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ARAB FEDERATION OF CLINICAL BIOLOGY AFCB

EFLM EUROPEAN FEDERATION OF CLINICAL CHEMISTRY AND LABORATORY MEDICINE

LM4MS

SECOND AFCB – EFLM CONFERENCE

LABORATORY MEDICINE FOR MOBILE SOCIETIES IN OUR AREA

ABSTRACTS BOOK

2 — 5 OCTOBER **2022**

Aquila - Atlantis Hotel, Heraklion, Crete, Greece.

BCLF

GSCC-CB GREEK SOCIETY OF CLINICAL CHEMISTRY CLINICAL BIOCHEMISTRY

Co-organized with the 20th GSCC-CB annual Congress and XXIX BCLF annual meeting

UNDER THE AUSPICES OF

IFCC

ΠΑΝΕΠΙΣΤΗΜΙΟ ΚΡΗΤΗΣ UNIVERSITY OF CRETE

Dr. Konstantinos Makris
Conference President

Dear Colleagues and Friends,

On behalf of the Organizing and Scientific Committee I have the special pleasure and honor to invite you to the joint **Meeting of BCLF, GSCC-CB** coorganized with the **AFCB- EFLM LM4MS Second Conference**. The main theme of this Conference will be **the Laboratory Medicine in the Mediterranean area.** The whole event is organized under **the auspices of IFCC** and of the **University of Crete**, and will take place **from October 2nd to 5th, 2022 in Heraklion, Crete, Greece**.

The Main Conference (and all the Satellite Symposia) will be conducted in a fully hybrid way (physical presence and zoom presentation streamed online) in order to comply the current Health Protocols for the organization of conferences and to allow to attend as much as possible delegates and presenters.

The aim of this conference is to initiate discussions between Laboratory Scientists and Clinicians that are in the front line of providing medical services to refugees and immigrants around the Mediterranean Area, and to propose to Health Authorities the best solutions for laboratory testing and health screening on the mobile societies settlements (hot spots, camps etc.).

This will help to evaluate precisely the health and medical needs of the mobile populations as also as to identify problems that arise to host countries and their populations, in order to propose solutions.

Target audience is specialized laboratory scientists (Biochemists, Chemists, Biologists, Microbiologists, Pathologists, etc.) from Greece and abroad, as well as clinicians involved in treating mobile and local populations and healthcare administrators.

During the conference there will be presentations of distinguished Greek and foreign scientists, Symposia, Round Tables, as also as exhibition of IVD instruments and reagents and Satellite Symposia organized from IVD Companies for the presentation of new methodologies and/or instrumentation.

Heraklion is the capital city of Crete, the big Island at the center of Eastern Mediterranean area, hosting a University, many Research Centers, big Hospitals and Clinical Laboratories with a broad spectrum of specialization and size.

Heraklion, also known as Iraklio, is a port city and is also known for the Palace of Knossos, just outside the city. The huge archaeological site dates back thousands of years to the Minoan civilization, and includes frescoes and baths. Guarding the city's Venetian port is the 16th-century Koules fortress. Heraklion Archaeological Museum has a large collection of Minoan art.

Looking forward to welcoming you in Heraklion in October 2022,

Konstantinos Makris

LABORATORY MEDICINE FOR MOBILE SOCIETIES IN OUR AREA

SCIENTIFIC PROGRAM COMMITTEE

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UNDER THE AUSPICES OF

ATTENDANCE:
Physical (expected): 250+ by streaming (expected) 2000+.

PUBLICATION:
A special issue of an indexed international Journal will be published with manuscripts related to each presentation.

CONFERENCE ORGANIZING COMMITTEE

Alexander Haliassos	President of the G.S.C.C.-C.B
Evgenia Konsta	G.S.C.C.-C.B Board Secretary
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Congress Secretariat: Sofia Kourbassi

ABSTRACTS

TESTING FOR MALNUTRITION AND SKELETAL HEALTH IN CHILDREN OF REFUGEES AND IMMIGRANTS

Dr Artemis Doulgeraki, MD, PhD, MRCPCH, FRCPCH

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Millions of people mainly from Syria, Afghanistan and Iraq fled their country because of war recently. The Mediterranean and the Balkan route towards Europe were used by 1,112,332 refugees during the period 2014-2019; one third of those were children.

In immigrant and refugee children, malnutrition and skeletal issues such as nutritional rickets are frequent, preventable and treatable and they should be diagnosed early. The CDC, EU Commission and the European Academy of Paediatrics have issued relevant guidelines.

Malnutrition leads to increased susceptibility to infections, impaired cognition and all-cause mortality. It includes fetal growth restriction, micronutrient deficiencies, wasting (acute malnutrition, weight for height Z-score), stunting (chronic malnutrition, height for age Z-score) and the anthropometric definitions are provided by WHO and CDC, including body mass index (underweight, overweight, obese). Useful clinical tools include height/length, weight and mean upper arm circumference. There are specific softwares for Z-score calculations, such as WHO Anthro (age<5y) and Anthro Plus (age >5y). Suggested laboratory indices (as a minimum) are complete blood count with differential, ferritin, lead levels and vitamin D. Iron deficiency anaemia in particular should be detected promptly and treated, because it is the most frequent nutritional deficiency encountered, along with vitamin D deficiency.

Regarding skeletal health, all children migrants are at risk for nutritional rickets (symptomatic, severe vitamin D deficiency, 25-OH-D <12 ng/ml) and manifests as irritability, fatigue, bone pain, skeletal deformities, enamel defects, delayed motor development, recurrent respiratory infections, stunting and fractures. If accompanied by severe hypocalcaemia, then tetany, seizures, laryngospasm, cardiomyopathy or even death may ensue. A thorough clinical assessment is mandatory. Vitamin D as a minimum should be checked in all migrants; then, depending on severity, other markers, such as Ca, P, ALP can be measured and X-rays of the wrist or knee can be performed.

To prevent malnutrition and impaired bone health, empirical supplementation of pregnant and/or lactating mothers and children<5y old with micronutrients, including iron and vitD, is recommended. Catch up growth is possible, therefore dedicated, culture-friendly paediatric units to offer screening, recording of health history, general preventative care and treatment are urgently needed.

TOXICOLOGY AND DRUGS OF ABUSE TESTING IN THE CAMP

Pr. Hedhili Abderrazek

DPM Tunisia

Migration has become one of the main indicators of health and social development in the world. Migration affects all regions of the world including Africa, Europe, the Mediterranean region, Asia, the Middle East, Latin America and North America. However, the past decades have shown that migration flows are increasingly undertaken with fake or

fraudulently obtained documents, to dangerous overland journeys across deserts and mountains.

In 2016, 2.5 million migrants were smuggled.

All too often, illegal migration is accompanied by profound shifts in values, identity, and status, and is eventually tied to drug abuse and terrorism.

The underlying reasons for drug use among immigrant and particularly immigrant children differ widely: availability of illicit chemicals, mental, physical and/or sexual exploitation and abuse, or trafficking, socioeconomic conditions, social marginalization, and expressions of anger or frustration due to the challenges of integration.

The high stakes behind the issue of illegal migration and its inextricable link to global health calls for widespread awareness of this phenomenon accompanied by concrete measures to safeguard public health and protect the main victims from potential abuse.

HOW TO APPLY THE USUAL QUALITY ASSESSMENT TOOLS TO AI LABMED APPLICATIONS

Damien Gruson 1,2

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Abstract

Laboratory Medicine is at the heart of a changing health ecosystem where emerging technologies, smart testing, remote monitoring, and data science are playing a central role. Clinical laboratories are also space for routine integration of multi-omics platforms (including genetics, epigenetics, transcriptomics, metabolomics, and proteomics), providing new services and pathophysiological insights to physicians. The resulting "augmented" laboratory medicine should have benefits for the management of patients and clinical outcomes.

To develop powerful integrative approaches, the use of artificial intelligence (AI) is evident and will improve the clinical decision support, risk modeling and treatment of complex diseases.

With the growing role of AI and mobile health devices, it is important to adopt the appropriated validation standards and carefully assess the quality of these complex data-driven AI algorithms before they are to be applied and disseminated in daily healthcare.

Clinical laboratories, specialists in laboratory medicine and coalition of caregivers are important actors to guarantee a safe and ethical use of AI and emerging technologies in healthcare.

Neonatal Diabetes in the ERA of Precision Medicine

Ioannis Papassotiriou[1,2,] PhD, EuSpLM, Nicolas C. Nicolaides[1,] MD, PhD, Christina Kanaka-Gantenbein[1,] MD, PhD

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Neonatal diabetes is a rare disease with an incidence that ranges between 1:100000 and 1:400000. It may occur alone or in the context of genetic syndromes. In its transient form, neonatal diabetes remits and eventually relapses in the next years, most commonly around puberty, whereas permanent neonatal diabetes does not remit at all. Finally, syndromic neonatal diabetes may exist as one of the several clinical features of a syndrome. Both the transient and the permanent form of neonatal diabetes contribute equally for the 90% of cases, while the syndromic form accounts for the rest 10%. Although, neonatal diabetes can be diagnosed within the first days of life, hyperglycemia in the neonatal period may have a long list of differential diagnosis. Indeed, sepsis, increased parenteral glucose administration, steroids and increased concentrations of counter-regulatory hormones represent typical causes of neonatal hyperglycemia that need to be ruled out. In the era of precision medicine, Next-Generation Sequencing (NGS) technologies are increasingly becoming useful tools in the diagnosis and identification of novel genetic causes of neonatal diabetes. It is now worldwide accepted that neonatal diabetes, albeit a genetically heterogeneous Mendelian disorder, has benefitted from the ever-increasing application of the targeted NGS, Whole-Exome Sequencing (WES) and Whole-Genome sequencing (WGS) in diagnostic, therapeutic and translational research settings. Novel mutations are identified and new genes are being reported, giving us a better understanding of the molecular (patho)physiology of β-cell function. Of great importance, some of the identified genes encode proteins that can be therapeutically targeted by per os administration of drugs, leading to the transition from insulin to sulfonylureas. However, it should be noted that all NGS methodology steps (wet lab, bioinformatics analysis and variant interpretation), should undergo quality management and standardization and should comply with the guidelines of the College of American Pathologists Laboratory Standards and the American College of Medical Genetics and Genomics, and/or other Societies. Nevertheless, the concurrent tremendous progress of bioinformatics and big data storage will offer a better clinical outcome for patients suffering from transient, permanent or syndromic form of neonatal diabetes.

POCT molecular biology tests for infectious diseases.

Pr Florence Doucet-Populaire, AP-HP, Université Paris-Saclay

Despite progress in terms of treatment and prevention, infectious diseases remain one of the main causes of morbidity and death. The situation even tends to reverse with re-emergence of "old" pathologies such us tuberculosis or syphilis; emergence of new infectious agents such as legionella, new influenza viruses, coronavirus (COVID-19) and emergence of multi-drug resistant micro-organisms such as carbapenemase-producing enterobacterales. Screening for certain infectious diseases is important and, if appropriately implemented, can be cost-effective and contribute to the prevention of disease in migrants and their host communities in Europe.

Molecular diagnostics are the key for a rapid diagnostic of infectious diseases. Molecular biology Point-of-care tests (POCT) provide rapid 'on-site' results at the site of care delivery, and in resource-limited settings, supporting timely and proper treatment. However, in view of the complexity of molecular diagnosis and the biosafety requirements involved, pathogen nucleic acid POCT is different from traditional blood-based physical and chemical index detection. According to the World Health Organization (WHO), POC tests that address infectious disease control needs should follow "ASSURED" criteria: affordable, sensitive, specific, user-friendly, rapid and robust, equipment-free and deliverable to end-users. This use of POCT in infectious disease surveillance, prevention and control has been supported by ECDC, for example in its guidance on infectious disease screening in migrants within EU/EEA Member States and the UK in which POCT in primary care settings is recommended, when appropriate, for HIV, hepatitis B and hepatitis C. The Next Phase of Molecular Diagnostics development is the Multiplex Molecular POCT for Syndromic Infectious Diseases with fast turnaround time including sample preparation, feasible at bedside without support infrastructure, or with minimal decentralized logistical and structural support (mini-l

ab). Ultimately, multiplexed molecular POCT leads to pathogen specific treatment and the use of narrow-spectrum antibiotics instead of excessive use of broad-spectrum antibiotics. In parallel, recent developments in smartphone-based POC tests for infectious diseases are promising. In conclusion, the molecular biology POCT are on-demand, near-Patient Technology allow diagnostic of infectious diseases with high portability and detection speed.

PROFICIENCY TESTING (EXTERNAL QUALITY ASSURANCE) - ENSURING QUALITY IN ALL ASPECTS OF POCT

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POCT is the most rapidly growing field in laboratory medicine. With increasing technological and analytical possibilities, an increasing number of analyses can now be carried out on POC instruments. Although the costs of POC instruments are less than hospital instruments, the number of users of POC instruments are much larger, ranging from wards in the hospitals, GP offices, nursing homes, pharmacies and last, but not least tests for self-measurements. With the increasing emphasis on patient empowerment, this is a wanted development.

Quality control as well as external quality control/proficiency testing are well-established routine in laboratory medicine. Since POCT is carried out in a different environment with different users and often with different performance specifications and different types on inbuilt controls, we have to re-evaluate how and which types of EQA schemes we should use. It is necessary to assure quality in all aspects in the different phases of the total testing process. An EQA provider for POC must therefore have schemes for pre-analytical, analytical and postanalytical phases of the testing process. In the lecture it will be addressed how this can be done in practice and how this is done for the 3600 POC users in Norway.

LABORATORY MEDICINE WITH REDUCED RESOURCES AROUND THE MEDITERRANEAN SEA

Dr. Ghassan Shannan, BSc, PhD

Syrian Clinical Laboratory Association Al Rasheed Private University

Laboratory medicine provide sizable and important services in the diagnosis and management of diseases in any health setting. Despite this fact, the average expenditure on IVD is less than 1% of Total Health Expenditure, THE. In addition, in low income, lower middle income and upper middle-income countries with limited resources the Total Health Expenditure, THE is very low; consequently, the IVD share is also dramatically low.

Health indicators such as life expectancy at birth, maternal maternity rate, health workforce and hospital beds have been compared between southern and northern countries of the Mediterranean. A clear gap is obvious between southern and northern countries.

A review of Medical Laboratory Practice in limited resources countries will be discussed. A strategy should be established which should include the followings:

1. Financing
2. Human Resources
3. Education & Training
4. Infrastructure
5. Equipment
6. Quality Management
7. Evidence Based Laboratory Medic

Challenges to pursue accurate and satisfactory service will also be discussed.

- The absence of essential infrastructure
- Laboratory supplies
- Basic equipment
- Skilled personnel
- Supply chain management including Cold Chain
- Equipment maintenance
- Reliance on empirical treatment
- Inadequate quality management systems
- No government standards for laboratory testing
- Limited Income of the Population in Developing Countries

We will shed some lights on various points for consideration, including but not limited:

- Delivering modern, high-quality, affordable pathology and laboratory medicine to low-income and middle-income countries.
- Ready- or Home-made Reagents
- New or Refurbished Equipment
- Closed or Open Systems??
- Equipment Donation??
- Organ Function Tests??
- Costly Tests for the sake of Diagnosis only with no Apparent Cure??
- Tests for extremely expensive treatment??

INTER-LABORATORY EXCHANGE OF KNOWLEDGE AND TECHNOLOGY AROUND OUR SEA - A MATTER FOR YOUNG SCIENTIST

Rania Abu Seir

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Introduction

In the modern era, scientific communication became the cornerstone of success in biomedical research, whether written or oral. Furthermore, good communication can encourage the participation of young scientists. Laboratories are an essential part of health systems and play a critical role in detecting, diagnosing, treating, and controlling diseases. The continuous expansion in knowledge, methodology, and technology is impacting the demands placed on medical laboratory scientists (MLSs). Research activities can develop critical thinking and problem-solving skills required for MLSs, promote teamwork and effective communication, and improve experience and skills required in different domains.

Aims

To explore the barriers to involvement in scientific activities among young medical laboratory scientists in Palestine.

Methods

We conducted an online cross-sectional survey among young medical laboratory scientists between the ages of 21-45

years. The questionnaire assessed involvement in scientific activities, self-reported research experience, and perceived barriers to participating in scientific research.

Results

Of 114 participants, 17.8% had a master's degree. The majority of participants (89.5%) reported having a self-interest in scientific research. A large proportion reported attending conferences (77.2%) and participating in continuing education courses (76.3%) and training workshops (52.6%) at least once a year. However, around 60% reported never participating in a conference and less than 15% reported being published authors. Personal barriers such as lack of sufficient time and overwhelming workload had the highest mean scores followed by administrative barriers such as lack of support from academic institutions, professional mentorship, and sufficient opportunities to pursue scholarly interests. Furthermore, we observed significant differences in research experience between holders of bachelor's degrees and master's degrees.

Conclusions

The continuous development of the existing types of scientific communication requires the establishment of a strong collaborative network for young scientists and the development of new ways of communication. In addition, to build a sustainable national laboratory information system, competency-based education that integrates generic and profes-sion-specific competencies into the curricula of MLS could be the initial and most significant step toward bridging the gap in scientific communication and involvement in scientific activities among MLSs.

SIGNIFCIANT IMPORTANCE OF LABORATORY MEDICINE IN THE HEALTH OF PALESTINIAN REFUGEES IN WEST BANK AND GAZA

By: Osama Najjar- Palestine

The Palestinian population is approximately 5.0 million living in two geographically separate re¬gions; the West Bank and Gaza Strip, ruled by the Palestinian Authority since 1994. The Palestinian Ministry of Health (MOH) is the pri¬mary provider of healthcare services for the Palestinian population. Other healthcare pro¬viders include non-governmental organizations (NGOs), United Nations Relief and Works Agency (UNRWA) for Palestine Refugees and Military Medical Services.

At present, 124 of the 139 UNRWA health facilities provide comprehensive laboratory services. The remaining 15 facilities provide basic laboratory support (blood glucose, blood hemoglobin and urine tests by dipstick) through competent nursing staff using basic laboratory equipment. A total of 6,810,889 laboratory tests were performed Agency-wide in 2022. The cost of laboratory services continued to be far below the host-country rates for equivalent services.

Most of the healthcare services provided for refugees need laboratory tests. Those, laboratory tests affect the quality of the healthcare services for refugees to a better health. For example continuous Hemoglobin measurements for children decrease the anemia among Palestinian refugees to a less than normal level among such population in other countries.

Laboratory services for refugees play a major role in better health outcomes and affect positively the quality of life among Palestinian refugees.

A LAB FOR A CAMP? TOP TIPS TO DECIDE BETWEEN YES AND NO

Myrna Germanos Haddad
St Joseph University, Lebanon

The poor hygiene and living conditions in the refugee camps along with the high burden of certain infectious diseases and limited access to care are important reasons to consider Point of care tests (POCTs). These assays do not require skilled personnel and allow the immediate processing of samples. They deliver results promptly in real-time and allow the expeditious diagnosis and treatment. POCTs can be a much more practical alternative to large laboratory equipments which

require trained staff, space and costly reagents. In addition to being more cost effective, POCTs offer refugees confidential screening tests where they reside without discrimination. The limitations of POCTs may be calibration and validation issues. However, these may be circumvented by regular check-ups and maintenance of the instruments.

ID:15530 | IMPROVING DIAGNOSTICS OF CARDIOMETABOLIC DISEASE THROUGH INTEGRATED MULTI-OMICS APPROACHES

Miron Sopic

Medical Biochemistry, Faculty of Pharmacy, University of Belgrade

Despite the progress of healthcare systems in the past half of the century, the latest epidemiological data suggest that cardiovascular diseases (CVD) are still the leading cause of morbidity and mortality worldwide. In order to improve the CVD outcomes, we need new strategies that incorporate the complex interplay of different driving forces behind atherosclerosis in addition to the traditional risk factors. In order to utilize the full power of precision medicine, it is crucial to acquire and integrate data spanning from genotypes to phenotypes. Considering the multifaceted nature of atherosclerosis, and the large number of potential contributors to endothelial dysfunction, plaque development and erosion, it is now vital to get holistic view and bring a systems biology approach to the forefront of CVD research. System biology represents a powerful tool for understanding the mechanisms connecting identified causal genetic variation to CVD, where many sources of variability are integrated into statistical models to identify key drivers and pathways that have the largest contribution to the disease. Recent advances in the different omics technologies facilitated novel discoveries, revealing numerous links between genetic, epigenetic, transcriptomic, proteomic, metabolomic data and different pathophysiological processes. Currently, main challenges in clinical utilization of these data include: 1) insufficient number of evidence-based conclusions drawn from large multi-centric omics studies; 2) complexity of technologies used; 3) lack of pipelines for multiomics data integration; 4) lack of appropriate standards in the wet lab and dry lab protocols; 5) lack of specialists that could cope with omics approaches in everyday clinical practice. Thus, we need experts from different fields to tackle with these challenges through research and educational concepts, bringing the interdisciplinary views and vision to the table. Combination of basic research with clinical expertise and complex bioinformatics is essential to go beyond state-of-the-art, and more importantly foster new generation of scientists competent for utilization of "omics" in clinically relevant settings.

ISRAELI NATIONAL NEWBORN SCREENING PROGRAM

Suha Daas

Newborn screening, ministry of health, Israel

The Newborn Screening Program in Israel is a national effort, and all samples are transferred, handled and analyzed at a single laboratory. On average, results are reported on the 4th day of life. The program policy consist of testing any sample received free of charge for the family.

The newborn screening laboratory has state-of-the-art highly automated technologies including Specimen Gate LIMS with unique specifications and algorithms, Dried Blood Spot punchers, tandem mass spectrometry (MSMS), AutoDelfia and PCR instruments.

Annually we test 200,000 newborns among them about 5,000 non-citizens. The non-citizens include newborns of African refugees or work immigrants, as well as Palestinian sick babies from Gaza and the West Bank.

The newborn screening panel comprises 50 disorders and on average we identify at least one sick baby per day! The Israeli newborn population consist of 75% Jews and about 23% Arab Muslim and 2% of others (Christians, Druze). However, more than 50% of sick babies identified by our program are from the Arab newborn population due to closely consanguineous marriages.

Normal newborn screening results are available on the Ministry of Health website for parents and medical stuff. Newborns with positive results are referred to medical specialist near their home or hospital of birth.

HOW TO COLLABORATE TO IMPROVE THE SCREENING POWER IN LOW OR MIDDLE INCOMES COUNTRIES: TF-NBS AND THE ROLE OF IFCC

Khosrow Adeli PhD, FCACB, DABCC, FAACC

President, IFCC

Newborn screening (NBS) reduces infant morbidity and mortality by initiating early detection, treatment, and management of newborns with congenital disorders. Unfortunately, only about one third of infants around the world receive screening at birth, leading to significantly increased risk of disease during childhood and/or adulthood. The IFCC has made NBS a major focus of its strategic plans and has created a new Taskforce on Global Newborn Screening. The taskforce has completed the selection of countries for IFCC's NBS Pilot Project via a call for participation to IFCC member societies using pre-defined selection criteria. A pilot program is expected to begin in 2023 in select countries in Asia, Africa and South America.

The key mandates of the IFCC Global Newborn Screening Program are to: a) Identify potential participating centres (healthcare facilities and clinical laboratories) and stakeholders (government ministries and/or agencies) within each Partner Country. b) Engage with these centres in order to conduct situation assessment (information in regard to NBS-related diseases as well as resources/barriers in the Partner Country). c) Identify ways in which the IFCC can provide lacking resources and support. d) Develop a detailed protocol for an initial Pilot Project specific to each Partner Country. The proposal will be presented to the Partner Country via the respective IFCC member society. The Pilot Project implementation must be conditional on the Partner Country's commitment to its continuation following IFCC's initial financial support. e) Implement the Pilot Project in participating centres in each Partner Country in collaboration with the IFCC member society and other stakeholders. f) Monitor and evaluate the progress of the overall IFCC NBS Initiative, as well as the progress of specific Pilot Projects, through data collection and on-site/virtual visits by dispatching scientific teams to each Partner Country. g) Plan for the expansion of the IFCC's NBS Initiative, as well as the expansion of each NBS Pilot Project.

In my presentation, I will provide an overview of the global NBS program and how IFCC is collaborating with International Society of Newborn Screening, Centre of Disease Control and AACC to develop a wider partnership to enhance the feasibility of developing and supporting NBS programs in developing countries around the world.

INFECTIOUS DISEASE VACCINE PLATFORMS: PAVING THE WAY FOR NEW VACCINE AND THERAPEUTIC NANOMEDICINE PRODUCTS

Sophia G. Antimisiaris[1,2]

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The application of nanotechnology in Pharmacy and especially in the development of sophisticated formulations for drug administration and/or targeted delivery, has led to the development of therapeutic systems or nanotherapeutics with exceptional advantages. The most recent and well-known development in the area is the successful development of mRNA vaccines against the virus that causes SARS-COV-2 (severe acute respiratory syndrome coronavirus 2).

Two important milestones that facilitated the development of liposomal mRNA vaccines are:

(i) The synthesis of new ionizable lipids that allow rapid lysis of liposomes after they enter the cytoplasm and before they are catalyzed by lysosomes (early lysosomal escape), and

(ii) The recent development of microfluidic mixing methodologies for industrial production of liposomes.

mRNA-liposomal vaccines will be presented from a formulation point of view, and the contribution of the milestones mentioned above analyzed.

Furthermore it will be explained how such technologies may pave the way for future therapeutic products, especially for overcoming recent bottleneck's in therapeutics, with some examples from the results of our studies.

EU Syllabus for training in Laboratory medicine

Assist.Prof. Evgenija Homšak, PhD, EuSpLM,
University Clinical Centre Maribor, Slovenia
EFLM Professional Committee, Chair

The European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) is the essential professional organization in the field of Clinical Chemistry and Laboratory Medicine (CCLM) that join and links together Professional National Societies and their members throughout Europe. One of the most important efforts through the Profession Committee (C-P) scope and tasks are to achieve automatic regulation and recognition of professional qualifications under European Union (EU) legislation based on the principles of the free movement of professionals within Europe. According to the »EU Directive 2013/55/EU on recognition of professional Qualifications«, this effort could be achieved with the harmonization of our profession on the EFLM level through Common Training Framework (CTF), based on a Common Syllabus and confirmed exit qualifications. This process started almost 20 years ago with the European Communities Confederation of Clinical Chemistry and Laboratory Medicine (EC4), which laid the base for rules and minimal criteria for harmonizing our diverse education system through EU Countries. Since 2016 EC4 has been transferred to the EFLM (C-P). Through these years, several (5) versions of the Syllabus for postgraduate training in CCLM were prepared, which present the education/training program with important areas of knowledge and essential competencies for our profession. It represents the cornerstone for establishing Equivalents of standards (EoS) for European Specialists in Laboratory Medicine (EuSpLM). According to determined criteria, EoS has been delivered to the countries whose education program/system for specialization postgraduate training fulfils and includes all essential parts of these established rules: education/training duration, program (polyvalent), final exam/exit qualification. To support the harmonization of postgraduate education of EuSpLM, EFLM has also developed important project Syllabus Courses. In 2021 under the EFLM Task Group for Syllabus Course, in cooperation with more than 200 experts from all over the world, it has been launched more than 40 modules and over 300 prerecorded webinars/lectures that cover all topics in different fields of laboratory medicine and four main sections of the EFLM Syllabus: the generic knowledge, skills and competencies, the specialist knowledge within each discipline, the skills required to carry out research, development and audit, and the leadership skills and competencies.

References:

The European Federation of Clinical Chemistry and Laboratory Medicine syllabus for postgraduate education and training for Specialists in Laboratory Medicine: version 5 – 2018. Jassam N, Lake J, Dabrowska M, Queralto J, Rizos D, Lichtinghagen R, et al. Clin Chem Lab Med doi.org/10.1515/cclm-2018-0344

POST DEGREE TRAINING IN LABORATORY MEDICINE IN TUNISIA

Dr Khalil BEN ABDALLAH
Private Biologist pharmacist
Treasurer of The Tunisian Society of Clinical Biology (STBC)
Treasurer of The Tunisian Union of Private Laboratory
Member of the JNBC AND BIOMED-J organizing committee

Medical biology in Tunisia is a profession practiced since 1985 with the release of the first promotion of pharmacist biologist, it is a well regulated profession, it is governed by a law (2002-54) of June 11, 2002.

Authorized to practice medical biology in Tunisia are people with the diploma of pharmacist or doctor after having passed an internship exam and then 4 years of internships between different specialties : microbiology, biochemistry, hematology, parasitology and immunology which are mandatory and 3 others to choose from with two semesters in the chosen specialty and one semester between toxicology, PMA, transfusion.

Pharmacists having to pass 4 exams microbiology, biochemistry, hematology and parasitology unlike doctors who pass an exam at the end of specialty.

Biologists in Tunisia can follow a hospital-university career, or an hospital career or a private career with the opening a private medical biology laboratory.

Continuing education in Tunisia is not obligatory, it is mainly provided by the STBC but also by other learned societies such as the STPI, the STM… and also the faculties of pharmacy and medicine.

STBC insure the continuing education with minimum 6 training days per year and the organization each year of the National Days of Clinical Biology JNBC in May and the Tunisian Days of Biology Practitioner JTBP in October.

Law 2002-54 commits clinical biology to a process of harmonization and quality, it has strengths and limits and twenty years after its publication, it's time to revise it.

As of February, 2022, we have in Tunisia 560 private laboratories and 388 public laboratories.

"Post Degree Training in Laboratory Medicine – The Young Scientists Point of View"

E. Konsta
Chemist, MSc in Clinical Chemistry, PhD, Laboratory of Microbiology, Department of Water Quality Control, EYDAP, Greece

In Greece, there are approximately 500 laboratories in public hospitals and 2500 private laboratories (doing microbiology, hematology, biochemistry, immunology and molecular techniques). Medical doctors (biopathology), scientists (chemistry, biology, biochemistry, molecular biology) and pharmacists are being able to practice in the field of clinical chemistry and laboratory medicine. In addition, in some laboratories, there are persons with academic (usually technological) education without post-graduate specialization.

In Greece, there is no officially organized training of the specialists in clinical chemistry and laboratory medicine. In 1973, the Greek State passed a law that was introducing the Clinical Chemistry specialty for scientists (chemists, biologists, biochemists, pharmacists; law 131/1973). This law is still active but was never implemented due to the strong opposition by the medical Biopathologists. The Greek Society of Clinical Chemistry-Clinical Biochemistry (GSCC-CB) via the NCCRC ({Greek} National Clinical Chemistry Registration Committee) decided to organize a voluntary specialists training (duration 5 years) for scientists and pharmacists. This training includes both theory and practice.

The theoretical education is based on the EC4 Syllabus. A voluntary examination is organized by the GSCC-CB and the

NCCRC on the content of the educational program. Success to the examination leads to a certificate of (theoretical) competence. The GSCC-CB encourages all the young scientists who enter the field of clinical chemistry and laboratory medicine to follow this educational program and also to follow the on-the-job training according to the log-book provided by the GSCC-CB. The practical training is being realized by means of a "professional training dossier (PTD)" (Logbook). The PTD describes all the laboratory procedures that the trainee has to go through.

Today, the Greek Register counts 235 members, whereas 65 of them have become members of the European Register.

EFLM Task Group Young Scientists

Aleksei Tikhonov, PhD, Gustave Roussy, Villejuif, France

EFLM Task Group Young Scientists (TG-YS) was established in 2021 and gathered young laboratory medicine professionals from all EFLM countries. The goal of the group is to raise awareness of young scientists in laboratory medicine, build a communication network of young specialists within the profession and with colleagues from related disciplines, and facilitate various professional and scientific exchange activities. Group members organize and conduct virtual and face-to-face social, scientific, and educational meetings to share experiences and foster collaboration. They actively promote EFLM activities through EFLM communication channels and on the national level. Active participation of the young laboratory medicine specialists in EFLM activities is essential for sustainable society evolution and effective professional development.

BCLF SESSION

CLINICAL MASS SPECTROMETRY - A MAJOR TECHNOLOGICAL ADVANCES DRIVER IN LABORATORY MEDICINE

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Introduction.

There is an extraordinary flood of new technologies in medicine nowadays - sophisticated diagnostics based on genome assays, mass spectrometry, magnetic resonance spectroscopy and cell sorting platforms are driving the technological transfer, enormously enhance the informstive value of laboratory medicine, and promote the entrance of individualized patient management in clinical practice. Aim. This work overviews the role of Clinical MS Mass spectrometry (MS) in and could be viewed as one of the major drivers that promote the development of precision medicine.

Methods.

While genetic testing allows the physician to check personal genetic program and choose appropriate medicine, the performance of MS assays provides the patient's actual phenotype, with all of the environmental, pharmacological and pathological variables. Therefore, MS is essential technology for ultimate personalization of patient management. LC-MS/MS (QQQ) is the today's most utilized analytical platform, but high-resolution MS systems are also employed to resolve challenging analytical demands.

Results.

The great technological advance of MS resulted in the introduction of methods with unprecedented identification power, extreme sensitivity, specificity and extended linearity range, which are based on the current reference analytical principles. Further, the ability to perform panel profiling with simultaneous measurement of bioactive compounds, their precursors and metabolites in a single sample, enormously amplifies the informative value of results, with ultimate improvement

of patient care. Typical examples include new born screening, TDM, toxicology, endocrinology, microbiology, clinical omics assays and others. Experience of over 18 years with clinical mass spectrometry in the field of immunosuppressive drug monitoring, analysis of individual steroids, vitamin D status, steroidomic diagnostics, dihydropyrimidine dehydrogenase phenotyping and pharmacokinetic bioanalysis for industry will be presented and discussed.

Conclusion.

It should be specially emphasized that clinical MS integrates chemical and anatomical pathology: MS imaging and I-knife-MS guidance in surgery, although still in research phase, open new horizons for personalized treatment and individualized patient care.

PHARMACOGENETIC ASPECTS OF ACENOCOUMAROL THERAPY IN PATIENTS WITH CARDIAC SURGERY: THE EFFECTS OF CYP2C9 AND VKORC1 GENE POLYMORPHISMS

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Introduction

Acenocoumarol therapy has been widely used for extracorporeal cardiac surgery patients in Bulgaria in order to prevent thromboembolic events. The variety of responses among patients to this drug are driven by genetic background.

Aim: To investigate, during the initial phase of acenocoumarol therapy, the effect of CYP2C9 variant alleles and VKORC1 haplotypes, single and in combination in patients after extracorporeal cardiac surgery in order to achieve effective and safe treatment.

Methods

Genomic DNA samples from 200 Bulgarian patients subjected to cardiac surgery with extracorporeal circulation were analyzed for VKORC1 1639G>A and CYP2C9*2&*3 polymorphisms by real-time polymerase chain reaction (PCR). Analyses included genotypes with a frequency ≥5%. Patients were divided into "normal respondents" with an INR in therapeutic range (2-3.5) and "hyperreactive respondents" with an INR> 3.5. Chi-square test and regression analyses were applied to compare the between group differences.

Results

There were a total of 133 (66.5%) men and 67 (33.5%) women. The median age was 63.9±10.8 years. Distribution for VKORC1 and CYP2C9 showed an A/G*1/*1 prevalence (27%), followed by G/G*1/*1 (24.5%), A/G*1/*2 (10%), G/G*1/*2 (9.5%), A/A*1/*1 (9.5%), G/G*1/*3 (7%) and A/A*1/*2 (5%). Polymorphic alleles of CYP2C9 or AG/AA haplotype had significantly higher INR and more "hyperreactive respondents" then those with wild CYP2C9 allele or GG haplotype (p = 0.015). The comparison between G/G *1/*1 (n = 49) and the overall cohort of "hyperreactive respondents" (n = 136) showed that on the third day of acenocoumarol treatment the G/G *1/*1 group had significantly lower INR (2.05 vs.2.42, p = 0.05) and almost three times higher relative risk of subtherapeutic anticoagulant effect (61% vs. 16.2%, p <0.001). Combined CYP2C9 mutant alleles and/or AG/AA haplotypes are associated with a higher incidence of "hyperreactive respondents" and bleeding than those with wild CYP2C9 alleles and GG haplotype.

Conclusion

Pretesting patients for CYP2C9 and VKORC1 genetic polymorphisms could aid in risk stratification and individualizing acenocoumarol therapy.

SPECIALIZATION IN MEDICAL BIOCHEMISTRY IN THE REPUBLIC OF NORTH MACEDONIA - FUTURE PERSPECTIVES

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Laboratory profession across Europe still differs in academic background of the specialists in clinical chemistry. In our country, specialists in medical biochemistry/clinical chemistry hold basic education in general medicine (MDs) or pharmacy (Ms PHs). The length of the graduate studies in general medicine is six years with 360 ECTS, and the length for general pharmacy is five years with 300 ECTS. Few common subjects are studied during graduate studies for both profiles that differ in the ECTS, such as: Chemistry (7 ECTS for MDs and 39,5 ECTS for Ms PHs); Anatomy and physiology (28 ECTS for MDs and 10 ECTS Ms PHs), Biochemistry (12,5 ECTS for MDs and 6 ECTS for Ms PHs), Biophysics (2 ECTS for MDs and 7 ECTS for Ms PHs), Molecular and Cell Biology and Genetics (10 ECTS for MDs and 6 ECTS for Ms PHs); Microbiology (10 ECTS for MDs and 7 ECTS for Ms PHs); Pathophysiology (11,5 ECTS for MDs and 6 for Ms PH); Pharmacology (7 ECTS for MDs and 6 for Ms PHs); Clinical Biochemistry (1,5 ECTS for MDs and 7 ECTS for Ms PHs). The postgraduate training in medical biochemistry, as a monovalent specialization, for both profiles, ranges 4 years. Although the graduate syllabus in pharmacology doesn't have any clinical subject, the specialization program for both graduate profiles is the same for both profiles, as well as the title, and was succeeded and revised several times after the succession of the former country. Presently, the total length of basic education for both profiles is 10.5 for MDs and 9,5 years respectively, and both profiles are registered in the European Register of Specialists in Laboratory Medicine. The question arising for the perspective vocational training for medical doctors and pharmacists is: do we need different specialization programs for these profiles, bearing in mind the previous education and the need of our health care system? The opinion of the members of the Department of Biochemistry and Clinical Chemistry (responsible for proposing and creating specialization syllabuses) is that, in our country, we need two different monovalent specializations in clinical chemistry/medical biochemistry, one for medical doctors and one for masters of pharmacy, which will result in two different profiles and responsibilities in the health care system in our republic.

ACCREDITATION FROM A POSITIVE LEADERSHIP PERSPECTIVE

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When lab staff is alone—no management or official representatives in sight—one of the things they complain about the most is accreditation. At worst, we consider it a huge waste of time and effort. It's not that we don't see the benefits of accreditation. We do. But we find that these returns do not satisfy us and are definitely not worth the costs associated with the process.

So if accreditation is unlikely to go away, is it possible to make lemonade out of this lemon and create a more positive outcome from what appears to be long-lasting,exhausting and sometimes boring process?

House cleaning

If we take the perspective, "If we have to do it anyway, how can we make it better? we could start by saying that accreditation gives us a rather rare opportunity to do some cleaning in the labs. By forcing us to regularly review our policies and procedures and the staffing of various tasks in our laboratories, we have an externally imposed reason to engage in a process that can lead to internally beneficial outcomes.

Become proactive

Just as we have learned to be more proactive when we conduct laboratory evaluations—not only evaluating past per-

formance, but also building on the past to set goals for the future—accreditation can help us become more proactive. Accreditation offers us a regular opportunity to ask ourselves where we want to go based on where we have already been. It gives us a chance to plan systematically by looking at our best practices and comparing our current results with those of the past.

Meeting the enemy

We are the ones who vote to approve, adopt or accept the standards. We may have met the enemy - and that is us. If some of us who feel constrained by outdated standards and processes used by accrediting agencies were a little more open in our meetings about why accreditation often hurts us more than it helps, maybe we could start to initiate some change. If we don't succeed, at least we wouldn't be worse off than we are now.

ESSENTIAL TRACE ELEMENTS: COMPLEX CROSS-TALKS IN INFLAMATION

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If one element is absolutely necessary for the physiological needs in human body, if it is not replaceable by another element and it exerts its effect directly but no by antagonism of another element at toxic level, it is referred as essential (vital). Certain trace elements are biologically classified as essential (Fe, I, Cu, Zn, Co, Cr, Mo, Se, Mn) and others might be considered as borderline candidates (As, B, Br, F, Li, Ni, Si, V -"conditionally-essential"). The main goal of this review is to present current data on the role of trace elements in various pathologies, associated with deficiency and accumulation (congenital and acquired). Special focus is placed on molecular knowledge about the complex role of the essential trace elements copper (Cu) and zinc (Zn) in inflammation. As robustly observed in other infections, systemic and chronic inflammatory responses are associated with significant biochemical and physiological alterations, thus affecting the concentration of plasma proteins, macro- and micro- nutrients. During infections or inflammation, abnormality in serum levels of Cu and Zn could be developed: one of the most common case is an elevation in serum Cu and a depression in serum Zn. These disorders can be expressed as Cu:Zn ratio, a clinically more useful index than only the concentration of either of these trace metals. The positive and negative reactants that are "players" in the acute and chronic inflammation, together with very important for Cu and Zn metabolism molecules as metallothionenins, are responsible, due to their specific "behaviour", for the alterations developed in inflammation. The impaired balance of every element should be interpreted in the view of complicated interactions between different elements taking into account possible epigenetic factors. The personalised approach in contemporary medicine, concerning not only rare diseases, but also numerous socially significant diseases and even common banal inflammations, is associated with clinical benefits of multi-elemental profile in one individual. The laboratory monitoring of the plasma proteins in combination with blood micronutrient levels could serve as a useful tool for a more comprehensive evaluation of inflammation. Better clinical cares could be achieved in such pathologies by supplementation in the case of deficiency occurred.

ORAL PRESENTATIONS

OP01

ASSOCIATION OF SERUM CARBOXYLATED AND UNDERCARBOXYLATED OSTEOCALCIN WITH INDICES OF OBESITY, GLYCEMIA, AND CORONARY ATHEROSCLEROSIS DISEASE IN SYRIAN MEN

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Background and Aims

The higher incidence of coronary events in obese subjects is only partly explained by conventional associated risk factors. Atherosclerosis is associated with circulating osteocalcin; however, this association is often conflicting and unclear. This study aimed to determine serum osteocalcin levels in overweight/obese men, and to investigate association between osteocalcin, indices of obesity, and lipid biomarkers in the development of coronary atherosclerosis disease (CAD).

Methods and Results

A cross-sectional study was conducted on 84 male: 20 early coronary atherosclerosis (ECA) patients, aged (55.1± 10.8 years), defined as patients with mild CAD (stenosis in any major epicardial arteries), and 38 late coronary atherosclerosis (LCA) patients, aged (59.0± 9.5 years), defined as patients with severe, multivessel CAD (>50% stenosis in at least one or more major epicardial arteries). Healthy control (HC) group included 26 healthy male subjects, aged (38.6±7.7 years). Participants to the study (HC+ECA+LCA) were stratified according WHO obesity criteria into two groups: Normal weight group, BMI 20-24.9 kg/m2 n=26, aged 51.05± 13.84 years, overweight/obese group, BMI ≥ 25 kg/m2, n=58, aged 53.95± 9.29 years. Carboxylated (cOC) and undercarboxylated osteocalcin (ucOC) were quantified using ELISA technique. According to the lipid profile, HDL-C was higher in non-obese than overweight/obese groups (33.77±10.96 vs 27.71±8.28 mg/dL, P=0.019). FBG was higher in obese groups than non-obese (179.43±104.98 vs 108.94±62.55 mg/dL, P=0.006). CAD was prevalence, as expected, in overweight/obese group than non-obese group (89.7% vs 23.1%, p=<0.01). cOC levels was higher in normal group than overweight/obese group (8.77±4.01 vs ng/dL6.66±3.58, P=0.026). ucOC levels was higher in normal group than overweight/obese group (4.28±2.73 vs 2.10±1.59 ng/dL, P=0.001). cOC was similar between high level HDL-C group and low level HDL-C group (8.51±4.08 vs 7.63±3.82 ng/dL, P=0.315). ucOC was higher in high level HDL-C group than low level HDL-C group (4.28±2.73 vs 2.10±1.59 ng/dL, P<0.01)

Conclusion

Our study showed that bone is not merely an endocrine target, but it may also play a role in controlling metabolic phenotypes such as obesity and HDL-C level. This study also provide further support for the hypothesis that ucOC is a protective factor for both CAD and obesity.

OP02

miRNA PROFILING AND WEIGHT LOSS: DIRECTED ACYCLIC GRAPHS PATHWAYS ON METABOLIC HORMONAL MARKERS IN BARIATRIC SURGERY

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Background

Epigenetic changes after bariatric surgery are of increasing interest to researchers. We evaluated the association of two circulating miRNAs (miR-222 and miR-146a) before and after bariatric surgery and the effects of weight loss on metabolic biomarkers including leptin, ghrelin, peptide YY, and glucagon peptide-1 (GLP-1).

Methods: We prospectively evaluated patients at pre-and six months post-bariatric surgery for % excess weight loss (EWL), microRNAs (miRNAs) (222 and 146) were determined by RT-qPCR, and metabolic biomarkers including; hormones (leptin, insulin, ghrelin, peptide YY, and GLP-1) were evaluated by ELISA, while lipid profile and fasting glucose by 7600 Hitachi automatic analyzer and HOMA-IR was calculated.

Results

% EWL correlated positively with miR146a (R:0.437, p:0.001) and negatively with miR222 (R: -0.509, p:0.001).

According to the directed acyclic graph (DAG) metabolic pathway model, decreased leptin levels correlated significantly with %EWL (R-.379, p.004) but not with % change in miRNA 222 (R.172, p.204). Decreased GLP-1 levels correlated significantly with % change in miRNA 222 (R.402, p.002) but not with %EWL (R-.214, p.114). Decreased ghrelin levels correlated significantly with %EWL (R-.274, p.041) and % change in miRNA 222 (R.471, p.0001). Decreased total (T) and low-density lipoprotein (LDL) cholesterol levels correlated significantly with % change in miRNA 222 and not with %EWL. Homeostatic model assessment of insulin resistance (HOMA-IR) levels correlated significantly with %EWL and not with % change in miRNA 222. % change in miR146 had no significant effect on the metabolic biomarkers.

Conclusions

Bariatric surgery resulted in changes in miRNA expression. The DAG pathway revealed that postoperative increase in %EWL was accompanied by % change in miR222, and weight loss had a direct effect on the metabolic biomarker.

OP03

PROTEOMIC ANALYSIS REVEALS POSSIBLE BIOMARKERS OF Q-FEVER

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Introduction

Coxiella burnetti, the causative agent of the worldwide zoonosis Q fever in humans,is an obligatory intracellular bacterial pathogen. Due to its surprisingly high infectious nature; Coxiella has been characterized as a world biological weapon agent, according to the Centers of Disease Control and Preventions; USA1. The aim of this study is to investigate how C.

burnetii, contributes to changes of the host cell proteome by using proteome-wide label-free quantification techniques. For this reason we analysed the whole proteome of infected and non infectedhuman MRC-5 fibroblasts cells across three time points of infection: 2 days (lag phase), 10 days (stationary phase) and 19 days (late stationary phase).

Materials and Methods

C. burnetii Nine Mile RSA 493 (phase II, clone 4) was propagated on MRC-5 cells (CCL-171™) in Dulbecco's Modified Eagle's Medium-high glucose medium with 10% fetal bovine serum.Peptides obtained from whole cells using the FASP protocol, were separated on a C18-Pepmap analytical column and analysed by a Fusion Lumos mass spectrometer. The bioinformatics and statistical analysis was carried out using MaxQuant, Perseus and the statistical computing software R.

Results

In total 6629 proteins have been identified in the infected and non-infected MRC-5 cells. A subset of more than 1100 proteins were matched to Coxiella's proteome, were identified only in the infected MRC-5 cells. The rest of about 5520 belong to host proteome. Applying stringent criteria for quantification, the number of reliably quantified proteins reduced to 1034 and 4331 for Coxiella's or the fibroblast's proteome, respectively. Furthermore, the results of this study suggest potential effectors proteins and proteins which could be used as biomarkers based on proteins that are known for their serodiagnostic potential[2].

Conclusions: This study revealed differential regulation of proteins associated with metabolism, protein synthesis, and transport and stress response in the host and Coxiella burnetii. Further, this study provides a list of 49 proteins with serodiagnostic potential based on their abundance. Lastly, MYDGF is the only protein that is up-regulated in 2 days. Based on a recent study3, MYDGF was proven to be a potential biomarker for the diagnosis of acute myocardial infarction.

Literature

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OP04

CONCURRENT MOLECULAR CHARACTERIZATION OF SAND FLIES AND LEISHMANIA PARASITES BY AMPLICON BASED NEXT GENERATION SEQUENCING

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Background

Phlebotomine sand flies are vectors of Leishmania parasites, which are the causative agents of leishmaniasis. Herein, we developed an amplicon-based next-generation sequencing (Amp-NGS) to characterize sand flies and Leishmania parasites simultaneously targeting partial fragments of 18S rDNA and ITS1 genes, respectively.

Methods

Our assay was optimized using reference sand fly (n = 8) and Leishmania spp. (n = 9) samples and validated using wild-caught sand flies from Palestine. The assay was highly specific, and all DNA references were successfully identified to the species level.

Results

Among the wild-caught sand flies (n = 187), Phlebotomus spp. represented 95% of the collected samples (177/187), including Ph. sergenti (147/187, 79%), Ph. papatasi (19/187, 10.2%), Ph. perfiliewi (3/187, 1.6%), Ph. Tobbi (2/187, 1.2%) and Ph. syriacus (6/187, 3.2%). Sergentomyia spp. represented only 5% (10/187) of the collected samples

and included S. dentata (n = 6), S. fallax (n = 2), S. schwetzi (n = 1) and S. ghesquiere (n = 1). The study observed strong positive correlation between sand fly identification results of the Amp-NGS and morphological identification method (r = 0.84, df = 185, P < 0.001). Some discrepancies between the two methods in the identification of closely related species (i.e. Ph. perfiliewi, Ph. tobbi and Ph. syriacus) were observed. Leishmania DNA was detected and identified as L. tropica in 14 samples (14/187, 7.5%).

Conclusions

Our assay was sensitive to detect (limit of detection was 0.0016 ng/reaction) and identify Leishmania DNA in sand flies, thus representing a new tool for studying sand flies and their associated Leishmania parasites in endemic areas.

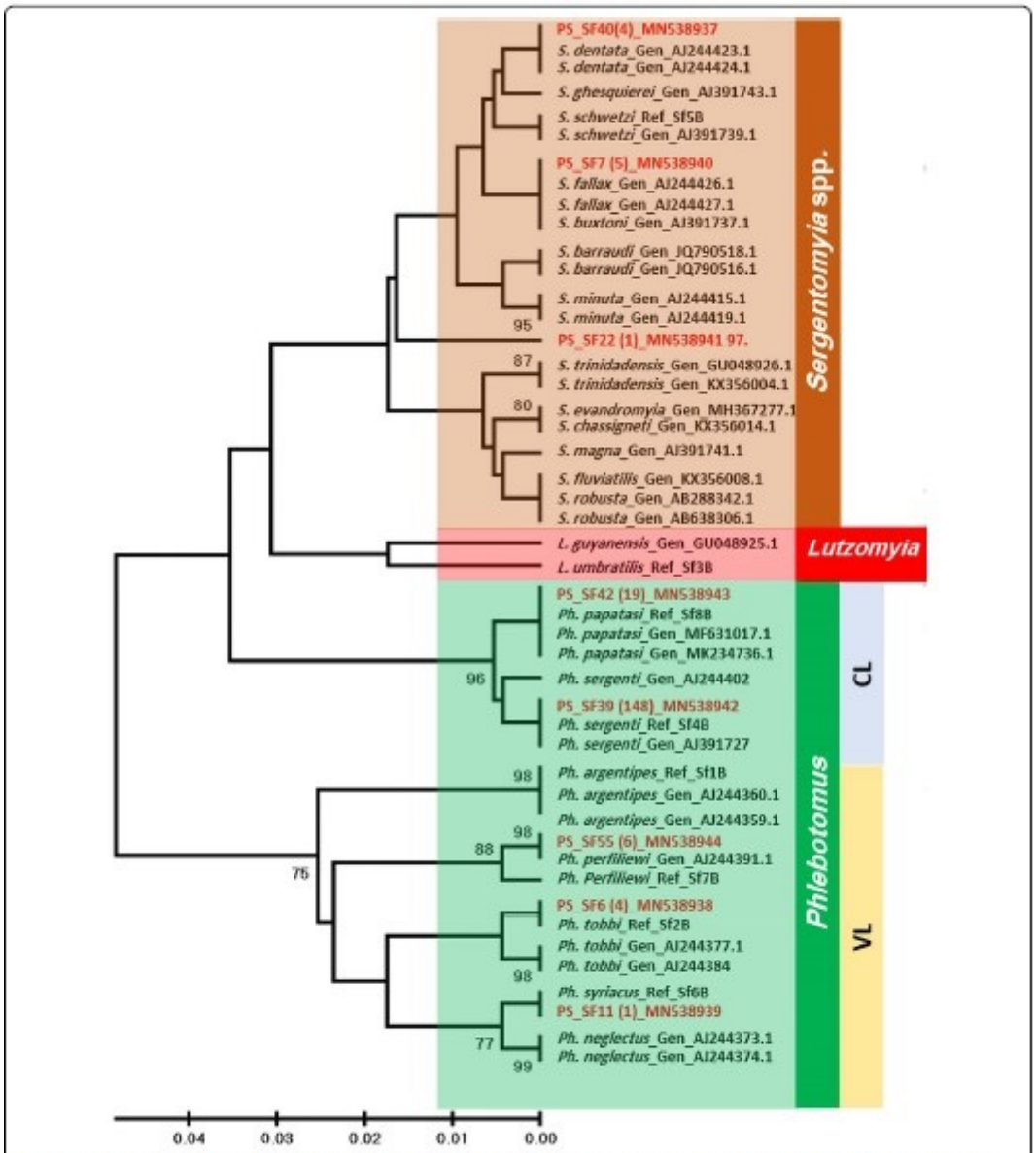

Fig. 5 Neighbor-joining (NJ) tree showing the relationships of the study and reference sandflies (*n* = 227) shown in bold red based on 150 bp of the 18srDNA gene sequences. MEGA X program was used for constructing the phylogenetic trees. DNA sequences were aligned using Clustal-W program. Bootstrap values are based on 1000 replicates [46]. Ph., *Phlebotomus*; S. *Sergentomyia*; L., *Lutzomyia*; Gen_AJ244374.1, GenBank accession number; Ref_sf6B, reference strain_sand fly number 6B; CL, cutaneous leishmaniasis; VL, visceral leishmaniasis; PS, Palestine with Palestinian sand fly code; numbers in parentheses indicate number of samples with identical genetic characters

OP05

SYSTEMATIC COMPARISON OF DIFFERENT DERIVATISATION REAGENTS FOR DETERMINATION OF MULTIPLE VITAMIN D3 METABOLITES USING LC-MS/MS - LONG-TERM STABILITY DETERMINATION OF CHEMICAL DERIVATIVES

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Introduction

Liquid chromatography/tandem mass spectrometry (LC-MS/MS) is firmly established today as the gold standard technique for analysis of vitamin D. The major vitamin D metabolites are 25-hydroxyvitamin D3, 1,25-dihydroxyvitamin D3, and 24,25-dihydroxyvitamin D3. Their quantification can be very challenging, in particular for the low abundant species. Chemical derivatisation can enhance the detection sensitivity by increasing the ionization efficiency, shifting the mass to higher m/z values with less isobaric noise and providing specific fragmentation patterns for MS/MS.

Aim

In this study, we compared the detection sensitivity and long-term stability of seven chemical derivatives of vitamin D3, 3β-25(OH)D3, 3α-25(OH)D3, 1,25(OH)2D3 and 24,25(OH)2D3 in human serum.

Methods

Four dienophile reagents (4-phenyl-1,2,4-triazoline-3,5-dione (PTAD), 4-[2-(6,7-dimethoxy-4-methyl-3-oxo-3,4-dihydroquinoxalyl)ethyl]-1,2,4-triazoline-3,5-dione (DMEQ-TAD), Amplifex, 2-nitrosopyridine (PyrNO)), two reagents for hydroxyl functions (isonicotinoyl chloride (INC), 2-fluoro-1-methylpyridinium p-toluenesulfonate (FMP-TS)) and a combination of both, including PTAD derivatisation and acetylation (PTAD-Ac) of the hydroxyl groups, were tested. The measurements were conducted using a Sciex QTRAP 6500+. Stability experiments were conducted after 1 and 3 months of storage at -20 oC.

Results

Derivatisations using Cookson-type reagents are well established. More than a single peak for each vitamin D analyte are present at the chromatogram as a result of diastereomer (R and S) formation during reaction which could have a negative impact on the sensitivity of the method. Derivatisation reagents targeting hydroxyl group can be an alternative choice. Depending on the stereochemical hindrance of each hydroxyl group and the amount of derivatisation reagent used, 1-3 different precursor ions depending on the structure of each analyte can occur. Better sensitivity was observed for the dienophile reagents, because of the more selective derivatisation and the presence of permanently charged moieties (Amplifex, FMP-TS). Regarding long-term stability, the most stable chemical derivatives were formed by Amplifex reagent even after 3 months of storage. DMEQ-TAD, FMP-TS, INC and PTAD-Ac derivatives showed good stability after 1 month for some metabolites.

Conclusions

No systematic comparison of multiple derivatising agents for MS analysis of vitamin D metabolites or long-term stability of the chemical derivatives has ever been presented. This information is important, as limited literature data show contradictory information.

OP06

WHAT IF WE ASK ABOUT AMMONIUM LEVELS? A CASE REPORT

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Introduction

Ornithine transcarbamylase (OCT) deficiency is the most common of the urea cycle diseases (ECU). It is an estimated incidence of 1 in 14 000 live births and men are usually more severely affected than women. ECU, which belong to inborn errors of metabolism, triggers neonatal hyperammonemia with a high rate of neurological sequelae and mortality.

Therefore, it is important to recognize and treat quickly to prevent cerebral oedema becoming established and irreversible.

We present the case of a neonatal patient hemizygous in the OCT gene. That case is considered of interest due to symptomatic hyperammonaemia in newborn is a medical emergency despite of its low frequency.

Aim

A 48-hour-old male who was referred from Neonatal Unit with respiratory complaints and neurological symptoms: hypoactivity, hypertonia in the limbs, and tremors.

Regarding personal medical history, healthy parents without family consanguinity.

Methods Not applicable.

Results

During admission, the patient suffered an episode of seizures and rapid neurological deterioration. Metabolic evaluation confirmed highly significant hyperammonemia (ammonium: 990 micromoles/L) without metabolic acidosis, with glycemia in the range and a negative ketone bodies levels in urine. The results led to the diagnostic suspicion of alteration in ECU.

Treatment was then started aimed at reducing exogenous and endogenous ammonia production, ammonia elimination by means of drugs that use alternative pathways to UC (phenylbutyrate and benzoate), cofactors that optimize UC and other metabolic pathways (arginine, N-carbamylglutamate, vitamin B12) and drugs that favor the elimination of toxins (carnitine). In addition to this, the patient underwent intermittent hemodialysis.

The patient presented hemizygosis for the variant in OTC gene (Xp21.1), NM_000531.5: c.443T>C, p.(Leu148Ser). This variant, which is described as pathological, is associated with an X-linked pattern of inheritance, with OCT deficiency.

Currently the patient has a favorable evolution and continues in multidisciplinary clinical follow-up.

Conclusions

Early diagnosis of OCT deficiency in the neonatal stage should be recognized and immediate treated to improve the vital and long-term prognosis of these children.

The genetic study of segregation of the variant in relatives of the maternal line and genetic counseling is recommended.

OP07

ELEVATED LUTEINIZING HORMONE DESPITE NORMAL TESTOSTERONE LEVELS IN NEWLY DIAGNOSED PROSTATE CANCER PATIENTS

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Introduction

Prostate specific antigen (PSA) is largely used as a tumor marker for prostate cancer. However, PSA is not specific therefore, other biomarkers might aid physicians to better assess cancer prognosis and monitoring. The most obvious candidate biomarkers which regulate the prostate are sex steroid and pituitary hormones

Objective

The main of this study was to investigate the role of pituitary hormones luteinizing hormone (LH) and follicle stimulating hormone (FSH) in prostate cancer. We also studied the relationship between LH and steroid hormones including total testosterone, free testosterone, bioavailable testosterone (BAT), sex hormone binding globulin (SHBG), estradiol (E2) and progesterone. Finally, we looked in to the relationship between all the study parameters and Gleason scores.

Subjects and methods: serum was withdrawn from newly diagnosed 35 prostate cancer patients and 30 controls. Using Roche-immunoassay analyzers we measured serum levels of: (PSA), total testosterone, free testosterone, (SHBG), (E2), BAT, (FSH), (LH) and albumin. SPSS statistical software used to perform students' t test and Spearman rho for association studies. P value of < 0.05 was considered significant.

Results

The study showed that 71% of prostate cancer patients had raised serum LH while having normal testosterone levels. A statistically significant increase ($P \leq 0.01$) in the level of PSA (128.9 ± 275.01 $P<0.0136$), LH (14.31 ± 13.84 $P<0.0108$), and very statistically significant in the level of total testosterone (4.60 ± 1.69 $P<0.0015$) and BAT (1.93 ± 0.95 $P<0.0012$) was observed when compared to control group.Despite these differences, the results of total testosterone, free testosterone and BAT were in the normal range, while mean LH levels in the prostate group was elevated. In addition, there was non- significant difference in the levels of free testosterone, SHBG, FSH and E2 when compared to the control group. Except for free testosterone, none of the above parameters correlated with Gleason score.

Conclusion

For the first time in prostate cancer patients, we report that despite raised serum LH, serum testosterone levels were normal. These finding suggest a mechanism of subclinical hypogonadism or compensated hypogonadism might play an important role in prostate cancer development, further investigation is needed to draw firm conclusions.

OP08

QUALITY ASSESMENT OF TOTAL TESTING PROCESS AND PREANALYTICAL ERRORS IN A LAB FOR A CAMP: WHERE ARE WE IN COVID-19 PANDEMIC?

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Background

Factors of quality such as: personnel, education and training of employees, dependence on adequate equipment, in-

novation of services, quality , standardization of performed services through the application of quality management system(QMS) is necessary.

Inadequate preparation of patients and skills of medical phlebotomists are sources of errors in preanalytical phases. The aim of this retrospective study is monitoring, documenting and preventing errors in pre-analytical phase in a two labs for a camp in Covid-19 pandemic for better health care of patients.

Methods

The study has been done from 2019 to 2021 years during corona pandemic and involves monitoring, documenting and preventing errors with aspect to phlebotomy in two laboratories for a camp of health care of patients. Errors are classified according to IFCC recommendation as quality indicators: insufficient sample volume, inappropriately labeled sample and sample damage.

Results

The study has shown that the most common errors are insufficient sample volume and sample damage (0.97 %). Inappropriately labeled samples were significantly lower and completely eliminated during period of study (2019 was 0.24 %, 2021 was 0 %; p<0, 01). No significantly decrease in number of sample damaged (2019- 0.40 % - 2021- 0, 30 %) was shown and insufficient sample volume (2019- 0.33% - 2021-0.22%) were constantly persisting during the period of study.

Conclusion

Through permanently improvement of the QMS, implementation of certification and accreditation of laboratories according to the ISO15189, 2018- (QM / QA) standard for medical laboratories with a special requirement of the entire laboratory testing and implementation of LIS (Laboratory Information System), the most important standard for POCT-ISO22870: 2006 Point of care testing, clear, transparent and available procedures, errors from pre-analytical phase in a lab for a camp can be minimized. Special attention should be paid on errors that continue to existing in the study. A smaller number of errors in pre-analytical phase mean more accurate, precise and valid results, correct and fast diagnosis, satisfied patients and principle of cost benefit with guidelines: " no blood sample is better than a bad blood sample" and "more is better".

Key words

Accreditation,laboratory testing in a camp, GLP,preanalytical errors ,health care, patient

OP09

THE ASSOCIATION OF SEMINAL PLASMA BIOCHEMICAL CONSTITUENTS LEVEL AND DIFFERENT SEMINAL CONDITIONS

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Introduction

Seminal plasma is a fluid of semen, secreted by epididymis and accessory glands before and during ejaculation. It is a very complex fluid and contains many different biochemical constituents such as proteins, enzymes, lipids, macro-and microelements, etc. Male factor infertility is often characterized by abnormalities on semen analysis such as low or absent sperm counts and low motility. Biochemical constituent levels in male infertility are poorly examined.

Aim: The aim of this study was to assess seminal biochemical constituents level in different seminal conditions.

Materials And Methodes: Two hundred sixty men samples were collected by masturbation after four to five days of sexual

abstinence. The samples were divided into five group, normozoospermia (N=67), asthenozoospermia (N=18), oligozoospermia (N=31), oligoasthenozoospermia (N=40) and azospermia (N=104). After cytological analysis, samples of all patients were centrifuged and lactate dehydrogenase (LDH), aspartate aminotransferase (AST), alanine aminotransferasa (ALT), gama-glutamil transferase (γ-GT), total protein (TP), total calcium (Ca), magnesium (Mg) and inorganic phosphate (P) were determined in seminal plasma using commercial tests. All values as median and interquartilerange (25th, 75th percentile).

Results: TP values in patients with azoospermia 35 (42-52) were significantly lower ($p<0.05$) than in those with asthenozoospermia 47 (51-57) and oligoasthenozoospermia 44 (50-57). AST acitivities in patients with azoospermia 130 (171-239) was significantly lower ($p<0.05$) compared to normozoospermia 257 (320-458), and asthenozoospermia 261 (314-382), while patients with normozoospermia had significantly higher AST activites ($p<0.05$) than those with oligozoospermia 190 (223-253) and oligoasthenozoospermia 170 (209-314). ALT acitivity in patients with azoospermia was significantly lower ($p<0.05$) 12 (17.-24) compared to patients with normozoospermia 17 (22-31). Patients with normozoospermia had significantly higher ($p<0.05$) GGT activity 8683 (11877-14700) compared with patients with azoospermia 7080 (9055-11925) and oligozoospermia 6500 (8500-11355). LDH in patients with normozoospermia 3240 (4473-6169) was significantly higher than in infertile men with azoospermia 2059 (2818-3947), oligozoospermia 2294 (2863-3616) and oligoasthenozoospermia 2228 (3050-4069). There was no statistical difference in Ca, Mg, P ($p>0.05$) between five examined groups.

Conclusions

Infertile man with azoospermia had significantly lower AST, ALT, GGT, LDH activities compared with those with normoazospermia, until patients with oligoasthenozooospermia had significantly lower AST and LDH compared with patients normoazospermia.

OP10

CORRELATIONS BETWEEN D-DIMER, C REACTIVE PROTEIN AND HEMATOLOGIC DATA IN OUTPATIENTS WITH COVID-19

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Introduction

Coronavirus disease (COVID-19) is an infectious disease caused by the SARS-CoV-2 virus. Most people infected with the virus will experience mild to moderate respiratory illness and recover without requiring special treatment. The main routine tests requested for COVID-19 patients include complete blood count (CBC), assays investigating coagulation and fibrinolysis cascades (PT,aPTT, D-dimer), and inflammation-related parameters (ESR, CRP, ferritin, procalcitonin).

Aim

To evaluate correlations between D-dimer, C reactive protein (CRP) and hematologic data (CBC) in outpatients with COVID-19. We aim to assess whether the correlations found in hospitalised patients also apply to outpatients.

Methods

We performed a retrospective study including 100 outpatients, 51 women and 49 men, mean age 51 ± 15 years, from August 2020 to January 2022, diagnosed with COVID-19 by RT-PCR or rapid test positive.

Results

Most affected age group with COVID-19 was 51-60 years with 29%, whereas 73% of outpatients were < 60 years. Hematologic data in most patients were normal, only 23% of them had an increased Neutrophil to Lymphocyte Ratio (NLR) and 22 % had decreased the lymphocyte percentage. Regarding inflammatory parameters 42% had CRP above reference range and only 22% had an increased value of D-dimer. We have found correlations between White blood cell (WBC)

and NLR (r=0.58, p<0.001), WBC and Platelets (r=0.45, p<0.004). WBC and CRP (r=0.42, p<0.01). D-dimer and Erythrocyte Sedimentation Rate (ESR) (r=0.42, p<0.009), D-dimer and CRP (r=0.39, p<0.02). CRP and WBC (r=0.42, p<0.011), CRP and NLR (r=0.73, p<0.001), CRP and ESR (r=0.62, p<0.001).

Conclusions

We conclude that the correlations found between the variables studied from us are also present in outpatients and therefore they can serve as good markers to decide performing further tests for the diagnosis of severe cases and prognosis of COVID-19.

Keywords: D-dimer, CRP, NLR, COVID-19, outpatients.

ePOSTERS

Analytical technologies and applications

eP01

DIFFERENCES IN URACILEMIA BETWEEN HIGH PERFORMANCE LIQUID CHROMATOGRAPHY AND LIQUID CHROMATOGRAPHY-MASS SPECTROMETRY

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Introduction

Fluorouracil is contraindicated in individuals who are known to have complete dihydropyrimidine dehydrogenase (DPD) deficiency, or when complete deficiency is suspected because of early-onset or unusually severe fluorouracil toxicity (Fluorouracil-FresseniusKabi).

The patients should be tested for the lack of the DPD-enzyme before starting cancer treatment with fluorouracil or related medicines (EMA/229267/2020).

DPD-phenotype is usually determinated with the analysis of the level of uracilemia by LCMS technology, a reference method available to few laboratories due to high technological cost. There are several publications that explore this analysis using HPLC-UV technology, which is less expensive, and widely distributed in hospitals.

Our group has modified uracilemia method for HPLC-UV proposed by Dèporte (Dèporte-PMID:16545990) using BenFredj sample preparation (BenFredj-PMID:18619742) with good results. However, when we compare LCMS versus HPLC results, we observe sometimes HPLC results are 2-3 times higher than LCMS.

Aim: To analyze the effect of differences in pH in the determination of uracilemia by HPLC-UV versus LCMS.

Methods: Serum samples of patients candidates to receive fluorouracil therapy were centrifugated, aliquoted into 2 vials and frozen at -80°C until analysis.

Uracilemia was measured in all samples by HPLC-UV and LMCS method/technologies. pH of all reagents and clinical samples was measured with a pH-meter (pH50-VioLab XS-Instruments). For HPLC-UV analyses Chromsystems HPLC (EasylineDataSystems-GeminyxIII) with UV-detector at 268nm and a C18-column (Teknokroma) was used. All reagents were from MerkMillipore and standards and controls were prepared in-house by gravimetry. Uracilemia by LCMS analyses was carried out in an external laboratory.

Results

All reagents and mobile phase for HPLC method had pH according to the method described (Dèporte-PMID:16545990).

Standards, controls and samples maintained the expected pH after extraction according to the method before HPLC analysis.

However, clinical samples before extraction showed disparity in pH. Alkalinzation was observed after uncapping extraction tube or delaying extraction process. When pH was >7.6, LCMS uracilemia results had two to three times lower values that HPLC results.

Conclusion

These results suggest that the method used to LCMS technique is very sensitive to changes in pH and that alkalinization by uncapping potentially underestimate the uracilemia determined by LCMS, but not determined by HPLC-UV.

eP02

BIOTIN INTERFERENCE IN IMMUNOASSAYS: IS STILL A DANGER PRESENT?

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Introduction

Immunoassays are important qualitative and quantitative analytical techniques and, despite the improvements that have been made, there is still the possibility that various substances interfere by changing the measurable concentration of the analyte or the binding of antibodies, resulting in erroneous and consequently dangerous clinical decisions. The main interferences in immunoassays are: anti-streptavidin antibodies, anti-ruthenium antibodies, anti-alkaline phosphatase antibodies, hook effect, cross-reactions, macro analytes as p.ex macro-TSH, heterophile antibodies, HAAA, RF, gammopathies. Among the most important interferences is that of biotin. Excess use of biotin, mainly in uncontrolled nutritional supplements, creates a significant problem in laboratory results, as in most immunoassays the biotin-streptavidin complex is used. The most serious reported incident of interference, is that of the death of a patient in the ER with symptoms of an acute myocardial infarction from a falsely low troponin measurement by an affected instrument.

Aim

The review of immunoassay interferences and mainly biotin, as well as the search for automated immunoassay systems affected by biotin.

Methods

Inserts from manufacturers, bibliographic search of articles related to interferences in immunoassays as well as communication with scientific specialists in assay performances of IVD manufacturers.

Results

We found increased number of reports related to interference from exogenous biotin and efforts by manufacturers to avoid it. The table summarizes the vulnerability to biotin interference in six of the most popular immunoassay systems that are used by laboratories participating to ESEAP.

Assay system		Percentage of vulnerability to biotin interference **TODAY** (*mid 2022*)
Siemens	*Centaur*	20%
	Attelica	20%
Beckman coulter *(but with **increased biotin threshold**)*	*Access*	9%
	DxI800	9%
Abbott	*Architect*	0%
	Alinity	0%
Roche *(but with **increased biotin threshold**)*		100%
Tosoh (*all current platforms*)		0%
SNIBE (*all platforms*)		1,2%

Conclusions

Great effort by manufacturers has been done to reduce biotin interference, mainly by using biotin-free reagents (Tosoh, Snibe), precomplexed biotin (Abbott, Siemens) or increased biotin threshold (Beckman Coulter, Roche). This leaded to the elimination of this serious interference.

eP03

IMPORTANCE OF IMMUNOASSAY METHODOLOGY IN THE ASSESSMENT OF FREE ANDROGENIC INDEX IN TRANSGENDER MEN

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Introduction

During gender affirmation therapy free androgen index (FAI) is often used to assess androgenisation. Its calculation depends on testosterone and sexual hormone binding globulin (SHBG) values. The gold standard is mass spectrometry análisis for testosterone determination, but currently the most widespread technique is immunoassay. Both determinations may differ depending on the antibody used.

In these patients, the estimation of the androgen index is one of the bases for lifelong follow-up. Consequently, the possibility of a change in the methodology used by the laboratory is considerable.

Aim

Our aim was to study if differences exist between two inmunoassay methods and if fact could affect the calculation of the ratio.

Material and methods

Sixty-four SHBG and testosterone samples from 40 transgender men were analysed based on the document "Statistical methods in the comparison of laboratory equipment" (AEBM, 2013). The autoanalysers Atellica IM from Siemens Healthineers and Maglumi 2000 from Snibe were used and FAI was calculated for each methodology. In addition, free testosterone was determined on the Maglumi2000 to analyse the correlation of FAIs with free testosterone.

Results

Passing-Bablok regression showed that FAI estimation using both methodologies is interchangeable (a=-0.002 CI95% [-0.01648 to 3.3305] and b=1.080 CI95% [1.000 to 1.259]) by including 0 in the Y-intercept interval and 1 in the slope interval. Nevertheless the results for testosterone and SHBG separately were not. Both methods were correlated with free testosterone analysed in the Snibe autoanalyser (p<0.001), (Maglumi ρ=0.885; Atellica ρ=0.759).

Conclusion

In transgender men FAI using one or the other methodology (Maglumi2000 vs Atellica) could be considered as equivalent. However, neither testosterone nor SHBG techniques are exchangeable if used separately. It is important to take into account the available methodology when assessing the evolution of these parameters.

ePO4

STUDY OF GASOMETRIES

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Introduction

The clinical laboratory is subject to the influence of a society that demands higher quality care and with an increasingly adjusted cost efficiency. For this reason, we must know how to value the analytical equipment that the different commercial houses propose to us when a technological renewal contest is launched.

The main objective of our study was to evaluate the variability of results of the different parameters measured by two RAPID POINT 500e gasometers, RADIOMETER ABL800FLEX used in our laboratory, according to the SEQC recommendations.

Material and methods

50 whole blood samples were randomly selected, from hospitalized patients and from the emergency area, which were processed instantly upon receipt in the laboratory and in parallel by both analyzers, firstly in the RADIOMETER equipment. ABL800FLEX and then in RAPID POINT 500e. For the statistical analysis, the Passing-Bablok linear regression , the Bland-Altman mean difference, the Pearson correlation coefficient and the intraclass correlation coefficient were studied using the statistical program MedCalc ® 16.4 .3

Results

All possible combinations were made two by two

Conclusions

In the comparison RAPID POINT 500e GASOMETERS, RADIOMETER ABL800FLEX according to the Passing-Bablok linear regression No systematic or constant differences were detected (the 95% CI of the ordinate at the origin contains 0), nor of the proportional type (the 95% CI of the slope contains 1), thus concluding that both methods are interchangeable and that there is a good correlation for the parameters studied between both teams, which would allow a change of equipment without the results being affected.

ePO5

A COMPARISION OF ESTIMATION OF ELECTROLYTES (NAMELY SERUM SODIUM AND POTASSIUM) BY A WET CHEMISTRY ANALYZER (EASYLYTE) WITH THAT BY A DRY CHEMISTRY ANALYZER (VITROS350)

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Introduction

The estimation of serum electrolytes by the Vitros 350 is based on the dry chemistry technology which has completely eliminated the need for water. Since the sample volume needed in the Vitros 350 is only 10 µl, the instrument is advantageous for paediatric and neonatal samples. A study was designed to compare the results of estimation of sodium and potassium obtained by the Vitros 350 with those of the wet reagent analyzer (Easylyte), the latter serving as the reference instrument.

Aim

The purpose of the study is to assess the agreement between results of electrolytes (serum sodium and potassium) estimated by a wet chemistry instrument with that obtained by a dry chemistry analyzer.

Materials and methods

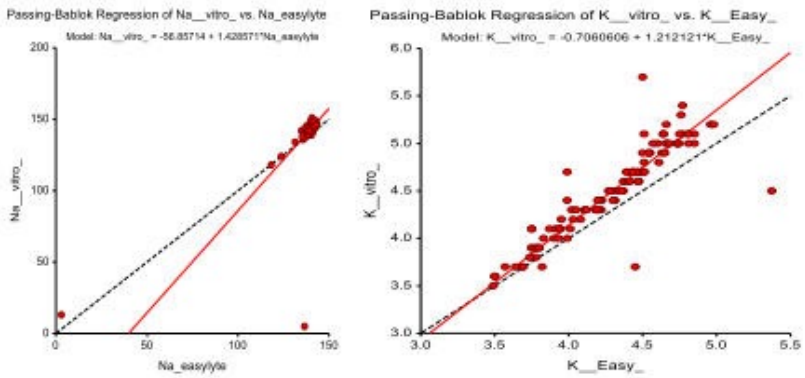
120 samples were selected randomly from the usual lab workflow.No exclusion criterion was applied except for grossly hemolysed, lipemic or icteric samples. All the samples were run on both the instruments.The results thus obtained were compiled, tabulated and statistically analyzed.

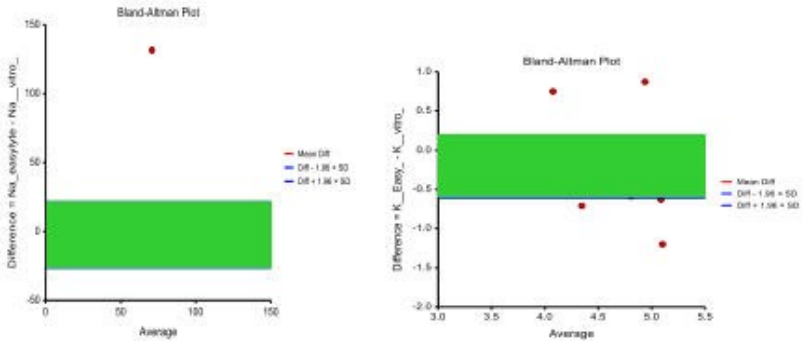
Results and Analysis

The Wilcoxon rank sum test revealed a significant difference in the means in the results obtained from the two different analyzers (Sodium-Z=-9.14,P<0.00001,Potassium-Z =8.84(P<0.00001).Kendall's correlation coefficient t (sodium)=0.50 and t (potassium)=0.83,suggested a strong correlation for potassium and a moderate correlation for sodium. Passing Bablock regression concluded that the methods are not equal. Bland Altman plots show a bias of -2.22 for sodium and -0.21 for potassium. Thus, statistical analysis revealed conflicting solutions and no comments were possible on clinical decision limits and linearity trends with this small sample size.

Conclusion

Statistical analysis revealed conflicting solutions because the methodologies used in the two analyzers differ greatly and this may affect the level of agreement in the measurements.





	Sodium (Easylyte)	Sodium (Vitros)	Potassium (Easylyte)	Potassium (Vitros)
Mean	139.1	141.1	5.37	4.5
Median	139.2	143	4.26	4.5
Mode	139.2	142	3.99	4.7
Standard deviation	2.99	3.88	0.36	0.45

Table showing the descriptive statistics of the Sodium and Potassium measurement by the Wet Chemistry instrument (Easylyte) and the Dry Chemistry instrument (Vitros 350).(units= meq/L)

ePO5

A COMPARISION OF ESTIMATION OF ELECTROLYTES (NAMELY SERUM SODIUM AND POTASSIUM) BY A WET CHEMISTRY ANALYZER (EASYLYTE) WITH THAT BY A DRY CHEMISTRY ANALYZER (VITROS350)

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Results and Analysis

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Conclusion

Statistical analysis revealed conflicting solutions because the methodologies used in the two analyzers differ greatly and this may affect the level of agreement in the measurements.

ePO6

ASSESSMENT OD DATA WORKFLOW AMONG CANCER HEALTHCARE PROVIDERS IN PALESTINE: REDESIGNING HEALTH INFORMATICS MODEL FOR CANCER REGISTRY

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Introduction

Cancer registries collect tumor-related data to monitor epidemiological incident or mortality rates and support cancer research in both studies hospital based and population based. Data workflow process is a common concern when applying hospital or population based registry data among healthcare providers, and only a few recent worldwide studies have systemically assessed cancer registry data workflow. To our knowledge there is a lack of systematic assessments of data workflow in Palestine and therefore there are no specific guidelines for reporting cancer cases through the National Cancer e-Registry in Palestine.

Aim

Our study aimed at assessing the efficacy of the data workflow in the cancer registry in Palestine as well as redesigning a model for such a data workflow.

Methods

We adopted a triangulation (quantitative and qualitative) study approach to assess the data workflow and used the International Agency for Research on Cancer (IARC) variables to subtask within the process cycle. We adopted a model designed by Indiana University and an assessment framework for health modeling that was developed by the European Network of Cancer Registries (ENCR) to collect data from cancer registrars and data collectors.

Results

Our results show an average completeness and validity of the data (95.56%), with barriers and facilitators categorized into six themes, with the most common barrier being related to localization of health services and not understanding the concepts related to cancer registry.

Conclusion and main findings

Our study shows that the most reported facilitators are related to benefits from previous experiences. Our redesigned workflow ensures facilitating communication between registrars and their access to information in the focal point centers. Our study confirms that a proper cancer registry is very important as a form of evidence-based data to improve quality of care and prevent cancer deaths in Palestine, which is fundamental to improve policies.

ePO7

CLINICAL CHARACTERISTICS OF HOSPITALIZED COVID -19 PATIENTS IN PALESTINE (Descriptive Analysis and Data Mining Approach)

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A retrospective review of 199 patient's medical records was conducted in the year 2020 to understand the clinical knowledge about COVID-19 patients in Palestine, using conventional statistical analysis and data mining tools, to develop a model that can predict patients experience of severe or non-severe symptoms during their disease course.

This study was conducted in two phases; First: a comprehensive descriptive analysis of the patient's demographic, comorbidities, complaints, and laboratory findings, examined the relationship between each variable with patient's health status and the severity of the symptoms experienced (severe, non-severe), This phase showed that the most common symptoms on admission were cough (51.5%), fever (41.7%), and shortness of breath (25.8%). Numerous differences were reported between severe and non-severe cases, including higher WBC, neutrophil, LDH, ferritin, BUN, and creatinine (P<.05), and lower lymphocytes percentage and SPO2 (P<.001). Relationships between CRP and severity level on the one hand and Ferritin level, and Monocytes on the other were statistically significant (P-value <0.05). Having diabetes, hypertension and Cerebrovascular Disease was significantly associated with the higher severity level of COVID-19 (P<.001). Second: The disease severity data from the first phase with artificial intelligence (AI) algorithms were used to develop an algorithm that can predict the patients' future condition. Four popular machine learning tools were used; Logistic Regression (LR), Support Vector Machine (SVM), Decision Tree (DT), and Artificial Neural Networks (ANNs). Models' accuracy levels were; 71%, 71,5%, 71,5%, and 74%, respectively, all considered high accuracy levels the models registered in spite of the limited available data used in models testing.

Conclusion

AI can be very effective to describe behavior of COVID-19 and inform clinical decisions and treatment plans.

ePO8

GENES SETS CLASSIFICATION BASED ON OPTIMIZED MULTICLASS LEAST SQUARES SUPPORT VECTOR MACHINES

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Gene expression datasets are large datasets, and they are rich source of valuable and informative genes. Thus, the identification of informative genes groups is difficult. Support vector machine (SVM) is one of the popular methods in machine learning since SVM have good results in terms of classification and prediction. Classification analysis is the process of finding the best model of the classifier to predict classes of data whose class label is not specified. In this study, the Least Support Vector Machine (LS-SVM) method is used to classify genes groups in gene expression data sets, since LS-SVM is better compared to standard SVM in computation processes, rapid convergence, and high precision. The data sets are gene expression data for leukemic cells. The data sets are preprocessed prior to the classification process. The LS-SVM has several parameters. Optimal parameter delivery to the LS-SVM method can affect the classification accuracy. Thus, for the optimal parameter selection method on LS-SMVS we used Tabu algorithm (TS). The study results show that the selection of optimal parameters can significantly increased classification accuracy and computational process times.

ePO9

ANTIBODIES AGAINST ANTIGENS OF THE MIDBODY REGION IN A PATIENT WITH RAYNAUD'S SYNDROME

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Introduction

Antibodies against antigens of the midbody region are a rare autoantibody pattern. The aim of our study is to present the detection of this pattern in a patient with Raynaud's Syndrome.

Case Presentation: A 67 year old woman, dialysis patient, showed localized pain in her hand fingers, characteristic "pins and needles" sensation, skin discoloration and sensation of cold. We performed biochemistry tests, hematology tests, serum protein electrophoresis and coagulation tests. The abnormal test results are presented in the following table. Indirect immunofluorescence showed anti-midbody at high titers of >1/1280 and ANA cytoplasmic pattern a high titers of >1/1280.

Discussion

The midbody is occurred in the final phase of cell division and includes microtubuli related to the spindle mid-zone, as well as certain associated proteins. Midbody pattern is related with Sjögrens Syndrome, Raynaud's Syndrome and gastrointestinal system cancer. According to some cases in literature, the presence of anti-midbody antibodies seems to be related to Pre-Systemic Sclerosis. The aforementioned patient was followed up for a long period of time and showed a later manifestation of Systematic Sclerosis.

TEST	RESULT	UNITS	NORMAL RANGE
ferritin	153,2	ng/mL	10-120
fibrinogen	468	mg/dL	200-450
igG	539,4	mg/dL	700-1600
CRP	32,7	mg/L	0-7
ESR	35	mm	0-20
reticulocytes	3,5	%	0-2
Hct	26,9	%	37-47
Hb	8,8	g/dL	12-16
RBC	3,13	M/μL	4,00-5,00

eP10

ANTINUCLEAR ANTIBODIES IN RHEUMATOID FACTOR SEROPOSITIVE PATIENTS IN ALBANIA

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Introduction

Autoantibodies are a common and characteristic feature of rheumatic autoimmune diseases.

Aim

This study aims to explore antinuclear antibodies (ANA) expression in Rheumatoid Factor (RF) seropositive patients in Albania.

Methods

This is an observational prospective study performed in Laboratory Department, University Hospital Center 'Mother Teresa' (UHCMT), Tirana from January 2022 to July 2022. In this study are enrolled 85 consecutive adult RF seropositive patients that were tested for ANA presence. Data (age, gender, anti-CCP, CRP, LDH, Albumin, complete blood count results) were electronically collected from laboratory information system of Laboratory Networks, UHCMT. Quantitative determination of RF was performed on Alinity-c analyzer,Abbott with immunoturbidimetric methodology. Semiquantitative determination of IgG ANA was performed with indirect immunofluorescence using Euroimmun Mosaic HEp-20-10/Liver kit. Titers equal or higher than 1:160 were considered positive. Comparison between ANA positive and negative patients for gender, age and laboratory parameters was performed using IBM SPSS Statistics 26. P-value<0.05 was considered statistically significant.

Results

82.4% were women and 17.6% were men. The average age was 58 +/- 14 years. 60 patients (70.6%) were ANA positive with the following titers: 1:160 in 38%, 1:320 in 31%, 1:640 in 15% and 1:1280 in 16 % of ANA positive patients. 88.6% of ANA positive patients had speckled fluorescence pattern, 5.4% had mixed nucleolar and speckled fluorescence pattern, 4% nucleolar pattern and 2% homogenous pattern. 52% of ANA positive patients showed mitotic fluorescence. 2% of the patients presented with cytoplasmic fluorescence. ANA positive patients were significantly younger than RF positive patients with negative ANA. ANA positive patients presented with significantly higher RF titers. Elevated CRP was found on 52% of the patients. ANA positive patients had significantly higher CRP than ANA negative patients. Anti-CCP were positive in 51% of patients. Anemia was present in 34.1% of patients. No significant differences were found among ANA positive and negative patients regarding gender, anti-CCP positivity and complete blood count parameters.

Conclusions

Antinuclear antibodies are a frequent finding among RF-seropositive patients in Albania. The most frequent fluorescence pattern is speckled. Further studies are needed to evaluate the role of antinuclear antibodies in the prognosis of rheumatic diseases.

eP11

ANTINUCLEAR ANTIBODIES WITH IMMUNOFLUORESCENCE AN IMPORTANT SCREENING TEST IN POLYMYOSITIS

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Introduction

Indirect immunofluorescence (IIF), in which HEp-2 cells are used as a substrate, is a very important technique for detecting antinuclear antibodies (ANAs) in autoimmune diseases. One of the rarest and most difficult autoimmune disease to diagnose is polymyositis, which presents with inflammation and muscle weakness.

Aim

To show the importance of ANA with immunofluorescence in diagnosis of polymyositis.

Method: ANA IFA was performed using ANA Hep-2 kit. ENA profile and ANA screen elements were measured using the ELISA method in Alegria. Anti-PM/SCL 100 and anti-MI-2 elements were measured using the EIA method.

Result

A 63-year-old women was presented to a rheumatological clinic with a three-month medical history of muscle pain and stiffness, shortness of breath, difficulty swallowing. The rheumatologist recommended an antinuclear antibody test.

The IIFA method was chosen because of the advantage of including the pattern of ANA. ANA IIFA Hep-2 resulted positive with a titer of 1:1280 and homogenous fine granular nucleolar pattern, typical in PM/SCL-100 antibodies found in polymyositis.

The laboratory findings showed high WBC at 11.6x103 mm3, elevated ESR at 40 and increased CPK at 210 U/L, rheumatoid factor was negative at 9.63, Anti-CCP was negative at 2.5. The autoantibodies resulted negative as follows: Anti-dsDNA 1.4, Anti-Jo-1 0.3, Anti-Scl-70 1.8, Anti-Sm 0.8, Anti-SS-A 0.1, Anti-SS-B 1.7, Anti-RNP-70 0.8.

Only two autoantibodies PM/Scl-100 3.0 and Mi-2 3.0 resulted positive, which supported the diagnose of polymyositis. The patient took an Electromyography and a muscle biopsy and with positive laboratory results the diagnosis of polymyositis was confirmed.

Conclusion

ANA immunofluorescence is a very important screening test of polymyositis. For better clinical relevance of polymyositis antibodies, we suggest a combined detection strategy based on ELISA, EIA and ANA-IIFA methods.

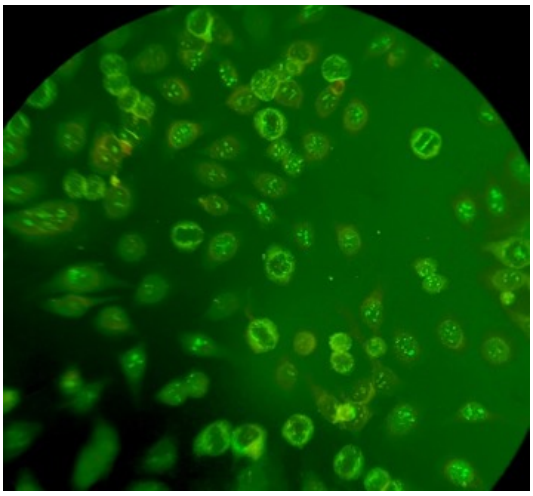

Fig1. ANA-Hep-2 slide positive ANA , nucleolar pattern.

eP12

DETECTION OF ANTIBODIES TO EXTRACTABLE NUCLEAR ANTIGENS (ENAS) IN PATIENTS WITH AUTOIMMUNE DISEASES

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Introduction

The aim of our study is to evaluate the frequency of different autoantibodies to extractable nuclear antigens (ENAs) in patients with autoimmune diseases and their correlation with ANA titer and fluorescence pattern

Materials and Methods

The study was performed via a review of the medical records of patients with autoimmune diseases. A total of 487 patients (73,1 % women and 36,9% men) were included. All patients were positive for ANA test and underwent anti-ENA examination as a complementary test. Anti-ENA evaluation was performed using an immunoblot assay (AESKUBLOTS ANA-17 Pro) for the qualitative determination of IgG autoantibodies against dsDNA, Nucleosomes, Histones, SmD1, PCNA, P0, SS-A/Ro60kD, SS-A/Ro52kD, SS-B/La, CENP-B, Scl-70, U1 snRNP, AMA M2, Jo-1, Pm-Scl, Ku and Mi-2 s in serum or plasma.

Results

78 out of the 487 ANA positive patients were anti-ENA-positive. The anti-SSA/Ro autoantibody exhibited the highest frequency in the group (57.7%; 45/78). 33 sera were exclusive for anti-SSA/Ro and among them 3 cases were SS-A/Ro52kD. Moreover, 4 sera were associated with both anti-SSA/Ro60kD and RNP, 7 sera were associated with both anti-SSA/Ro60kD and SS-B/La and 1 serum with both anti-SSA/Ro60kD and Scl-70. RNP exhibited a frequency of 20, 5% (16/78), SS-B/La 12,8% (10/78). Sm and Scl-70 6,4% each (5/78) and Jo-1 a frequency of 1,2% (1/78). Anti-dsDNA exhibited a frequency of 7.7% (6/78). The following ANA patterns occurred in patients with positive anti-ENA: speckled (44/78), centromere (3/78); homogeneous (29/78); and nucleolar pattern (2/78). The association results indicated a significant positive relationship between ANA titer and the presence of anti-ENA.

Conclusions

Screening for autoantibodies against ENA seems mandatory in patients with autoimmune diseases, especially when ANA titer is high. Immunoblotting was introduced in the 1980s and has been a very useful tool in understanding the spectrum of anti-ENA, and in particular, the dissection anti-SSA/Ro reactivity into 60 kDa and 52 kDa subsets.

eP13

APPLICATION OF TISSUE MICROARRAY (TMA) TECHNIQUE FOR IMMUNOHISTOCHEMISTRY ON BREAST CANCER TISSUE

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Introduction

Tissue microarrays (TMA) is a useful technique for studying the expression of specific proteins in tumors and confirming diagnosis under standard conditions on a single slide, and providing maximum preservation and archiving of tissue samples as well as to use smaller amounts of antibodies and reagents.

Aim

Tissue microarray (TMA) is not yet applied in Syria, so we aimed in this research to first establish and optimize this technique at the department of pathology, Tishreen university hospital, Latakia, Syria.

Methods

88 blocks of breast cancer patients were selected, and the tissue specimens of breast cancer donors were placed in the block using tissue microarray (EZ-TMA™) by punching 3 cores from each donor. The first TMA block contained 42 breast cancer cases and the other 46 breast cancer cases.

Three different conditions were tested for merging: 43°C for 2 hours, 46°C for 2 hour): The TMA block was placed with cores facing upward for two hour in a 46°C and 43°C for 24 hours. Then sectioning using (medite®) Microtome with three different sections' thickness 3 micrometer, 5 micrometer and 7 micrometer.

Iimmunohistochemistry was performed to detect CD3 using Anti CD3 antibodies (Bio SB Inc®) to detect tumor infiltrating lymphocytes TILs.

Results

The following parameters proved effective and high quality: incubating at 43 °C for 24 hours then cooling to room temperature, cutting with a thickness of 7 µm gave the best stained slides. CD3 staining showed TILs CD3+ low or no expression among 69.5% of patients, medium expression among 19.5% of patients and high expression among 11%. High expression of TILs was observed in triple negative breast cancer and this finding is similar to Rashed et. Al, 2021 findings.

Conclusions

This study resembles the first experiment on tissue microarray in Syria. Breast cancer tissues provide invaluable resources for TMA creation. TMA should be optimized to have precise assessment. Multiple patient samples can be studied for many cancer biomarkers using TMA technique effectively using the minimum amount of reagents under the same standard conditions.

eP14

INTEGRATIVE OMICS APPROACH IN BREAST CANCER MANAGEMENT

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Introduction

As the most frequent malignancy and a leading cause of cancer death among women worldwide, breast cancer (BC) remains a huge challenge for multidisciplinary approaches in advancement of the diagnostic and treatment strategies.

Aim

To evaluate and highlight the potential of integrative omics approach in BC management.

Methodology

Comprehensive literature survey.

Results

The advanced genomic methodologies like next-generation sequencing, complemented with the novel insights in transcriptomics, epigenomics, proteomics, metabolomics and microbiomics, have set the scene for large-scale research aimed at discovery and clinical application of novel, highly specific laboratory markers to enable early diagnosis, precise prognosis and classification, timely and personalized treatment, effective monitoring or design of a targeted pharmacotherapy.

Apart from breast tissue, specimens like urine, tumor interstitial fluid, blood, serum or plasma, saliva and the nipple aspirate fluid are frequently used as sources of biomarkers in clinical research involving various 'omics approaches. The set of laboratory markers currently implemented in clinical practice, including CA 15-3, CEA, ER-, PR-, HER2-expression, BRCA1 and 2 mutations, is expected to be supplemented with novel promising markers like microRNA molecules (oncomiR and TS-miRNAs), circulating RNAs, circulating tumor DNA, circulating cell free DNA, CA27.29 molecules, but potential is also identified for EZH2, GP88, Cyclin-E, B-Myb, Twist, DMP1β, RB1, novelty discovered hub genes like CENPL, ISG20L2, LSM4, MRPL3, and a number of metabolites, amino acids and lipids.

The available 'omics databases like PRIDE, COSMIC, GENIE, TCGA, CPTAC, GXB, GEO, HUPO and TACCO; the molecular assays including OncotypeDx and MammaPrint; cancer metabolic biomarker database (CMBD); the Human OncoBiome Database; the AURORA program, the BC epigenomics track hub, have aided substantially the integration of results and their in silico analysis, data access to independent research groups enabling evaluation of previously detected or identification of novel markers with clinical relevance including: genes, proteins, metabolites, microbiota-constituents.

Conclusion

The novel 'omics technologies have dramatically changed our understanding of the mechanisms underlying cancer. Their integrative application holds huge potential in characterization of BC heterogeneity in a therapeutically meaningful way as well as identification of potential biomarkers to be used as hallmarks of the disease or targets for personalized drug design.

eP15

THE ROLE OF LABORATORY IN DIFFERENTIATING PROSTATE CANCER FROM BENIGN PROSTATIC HYPERPLASIA

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Background/Aim

Prostate cancer (PC) and Benign prostatic hyperplasia (BPH) are common diseases in males older than 60 years old. Their early diagnosis improves prognosis and quality of life. PSA is a biomarker organ specific, but not disease specific, and is necessary to study other biomarkers to distinguish these diseases from each other.

Methods

We have studied retrospectively 102 cases, during the period June 2007-October 2020, 63 of them with BPH and 39 with PC. Whole blood K3-EDTA samples were analyzed with ABX MICROS 60 and serum separator gel samples for measuring PSA, free PSA were analyzed with ARCHITECT 2000.

Results

There was a significant correlation between PC and BPH cases for age, total PSA, ratio f PSA /t PSA, MPV, PDW. (Table nr.1). In order to deepen our analysis, we evaluated the sensitivity and specificity of the tumor marker PSA, ratio f PSA/ t PSA, hemogram parameters and the combination of these parameters in order to make a better differential diagnosis between PC and BPH. During this analysis, with ROC curves,use of PDW and MPV in addition to PSA in the diagnosis of PC increases sensitivity at 81.2% and specificity at 91.6%. The use of ratio f PSA/t PSA in the diagnosis of PC has sensitivity 72.3% and specificity 93.6%, higher than PSA alone.

Conclusions

Use of ratio f PSA/t PSA instead of total PSA has a higher accuracy in diagnosing PC. Also measurement of laboratory parameters like MPV and PDW can elevate sensitivity and specificity of total PSA measurement alone in PC and BPH cases.

Table nr.1

	DIAGNOSIS	MEAN	STANDART DEV.	P
AGE	BPH	70.43	7.420	**0.002***
	PC	66.00	5.620	
PSA	BPH	6.6465	2.49259	**0.000***
	PC	12.5723	6.64408	
f PSA/t PSA	BPH	0.3247	.1791	**0.000***
	PC	0.1275	.0578	
PDW	BPH	12.3349	1.27498	0.009*
	PC	13.9487	1.87070	
MPV	BPH	7.8397	1.19063	**0.017***
	PC	6.4462	1.24580	

eP16

THE NEW BIOMARKERS [-2]proPSA AND PHI (PROSTATE HEALTH INDEX) IN THE DIAGNOSIS OF PATIENTS WITH PROSTATE CANCER

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Introduction

A low concentration of FreePSA has been shown to be associated with prostate cancer. However the past years several isoforms of fPSA have been identified as precursor forms of PSA. The [-2]proPSA form has been determined to be the most specific for prostate cancer. When [-2]proPSA measurements are combined with PSA and fPSA, the resulting index, known as Prostate Health Index (phi) is a patient's personalized prostate cancer risk assessment.

Aim

The aim of the study is to provide improved clinical specificity, leading to a potential reduction of unnecessary biopsies, and individualized risk assessment for prostate cancer.

Methods: The concentration of [-2]proPSA was measured in the serum of 23 patients using the new immunoassay of Beckman Coulter. The patients aged 50 years and older, with non-suspicious digital rectal examination findings and their PSA was 2-10ng/ml.

Results

From the concentration of [-2]proPSA , fPSA and PSA we calculated the phi. The results are represented in the table below.

As it is shown, 3/23 patients(13,04%) had a ratio<0,25 and a phi 0-20,9, which mean that they were in low risk of prostate cancer. 15/23 patients (65,21%) had a ratio <0,25 and phi 21-39,9,which classifies them in the moderate risk group of prostate cancer and 4 of them (17,39%) had a ratio<0,25 and phi >40, which means that they belong in the high risk group.

Conclusions

The measurement of the [-2]proPSA and the phi, may provide improve discrimination between prostate cancer and benign disease in men with a PSA concentration 2-10ng/ml and negative digital rectal examination, as it classifies the patients in the low, moderate and high risk group of prostate cancer. Generally, phi may have a place in clinical decision strageties including mpMRI, in order to reduce unnecessary negative initial or repeated biopsies, which in order to be proven, needs further research.

PSA 2-10ng/ml	phi 0-20,9	phi 21-39,9	phi 40+
fPSA/PSA >0,25	1		
fPSA/PSA <0,25	3	15	4

eP17

THE SIGNIFICANCE OF NEUTROPHIL-LYMPHOCYTE RATIO AND PLATELET- LYMPHOCYTE RATIO IN THE PROGNOSIS OF CARDIOVASCULAR COMPLICATIONS IN COVID-19 PATIENTS

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Introduction

The main characteristic of COVID-19 disease is the development of severe acute respiratory syndrome. In addition, a massive inflammatory host response and multiorgan dysfunction may occur. Uncontrolled inflammatory cascade can cause numerous complications, including cardiovascular events, contributed by direct viral invasion of the cardiomyocytes as well as severe hypoxia.

Aim

The aim of our study was to evaluate the significance of neutrophil-lymphocyte ratio (NLR) and platelet-lymphocyte ratio (PLR) in the prognosis of cardiovascular complications in COVID-19 patients. We correlated NLR and PLR with the concentrations of certain biomarkers of cardiomyocyte damage.

Methods

This case-control clinical study enrolled 100 patients with COVID-19, who were hospitalized at the University Clinical Center in Kragujevac during the first quarter of 2021. The control group comprised 60 healthy controls. Data on laboratory parameters, determined by standard accepted methods in Laboratory diagnostic service, were collected and analyzed: (1) markers of inflammation (NLR, PLR, and C reactive protein (CRP)), and (2) markers of cardiomyocyte damage (creatine kinase (CK), CK-MB isoform, lactate dehydrogenase (LDH), pro-B-natriuretic peptide (proBNP), and high sensitive troponin I (hs-TnI)).

Results

The average age of COVID-19 patients was 59.97 ± 11.78ys, and 58.91 ± 17.71ys in control subjects (p=0.436). Significant differences in NLR (8.21 ± 4.38 vs. 2.63 ± 1.38, p<0.001) and PLR (236.18 ± 185.13 vs. 131.89 ± 54.75, p=0.001) values between COVID-19 patients and controls were established. It has also been shown that COVID-19 patients had significantly higher concentrations of CRP (61.56 ± 68.84 vs 3.1 ± 1.98 mg/dL), LDH (842.16 ± 666.49 vs 283.95 ± 58.47 U/L), proBNP (753.28 ± 579.49 vs 56.96 ± 52.49 pg/mL) and hs-TnI (0.36 ± 1.0 vs 0.01 ± 0.01 ng/mL) compared to controls (p<0.001). Bivariate correlation test confirmed a significant relationship of NLR with CK-MB (r=0.517, p<0.001), proBNP (r=0.377, p<0.001) and hs-TnI (r=0.249, p=0.041) in COVID-19 patients. PLR was positively correlated with CK-MB (r=0.322, p=0.005). There were no significant relationships of NLR and PLR with biomarkers of cardiomyocyte damage in the group of healthy subjects.

Conclusion

NLR and PLR could be a useful biomarkers in the prognosis of cardiovascular complications in COVID-19 patients.

Key words

neutrophil-lymphocyte ratio, platelet-lymphocyte ratio, COVID-19, cardiovascular complications

eP18

THE SIGNIFICANCE OF THE DE RITIS RATIO IN ACUTE MYOCARDIAL INFARCTION

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Introduction

An increase in the De Ritis ratio in patients with acute myocardial infarction is a consequence of organ damage and systemic hypoperfusion, and according to literary data, its value significantly correlates with the severity of the clinical picture and has prognostic significance.

Aim

The aim of our research was to determine the significance of the De Ritis ratio in patients with acute myocardial infarction, as well as its correlation with biomarkers of inflammation and heart muscle damage.

Methods

The research was designed as a cross-sectional retrospective observational study that included 50 patients with acute myocardial infarction, treated at the Cardiology Clinic of the University Clinical Center Kragujevac, in the period from January to April of 2021. Using standard laboratory methods, serum concentrations of: high-sensitivity troponin I (hsTNI), NT-pBNP and CRP were determined in all subjects; enzyme activity: CK, CKMB, AST, ALT; the total number of neutrophils, the number of lymphocytes, and the De Ritis ratio as the ratio of neutrophils to lymphocytes (N/L ratio) was monitored.

Results

Based on the results of the correlation analysis in patients with acute myocardial infarction, a statistically significant correlation of the value of the De Ritis ratio was shown with: hs TNI concentration ($r = 0.545$, $p < 0.001$); CK activity ($r = 0.578$, $p < 0.001$) and CKMB activity ($r = 0.634$, $p < 0.001$). Likewise, a statistically significant correlation between the N/L ratio and the concentration of NT-pBNP ($r = 0.346$, $p = 0.015$) was shown, as well as the concentration of CRP with the concentration of hs TNI ($r = 0.514$; $p < 0.001$). Univariate logistic regression showed that the De Ritis ratio is the most significant predictor of the development of acute myocardial infarction (OR=8.8; p<0.001) in the examined group of patients in relation to AST (OR=1.15); ALT (OR = 1.03); CK (OR=1.01).

Conclusion

The De Ritis ratio is a significant predictor of the development of acute myocardial infarction and is correlated with biomarkers of cardiomyocyte damage.

Key words: De Ritis ratio, acute myocardial infarction, biomarkers of cardiac damage, prediction, inflamation

eP19

HOMOCYSTINEMIA AND POLYMORPHYSM OF THE GENE FOR METHYLEN TETRAHYDROFOLATE REDUCTASE (C677T) IN PATIENT WITH CORONARY ARTERY DISEASE

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Background

Hyperhomocysteinemia, which commonly occurs either as a result of a methylene tetrahydrofolate reductase (MTHFR) gene mutation or vitamin B12 and folic acid deficiency, has been reported as a risk factor for coronary artery disease (CAD).

The goal of this study was to determine the concentration of total homocysteine ??(tHcy) and the prevalence of C677T mutation of MTHFR in healthy subjects and CAD patients, and moreover have been analyzed, as a risk factor that may affect this disease.

Material and Methods

The study included 123 healthy subjects as control group and 81 patients with diagnosis CAD confirmed coronarography. Plasma tHcy concentration was determined by a cyclic enzyme method, and MTHFR gene polymorphism was analyzed by Polymerase Chain Reaction (PCR) and fragment length polymorphism (RFLP) by Schneider.

Results

The plasma tHcy concentration in healthy subjects and in CAD patients for female were (8.3 ± 2.3 μmol / L vs. 18.73 ± 5.71 μmol / L), whereas in men were (11.1 ± 5, 0 μmol / L v.s 14.5 ± 6.16 μmol / L). The statistically significantly higher tHcy concentrations was in CAD patients than in healthy subjects ($p < 0.001$). The values ??of X2test ($x2 = 35.48$ and $p < 0.001$) for both groups showed a significant correlation between the tHcy concentration and development of CAD.

In healthy subjects and CAD patients for mutation of MTFHR gene C677T, genotype heterozygous CT was with highest frequency (46% vs. 50%), then wild homozygous genotype CC (44% vs. 33%), and with the lowest frequency was TT genotype (10% vs.17%). The most important is that homozygous TT is responsible for highest concentration of tHcy and analysis of differences showed that it is a not reasion for development the CAD ($p > 0.05$).

Conclusions

There is a significantly higher plasma level of tHcy in patients with CAD than in healthy subjects which represents risk factor for CAD. Our findings have also shown that current mutations in the MTHFR C677T gene affect tHcy levels, but these mutations are not a risk factor for the development of CAD.

Keywords

total homocysteine, methylene tetrahydrofolate reductase, coronary artery disease

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eP20

DEMAND FOR hs-cTnT DETERMINATION IN THE DEPARTMENT OF BIOPATHOLOGY OF A RURAL HOSPITAL, IN A 5-YEAR PERIOD

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Introduction

High-sensitivity troponin (hs-cTnT) is a useful biomarker for the assessment of heart failure, for the pathogenesis of sub-clinical and acute myocardial infarction. The high-sensitivity assay improves the analytical limits of detection so that the detection of hs-cTnT in healthy subjects is feasible, allowing a better assessment of its levels to determine more precisely what constitutes a clinically significant change in its concentration and therefore, to more successfully identify acute cases. An increase in hs-cTnT is also observed in patients with COVID-19 due to the myocardial damage that accompanies this infection.

Aim

The study of the demand of hs-cTnT determination at the Department of Biopathology of our Hospital, for the period 05/2017- 05/2022.

Methods

We ran a statistical analysis of the data regarding the hs-cTnT determinations that were requested and performed at the Department of Biopathology of the General Hospital of Amfissa. The tests were performed on the Alinity immunoassay analyzer (Abbott). The data were obtained from the database of the CTEAM LIS information system of the Computer Team company, which is used by our laboratory.

Results

The data analysis showed that in the aforementioned period of time, there were performed 8301 determinations of hs-cTnT in a total of 964956 laboratory tests . In Table 1, we present the results per semester.

Conclusions

During this 5-year period, the demand for hs-cTnT testing was gradually increased, but showed an even more rapid raise after the onset of the COVID-19 pandemic and remains, until today, at a high level. This fact makes it difficult to effectively predict the needs for reagents and consumables for the laboratory as well as sometimes, their supply, as, due to increased consumption worldwide, sometimes there is a shortage of them in the trade.

Table 1

hs-cTnT

Semester	1st	2nd	3rd	4th	5th	6th	7th	8th	9th	10th	Total
hs-cTnT	478	516	512	636	694	735	757	1099	1485	1249	8161
Total Tests	95115	92896	93623	98654	127884	86795	96627	94417	99474	79198	964683
hs-cTnT /10.000 tests	50,25	55,55	64,47	54,27	84,68	78,34	116,40	118,1	149,29	157,71	84,60

eP21

DEMAND FOR D-DIMERS DETERMINATION AT THE DEPARTMENT OF BIOPATHOLOGY OF A RURAL HOSPITAL, IN A 4-YEAR PERIOD

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Introduction

D-Dimers(D-dimers, fibrin degradation products) are an indirect biomarker of fibrinolysis. This molecule presents unique properties as a biomarker in cases of hemostatic disorders as well as an indicator of intravascular thrombosis.Mainly,D-Dimers have been extensively studied to rule out the diagnosis of venous thromboembolic disease. In addition, they are evaluated to determine the ideal duration of antithrombotic therapy in patients with venous thromboembolic disease, to diagnose and monitor disseminated intravascular coagulation and other pathological conditions that pose risks of bleeding or thrombosis. Patients with COVID-19 are one such group of high-risk patients, in whom the determination of D-Dimers has significant prognostic value.

Aim

The study of the demand of D-Dimers determination at the Department of Biopathology of our Hospital, for the period 06/2018- 05/2022.

Methods

We ran a statistical analysis of the data regarding the D-Dimers determinations requested and performed at the Department of Biopathology of our hospital. The data were obtained from the database of the CTEAM LIS information system of the Computer Team company, which is used by our laboratory..

Results

The statistical analysis of the retrieved data revealed that in the above period 2736 D-Dimers determinations were carried out in a total of 776945 biopathological tests. In Table 1, the results per semester are presented in detail.

Conclusions

According to our study, during the period 06/2018-05/2022, the demand for D-Dimers showed a steep increase. These results indicate on the one hand the rapid response on the part of clinicians, from the moment it became available the conduct of this particular examination, on the other hand, that the peak of demand coincides in time with periods of outbreaks of the disease COVID-19. D-Dimers are a valuable tool against the disease COVID-19, however the ever-increasing demand may be challenge for the financial management of laboratory resources.

Table 1

D-Dimers

Semester	1st	2nd	3rd	4th	5th	6th	7th	8th	Total
D-Dimers	61	96	107	155	179	481	823	834	2736
Total Tests	93623	98654	127884	86795	96627	94417	99747	79198	776945
D-Dimers /10.000 tests	6,52	9,73	8,37	17,86	18,52	50,94	82,51	105,31	35,21

eP22

ERYTHROCYTE MEMBRANE FATTY ACID CLUSTER AND COMPLIANCE WITH MEDITERRANEAN-STYLE DIETARY PATTERN AMONG TESTICULAR GERM-CELL TUMOR SURVIVORS

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Introduction

Although considered an uncommon cancer accounting for approximately 1% of all oncopathologies affecting men, testicular germ-cell tumors (TGCT) represent the most commonly diagnosed solid malignancy in male population aged 15-40 years. Remarkably high survival rate and relatively young age of patients direct the clinical and research attention towards survivorship care and long-term treatment-induced health repercussions. Due to chemotherapy-related toxicities and potential gonadal dysfunction, testicular germ-cell tumor survivors (TGCTS) may exert an increased susceptibility to developing metabolic disturbances and cardiovascular sequelae. Copious scientific evidence support health advantages of the Mediterranean-style Dietary Pattern (MedDP) for both primary and secondary prevention of cardiovascular diseases, including surrogate risk factors and the overall morbidity and mortality burden.

Aim

This observational, cross-sectional study recruited a sample of TGCTS attending follow-up care at the Clinic of Urology, University Clinical Center of Serbia aiming to investigate the relationship between the compliance with MedDP and erythrocyte membrane fatty acid (FA) profile featuring integrative measure of genetic factors, endogenous metabolism and dietary intake.

Methods

MedDP adherence was assessed with validated semi-quantitative MedDietScore index based on consumption frequency for 11 food groups (wholegrain cereals, fruits, vegetables, legumes, red meat, poultry, fish, dairies, olive oil and alcohol). Erythrocyte FA cluster was determined by gas chromatography.

Results

Based on self-reported dietary intake and predefined scoring system, the study cohort (n=52, age x?=36.44±8.48 years) displayed low-to-moderate MedDP adherence (MedDietScore: x?=26.61±6.04, range 17-42). Significant positive association was observed between the MedDietScore and eicosapentaenoic acid (EPA, C20:5n-3) ($r=0.344$; $p<0.05$), docosahexaenoic acid (DHA, C22:6n-3) ($r=0.401$; $p<0.01$), Omega-3 Index, i.e. %EPA+%DHA ($r=0.391$; $p<0.001$), and polyunsaturated FA (PUFA) balance, i.e. Omega-3 Index/total PUFA×100 ($r=0.307$; $p<0.05$). Overall, 55.77% of subjects had Omega-3 Index under 4% indicating high cardiovascular hazard, while the rest were allocated in the intermediary risk-category. MedDietScore correlated inversely with total saturated FAs ($r=-0.312$; $p<0.05$), and saturated FAs to monounsaturated FAs ratio ($r=-0.224$; $p<0.05$).

Conclusions

Given the importance of cardiovascular health promotion among TGCTS, and beneficial effects of MedDP, tailored interventional programs addressing dietary behavior as major modifiable risk factor may contribute to achieving and preserving a favorable cardiometabolic status in this vulnerable group.

eP23

CORRELATION OF VITAMIN D AND C-REACTIVE PROTEIN IN NEWLY DIAGNOSED ESSENTIAL ARTERIAL HYPERTENSION

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Introduction

Arterial hypertension is a complex, multifactorial disease, the major risk factor for cardiovascular disease.

Aim

The aim of the study was to determine the correlation between vitamin D and C reactive protein levels in patients with newly diagnosed essential arterial hypertension.

Methods

Study included 95 participants of both sexes, divided in two groups. First group consisted of 45 respondents with newly diagnosed essential arterial hypertension and the second group was age ande gender matched control group, consisted of 50 clinically and biochemically healthy examinees. The concentrations of highly sensitive C-reactive protein (hsCRP) and vitamin D (25(OH)D) were determined on Alinity i analyzers, Abbott Dg., for all subjects included in the study.

Results

Vitamin D levels were statistically significantly lower (40.90±17.8 vs. 52.47±19.5; p<0.01), while hsCRP levels were significantly higher in newly diagnosed hypertensive patients (2.95±0.5 vs. 1.60±0.3p<0.01). Significant negative correlation was found between vitamin D and hsCRP in hypertensive group of patients (r = - 0.605; p <0.01).

Conclusion

Vitamin D levels are statistically significantly lower, and hsCRP levels are statistically significantly higher in patients with newly diagnosed essential hypertension compared to the healthy controls. There is a statistically significant negative association between viamin D and hsCRP in patients with newly diagnosed essential hypertension.

eP24

INTERFERENCES LEADING TO ANALYTICAL ERRORS IN THE BIOCHEMICAL LABORATORY THE EFFECT OF MACROMOLECULES ON THE DETERMINATION OF HSTNI

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Introduction

During a cycle of laboratory testing, which consists of three main phases, the pre-analytical, analytical and post-analytical, there is the possibility of errors occurring, which alter the values ??of biochemicals and immunological tests, resulting in the clinician misdiagnosing and in inappropriate treatment. Several studies have reported the presence of macroforms in the serum of patients. Macroforms are high molecular mass complexes of immunoglobulins with enzymes, proteins, and hormones. Their clearance rate in the blood is slow, giving false positive results in immunoassays.

Aim

The aim of this work was the review of the interferences that can lead to analytical errors in a biochemical laboratory, with

particular emphasis on the interferences against immunoenzymatic reactions such as heterophilic antibodies, autoantibodies, macroforms, etc. We focused on the detection of interference due to the presence of macroforms and heterophilic antibodies in hsTnI (high sensitivity troponin I) test in patient serum.

Methods

We collected random consecutive samples for two population groups of hsTnI, control group (values: 100-2000 ng/ml) and study group (values: >100 ng/ml), which were treated with saline and with polyethylene glycol to precipitate immunoglobulin complexes. For the detection of heterophilic antibodies, we use the heterophilic blocking tubes (HBT) kit.

Results

The highest relative frequency of the samples (40/132)shows approximately 80-90% percentage of precipitation with polyethylene glycol. The diagram of PPhsTnI (percentages of PEG precipitated hsTnI) of the control group follows a normal distribution, on the contrary, the diagram of the study group does not. Also, using the HBT kit we confirmed the presence of heterophilic antibodies in 1 of 8 samples hsTnI.

Conclusions

During the immunoassay of the hsTnI test and comparing their two population groups, based on the literature we confirmed the presence of interference due to macroforms and heterophilic antibodies.

Education and Training in Laboratory Medicine

eP25

DIAGNOSIS OF MACROAMYLASEMIA WITH A CRITICAL VALUE OF SERUM AMYLASE IN A PATIENT FROM PRIMARY SCHOOL

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Introduction

Macroamylasemia, or the presence of amylase polymers or amylase complexes with immunoglobulins (mainly IgA) or other plasma proteins, has a prevalence of 0.5-2% in the adult population, being more frequent in the age range of 50-70 years. It is a benign alteration that does not require treatment, although it has been frequently associated with autoimmune diseases, especially celiac disease. It is important to distinguish it from other cases of hyperamylasemia to avoid warnings of critical values that lead to unnecessary studies or treatments.

Clinical case

We received a request for a complete analysis: complete blood count, coagulation study, biochemistry (liver, lipid, iron, hormonal profile, proteinogram, glycosylated, elemental and sediment hemoglobin and microalbuminuria) from the health center, without a presumptive diagnosis, of a 49-year-old patient years of age, of which there is no history in the hospital, the last episode reflected in the primary history being a consultation for dermatitis. We found a marked amylasemia, 34 times the upper limit of normality: 4064 U/l, slight increase in transaminases and hypercholesterolemia. Given this critical value, the responsible physician adds a determination of amylase in urine and serum lipase. Given the results of amylase <20 U/l and normal lipase, a precipitation study was performed with polyethylene glycol and measurement of the remaining enzymatic activity, the results of which confirmed our suspicion of macroamylasemia in the sample.

Conclusions

When faced with amylase elevations, especially without pancreatic symptoms, we must rule out the presence of amylase

macrocomplexes , which, due to their large size, do not filter through the glomerulus and accumulate in the blood without there being a real increase in amylase secretion, to avoid putting protocols for warning of unnecessary critical values are in place that would lead to unnecessary examinations and hospitalizations.

Some authors suggest screening for celiac disease in the presence of macroamylasemia, as this is a paucisymptomatic disease , and in the face of the episode described in the primary history of dermatitis, also frequently associated with celiac disease, it might be useful to complete the requested study with a screening of celiac disease

Endocrinology

eP26

ADVANCED LABORATORY METHODS TO IMPROVE THE DIAGNOSTIC AND FOLLOW-UP OF THE PATIENTS WITH ENDOCRINE DISORDERS

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The aim of the paper is to present how technologically advanced laboratory methods or smart investigation protocols can improve the diagnostic and follow-up of the patients with endocrine disorders

In hormonology, the limits of hormone immunoassay can be overcomed by mass spectrometry.

For chromosomal abnormalities classical cytogenetics is complemented by molecular cytogenetics through in situ hybridization (FISH) and comparative genomic hybridization (aCGH).

Mutations, deletions, duplications are detected by MLPA and sequencing, including next generation sequencing.

The availability of these methods results in comprehensive testing of patients, better diagnosis and care.

eP27

MONOCYTES TO HDL-CHOLESTEROL RATIO IN TYPE 2 DIABETES MELITUS

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Introduction

Type 2 diabetes mellitus (T2DM) is a chronic condition of hyperglycemia characterized by an increased risk of atherosclerosis. Low-grade chronic inflammation and the accumulation of certain lipid particles are the basis of the development of atherosclerosis. The pro-inflammatory and pro-oxidant effects of monocytes may be attenuated by HDL-cholesterol activity.

Aim

The aim of this study was to examine monocytes to HDL–cholesterol ratio (MHR) in patients with T2DM.

Methods

A retrospective study included 50 patients with T2DM and 30 healthy, age and gender matched subjects. Basal glycemia, the index of insulin resistance (HOMA-IR), glycated hemoglobin (HbA1c), parameters of lipid status, and number of white blood cells were taken from the database of the Center of Laboratory Medicine, Clinical Center of Vojvodina. MHR indexes were calculated.

Results

The T2DM group showed statistically higher glycemia (9.00±2.40 vs. 5.2±0.4 mmol/l; p=0.00); HbA1c (57.16±20.72 vs. 34.70±2.21 mmol/mol; p=0.000), HOMA-IR (2.82±1.46 vs 1.19±0.43) and triglyceride values (2.09±1.30 mmol/l vs. 1.58±1.08 mmol/l; p=0.004) as well as significantly lower HDL-cholesterol values (1.25±0.41 mmol/l vs. 1.42±0,38 mmol/l; p=0.000), compared to the control group. MHR was significantly higher in the T2DM group (10.34±1.31 vs. 9.28±0.93; p=0.05). A significant positive correlation was found between MHR and HbA1c (r=0.244; p=0.05); MHR and HOMA-IR (r =0.264; p=0.05) as well as between MHR and triglycerides (r=0.411; p=0.01).

Conclusions

The group of T2DM patients had significantly higher values of the calculated MHR values.

eP28

siMS AND siMS RISK SCORE ARE ASSOCIATED WITH RISK FACTORS FOR CARDIOVASCULAR EVENTS IN UNIVERSITY STUDENTS WITH METABOLIC SYNDROME

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Background

The aim of this study was to examine the validity of siMS and siMS risk score in quantification of cardiovascular risk and to correlate these scores with parameters of inflammation, indicators of obesity and lipid status in obese and non-obese University students.

Methods

The study included 238 University students, aged of 22.3±1.85, of both genders, grouped into three tested groups: Metabolic syndrome (MS), Pre-Metabolic syndrome (Pre-MS) and control group. The following anthropometric, oxidative stress, inflammatory and lipid parameters were measured: body mass index (BMI), waist circumference (WC), hipp circumference (HC), waist-to-hipp ratio (WHR), high sensitive C-reactive protein (hsCRP), total Cholesterol (TC), HDL-cholesterol (HDL-c), LDL-cholesterol (LDL-c), VLDL-cholesterol (VLDL-c), non-HDL-cholesterol (non-HDL-c) and triglycerides (TG). siMS and siMS risk score were calculated using the appropriate formulas developed by Soldatovic et al.

Results

Significantly increased values of siMS and siMS risk scores were obtained in the MS and Pre-MS groups compared to the controls (p<0.0001). Significant strong correlations of calculated scores were obtained with the anthropometric (BMI, WC, HC, WHR) and lipid parameters (HDL-c, VLDL-c, non-HDL-c, TG), while moderate correlations were obtained with

TC, LDL-c, and hsCRP (p<0.0001). ROC analysis showed that both scores have high discrimination power for metabolic syndrome but not for Pre-MS.

Conclusions: Continuous siMS and siMS risk scores represent a practical and accurate parameters for the evaluation of MS and cardiovascular risk among obese young subjects.

eP29

A CASE OF A NON-DIABETIC PATIENT WITH HIGH GLYCATED HEMOGLOBIN BECAUSE OF IRON DEFFICENCY ANEMIA

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Introduction

Glycated hemoglobin HbA1c is an important indicator of glycemic control in diabetic patients and reflects patient's glycemic status over the previous 3 months. HbA1c higher than 6.5% is usually found in diabetes, but it is also observed in other conditions with a prolonged erythrocyte life span as alcoholism, hyperbilirubinemia and anemia. This is a case of a non-diabetic female with high HbA1c and later discovered that the cause was iron deficiency anemia.

Aim

To show the correlation between Iron deficiency Anemia and elevated HbA1c in a patient with normal plasma glucose levels.

Methods

The patient HbA1c was measured with High Performance Liquid Chromatography using BIO-RAD D-10. The patient ferritin was measured with CLIA method in MAGLUM 4000, and complete blood count was measured in a hematology analyzer. Fasting plasma glucose, serum iron and TIBC were measured in a chemistry analyzer (Cobas Integra).

Results

A 34 years old female patient was presented in laboratory for an annual check-up with a one month medical history of fatigue, shortness of breath and cold hands and feet. The patient did not refer any medical disease or taking any medication recently.

The complete blood count showed: Hb 9.2 g/dl, RBC 3.68x106 mm3, WBC 5.6x103 mm3, HCT 26 %. In blood smear was seen hypochromic anemia.

Fasting plasma glucose was 107 mg/dl, AST 18 U/L, ALT 25 U/L, ALP 48 U/L, bilirubin 0.9 mg/dl, creatinine 0.8 mg/dl, urea 35 mg/dl, ferritin 3.5 ng/dl, HbA1c 7.2 %. The hematologist recommended serum iron and TIBC test, which resulted respectively 22 μg/L and 510 mcg/dl.

The patient took two months of iron supplementation therapy and after repeating again the laboratory tests the HbA1c decreased to 6.2% while Hb increased 11.4 %.

Conclusions

Iron deficiency Anemia increases HbA1c level in nondiabetic patients and it should be interpreted carefully in all iron deficiencies patients so it should not lead to misdiagnosis.

eP30

EVALUATION OF HORMONAL PARAMETERS AND PHYSICAL DEVELOPMENT IN PREMATURE PUBERTY AS AN AID FOR DIAGNOSIS AND TREATMENT

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Introduction

Premature puberty is a rare pathology and the most common type is GnRH-dependent puberty. The cause of precocious puberty often can't be found. Rarely, certain conditions, such as infections, hormone disorders, tumors, brain abnormalities or injuries may be the cause. Treatment for precocious puberty includes medication to delay further development.

Aim

The aim of our study is to explore premature puberty based on physical and laboratory data, in order to establish the diagnosis and evaluate the effect of therapy on hormonal parameters and physical development for a period of six years of patients treated at the hospital University "MOTHER TERESA".

Methods

This is a retrospective time study which include 20 patients, 19 females and one male, mean age 6.8 ± 2.8. Patient evaluation was based on physical examination, laboratory data, radiology imaging and therapeutic data followed by a statistical analysis performed with SPSS 22.

Results

Most of patients 45%, are classified on TANNER 3 stage of development at the beginning of treatment. The mean LH level at the beginning of treatment was 3.05 +/- 3.28 and at the end 1.26 +/- 2.14, p = 0.002. While FSH varies from 4.58 +/- 3.4 at the beginning of treatment to 1.8 +/- 1.3 at the end of treatment, p = 0.005. The standard length deviation (SDS) refers to the average length of all patients, at the beginning of treatment is 1.2 +/- 1.5 and changes to 0.69 +/- 1.5 at the end of treatment p = 0.024. SDS for the average total BMI goes from 1.16 +/- 1.19 to 1.18 +/- 1.01 at the end of treatment, p = 0.93. LH values ??reach a peak in min 30 of the LHRH stimulation test, LH> 5UI / L. At the end of treatment most cases have the same biological age as bone age.

Conclusions

Hormonal and physical evaluation is very important in establishing the diagnosis in children with premature puberty and also in evaluating the effects of therapy on their further development.

eP31

COMPARISON OF ION-EXCHANGE HPLC TECHNOLOGY AND DIRECT ENZYMATIC CHEMISTRY ASSAY FOR MEASUREMENT OF HbA1c

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Introduction

The global prevalence of diabetes mellitus has been increasing steadily over the past seceral decades. Monitoring of HbA1c is very important for the management of patients with diabetes There are different methods for its measurement.

Aim

This study compare two methods: ion-exchange HPLC technology on BIORAD D10® and direct enzymatic chemistry assay on Alinity c®

Methods

Sixty patients were tested for HbA1c concentration. EDTA was used as anticoagulant and HbA1c was measured on both analyzer simultaneously within 12 hours from sampling. Testing was perform in accordance with manufacturer's instructions. Statistical analyses were performed using MedCalc Statistical Software version 19.0.7. The difference of measurements was estimated by calculating the bias using Bland-Altmann plots. Comparison of assays was performed using Passing-Bablok regression analysis.

Results

Concentrations measured on Alinity® were between 4.5 and 11.9% (25.7and 106.6 mmol/mol), while on BIORAD® they ranged in 4.5-12.2% (25.7 and 109.8 mmol/mol). Bland-Altman analysis (where the differences of the two paired measurements are plotted against the mean of both measurements) for HbA1c concentration showed that more than 95% of points are between ±1.96 SD of the mean (limits of agreement were between -0.87 to -0.07 and the mean difference was 0.47). samples with HbA1c<6% difference was 0.37 (0-0.7) % and in those above this cut-off 0.56 (0.3-0.8) %. Passing-Bablok analysis on the whole group showed intercept (95% CI) of -0.094 (-0,1833 to 0,4113) with slope (95% CI) of 1,0588 (1,0161 to 1,0952). For samples with HbA1c<6% analogous values were -1,5875 -2,5929 to -0,2937) and 1,3750 (1,1250 to 1,5714), while for those above this concentration exceeded 0,6000 (-0.365-0.345) and 1.000 (0,6000 to 0,9643).

Conclusion

Results of this research showed acceptable agreement between glucose concentration measurement on ion-exchange HPLC technology on BIORAD D10® and direct enzymatic chemistry assay on Alinity c®. These findings should be evaluated in larger studies including samples containing haemoglobin variants.

eP32

THE PREVALENCE OF THYROID AUTOIMMUNITY IN PREGNANCY IN BENUE, A NORTH-CENTRAL STATE IN NIGERIA

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Background

Thyroid dysfunction in pregnancy is often associated with an increased risk of adverse maternal and foetal outcomes. The instability of the thyroid gland may complicate the outcomes of pregnancy. Some may present with thyroid autoimmunity which is rare because thyroid autoimmune activity decreases as pregnancy advances, but thyroid autoimmunity may be present in a normal pregnancy without overt signs of thyroid dysfunction.

Methodology

This was a hospital-based cross-sectional study with two hundred and fifty (250) healthy pregnant women who were enrolled for the study. Participants' blood specimens were collected and analysed for serum Thyroid stimulating hormone (TSH) and anti-Thyroperoxidase antibodies(anti-TPOab) using the Enzyme-Linked Immunosorbent Assay (ELISA). Statistical Package for Social Sciences (SPSS) version 21 was used for statistical analysis.

Results

31(12.4%) participants were identified to have thyroid dysfunction and out of the participants with thyroid dysfunction, a total of 6(2.4%) participants had thyroid antibodies present. 4 (1.6%) being hypothyroid and 2 (0.8%) euthyroid in thyroid status.

Conclusion

The prevalence of thyroid autoimmunity in pregnancy was at 2.4%, and the assay of anti-TPO antibodies and thyroid stimulating hormone were reliable markers in identifying thyroid dysfunction in pregnancy. Most participants with AITD in this study were hypothyroid. In addition, the early detection of thyroid antibodies reduces the risk of adverse maternal and foetal outcomes, while Levothyroxine is a beneficial treatment in established AITD

eP33

THE INFLUENCE OF GLYCEMIA AND THE CHANGES EXPECTED IN THE MOTHER AND FETUS DURING GESTATIONAL DIABETES

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Introduction and aim

Gestational diabetes mellitus (GDM) occurs during pregnancy and they are several risk factors for its occurrence such as higher body weight ,age history o diabetes in the family, previous complications in pregnancy associated with diabetes. OGTT (oral glucose tolerance test) is performed at 24 -28 weeks of pregnancy as a screening for all pregnant women to diagnose in time and prevent complications that can threaten the life of the mother and the fetus.

Material and Methods

Venous blood from 30 respondents (pregnante women) in which blood was taken after 12 h of fasting for testing fasting glycaemia and HbA1C.

Methods

For determining glycaemia in venous blood we used photometric enzyme methods, and to determine HbA1C we used turbidimetric methods.

Results

For fasting glycemia and HbA1C in pregnant women in the seventh month of the last trimester, we get the following results :6.05 ± 2.66 mmol/L and 6.73±1.28 %. In the next eighth month, the average glycemia decreased 5.87±2.16 mmol/L, while the HbA1C level increased 6.86±0.96 %. In the last month of pregnancy before deliveri, the glycemia moves in the limits of this interval, t.e decreased 5.79 ±1.93 mmol/L. HbA1C was in the same range as the previous month between 6.82±0.69 %.

Conclusions

Dietary modification and increased physical activity are the primary treatments for GDM, but pharmacotherapy, usually insulin, is used when normoglycaemia is not achieved. Optimal management of mother and infant during long –term follow-up remains challenging with very limited implementation of preventive strategies in most part of the world.

Key words: pregnant women, gestational diabetes. complications. venous blood.

eP34

STEROID HORMONE DEHYDOEPIANDROSTERONE AFFECTS THE PRO-INFLAMMATORY PHENOTYPE OF HUMAN ADIPOCYTES AND MOUSE 3T3L1 ADIPOCYTES

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Introduction

Obesity is the result of excess fat storage that is accumulated in white hyperplastic adipocytes and the obese people are at risk of developing metabolic disorders with a common characteristic, the low grade-metabolic inflammation.

Dehydroepiandrosterone (DHEA) is a steroid hormone that is found in large quantities in the adipose tissue. In aging, the declining levels of DHEA are correlated with increased adiposity. Studies in rodents have shown that DHEA inhibits pre-adipocyte proliferation and affects the white adipogenesis process while, the role of DHEA in human white adipocytes physiology in not known.

Aim

The aim of this study was to investigate the role of DHEA in the accumulation of lipids and the inflammatory profile of human and mouse white adipocytes.

Methods

Visceral adipose tissues from human subjects undergoing general surgery were obtained and processed to isolate mature white adipocytes. Also, the mouse pre-adipocytes 3T3L1 were differentiated in vitro to white adipocytes. DHEA (10-8 M) was added at different time points in adipocytes in culture. Intracellular lipids accumulation was measured by Oil-Red-O staining and the levels of inflammatory markers by Luminex and ELISA. The mRNA levels of adiponectin were measured by qPCR.

Results

Human white mature adipocytes incubated with DHEA (10-8 M) for 30hrs showed decreased intracellular lipids accumulation. A reduction in IL1b secretion was observed at 24hrs while at the same time the mRNA levels of adiponectin were significantly increased. 3T3L1 cells differentiated in vitro and stimulated with DHEA for 3hrs showed reduced IL6 secretion. Incubation of 3T3L1 cells with DHEA during the differentiation period successfully inhibited the white adipogenesis process and upregulated the mRNA expression levels of adiponectin.

Conclusions

Our data show that DHEA was capable of inhibiting the white adipogenesis process and at the same time of increasing the mRNA levels of adiponectin that was followed by a less inflammatory profile of adipocytes. These results are confirming the beneficial role of DHEA in both human and mouse adipocytes.

Gastrointestinal diseases, including hepatic and pancreatic diseases

eP35

IS A CHANGE IN THE METHOD OF LIVER ENZYMES NECESSARY IN OUR COMMUNITY?

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Introduction

The recommended method for the analysis of liver enzymes (ALT/AST) is that supplemented with pyridoxal phosphate. Its importance lies in those patients who have a deficit of this enzyme since in the method without supplementation they will give us falsely diminished results.

Goal

To analyze if there are differences in the classification of patients based on the use of two different methods.

Material and methods

132 serum samples were analyzed in parallel for the determination of ALT and AST with and without pyridoxal phosphate, of which 60 were from the digestive service and 72 from other services. 50% of the patients were selected based on bio-

chemical parameters indicative of malnutrition and/or deficits, such as folic acid, transferrin and ZInC .

The samples were categorized according to the reference value in normal and pathological and the methods according to whether or not they were activated with pyridoxal phosphate. To demonstrate whether there was a relationship in the categorization of patients according to the analytical method used, the chi -square statistical test with Yates correction was applied, considering p <0.05 to be significant.

Results

No statistically significant differences were found neither in the total group of patients nor in the group of digestive patients.

Conclusion

Despite not being statistically comparable techniques, the pathological categorization of patients does not vary using one technique or another. And although false negatives have been described in the determination of transaminases with the test without pyridoxal phosphate in patients with a deficiency of the same, especially in 50% of severe alcoholics; In our study, in which we have included alcoholic patients, patients with malnutrition, etc. , we have indirectly verified that there is apparently no deficiency of pyridoxal phosphate in our environment that makes it necessary for us to make a change in the liver enzyme method.

Haematology, including haemostasis

eP36

A CASE REPORT OF PSEUDOTHROMBOCYTOPENIA DUE TO PLATELET CLUMPING

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Introduction

Pseudothrombocytopenia is a common laboratory phenomenon that results from an underestimation of the platelet count as measured by an automated cell counter.The most common mechanism is platelet clumping.Clumping is most often due to the anticoagulant EDTA and it has an incidence of aproximately 0.1%. in the general hospital population. Although automatic blood analyzers are widely used, peripheral blood smear is the hematological gold standard for definitive diagnosis.

Aim

The importance of recognizing EDTA induced pseudothrombocytopenia during the laboratory practice to avoid inappropriate diagnosis and treatment.

Methods: Venous blood samples were obtained in 3 different tubes containing EDTA,sodium citrate and heparin anticoagulant. A peripheral blood smear was prepared in each case to confirm the results of automated analyzer.

Results

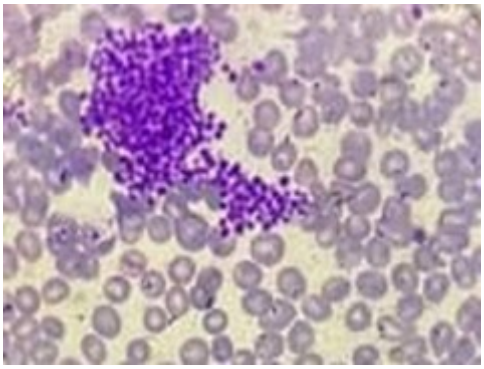
Case Report: A 65-year old man was referred to our clinic due to thrombocytopenia .The patient's platelet count was determined by a different laboratory to be 8 x 103 µL. The patient had no history of drug intake, blood trans-fusion, systemic disease or recent viral infection. Physical examination was normal. Laboratory findings were as follows: PLT: 13 x 103 µL ; Hb: 13.6 g/dL; WBC: 4.25 x 103 µL. A peripheral blood smear was prepared to confirm the results taken from the automated analyzer and clumping was observed. Repeat testing on a sample collected in sodium citrate showed a PLT count 60 x 103 µL and the peripheral smear clumping too.

EDTA-dependent pseudo thrombocytopenia was suspected and the clinician was advised to send a sample in a heparin

tube. Repeat testing of the new sample collected in heparin tube showed normal PLT of 220 x 103 µL.

Conclusion

Any case of thrombocytopenia found on an automated Cell-counter should be confirmed by examination of peripheral smear prior to further investigation or treatment.



eP37

IMPORTANCE OF BLOOD SMEAR EVALUATION IN THE DIAGNOSIS OF LEUKEMIA

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Introduction

Automated hematology analyzers provide reliable diagnostic information. However, microscopic evaluation of peripheral blood smear affords clinicians additional data that may be used to guide diagnosis and treatment.With the development of automated blood-cell analyzers, the proportion of bloodcount samples that require a blood smear has steadily diminished and in many clinical settings is now 15 percent or less.

Aim

Blood smear review remains an important tool in diagnosis of leukemia and should always be taken in consideration based on the information given from the hematology analyzers.

Methods

Venous blood sample was obtained from the patient in K3 EDTA. Peripheral blood smear was prepared to confirm the results of automated analyzer.

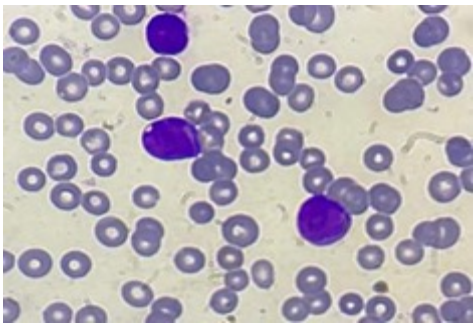
Results

Case report: A 77 years old woman came to our clinic for a routine checkup. She complained of fatigue, tiredness and fever for the last month to the physician. Physical examination revealed no significant abnormality apart from pallor. She had no history of hematological disorders. She referred only for a past urinary infection last month. Blood count was ordered, which showed leukocitosis, anemia and thrombocytopenia. The evaluation from the hematology analyzer showed a Hb value 9.7 g/dl ,Plt count 22.000/mm3 and WBC 57.62 10^3/µL .The hematology analyzer couldn't measure WBC differential count and reported a flag. A peripheral blood smear was prepared for further evaluation and the Wbc differential count indicated acute leukemia with results as follow: Segmented neutrophils 1%,Band neuthrophils 1%,Lymphocytes 14%, Basophils 2% and Blasts 82% (Fig1).

With these results the patient took an urgent recommendation to a hematologist in a tertiary hospital center for further evaluation and examinations.

Conclusion

Peripheral blood smear is essential in diagnosing leukemia or other hematologycal disorders and should always be taken in consideration despite the development of sophisticated blood cell analyzers.



eP38

SERUM ERYTHROFERRONE QUANTIFICATION IN BULGARIAN POPULATION

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Introduction – Aim

Erythroferrone was discovered in year 2014 as regulator of hepcidin synthesis and is synthesized by erythroblasts. Erythroferrone main function is to suppress hepcidin synthesis during inflammation, thereby regulates iron homeostasis. We aimed to establish erythroferrone reference interval for Bulgarian population using validated ELISA method.

Methods

170 healthy volunteers were included; intima-media thickness (IMT), ankle-brachium index (ABI), bodi-mass index (BMI) were calculated. Biological specimen (venous blood) was taken for evaluation of red blood cells count, hemoglobin, hematocrit, high sensitive CRP, serum iron and TIBC, ferritin, haptoglobin, hepcidin, TNF-α, and IL-6. Included volunteers were divided into two age borders – ?) from 18 to 50 years old and b) above age of 50.

Results

For calibration curve we use recombinant human ERFE with level 10 ng/mL, from which after proper dilutions the required clinically relevant values were established. Diagnostic sensitivity was 0.057 ng/ml. After applying specific statistical analyses and parametric distribution we found 6.9 – 16.1 ng/mL as reference range for Bulgarian population.

Conclusions

The immunological ELISA method we choose for serum erythroferrone quantification showed high specificity and sensitivity during validation process. It is new parameter for Bulgarian clinical laboratory practice and hematology specialty.

Acknowledgements

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eP39

THE NEED FOR EASY ACCESS TO A HEMATOLOGIST IN RURAL AREAS- A BIOPATHOLOGIST'S VIEW

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Introduction

Many chronic hematological disorders are eluding correct hematological diagnosis and treatment due to the difficulty of patient access to hematology clinics in rural areas.

Background

Early diagnosis of hematological disorders is a challenge that most biopathology laboratories face in every day practice, especially in rural areas. Whereas acute hematological disorders are usually followed by overwhelming clinical manifestations and laboratory findings which lead to a fast diagnosis and an even faster transfer to a hematology clinic, chronic, initially non-threatening, hematological diseases present an every day riddle, especially for elderly patients whose quality of life is affected consequently. The slow development of diseases such as CLL or myelodysplastic syndromes, even in the presence of strong indications that dictate referral to a hematologist, allows the patients to form their own decisions, often against visiting the recommended specialist, and much more so when it is necessary to travel to some urban center for this purpose. This negligence often turns out to be a burden on their health, thus jeopardizing the therapeutical outcome, when they finally seek help. Unfortunately, the access to a hematologist can be difficult for patients living in rural areas and very often it leads to an underdiagnosis of chronic conditions.

Conclusion

The recognition of the incidence of chronic hematological diseases requires the facilitation of patient access to a hematologist, especially in areas outside urban centers. The biopathologist can play a significant role to the early diagnosis of these diseases but the hematologist is the medical expert who makes the final diagnosis and offers essential and targeted health care. Therefore, care should be taken for easy access to a hematologist, especially for residents of non-urban areas.

eP40

COLD AGGLUTININ DISEASE DIAGNOSED BY HAEMOGRAM ALTERATIONS

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Introduction

Cold agglutinin syndrome is a rare form of autoimmune hemolytic anemia (AIHA), in which red blood cells lysis is mediated by IgM-type immunoglobulins directed against polysaccharide antigens on the red blood cells membrane. This hemolysis occurs at temperatures below 37 °C.

The presentation can be idiopathic or secondary to infections or neoplasms.

In some cases, the diagnosis is made by chance when performing a complete blood count, by detecting an abnormal aggregation of red blood cells.

Aim

The case of a patient with an altered blood count due to the presence of these immunoglobulins is presented.

Methods And Results: A 75-year-old patient came to the emergency room due to fever measured at 39°C with a dysthermic sensation, in addition to intense asthenia and hyporexia.

In the emergency analysis, severe anemia (5.9 g/dl) and an abnormal number of red blood cells (190,000/µL, NV: 4 x106 / µL) standed out. The analyzer alerted interference in all the parameters, highlighting a corpuscular hemoglobin concentration (MCHC) value greater than the measurement range. A smear review was performed, with presence of agglutinated red blood cells.

With high suspicion of the presence of cold agglutinins, it is decided to heat the sample to 37 °C and process quickly. With this, the interferences dissapeared and more reliable results, although pathological, were achieved.

The suspicion of AIHA due to cold agglutinins, further supported by findings of elevated indirect bilirubin and LDH, was confirmed by Coombs and antiC3d tests. In addition, the proteinogram and subsequent serum immunofixation confirmed the presence of IgM-type immunoglobulins.

Although the patient presented fever on admission, bacterial cultures resulted negative, classifying the AIHA as idiopathic.

After treatment with heater transfusions and Rituximab, the patient improved, progressively increasing her Hemoglobin.

Conclusions

The analyzers (DxH 900, Beckman Coulter) classify and count blood cells based on the principle of impedance, which depends on the size of these cells.

In AIHAs due to cold agglutinins, the binding of the IgM antibody to the red blood cell at low temperatures forms large aggregates that interfere with the analyzer channels, giving rise to incorrect results in both cell counting and classification.

eP41

BICLONAL MULTIPLE MYELOMA

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Monoclonal gammapathies are disorders caused due to the proliferation of a clone of B cells that produces a monoclonal immunoglobulin, giving rise to the appearance of a paraprotein or monoclonal component (MC). Within this group of pathologies we find multiple myeloma (MM), Waldenström's macroglobulinemia and amyloidosis.

Multiple Myeloma accounts for approximately 10-13% of all hematological cancers, with an incidence of 4-7 cases per 100,000 inhabitants.

Some of the signs that may lead to suspect the existence of MM are: bone pain, anemia, hypercalcaemia, presence of MC in serum or urine, recurrent infections or kidney failure.

The most indicated laboratory tests for MM screening are a complete blood count, electrophoresis of serum and concentrated urine, quantification of immunoglobulins and determination of creatinine, calcium, albumin and total proteins.

Aim

In this work we present a case of biclonal multiple myeloma, characterized by the presence of two MC in serum, whose incidence is only 2% of all multiple myelomas.

Methods And Results

An 87-year-old patient who went to the emergency room due to general syndrome characterized by bone pain, decreased intake and difficulty urinating.

In the emergency analysis, renal failure (Creatinine 2.37 mg/dl) and hypercalcaemia (15.4 mg/dl), as well as anemia (Hb 9.8 g/dl) are highlighted. The patient was admitted to Internal Medicine with a diagnosis of exacerbated CKD and anemia. In view of the results, a proteinogram was extended from the laboratory due to high suspicion of Multiple Myeloma. In it, two possible monoclonal peaks were visualized in the β2 and γ regions. To confirm, both immutipation and immufixation were performed, where an IgA Lambda monoclonal band was identified in the β2 region as well as IgG Kappa in the γ region. Therefore, the patient was finally admitted to Hematology with a diagnosis of biclonal multiple myeloma.

Ultimately, the patient died due to severe hypercalcaemia and the rapid progression of the disease.

Conclusion

Biclonal MM are rare and characterized by the simultaneous appearance of 2 types of MC, although the clinical manifestations are similar to other types of MM. The most frequent is founding a combination of IgA/IgG, as in our case (53%) . It is not clear whether these two components originate from a single clone or from two clones of B cells.

ePP42

SPURIOUS INCREASED MCHC AS ANALYTICAL ALARM TO ENSURE DELIVERY OF THE RIGHT RESULTS TO THE CLINICIANS

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Introduction

Hemogram abnormality related to one or more of the measured parameters red blood cells (RBC), hemoglobin (HGB) or hematocrit (HCT) leads to abnormal calculated RBC indices, especially mean corpuscular hemoglobin concentration (MCHC), which is one of the most indicative abnormalities generated by hematology analyzers, alarming the users about a spurious result. Different etiologies lead to spurious results of RBC, HCT or HGB measurements like as : cold agglutination (CA) or RBC disease, the presence of abnormal plasma: as lipemic, icteric or hemolytic situation.

The aim

It is to evaluate whether the increase of MCHC is an artifact or a pathological situation

Methodology: Case-control study, the proportion expected from the exposure factor considered, which is the increase of MCHC > 37.0. As well as the change that the hemograma undergoes after the replacement of plasma, that is, after we have removed the interfering factor by obtaining normal values ??of MCHC. In the study, probabilistic samples were selected referring to the increase of MCHC and not to another systematic selection index.

Results

It can be observed that, after the normalization of MCHC, the average values ??of all laboratory parameters, with the exception of WBC, have changed in a statistically significant way. For Lymphocyte, RBC, HCT, MCV and PLT parameters the mean value after MCHC normalization is higher than the corresponding mean value before MCHC normalization; this means that for these parameters, the adjustment of MCHC leads to their statistically significant (fictitious) decrease compared to their real value. The opposite was evident for the laboratory parameters Mono, Hemoglobin and MCH, whose mean value after MCHC normalization is lower than the corresponding mean value before MCHC normalization; this means that for these parameters, the adjustment of MCHC leads to their statistically significant (fictitious) increase compared to their real value.

Conclusions

In daily practice in hematology laboratories, spurious increased MCHC should be induces an analytical alarm and needs prompt corrective action to ensure delivery of the right results to the clinicians.

Haematology, including haemostasis

eP43

THE IMPACT OF URIC ACID AND PLASMA TOTAL ANTIOXIDANT CAPACITY ON DEVELOPMENT OF AGE –RELATED MACULAR DEGENERATION

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Background

Recent epidemiological data demonstrated that altered uric acid metabolism could play an important role in the pathogensis of age-related macular degeneration (AMD). The aim of this study was to analyze the plasma concentration of uric acid and total antioxidant status (TAS) in order to find the possible association of these parameters with occurence of AMD, and to assess whether these parameters could serve as potential predictors for AMD.

Material and methods: A cross-sectional study included 110 AMD patients aged 71.3±6.3 yr., classified into three categories: early AMD, advanced-dry (geographic athrophy) and advanced-wet AMD (choroidal neovascularization) and 85 age matched control subjects.

Results

Significantly lower TAS ($p<0.001$) and uric acid values ($p=0.006$) were obtained in the whole group of AMD patients compared to the controls. TAS and uric acid were much more reduced in the group of advanced-wet AMD compared to early AMD and to the controls ($p<0.05$). Lower values of tested parameters were obtained in female subgoup compared to males in both tested groups, as well as in the subgroup of hypertensive and diabetic AMD patients compared to appropriate subgroup of the controls ($p<0.001$). Significant association of tested parameters was found with occurence of AMD ($p<0.05$). According to ROC analysis TAS concentration lower than 1.25 mmol/L have sufficient predictive ability for AMD (OR:193.2; 95% CI: 27.7–1349.1; $P<0.000$).

Conclusion

Significantly reduced plasma total antioxidant status and uric acid concentration found in AMD, could have an important role in development of increase oxidative stress and occurence of AMD.

eP44

CLINICAL-LABORATORY CONSTELLATIONS IN SEPTIC COMPLICATIONS AFTER VASCULAR RECONSTRUCTIONS

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Introduction

The first report of the use of synthetic material for reconstructive vascular surgery was in 1950. Since then, infection of the vascular grafts, although not frequent, remains a serious complication associated in some situations with a fatal outcome. Septic complications in vascular surgery are accompanied by complex pathophysiological mechanisms, which are clinically established through laboratory studies. The clinical-biochemical results confirm to a large extent the septic complications after vascular reconstructions, therefore timely accurate assessment, and interpretation of the values ??especially of C-reactive protein and IgA in patients with a history of past vascular interventions is important.

Aim

The aim of our study is the discovery of potential biomarkers in septic complications in vascular surgery.

Methods

We present four clinical cases of patients after vascular reconstruction in the aorto-iliac segment, admitted in clinic of vascular surgery of the Military Medical Academy due to graft infection and their biochemical results. The included patients were on average age of 68.7 ± 4.5 years; 75% males.

Results

In all four cases, an increase in immunoglobulin A (aver 20.6 g/L ± 4.2 g/L) and C-reactive protein (74.5 mg/L ± 48.7 mg/L) was observed. A strong positive correlation was found between Immunoglobulin A and CRP results ($r=0.846$, $P<0.05$). Serum iron concentrations were decreased (3.8 μmol/L ± 0.95 μmol/L, which correlates positively to CRP levels ($r=0.644$, $P<0.05$).

Conclusions

Infection involving the vascular prosthesis is difficult to treat. If not diagnosed and treated in time, it leads to catastrophic consequences because of sepsis, haemorrhage, or thrombosis with ischemia. Clinical biochemical tests are extremely important for timely diagnosis and correct complex treatment.

Key words

septic complication, clinical-biochemical results, vascular reconstruction, potential biomarkers.

eP45

HAPTOGLOBIN PHENOTYPE IN BULGARIAN POPULATION CAUSES ATHEROSCLEROTIC CHANGES IN HUMAN ORGANISM

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Introduction - Aim

Human gene of haptoglobin is presented by two alleles. Haptoglobin types are 1-1, 1-2 and 2-2. Different studies show role of type 2-2 in cardio-vascular disease occurrence during diabetes. Haptoglobin type 1 is known to suppress hemoglobin-based oxygenation of HDL and LDL, acting like antioxidant.

Methods: According to a previous trial, Bulgarian population is with haptoglobin 2-2 type, which causes frequent morbidity by systematic diseases, such as atherosclerosis, diabetes, diabetic nephropathies, gestational diabetes, anemia, etc. 78 volunteers were included, age 29.7 ± 5.4. In all included volunteers IMT, ABI, CBC, iron homeostasis, hsCRP and haptoglobin phenotype was performed.

Results

Elevated serum hepcidin concentrations were found in patients with atherosclerotic a. carotis evidences (105.4 ± 9.9 μg/L) compared to healthy volunteers (20.1 ± 2.2 μg/L), $P<0.005$. In population with haptoglobin phenotype 2-2 we found strong positive correlation between serum hepcidin concentration and impared IMT and ABI parameters ($r=0.855$, $r=0.821$, resp.; $P<0.01$). In nine volunteers were found haptoglobin phenotype 2-1 which was presented with no serum hepcidin levels and no atherosclerotic IMT and ABI changes.

Conclusions

The main reason for acute coronary thrombosis is atherosclerotic plaque rupture. Extra-vascular hemoglobin plays role as start mechanism for inflammation in the plaques. Important contra-active mechanism is played by haptoglobin. Thus, it prevents kidney injury from free hemoglobin. Hepcidin regulates iron homeostasis by its interaction with intracellular iron exporter ferroportin.

Acknowledgements

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eP46

EFFECTS OF DIESEL EXHAUST FUMES EXPOSURE ON ARTERIAL WALL PROPERTIES, INFLAMMATORY PROCESS AND FIBROSIS-FIBRINOLYSIS STATUS

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Introduction

Exhaust fumes from diesel engines are a complex mixture of toxic compounds with a wide variety of harmful effects. The acute effects of diesel exhaust fumes on the cardiovascular system are well-known. However, their short-term impact has not been thoroughly studied.

Aim

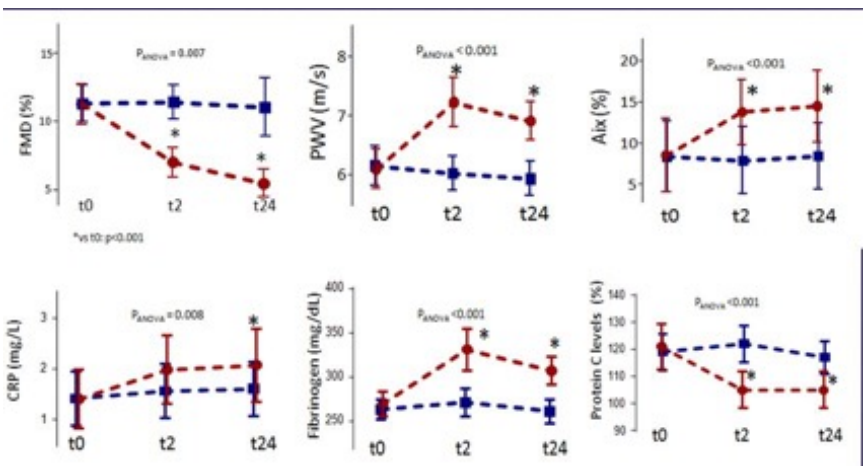
To study the acute and short-term (24 h) effects of diesel exhaust particles (DEPs) on endothelial function, arterial wall properties, inflammatory process and fibrosis-fibrinolysis status

Methods

In this blind cross over study, 40 healthy volunteers (median age 40 years old) have been exposed to diesel exhaust particles (DEPs) and then to filtered air (FA) over a 2-hour session with a wash out period of 4 weeks. Exposure to DEPs was calibrated based on the mass of microparticles less than 2,5 microns in diameter (PM 2,5). Flow-mediated-dilation (FMD) was used to estimate endothelial function. Pulse wave velocity (PWV) and augmentation index (AIx) assessed central aortic stiffness and arterial reflected waves respectively. C reactive protein (CRP) was measured to determine the inflammatory status, as well as fibrinogen and protein C levels to evaluate the impact on the coagulation cascade. All measurements were performed before each session (T0), at the end of the 2 hours exposure session (T2) and 24 hours after completion of each session (T24). Variables with normal distribution are presented as mean±SD otherwise as median±SEM.

Results

At T0 of DEP and FA exposure there was no significant difference in FMD, PWV, AIX, CRP, protein C and fibrinogen levels. Exposure to DEP decreased significantly FMD (T0:11.97±4.61%,T2:7.71±3.36%, T24:6.17±3.19%, p<0.001) and increased PWV (T0:6.09±1.03m/sec, T2: 7.22±1.31m/sec, T24: 6.90±1.03m/sec, p<0.001), AIx (T0:8.17±3.19%, T2: 12.71±3.36%, T24: 13.17±4.61%, p<0.001), CRP (T0:1.41±0.18 mg/L, T2: 1.99±0.21mg/L, T24: 2.08±0.24mg/L, p=0.04) and fibrinogen levels (T0:269±44 mg/



dL, T2: 331±75 mg/dL, T24: 307±51 mg/dL, p=0.002). Protein C was significantly reduced (T0:121±26%, T2: 104±21%, T24: 105±20%,p=0.003). Exposure to FA had no significant impact on the study parameters.

Conclusions

Exposure to diesel exhaust fumes may have significant adverse effects on the cardiovascular system with impairment of endothelial function, arterial wall properties, inflammatory status and fibrosis-fibrinolysis parameters not only during the exposure period but as far as 24 hours after exposure.

Kidney diseases

eP47

SERUM CYSTATIN C AND URINARY BIOMARKERS IN THE EVALUATION OF CHILDHOOD CHRONIC KIDNEY DISEASE

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Introduction

Chronic kidney disease is a modern epidemic of developed society today in the world. In recent years, this disease has become a serious problem in childhood, because the incidence of children with terminal kidney disease has increased

Aim: To evaluate the value of serum cystatin C and urinary biomarkers (b2 microglobulin and urinary neutrophil gelatinase-associated lipocalin (NGAL) in early prediction of chronic kidney disease.

Methods

It is a retrospective-prospective study, in which 70 patients will be evaluated in the period from January 1, 2020 to June 30, 2022, who came to the University Clinic for Pediatric Diseases, with clinical signs, symptoms, laboratory tests, and imaging studies for chronic kidney disease.The examined group will be divided into three groups: group I will include patients with congenital anomalies of the kidneys and urinary tract, group II will include patients with tubulopathies and metabolic diseases with renal affection, group III will include patients with glomerulopathies.

Results:, The study showed female predominance in three of groups (71% vs 54%) and with a mean age of 7.37±2.21 years. Serum cystatin C were statistically significantly increased in patients in group I with congenital anomalies of the kidneys and urinary tract ($p < 0.001$). A statistically significant positive correlation was found between serum cystatin C in correlation with urinary NGAL($p < 0.001$) in the three groups , as well as between serum cystatin C in correlation with b2 microglobuline ($p < 0.001$) in the three groups.

Conclusion

Serum cystatin C is a sensitive and predictive marker for early prediction of chronic kidney disease

Keywords

Serum cystatin, Neutrophil gelatinous associated lipocalin (NGAL), b2 microglobulin, chronic kidney disease

eP48

QUALITY INDICATORS AND SIX-SIGMA PERFORMANCE OF THE ANALYTICAL PHASE IN A BIOCHEMISTRY LABORATORY

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Introduction

Quality indicators (QIs) are important tool for the monitoring and evaluation of laboratory performance. The aim of our study was to evaluate QIs of the analytical phase in a Biochemistry laboratory.

Materials and Methods

The relevant data was collected from January 2017 to December 2021. Data recording was performed on daily basis. Six sigma performance was evaluated by the Westgard calculator. The QIs are proposed by IFCC and characterized as Key process Indicators Priority 1 (Mandatory).

- Intra-IQC Percentage of: Number of tests without IQC /Total number of tests in the menu.
- Intra-UnIQC Percentage of: Number of IQC results outside defined limits / Total number of IQC results
- Intra-EQA Percentage of: Number of tests without EQA-PT control / Total number of tests in the menu
- Intra-Unac Percentage of: Number of unacceptable performances in EQAS-PT Schemes, per year/ Total number of performances in EQASchemes, per year.

Results

The results are presented in the following tables:

Conclusion

Intra-UnIQC presents a satisfactory performance level. The same is observed about Intra-EQA, except from 2019, when many equipment failures were detected. Equipment was eventually replaced. Intra-Unac and Intra-IQC show a gradual improvement through the years. For the 2 tests remaning without IQC and EQA-PT control, alternative control methods are applied until the problem can be solved. Monitoring and evaluation of QIs can identify problematic area in the processes and helps improving the quality and effectiveness of laboratory testing.

QI	YEAR	NUMBER OF UNACCEPTABLE PERFORMANCES	TOTAL NUMBER OF PERFORMANCES	DPMO	SIX SIGMA
Intra-UnIQC	2017	52	23775	2187	4,4
	2018	43	23775	1809	4,5
	2019	31	23775	1304	4,6
	2020	32	23775	1346	4,6
	2021	18	23775	757	4,7
Intra-EQA	2017	3	2040	1471	4,5
	2018	3	2040	1471	4,5
	2019	29	2040	14216	3,7
	2020	4	2060	1942	4,4
	2021	4	2060	1942	4,4

QI	YEAR	TESTS WITHOUT IQC OR EQA	TOTAL MUMBER OF TESTS IN THE MENU	%
Intra-Unac	2017	8	62	12,90
	2018	8	62	12,90
	2019	8	62	12,90
	2020	2	70	2,86
	2021	2	70	2,86
Intra-IQC	2017	3	62	3,00
	2018	3	62	3,00
	2019	3	62	3,00
	2020	2	70	2,00
	2021	2	70	2,00

eP49

DIFFERENCE IN THE DETERMINATION OF HAEMOGLOBIN AS AN INTERNAL QUALITY CONTROL IN BLOOD GAS ANALYSIS

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Introduction

Shaking of the sample prior to blood gas analysis is a common source of pre-analytical error that can affect the results. In our centre we detected irregular processing by staff prior to analysis after a laboratory refurbishment.

Aim: Our work aims to analyse the usefulness of the differences between the haemoglobin of blood gases and the haemoglobin of the haematological counter in the same analytical request:

1.- As a tool for detecting pre-analytical error in blood gas analysis and the need for staff training/updating.

2.- As a follow-up of the training efforts applied.

Methods

Haemoglobin values were extracted between 07/2018 (the period in which the emergency laboratory staff were renewed) and 12/2021 in those requests that included simultaneously, in the same extraction and request, the determination of haemoglobin (in haemoglobin in EDTA sample) by a Beckman CoulterDxH800 analyser and haemoglobin in venous blood gases by a Radiometer ABL 800Flex blood gas analyser. Absolute differences between the two values were calculated and the monthly and half-yearly arithmetic mean was calculated. A regression line was calculated with the values obtained monthly.

Results

The values obtained were for the first semester: 0.739, second semester: 0.669, third semester: 0.548, fourth semester: 0.509, fifth semester: 0.479, sixth semester: 0.400. The regression line showed the following equation: y=0.78 - 0.0147X (R2=0.799).

The results obtained showed a downward trend from the beginning of the intervention, showing better results in the first year of the intervention, detecting at present, although in a milder form, a progressive improvement.

Conclusions

Differences in simultaneously determined haemoglobin values are a good indicator for sample homogenisation quality. They serve as a control of staff training and as an internal quality control in laboratories where it is possible to perform simultaneous haemoglobin determination by another method on a sample from the same extraction, as in the case of a haemogram.

eP50

ASSESSING SIX SIGMA SCORE OF BIO-RAD HbA1c PROGRAM BASED ON DIFFERENT TOTAL ALLOWABLE ERROR FROM CLIA AND NGSP

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Introduction

HbA1c is an important biomarker, needed to evaluate long term outcomes of diabetes and analytical reliability of HbA1c should be obtained by clinical laboratories. Reliability is obtained by Internal Quality Control (IQC) and External Quality Control (EQC) using analysis of data using statistical methods. As standardisation of method is either done based on CLIA, NGSP or DCCT guidelines, we have calculated our six sigma scores based on TEa provided by CLIA and NGSP guidelines

Aim

To determine Six sigma score of BioRad D-10 HbA1c analyzer using different Total Allowable error (TEa) taken from CLIA and NGSP over a period of 5 months

Methods: Mean and SD was calculated using SPSS. CV, Coefficient of Variation was determined from calculated laboratory mean and calculated standard deviation, obtained from 5 months of IQC and RIQAS data, TEa taken from CLIA and NGSP guidelines. Sigma metrics for each parameter was calculated

Result

Six Sigma score was calculated for both levels of QC and RIQAS with TEa 10 and 6 from CLIA and NGSP guidelines respectively which can be seen in table below. On operator specific chart plotted for TEa 6%, most of our result were between 2-3 sigma while plotting in TEa 10%, our results were between 5-4 sigma.

Conclusions

Our BIORAD D-10 from above data seems to have an acceptable mean sigma level at both TEa of 10% (CLIA) and 6% (NGSP). We plan to implement westgard rejection rules based on our six sigma results for TEa 6% so that our results will bring more confidence in monitoring of Diabetes Mellitus.

	November 2020	December 2020	January 2021	February 2021	March 2021
TEa (CLIA-88)	10	10	10	10	10
Sigma (CLIA-88)	6.2	4.8	5.7	4.4	6
TEa (NGSP)	6	6	6	6	6
Sigma (NGSP)	3.3	2.2	3.2	2.6	3

Microbiology - Infectious diseases including COVID 19

eP51

WATERBORNE OUTBREAKS IN THE MEDITERRANEAN REGION

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Introduction

Each year, waterborne diseases affect hundreds of millions of people, primarily those living without safe, accessible water. The purpose of this study is the presentation of waterborne outbreaks recorded in the Mediterranean countries between 2008 and 2021.

Materials and Methods

We performed an extensive bibliographic review in Pubmed, Embase, Google Scholar, ECDC and WHO. Information was not available for all countries, eg Libya, Tunisia, Morocco and Malta, probably due to the differences in the epidemiological surveillance systems between countries. Waterborne outbreaks we categorized according to the country and to the transmission mode (drinking water, bottled water, insects hatching or living near water collections, water used in health centers, recreational water and consumption of food treated with contaminated water).

Results

The results are presented in the following Table.

Discussion

Waterborne outbreaks are observed in all Mediterranean countries, due to all types of pathogens (bacteria, viruses, helminths and protozoa). The increase in their incidence observed in the last years is attributed to water supply and sewerage systems aging, to socio-economic and demographic factors, such as overpopulation, population aging, increase in immunocompromised individuals, lifestyle changes, and to climate change and extreme weather phenomena. In addition, during the recent years there have been significant humanitarian crises in the region as a result of the ongoing civil and war conflicts, with mass population displacement, food insecurity, lack of access to health infrastructure, and immigration. Harmonization of the surveillance systems will contribute to a better evaluation of the outbreaks.

COUNTRY	REGION	YEAR	DISEASE	WATER TYPE	PATHOGEN	MODE OF TRANSMISSION
ISRAEL	NORTH ISRAEL	2018	LEPTOSPIROSIS	RECREATIONL WATER (KAYAKING AND RAFTING)	LEPTOSPIRA	CONTAMINATED WATER BY ANIMAL URINE
TURKEY	HAMAMKOY-IZMIR	2014	TULAREMIA	DRINKING WATER	Francisella tularensis	FROM ENVIRONMENTAL RESERVOIR
	Keçiborlu province	2010	ACUTE GASTROENTERITIS	DRINKING WATER	Norovirus	CONTAMINATED WATER SUPPLY SYSTEM
	NIGDE CITY	2014	ACUTE GASTROENTERITIS	DRINKING WATER	Rotavirus	CONTAMINATED WATER SUPPLY SYSTEM
	SAMSUN PROVINCE	2012	ACUTE GASTROENTERITIS	DRINKING WATER	Shigella sonnei	CONTAMINATED WATER SUPPLY SYSTEM
	ANATOLIA	2011	PHARYNGITIS/RESPIRATORY SYSTEM INFECTION	DRINKING WATER	Francisella tularensis	CONSUMPTION OF WATER FROM UNDERGROUND SOURCE
FRANCE	Pérignat-lès-Sarliève	2010	ACUTE GASTROENTERITIS	DRINKING WATER	Campylobacter jejuni	FECAL CONTAMINATION OF WATERS SUPPLY SYSTEM
	Pleaux	2012	ACUTE GASTROENTERITIS	DRINKING WATER	Norovirus	FECAL CONTAMINATION OF WATERS SUPPLY SYSTEM
	SOUTHWEST FRANCE	2014	ACUTE GASTROENTERITIS	DRINKING WATER	ENTEROCOCCI AND COLIFORMS	CONTAMINATED WATER SUPPLY SYSTEM BY SEWAGE
	MONTPELLIER	2014	Chikungunya Fever	HEAVYRAINFALL WATER	Chikungunya Virus	Aedes albopictus
	BRETAGNE	2015	ACUTE GASTROENTERITIS	SEA FOOD CONSUMPTION	Norovirus	SEAFOOD CULTURE IN CONTAMINATED WATER
	SOUTHWEST FRANCE	2016	ACUTE GASTROENTERITIS	DRINKING WATER	ENTEROVIRUS	
	VILAINE	2016	LEPTOSPIROSIS	RECREATIONAL WATER (RAFTING AND KAYAK)	Leptospira kirschneri	CONTAMINATED WATER BY ANIMAL URINE
	SOUTHEAST FRANCE	2017	Chikungunya Fever	RAINWATER COLLECTION TANK	Chikungunya Virus	Aedes albopictus
	SOUTHWEST FRANCE	2017	ACUTE GASTROENTERITIS	DRINKING WATER	Shigella sonnei (Multiresistant strain)	
	Provence-Alpes-Côte d'Azur	2018	DENGUE FEVER	HEAVYRAINFALL WATER	Dengue Virus	Aedes spp
ITALY	ROME	2009-2010	DERMATOLOGICAL INFECTION	RECREATIONL WATER (SWIMMING POOL)	Mycobacterium chelonae complex	
		2011	ACUTE GASTROENTERITIS	DRINKING WATER	Salmonella napoli	WATER SUPPLY SYSTEM MAINTENANCE
LEBANON	Becharreh	2010	ACUTE GASTROENTERITIS	DRINKING WATER	ENTEROCOCCI AND COLIFORMS	WATER SUPPLY SYSTEM MAINTENANCE
SLOBENIA	Mengeš	2010	ACUTE GASTROENTERITIS	DRINKING WATER	NOROVIRUS KAI ROTAVIRUS	CONTAMINATED WATER SUPPLY SYSTEM BY SEWAGE
MONTENEGRO	Podgorica	2008	ACUTE GASTROENTERITIS	DRINKING WATER	NOROVIRUS	CONTAMINATED WATER SUPPLY SYSTEM BY SEWAGE
CROATIA	Šibenik	2014	ACUTE GASTROENTERITIS	DRINKINGWATER	Salmonella enterica subsp. enterica serovar Enteritidis	WATER SUPPLY SYSTEM FAILURE
GREECE	PELOPONESE	2009	ACUTE GASTROENTERITIS	DRINKING WATER	Salmonella spp	WATER SUPPLY SYSTEM FAILURE
	CRETE	2009		DRINKING WATER	Campylobacter jejuni	
	THESSALY	2010		DRINKING WATER	NOROVIRUS	
	NORTH AEGEAN	2010		SEA-FOOD CONSUMPTION	NOROVIRUS	SEAFOOD CULTURE IN CONTAMINATED WATER
	SOUTH AEGEAN	2010		DRINKING WATER	NOROVIRUS	WATER SUPPLY SYSTEM FAILURE
	ATTICA	2011		DRINKING WATER	NOROVIRUS	
	ATTICA	2012		DRINKING WATER	NOROVIRUS	
	MACEDONIA	2012		DRINKING WATER	NOROVIRUS	
	MACEDONIA	2012		DRINKING WATER	NOROVIRUS	
	THESSALY	2012		DRINKING WATER	NOROVIRUS	
	MACEDONIA	2015		DRINKING WATER	NOROVIRUS	
	SOUTH GREECE	2018		DRINKING WATER	UNKNOWN	
	NOTHERN GREECE	2019		DRINKING WATER	E.coli (EHEC)	
	PELOPONESE	2020		DRINKING WATER	E.coli (STEC AND O157)	
ALGERIA	TIRAZA	2018	CHOLERA	DRINKING WATER	Vibrio cholerae Ogawa	
CYPRUS	NIKOSIA	2008	LEGIONELLOSIS	HUMIDIFIER WATER IN PEDIATRIC HOSPITAL	Legionella pneumophila	Aerosol produced by cold-mist humidifier that was filled with contaminated tap water
SPAIN	Gipuzkoa	2008	PHARYNGITIS	RECREATIONAL WATER	adenovirus type 4	
	Valencia	2011	LEGIONELLOSIS	RECREATIONAL WATER	Legionella pneumophila	BIOFILM FORMATION IN WATER SUPPLY SYSTEM
	CATALUNA	2016	ACUTE GASTROENTERITIS	BOTTLED WATER	Norovirus	
	CATALUNA	2018	DENGUE FEVER		Dengue Virus	Aedes spp
	ISLAS CANARIAS	2018	PNEUMONIA	WATER IN HEALTH CLINIC	Klebsiella pneumoniae ST392	

eP52

SARS-CoV-2 MODULATES INFLAMMATORY RESPONSES OF ALVEOLAR EPITHELIAL TYPE II CELLS VIA PI3K/AKT PATHWAY

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Introduction

SARS-CoV-2 infects through the respiratory route and triggers inflammatory response by affecting multiple cell types including type II alveolar epithelial cells. SARS-CoV-2 triggers signals via its Spike (S) protein, which have been shown to participate in the pathogenesis of COVID19.

Aim

In the present study we investigated the effect of SARS-CoV2 on type II alveolar epithelial cells, focusing on signals initiated by its S protein and their impact on the expression of inflammatory mediators.

Results

The results showed that SARS-CoV-2 S protein decreased the expression and secretion of IL8, IL6 and TNFα, 6 hours following stimulation, while it had no effect on IFNα, CXCL5 and PAI-1 expression. We further examined whether SARS-CoV-2 S protein, when combined with TLR2 signals, which are also triggered by SARS-CoV2 and its envelope protein, exerts a different effect in type II alveolar epithelial cells. Simultaneous treatment of A549 cells with SARS-CoV-2 S protein and the TLR2 ligand PAM3csk4 decreased secretion of IL8, IL6 and TNFα, while it significantly increased IFNα, CXCL5 and PAI-1 expression. To investigate the molecular pathway through which SARS-CoV-2 S protein exerts this immunomodulatory action in alveolar epithelial cells, we measured the induction of MAPK/ERK and PI3K/AKT pathways and found that SARS-CoV-2 S protein induced the activation of the serine threonine kinase AKT. Treatment with the Akt inhibitor MK-2206, abolished the inhibitory effect of SARS-CoV-2 S protein on IL8, IL6 and TNFα expression, suggesting that SARS-CoV-2 S protein mediated its action via AKT kinases.

Conclusions

The findings of our study, showed that SARS-CoV-2 S protein suppressed inflammatory responses in alveolar epithelial type II cells at early stages of infection through activation of the PI3K/AKT pathway. Thus, our results suggest that at early stages SARS-CoV-2 S protein signals inhibit immune responses to the virus allowing it to propagate the infection while in combination with TLR2 signals enhanced PAI-1 expression, potentially affecting the local coagulation cascade.

eP53

BACTERIURIA DUE TO S. AGALACTIAE IN A ROMA DRUG AND ALCOHOL USER- A CASE REPORT

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Introduction

S. agalactiae,a common commensal of the human gastrointestinal and genitourinary tracts,causes a broad range of infectious diseases in neonates,pregnant women,elderly and immunocompromised patients.Moreover,S. agalactiae is responsible for approximately 2–3% of all urinary tract infections(UTIs),including asymptomatic bacteriuria,cystitis and pyelonephritis[1].

Aim

We present a case of S.agalactiae bacteriuria in a Roma chronic drug and alcohol abuser.

Case report

A 55-year-old Roma male patient was admitted to ER our hospital complaining of diffuse abdominal pain.No symptoms related to the urinary tract were present.The patient was a chronic alcohol and drug user and reported a history of stomach rupture.The laboratory test ordered revealed the following:1)CBC WBC:8710/μL (NEU 77,1%, LYMPH 14,4%),Hg 14,2gr/dL,Ht 42,7%(Celldyn Ruby,Abbott)2)Clinical Chemistry test:GLU:86 mg/dL,UREA: 44 mg/dL,CREA: 2,11 mg/dL,K: 4,2 mmol/L,Na:136 mmol/L,SGOT: 16 U/L,SGPT:11 U/L,γGT: 27 U/L,ALP:74 U/L,AMY: 122.7U/L,TBIL: 0.41mg/dL,DBIL: 0,07mg/dL,CRP: 3.68 mg/dL(Beckman Coulter AU680,Leriva).3)Urinalysis:There were no pathological findings in the chemical analysis(iChem Velocity Urinalysis Analyzer, Leriva)and the sediment microscopy revealed the absence of pyuria and the presence of a few cocci.The urine culture was performed using conventional methods.After a 48-hour incubation,a Gram(+)positive,beta-haemolytic coccus was grown on blood agar(>105cfu/mL). The microorganism was identified as Streptococcus agalactiae(Microscan Autoscan,Siemens,Microscan ID:98.54%)and found susceptible to all the antibiotics tested according to the EUCAST criteria.

Conclusions

Alcoholism has been described as a predisposing factor for S.agalactiae infections,even in immunocompetent patients[2,3].In our case,although the characteristic clinical symptomatology of UTI was absent,the S.agalactiae bacteriuria we found might be considered relevant to the patient's history.

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eP54

THE ADDED VALUE OF THE LABORATORY SPECIALIST IN THE ANALYSIS OF BIOLOGICAL FLUIDS

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Introduction

The analysis of biological fluids is essential in clinical practice. The reference method for cell counting is the haemocytometric chamber (Neubauer/Fuchs-Rosenthal). There is a growing trend to incorporate cytometric autoanalysers, not without limitations, to reduce the workload in the laboratory.

Aim

To demonstrate the added value of optical microscopic visualisation by a laboratory specialist.

Methods

A 55-year-old man, bipulmonary transplant recipient due to familial pulmonary fibrosis (FOPD) and emphysema, attended the emergency department for dyspnoea and pleuropericardial effusion under study. In that moment he patient was receiving prophylactic antibacterial treatment.

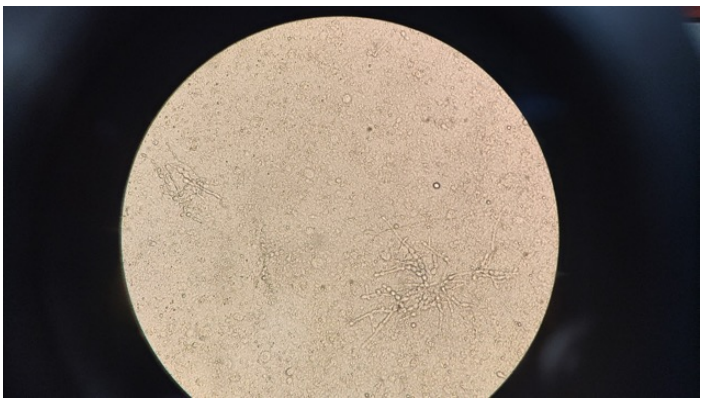
A pleural fluid sample was received for analysis of pH (ABL800 from Radiometer); automated cell count (DxH900 from Beckman Coulter); and biochemical parameters (Atellica CH from Siemens Healthcare).

Results

The macroscopic appearance was cloudy and whitish, pH=7.36. The sample could not be analysed in the cytometer due to the dense consistency of the sample. The laboratory specialist decided to visualise it in a haemocytometric chamber and reported a very purulent liquid appearance (possible empyema), no possibility of exact cell counting, and the presence of abundant hyphae and detritus.

Biochemical parameters after difficult centrifugation: glucose=20 mg/dL, triglycerides<30 mg/dL, LDH=11701 U/L, ADA=128 U/L, and total proteins, albumin and cholesterol: rejected due to analytical interference.

The clinician was notified by telephone of the unexpected result. The empirical treatment was changed to piperacillin-tazobactam, and selective antifungals were added for the treatment of filamentous fungi due to our visualisation of abundant hyphae, 48 hours before the diagnosis of Candida albicans by microbiological culture.



Conclusions

Direct visualisation of the fluid, and instant result communication to the clinician, allowed the diagnosis and treatment of unsuspected pleuro-pericardial mycosis in a patient, immunosuppressed by bipulmonary transplantation to be brought forward by 2 days.

Biological fluid analysers represent an important advance in laboratory automation. However, they do not replace the need for a review by the laboratory specialist to check and complete the auto-analyser report.

The establishment of alert results, with quick reporting results, should be proposed by the laboratory specialist, in collaboration with clinicians, although in daily laboratory practice each clinical case must be evaluated individually.

eP55

THE EFFECT OF LIPOSOME PREPARATION METHOD ON PHYSICOCHEMICAL PROPERTIES AND ANTIMICROBIAL ACTIVITY OF LIPOSOMAL MOXIFLOXACIN

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Two different liposome (LIP) preparation methods were used, the DRV and the active loading (AL) [1] methods to study their effect on antimicrobial efficacy (AE) of MOX-LIPs. LIPs were compared for MOX encapsulation (EE%), release kinetics, physicochemical properties (DLS, Cryo-TEM, XRD) and antimicrobial efficacy. LIP AE towards planktonic & biofilm bacteria was investigated.

AL method conferred liposomes with higher MOX EE%, that retained MOX for significantly longer periods compared to DRVs. All MOX LIP types (DRV & AL) had spherical morphology in nano-scale (100-150 nm), while most AL liposomal formulations and especially the ones with high EE% revealed some MOX crystallinity (XRD experiments).

Interestingly, AL LIPs demonstrated significantly higher AE towards planktonic S. epidermidis growth (Figure A,B) and biofilm susceptibility of the same bacterial strain, compared to corresponding DRV LIPs, indicating the importance of MOX retention in LIPs for their activity.

In conclusion, the liposome preparation method/type determines the rate of MOX release from LIPs and modulates their antimicrobial potential, a finding that deserves further in vitro and in vivo exploitation.

Acknowledgements

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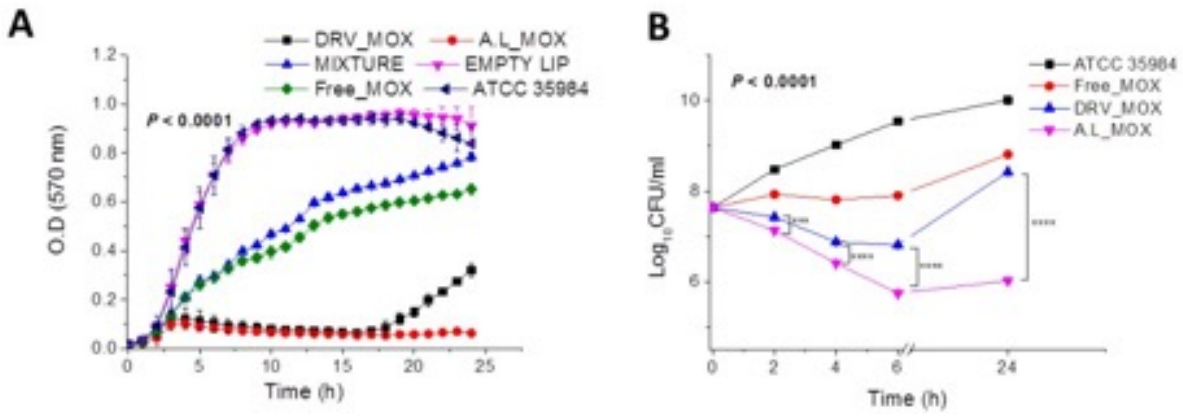

eP56

MONITORING OF SARS-CoV-2 NEUTRALIZING ANTIBODIES IN HEALTH CARE WORKERS AFTER VACCINATION WITH BNT162b2

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Intro

The health-care workers carried significant burden during the emergence of the SARS-CoV-2 pandemic. It is estimated that up to 11% of all samples from this population were positive in 2020, making them priority group for immunization.

Aim: It has been shown the levels of specific neutralizing antibodies are good predictors of vaccine efficacy. The aim of the present report is to estimate the early efficacy of the immunization process by determining the levels of specific neutralizing antibodies (NAbs).

Methods

131 HCWs enrolled to the earliest immunization program during the second peak of cases and accepted the call for vaccination with the BNT162b2 vaccine were included. At baseline, the participants were evaluated for past positivity for the virus via their electronic health care records and self-report. Patients were examined 6 months after on a conclusive visit, when blood samples were obtained for determining the presence of NAbs. The blood samples were analyzed by the CLIA method with the MAGLUMI 800 analyzer.

Results

The study sample included 131 health workers, out of which 85 patients were female. The median age of the sample was 36 years (IQR 32-42). From the whole sample, 32 patients reported previous known SARS-CoV-2 infection (24.4%). Six months after the vaccination, all patients with previous infection achieved NAbs above the threshold value of 0.3 , while from the other group only two patients had NAbs levels below 0.3. The median value of the NAbs in the whole sample was 1.56 (IQR 0.42 – 5.73), while patients with previous SARS-CoV-2 infection had median value of 6.45 (IQR 4.16 – 9.03), reaching striking difference when compared to patients without previous infection ($p < 0.001$).

Discussion

In a convenience sample consisted of health workers, the immunization with BNT162b2 produced NAbs titer above the threshold value in 98.5% of the participants, six months after the second dose. Participants with previous documented infection had substantially higher titer of NAbs, leaving room for further exploration on the best practice for the immunization process during the pandemic.

Keywords

MAGLUMI 800 ; neutralizing antibodies ; SARS-CoV-2 ; BNT162b2 ; immunization ; vaccine ;

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ASSOCIATION OF SERUM IL-6 WITH BIOCHEMICAL AND CLINICAL PARAMETERS AS PREDICTOR FOR COVID-19 DISEASE PROGRESSION

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Introduction

COVID19 is characterized by increased and de-regulated inflammatory response and development of cytokine storm, which has been linked to lung tissue damage and requirement of the patients for mechanical ventilation. Among the cytokines induced is IL-6, which has been used as biomarker for prediction of disease outcome and requirement for mechanical ventilation.

Aim

Aim of the work was to evaluate the levels of IL-6 in covid patients, determine its association with other biochemical and clinical parameters, and compare their value as predictors of requirement for mechanical ventilation and mortality in COVID-19 patients.

Methods

Serum levels of IL-6, CRP, Ferritin, D-Dimers, LDH, Creatinine, Troponin, and other biochemical parameters, hematological parameters such as WBC, Lymphocytes, blood gasses such as PaO2, PaCO2, pH and estimation of the requirement of ventilation as measured by PO2/FiO2, ROX, Respiratory Rate, was measured in COVID-19 patients admitted in the University Hospital of Heraklion. Disease outcome was monitored as requirement for ICU admission, requirement for High Flow Oxygen Therapy (HFOT) and survival.

Results

The results showed that IL-6 serum levels at the stage of admission was strongly associated with survival, requirement for mechanical ventilation and requirement of HFOT. The association was even stronger with the maximum value of serum IL-6. This association was stronger in men than women. Blood gas values were also associated with survival, mechanical ventilation ad HFOT, in a similar manner to this of IL-6.

Conclusions

Measurement of IL-6 upon admission of COVID-19 patients is a strong predictor of disease progression and outcome, but not stronger than measurement of blood gas parameters.

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URINARY TRACT INFECTION DUE TO HAFNIA ALVEI IN A PATIENT WITH MULTIMORBIDITY AFTER COVID-19

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Introduction

Hafnia alvei is a Gram negative, facultative anaerobic bacterium, belonging to the Enterobacteriaceae family and colonizes the gastrointestinal tract of bees, birds, fish and mammals. H. alvei has been recovered from the oropharynx and the gastrointestinal tract of humans, but has rarely been reported as a pathogen. A few cases of H. alvei infection have been reported so far, and the majority of them in patients with multimorbidity. We present a case of Hafnia alvei urinary tract infection in a patient with multimorbidity, 45 days after her hospitalization for COVID-19 pneumonia.

Case Report

A 75-year-old female patient was admitted to the Department of Internal Medicine of our hospital due to fever and dyspnea. From her history, type 2 diabetes, hypothyroidism, hypertension, heart failure, idiopathic tremor and hyperuricemia are mentioned as well as colon polyps. The clinical examination revealed: blood pressure 120/80mmHg, SO2 94%, temperature 36,60C, paleness, diminished breath sounds, and bilateral edema of lower extremities. The diagnostic tests were as below: Hb 7.9gr/dL, Ht 24.8%, MCV 108.4fL, PLT 120K/μL, CRP 5.97mg/dL, D-Dimers 2.14μg/mL. Urinanalysis showed protein 50mg/dL, urobilinogen 2mg/dL, red blood cells 4-6/hpf, white blood cells 100-120/hpf and abundant bacteria. Urine culture revealed a growthf>105cfu/mL of a Gram (-) negative bacterium, on blood agar and MacConkey No2 agar. The identification and the antimicrobial susceptibility testing were performed using the Microscan Autoscan-4 System (Siemens) and the microorganism was identified as Hafnia alvei (Microscan ID 99,99%). The strain was found susceptible to all the antibiotics tested according to the criteria of the EUCAST. The patient received empiric IV ciprofloxacin for two days, but due to the deterioration of the inflammation markers (CRP 14,26mg/dL), she was switched to IV ceftriaxone. After eight days, the patient's symptoms subsided, and she was discharged from hospital

Conclusions

H.alvei is an uncommon human pathogen, which should always be taken into account and evaluated, especially in cases like the one we presented herein. The occurence of Hafnia alvei infections is of interest in people who have or have had COVID-19[1,2].

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CLINICAL VALIDATION OF SNIBE'S NEW RAPID ANTIGEN TEST FOR THE DETECTION OF SARS-CoV-2

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Introduction - Aim

Lateral Flow Chromatography devices have been extensively used as rapid tests for the detection of Nucleocapsid antigen of SARS-CoV-2. In this study we aimed to validate the new rapid tests: RAPISAFE SARS-CoV-2 Antigen Rapid Test for SARS-CoV-2 diagnosis, released by the Shenzhen New Industries Biomedical Engineering co. Ltd. (SNIBE, Shenzhen, P. R. China).

Material and Methods

The RAPISAFE test detects the presence of SARS-CoV-2 N (Nucleocapsid) antigen on nasopharyngeal or oropharyngeal swabs taken from patients suspected for COVID-19.

As reference PCR method for the comparison, the Convergys RT-PCR COVID-19 Detection Kit on Convergys POC analyzer (Convergent Technologies GmbH & Co. KG Coelbe, Germany) was used.

In 526 pair of swab samples (nasopharyngeal and oropharyngeal) collected prospectively from 526 subjects, RAPISAFE test and Convergys RT-PCR was performed simultaneously. The mean (±SD) age of subjects was 38.4±18.4 years, range 1 to 87 years. 163 subjects were symptomatic for COVID-19, 87 had a close contact with an infected person and the remaining 286 didn't have any contact. The study was performed between January and March 2022.

Results

The sensitivity (SE) of the RAPISAFE test in all 526 nasopharyngeal samples was 90.4% and the specificity (SP) was 100%. In the corresponding oropharyngeal samples, the SE was 64.1% and the SP 98.5%.

For the samples with PCR Cts ≤25 the SE was 95.9% and SP 100% for nasopharyngeal samples and 70.9% and 98.5% respectively for the oropharyngeal samples.

The SE of the RAPISAFE test was higher in the symptomatic subjects or subjects with close contact compared to subjects with not known contact for both nasopharyngeal and oropharyngeal swabs.

Mean number of PCR cycles in positive nasopharyngeal swabs (15.7±5.1; n=218) was significantly lower ($p<0.001$) than in positive oropharyngeal swabs (20.5±3.4; n=198).

Conclusion

The SE and SP of SNIBE's RAPISAFE SARS-CoV-2 Rapid Antigen Test in nasopharyngeal swabs covers the EU specifications for clinical performance criteria (SE > 80% and SP > 98%). Also covers the clinical performance criteria (SE > 90%) for samples with PCR Cts ≤ 25.

The performance (SE, SP) of the RAPISAFE test in oropharyngeal swabs does not cover the EU specifications.

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B-NATRIURETIC PEPTIDE (BNP) AS A BIOMARKER FOR MULTISYSTEM INFLAMMATORY SYNDROME IN CHILDREN: A PRELIMINARY REPORT

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Introduction

COVID-19 disease caused by SARS-CoV-2 has a significant impact on everyday-life globally. The disease affects mostly, adults exhibiting great clinical heterogeneity. 40% of adults are asymptomatic. Nevertheless, severe COVID-19 cases exhibit pneumonia, which may be complicated by ARDS. Children represent about 19% of all COVID-19 cases. They are mostly asymptomatic or have indolent disease and are less likely to become severely ill and be hospitalized, compared to adults. Nevertheless, severe disease and sudden death are reported. The most dangerous complication in children is the multisystem inflammatory syndrome(MIS-C). Children with MIS-C have fever, multisystem and cardiovascular manifestations and up to 50-80% require Intensive-Care-Unit(ICU) hospitalization. Recent studies reported elevated CRP, troponin and decreased platelets and lymphocytes. B-natriuretic peptide(BNP) is a biomarker of heart failure diagnosis and treatment and for Acute Respiratory Distress Syndrome(ARDS) prognosis. More and more scientists study its role in MIS-C.

Aim

To determine and evaluate BNP levels in children with COVID-19 and in children with MIS-C. To estimate the possibility that BNP distinguishes children with increased risk of developing MIS-C.

Methods: 41 children were admitted to the emergency department(ED) of a tertiary hospital of northern Greece, between 1/2/2020-30/6/2022. All children were hospitalized and diagnosed with COVID-19 disease, confirmed by RT-PCR, some were tested positive for SARS-CoV-2 antibodies, upon admission. Their median age was 9 years(3months-15years) and 25(60.98%) were male. 17(41.56%) of them were diagnosed with MIS-C according to the current diagnostic criteria. BNP was determined on the UniCell DXI 800 Access Immunoassay System (paramagnetic particle solid-phase and enhanced chemiluminescent method). Mood's median test compared BNP levels between children with COVID-19 disease and children with MIS-C.

Results

Median BNP levels regarding the 24 children with COVID-19 disease were 30(8-187)pg/ml. The 17 children with MIS-C had median BNP levels 216(55-4161)pg/ml. BNP levels upon admission were statistically significant higher, regarding children with MIS-C, compared to children with COVID-19 disease($p<0.05$).

Conclusion

MIS-C is a life-threatening complication of COVID-19 in children. Main manifestations are cardiovascular complications, resulting in elevated BNP levels. Close clinical and laboratory monitoring of severely ill children attending the ED, including BNP levels determination are necessary for prompt MIS-C diagnosis and treatment.

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PROCALCITONIN LEVELS AND ITS CLINICAL SIGNIFICANCE REGARDING COVID-19 PATIENTS OF A TERTIARY HOSPITAL OF NORTHERN GREECE

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Introduction

Coronavirus disease-2019(COVID-19) is a novel disease with significant severity and mortality, was declared a pandemic on March 2020 and exhibits heterogeneous clinical course with patients being asymptomatic or with indolent disease, while others suffer severe disease with fatal outcome. It is crucial to have prognostic factors for optimal patients' management and treatment. Low WBC and PLTs, elevated CRP and D-Dimmers are correlated with severe disease and mortality. Numerous studies try to identify new prognostic markers. Procalcitonin(PCT) is a biomarker of inflammation, elevated in bacterial infections and normal, regarding viral infections.

Aim

To determine and evaluate PCT levels in COVID-19 patients, in critically ill COVID-19 patients and estimate the probability that PCT predicts disease severity by investigating whether PCT distinguishes patients admitted to the Intensive-Care-Unit(ICU).

Methods

717 patients admitted to a tertiary hospital of northern Greece between 1/11/20–31/5/21 (second-third pandemic wave) were studied. 597(83.26%) had RT-PCR confirmed COVID-19 disease, mean age 65.1±13.94years, 398(66.6%) were male. 120(16.74%) had other febrile infections, mean age 67.7±18.2years, 52(43.33%) were male. Patients with renal disease were excluded. PCT was determined upon admission or within the first 48 hours of hospitalization, excluding hospital infections. PCT was determined using UniCell DXI800 Access Immunoassay System (paramagnetic particle solid-phase enhanced chemiluminescent assay). The Mann-Whitney U-test compared COVID-19 and nonCOVID-19 patients' PCT levels. Mood's median test compared ICU and Internal Medicine Department(IMD) COVID-19 patients' PCT levels.

Results

Regarding 597 COVID-19 patients, median PCT was 0.35ng/ml, 116(19.43%) had abnormal PCT levels(PCT>0.5ng/ml), 160(26.8%) were admitted to ICU and 437(73.2%) to IMD. The 120 nonCOVID-19 patients had median PCT 1.95ng/ml, which was significantly higher compared to COVID-19 patients, 48(40%) exhibited PCT>0.5ng/ml. COVID-19 patients had statistically significant lower PCT levels compared to nonCOVID-19($p<0.05$). PCT levels upon admission were statistically significant higher, regarding ICU admitted COVID-19 patients, compared to IMD admitted COVID-19 patients($p<0.05$).

Conclusion

COVID-19 is a viral infection and PCT, when elevated, indicates secondary bacterial infection complicating the clinical course. COVID-19 patients exhibiting elevated PCT upon admission require additional monitoring and therapeutic intervention because they are likely to suffer from bacterial co-infection and more likely to have worst clinical course and be hospitalized in the ICU.

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S. LUGDUNENSIS ISOLATION FROM BODY FLUID CULTURES DURING A 6-YEAR PERIOD (2016-2022)

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Introduction

S. lugdunensis belongs to the coagulase negative (-) staphylococci (CoNS) and is part of the normal flora of the skin, usually colonizing the perineum. Recently, S. lugdunensis has emerged as a human pathogen involved mostrly in skin, soft tissue, bone, joint, prosthetic joint infection and also as a cause of aggressive endocarditis , urinary tract infections and peritonitis[1]. S. lugdunensis differs from other CoNS because its pathogenicity is similar to that of S. aureus and because it remains sensitive to most antibiotics [2]. The purpose of this work is the study of S. lugdunensis strains isolated from several body fluid cultures at the Department of Clinical Microbiology of our Hospital during the period 2016-2022.

Methods

We recorded the strains of S. lugdunensis isolated from cultures of body fluids and other biological materials at the Department of Clinical Microbiology of the General Hospital of Amfissa, during the period 2016-2022. Cultures were performed using conventional microbiological methods, while identification and antibiotic susceptibility testing were performed using the semi-automated Microscan Autoscan system (Siemens).

Results

In total, six (6) strains of S. lugdunensis were isolated, one (1) from urine, one (1) from synovial fluid, and three (3) from purulent secretions. The four (4) strains were isolated from male patients, while the remaining two (2) from female patients. Of the six strains, two were resistant to oxacillin, tobramycin and daptomycin, while one of them also showed resistance to quinolones. Three of the strains were multisensitive.

Conclusions

Given the pathogenicity of S. lugdunensis strains, their management when isolated as causes of infections is analogous to that of S. aureus. For this reason, it is important to correctly identify the specific microorganism as well as monitor its resistance profile.

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LOOKING FOR A RELATIONSHIP BETWEEN BNP AND COVID-19 DISEASE SEVERITY AND HOSPITALIZATION

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Introduction

SARS-CoV-2 is the cause of COVID-19 disease, a new pandemic with clinical heterogeneity. Patients suffer mild disease or are asymptomatic but sometimes, through the activation of inflammatory mechanisms, patients develop severe disease with fatal outcome. Many laboratory tests have been studied to optimize patient management, such as CRP, D-Dimmers, fibrinogen, ferritin, lymphocytes and platelets count. B-natriuretic peptide(BNP) is a widely used biomarker both in the diagnosis and treatment of heart failure, including patients presenting at the emergency department(ED) suffering from Acute Respiratory Distress Syndrome(ARDS). Regarding patients hospitalized in the Intensive-Care-Unit(ICU), BNP can identify those with high probability for ARDS and predict mortality.

Aim

To investigate whether BNP can predict which COVID-19 patients need ICU hospitalization.

Methods

BNP was determined by enhanced chemiluminescence on the UniCell DXI800 immune system in 104 RT-PCR confirmed COVID-19 patients, admitted to a tertiary hospital of Northern Greece, during the second pandemic wave. Patients with cardiac and/or renal failure were excluded. Patients' median age was 70(57-94)years, 68(65.38%) were male. 33(31.73%) patients were hospitalized in the ICU and 71(68.27%) in the Internal Medicine Department(IMD). The non-parametric Mood's median test compared BNP levels between COVID-19 patients hospitalized in the ICU and those hospitalized in the IMD.

Results

In 104 patients with COVID-19 disease, the median BNP value was 72.5(9-1382) pg/ml. The patients hospitalized in the ICU, had median BNP value 72pg/ml, while those hospitalized in the IMD had 73pg/ml. No statistically significant difference in BNP levels was found between the COVID-19 patients hospitalized in the ICU and the IMD(p=0.369).

Conclusion

In patients with pneumonia, BNP levels increase due to the cumulative release of cytokines that induce inflammation. Several studies regarding COVID-19 disease report excessive activation of inflammation correlated with elevated BNP levels. Studies also correlate BNP with disease severity and mortality in COVID-19 patients. We investigated whether BNP upon admission is an indicator for patients' hospitalization in the ICU. In this cohort of patients with COVID-19 disease, BNP was unable to distinguish which patients had severe disease and required ICU hospitalization. Larger number of patients may be more likely to clarify the role of BNP as a prognostic marker in COVID-19 disease.

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COMPARISON OF OMICRON AND DELTA SARS-COV-2 VARIANTS LABORATORY TESTS: A PRELIMINARY REPORT

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Introduction

The Omicron variant was first identified on 9/11/2021 in South Africa and was declared a variant of concern(VOC) on 26/11/2021 by the World Health Organization(WHO). Omicron is more contagious and evades vaccination. Recent studies showed that the Omicron variant causes indolent disease and patients are less likely to be hospitalized. Regarding hospitalized patients, they do not have severe pneumonia and hyperinflammation syndrome, as often as in previous waves of the pandemic. Consequently, laboratory tests including inflammatory markers are different and their study is remarkably challenging.

Aim

To compare the inflammatory markers ferritin, procalcitonin(PCT) and fibrinogen between Delta and Omicron variant waves of the pandemic in hospitalized COVID-19 patients.

Methods

414 COVID-19 patients, RT-PCR confirmed, 213(51.45%) male, median age 68(17-98)years were admitted to a tertiary hospital, 240(57.97%) within Delta variant wave and 174(42.03%) within Omicron variant wave. PCT was determined upon admission or within the first 48 hours of the admission, to exclude hospital infections. Patients with renal and/or cardiac failure were excluded. Ferritin and PCT were determined on UniCell DXI800 Access Immunoassay System, a paramagnetic particle solid-phase and enhanced chemiluminescent assay. Fibrinogen was determined on BCS® XP Siemens with a modified Clauss method. Mood's median test compared laboratory tests between Delta and Omicron variant waves.

Results

Ferritin was determined in 95 patients, 66 within Delta variant wave, median 540,4(31,4-5558)ng/ml and 29 within Omicron variant, median 166,7(28-4457)ng/ml. PCT was determined in 193 patients, 124 within Delta variant, median 0,1(0,01-34,1) ng/ml and 69 within Omicron variant, median 0,07(0,02-5,7)ng/ml. Fibrinogen was determined in 404 patients, 237 within Delta variant, median 548,3(42,3-1560)mg/dl and 167 within Omicron variant, median 437,3(20-1042,5)mg/dl. Ferritin, PCT, fibrinogen levels upon admission were statistically significant lower, regarding patients admitted during the Omicron variant pandemic wave, compared to patients admitted during the Delta variant pandemic wave($p<0.05$).

Conclusion

Omicron variant is more contagious and evades vaccination. Nevertheless, Omicron causes mild disease. This is reflected in laboratory tests. Patients with the Omicron variant have lower inflammatory markers levels including ferritin, procalcitonin and fibrinogen. Vaccination is also likely to reduce the incidence of severe illness. Nevertheless, people with underlying diseases still remain more vulnerable.

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LIPID PROFILE ALTERATIONS IN COVID -19 PATIENTS

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Background

Covid-19 is an infectious disease caused by SARS COV-2, firstly identified in Wuhan, China in December 2019 and quickly spread worldwide, resulting in COVID-19 pandemic. The virus initiates a systemic inflammatory syndrome shown by the elevation in inflammatory markers' levels.

Aim

Among other alterations this study investigates the alterations in the lipid profile of COVID-19 patients during the acute phase of the infection.

Methods

A retrospective study was performed to examine serum levels of C reactive Protein(CRP), Total Cholesterol, LDL-Cholesterol and Triglycerides and the plasmatic level of D-Dimer on 95 ambulatory patients diagnosed with COVID-19 by RT PCR, during the acute phase of the infection in a 8 month period, between April and December 2021. The data gathered were processed by SPSS.

Results

87.5 % of the patients were found to have high CRP serum values. 86.3 % of the patients were found to have high DDimer values.62.2 % of the patients were found to have high Total cholesterol serum values. 41% of the patients were found to have high Triglycerides values and 41.1% of the patients were found to have high LDL cholesterol values. DDimer has a statistically significant correlation with Total Cholesterol $r=0.31$, $N=95$, $p<0.01$; LDL Cholesterol $r=0.34$, $N=95$, $p<0.01$ and Triglycerides $r=0.35$, $N=95$,$p<0.01$. CRP has a statistically significant correlation with Total Cholesterol $r=0.35$, $N=95$, $p<0.01$;LDL Cholesterol $r=0.37$, $N=95$, $p<0.01$ and with Triglycerides $r=0.24$, $N=95$, $p<0.05$. In 18.2% of the patients inflammation markers CRP and DDimer have a predictive effect on Total Cholesterol statistically significant at $p<0.01$. In 21.2% of the patients inflammation markers CRP and DDimer have a predictive effect on LDL Cholesterol statistically significant at $p<0.01$.

Conclusion

Total Cholesterol, LDL-Cholesterol and Triglycerides were found to be significantly higher in patients with more severe inflammatory response shown by higher levels of CRP and D-Dimer.

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FIRST TRIMESTER PRISCA SCREENING FOR DOWN AND EDWARD SYNDROME IN UNSELECTED PREGNANCIES - REPORT OF A 6-YEAR EXPERIENCE

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Introduction

First-trimester combined screening based on maternal age, fetal nuchal translucency thickness and the serum markers free β-hCG and PAPP-A is effective screening for trisomy 21, with a detection rate of about 90–95% for a false-positive rate of 3–5%.

Aim

Since April 1999, the first trimester combined test has been offered to every pregnant woman who came to Cathay General Hospital, Taipei, during 10–13 weeks of gestation. The aim of this study was to evaluate the performance of the first trimester combined test for Down syndrome detection

Methods

Down syndrome risk was calculated using PRISCA software screening which include measuring of fetal nuchal translucency and maternal serum levels of free β-hCG and PAPP-A by using a Immulite 2000 xpi analyzer.

Results: The participants comprised 2975 women who participated in the screening voluntarily. The mean gestational age, maternal age, and maternal weight were 86 days, 28 years and 67 kg, respectively. There were 257 (9%) women >35 years of age and 54 (2%) women with multiple gestations - twins. In total, 215 cases of chromosomal abnormality were identified,among them, there were 165 cases of trisomy 21 and 50 cases of trisomy 18. The detection rates for trisomy 21 and trisomy 18 were 77% and 23%. In trisomy 21, the MoM of maternal serum PAPP-A is 0.93 and free β-hCG is 2.43. In trisomy 18, the mean MoM of PAPP-A is 0.55 and free β-hCG is 1.21.

Conclusions

First trimester screening for aneuploidy is a valuable tool for the obstetrician and indications and methods of screening have evolved over the last quarter century. The primary goal of first trimester screen is to identify higher risk women for fetal aneuploidy and give them the option to pursue diagnostic testing in a timely manner if desired. The first trimester combined test is an effective screening tool for Down syndrome detection, with an acceptable low false positive rate. The best timing of screening will be between 11 and 12 weeks' gestation.

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PRE-ECLAMPSIA –PREDICTIVE VALUES OF STANDARD BIOCHEMICAL MARKERS IN FIRST TRIMESTER OF PREGNANCY

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Introduction

Pre-eclampsia is pregnancy-specific, multi-systemic disorder that occurs after 20 weeks of gestation.

Aim: The aim of this study was to evaluate the predictive value of standard biochemical analysis such as creatinine, urea, uric acid and C-reactive protein (CRP) in the first trimester of pregnancy for pre-eclampsia (PE), in comparison to Pregnancy associated plasma protein-A (PAPP-A).

Methods

This prospective study included 403 women over 18, in the first trimester of pregnancy (11 to 14 g.w.), with a singleton pregnancy. All patients were included in the first trimester screening program for fetal chromosomal abnormalities and possible monitoring of high-risk pregnancies, at the Clinical Center of Vojvodina. The participants were followed until the end of pregnancy, and divided into two groups: a group with subsequently developed PE (PE group) (n=61); and a control group (CG group) with no complications and normal outcomes (n=342). Serum values of urea, creatinine, uric acid and CRP were determined by standard biochemical methods, in patient's sera. in both groups. PAPP-A was determined using ECLA method on Roshe Cobas e411.

Results

Serum creatinine level is significantly higher in PE group than in control group ($p<0.001$) (62.3±9.6 vs. 49.2±9.6 μmol/L), as well as level of uric acid ($p<0.001$) (238.2 ± 54.3 vs. 197.5 ± 36.74mmol/L). Serum CRP (4.67±6.01 vs. 3.05±6.80 mg/L) and urea levels (3.3 to 3.4 mmol/L) doesn't' show significant difference between groups. According to our results, there was no significant decrease in PAPP-A levels in PE group, compared to control group (1.26 vs 1.06 MoM). In the detection of women with PE, serum creatinine level ≥ 53 μmoll/L had a sensitivity of 85.2 % and specificity of 71.3 % and positive likehood ratio of 2.97. The level of serum creatinine is in a negative significant correlation ($p<0.05$) with the week of gestational outcomes ($R=-0.597$), birth weight ($R=-0.534$) and Apgar score ($R=-0.374$) of newborns. ROC analysys showed that the best diagnostic accuracy in PE prediction is detected for serum creatinine (AUC 0,837), compared to uric acid (AUC)

Conclusions

Results of our research show that serum concentration of creatinine in first trimester could have a significant predictive value for PE.

eP68

CEREBROSPINAL FLUID YKL-40 AS A POTENTIAL BIOMARKER FOR THE DIAGNOSIS OF PRECLINICAL STAGES OF ALZHEIMER DISEASE: A SYSTEMATIC REVIEW AND META-ANALYSIS

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Introduction

Alzheimer's disease belongs to a class of neurodegenerative conditions that cause dementia. Dementia refers to a condition in which impairment of mental functions are observed which include speech, perception and memory. Psychological and behavioral disorders may also appear as symptoms in dementia. It is estimated that the onset of the disease begins about 20 years before the onset of symptoms. Therefore, the prognosis of the disease in the early stages is of great importance as it will also signal the early beginning of the treatment.

Aim

The purpose of this study was to investigate the role of a biomarker, namely the inflammatory biomarker YKL-40, for the prognosis of Alzheimer's disease in the preclinical and prodromal stages of the diseases (i.e. early stages).

Methods: All bibliographic data related to the measurement of the biomarker in the cerebrospinal fluid of healthy individuals and patients at different stages of Alzheimer's disease, were collected. To investigate the correlation of the above data, a systematic review and a meta-analysis were employed. PubMed and Google scholar databases were searched up until May 2021. The random effects model was used in the meta-analysis utilizing the mean difference (MD). Heterogeneity was quantitatively assessed using the Cohran's Q and the I2 index. Sensitivity analysis, cumulative meta-analysis and assessment of publication bias were also performed.

Results

From a total of 35 studies identified by the searching procedure, 10 were finally included in the meta-analysis. The meta-analysis yielded statistically significant results for the prognosis of the preclinical stage of Alzheimer's disease (mean difference= 41.207, 95%CI= 25.42-56.99). Considerable heterogeneity was present among the studies (I2>75%), but no evidence of publication bias (Egger's test: P=0.089).

Conclusions

The results of this meta-analysis suggest that YKL-40 can be considered as valuable biomarkers for the detection of Alzheimer's disease in the preclinical stages.

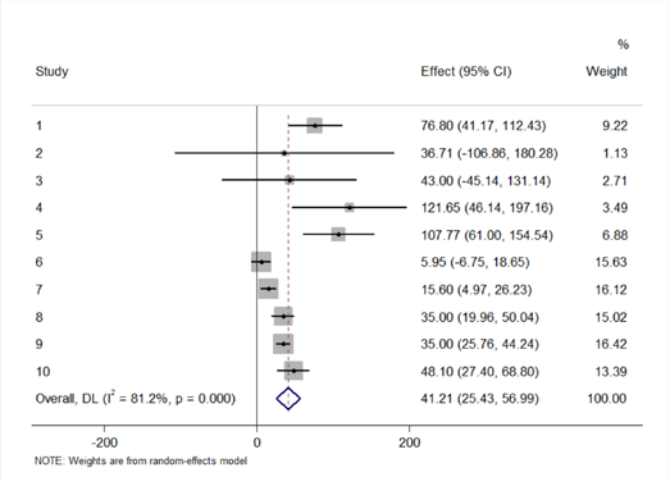

Figure 1. Forest plot of CFS YKL-40 for he diagnosis of preclinical stages of Alzheimer's disease

eP69

THE LABORATORY ROLE IN THE SPORADIC CREUTZFELDT-JAKOB DISEASE (CJD) DIAGNOSIS. A CASE REPORT

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Introduction

Sporadic Creutzfeldt-Jakob disease (sCJD) is a rare and fatal neurodegenerative disease included within transmissible spongiform encephalopathies. These diseases are associated with the deposition within the brain of a prion protein (PrPC) whose PrPC misfolding induces aggregation into fibrils and plaques. Most patients die within 12 months after the onset. The sCJD diagnosis is associated with a rapidly progressive dementia, ataxia, myoclonus, or other neurologic signs and hypersignal in the basal ganglia or, at least, two cortical regions on brain magnetic resonance imaging (MRI), and positivity cerebrospinal fluid (CSF) biomarkers for 14-3-3 and Real-Time Quaking-Induced Conversion (RTquIC).

Aim: A 80-years-old man who was referred to a 1-month history of rapidly progressive dementia. The initial symptoms including increased forgetfulness, behavioral changes, dysarthria and apraxia.

Methods.

Not applicable.

Results

The neuropsychological examination revealed unequivocal deterioration of cognitive functions such as severe slurring of speech, short-term forgetting episodes and apraxia. He demonstrated loss of ability to obey simple commands. Babinski reflexes were absent. No myoclonic jerks were identified.

His general conditions worsened significantly by few weeks since admission. We observed general myoclonus, and he was only able to move with double support due to the ataxic gait.

The laboratory tests, including blood tests, glucose levels, renal, hepatic, and thyroid function tests, electrolytes, C-reactive protein (CRP) and B12 vitamin levels were normal. The autoimmune screens were negative, and the results of CSF were irrelevant.

MRI brain revealed no stroke but cortical and left caudate diffusion restriction, suggestive of sCJD.

The EEG showed an abnormal pattern of periodic sharp-wave complexes, which, in that clinical context of rapidly progressive dementia, could be characteristic of sCJD. CSF study showed raised protein 14-3-3, a neuronal death biomarker, and RT-QuIC analysis, a new more sensitive and specific diagnostic method which has recently emerged, was also positive.

Conclusions

The clinical symptoms of sCJD can be masqueraded by other neurological syndromes. Therefore, we would like to underline the difficulties in reaching the diagnosis and the need for cutting-edge diagnostic methods in which the laboratory role is essential.

eP70

25(OH)VIT-D DETERMINATION AT THE DEPARTMENT OF BIOPATHOLOGY OF A RURAL HOSPITAL IN A 5-YEAR PERIOD (2017-2022)

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Introduction

For decades, vitamin D has been known for its contribution to the development, function and maintenance of bone health, through the regulation of calcium homeostasis, throughout life. Sufficient levels of vitamin D prevent rickets in children and osteomalacia in adults. Furthermore, the adequate intake of vitamin D and calcium is the basis for the prevention and treatment of osteoporosis. But in addition to the role it plays in the metabolism of the skeletal system, vitamin D is involved in the functioning of the immune and cardiovascular systems, as well as in cell growth and division. The determination of 25(OH)vit-D is the indicated test to evaluate the levels of vitamin D in the body.

Aim

The study of the demand of 25(OH)Vit D determination in the medical laboratory department of a rural hospital in a 5-year period (05/2017-05/2022).

Methods

We ran a statistical analysis of the data regarding the 25(OH)vit-D determinations that were requested and performed at the Medical Laboratory Department of the General Hospital of Amfissas. The tests were performed on the Alinity immunoassay analyzer (Abbott). The data were obtained from the database of the CTEAM LIS information system of the Computer Team company, which is used by our laboratory.

Results

The data analysis showed that in the aforementioned period of time, there were performed 10183 determinations of 25(OH)vit-D in a total of 964956 laboratory tests . In Table 1, we present the results per semester.

Conclusions: The demand for 25(OH)vit-D testing was almost doubled, up to the end of 2018. Subsequently, demand remained at relatively steady, high levels through May 2022. These findings reflect increased awareness among both prescribing physicians and patients of the importance of vitamin D for health.

Table 1

25(OH)Vit D											
Semester	1st	2nd	3rd	4th	5th	6th	7th	8th	9th	10th	Total
25(OH)Vit D	620	841	1139	1069	1348	918	1150	1115	1149	834	10183
Total Tests	95115	92896	93623	98654	127884	86795	96627	94417	99747	79198	964956
25(OH)VitD/10.000 tests	65,18	90,53	121,66	108,4	105,41	105,8	119	118,1	115,19	105,3	105,53

Personalised Medicine, including pharmacogenetics

eP71

A NOVEL MITOXANTRONE-GNRH ANALOGUE CONJUGATE WITH ANTITUMOR EFFECTS

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Introduction

The gonadotropin releasing hormone (GnRH) plays a key role in the function of the reproductive system through its interaction with its receptor, GnRH-R. GnRH-R is also highly expressed in various types of tumor cells, including the ovarian ones. Previous studies have shown that GnRH analogues exert antitumor effects on ovarian and other cancer cells, through their interaction with the GnRH-R expressed in these cells.

Aim

In this study we aimed to develop novel GnRH analogues with antitumor actions, by conjugating them with the cytotoxic agent, mitoxantrone, thus creating the analogue Con3. Con3 is anticipated to release mitoxantrone into cancer cells after its binding to GnRH-R and subsequent internalization of the GnRH-R/Con3 complex.

Methods

To create the con3 analogue we chemically modified the GnRH analogue, leuprolide and conjugated it with mitoxantrone. Evaluation of the pharmacological properties of the con3 was performed in competition radioligand binding studies using [125I]-DTyr6-His5-GnRH as radioligand and membrane homogenates from HEK 293 cells stably expressing the GnRH-R. The apparent binding affinities (IC50 values) were obtained by fitting the binding data to a one-site competition model. Further assays to examine the antiproliferative effects of con3 were performed by incubating the ovarian cancer cell line, SK-OV-3, with con3 at different concentrations for 1-4 days and using the MTT assay.

Results

Con3 binds to GnRH-R in a dose-response manner, with the high affinity of 0.06 nM, thus allowing us to characterize its antiproliferative actions. This was accomplished by testing its ability to inhibit the proliferation of SC-OV-3 cells. The results have shown that the proliferation of SC-OV-3 cells was inhibited by Con3 in a time-dependent manner (1-4 days). Interestingly, Con3 inhibited the proliferation of SC-OV-3 cells in a dose-dependent manner. The antiproliferative potency of con3 after exposure of SC-OV-3 to peptide for 2,3 and 4 days, was 1.4 μM, 0.85 μM, 0.97 μM and 1,1 μM, respectively.

Conclusions

1. Con3 binds to GnRH-R with high affinity (0.06 nM)
2. Proliferation of SC-OV-3 cells was inhibited by Con3 in a time and dose dependent manner.

eP72

INTERFERENCES LEADING TO ANALYTICAL ERRORS IN THE BIOCHEMICAL LABORATORY THE EFFECT OF MACROMOLECULES ON THE DETERMINATION OF FOL AND B12

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1. B12 and FOL

Introduction

During a cycle of laboratory testing, which consists of three main phases, the pre-analytical, analytical and post-analytical, there is the possibility of errors occurring, which alter the values ??of biochemicals and immunological tests, resulting in the clinician misdiagnosing and in inappropriate treatment. Several studies have reported the presence of macroforms in the serum of patients. Macromolecules-macroforms are high molecular mass complexes of immunoglobulins with enzymes, proteins, and hormones. Their clearance rate in the blood is slow, giving false positive results in immunoassays.

Aim

The aim of this work was the review of the interferences that can lead to analytical errors in a biochemical laboratory, with particular emphasis on the interferences against immunoenzymatic reactions such as heterophilic antibodies, autoantibodies, macroforms, etc. We focused on the detection of interference due to the presence of macroforms in two types of tests, vitamin B12 and folic acid FOL in patient serum.

Methods

We collected random consecutive samples for two population groups, control group (B12 values: 400-1400 ng/ml; FOL: 4-20 ng/ml) and study group (B12 values: >1400 ng/ml; FOL: >24 ng/ml), which were treated with saline and with polyethylene glycol to precipitate immunoglobulin complexes.

Results

For the test of B12, the highest relative frequency of the samples (30/90) shows approximately 30% percentage of precipitation with polyethylene glycol. The diagram of PPB12(percentages of PEG precipitated B12) of the control group follows a normal distribution, on the contrary, the diagram of the study group does not. For the test of FOL, the highest relative frequency of the samples (40/132) shows approximately 20% percentage of precipitation with polyethylene glycol. The diagram of PPFOL (percentages of PEG precipitated FOL) of the control group follows a normal distribution, on the contrary, the diagram of the study group follows a positive skew distribution.

Conclusions

During the immunoassay of the B12 and FOL tests and comparing their two population groups, based on the literature we confirmed the presence of interference due to macroforms in the samples of B12 test and not in the FOL test.

Toxicology, including therapeutic drug monitoring

THE RELATIONSHIP BETWEEN BLOOD AND URINE CADMIUM CONCENTRATION IN WORKERS WITH NICKEL-CADMIUM BATTERIES IN BULGARIA

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Introduction – Aim

Cadmium is a toxic metal. When it builds up in the body over time or in a short period of time, health problems may occur. Cadmium toxicity can affect any of the internal organs, especially the lungs when inhaled, as well as the kidneys and liver. The purpose of this study is to investigate exposure parameters such as blood and urine cadmium, and their correlation to proteinuria, amino acids, glucose, β2-microglobulin, retinol-binding protein, albumin, serum β2-microglobulin, creatinine clearance, and tubular reabsorption of beta 2–microglobulin. The concentrations of cadmium in whole blood (Cd-B) and in urine (Cd-U) were determined in more than 100 workers exposed to cadmium and in a comparable group of unexposed workers.

Methods

Analysis of cadmium in blood and urine were performed using a graphite furnace atomic absorption spectrophotometer (GF-AAS) PinAAcle 900Z. All chemicals are with analytical or certified reagents. A standard cadmium solution Certipur® (Merck) (1000 mg/L Cd) was used. Working solutions were prepared daily in 1% nitric acid and 0.3% ammonium dihydrogen phosphate. Concentrated nitric acid (Merck) with cadmium content (<0.00005%) below the detection limit of GF-AAS was used.

Results

Urinary cadmium levels can be used to estimate body cadmium burden. In chronic exposures, the kidneys are the primary target organ. Symptoms associated with cadmium toxicity vary depending on the route of exposure and may include tubular proteinuria, fever, headache, dyspnea, chest pain, conjunctivitis, rhinitis, sore throat, and cough. Ingestion of cadmium in high concentration can cause vomiting, diarrhea, salivation, cramps, and abdominal pain. Based on the data, it is concluded that the health effects of cadmium can be assessed using the following critical levels: cadmium in blood – 10 µg/L, cadmium in urine – 10 µg /g creatinine; urinary beta 2-microglobulin – 2000 µg /g creatinine, urinary retinol-binding protein – 200 µg /g creatinine, and urinary β2-microglobulin to albumin ratio of 0.001. Biological threshold limits for cadmium in urine and blood are based on the correlation of biological levels for renal dysfunction. The use of markers of high and low molecular weight proteinuria should be integrated into the health surveillance of workers exposed to cadmium. Urinary Cd concentrations were significantly higher in exposed workers compared to controls, although no statistical difference was observed in urinary β2-microglobulin content between the two groups. Mean concentrations of Cd in blood and urine did not exceed the recommended reference values ??of 10 µg/L in blood and 10 µg/g creatinine in urine.

Conclusions

In exposed workers, both Cd-U and Cd-B correlated with exposure intensity but not exposure duration. Cd-B is assumed to reflect current exposure, but Cd-U may reflect body burden of cadmium when exposure is low and current exposure when exposure is high (industrial situations). The best screening and diagnostic test for chronic cadmium exposure is a 24-hour urinary cadmium level normalized to creatinine excretion. Excretion of metallothionein and β2-microglobulin in the urine can be related to the long-term cadmium exposure.

eP74

EVALUATION OF D-DIMERS LEVEL, SERUM CRP CONCENTRATION AND LACTATE DEHYDROGENASE ACTIVITY IN PATIENTS WITH MILD COVID-19

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Introduction and aim

This study retrospectively analysed the clinical value of three biochemical parameters in patients with mild COVID-19. D-dimers level, serum C-reactive protein (CRP) and lactate dehydrogenase activity (LDH) were examined in patients three days after detection of infection with Sars-Cov-2.

Methods

A total of 278 patents from 20-80 years old, divided in three subgroups, were included. D-dimers level as a possible marker of extravascular fibrinolysis was determined by fluorescent enzymatic method, CRP concentration with turbidimetric method and serum lactate dehydrogenase by spectrophotometric method.

Results

In the first subgroup from 20-40 years old, from 75 patients, D-dimers level was increased for about 50% above normal in 7 patients and two-fold increase above normal in 4 patients. CRP concentration was increased two to ten-fold above normal in 6 patients and LDH activity was increased for about 50% above normal in two patients and two- fold increase in 8 patients. In the second subgroup from 41-60 years old, from 112 patients, D-dimers level was increased in 33 patients from about 50% above normal to two-fold above and five-fold above only in one patient. CRP concentration was increased two to ten-fold above normal in 34 patients and twenty- fold increase in only two patients. LDH activity was increased for about 50% above normal to two-fold above in 43 patients. In the third subgroup from 61-80 years old patients, from 91 patients, D-dimers level was increased in 52 patients from 50% above normal to four-fold above and twenty fold above only in one patient. CRP concentration was two to twenty five-fold above normal in 39 patients. LDH activity was increased from 50% above normal to three-fold above in 54 patients.

Conclusion

The results have shown that highest values for evaluated biochemical parameters were detected in age subgroup from 61-80 years old, which confirmed the finding that old age is the first risk factor for unfavourable course of the disease together with underlying comorbidities. The levels of above mentioned biochemical parameters may facilitate the assessment of disease severity and might be useful in indicating progression from mild to severe COVID-19.

Table 1. Results for D-dimers and CRP levels and LDH activity in different age groups of patients with mild Covid-19

A total of 278 patients	D-dimers increase above normal			CRP increase above normal		LDH increase above normal	
20- 40 years old (75 patients)	50% (7 patients)	two- fold (4 patients)		two to ten- fold (6 patients)		50% (2 patients)	two- fold (8 patients)
41- 60 years old (112 patients)	50% (33 patients)	two- fold (1 patient)	five- fold (1 patient)	two to ten-fold (34 patients)	twenty-fold (1 patient)	50% to two-fold (43 patients)	
61- 80 years old (91 patients)	50% (52 patients)	four- fold (1 patient)	twenty- fold (1 patient)	two to twenty five- fold (39 patients)		50%-three-fold (54 patients)	

eP75

COLD AGGLUTININ DISEASE, A LABORATORY CHALLENGE

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Introduction

Cold agglutinin disease (CAD), is a rare autoimmune disorder characterized by the premature destruction of red blood cells. (hemolysis). More specifically CAD is a subtype of autoimmune hemolytic anemia. It is termed "cold" because the antibodies are active and cause hemolysis at cold temperatures, usually at 3-4 °C. Cold agglutinin disease can be primary(unknown cause) or secondary, due to an underlying condition

Aim

The importance of knowledge of cold agglutinin interference and effects on laboratory tests.

Case presentation

Method

A 58 years old woman presented at laboratory for hematology profile evaluation. Blood sample was collected into anticoagulated tube for CBC and analyzed with sfri-hemix automated hematology analyzer

Results

CBC showed invalid findings wi. Checking the laboratory process showed precipitation residue sticky to the sides of the tube. The automated analyzer results indicated low Hct (17,4%) RBC count (1.900.000/mm3) MCV (92fl), MCH (74,1pg),MCHC (81.4g/dl).The sample was reanalyzed after warming in a bain-marie for 25 minutes to 37°C. The CBC result improved with Hct (37.6%),erythrocyte count (4.000.000/mm3),MCV(92fl),MCH(34,2pg) and MCHC (37g/dl).

Conclusion

Knowledge of this phenomena can help prevent wasting too much time releasing an accurate laboratory result and make an early and accurate diagnosis.

eP76

STUDY OF BIOCHEMICAL INDICATORS RELATED TO METABOLIC SYNDROME IN PRISONERS

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Introduction-Aim

Prisoners are a population on which relatively few studies have been conducted and,therefore,insufficient information is available.The purpose of this work is the retrospective study of the biochemical indicators related to the metabolic syndrome in inmates of the detention centers of Fokida Regional Unit.

Methods

The results of the biochemical tests performed on the above population for the period 01/01/2021-31/12/2021 were collected and recorded as a Microsoft Excel 2010 file.Specifically,the following biochemical tests were recorded:fasting glucose

(GLU),uric acid(URIC ACID),Total cholesterol(TCHOL),HDL,LDL,Triglycerides(TRG) and Glycosylated hemoglobin(HbA1C). Moreover,TyG index was calculated(TyG=Ln [TRG(mg/dl)*GLU(mg/dl)/2].The biochemical tests were performed with the AU680 Beckman Coulter biochemical analyzer(Leriva).A total of 100 male prisoners aged 22-83 were examined.The statistical analysis for the difference in mean values between the uric acid quartiles was done using one-way ANOVA,whilst the correlations between the variables were assessed using Spearman's RHO.

Results

The results are presented in Tables 1 and 2.In Table 1 we note that in all four quartiles,the average value of fasting glucose exceeds the value of 125 mg/dL, while high total cholesterol and LDL values are observed (>170 mg/dL and >100 mg/dL, respectively). It is interesting to note that in all four quartiles the average HbA1C value ranges from 6.58% to 6.94%. In Table 2, we observe that, although we might expect correlations to emerge, we found none, probably due to weaknesses in the design of the research.

Conclusions

This study indicates that factors related to the manifestation of metabolic syndrome may be present with an increased incidence in the examined population. Due to the peculiarities and the special conditions under which prisoners live, a new, larger scale study which it would also take into account somatometric indicators, may offer important information about the health profile of this special population.

Table 1	Q1	Q2	Q3	Q4	p-value
AGE	50,26±11,63	53,31±13,05	50,76±14,89	56,53±14,83	0,310
GLU	125,63±45,20	130,35±48,56	129,85±53,53	138,97±63,68	0,820
URIC ACID	3,83±0,70	5,39±0,25	6,01±0,23	7,48±0,73	**<0,01**
TCHOL	179,75±52,26	186,88±51,20	185,42±30,89	195,97±41,22	0,563
TRG	98,92±31,95	138,18±107,82	116,65±48,80	132,97±55,59	0,134
HDL	48,13±12,33	48,47±11,18	46,58±11,75	44,79±6,17	0,553
LDL	111,84±46,31	110,78±44,98	115,52±26,78	124,59±37,25	0,541
HbA1C	6,87±2,04	6,58±1,64	6,68±1,85	6,94±1,67	0,900
TyG	4,66±0,26	4,77±0,36	4,73±0,33	4,83±0,30	0,244
TG/HDL-C	2,21±0,93	3,22±3,13	2,78±1,61	3,06±1,44	0,243
N	24	17	26	33	
P	0,24	0,17	0,26	0,33	

Table 2	AGE	GLU	URIC ACID	TCHOL	TRG	HDL	LDL	HbA1C	TyG	TG/HDL-C
ΗΛΙΚΙΑ	1									
GLU	0,224	1								
URICA	0,104	0,09	1							
TCHOL	0,082	0,095	0,095	1						
TRG	0,081	0,127	0,189	0,375	1					
HDL	0,047	-0,216	-0,182	0,198	-0,302	1				
LDL	0,053	0,123	0,094	0,952	0,177	0,055	1			
HbA1C	0,143	0,782	0,017	0,008	0,088	-0,253	0,047	1		
TyG	0,240	0,695	0,197	0,32	0,745	-0,363	0,215	0,556	1	
TG/HDL-C	0,068	0,179	0,172	0,295	0,943	-0,531	0,166	0,145	0,723	1

eP77

GnRH ANALOGUE CONJUGATED WITH MITOXANTRONE: A NOVEL AGENT WITH ANTITUMOR ACTIVITY

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Introduction

Breast cancer is one of the most common types of cancer in women. Even though many new treatments have been developed, the incidence of this disease is increasing every year.

The gonadotropin releasing hormone GnRH is a decapeptide that interacts with the GnRH receptor (GnRH-R). Importantly, GnRH-R is highly expressed in various types of cancer cells, such as breast cancer cells. According to studies, GnRH analogues have antiproliferative effect in breast cancer cells, through their interaction with the GnRH-R.

Aim

The aim of this study was to develop novel GnRH analogues with increased antiproliferative effects, by conjugating them with the cytotoxic agent, mitoxantrone. Thus, we created the analogue Con7. Con7 was expected to release mitoxantrone into cancer cells after its binding to GnRH-R and as a result internalize the GnRH-R/Con7 complex.

Methods

To create the Con7 analogue we chemically modified the GnRH analogue, leuprolide and conjugated it with mitoxantrone. We incubated the breast cancer cell line MDA-MB-231 with increasing concentrations of Con7 for four days, in order to examine its antiproliferative properties. The MTT assay was used to measure the proliferation rate of the cells.

Results: The novel GnRH analogue Con7 inhibited the proliferation of MDA-MB-231 cells in a time-dependent manner (1-4 days). Con7 showed its antiproliferative effect at day 3 and 4. Con7 reduced the proliferation rate at M.

Conclusions

Proliferation of MDA-MB-231 cells was inhibited by Con7 in a time-dependent manner, reaching its maximum inhibitory effect at day 4. Moreover, the inhibitory effect of Con7 was comparable to that of unconjugated mitoxantrone, a known anticancer drug. Additional studies are in progress to investigate the effect of Con7 on the migration rate of breast cancer cells.

eP78

EARLY URINARY MARKERS FOR RENAL INJURY IN OVERWEIGHTED AND OBESE CHILDREN AND ADOLESCENTS

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Introduction

Recent data indicate that overweight and obesity contribute to increase of chronic kidney disease (CKD) incidence in children and adolescents.

Aim

To examine of several markers for early detection of renal injury in overweight and obese children and adolescents.

Material and methods

In total 64 overweight and obese children and adolescents with BMI (body mass index) higher than 25 Kg/m2 and 50 healthy children and adolescents with normal body weight, 5-19 years old, matched for gender and age, were enrolled in the study. In the second morning urine levels of KIM-1 (kidney injury molecule-1) were determined with ELISA method, the activity of the enzyme NAG (N-acetyl-beta-D-glucosaminidase) with spectrophotometric method and microalbumin with chemiluminescent method. Serum concentrations of creatinine, urea, uric acid, fasting glucose and insulin, and parameters of lipid profile, and urine creatinine level were determined by using of standard biochemical methods. Insulin resistance was estimated using homeostasis model (HOMA-IR).

Results

No significant difference were found between two groups of subjects for serum creatinine, glucose, urea, uric acid and for lipid profile parameters ($p > 0.05$). Urine KIM-1/Cr and NAG/Cr ratio were found significantly higher in the group of overweight and obese children and adolescents. The modest significant correlation was detected in this group between urinary microalbumin/Cr ratio and KIM-1/Cr ($r=0.53$, $p<0.05$). There was also significantly high correlation ($r=0.83$, $p<0.05$) between KIM-1/Cr and NAG/Cr ratio, as well as between microalbumin/Cr and NAG/Cr ratio ($r=0.76$, $p<0.05$). Insulin resistance was detected in 53% of overweight and obese children and adolescents. No significant difference was detected between subjects with or without insulin resistance for KIM-1, NAG and microalbumin.

Conclusion

The results have shown that urinary KIM-1, NAG and microalbumin are promising markers for detection of early renal injury in overweight and obese children and adolescents, that could be used with aim to prevent development and progression of chronic kidney disease.

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REFUGEES: A CHALLENGE TO THE HEALTHCARE SYSTEM

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Since more than two years, Lebanon has been facing one of the most difficult economic crises while hosting more than two million refugees. The healthcare system has been particularly impacted and is trying to survive and fulfill the needs of citizens while also being burdened by the health requirements of refugees. Laboratories which are a cornerstone of the Lebanese health system have, in turn, witnessed an increase in the number of tests amid a major increase in costs and a limitation of resources. Our aim is to determine the most commonly ordered laboratory tests for refugees and their impact on the healthcare system.

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