretation of serum follicle-stimulating hormone (FSH) levels as macro-FSH, which plays a critical role in the diagnosis, monitoring, and treatment process of clinical diseases.

**Methods:** Laboratory tests were requested for a 68-year-old male patient diagnosed with Benign Prostatic Hyperplasia (BPH) who presented to our urology outpatient clinic for routine follow-up without any complaints (FSH/LH: 12.70/11.2 IU/L, Total Testosterone: 9.95 ng/mL). Upon detection of high follicle-stimulating hormone (FSH), luteinizing hormone (LH), and Total Testosterone levels, the patient was referred to the endocrinology and metabolism outpatient clinic. Subsequent laboratory measurements also revealed high follicle-stimulating hormone (FSH) and luteinizing hormone (LH) levels. The patient had no history of exogenous use. Pituitary and pituitary gland imaging were performed based on the preliminary diagnosis of Hypophyseal adenoma - Gonadotropinoma (FSHoma).

**Findings:** On pituitary MRI imaging, the sella cavity was of normal width, and there was no evidence supporting the diagnosis of Gonadotropinoma upon natural visualization of the pituitary gland. The patient had no clinical symptoms. For the FSH and LH levels that were high in the second measurement, they were re-measured after precipitation with PEG in the laboratory (FSH/LH: 15.70/12.2 IU/L).

**Conclusion:** A decrease in FSH and LH parameters was observed following precipitation with polyethylene glycol (PEG) (FSH/LH: 7.94/1.87 IU/L). According to research, when the recovery is above 40% after polyethylene glycol (PEG) precipitation, it is considered that serum analyte levels are high due to macro-FSH. Macro-hormones are diagnosed by low recovery after precipitation with PEG (high sedimentation), high binding to protein G or protein A columns, and the presence of large molecular weight hormones in gel filtration chromatography (GFC). In clinical practice, tests based on hormone-dependent interpretation require careful evaluation for the diagnosis of macro-hormones. The most commonly used method in our daily practice is precipitation with polyethylene glycol (PEG). Anti-follicle-stimulating hormone (FSH) antibodies may be associated with macro-follicle-stimulating hormone (FSH). In rare clinical situations, contacting a medical biochemist prior to evaluating unexpected results can prevent unnecessary advanced investigation methods.

**Keywords:** Gonadotropinoma, Macro-FSH, Rational laboratory

## OA-15

### Performance Comparison of 2 Hematology Analyzers: Urit BH 6800 and Mindray 6000

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**Aim:** Complete blood count is at the top of the list of tests ordered in both routine and follow-up diagnosis and treatment of patients. The development of technology in recent years has facilitated the entry of new companies into the sector. It is very important to determine the analysis performance of new analyzers to be accepted into the laboratory by comparing them with existing devices. In this view, we aimed to compare the analytical performance of the Urit BH 6800, which we have recently started to use in our laboratory, with the Mindray 6000. Method: For repeatability, peripheral blood samples of 3 randomly selected patients were collected and each ran 20 times in a row. For reproducibility, 3 levels of control material were run 5 times for 5 days consecutively according to CLSI 15-A3 guideline. 527 different samples run on both analyzers one after the other. Samples were kept in K2 EDTA tubes at room temperature and run on both devices within 2 hours of arrival. WBC, LYM, MON, NEU, EOS, BASO, IMG, RBC, HGB, HCT, MCV, MCH, MCHC, PLT, MPV, PDW, PCT, PLCR, RDW\_CV, RDW\_SD parameters were evaluated. Correlation, Passing-Bablok regression and Bland-Altman statistical analyses were performed by using MedCalc and Excel software. Results: Both within-run and between-run %CV values are in the specified range that is indicated in European Federation of Clinical Chemistry and Laboratory Medicine's biological variation database. Besides all tests showed well correlation ( $r=0.433$  to  $0.998$ ) and the significance level was  $p<0.001$ . Conclusion: Due to the fact that there is a good correlation between the tests, %CV values used for assessment of repeatability and reproducibility are within the defined limits and the difference between compared measurements is minimal, Urit BH 6800 and Mindray 6000 hematology analyzers can be used interchangeably. Since there is limited information in the literature on this model of the newly introduced Urit BH 6800 analyzer, it is recommended to confirm the information we have obtained with more comprehensive new studies.

**Keywords:** complete blood count; hematological analyzer; comparison; Urit BH 6800; Mindray 6000



## OA-16

### MiR-181a-3p Expression Reduced in Aortic Tissues of the Patients with Ascending Aortic Aneurysm

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**Aim:** Ascending aortic aneurysm, defined as bulging in the ascending part of aorta, is a serious health problem which can cause life threatening complications such as aortic dissections or ruptures and mostly silent in early stages. Recent studies suggest that non- protein coding DNA and non-coding RNAs play an important role in the execution of complex biological functions. Among these, microRNAs (miRNAs; 21-25 nucleotides) are one of the most researched groups which are identified as key regulators in governing physiological and pathological processes. miR-181a-3p is known for its role in regulation of angiogenesis and inflammation on the vessel walls. However, there is no knowledge about the relationship between miR-181a-3p and ascending aortic aneurysm.

**Methods:** Our study included patients planned for coronary bypass surgery as control subjects (n=25, mean age: 63±10 years) and patients diagnosed with ascending aortic aneurysm as experimental group (n=25, mean age: 58±10 years) who were admitted to 3rd step education and research hospital between April 2023 and April 2024. Experimental group subjects were selected amongst patients scheduled for elective aortic aneurysm surgery with aortic diameter higher than 5,5 centimeters. miRNA expression levels were measured by simultaneous Reverse-Transcriptase Polymerase Chain Reaction (qRT-PCR). This study received approval from the Ethical Committee of Haydarpaşa Numune Training and Research Hospital, Istanbul, Türkiye. Results were evaluated using chi- square, logistic regression analysis, ROC analysis and Mann-Whitney U tests.

**Results:** No significant difference was found between the patient and control groups in terms of age, gender distribution, blood pressure, renal function and ejection fraction. When the patient group was compared with the control group, miR-181a-3p expression was significantly lower (p=0.018). The ROC analysis shows that when the cut-off point of the miR-181a-3p expression was 0.16, the sensitivity and specificity were 52% and 89%, respectively (area under the curve 0.717) for demonstrating ascending aortic aneurysm. However, there was no relation between aortic insufficiency and miR-181a-3p levels in both groups.

**Conclusion:** According to our findings, miR-181a-3p gene expression was decreased in the aortic tissues of the patients with ascending aortic aneurysm. However, our study continues to investigate miRNA levels in patient serums to understand its diagnostic value.

**Keywords:** Ascending aortic aneurysm, miRNA, miR-181a-3p

## OA-17

### Investigation of HbA2 Levels and Beta Thalassemia in Carriers of Hemoglobinopathy

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**Aim:** Hemoglobinopathies and beta thalassemia are health problems that cannot be ignored in our country. Although the rate of beta thalassemia carriage is 2% in Turkey, this rate is reported to be up to 10% in some regions. The prevalence of hemoglobinopathy in Turkey is approximately 1%. Although their individual prevalence is well known, there is insufficient data on their association. In this study, we aimed to examine HbA2 levels and association with beta thalassemia in carriers of hemoglobinopathy.

**Methods:** Hemoglobin variant results obtained from family medicine centers' patients from 01/01/2021 to 01/10/2023 and analyzed by high-performance liquid chromatography (HPLC) method on Arkray Adams HA8180T analyzer in the central laboratory of our hospital were included in the study. HbE carriers were excluded because HbA2 levels could not be determined due to similar elution times. The prevalence of elevated HbA2 (>3.5%) and the percentages of HbA and variant hemoglobins of HbS, HbD and HbC carriers were evaluated as well as complete blood count results.

**Results:** We observed that 49 (73%) of 67 HbC variant carriers and 33 (22%) of 153 HbS carriers had elevated HbA2 levels, while there was no patient with elevated HbA2 in 131 HbD carriers. The prevalence of elevated HbA2 in HbC carriers was higher than HbS carriers ( $p < 0.001$ ). When the HbA percentages of HbC and HbS carriers who had elevated HbA2 were analyzed; only two (4%) of HbC carriers and two (6%) of HbS carriers have lower HbA percentages than their variant percentages. The complete blood count results also supported the beta thalassemia carriage in these patients. On the other hand, no patient has an HbA percentage lower than the HbD percentage in HbD carriers.

**Conclusion:** In our study, we found that the prevalence of elevated HbA2 in HbC and HbS carriers was very high but most of their percentages of HbA suggest that there is no association of beta thalassemia as compound heterozygous. These results indicate that the use of HbA2 >3.5% as a cut-off value for association with beta thalassemia in HbC and HbS carriers is not appropriate by HPLC method. The lack of elevated HbA2 levels in HbD carriers suggests that the HbA2 levels were not falsely elevated by HPLC method in HbD carriers. However, since there are no compound heterozygous HbD/beta thalassemia patients according to HbA percentages, it remains unclear whether >3.5% can be used as the HbA2 cut-off value for association with beta thalassemia in HbD carriers by HPLC method. Further studies including capillary electrophoresis and genetic analysis are needed in this regard.

**Keywords:** HbA2, Beta Thalassemia, Hemoglobin variants, HPLC

## OA-18

### A Retrospective Evaluation of Serum Immunofixation Electrophoresis for the Percentage of IgD Monoclonal Gammopathy

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**Aim:** Multiple myeloma is a neoplastic disease characterised by the proliferation of plasma cells in the bone marrow that produce monoclonal immunoglobulins. The presence of a serum IgD monoclonal protein, M protein or paraprotein is observed in less than 1% of patients with monoclonal gammopathies and is generally indicative of a malignant plasma cell disorder. In this study, we analysed the results of serum immunofixation electrophoresis performed in our laboratory over one year and determined the percentage of IgD monoclonal gammopathy.

**Methods:** This retrospective study includes the results of serum immunofixation electrophoresis performed in a city hospital laboratory from 30 April 2023 to 30 April 2024. We routinely screen for IgG, IgM, IgA, kappa, and lambda gammopathies by serum immunofixation electrophoresis, and perform further testing for IgD and IgE in patients with free kappa or lambda light chains on serum immunofixation electrophoresis.

**Results:** of the 4336 patients tested by serum immunofixation electrophoresis, 959 were identified as having monoclonal gammopathy, and 48 (5%) had free kappa or lambda light chains. Only 2 (0.2%) of these 959 patients were diagnosed with IgD monoclonal gammopathy. The first patient was a 56-year-old man with anaemia and bone lesions. Serum protein electrophoresis revealed an M peak in the gamma globulin region, quantified at 3.4 g/L. Serum immunofixation electrophoresis revealed IgD lambda and free lambda monoclonal gammopathies. Clinical information was not available as the patient was treated at an external centre. The second patient was a 51-year-old man who presented with left shoulder pain of three months' duration. Serum protein electrophoresis showed an M peak in the gamma globulin region quantified at 4.7 g/L. Serum immunofixation electrophoresis identified a monoclonal IgD kappa band, with an IgD level measured at 9786 mg/dL. The patient was started on bortezomib, lenalidomide and dexamethasone. After three cycles of chemotherapy, the patient was scheduled for autologous stem cell transplantation.

**Conclusion:** IgD multiple myeloma is a rare variant of myeloma. Although the overall survival of patients with IgD multiple myeloma is shorter compared to other multiple myeloma subtypes, patient outcomes are improving with the use of novel agents and autologous stem cell transplantation. The proteasome inhibitor bortezomib and the immunomodulatory agent lenalidomide have been shown to be effective against the disease. The diagnosis of patients with IgD multiple myeloma is therefore extremely important. If myeloma is suspected and only one monoclonal light chain is detected in the serum, the patient should be tested for the presence of IgD and IgE monoclonal proteins.

**Keywords:** IgD, monoclonal gammopathy, serum immunofixation electrophoresis

## OA-19

### Are there Relationship Otosclerosis with Serum HE4 and CA125 Level? : A pilot Study

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**Aim:** Otosclerosis is a hearing disorder caused by sclerotic bone in the ear that forms as a result of repeated cycles of osteolysis and osteogenesis. There are recent studies that serum cancer antigen-125 (Ca 125) and human epididymis protein 4 (HE4) levels has been identified as biomarkers in some fibrosis diseases. The purpose of this pilot study was to evaluate the value of HE4 and CA-125 in otosclerosis patients.

**Method:** The study population consisted of 60 people (30 otosclerosis patients, 30 control group). We collected blood samples for HE4 and CA-125 levels. Serum HE4 and CA-125 levels were measured by enzyme-linked immunosorbent assay (ELISA). We compared the results between otosclerosis patients and the normal subject. The area under the curve (AUC) of the receiver operating characteristic (ROC) curve was performed to evaluate the diagnostic value.

**Results:** There was no differences in CA-125 level between the otosclerosis (20.3 U/mL [10.4-42.1] and control group (19.3 U/mL [15.3- 49.8]) ( $p>0.05$ ). HE4 level was significantly higher in the otosclerosis group (60.9 pmol/L [32.1-101.8]) than the control group (25.3 pmol/L [12.4-91.9]) ( $p<0.001$ ). The AUC in ROC analysis of HE4 was 0.768 ( $p<0.001$ ).

**Conclusion:** This is the first clinical study to evaluate the diagnostic values of serum HE4 levels in diagnosed otosclerosis patients. Serum HE4 level may be a useful biomarker in otosclerosis. Further studies with a larger number of patients are required to confirm our pilot results.

**Keywords:** otosclerosis, serum cancer antigen-125, human epididymis protein 4, fibrosis diseases

## OA-20

### Evaluation of the Diagnostic Utility of Inflammatory Indices in Cases of Acute Appendicitis

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**Objective:** Acute appendicitis (AA) is one of the most common causes of hospitalization among patients presenting with abdominal pain to the emergency department (1). The clinical diagnosis of acute appendicitis relies on the patient's history, physical examination, laboratory evaluation, and imaging findings (2). In recent times, laboratory findings have gained prominence in the diagnosis of acute appendicitis due to their easy accessibility and rapid turnaround time (3). Indices calculated from values obtained through hemogram analysis indicate the presence of inflammation. Inflammatory indices that evaluate different components of complete blood count together, such as Neutrophil Lymphocyte Ratio (NLO), Platelet Lymphocyte Ratio (PLO) and the systemic immune inflammation index (SII), have been shown to be useful in predicting the prognosis and/or monitoring of various diseases (4). In this study, we aimed to determine the usefulness of inflammatory indices such as SII, NLO, and PLO as effective parameters in the diagnosis of acute appendicitis.

**Method:** The study was conducted retrospectively and at a single center between November 2023 and May 2024 at the Emergency Department of Konya City Hospital. It included 60 patients diagnosed with acute appendicitis based on clinical findings and radiological imaging, who met the inclusion criteria, and a control group of 60 healthy individuals who presented for routine health examination and were evaluated as healthy. Demographic data, medical history, WBC, platelet count, neutrophil count, lymphocyte count were obtained from the Laboratory Information Management System (LIMS) and recorded in the created dataset. SII was calculated using the platelet count  $\times$  neutrophil/lymphocyte count formula. Data were analyzed using SigmaPlot 12.0 Program.

**Results:** The neutrophil values of patients with acute appendicitis ( $9.78 \pm 0.5$ ) were statistically significantly higher compared to the neutrophil values of the control group ( $4.1 \pm 1.1$ ) ( $p < 0.001$ ); while the lymphocyte values ( $2 \pm 0.7$ ) of patients with acute appendicitis were statistically significantly lower compared to the lymphocyte values of the control group ( $2.33 \pm 0.5$ ) ( $p < 0.005$ ). The SII values of patients with acute appendicitis ( $1571.1 \pm 1335.6$ ) were statistically significantly higher compared to the SII values of the control group ( $500.5 \pm 227.2$ ) ( $p < 0.001$ ). The NLO values of patients with acute appendicitis ( $5.99 \pm 4.9$ ) were statistically significantly higher compared to the NLO values of the control group ( $1.84 \pm 0.69$ ) ( $p < 0.005$ ); whereas there was no statistically significant difference between the PLO values of patients with acute appendicitis ( $146.2 \pm 75.4$ ) and those of the control group ( $121.4 \pm 39.2$ ) ( $p = 0.02$ ).

**Conclusion:** Considering the findings of the study, we believe that SII and NLO can be used as effective parameters to support the diagnosis of acute appendicitis. Evaluation of inflammatory markers along with clinical findings will reduce both misdiagnoses and rates of unnecessary surgery. Additionally, this approach may reduce the need for diagnostic imaging tests that expose the patient to radiation and incur additional costs, such as contrast-enhanced computed tomography. These indices, being easy to calculate and cost-effective, relying solely on complete blood count, and not based on subjective findings, have the potential to provide more accurate results in the diagnosis of acute appendicitis (AA).

**Keywords:** Acute appendicitis, SII, NLO, PLO

## OA-21

### Analytical Evaluation of the Trinity Biotech Premier Hb9210 and Comparison with the Sebia Capillarys 3 Tera for HbA1c Determination

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**Aim:** Glycated hemoglobin measurements are valuable for long-term glycemic control and the diagnosis of diabetes. The methods of choice for HbA1c measurement in clinical practice are the following: ion-exchange chromatography; affinity chromatography, capillary electrophoresis, immunoassay and enzymatic. The aim of this study was to evaluate analytical performance and compare two different assays for HbA1c measurement, Trinity Biotech Premier Hb9210 boronate affinity high- performance liquid chromatography (HPLC) and capillary electrophoresis system Capillarys 3 Tera (Sebia, France) in different patient groups.

**Methods:** We assessed imprecision, estimation of bias, linearity and carryover of Hb9210 instrument with respect to the Clinical and Laboratory Standards Institute (CLSI) guidelines EP15-A3, CLSI EP10-A3, and EP06-A protocols. The method comparison study was performed with Capillarys 3 Tera (Sebia, France) by using Deming regression and the Bland-Altman plot methods as recommended in EP09-A3. A total of 120 patients with non-diabetes, pre-diabetes or diabetes mellitus were also included in the study for method comparison. The precision study conducted with patient samples and internal quality control (IQC) materials were evaluated according to imprecision limits sourced from The American Diabetes Association (ADA) guidelines. Other acceptance criterias were based on manufacturer technical specifications and the EFLM Biological Variation Database.

**Results:** Imprecision estimates in form of repeatability %CV (CVR) and within laboratory %CV (CVWL) were below the analytical allowable imprecision (CVA) defined as <2% for National Glycohemoglobin Standardization Program [NGSP] units and <3% for International Federation of Clinical Chemistry and Laboratory Medicine [IFCC] units in ADA guidelines for all samples in both methods. The estimated bias was within verification limits. However, the method comparison study showed concentration-dependent variations according to Deming regression. In the comparison study, the Bland-Altman plot used to compare the results of the methods showed good agreement and mean % difference is below desirable bias according to biological variation (BV) limits. This difference is greater at higher concentrations. Carryover effects were within acceptable criteria manufacturer's claims. The method proved to be linear in the concentration range of 4.5- 14.6%. Conclusion: The two HbA1c methods commonly used with commercial analyzers showed a good reliability and comparability. However, laboratories should be aware of the limitations of their methods and the availability of more accurate and precise HbA1c determination methods.

**Keywords:** Analytical performance, method comparison, hemoglobin A1c, boronate affinity chromatography, capillary zone electrophoresis

## OA-22

### Comparison of the Effects of BD Vacutainer® Plus and Vacusera® Urine Preservative Tubes on Routine Urinalysis

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**Aim:** A complete urinalysis is one of the most frequently performed tests in clinical laboratories. Despite the advances in the performance of fully automated urine analyzers in recent years, the pre-analytical phase of modern analysis still requires improvement. According to the CLSI GP16A3 guidelines, the recommended analysis time for urine samples is within 2 hours after collection. However, if the analysis exceeds this period, the samples must be stored under controlled temperature or preservatives must be used for accurate results. The aim of this study is to evaluate the stability of chlorhexidine-based preservative tubes and no-additive urine tubes stored at +4 degrees on strips and microscopy.

**Method:** In this study, 44 pathological urine samples were included. Fresh urine samples were filled into non-additive tubes, and analyses were done within 1 hour. Each sample (8 mL) which was aliquoted into Becton–Dickinson Vacutainer® Plus Urinalysis Preservative (BD-UAP) tubes and VACUSERA® Urine Tube - Chlorhexidine tubes, were evaluated after 4, 8, 12, 24, 48, and 72 hours of storage and compared with samples refrigerated in no additive polystyrene tubes (PS) after 4 and 8 hours. All analyses were performed on H- 800 and FUS-200 automatic modular urine analyzers (Dirui Industry, Changchun, China). The automated classification was visually checked on the screen, and corrected if necessary by the same laboratory specialist. Cohen's kappa coefficient ( $\kappa$ ) was calculated to assess the agreement of results. A  $\kappa > 0.80$  was accepted as perfect agreement, indicating that the analytes were stable.

**Results:** Specific gravity, pH, glucose, and nitrite were stable in all tubes and conditions. Although all bilirubin results were negative, had 100% agreement in all tubes and conditions. Hemoglobin and urobilinogen showed perfect agreement between all the tubes and remained stable up to 8 hours. Protein remained stable in VACUSERA and BD-UAP tubes for up to 8 hours, but not in PS tubes. Ketone and erythrocyte remained stable up to 8 hours in the VACUSERA tubes but was stable only up to 4 hours in PS tubes. However, ketone and erythrocyte was not stable in the BD-UAP tube at any time period. Bacteria was well preserved in PS and VACUSERA tubes up to 8 hours whereas in BD-UAP tubes up to 4 hours. Leukocyte esterase were stable up to eight hours in PS and BD-UAP tubes whereas remained stable for up to 48 hours in VACUSERA tube. Leukocyte did not remain stable in any tube at any hour.

**Conclusion:** The first important finding of this study is that specific gravity, pH, glucose, and nitrite can be stored for up to 72 hours in both chlorhexidine-based preservative tubes. The second notable finding of this study is that chemical parameters generally show better agreement and stability than microscopic parameters. Another remarkable finding is that the VACUSERA tubes have a longer stability period than BD tubes for ketone, erythrocyte, leukocyte esterase and bacteria parameters. In conclusion, using a preservative tube offers comparable results with refrigeration for the majority of parameters for 4 hours and up to 8 hours.

**Keywords:** Urinalysis, preanalytical phase, preservative tube, chlorhexidine

## OA-23

### Evaluation of Total Oxidant and Total Antioxidant Status During Clinical Follow-Up In Patients with Membranous Nephropathy

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**Introduction:** Membranous nephropathy is one of the common causes of adult-onset nephrotic syndrome. This autoimmune disorder primarily targets the kidneys, leading to the accumulation of subepithelial glomerular immune complexes containing immunoglobulin G autoantibodies and complement proteins.

**Objective:** Oxidative stress occurs in response to the oxidative damage caused when the body's antioxidative and scavenging activities cannot cope with the active oxidants produced by a harmful stimulant. Oxidative stress involves macromolecular oxidative damage, induces tissue protein denaturation, DNA damage, and lipid peroxidation, and interferes with the body's normal metabolic activity, leading to the occurrence and/or development of diseases. This study included 18 patients (9 males, 9 females) diagnosed with membranous glomerulonephropathy based on biopsy results and was designed to investigate the level of oxidative stress in membranous nephropathy patients.

**Methods:** 18 patients (the mean age was  $61.5 \pm 12.6$  years) were enrolled in this study. Blood samples (at baseline and at the 3rd month of the clinical follow-up process) were collected and separated from the cells by centrifugation at 4000 rpm for 10 min and were stored at  $-80^{\circ}\text{C}$ . Total Antioxidant Status (TAS) was measured using the colorimetric method which was based on the color bleaching of reduced radical molecules after oxidation with hydrogen peroxide. Total Oxidant Status (TOS) was measured using the Erel method which was based on the oxidation of ferrous ion to ferric ion in the presence of various oxidative species under acidic conditions. Spectrophotometric monitoring was used for TAS (660 nm) and TOS (530 nm). Oxidative Stress Index (OSI) was calculated by using the following formula:  $\text{OSI (Arbitrary Unit)} = (\text{TOS, } \mu\text{mol H}_2\text{O}_2 \text{ Eq/L}) / (\text{TAS, } \mu\text{mol ascorbic acid Eq/L})$

**Results:** At baseline, Total Oxidant Status and Total Antioxidant Status levels were  $0.9358 \pm 0.3392$  and  $3.2192 \pm 0.1410$ , respectively. Corresponding values at the 3rd month were  $0.6273 \pm 0.2189$  and  $3.1660 \pm 0.1953$ . No significant differences were observed across these periods ( $p=0.628$ ,  $p=0.067$ ). Oxidative Stress Index levels (non-normally distributed) were  $0.2797 \pm 0.1553$  at baseline and  $0.1840 \pm 0.0797$  at the 3rd month. The latter values were significantly 2.59 times lower compared to baseline ( $p=0.01$ ).

**Conclusion:** Although there was no significant difference between the levels of TAS and TOS, it is important to investigate the mechanisms underlying the differences in the oxidative stress index. This difference may indicate that the levels of antioxidants and oxidants need to be evaluated not only individually but also in relation to each other. Considering that oxidative stress is a complex process and involves multiple factors, it may be associated with the progression and status of the disease. Perhaps disease duration, treatment response, age groups, gender or other clinical variables could explain these differences in the oxidative stress index. To support the obtained findings and provide further insights, more comprehensive and long-term research can be conducted. These studies can help us better understand the complexity of oxidative stress and utilize this knowledge in clinical practice.

**Keywords:** Membranous nephropathy, Total Antioxidant Status, Total Oxidant Status, oxidative stress



## OA-24

### Hypertension Induced Thrombotic Microangiopathy: Case Report

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**Aim:** Thrombotic microangiopathies are rare but life-threatening disorders characterized by microangiopathic hemolytic anemia, thrombocytopenia, and microthrombus formation leading to tissue damage. Malignant hypertension may induce these conditions through microvascular thrombosis. It's important to distinguish thrombotic microangiopathy from conditions such as thrombotic thrombocytopenic purpura and hemolytic uremic syndrome. Systemic conditions associated with thrombotic microangiopathy include severe hypertension, systemic lupus erythematosus, pre-eclampsia, and HELLP syndrome. It can complicate transplantation, and untreated cases carry a high risk of mortality, emphasizing early detection. We present a case of hypertension-induced thrombotic microangiopathy in a 20-year-old man that progressed to end-stage renal disease, highlighting the diagnostic challenges and the need for early intervention.

**Methods:** A 20-year-old male presented to our hospital's emergency department with abdominal pain, nausea, vomiting, and dyspnea. In his medical history, it was stated that two months ago he went to another medical center because of weakness and syncope, where he was diagnosed with acute kidney injury and hypertension. However, after the first treatment, he refused to undergo further tests. He had no notable surgical or familial history. Examination at our emergency clinic revealed a blood pressure of 190/110 mmHg, consistent pulses, respiratory rate of 25 breaths per-minute and tachycardia. Further investigations showed serum urea 185.2 mg/dL, creatinine 10 mg/dL, hemoglobin 8.2 g/dL, ldh 398 IU/L and total bilirubin 0.36 mg/dL. The patient received antihypertensive treatment and emergency hemodialysis. He was then referred to the nephrology clinic for evaluation. Differential diagnoses included paroxysmal nocturnal hemoglobinuria, platelet dysfunction, sickle cell anemia, congenital dyserythropoietic anemia and infectious etiologies. Renal ultrasound and biopsy were planned.

**Results:** The Coombs test was negative and the peripheral smear showed schistocytes, indicating microangiopathic hemolytic anemia. Additionally, the patient had thrombocytopenia with a platelet count of 105,000 per-microliter. These findings, along with severe hypertension on admission, strongly suggested hypertension-induced thrombotic microangiopathy. Further tests included normal ADAMTS13 activity, negative blood and urine cultures, and screenings for various infections. No infection that could have caused this picture was found in the patient. Hemoglobin electrophoresis showed no abnormalities, and both platelet function tests and paroxysmal nocturnal hemoglobinuria flow cytometry panel results were normal. Renal ultrasound ruled out other causes of renal dysfunction, while serum and urine protein electrophoresis were negative for monoclonal paraproteinemia. Renal and portal Doppler imaging found no signs of thrombosis. The renal biopsy revealed glomerular sclerosis, hyperplastic vessels, arteriolar lipid changes suggestive of thrombotic microangiopathy, and thickening of the glomerular basement membrane. Renal failure was deemed chronic, and after excluding other etiologies, the diagnosis of hypertension-induced thrombotic microangiopathy was confirmed.

**Conclusion:** This case highlights a young patient with uncontrolled hypertension leading to secondary thrombotic microangiopathy. These rare, life-threatening disorders require early diagnosis and management. For hypertension-induced thrombotic microangiopathy, early and consistent treatment is crucial for prognosis. Uncontrolled hypertension increases the risk of end-stage renal disease and negatively impacts the economy. Distinguishing this condition from other etiologies like thrombotic thrombocytopenic purpura and hemolytic uremic syndrome is essential. Multidisciplinary approach is necessary to recognize and treat these conditions, emphasizing renal function and diagnostic challenges.

**Keywords:** Thrombotic Microangiopathy, Hypertension Induced Tma, Thrombotic Thrombocytopenic Purpura, Hemolytic Uremic Syndrome, ADAMTS13, Hypertensive Emergency, Thrombotic Microangiopathy

## OA-25

### A Case of Alkaptonuria with Mismatched Protein Results in Urine

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**Aim:** Alkaptonuria is a rare autosomal recessive disorder characterized by the inability to metabolize homogentisic acid and its accumulation in the body in the absence of the enzyme homogentisic acid oxidase. One of the earliest signs of the condition is dark-stained diapers, as homogentisic acid causes urine to turn black. The kidneys play a critical role in the elimination of homogentisic acid. However, the presence of renal disease in alkaptonuria is extremely rare and usually occurs with the progression of the disease. In our study, we aimed to describe a 9-year-old boy with alkaptonuria who was asymptomatic and had unexpected proteinuria.

**Methods:** A 9-year-old male patient who was diagnosed with alkaptonuria due to darkening of urine color at the age of 3 months was admitted to our hospital. No obvious abnormality was noticed on physical examination. Biochemical analysis of urine revealed 94.2 mg/dl (N:1-15 mg/dl) protein in the random urine of the patient. Similarly, the protein/creatinine ratio of 1139 mg/g creatinine (N:0-200 mg/g creatinine) was found to be increased in random urine. The patient was referred to the pediatric nephrology department to search for the etiology of proteinuria and was hospitalized with a biopsy plan.

**Results:** In the laboratory findings of the patient, renal function parameters were within normal ranges and proteinuria was not detected in a complete urinalysis performed with the colorimetric urine strip test method (URIT UC 1800, BT products Turkey). However, biochemical analysis of 24-hour urine by turbidimetric method based on the principle of creating turbidity by adding benzethonium chloride showed an increased 24-hour urine protein of 895 mg/24 hours (N:0-140). (Cobas c 720, Roche Germany) Due to mismatched urine protein results, urine protein electrophoresis was performed for confirmation and no proteinuria was found in consistent with the urine strip test. Proteinuria was not considered after evaluation of the patient's history, physical examination and laboratory results. Considering the data related to the patient's mismatched urine protein results, it was thought that homogentisic acid may have caused interference with benzethonium chloride and false positive results in urine biochemistry analysis by turbidometric method.

**Conclusion:** The most commonly used urinalysis method is the chemical analysis, which can be performed manually or automatically by using urine strips. The principle of the urine strip test for complete urinalysis is based on the error created by proteins on a pH indicator. However, the turbidometric method used in the biochemical analysis of the urine is based on the principle that absorbance increases when urine protein is precipitated by benzethonium chloride in an alkaline medium. It has been previously shown in the literature in a few cases that oxidation of homogentisic acid by alkaline conditions in the benzethonium chloride method causes interference and false positivity. Our observation led us to point out that homogentisic acid may interfere with urine protein analysis and give false positive results. This should be kept in mind to avoid unnecessary invasive procedures in patients with alkaptonuria.

**Keywords:** alkaptonuria, homogentisic acid

## OA-26

### Systemic Inflammatory Response Index and Vitamin D in Newly Diagnosed Pediatric Celiac Patients

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**Aim:** Celiac disease, triggered by gluten exposure in genetically susceptible individuals, is a chronic autoimmune condition. While small intestine biopsy remains the diagnostic 'gold standard,' serological tests like tissue transglutaminase antibodies have gained importance. However, their high cost and limited availability present challenges. Inflammatory cytokines play a role in celiac disease, with monocytes expressing them notably. Some studies suggest increased activation of circulating monocytes in celiac patients, possibly linked to intestinal epithelial damage. Vitamin D, beyond its classical role, regulates cell differentiation and proliferation, exhibiting anti-inflammatory effects. Research indicates its potential in reducing inflammatory factors secreted by monocytes, possibly protecting against inflammation. Studying complete blood count parameters for systemic inflammation and disease activity has become increasingly common due to easy affordability and accessibility. The Systemic Inflammatory Response Index evaluates overall inflammatory response. This study aims to evaluate Systemic Inflammatory Response Index and vitamin D in Celiac disease.

**Methods:** The study included 46 patients recently diagnosed with Celiac disease (25 girls and 21 boys; mean age: 10.89±4.40 years) and 52 healthy controls (28 girls and 24 boys; mean age: 10.96±3.02). The complete blood count was performed using the Sysmex XN-1000 automated hematology analyzer (Sysmex Inc, Kobe, Japan). Systemic Inflammatory Response Index was calculated by (neutrophil count×monocyte count)/lymphocyte count. Vitamin-D levels were detected by electrochemiluminescence immunoassay on Roche Elecsys system.

**Results:** The total leukocyte counts (x1000/uL) were 7.90 ± 2.58 and 6.96±1,62 (p=0.03), the neutrophil counts were 4.29 ± 2.34 and 3.44 ± 1.19 (p=0.02), the lymphocyte counts were 2,78 ± 0,96 and 2,61 ± 0,85 (p >0.05) and monocyte counts were 0.59 ± 0.21, and 0.43 ± 00.14 (p<0.0001) in patient group and control group, respectively. Systemic Inflammatory Response Index was determined as 1,19 ± 1,31 in the celiac group and 0.62 ± 0.34 in the control group (p=0.003). When evaluating the diagnostic significance of monocyte levels and Systemic Inflammatory Response Index measurements, the area under the ROC curve were calculated as 0.73 and 0.63, respectively. Although the vitamin D level (µg/L) was lower in the patient group than control group, there was no significant difference between the groups (20.61 ± 7.92 and 21.61 ± 6.89 (p>0.05)). There was a negative correlation between vitamin D levels and % monocyte counts (r:-0.270, p=0.07). However, this relationship was not statistically significant.

**Discussion:** In our study, the systemic inflammatory response index and monocyte values were found to be significantly higher in the celiac patient group. With this finding, considered that these parameters could be used for the diagnosis of celiac disease in situations where tissue transglutaminase-IgA cannot be measured. In the celiac group, a negative correlation was observed between vitamin D and monocyte percentage; however, this correlation was not found to be statistically significant. Nevertheless, it is thought that re-evaluating this relationship in studies with increased sample sizes would be beneficial.

**Keywords:** Celiac disease, Systemic Inflammatory Response Index, Vitamin D, Monocyte

## OA-27

### Development of a Decision Support System for Laboratory Experts in CSF Oligoclonal Band Analysis: Preliminary Data

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**Aim:** This study aimed to develop an image analysis system to assist laboratory experts in interpreting CSF oligoclonal band analysis membrane images for the determination of intrathecal IgG synthesis.

**Methods:** Membrane images of serum and CSF samples from a total of 1044 patients between 2010 and 2022 at the Central Laboratory of Dokuz Eylul University were evaluated using isoelectric focusing and immunoblotting methods (Interlab G26 Electrophoresis). All membranes were scanned at 600 dpi resolution. After cropping the date and information part, the digital images were converted to grayscale for analysis. The CLAHE algorithm was applied to enhance contrast, and the Rolling Ball algorithm was used to correct background variations. The row mean of the images was taken, and peaks were identified using a peak finding algorithm after applying a negative transformation. Possible oligoclonal band presences were evaluated using our custom-developed band detection software. The results obtained with our software were compared with the laboratory experts' evaluations.

**Results:** When categorizing patient results with type 1 and type 4 patterns as oligoclonal band negative and results with type 2 and type 3 patterns as oligoclonal band positive, the diagnostic performance of the semi-automatic system was found to have 84% specificity and 82.61% sensitivity. However, the system was not yet sufficient in distinguishing between type 3 and type 2.

**Conclusion:** In the diagnosis of Multiple Sclerosis, as stated in the 2017 McDonald criteria, the demonstration of specific oligoclonal bands in the cerebrospinal fluid is diagnostic. The demonstration of bands using the immunoblotting technique following immunoglobulin G isoelectric focusing is the current gold standard method. However, interpreting the results by laboratory experts is time-consuming and not standardized. In this study, we developed an image analysis system to assist laboratory experts in interpreting CSF oligoclonal band analysis membrane images in a semi-automatic manner. However, further studies are needed to improve the diagnostic performance of this system, particularly in differentiating between types.

**Keywords:** Oligoclonal Band, Image Analysis, Isoelectric Focusing, Diagnostic Analysis

## OA-28

### The Role of Serum Total Tau and Orexin Biomarkers in Early Diagnosis of Alzheimer's Disease

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**Aim:** Alzheimer's disease (AD), a neurodegenerative disorder, is the most common cause of dementia (50-75%). While amyloid plaque accumulation and neurofibrillary tangle formation play a fundamental role in the pathophysiology of AD, impaired protein clearance is the primary pathogenesis in spontaneous AD. Currently, non-invasive biomarkers targeting underlying pathologies of AD are being investigated to facilitate early diagnosis. Tau, reflecting axonal neurodegeneration, and orexin, reflecting protein clearance dysfunction, are among the markers associated with AD pathology. In this study, we aimed to examine the relationship between serum total tau and orexin levels and the risk of AD.

**Method:** Between February 2023 and February 2024, 86 patients diagnosed with Alzheimer's disease and 30 healthy individuals were included in our study, conducted at the Department of Neurology, Dementia Clinic, Prof. Dr. Mazhar Osman Mental Health and Neurological Disorders Training and Research Hospital. Mini-Mental State Examination (MMSE) and Clinical Dementia Rating Scale-Sum of Boxes (CDR-SOB) scores were recorded for the patient group. Serum samples obtained from both patient and control groups were analyzed for levels of total tau and orexin using the enzyme-linked immunosorbent assay (ELISA) method. Receiver operating characteristic (ROC) analysis was employed to evaluate the ability of these biomarkers to discriminate between Alzheimer's patients and the control group.

**Results:** According to the clinical dementia staging scale, the 86 patients were divided into three groups: 33 in the mild stage, 29 in the moderate stage, and 24 in the severe stage. The patient and control groups were comparable in terms of age ( $p=0.377$ ) and gender distribution ( $p=0.518$ ). Levels of total tau ( $p<0.001$ ) and orexin ( $p=0.002$ ) in the serum of Alzheimer's patients were significantly higher compared to the control group. When the ROC curve was plotted, it was observed that serum total tau values alone had the highest sensitivity and specificity. When orexin was combined with total tau, it was found to have higher sensitivity and specificity compared to orexin alone.

**Conclusion:** Our findings suggest that total tau alone is sufficient for the early diagnosis of Alzheimer's disease. However, the combination of total tau and orexin biomarkers could also be used as a screening test for AD.

**Keywords:** Alzheimer's disease, tau, orexin, early diagnosis

## OA-29

### Can Albumin Concentrations Obtained by Serum Protein Electrophoresis Be Used Instead of Direct Measurement?

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**Aim:** Serum protein electrophoresis is frequently used for the detection of monoclonal gammopathies. In serum protein electrophoresis, the concentrations of albumin and other bands can be determined using total protein results. Serum albumin measurement can be used for many purposes such as assessment of nutritional status, liver and kidney function, and prediction of corrected calcium results. Additionally, low albumin levels are associated with high mortality, and appropriate therapeutic use has been shown to improve prognosis. In this study, we aimed to investigate whether albumin concentrations obtained in different serum protein electrophoresis patterns can be used instead of albumin concentrations determined by direct measurement.

**Method:** The data used were obtained from 373 patients aged 18-90 years with serum protein electrophoresis, total protein, albumin and immunoglobulin results with the same barcode number analyzed between 01.09.2023 and 01.09.2024 in the Medical Biochemistry Laboratory of Eskişehir Osmangazi University Hospital. Patients with hemolysis index>500, ictericia index>20, lipemia index>550, CRP>5 mg/L and patients with biclonal gammopathy were excluded. Serum protein electrophoresis was performed with Helena V8 Nexus (Helena Biosciences, Gateshead, UK) capillary electrophoresis. Total protein, albumin and immunoglobulin levels were analyzed on a Cobas c702 (Roche Diagnostics, Mannheim, Germany) autoanalyzer using the biuret method, bromocresol green and immunoturbidimetric method, respectively. The relationship between serum protein electrophoresis and direct albumin measurements was evaluated by Method comparison & evaluation Bland-Altman plot analysis and Passing-Bablok regression analysis using MedCalc V22.023. In addition, the analysis was done in the subgroups that were created according to concentrations of total protein, Ig G and gamma band and presence/absence of monoclonal gammopathy. To assess clinical significance, the total acceptable error that calculated with intra-individual and inter-individual variations was obtained by median of direct albumin concentrations. The mean biases compared with the total acceptable error.

**Results:** When all patients were evaluated, although showed no significant deviation from linearity between serum protein electrophoresis and direct albumin measurements in Passing-Bablok regression analysis [ $P=0.49$ ,  $y = -0.61 + 1.08 x$ , (slope: 1.04 to 1.12, 95% CI), (intercept: -0.76 to -0.46, 95% CI)], mean bias 0.28 g/dL and the difference between the two methods was highly significant in Bland-Altman plot analysis ( $P<0.0001$ ). Since the mean bias was above the total acceptable error, it was considered clinically significant. Although Passing-Bablok regression analysis was linear in all other subgroups ( $P>0.05$ ) and intercept confidence interval included 0 and slope confidence interval included 1 in some groups, the mean bias in Bland-Altman plot analyses was found to be above the clinical significance level.

**Conclusion:** Regardless of total protein, Ig G, M-spike and gamma band levels, albumin concentrations calculated by serum protein electrophoresis were lower than those measured by bromocresol green in all groups and were considered clinically significant. Therefore, albumin levels calculated by serum protein electrophoresis cannot be used as a substitute for direct albumin measurement using bromocresol green method.

**Keywords:** Blood protein electrophoresis, serum albumin, bromocresol green

## OA-30

### Effect of Vacuum and Non-Vacuum Urine Tubes on Urinary Analysis

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**Objective:** Vacuum urine collection systems are widely used for sediment and chemical strip analysis of urine. Microscopic examination of urine sediments has become one of the most commonly used diagnostic tests because it is easily available, inexpensive and non-invasive. Moreover, it plays a critical role in the diagnosis, treatment and prognostic evaluation of many diseases, especially renal and urinary system diseases. Vacuum urine collection systems are used for these analyses because they are user-friendly, standardized, shorten the turnaround time and have a low risk of contamination. In our study, we aimed to investigate the effect of negative pressure in vacuum urine tubes on sediment and chemical strip analysis results.

**Method:** The study included 124 urine specimens collected from patients admitted to the urine laboratory at Istanbul Bakırköy Dr. Sadi Konuk Training and Research Hospital. Urine samples were collected with the collection cups used for vacuum tubes and then aliquoted to the collection cups used for non-vacuum tubes. The samples in the first collection cups were transferred to vacuum tubes with negative pressure and the samples in the other system were transferred to non-vacuum tubes. They are both analyzed on a Dirui FUS-200 sediment analyzer and Dirui H-800 strip analyzer (DIRIU Industrial Co., China). All samples were run consecutively by the same operator within 1 hour after the time of collection. Microscopic verification of the results obtained was performed on a Dirui FUS-200 sediment analyzer.

**Results:** The results of sediment and chemical strip analyses performed with vacuum and non-vacuum tubes were compared with Wilcoxon Signed Ranks Test using Statistical Package for the Social Science Version 26 for Microsoft Windows. In sediment analysis, there was no significant difference between vacuum and non-vacuum tubes for the results of leukocyte, squamous epithelium, non-squamous epithelium and hyaline casts, whereas there was a significant difference for the results of erythrocyte, crystal and granular casts ( $p < 0.05$ ). In chemical strip analysis, there was no significant difference between vacuum and non-vacuum tubes for the results of hemoglobin, bilirubin, ketone, urobilinogen, protein, nitrite, leukocyte, glucose and pH; however, there was a significant difference for the results of specific gravity ( $p < 0.05$ ).

**Conclusion:** In our study, we concluded that negative pressure tubes in urine collection systems may cause statistically significant changes in some parameters and that these should be evaluated by considering the patient's clinic. However, we believe that more studies including larger sample sizes with different patient groups, especially nephrology patients concerning the significance of these parameters in the management of these patients, are needed to determine whether the differences in these parameters affect the diagnostic and therapeutic course.

**Keywords:** Urine specimen collection, sediment analysis, urinalysis, strip analysis, vacuum



## OA-31

### The Role of Agarose Gel Immunofixation Electrophoresis as a Reflex Test to Capillary Electrophoresis Immunotyping

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**Introduction:** Protein electrophoresis is a laboratory method used to separate protein molecules based on their size and electrical charge. Agarose gel or capillary zone electrophoresis are main techniques used to identify monoclonal gammopathies and detect serum protein disorders. Capillary electrophoresis immunotyping (IT) and gel Immunofixation electrophoresis (IFE) are used to confirm the presence of M-protein and to determine its subtype. Although IFE is a gold standard technique, capillary electrophoresis IT test is easier to perform and less time-consuming. The aim of this study was to detect the number of IT results which was later referred to be analyzed in gel IFE. Moreover, to analyze the importance of this referral to the final report of the result.

**Method:** In this retrospective study, the results of patients who applied to our hospital between March 2023 and February 2024 for IT were evaluated. According to the IT assessment by at least 4 clinical biochemistry specialist/residency student, results which required further evaluation by gel IFE to be reported, were classified into two groups. Group I for patients' results included suspicion of monoclonal gammopathy. According to their suspicion, sub-groups were categorized as; (Ia) samples with hypogammaglobulinemia, (Ib) samples that were thought that they are uncertain/difficult to evaluate by only one specialist and (Ic) by two or more specialists. Group II for samples with gammopathy that couldn't be subtyped by IT technique. Sub-groups were categorized as; (IIa) samples with the suspicion of biclonality, (IIb) samples need to be analyzed to detect whether IG D or IG E monoclonal gammopathy is present or not and (IIc) samples which require processing with mercaptoethanol for certainty of biclonality.

**Results:** In total 1056 IT results were analyzed, 46.7% of the results required gel IFE. According to the IFE results: Monoclonal gammopathy was detected in 49.6% of group I, the results were distributed as: 54.1% in (Ia), 40.4% in (Ib) and 53.9% in (Ic), to subgroups. Biclonal gammopathy was detected in 59.6% of group (IIa), IG D monoclonal gammopathy was detected in 20% of group (IIb) and dimeric gammopathy was detected in 37.5% of group (IIc).

**Conclusion:** It's important to note that, evaluating IT combination with gel IFE is fundamental to detect all challenging gammopathies. In addition, it's beneficial to work as a team with multiple experienced specialists to distinguish minimal gammopathies within IT evaluation.

**Keywords:** Blood protein electrophoresis, capillary electrophoresis, immunoelectrophoresis, monoclonal gammopathies.

## OA-32

### Effect of lncRNA- HOXA-AS2 Gene Expression on Aortic Tissues of the Patients with Ascending Aortic Aneurysm

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**Aim:** Ascending aorta aneurysm (AsAA), defined as dilatation in the ascending part of aorta, presents a special challenge to cardiac surgeons because they remain asymptomatic until they present with either dissection or rupture. Studies have begun to reveal the relationships between the expression of long non-coding RNA molecules (lncRNA) and various pathological conditions such as neurodegenerative diseases, cardiovascular diseases and cancer. HOXA Cluster Antisense RNA 2 (HOXA-AS2) is a 1048-bp long noncoding RNA located on human chromosome 7p15.2 between the HOXA3 and HOXA4 genes in the HOXA cluster. There are several studies that investigate lncRNA expression in aortic tissues with aneurysm. However, there is no knowledge about the relationship between lncRNA-HOXA-AS2 expression levels and AsAA. In the present study, our aim was to compare the aortic tissue lncRNA-HOXA- AS2 expression levels in patients with AsAA and controls.

**Materials and Methods:** Our study included patients planned for aorta coronary bypass surgery as control subjects (n=25, mean age: 63±10 years) and patients diagnosed with AsAA as experimental group (n=25, mean age: 58±10 years) who were admitted to third step education and research hospital between 2023 and 2024. Total RNA was purified from samples using miRNeasy Tissue/Cells Advanced kit following manufacturer's instructions. cDNA was synthesized using miRCURY RT<sup>2</sup> First Strand Kit. lncRNA HOXA-AS2 expression was quantified on a Qiagen Rotor-Gene Q quantitative real time polymerase chain reaction (qRT-PCR) device. Primers used in the study were obtained from Qiagen Technologies. Expression levels were calculated using 2<sup>-ΔΔCT</sup> method and the relative expression of each gene was normalized to that of GAPDH. For the statistical analyses, Pearson's Chi-Squared ( $\chi^2$ ), Mann-Whitney U and logistic regression model tests were used where appropriate.

**Results:** No significant difference was found between the patient and control groups in terms of age, gender distribution, blood pressure, renal function and ejection fraction. lncRNA-HOXA-AS2 expression level in aortic tissues with aneurysm was not significantly different compared to control tissues (p=0.203). Moreover, there was no correlation between the lncRNA-HOXA-AS2 expression level and ascending aorta diameters.

**Conclusion:** Our findings suggest that lncRNA- HOXA-AS2 expression levels may not be related to ascending aorta aneurysm diagnosis.

**Keywords:** Ascending Aortic Aneurysm, HOXA-AS2, qPCR

## OA-33

### Investigation of Long Non-Coding RNA-ATB and MiR-200c expressions in Bladder Cancer Patients

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**Introduction and Aim:** The incidence of bladder cancer (BC) is widespread worldwide, but the detailed mechanisms of its occurrence and development are not yet fully understood. Genetic alterations are strongly associated with BC development. Recently, non-coding RNAs (ncRNAs) have been shown to play an important role in many different diseases, making them a promising area in cancer research. The two main classes of ncRNAs are microRNAs (miRNAs) and the more recently identified long ncRNAs (lncRNAs). Both classes of ncRNAs have been shown to be involved in cancer formation and development. The role of ncRNAs in the carcinogenesis and progression of BC and the underlying molecular mechanisms remain largely unclear. Demonstration of differential expression of ncRNAs between tumor and adjacent normal tissues of patients with BC may result in the discovery of new diagnostic and prognostic markers. Therefore, the aim of this study was to determine the expression of TGF- $\beta$ -activated lncRNA (lncRNA-ATB) and microRNA-200c (miR-200c) in tumorous tissues of BC patients and to investigate the relationship between these ncRNAs and histopathological and clinical features of patients with BC.

**Methods:** Between May 2022 and April 2023, 50 patients diagnosed with BC who were admitted to the Department of Urology, Division of Urologic Oncology, Istanbul Faculty of Medicine, Istanbul University were included. Tumorous tissues and adjacent normal tissues were obtained during transurethral bladder tumor resection (TUR-BT) procedure. lncRNA-ATB and miR-200c expressions were determined by quantitative real-time polymerase chain reaction (qRT-PCR).

**Findings:** In our study, we found that lncRNA-ATB expression in tumor tissues of patients with BC was statistically significantly increased compared to normal tissue adjacent to the tumor. There was also a statistically significant increase in lncRNA-ATB expression in high-grade tumors compared to low-grade tumors. An increase was found in miR-200c expression as in lncRNA-ATB. In contrast to lncRNA-ATB, the expression level in those with high-grade tumors was statistically lower than in those with low-grade tumors. When the changes of both ncRNA expressions at different stages of the tumor were examined, no statistically significant difference was found. ROC analysis showed that lncRNA-ATB had 72% sensitivity and 68% specificity and miR-200c had 68% sensitivity and 64% specificity in distinguishing tumor tissue from adjacent normal tissue. Despite these findings, no correlation was observed between lncRNA-ATB and miR-200c expression in tumor tissues.

**Conclusion:** Our study is the first study in which lncRNA-ATB and miR-200c expressions were evaluated together. It suggests that lncRNA-ATB and miR-200c may be an early biomarker for non-muscle invasive BC patients. However, more comprehensive studies including patients with muscle invasive, near and distant metastases are needed.

**Keywords:** bladder cancer, miRNA, lncRNA, ncRNA, miR-200c, lncRNA-ATB, biomarker

## OA-34

### Relationship of MiR-29b-3p Expression with Clinical and Histopathological Parameters in Non-Muscle Invasive Bladder Cancer

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**Aim:** Bladder cancer is the 10th most common cancer type worldwide. Cystoscopy represents the gold standard for bladder cancer diagnosis, but this procedure is invasive and painful, hence the need to identify new biomarkers through noninvasive procedures. Currently, there is no specific biomarker available for the diagnosis and monitoring of bladder cancer. microRNAs (miRNAs) are considered to be promising diagnostic molecules, because they are very stable in biological fluids (including urine) and easily detectable. The miR-29 family exerts tumor suppressor effects by inhibiting the proliferation and migration of cancer cells via downregulation of oncogenes and/or upregulation of tumor suppressor genes. We aimed to investigate the relationship between tissue levels of miR-29b-3p with clinical and pathologic variables of non-muscle invasive bladder cancer (NMIBC).

**Material and Methods:** Forty patients diagnosed with non-invasive bladder cancer between May 2022 and October 2023 in the urologic oncology clinic of our institution were included in the study. Written consent was obtained from the patients prior to the transurethral resection of the bladder tumor (TUR-BT) procedure. Tumor and adjacent non-tumor tissue samples obtained during resection were evaluated. Total RNA was purified from samples using miRNeasy Tissue/Cells Advanced kit following manufacturer's instruction. cDNA was synthesized using miRCURY™ LNA RT Kit. miR-29b-3p expression was quantified on a Qiagen Rotor-Gene Q quantitative real time polymerase chain reaction (qRT-PCR) device. Primers used in the study were obtained from Qiagen Technologies. Expression levels were calculated using 2<sup>-ΔΔCT</sup> method and the relative expression of each gene was normalized to that of U6 snRNA.

The median age of the patient group at diagnosis, the body mass index, smoking behavior and presenting complaint were evaluated. The study group consisted of patients with tumor stages of Ta (n=22, 55%) and T1 (n=18, 45%). 15 of the patients (37.5%) were low grade, while 25 (62.5%) were high grade. The presence of CIS accompanying the tumor was detected in 12 (30%) of the patients, and accompanying variant pathology was detected in 3 (7.5%) patients. For the statistical analyses, Pearson's Chi-Squared ( $\chi^2$ ), Mann-Whitney U and logistic regression model tests were used where appropriate.

**Results:** miR-29b-3p expression level in tumor tissues of BC patients was not significantly different compared to control tissues adjacent to the tumor. Furthermore, miR-29b-3p expression level was not statistically different between tissues with different stages (p=0.515), grades (p=0.300) or histopathologies (p=0.759). When the relationship between the patients' clinical parameters and miR-29b-3p expressions was examined, although there was a decrease in miR-29b-3p expressions in the smoking group compared to the non-smoking group, this difference was not found to be significant (p = 0.711). No significant correlation was found between age, BMI, macrohematuria, microhematuria and lower urinary tract symptoms.

**Conclusion:** Our findings suggest that miR-29b-3p expression levels may not be related to non-muscle invasive bladder cancer diagnosis and prognosis.

**Keywords:** bladder cancer, miRNA, miR-29b-3p, tumor biomarkers

## OA-35

### Evaluation of Serum Epiregulin Level and Relationship of Epiregulin Level with Laboratory Parameters in Patients with Newly Diagnosed Type 2 Diabetes Mellitus

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**Aim:** Type 2 diabetes mellitus (T2DM) is characterised by increased insulin resistance and functional failure of pancreatic  $\beta$ -cells. Epiregulin is a polypeptide belonging to the epidermal growth factor family and is involved in many biological processes such as cell growth, differentiation and tissue repair. In a study, it was observed that epiregulin stimulates the growth of pancreatic  $\beta$ -cells and insulin secretion. In this study, we aimed to determine whether serum epiregulin levels are different in patients with newly diagnosed T2DM compared to healthy controls and to investigate the relationship between serum epiregulin levels and metabolic parameters.

**Method:** Our study is a case-control study. Newly diagnosed T2DM patients aged 18-65 years and healthy control group were randomly included in the study according to the eligibility criteria and voluntary participation in the Internal Medicine Outpatient Clinic of our hospital. Biochemical and hormonal parameters and HbA1c levels of the participants were analysed. Serum epiregulin levels were analysed by sandwich ELISA method on a Multiskan GO model ELISA reader (Thermo Fisher Scientific, Finland). Epiregulin concentrations were expressed as pg/mL. Statistical Package for Social Sciences (SPSS) 26.0 (IBM, USA) was used for statistical analyses and calculations. The distribution of variables was evaluated by Kolmogorov-Smirnov test. Depending on whether the distributions were normal or not, Student's T test or Mann-Whitney U test was used for comparison. The relationship between variables was determined by Pearson or Spearman correlation analysis. The significance level was set as  $p < 0.05$ .

**Results:** of the samples, 38 were female (19 patients, 19 controls) and 39 were male (20 patients, 19 controls). The median values of serum epiregulin levels were significantly higher in patients with newly diagnosed T2DM than in the control group (416 pg/mL, 182 pg/mL, respectively) ( $p=0.005$ ). Although the mean serum epiregulin levels were higher in women ( $298 \pm 235$  pg/mL) than in men ( $341 \pm 224$  pg/mL), no significant difference was observed ( $p=0.408$ ). When the relationship between epiregulin and laboratory parameters was analysed, a significant positive correlation was found between fasting and postprandial blood glucose.

**Conclusion:** The elevated levels of epiregulin and its correlation with blood glucose suggest that epiregulin has an effect on the pathogenesis of T2DM. Randomised controlled studies in larger populations are needed to clearly demonstrate the role of epiregulin in the development of T2DM.

**Keywords:** Diabetes mellitus, Epiregulin, ELISA

## OA-36

### The Relationship Between Ghrelin and Iron Metabolism in the Beta Thalassemia Patient Group

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**Objective:** Studies on HIF2 $\alpha$ , hepcidin and ferroportin molecules in beta thalassemia major ( $\beta$ -TM) patients show that there are disorders in these pathways. In recent years, the relationship between the ghrelin hormone and iron metabolism has been investigated independently of patients with iron deficiency anemia. We aimed to contribute to the etiopathogenesis of these successful  $\beta$ -TM patients by examining the changes in ghrelin hormone levels.

**Methods:** 52  $\beta$ -TM and 23 controls were included in our study. Blood counts, routine biochemical parameters, HIF2 $\alpha$ , hepcidin and ghrelin levels were studied in blood samples taken from volunteers. Serum HIF2 $\alpha$ , hepcidin and ghrelin levels were measured by ELISA method. Results: Erythrocyte indices, serum total bilirubin, direct bilirubin, iron, unsaturated iron binding capacity, total iron binding capacity and ferritin levels showed significant differences between the two groups ( $p < 0.05$ ). There was no significant difference in serum HIF2 $\alpha$  and hepcidin levels between the two groups. When the patient group was compared with healthy controls, serum ghrelin levels were found to be significantly higher in the patient group ( $p < 0.05$ ). There was a significant positive correlation between serum ghrelin and ferritin levels in the patient group ( $r = 0.401$ ) ( $p < 0.05$ ).

**Conclusion:** It suggests that high ghrelin levels may have an important role in regulating impaired iron metabolism in patients with  $\beta$ -TM. The positive correlation between serum ghrelin levels and ferritin suggests that serum iron may have an important role in ghrelin synthesis and that increased ghrelin levels may be a factor that increases ferritin synthesis.

**Keywords:** Beta thalassemia major, iron metabolism, ghrelin

## OA-37

### Comparison of HbA1c Measurement with Immunoturbidimetric Method and High Performance Liquid Chromatography Method

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**Objective:** Diabetes mellitus is becoming increasingly common worldwide. HbA1c measurement has been defined as a diagnostic criterion for diabetes mellitus. However, since various methods and devices have been developed for HbA1c measurement, standardization has become necessary. The aim of this study was to compare HbA1c levels measured by immunoturbidimetric method and high performance liquid chromatography method, which is accepted as the reference method, in 101 samples. Comparison studies between methods provide evidence-based information about the compatibility of test results.

**Material Method:** The study included 101 Ethylenediamine tetraacetic acid whole blood samples from routine HbA1c test orders. HbA1c levels in the samples ranged from 4.6% to 14.3%. Each sample was analyzed by two different methods, with the analysis time not exceeding one hour. High performance liquid chromatography was performed on HLC-723 TOSOH-G11 (Tosoh Bioscience, Inc., Tokyo, Japan) and immunoturbidimetric analysis was performed on Beckman Coulter AU5800 (Beckman Coulter Inc, Mishima, Japan) biochemistry autoanalyzer with Archem Diagnostic HbA1c II reagent (Archem Health Industry Trade Inc., Istanbul, Turkey). The results were statistically evaluated using SPSS 25.0 program.  $p < 0,05$  was accepted for statistical significance. The compatibility between the two methods was evaluated with correlation analysis. The relationship between the dependent variable and the independent variable was determined by linear regression analysis.

**Results:** While the average value for the high performance liquid chromatography method was %7.13, it was %7.29 for the immunoturbidimetric method. According to Pearson correlation test, there was a very high positive correlation between the methods ( $p < 0.001$ ,  $r = 0.99$ ). According to simple linear regression analysis,  $y = 0.45 + 0.96x$ ,  $r^2 = 0.98$ . In regression equation analysis, the regression coefficient was 0.96 [%95 confidence interval 0.931-0.985] and the intercept was 0.45 [%95 confidence interval ( 0.256-0.654)]. The mean difference between both methods was 0.15 [%95 confidence interval ( 0.093-0.215)] ( $p < 0.001$ ).

**Conclusion:** Although there is a very high agreement between the two methods in the HbA1c results according to the correlation analysis, a proportional systematic error is noticeable in the HbA1c results measured by the immunoturbidimetric method compared to HPLC. The same method should be used when monitoring HbA1c levels in patients.

**Keywords:** HPLC, immunoturbidimetric, HbA1c, method comparison

## OA-38

### Laboratory Perspective of Cold Agglutinin Syndrome: A Case Study

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**Aim and background:** This is a case report of a Cold Agglutinin Syndrome presenting to the laboratory with interferences of cold agglutinins on CBC analysis. Besides resolving the cold agglutination, we emphasized on the potential support of laboratory on diagnosis of patient.

**Case presentation:** During our routine laboratory workload, we encountered a blood sample from a 79-year-old patient diagnosed as colon cancer one year ago and was in remission at the date. CBC analysis of the sample showed many flags by the automated hematology analyzer (Sysmex Corporation, Kobe, Japan). A mismatch in the Rule of Three with spuriously high MCHC and MCV was recorded. Lipemia and hemolysis were evaluated for possible interferences. Samples exhibited neither hemolysis nor lipemia. Peripheral blood smear examination of the sample revealed clusters of RBC agglutination. Patient was called back to the laboratory and three fresh blood samples were collected into 3.0 mL K2-EDTA tubes in our laboratory. Pre-warming of tubes and sample treatment with 2-mercaptoethanol (2-ME) procedures were applied. Both prewarmed and 2-ME added tubes did not show agglutination as checked by peripheral blood smear. Direct antiglobulin test was positive for C3d and negative for Ig G. Hemolysis related biochemistry parameters LDH, AST, potassium, total and direct bilirubin, haptoglobin were in normal range; there was no evidence of intravascular hemolysis. Quantitative determination of immunoglobulins G, A, and M were all in normal range. Any kind of paraproteinemia was not detected by serum protein electrophoresis and immune fixation electrophoresis. Free kappa and Lambda light chains analysis were also in normal range. The patient was identified as cold agglutinin syndrome secondary to a malignancy.

**Conclusion:** This case study provides a step-by-step approach for laboratory management and diagnosis of cold agglutination.

**Keywords:** Cold agglutinin syndrome; complete blood count; 2-mercaptoethanol



## OA-39

### Can Earlobe Capillary Blood Be An Alternative to Arterial Blood in Blood Gas Analysis?

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**Purpose:** The 'gold standard' sample for blood gas analysis is arterial blood. Arterialized blood from dilated capillaries of the earlobe is suggested to be a good predictor of arterial blood content. Earlobe sampling has several advantages compared to arterial puncture, such as being less invasive, less risk of complications, and better patient tolerance. However, since conflicting results were obtained in studies regarding the accuracy of this technique, it has not been used routinely in current clinical practice. Therefore, our study aimed to compare arterial and capillary blood gas parameters.

**Method:** Forty-three adult patients hospitalized in the Chest Diseases ward of our hospital were included in the study. Capillary blood samples from the earlobe and arterial blood samples from the radial artery were taken randomly from the patients at rest and blood gas measurements were performed on the same analyzer (ABL 800, Radiometer) without waiting. Capillary and arterial pH, pCO<sub>2</sub>, pO<sub>2</sub>, and lactate results were compared by Wilcoxon test because they were not normally distributed; hemoglobin, hematocrit, potassium, sodium, and calcium results were compared by Paired sample t-test. Bias / %bias values of the tests were calculated and evaluated according to target values (CLIA 2024, Rilibak, and RCPA).

**Results:** Arterial pH and pO<sub>2</sub> values were significantly higher than capillary values; pCO<sub>2</sub>, lactate, hemoglobin, hematocrit and potassium values were found to be significantly low. Although there was a statistically significant difference between capillary and arterial blood gas pH, pCO<sub>2</sub>, pO<sub>2</sub>, lactate, hemoglobin, hematocrit, and potassium values, the bias / %bias values of pH and PCO<sub>2</sub> parameters were found to be at an acceptable level compared to the target values. Capillary and arterial sodium and calcium results were found to be statistically and clinically similar.

**Conclusion:** It was observed that capillary blood gas can be a reliable alternative to arterial blood gas for pH, PCO<sub>2</sub>, sodium, and calcium parameters. However, for PO<sub>2</sub>, lactate, hemoglobin, hematocrit, and potassium parameters, capillary blood gas analysis from the earlobe is not suitable for clinical evaluation.

**Keywords:** blood gas, arterial, capillary, earlobe

## OA-40

### Evaluation of the Effect of Various Storage Conditions on Ethanol Level

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**Aim:** This study investigates the influence of storage time and temperature on blood ethanol levels in stored samples, crucial for maintaining result reliability due to the impact of various preanalytical variables.

**Materials and Methods:** Blood samples were collected from 400 volunteers into sodium fluoride-containing tubes, centrifuged to form a plasma pool. This pool, confirmed to have an ethanol concentration <10 mg/dl, was supplemented with ethanol to final concentrations of 50, 100, 200, and 400 mg/dL, resulting in five distinct concentration pools. These were further divided into eight subgroups (seven portions each), with some stored at room temperature for up to 24 hours and others stored for 1, 3, and 6 months at -20 °C and -80 °C to facilitate later ethanol level measurements.

**Results:** No significant statistical difference ( $P>0.05$ ) was observed in ethanol levels between samples stored at room temperature for 0 and 24 hours. However, significant differences ( $P<0.05$ ) were noted between the 0-hour baseline and those stored for extended periods (1, 3, and 6 months) at both -20°C and -80°C. The analysis also considered the maximum allowable difference (MIF) for ethyl alcohol, set at "±10%" by the Royal College of Pathologist Australasia. Comparing this with the percentage change (%PD) values from all groups (-0.64 to -5.47) indicated no analytically significant deviations.

**Conclusion:** The study confirms that witness samples in forensic cases can be stably stored at -20°C without any loss in stability for the legal storage duration. Furthermore, it is recommended that laboratories develop an "instability equation" for ethanol samples and calculate stability limits based on their own specific performance characteristics, adhering to clinically determined MIF values.

**Keywords:** Ethyl alcohol, stability, storage temperature, storage time, instability equation, maximum allowable difference, stability limit

## OA-41

### Lithium Heparin Blood Collection Tubes Have Insignificant Lot-To-Lot Variations

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**Aim:** Blood collection tubes (BCTs) are critical in the pre-analytical phase. Hospitals may use different lots of the BCTs, therefore it's important to compare them. In this study, we compared different lots of the Greiner Vacuette Lithium Heparin tubes.

**Methods:** Blood was collected from volunteers using two different lots of the Greiner Vacuette Lithium Heparin tubes. We then analyzed nine clinical chemistry and four immunoassay analytes simultaneously on the Roche Cobas analyzer. We compared the BCTs according to the recommendations of the CLSI GP34-A guideline. We used Passing-Bablok regression analysis and Bland-Altman plots to evaluate them using MedCalc® version 22.013 (MedCalc Software Ltd, Ostend, Belgium). We set desirable bias goals obtained via the EFLM Biological Variation Database, and statistical significance was set at  $p < 0.05$  with a 95% confidence interval (CI).

**Results:** The results showed no statistically significant difference in Passing Bablok analysis of the 13 analytes. All analytes' mean percentage differences (MPD) did not exceed the bias goals. However, some analytes had 95% confidence intervals (CIs) of the MPD that exceeded the desirable bias goals. For example, Sodium had an MPD of 0.09% and 95% CIs of -0.200% to 0.388%, with a desirable bias of 0.2%. Potassium had an MPD of -0.48% and 95% CIs of -1.721% to 0.756%, with a desirable bias of 1.6%. Glucose had an MPD of -1.63% and 95% CIs of -3.703% to 0.434%, with a desirable bias of 2.3%. Ferritin had an MPD of -0.37% and 95% CIs of -14.748% to 14.015%, with a desirable bias of 3.2%.

**Conclusions:** According to the CLSI GP34A guideline, different lots of the Greiner Vacuette Lithium Heparin BCTs were found to be interchangeable, with negligible lot-to-lot variations. However, the Biological Variation goals could be considered strict. In-vitro diagnostic manufacturers and laboratories should carefully monitor the BCTs' lot-to-lot variation.

**Keywords:** Blood specimen collection, blood collection tube, lot-to-lot variation, pre-analytical phase

## OA-42

### Comparison of 3 Different Blood Collection Tubes for Valproic Acid and Carbamazepine Analyte Measurement

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**Objective:** Verification of sampling tubes, one of the sources of preanalytical errors, is crucial for accurate test results and helps to reduce preanalytical errors. The aim of this study was to compare the results of valproic acid and carbamazepine analytes in three different sampling tubes.

**Material and method:** Valproic acid-treated 25 volunteers and carbamazepine-treated 21 volunteers agreed to participate in the study. Blood samples were collected in 4 separate tubes: a tube without additives and gel (Z tube) and three different tubes with clot activator (vacusera with gel, vacusera without gel and vacutainer with gel). In addition to the comparison with the Z tube, stability analyses of the analytes in all three tubes were performed after the samples were stored at +4°C for 7 days. The bias % was calculated and the results were evaluated according to a bias level of 10%. Our study was conducted according to the CLSI GP 34A guideline. As stated in the guideline, our sample number was planned to meet the minimum requirement of 20 and the order of control and comparison tubes was randomised during blood collection. Valproic acid and carbamazepine analytes were measured by chemiluminescent microparticle immunoassay using ARCHITECT i1000SR (Abbott Laboratories, Chicago, IL, USA).

**Results:** The bias% levels of valproic acid and carbamazepine analytes were 0.61-8.03 in the vacusera tube with gel, 2.60-4.08 in the vacusera tube without gel and 2.10-2.55 in the vacutainer tube with gel, respectively. After seven days, three tubes were stable.

**Conclusion:** Various blood collection tubes with different contents may cause clinically significant differences in test results. In our study, we found that the three tubes met the performance targets for valproic acid and carbamazepine analytes. It was concluded that all three tubes can be safely used for these two analytes.

**Keywords:** Valproic acid, Carbamazepine, Verification, Preanalytical error

## OA-43

### Is the Serum Uric Acid /HDL Cholesterol Ratio Associated with Diabetic Nephropathy?

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**Objective:** Diabetic nephropathy(DN) is a serious microvascular complication of T2DM. Markers are important for diagnosis of this complication. The uric acid to HDL ratio(UHR) has been reported to be associated with inflammatory and metabolic diseases. In our study, we aimed to demonstrate the diagnostic role of UHR, which is a marker of chronic and low-grade inflammation, for DN.

**Methods:** We retrospectively analyzed the data of 521 patients who applied to the Internal Medicine outpatient clinic of Başakşehir Çam and Sakura City Hospital. The study population was divided into two groups according to the presence of DN. Laboratory data, including UHR levels, of diabetic patients with DN were compared with patients without DN.

**Results:** Significant difference was observed in UHR levels between T2DM patients with DN( $0.12 \pm 0.54$ ) and those without DN( $0.11 \pm 0.46$ ) ( $p = 0.003$ ). In DN group, creatinine( $1.0 \pm 0.51$ ,  $0.82 \pm 0.21$ ,  $p < 0.001$ ), uric acid( $5.29 \pm 1.62$ ,  $4.77 \pm 1.51$ ,  $p < 0.001$ ), and HbA1c( $7.91 \pm 2.44$ ,  $7.22 \pm 1.91$ ,  $p = 0.021$ ) levels significantly elevated compared to non-DN group. Positive correlation was observed between UHR and creatinine( $r = 0.516$ ,  $p < 0.001$ ), uric acid( $r = 0.805$ ,  $p < 0.001$ ), and microalbumin levels( $r = 0.776$ ,  $p < 0.001$ ).

**Conclusion:** The results of our study showed that UHR increased significantly in diabetic patients with DN and that this increase was correlated with UACR levels. This study showed that increased UHR levels in diabetic patients are an important marker for the presence and severity of microalbuminuria and thus may be important in the early detection of renal dysfunction in T2DM. We think that even in the early stages of microalbuminuria, it can be used as an important diagnostic marker in the follow-up of diabetic patients. Therefore, we recommend that UHR be routinely evaluated in addition to biochemistry parameters in DN patients.

**Keywords:** Type 2 diabetes mellitus; diabetic nephropathy; uric acid-to-HDL cholesterol ratio; complication; inflammation

## OA-44

### Reference Intervals for Urea, Creatinine and Uric Acid in Pregnancy

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**Aim:** In this study, we aim to examine the differences in serum urea, creatinine, and uric acid levels between pregnant and non-pregnant women, and to explore the need for trimester-specific reference intervals in pregnancy.

**Methods:** Serum urea, creatinine, and uric acid measurements of 1,100 pregnant women who visited the outpatient clinics of Lutfi Kırdar City Hospital for pregnancy screening tests between January 1, 2022, and January 1, 2024, were obtained from laboratory data. All measurements were conducted using the Cobas c 702 (Roche Diagnostics, Penzberg, Germany). The serum levels of urea, creatinine, and uric acid were compared among four groups: non-pregnant women and the three trimesters of pregnancy. Statistical significance was tested with the standard normal deviation test (Z-test), and clinical significance was assessed using the corresponding reference change values (RCV) of the analytes. Reference intervals were determined using non-parametric methods in accordance with the guidelines provided by the Clinical and Laboratory Standards Institute (CLSI) guideline C28-A3.

**Results:** Compared to non-pregnants, pregnant women showed lower levels of serum urea, creatinine, and uric acid with statistical and clinical significance. Urea and creatinine levels did not show any significance among trimesters. Uric acid levels in the first and second trimesters also did not show a significant difference between each other while third trimester values were found different. Therefore reference intervals for uric acid were partitioned as the first two trimester and the third trimester. As a result, the reference intervals for pregnant women were determined to be 9-23 mg/dL for urea, 0.35-0.63 mg/dL for creatinine; and for uric acid, 1.8-4.1 mg/dL for first two trimesters and 2.1-5.1 mg/dL for the third trimester.

**Conclusion:** Pregnant women had significantly lower values for serum urea, creatinine, and uric acid compared to non-pregnant women. For accurate interpretation of renal functions, specific reference intervals should be used in pregnancy.

**Keywords:** urea, creatinine, uric acid, reference intervals, pregnancy

## OA-45

### Determination of Zinc Reference Interval by Indirect Method in Adults

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**Objective:** In our study, we aimed to determine the reference intervals of zinc parameters in the adult population according to sex by using the indirect method of truncated maximum likelihood (TML).

**Method:** In our study, the results of 51,063 patients with zinc test requests between 20.05.2021-24.05.2024 were retrospectively analysed through the laboratory information system. Patient results were anonymised and converted into a data set in Excel software. Tukey test was used for outlier detection and Kolmogrov-Smirnov test was used for distribution normality. Patient data under the age of 18 years and results that could be due to preanalytical error were discarded from the raw data. After these procedures, the remaining 38,729 data were separated according to gender. The reference intervals of both sex groups were determined using the TML method and confidence intervals were calculated. For the TML method, the RLE v49 software shared by the German Society for Clinical Chemistry and Laboratory Medicine on its website was used. The reference intervals we determined were compared with the current reference intervals used in our laboratory.

**Results:** 51,063 patients were screened between the specified dates. 414 patients were discarded due to preanalytical errors. of the remaining 50,649 patients, 11,774 data were excluded due to being under 18 years of age. Outliers were then extracted from the remaining patient data. As a result, a total of 38,729 patients, 30,033 females and 8696 males, were included in the study. According to the Kolmogrov- Smirnov test, the data did not fit the normal distribution. The reference ranges stated in the kit package insert were 72.6 -127 µg/dL for men and 70-114 µg/dL for women. The reference range values obtained by indirect method in our study were 65.3-136.6 µg/dL for men and 63.2-131.7 µg/dL for women.

**Conclusion:** When the reference ranges obtained in our study are compared with the reference ranges stated in the kit package insert, it is seen that the reference ranges we found have a wider range. In addition, in our study; similar to the reference range stated in the kit package insert, the zinc reference range was lower in females compared to males. We conclude that it is important for each laboratory to determine its own reference ranges for a more accurate evaluation of laboratory results.

**Keywords:** Indirect method, zinc, reference range, truncated maximum likelihood

## OA-46

### The effect of *Myrtus communis* extract on lncRNA-PVT1 expression in A549 lung cancer and BEAS-2B normal bronchial cells

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**Objective:** Approximately 12.3% of all cancer cases worldwide are lung cancer, making it the most prevalent type of cancer. Finding sensitive biomarkers that can be utilized as targets for therapy and for diagnosis has long been a top priority in cancer research. Long non-coding RNAs (lncRNAs) have been shown in numerous studies to play a role in the emergence of cancers. One of these lncRNAs, plasmacytoma variant translocation 1 (PVT1), looks quite interesting and is implicated in several cancers. PVT1 is a lncRNA that is encoded by the human PVT1 gene. According to recent research, PVT1 expression is higher in several cancer types as compared to the equivalent neighboring non-tumor tissues. While *Myrtus communis* (MC) has been studied extensively for its medicinal benefits. However, no research has looked at how its essential oil and other MC components affect PVT1 expression levels in lung cancer cells.

**Material & Method:** *Myrtus communis* leaf ethanol extracts (MCE) were added to cells at varying concentrations (0.01–1000 µg/ml) and left for a full day at 37 °C. The Sulforhodamine B (SRB) assay was used to quantify the cytotoxic effect of MCE on A549 and BEAS-2B cells. Reverse transcriptase polymerase chain reaction (qRT-PCR) was used to determine the impact of MCE on the level of PVT1 expression in A549 and BEAS-2B cells. A control group of healthy cells was BEAS-2B cells. 2-ΔΔCT was used to evaluate the PVT1 expression levels.

**Results:** According to the SRB results, MCE inhibited the proliferation of A549 and BEAS-2B cells with an IC<sub>50</sub> value of 100 µg/ml and 1000 µg/ml, respectively. For PVT1 expression analysis, cells were treated with MCE at IC<sub>50</sub> and IC<sub>25</sub> doses. When MCE was given to A549 cells, PVT1 expression levels dropped in comparison to the control group. At the 100 µg/ml MCE dose, this decrease was observed to be 1.67 times; however, it was not statistically significant. Applying 500 µg/ml MCE to BEAS-2B cells resulted in a 2-fold decrease in PVT1 expression level when compared to the control, indicating a significant difference. PVT1 expression level was found to rise 1.85 times compared to the control when the MCE dose of 100 µg/ml was applied but this increase was not statistically significant. PVT1 expression levels were found to be lower in the group that received higher doses of MCE for both cell lines than in the group that received lower doses.

**Conclusion:** This study is an important preliminary study in terms of guiding future studies and is the first study to examine the effects of MCE on PVT1 expression levels in lung cancer cells. The results show that MC may have a significant anti-cancer effect on A549 cancer cells. Future studies to reveal the compounds responsible for the effect through future bioactivity-directed fractionation (BAYF) studies, to standardize the extract based on these compounds, and to evaluate the effect of the extract with in vivo studies may aid in the evaluation of the effect of this compound on lung cancer.

**Keywords:** Lung cancer, lncRNA-PVT1, *Myrtus communis*



## OA-47

### A Rare Case of Metabolic Disease: Holocarboxylase Synthetase Deficiency

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**Objectives:** Multiple carboxylase deficiency is an autosomal recessive disorder of biotin metabolism. The underlying mechanism is biotinidase or holocarboxylase synthetase (HCLS) deficiency. Holocarboxylase synthetase is an enzyme that catalyzes the biotinylation of carboxylases, including propionyl-CoA carboxylase, 3-methylcrotonyl-CoA carboxylase, pyruvate carboxylase, and acetyl-CoA carboxylase. Holocarboxylase synthetase deficiency occurs in approximately 1 in 200,000 births worldwide. Symptoms of biotinidase and holocarboxylase synthetase deficiency are similar, and clinical distinction is often challenging. Both disorders typically respond to biotin therapy. In our study, we aimed to evaluate the laboratory data of a case of holocarboxylase synthetase deficiency.

**Materials and Methods:** The 2-year-old male patient presented to our pediatric emergency clinic, experiencing shortness of breath and intensified respiratory effort beginning in the morning and worsening by evening. The patient was transferred to the pediatric intensive care unit due to the presence of metabolic acidosis findings in the blood gas analysis and persistent respiratory distress. Due to neurological deterioration and persistent metabolic acidosis despite treatment, the patient was evaluated for metabolic diseases. The quantitative amino acid analysis in blood and urine, urine organic acid analysis, carnitine/acylcarnitine profile analysis were requested.

**Results:** Laboratory findings included mildly elevated ammonia (93  $\mu\text{mol/L}$ , N:16-60), hyperglycemia (137 mg/dL, N:74-100), and ketones in urine (80 mg/dL, N:0-5). In the carnitine/acylcarnitine profile, elevated levels of C2 acetyl carnitine, C5 3OH isovaleryl carnitine, valine, leucine were detected. In the quantitative amino acid analysis in blood (LC-MS/MS method), increased levels of alanine, alpha-aminobutyric acid, valine, proline, leucine were detected. Quantitative analysis of amino acids in urine (LC-MS/MS method) showed elevated levels of phosphoethanolamine, valine, cystine, leucine, proline, and alloisoleucine. In the urine organic acid analysis; increased levels of lactic acid, 2-hydroxybutyric acid, 3-hydroxypropionic acid, pyruvic acid, 3-hydroxybutyric acid, 3-hydroxyisovaleric acid, acetoacetic acid, acetate, tiglylglycine, 3-methylcrotonyl glycine, methylcitric acid, 4-hydroxyphenyl lactic acid, and 4OH-phenyl pyruvic acid were detected. The patient's biotinidase enzyme activity level was 4.01 U/L. (N:3.5-13.8).

**Conclusion:** In holocarboxylase synthetase deficiency, carboxylase enzyme metabolites are abnormally excreted in urine. These metabolites suggest deficiencies: 3-methylcrotonylglycine and 3-hydroxyisovalerate indicate methylcrotonyl-CoA carboxylase deficiency; 3-hydroxypropionate, methylcitrate, tiglylglycine suggest propionyl-CoA carboxylase deficiency; and lactate suggests pyruvate carboxylase deficiency. This pattern is typical of multiple carboxylase deficiency due to holocarboxylase synthetase or biotinidase deficiency. Thus, a late-onset, biotin-responsive holocarboxylase synthetase deficiency was suspected in this patient based on typical LC/MS and tandem mass study findings, and normal biotinidase activity. After biotin administration, the patient showed clinical improvement and blood gas analysis was normal. Repeat carnitine/acylcarnitine and blood amino acid analyses showed all values within normal range. Genetic analysis for HLCS was requested and the patient was discharged in good general condition. If these metabolites reflecting multiple carboxylase deficiency are found in urine, one should consider the possibility of

holocarboxylase synthetase deficiency, which is relatively uncommon. Especially the elevation of tiglylglycine, which we rarely see in urine organic acid analysis, and the significantly elevated level of 3-hydroxyisovaleric acid are noteworthy. In this case, the presence of normal biotinidase enzyme activity in the serum supports the diagnosis of holocarboxylase synthetase deficiency.

**Keywords:** holocarboxylase synthetase, biotin

## OA-48

### Inappropriate Test Requests for Tumor Markers: A Single-Center Experience

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**Introduction:** Despite accounting for only 3-5% of total medical costs, laboratory tests play a crucial role in directing 70% of medical decisions. Generally, it is estimated that 20-30% of all test requests are inadequate, unnecessary, or improperly used. Guidelines at national and international levels have been established to determine when, under what circumstances, at what intervals, and according to which algorithms laboratory tests should be performed; requests made outside these guidelines are defined as inappropriate. Appropriate test requests involve requesting tests "from the right patient, using the right test, with the right method, at the right time, at the right cost, and producing the right result." The aim of this study is to determine the rate of inappropriate test requests for tumor markers (CA15.3, CA19.9, CA125, and free PSA) among patients presenting to the Central Laboratory of Balcalı Hospital in 2022 through a retrospective review of the system.

**Materials and Methods:** Statistical data were calculated annually based on the requests for tumor markers performed between January 1, 2022, and December 31, 2022. Data related to tumor markers were retrospectively reviewed via the Laboratory Information Management System (LIMS) and Hospital Information Management System (HIMS) to identify and evaluate inappropriate test requests based on objective information provided in the guidelines.

**Results:** CA19.9 was requested in 69% of patients who had CA15.3 requested, in 65.34% of patients who had CA125 requested, and in 77.35% of patients who had both CA15.3 and CA125 requested. The rate of requesting two tumor markers together was calculated as 70.56% in average. The rate of co-requesting three markers was determined as 59.44%. fPSA was requested in 88.48% of requests with PSA levels either <4 ng/mL or >10 ng/mL.

**Conclusion:** Elevated levels of tumor markers can be observed in various organ pathologies. Guidelines recommend requesting CA15.3 for breast, CA125 for ovarian, and CA19.9 for gastrointestinal pathologies. To distinguish between prostate cancer and benign prostatic hyperplasia (BPH) and prevent unnecessary prostate biopsies, it is recommended to measure fPSA when the total PSA level is between 4- 10 ng/mL. In our study, the high rate of co-requesting tumor markers with the same clinical diagnosis and inappropriate requests for fPSA impose unnecessary stress on patients and burden laboratory expenses. The aim of the next phase is to take necessary measures, interventions, and reduce unnecessary requests to decrease unnecessary costs on the laboratory budget.

**Keywords:** inappropriate testing, tumor markers, laboratory medicine

## OA-49

### Comparison and Reporting of Measurement Uncertainty Results Calculated on Three Different Abbott Architect C16000 Autoanalyzers

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**Purpose:** Biochemistry tests are widely used worldwide as indispensable tools by clinicians to determine clinical progression, and the test results are crucial for the decision-making process of clinicians. However, as with all measurement processes, biochemical measurements are not absolute, and each test result has a measurement uncertainty. Measurement uncertainty is a statistical parameter that indicates the distribution of probabilities that can be attributed to the reported measurement result. Measurement uncertainty is important in biochemistry laboratories to define test validity and demonstrate the clinical relevance of the test. Lower values are associated with more reliable results, whereas higher values decrease the reliability of the results. Therefore, laboratories should minimize test uncertainties as much as possible. Reporting the measurement uncertainty values to clinicians can facilitate more effective management of the decision-making processes. In this study, we aim to calculate the measurement uncertainty of analytes in our three different biochemistry autoanalyzers and compare our laboratory performance with the allowable total error (TEa) values set by both the Ministry of Health and the Clinical Laboratory Improvement Amendments (CLIA). Additionally, we aim to examine how measurement uncertainty should be reported to clinicians.

**Materials And Methods:** In our laboratory, low and high-level internal quality control samples for 19 analytes measured over 34 days between March 6, 2024, and April 30, 2024, were used across three different Architect C16000 (Abbott Diagnostics, Illinois, USA) autoanalyzers. Bias data were obtained from the Klinik Biyokimya Uzmanları Derneği External Quality Control Programme (KBUDEK) (Istanbul, Türkiye). As internal quality control samples, low-level control lot number 12911222 and high-level control lot number 12505223 of the Multichem S Plus (TECHNOPATH, Ballina Co. Tipperary, Ireland) brand were used. Measurement uncertainty values were calculated using internal quality and external quality control data according to the Nordtest guidelines and expanded with a 95% confidence interval.

**Results:** Albumin and sodium were found to exceed the %TEa values of CLIA for devices A and C, while they were found to be compatible with the %TEa values stated by the Ministry of Health. Similarly, blood urea nitrogen is higher than the %TEa values of CLIA for devices A, B, and C, yet it is compatible with the Ministry of Health data. The measurement uncertainty of the other analytes calculated includes alanine transaminase (ALT), alkaline phosphatase (ALP), aspartate transaminase (AST), glucose, high-density lipoprotein (HDL), calcium, chloride, creatine kinase, creatinine, magnesium, potassium, total bilirubin, total cholesterol, total protein, triglyceride, and uric acid. The calculated measurement values for these analytes were found to be compatible with both the %TEa limits stated by CLIA and the Ministry of Health.

**Conclusion:** Even when using the same brand of device, kit, and controls, different measurement uncertainty percentages may arise in devices due to other factors. In this case, the issue arises as to which results should be communicated to clinicians. Laboratories using multiple autoanalyzers for the same test should include the measurement uncertainty of the device on which the analyte is measured in the report.

**Keywords:** Biochemical marker, Quality management, Measurement uncertainty

## OA-51

### Determination of Measurement Uncertainty Using Clinical Decision Limits and Population Data

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**Objective:** Analytical Performance Specifications (APS) are standards for evaluating the reliability and accuracy of clinical lab tests, starting from a 1999 Stockholm classification and simplified in 2014 at the EFLM conference in Milan. Various models and methods are used to assess analytical performance, posing challenges due to their link to clinical decisions, biological variations, and laboratory performance. A data-driven tool using population data has been developed to facilitate this process. The aim of our study is to determine APS related to the measurement uncertainty in laboratory tests using test results specific to our population.

**Methods:** A1c, total cholesterol, vitamin D, and folate results obtained from outpatient patients aged between 18 and 65, with samples collected before noon, were used in the study. The HbA1c test was performed on the H9 Hemoglobin Analyzer, the total cholesterol, vitamin D, and folate tests were performed on the Roche Cobas 8000 platform. Decision limits of 5.7-6.4% for HbA1c, 200-230 mg/dL for total cholesterol, 10-20-30 ng/mL for folate, and 3-6-10 ng/mL for vitamin D were applied. Patient results were obtained using a single instrument (the same reading cell for Vitamin D and Folate) and the same lot (single column usage for HbA1c). The online application developed by Çubukçu at <https://hikmetc-apscalculator.streamlit.app/> calculated APS' relative standard measurement uncertainty (urel (%)).

**Results:** In our study, data from 1462 patients for HbA1c, 2829 for total cholesterol, 2291 for folate, and 2628 for vitamin D were used for APS calculation. Minimum (90% compliance) urel was determined as 3.9% for HbA1c, 6.4% for total cholesterol, 11.5% for folate, and 9.2% for vitamin D based on these calculations.

**Conclusion:** The use of a data-driven computer-assisted application in determining APS enables laboratory professionals to calculate measurement uncertainty using their own values, enhancing result evaluation and patient management in today's laboratory practices, as we believe in our study.

**Keywords:** measurement uncertainty, analytical performance, clinical decision limit, data

## OA-52

### Evaluation of the Diurnal Reference Intervals of Thyroid Function Tests Using Two Different Indirect Methods

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**Introduction:** Reference intervals (RI) are the most commonly used tool for medical decision-making and interpretation of test results. It is recommended that each laboratory should determine the RI specific to its method and local population. RI can be calculated by direct and indirect methods. Due to biological variation, it may not be appropriate to evaluate some laboratory tests with a single RI. The aim of this study was to determine the reference intervals of thyroid function tests, known to exhibit biological variation, at different times of the day using two different indirect methods.

**Methods:** Over the past 7 years (01.01.2016-31.12.2023), TSH (mIU/L), fT4 (ng/dL), and fT3 (pg/mL) values of 13,930 individuals aged 18- 65, who were admitted to the laboratory between 07:00 and 17:00 and had negative anti-TPO and anti-Tg results were utilized. Samples were partitioned into two sections according to the time of laboratory acceptance: before noon (07:00-11:59) and afternoon (12:00-17:00). The Tukey method was used to exclude outliers. After excluding outliers, reference intervals and 90% confidence intervals were calculated by the non-parametric and Battacharya methods using at least 9603 values for before noon (am), at least 3623 values for after noon (pm) and at least 13226 values for the entire day (ed). Bias Ratio (BR) was used to compare the reference intervals. The significance level in BR value was accepted as >0.375.

**Result:** The reference intervals were determined using the non-parametric method as follows: TSHed (0.19-5.34), TSHam (0.16-5.71), TSHpm (0.28-4.32), fT4ed (0.6-1.18), fT4am (0.6-1.18), fT4pm (0.62-1.19), fT3ed (2.6-4.26), fT3am (2.59-4.26), fT3pm (2.63-4.27) and using the Bhattacharya method as follows: TSHed (0.15-5.53), TSHam (0.13-5.86), TSHpm (0.25-4.52), fT4ed (0.61-1.19), fT4am (0.6- 1.17), fT4pm (0.61-1.19), fT3ed (2.56-4.32), fT3am (2.58-4.31), fT3pm (2.6-4.15).

**Discussion:** It has been observed that the RIs calculated by the two different methods were similar in all tests. It was observed that the RIs for TSH before and after noon were different in, while they were consistent in fT4 and fT3. It is expected that this difference will produce changes in clinical interpretation. Therefore, while we consider that the RIs for the TSH should be given separately for the before and after noon, it is clear that this is a preliminary study and further research is necessary to confirm the results.

**Keywords:** Reference Interval, Indirect, Biological Variation, Thyroid Hormones

## OA-53

### Preferences in Gestational Diabetes Mellitus Screening Tests: A Comparison of One-Step and Two-Step Approaches

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**Objective:** Gestational diabetes mellitus (GDM) is a metabolic disorder that occurs during pregnancy, increasing the risk of complications for both the mother and the baby during pregnancy as well as in the postpartum period. Identifying high-risk pregnancies is crucial for improving maternal and fetal health in both the short and long term. Two different approaches are commonly used for screening pregnant women and diagnosing GDM. In the two-step screening conducted between the 24th and 28th weeks of pregnancy, a 50-gram glucose challenge test is initially administered. Pregnant women with glucose levels of 140-180 mg/dl then undergo a 100-gram, 3-hour oral glucose tolerance test (OGTT). According to the Carpenter-Coustan criteria, GDM is diagnosed if 2 or more threshold values are exceeded. In the one-step screening, a 75-gram OGTT is administered between the 24th and 28th weeks of pregnancy, and GDM is diagnosed if at least one threshold value is exceeded according to the IADPSG criteria (fasting  $\geq 92$  mg/dl, 1-hour  $\geq 180$  mg/dl, 2-hour  $\geq 153$  mg/dl). Each approach has its advantages and disadvantages. The aim of our study is to examine which method is more commonly used for GDM screening in our hospital, the positivity rates, and clinicians' preferences.

**Materials and Methods:** In this study, data from a total of 6623 pregnant women who underwent one-step and two-step GDM screening tests in our hospital between 2020 and 2023 were retrospectively analyzed.

**Results:** The two-step screening test was applied to 3445 pregnant women, and 2969 completed the screening, with a GDM diagnosis rate of 5.85% (174 pregnant women). Out of 476 pregnant women with a 50-gram glucose challenge result of 140-180 mg/dl, none proceeded to the 100-gram glucose challenge test. The one-step screening test was applied to 3178 pregnant women, with a GDM diagnosis rate of 26% (827 pregnant women).

**Conclusion:** Although there is no consensus among clinicians regarding the approach to GDM diagnosis and screening, the one-step screening test application is increasingly becoming prevalent in line with the ADA/IADPSG guidelines published in 2011. The current guidelines of the Turkish Endocrinology Society recommend both one-step and two-step approaches for GDM screening. In our hospital, both methods are used for GDM screening in pregnant women. We believe that the one-step screening test might be more suitable due to the lower diagnostic rate and the challenges in implementing and monitoring the two-step screening test compared to the one-step screening test.

**Keywords:** OGTT, GDM, screening test

## OA-56

### Comparison of Osmolality Estimates Based on Conductivity and Biochemical Formula with Reference Method

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**Aim** Urine osmolality is evaluated to determine the body's hydration status and concentration ability of the kidney. However, since osmometer analyzers are difficult to obtain, alternative methods have been developed to estimate osmolality. In this study, we aimed to compare the conductivity-based method and the osmometric method for urine osmolality determination.

**Method** Urine samples of 56 patients were included in the study. For urine osmolality estimation, conductivity-based osmolality calculation and biochemical osmolality calculation formulas were used on the Zybio U2610 (Zybio Ins., China) device for routine complete urinalysis. The osmometric method (Osmomat 3000, Gonotec GmbH, Germany) was used for the reference method. In the literature, the formulas were determined as follows; Formula 1:  $[1.07 \times \{(2 \times \text{urine sodium, mmol/L}) + (\text{urine urea nitrogen, mg/dl} / 2.8) + (\text{urine creatinine, mg/dl} \times 2/3)\} + 16]$ , Formula 2:  $[2 \times (\text{urine sodium, mmol/L} + \text{urine potassium, mmol/l}) + (\text{urine urea, mg/dl} / 5.8)]$ . Biochemical analytes were measured on the Cobas c702 (Roche Diagnostics, USA) device. The repeatability study on the Zybio device was carried out with two levels of internal quality control samples for 5 days and 5 repetitions. Statistical and clinical evaluation was made for method comparison.

**Results** In the Zybio device, intra-day coefficients of variation (CV) were found to be 1.1% and 1.3% for the 1st and 2nd levels, respectively. Between-day CV was found to be 2.5% and 2.7%. The correlation coefficients between Osmomat, Zybio, Formula 1 and Formula 2 were found to be as  $r = 0.866$  ( $p < 0.001$ ),  $r = 0.923$  ( $p < 0.001$ ) and  $r = 0.948$  ( $p < 0.001$ ), respectively. There was a significant difference between the results of Osmomat and Zybio ( $p < 0.001$ ). But there was no significant difference between Osmomat and Formula 1, 2 ( $p = 0.271$  and  $p = 0.936$ , respectively). Bias% between Osmomat and Zybio, Formulas 1 and 2) were -53.7%, 14.2% and 3.8%, respectively. Bias % of Formula 1 and 2 were found to be acceptable according to biological variation limits (16.1%) except for Zybio.

**Conclusion** It has been determined that the Zybio device, which measures conductivity-based osmolality, cannot be used instead of reference osmolality measurement, and the formulas remain valid estimating urine osmolality. Therefore, there is a need to improve the Zybio conductance-based osmolality measurement.

**Keywords:** Automated urine analyzer, osmolality, conductivity



## OA-57

### An Interference Case in Free Prostate-Specific Antigen Analysis

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**Objective:** Prostate-specific antigen (PSA) is used for the early diagnosis, treatment monitoring, and staging of prostate cancer. Free PSA represents the unbound fraction of PSA in the bloodstream. The percent free PSA (proportion of free PSA to total PSA) is used to differentiate between benign and malignant prostate disorders. In this case, we investigated a discordant-free PSA result in terms of interference.

**Methods:** An 83-year-old male patient, diagnosed with sarcomatoid bladder cancer, underwent repeated free PSA testing using two different UniCel DxI 800 (Beckman Coulter, USA) analyzers with an unexpected elevation of percent free PSA. For interference analysis, total and free PSA measurements were repeated with an alternative analyzer (Siemens Advia Centaur XPT, Germany), the sample was treated with a heterophilic blocking tube (HBT, Scantibodies Laboratory, USA) and precipitated with 25% polyethylene glycol solution (PEG 6000, Merck, Germany). Additionally, the rheumatoid factor (RF) level was measured by an immunoturbidimetric method (Cobas 6000, Roche Diagnostics, Germany).

**Results:** Repeated free PSA results were 1.59 and 2.19 ng/ml and total PSA results were 0.14 and 0.17 ng/ml respectively. The free and total PSA results on the Siemens Advia Centaur XPT analyzer were 0.04 and 0.18 ng/ml, respectively. After treatment with the heterophilic blocking tube, the free and total PSA results were 2.28 and 0.14 ng/ml, respectively. Following PEG precipitation, the free and total PSA results were 0.02 and 0.20 ng/ml, with a recovery of 2.5% for free PSA. The RF result was 22 IU/mL and it was higher than the RF cut off (<14 IU/mL).

**Conclusion:** It is crucial to evaluate potential interference causes in unexpected and inconsistent laboratory test results. It should be kept in mind that especially in elderly patients high RF levels may be the cause of discordant immunoassay test results.

**Keywords:** interference, prostate-specific antigen, rheumatoid factor

## OA-58

### Experience with Immunofixation Electrophoresis: Evaluation of Our Initial Data

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**Objective:** Monoclonal gammopathies result from the clonal proliferation of plasma cells or B-lymphocytes. The presence of an isolated protein band, referred to as an M peak, which indicates monoclonal protein production in serum protein electrophoresis, suggests the potential existence of a proliferative disease related to plasma cells. In such cases, it is also necessary to conduct immunofixation electrophoresis (IFE) on serum and urine samples in addition to protein electrophoresis. The presence of findings indicating monoclonal immunoglobulin production in serum immunofixation electrophoresis is termed paraproteinemia. In addition to confirming the diagnosis, immunofixation electrophoresis is the gold standard method for determining the class of produced monoclonal protein. IFE analysis began at the Biochemistry Laboratory of Aydın Adnan Menderes University Faculty of Medicine in December 2023. This study aims to retrospectively evaluate all IFE reports for patients analyzed in our laboratory during the period up to June 2024.

**Methods:** Immunofixation electrophoresis of patients' samples, suspected of having monoclonal gammopathy and sent to our biochemistry laboratory, was conducted using the Interlab G26 device. In our study, all patient results from the six-month period in which electrophoresis was conducted were retrospectively reviewed using the Laboratory Information System.

**Results:** A total of 396 serum IFE analysis reports were evaluated. A paraprotein band was detected in the serum of 110 patients (27%). Among these, IgG lambda was found in 42 patients (38%), IgG kappa in 38 patients (34%), IgA kappa in 16 patients (14%), IgM kappa in 10 patients (9%), IgA lambda in 8 patients (7%), and IgM lambda in 6 patients (5%). Our study also reviewed 254 urine IFE results, with a paraprotein band present in 43% of these cases.

**Conclusion:** The immunofixation electrophoresis results for patients who presented to our hospital's hematology clinic with a preliminary diagnosis of monoclonal gammopathy provide insights into the frequency of monoclonal gammopathy and predictability of paraprotein type in these patients. IFE is a simple and effective method for the diagnosis of monoclonal gammopathies.

**Keywords:** Immunofixation electrophoresis, paraproteinemia, monoclonal gammopathy

## OA-60

### Predictive Role of Indoleamine 2,3-dioxygenase-Mediated Tryptophan/Kynurenine Pathway in Acute Renal Allograft Rejection

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**Introduction:** Renal transplantation is performed to prolong and improve the lives of those with end-stage renal disease. Despite advances in immunosuppressive therapy and treatment of infections, acute rejection still remains a problem following kidney transplantation. Indoleamine 2,3-dioxygenase is a rate-limiting enzyme in the degradation of the tryptophan via the kynurenine pathway. It has been proposed as an immunomodulatory enzyme and its activity is expressed by kynurenine/tryptophan ratio. The activation of Indoleamine 2,3-dioxygenase is involved in regulating immune responses. Its role in solid organ transplantation is still unclear. This prospective observational study aimed to assess the role of Indoleamine 2,3-dioxygenase-mediated tryptophan/kynurenine pathway metabolites for predicting acute renal allograft rejection.

**Patients and Methods:** This study included 66 renal transplant recipients (15 female, 51 male; mean age:  $41.98 \pm 12.88$ ) from living related donors. Blood samples were collected after transplantation at day 1 and 7. Serum creatinine levels were analysed by modified Jaffe method in Cobas 8000 analyser. Glomerular filtration rate (eGFR) was estimated by The Chronic Kidney Disease Epidemiology Collaboration equation. Serum tryptophan, kynurenine, kynurenic acid, anthranilic acid, 3-hydroxykynurenine, 3-hydroxyanthranilic acid, picolinic acid, quinolinic acid and xanthurenic acid levels were measured by using a multiplex LC-MS/MS Analysis kit (Jasem Tryptophan and Its Kynurenine Pathway Metabolites Kit, Altium International R&D Centre, Istanbul, Turkey). These measurements were performed by triple quadrupole LC-MS/MS system (6470A, Agilent Technologies, Santa Clara, CA, USA). Patients were assigned to 2 groups depending on their history and clinically diagnosed acute rejection [AR(+)] and without acute rejection [AR(-)]. We compared kynurenine/tryptophan levels and other metabolites between the groups using both Mann-Whitney U and Student's t tests. Data were expressed as mean  $\pm$  Standard deviations. SPSS was used to statistical analysis.

**Results:** Among 66 recipients, 35 had AR(+) and 31 had AR(-). Mean serum creatinine levels and GFR values were statistically different among these groups at all time points. Serum creatinine levels were found as increase while eGFR values decreases in the acute rejection group ( $p < 0.0001$ ). Serum 3-hydroxyanthranilic acid levels were significantly higher while 3-hydroxykynurenine and quinolinic acid levels were lower on day 7 compared to day 1 in the acute rejection group ( $p < 0.05$ ). Serum kynurenine/tryptophan ratios were increased significantly on day 7 compared to day 1 in AR(-) patients. 3-hydroxyanthranilic acid levels were significantly higher while 3-hydroxykynurenine, picolinic, kynurenic and quinolinic acid levels were significantly lower on day 7 in AR(-) patients.

**Conclusions:** The findings from our study have revealed interesting insights into the role of indoleamine 2,3-dioxygenase in kidney transplantation, specifically focusing its potential impact on acute rejection episodes. These associations suggest that it serves as a suppressor mechanism used by T-regulatory cells and may play a protective role in kidney transplant rejection. Indoleamine 2,3-dioxygenase modulation could offer novel therapeutic targets to enhance graft survival. Further research is needed to fully understand the mechanisms behind its immunomodulatory functions and its potential clinical value.

**Keywords:** Acute Renal Allograft Rejection, Tryptophan/Kynurenine Pathway, Indoleamine 2,3-dioxygenase

## OA-61

### Comparison of Anemia Incidence and Neutrophil/Lymphocyte Ratio in Alzheimer's Disease and Healthy Groups

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**Aim** Alzheimer's disease is considered the most common form of dementia, contributing to 60-70% of cases. Neurodegenerative diseases often emerge with cellular iron deficiency, energy depletion, and inflammation. Significant evidence suggests that inflammation plays a role in the pathophysiology of Alzheimer's disease. This study aims to test the differences between Alzheimer's patients and control group in terms of Neutrophil/Lymphocyte Ratio (NLR) as an inflammation marker and anemia parameters.

**Method** This research was designed as a retrospective study. The data of 493 patients diagnosed with Alzheimer's between 28.02.2022 and 01.01.2023 were reviewed for complete blood count (CBC) parameters. Sample size was calculated using G Power. The results of the patient group were compared with 80 healthy control subjects. CBC were determined in the Central Laboratory using the Sysmex XN-10 Automatic Hematology Analyzer (Kobe 651-0073 Japan) with EDTA-containing tubes. NLR was calculated using neutrophil and lymphocyte counts as follows:  $NLR = \frac{\text{Neutrophil count}}{\text{Lymphocyte count}}$ .

**Results** It was found that the values of RDW CV and NLR in Alzheimer's patients were significantly higher compared to the control group ( $p=0.034$  and  $p=0.025$ , respectively), whereas the values of RBC, HGB, HCT, and LYMPH % were significantly lower ( $p<0.001$ ,  $p<0.001$ ,  $p<0.001$ , and  $p=0.016$ , respectively). There were no statistically significant differences between the groups in terms of WBC, NEUT, LYMPH, MONO, EO, BASO, MONO %, MCV, MCH, MCHC, PLT, RDW SD, PDW, MPV, P-LCR, PCT, NEUT %, and BASO % values ( $p>0.05$ ).

**Conclusion** Overall, irregular iron metabolism is associated with cognitive impairments, including memory and attention deficits, as well as anemia or at least iron deficiency. It has been reported that amyloid precursor protein and tau have physiological roles in neuronal iron homeostasis. Neuropathological and neuroimaging studies have reported that brain iron levels are associated with plaque and tangle pathology. This study showed that the incidence of anemia and NLR as an inflammation marker were significantly higher in Alzheimer's disease compared to the control group.

**Keywords:** Alzheimer's disease, complete blood count, anemia, Neutrophil/Lymphocyte Ratio

## OA-62

### Laboratory and clinical physicians' collaborative approach: two cases with intravascular haemolysis post dialysis

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**Introduction:** Haemolysis during dialyses may occur due to reasons such as water-borne toxins, centralized dialysis equipment failure, blood tubing defects or from inadequate medical dialytic therapy. Progression of this condition within patients is a severe life-threatening condition. Haemolysis interference lead to incorrect laboratory results, hence, if this goes unnoticed, patients may be misdiagnosed and treated incorrectly.

**Case Presentation:** In this case report, two female patients in their 70's were admitted to our emergency department within 6 months period, post their haemodialysis session from the same dialysis centre. Case 1 applied with symptoms such as nausea, abdominal pain and vomiting. After a while ecchymosis and icterus developed. On the other hand, Case 2 had similar symptoms with an addition of sudden skin darkening. Following primary evaluation, Case 1 was referred to Internal Medicine Intensive Care Unit with pre-diagnosis of thrombotic thrombocytopenic purpura. Case 2 was referred to Gastroenterology department with pre-diagnosis of liver failure or cholestasis and ERCP was considered. Both patients' blood specimen was rejected due to high haemolysis index by laboratory staff. However, after second blood specimens were evaluated by laboratory physician, in vivo haemolysis was contemplated. Primarily, in vitro haemolysis reasons were eliminated and clinicians were informed that the patients may have in vivo haemolysis, therefore the followings tests were required; peripheral blood smear, reticulocyte count, Coombs test, haptoglobin and coagulation test. Blood samples were tested using dilution factors to exclude interference caused by high haemolysis index. Case 1 and Case 2 results respectively: Haemoglobin: 10.5 g/dL, 9.8 g/dL; Reticulocyte count: 0.0542 106/uL, 0.0704 106/uL; Reticulocyte percentage: 1.89 %, 2.47%; Platelet count: 82 103/uL, 148 103/uL; WBC count: 22.66 103/uL, 9 103/uL; Lactate dehydrogenase: 6980 U/L, 3279 U/L; Haptoglobin: 0,5 g/L, 0,5 g/L; Total bilirubin: 6.8 mg/dL, 2.4 mg/dL; Unconjugated bilirubin: 5.4 mg/dL, not detected; AST: 770 U/L, 280 U/L ; ALT: 290 U/L, 90 U/L; Amylase <3 U/L, 150 U/L; Lipase 190 U/L, 349 U/L; for both cases Coombs test: Negative and Schistocytes: Not detected. According to the assessment of both patients clinical follow up including variety of tests, in vivo haemolysis was verified; pre-diagnose of thrombotic thrombocytopenic purpura and ERCP procedure was eradicated for case 1 and 2 respectively.

**Conclusion:** In both cases, patients were treated and were discharged after their recovery. These case examples demonstrate that pre-analytical factors such as haemolysis affect laboratory results and laboratory physicians must be aware of these possible faulty interferences. Thus, importance of collaborative work between clinicians and laboratory physicians is highly important to obtain the optimum outcome for patients.

**Keywords:** Intravascular Haemolyses, Dialysis, Interdisciplinary Communication

## OA-63

### Impact of Diabetes on Immune Responses in COVID-19 Intensive Care Patients: An Observational Study

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**Aim:** Patients with diabetes have a known risk of severe COVID-19-related complications. We evaluated the effects of diabetes on the severity of infections and immune responses in COVID-19 patients admitted to intensive care units, aiming to clarify how underlying metabolic disorders influence COVID-19 progression and outcomes.

**Method:** We conducted this prospective analysis in the operational intensive care unit of a tertiary care hospital in Turkey. We involved patients diagnosed with COVID-19, categorized into diabetic and non-diabetic groups. We collected data on inflammatory markers, immune response, and diabetic parameters within the first 24 hours of admission. Between these two groups, we also assessed the presence of bacterial infection, the acute physiology and chronic health evaluation score II, the Glasgow Coma scale, and the use of mechanical ventilation. Statistical analyses were performed using GraphPad Prism version 10.2.2 and included the Mann-Whitney U test, Chi-square test, and t-tests, with a significance level set at  $p < 0.05$ .

**Results:** The study included 73 adult patients: 34 diabetic and 39 non-diabetic. The overall mortality rate was 48% ( $n = 35$ ) and was comparable between groups, with 23.2% in diabetics and 24.6% in non-diabetics ( $p = 0.245$ ). The diabetic group had a higher average age (68 years) and a longer median hospital stay (11.5 days), but these differences were not statistically significant ( $p = 0.363$ ,  $p = 0.154$ , respectively). The incidence of bacterial infections was nearly identical between the groups (35% in diabetics and 33% in non-diabetics). Mechanical ventilation usage rates were similar, with 65% in diabetics and 74% in non-diabetics ( $p = 0.446$ ). The Acute Physiology and Chronic Health Evaluation II scores were similar between groups, with diabetics averaging 16.8 and non-those with diabetes 18.3 ( $p = 0.393$ ). Both groups typically had a median Glasgow Coma Scale of 15 ( $p = 0.943$ ). Diabetic patients had a significantly higher neutrophil-lymphocyte ratio of 17.3 compared to 10.7 in non-diabetics ( $p = 0.020$ ). Diabetic patients also showed a significantly higher median neutrophil count of  $9.2 \times 10^3/\mu\text{L}$  versus  $7.3 \times 10^3/\mu\text{L}$  in non-diabetics ( $p = 0.037$ ). The percentage of lymphocytes was significantly lower in diabetic patients, at 5.4% compared to 7.5% in non-diabetics ( $p = 0.034$ ).

**Conclusion:** Our study confirms that diabetes significantly worsens both inflammatory and immunological responses in COVID-19 patients in intensive care settings. Increased neutrophil-lymphocyte ratios and neutrophil counts, along with reduced lymphocyte percentages, indicate a pronounced inflammatory response and weakened immune system in diabetics. Although there was no direct link to higher mortality or increased need for mechanical ventilation, these immunological alterations highlight the increased risk of severe outcomes in diabetic patients. This underscores the critical need for personalized medical strategies tailored specifically to diabetic patients. Understanding the relationship between diabetes and COVID-19 is essential for developing effective therapeutic interventions, aiming to optimize care and improve outcomes for this vulnerable population.

**Keywords:** diabetes, COVID-19, inflammatory response, immunological alterations, intensive care

## OA-64

### **An Indirect Data-mining Approach to Determine the Reference Interval of Activated Partial Thromboplastin Time in Adults**

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**Aim:** Routine coagulation assays, vital for preoperative screening, bleeding risk assessment, and anticoagulant therapy monitoring. Reference intervals are crucial for determining normal test results and are fundamental for disease diagnosis and clinical decision-making. Direct methods of establishing reference intervals are costly and have some ethical issues. On the other hand indirect methods like Hoffman, KOSMIC and refineR are more practical. In our study we aimed to determine the reference interval of activated partial thromboplastin time by refineR indirect method.

**Methods:** A total of 410.988 test results of the activated partial thromboplastin time results which were measured by Cobas t511 and t 711 analyzers with aPTT Screen reagent were extracted from the Laboratory Information System with the age (18-90), gender, and location of the patients between 1 January 2024 and 31 May 2024 and performed according to CLSI C28-A3 guideline. After the exclusion of invalid test results, multiple test results belonging to single patient, results from the certain departments (e.g., oncology, intensive care unit), and with diseases including coagulation disorders and diseases of the blood system, thromboembolic diseases, hemorrhagic diseases, heart and cerebrovascular diseases, malignant tumors, abnormal liver and renal function, 1253 unique activated partial thromboplastin time results were used. The expected pathological fraction was below 20% of the dataset. The refineR algorithm employs the default 1-parameter Box-Cox transformation to model the non-pathological distribution for each partition. This process uses this cleaned data set and incorporates bootstrapping to generate confidence intervals. Reference interval was calculated by using refineR (v1.6.1).

**Results:** We computed reference intervals for activated partial thromboplastin time results using the refineR algorithm. Upper and lower RIs for activated partial thromboplastin time results were 23,1 (95% confidence interval; 22,7-23,5) and 32,3 seconds (95% confidence interval; 31,1-32,6), respectively.

**Conclusion:** Reference intervals estimated using refineR method were compared with the reference interval (23.6-30.6 seconds) established by manufacturers' package insert. The computed reference interval for the Roche t 511 and 711 were basically consistent with the ones in the instructions, except the upper reference interval which was slightly higher. Laboratories should establish their own reference intervals according to the assessment of their patient database will provide better guidance for the diagnosis, monitoring and prognosis of patients.

**Keywords:** indirect reference interval, aPTT, refineR

## OA-65

### The Role of Long Non-Coding RNA-NEAT1 Expression in Metastatic Potential in Prostate Cancer cell lines

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**Aim:** Prostate cancer (PCa), one of the most common malignant tumors in the male urinary system, is a malignant tumor that originates in the prostate epithelium. According to the latest global epidemiological statistics in 2019, the prevalence of PCa has surpassed lung cancer, ranking first. Although significant progress has been made in the diagnosis and treatment of PCa, the long-term prognosis remains poor and it is still the second most mortal cancer in men. Therefore, there is an urgent need to explore new molecular mechanisms in PCa progression to design more effective treatment strategies. Increasing evidence suggests that long non-coding RNAs (lncRNAs) play significant roles in cancer development. LncRNA NEAT1 functions as an oncogene in many cancers. However, the roles of NEAT1 in PCa remain largely unknown. The present study aims to investigate the role of NEAT1 in the development of PCa.

**Methods:** The expression levels of NEAT1 were detected in PCa cell lines with high metastatic potential (PC-3), moderate metastatic potential (DU-145), and normal prostate epithelial cell line (PNT1A). The expression levels were measured using quantitative real time polymerase chain reaction assay (qRT-PCR). The relative expression of NEAT1 was calculated using the  $2^{-\Delta\Delta Ct}$  method. Fold regulation was calculated using the formula: Fold Regulation =  $(-1 / \text{fold change value})$  for fold change values less than 1. Upregulation was defined as having a fold regulation value of  $\geq 2$  and downregulation as having a value of  $\leq -2$ . Group differences were evaluated using the Student's t-test, with significance set at  $p < 0.05$ . Results: In this study, the expression levels of NEAT1 were examined in PC-3, DU-145 and PNT1A. Our findings show that NEAT1 is significantly increased in PC-3 prostate cancer cell line with high metastatic potential ( $p < 0.05$ ). No significant difference was observed in NEAT1 levels between the DU-145 cells with moderate metastatic potential and the normal cell line PNT1A ( $p > 0.05$ ).

**Conclusion:** The high expression of NEAT1, correlated with increased metastatic potential, strengthens the hypothesis that NEAT1 may function as an oncogene supporting the invasion and metastasis capabilities of cancer cells. The observed increase in NEAT1 expression, particularly in PC-3 cells, suggests that NEAT1 may play a critical role in the progression of prostate cancer. This preliminary finding promotes the hope for the development of therapeutic strategies targeting NEAT1 in the treatment of prostate cancer. In conclusion, this study reveals that NEAT1 is a potential oncogene in prostate cancer and may play a significant role, especially in cancer cells with high metastatic potential.

**Keywords:** prostate cancer; lncRNA-NEAT1; oncogene; qRT-PCR; metastasis



## OA-66

### **Human Papillomavirus and Cervical Cancer: Determining Prevalence and Genotype Distribution of HPV Infection Among Women in Azerbaijan Using Real-Time PCR Method.**

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**Abstract:** Human Papillomavirus (HPV) is the most common sexually transmitted infection. HPV are the etiological agents of numerous genital cancers. The Papillomaviridae is a family of small, non-enveloped viruses with double-stranded DNA genomes. Based on their association with cervical cancer HPV can be categorized as high- and low-risk HPV types. According to WHO cervical cancer ranks fourth most commonly occurring cancer in woman and the seventh most common cancer overall. Almost all sexually active people had be infected at least once in their lives. In the most cases, immune system eliminates HPV from the body. Otherwise persistent infection with high-risk Papillomavirus cause abnormal cells to develop, which can transform to cancer. That is why early screening for the presence “low risk” and “high-risk” types of HPV in the cervical epithelium very important for prevention malignant transformation in cervix in future.

**Aim:** This study aimed to determine the most common high-risk genotypes of HPV using real-time PCR method among women of Azerbaijan.

**Method:** During the study, cervical swabs were collected from 337 women aged 22–65 years at the National Oncology Center, from January 2020 to May 2024, and were tested for presence of Papillomavirus in cervical epithelium. For the detection of HPV DNA we used real time PCR test system (DNA-Technology). At first HPV DNA was extracted by the steps in specification using PREP-NA PLUS extraction kit (DNA-Technology) for manual HPV DNA isolation. Then we used HPV QUANT-21 Quantitative RT-PCR Kit, which is intended for the specific identification and quantification of 21 genotypes of HPV (3 type low-risk: 6, 11, 44; and 18 type high-risk: 16, 18, 26, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 68, 73, 82) in human biological samples. For amplification and detection of results was used 5-channel Detecting Thermocycler (DT 96, DNA-Technology).

**Results:** 201 tested samples (59.6%) were HPV DNA negative and in 136 samples (40.4%) HPV DNA was detected. Due to the fact that using method allows make screening and genotyping at the same time, we could analyze prevalence and distribution of HPV genotypes in our study group. So, among 136 women with positive result 64 (47.1%) of them was infected by one genotype of HPV (monoinfection), 72 (52.9%) of them infected by two or more HPV genotypes (multiple infection). The seven most common high-risk genotypes were 16 (23.5%), 68 (16%), 18 (15.4%), 56 (11.7%), 51(10.3%), 39 (10.3%), 31 (9.5%).

**Conclusion:** From an oncological point of view, cervical cancer is one of the most preventable and curable cancers worldwide, early screening and vaccination must be primary and secondary cervical cancer prevention strategies. But the most prevalent HPV genotypes among women can vary in different regions. So, in our study, we obtained results, that HPV genotypes specific for our region are 16, 68, 18, 56, 51, 39, 31. Therefore, specific HPV prevalence data in a specific region are closely related to future vaccine developments.

**Keywords:** HPV, Human Papillomavirus, Cervical Cancer, high-risk, Real-time PCR

## OA-67

### Routine Antibiogram of Clinical Enterococcal Isolates: A Speculation

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**Aim:** This study aimed to speciate, characterize, and detect antibiotic resistance in 200 Enterococcal isolates from various clinical specimens, excluding samples from commensal sites like stool and respiratory specimens. **Methods:** Phenotypic identification was performed using automated Vitek 2 Compact System and standard biochemical properties, with antibiotic sensitivity tested by Kirby Bauer disk diffusion method and Vitek 2 system, using a 628 AST card. Vancomycin resistance was genotypically detected by VanA and VanB gene testing. **Result:** A significant prevalence of Enterococcal infections was found among patients aged 21-40 years (45.5%), with a median age of 30.5 years, predominantly affecting outpatients (64%). *Enterococcus faecalis* was the most common species in outpatient settings (73.2%) and *Enterococcus faecium* in inpatient settings (56.3%), with the highest isolation from urine (64.5%), followed by pus (29.5%), body fluids (3%), and blood (3%). The antibiotic susceptibility pattern showed high sensitivity to linezolid (96.5%) and vancomycin (91.5%), but notable resistance to ciprofloxacin (73.7%), doxycycline (70.5%), erythromycin (70.4%), and high level gentamicin (56.5%), with 8.5% of isolates being vancomycin resistant and 2.5% linezolid resistant. *Enterococcus faecalis* demonstrated higher sensitivity to linezolid (98.6%) and vancomycin (94.9%) than *Enterococcus faecium*.

All urinary isolates were susceptible to fosfomycin, with 96.5% sensitive to linezolid and 91.5% to vancomycin. Ciprofloxacin resistance was noted in 73.6% of urinary isolates, followed by doxycycline (67.4%). High level gentamicin resistance was present in 58.1% of urinary isolates, and high level streptomycin resistance in 21.7%. Pus isolates showed 96.6% sensitivity to linezolid and 93.2% to vancomycin, with doxycycline being the most resistant antibiotic (77.9%), followed by erythromycin (69.4%). High level gentamicin resistance in pus isolates was 52.5%, and high level streptomycin resistance was 20.3%.

All blood isolates were sensitive to linezolid, and 83.3% were sensitive to vancomycin. penicillin and ampicillin were sensitive in 66.7% of blood isolates, while erythromycin was the most resistant (83.3%), followed by doxycycline (66.7%). Among six isolates from sterile body fluids, all were sensitive to linezolid, half were sensitive to ampicillin and penicillin, and one was resistant to vancomycin. Erythromycin and doxycycline resistance was seen in 66.7% of isolates. Vancomycin resistance (8.5%) was confirmed both phenotypically and genotypically showing VanA gene positivity in most vancomycin resistant enterococcal isolates. High level aminoglycoside resistance was present in 60.5% of isolates, with high level gentamicin resistance (56.5%) more prevalent than high level streptomycin resistance (21.5%).

**Conclusion:** The increasing prevalence of vancomycin and linezolid resistance in *Enterococcus*, as observed in our study, provide very few treatment options. These resistant strains are more common among admitted patients, where as high level aminoglycoside resistance is predominantly seen in outpatients. The study underscores the increasing antibiotic resistance in *Enterococci*, particularly in inpatient settings, with *Enterococcus faecium* showing higher resistance. The findings highlight the critical need for strict hospital hygiene practices, accurate species identification, and ongoing monitoring of resistance patterns to manage Enterococcal infections effectively.

**Keywords:** Vancomycin resistant Enterococci, Antibiotic resistance, *Enterococcus faecium*, *Enterococcus faecalis*

## OA-68

### Mixed Connective Tissue Disease or Not: The Dilemma with Positive anti-RNP Antibodies

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**Aim:** Mixed connective tissue disease(MCTD) is a rare autoimmune disease where there is positivity to anti-ribonucleoprotein(RNP) antibodies along with overlapping features of systemic lupus erythematosus, scleroderma and polymyositis. The occurrence of these antibodies in disorders like systemic lupus erythematosus pose a diagnostic dilemma where there are overlapping features of lupus in MCTD. The aim of this study was to understand the presentation and consequence of patients with anti-RNP antibodies to identify aspects that differentiate them.

**Method:** This was a retrospective cohort study of patients who visited the lab over a period of four years of 2018-2022 and had a positivity of anti-RNP antibodies by the line immunoassay (Euroimmune) at 1:100 sample dilution. Corresponding immunofluorescence patterns were also obtained from lab records. The symptoms at presentation and organ systems involved were recorded. The Clinical data was obtained from electronic data base and where not available the patients were contacted to obtain data. The study was started after Institutional Ethics Clearance and was conducted as per the Declaration of Helsinki.

**Results:** ANA profile positivity was observed in 697 out of 1616 individuals. Out of them anti-RNP was positive in 246(35.29%) individuals. They were predominantly females with a mean age in years of 28.5±6.31. The predominant pattern by immunofluorescence was the characteristic nuclear large/coarse speckled followed by nuclear fine speckled and nuclear homogenous pattern. Out of these 246 positive patients 21 were diagnosed with MCTD and undifferentiated MCTD. The involvement of musculoskeletal, skin and mucosa and gastrointestinal were in the order of 84.3%, 77.1%, 62.9% respectively. The antibody profile revealed rheumatoid factor positivity in 21.5%, 56% each positivity for anti-Sm and anti-SSA and 47% positivity for anti-Ro-52.2. Out of 246 individuals were 98 anti-RNP positive were diagnosed as per SLE criteria and showed positivity for anti-RNP. However, these SLE patients were clustered with anti-RNP and ds-DNA positivity and another with anti-RNP, anti-Sm and Anti-Ro52. Based on the cluster there were differential clinical features which when placed into the SLE criteria and the Kahn and Alarcón-Segovia criteria helped in improving the diagnosis of MCTD.

**Conclusion:** MCTD patients had more clinical features pertaining to joint pain and generalised weakness, edema in hands and skin lesions as compared to SLE who has more involvement of kidney. Anti-RNP antibodies are not the uniqueness of MCTD diagnosis as the association is also observed in SLE. However, SLE patients had clustering of antibodies along anti-RNP which also helped in ruling out MCTD from SLE. Further as already known the Kahn and Alarcón-Segovia criteria can be used to rule in the disease.

**Keywords:** Alarcón-Segovia, Anti-Sm, Coarse speckled, Kahn, Systemic lupus erythematosus

## OA-69

### Investigation of mirna-193b Expression and Stathmin 1 Level in Serum of Patients with Epithelial Ovary Cancer

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**Purpose:** Epithelial ovarian cancer (EOC) is the worst tumor among gynecological cancers due to its poor early detection rate and unfavorable prognosis. Early diagnosis of cancers, determination of prognosis and follow-up are vital for managing these diseases and restoring patients' health. For this purpose, it becomes important to identify biomarkers that will provide clinical information and prediction to healthcare professionals. There is no biomarker in management of EOC except CA125 for the follow-up of the patients. Considering the relationship of Stathmin 1 (STMN1) with EOC, it stands out as a candidate biomarker. Although the relationship between miR-193b, STMN1, and EOC has been demonstrated at the tissue and cell level, this relationship has not been investigated in sera of EOC patients. Therefore, in our study, the relationship between serum levels of STMN1 and miR-193b and EOC was investigated. Furthermore, their relationship with the clinical and pathological variables related to the disease and prognosis were evaluated.

**Materials and Methods:** 76 patients diagnosed with EOC and 79 healthy individuals who were followed up by the gynecology and gynecological oncology outpatient clinics of Istanbul University Istanbul Faculty of Medicine were included in our study. Relative expressions of mir-193b and STMN1 mRNA levels were measured by quantitative real time polymerase chain reaction assay (qRT-PCR); STMN1 protein levels were measured using the ELISA method. Results were evaluated using chi-square, Mann-Whitney U, Spearman correlation, ROC analysis.

**Results:** No significant difference was detected between the patient and control groups in terms of age, smoking habit, and BMI. There was no significant difference between the EOC and the control group regarding serum miR-193b and STMN1 mRNA expression and STMN1 protein levels. No significant relationship was found between serum miR-193b values and clinical parameters. Although there was no relationship between serum STMN1 values and grade, STMN1 expression and protein levels were significantly higher in advanced-stage patients compared to early-stage patients. There was no significant correlation between serum miR-193b and STMN1 levels, as well as between these parameters and CA125, CEA, and CA19.9 levels.

**Conclusion:** Our findings did not find the relationship between EOC and STMN1 or miR-193b at serum level, which was reported to exist at tissue and cell levels. However, STMN1 serum mRNA and protein levels were observed to be higher in advanced stages of the disease. New studies will contribute to determining the roles of both STMN1 and miR-193b in clinical diagnosis, follow-up and prognosis of EOC patients.

**Keywords:** Epithelial ovarian cancer, miRNA, mRNA, miR-193b, STMN1

## OA-70

### The Effect of Glycine on Cyclosporine Induced Oxidative Stress and Hepatotoxicity in Rats

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**Aim:** Cyclosporine is an immunosuppressive drug widely used in organ transplantations for preventing graft rejection and some autoimmune diseases. However, long-term use of cyclosporine may cause toxic effects such as nephrotoxicity, cardiotoxicity, and hepatotoxicity. Several investigators have suggested that increased reactive oxygen species formation and inflammation are effective in cyclosporine-induced hepatotoxicity. Therefore, antioxidant therapy seems to be a useful method to prevent hepatotoxicity induced by cyclosporine. Glycine is an amino acid with antioxidant, anti-inflammatory and immunomodulatory effects. In this study, we aimed to investigate whether glycine treatment has a protective role against cyclosporine-induced hepatotoxicity.

**Methods:** Sprague Dawley rats were divided into control, cyclosporine, cyclosporine+glycine-low dose, cyclosporine+glycine-high dose and control glycine groups. Cyclosporine (20 mg/kg/day; subcutaneously) was administered to rats along with glycine injections (250 or 1000 mg/kg; i.p) for 21 days. The level of total bilirubin and activities of alanine aminotransferase, aspartate aminotransferase and  $\gamma$ -glutamyl transferase were determined in serum. Hepatic levels of reactive oxygen species, thiobarbituric acid reactive substances, advanced oxidation protein products as prooxidant parameters, glutathione and ferric reducing antioxidant power as antioxidant parameters were measured by spectrophotometric methods. Tumor necrosis factor- $\alpha$  level and myeloperoxidase activity were determined in the liver tissue for the assessment of inflammation together with histopathology.

**Results:** Cyclosporine caused significant increases in liver function tests in serum, and hepatic prooxidant and inflammatory parameters as compared to control. Although hepatic glutathione level increased, ferric reducing antioxidant power did not alter in cyclosporine treated rats. Cyclosporine also caused degeneration in hepatocytes and sinusoidal spaces. Glycine, especially its high dose, was detected to alleviate cyclosporine-induced deterioration of hepatic function tests, histopathological alterations, oxidative stress and inflammation in rats.

**Conclusion:** Our results indicate that glycine may be useful for the prevention of cyclosporine-induced hepatic toxicity.

**Keywords:** Cyclosporine, glycine, hepatotoxicity, oxidative stress, inflammation

## OA-71

### **Proposal for a Patient-Centered Management Model: Integrated Cervical Pathology Unit for Women at High Risk of Cervical Cancer at DIPRECA Hospital**

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**Aims:** 1. Evaluate the cervical cancer prevention program: Screening coverage, access gaps, and management along the patient journey in Policemen Healthcare system in Chile. 2. Propose the formation of an Integrated Cervical Pathology Unit, based on value and focused on patients, with which the diagnosed gaps are solved.

**Methods:** A retrospective analysis of the screening program with the HPV molecular test, performed in cobas 4800 (detection of HPV 16, 18 and 12 clustered high-risk genotypes) is presented. The % coverage was determined based on the target female population of 7,500 women in the Metropolitan Region and the genotypes detected in women over 30 years of age. A survey of information was applied to understand the clinical route that women experience, conducting in-depth interviews with patients, health personnel who are in the process of care and also professionals and experts in the field in the Chilean public system. With this methodology, gaps, processes to be improved and costs associated with screening, confirmation and treatment were identified. This diagnosis led to a value proposition of an optimized clinical pathway for patients in an integrated Cervical Pathology Unit model, based on the principles of Value-Based Healthcare

**Results:** The coverage achieved in screening was 73% between January 2023 and June 2024, meeting the coverage goal with a high- sensitivity test, proposed by WHO. The HPV positivity in women 30 years or older was 16%, distributed in 157 women with HPV16+, 32 with HPV18+ and 560 with VPH12AR+. Diagnosis of the program: In the analysis of the patients' journey from screening, 5 moments with opportunity for improvement were identified, either in management, access or the experience of women. The two most relevant gaps were: a) access to gynecology- oncology after the identification of a positive high-risk result and b) the need for emotional support at the time of receiving the positive HPV result.

**Proposal:** to form a multidisciplinary unit, composed by gynecologist-oncologist, midwife, psychologist, in direct relationship with Laboratory, Anatomopathology and IT, working in coordination around the needs of patients with high risk of CC, based on standardized clinical protocols. This model includes the reporting of clinical outcomes, identified by the team and those reported by the patients. In addition, the costs of the cycle were estimated, with expected savings of 76USD per woman/year, and a projected reduction in cases of invasive cancer of 3/year.

**Conclusions:** The Cervical Cancer Prevention Program has managed to achieve screening coverage in accordance with what the WHO stipulates to eliminate this cancer. Positivity in women aged 30 years or older was higher than expected by previous studies in the same region. The gaps in access to colposcopy are common to all care programs and these figures allow us to better estimate the real demand based on the achievement of coverage, confirmation and treatment goals. The value proposition of the care model based on value-based-healthcare principles, considers women at the center of management and provides tools to achieve better health outcomes, increase patient satisfaction and generate savings for the healthcare system.

**Keywords:** HPV screening, Value-based healthcare, cervical cancer, laboratory medicine outcomes, patient-centered

## OA-72

### Measurement Uncertainty of the Albumin to Creatinine Ratio in Urine: Application to the Interpretation of Laboratory Results

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**Aim:** Urine samples are frequently used to diagnose, treatment and follow up various diseases. Different types of urine samples, such as random spot urine and 24-hour urine, are used in routine practice, but spot urine are preferred due to the ease of collection. Spot urine analytes are reported as a ratio to creatinine in routine clinical practice. Albumin and the ratio of albumin/creatinine measurements in the urine are essential in evaluating kidney pathologies. Therefore, urine biochemistry test results are crucial in clinical decision-making. Despite method validation, internal quality control, and proficiency tests to ensure the reliability of analytical results, there is always an uncertainty associated with the measured value. Measurement uncertainty refers to the quality of the analytical result and provides comparability between analytical results and specification limits. We aimed to calculate the uncertainty of measurement for quality management in the analytical phase for urine albumin, creatinine, and albumin/ creatinine ratio.

**Methods:** Measurement uncertainties of urine albumin, urine creatinine, and albumin/creatinine ratio were estimated using the ISO/TS 20914:2019 guideline. The Roche Cobas 8000 system was used for measurement of parameters. Expanded uncertainties were compared with the maximum allowable measurement uncertainty of the European Federation of Clinical Chemistry and Laboratory Medicine Biological Variation Database.

**Results:** The expanded measurement uncertainty estimated for level 1 of albumin, creatinine and albumin/ creatine ratio were 12.1%, 6.06 % and 13.16%, and level 2 were 5.13%, 4.52% and 6.28%, respectively.

**Conclusion:** Measurement uncertainties of albumin and creatinine have met the specification limits. These uncertainties should be considered when interpreting laboratory results, especially with the values that are close to the 30 mg/g decision threshold.

**Keywords:** Albumin, creatinine, uncertainty of measurement

## OA-73

### Impact of Pre-analytical Factors on the Diagnosis of Von Willebrand Disease

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**Introduction:** Von Willebrand disease (vWD) is the most common (1%) inherited bleeding disorder caused by deficiency or dysfunction of von Willebrand Factor (vWF) (1). Von Willebrand factor (vWF) is a multimeric glycoprotein defined as an acute phase reactant, synthesized in megakaryocytes and endothelial cells. VWF carries Factor VIII (FVIII) and enables the formation of the primary platelet plug to contribute hemostasis (2,3). Initial screening tests to diagnose VWD should include complete blood count, prothrombin time (PT), activated partial thromboplastin time (aPTT) and platelet function tests. In addition, vWF antigen (vWF:Ag), ristocetin cofactor test (vWF:RCo) that correlates with vWF:Ag, vWF:RCo/vWF:Ag ratio and FVIII activity should be performed specifically (1). While aging, pregnancy, hypertension, diabetes mellitus, stress, exercise and estradiol induces vWF release, hypothyroidism decreases vWF levels. Also 20-30 % lower vWF:Ag levels are seen in blood group O individuals compared with non-O individuals (4). Guidelines recommend that vWAg levels should be analyzed in completely healthy conditions (2). Factors increase VWF:Ag levels are shown in figure 1

**Aim:** After receiving feedback from clinicians that the low vWF:Ag and vWF:RCo results studied in our laboratory didn't correlate with each other and clinical findings of the patients, we aimed to examine all vWF:Ag and vWF:RCo results analyzed in 2021 in our study.

**Methods:** We examined the results of 100 patients in whom vWF:Ag and vWF:RCo were analysed in 2021. of these, fifty-seven patients with vWF:Ag levels below 50 IU/dL and/or uncorrelated vWF:Ag-vWF:RCo levels were included the study. VWF:Ag analysed by immunoturbidimetric method (ACLTOP700, Bedford, MA, USA) and vWF:RCo analyzed by light transmission aggregometer (CHRONO-LOG, Havertown, PA, USA). Patients' laboratory results and clinical data obtained retrospectively. The study population distributed into 3 groups according to the classifications in the guidelines (2): VWF:Ag levels below 30 IU/dL (VWD), vWF:Ag levels between 30-50 IU/dL (low vWF) and uncorrelated vWF:Ag-vWF:RCo levels.

**Results:** We observed that 3 patients' blood groups were O and one patient received hormone therapy during the analysis in the group of VWF:Ag levels below 30IU/dl (n:8). Three patients' blood groups were O and one had a serious infection during the analysis in the group of VWF:Ag levels of 30-50IU/dL (n:9). In the group of uncorrelated VWF:Ag-vWF:RCo levels (n:40); one patient was diagnosed with Type 2 vWD, 10 patients didn't have factors that could affect the test results, the remaining patients (n:29;72,5%) had conditions that could affect the results due to blood type, systemic disease in the history, pregnancy and hormonal reasons. Clinical characteristics and the classification of the patients according to laboratory findings are shown in table 1.

**Conclusion:** VWF is an unstable protein and there are many factors that affect its releasing pattern. Therefore, vWF:RCo and VWF:Ag levels may not correlate during the analysis. When evaluating vWF:Ag results to diagnose vWD, preanalytical factors that affect vWF:Ag levels and optimal time for sampling should be considered.

**Keywords:** Von Willebrand Factor, Von Willebrand disease



## OA-74

### **Building a Sustainable Laboratory Culture: The Power of Awareness and Strategic Training Programs**

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**Aim:** Global warming significantly affects temperature and seasonal patterns. The Paris Agreement targets limiting global temperature rise to 1.5°C above pre-industrial levels, with a 43% reduction in greenhouse gas emissions by 2030. The European Union aims to achieve climate neutrality by 2050. Laboratory medicine must support a sustainable healthcare system by using resources efficiently while delivering high-quality services. Labs, as major energy consumers, contribute significantly to carbon emissions and should work to reduce their carbon footprint as part of their societal responsibilities. To promote sustainability, we have organized a training program focused on carbon footprint reduction, green chemistry, and resource efficiency, aiming to adopt sustainable practices and create a more livable future.

**Method:** A face-to-face training program was conducted for 6 academicians, 5 biochemistry specialists, 4 resident doctors, 3 biologists, 22 laboratory technicians, 1 medical secretary, and 2 cleaning staff members working at the Antalya Training and Research Hospital Medical Biochemistry Laboratory. Additionally, an online training was provided to 11 individuals from the Medical Biochemistry Department of Bakırköy Dr. Sadi Konuk Training and Research Hospital, including 2 academicians, 1 biochemistry specialist, and 8 resident doctors. The program consisted of a total of 4 sessions, with 3 conducted in person and 1 online. The same training program was repeated for each group. A pre-test was administered to assess the baseline knowledge of the sample group, and a post-test was conducted to evaluate the effectiveness of the training. The pre- and post-tests, consisting of the same 14 questions, were used for evaluation. Questions 2 and 5 were scored at 8 points each, while the remaining questions were scored at 7 points each, with a total possible score of 100 points. Statistical analysis was performed using SPSS (version 15.0) and Prisma software.

**Results:** A statistically significant difference was observed when comparing the pre-test and post-test results of all participants. This difference was more pronounced in the assistant doctor and technician groups. The training was found to be effective, leading to a notable increase in awareness within the group. When comparing between groups, assistant doctors performed better in the pre-test. Additionally, the awareness level of assistants in the pre-test was significantly higher compared to other laboratory staff and technicians ( $p < 0.05$ ). In the post-test, the success of assistant doctors was significantly higher than that of technicians ( $p < 0.05$ ). Both assistant doctors and technicians showed substantial improvement in the post-test. No significant difference was found between face-to-face and online training ( $p = 0.137$ ).

**Conclusion:** The study aimed to raise awareness among laboratory staff about sustainability and climate change. Following this, our laboratory progressed towards the goal by obtaining the Green Lab Certification. The pre- and post-test method effectively measured the training's impact, highlighting strengths and areas for improvement. For a sustainable future, laboratories must enhance energy efficiency, reduce their carbon footprint, and adopt green chemistry practices. Ongoing education and awareness are crucial, supported by relevant policies, to minimize environmental impact and contribute to a sustainable healthcare system.

**Keywords:** Sustainability, climate change awareness, green laboratory, pre-post test, carbon footprint

## OA-75

### Preparation of Honey Extract Additive Sodium Alginate Hydrogels and in Vitro Wound Healing

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**Aim:** Conventional wound dressings have limited wound absorption capacity, are permeable to microbes and can adhere to wounds, leading to secondary injuries. Hydrogels are promising alternative dressings to overcome these challenges. The aim of this study was to develop hydrogel films based on sodium alginate loaded with honey extract.

**Methods:** These films were prepared by cross-linking method and evaluated for wound healing activity.

**Results:** The prepared films were 0.05-0.096 mm thick and flexible, indicating good physical properties. The optimized formulations showed successful loading of the extract into the film matrix without any interaction as confirmed by FTIR. The maximum zone of inhibition against Gram-positive and Gram-negative bacteria was achieved with the optimum formulation, i.e. 20 mm and 10 mm, respectively, with >89% removal activity. Moreover, this optimal formulation was able to achieve 88% wound healing in fibroblast cells. The histograms of the group treated with the optimized formulation also revealed a complete re-epithelialization of the wounds.

**Conclusion:** In conclusion, our honey extract-loaded hydrogel dressing successfully demonstrated its potential for wound healing.

**Keywords:** Sodium alginate, Hydrogel, Honey Extract, In Vitro Wound Healing

## OA-76

### Effect of Ramadan Diurnal Intermittent Fasting on the Metabolic Syndrome Components from a Laboratory Perspective

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**Aim:** The physiological changes during intermittent fasting and the potential effects of delayed or shortened sleep durations on human metabolism have not yet been fully determined. This study was designed to investigate the potential therapeutic effects of Ramadan fasting on the components of metabolic syndrome, given the alarming increase in metabolic syndrome cases worldwide.

**Method:** A total of 71 individuals, 16 with metabolic syndrome and 55 without metabolic syndrome, who agreed to participate in our study in the endocrinology department of our hospital; anthropometric measurements (weight, waist circumference) and blood parameters related to lipid and glucose metabolism (total cholesterol, triglyceride, HDL cholesterol, LDL cholesterol, glucose, insulin) were recorded in the first and last weeks of Ramadan. The volunteers, with an average age of 53,8±10,5, reported fasting for 3-4 weeks during the month of Ramadan. The two results obtained were evaluated by comparing them with the reference change values in our laboratory.

**Findings:** When we compared the means of all parameters at the end of Ramadan in individuals with metabolic syndrome (n=16), decreases were observed except for HDL cholesterol concentration. The percentage changes were 2% for weight, 3.2% for waist circumference, 4.5% for total cholesterol, 20.5% for triglyceride, 7.8% for HDL cholesterol, 4.8% for LDL cholesterol, 1.4% for glucose and 40.6% for insulin. When the means of all individuals participating in the study (n=71) were compared, decreases were observed except for glucose and HDL cholesterol. The percentage changes were 2.2% for weight, 2% for waist circumference, 1.8% for total cholesterol, 20.7% for triglyceride, 3.7% for HDL cholesterol, 2.5% for LDL cholesterol, 4.2% for glucose and 28% for insulin. To determine whether these changes were clinically significant, the reference change values of the tests at the 95% confidence level were calculated. Our reference change values were found to be 13% for total cholesterol, 38% for triglyceride, 15% for HDL cholesterol, 18% for LDL cholesterol, 11% for glucose, and 38.5% for insulin in serial measurements in our laboratory, and these were then compared with the % change values in our study. Except for the % change in insulin concentration in the individuals with metabolic syndrome group, no % change exceeded the reference change value of the relevant test. This suggests that the % changes in tests other than insulin in our study could be due to individual variation or preanalytical/analytical processes.

**Conclusion:** When the % changes were examined according to the reference change values, the fact that insulin showed a change exceeding the reference change value at the end of Ramadan in individuals with metabolic syndrome who fasted indicates the effectiveness of Ramadan fasting. The most widely accepted pathophysiological cause of metabolic syndrome development is insulin resistance, and in these patients, the incidence rates of hyperlipidemia, diabetes mellitus, hypertension, coronary artery disease, and non-alcoholic fatty liver disease have increased. In managing such a significant disease, there is a need for an affordable strategy that patients can easily adhere to, and intermittent fasting could be considered as an alternative to medical treatment for overcoming insulin.

**Keywords:** reference change value, RCV, intermittent fasting, metabolic syndrome

## OA-78

### Assessment of Mitochondrial Respiratory Capacity in Hemoglobinopathies by the Oroboros O2k-FluoRespirometer

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**Aim:** Thalassemia and sickle cell anaemia are among the most common rare inherited haematological diseases and are an important public health problem in Turkey and in the world. The only curative treatment is hematopoietic stem cell transplantation, but more frequently conventional treatments such as transfusion and iron chelation are applied. Many pathologies during the treatment are linked to mitochondrial dysfunction and oxidative stress (OS). Mitochondrial cofactors, known as 'mitochondrial nutrients' (MN)— including riboflavine, Coenzyme Q10 (CoQ10), and L-carnitine (CARN) have been evaluated in several clinical trials. This study aims to evaluate the effect by measuring the mitochondrial respiration rate in patients with thalassemia and sickle cell anemia in basal conditions and after the in vitro treatment of a cocktail containing NAD, coenzyme Q10, carnitine, and riboflavin to the cells.

**Methods:** The study included 12 sickle cell disease and 34 thalassemia patients followed up at the Istanbul Medical Faculty Pediatric Hematology Clinic. Cell free mt DNA levels were determined as a mitochondrial damage marker in plasma-derived DNA by RT-PCR. Patients with the highest level of damage were selected for further analysis. The final study was conducted with six patients diagnosed with hemoglobinopathies. Capricorn Scientific Lymphocyte Separation Medium LSM-A (500ml) was used for in vitro isolation of mononuclear cells (lymphocytes and monocytes) from peripheral blood. Oxygen consumption of mitochondria in lymphocytes was measured using an Oroboros O2k-FluoRespirometer (Oroboros Instruments, Innsbruck, Austria). Age- and gender-matched healthy control lymphocytes were also added to the analysis during each experiment. Basal and treatment forms of lymphocytes were analyzed with B2 (Riboflavin) 250 µM, Coenzyme Q10 50 µM, NAD 250 µM, L-Carnitine 250 µM concentrations. Results: When compared to the control group, both thalassemia and SCA patients had significantly higher levels of ccfmtDNA ( $p < 0,01$  and  $p < 0,001$  respectively). According to the Oroboros analysis results, it was determined that the oxygen consumption rate increased when basal and post-treatment respirations were compared ( $p=0.019$ ). The percentage of respiration increase in the respiratory chain compared to the baseline value was significantly higher than in the control group ( $p=0,0369$ ).

**Discussion:** Recent studies showed that supplementation of NAD<sup>+</sup> precursors in the salvage pathway is therapeutic for multiple pathologies. Maintaining the NAD<sup>+</sup> pool by delivering NAD<sup>+</sup> precursors can treat many disorders by improving mitochondrial function. Our study showed the results of a non-invasive method determining the effects of NAD<sup>+</sup> and other supplements used in hemoglobinopathies on oxidative phosphorylation. We suggest this non-invasive method to demonstrate several supplements' effects on oxidative phosphorylation. Oroboros is a fast and reliable analytical method using peripheral blood cells as the sample type. Secondly, ccfmtDNA is an erythrocytic DAMP (Damage Associated Molecular Patterns) that highlights the role of mitochondria in the pathology of inherited hemolytic anemias.

**Keywords:** Thalassemia Sickle Cell Anemia Mitochondrial respiratory capacity FluoRespirometer

## OA-79

### Prediction of Blood Tacrolimus Level After Liver Transplantation with Machine Learning Models Using Routine Laboratory Tests

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**Aim:** Tacrolimus is frequently used for immunosuppressive purposes to prevent tissue rejection after liver transplantation. Due to its narrow therapeutic index, close monitoring of blood level is crucial. However, tacrolimus level measurement can only be done in certain laboratories and patients can not always have easy access to them. Therefore, the prediction and relationship of tacrolimus level with laboratory tests more accessible were investigated.

**Method:** The study was conducted retrospectively by examining the Turgut Ozal Medical Center laboratory information system records. The tacrolimus level of liver transplantation patients over the age of 18, who were visited clinic for control purposes between 01.01.2019-31.12.2023 were used. According to the blood tacrolimus level, they were divided into four groups <5 (ng/mL, n = 7.682), 5-10(n = 5.449) , 10-15(n = 710) and > 15(n = 137).

**Results:** In comparisons among groups, there was a significant difference among age, ALT, AST, direct bilirubin, total bilirubin, ALP, albumin, hematocrit, hemoglobin, lymphocyte, MCV, erythrocyte, monocyte, MPV, neutrophil, PCT, PDW, PLCR, platelet and leukocyte level. The performance of machine learning models for predicting tacrolimus level was found to be highest for Cubist, followed by Radial Basis Function Support Vector Machines, Extreme Gradient Boosting (XGBoost), and Decision Trees, with R<sup>2</sup> values of 0.171, 0.137, 0.121, and 0.038 respectively. The Cubist model's prediction performance was most significantly influenced by the parameters of erythrocytes (100%) and age (74.8%).

**Conclusion:** Blood tacrolimus level affect and/or are affected by biochemical and hematological parameters. Blood tacrolimus level can be predicted using routine laboratory tests with machine learning models,

**Keywords:** tacrolimus, liver transplantation, machine learning

## OA-80

### Identification of New Psychoactive Substances (NPS) Abused in Turkish Population

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**Aim:** Next Generation Psychoactive Substances (NPS) are synthetic drugs that cause adverse events and deaths. Therefore, knowing and analyzing NPSs is crucial for public health. Identification of newly synthesized NPSs is analytically difficult and complex. The use profile of NPSs in Turkey needs to be rapidly revealed and included in routine analyses. Our study is based on biological samples (reasonable suspects for NPS use) obtained from psychiatric hospitals operating in Istanbul. Our aim is to find currently used NPSs, compare them with a library database, obtain certified reference materials for the analyte of interest, validate them and thus include them in routine analyses and accelerate related studies.

**Method:** In our study, 11 samples from patients known or suspected to use NPS were included in the analysis. High Resolution LC MS/MS (Thermo Q Exactive Focus, Orbitrap) was used for analysis. Samples were run twice using solid phase extraction and dilution. Samples were run twice after enzymatic hydrolysis by solid phase extraction and dilution. The possible analyte peak obtained in both runs for the same sample was evaluated in terms of mass to charge ratio (m/z), rt (retention time), isotope, fragment and area. Furthermore, the peak of interest was not detected in blank samples. The possible analyte peak was then entered into an Excel program for analysis. The m/z value, area, rt and fragments of the peak obtained by dilution were included in the evaluation program. In the program, percentage scores were determined for the processed rt, area and fragments. The data obtained were compared with an in-house library database containing 450 analytes.

**Results:** According to the data obtained, one of the 11 samples analyzed was positive for DIETHYLONE, 4 were positive for both 2- METHYLETHCATHINONE (2-MEC) and DIETHYLONE and 3 were positive for 2-MEC only. As a result of this study, the positivity rate for DIETHYLONE and 2-MEC analytes was high. Following benchmarking, the aim is to purchase certified reference materials (CRM) to include mass and pattern matched analytes in routine analysis. This will enable validation for the analyte of interest.

**Conclusion:** In conclusion, we believe that our study will be instructive in terms of obtaining CRM for the analytes with the available data and including these two analytes in routine analyses after verification and validation studies in a larger group and shedding light on many similar studies.

**Keywords:** new psychoactive substance, synthetic drugs, methylethcathinone, diethylone

## OA-81

### Importance of Pro-Oxidant/Antioxidant Balance in the Pathogenesis of Arthropathy with Hemophilia

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**Aim:** Hemophilia is associated with early onset and recurrent bleeding complications such as hemarthrosis which results in intra-articular inflammation, angiogenesis and the joint deformities. The deposition of iron-rich products, hemoglobin, hemosiderin and iron on the joint surface triggers the generation of pro-inflammatory cytokines and oxygen free radicals in the joint, and the iron together with oxidative stress has been shown to contribute the cartilage destruction. However, the pathogenesis of arthropathy in the hemophilic patients has not been clarified yet and no many studies about the effect of oxidative stress on the hemophilic arthropathy. The aim of this study was to investigate the role of pro-oxidant (reactive oxygen species, thiobarbituric acid reactive substances, advanced oxidation protein products, advanced glycation end products), antioxidant (total-thiol content and ferric reducing antioxidant power), and inflammatory (interleukin-1  $\beta$ ) markers in hemophilia cases with and without arthropathy and to elucidate the possibilities of supportive treatment that will increase the quality of life of these patients.

**Methods:** The study group consisted of 18 patients without arthropathy [10 (3-36) years] and 38 patients with arthropathy [31 (13-65) years] who were previously diagnosed with hemophilia (A/B) and were followed up at the Department of Pediatric Hematology and Oncology, Oncology Institute, Istanbul University. The patients were also evaluated according to body mass index and smoking habit. Patients with acute infection, chronic hepatitis, renal cardiac or autoimmune/rheumatic diseases were excluded from the study group. Serum concentrations of thiobarbituric acid reactive substances, advanced oxidation protein products, advanced glycation end products, total-thiol content and ferric reducing antioxidant power were measured by spectrophotometric method, while the reactive oxygen species was detected by fluorometry. The concentration of interleukin-1  $\beta$  was quantified by enzyme-linked immunosorbent assay.

**Results:** In this study, when the serum reactive oxygen species, thiobarbituric acid reactive substances and advanced oxidation protein products levels of patients with arthropathy were compared to those without arthropathy, no significant differences were observed, while the only advanced glycation end products levels were significantly increased [183.1(146.5-221.8) vs 208.8 (104.2-287.2), p=0.002]. Although interleukin-1  $\beta$  levels increased in those with arthropathy, 15% of increase was statistically insignificant. Ferric reducing antioxidant power levels, which indicate total antioxidant capacity, were also significantly increased in the arthropathy group [292.6 (189.0-422.1)] vs 329.2 (240.8- 533.4), p=0.04], while only 10% of increase was obtained in total-thiol content, insignificantly. When adjustment was made for smoking, which is known to stimulate oxidative stress, no statistical changes were observed.

**Conclusion:** According to our results, no significant increase was detected in oxidative stress and inflammatory parameters other than advanced glycation end products, while ferric reducing antioxidant power and total-thiol content indicate that the antioxidant defense mechanism is induced to

keep the oxidative stress in balance. These results suggested that antioxidant treatment may contribute to the prevention of the destructive joint deformities by suppressing of the onset and development of the complications.

**Keywords:** Key Words: Hemophilia A/B; Pro-oxidant/Antioxidant balance; reactive oxygen species; inflammation



## OA-82

### Evaluation of Analytical Performance of Anti-Müllerian Hormone By Sigma Metric Method

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**Aim:** Anti-Müllerian Hormone (AMH) is a key marker for evaluating ovarian reserve in the clinic setting. The Six Sigma methodology is a quality measurement method used to assess laboratory performance. This study aimed to evaluate the analytical performance of AMH using the Sigma metric method, incorporating both internal quality control (IQC) and external quality control data (EQA).

**Methods:** The retrospective study calculated sigma metrics using internal quality control and external quality assurance data collected between February and July 2024. The Coefficient of Variation (%CV) was calculated from low and high IQC data obtained from analyses performed on the Cobas 8000 (ROCHE Diagnostics, Mannheim, Germany) platform, and % bias was obtained from the NEQAS EQA program. Total allowable error (TEa) was derived using EFLM (European Federation of Clinical Chemistry and Laboratory Medicine) Biological Variation database. Sigma values were calculated separately for the low quality control and high control levels using the formula  $\Sigma(\sigma) = (TEa - \%Bias) / \%CV$ . Sigma values were categorized as follows:  $\leq 3$  (low), 3-6 (acceptable), and  $\geq 6$  (good).

**Results:** The Sigma values for low and high control levels of AMH were calculated as 4.37 and 6.72, respectively.

**Conclusion:** The Sigma metric method is a valuable tool for evaluating analytical methods and enhancing laboratory performance, enabling the correction of errors based on the results. The analytical performance of AMH was found to be the acceptable for the low-level control and good performance for the high-level control.

**Keywords:** anti müllerian hormone, six sigma, quality

## OA-83

### Indirect methods for cortisol reference intervals: A comparison of Bhattacharya, Hoffmann, Kosmic, RefineR methods with the ReflimR method

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**Aim:** Immunoassays and chromatographic methods for serum total cortisol measurement differ in terms of their analytical performance, reference intervals, diagnostic specificity, and sensitivity. This requires the definition of laboratory-specific reference interval(RI) or cut-off values. The Endocrine Society guidelines accept these differences in test performance, which can lead to misinterpretations. The RIs obtained using direct methods is costly and has ethical and practical challenges. To overcome these challenges, indirect methods such as Hoffmann and Bhattacharya, as well as software tools like KOSMIC, RefineR, and ReflimR that utilize different indirect methods, are used. Our aim is to calculate RIs for serum total cortisol using the current software, ReflimR, and to compare these with RIs obtained by other methods and those recommended by the manufacturer.

**Methods:** The study includes the cortisol results of 1,763 patients(aged 18-65) who admitted to outpatient clinics and whose blood samples were taken between 8-10 am in 2021 and 2022 years. Serum cortisol levels were analyzed using the ECLIA method on the Roche Cobas e801 analyzer with the Elecsys Cortisol II kit. Results outside the analytical range and outlier results were excluded from the study. Differences between genders were statistically evaluated. RI limits were calculated using indirect methods (Hoffmann, Bhattacharya methods and KOSMIC, RefineR, and ReflimR tools). The RIs obtained from ReflimR were compared with RIs from other methods/tools and RIs recommended by manufacturer. Results: After excluding outliers, a total of 1,432 cortisol data points (Females:1,089, Males:343) were obtained. For all individuals, the manufacturer's lower limit(LL) and upper limit(LL)(4.82-19.5 µg/dL) were not within tolerance limits of LL and UL calculated by ReflimR(6.8-26.9 µg/dL). The RIs for Hoffmann, Bhattacharya, Kosmic, and RefineR were 5.1-23.6 µg/dL, 6.4-21.5 µg/dL, 5.1-23.6 µg/dL and 5.5-23.6 µg/dL, respectively. The UL of Hoffmann, Kosmic and RefineR, and LL of Bhattacharya were overlapped with limits of reference limits calculated by ReflimR. As there is a statistically significant difference was found between genders(p=0.011), RIs were separately calculated for males and females. For males, the RIs for Hoffmann, Bhattacharya, Kosmic, RefineR and ReflimeR were 5.0-22.4 µg/dL, 6.2-22.3 µg/dL, 5.9-21.6 µg/dL, 5.3-21.6 µg/dL and 4.6-21.5 µg/dL, respectively. In males, while the UL at ReflimR overlapped with those at Hoffmann, Bhattacharya, Kosmic and RefineR, the LL at ReflimR overlapped with LL at Hoffmann and RefineR. For females, the RIs for Hoffmann, Bhattacharya, Kosmic, RefineR and ReflimeR were 5.2-24.0 µg/dL, 5.5-23.2 µg/dL, 5.6-24.5 µg/dL, 5.8-24.4 µg/dL and 6.7-27.7 µg/dL, respectively. In females, the UL at ReflimR overlapped those at Hoffmann, Kosmic and RefineR, while the LL at ReflimeR overlapped with those at Kosmic and RefineR.

**Conclusion:** The difference between the manufacturer's recommended RI for serum total cortisol measurement and the RI calculated with ReflimR was found. In both genders, the reference limits obtained by ReflimeR and RefineR were closer to each other than those obtained by the other methods. Indirect methods can be used instead of direct methods to determine or verify laboratory-specific RIs. However, differences in the selection and application of the indirect methods used should be considered.

**Keywords:** Reference interval, reference limits, cortisol, indirect method

## OA-84

### Verification of Enzyme Reference Intervals by Indirect Method

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**Aim:** Reference intervals (RIs) are expressed as the central 95% of the results obtained from healthy individuals using direct methods. Although direct methods are considered the 'gold standard', they are difficult to perform on a sufficient number of reference individuals due to ethical, time and financial difficulties. Although the guidelines recommend a simplified verification using 20 healthy individuals, this approach may not be sufficient to verify a very wide RIs. Therefore, indirect methods based on advanced statistical models that include a larger number of pathological and non-pathological results have been developed. Our aim was to calculate RIs for 8 different serum enzyme levels in our laboratory using the indirect method and to compare these data with the manufacturer's recommended RIs and RIs obtained using the direct method.

**Method:** Patients (18-65 years old) who were admitted to our hospital as outpatients in 2021 and 2022 and whose blood samples were taken between 8 and 10 am were included in the study. Patients' data were the test results at first admission of patients to hospital. Serum alkaline phosphatase (ALP), alanine transaminase (ALT), amylase, aspartate transaminase (AST), creatinine kinase (CK), gamma-glutamyl transferase (GGT), lactate dehydrogenase (LDH) and lipase enzyme activities were measured using a Cobas c702 analyser (Roche Diagnostic, Germany). Data outside the analytical measurement range and outliers were not included in the analysis. The difference between the genders was evaluated statistically. The R-based packages RefineR and ReflimR were used for the indirect method. The data obtained with ReflimR were compared with the manufacturer's recommended RIs and the RIs obtained with RefineR and the direct method (parametric and non-parametric).

**Results:** Male and female RIs were calculated separately by ReflimR and RefineR due to the statistical difference between the genders, except for amylase, LDH and lipase. Also, in all tests calculated by ReflimR, there were no overlapping between CI of reference limits of males and females. Despite the statistical difference, only ALT and GGT reference limits did not overlap in RefineR in men and women. All reference limits (96.2%) were in agreement except for the CI of the upper limit of GGT estimated by ReflimR and RefineR. 42.9% of the RI recommended by the manufacturer, 51.7% and 64.3% of the RI obtained using parametric and non-parametric methods, respectively, overlapped with those of ReflimR.

**Conclusion:** ReflimeR results were similar to those of the more complex and time-consuming RefineR method. However, many of the reference limits recommended by the manufacturers could not be verified. ReflimR results showed better agreement with the non-parametric method than with the parametric method. Considering the results, ReflimR can be recommended for the verification of existing RI rather than for the determination of denova RI.

**Keywords:** Reference interval, enzyme activity, indirect method, direct method

## OA-86

### Investigation of the Relationship Between Growth Retardation and Serum Stanniocalcin-2 /PAPP-A2/ IGFBP3 Axis in Children with Transfusion-Dependent Thalassemia

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**Aim:** Thalassemia is a hereditary disorder resulting from the lack of synthesis of one or more globins in the hemoglobin molecule. The primary treatment method for thalassemia is regular blood transfusions. Over time, complications related to the endocrine system develop in thalassemia patients due to these transfusions, with growth retardation being the most common endocrine disorder. This study aims to investigate the relationship between growth retardation and the serum Stanniocalcin-2 / PAPP-A2 / IGFBP-3 axis in children with transfusion- dependent thalassemia major.

**Methods:** The study was conducted with 42 children aged 7-13 years with thalassemia major who were being followed at the Pediatric Hematology Department of Ataturk University Faculty of Medicine. These children were then divided into two groups: those diagnosed with short stature (n=21) and those with normal growth (n=21). A control group consisting of 21 healthy children who presented to the outpatient clinic for routine check-ups was also included. The demographic data, anthropometric measurements, and laboratory tests of the patients were recorded. Serum Stanniocalcin-2, PAPP-A2, IGFBP3, and IGF-1 levels were measured using the enzyme-linked immunosorbent assay (ELISA) method. The study was supported by the Ataturk University “ Individual Research Project” under project code no. TYL-2023-13351.

**Results:** In patients with transfusion-dependent thalassemia major, we found significantly higher levels of Stanniocalcin-2, IGFBP3, and IGF-1, and lower levels of PAPP-A2 compared to the healthy control group. In thalassemia major patients diagnosed with short stature, Stanniocalcin- 2, total IGFBP3, and total IGF-1 levels were significantly higher, while PAPP-A2 levels were lower, compared to those with normal growth and the healthy control group. When the groups were evaluated as a whole, there was a significant and high negative correlation between serum Stanniocalcin-2, IGFBP3, and IGF-1 levels and height SDS, while there was a significant and high positive correlation with PAPP-A2.

**Conclusion:** In conclusion, our study suggests that there may be a relationship between short stature and the Stanniocalcin-2/PAPP- A2/IGFBP3 axis in patients with transfusion-dependent thalassemia major. Measuring the levels of these axis components at an early stage could be useful in predicting short stature in thalassemia patients.

**Keywords:** Blood transfusion, IGF-1, IGFBP3, PAPP-A2, short stature, Stanniocalcin-2, thalassemia major

## OA-87

### Sirt1 Levels in Behcet Syndrome

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**Aim:** Behcet syndrome is a variable vasculitis that can affect multiple organ systems. SIRT1 plays a role in regulating biological processes, including energy balance, inflammation, oxidative stress, etc... Most studies have demonstrated the importance of SIRT1 in the pathogenesis of autoimmune diseases, including rheumatoid arthritis, systemic lupus erythematosus, and others. We aimed to ascertain the levels of SIRT1, IL-6 and TNF- $\alpha$  in the serum of patients with Behcet syndrome, to compare these levels with those of healthy individuals and to elucidate the relationships with disease activity scores. Additionally, the objective was to identify if SIRT1 as a biochemical marker for Behcet syndrome.

**Methods:** The study population comprised 40 patients and 40 healthy age- and gender-matched volunteers. The disease activity status of patients was determined according to the Behcet Disease Current Activity Form (BDCAF). The score of  $\geq 4$  were considered as active and  $< 4$  were considered inactive.

**Results:** CRP, IL-6 and TNF- $\alpha$  values were found to be statistically significantly higher in patients than in the control group. SIRT1 values were higher in the patients than in the controls, but there was no statistical significance. A comparison of patients using and not using TNF- $\alpha$  blockers revealed significantly elevated TNF- $\alpha$  values in the former group. According to BDCAF, 12 patients were considered active and 28 patients were considered inactive. No significant difference was observed in SIRT1 and IL-6 values between these two groups. The levels of CRP and IL-6 were found to be statistically significantly higher in patients than comparison to the control group but no statistically significant difference was identified between the active and inactive patient groups. TNF- $\alpha$  levels were found to be statistically significantly higher in patients with active disease compared to those with inactive disease and the control group. On the other hand, TNF- $\alpha$  levels were also found to be statistically significantly higher in the inactive patient group compared to the control group. When the median values were taken into account, SIRT1 values were found to be higher in active patients than in the control group and lower in the control group than inactive patients, but no statistical significance was found between these values. When patients using and not using TNF- $\alpha$  blockers were compared, it was found that TNF- $\alpha$  values were significantly higher in patients using TNF- $\alpha$  blockers. No difference was found in SIRT1 and IL-6 values. However, when the median values were examined, it was seen that IL-6 and SIRT1 values were higher in patients using TNF- $\alpha$  blockers.

**Conclusion:** In our study, we could not establish a correlation between SIRT1 levels and disease activity. This may be related to the small patient number of our study and may also be related to the fact that our patients were receiving active medical treatment. For this purpose, investigating SIRT1 levels in newly diagnosed Behcet syndrome patients before the initiation of treatment may pave the way for new studies.

**Keywords:** BEHCET, SIRT1

## OA-88

### Opioids Analyses on LC-MSMS

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**Aim:** The threshold concentration for legal conviction for urine opiate is 2000 ng/ml. In the analyzes carried out by immunochemical method, the antibody found in ready-made commercial kits for opiate level was created against morphine and was compared with morphine and derivatives. Opiate derivatives are heroin, morphine, codeine, norcodeine, dihydrocodeine, hydrocodeine, morphine 3 glucuronide (M3G), morphine 6 glucuronide (M6G), hydromorphone. In the analyzes performed by the immunochemical method, the kit produces a result regardless of the opiate substance. And this result is above 2000 ng/ml, it is considered positive. Although drug and stimulant screening analyzes in urine are commonly performed by immunochemical method, biochemistry laboratories using LC-MSMS has been increasing in the last years. However, in the screenings carried out with this method, the analytes which a calibration curve is drawn with the standard can be scanned, and substances for which standards are not used cannot be identified. Opiate-derived analytes include morphine, morphine derivatives, codeine, codeine derivatives, diacetyl morphine, 6 monoacetyl morphine. In this case, if not all morphine conjugates are examined, there may be a possibility of giving false negative results with the chromatographic method. The aim of this study is to draw attention to the necessity of not missing the substances that need to be analyzed when applying chromatographic methods in which opiates and their derivatives are analyzed one by one.

**Method:** Although chromatographic methods are accepted as the reference method, to understand the reasons why the opioid results were above the threshold value by the immunochemical method and same samples were found to be below, in the screening and confirmation analyzes. 125 samples, were selected by random among the samples reported as opiate results 2000-5900 ng/ml by immunochemical screening. They were re-studied chromatographically, it was seen that in 82% (103 of 125) of these samples, the results of all opiate-related substances; morphine, codeine, norcodeine, hydrocodeine, dihydrocodeine and diacetylmorphine levels did not exceed 300 ng/ml totaly. All technical reasons were excluded and it was determined, only untreated derivative opiates could cause this. These samples were re-studied by adding M3G, M6G and hydromorphone to our chromatographic analysis method.

**Findings:** When the same samples were evaluated under the standard curve containing M3G, M6G and hydromorphone analytes, it was observed that M3G and M6G levels were at least 1650,0 average of 2700,0 ng/ml.

**Conclusion:** M3G and M6G, generally are not screened using the mass measurement method. Morphine is converted in to its main derivatives, M3G and M6G in the liver. Even though the threshold value of opioid analytes for screening samples analyzed with the LC-MSMS device is 300 ng/ml, it is seen that many samples do not exceed this threshold value. Moreover according to the health practice communiqué (SUT), it can be seen that M3G and M6G analysis are not included among the verification analysis codes. In this case, it is concluded that all laboratories performing substance screening and verification analysis with the LC-MSMS method must add M3G and M6G tests to their test panels.

**Keywords:** Opioid screening and confirmation analyzes, LC-MSMS

## OA-89

### Necessity of Drug and Stimulant Control in Traffic

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**Aim:** At the traffic alcohol examination, after quantitative measurements of ethanol levels in the blood in medical biochemistry laboratories, private car drivers whose blood ethanol levels are above 50 mg/dl are given administrative fines and their driver's licenses are suspended for six months. In the event of a second violation of the prohibition on driving under the influence of alcohol, the driver's license will be suspended for two years; in the event of three or more violations, the driver's license will be suspended for five years each time. However, driving under the influence of drugs and stimulants is not questioned during traffic stops or is rarely questioned. Probation cases are those who have substance use and who after being caught once, go to a hospital where there is a probation outpatient clinic three times, two weeks apart, and ask for analysis of their urine sample. Approximately 90% of these cases give samples six times at intervals of one to two weeks after the first three checks, and some are even referred to advanced probation outpatient clinics and give samples six more times. And throughout all these processes, people's driver's licenses are not taken. The aim of this study is to report the rate of drug positivity in urine samples of these cases followed up in the probation clinic during repeated hospital admissions and to draw attention to the fact that these individuals, who should not have a motor vehicle license, are still actively working as drivers.

**Method:** The consecutive results of the individuals who underwent urine screening analysis for narcotics and stimulants and who were under surveillance by the probation department were examined. All data were taken from the laboratory information system. In the last five years, out of a total of 38,000 samples of people who had their urine screened for substances and gave consecutive samples at least two weeks apart,

**Findings:** 80% of the consecutive controls of the samples belonging to the same person were found to be positive in at least half of their visits. The most common and almost constantly used substance was methamphetamine, followed by pregabalin, cannabis. More than one substance was used together in approximately 50% of the positive samples.

**Conclusion:** According to Article 48 of the Highway Traffic Law No. 6047, technical devices are used by law enforcement officers to determine the amount of alcohol in the blood at the during traffic inspections. However, there is no routine practice for drug and stimulant use on highways. People who are under probation, their driver's license are still active in traffic. In probation clinics, people who continue to actively use amphetamine and derivative stimulant drugs, euphoric drugs such as pregabalin, or some hallucinogens, or narcotic agents such as heroin and other opiate derivatives are monitored. During probation follow-ups, their driver's licenses should canceled by administrative institutions without waiting for traffic control, just like people with alcohol use disorder.

**Keywords:** Probation cases, Use of narcotic and stimulant substances, driving licence

## OA-90

### Decreased Serum Tryptophan and Quinolinic Acid Levels May Be Related to Generalized Anxiety Disorder in Children and Adolescents

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**Aim:** Generalized Anxiety Disorder is one of the most prevalent anxiety disorders among children and adolescents. Imbalances in the tryptophan/kynurenine pathway, particularly an excess or change in the metabolites ratio of specific neuroactive features, can be linked to some neurological and psychiatric conditions. To our knowledge, this study is the first investigation of the relationship between the tryptophan/kynurenine pathway and generalized anxiety disorder in children and adolescents. This study aims to investigate the potential impact of serum levels of tryptophan, kynurenine, kynurenic acid, and quinolinic acid levels, known as tryptophan/kynurenine pathway markers, on the etiology and clinical course of Generalized Anxiety Disorder.

**Methods:** This study was conducted between May 2022 and October 2023 at the Gazi University Faculty of Medicine Hospital. Participants were selected from Child and Adolescent Psychiatry Outpatient Clinic and samples were analyzed at the Department of Medical Biochemistry of the hospital. A total of 26 treatment-naïve children with Generalized Anxiety Disorder and 38 healthy controls were included in the present study. All participants were interviewed with Schedule for Affective Disorders and Schizophrenia for School-Age Children by certified psychiatrists to confirm Generalized Anxiety Disorder diagnosis and exclude comorbid psychiatric disorders. The severity of Generalized Anxiety Disorder symptoms was assessed by revised Child Anxiety and Depression Scales. Venous blood samples were collected and centrifuged at 1.300g for 10 minutes to obtain the separated serum. The serum tryptophan, kynurenine, kynurenic acid, and quinolinic acid levels were measured using original commercial ELISA kits.

**Results:** 26 patients with Generalized Anxiety Disorder (7 male, 19 female; median (IQR 25-75) of age 170 (138-190) months) and 38 healthy controls (18 male, 20 female; age 154 (123-180) months) presented comparable age and gender. Serum Tryptophan and Quinolinic acid levels of the Generalized Anxiety Disorder group were significantly lower than the control group. Tryptophan (median [IQR], 62.8 (19.7- 82.3) vs. 210 (58-264) mg/L,  $p = 0.001$ ) and Quinolinic acid (median [IQR], 9.9 (7.8-11.4) vs. 11.9 (10-17) ng/mL,  $p = 0.004$ ). The groups had no significant difference regarding kynurenine and kynurenic acid levels ( $p > 0.05$ ). Serum levels of tryptophan, kynurenic acid, and quinolinic acid levels were negatively significantly correlated with Generalized Anxiety Disordersymptom severity ( $p=0,008$ ,  $p=0,047$ ,  $p=0,002$  respectively).

**Conclusions:** The present research explored whether the kynurenine pathway imbalances can be the underlying cause of Generalized Anxiety Disorder in childhood. It is thought that the kynurenine pathway has been proposed to play an essential role in neuronal inflammation and alterations in the central nervous system. Imbalances in kynurenine pathway cause an excess or change in the proportion of metabolites with neuroactive compounds. The findings suggested that children with Generalized Anxiety Disorder had lower serum tryptophan and quinolinic acid levels than controls. There were also significant correlations between serum tryptophan and quinolinic acid levels with Generalized Anxiety Disorder symptom severity. These findings point that serum tryptophan oxidation pathway marker levels may be related to Generalized Anxiety Disorder etiopathogenesis, depending on Generalized Anxiety Disorder symptom severity.

**Keywords:** Generalized Anxiety Disorder, tryptophan/kynurenine pathway, tryptophan, kynurenine, kynurenic acid, quinolinic acid



## OA-91

### Lifotronic H9 HbA1c Analyzer User Verification

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**Purpose:** Laboratory services are vital at every stage within healthcare organizations, from diagnosis to treatment. The accurate assessment of HbA1c, which can influence clinical decisions with slight variations, requires medical laboratories to play a critical role. Hence, the reliable measurement of laboratory tests is of utmost importance. In this study, user verification of the newly introduced HbA1c analyzer in our hospital was performed.

**Method:** Our study employed precision and accuracy values for the user verification of the Lifotronic H9 device, in accordance with the CLSI's EP15-A3 and EP09 guidelines. These values were obtained through a method comparison with the BioRad VARIANT II device. The device's acceptability criteria were established based on the technical specifications provided by the manufacturer. Patient samples were tested with both the Lifotronic and the routinely used BioRad devices. The concordance between the two methods was assessed using Passing-Bablok and Bland-Altman analyses.

**Result:** The calculated repeatability and within-laboratory coefficient of variation (CV<sub>r</sub> and CV<sub>wl</sub>, respectively) were 0.69% and 1.19%, which fell within the verification limits when compared to the manufacturer's specified values of 5% and 10%, respectively. The Bland-Altman plots revealed a heterogeneous distribution of measurement differences around zero. It was also determined that the Lifotronic device measured lower values than the BioRad device for HbA1c measurements at low and normal concentration levels. Upon reviewing the Passing-Bablok analysis, it was observed that the confidence intervals were closely aligned with the desired values.

**Conclusion:** Despite some observed bias in concentration-related results, this bias remained below the permissible total error, and the overall analytical performance of the Lifotronic H9 device was considered satisfactory. The Lifotronic H9, suitable for our laboratory, stands out for its superior analytical performance, accuracy, and high sensitivity.

**Keywords:** Bias, Glycated Hemoglobin A, Methods