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INTRODUCTION

Dear colleagues and friends,

The Association of Medical Biochemists of Bosnia and Herzegovina has the pleasure of inviting You to the 23rd Meeting of the Balkan Clinical Laboratory Federation to be held in Sarajevo, on October 7-9, 2015, joined together with the 2nd Congress of Medical Biochemists of Bosnia and Herzegovina.

The Congresess will be held under the auspices of the International Federation of Clinical Chemistryand Laboratory Medicine (IFCC), European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) and Balkan Clinical Laboratory Federation (BCLF).

This is the first time that our Association of Medical Biochemists of Bosnia and Herzegovina acts as the organizer of the BCLF Meeting.

Sarajevo is the capital and largest city of Bosnia and Herzegovina, a place where every building tells a story be it serene or of strife. It is a place where the old world meets the new world; where The East meets The West. The city is famous for its traditional, cultural, and religious diversity, with adherents of Islam, Orthodoxy, Catholicism and Judaism, coexisting there for centuries. Due to this long and rich history of religious and cultural variety, Sarajevo was sometimes called the "Jerusalem of Europe".

In 1885, Sarajevo was the first city in Europe and the second city in the World to have full-time electric tram network running through the city. In 1914, it was the site of the assassination of the Archduke of Austria-Hungary which sparked World War I. Seventy years later, it hosted the 1984 Winter Olympics. Ever since the Olympic games Sarajevo has been a popular tourist attraction.

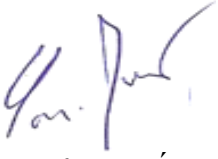
Nowadays Sarajevo is the leading political, social and cultural center of Bosnia and Herzegovina, a prominent center of culture in the Balkans, with its region-wide influence in entertainment, media, fashion, and the arts.

The Sessions of 20th BCLF Meeting will be devoted to advanced technology, biological variation, blood gases, biomarkers of renal function, cancer and tumor markers, cardiovascular diseases, clinical application of quantitative spectrometry, critical care/ emergency lab, education and training in laboratory medicine, endocrinology, hematology, hemostasis, laboratory errors and patient safety, laboratory safety, metrology and standardization in laboratory medicine, osteoporosis and bone metabolism, pediatric and elderly laboratory testing, point – of – care testing , preanalitical variables, quality and accreditation in laboratory medicine, reference ranges and decision levels toxicology and therapeutic drug monitoring, trace elements and much more.

Lectures and sessions of the 23rd BCLF Congress will be dedicated to the most recent findings in biochemistry and related sciences. The 23rd BCLF Congress will host many distinguished scientists and lecturers with the intention to increase the participation of clinical chemists from all the Balkan countries, as well as from the neighboring states.

We are looking forward to meeting you in Sarajevo this October. Join us and enjoy the 23rd BCLF Meeting and your stay in Sarajevo.

President of the 23rd BCLF Meeting



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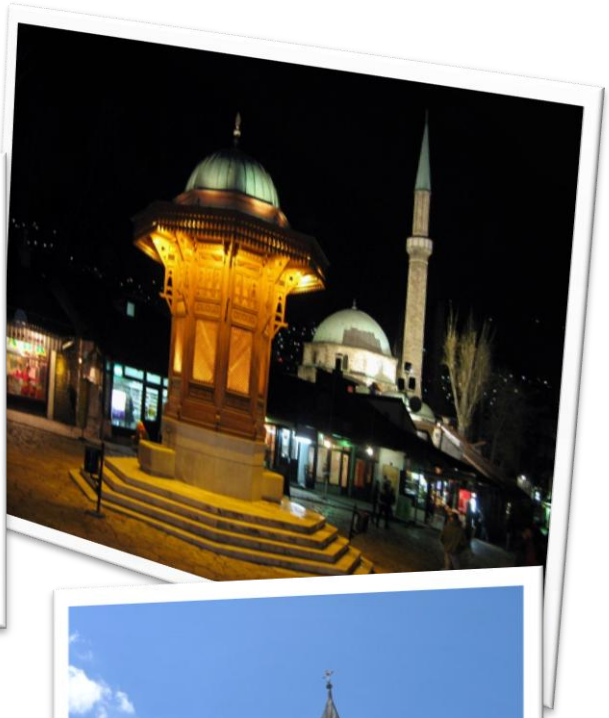

www.bclf2015.org

AUSPICES

23rd Meeting of the Balkan Clinical Laboratory Federation
2nd National Congress of the Association of Medical Biochemist in Bosnia and Herzegovina

is organized under the Auspices of the

International Federation of Clinical Chemistry and Laboratory Medicine (IFCC)
European Federation of Clinical Chemistry and Laboratory Medicine (EFLM)
Balkan Clinical Laboratory Federation (BCLF)
Local authorities

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Association of Medical Biochemist in Bosnia and Herzegovina

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23rd MEETING OF THE BALKAN CLINICAL LABORATORY FEDERATION

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LECTURE PRESENTATIONS

DEFINING PERFORMANCE SPECIFICATIONS IN LABORATORY TESTING

Mauro Panteghini

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Defining analytical performance specifications for each analyte is essential to make its determination clinically usable and to ensure that the measurement error does not prevail on the result. These performance goals should be established by the laboratory profession according to recognized and widely accepted scientific models. Particularly, the hierarchy of sources for deriving analytical goals of a laboratory measurement has been recently updated in a conference held in Milan on November 2014. Although the essence of the hierarchy originally established in 1999 in Stockholm was supported, new perspectives have been forwarded prompting simplification and explanatory additions. Basically, the recommended approaches for defining analytical performance specifications should rely on the effect of analytical performance on clinical outcomes or on the biological variation of the measurand. The attention is primarily directed towards the measurand and its biological and clinical characteristics, some models being therefore better suited for certain measurands than for others. For instance, the model based on biological variation is probably not appropriate for analytes showing high individuality: for those analytes, outcome-based data or, in their absence, the state of the art of measurement quality are the models to derive analytical specifications. Proposed analytical performance specifications should therefore always be accompanied by a statement of the rationale, the source and the quality of the evidence behind the recommendation.

CANCER EPIGENETICS IN THE CLINICAL PRACTICE

Angeliki Magklara,

Department of Medicine, School of Health Sciences, University of Ioannina, Greece

There has been an explosion of knowledge in the field of Epigenetics in the last two decades. We have witnessed the emergence of a new biological code, the "epigenetic code", as an equally important determining factor of phenotypic variation in health and disease. With the development and application of new powerful technologies the field of epigenomics has revealed distinct epigenetic profiles in different cell types, as well as, numerous epigenetic aberrations in a growing number of human disorders including cancer. The progressive elucidation of the "cancer epigenome" and the mechanisms that govern it has started revealing the complex interactions between epigenetic and genetic changes and how they contribute to the initiation and progression of cancer. Of great consequence is the fact that the epigenetic marks are reversible, which makes them the perfect targets for the development of therapeutic schemes that aim to re-establish the normal epigenetic landscape and opens up new promising possibilities for the fight against cancer. As a result, the first epigenetic therapies for cancer treatment have arrived in the clinic and many more are, currently, in clinical trials. In parallel, the newly gained knowledge on cancer epigenomes has promoted the development of new potential cancer biomarker strategies and has supported a more efficient cancer subtyping. Examples of such recent advances in the use of cancer epigenetics in the clinical practice will be discussed.

MASS SPECTROMETRY TECHNIQUES IN LABORATORY PRACTICE

Ali Ünlü

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Clinical chemistry laboratories routinly use molecular or atomic absorbtion spectrometry, electrochemical methods such as potentiometry, amperometry, immunassay methods, immunnephelometry and separation techniques such as electrophoresis and chromatography.

Increase in the borderline cases due to early diagnostic techniques and development of the health care systems require more sensitive, specific and reliable techniques than routinly used methods. Development of the mass spectrometry (MS) techniques became an useful tool for the measuring of analytes. Mass spectrometry integrated with liquid (LC/MS) or gas chromatography (GC/MS) provides higher sensitivity and specifity. Because of having unequalled sensitivity, lower detection limits and diversity of its applications mass spectrometry has an outstanding position among analytical methods.

MS methods are commonly used in endocrinology, clinical and forensic toxicology, inborn error of metabolism, therapeutic drug monitoring and emerging clinical biomarkers. Steroids measurements are one of the main focus in MS laboratories in Endocrinology section. MS analysis recommended hormones are; estradiol in male, prepubertal ages and postmenapausal term, free testosterone, aldosterone, 17-OH progesterone, Deoksicorticosterone, 25-OH vitamin D_2 ve D_3, 25-OH vitamin D_3/3-epi-25-OH vitamin D_3, 1,25-dihydroxy vitamin D_3 ve 24,25 dihydroxy vitamin D_3. MS analysis is also found to be superior against immunassay in the measurement of free tyroxine and tyriodotronine levels.
The chemical structures of therapeutic drugs metabolites are quite similar to the parent drug, therefore, it is difficult if not impossible to construct an immunoassay that recognizes the parent compound without some degree of cross-reactivity towards one or more of the metabolites. MS based assays have been developed for immunosuppressants and are widely used in clinical practice.

MS has been used for clinical and forensic toxicology for two main reasons. Immunoassays are platforms used as a screening test because they provide faster results and tests are commercially available. Due to the specificity limitations of immunoassays, MS is used to confirm false positive results from the screening immunoassay systems. As such, these MS assays are designed to find particular drugs or their metabolites. MS analysis is also used for comprehensive drug screening.

For the use of inborn errors of metabolism, analyzing for amino, organic, and fatty acids has undergone a series of developments to the technology. LC–MS/MS is now recognized as one of the most definitive analysis procedures for measuring these analytes. LC–MS/MS system is capable of measuring all of the analytes within a group in a single run. So MS is called a "multiplex" testing.

Popular analytes that may have clinical importance are being discovered each year by proteomic and metabolic research. LC–MS/MS is a viable option for the measurement of clinically relevant analytes, particularly if they are within the molecular weight range that is suitable for MS analysis as methylarginines.
Development of the MS and chromatographic techniques have lead to great success to quantification and characterisation of proteins. This achivement has played a key role in the birth of the field of proteomics. Metabolomics is also, outgrowths of research and developments in MS. A draft of human proteomics was published in Nature in 2014. The diagnosis of infectious pathogens presents the range of application of mass spectrometry and its growing potential to contribute to clinical diagnostics.

Low solvent volumes, high throughput, providing clinically stable results with deuterated internal standards, minimizing the specifity problems, high analytical

range, improved sensitivity, multiplex testing in a single run are the most important advantages of MS systems.

Requirement of experience for method development and procedures, time consuming application and method validation progress, long turn around times due to preanalitycal steps, difference in calibrator and methods, lack of standardisation of solvents and stability issues are the main disadvantages.

The cost of analysis is also of critical importance, which is closely related to the number of samples analyzed. Development of analytical techniques is always expensive, time-consuming and needs expertise. However MS enstruments are powerfull tools and can be cost-effective after 1-2 years in clinical laboratories. The cost of test is lower than other methods for high throughput experiments.

THE SERUM VALUE OF URIC ACID IN PATIENTS WITH CEREBROVASCULAR DISEASE (ISCHEMIC STROKE AND VASCULAR DEMENTIA)

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Background: In our study we have investigated uric acid in 150 patients diagnosed with the first ischemic brain stroke, where blood samples were taken during the acute phase and post acute phase, 50 patients diagnosed with vascular dementia developed as a consequence of ischemic brain stroke, i.e. of many small ischemic focus of various age and 50 subjects at healthy group
Methods: Uric acid was determined by DIMENSION LxR automatic analyser of DADE BEHRING Company, using enzymatic method. Collected data were statistically analyzed using programs SPSS version 18.0 and Microsoft Office Excel 2012.
Results: The patients with ischemic stroke had hyperuricemia in 30 % and vascular dementia in 8%. Our results showed that uric acid increased to 7 days after ischemic stroke by 5.3%, after 14 days of 9.5%. Using the Wilcoxon signed ranks test (Z = -2.736, p=0.006) was found statistically significant difference between the average concentration of uric acid after 24-48 hours and 14 days of ischemic stroke. Using the Mann-Whitney test (Z = -1.837, p=0.066) it was not a significant difference between concentrations of uric acid in the acute phase (24-48 hours of ischemic stroke) and control groups. According to the same test (Z = -2.837, p=0.005; Z = -2.734, p=0.006) it was a significant difference between concentrations of uric acid after 7 and 14 days of ischemic stroke and control groups. Using the same tests, no significant difference between the average concentrations of uric acid in acute (Z = -0.458, p=0.647; Z= -0.614; p=0.539) and post-acute phase (Z = -0.700, p=0.484) of ischemic stroke and vascular dementia groups with the significance level of $p < 0.05$. Using the Mann-Whitney test (Z=-2.241, p= 0.025) was significant difference between the average concentrations of uric acid in vascular dementia and control group with the significance level of $p < 0.05$.
Conclusions: Uric acid concentration is higher in the group with ischemic brain stroke and vascular dementia than in control group. It is possible that increases uric acid reflect renovascular atherosclerosis and tissue hypoxia. Therefore monitoring of uric acid at patients with ischemic stroke is important because uric acid is more harmful than protective.

BIOCHEMICAL METHODS IN DIAGNOSE AND MANAGEMENT OF DIABETES TYPE 1 IN KOSOVO

Dr. Afrim Kotori,

Kosovo

Aim of the presentation was to present the diagnostic approach related to Diabetes type 1 in children and adolescents in countries in development such as Kosovo.

Introduction: The incidence of type 1 Diabetes is increasing worldwide and in Kosovo too. For that reason it was a need to be rationale in diagnostic methods. First objective was to make a proper diagnose in time to prevent DKA (diabetic ketoacidosis) in new cases with diabetes and second to ensure appropriate testing to prevent complications of diabetes in youth.

Methods: I In all cases with diabetes in children and adolescents it was done testing of: acid base state (pH, bicarbonates), glycaemia, electrolytes, HbA1c, C peptide, insulin, antibodies-ICA(Insulin cell antibodies), GADA (glutei acid decarboxylase antibodies) and IA2 , fT4, TSH, antiTPO, tTg IgA/IgG, DGP IgA/IgG, IgA total and genetic testing.

Results: From y. 2009 up to 2014 they were 196 new cases of diabetes in youth in Kosovo. In the state of Diabetic ketoacidosis they were 118 new cases of diabetes type 1 and from them 75 with severe DKA. 2 children were diagnosed with MODY diabetes and 2 adolescents with Diabetes type 2. 4 cases with neonatal diabetes and three of them were diagnosed in the state of DKA.
From 196 new cases of diabetes type 1, 22 children were diagnosed with celiac disease, 12 females and 10 males and 19 children with autoimmune thyroiditis, 11 females and 8 males.

Conclusions: In Kosovo the incidence of diabetes type 1 is increasing and in the last five years we have three times more cases with diabetes type 1. These finding gave us as a Clinical professionist to be more active in diagnostic methods and follow up of patients with Diabetes in countries in development.

ANALYTICAL CHALLENGES IN THE IDENTIFICATION OF „NEW PSYCHOACTIVE SUBSTANCES"

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University clinical centre Maribor, Clinical Chemistry department, Maribor, Slovenia

The role of laboratory, and specially the role of clinical chemist, in detection of disease based on action of toxins (intoxication), is not well defined. The clinical staff usually shares the opinion that the laboratory analytics is performed only to confirm their judgment and to fill the patient's administrative documentation, but has no influence on the course of the treatment. An additional issue is the lasting education and the lack of properly educated personnel for toxicological analytics. This situation is being changed because the techniques and knowledge in (clinical) laboratories has developed.
Over the past five years there has been an enormous increase in the number, type and availability of new psychoactive substances (NPS) in Europe. *An NPS is defined as 'a new narcotic or psychotropic drug, in pure form or in preparation, that is not controlled by the United Nations drug conventions, but which may pose a public health threat comparable to that posed by substances listed in these conventions'.* During 2014, 101 new psychoactive substances were reported to the EU Early Warning

System for the first time. This brings the total number of substances being monitored by the European Monitoring Centre for Drugs and Drug Addiction (EMCDDA) to more than 450.This increasing prevalence remains a problem worldwide. NPS may have a depressant, stimulant and/or hallucinogenic effect and tend to mimic existing, popular illicit drugs, such as cannabis and MDMA. NPS, with their continuous widespread availability and unpredictable effects, especially when used in combination with prescribed drugs, make them an important public health problem.

Screening procedures in toxicology represent a set of tests that are focused, performed quickly and easily. The basic aim is to obtain useful information in the shortest time. Because screening procedures are oriented to well-defined substances, their use in detection of NPS is negligible. More sophisticated approach (chromatography) is therefore needed to detect and identify these novel products. Such methods represent highly demanding techniques on very expensive analytical tools. They have their drawbacks, which are mainly reflected in lower robustness, requiring highly trained personnel and are not automated.

Toxicological analysis is an indispensable tool in the identification of this novel drugs, as patients commonly present with nonspecific signs and symptoms when intoxicated.

FACTOR ANALYSIS AND ASSOCIATION OF LIPID, INFLAMMATORY, CARDIAC AND RENAL BIOMARKERS WITH C-REACTIVE PROTEIN IN CARDIOVASCULAR RISK CATEGORIZATION

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Background: Atherosclerosis is a disease conditioned with multiple factors followed by chronic low-grade inflammation and dyslipidemia. Thanks to substantial evidence that C-reactive protein (CRP) strongly and independently predicts cardiovascular complications, the use of CRP in clinical practice is recommended by several institutions for risk assessment in primary prevention. Also, there is evidence of other factors, contributing to and maintaining the intensity of atherosclerotic processes, which might identify cardiovascular risk contribution not originated from traditional risk factors. The aim of this study was to examine, using factor analysis, the nature of influence of biomarkers of inflammation, lipid metabolism, renal, and cardiac function on cardiovascular risk, and their possible connection and relations to CRP values.

Methods: To examine and analyze clustering of inflammatory markers [serum amyloid A (SAA), fibrinogen, α_1-acid glycoprotein (A1AGP), haptoglobin, C3 and C4 complement components], markers of lipid metabolism [total, HDL, non-HDL and LDL cholesterol, triglycerides, apolipoprotein A-I (apo A-I), apolipoprotein B (apo B), lipoprotein (a) (Lp(a))], renal [creatinine, cystatin C (Cys-C), estimated glomerular filtration rate (eGFR)], and cardiac function [N-terminal pro-natriuretic peptide type B (NT-proBNP), high sensitivity cardiac troponin T (hs-cTnT)], as well as traditional cardiovascular risk factors [age, gender, body mass index (BMI), systolic blood pressure (SBP)], principal component analysis was used.

Results: Factor analysis identified five clusters, i.e. principal components (factors), which explained 65.3% of the

total variance (29.0% factor 1, 13.2% factor 2, 9.0% factor 3, 8.5% factor 4, and 5.6% factor 5). Based on factor loading of ≥ 0.5 clasters were interpreted as 1) „systemic inflammation" (fibrinogen, SAA, A1AGP, haptoglobin, C3 and C4 complement components); 2) „cardiorenal factor" (creatinine, uric acid, Cys-C, hs-cTnT and gender); 3) „atherogenic cholesterol" (LDL and non-HDL cholesterol); 4) „hemodynamic factor" (age and NT-proBNP); and 5) „metabolic factor" (triglycerides and HDL cholesterol). The Kaiser-Meyer-Olkin measure of sampling adequacy was 0.75. In multiple regression analysis, five factor model had the best predictive value for CRP concentrations >1 mg/L (OR 6.53, 95% CI 4.06–10.50, *P*<0.0001 for "systemic inflammation"; OR 1.44, 95% CI 1.04–2.00, *P*=0.028 for „cardiorenal factor"; OR 1.76, 95% CI 1.23–2.5, *P*=0.002 for „atherogenic cholesterol"; OR 1.91, 95% CI 1.33 – 2.73, *P*<0.0001 for „hemodynamic factor";OR 1.90, 95% CI 1.33 – 2.73, *P*<0.0001 for „metabolic factor"), while „cardiorenal factor" and „atherogenic cholesterol" completely lost their significance (*P*>0.05) for predicting CRP concentrations >2 mg/L and >3 mg/L. The ability of the factor-based logistic regression model was compared with multivariable logistic model containing all 25 variables in predicting the presence of CRP concentrations >1 mg/L, >2 mg/L, and >3 mg/L. The area under the receiver operator characteristics curve (AUC) of the five factor model was 0.889 and was not statistically significantly different from the 25 variable model (AUC=0.922) (P=0.2113). However, the differences between the two models examined were statistically significant in predicting the values of CRP>2 mg/L and CRP>3 mg/L.

Conclusion: Systemic inflammation, cardiorenal function, atherogenic lipid profile, hemodynamic and metabolic status might independently contribute to the pathophysiology of chronic, subclinical inflammation in atherosclerosis. They might represent underlying dimensions accompanying the elevation of CRP concentration and increased cardiovascular risk.

REVESE CHOLESTEROL TRANSPORT: THE ROLE OF HIGH DENSITY LIPOPROTEIN

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Revese cholesterol transport (RCT) is a multi-step process resulting in the net movement of cholesterol from peripheral tissues back to the liver via the plasma.
The cardiorotective role of HDL is thought to be related at least in part to the role of HDL in RCT. In addition, HDL has important anti-oxidative and anti-inflammatory properties and may prevent adhesion of monocyes to the endothelial cells.

HDL comprise a heterogeneous class of lipoproteins.
HDL can be subdivided according density in HDL2, the less dense subfraction and HDL3.

HDL formation is regarded to begin with pre β-HDL. As the particles mature they become spherical with a α-electrophoretic mobility with HDL2 originating from HDL3.

HDL particles are constantly remodeled as a result of many interrelated processes. Several enzymes including lipoprotein lipase (LPL), hepatic lipase (HL), endothelial lipase (EL) and lecithin cholesterol acyltransferase (LCAT), as well as lipoprotein transfer proteins including cholesteryl ester transfer protein (CETP) and phospholipid transfer protein (PLTP) are involved in HDL metabolism.

LPL hydrolyses triglycerides contained within lipoproteins. By hydrolyzing HDL triglycerides as well as HDL phospholipids, HL contributes to the metabolism of HDL.

EL shows primarly phospholipase activity and relatively little triglyceride lipase activity.

LCAT catalyzes the transfer of 2-acyl groups from lecithin or phosphatidylethanolamine to free (unesterified) cholesterol. The LCAT reaction results in the generation of cholesteryl ester molecules that by their hydrophobic nature are retained in the core of HDL particles.

CETP enables the mass transfer and exchange of cholesteryl ester and triglyceride molecules among lipoproteins.

PLTP facilitates phospholipid and free cholesterol transfer between lipoproteins during lipolysis. Moreover, PLTP is able to convert HDL in larger and smaller particles.

There are abnormalities and alterations in HDL remodeling in relation to lipases, LCAT and
CETP –mediated process of neutral lipid transfer in insulin resistance and type 2 diabetes mellitus.

THE GLOBAL DIABETES EPIDEMIC AND ROLE OF GLYCATED HAEMOGLOBIN

Lennartz Lieselotte

Germany

Currently there are more than 346 million people worldwide living with diabetes, and World Health Organization (WHO) expects this to double by 2030. Unlike type 1 diabetes, an autoimmune disease with sudden stop of insulin production, onset of type 2 diabetes, which makes up about 90 percent of cases worldwide, is directly related to lifestyle and linked to obesity.

Monitoring of glucose only provides a snapshot overview about the patient's glycemic status and the golden standard for monitoring long-term glycemic control of diabetic patients is the measurement of glycated haemoglobin (HbA1c).

Known complications of uncontrolled diabetes can be reduced by an early start of optimized glucose control using HbA1c to guide therapy.
HbA1c is now also in use as early marker for diagnosing diabetes together with fasting plasma glucose (FPG) levels and/or an oral glucose tolerance test (OGTT).
HbA1c testing for diagnosing diabetes mellitus offers several advantages over the traditionally used testing methods including standardization from a clinical and metrological point of view, the better reflection of long term glycemic status, the less biological variation and the robustness to pre-analytical factors

WHETHER MMP-9 MAY BE AN EARLY BIOMARKER FOR ACUTE CORONARY SYNDROME

Emina Čolak

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Background: Matrix metalloproteinases (MMPs) are a family of Zn^{2+}-dependent endopeptidases capable of cleaving components of extracellular matrix. Several lines of evidence indicate that MMPs influence the process of atherosclerotic lesion formation and degradation of vulnerable atherosclerotic plaque. Such markers for plaque instability or rupture would establish the diagnosis of ACS at the earliest stage and may predict the onset.The aim of this study was to determinate and analyze the activity of total and active form of MMP-9 in patients with acute coronary syndrome in order to determinate the possibility of using these parameters as predictors for future coronary hart events.

Material and methods: A total of 108 subjects were included in this study, aged of 45 to 65 yr.78 subjects were having acute myocardial infarction (37 patients diagnosed as type 2 diabetes mellitus, and 41 as non-diabetics with AMI), and 30 healthy subjects (CG).

Results: Statistical processing data revealed significantly higher activities of total and active MMP-9 in AMI patients compared to the controles (p=0.016 and p=0.001), especially in diabetics patients compared to non diabetic patients with AMI (p=0.006; p<0.001). ROC curve showed high sensitivity (SE) and specificity (SP) of used diagnostic test (T.MMP9: SE=94.5% and SP=45.5%; A.MMP9: SE=83.3% and SP=81.8%). The estimated cut-off values were 27.6 ng/mL for T.MMP-9 and 23.8 ng/mL for A.MMP-9. The Multiple Regression analysis model weighted by A.MMP-9 lower and higher than 23.8 ng/mL, indicated that both parameters were significant predictors of AMI (β_{AMMP9}=0.783;t=8.386; p<0.0001; β_{TMMP9}=0.175;t=2.53,p=0.016).

Conclusion: Based on the obtained results it can be concluded that both parameters could be used as predictors of the early onset of acute myocardial infarction, especially in those subjects under higher risk for AMI such as diabetes mellitus type 2.

CARDIOVASCULAR RISK IN PATIENT ON HEMODIALYSIS: NOVEL PREDICTORS AS WELL AS NEW LOOK ON SOME OLD MARKERS

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Cardiac troponin is indicator of cardiomyocyte damage in various clinical settings. Chronic renal failure (CRF) is characterized by changes in lipid status, nutritional status, oxidative stress, antioxidant protection, inflammation parameters and also by increased basal levels of cardiac troponin regardless the presence of acute coronary syndrome.In this study, we investigated whether levels of hsTnI, markers of inflammation, nutrition and oxidative stress and lipid markers are predictive of death.

We were monitoring 62 patients (30 males and 32 females) who were on regular hemodialysis treatment more

than 3 months. hsTnI was measured on Architect i2000, Abbott, USA; prealbumin, retinol-binding protein (RBP) and hsCRP on BNII nephelometer; Siemens, Germany; albumin, cholesterol, triglycerides, HDL-cholesterol and LDL-cholesterol were measured on Cobas c6000, Roche, Germany; nonHDL-cholesterol was calculated, prooxidant-antioxidant balance (PAB), total antioxidant status (TAS) and superoxide dismutase in erythrocytes (SOD) were determined by spectrophotometry methods and resistin by ELISA. We observed these parameters in hemodyalisis patients who survived and those who died 15 months after sampling. To examine the effect of all parameters on survival we used a logistic regression and Kaplan-Meier survival curves.

Our data, analyzed by logistic regression, suggested that hsTnI, albumin, prealbumin, RBP, cholesterol, LDL-cholesterol, nonHDL-cholesterol, hsCRP, PAB ($p<0.05$) and TAS ($p<0.1$) were predictors of mortality in this group of patients. Triglycerides, HDL-cholesterol, SOD and resistin were not significant predictors of mortality. The strongest predictors of mortality were prealbumin ($p=0.001$), RBP ($p=0.003$) and hsCRP ($p=0.009$). Based on our data we concluded that parameters of nutritional, lipid status (cholesterol, LDL-cholesterol, nonHDL-cholesterol) and TAS were lower in the patients who died and PAB, hsCRP and hsTnI were higher. We analyzed Kaplan-Meier survival curves for tertiles of prealbumin and hsCRP and quartiles of hsTnI and PAB and these results showed the greatest mortality risk in the lowest tertile of prealbumin (≤0.269 g/L; $p<0.05$), highest tertile of hsCRP (≥7.07 mg/L; $p<0.001$), highest quartile of hsTnI (≥39.7 ng/L, $p<0.05$) and highest PAB quartile (≥38.2 HKU, $p<0.05$). We also analyzed our data using Spearman correlation coefficient. These results indicated that there is a significant linear relationship between hsTnI and cholesterol, HDL-cholesterol, LDL-cholesterol ($p<0.05$), prealbumin, RBP, hsCRP and PAB ($p<0.001$).

Based on these results, we can conclude that the patients who died were in a state of more intense inflammation and oxidative stress compared to surviving patients and their nutritional status was worse. hsTnI in these study was good predictor of overall mortality. The question is if active intervention for patient identified to be at high risk with this biomarker offers any benefit for long-term survival.

MARKERS OF INFLAMMATION AND ANTIOXIDANT ENZYME ACTIVITIES IN RESTENOSIS FOLLOWING PERCUTANEOUS CORONARY INTERVENTION

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Background: Angiographic benefit of percutaneous coronary intervention (PCI) is often compromised by the need for repeated revascularization, due to the development of in-stent restenosis. Numerous studies have tried to establish the predictive value of different biochemical markers of restenosis, but the results were often controversial.

The aim of this study was to assess the prognostic significance of antioxidant, lipid markers and markers of inflammation in the development of in-stent restenosis(ISR) afterPCI.

Material and methodes: Serum, high sensitivity CRP (hs-CRP), soluble intracellular adhesion molecule-1 (ICAM-1), transforming growth factor-beta (TGF-β), oxidized low-density lipoprotein (oxLDL), ceruloplasmin levels and serum catalase(CAT) activity were determined in 44 patients before stent implantation

procedure as well as 6 months after the monitoringof PCI.

The results of the follow-up, they found that patients who developed angiographic ISR,increase the level of hs-CRP was significantly higher than inpatients without stenosis. Stent implantation induces a compensatory increase in serum antioxidant protection during follow-up with a much lower CAT activity in patients with ISR,which probably contributes to its development. No significant changes in the circulation at the level of ICAM-1, TGF-β, oxLDL and ceruloplasmin were observed between the groups.
In conclusion, serum hs-CRP andCAT activity may be considered as useful predictive biomarkers for monitoring patients during follow-up after stent implantation.

TROPONIN – HOW LABORATORY FINDINGS CAN HELP US IN THE TREATMENT OF OUR PATIENTS

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Three types of troponins exist – troponin I, T and C. Raised troponin levels are prognostically important in many of the conditions in which they are used for diagnosis. Causes of its elevation other than in miocardial infarction included many conditions such as cardiac surgery, myocarditis, cardiac contusion/trauma, endocarditis, chronic severe heart failure, cardiac arrhythmias (tachy/bradyarrhythmias, heart blocks), pulmonary embolism, stroke, CPR, sepsis, renal failure, burns, medication and toxins, critical illness, etc. Troponin elevations are quite common in intensive care unit patients such as the one with diagnosis mentioned above.
Certain subtypes of troponin, such as cardiac I and T, are very sensitive and specific indicators of damage to the heart muscle, or can be used to monitor drug and toxin-induced cardiomyocite toxicity. Elevated troponin levels should always be evaluated in a clinical context. The absolute abnormal values varies depending on the clinical settings in which the patient is evaluated and the assay used. In the cardiac surgery, troponin should be accompanied by other cardiac markers (CK, CK-MB, myoglobin, hs-CRP, BNP). Using high-sensitive troponin tests (hs-cTn) we are able to detect sooner acute coronary syndrome, and to adjust proper therapy.
So, troponin can be use for diagnostic procedures, for therapy protocol and to predict treatment outcome. This test is already routinely used at our Clinic.

IMMUNOLOGICAL PARAMETERS IN DIAGNOSTICS OF MULTIPLE SCLEROSIS

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Background: Multiple sclerosis is a chronic inflammatory, progressive, multifocal, demyelinating autoimmune disease of the central nervous system, which manifests itself in different symptoms of the nervous system. Symptoms can occur in attacks (85-90%) or slowly progressive over time.

The cause and pathogenesis of multiple sclerosis are not known.

Matherial and methods: On Clinical Center University of Sarajevo Department of Clinical Immunology we analyzed 255 samples of CSF and serum samples which were sent from the Department of Neurology Clinical Center University of Sarajevo. The objectives were to determine the presence of specific immunological parametres such as oligoclonal IgG bands in CSF and serum, the values of albumin and immunoglobulin IgG in CSF and serum, to determine quotient (CSF / serum) of albumin and immunoglobulin IgG, include the value of the quotient of albumin and immunoglobulin IgG in Reibergram, Isoelectric focusing method uses to determine the presence of IgG oligoclonal bands. We analised the correlation between intrathecal synthesis of IgG obtained using Reibergram and the presence of IgG oligoclonal bands on agarose gel. We used nephelomety method to determine albumin and immunoglobulin values.

Results: From a total of 254 analyzed samples of serum and cerebrospinal fluid of patients, 179 had a negative intrathecal IgG synthesis by Reibergram, while 75 had a positive intrathecal IgG synthesis. Among all analised samples 174 was negative on gel, we did not observe the presence of IgG oligoclonal bands of type 2 or type 3, and 80 samples of CSF and serum of patients showed oligoclonal bands on the gel type 2 or type 3. Type 4 is detected in 2 patients, and type 5 in one patient (patient was sent from infectious clinic to diagnose monoclonal gammapathy). We analyzed the quotients values of Qalb and QIgG in all samples compared to patients who had oligoclonal IgG bands present on the gel. We came to the conclusion that values are largely similar, with slight variations that were not statistically significant.

Conclusion: Due to the high sensitivity of oligoclonal bands in CSF and high specificity with the appropriate clinical conditions, testing of oligoclonal bands in the presence of CSF IgG is extensively recommended to support the diagnosis of multiple sclerosis.

GLUCOSE-6-PHOSPHATE DEHYDROGENASE DEFICIENCY AND ALZHEIMER`S DISEASE: PARTNERS IN CRIME?

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Alzheimer's disease is a multifaceted brain disorder which involves various coupled irreversible, progressive biochemical reactions that significantly reduce quality of life as well as the actual life expectancy. Aging, genetic predispositions, head trauma, diabetes, cardiovascular disease, deficiencies in insulin signaling, dysfunction of mitochondria-associated membranes, cerebrovascular changes, high cholesterol level, increased oxidative stress and free radical formation, DNA damage, disturbed energy metabolism, and synaptic dysfunction, high blood pressure, obesity, dietary habits, exercise, social engagement, and mental stress are noted among the risk factors of this disease.

In this review I would like to draw the attention on glucose-6-phosphate dehydrogenase deficiency and its relationship with Alzheimer`s disease. This enzymopathy is the most common human congenital defect of metabolism. Glucose-6-phosphate dehydrogenase deficiency is defined by decrease in NADPH+H+ and reduced form of glutathione concentration and that might in turn, amplify oxidative stress due to

essentiality of the enzyme. This most common enzymopathy may manifest itself in severe forms, however most of the individuals with this deficiency are not essentially symptomatic. To understand the sporadic Alzheimer's disease, the writer of this paper thinks that, looking into a crystal ball might not yield much of a benefit but G6PD deficiency could effortlessly give some clues.

PREVENTION OF HEMOGLOBINOPATHIES IN TURKEY

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Hemoglobinopathies are the most common genetic disorders in Turkey. The incidence of beta thalassemia and sickle cell trait (HbAS) is 2.0% and 0.3% respectively. In addition to HbS, 51 abnormal hemoglobins and 42 different beta thalassemia mutations have been detected by DNA analysis. In Turkey, beta thalassemia and sickle cell anemia cause major health problems. For thirty years, screening programs for carriers, genetic counseling and prenatal diagnosis have sought to prevent hemoglobinopathies.

In 1983, the first prenatal diagnosis center was established for sickle cell anemia and beta thalassemia at Hacettepe University, Ankara. After many population-screening studies, a law was passed in 1993 by the Turkish Parliament for the eradication of hemoglobinopathies. Forty-one premarital screening centers were set up by the Ministry of Health in the 33 provinces where most of the transfusion-dependent thalassemic patients live. The mothers at risk for hemoglobinopathies were given genetic counseling and directed to prenatal diagnosis centers. Since 1990, four prenatal diagnosis centers have been established at university hospitals in Adana, Antalya, İstanbul and İzmir.

A total of 5255 prenatal diagnoses have been made for sickle cell anemia and beta thalassemia in 5 centers; 1338 fetuses have been diagnosed as homozygous or compound heterozygotes for hemoglobinopathies. Prenatal diagnosis was performed on families who had decided to terminate the pregnancy if it were to be found that the fetus was affected.

CELL-FREE MRNA IN PLASMA AS A NOVEL RESEARCH TOOL IN LABORATORY MEDICINE - OUR EXPERIENCE

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Extracellular nucleic acids, discovered by Mandela and Metais in 1947, are found in plasma, serum, as well as in other biological samples outside the cell. Concentration of the cell-free nucleic acids in body fluids is a result of the balance mechanisms of release from cells (necrosis, apoptosis, lysis, or not yet sufficiently elucidated mechanism of active secretion) and removal mechanisms (removal of the reticuloendothelial system cells in macrophages, degradation by endonucleases). In the plasma of healthy

subjects the level of RNA concentration is up to 250 mg/L, while in unwell patients RNA level can exceed 3000 mg / L. Nowadays, especially in oncology, for early detection and diagnosis of various cancers, the analysis of mRNA in plasma has shown useful. Quantitative analysis of cell- free mRNA of the gene β-globin in plasma of patients with acute coronary syndrome in the field of coronary atherosclerosis has a prognostic value. Analysis of cardiac-specific miRNA in plasma, i.e. miR-208a, was proposed as a biomarker of cardiomyocyte damage, with a potential for early diagnosis of ACS . In plasma of healthy subjects there was no miR-208a, but it was detected in all ACS patients within 1 to 4 hours of the onset of chest pain, exhibiting higher sensitivity than troponin I.

Our experience: Plasma mRNA analysis is a novel, intriguing field for the non-invasive *in vivo* assessment and monitoring of pathologic processes in their tissues of origin. The analytical approach represents an alternative or supplement to protein analysis. Using fully optimised analytical procedures, it is possible to measure the level of plasma mRNA from genes which play an important role in development and destabilisation of atherosclerotic plaque with a between-run imprecision expressed as the coefficient of variation of threshold cycle of less than 2% for the whole analytical procedure from isolation to quantification. As published previously by Cerne D *et al.*, in plasma of patients with coronary atherosclerosis, as well as in healthy volunteers, it is possible to quantify mRNA of genes encoding cathepsin S (CTSS), cathepsin B (CTSB), CD40, chemokine (C-C motif) ligand 2 (*CCL2*, encoding monocyte chemoattractant protein-1, MCP-1), death- associated protein kinase 1 (DAPK1), matrix metallopeptidase 9 (MMP9) and vascular cell adhesion molecule-1 (VCAM1) in plasma.
Likewise, plasma mRNA analysis, as a novel research tool for non-invasive assessment of gene expression profiles in vascular beds, may be used to investigate statin pleiotropy *in vivo*. In the study Mirjanic-Azaric *et al.,* in patients with stable angina pectoris atorvastatin (20 mg/day, 10 weeks) consistently diminished plasma mRNA levels from statin pleiotropy-target genes *CCL2* and intercellular adhesion molecule-1 (*ICAM1)*, but did not change heme oxygenase-1 (*HMOX1)* and *CTSS* expression. Plasma mRNA analysis as a supplement to protein analysis showed association of genes with their proteins, and their protein activity products and their sensitivity to atorvastatin.
The mRNA analysis shows enormous potential in diagnostics as well as in the monitoring of therapy in the field of atherosclerosis. mRNA analysis is an important research tool that offers an interesting scientific challenge.

POCT - HOW TO IMPLEMENT AND MAINTAIN QUALITY CONTROL

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Point of care testing or in short POCT (allso called bedside testing, near patient testing) is defined as any *in vitro* diagnostic procedure done outside the dedicated laboratory environment.. As it is always done near the patient, it has become an additional job for clinical staff, mainly nurses. Since POCT represents the fastest growing segment of in vitro laboratory diagnostics worldwide, the maintanance of quality standards remains an ongoing challenge for laboratory professionals. By its nature, POCT falls within the interest and responsibility scope of the laboratory personnel, however by its location and means of use it belongs to other areas of patient care. Only the successful blending of these two determinant factors ensures adequate testing quality

and results reliability. Within hospital environment (which represents about 70% of POCT market) it has become an indispensable part of any clinical situation that requires immediate access to results, whether for medical or logistic reasons. It is of utter importance that laboratory staff remains recognized and involved in decision making about hospital POCT at all stages. The optimal approach towards this goal is establishing a hospital POCT committee which should consist of all interested parties' representatives: physicians who need the result, nurses who will do the testing, laboratory professionals who possess the required knowledge about technologies and operating procedures and last but not least, hospital administration who has to cover the costs. In terms of financial issues, it is highly advisable that cost coverage is clarified before any equipment is obtained – in other words, who will be covering the cost of equipment and/or consumables – under most circumstances, the financial responsibility should not be taken by the laboratory but by the hospital department that needs POCT services. POCT instruments and equipment should be carefully chosen only after the committee has reached a consensus regarding specific clinical needs and available means of fulfilling them. The direct vending approach, whereby IVD industry representatives offer their POCT product directly to clinical staff should be strongly discouraged or if possible explicitly forbidden. When a particular POCT equipment has been chosen through the POCT committee, the laboratory should initially verify its performance and then organize education and training on site. If a parallel central laboratory procedure exists, the results comparability has to be checked and communicated to the end users. Clinical staff needs to understand the possible consequences of inadequate sample handling, as well as main interferences possibilities and other existing constraints (9). User identification should be introduced as an indispensable part of the training, since clinical staff has to stay aware of the fact that laboratory people will be responsible for the functionality of the instrument, while they will be held accountable for each particular patient result. Regarding instrument functionality, it can stay implemented usually mainly through continuous endeavours by laboratory POCT dedicated staff to maintain the same quality standards applied within the central laboratory – internal and external quality control, remote review of test and instrument data, supervising the regularity of cleaning and maintenance procedures together with constant education and reeducation of clinical personnel. From this emerges the last but far from least general rule of good POCT hospital quality network or any other kind of POCT testing: it is continuous communication between all parties involved (laboratory and clinics) who are interested in obtaining true and reliable results.

METABOLIC SYNDROME IN PATIENTS WITH MYOTONIC DYSTROPHIES

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Introduction: Metabolic syndrome (MetS) is a set of metabolic and hemodynamic impairment that multiplies the risk of atherosclerotic disease and

diabetes mellitus type 2. There are no data of the frequency of MetS in patients with myotonic dystrophies (DM).

Objective: To determine frequency and importance of MetS in patients with DM type 1 and 2 (DM1 and DM2).

Method: The study included 66 DM1 patients (50% men, age 41.9 ± 10.5, disease duration 19.3 ± 8.6 years) and 47 DM2 patients (31% men, age 51.9 ± 11 1, disease duration 15.0 ± 13.7 years). Consensus criteria for MetS from 2009 were applied.

Results: Prevalence of MetS in patients with DM1 was 17%, while the individual components of MetS were presented with the following frequencies: hypertriglyceridemia 67%, low HDL cholesterol 35%, hypertension 18%, central obesity 14% and hyperglycemia 9%. Prevalence of MetS in patients with DM2 was 49%. Frequency of the individual components of MetS was as follows: elevated blood pressure 60%, central obesity 57%, hypertriglyceridemia 47%, hyperglycemia 32% and low HDL cholesterol 28%. The presence of MetS was not associated with age and gender of patients, disease duration and degree of muscular disability in any of two patient groups ($p>0.05$). DM1 and DM2 patients with MetS had significantly lower quality of life as measured by SF-36 questionnaire compared with patients who did not have MetS ($p<0.05$).

Conclusion: Patients with DM2 had a higher frequency of MetS compared to DM1 patients. Dyslipidemia was the most common component of MetS in DM1 patients, and arterial hypertension and central obesity in DM2. Quality of life was significantly lower in DM patients with MetS.

STABILITY OF URINE SPECIMENS STORED WITH AND WITHOUT PRESERVATIVES AT ROOM TEMPERATURE AND ON ICE PRIOR TO URINALYSIS

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Background : Laboratories determine the most appropriate approach for the collection and transport of urine specimens. We investigated the effect of a chlorhexidine-based preservative tube on sample stability, compared the results of refrigerated polystyrene tubes with no additives, and investigated the effect of temperature on the performance of preservative tubes.

Subjects and methods: Fresh urine specimen (n=48) aliquots in BD Vacutainer® Plus Urinalysis Preservative Tubes and polystyrene tubes were analyzed on an Iris Diagnostics iQ200. Samples in polystyrene tubes were refrigerated for 4 and 8 h. Four aliquots in preservative tubes were kept at room temperature for 4, 8, 24, and 72 h, while two aliquots were kept on ice for 4 and 8 h.

Results: There was good agreement for all chemistry and microscopy parameters with the exceptions of white blood cells (WBCs) at 24 and 72 h and red blood cells (RBCs) at 72 h. Preservative tubes on ice showed a significant decrease in concordance of WBCs and calcium oxalate (CaOx) parameters compared with the results at room temperature.

Results of refrigerated polystyrene tubes showed good agreement with the exceptions of WBC clumps and amorphous crystal at 8 h.

Conclusions: A chlorhexidine-containing preservative tube seems advantageous for urine sample transport from outside healthcare services. A preservative tube offers comparable results with urine samples kept in a refrigerator for 4–8 h for the majority of parameters. Keeping samples at room temperature is recommended when preservative tubes are used because ice produces a negative effect on WBCs and CaOx.

NEW (DIRECT) ORAL ANTICOAGULANTS (N(D)OAC) - A LABORATORY PERSPECTIVE WHEN AND HOW TO TEST N(D)OAC

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The new (direct) oral anticoagulants (N(D)OAC), represented by the thrombin-inhibitor - dabigatran and the Xa - inhibitors rivaroxaban and apixaban, are now approved in many countries, for the prevention of stroke in patients with nonvalvular atrial fibrillation (AF) and for secondary prevention of venous thromboembolism (VTE).

Routine laboratory monitoring is currently not recommended for N(D)OAC. However, there are situations in which measurements of the drugs and their effect are desirable (e.g. preparation for surgery; major bleeds, to check compliance and/or effect, renal and/or liver impairment). Renal impairment increases the risk of suffering major bleeds particularly in patients treated with dabigatran and rivaroxaban (the risk is lower for apixaban), and measurements of drug levels and/or anticoagulant effects may therefore be of particular interest in those patients. The lack of a specific antidote makes this issue even more important.

Chromatography with tandem mass spectrometry (LC-MS/ MS) is the method of choice for the detection and quantification of those drugs. However, this method will probably be available for routine use only in specialized centers, and in such centers perhaps only during weekdays. Therefore, functional coagulation tests that indirectly estimate plasma concentrations of N(D)OAC may be used instead.

Considering dabigatran, diluted thrombin time assays (e.g. Hemoclot thrombin inhibitor (HTI)) and ecarin clotting assay (ECA) are the assays of choice. However, neither of these assays can be used to monitor low levels or infer the total absence of dabigatran. The classical coagulation assays, PT-INR and aPTT, have no or very limited utility in this context. aPTT may be used for qualitative assessment if HTI and ECA are not available, with the precaution that a normal aPTT may be observed even with therapeutic levels of dabigatran.

In the case of rivaroxaban, anti-Factor Xa chromogenic assay with specific rivaroxaban calibrator in which results are expressed as rivaroxaban concentration (μg/l) can provide accurate results. The classical coagulation assays, PT-INR and aPTT, have no or very limited utility either. PT may be used for qualitative assessment if the anti-Xa chromogenic assay is not available, but this assay cannot be used for the determination of rivaroxaban level.

Limited experience is available for the laboratory testing of apixaban. The anti-FXa chromogenic assay should be the assay of choice preferably with a specific plasma calibrator which is currently available. The classical coagulation assays, PT-INR and aPTT, seem to be poorly responsive and should not be used for laboratory testing of apixaban.

When laboratory testing of N(D)OAC is necessary the obtained results should be carefully interpreted since plasma concentrations vary considerably even in healthy individuals. N(D)OAC have a relatively short half life (9–15 hours) with peak concentrations reached after 2-4 hours. Therefore, the results of laboratory tests will be difficult to interpret without the knowledge of the exact time when the blood sample was collected related to the last dose of medication. Furthermore, plasma concentration does not fully correspond with the anticoagulant *ex-vivo* effect. It is therefore of particular importance to correlate laboratory findings with clinical outcomes before potential recommendations for desired concentrations of NOAC.

COMPARISON OF DIFFERENT STATISTICAL DESIGNS FOR INTERPRETATION OF AN EXTERNAL QUALITY ASSESSMENT SCHEME

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Background: Proficiency testing is defined as "determination of laboratory performance by means of interlaboratory comparisons" in International standard ISO 13528:2005(E). The standard states the statistical methods used for determining the assigned value, standard deviation and calculation of the performance scores for interlaboratory comparisons. Parametric statistics are the more commonly known approach in an external quality assessment program. However, due to the variability of laboratory performances, the distribution of data is often abnormal, including some outlying values. Robust statistical methods, which are based on weighted data and are resistant to the presence of outliers can be used alternatively for EQA evaluation. In this study, we compared the performance scores obtained by using different statistical methods: Parametric statistics (arithmetic mean and standard deviation) and non-parametric robust statistics (median and robust standard deviation).

Material and method: A total of 1008 glucose, 842 lactate dehydrogenase (LDH) and 846 sodium results of three lots at different concentrations, submitted through the Kbudek-EQA reporting system, were collected and analyzed by Kbudek-EQA evaluating system and SPSS 17.0. Participants were evaluated in their peer groups. The data distributions of three sets were tested by Kolmogorov-Smirnov Z test. "Consensus of Participants' Results" approach was used to determine the assigned value for a test material. z-scores were calculated using both standard deviation and robust standard deviation. The z-score values outside the ±3 standard deviation (SD) were evaluated as a procedure that needs investigation. These were considered unsatisfactory, while the values inside the ±2 SD limits were considered satisfactory, and z-scores outside the ±2 SD limit but inside the ±3 SD limits were considered questionable.

Result: We found the rate of the unsuccessful and questionable performance scores of the three lots for glucose to be 4.9%, 10.1%, 11.1% respectively, by using robust statistics and 2.1%, 6.8%, 7.9% by using parametric statistics. While the rate of the unsuccessful and questionable performance scores for sodium were found to be %12.4, 8.1%, 7.2% by using robust statistics and 8.5%, 5.7%, 4.9% by using parametric statistics; the rate of the unsuccessful and questionable performance scores for LDH were found to be 10.1%, 7.9 %, 11.6% by using robust statistics and 7%, 5.7%, 6% by using parametric statistics. Two methods of evaluation of performance didn't show statistically very good agreement for three parametres (McNemar's test, $P<0.05$).

Conclusion:
External quality assessment including extensive participants often result in abnormal distribution. Robust statistical methods are insensitive to slight deviation for a given probability model and can estimate the population parameters utilizing robust algorithm. We can conclude that robust statistical method may be more appropriate for EQA result analysis in selected cases.

CHALLENGE, SOLUTION, IMPROVEMENT – THE IMPORTANCE OF ACCREDITATION IN PRIVATE PRACTICE

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The first private laboratories in Serbia were founded over 20 years ago as a support to private doctor's offices and polyclinics. Thanks to constant tracking of the patients needs and fast adaptation to those demands, the development of private practice in all areas had lead to the fast development of the private laboratories. Rapid development and conquering new technologies, expanding the choice of services which is offered to customers and stiff competitors have forced the need for continuous advancement of quality of work of the laboratories. The process of accreditation was a logic path to realization of a set goal: ensuring the trust in quality and competence.

During preparations for accreditation according to SRPS ISO 15189 standard, the laboratory was faced with numerous challenges. The quality manual was created to define the politics and general goals, legal status and main tasks, inner organization, the tasks of certain organizational cores and functions, the system of supervision of functioning of the systems management and the form of evaluation of the degree of effectiveness. The rule book which regulates internal organization and systematization was created, the organizational scheme was defined and detailed job descriptions for every position were issued. The mold of responsibility was made according to standard's demands. The numerous procedures and instructions of systems of management, by which the process of work in laboratories were managed, were defined. The code of conduct alongside with the strict respect of code of ethics were defined. Security measures, which protect the privacy of customers according to the regulations of law and standard's demands, were defined. All of the technical regulations in the work of a laboratory, starting with provision of environmental conditions, handling of equipment, supervision of staff and inclusion in programs of external quality control, were arranged. There were several key points which demanded significant improvement. Informational system was the most important project that started in 2009. and is still in development. Today, laboratory equipment is connected to LIS via bidirectional communication between each instrument and host computer.

Accreditation improves the laboratories quality of service, increases the quality of results, motivates the employees and increases the satisfaction of customers. Also it ensures the optimal usage of resources as well as continuous tracking and constant improvement of efficiency of defined processes. The accredited laboratory is unavoidable partner of clinical staff responsible for treatment and care of patients, and laboratory results represent reliable support to making a decision about a diagnosis, treatment and prognosis of disease.

LECTURE PRESENTATIONS

NATIONAL CONGRESS OF THE ASSOCIATION OF MEDICAL BIOCHEMIST IN BOSNIA AND HERZEGOVINA

IMPACT OF THE EN ISO 15189:2012 ON HARMONIZATION OF LABORATORY TESTING

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For a medical laboratory, receiving accreditation means appropriate laboratory processes are in place. Today is quality management system in medical laboratories implemented by application of international norm EN ISO 15189:2012, Medical laboratories – Requirements for quality and competence. The EN ISO 15189:2012 specifies management and technical requirements for medical laboratory testing.

Harmonization has become a major topic in laboratory medicine over recent years. Efforts for harmonization must be made in the total testing process, from test requesting to communication of the laboratory test results and its consequences to the patient. The complete picture of harmonization in laboratory testing goes beyond method and analytical results harmonization and includes all other aspects of laboratory testing.

Accreditation of medical laboratories according to the EN ISO 15189:2012 standard introduces among requirements the provision of "advice to those requesting information about the choice of tests, the use of laboratory services, and the interpretation of laboratory data" which is important issue of strategies to improve appropriateness in test request and harmonization current practices. Also, the EN ISO 15189:2012 contains requirements regarding report transmission, reference intervals, communication of critical values and harmonization of consultant advisory service.

Laboratory accreditation is the internationally accepted framework for increasing test quality, reducing the frequency of laboratory errors and achieving harmonization of the total testing process.

HARMONIZATION IN THE ANALYTICAL PHASE OF LABORATORY TESTING: MANUFACTURERS DECLARATIONS

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Even though harmonization of laboratory testing includes all phases of laboratory work, harmonization of laboratory procedures and methods is still a main prerequisite for interchangeable laboratory results. Application of different methods and reagents for the same test can result with significantly different result and clinical outcome for the patient. In order to determine the agreement between different methods, laboratory specialists have to be familiar with all method characteristics. The main sources of information for laboratory methods are manufacturers declarations.

EU Directive on in vitro diagnostic medical devices – IVD 98/79/EC regulates responsibilities of manufacturers of laboratory reagents. According to the document, manufacturers are obliged to perform extensive validation of all reagents and analytical systems that are intended for diagnostic purposes. This data has to be presented in an accurate way and available to the users. Unfortunately,

manufacturers declarations are often incomplete, inaccurate and not harmonized. Moreover, information from the declarations often cannot be verified in the routine lab conditions.

Harmonized data in manufacturers declarations would allow objective comparison of different reagents. Manufacturers generally meet required criteria and most declarations contain data on precision, linearity or interferences. However, there is a great heterogeneity in data presentation. For example, in precision studies, manufacturers use different materials (control or patient samples), different number of replications, different concentration levels, different formula for calculation precision measures, or even different precision measures. When presenting the data on hemolysis, icteria and lipemia, manufacturers should describe protocols for creating samples with interferences, interferent concentrations, analyte concentrations and measured bias. Declarations are often not complete and lot of these data is missing. It is therefore very hard to compare the data between different reagents or even replicate the data in verification studies. Reasons for this probably lie in the differences between manufacturers labs and routine labs worldwide where lot of people are included into working processes or because of the deterioration of measuring or supporting equipment. Additionally, different protocols are sometimes used for verification.

It is therefore obligation of manufacturers to use up-to-date standards when performing validation studies, to update the data when new protocols are released and to present all relevant data to their users. Laboratory specialists have to verify that declared specifications can be confirmed in the routine conditions and together make efforts to improve harmonization of all phases of laboratory work.

QUALITY INDICATORS IN POST-ANALITICAL PHASE OF LABORATORY TESTING

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Laboratory testing, a highly complex process comonly called the total testing process (TTP), is usually subdivided into three traditional analytical phases. Activities that fall into the post-analytical phase include result reporting, critical value notification, manual transcription of results and subsequent data entry, and analysis of turnaround time (TAT).Traditionally, the laboratory community has viewed post-analytical errors as errors that occur after the analysis is complete but within the confines of the 4 walls of the laboratory itself and under the control of laboratory. Post-analytical phase of the TTP has been considered to be less prone to error than pre-analytical processes because of the widespread adoption of laboratory automation and interfaced laboratory reporting. However, the explosion of environments in which laboratory results are displayed, such as smart phones, tablets, and patient portals, make the management of test results an increasingly error-prone process. Laboratories must become advocates for patient safety by developing new quality monitors to ensure that results posting in electronic health records are interpretable and are received and reviewed by responsible providers of care.

Establishing reference intervals is a major step in the post-analytical phase of the testing process, enabling proper interpretation and usage of laboratory data.

Laboratory testing plays an indispensable role in health care, as 80–

90% of all diagnoses are made on the basis of laboratory test results. First, specimens are collected by a nurse and delivered to the laboratoryby a courier. Second, the clinical pathologist uses the specimenas a surrogate for the patient to performthe test and returns the resultsto the requesting physician. Finally, the patient is diagnosed or treatedaccording to the physician's judgment,which is informed by the test results. It is clear that any failure in this series of events may result indelayed or misinformed health care, with the potential for great financialor physical costs to patients.

Timely delivery of correct results has long been considered as the goal of quality management inclinical laboratory.Quality monitors and controls for the post-analytical process have focused on critical result notification, meeting established turnaround time goals, and review of changed reports. The rapid increase in the adoption of electronic health records has created a new role for laboratory professionals in the management of patient test results. Laboratory professionals must interface with the clinical side of the health care team in establishing quality control for the post-analytical processes, particularly in high-risk transition of care.

-verification for analytical results:
assessment and confirmation of the integrity of the results,
identification of any outliers,
exclude any outliers caused by pre-analytical error,
countercheck any questionable results
-Reflective/reflex testing
-Units and reference intervals
-Interpretation and commenting on test results
-Labor
-result reporting:
identify critical results and inform the physician (reporting of critical results), release the report in a timely manner, verify that all required tests were completed

-Post-post-analytical
interpretation of test results in the clinical context and action

It has been roughly estimated that approximately 70% of all majorclinical decisions involve consideration of laboratory results. Ina ddition,approximately 40-94% of all objective health record data are laboratory results. Undoubtedly, accurate test results are essential for major clinical decisions involving disease identification, classification, treatment, and monitoring. Factors that constitute an accurate laboratory result involve more than analytical accuracy and can be summarized as follows: The report reaching the clinician contained the right result, together with interpretative information, such as a reference range and other comments, aiding clinicians in the decision-making process.
Post-analytical steps, involving the transmission of the laboratory data to the clinical provider, who uses the information for decision making.
Although minimization of analytical errors has been the main focus of developments in laboratory medicine, the other steps are more frequent sources of erroneous results. An analysis indicated that in the laboratory, pre-analytical errors accounted for 62% of all errors, with post-analytical representing 23% and analytical 15% of all laboratory errors.
Importantly, 92% of the pre-analytical, 88% of analytical, and 14% of post-analytical errors were preventable.

REFERENCE INTERVALS FOR BIOCHEMICAL PARAMETERS HEALTHY STUDENTS IN BOSNIA AND HERZEGOVINA

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The clinical chemistry reference interval is one of the most important decision making tools used to distinguish between healthy and diseased individuals. A variery of factors can influence reference ranges. Different laboratory methods often yield significantly different results and therefore require different reference ranges. Beceause of differences in age, genetic background, or exposure to environmental factors, different populations may need different reference ranges for certain laboratory analytes. Results of laboratory investigations have limited meaning on medical decisions without reference intervals (RIs). Biochemical values determined in the present study serve as reference range for student population and can be used for helathy control and diagnosis of diseases. The reference group consisted of 98 healthy subjects (54 famales and 44 males, between 20-25 years of age) selectiong aposterior according to the strictly defined criteria. Sample were analyzed on Cobas 6000 and Architect platforms.The reference intervals were validated as recommended by the Clinical laboratory Standard Institute (CLSI). For derivation of RIs, both parametric and non parametric methods were applied. Comatasion of biochemical ranges, obtained for healthy students, with reference ranges from other nationalities, suggested that most values were similer.

MANAGING THE DEMAND FOR LABORATORY TESTING- SELECT APPROPRIATE TESTS

Emir Hondo

Medical laboratories play an important role in disease prevention, detection and treatment of patients, but diagnostic errors are often associated with laboratory testing.

After a huge progress in the analytical phase and reducing its share in the total percentage of errors in the entire testing process, with help above all from automation and harmonization assays, and the increasing efforts to improve the quality of the pre- and postanalytical phase related to laboratory work, in the last ten years there is an increasing awareness of participation of "Non-laboratory" errors in the total number of diagnostic errors .

This is especially due to irrational demands of clinicians for laboratory tests, their inadequate interpretation and lack of understanding / ignorance about opportunities / limitations of certain laboratory tests.

Taking all of the above into account, the laboratory specialists have a big responsibility in close cooperation with clinicians to improve management requirements for laboratory testing, and thereby improve patient safety without diminishing the significance of direct and indirect material savings that would result from improving the quality of the entire testing process.

SMART LABORATORY

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Background: Past five years can be described as "IT revolution" in healthcare in general, that wave was lead by laboratory departments in general.

Methods: Every single one of laboratory departments are submited to the order by physician that send samples for testing for test that they believe that is the best for the moment and for the amount of information that they have.

Results: We now have laboratories that are acting as a service for doctor with little or no influence in treating patient. Also laboratories are commonly considered one of the most spending departments in healthcare institutions.

Conclusion: Laboratory processes can be smarter than that and laboratory staff must be more involved for treating patient and usual attitude that laboratory is just service for doctors is everything but that because all medical staff are in service for patient health.

CONTRIBUTION TO FIGHT AGAINST DOPING IN BOSNIA AND HERZEGOVINA: ROLE OF AGENCY FOR ANTI-DOPING CONTROL

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Background: Agency for anti-doping control of Bosnia and Herzegovina was established in the Law of sports by the Parliamentary Assembly of Bosnia and Herzegovina in 2008. First doping controls of urine of Bosnian athletes under legislative of the Agency started in 2010. The Agency has Code compliance with all regulating documents issued by World Anti-Doping Agency (WADA).

Materials and methods: Data about the number of samples collected by the Agency and number of positive doping cases in human and animal sports were analyzed.

Results: In 2010 the Agency collected only 72 urine samples and analyzed them for doping substances in an accredited laboratory in Bucharest. Nowadays Agency collects around 300 samples per year. Doping positive cases in a period 2010-2015among Bosnian athletes in following sports are: Kick boxing- 2; Weightlifting-2; Karate-1; Handbal-2; Kayak-2; Substances that had been detected among athletes are: Boldenone, Methandienone, Amphetamine, THC, Ephedrine.
In years 2011- 2012,The Agency also collected 15 samples in animal sports, where 3 positive cases were found and one refusal of doping control, which is also sanctioned as a positive case. Forbidden substances found among tested horses are: Triamcinolone-acethonide, Phenyl-buthasone, Teophylline, Dexamethasone.

Conclusions: Certain sports have higher risk of forbidden substance abuse among athletes. Percent of doping positive cases among Bosnian athletes was between 1-1.5 %, similar to global doping percent. We found 20 % of doping positive cases among horse samples, and this problem should be addressed, among decision makers in Bosnian animal sport.

POSTER PRESENTATIONS

PT01
BODY FLUID CELL COUNTING: COMPARISON BETWEEN TWO AUTOMATED HEMATOLOGY ANALYSERS AND THE MANUAL MICROSCOPE METHOD

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Background: Laboratory testing can be performed on many types of fluids other than blood. Up to now the microscopic counting and the differentiation of white blood cells (WBC) in a body fluid smear have been used as a reference. The aim of our study is to prove if there is any difference between number of body fluids cell counting on analysers or manual and to evaluate the correlation between these techniques.

Materials and methods: We have estimated 30 body fluid samples: 21 cerebrospinal fluid (CSF) samples (70%), 6 pleural fluids (20%) and 3 abdominal fluid samples (10%). Total counting of each sample has been conducted with two automated systems: "Sysmex XT-4000i" and "Sysmex XE-5000" and manual method. For device counting we have used body fluid mode with differentiation of mono andpolymorphonuclear WBC**.** Fuchs-Rosenthal chamber was used for CSF cell counting and Neubauer chamber for cell counting in all other body fluids. For visualisation and cell staining, we have used: methyl violet and Türk'ssolution.

Results: We found statistically significant positive and strong correlation between "Sysmex XT- 4000i" and "Sysmex XE- 5000" and between these two types of Sysmex counters and manual method. Correlation coefficient in both cases was 0,992 (significance level $p<0,0001$). There is positive correlation in differentiation of mono andpolymorphonuclears on two types of "Sysmex" counters, coefficient 0,823 and with manual techniques of counting, coefficient was 0,721. We found positive strong relation in all CSF samples: low cell count (< 5), medium (between 6 and 50) and high cell count samples (> 50) with coefficient >0,95. The results for abdominal and pleural fluids were similar. We examined 4 samples with <500 cells and 5 samples with cell count >500. In both group correlation coefficient was higher than 0.985.

Conclusion: The introduction of automated method for body fluid cell analysis is very useful in the routine laboratory analysis examination. We found very strong and positive correlation between "Sysmex XT- 4000i", "Sysmex XE- 5000" and manual reference method for body fluid cell counting and for mono andpolymorphonuclear WBC differentiation, both at lower and at higher cell counts.

PT02
HIGH GLUCOSE ENHANCES THE PROLIFERATION OF VASCULAR SMOOTH MUSCLE CELLS AND ANGIOTENSIN II-INDUCED P44/42 MAPK AND P38 MAPK PHOSPHORYLATIONS

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Background/aim: Angiotensin II (Ang II) signaling pathways under high glucose (HG) conditions responsible for the accelerated cardiovascular disease in

diabetes mellitus. Moreover, the effects of Ang II under HG condition in vascular smooth muscle cells (VSMCs) is still obscure. In this study, we examined p44/42 MAPK (ERK1/2) and p38 MAPK activation following to Ang II stimulation in high glucose conditioned-media (25 mM). In addition, the effects of HG and Ang II on proliferation of VSMCs were also evaluated.

Materials and methods: VSMCs were isolated from male Wistar rat thoracic aorta and cultured. Growth-arrested VSMCs were placed in either normal glucose (NG: 5.5 mM) or high glucose (HG: 25 mM) media for 48 h and then the cells were stimulated with Ang II (100 mM) in a time course. The phosphorylation of p44/42 MAPK and p38 MAPK were determined by immunoblotting. To determine the effect of Ang II on VSMC proliferation, the cells were incubated in NG and HG medium for 24 h, and then cells were induced with Ang II for 24 h in the same media (Total incubation time was 48 h). The cell proliferation assay was performed by using an MTT-based technique.

Results: Ang II stimulated both p44/42 MAPK and p38 MAPK phosphorylation more robustly in HG media for 2, 5, 10 min compared to NG media in VSMCs. Incubation of VSMCs in high glucose for 48 h increased VSMC proliferation compared to normal glucose incubation. Additionally, proliferation of VSMC induced with Ang II for 24 h in HG was increased more effectively compared to NG.

Conclusion: Our results highlights that proliferative effect of HG and Ang II on VSMC may be associated with augmented p44/42 MAPK and p38 MAPK phosphorylations. In conclusion, the restrict control of high glucose level should be among the primary targets to encounter vasculopathies in diabetic patients.

PT03
THE FIRST STUDY OF FREQUENCY CCR5Δ32 MUTATION IN BOSNIAN AND POLISH PATIENTS WITH CROHN'S DISEASE.

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Background: Crohn's disease (CD) is a multifactorial disease with an environmental factors and genetic background. In the recent period the available data on genetic factors and immune system in susceptibility in CD attracts great attention. Chemokines and their receptors participate in the pathogenesis of various inflammatory diseases, where play an important role in migration and activation of monocytes and macrophages. Several studies have confirmed that genetic variants in the chemokine receptor *CCR5* gene correlate with susceptibility to CD. Data on prevalence and phenotypic consequences of polymorphism *CCR5* gene in CD in Poland and Bosnian populations are sparse or nonexistent. Therefore, the aim of our study was to assess the frequency of Δ32 allele and their association with phenotypic expression of the disease, in population of Polish and Bosnian patients with CD.

Subjects and methods: We recruited 86 CD patients mean age 34.1 (±13.0) and 83 controls mean age 35.4 (±12.8) years in Poland and 30 CD patients mean age 44.1 (±14.5) and 30 controls mean age 61.3 (± 15.2) years in Bosnia and Herzegovina; 229 participants in total. We determined the prevalence of *CCR5*Δ32mutation and its association with phenotypic expression of the disease according to Montreal classification. Participants were genotyped for *CCR5*Δ32mutation by polymerase chain reaction (PCR) and follow-up using the Statistical Analysis Package IBM SPSS Statistics (version 21).We verified the correctness of results by performing re-genotyping of randomly selected samples.

Results: We identified 2 heterozygotes in Bosnian and 8 heterozygotes and 2 homozygotes in Polish CD patients, with mean Δ32 allele frequency 3.3% and 7.0%, respectively. In Bosnian and Polish control group we found 8 and 16 heterozygotes, with mean Δ32 allele frequency 13.3% and 9.8%, respectively.

Conclusion: Increased frequency of the Δ32 allele in CD patients from Poland (p=0.048) but not from Bosnia was observed. In Polish CD patients the age of diagnosis between 17 and 40 y and ileocolonic location were related with higher frequency of *CCR5*Δ32 mutation, while in Bosnian CD patients the correlation was with the age of diagnosis above 40 y (both p>0.05). Further studies with larger samples in both countries are warranted.

PT04
SALIVARY INTERLEUKIN-6 AND INTERLEUKIN-10 LEVELS IN SUBJECTS WITH OBESITY AND GINGIVITIS

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Background: Gingivitis is a frequent inflammatory process of the gingival tissues that is mainly caused by the accumulation of plaque. Interleukin IL-6 (IL-6) is known to be a major modulator of inflammation in inflammatory reactions. Interleukin IL-10 (IL-10) has been found to regulate and control the balance between innate inflammatory and acquired humoral responses. In this study we aimed to evaluate the association between obesity and periodontal disease in regarding by cytokines IL-6 and IL-10 in saliva.

Methods: The study was carriedout in 40 subjects: 20 obese patients with gingivitis and 20 non-obese patients with gingivitis who are controls. Periodontal parameters such as gingival index (GI) and plaqueindex (PI) of patients were recorded. Saliva samples were used for measuring IL-6 and IL-10 levels by ELISA method.

Results: Saliva IL-6 levels weres ignificantly higher in obese patients than those of non-obese subjects (p=0.002). When two groups were evaluated together, negative significant correlation between GI and salivary IL-10 levels (r=-0.452, p=0.003) and positive correlations between salivary IL-6 level and body mass index (BMI) (r=0.369, p=0.019) were found. There was a negative correlation between the GI and salivary IL-10 levels in obese patients (r=-0.548, p=0.012). Also there was a

positive correlations between the salivary level of IL-6 and IL-10 in obese patients (r=0.594, p=0.006).

Conclusion: In our study, it was demonstrated that obese patients with gingivitis has increased salivary IL-6 levels compared with non-obese patients. Obesity and BMI may also affect the inflammatory response by altering the IL-6 level

PT05
THE DETERMINATION OF CYCLOSPORINE IN WHOLE BLOOD AT PATIENTS AFTER KIDNEY TRANSPLANTATION

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Background: The specific pharmacokinetic profile of cyclosporine is low therapeutic index and potential interactions with numerous medications indicate the fact that monitoring of immunosuppressive therapy is the essential part of therapy protocol in transplant patients.

Material and methods: The patients have got a cyclosporine microemulsion formulation (Neoral®; Novartis Pharma) in dose 100-250 mg/mL at the first 7 days after kidney transplantation. In the study we have determined blood concentration of cyclosporine at 30 transplantation patients, who were hospitalized at Department of Nephrology at the University Clinics Centre of Sarajevo. The cyclosporine concentration of 100 blood samples was determined using CMIA (chemiluminesecent microparticle immnoassay) Architect i 2000 and FPIA (fluorescence polarization immunoassay) AxSYMAbbott diagnostic. The reference blood range of cyclosporine for kidney organ transplantation at the first 7 days for initiation dose lies between 150 and 250 ng/mL.The quality control, precision and accurancy of Architect i 2000 were assessed.

Results: The quality control was done using quality control serums for low ($\bar{x}$ = 95 ng/mL), medium ($\bar{x}$ = 338 ng/mL) and high ($\bar{x}$ = 819 ng/mL). We have used commercial BIORAD controls and got reproducibility CV 5.83 % to 13 % for Architect i 2000. It was established that the main difference between Architect i 2000 and AxSYM and it was statistically significant for $P < 0.05$ according to Student t-test. Correlation coefficient was r = 0.935.

Conclusion: The Architect CMIA immnoassay has low functional sensitivity and lower elimination of interferences: haematocrit, high values of cholesterol, triglycerides, bilirubin, total protein and uric acid. It has reduced interference relative to other immunoassay and is a convenient and sensitive automated method to measure cyclosporine in whole blood.

PT06
COMPARISON OF THE RESULTS OF ROUTINE URINEANALYSIS AND THE URINE CULTURE

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Background: Routine urine analysis and the urine culture are most commonly done analyses in the case of urinary tract infection.The study aim was to compare results of the routine urineanalysisand urine culture.

Methods: Routine urinanalysis and urine culture were performed on 100 urine samples..The positive results of leukocyte esterase obtained by chemical examination of urineand the number of leukocytes over 5 leukocytes per each high power fieldobtained by microscopic examination were taken as possible markers or urinary tract infection.

Results: Results were organized into two groups: the first group with the negative results of leukocytes (below 5 leukocytes per each high power field), and the second group with positive results of leukocytes (over 5 leukocytes per each highpower field). In the first group there were 70 urine samples with negative results of leukocyte esterase. From 70 urine samples with negative results of leukocytes, pathogenic bacteria were detected in 14 urine samples (20 %). In the second group there were 30 urine samples with the positive leukocyte esterase. From the 30 urine samples with positive results of leukocytes, in 27 of them pathogenic bacteria were isolated in urine culture (90%).

Conclusions: In the group with the positive results of leukocytes it was justified to include urine culture, because in 90 percent samples with positive results of leukocytes pathogenic bacteria were isolated. In the group with negative results of leukocytes in 80 percent pathogenic bacteria were not isolated, which indicates that the urine culture was not justified since there were no clinical indications.

PT07
MPV VALUES IN PATIENTS WITH PSORIASIS

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Background: Psoriasis is a common chronic inflammatory, immune-mediated disease, found worldwide; its frequency varies widely from 0.2 to 11.8%. Psoriasis is a chronic inflammatory skin disease with a complex etiology involving genetic and environmental factors. The relationship between psoriasis and other diseases has drawn increasing interest in recent years. Psoriasis is a chronic inflammatory skin disease with a complex etiology involving genetic and environmental factors. The relationship between psoriasis and other diseases has drawn increasing interest in recent years. Platelets are known to play important roles in inflammatory reactions and immune responses. Platelets can be activated by various stimuli, and its activity is known to mediate immune-inflammatory process. The activity of platelets is assessed by measuring various platelet-derived secretory molecules, such as adhesion proteins, growth factors, chemokines, cytokines and coagulation factors. Besides, mean platelet volume (MPV) and platelet distribution width (PDW) have been extensively studied and reported as platelet activation markers.

Mean platelet volume (MPV) has been used as a marker of platelet activation. MPV indicates the size of platelets, and its increase is an indicator of larger, more reactive platelets resulting from an increased platelet turnover. The aim of this study was to investigate the MPV values in patients with psoriasis

Materials and methods: Whole blood samples were collected from 90 healthy control and 130 patients with Psoriasis. The mean age for controls and patients were 43±16 and 39±17years, respectively. Patients with chronic disease and inflammatory disorders were excluded. MPV levels were calculated with Abbott Cell Dyne heamotolgy analyzer. Statistical analysis was performed with SPSS v15.

Results: The mean of MPV values in patients with psoriasis (9.00±1.82) were significantly higher compared to control group (8.05±1.48) ($p<0.001$).

Conclusions: According to this study's results, MPV is increased in psoriasis patients and correlates with disease severity. Therefore, MPV levels may be considered as a marker of disease severity of psoriasis.

PT08
COMPARISON OF METHODS OF FLOW CYTOMETRY ON THE CELL DYN SAPPHIRE ANALYZER AND SUPRAVITAL STAINING WITH BRILLIANT CRESYL BLUE FOR THE DETERMINATION OF RETICULOCYTE COUNT INTHEBLOOD

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Background: Determining the number of reticulocytes in the blood is an important test in the study of erythropoietic activity of bone marrow. The most commonly used methods for the counting of the absolute number of reticulocytes is microscopic, following coloring with the brilliant cresyl blue. It is a time consuming, inaccurate and imprecise method because of the subjective morphological determination of reticulocytes. The number of reticulocytes can also be determined on the hematologic Analyzer Cell Dyn Sapphire, using the method of flow cytometry, tying fluorescentcolor to the RNA from reticulocytes. The purpose of this researchwas to introduce an automatic method of flow cytometry to determine the number of reticulocytes as a routine hematologic test.

Materials And Methods: With the flow cytometry method and supravital staining with brilliant cresyl blue, it is possible to simultaneously determine the reticulocytes in 123 blood samples from subjects of the Hematologic clinic of the University Clinical Center Sarajevo. Resulting values of the number of reticulocytesare divided in three groups: A (N=19)100x10e9/L; group B (N=52) 22-99 x10e9/L and group C (N=52) 19x10e9/L. Compliance between the two methods is verified by a statistical correlation test.

Results: A satisfactory correlation of the two test methods was reached: for the total number of samples ($r=0.9755$, $p<0.0001$); group A ($r=0.9581$, $p<0.0001$); group B ($r=0.7639$, $p<0.0001$) and group C ($r=0.4948$, $p=0.0002$).

Conclusion: Based on obtained results, it is possible to make a determination of reticulocytes using the method of flow cytometry on the Cell Dyn Sapphire Analyzer, and should be brought into routine hematologic test application.

PT09
COMPARISON OF THE RESULTS OF SODIUM AND POTASSIUM SERUM LEVELS IN THE TWO DIFFERENT ANALYTICAL SYSTEMS

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Introduction: Potassium (K) is the major intracellular, and sodium (Na)is the main extracellular cation. The aim of the study was to compare the serum levels of K and Na obtained by using the biochemical analyser BT 3500 (Biotehnica Instruments, Italy),andthe acid-base analyser Cobas b 221 (Roche Diagnostics, Germany), with the purpose of their comparative use in laboratory work.

Materials and methods: Analysers BT 3500 (indirect potentiometry) and Cobas b 221 (direct potentiometry) were used for measuring serum level of K and Na. Sixty-one (n=61)serum samples were analysed with both analysers comparatively. Checking of the accuracy and precision of the analysers was carried out. The results were statistically analysed usingMedCalc software.

Results: The lowest Na level measured with the analyser BT 3500 was 117.0 mmol/L, and the highest level was 146.0 mmol/L. The lowest K level measured with the analyser BT 3500 was 3.10 mmol/L, and the highest level was 5.90 mmol/L. The lowest Na level measured with the Cobas b 221 was 115.0 mmol/L, and the highest level was 145.0 mmol/L. The lowest K level measured with the Cobas b 221 analyser was 2.90 mmol/L, and the highest level was 5.80 mmol/L. Passing-Bablok regression equation for comparison the Na level was $y = 0.000000 + 1.000000 x$, and for K level $y = - 0.10000 + 1.000000 x$. The results indicate that there are no constant and proportional difference in the comparative measurement of Na and K using both the analytical systems. Cusum test did not give a significant deviation in the linearity for K ($P = 0.70$) and Na ($P = 0.82$). Bland Altman graph showed distribution of the values of K and Na within ±1.96.

Conclusion: Both analytical systems BT 3500 and Cobas b 221 can be used comparatively for the analysis of Na and K in routine laboratory practice.

PT10
THE EVALUATION OF REPEATING TESTS AND REPEATED NOTIFICATIONS FOR CRITICAL TEST VALUES IN BIOCHEMISTRY AND HEMATOLOGY LABORATORIES; A MISSING LINK.

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Background: Critical values are the results of laboratory tests that significantly outside the normal limits for the patients indicating a life-threatening situation and should be reported to treating physician immediately. Repeat testing and repeated notifications for critical test results are some of delaying factors in critical value reporting and may cause overloading clinical staff. In

this study we aimed to investigate the proportions of repeated chemistry and hematology critical values that differ significantly from the original value and the occurrence and distribution of repeated notifications in our central biochemistry laboratory.

Materials and methods: All data obtained from laboratory information system (LIS) and written documents used in critical test reporting during 12 months in central biochemistry laboratory of Mustafa Kemal University Hospital. Critical test name, critical value, reporting time, reported person and recipient's name is also recorded to the critical test forms. We calculated the percent difference using formula, % Difference= (Initial test value-repeated test value)/initial test value (data not shown). Results were evaluated using Total Allowable Error (TAE) limits from Westgard database. "Acceptable results" represent the percentage of repeats in which the difference between initial and repeated values does not exceed TAE for allowable error criteria.

Results: During one-year study, 17024 critical tests were observed in central biochemistry laboratory. The most of the critical test results were reported form inpatients (68.8 %). Potassium (19.1 %), hemoglobin (17.2 %) and WBC (13.1 %) were the main reported critical tests and the most common repeated notification was observed in white blood cell (WBC) count (9%).

Conclusions: According to TAE limits, there is no need for repeating critical test value for ALT, AST, troponin, urea, total bilirubin, high glucose and WBC values but other tests especially low critical PLT values and high critical sodium values should be re-evaluated before critical test reporting. According to the RCV only potassium and sodium values need to be repeated before reporting.

PT11
COMPARISON OF TWO CHEMILUMINESCENT IMMUNOASSAY (CMIA I ECLIA) FOR DETERMINATION OF TUMOR MARKER CA 15-3 IN HUMAN SERUM

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Background: In our study, we have investigated the level of CA15-3 in 90 patients with benign and malignant breast disease using two immunoassays electrochemiluminescence immunoassay (ECLIA) and chemiluminescentmicroparticle immunoassay (CMIA).

Materials and methods: The COBAS e 601 (Roche) uses an ECLIA with cut off 0-25 U/mL and Architect i2000 analyzer (Abbott) uses CMIA with cuf off 0.0-31.3 U/mL for determination of tumor marker CA 15-3 in human serum. The statistical significance was set at $p < 0.05$.

Results: Comparison of CA15-3 on COBAS e 601 (Roche) with the Architect (Abbott) showeda correlation coefficient R = 0.998 for patients with malignant tumor disease. The patients with benign disease in comparing between the immunoassays had correlation coefficient R = 0.925. The mean concentration of CA15-3 in ECLIA method was 273.43 U/mL and using CMIA method was 334.41 U/mL in patients with malignant tumor disease. In the group of patients with benign disease mean concentration of CA 15-3 using ECLIA method was 14.48 U/mL and CMIA method was

14.94 U/mL.The results were higher in CMIA method for 3.09-18.21% in comparison with ECLIA. The quality control was done using quality control serums for low ($\bar{x}$ =21.7 U/mL) and high ($\bar{x}$ = 103 U/mL) control. We have used commercial controls and got reproducibility CV 2.93 % to 6.47 % for COBAS e 601 and for CV 4.78 % to 6.36 % Architect i 2000.

Conclusions: Patients should be monitored on a single method to avoid differences in the results. The various immunoassay techniques for detection of CA 15-3 tumor marker use different monoclonal antibodies leading to different results. Different antibodies recognize different parts of the molecule, and antigen heterogeneity may account in part for intermethod differences.

PT12
SERUM ANNEXIN A1 AND LAMININ CONCENTRATIONS IN PATIENTS WITH BLADDER CANCER

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Background: Bladder cancer is the second most common cancer of the genitourinary system. In recent years, it has become clear that various extracellular substrates including laminin play an important role in the process of invasion and metastasis of malignant tumors. Annexin A1 (ANX A1) is a member of the calcium- and membrane-binding family and plays an essential role in tumori genesis and apoptosis. The aim of this study was to evaluate serum levels of laminin and ANX A1 in patients with bladder cancer.

Materials and Methods: Forty three patients diagnosed with bladder cancer following cystoscopic tissue resection were enrolled as group 1. Thirty seven healthy individuals were enrolled as the control group, group 2. Laminin and ANX A1 levels in serum samples were measured with enzyme linked immunosorbent assay.

Results: In group 1, serum ANX A1 levels were higher than in group 2 (p=0.001), There was no significant difference in serum laminin concentration between two groups. Also correlation between ANX A1and laminin was not found. When patients were divided in to two subgroups according to tumor grade, serum ANX A1 levels exhibited no statistically significant difference between the low and high grade groups.

Conclusion: Because serum ANX A1 increases in patients with bladder cancer, this marker may be used in diagnosis of bladder cancer. But further studies on larger groups are needed to confirm this finding.

PT13
DISTURBANCES OF ACID BASE BALANCE IN PATIENTS WITH ACUTE LEUKEMIA

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Background: Acute leukemia occurs when a hematopoietic stem cell undergoes malignant transformation into a primitive, undifferentiated cell with abnormal longevity. Disturbances of acid base balance and electrolyte abnormality are commonly seen in patients with acute leukemia, due to eitherleukemic processes, organ infiltration, and cell death or to adverse effects of cytotoxic drugs. The aim of thisstudy was to determinethe acid baseand blood gas parameters, as well as theactivity of theenzymelactatedehydrogenase (LDH) in thebloodofpatientswith acute leukemia.

Subjects and methods: Our study group consisted of 35 patients with acute leukemia and 18 healthy subjects as the control group. Ten of our patients were male and 15 were female. All acid base and gas parameters were monitored on a blood gas analyzer.

Results: Laboratory data on admission were as follows: white blood cells (WBC) count was elevated ($25x10^9$ g/L); the activity of LDH was significantly higher (2487.4±1463 U/L), opposite to control (358±24.58 U/L; $p<0.001$). Arterial blood gas analysis revealed significantly high level pH (7.45±0.01) than healthy donors (7.41±0.01; $p<0.05$) Marked hypoxia Po_2 (71.54±4.02 mmHg) and hypocapnia (31.08± 0.92 mmHg) were found in leukemic patients, versus to control group (96.18±3.01 mmHg and 35.70±0.82 mmHg respectively; $p<0.001$). In patients with acute leukemia we noted significantly high $AaDO_2$ gradient (38.18±4.71), opposite to control (6.19±3.55; $p<0.001$), respectively significantly depleted a/A gradient(0.63±0.04) compared to healthy individuals (0.86±0.09; $p<0.001$). There was significant positive correlation between the activity of LDH and acid base parameters - pH, bicarbonates (HCO_3) and base excess (BE) ($p<0.001$ for all three).

Conclusion: These findings suggest that combination of respiratory alkalosis (due to hyperventilation) and metabolic acidosis (possibly due to lactic acidosis) was associated in mismatching of ventilation to perfusion increases venous admixture in leukemic patients.

PT14
EVALUATION OF AMINO ACIDS LEVELS MEASURED IN OUR LABORATORY

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Background: Amino acids play a variety of roles in metabolism, as components of proteins. Liquid chromatography-tandem mass spectrometry (LC-MS/MS) has greatly increased the screening possibilities by monitoring levels of amino acids.

In this study admitted to our hospital between April-August 2015, 58 male, 56 female (757.53 ± 1286.019 day)

amino acids results in 114 patients were evaluated.

Methods: 12 amino acid levels were evaluated in this study; Alanine, arginine, aspartic acid, citrulline, glutamic acid, glycine, leu-ileu, methionine, ornithine, phenylalanine, valin and tyrosine. Amino acids were analysed in dried blood spots using liquid chromatography-tandem mass spectrometry (LC-MS/MS) in male and female patients.

Results: In conclucion, amino acid levels of patients were 318.93 ± 190.896 (alanine), 19.45 ± 18.807(arginine), 59.35 ± 29.178(aspartic acid), 20.73 ± 9.470 (citrulline), 206.41 ± 75.119(glutamic acid), 377.78 ± 235.722 (glycine), 179.38 ± 262.289 (leu-ileu), 23.01 ± 11.552 (methionine), 202.80 ± 158.462 (ornithine), 54.20 ± 28.120 (phenylalanine), 185.12 ± 85.164 (valin), 67.99 ± 59.740(tyrosine) umol/L. respectively (mean ± standart deviation). The reference ranges for these amino acids; (130 to 542), (10-130), (10-100), (3-46.7), (20-273), (106-551), (43-205), (6-58.8), (20-280), (16-129), (52-234), (32-275), respectively.
12 of 114 patients have Alanine which is above the reference levels (arginine and citrulline 1,aspartic acid and methionine 3, tyrosine 4, phenylalanine 2, glycine16, glutamic acid 18, leu-ileu 21, ornithine 24, valin 22) and 6 patients alanine levels is below the reference levels (arginine 31, aspartic acid and valin 1, tyrosine 16).

Conclusions: This study is important for Konya region amino acid metabolism disorders studies subject areas. This study is the first study to assess aminosit levels of patients living in our region. We believe that our results will be useful for future studies.

PT15
EFFECTS OF DIFFERENT DOSES ZINC GLUCONATE ON GLYCEMIAAND ANTIOXIDANT PARAMETERS IN TYPE 2 DIABETICPATIENTS

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Background: Researches with micronutrients are getting more and more important in science and also in practice. In this view, zinc is having a special role in preventing micro- and macrovascular diabetic complications, as integral componentof the antioxidant enzyme (superoxide dismutase) and also as cofactor of enzymes and hormones involved in the metabolism of glucose. The aim of this study was to investigate the effects of different doses of zinc gluconate on glycemia and parameters of antioxidative defense: superoxide dismutase (SOD) and total antioxidant status (TAS).

Subjects and methods: Our study included 12 subjects with type 2 diabetes mellitus and 12 healthy individuals. All participants had not taken vitamin or mineral supplements for at least 3

months before sampling. Blood samples were drawn after an overnight fasting in both groups, before and 24 hour after administration of zinc gluconate in three different single doses (15, 25 and 50 mg) with washout period (10 $t_{1/2}$) between treatments.

Results: In type 2 diabetic patients and healthy individuals different single doses of zinc gluconate didn't cause any significant changes in serum glucose level and TAS. After administration of zinc gluconate in different single doses (15, 25 and 50 mg) the activity of SOD in hemolysate of erythrocytes was increased but not statistically significant only in type 2 diabetic patients. As expected zinc concentration in urine was significantly higher in diabetic patients compared to the healthy subjects ($p=0.0269$). Zinc concentration in serum was not significantly different between this two groups. Glucose concentrations were negatively correlated with superoxide dismutase activity.

Conclusions: In conclusion, different single doses of zinc gluconate don't inducestatistically significant changes in glucose level and antioxidant status parameters (TAS and SOD)in type 2 diabetic patients.

PT16
THE INVESTIGATION OF THE EFFECTS OF WALNUT AND HOMEMADE ALCHOHOL (BOGMA RAKI) CONSUMPTION ON LEARNING AND MEMORY WITH MORRIS WATER MAZE TEST.

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Background: Homemade alcohol (Bogma Raki), which is a homemade alcoholic beverage, optionally produced from various fruits such as raisins, dates, figs, mulberry and plum especially in Cukurova region. Walnut is a nutritious essential for the body which has rich antioxidant content such as melatonin and contains a high proportion of polyunsaturated fatty acids such as omega-3 and omega-6. Walnut also includes minerals essential for brain functions. In this study, we investigated the effects Bogma Raki and Walnuts consumption on learning and memory with Morris Water Maze (MWM).

Materials and methods: The subjects were divided into 4 groups, including 12 animals in each group; Group 1: Control group; Group 2: Walnut group (10 g / kg / day); Group 3: Bogma Raki group (30% v / v, 9.2 ml / kg / day); Group 4: Bogma Raki + Walnut group. At the end of this four-week study, spatial learning and memory functions related to hippocampus of all subjects were evaluated by MWM test.

Results: In the MWM test, We evaluated the Platform Finding Time (PFT) and found that at the 1st day of the study, PFT were significantly longer in Bogma Raki + Walnut group ($p<0.01$) compared to Bogma Raki group. During Probe test, the number of passes from the target quadrant was examined and significant increases were observed in both Walnut + Bogma Raki group and Walnut group compared to Bogma Raki group ($p<0.05$). In recall period, the time point of first pass to target quadrant were significantly shorter in Walnut group compared to Bogma raki group ($p<0.05$).

Conclusions: We found that walnut consumption can help developing reference memory, spatial learning and improve memory damages related to Bogma Raki consumption.

PT17
ESTIMATING GLOMERULAR FILTRATION RATE BY DIFFERENTFORMULAS

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Background: Since 2012, as recommended by KDIGO and NKF, CKDsare divided into 6 stages based on the three criteria: cause, GFR, albuminuria. Despite the need ofestimating GFR, there is no practical method of direct measurement. In recent decades a new protein markercystatin C,has been used to estimate GFR.Formulasfor estimating GFR - MDRD and CKD-EPI with creatinine only, cystatin C only or with both biomarkers are continuously modified as they include age, gender, race,and marker concentration.

Materials and methods: GFRhas been estimated in 617 individuals: 153 clinically healthy persons; 152 patients with type 2 diabetes mellitus without hypertension; 150 patients with essential hypertension; 162 patients with type 2 diabetes mellitus andconcomitant hypertension. Allpeople enrolled in the study have beentested for: albumin in urine, albumin% in total protein, albumin/creatinineratio (ACR), protein/creatinineratio (PCR); serum creatinine (Jaffe method) and cystatin C (PETIA). To estimate GFR the following equations are used: MDRD with creatinine only, CKD-EPI with creatinineonly, CKD-EPI with cystatin only, CKD-EPI with creatinine and cystatin C.

Results: We found out differences between the different equations, but the mean value of GFR was reduced in all patients. The formula with cystatin C only was with the highest values in control group and in patients with diabetes mellitus and hypertension. The equations with creatinine showed underestimation in low levels and overestimation in high levels of GFR, from 5 to 11%. Clinical reliability for the accurate measurement of GFR wasalso complemented by ROC curves in patients. The combined equation was with the highest diagnostic efficacy based on ROC curves. GFR correlation with albumin is strongest with the combined formula and weakestwith MDRD.

Conclusion:Upon data verification it was found that the simultaneous use of creatinine-cystatin Cequation is more effective than applying equations using these biomarkers alone. It was found that formulas with creatinine were not precise enough and lead to over-diagnosis of CKD.

PT18
THE DETERMINATION OF THE HEMOLYSIS INDEX IN DIFFERENT TUBE BRANDS

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Background: Hemolysis is the distribution of the red cell membrane resulting in the release of hemoglobin and may be the consequence of intravascular events or may ocur subsequent to or during blood collection. Hemolysis is one of the most common preanalytical errors affecting test results. We aimed to investigate whether tube brand is affect hemolysis or not.

Materials and Methods: Blood specimens were collected by venipuncture using a 21G needle in to 99

tube sets. Each set consisting of three different brand tubes: HemaTube (First tube), BD Vacutainer® SST™ II Advance (second tube), Greiner VACUETTE® (third tube). All tubes were mixed by 3 inversions after fill and allowed to clot for 30 minutes. All tubes were centrifuged at 1800xg, for 10 minutes in swing bucket centrifuges at 22°C. After centrifugation, serum from each tube was analyzed on Cobas 6000 Roche chemistry analyzer for serum index including hemolysis. Data was analyzed using one-way ANOVA for repeated measures.

Results: There were significant differences among three tubes regarding to hemolysis ($p=0.0001$). Hemolysis was higher in first tube ($p=0.016$) and third tube ($p<0.0001$) than in second tube. Also in first tube, hemolysis was lower than third tube ($p=0.032$).

Conclusion: Finally, we can say that there were significant differences between different tube brand regarding to hemolysis, and this situation can be considered when the test results are evaluated.

PT19
THE VITAMIN D EFFECT ON THE ENERGY LEVEL OF MCF-7 BREAST CANCER CELL LINE

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Vitamin D is well known as a modulator of calcium and bone metabolism. For the past three decades, abundant evidences have been accumulated to indicate that the active form of vitamin D (1*α*,25-dihydroxyvitamin D_3,1*α*,25$(OH)_2D_3$) or calcitriol, possesses many actions which werenot associated with calcium and bone metabolism. They include anticancer effects such as antiproliferation, antiangiogenesisproapoptosis andprodifferentiation.We questioned howthese effectschanged energy levels of MCF-7 cells.

ATP is the primary energy transporter for most energy-requiring reactions that occur in the cells.The AMP levels within the cytosol provide a better indicator of the rate of ATP utilization than the ATP concentration itself. The concentration of AMP in the cytosol is determined by the equilibrium position of the adenylate kinase reaction.2ADP ↔AMP+ATP

Vitamin D applied MCF-7 cellswere collected at 24th, 48th and 72th hours after Vit. D treatment.ATP,ADP and ADP results were obtained ,through HPLC device and standardized byusing cell lysate protein levels..We observed thatin the groups to which was applied vitamin D,i.e. the 24hr, 48hr and 72hr groups the AMP levels were increasing by time in comparison to control groups therefore highlighting an ATP consumption. Asaconsequence when vitamin D is considered as anti-proliferative, the increase in energy consumption makes us to think that this decrease in ATP level may be led by apoptotic process.

Understanding the features and complexity of the cancer energy metabolism will help to develop new approaches in target therapy of breast cancer.

PT20
EVALUATION OF ARCHITECT (CMIA) AND STRATUS CS (FEIA) FOR MEASUREMENT OF CARDIAC MARKERS HS TROPONIN I AND TROPONIN I

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Background: In our study we have investigated the level troponin I (TnI) and hs troponin I (hsTnI) in patients with acute coronary syndrome (ACS) using two methods chemiluminescentmicroparticle immunoassay (CMIA) Architect (Abbott) and fluorimetric immunoassay (FEIA) Stratus CS (Dade Behring).

Subjects and methods: Blood samples of 40 ACS patients have been analysed 3 hours after hospital admission.The Architect (Abbott) used CMIA method with cut off for hsTnI 0.0-35 pg/mL and Stratus CS (Dade Behring) usedFEIA method withcut off for TnI 0.0-0.07 pg/mL.

Results: Correlation between TnI and hsTnI blood levels was r = 0.941. Regression equation revealed a slope of 0.0012 and a y axis intercept of 0.0026.. The mean serum concentrations of TnI and hsTnI were 0.01 pg/mL and 10.82 pg/ml, respectively.The five of all 40 serum samplers were likely to be true Stratus CS negatives because cardiac events during follow-up were not detected.The mean concentration of hsTn I with no detected cardiac events in Architect assay was 5.7 pg/mL and for TnI Stratus CS was 0.0 pg/mL. The mean concentrations of hsTnI (Architect assay)and TnI(Stratus CS)in patients with detected cardiac eventswere 16868.2 pg/mL and 0.83 pg/mL, respectively.

Conclusions: Use of hsTnI and cTnI assays in patients with suspected ACS provides useful diagnostic information. Application of the relative change in hsTnI or cTnI concentration within 3 hours after admission in combination with the 99th percentile diagnostic cut off value on admission improves specificity and may facilitate an accurate early rule-in of ACS.

PT21
EFFECT OF FLUOROSIS OVER PARAOXONASE ENZYME ACTIVITY

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Background: The study was aimed to investigate the effect of fluorosis over paraoxonase enzyme activity. The outcome of taking high amount of fluorine, also known as fluorine poisoning, is called 'fluorosis'. Paraoxonase, a calcium-dependent hydrolase enzyme, is A group of orildialkilphosfotase according to Aldrige classification system.

Materials and methods: In this study, 15 sheep, between 3-4 years old, carrying fluorosis signs was named as fluorosis group, 10 healthy sheep as control group. Paraoxanase enzyme activity and lipoprotein levels were determined from serum samples.

Results: According to the results of analysis, paraoxanase activity in fluorosis group compared to control group was statistically higher. ($p<0,05$).

There was a significant increase in triglyceride level in fluorosis group compared to the control group. ($p<0,001$). Cholesterol level, one less in fluorosis group compared to the control group ($p<0,001$); VLDL level, one more in fluorosis group compared to control group ($p<0,05$); LDL level, significantly higher in control group compared to fluorosis group ($p<0,005$) were observed. Also HDL level was not significant change ($p>0,05$).

Conclusions: Consequently, paraoxonase enzyme activity and serum lipid profile are influenced in sheep with fluorosis; triglyceride, cholesterol and LDL levels are less; VLDL level and paraoxonase enzyme activity are higher; however there is no significant change in HDL levels.

PT22
EVALUATION OF LIPID PEROXIDATION AND ANTIOXIDANT ENZYME ACTIVITIES IN HEMODIALYSIS PATIENTS

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Background: Intravenous application of iron preparations which is a routine treatment of anemia in hemodialysis patients with end-stage renal disease can lead to iron overload in the body. Redox-active iron can catalyse the formation of hydroxyl radicals and initiation of lipid peroxidation, increase oxidative stress and speed up the development of complications in these patients. In this study, we determined the markers of lipid peroxidation, protein oxidation and antioxidant enzyme activities (superoxide dismutase, glutathione peroxidase, glutathione S-transferase) in serum of patients with end-stage renal disease on hemodialysis, who had received repeated treatment of iron supplementation.

Patients and methods: The study included 29 patients undergoing regular hemodialysis treatment. These patients were divided into three groups according to the serum ferritin levels: group I (serum ferritin between 100 and 300 μg/L); group II (serum ferritin between 301 and 600 μg/L), and group III (serum ferritin above 601 μg/L).

Results: The serum of patients with the highest concentration of serum ferritin and iron contained significantly higher levels of lipid peroxidation products, total hydroperoxides and malondialdehyde and advanced oxidation protein products and the lowest concentration of sulfhydryl groups, reduced glutathione and total antioxidant capacity.

Conclusion: Based on the obtained results, it can be concluded that iron supplementation in hemodialysis patients and consequently body iron overload of exacerbated oxidative stress have already been present in these patients.

PT23
THE EFFECT OF THYMOQUINONE TREATMENT ON NUCLEAR FACTOR KAPPA B (NF-KB) AND DNA DAMAGE ON EXPERIMENTAL DIABETIC RATS

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Background: In this study, effects of nuclear factor kappa B and formation of DNA damage on detection of the possible occurrence of complications depending on experimental diabetics were investigated. The evaluation of the possible role of thymoquinone (TQ) was aimed in the prevention and treatment of these complications.

Materials/subjects and methods: For this purpose, 28 male Wistar-Albino rats weighing between 200-250 g were used. Each containing of seven rats, the rats were divided into four groups of control (C), TQ (T), diabetes (D) and diabetes + TQ (DT). TQ was administered as 30 mg/kg/day by oral gavage to the rats in DT and T groups. In the blood samples collected after 21 days of trial, the values of glucose, HbA1c, ALT, AST, GGT, urea, uric acid, creatinine were measured as well as the quantities of NF-KB and 8-OHdG.

Results: It was determined that glucose levels were increased significantly in D group ($p < 0.05$), decreased significantly and approached to control group in DT ($p < 0.05$) and decreased in the group in T group was compared to the control group ($p < 0.05$). HbA1c levels were significantly increased only in the diabetic group ($p < 0.05$), and decreased in DT and approached to the control. It was observed that ALT and AST activities were increased significantly in D group ($p < 0.05$), while significantly decreased in DT group closing to the control. GGT activity was the highest in the D group ($p < 0.05$) but decreased significantly in DT group compared to D group ($p < 0.05$). Urea concentrations were the highest in D and the lowest in T ($p < 0.05$) while decreased significantly in DT group compared to the D group ($p < 0.05$). DNA damage were increased in both of the diabetics, but, was not statistically significant. NFκB levels were the highest in the diabetic group ($p < 0.05$), while there was no important difference in TQ and DT groups compared to the diabetic group.

Conclusions: As a result, it was observed that increased glucose and HbA1c levels by STZ-induced diabetes and indicators of liver and kidney damages were decreased significantly and approached to the control group following the administration of TQ. It was determined that 8-OHdG which is an indicator of DNA damage and NFκB levels were increased in the D group.

PT24
FOLLOWING THE SERUM LEVELS OF CREATINE KINASE IN OVERT AND SUBCLINICAL HYPOTHYROIDISM

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Background: The aim of this study was to determine serum levels of creatine kinase (CK) in overt and subclinical hypothyroidism. To investigate the change in CK levels with treatment and to evaluate the relationship between free triiodsothyronine (FT3), free thyroxine (FT4), and thyrotropin (TSH) levels and the degree of skeletal muscle involvement, as determined by serum CK

levels. Patients with other causes of CK elevation were excluded.

Subjects and methods: We included 26 patients (24 women and 2 men, ages 40.65 +/- 12.55 years) with overt hypothyroidism, 36 patients (35 women, 1 man, ages 41.55 +/- 10.45 years) with subclinical hypothyroidism, and 30 age- and gender-matched controls (27 women, 3 men, ages 40.81 +/- 11.20 years) in the study. Serum levels of TSH, FT4, FT3, and CK were measured in all subjects.

Results: CK elevation was found in 17 patients (58%) with overt hypothyroidism and in 4 patients (10%) with subclinical hypothyroidism. Although a statistically significant elevation of CK levels was found in patients with overt hypothyroidism when compared with patients with subclinical hypothyroidism and controls (p=0.0001, p=0.01, respectively), no difference was found between the subclinical hypothyroidism and control groups (p = 0.14). In hypothyroid (overt and subclinical) patients, a positive correlation was found between CK and TSH (r = 0.422; p = 0.04), and a negative correlation between CK and FT3 (r = - 0.526; p = 0.002) and between CK and FT4 (r =0.437; p = 0.04).

Conslusions: CK levels decreased to normal levels after thyroid function normalized with treatment. In conclusion, skeletal muscle is affected by hypothyroidism more profoundly in cases of overt hypothyroidism, less so when subclinical hypothyroidism is present.

PT25
EVALUATION OF OXIDATIVE STRESS STATUS IN SCHIZOPHRENIA PATIENTS SUBJECTED TO ANTIPSYCHOTIC DRUG TREATMENT

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Background: Schizophrenia is a mental disorder that affects almost 1% of the world population. Oxidative stress has been suggested to contribute to the pathophysiology of schizophrenia. Antipsychotics are the cornerstone of pharmacological treatment for schizophrenia.

Materials and methods: We investigated the effects of antipsychotics on serum oxidative stress status. 44 schizophrenia patients were enrolled in the study whose serum samples were obtained on admission before treatment, and after treatment with antipsychotics. The levels of oxidative stress status biomarkers such as protein carbonyl (PCO), total thiol (T-SH), protein thiol (P-SH), non-protein thiol (NP-SH), advanced oxidation protein products (AOPP), lipid hydroperoxides (LHP) and malondialdehyde (MDA) were measured.

Results: The levels of PCO and AOPP as the protein oxidation biomarkers and the levels of LHP and MDA were found to be significantly lower with antipsychotic treatment. On the other hand, in redox sensitive thiol group containing biomarkers; a slight decrease in NP-SH levels and a slight increase in P-SH levels were found in serum samples obtained after treatment

Conclusions: In conclusion, antipsychotics seem to be effective on serum oxidative stress status and the decreased antioxidant defense can be suggested as probably existing later in patients on chronic treatment with antipsychotics which may not have a direct effect on antioxidant defense system. In this respect, identifying valid therapeutic strategies to reduce oxidative stress could be another therapeutic target in the clinical course of the disorder.

PT26
GLUTATHION PEROXIDASE ACTIVITY AND INFLAMMATION MARKERS IN METABOLIC SYNDROME

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Background: Metabolic syndrome (MetS) is a risk factor ofdiabetes and cardiovascular disease, both of which are associated with increased oxidative stress and inflammation. Therefore, we aimed to determine glutathion peroxidase (GPx) activity and inflammation markers in overweight/obese women with MetS and to investigate their potential association with MetS components.

Subjects And Methods: A total of 100 overweight/obese women, mean age 56.7± 4.8 years (49 without MetS and 51 with MetS), and 50 age-matched normal weight controls were included. MetS was defined using International Diabetes Federation criteria. Blood pressure, anthropometric and biochemical parameters (GPx, glucose, insulin, lipid and inflammatory markers (C-reactive protein (CRP), fibrinogen, white blood cell count (WBC)) were measured. Insulin resistance (HOMA-IR) was calculated.

Results: Overweight/obese women with MetS displayed decreased GPx activity ($p=0.040$), but higher WBC, CRP and fibrinogen level ($p=0.035$, $p<0.001$, and $p=0.020$), compared with the group without MetS and with normal weight group. In multiple regression analysis waist circumference correlated independently with CRP (Beta=0.410, $p<0.001$). HOMA-IR (Beta=0.225, $p=0.007$) and triglycerides (Beta=0.188, $p=0.023$) were independent predictors of fibrinogen, and triglycerides were independent predictor of WBC (Beta=0.250, $p=0.012$). GPx did not correlate with any of MetS components.

Conclusion: Decreased antioxidant defence and association between inflammation biomarkers and MetS components in overweight/obese women were observed. Weight loss programs may be of benefit in reduction of inflammation and decreasing the prevalence of MetS.

PT27
THE EFFECT OF RAMADAN FASTING ON SERUM LIPID PROFILE AND INSULIN RESISTANCE IN HEALTHY SUBJECTS

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Background: The effects of Ramadan fasting on serum lipid profile are contraversial. In this study, we aimed to investigate the effects of Ramadan fasting on lipid profiles and insulin resistance (IR).

Materials and Methods: The study included 39 healthy subjects (19 women, 20 men, meanage: 33.1±5.9 years). Blood samples were obtained on the first and last days of Ramadan fasting. Serum insulin, glucose, total cholesterol (TC), high density lipoprotein cholesterol (HDL-C), low density lipoprotein-C (LDL-C) and triglyceride (TG) levels were measured. Homeostasis model assesment-IR (HOMA-IR) was calculated to evaluate IR.

Results: While TC and LDL-C levels on the last day of Ramadan fasting were significantly higher compared with the first day value (p=0.001 and p=0.0001respectively), there were no significant diffrences in TG and HDL-C levels between the first and last days of Ramadan. In the first day of Ramadan, HOMA-IR was lower than in the last day (p=0.03). BMI values did not change in the last day of Ramadan when compared to first day values.

Conclusion: In Ramadan fasting, an increase in TC, LDL-C and IR may be seen. These results may be due to changes in the food habits and the amount and composition of meal consumed by the subjects in our region at Ramadan. Our people increase their intake of carbohydrate and fat during Ramadan. Thus, it is important to pay attention to amount and content of diet consumed in Ramadan.

PT28
SERUM HEPCIDIN LEVELS IN B–THALASSEMIA PATIENTS

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Background: Systemic administration of blood transfusions is the reason for the growing number of adults with congenital anemia and iron overload. β-thalassemia is a genetic disorder that is due to a reduced or complete absence of synthesis of β-globin chain in a molecule of hemoglobin. Disproportion in the synthesis of α- and β-chains leads to the formation of unstable aggregates followed by precipitation, haemolytic anemia and ineffective erythropoiesis.The aim of our study was to determine serum levels hepcidin in patients with β-thalassemia and certain diagnostic his importance to the therapeutic approach and its correlation with damage to the cardiovascular system.

Subjects and methods: For a period of one year we quantified serum hepcidin levels using verified method for Bulgarian population. We included 51

patients of average age 29.9 ± 3.7 (M 62.5%, 37.5% G).

Results: We detected a statistically significant decreases of hepcidin to the control group (P <0.001). Hepcidin concentration of serum in patients with β-thalassemia is 0.77 μg/L ± 0.2, compared to the control group, 19.9 μg/L ± 1.7.

Conclusion: Established low serum hepcidin levels in patients with β-thalassemia, directed to the correct therapeutic behavior of hepcidin agonists supporting chelating therapy. Transfusion therapy must be balanced. Its individualization can be done by monitoring the serum levels of hepcidin and determining the indices relative to the soluble transferrin receptors.

PT29
HEMATOLOGIC PARAMETERS IN HEALTHY SUBJECTS WITH RAMADAN FASTING

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Background: The aim of this study was to evaluate the hematologic parameters in healthy subjects with Ramadan fasting.

Materials and Methods: Thirty eight healthy subjects were recruited and evaluated on the first and last days of Ramadan fasting in terms of hematologic parameters including erythrocyte sedimentation rate (ESR), red blood cell (RBC), white blood cell (WBC), platelet (PLT), hemoglobin (Hb),and PLT indices. Paired t test or Wilcoxon test were used to compare hematologic parameters between the first and last days of Ramadan.

Results: In the first day of Ramadan, ESR, RBC, PLT and Hb values were similar to the values of the last day of Ramadan. WBC count was significantly higher in day 1 than in the last day of Ramadan (p=0.0001). Mean platelet volume (MPV) values was signigicantly higher in the first day than in the last day of Ramadan (p=0.02).

Conclusion: Because MPV is an indicator of PLT reactivity, in Ramadan, fasting patients with coagulation disorders should pay attention to this issue.

PT30
SCREENING FOR CHRONIC KIDNEY DISEASE IN ADULTS IN VOJVODINA

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Background: Chronic kidney disease (CKD) is often underdiagnosedand represents significant medical problem. The aim of the study is to screenfor CKD in Vojvodina in the active working population.

Subjects and methods: The study included3153 male i 429 femalesubjectsbetween 18-65 years of age who performedphysical examinations in The Institute of Occupational Health, Novi Sad.All the patients underwent measurement of arterial blood pressure, chemical(commercial test strips Acon

Laboratories Inc., USA)and microscopic examination of first morning urine sample,determination of serum levels of creatinine, blood urea nitrogen, uric acid and glucose (standard biochemical methods). In study participants who were at an increased risk for CKD, as well as in a number of study subjects without risk factors, ratio albumine/creatinine in urine (A/C ratio) was performed. Glomerular filtration rate (GFR (ml/min/1.73 m^2)) was calculated by using CKD-EPI formula.

Results: Pathological result of first morning urine test was present in about 19.3% subjects. Glomerular filtration rate below 60 ml/min/1.73 $m^2$was present in 1.6%, 60-89ml/min/1.73 m^2in 43%, above 90 ml/min/1.73 m^2 in 55.4% of the subjects.A/C ratio (mg/mmol) was >3 in 9.5% subjectsand the most of the subjects (86.6%) had A/C ratio of 3-30 mg/mmol. Most the of subjects (75%) with GFR<60 ml/min/1.73 m^2 and/or A/C>3 mg/mmol and/or pathological result of first morning urine test have already had arterial hypertension (HTA) and/or diabetes (DM).Of the remaining one fourthof the participants, half had high blood pressure and/or fasting glycemia>7 mmol/l at the time of test without diagnosis ofHTA or DM.23.3% of all the subjects already had diagnosed HTA, and 3.7% of them had DM. 4% of subjects without any earlier diagnoses have had fasting glycemia ≥7 mmol/l.

Conclusions: Pathological result of first morning urine is present in about 1/5, while GFR<60 ml/min/1.73 $m^2$is present in 1.6% of population of Vojvodina. The most subjects with GFR<60 ml/min/1.73 m^2suffernig from HTA and/or DM. Detection of patients with increased risk for CKD,primarily with HTA and DM,and examination of their renal function should be priority in prevention of devlopment and progression of CKD.

PT31
ANALYSES OF SAMPLES REJECTED IN PREANALYTICAL PROCESS

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Introduction and Aim: The preanalytical phase is the most common source of laboratory errors. Continuous monitoring and management of preanalytical errors is therefore crucial to the quality of laboratory performance, and also required for all clinical laboratories accredited in accordance with ISO 15189 quality standard.Preanalytical phase errors are waste of time and money which are; improper labeling, hemolyzed samples, lipemic samples, clotted samples, inappropriate container, insufficient sample and damaged sample. Based on these errors quality indicators are determined. Process performance is evaluated by comparing calculated quality indicators with target values. In this study, our aim is to evaluate common errors in pre-analytical process and use them as quality indicators.

Materials and Method: Data were collected monthly for the period of January 2015–April 2015. For every type of error monthly percentages have been calculated and evaluated according to the Quality Indicators (QIs) developed by the IFCC Working Group on ''Laboratory Errors and Patient Safety'' (WG-LEPS).

Results: Quality indicators calculated according to each error type in pre-analytical process have been detected

above "optimum performance" level according to quality targets. The highest error rates; "clotted sample" was in the first, "hemolyzed sample" was in the second place among the highest error rates for january, "hemolyzed sample" was in the first, "clotted sample" was in the second place among the highest error rates for february and march. The lowest error rate belonged to"the damaged sample"for each month.

Conclusion: Our results showed that quality indicators may be useful for evaluation of pre-analytical process. According to the quality indicators that could not achieve the target, the origin of the errors can be determined, corrective and preventive actions can be carried out.It is important to monitor the complete testing process, including the pre-and post-analytical phases, where many errors occur.

PT32
BNP RELEASE IN ACUTE DECOMPENSATED HEART FAILURE WITH REDUCED AND PRESERVED EJECTION FRACTION

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Background*:* Recently, it has been recognised that up to 50 % patient with heart failure have preserved ejection fraction. BNP is elevated in acute heart failure and aim of this work was to investigate possible differences in BNP release in patient with reduced and preserved ejection fraction (HFREF and HFPEF).

Subject And Methods*:* In our study 42 patients were hospitalised in Intensive Care Unit at Cardiology Clinic inacute heart failure, two of them were exluded from study because of intrahospital death within 72 hour from admission. Blood for BNP was taken twice-on admission (BNP1) and on the discharge (BNP2). We also calculated percentage of BNP reduction at discharge compared to admission values.

Results: Among the whole population,40 patients had reduced sistolic function (%) with mean LVEF 39.27 +/- 10.95 and 41 patient had preserved ejection fraction, mean LVEF 35.85 +/- 13.40. BNP1 mean 1658.43 +/- 1497.23 pg/mL was higher in HFrEF compared to HFpEF, as well as discharge mean BNP2 was 667 +/- 704.84 pg/mL.Percentage of BNP reduction (BNP1-BNP2/100) was from 10 to 93 % and mean 56.2% +/- 23.64 and it was higher in HFrEF compared to HFpEF. The main difference between BNP1 and BNP2 was statistically significant for $P < 0.05$ according to Student t-test. Correlation coefficient was $r = 0.935$ and the regression equation revealed a slope of 0.2921 and y axis intercept of 122.48.

Conclusion: In acute heart failure BNP levels werehigher in patients with reduced EF compared to patients with preserved EF. Percentage of BNP reduction at discharge was higher in HFrEF compared to HFpEF. The acute heart failure neurohumorale activation is more pronounced in subpopulation of patient with reduced ejection fraction.

PT33
ANALYSES OF SAMPLES REJECTION ACCORDING TO SIX SIGMA METHODOLOGY

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Background: In clinical laboratories, preanalytical process included all processes from the time a laboratory request is made by a physician until the sample is ready for testing. If an error is detected, samples must be re-collected, procedures are delayed, and time is wasted. According to six sigma methodology process sigma level is the indicator of efficiency and cost effectiveness and makes a holistic point of view to the process. Preanalytical process sigma levels are calculated according to pre-analytical process errors. These errors are waste of time and money which are; improper labeling, hemolyzed samples, lipemic samples, clotted samples, inappropriate container, insufficient sample and damaged sample. In this study, we aimed to assess the pre-analytical process performance according to six sigma methodology.

Materials and method: Preanalytical process error data between January 2015–April 2015 were obtained from the laboratory information system. Monthly process sigma levels were calculated for every type of error. We have specified the target quality performance level as 4.6 sigma which has a waste rate of%10 with 1000 DPM.

Results: For each error type, process sigma levels were found above the target for each month (4.6). Pre-analytical process error with the lowest process sigma level was "hemolyzed sample", the highest was "damaged sample".

Conclusion: Our study showed that errors with low process sigma levels in our laboratory can be determined and these errors may be evaluated as a whole with analytical and post analytical processes. Six sigma methodology may provide a detailed assessment of measurement processes with preanalytical process sigma levels and controlling the variables. The origin of the errors that have low sigma levels can be determined, corrective and preventive actions can be carried out immediately.

PT34
CORRELATION BETWEEN LIVER ENZYMES AND PALMITIC ACID REGARDING GLYCEMIC CONTROL IN TYPE 2 DIABETIC PATIENTS

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Background: It has long been proposed that elevation of liver enzymes including alanine aminotransferase (ALT), aspartate aminotransferase (AST) and γ-glutamyltransferase (GGT) and increase in free fatty acids (FFAs) levels may be associated with insulin resistance (IR) and Type 2 *diabetes mellitus* (T2DM). Limited number of studies have examined the association of liver enzymes and FFAs with T2DM control. In this study we examined correlation of liver enzymes activities and palmitic acid (saturated fatty acid, SFA) levels in T2DM patients with good and poor control.

Subject and methods: We analyzed the activities of ALT, AST (aspartate amino transferase), GGT and palmitic, C16:0 levels, fasting plasma glucose (FPG) in 71 T2DM patients (36 good and 35 poor control), and 40 age-matched healthy controls. All subjects included in this study were free of evidence of hepatitis, viral infection, or active liver and kidney damage. Standard IFCC enzyme protocols were used to determine enzyme activities on the Alcyon analyzer, while concentrations of palmitic acid were determined by gas chromatography.

Results: As espected, the results showed a significant positive correlation that was observed between liver enzyme activity and palmitic acid levels in Type 2 diabetic patients ($p < 0.05$). Interestingly, significant correlation was observed between palmitic acid and ALT activity in diabetic patients with good control of T2DM ($p < 0.05$) while, a negative correlation between palmitic acid and GGT ($\rho = -0.012$) was demonstrated in patients with poor control of T2DM.

Conclusion: Our data suggest relevance of sinchronous monitoring of liver enzymes particularly ALT and palmitic acid levels, in order to achieve proper control of the disease.

PT35
ASSOCIATION BETWEEN PLASMA MPO AND MMP-9 LEVELS AND RISK OF CORONARY ARTERY DISEASE

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Background: Coronary artery disease (CAD) is the most common cause of atherosclerosis lipoprotein particles in a solid and/or through the intima layer of the accumulation of dysfunctional vascular endothelium is an inflammatory event. Myeloperoxidase (MPO) is heme peroxidase, which is produced from

granular leukocytes. It also promotes oxidative damage of host tissues at sites of inflammation, including atherosclerotic lesions. We investigated the association between plasma myeloperoxidase (MPO) andMatrix Metalloproteinase-9 (MMP-9) levels in CAD patients and control subjects.

Materials and Methods: The case group consisted of 88 patients who had angiographic ally proved atherosclerotic plaques in their coronary arteries. The control group consisted of 88 individuals who did not have a history of coronary artery disease. The MPO and MMP-9 levels were measured usingEnzyme-Linked Immuno Sorbent Assay (ELISA) method.

Results: When compared to the controls, significant increases in systolic blood pressure, total cholesteroland LDL-Clevels were found in CAD patients. Gender, diastolic blood pressure, hypertension, triglycerides, HDL cholesterol, and VLDLcholesterol levelsdid not differsignificantlybetween the groups. Plasma MPO levels were significantly greater in patients with CAD than in controls ($p < 0.05$). Plasma MMP-9 levels isdecreased in patients withCAD than controls andthis changewas notstatistically significant.

Conclusion: Elevated levels of plasma MPO are associated with the presence of CAD. These findings support a potential role for MPO as an inflammatory marker in CAD and may have implications for atherosclerosis diagnosis and risk assessment.

PT36
COMPARISON OF INTACT PARATHYROID HORMONE (IPTH) ON VITROS ECIAND ABBOT ARCHITECT CI 8200

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Background: We compared the values of IPTH, obtained on Vitros ECI (Orhto Clinical Diagnostics, Jonson and Jonson, UK) and Architect ci 8200(Abbott Diagnostics) as a part of method validation for the new analyzer.

Materials and methods: The study included 49 serums for IPTH. We evaluated concentration level between two analyzers that works on chemiluminescence (CMIA). Vitros ECI is immunochemical analyzer that works on indirect CMIA, but Abbott –Architect ci8200 is immunochemical analyzer that works on direct CMIA. We performed statistical comparative analysis using program SPSS v.21

Results: Thecorelation coefficient(r) between VitrosECI and Architect ci 8200 was: IPTH (N=49; r=0,98)

Conclusion: Data obtained by Passing-Bablok regression showed that the results are completely comparable and they follow the same linearity

PT37
DETERMINATION OF METHYLMALONIC ACID IN BIOLOGICAL FLUIDS BY LIQUID-CHROMATOGRAPHY-MASS SPECTROMETRY (LC-MS/MS)

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Background: Methylmalonic acid (MMA) is a metabolic intermediate in the conversion of propionic acid to succinic acid (SA). Measurement of MMA has become an important diagnostic procedure in clinical laboratories because of the increased evidence that elevated MMA is a marker of cobalamin deficiency.

Materials and methods: For serum methylmalonic acid measurement, 100 µL of internal standard (d3-methylmalonic acid) in water was added to 1000 µL standart or serum and centrifuged at 13.000 rpm for 10 minutes to remove the precipitated proteins. The supernatant was collected and dried under a nitrogen gas flow at 60 ◦C. Derivatisation step was performed dissolving the dried extract in 200 µLof a freshly prepared butanol solution containing 5% (v v−1) acetyl chloride and kept at 65 ◦C for 15 minutes. The solvent was removed by evaporation under nitrogen flow at 65 ◦C. The derivatised samples were dissolved in 100 µLof water–methanol (90:10, v v−1) containing 0.1% (v v−1) formic acid and 40 µL was injected into the ultra performance liquid chromatography analytical column for chromatography.

Results: The methylmalonic assay was linear up to 200 µmol/L. Interassay CVs were 6.7%, 5.0%, and 5.0% for mean concentrations of 0.15, 0.36, and 0.65 _mol/L, respectively.

Conclusions: This method is accurate and precise. The short and fast run time, the feasibility of high sample throughput and the small amount of sample required make this method very suitable for routine analysis in the clinical setting. This method may be used for determining the vitamin B12 deficiency. Also urine levels of methylmalonic acid may be used in follow-up of methylmalonic acidemias by this method.

PT38
EVALUATION OF LEVELS OF TUMOR MARKER CA-125 IN PATIENTS OPERATED BECAUSE OF OVARIAN ENDOMETRIOMAS

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Background: Carcinoembrional antigen (CA-125) is the most widely used serum marker of endometriosis. Endometriosis is defined as the presence of endometrial-like tissue outside the uterus. Ovarian endometrioma is cystic manifestation of endometriosis.The aim of this study is to evaluate levels of CA-125 before, three and six months after laparoscopic operation because of ovarian endometrioma.

Subjects and methods: In a prospective study serum levels of tumor marker for endometriosis (CA-125) were evaluated for 50 patients underwent laporoscopic surgery at University Clinic for gynecology and obstetrics in Skopje in period from 01.01.2013 to 01.01.2015. Immulite 2000 OM-MA chemiluminescent immunometric assay was used for measurement of serum

levels of CA-125.A paired Student's t-test was used to test differences between levels before and three and six months after surgery. P value less than 0.05 was considered as statistically significant. SPSS ver.12 was used in this analysis.

Results: The mean age of the patients was 30.57±5.59 years. The mean level of CA-125 before surgery was 32.4±9.8 mIU/ml and was reduced to 19.5±2.5mIU/ml three months after and 15.4±2.8 six months after surgery.The difference was statistically significant (t_1=5.05, $p_1 \leq 0.01$; t_2=6.77, $p_2 \leq 0.01$).

Conclusion: This study results suggest that serum levels of CA-125 were reduced after laparoscopic surgery with extirpation of ovarian endometrioma and could be used for investigation of these patients in the future.

PT39
DETERMINATION OF SERUM ANDROSTENEDIONE BY LIQUID-CHROMATOGRAPHY-MASSSPECTROMETRY (LC-MS/MS)

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Background: Adrenal glands are the major source of androgens and androgen hormones in reproductive- aged and postmenopausal women. Androstenedione is produced in large vast by both the adrenal glands and gonads. Overproduction of androstenedione can be caused by the lack of adrenal steroid biosynthesis, tumors of ovarian and adrenal origin, polycystic ovarian syndrome, increased peripheral sensitivity to androgens, and increased peripheral production of androgens. In addition to improved accuracy and sensitivity for steroid measurements, liquid chromatography tandem mass spectrometry (LC-MS /MS) can distinguish compounds and can therefore be used to quantify multiple steroids from one sample. The aim of this study was to determine androstenedione by LC-MS/MS system.

Materials and methods: For serum androstenedione measurement, 50 µL of internal standard (d5- 11 deoksikortizol) in methanol was added to 250 µL standart or serum and centrifuged at 13.000 rpm for 10 minutes to remove the precipitated proteins. Supernatant was transferred to clean tubes and this procedure was performed twice. The supernatant wasc ollected and dried under a nitrogen gas flow at 60 ◦C and dissolved in mobile phase. 60 µL was injected in to the ultra performance liquid chromatography analytical column for chromatography.

Results: The androstenedione assay was linear up to50µmol/L. Lower limit of quantitation and lower limit of detection were 0.195 ng/mL (signal to noise ratio=16.2) and 0.097 ng/mL (signal to noise ratio=7.4), respectively.

Conclusions: By this method, accurate and sensitive measurement of androstenedione was analyzed with LC-MS/MS system. This method has advantages like improved sensitivity and linearity. Data from calibration curves reveal that this method can be used in routine procedure.

PT40
POSTGRADUATE TRAINING IN MEDICAL BIOCHEMISTRY IN MACEDONIA

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Medical Doctors and Masters of Pharmacy may apply to postgraduate training in medical biochemistry in Macedonia. According to Bologna ECTS,MDs have 6- year and pharmacist 5-year undergraduate studies .
Postgraduate training, according to EC4European Syllabus for Post-Graduate Training in Clinical Chemistry and Laboratory Medicine, lastsfor 4 years and includes mandatory and elective courses. Candidates are required to attend 270 hours ofthe mandatory theoretical courses (medical and clinical biochemistry, haematology, transfusion, microbiology, etc.), 42 months of practice in different laboratories at the Medical Faculty, UKIM Skopje, and to prepare a specialization paper and to defense it at the day of taking the examination.
The specialization program includes courses in medical and clinical chemistry, haematology, transfusion, microbiology, genetics, biochemical aspect of pathology and oncology, and pharmacology, laboratory management and quality control in laboratories. This training program is governed by of the Ministry of Health.
After getting a diploma for specialist of medical biochemistry, the candidate is obliged to undergo a program of Continuing Medical Education (CME) and to renew the licence every 7 years at Doctors Chamber and Pharmacy Chamber of Macedonia, respectively.

PT41
DETERMINATION OF SERUM BETA-SITOSTEROL BY GAS CHROMATOGRAPHY-MASS SPECTROMETRY (GC-MS)

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Background: β-Sitosterol, an important phytosterol found in plant food, is known to exert antiatherosclerotic activity. Sitosterolaemia is a rare autosomal recessive disorder characterised by elevated plasma levels of plant sterols. The aim of this work was to develop a simple, fast and accurate gas-chromatography-mass spectrometry method for determination and quantification of β-Sitosterol.

Material and methods: For serum β-Sitosterol measurement, 100 µL of internal standard (5α-cholestane) in toluene was added to 200 µL standart or plasma in a glass tubes containing 1ml of potassium hydroxide in ethanol (1.0 mol/L). Tubes were well mixed, flushed with N2 and heated at 70 ◦C for 60 min in the dark. The reactions were stopped by cooling the tubes under running cold water. After cooling, the solution was diluted with water (1 ml) and the lipids were extracted twice with 2ml of a solution of hexane and absolute ethanol (20:1, v/v), containing 12.5 mg l−1 butylated hydroxytoluene. The samples were vortexed and then centrifuged at 3500 rpm at 20 ◦C for 10 min to accelerate phase separation; the organic phase was transferred to small glass vials, dried completely under a steam of N2. The lipid extractwas derivatized with 200 µL freshly prepared pyridine-MSTFA with (1:1, v/v). Samples were then incubated at 70 ◦C for 60 min, and finally analyzed by GC–MS.

Results: We developed an optimised analytical method for the simultaneous analysis of β-sitosterol in serum using gas chromatography-mass spectrometry (GC–MS) with multiple selected ion

monitoring (SIM). This method is based on the alkaline hydrolysis of sterol esters, extraction of free sterols and derivatization. The beta-sitosterol assay was linear up to 200 µmol/L. Interassay CVs were 8%, 6.2%, and 5.0% for mean concentrations of 1, 5, and 10 µmol/L, respectively.

Conclusions: Serum β-Sitosterol measurement can be easily performed by GC-MS system to identify the patients with atherosclerotic risk factors. This method allows accurate the pathological levels. Although long experimental procedure, this method has advantage to seperate all the peaks in a chromatogram.

PT42
PROSTATE CANCER ANTIGEN 3 (PCA3) A NEW BIOMARKER IN DETERMINATION OF PROSTATE CANCER (PCa), IN OUR EXPERIENCE

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Introduction: Prostate Cancer (PCa) is the most common male cancer in Europe. PSA test is a screeningtest for men in early detection of prostate cancer. However numerous false positive results and many unnecessary biopsies has lied to searchothermethods, one of the most promising is the Prostate Cancer Gene 3 (PCA3) a non-coding mRNA gene, whose level increases in PCa tissues.The PCA3 result is not elevated in prostate diseases, such as benign prostatic hyperplasia (BPH) or inflammation of the prostate (prostatitis). A non-invasive test for PCA3 is developed using urine collected after a digital rectal examination (DRE).The PCA3 secure to avoid many unnecessary biopsies and help to make more decisions in Prostate cancer (PCa).

Methods: This study includes164 male patients with PSA levels higher than ≥4 ng/ml (Elecsys 2010),withnegative prostate biopsy and continuing elevated values PSAhigher than ≥4 ng/mL, which are recommended to repeat prostate biopsies. Our hospital recommend PCA3 test. The PCA3 assay is a simple test, requires collection of 20-30 mL of urine samples, after an intensive prostatic massage. The PCA3 mRNA levels are determined using transcription-mediated amplification.

Results: The PCA3 test measures the PCA3 mRNA concentration in urine samples.The cut-off for a Positive PCA3 score is≥ 35. Men with PCA3 ≥ 35 have more probability to have a positive prostate biopsies. PCA3 detection in urine presented a sensitivity to 58%, and specificity 72%.

Conclusions: Prostate cancer gene 3 (PCA3) results have shown the high specificity of this test.The PCA3 assay may help to avoid many unnecessary biopsies, potential discomfort and complications (pain, bleeding and infections) for the men involved.However the decision to perform a prostate biopsy does not only depend on the PCA3 Score but should be made in combination with other prostate cancer risk factors such PSA level etc.

PT43
EFFECTS OF HİGH-FAT DİET AND ACRYLAMİDE TREATMENT ON TİSSUE OXİDANT AND ANTİOXİDANT LEVELS İN RATS

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Background: Acrylamide (ACR) is an organic chemical which occurs in foods widespreadly consumed in diets worldwide. ACR promotes the generation of ROS and the depletion of anti-oxidants. The aim of this study was to investigate liver, brain and kidney of tissue total antioxidant status (TAS), total oxidant status (TOS) and liver and brain of tissue oxidized LDL (ox-LDL) levels in long term ACR given rats, compared to control rats.

Material and methods: Forty eight male Wistar rats (5-6 weeks of aged) were segregated into eight groups (8 rats per group) and the rats in first four groups were fed with a high-fat diet (crude fat 20%) and the rats in second four groups were fed standard died (crude fat 2.7 %). Animals in each of two diet groups were given acrylamide at the doses of 0, 2, 10 and 20 mg/kg bw/day via drinking water for 28 days. At the end of the experiment tissue samples were analyzed for TAS, TOS and ox-LDL.

Results: Ox-LDL and TOS levels were increased as doses of acrylamid were elevated in liver, brain and kidney tissue, no significant difference was present at levels of ox-LDL/protein ($p=0.087$) and TOS/protein ($p=0.751$) in liver tissue among eight groups. While no significant difference was observed at levels of ox-LDL/protein in brain tissue ($p=0.808$) among eight groups, TOS/protein levels in brain tissue ($p<0.001$) became increased. No significant difference was present at level of TOS/protein ($p=0.052$) in renal tissue among eight groups. As doses of acrylamid were increased, TAS/protein levels in brain, liver and renal tissue became decreased, and a significant difference was present at TAS/protein levels among the groups ($p<0.001$).

Conclusions: Our findings show that long term treatment with 2, 10 and 20 mg/kg doses of only ACR and high-fat diet-ACR treatment led to a significant depletion of tissue TAS levels and overproduction of tissue TOS and ox-LDL levels, consequently, to an increase in oxidative stress.

PT44
ASSOCIATION OF *TCF7L2* GENETIC VARIANT (RS7903146) WITH TYPE 2 DIABETES IN POPULATIONS FROM BOSNIA AND HERZEGOVINA AND KOSOVO

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Background: Type 2 diabetes (T2D) is chronic, incurable, complex polygenicdisease, characterized by hyperglycemia. *TCF7L2,* member of Wntsignaling pathway, is a transcription factor influencing the transcription of several genes thereby exerting a large variety of functions within the cell. Selected variant of candidate gene, *TCF7L2* (rs7903146), that we analyzed in our study, is associated with dysfunction of pancreatic β cells.

Subjects and methods: The study included 638 patients with T2D and prediabetes and 360 healthy subjects as control group recruited at the Clinical Centre University of Sarajevo, University Hospital of Clinical Centre in Banja Luka, General Hospital in Tešanj and Health Centre in Prizren, both sexes, aged from 40 up to 65 years. The study examined the differences of genotype frequencies of the analyzed polymorphism between patients with T2D, prediabetes and healthy population. Also, we analyzed association of genotypes of selected candidate gene polymorphisms with clinical and biochemical parameters of T2D. Analysis of all biochemical parameters were determined by standard IFCC protocols. For isolation of genomic DNA was used Miller's protocol and Qiagen commercial test kit. Genotyping of analyzed polymorphisms was performed by MassArraySequenomiPlex platform and RT-PCR method.

Results: Result of logistic regression analyses are significant for rs7903146 polymorphism, and showed that the risk T allele was significantly associated with increased risk of T2D (OR=2.425, 95% CI 1.752-3.335, $p<0.001$). T allele of *TCF7L2* gene polymorphism - rs7903146 showed significant association with anthropometric parameters (BMI and hip circumference), blood pressure, inflammatory markers (CRP, fibrinogen and leukocytes), total proteins and GGT enzyme activity. Also it is important to note the tendency of association of T allele with insulin, HOMA IR index, waist circumference, triglycerides and total cholesterol levels.

Conclusions: Results of our study showed that rs7903146 polymorphism is significantly associated with increased risk of T2D, and with important markers of this disease.

PT45
EFFECTS OF THALİDOMİDE AND ETANERCEPTTHERAPY İN LPS INDUCED SEPSİS MODEL İN RATS

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Background: Sepsis is an excessive inflammatory systemic response created by the organism against endotoxin-producing bacteria. Hemodynamic and metabolic changes that occur in the cell as a result of sepsis lead to multiple organ failure and death by creating both endothelial damage and intracellular damage. Angiogenic factors have been implicated in the pathophysiology of sepsis. Endoglin(ENG),is a Transforming Growth Factor-β (TGF-β) co-receptor and essential for vascular development and angiogenesis, increases the formation of extracellular matrix (ECM) by triggering cell chemotaxis during inflammation. In experimental models of sepsislipopolysaccharide(LPS), there is increased expression of vascular endothelial growth factors (VEGF). Our aim was to indicate a protective role of thalidomide and etanercept, which have anti-TNF-α activityon excessive inflammatory response in sepsis-induced liver injury.

Materials and methods: Thirty rats were randomized into five groups. In Group 1, rats were sham operated. In Group 2, sepsis was induced by LPS. In Group 3, sepsis was induced and as a therapeutic agent thalidomide was given. In Group 4, sepsis was induced and as a therapeutic agent etanercept was given. In Group 5, sepsis was induced and both thalidomide and etanercept were given as a therapeutic agent. Hepatic tissue matrix metalloproteinase-9 (MMP-9) and VEGF levels were determined by Enzyme-Linked Immuno Sorbent Assay (ELISA) methodin all groups.Quantitative assessment of endoglin protein expression was performed on hepatic tissue specimen by Western blot analysis.

Results: In Group 2, tissue MMP-9, VEGF and ENG levels were statistically significantly higher than in Group 1 ($p<0.05$). Pretreatment with etanercept also significantly decreased the levels of MMP-9 and ENG in sepsis induced rats. Additionally, VEGF and ENG levels dramatically decreased in thalidomide-induced rats compared to Group 2 ($p<0.005$).

Conclusion: Thalidomide and etanercept therapy significantly improved ECM formation and angiogenesis in LPS-induced sepsis model. We expect that these treatment agents will give new perspectives into the treatment of sepsis.

PT46
LACTATE LEVELS PREDICTING DEATH OUTCOME IN SEPTIC PATIENTS

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Background: Sepsis is defined as the presence (probable or documented) of infection together with systemic inflammatory response to infection. It is life–treating illness wich causes millions of death globally each year. Lactate is one of the first and most often used biomarkers of sepsis.

Subjects and methods: In the prospective study were enrolled 86 patients who met two or more positive

criteria for Systemic Inflammatory Response Syndrome (SIRS) with the aim to analyze predictive value of lactate in sepsis. Lactate levels were measured from capillary blood samples and compare between SIRS positive noninfected (n=20) vs. patients with documented sepsis (n=66), and between septic patients with positive (n=45) vs. negative blood culture (n=21). (SOFA) and (APACHE) II score were obtained from al patients in the same time and compare with lactate levels. Lactate levels were used to predict outcome in septic patients.

Results: There was significant differences between SIRS positive noninfected patients 1,495 ±0,063 mmol/L vs. patients with documented sepsis 2,858±0,255 and between patients with sepsis and positive blood culture (HEM+) 3,243±0,348 vs. blood culture negative patients (HEM-) 1,924±0,1411. Lactate levels shown good positive correlation with SOFA (r=0,75) and APACHE II score (r=0,58). With cut off value 2,05 mmol/L (AUC 0,87) lactate levels had positive predictive value (PPV) 100% and negative predictive value (NPV) 57,14% in predicting sepsis; and at cut off 2,35 mmol/L (AUC 0,79) had PPV 85,71%, NPV 50,0%. Lactate levels had good correlation with death outcome (r=0,66); at cut off value 3,25 (0,95) had PPV 90,05 and NPV 89,47% predicting death outcome in septic patients.

Conclusions: Lactate is useful biomarker in septic patients. It is good parameter severity of sepsis, and can predict death outcome in septic patients.

PT47
ENDOTHELIAL LIPASE AND INFLAMMATION MARKERS IN CORONARY ARTERY DISEASE

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Background: Coronary artery diseases (CAD) greatly threaten the lives of people in developing countries. As a complex and multifactorial polygenetic disorder, the development of CAD is depend on multiple-risk factors. Endothelial lipase (EL) is a new member of the triglyceride (TG) lipase family, which also includes lipoprotein lipase (LPL) and hepatic lipase (HL). EL may play an important role in the pathogenesis of CAD. The aim of the present study was to investigate the effects of endothelial lipase and inflammation markers (high sensitive C - reactive protein (hs-CRP), Interleukin-6 (IL-6) and Interleukin-10 (IL-10)) in the CAD patients.

Materials and methods: This case-control study included 88 patients and 88 healthy individuals. Plasma hs-CRP, IL-6, IL-10 and ELlevels were determined by Enzyme-Linked Immuno Sorbent Assay (ELISA) method.

Results: When demographic characteristics were compared between both groups; total cholesterol levels were found increased ($p < 0.001$), whereas smoking, systolic blood pressure, and LDL cholesterol levels were found decreased ($p < 0.05$). In terms of gender, diastolic blood pressure, hypertension, triglycerides, HDL cholesterol and VLDL cholesterol levels, there was no significantly difference between the two groups. When compared to the controls, statistically significant increases in IL-6 and hs-CRP levels were found in CAD patients. Plasma IL-10 were significantly greater in controls than in patients with CAD ($p < 0.05$). Plasma

EL levels were not statistically significant differences ($p>0.05$).

Conclusion: We report that human plasma inflammation markers are significantly associated with CAD but we were not determined the relationship between EL concentrations and CAD.

PT48
ANALYSIS OF THE PLASMA FREE FATTY ACIDS COMPOSITION IN PREDIABETES

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Background: Prediabetes represents a condition of impaired fasting glucose or impaired glucose tolerance, both being risk factors for type 2 diabetes (T2D). Impaired glycemia in prediabetes is associated with insulin resistance (IR), accompanied with altered fatty acid metabolism and inflammation. Elevated free fatty acids (FFAs) disturb the normal glucose homeostasis, decrease insulin release and reduce the efficiency of glucose uptake in insulin-sensitive tissues. Studies related to FFAs composition in prediabetic individuals are lacking and therefore in this study we analysed their plasma concentration and possibly specific composition in prediabetes.

Subjects and methods: Total of 85 subjects (45 prediabetics and 40 patients classified as controls on the basis of glucose tolerance test), with no evidence of hepatitis B or C viral infection or active liver and kidney damage were recruited in this study. Classification of patients was made according to criteria used by WHO and European Association for the Study of Diabetes. Standard IFCC protocols were used for analysing glycatedhemoglobin, glucose and other biochemical parameters, while FFA composition and concentrations were determined by gas chromatography with mass spectrometry detection.

Results: The FFA compositions of the controls and prediabetic subjects were different for 5 of 14 fatty acids with 14 to 22 carbons. The most common fatty acids in both groups of patients were C16:0, C18:1 and C18:2. Compared to controls, total FFA (saturated and monounsaturated fatty acid) concentrations were higher in prediabetic patients. Interestingly in prediabetic population, C22 polyunsaturated fatty acids were not detected. Also, there were no significant differences between male and female patients. In addition, our data also showed a significant difference in C16:0 levels in prediabetics with poorly and well-controlled disease.

Conclusions: These observations indicate that changes in composition and concentrations of FFA are associated with the development of the disease, opposing the view of them being a causative factor of poor glucose control and insulin insensitivity.

PT49
IN DIABETIC AND AMINOGUANIDIN USING RATS GLYCATION EFFECTS ON CORPUS CAVERNOUS COLLAGEN TYPES

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Background: Diabetes mellitus having chronic complications, is a disease with high morbidity and mortality rate. Microvascular diabetic complications and increased advanced glycation products are esp important in diabetes and erectile dysfunction is a common problem in diabetes. We aomed to search if the increased collagen glycation in diabetes is the reason of erectile dysfunction.

Subjects and methods: We evaluated effects of aminoguanidin In stz induced diabetic 8 week old rats . By this means serum glucose, HbA1c, kidney functions, total cholesterol and triglyceride levels, osmolarity, and penile tunica albuginea 5HMF levels, to determine tissue glycation level,collagen levels and types were measured to see if the proportion of collagen types were changed.. And its relation with erectile dysfunction was evaluated.
31 Wistar male rats were included in the study, 9 control, 10 diabetic and 12 diabetic treated with aminoguanidin.

After 8 weeks of diabetes, the biochemical parameters were measured 5HMF levels are determined photometrically and collagen levels and types were measured by SDSPAGE.

Results: Serum G and HbA1c levels were higher in diabetic group than the controls. and these levels decreased after aminoguanidin application. Bun and creatinine levels were increased in diabetics and bun levels decreased but creatinine levels did not change in ag using group. Cholest and tg levels were higher in diabetics and tg decreased in ag group. 5Hmf levels were higher in diabetic penile tunica albuginea tissue and they significantly decreased in ag group.
Total and type 1 collagen were lower in diabetics but ag usage increased them. Type3 collagen levels were not different significantly.

Conclusions: We conclude that the undesirable effects of age for penile erection can be reversed by ag. Also the breakup of type1/type3 collagen levels proportion can have an effect on erectile dysfunction.

PT50
ANTHROPOMETRIC CHARACTERISTICS AND HIGH SENSITIVE C-REACTIVE PROTEIN

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Background: C- reactive protein of high sensitivity (hsCRP) is an inflammation marker. Discovery of hsCRP showed an important connection between inflammation and higher risk for cardiovascular disease. Obesity is a disease of modern age and one of the

main risk factors for development of cardiovascular diseases in adults.

Subjects and methods: The investigation included 82 metabolically healthy examinees, of both sexes, who are older than 18. In all examinees, the following anthropometric parameters were measured: height, weight, waist and calculated BMI. Concentration of hsCRP (mg/L) was determined by immunoturbidimetric method on latex particles, using biochemical analyzer Architect c 4000 made by the Abbott company. On the basis of waist measure and BMI, the examinees were divided into two groups: the group with normal weight and the group of obese.

Results: Average value of BMI (kg/m²) and waist measure (cm) in obese and in the group with normal weight were (29.47±2.87, 22.32±2.35 kg/m², respectively) and (100.83±8.12; 74.68±9.35cm, respectively). In the group of obese, average concentration of hsCRP was 5.69±1.96 mg/l and all examinees had the value higher than 3mg/L. Average value of hsCRP in the group with normal weight was 0.82±0.44 mg/L. By comparing the values of hsCRP in both groups using the Student's t test significantly higher concentration was recorded in the group of obese ($t=11.191; P<0.001$).

Conclusion: All examinees in the group of obese has the hsCRP higher than 3.0 mg/L, which is in accordance with the assumption that obesity belongs to highly risk factor for the development of cardiovascular diseases.

PF01
INVASION CAPACITIES OF MCF-7 AND MDA-MB-435 WITH REAL-TIME CELL ANALYSIS

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Background: Invasion and migration comprise mechanisms involved in both physiological processes and pathological processes incluiding tumor cell metastasis.The aim of this study was by continuous monitoring to assay the invasion capacity of human breast cancer (MCF-7 and MDA-MB-435) celll in esusing the xCELLi gence system, real time cell-analyser.

Material and methods: The rate of cells invasion were monitored in real-time with the xCELLigence system CIM-plates (n=6)by cell index (CI) impedance measurements.
The upper chamber of the CIM-plates was coated with 20 ula 1:20 solution of Matri gel and incubated for 4 hours. Then160 µL 10 % FBS DMEM and 30 µL serum-free DMEM was added to the lower and upper chambers respectively. 24 h serum-starved $2X10^4$ cells were seeded to the upper chamber. Control group was composed of uncoated wells. After this incubation period, a measurement step was performed as a background signal, generated by cell-free media. After cell addition, CIM-plates were incubated during 30 minutes at room temperature in the laminar flow hood to allow the cells to settle on to the membrane according to the manufacturer's guidelines. The impedance value of each well was automatically monitored by the xCELLigence system every 15 minutes for 24 h and expressed as a CI value.

Results: Data demonstrated that the impedance CI of coated well on MDA-MB-435 sharply increased after seeding up to reach its maximum from 5 to25 h. The CI initial of MDA-MB-435 cells was determined a minimum at 5h to increase again to a maximum at 25 h. The control group and the experimental group was determined to be same level at 25 hours. The CI of MCF-7cells showed a minimum at 5h and reached its second maximum

at 28h. The strong correlations with conventional method imply a similar observation of cells ınvasion activities. In all, we conclude that the invasion capacity of MDA-MB-435 experiments reflects to be higher than invasion capacity of MCF-7.

Conclusion: Our data suggests that the xCELLigence live cell analysis system provides an accurate and rapid platform for migration and invasiveness of the cancer cells.

PF02
ASSESSMENT OF COMPARABILITY OF THE TWO ANALYTICAL SYSTEMS FOR MEASURING THE HAEMOGLOBIN LEVELS IN THE BLOOD

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Introduction: The haemoglobin (Hb) level is the most commonly used parameter for screening of blood donors (BD) for the presence of anaemia. The aim of the research was to compare the Hb levels in venous blood obtained by measurements performed on a Siemens Advia 2120 analyser with the results obtained from capillary blood samples with the device EKF Hemo Control (EKF HC).

Materials and methods: 41 blood samples of BD were included in study.Capillary blood Hb levels of subjects whose values met the criteria for the selection of blood donorswere routinely measured using the EKF HC (optical absorption photometry, haemoglobincyanide method). An abbreviated evaluation of Siemens Advia 2120 was carried out (using haemoglobincyanidespectrophotometric method) for Hb parameter. The results were statistically analysed in MedCalc software.

RESULTS: The Hb levels measured with the instrument EKF Hemo Control and the Siemens Advia 2120 analyser were 141-167 g/L,143-169 g/, respectively.
Hb levels obtained using Siemens Advia 2120 analyser and EKF HC device underwent Passing-Bablok regression analysis. The regression equation was y= 2.000000 + 1.000000 x. Cusum linearity test did not give a significant deviation in linearity (P=0.86). Bland-Altman graph showed values distributed within ± 1.96.

CONCLUSION: Comparing the two devices for measuring Hb levels in the blood, Siemens Advia 2120 analyser as a reference with EKF HC, we concluded that these two methods are comparable. A small constant difference without proportional error is present between them. On the basis of this study we can conclude that the EKF HC device is suitable for screening of Hb levels in BD.

PF03
MASS SPECTROMETRIC MEASUREMENT OF URINARY NETRIN-1 IN RENAL TRANSPLANTATION

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Background: Netrin-1 can be a useful early diagnostic biomarker of acute kidney injury (AKI) after renal transplantation. The use of netrin-1 in clinical practice requires that this biomarker be associated with an analytical method that combines specificity, accuracy and robustness. This study aimed to develop an optimized multiple reaction monitoring (MRM) method using ultrafast liquid chromatography coupled with tandem mass spectrometry to measure urinary netrin-1 levels in renal transplant recipients.

Materials and methods: Purified recombinant human netrin-1 tryptic standard was analyzed by Matrix-Assisted Laser Desorption Ionization-Time of Flight Mass Spectrometry (MALDI-TOF) MS/MS and LC-MS/MS to select for peptides that provided specificity and adequate response in developing an MRM method for urinary netrin-1 quantification. Human urine samples collected from kidney transplant recipients were isolated, concentrated, precipitated and trypsin digested before mass spectrometric analysis of netrin-1. Netrin-1 levels were also measured in urine samples by enzyme immunoassay.

Results: The tryptic peptide ion $MH^{+2}$of ^{270}DSYFYAVSDLQVGGR^{284} (*m/z* 839) provided an adequate signal and was used for quantification of netrin-1 under conditions employed for LC-MS/MS analysis. MALDI-TOF MS/MS spectra obtained by collision-induced dissociation of the parent MH^{+2} ion ^{270}DSYFYAVSDLQVGGR^{284}resulted in*y*8, *y*9 and *y*11 product ions that were used for quantitative analysis by MRM method. Urinary Netrin-1 content measured by LC-MS/MS after transplantation was significantly higher compared to before transplantation levels. The Spearman correlation coefficient between the two methods was statistically significant. Intra-day and inter-day coefficient of variation provided good repeatability and reproducibility for validation of LC-MS/MS analysis.

Conclusions: LC-MS/MS quantification of Netrin-1 may provide a new reference method to determine changes of this potential biomarker in human kidney transplant patients.

PF04
ASSOCIATION OF METHYLENETETRAHYDROFOLATEREDUCTASE GENE POLIMORPHISMS C677T ON THE HOMOCYSTEINELEVELS IN CORONARY ARTHERYDIASEASE

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Background: The aims of this paper were to determine the concentration of the total homocysteine(tHcy), to find whether the increased level of tHcy is

associated with mutation of the C677T methylenetetrahydro- folatereductase gene (MTHFR). Also, we wanted to show whether the determination of the polymorphisms of this gene is particularly involved in the establishing of the diagnosis of coronary artery disease (CAD), and whether this genetic analysis is the choice for this most common disease.

Subjects and methods: The study included 84 subjects divided into two main groups: 43 healthy subjects as control group and 41 patients with CAD. The concentration of tHcy was determined by a cyclic enzymatic method, and the mutation of the MTHFR C677T gene was examined by a polymerase chain reaction.

Results: The concentration of tHcy plasma in the patients with CAD was significantly higher (18,72 ± 5,31 μmol / L) compared to the control group (11,11 ± 3,23 μmol / L) ($p < 0.001$). The statistical analysis with multiple regression showed that at the genotypes CT and TT of the MTHFR (C677T), the tHcy level is not significantly higher than the CC genotype of MTHFR (C677T) nor in the control group nor in the patients with CAD.

Conclusion: The analysis of the results showed that the polymorphism of the MTHFR (C677T) gene is not associated, in most cases, with the mild to moderate hyperhomocysteinemia.

PF05
MEAN PLATELET VOLUME VALUES IN PATIENTS WITH RHEUMATOID ARTHRITIS

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Background: Rheumatoid arthritis (RA) is characterized by inflammatory path-ways that lead to proliferation of synovial cells in joints. Like many autoimmune diseases, the etiology of RA is multifactorial. platelet-derived markers of inflammation and thrombogenicity are least studied, and their association with other risk factors in RA still remains obscure. The aim of this study was to investigate the mean platelet volume (MPV) values in this disease.

Materials and methods: Whole blood samples were collected from 60 healthy control and 119 patients with rheumatoid arthritis. The mean age for controls and patients were 45±11 and 45±3 years, respectively. Patients with chronic disease and inflammatory disorders were excluded. MPV levels were calculated with Abbott Cell Dyne heamotolgy analyzer. Statistical analysis was performed with SPSS v15.

Results: The median of MPV values in patients rheumatoid arthritis [9.5 (5.39-12.5)] were significantly higher compared to control group [7.5 (5.91-11.3)] ($p<0.001$).

Conclusions: MPV is a component of the CBC test. Although clinical utility and validity of MPV have not been established yet, some authors argue its use in inflammatory disorders. According to this study's results, MPV values might present the inflammation and must be established in large scale patient populations.

PF06
ANTIOXIDANT EFFECT OF DOPAMINE AGONISTS BROMOCRIPTINE ON COLD RESTRAINT STRESS- INDUCED GASTRIC ULCER IN RATS

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Background: Oxidative stress plays an important role in pathogenesis of several diseases. The role of reactive oxygen species (ROS) in the generation of gastric injury is also well known. Bromocriptine, a dopamine D_2 receptor agonist and mild D_1 receptor antagonist, have been shown to be effective protective factors against stress ulcerogenesis. Recent observations suggest that bromocriptine is an antioxidant that inhibits free radical formation and acts as a strong free radical scavenger *in vitro* and *in vivo*. The aim of this study was to investigate the antioxidant effect of bromocriptine on cold restraint stress-induced gastric ulcer in rats.

Materials and methods: Sexually mature male laboratory Wistar rats, with an average body weight of 200-230 g and aged up to three months,were starved for 24h and restrained inside individual close-fitting tubular cages and exposed to 4°C for 3h. Animals were pretreated with bromocriptine 24 h before the stress (25 mg/kg b.w.), and also the next day 1.5 h prior to the cold restraint stres (12.5 mg/kg b.w). Control group was only exposed to cold restraint stress. Animals after treatments were decapitated and the stomach were extracted. We determined the intensity of lipid peroxidation, content of reduced glutathione, activities of xanthine oxidase, catalase, glutathione reductase, glutathione peroxidase and peroxidase in homogenised stomach.

Results: Bromocriptine in combination with stress caused a statistically significant increased of the intensity of lipid peroxidation and activities of xanthine oxidase, catalase and glutathione reductase, significantly decreased activity of peroxidase, while the activity of glutathione peroxidase and content of reduced glutathione remained unchanged compared to the control group.

Conclusion: Significantly enhanced activities of catalase and xanthine oxidase and the intensity of lipid peroxidation in bromocriptine group compared to the control indicates that bromocriptinethere is noantioxidant effect on cold restraint stress- induced gastric ulcer in rats.

PF07
MEASUREMENT UNCERTAINITY OF HEMOGLOBIN A2 IN PUBLIC HEALTH LABORATORY

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Objective: High Performance Liquid Chromatography (HPLC) is widely used to classify hemoglobin fractions in screening of hemoglobinopathies. In this study we aim to detect the "measurement uncertainty" of the

Hemoglobin A2 in thalassemia screening and assume its value in the laboratory test results.

Materials-methods: HbA2 analysis was measured by HPLC analyser Biorad Variant-II instrument, which works in cation exchange principle, in Uşak Public Health Thalassemia Laboratory. Measurement uncertainty calculations were made according to the Nordtest manuals, using internal, and external quality control values.

Results: The measurement uncertainty for HbA2 was ±12% within a 95% coverage probability. The cut-off value used in our laboratory for a positive screening result is 3.5%. Our considerations show that; we should present the cut-off value of Hemoglobin A2 as 3,1-3,9.

Conclusions: Although HbA2 measurement is important parameter in the screening of thalassemia, the literature lacks studies of measurement uncertainty of this parameter. Each laboratory should define their own measurement uncertainties not only to set their quality goals, but also in order to verify their capability to aid in clinical decisions.

PF08
ANALYSIS OF THE ACID-BASE STATUS OF THE BLOOD IN PATIENTS WITH CHRONIC RESPIRATORY FAILURE OF DIFFERENT ETIOLOGY

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Background: Analysis of the acid-base status of arterial blood provides help in the diagnosis of chronic respiratory failure (CRF), monitoring the disease, the efficiency of gas exchange and the effect of the treatment.The aim of this study was to analyze the acid-base status of the blood in patients with CRF of different etiology, including patients with chronic obstructive pulmonary disease (COPD), bronchial asthma and pneumonia..

Subjects and methods: The study was conducted at the Department of Pulmonary Diseases and Tuberculosis Podhrastovi, KCU Sarajevo, and included 90 patients with CRF of different etiology, of both sexes, aged 21-86 years. Arterial blood gas analysis included: pH, pCO_2, pO_2, HCO_3, BE (B), sO_2. Investigated spirometry parameters were: forced expiratory volume in the first second (FEV1), forced vital capacity (FVC), the ratio of FEV1 and FVC (FEV1/FVC%) and the largest expiratory flow (PEF).

Results: pH of arterial blood in the COPD group was statistically significantly lower than the pH of the bronchial asthma group and pneumonia. Value of pCO2 in arterial blood was highest in patients with COPD while PO_2 of arterial blood was highest in patients with bronchial asthma. The concentration of HCO_3 in arterial blood in the COPD group was statistically significantly higher than the values in the group of bronchial asthma and pneumonia group. In the group of COPD, pH and pO_2 after treatment were significantly higher than the values on admission. In the group of patients with bronchial asthma, pH was significantly higher after therapy administration. In patients with pneumonia, value of pCO_2 was also significantly higher after administration of therapy.Patients with COPD had statistically significantly lower values of FEV1% and FVC%, compared to patients with bronchial asthma and pneumonia, and statistically significantly lower PEF% compared to PEF% of patients with

bronchial asthma. FEV1/FVC% in both COPD and bronchial asthma were statistically significantly lower than the value of the group pneumonia.

Conclusion: There are significant differences in acid-base status of arterial blood in patients with CRF of different etiology. In patients with COPD hypoxemia with hypercapnia is usually expressed, while in other groups there is only hypoxemia.

PF09
OXIDANT AND ANTIOXIDANT STATUS IN SYSTEMIC LIPOPOLYSACCHARIDE INDUCED RATS

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Background: Sepsis is a complex series of systemic inflammatory reaction condition. Foreign bacterias and Lipopolysaccharides (LPS) lead to the release of many cytokines as a result of the stimulation of various immune cells. Reactive oxygen species (ROS) and oxidative stress are thought to play a central role in potentiating macrophage activation, causing excessive inflammation, endothelial damage in many tissues, and sepsis. Thus, increasing evidence demonstrates that reactive oxygen species plays important roles in LPS induced septic injury. The aim of this study was to investigate the actions of treatment agents, which have antiinflammatuar and anti-TNF activity, on hepatic oxidative stress in LPS induced sepsis rat model.

Materials and methods: Thirty rats were randomly divided into five experimental groups: sham (group 1), sepsis (group 2), sepsis + thalidomide (group 3), sepsis + etanercept (group 4) and sepsis + thalidomide+ etanercept (group 5), n = 6 each. LPS was administered intraperitoneally to group 2. Liver tissue total antioxidant capacity (TAS), total oxidative status (TOS) and malondialdehyde (MDA), which is the end product of lipid peroxidation, levels were measured in all groups. To determine TOS and for the quantitative evaluation of TAS were used Enzyme-Linked Immuno Sorbent Assay (ELISA) kit. MDA measurements were evaluated using High-performance liquid chromatography (HPLC) system.

Results: The levels of tissue MDA and TOS in the sepsis group were much higher than those in the sham group. At the same time, the MDA levels in group 5 were significantly lower than sepsis group ($p<0.001$). However, the levels of TOS in the sepsis group was significantly higher than group 4 and 5 ($p<0.001$, both of them). Additionally, TAS levels in the sepsis group were lower than sham group, but not statistically significant. At this time, the levels of tissue TAS in group 5 were noticable higher than sepsis group ($p=0.044$).

Conclusion: Our results demonstrate that both etanercept and etanercept+ thalidomide administration may have a beneficial effect on oxidative stress parameters induced in experimental sepsis rat models.Therefore, as a treatment agent, etanercept alone or in combination with thalidomide can be used to avoid devastating effects of sepsis.

PF10
KLOTHO GENE 352F>V (RS9536314) POLYMORPHISM IN HEMODIALYSIS PATIENTS FROM BOSNIA AND HERZEGOVINA – PRELIMINARY REPORT.

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Background: Recently proposed that polymorphism 352F>V of the *KLOTHO* gene may be one of the factors linked with deficiency production of KLOTHO protein in the chronic kidney disease (CKD). The Klotho-protein deficiency (linked with variant 352V) may initiate or intensify disturbances mineral metabolism which is involved in CKD patients with different etiologies such as: obstructive nephropathy, diabetic nephropathy, minimal-change nephritic syndrome, IgA nephropathy and chronic glomerulonephritis progression and development of extra renal complications. It was found that deficiency of Klotho promotes renal fibrosis, delay kidney regeneration and renders the kidney more susceptible to injury and eventually promotes chronic progression. Data on prevalence and phenotypic consequences of *KLOTHO* variants are sparse, in population of Bosnia and Herzegovina are nonexistent. Therefore, the aim of our study was to estimate the frequency of 352V allele in hemodialysis patients and in healthy controls.

Subjects and methods: We recruited 64 hemodialysis patients (40 male and 24 female, mean age 55±14.6 years), and 15 healthy controls (4 male and 11 female, mean age 39.93±9 years); 79 participants in total. Duration of hemodialysis treatment was 85,2±17month. We determined the prevalence of a common T>G polymorphism, resulting in a phenylalanine (F) to valine (V) substitution at aminoacid position 352 and their association with CKD patients. We used PCR-RFLP techniques.

Results: In hemodialysis patients we identified 13 FV heterozygotes and 2 VV homozygotes, but in healthy controls 1 heterozygotes and none homozygotes. The average frequency of 352V allele was 21% and 6% in hemodialysis patients and healthy controls, respectively.

Conclusions: This is the first report on the prevalence of 352F>V KLOTHO gene from Bosnia and Herzegovina. The clinical value of *KLOTHO* determination in monitoring complications in CKD with larger samples is warranted.

PF11
PARAOXONASE ACTIVIY AND OXIDATIVE STRESS IN CHILDREN WITH DIABETIC KETOACIDOSIS

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Background: The aim of our study was to investigate serum paraoxonase 1 (PON1) activity, an antioxidative enzyme, total antioxidant status (TAS), and total oxidant status (TOS) in children with diabetic ketoacidosis (DKA).

Materials/Subjects and methods: The study included 30 diabetic children with DKA (17 females and 13 males; mean age 8.48 ± 4.55 years) and 27 healthy controls (17 females and 10 males; mean age 8.20 ± 4.59 years). Serum PON1 activity, TAS and TOS levels were measured upon admission and 24 hours later in the patients and the controls.

Results: The difference between PON1 activity of diabetic children with DKA on admission and 24 hours later was not significant. Also, PON1 activity of diabetic patients with DKA on admission and 24 hours later was not significantly different compared to that of the controls. The diabetic children had significantly lower TAS values than the controls on admission ($p<0.001$) whereas TAS values were significantly higher than the controls 24 hours later ($p<0.05$). The diabetic children had significantly higher TOS values than that of the controls on admission ($p<0.01$) and 24 hours later ($p<0.05$). TAS levels of diabetic children with DKA were increased significantly ($p<0.001$) whereas TOS levels were decreased significantly ($p<0.05$) 24 hours later compared to the values on admission.

Conclusions: Our findings demonstrated that diabetic children with DKA had significantly increased oxidative stress that could be effectively treated by intensive insulin therapy. Also, decreased TAC levels might have resulted from increased oxidative stress which may lead to consumption of antioxidant molecules. But, PON1 activity was not significantly changed either in DKA or after therapy in the diabetic children.

PF12
LIPID BODY TYPES IN HEAVY PROTEINURIA URINE SAMPLES

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Background: The presence of a double refractive urine lipid bodies (ULB) in the urinary sediment is indicator of severe renal dysfunction. ULB are common in patients with renal glomerular diseases causing heavy proteinuria. They are unexpected finding in those patients without heavy proteinuria and rare finding in patients with nonglomerular renal diseases. Determination of a ULB

presence is very important for the diagnosis as well as for the evaluation of severity and course of the renal disease. The aim of the study was to determine the presence and the most frequent type of ULB in patients with heavy proteinuria.

Materials and methods: The cross-sectional study included 36 patients with heavy proteinuria (>3g/24h), both sexes (22 males, 14 females). Patients were hospitalized at the Clinical Centre University of Sarajevo from April 2014 to February 2015. Microscopic analysis of the urine sediment on the glass slide covered with cover slip obtained from the native, first morning urine were performed at the Clinical Chemistry and Biochemistry, Clinical Centre University of Sarajevo. All microscopic specimens were first examined under low and high power by the phase contrast light microscopy to identify particles as recommended by international guidelines. Verifications that were carried out by using filters for polarized light microscopy (Nicol prisms), mandatory for the correct identification of lipids, crystals and contaminants, showed a double refractive ULB, made from cholesterol esters, as „Maltese crosses" in the urine sediment with a presence of lipiduria .

Results: ULB were present in 34 (94%) patients. Among them, the most frequent were fatty casts, presented in 94%.of urine samples. Oval fat bodies and free fat drops were noted also (32%, 32% respectively). The lowest frequency was noted for cholesterol crystals (3%). Despite of heavy proteinuria in 6% of urine samples ULB were not detected.

Conclusions: ULB are common finding in patients with heavy proteinuria and the most frequent ULB type are fatty casts.

PF13
SERUM LEVEL OF SOLUBLE UROKINASE-TYPE PLASMINOGEN ACTIVATOR RECEPTOR (SUPAR) AS A NEW INFLAMMATION MARKER IN CHILDHOOD OBESITY

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Background: Obesity is a growing worldwide health problem affecting children and associated to low-grade inflammatory state with enhanced production of inflammatory mediators in childhood. Soluble urokinase plasminogen activator receptor (suPAR) can be generated as a proinflammatory marker. We aimed to evaluate suPAR, leptin, adiponectin, interleukin-6 (IL6), high sensitive C-reactive protein (hsCRP) and fibrinogen in the childhood obesity.

Material and Methods: This study was performed in 50 (19 males, 36 females) obese children aged 10 to 17 years (14.29 ± 1.70) and 45 (18 males, 25 females) control subjects aged 10 to 17 years (14.26 ± 1.80). Serum suPAR, IL-6, leptin and adiponectin were measured by using ELISA method.

Results: suPAR ($p<0.05$), IL-6 ($p<0.05$), leptin ($p<0.001$), hsCRP ($p<0.001$) and fibrinogen ($p<0.001$) values of the obese subjects were significantly higher than those of the controls. Adiponectin ($p<0.01$) values of

the obese subjects were significantly lower than those of the control group.

Conclusion: suPAR may be a good novel biomarker for systemic subclinical inflammation and immune activation linked to childhood obesity. We consider that the combined evaluation of suPAR and other parameters is a better therapeutic aid, and careful management of lifestyle from childhood may contribute to the prevention and progression of Metabolic Syndrome in the future.

PF14
THE ASSOCIATION BETWEEN LIPID PROFILE AND INFLAMMATORY PARAMETERS IN THE BLOOD DEPENDING ON THE ACHIEVED GLYCEMIC CONTROL IN TYPE 2 DIABETES MELLITUS

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Background: The synergistic effects of dyslipidemia and low-grade inflammation, trigger and accelerate development of atherosclerotic complications in diabetes mellitus type 2 (DMT2) patients. The aim of the study was to evaluatethe association between lipid profile and inflammatory parameters such as C-reactive protein (CRP) and fibrinogen, and the impact of achieved glycemic control on their association in DMT2.

Subjects and methods: A cross sectional, observational study included 80 sex matched DMT2 subjects, aged 46-82 years. The subjects treated at Primary Health Care Center Kladanj in January/February 2015 were distributed according to glycated hemoglobin level (HbA1c) into two groups: HbA1c ≤ 7% and HbA1c>7%. Lipid profile analysis was performed in Primary Health Care Center Kladanj but testing of inflammatory markers was performed in Laboratory unit of University Clinical Center Tuzla, both using standard methods. The accepted level of statistical significance was $p<0,05$.

Results: The subjects of HbA1c>7% group had higher blood values of total cholesterol (TC), triglycerides (TGc), low density lipoprotein cholesterol (LDLc) ($p<0,0005$), very low density lipoprotein cholesterol (VLDLc) ($p=0,007$), and lower values of high density lipoprotein cholesterol (HDLc) ($p=0,043$) compared to HbA1c ≤ 7% group. Besides a negative correlation between CRP and HDLc ($rho=-0,46$; $p<0,01$), the positive correlations of CRP and fibrinogen with TC ($rho=0,52$; $p<0,01$and $rho=0,71$; $p<0,0005$, respectively) and LDLc ($rho=0,35$; $p<0,05$ and $rho=0,42$; $p<0,01$, respectively) in HbA1c>7% group were found.There were the positive correlations of TGc ($rho=0,47$ $p<0,01$); TC, LDLc, VLDLc ($rho=0,37$, $rho=0,31$, $rho=0,46$; $p<0,05$, respectively) as well as the negative correlations of HDLc ($rho=-0.33$; $p<0,05$) with fibrinogen in the HbA1c<7% group. In this group, CRP also correlated positively with TC, TGc, LDLc, and VLDLc ($rho=0,50$, $rho=0,44$, $rho=0,44$,$rho=0,43$; $p<0,01$, respectively).

Conclusion: Significantly different values of inflammatory markers and lipid profile parameters depending on glycemic control as well as their relationships in DMT2 indicate the importance of good glycemic control achievement as key factors in development of DMT2 complications.

PF15
SERUM LEVELS OF CYTOCHROME C (CYCS) AND FAS LIGAND (FASL) IN THE RATS FED HIGH-FAT DIET AND ACRYLAMIDE

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Background: Acrylamide an organic chemical occurs in foods widespreadly consumed in diets worldwide. Fas ligand and cytochrome *c* are key elements in the process of apoptosis. The aim of this study was to investigate serum cytochrome *c* and Fas ligand levels in the rats fed high-fat diet and acrylamide, compared to control rats.

Materıals and methods: Forty-eight male Wistar rats (5-6 weeks of aged) were segregated into two diet groups and fed with a high-fat diet (crude fat 20%) or standard diet (crude fat 2.7%), respectively; and animals in each diet groups were exposed to acrylamide at the dose of 0, 2, 10 or 20 mg/kg bw/day via drinking water for 28 days. At the end of the experiment, serum samples were analyzed for cytochrome *c* and Fas ligand with the ELISA method.

Results: Serum Fas ligand levels were significantly higher at concentrations of 10 mg/kg bw/day acrylamide and 20 mg/kg bw/day acrylamide in the rats when compared with those of the control rats. No differences were found in serum Fas ligand levels between the control rats and the rats fed high-fat diet. On the other hand, there were no difference between groups in terms of serum cytochrome *c* levels.

Conclusions: Our findings show that acrylamide exposure may lead to apoptosis and high-fat-intake reverses the effects on acrylamide -induced apoptosis.

PF16
ASSOCIATION OF GAMMAGLUTAMYLTRANSFERASE (GGT) WITH THE TYPE OF ACUTE CORONARY SYNDROME

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Background: The elevatedserum GGTlevel is independently correlated with conditions such as alcohol consumption, elevated serum cholesterol, obesity, high blood pressure andmyocardial infarction. It is also known that serum GGT activity is an independent risk factor for myocardial infarction and cardiac death in patients with acute coronary syndrome (ACS).The study aim was to assess serum GGT activity in patients with ACS and apparently healthy subjects, and the association of GGTwiththe type of ACS in the patients group.

Subjects and methods: A total of 80 subjects was recruited to the study, 39

women and 41 men, divided into two groups, a control group with apparently healthy subject, and the group of 41 patients with ACS who were admitted and hospitalized in the intensive care at the unit of the Clinic for heart disease and rheumatism University Clinical Center Sarajevo. The type of ACS was diagnosed according to the clinical presentation, ECG and the cardiac troponin I. Data from both groups were analyzed using SPSS statistical software version 19, and the accepted level of statistical significance was $p<0.05$.

Results: From the total of 41 patients, 48.8% had ST segment elevation myocardial infarction (STEMI), 26.8% had non-ST segment elevation myocardial infarction (NSTEMI) and 24.4% had unstable angina.The most common risk factors for ACS were hypertension (75.6%), dyslipidemia (58.5%) and cigarette consumption (51.2%). The difference between serum GGT activity of patients with ACS was significantly higher compared to the control group ($p<0.05$). Statistically significant difference in GGT activity among all groups (control groups and types of ACS) was noted ($p<0.05$). Patients with unstable angina and STEMI type had higher GGT activity, compared to healthy subjects ($p>0.05$, respectively). Significant gender difference between control and ACS group, was noted ($p<0.05$).

Conclusion: GGT is significantly higher in patients with ACS than in the control group. The difference in GGT activity among different types of ACS was also noted. Gender difference in enzyme activity between groups was found. Monitoring the values of this parameter could be of great importance as a risk factor in tracking the progress of coronary disease.

PF17
SERUM TRACE ELEMENT STATUS OF PATIENTS WITH PRIMARY HYPERLIPIDEMIA

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Background: Lipid and lipoprotein abnormalities are common in the general population, and are regarded as a modifiable risk factor for cardiovascular disease due to their influence on atherosclerosis. Organism is established by organic and inorganic matters. Trace minerals including inorganic matters and they found very little amounts in human body. Although this little amounts trace minerals have vital function whether directly or indirectly like vitamin synthesis, hormone production, cell respiration etc. Trace elements participate in biological systems as components of enzymes or as catalysts carrying out some chemical reactions in living cells. The aim of the present study was to compare serum trace elements levels (Fe, Cu, Zn, Mn, Cr, Se, Co, Ni, Mo, Cd) in patients with primary hyperlipidemia and healthy subjects.

Materials and methods: This study was performed on 46 primary hyperlipidemic subjects and 33 healthy subjects. Inductively coupled plasma mass spectrometry (ICP-MS) was used

for the determination of serum trace element concentrations.

RESULTS: The levels of Co ($p<0.001$), Ni ($p<0.01$), Cd ($p<0.001$) were significantly higher and the level of Cr ($p<0.01$), Fe ($p<0.05$), Mn ($p<0.01$), Se ($p<0.001$) and Mo ($p<0.001$) were significantly lower in the primary hyperlipidemic subjects than the healthy subjects. Also, the serum levels of Zn and Cu of both groups were not significantly different from each other.

CONCLUSIONS: Our findings show that there are clear associations between Cr, Mn, Se, Mo and Fe deficiencies and primary hyperlipidemia. In addition, our results show that, there is no direct association between serum Zn and Cu levels and the state of primary hyperlipidemia.

PF18
ATHEROGENIC INDEX OF PLASMA IS MORE SENSITIVE THAN CASTELLI INDEX II IN ATHEROGENIC RISK CLASSIFICATION OF TYPE 2DIABETESMELLITUS PATIENTS

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Background: Lipid and lipoprotein testing providethe valuable information on an atherogenic risk in diabetes mellitus type 2 (DMT2) patients. The more pratical way to classify patients into low and high risk atherogenic groups is to calculate lipoprotein ratio.The most frequentlyused ratio in Bosnia and Herzegovina biochemistry laboratories is Castelli index II (low density lipoprotein cholesterol (LDLc) and high density lipoprotein cholesterol (HDLc) ratio).The study aim was to calculate and assess the atherogenic risk level in DMT2 patients by using Castelli index 2 and atherogenic index of plasma (AIP) and to find out if the difference in frequency exists.

Patients and methods:A cross sectional study included 80 DMT2 patients, aged 46-82 years. Lipid and lipoprotein analysis was performed in overnight fastingpatientsin Primary Health Care Center Kladanj.Castelli index II was calculated from the molar concentration of LDLc and HDLcserum values.A logarithmically transformed ratio of the molar concentrations of triglycerides to HDLc was the atherogenic index of plasma.Patients were classified into low and high risk, according to Castelli index (values less or more than 4, respectively). Low, moderate and high risk groups were formed according to AIP values (<0.11, 0.11-0.21 and >0.21, respectively).

Study results: The average values of Castelli index II and AIP in DMT2 patients were 4.1 (3.25-4.85) and AIP 0.34±0.14,respectively. From a total 80 diabetic patients, according to Castelli index II, 34 (42.5%) of them were classified into low risk and 46 (57.5%) in high risk groups.The statistically significant difference was found in comparison withAIP classification ($p=0.021$); 78 (97.5%) patients were classified into intermediate and high risk (17 (21.25%), 61 (76.25%), respectively) but just 2 (0.025%)patients were in the low risk group.

Conclusion: Atherogenic index of plasma is more sensitive than Castelli index in atherogenic risk assessement regarding on its strong correlation with lipoprotein particle size.

PF19
THE COMPARISON OF TWO DIFFERENT ANALYZERS ON THE EVALUATION OF HBA1C RESULTS

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Aim: HbA1c is widely used in evaluating retrospectively the long-term control of blood glucose concentrations in diabetic patients. Furthermore an HbA1c of 6.5% is recommended as the cutoff point for diagnosing diabetes. In this study we assessed the correlationamong HbA1c values measured using 2 different widely used HPLC instruments.

Method: The evaluation of HbA1c for 300 out patients who visited Turgut Ozal University Hospital clinics with the suspicion and monitoring of diabetes were included in to the study randomly without the distinction of sex. Shimadzu and Zivak Technologies trade marked devices were used to analyze EDTA-treated plasma samples. Correlation assessment was performed using SPSS 16.0 software (Chicogo, IL, USA).

Result: The correlation coefficient of HbA1c was calculated as 0.974 between the two devices, which revealed strong correlation. The difference between the results obtained from the two HPLC instruments were found to be statistically insignificant (p=0.295).

Conclusion: HbA1c is universally used for both diagnosing and monitoring diabetes. HPLC is the mostly preferred technique for measuring HbA1c. A correlation analysis is required for the validation of results in case there is a renewal of routinely used devices with different devices. Therefore we compared the measurements of HbA1c obtained from the both devices to test the usability of the new device and as a result we found out strong correlation between the two devices.

PF21
THE COMPARISON OF TWO DIFFERENT HPLC INSTRUMENTS ON THE EVALUATION OF VITAMIN D LEVELS

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Aim: The aim of method review studies is to assess the analytical error ratio of the newly used instruments in laboratories.
Vitamin D is a prehormone and its two major forms are ergocalciferol (vitamin D_2), and cholecalciferol (vitamin D_3). With in the epidermal layer of skin, 7-Dehydrocholesterol, a zoosterol, undergoes an electrocyclic reaction as a result of 290-310 nm wave length UV radiation, this is the major resource of endogen vitamin D synthesis. The correct measurement of 25-hydroxycholecalciferol ($25(OH)D_3$) levels is of great clinical significance, therefore a standardization is required. Methods other than Tandem mass

spectrometry (LC-MS/MS) include High-performance liquid chromatography (HPLC), Radioimmunoassay (RIA) and Chemiluminescence methods. Thus we used LC-MS/MS method, which is the golden standard, to compare the results of two different HPLC instruments, a Shimadzu trademarked instrument that is currently used in our laboratory and a Zivak Technologies trademarked instrument that is planned to be newly installed.

Method: The plasma samples of 65 out patients who visited Turgut Ozal University Hospital clinics, without the distinction of diagnosis, were analyzed to measure vitamin D levels using the both HPLC instruments and the LC-MS/MS instrument.

Result: The correlation coefficients of vitamin D levels were 0.9359 for Zivak Technologies HPLC instrument vs. Zivak Technologies LC-MS/MS instrument, 0.9015 for Shimadzu HPLC instrument vs. Zivak Technologies LC-MS/MS instrument and 0.9219 for Zivak Technologies HPLC instrument vs. Shimadzu HPLC instrument. The differences between all three instruments was found to be statistically insignificant ($p>0.05$).

Conclusion: For the last 35 years, assessment of $25(OH)D_3$ metabolite levels has had a great significance in the evaluation of calcium homeostasis. Even though HPLC is not the golden standard, the results obtained are the closest to the measurements of LC-MS/MS instruments, compared to the other methods. Moreover LC-MS/MS instruments are relatively more expansive therefore are not routinely used in most of the laboratories. As a conclusion our study showed correlation between the two HPLC instruments which suggests tha tboth can be used routinely in laboratories.

PF22
NEUTROPHIL TO LYMPHOCYTE RATIO – A GOOD INDICATOR OF SUBCLINICALINFLAMMATION LEVEL IN TYPE 2 DIABETES MELLITUS

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Background: Type 2 diabetes mellitus (DMT2) is a major public health problem worldwide, resulting as a consequence of insulin resistance or inadequate production of insulin by the beta cells of the pancreas. Several studies have confirmed the link between systemic inflammation and insulin resistance, pointing to the importance of neutrophil to lymphocyte ratio (NLR), as marker of subclinical inflammation. Aim was to examine the NLR in DMT2 patients with good (HbA1c<7%) and the poor (HbA1c>7%) glycaemic regulation, and examine its relationship withother inflammatory parameters.

Patients and methods: Cross-sectional study included 50 patients, suffering from DMT2 (17 males and 33 females, aged 67-80 years). Patients were divided into two groups: with good (HbA1c<7%, n = 25), and poor glycaemic regulation (HbA1c>7%, n = 25).Biochemical analyses for glycatedhaemoglobin (HbA1c), erythrocyte sedimentation rate, the serum concentration of total protein, albumin and globulin, were performed at the Department for Clinical Chemistry and Biochemistry, Clinical Centre University of Sarajevo, with calculated albumin/globulin ratio, and NLR

calculated from white blood cells differential count.

Results: Significantly higher values of the NLR ($p<0.0005$) were observed in patients with poor vs. patients with good glycaemic regulation. Significant positive correlation of HbA1c with NLR (rho = 0.432; $p=0.002$) was obtained, and significant negative correlation of NLR with albumin, A/G ratio and total serum protein in patients with DMT2 (rho = -0432, $p= 0.002$; rho = -0379, $p= 0.007$; rho = -0285, $p= 0.045$, respectively).
Conclusion: Significantly higher values of the NLR were observed in patients with a poor vs. patients with good glycaemic regulation, pointing to NLR as a good indicator of subclinical inflammation level, associated with achieved glycoregulation.

PF23
THE EFFECTS OF ROSMARINIC ACID ON THE COLON TISSUE IN COLORECTAL CANCER MODEL CREATED BY AZOXYETHANE

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Background: Colorectal cancer (CRC) is one of the most common cancers in men and women. The anti-inflammatory or antioxidant mediators are known to be effective against CRC. In this study, the rosmarinic acid was usedto investigate the anticancer effect against CRC induced by azoxymethane (AOM) in rats.

Materials and Methods: The rats were divided into 3 groups. In addition to the standart diet of the all groups 15,8% peanut oil was added throughout the experiment. The control group received 0,9% saline (ip, weekly) for 4 weeks. Group 2 received 15 mg/kg AOM (ip, weekly) for four weeks. The azoxymethane+rosmarinic acid group was induced colon cancer by injecting 15 mg/kg AOM (ip, weekly) for four weeks and was formed by giving oral daily 5 mg/kg rosmarinic acid. The all rats were sacrificed at the end of 30 weeks. Biochemical and histopathological examinations were performed in colorectal tissue samples.

Results: The results of the histopathological adenocarcinoma 87.5% and dysplasia 12.5% were observed in the AOM group. In treatment groups, a decrease was observed in adenocarcinoma rate. Adenocarcinoma 66.7% and dysplasia 33.3% were observed in the in the treatment group. When compared to the controls, significant decreases in Total Antioxidan Species and adiponectin levels, significant increases in Total Oxidant Species and Monocyte chemoattractant protein-1 (MCP-1) levels were found in AOM group. Significant decreases in tissue TOS and MCP-1 levels were found in the treatment group compared to AOM group.

Conclusion: In conclusion, it was confirmed by biochemical and histopathological results that rosmarinic acid decreases in the incidence of tumor.

PF24
CHANGES IN SERUM LIPOPROTEINS AND SIGNIFICANCE OF LIPOPROTEIN RATIOS IN ISCHEMIC CEREBROVASCULAR DISEASE

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Background: Disorder in lipid metabolism is one of the risk factors for atherosclerosis development, which plays most important role in ischemic cerebrovascular disease onset. The aim of study was to determine changes in levels, and sex differences in serum values for total cholesterol, triglycerides, HDL-c, LDL-c, VLDL-c TC/HDL-c and LDLc/HDLc ratio in patients with ischemic cerebrovascular insult.

Subjects And Methods: The study included patients from Neurology Clinic at the Clinical Center University of Sarajevo, hospitalized for the first time, in the period from January 2004 to November 2007. After the inclusion and exclusion criteria were met, the total study consisted of 217 (100 female and 117 male) patients. The study was designed as a retrospective study. Laboratory test were evaluated with special focus on serum lipids concentration: total cholesterol (TC), triglycerides, high density lipoprotein (HDL-c), low density lipoprotein (LDL-c), very low density lipoprotein (VLDL-c), total cholesterol/HDL cholesterol (TC/HDLc) and LDLc/HDLc ratio.

Results: Our results showed lower levels of serum HDL-c in 80% of patients compared to reference range values, with average of 0,81 mmol/L. TC/HDLc ratio was higher at 87% of patients compared to reference range values, with average of 6,4. LDLc/HDLc ratio was higher at 89% of patients compared to reference range values, with average of 5,3.
Significantly different were values for TC (5,60 mmol/L vs. 6,08mmol/L), HDL-c (0,99 mmol/L vs. 1,37 mmol/L) and LDL-c (3,77 mmol/L vs. 4,15 mmol/L); ($p<0,05$) between male and female subjects, while triglyceride and VLDL-c values showed no significant difference between male and female subject values (1,88 mmol/L vs. 1,86 mmol/L and 0,85 mmol/L vs. 0,82 mmol/L, respectively);($p>0,05$).

Conclusions: TC/HDL-c and LDL/HDL ratio, as well as HDL-c serum levels have a greater importance in ischemic cerebrovascular disease, and are therefore better predictors of cerebrovascular disease than other routinely used, simple lipid parameters.

PF25
THE EVALUATION OF SERUM TOTAL OXIDANT/ANTI-OXIDANT STATUS AND ISCHEMIA-MODIFIED ALBUMIN LEVELS IN PATIENTS WITH ACUTE PANCREATITIS

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Background: Acute pancreatitis (AP), an inflammatory disorder of the pancreas, is associated with significant morbidity and mortality. The pathogenesis of AP has been suggested to involve high oxidative stress (OS) combined with inadequate antioxidant status. The degree of oxidant/anti-oxidant balance correlating with the clinical severity of AP. Here, we aimed to investigate the levels of serum total anti-oxidant status (TAS), total oxidant status (TOS) and ischemia-modified albumin (IMA) in patients with mild AP.

Material and Methods: Thirty subjects with mild AP and 29 healthy controls were enrolled into the study. The levels of TAS, TOS and IMA, C-reactive protein (CRP), high sensitivity CRP (hs-CRP) and fibrinogen were measured in both groups.

Results: TAS levels were significantly lower (p=0.037), while IMA levels were significantly higher (p<0.001) in patients, compared to controls. TOS levels were similar between two groups. Fibrinogen, CRP and hs-CRP levels were significantly higher in patients than controls (p<0.001 for all parameters). IMA levels were positively correlated with amylase and lipase levels (r=0.448, p=0.001 and r=0.469, p=<0.001, respectively); further, TOS levels were also positively correlated with those of lipase (r=0.261, p=0.048). There was a negative correlation between TAS levels, and amylase and lipase levels (r=-0277, p=0.035 and r=-0.278, p=0.034, respectively).

Conclusions: In literature, OS is reported to be associated with the inflammatory process and the severity of AP. The measurements of TAS and TOS are used to predict OS, and IMA is considered a non-specific biomarker in the evaluation of OS. In our study, while IMA levels were increased in patients with mild AP, TAS levels were seen to be decreased.

PF26
SERUM CREATININE VS CREATININE CLEARANCE IN THE ASSESSMENT OF RENAL FUNCTION

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Background: Serum creatinine and creatinine clearance are the most often used in clinical practice for estimation of renal function. Today, different set of formulas is at the disposal for estimation of the glomerular filtration rate (GFR) as the basic function of the kidney. One of the most common is the Cockcroft-Gault formula, which although originally intended for determination of creatinine clearance, can be used to estimate GFR. Study evaluated the benefits of determining creatinine clearance through a 24 hour creatinine clearance and the Cockcroft-Gault method adjusted for the body surface area (BSA) and body mass index(BMI).

Subjects And Methods: The study was conducted as a cross sectional at the Endocrinology Clinic, the Clinical Centre of Sarajevo, from April 2013 to January 2014. Study included 90 patients suffering from diabetes mellitus type 2, aged 33-85 years, 47 women and 43 men. Serum creatinine was determined using Jaffe colorimetric method.

Results: The average age of women was 62.0±11.3 and men 60.5±13.4. Correlation values of serum creatinine with creatinine clearance adjusted for BSA of 1.73 m^2 were r=-0.375 for a 24 hour creatinine clearance ($CC_{STAND/1.73m^2}$), r=-0.714 for the Cockcroft-Gault ($CC_{CG/1.73m^2}$) and r=-0.723 for the Cockcroft-Gault corrected to BMI ($CC_{CG\ BMI/1.73m^2}$). There were examinees with referral values of serum creatinine and abnormal values of creatinine clearance in the following groups – a 24 hour creatinine clearance ($CC_{STAND/1.73m^2}$) group with 41 patients (45.6%), the Cockcroft-Gault ($CC_{CG/1.73m^2}$) group with 27 patients (30.0%), and the Cockcroft-Gault group adjusted for BMI with 34(37.8%).

Conclusions: Since the highest level of negative correlation occured between the values of serum creatinine and creatinine clearance obtained by the Cockcroft-Gault method corrected for BSA and BMI, this method is recommended to be used.

PF27
THE FIRST STUDY OF FREQUENCY CCR5Δ32 MUTATION IN BOSNIAN AND POLISH PATIENTS WITH CROHN'S DISEASE

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Background: Crohn's disease (CD) is a multifactorial disease with an environmental factors and genetic background. In the recent period the available data of genetic factors and immune system in susceptibility in CD attracts great attention. Chemokines and their receptors participate in the pathogenesis of various inflammatory diseases, where play an important role in

migration and activation of monocytes and macrophages. Several studies have confirmed that genetic variants in chemokine receptor *CCR5* gene are correlated with susceptibility to CD. Data on prevalence and phenotypic consequences of polymorphism *CCR5* gene in CD in Poland and Bosnian populations are sparse or nonexistent. Therefore, the aim of our study was to assess the frequency of Δ32 allele and their association with phenotypic expression of the disease, in population of Polish and Bosnian patients with CD.

Subjects and Methods: We recruited 86 CD patients mean age 34.1 (±13.0) y and 83 controls mean age 35.4 (±12.8) y in Poland and 30 CD patients mean age 44.1 (±14.5) y and 30 controls mean age 61.3 (± 15.2) y in Bosnia and Herzegovina; 229 participants in total. We determined the prevalence of *CCR5*Δ32 mutation and its association with phenotypic expression of the disease according to Montreal classification. Participants were genotyped for *CCR5*Δ32 mutation by polymerase chain reaction (PCR) and follow-up using the Statistical Analysis Package IBM SPSS Statistics (version 21). We verified the correctness of results by performing re-genotyping of randomly selected samples.

Results: We identified 2 heterozygotes in Bosnian and 8 heterozygotes and 2 homozygotes in Polish CD patients, with mean Δ32 allele frequency 3.3% and 7.0%, respectively. In Bosnian and Polish control group we found 8 and 16 heterozygotes, with mean Δ32 allele frequency 13.3% and 9.8%, respectively.

Conclusion: Increased frequency of the Δ32 allele in CD patients from Poland ($p=0.048$) but not from Bosnia was observed. In Polish CD patients the age of diagnosis between 17 and 40 yo and ileocolonic location were related with higher frequency of *CCR5*Δ32 mutation, while in Bosnian CD patients the correlation was with the age of diagnosis above 40 yo (both $p>0.05$). Further studies with larger samples in both countries are warranted.

PF28
APOLIPOPROTEIN E GENE POLYMORPHISMS (RS429358 AND RS7412)IN THE HEALTHY BOSNIAN POPULATION AS A COMPLEMENT THE EUROPEAN DATA.

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Background: The alleles of *APOE* gene such as: Ɛ2, Ɛ3, Ɛ4 have been associated with a number of diseases in different populations. The frequency of the *APOE* alleles varies worldwide and differences are observed between geographic locations and ethnic populations. In Europe, in Northern countries the average frequency of alleles Ɛ2, Ɛ3 and Ɛ4 is 8.5%, 75.5% and 16.0%, whereas in Middle and Western countries is 8.6%, 76.2% and 15.3%, respectively. In Southern countries the average frequency of alleles Ɛ2, Ɛ3 and Ɛ4 is 7.5%, 80,2% and 12.2%, respectively. Due to important role in metabolism and clinical importance, allelic variation in different populations is analyzed to evaluate the usage of *APOE* in a clinical and evolutionary context. All in all, data on frequency of alleles are numerous but

reports in population of Bosnia and Herzegovina are nonexistent. Therefore, the aim of this study was to estimate the allele frequency for the *APOE* polymorphism in healthy Bosnian subjects and to compare to data in other European countries.

Subjects and methods: We recruited 170 subjects (108 female and 62 male) from Bosnia and Herzegovina, mean age of cohort was 53.0 (±5.0) years. Genotypes were determined by real-time PCR using StepOneTM Real-Time PCR System, Applied Biosystems (AB) and TaqMan SNP Genotyping Assays for two *APOE* polymorphisms including rs429358 and rs7412 (Life technologies, Assay ID: C_3084793_20, C_904973_10, respectively) (AB). The data were analyzed with Taq Man Genotyper Software v. 1.0.1. Electronic databases including Medline and Embase were searched from 1997 to April 2015.

Results: In our group the prevalence of heterozygotes: Ɛ2Ɛ3, Ɛ2Ɛ4 and Ɛ3Ɛ4 was 20.6%, 3.5% and 12.9%, respectively, while prevalence of homozygotes: Ɛ2Ɛ2, Ɛ3Ɛ3, Ɛ4Ɛ4 was 0.6%, 61.2% and 1.2%, respectively, with mean Ɛ2, Ɛ3, Ɛ4 allele frequency of 12.6%, 78.0%, 9.4%, respectively.

Conclusions: The findings of the investigation in Bosnia show higher frequency of Ɛ2 allele than Northern, Middle and Western and Southern European countries, frequency of Ɛ3 allele was lower only in comparison with Southern countries, while frequency of Ɛ4 allele was lower than in all these regions of Europe. For frequency of Ɛ4 allele decreasing linear gradient from North to South was observed.

PF29
THE FIRST SCREENING RESULTS OF SIX LYSOSOMAL STORAGE DISORDERS BY USING A HPLC-MS/MS MULTIPLEX ASSAY IN TURKEY

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***Background*:** Mass spectrometry has been used for the diagnose of lysosomal storage disorders (LSD) such as Pompe, Fabry, Gaucher, Krabbe, Niemann-Pick A/B and mucopolysaccharidosis I in dried blood spots (DBS). Diminished enzyme activities can be simultaneously evaluated by MS/MS determination of the products obtained after incubation with specific substrates. In this study we aimed to investigate a HPLC-MS/MS method for multiplex screening of LSDs in dried blood spots in Turkey.

***Methods*:** Dried blood spots (3.2-mm) were incubated for 20 h with cocktails containing substrates and internal standards. We determined the resulting product and internal standard using LC-MS/MS (Shimadzu 8030 Triple Quadrupole Liquid Chromatograph Mass Spectrometer, Shimadzu Scientific Instruments, Japan). The method did not require offline sample preparation such as liquid-liquid and solid-phase extraction. Between- and within-run imprecision, carryover, limits of detection and quantification were determined. We also analyzed CDC QC samples and 10 samples from patients with known LSDs.

***Results*:** A total of 1000 dried blood samples were analyzed for the lysosomal α-glucosidase, β-glucocerebrosidase, α-galactosidase, acid sphingomyelinase, galactocerebrosidase, and α-L-iduronidase activities. Affected patient's enzyme activities were found as significantly lower. Carryover were not observed, whereas between -run and within-run imprecision were <10%.

***Conclusions*:** Our data shown that the mass spectrometric techniques can be easily used for the screening of lysosomal storage diseases which presents remarkable technical advantages compared with traditional methods. This method allows to significant decreases in sample preparation and analytical times and reagent costs. The screening for several LSDs simultaneously is appropriate for use in high-throughput screening laboratories.

PF30
CHANGES OF SOCCER PLAYERS BLOOD ACID-BASE VALUES IN ONE TRAINING UNIT DURING PREPARATION PERIOD

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Background: The body's acid–base balance is tightly regulated by numerous mechanisms keeping the arterial blood pH between 7.35 and 7.45. Diseases as well as physical stress lead to disturbances of acid base balance, that lead to changes in blood pH and the parameters of blood acid base values (ABS). The aim of this study was to examine changes in the parameters of ABS before and after training (physical activity).

Material and methods: Twenty three male professional soccer players, aged 25,0±4,5 years, participated in this study. Capillary blood samples were collected before and after training to measure concentrations of lactate ([La]), hydrogen ions ($[H^+]$), bicarbonate ions ($[HCO_3^-]$), base excess and the arterial partial pressure of CO_2.

Results: Lactate concentration increased from 2,85±0,70 mmol/L before training to 3,58±1,26 mmol/L after training (p=0,031). Comparing the results for blood pH, measured before and after training, we observed no significant changes (p>0,05). Base excess values and concentration of HCO_3^- decreased in the body after training, resulting in compensatory metabolic acidosis. Training did not affect the concentration of hemoglobin, and consequently the blood oxygen saturation remained unchanged.

Conclusion: The results of the present study show that increased physical activity has resulted in increase of the concentration of lactate in the players muscles. At the same time, no significant changes in blood pH were observed because all measurements were performed on athletes who have metabolic and physiological adaptations of muscles.

PF31
THE INVESTIGATION OF CIRCULATING MICRORNAS ASSOCIATED WITH LIPID METABOLISM IN CHILDHOOD OBESITY.

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Background: Increasing obesity prevalence seen in children and adolescents is one of the most significant worldwide health challenges reaching epidemic proportions. microRNAs (miRNAs) are noncoding short RNA molecules regulating multiple biological processes linked to obesity. We aimed at evaluating the association between circulating miRNAs levels and lipid metabolism in obese and nonobese children and adolescents.

Material and Methods: This study was performed in 45 obese children and adolescents aged 9 to 17 years and 41 lean children and adolescents aged 9 to 17 years. Using real-time quantitative PCR (RT-qPCR) analysis, circulating miRNAs were evaluated in both groups.

Results: Whereas circulating miR-27 ($p=0.032$), miR-378 ($p<0.001$) and miR-370 ($p=0.045$) in obese patients were significantly higher than those of controls, circulating miR-335 ($p<0.001$), miR-143 ($p=0.001$) and miR-758 ($p=0.006$) in the obese were significantly lower, compared to those of controls. In addition, circulating miR-33 in obese children was higher than those of controls, but not significantly different ($p=0.687$).

Conclusion: Our findings show that a significant association is present between circulating miRNAs values and childhood obesity. Anti-miRNAs therapy may be beneficial in the treatment of dyslipidemias in obesity, a risk increasing cardiovascular diseases.

PF32
CIRCULATING LEVELS OF FIBRINOGEN AND C-REACTIVE PROTEIN IN WOMEN WITH BREAST CANCER

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Introduction: The aim of our study was to compare circulating levels of fibrinogen and CRP before and after surgery in all patients with clinical and morphological changes in breast that indicate breast cancer, compared to the levels of fibrinogen and CRP in patients with breast cancer without metastases , with those in women with breast cancer with metastases. We examined the association between levels of fibrinogen and CRP with histology parameters (size of the primary tumor, SHD, TNM).

Materials And Methods: The levels of fibrinogen in plasma were determined by immunonephelometric method and levels of CRP in serum were determined by immunoturbidimetry. We divided patients into two test groups: 1.patients with histologically confirmed diagnosis of breast cancer (50), 2. patients with histologically confirmed diagnosis of benign breast disease (35).

Results: We found a significant difference in preoperative and postoperative values of fibrinogen (p<

0.0001), and CRP (p=0.001), of the whole group, before and after surgery, Mann-Whitney test . There was a significant difference between preoperative CRP (p=0.026), and postoperative (p= 0.036) in the group with metastases. Fibrinogen was significantly higher in patients with metastasis (p= 0.04). The preoperatively fibrinogen values were not significant different between the groups with and without metastases (p= 9.177). There was not significant difference among other values: postoperative fibrinogen (p= 0.553) and CRP (p= 0.869), preoperative CRP (p=0.929). Preoperative and postoperative CRP and fibrinogen were not significantly correlated with tumor size, (SHD) and (TNM).

Conclusion: These biomarkers can give early information about the risk of disease relapse in patients without metastasis and no evidence of recurrence. They can also help in preoperative identification of patients who are at potential risk of incomplete surgical resection of the tumor.

PF33
GINGIVAL CREVICULAR FLUID ALKALINE PHOSPHATASE ACTIVITY AS AN INDICATOR OF SKELETAL MATURITY

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Background: Orthodontic treatment timing has a significant role in the outcomes of orthopaedic treatment in growing patients, especially when there are skeletal disharmonies. The most favourable time for the orthodontic treatment of skeletal disharmonies is before and/or during pubertal growth spurt. Individual skeletal maturity can be assessed by means of several more or less reliable parameters, with the most common being the radiographly-based, such as hand-wrist analysis and the cervical vertebral maturation method. Biochemical markers provide new possibilities in the assessment of skeletal maturity. Biochemical markers avoid x-ray exposure and represent agents that are directly involved in bone growth and remodeling. Alkaline phosphatase is essential enzyme for bone mineralization.

The aim of this study was to assess the gingival crevicular fluid alkaline phosphatase activity in growing subjects in relation to their stages of individual skeletal maturity, as recorded through the cervical vertebral maturation method.

Subjects and methods: Ninety-nine healthy growing subjects (47 boys and 52 girls), aged from nine to sixteen years (mean age 12.2 ± 1.94) were included in this study. Samples of gingival crevicular fluid, for assessment alkaline phosphatase activity, were collected from each subject at distal site of the maxillary right central incisor. Enzymatic activity was determined spectrophotometrically. Skeletal maturity was carried out through the cervical vertebral maturation method on lateral cephalograms (CVMI stages).

Results: The relationship between gingival crevicular fluid alkaline phosphatase activity and cervical vertebral maturation stages was significant ($p<0.01$). Significantly greater enzyme activity was seen in pubertal stages (CVMI 3 and 4), compared to the pre-pubertal (CVMI 1 and 2) and post-pubertal (CVMI 5 and 6) stages.

Conclusion: The assessment of gingival crevicular fluid alkaline phosphatase activity is reliable, non-invasive clinical biomarker for the identification skeletal maturity.

PF34
THE INVESTIGATION OF EFFECTS ON DNA DAMAGE IN BLOOD OF HIGH FAT DIET AND ACRYLAMİDE IN THE RATS

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Background: 8-Hydroxy-2′-deoxyguanosine (8-OHdG) is one of the major reactive oxygen species-induced DNA base-modified products that is widely accepted as a sensitive marker of oxidative DNA damage. Poly (ADP-ribose) polymerase (PARP) facilitates DNA repair by binding to DNA breaks and attracting DNA repair proteins to the site of damage. The aim of this study was to investigate serum 8-OHdG and PARP levels in the rats fed high-fat diet and acrylamide, compared to control rats.

Materıals and methods: Forty-eight male Wistar rats (5-6 weeks of aged) were segregated into two diet groups and fed with a high-fat diet (crude fat 20%) or standard diet (crude fat 2.7%), respectively; and animals in each diet groups were exposed to acrylamide at the dose of 0, 2, 10 or 20 mg/kg bw/day via drinking water for 28 days. At the end of the experiment, serum samples were analyzed for 8-OHdG and PARP with the ELISA method.

Results: Serum PARP levels were significantly higher at concentrations of 2 mg/kg bw/day acrylamide, 10 mg/kg bw/day acrylamide and 20 mg/kg bw/day acrylamide in rats when compared with those of the control rats. In addition, serum PARP levels of high fat diet and 20 mg/kg bw/day acrylamide in rats were significantly higher than those of the control rats. On the other hand, there were no difference between groups in terms of serum 8-OHdG levels.

Conclusions: The results of this study are the first to demonstrate that exposure to acrylamide and high fat diet, generates DNA damage.

PF35
LEVELS OF ERYTHROPOIETIN AND HEMOGLOBIN IN PATIENTS WITH VARIOUS DEGREES OF RENAL INSUFFICIENCY

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Background: Erythropoietin(EPO) is aglycoproteinhormonethat controls erythropoiesis.EPO stimulates the proliferation and differentiation of

erythroid precursor cells in bone marrow. Chronic renal failure leads to hyporegenerativeanemia due to a lack of erythropoietin. This study attempted to establish a quantitative association between erythropoietin and haemoglobinlevels at different degrees of renal insufficiency.

Subjects and methods: This study included 365 patients with different degrees of renal insufficiencyand 98 healthy subjects as the reference group. The examinees were divided in four groupsaccording to the degree of renal insufficiency. For testing, we used the serum and blood samples. We determined erythropoietin, creatinine and haemoglobin, using immunochemical and colorimetric methods. Also, we calculated creatinine clearance.

Results:The results showed the anemia presented in the early stage of renal disease. The patients with the lowest renal function,confirmed with the creatinine clearance less than 15 ml/min/1.73m2 (42% of patients), had the highest prevalence of anemia. This group had the lowest value of the EPO compared to the reference group ($p<0.01$). Patients with decreased renal function (creatinine clearance 30 to 59.99 and 60-89.99 ml / min /1.73m2) had a significantly higher values of erythropoietin compared to the reference group ($p <0.001$; $p <0.01$, respectively).

Conclusion: The results showed that to some extent there is a link between erythropoietin, hemoglobin and creatinine clearance.

PF36
THE RELATIONSHIP BETWEEN LCAT 4886 C/T GENE POLYMORPHISM AND THE RISK OF CORONARY ARTERY DISEASE

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Background:
Lecithincholesterolacyltransferase (LCAT) has a pivotal role in the formation and maturation of HDL-C and in reverse cholesterol transport. The aim of present study was to identify role of LCAT gene polymorphism in patients with coronary heart disease (CHD) and healthy volunteers.

Material and methods: The study included 200 patients aged 18-80 years who were diagnosed as CAD based on angiography and 100 healthy individuals. In this study, 3 groups were formed. Groups; 1) control 2) a single vascular lesion 3) multiple vascular lesion. In this study; demographic characteristics of all individuals were determined. In DNA samples isolated from patients and healthy individuals in the study population, LCAT 4886 C/T gene polymorphisms were detected by real time polymerase chain reaction (RT-PCR). Serum LCAT levels were measured by ELISA method. Serum lipid levels were measured by an autoanalyzer.

Results: Serum tryglyceride, levels were found to be significantly higher while HDL and LCAT levels were found to be significantly lower in CAD group when compared to control group. When LCAT 4886 C/T gene polymorphism was

assessed, there was homozygous genotype in 55, heterozygous genotype in 33 and mutant genotype in 12 of the controls, while there was homozygous genotype in 23, heterozygous genotype in 30 and mutant genotype in 17 of 70 patients with single-vessel disease. However, there was homozygous genotype in 35, heterozygous genotype in 60 and mutant genotype in 35 of 130 patients with multi-vessel disease. Genotype frequencies showed a statistically significant difference between patient and control groups ($p<0.001$).

Conclusions: Our findings suggest that lower serum LCAT levels and higher frequency of TT genotype the mutant genotype in LCAT 4886 position might play a potential role in the susceptibility to coronary artery disease in the Turkish population.

PF37
MEAN PLATELET VOLUME PREDICTS THE GLYCEMIC CONTROL DETERIORATION IN DIABETES MELLITUS TYPE 2 PATIENTS

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Background: Inadequate glycemic control enhances platelet activation in Diabetes mellitus type 2 (DMT2) patients. The glycosylated hemoglobin (HbA1c) value should be kept below 7% in order to prevent the development of diabetes complications. The study aim was to investigate the association of mean platelet volume (MPV), a surrogate marker of platelet activation with the short or long term glycemic control as well as to find out whether MPV could be used as a predictor of deterioration of glycoregulation in DMT2 patients.

Subjects and methods: The cross-sectional study enrolled 117 DMT2 patients, both sexes (75 females, 42 males), with a mean value of age 62 years (SD=12) treated at the Primary Health Centre in Zenica from March to May 2015. According to the HbA1c values patients were distributed into groups: A (n=49, HbA1c ≤7.0%) and B (n=68, HbA1c>7.0%). Fasting blood glucose (FBG), HbA1c and MPV were measured using standard laboratory methods. Binomial logistic regression analysis was performed to estimate the relationship between glycemic control, as dichotomous outcome and MPV as the main predictor.

Results: The significant, higher values of FBG, HbA1c and MPV were observed in the group B compared to the group A [FBG (11.4 (9.9-14.8) vs. 8.0 (7.2-8.9) mmol/L, P<0.001; HbA1c (8.8 (8.0-10.0) vs. 6.8 (6.4-7.0) %, P<0.001; MPV (10.5 (9.8-11.5) vs. 9.5 (9.0-10.1) fL, P<0.001, respectively]. There were no correlations of FBG and HbA1c with MPV in the group A diabetic patients (P>0.05) while HbA1c correlated positively with FBG and MPV (rho=0.372, P=0.002; rho=0.279, P=0.021, respectively) in the group B. Glycosylated hemoglobin correlated with FBG (rho=0.310; P=0.030) in the group A. Mean platelet volume was positively associated with the risk of inadequate glycemic control, with 2 times increased odds of inadequate glycemic control per femtoliter greater MPV (P<0.0005; Exp (B) =2.13; 95% CI=1.47 - 3.10) .

Conclusions: Mean platelet volume correlates with HbA1c in poorly controlled DMT2 patients. It could be used as a simple, effortless and cost-effective predictor of deterioration in long term glycoregulation.

PF38
URINARY NETRIN-1 LEVELS IN PREECLAMPTIC PREGNANTS

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Backgound: Preeclampsia is the most common cause of maternal and perinatal morbidity and mortality. Kidneys are organs where netrin-1 is highly expressed. Although the role of netrin-1 in kidney pathophysiology is not fully understood, studies have shown that netrin-1 suppressed ischemia-reperfusion-induced apoptosis. The aim of the present study was to determine whether tubular injury marker urinary netrin-1 level is increased in patients with preeclampsia and correlates with severity of disease.

Material and methods: 35 preeclamptic (based on American College of Obstetricians and Gynecologists) and 35 normotensif pregnant included in our study. All paarticipants were in third trimester before labor. Serum and spot urine samples were stored in aliquots at -80°C until analysis. Urinary netrin-1 levels specified in the project was studied by ELISA. Other biochemical analysis were determined by routine biochemical methods.

Results: We measured higher urinary netrin-1 levels in preeclamptic group than normotensive group ($p>0.05$). But we could not find any correlation between urinary netrin-1 levels and preeclampsia grade ($p>0.05$). In addition, siginificant differences were found between groups by means of serum urea, creatinine levels and birth weight ($p>0.05$). Moreover, we found the perinatal outcome was evidently worse for the preeclamptic ones.

Conclusion: Although changes in renal morphology occurs in glomerular endothelial cells, we found increased levels of the tubular injury marker netrin-1 in urine in patients with preeclampsia. Our results suggest that tubular damage might be in preeclampsia pathogenesis.

PF39
RISK MANAGEMENT IN MEDICAL LABS

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Background: Organizations of all types and sizes face internal and external factors and influences that make it uncertain whether and when they will achieve their objectives. The effect this uncertainty has on an organization's objectives is the "risk".
According to the accreditation standards of ISO 15189: 2012, the laboratory shall evaluate the impact of work processes and potential failures on examination results as they affect patient safety, and shall modify processes to reduce or eliminate the identified risks and document decisions and actions taken (Clause 4.14.6).

Materials and methods: For a medical lab to be accredited, a complete risk management plan must be prepared, documented, communicated and strictly implemented everywhere in the laboratory. Risk management is not a stand-alone activity separated from the main activities and processes of the lab. It is a part of the responsibilities of

management and an integral part of all lab processes, including strategic planning and change management processes.

Results: The main purpose of the risk management plan in a medical lab is to secure the work flow and to tightly control the analytic steps to get a better performance that could help the appropriate diagnosis of patients. Moreover, Risk management plan aims to manage all foreseeable risks (both opportunities and threats) in a manner which is proactive, effective and appropriate, in order to maximize the likelihood of the lab achieving its objectives, while maintaining risk exposure at an acceptable level.

Conclusion: In order to establish a risk management plan, several steps should take place sequentially namely; establishing the context, risk assessment, risk identification, risk analysis, risk evaluation and risk treatment. There are two more steps that are taking place along with the previously mentioned steps; those are communication and continuous monitoring.
The establishment of a medical lab risk management plan will be briefly discussed and illustrated.

PF40
SERUM CYSTATIN C LEVELS IN PATIENTS WITH CHRONIC RENAL DISEASE

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Background: Cystatin C is a cystatine protease inhibitor. In most conditions it is produced at a constant rate. Catabolism is almost completely by glomerular filtration and proxsimal tubular degradation. Cystatin C concentration in serum depends mainly upon the glomerular filtration rate (GFR). Accordingly, serum cystatin C is favorable endogenous parameter for the measurement of glomerular filtration rate. Our aim to demostrate that cystatin C could be proposed as an alternative for screening of chronic kidney disease.

Methods: Serum creatinin and cystatin C were measured simultaneously. Creatinine was determined on the RxLDimension system. Measurements of cystatin C were performed by turbidimetric immunoassay on Roche-Cobas c 501 analyzer. GFR was calculated using MDRD formula. Analytical assessment of cystatin C determination was comprised of within-run and between-run imprecision. For quality control we use Control 1 and 2. Cystatin C was measured in sera obtained from 45 healthy subjects and 32 patients with chronic renal failure (CRF). Median age of CRF patients was 52 year old–and included 19 females and 13 males.

Results: Within-run imprecision on the commercially controls for Control 1 is 4,6% and 3,6 for Control 2; between-day imprecision for Control 1 is 7.5% and 8.2% for Control 2 respectively. Median values for Cystatin C were significantly higher in patients comparedto control group (0.42 vs. 1.92 mg/l, $p<0,0001$). Mean value of serum creatinine was 178.2,Cystatin C ±49.3 µmol/L, and mean value of creatinine clearence was 43.2±19.3 ml/min. In CRF patient there was a high statistically significant positive correlation between creatinine concentration and cystatin C concentration ($p<0.01$) and there was a high statistically significant negative correlation between MDRD clearence and cystatin C concentration ($p<0,01$).

Conclusion: The presented results of the analytical evaluation methods for the determination of cystatin C on the Roche-Cobas c 501 analyzer showed an acceptable accuracy and precision. Cystatin C is good parameter, to determine the glomerular filtration rate in CRF patients.

PF41
SERUM HOMOCYSTEINE LEVELS AND METHYLENETETRAHYDROFOLATEREDUCTASE GENOTYPES IN COUPLES WITH IDIOPATHIC RECURRENT MISCARRIAGE

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Background: Hyperhomocysteinemia and methylenetetrahydrofolate-reductase (MTHFR) gene mutation have been postulated as a possible cause of recurrent miscarriage (RM).The aim of this study was to investigate the association between MTHFR genotypes and serum homocysteine levels in Macedonian couples with idiopathic recurrent miscarriage.

Material and methods:In this study were included 29 couples with idiopathic recurrent miscarriage. After genotyping all 58 individuals were divided into three groups. First group, controls (N=28) (no more than one mutation in both loci of MTHFR gene C667T and A1298C).Second group (N=13), homozygous (both copies of either the C677T mutation, or the A1298C mutation). Third group (N=17), compound heterozygous (one copy of the C677T mutation and one copy of the A1298C mutation). Genotyping was performed by reversal hybridization with CVD strip assay manufactured by Vienna Lab - Austria. Serum homocysteine levels were measured by chemiluminescent immunoenzyme assay.

Results: In the first group we found 5 individuals with elevated homocysteine level (>15 μmol/L) (17,8%), in the second group 3 individuals (23%), in the third group 6 individuals were with elevated homocysteine level (35%). The level of homocysteine was: first group 12,46±7,32; second group 13,12±6,91; third group 13,94±7,16. No significant differences were observed in serum homocysteine levels between the three studied groups($p>0.05$).

Conclusions: We found that elevated level of homocysteine is most frequent in the third group, in compound heterozygous. In the present study RM is not associated with hyperhomocysteinemia.

PF42
SERUM VITAMIN E IN PATIENTS ON MAINTENANCE HEMODIALYSIS

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Background: Vitamin E is an essential nutrient and important component of nonenzymatic antioxidant defense. Although an indiscriminate supplementation with antioxidant vitamins has been shown to have even harmful effects in the general population, a recent meta-analysis demonstrated that chronically hemodialyzed patients might benefit from vitamin E supplementation. Vitamin E increases LDL resistance to oxidation, reduces the number of muscle cramps, and improves erythropoietin responsiveness in chronically hemodialyzed patients. However, a broad consensus about the dose and length of vitamin E supplementation is lacking, especially in relation to the basal vitamin E concentrations.

Subjects and methods: Sixteen end-stage renal disease patients who had been on maintenance hemodialysis (HD) with a protocol of 3 HD sessions per week for more than two years were included in this study. Blood for analysis was taken immediately before and after a single HD session.
Twenty healthy individuals, nonsmokers, non-obese, without any acute or chronic disease, who did not take any medications, vitamins or supplements, were included in the study as a control group.
Alpha- and gamma-tocopherol were measured with a HPLC-fluorescence method.

Results: Alpha-tocopherol is major vitamin E component in both HD patients (alpha-tocopherol: 37.4 ± 7.3 μmol/L, gamma-tocopherol: 2.0 ± 1.5 μmol/L, both measured before the single HD session) and healthy subjects (alpha-tocopherol: 36.5 ± 4.7 μmol/L, gamma-tocopherol: 2.0 ± 0.6 μmol/L). A single HD session slightly, but significantly increases both components (42.4 ± 8.2 μmol/L and 2.2 ± 1.4 μmol/L), which can be attributed to the hemoconcentration resulting from HD. There is not a statistically significant difference of the serum tocopherols between the patients before HD session and the control subjects.

Conclusions: Severe vitamin E deficiency is not present in the study group of chronically hemodialyzed patients. Given the increased oxidative stress in chronically hemodialyzed patients, those with clinical manifestations of muscle cramps and/or hypo-responsiveness to the erythropoietin treatment, and with serum vitamin E concentrations within lower reference range, might be considered for vitamin E supplementation.

PF43
URINARY BETA-N-ACETHYLGLUCOSAMINIDASE ACTIVITY DURING STRESS TOLERANCE TEST IN INDIVIDUALS WITH EXERCISE INDUCED PROTEINURIA

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Background: Exercise induced proteinuria is a type of asymptomaticproteinuria and represent a common condition in school-age children and teenagers, related with increased physical effort. The aim of this study was to assess the variability of urinary protein excretion and beta-N-acethylglucosaminidase activity (beta-NAG) during stress tolerance test in young individuals with exercise induced proteinuria.

Methods: Theevaluation of the changes in qualitative and quantitative composition of urinary proteins, with SDS-PAG electrophoresis, in young individuals 7-24 years old, enabled us detection of subjects with exercise induced proteinuria. Five urinary samples were used excreted during stress tolerance test: two samples of first morning urine, two samples of daily urine and one sample of urine excreted after physical effort. The activity of the enzyme beta-NAG, a sensitive marker of tubular damage, was determined in all five urinary samples in 30 individuals with and 20 without exercise induced proteinuria, aged matched. Beta-NAG activity and creatinine concentration in urine samples were determined using spectrophotometric methods. Enzyme activity was expressed in U/g creatinine.

Results: In subjects with and without orthostatic proteinuria, the highest mean values for beta-NAG activity were detected in second morning urinary samples (4.4 U/g creatinine) and the lowest mean values were detected in samples excreted after physical effort (3.3 U/g creatinine). Besides variations in beta-NAG activity in five samples urine excreted during stress tolerance test, the activity of beta-NAG in all individuals were within the reference intervals.

Conclusion: The results lead to conclusion that there is no significant tubular damage in individuals with exercise induced proteinuria.

PF44
COMPARISON OF CD34 FLOW CYTOMETER BECMAN-COULTER FC-500 VALUES WITH WBC VALUES OBTAINED FROM SYSMEX XE-2100 HAEMATOLOGY ANALYSER AND OPTICAL MICROSCOPY OF MONONUCLEAR CELLS FROM THE APHERESIS PRODUCT

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Background: Autologous and allogeneic types of hematopoietic stem cell transplantation are used for treatment of malignant and non-malignant haematological diseases.The objective of this paper is to explore the correlation between CD34 performed on BECMAN-COULTER FC-500 flow cytometer, WBC performed on haematology analyser Sysmex XE-2100, (TOA Medical

Electronics, Kobe, Japan) and optical microscopy of mononuclear cells from the apheresis product (MNC).

Examinees and methods: Data were collected in the period from February 2013 to December 2014. In the given period, at UCC Tuzla conducted were 33 stem cell transplantations. WBC number from the apheresis material, together with CD34 number and mononuclear cells count performed by optical microscopy for all patients were determined.

Results: By using Pearson's linear correlation, values ratio of variables CD 34, % MNC and WBC was explored. Statistically significant correlation was not detected between variables % MNC and CD 34 of values with the probability of $p<0,772$ ($r= -0,056$). No statistically significant correlation was also detected in the correlation of WBC and CD 34 values ($p<0,149$, $r=0,275$). However, statistically significant moderated negative correlation was obtained ($p<0,010$) ($r= -0,444$) between WBC and % MNC values.

Conclusion: Testing showed discrepancies between WBC, CD34 and MNC in apheresis product. WBC values obtained from the haematology analyser Sysmex XE-2100 and MNĆ values obtained by optical microscopy of the apheresis product were in significant correlation.

PF45
NEOPTERIN IN PREDICTION AND THE OUTCOME OF NEONATAL INFECTION

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Introduction: Neonatal mortality from infections despite early diagnosis and optimal treatment is still high and does not decrease despite a new generation of antibiotics and the improvement of neonatal care.After treatment,neurological sequelae can be expected in approximately 20-50 % of children . There are many attempts to develop screening tests that can help detection of infected infants at a time when it is necessary to give an initial therapy.

The Aim of study : Examine the incidence of infection, neopterin values, haematological parameters in bouth groups. Evaluate predictive value of neopterin in the diagnosis and outcome of neonatal sepsis.

Materials and methods : A prospective study conducted at the Clinic for children's diseases and the Department of Obstetrics and Gynecology of the University Clinical Centre in Tuzla , included 66 infants of both sexes, gestational age 28-42 weeks . This study analyzed data on pregnancy and childbirth,newborn characteristics and values of newborn 's neopterin ,CBC and CRP .

Results: Neopterin was studied in 66 infants, of which 34 had neonatal infection, and 32 had no symptoms of disease. A positive value - neopterin above 10nmol/ L in the diagnosis of neonatal infection had a 100% positive predictive value, high negative predictive value of 94%, sensitivity of 54% and specificity of 100%. The diagnostic value of the test was 97%. The positive predictive value of neopterin in predicting the outcome of infants with neonatal infection was 9% with the high sensitivity of 100% and specificity of 51%. The diagnostic value of the test was 5%. Neopterin had higher sensitivity than CRP and hematology parametars.

Conclusion : Our study found that neopterin is more specific marker of neonatal infection than CRP and hematology parameters.

PF46
RETICULATED PLATELETS IN PATIENTS WITH TROMBOCYTOPENIA AND THEIR CORRELATION WITH COAGULATION FACTORS

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Background: Thrombocytopenia is a disorder in which there is an abnormally low platelets count.

In the evaluation of thrombocytopenia, quantification of reticulated platelets may be used as an index of megakaryopoiesis. Reticulated platelets are immature platelets that circulate the blood, their number reflects the thrombopoietic activity in the bone marrow and they are analogous to the red cell reticulocyte count in the evaluation of anemia. Aim of this study was to evaluate reticulated platelets percentage (RP%) in the blood of patients with trombocytopenia and to investigate possible correlations with coagulation factors.

Materials/subjects and methods: Study was designed as a retrospective observational study and it included 32 patients; 12 male and 20 female subjects of the Hematologic clinic, University Clinical Center Sarajevo. All subjects were diagnosed with thrombocytopenia. All study subjects have undergone a serie of laboratory tests: Coagulation tests were performed on Siemens BCS Analyzer and included INR, a-PTT, fibrinogen, coagulation factors (II, V, VII, VIII, IX, X, XI, XII, XIII), D-dimer, protein S, protein C and antitrombin III; Hematologic tests and RP% were performed on CELL-DYN Sapphire Analyzer using flow cytometry. Correlation between blood values of laboratory parameters was determined by Pearson correlation coefficient.

Results: Average percentage of reticulated platelets in thrombocytopenic patients was 5.68% (MIN 0.46%; MAX 16.3% with STDEV= +/- 4.38%). RP% correlated strongly with platelet count as a negative correlation ($p<0.00001$) meaning that RP% tends to be higher in patients with lower platelet count, and vice versa. RP% showed no statistically significant correlation to coagulation factors, except Factor VIII, and Protein S. Correlations between Factor VIII - RP% and Protein S - RP% were positive correlations ($p< 0.001$ and $p<0.0068$ respectively).

Conclusions: RP% is a relatively new parameter that needs extensive research in its clinical application, esspecially in patients with thrombocytopenia. RP% also needs to be investigated further in correlation with other laboratory parameters such as coagulation factors since therapy for thrombocytopenia is closely related with anticoagulant therapy.

PF47
IDENTIFICATION OF THE PREANALYTICAL ERRORS IN THE HOSPITAL LABORATORY-6 MONTHS STUDY

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Background: Pre- and post-analytical errors are estimated to constitute 93% of errors. Errors at any stage of the collection, testing and reporting process can potentially lead to a serious patient misdiagnosis. Errors during the collection process are not inevitable but can be prevented with a diligent application of quality control, continuing education and effective collection systems.

Materials and methods: A perspective analysis of the results obtained from the biomedical laboratory of Clinical center of Nis, Serbia for errors of the preanalytical phase has been carried out to summarize data. Laboratory personel were asked to register rejections, and causes for rejection of wards.

Results: Out of the 48.328 blood collection tubes screened over a period of 6 months, pre-analytical errors were observed in approximately 1.9% of the total number of samples received. The distribution of the different types of errors was then calculated. The majority of the rejected samples were hemolyzed, which accounts for 1.1% of the total number of samples received during this period. The amount of blood was insufficient for complete analysis in 0.08%. Another factor leading to rejection of blood samples in this study was insufficient blood volume(0.08%). A total of 0.4% samples in the wards were accompanied by innapropriate requisition slips.

Conclusion: The human role in samle collection makes complete elimination of errors associated with laboratory testing unrealistic. However, good practise and compliance with the new strategies for error prevention can lead to a substantial reduction in pre-analytical errors. A practice of keeping a record of the errors at all stages of analysis and then divising corrective strategies for their prevention can gradually free a laboratory from such errors.


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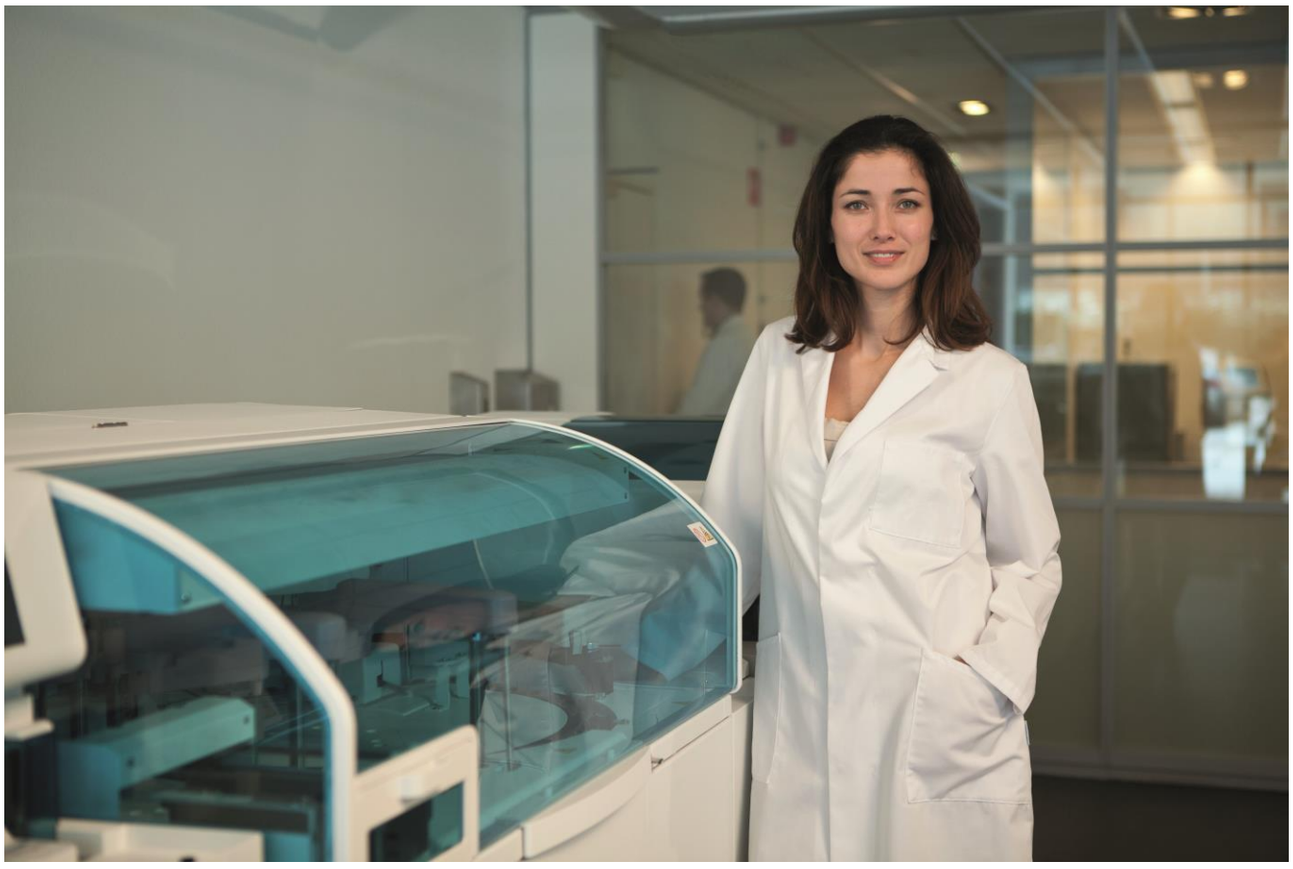

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