



# Laboratory & Diagnosis



Official Journal of the Iranian Association  
of Clinical Laboratory Doctors

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Iranian Scientific Association of  
Clinical Laboratory Doctors

## CONGRESS ABSTRACTS



# Laboratory & Diagnosis

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**Official Journal of Iranian Association of Clinical Laboratory Doctors  
Supplement Issue for IQC 19**

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## Message of Congress Chairman



**Dr. Sh. Hemmati**  
**DCLS**

### **In the Name of God**

The 19th Congress on Quality Improvement in Clinical Laboratories is undoubtedly one of the largest and most important congresses in field of laboratory sciences, which is held every year on the occasion of Laboratories Day. With the outbreak of COVID-19 in 2021, this congress was held virtually and was welcomed as the first virtual congress across the country. In spite of the shortcomings in its establishment, mostly due to weaknesses and uneven availability of communications infrastructure, post-event evaluation surveys proved the conspicuous success of the event as being complete and satisfactory in all respects. We hope to hold a hybrid congress in 2022 so that our colleagues can avail themselves of the workshops as well. In the current congress, several international scientists and experts in various fields of laboratory sciences are invited to have lecture on a number of topics. Thanking the pioneers and colleagues who have held the previous Quality Improvement Congresses so far, the motto of the first congress entitled “The quality has got no end” was suggested by our dear colleague Dr. Farsi, and in my opinion, the words added to its scientific richness inasmuch as over several thousand people annually visit the congress and its side exhibition. Based on cutting edge scientific discussions about new approaches in diagnosis in the scientific panels of the Congress, clinical diagnostic equipment and kits have been imported into the country in the soonest possible time by the representatives of reputable companies and are now being used by laboratory colleagues. The pace of growth and quality development of the country’s laboratories has been accelerated in such a way that in a short period of time, our colleagues established the setups and devices in all parts of the country to reduce the suffering of the people in depressed areas and prevent them from referring to the provincial centers. Unfortunately, in the last two years, due to the COVID-19 epidemic, illegitimate and cowardly sanctions, consequent unbridled inflation, and lack of a proportional increase in laboratory tariffs, the import of new devices and technologies and diagnostic kits to Iran has decreased. Moreover, some obstacles pertain to depreciation and wearing



out of equipment and machinery in conjunction with the multiplication of the kits and consumables prices, which may culminate in extended closure of small and medium-sized laboratories; therefore, the qualitative and quantitative growth that has occurred in the last twenty years will be futile. The terrible consequence is serious crisis in deprived areas in accessibility of laboratory services. In the 19th Congress, important scientific issues, including the big data project on the study of the reference range, being carried out by Iranian Association of Clinical Laboratory Doctors for the first time in the country, will be examined by domestic and foreign experts. Other topics such as pathogenic, diagnostic, and epidemiological aspects of COVID-19 and other topics in the field of hematology will also be commented and discussed. I wholeheartedly appreciate the assistance of Dr. Zarnani who delicately selected topics and arranged the panels and Dr. Bagheri, the esteemed executive secretary of the congress, who planned and implemented the preliminary arrangements of the event. One of the prominent aspects of this congress is that in each panel, clinical colleagues together with laboratory colleagues will discuss the issues raised from clinical and laboratory aspects. In the end, I wish God Almighty bestow health and success to colleagues and all participants of the congress who incessantly work and conduct research for the wellbeing of individuals.

## Message of Congress Secretaries



**Dr. A. H. Zarnani**  
**DCLS, PhD**



**Dr. S. M. Boutorabi**  
**DCLS, PhD**

### In the Name of God

Congresses on Quality Improvement in Clinical Laboratories (QICL) are the most influential and most visited scientific events in laboratory sciences in the country, which bring together the lab staff and provide an ideal opportunity to exchange the latest scientific findings. Since establishment of the congress in 2002, the main goal has gyrated around the improvement in quality of services in medical diagnostic laboratories and localizing the knowledge relevant to the field of laboratory sciences through an organized system of continuing medical education. The COVID-19 pandemic in early 2020, in addition to the dramatic economic and social effects, has had undeniable effects on the attendance to scientific events. This caused the 18th congress to be held virtually with one year delay in 2021. Now with continuation of COVID-19 pandemic, the 13th international & 19th national QICL congress will again be held online in 2022. Although virtual congresses have their own shortcomings including impossibility of interactive and face to face scientific discussions, they have some undeniable advantages. Energy saving, lowering carbon footprint, increased attendance; especially from countries with low-to-middle income and promotion of the diversity of participants and speakers are among superiorities that virtual congresses could potentially have over traditional face-to-face meetings. The 13th international & 19th national QICL virtual congress gives us great pleasure to invite national as well as participants from all over the world to exchange knowledge on different aspects of clinical laboratory sciences. This congress will bring together clinical laboratory managers, technicians, students, researchers and clinicians to maximize our mutual understanding of medical laboratory sciences and clinical dimensions. This congress, as an integrated event, will leverage the expertise of faculty speakers and the scientific committee allowing delegates to benefit from a wealth of knowledge in clinical laboratory sciences. Under the theme “Big Data in Laboratory Medicine” this congress will try to draw attention of clinical laboratory professionals and authorities for taking advantages of millions of clinical laboratory data generated each day for figuring a better health program out.



The project “IRLAR” (Iranian laboratory reference intervals) was approved and launched in 2020 by Board of Directors, The Iranian Association of Clinical Laboratory Doctors (IACLD). It is our great pleasure to present the results of this project for popular biochemical analytes in QICL 2022. The 13th international & 19th national QICL congress, besides providing an opportunity for participants to benefit from the latest lab achievements in all aspects of clinical laboratory sciences, will also focus on laboratory challenges in diagnosis of COVID-19 and vaccine-induced protective immune responses. The congress is strongly supported by many healthcare professionals, institutions, research centers and medical universities and companies actively involved in production of laboratory equipment and products. We are eagerly waiting to welcome you in QICL 2022 and your contribution to the success of the 13th international & 19th national congress on quality improvement in clinical laboratories.

## Message of Congress Executive Secretary



**Dr. K. Bagheri**  
**DCLS**

The 19th Congress on Quality Improvement in Clinical Laboratories will be held while we have been facing colossal challenges to overcome; late issuance of the permit for establishing continuing medical education credits and programs and the permit for side exhibitions of laboratory equipment from Food and Drug Administration of Iran on the one hand and on the other hand limited capacity of Milad Tower Conference Center in providing suitable accommodation commensurate with the requirements of the congress besides the need for swift adjustments and modifications in infrastructures of Iran Mall Event and Convention Center to set the stage for conducting some topics in virtual format have all made the involved professional team to work incessantly, day and night, to organize the congress in the best possible way for the audience. Therefore, after two years of interruption in holding the Congress in physical format due to COVID-19 restrictions in the community, the opportunity is now provided to establish the event in a suitable, calming, and relaxing environment. Unfortunately, the establishment of Congress in 2022 coincides with lack of allocation of foreign exchange by government to kits, supplies, and laboratory equipment, as well as a huge increase in daily costs of laboratories in all aspects, including personnel, energy carriers, and rising prices of consumables. If the laboratory tariff is not adjusted in proportion to these costs, medical diagnostic laboratories and companies supplying the equipment will soon encounter irreparable harms and deplorable conditions, and it is predicted that the field of laboratory diagnostics and subsequent treatments will suffer from a major failure. Therefore, we deeply believe that the 19th Congress on Quality Improvement in Clinical Laboratories will play a fundamental role in reflecting the views and problems of the laboratory community through the large participation of colleagues, private and public sector officials, news agencies, and mass media.



## Congress Main Topics

### Main Topics

### Coordinator

Advances in QC tools and techniques in medical laboratory

Dr. H. Bayat

Artificial intelligence, data science and laboratory medicine

Dr. M. Fatehi

Autoimmune disorders of coagulation

Dr. A. Shamsavari

Biomarkers of drug abuse in clinical laboratory  
(alcohol, opioids and psychotropic substances)

Dr. A.R. Timcheh-Hariri

Challenges in laboratory diagnosis of STD

Dr. T. Soori

Chemical-microscopic testing of biological liquids

Dr. M. Vanaki

Clinic and laboratory dialogue: current trends  
in the lab diagnostics of autoimmune disease

Dr. N. Kianmehr

Clinical and fundamental studies of tumor markers

Dr. A.R. Lotfikian

COVID-19 vaccination updates with emphasis  
on medical laboratory aspects

Prof. A. Harandi

Diagnosis of systemic mycoses and quality control in mycology lab

Dr. M. Ghahri

Ensuring biosafety in the activities of clinical diagnostic laboratories

Dr. K. Khodaverdian

Geriatric medical laboratory

Dr. F. Mirzadeh

Hematologic neoplasm (ALL, AML)

Dr. S. Hosseini



### Main Topics

### Coordinator

Lab investigation of prenatal genetic disorders

Dr. D. Omrani

Laboratory Investigation of endocrine disorders in pregnancy

Dr. H. Chehreh-Gosha

Lesson learned from COVID 19: from bench to bed Side

Prof. A.R. Ranjbar

Lipid profiling in clinical laboratory

Prof. M. Doosti

Microbiological diagnostic of tuberculosis and mycobacteriosis

Dr. GH.R. Hamzehloo

Modern technologies in laboratory hematology

Prof. A. Gharehbaghiyan

Omics technologies in medical laboratory

Prof. H.R. Khorram-Khorshid

Sepsis and systemic inflammation:  
laboratory verification of diagnosis

Dr. M. Mardani

Serologic diagnosis of infectious disease  
(H. Pylori and Brucellosis)

Dr. M. Douraghi

The power of big data in laboratory medicine:  
establishing reference intervals

Dr. R. Mohammadi

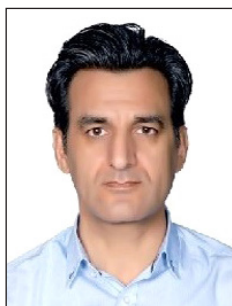
Updates on hepatitis B and C vaccine and laboratory diagnosis

Prof. F. Shokri

New findings laboratory science (Young scientists session)

Dr. F. Aziz Mohseni

## Coordinators of Congress Main Topics (in Alphabetic Order)



**Dr. H. Bayat**  
DCLS



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MD



**Dr. A. Shahsavari**  
PhD



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DCLS, PhD



**Dr. T. Soori**  
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**Dr. M. Vanaki**  
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**Dr. M. Doosti**  
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**Dr. Sh. Hemmati**  
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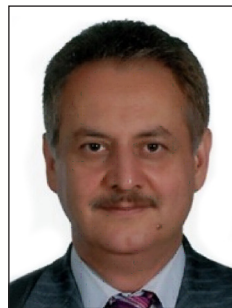
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**Dr. A. Gharehbaghian**  
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**Prof. H.R. Khorram-Khorshid**  
**MD**



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**Dr. R. Mohammadi**  
**PhD**



**Prof. F. Shokri**  
**PhD**



**Dr. F. Aziz Mohseni**  
**DCLS**



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# ABSTRACTS

**The 13th International & 19th National Congress  
on Quality Improvement in Clinical Laboratories**



# Oral Presentations

# Advances in QC Tools and Techniques in Medical Laboratory

## O1 – O4

Different aspect of medical laboratory, including quality control, are experiencing a fast improvement and evolution. New concepts and approaches, hard-ware and soft-ware improvements in IT, increased ability in manufacturing reference methods/instruments and material, as well as executing various standardization/harmonization programs are of the factors leading to the recent improvements in quality control in medical laboratory. The session “Advances in QC tools and techniques in medical laboratory” will address some parts of these improvements.

Professor Egon Amann, co-chair of the IFCC TF-GLQ (Task Force on Global Laboratory Quality), will present the goals, activities, and achievements of the Task Force.

Professor Marc Thelen, chair of the Netherlands’ EQA program (SKML), having a vast knowledge and expertise in establishing and improving one of the most credible EQA program, will present on how to plan EQA programs according to the intended purpose.

Dr. Huub V. Russom, a member of the IFCC work group on patient-based QC, will lecture about the recent improvements, especially via using computer simulation, in moving average technique using patients’ results, and will address advantages and shortcomings of this approach.

Hassan Bayt, will talk about the ISO/TS 20914(2019) document for estimating analytical measurement uncertainty in medical laboratory, and will address different characteristics of this procedure, especially its simple and practical approach.

O1

### **IFCC's Global Initiative to Enhance Clinical Laboratories' Quality in Developing Countries**

**Egon Peter Amann 1 \***

1- Philipps University Marburg, Germany and University of Applied Sciences, Hamm-Lippstadt, Germany

Background One of IFCC's important goals is to improve general lab quality in Clinical & Medical Laboratories worldwide, and in particular in developing countries. In previous years, the "Committee of Analytical Quality" (C-AQ) and the Working Group "Developing Quality Competence in Medical Laboratories" (DQCML) were chartered with this task. At the end of 2020 both initiatives were terminated and replaced by the "Task-Force on Global Lab Quality" (TF-GLQ). The TF-GLQ was chartered to identify countries around the world willing to improve their IQC lab standards and to participate in EQA/Proficiency Testing programs. Initially, ten countries with five labs each were identified for a pilot phase, in which IQC and EQA materials would be provided by the IFCC free of charge for the participating laboratories. On-site, hands-on training courses, conducted by TF members, are planned during the 1-year pilot phase. After the 1-year pilot phase, an in-depth analysis – based on pre-defined performance indicators (KPI) – shall be conducted. If the program appears to be successful, IFCC intends to roll out an expanded program to additional countries & labs. Achievements In 2021, a global IQC & EQA survey was conducted, in order to identify suitable countries & labs participating in the initial pilot phase. Based on the result of the survey, a priority list of ten countries willing to participate in the 1-year pilot phase was developed. Country coordinators were identified serving as a "country – IFCC" liaison managers for the pilot program. A list of assays of high probability of demand/usefulness for the 1-year pilot phase, considering country specific needs. Potential vendors for the IQC and EQA programs were identified and two rounds of bidding processes were performed. After a thorough analysis, a commercial vendor was chosen as the preferred IFCC partner for the EQA pilot program. For the IQC materials (these require daily laboratory applications and thus are more expensive compared to the EQA materials, which require only 4 x a year shipment to labs) it was decided to investigate first the individual conditions and needs of the participating labs and to devise subsequently a more customer-oriented approach. Plans for 2022 Develop jointly with the commercial vendor the EQA program for the 1-year pilot phase. Develop and conduct general IQC and EQA training programs for participating countries. (Prioritization, number and duration of such trainings needs to be worked out). Develop and conduct WEBINARS on IQC and EQA for participating countries. Draw conclusions & learnings from the 1-year-pilot phase to be applied for an expanded roll-out of the program to additional countries.

**O2**

## How to Make EQA Fit for Purpose

**Marc Thelen 1 \***

1- European Specialist in Laboratory Medicine, Clinical Chemist Amphia Hospital, Breda, the Netherlands

ISO 15189 requires laboratories to participate in a between-laboratory comparison, but what is the thought behind that? Between-laboratory comparison is not a purpose by itself. Its role is to reduce the risk for remaining inaccuracy after other measures to manage accuracy have been implemented. The first step to control accuracy within acceptable limits for the intended use is set during the validation or verification of method. Then the responsible specialist in laboratory medicine defines the analytical performance specifications (APS) for a measurand for a particular intended use and then determines whether the found analytical performance characteristics (APC) meet the APS. Once that is achieved the method can be accepted for patient care knowing that accuracy meets the set requirements. To establish that the performance as verified continues to meet the APS, ISO 15189 requires laboratories to use a quality control program. If such internal quality control procedure would have no risk to miss or misinterpret the introductions of new sources of inaccuracy, no extra policies like EQA would be necessary. But internal control policies are not perfect and do tend to miss or misjudge sources of inaccuracy in some occasions. Especially immunoassays are vulnerable to the incommutability of commercial IQC materials. Lot changes in reagent or calibrator could introduce a shift in patient results that is missed in IQC results. That is where EQA comes to stage, but EQA is not less vulnerable to the shortcomings of IQC if no attention is given to requirement for its role in the quality system. To be useful, the EQA needs materials that are commutable, which means that they have between method differences that are identical of that patient samples. Such materials would deserve value assignment in a reference method which would allow laboratories to judge the persistence of the metrological traceability of their method. In order to take full profit of the information provided by measuring commutable sample with value assignment in reference methods also a report format is needed that discloses all the information within the data. Optimal EQA reports show calculations of regression statistics between participants' data and target values as a measure for trueness and also calculates the dispersion around this regression as a measure for imprecision. Such EQA reports give direction to required corrective actions of a laboratory and also allows laboratories to judge whether previous corrective action haven been successful.

O3

### Technical Quality Assurance and Control Using Patient Moving Average

Huub Van Rossum 1 \*

1- Specialist in Laboratory Medicine and Clinical Chemistry at the Netherlands Cancer Institute

In the last couple of years new insights in the understanding and practical application of patient based real time quality control (PBRTQC) have been obtained. PBRTQC differs in several ways from statistical internal QC and combining both techniques allows improved analytical quality assurance and a more (cost-) efficient QC plan. Challenges for laboratories are how to obtain proper laboratory specific settings, how to intergrade these new techniques with standard QC and how to operate PBRTQC in routine practice. Recently, tools and documentation that addresses these issues has become available for medical laboratories, amongst others via an IFCC working group. This talk will briefly overview these recent developments, increase the understanding and potential application of PBRTQC techniques and to provide guidance when interested in implementing patient-based real-time QC on medical labs.



O4

### Measurement Uncertainty: ISO/TS 20914:2019 Document

Hassan Bayat \*

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In the recent years, especially after being considered mandatory in the ISO15189 document, the estimation of uncertainty of results in the medical laboratory has been a challenge and hot topic for the laboratorians. In respond, over the past years different organizations and expert bodies have introduced several protocols each having pros and cons; some not appropriate for the medical laboratories. To address this challenge, in 2019 the ISO organization published ISO/TS 20914(2019) document specifically devoted to estimating the uncertainty of analytical performance in medical laboratory. The protocol recommended in this document is based on a practical approach that is tailored appropriate to the abilities and the information accessible to laboratories. It lets the laboratories, using the IQC results and the uncertainty of calibrators presented in the calibrators certificates, estimate uncertainty without needing complicated mathematical calculations.



# Autoimmune Disorders of Coagulation

05 – 06

**O5****Laboratory and Blood Bank Aspects in Acquired Hemophilia A****Nader Vazifeh Shiran \***

1- School of Paramedical Sciences, Shahid Beheshti University of Medical Sciences, Tehran, Iran  
Nadershirani@yahoo.com

The main pathway of coagulation in the body is the internal pathway, the most important clinical factor of which is F-VIII. Factor VIII has the lowest amount and half-life in plasma and because it has a large gene with 26 exons and is encoded on the X chromosome, its mutation in males can significantly reduce the amount of F-VIII and Causes hereditary hemophilia. With the same approach, F-VIII has antigenetic properties due to its glycosylated domain B and A2 and binding to the antigenetic VWF, and its autoantibody or alloantibody can also cause dysfunction and symptoms of acquired hemophilia. Diagnosis, treatment and monitoring of acquired hemophilia are different from inherited type and in acquired hemophilia it is also important to evaluate the level of Bethesda, because if it increases above 10 Bethesda, the type of treatment change from injection of factor VIII to plasmapheresis, immunosuppression and injection of FEIBA and IVIG. In this lecture, we try to discuss the laboratory aspects, blood bank and monitoring of acquired hemophilia.

O6

### **Rotational Thromboelastometry (ROTEM)-Based Coagulation Management in Cardiac Surgery and Major Trauma**

**Robert Haywood Pyle \***

For major bleeding related to severe trauma, major surgery, or chronic anticoagulation, a rapid assessment of hemostatic function is crucial so that optimal fluid replacement and blood transfusion can be administered without delay. Obtaining conventional laboratory tests, such as the prothrombin time (PT), activated partial thromboplastin time (aPTT), and fibrinogen level, during acute bleeding may not be particularly useful in predicting bleeding after trauma or invasive procedures. The ROTEM analyzer is used to guide physicians in patient blood management. It can reduce unnecessary transfusions, re-operation and infection rates, hospital length of stay and total hospital costs and mortality. As a result, hospital efficiency and patient care are enhanced. Thromboelastography facilitates the management of bleeding episodes as they can occur in clinical situations such as: Cardiac and vascular surgery, Organ transplantation, Trauma, Abdominal surgery, Tumor removal. This discussion will provide an introduction into the technique of Thromboelastography and its application and advantages in patient care and transfusion practices.

# **Biomarkers of Drug Abuse in Clinical Laboratory (Alcohol, Opioids and Psychotropic Substances)**

## **07 – 09**

Drug and alcohol addiction is a common phenomenon in the contemporary world and is currently one of the most costly problems in societies, especially in Iran. Statistics show that there are two million addicts addicted to drugs and psychotropic substances and 6 million addicts in Iran, but according to the evidence, the statistics are there are about 80,000 drug-related prisoners in Iran, or about half of the prison population.

Regarding alcohol addiction, according to statistics, in 2015, only 10,265 deaths due to traffic accidents occurred due to alcohol consumption in drivers in the United States, and in general, one third of traffic accidents are due to alcohol consumption. In many developed countries, by adopting a medical model, a new situation has been provided for addicts to be able to change the existing situation by using medical services. However, this issue depends on the recognition of biomarkers and the correct diagnosis of alcohol, drugs and addictive drugs. This panel aims to discuss the markers of drug and alcohol abuse as well as legal and regulatory challenges.

O7

### **Biomarkers and Laboratory Methods of Alcohol Abuse**

**Alireza Timchehhariri 1 \***

1- Head of Khorasan Razavi Forensic Medicine Laboratory

Methods of measuring liquids and alcoholic products from the point of view of laboratory sciences on the one hand, as well as legal restrictions and establishing this relationship to provide a scientifically documented and defensible laboratory answer in these examples are presented and discussed. In what samples do we want to examine the presence of alcohol, including non-biological samples of liquids and biological samples Depending on the above categories, laboratory methods are different, including screening methods and confirmatory methods. Diagnosis methods depend on the type of sample that is non-biological or biological and includes living or corpses that are different. How to diagnose and identify alcohol according to the tests and diagnostic methods in the laboratory and under what circumstances we should perform these diagnostic methods Suspicion of alcohol use, trauma related to injury, monitoring of the disease being treated for alcohol use. What criteria and tests can guide you? Determination of alcohol in blood by chemical and chromatographic methods as well as measurement of alcohol in exhaled air are among these methods. Methods for detecting alcohol in non-biological fluids as well as methods for determining the percentage of alcohol each are discussed separately

**Keywords:** Suspicion of Alcohol Use, Trauma Related to Injury, Monitoring of the Disease Being Treated for Alcohol Use

O8

## Biomarkers of Substance use Disorder: A Review and Update

**Kambiz Soltaninejad 1 \***

1- Department of Forensic Toxicology, Legal Medicine Research Center, Legal Medicine Organization, Tehran, Iran

**Background:** Substance Use Disorder (SUD) refers to a range of medical condition involves heavy or frequent substance use even when it causes mental and physical problems. The use of sensitive and specific biomarkers to diagnose SUD is necessary due to the growing concern of this problem around the world. **Method:** A literature search was performed on PubMed and Google Scholar databases. The terms used for the search were: “Alcohol Use Disorder”, “Substance Use Disorder” and “Biomarker”. The search was limited to years 2010 to March 2021. **Results:** Among the specific biomarkers of ethanol consumption such as ethyl glucuronide/ sulfate levels (in blood, urine, hair and nails), the concentration ratio of 5-hydroxytryptofol to 5-hydroxy-indole acetic acid (5-HTOL / 5-HIAA) (in urine), phosphatidyl ethanol (in blood), fatty acid ethyl esters (in tissue) and level of phosphatidyl ethanol in blood as a specific and sensitive biomarker for identification of people with high and heavy alcohol intake. New studies suggest that DNA methylation in peripheral blood monocytes and blood cortisol level are considered as new biomarkers for diagnosis of alcohol use disorder. Besides, dynorphin and kappa-opioid receptor level in the lymphocytes and plasma and metabolic phenotyping are suggested as biomarkers of opioid use disorder in recent years. **Conclusion:** Considering the prevalence of SUD in the community, the need to familiarize and method development for measurement of specific and sensitive biomarkers for the diagnosis of SUD in the country seems necessary.

**Keywords:** Substance Use Disorder, Ethanol, Opioid, Biomarker

O9

### Drug and Alcohol Abuse: Legal Challenges

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1- General Department of Forensic Medicine of Khorasan Razavi, Laboratory, Mashhad, Iran

**Introduction:** Criminal law deals with the deterrent aspects and civil law with its consequences for the use of drugs and alcohol, such as the restriction of individual and social rights of drug users. Proof of drug and alcohol use in the Iranian judiciary is one of the important cases in which neglecting the proper process will lead to the violation of the civil rights of the accused and the creation of possible legal responsibilities for the authorities and public and private laboratories. These defendants should be referred to the competent authorities, such as a forensic medical laboratory, in accordance with legal procedures and pre-determined. **Method:** This article contains texts from the Civil and Criminal Laws of the Islamic Republic of Iran and instructions and sections of the Ministry of Health and Medical Education and the Forensic Medicine Organization, as well as available and published online resources such as Google Scholar and Magiran databases to search for keyword related articles. Drug abuse, criminal law, civil law, drug testing, alcohol testing, forensic laboratory have been used. **Results:** Judicial authorities, based on the results of the experiments of the laboratories of the Forensic Medicine Organization, issue a verdict on the use of drugs and alcohol. Familiarity with the rules and procedures in conducting these tests will have a great impact on reducing responsibilities and preventing legal challenges. **Conclusion:** Awareness of the process of proving drug and alcohol use in competent laboratories and the legal effects and consequences of introducing, accepting, sampling, testing and issuing answers to defendants will reduce legal and regulatory challenges in public and private medical diagnostic laboratories.

**Keywords:** Drug Abuse, Criminal Law, Civil Law, Drug Testing, Alcohol Testing, Forensic Laboratory

# Challenges in Laboratory Diagnosis of STD

## O10 – O13

Sexually transmitted infections (STD) are increasing in the last decades. It is obvious that sexually transmitted infections prominently impact the health of the communities.

We face a wide range of complications from stigmatization to mortality, morbidity due to acute and chronic pelvic inflammatory diseases, reproductive problems and also infections in new-borns.

some of them induce ulcers in the genital areas, among them infection with human papilloma virus (HPV) is the most common sexually transmitted infection in the world. this infection can be presented as asymptomatic infections or as genital wart .Genital herpes is cause by herpes simplex virus. This virus have types 1 and 2. Molluscum contagiosom is a viral infection may be transmitted by sexual ways. Some of them cause discharge such as gonorrhoea, chlamydia, trichomoniasis. In this panel we discuss the challenges in different diagnostic tests for detecting sexually transmitted organisms. Syphilis ,chancroid, lymphogranuloma venereum are STIs that we discuss their clinic-laboratory challenges. The overall goal of HIV testing and counseling (HTC) for a national program is to identify more and more people living with HIV as soon as possible and to successfully connect them to care and treatment services.HTC is never mandatory, and stigma and discrimination should always be avoided, and satisfaction, reassurance, advice, accurate testing methods, and connection to care centers should be considered for each individual.

O10

### **Challenges in Clinical-Laboratory Diagnosis of STD s with Discharge (Chlamydia, Gonorrhoea, Trichomoniasis)**

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We face a wide range of complications from stigmatization to mortality, morbidity due to acute and chronic pelvic inflammatory diseases, reproductive problems and also infections in newborns. Because of the expanding border of microbiological tests and diversities in costs and geographically availability in different countries and in order to choose proper antibiotics according to stewardship rules, clinicians have to be oriented to the exact types of laboratory tests for suspected infections and also know how to analyse the conflicting results in some cases. It is planned to discuss the challenges in different diagnostic tests to detect secreting STIs (gonorrhoea, chlamydia, trichomoniasis). Testing for chlamydia trachomatis is indicated in patients with urogenital, anorectal, or ocular symptoms. Diagnostic tests include direct methods: culture, antigenic test like (EIA), direct fluorescent antibody, immune chromatographic RDT, nucleic acid hybridization and indirect methods depend on detection of antibodies. Diagnosis of Neisseria Gonorrhoea (Ng) presently includes different techniques. Microscopies have rapid results but lack of sensitivity. The most sensitive test to detect Ng is nucleic acid amplification test (NAATs). New development of molecular techniques represents rapid detection. Trichomoniasis is an infection with purulent foul-smelling vaginal discharge, dysuria, and dyspareunia. On the old, most cost effective and reliable test, wet mount examination, jerky, flagella movement of organism can be found. Culture, the most specific, nucleic acid detection, the most sensitive test and antibody-based assays with determining recent from remote infection, are different techniques to reveal trichomoniasis.

**O11****Challenges in Clinical-Laboratory Diagnosis of STD s with ulcer. First Part  
(Syphilis, Chancroid, LGV)****Marjan Sohrabi 1 \***

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Syphilis is a systemic disease caused by a spirochete called *Treponema pallidum*; the most common mode of transmission has always been through sexual encounters nowadays transmission through transfusion has been reduced. Clinical stages of syphilis are divided in the order below: primary, secondary, tertiary syphilis and latent syphilis. Diagnostic and laboratory methods, serological tests, as well as the treatment methods used today will be discussed later. Chancroid is caused by a gram-negative bacillus called *Haemophilus ducreyi*, which causes one sole lesion with irregular borders, purulent and necrotic webs, or multiple ones but with a softer consistency compared to syphilitic canker. These lesions are accompanied with lymphadenopathy but without inflammation and they eventually leave a scar. Direct smear of lesion's exudate or biopsy is used to diagnose chancroid. Staining is not helpful in distinguishing genital lesions. This microorganism in the specific agar medium of Mueller-Hinton is seen in the form of Coccobacillus next to each other in a row. Lymphogranuloma Venereum (LGV) is caused by *Chlamydia Trachomatis*, in its early stages LGV's lesion resembles Herpes lesions; or its small and often asymptomatic papules. The lesion usually resolves before LGV being diagnosed therefore the patient presents only with inguinal lymphadenopathy which has become purulent and we will drain it in order to send the drained material for cultivation or molecular studies for diagnosing the disease; it is rarely detected by the shape of the lesions

O12

### Clinical and Laboratory Challenges in HIV/AIDS Diagnosis

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The overall goal of HIV testing and counseling (HTC) for a national program is to identify more and more people living with HIV as soon as possible and to successfully connect them to care and treatment services. There are several community-based and facility-based models for running HTC. Using different models to do HTC can bring us closer to the goal of public access to HIV prevention, testing, care and treatment services in all HTC models individual rights should be respected. HTC is never mandatory, and stigma and discrimination should always be avoided, and satisfaction, reassurance, advice, accurate testing methods, and connection to care centers should be considered for each individual. Today, the most important strategy to control the HIV epidemic is to treat PLWH. If PLWH are treated with antiretrovirals, they can live a healthier life by reducing the viral load to zero, and the risk of transmitting the disease to others is dramatically minimized. Some of the most important diagnostic challenges in the clinical and laboratory context include the following: The importance of using a single diagnostic algorithm to identify patients The difference between several diagnostic methods and the role of each method The importance of paying attention to the conditions and clinical findings in PLWH for identifying patients Diagnostic challenges in infants In this presentation, we try to address all of the above.

**O13****Challenges in Clinical-Laboratory Diagnosis of STD s with ulcer:  
(HPV, HSV, Molluscum)****Tahereh Soori 1 \***

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Sexually transmitted infections (STD) are increasing in the last decades. some of them induce ulcers in the genital areas, among them infection with human papilloma virus (HPV) is the most common sexually transmitted infection in the world. this infection can be presented as asymptomatic infections or as genital wart. Its diagnosis is mostly clinical. although sometimes another methods such as PCR or [atologic evaluation may be used. high risk types can be in association with genital cancers in the regions of cervix, vulve, penis and anal and also nasopharyngeal cancers. There are vaccins for preventing some types. Genital herpes is cause by herpes simplex virus. This virus have types 1 and 2. genital lesions is usually due to type 2. HSV infection remain permanently in the body and can be asymptomatic or presented as painful grouped vesicular lesions in genital areas. relaps is common. diagnosis is usually clinical ,although serologic and molecular methods(PCR) can be used in some cases .aktivirals such as acyclovir or valaciclovir can be used in symptomatic cases. Molluscum contagiosom is a viral infection may be transmitted by sextual ways. Diagnosis is mostly clinical .it has no definitive treatment and vaccine.

# Chemical-Microscopic Testing of Biological Liquids

## O14 – O17

Recognition of diagnostic challenges is a prerequisite for Standardization and harmonization of Body fluids Analysis in a medical laboratory. This study explains the challenges regarding the pre-analytic, analytic, and post analytical variables in body fluid analysis, and finally, provides guidance on how deviations might affect final interpretation of test results and change clinical decisions. Pre-analytic challenges include: Different approach of medical laboratories for acceptance and rejection criteria for body fluid samples, and Concession for acceptance of invasive body fluid, insufficient knowledge of clinical physician and nurses about body fluid specimen collection procedures, distribution in proper anti-Coagulant tubes, proper keeping body fluid samples in clinical wards and proper transportation and sample delivery to labs. The major Analytical challenge is the lack of competent staff with sufficient practical knowledge and awareness about unconformities related to body fluids, e.g. weakness of staff in differential diagnosis of microorganisms with artifact elements in gram stain in CSF analysis. Also, weakness of Education department of medical labs for planning effective training course, and obvious deviation between the results of manual and automated methods, especially in field of cytology of body fluid analysis are among the main challenges. The main challenge in post-analytical phase is lack of standard reference interval for types of body fluids that may cause misinterpretation of physician.

Conclusion: The main prerequisite for standardization and harmonization of body fluid analysis is recognition of unconformities in this field.

**O14****Diagnostic and Quality Challenges in Body Fluid Analysis****Mehrdad Vanaki 1 \***

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**Introduction:** Recognition of diagnostic challenges is a prerequisite for Standardization and harmonization of Body fluids Analysis\*[1] in a medical laboratory. This study explains the challenges regarding the pre-analytic, analytic, and post analytical variables in body fluid analysis, and finally, provides guidance on how deviations might affect final interpretation of test results and change clinical decisions. Pre-analytic challenges include: Different approach of medical laboratories for acceptance and rejection criteria for body fluid samples, and Concession for acceptance of invasive body fluid, insufficient knowledge of clinical physician and nurses about body fluid specimen collection procedures, distribution in proper anti-Coagulant tubes, proper keeping body fluid samples in clinical wards and proper transportation and sample delivery to labs. The major Analytical challenge is the lack of competent staff with sufficient practical knowledge and awareness about unconformities related to body fluids, e.g. weakness of staff in differential diagnosis of microorganisms with artifact elements in gram stain in CSF analysis. Also, weakness of Education department of medical labs for planning effective training course, and obvious deviation between the results of manual and automated methods, especially in field of cytology of body fluid analysis are among the main challenges. The main challenge in post-analytical phase is lack of standard reference interval for types of body fluids that may cause misinterpretation of physician. **Conclusion:** The main prerequisite for standardization and harmonization of body fluid analysis is recognition of unconformities in this field. [1] Body fluids Include CSF, Serous Body Fluid (Pleural, peritoneal, pericardial), Synovial fluids, etc.

**Keywords:** Body Fluid Analysis, Unconformity, Pre-Analytic, Analytic, Post-Analytic

O15

### Biochemical Analysis of Cerebrospinal Fluid, Quality Control and Its Clinical Applications

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Cerebrospinal fluid (CSF) is a clear, colorless fluid with a specific gravity of 1,007 that is produced from the choroid plexus of the ventricles or ventricles of the brain and circulates around the brain and spinal cord through the ventricular system. This fluid has various function, protects the brain and can remove harmful substances, including drugs. Helps maintain pressure in the cranial cavity and spinal canal at constant levels. It also carries hormones from where they are produced to the part of the brain where they are needed. The composition of CSF is similar to other extracellular fluids, but the concentrations of the constituents in these fluids vary. Laboratory reports of CSF degradation usually provide information on color, specific gravity, protein count, white blood cell count, glucose, and other electrolytes. In addition, CSF may be tested for immunoglobulins or lactate. Normal CSF contains a small number of white blood cells and no red blood cells. Autoimmune disorders, such as MS and Guillain-Barre' syndrome, can lead to an inflammatory reaction that can be detected in the CSF despite autoantibodies. CSF analysis for primary and metastatic cancerous tumors in the central nervous system, as well as by measuring the level of amyloid beta 1-42(A1-42) and p- and t-tau proteins, to diagnose Alzheimer's disease and to diagnose diseases Infections that cause meningitis and encephalitis are used.

**Keywords:** Biochemical Analysis, Cerebrospinal Fluid, Quality Control

**O16****Microbiological Study of Cerebrospinal and Other Sterile Body Fluids****Mohammad Rahbar 1 \***

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Infections of normally sterile body fluids such as cerebrospinal, pleural, peritoneal and synovial results in sever morbidity and mortality. Early detection of etiology agents of such infections is a very important issue for establishment of proper antimicrobial therapy and increasing of survival of patients. Care must be taken during specimen collection and transport to ensure that the specimen is not contaminated. Gram stain, culture and performance of molecular methods are the most important procedure in clinical microbiology laboratories. Microscopic gram stain examination of sterile body fluids remains the initial diagnostic test in the processing of specimens in the clinical microbiology laboratory. Culture of the specimen should include media for the detection of the most likely organisms to cause infection at this body site. In addition, surveillance of antimicrobial susceptibility is necessary to combat the emergence of resistance. The advent of molecular methods such as PCR and real- time PCR have resulted for the rapid identification of a number of infectious agents. Molecular methods have become increasingly incorporated into the clinical microbiology laboratory, particularly for the detection and characterization of virus and fastidious bacteria in sterile body fluids. The advantages of rapid turn-around time and high sensitivity and specificity are appealing however these methods must be matched by rigorous validation and quality control. Recently other new methods such as Matrix-Assisted Laser Desorption/Ionization Time-Of-Flight mass spectrometry (MALDI-TOF-MS) has been used for identification of microorganisms cultured on solid media in microbiology laboratories.

**Keywords:** Body Sterile Fluids, Molecular Methods

O17

### Cytological and hematological analysis of biological body fluids

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Biological body fluids include CSF and pleural, peritoneal, and synovial serous fluids, which are affected by transudates, exudates, infections, cellular infiltrates, and internal bleeding and undergo biochemical, cellular, and microbial changes. Cytological analysis of biological fluids is a topic that is less considered and taught, and because of its significant help in diagnosing of related diseases, it should be discussed and taught more. For cytological examination of fluids, manual counting by hemacytometer, Smear by cytospin, staining by wright Giemsa and methylene blue stain and microscopic differentiation were used. Traumatic tap diagnosis was performed based on the pattern of samples and cell count and true leukocyte correction was performed in case of traumatic sampling and cell differentiation was performed based on normal age range. In all body fluids, the number of leukocytes was less than 5/ul, but in the case of RBC, their number in CSF was less than 30/ul in infants, less than 5/ul in adults, and their number in serous fluids was less than 200/ul. Was. Diff monocytes and macrophages in infant CSF and serous fluids were often 65-75% and in adult CSF 15-45%. Neutrophil percentage was below 8% in infant and adult CSF and below 25% in serous fluid diffusion. Neutrophil percentage was 10-20% and 40-80% in neonatal and adult CSF and less than 5% in serous fluids, respectively. In case of traumatic tap during CSF sampling, WBC count should be corrected according to the formula: Corrected CSF WBC=  $WBC_f - (WBC_b \times RBC_f / RBC_b)$ . Cellular differentiation in adult CSF is different from that in infants, and serous fluid differentiation and CSF are different from blood differentiation, so that in all body fluids, the percentage of monocyte cells is higher than that of neutrophils. In serous fluids, due to exposure to the mesoderm membranes, mesothelial cells are sometimes seen, which must be differentiated from monocytes, and at the same time their malignancies must be distinguished from normal cells.

**Keywords:** Biological Fluids, Peritoneum, Synovial, Pleura, Cerebrospinal Fluid

# Clinic and Laboratory Dialogue:

## Current Trends in the Lab Diagnostics of Autoimmune Disease

### O18 – O21

Idiopathic inflammatory myopathies (IIMs) are a heterogenous group of autoimmune diseases with shared features of proximal muscle involvement, increased muscle enzymes and evidence of muscle inflammation on biopsy. The causes of IIMs are unknown. Patients typically present with subacute to chronic proximal muscle weakness.

Most of the patients have extra muscular features that may involve the skin, the esophagus, the lung, the heart and the joints.

IIM can be classified as primary and secondary.

Primary IIM are sub grouped as following categories:

Inclusion body myositis, immune-mediated necrotizing myopathies, dermatomyositis, polymyositis and anti-synthetase syndrome

IIM may be overlapped with some systemic rheumatic diseases such a systemic sclerosis and systemic lupuserythematosus.

In a minority of IIM, a malignancy with its specific feature may coexist.

Clinical features, laboratory investigation, Magnetic resonance imaging and electrodiagnostic study may help confirm the illness, although muscle biopsy remains the gold standard modality to differentiate other conditions with similar clinical manifestation.

This panel will provide an overall review on clinical assessment, serology and differential diagnosis of IIM.

O18

### Idiopathic Inflammatory Myopathies: Clinical Manifestations

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Idiopathic inflammatory myopathies (IIMs) are a group of autoimmune diseases with shared features of proximal muscle involvement and evidence of muscle inflammation. Muscle weakness is typically symmetric, proximal and can be very severe to very mild. IIM is a multisystem disorder with a different clinical symptom. Most of the patients have extra muscular features that may involve the skin, the esophagus, the lung, the heart or the joints. IIM can be classified as primary and secondary. Primary IIM are subgrouped as following categories: Inclusion body myositis, immune-mediated necrotizing myopathies, dermatomyositis, polymyositis and anti-synthetase syndrome IIM may be overlapped with some systemic rheumatic diseases such a systemic sclerosis and systemic lupus erythematosus. In a minority of IIM, a malignancy with its specific feature may coexist.



O19

## Idiopathic inflammatory myopathies, Differential diagnosis

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In patients suspected of Idiopathic inflammatory myopathies, initial evaluation including history, physical examination, selected laboratory tests as well as imaging and muscle biopsy may be needed to establish the diagnosis and rule out other conditions with similar manifestations.

O20

### Idiopathic Inflammatory Myopathies: Laboratory Investigation

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**Background:** Polymyositis (PM) and dermatomyositis (DM) are autoimmune myopathies with the systemic inflammatory condition that usually affects the muscle and also skin tissue for DM. **Material and methods:** The diagnosis of PM and DM isn't plain and displays a challenge for clinicians, it is based on the integration of several results including medical history, clinical presentation, and laboratory findings such as the presence of some autoantibodies and enzymes. **Results:** The autoantibodies related to PM and DM can be found at other inflammatory connective tissue diseases without myositis and muscle and skin involvement. Laboratory tests help in the diagnosis of PM and DM but they should always be assessed in the context of the clinical condition and other clinical tests and final decisions about diagnosis and treatment will be established on the incorporation of all results. **Conclusions:** This presentation focuses on laboratory findings including biochemical molecules and enzymes, immunogenetics of myositis, and emphasizes the importance of autoantibody detection in defining disease-specific phenotypes, helping to elucidate disease mechanisms, and definitive and rapid diagnosis of patients.

**O21****Myositis and malignancies****Anousheh Haghighi 1 \***

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The association of myositis with malignancy has been proven in various studies. Different types of malignancies are more prevalent in patients with myositis than in general population. This association is most commonly seen with dermatomyositis, but other types of myositis should be evaluated for underlying malignancy too. The most common prevalent malignancies associated with dermatomyositis are breast, ovarian, lung and pancreatic cancers. The onset of malignancy may be earlier, concurrent or following the onset of dermatomyositis. Recent studies show that the highest risk of malignancy is in the first year after dermatomyositis diagnosis. This risk remains high for the next two years and then decreases, but is still more than general population. Inclusion- body myositis is the least associated with malignancy among other types of myositis. It is recommended that all patients with myositis, especially dermatomyositis, be evaluated for underlying malignancy.



# Clinical and Fundamental Studies of Tumor Markers

## O22 – O26

Distant past, each type of cancer was treated the same in different patients. But today research has shown that each tumor has a unique features, even if it is a common type of cancer. Oncologists are increasingly using cancer bio markers to predict successful treatment for each cancer exclusively. Cancer biomarkers are biomolecules produced by the body or tumor in cancer patient. Testing biomarkers has revolutionized the treatment of many cancers. Biomarkers help identify tumor alterations. Biomarkers may be protein, DNA, RNA or metabolomic profiles that are specific to the tumor. Biomarkers testing can include the DNA sequencing, DNA or RNA tests to look for gene fusions, or tests to detect RNA or protein levels. Biomarkers are identified and measured for the following purposes: Individual risk assessment of tumor developing, Determining the individual risk of cancer recurrence, Predict treatment success for each specific patient, and treatment monitoring.

O22

## Cinnamaldehyde Reduces Glycolysis Pathway Through Inhibition of ErbB2/HSF1/ LDHA Pathway in 5637 Cell Line of Bladder Cancer

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**Background:** The growing prevalence of bladder cancer worldwide has become a major concern for researchers; and side effects of chemotherapy drugs have always been a major problem in the treatment of cancer. Cinnamaldehyde, the active ingredient in Cinnamon plant, has long been considered with anti-oxidant and anti-inflammatory effects. **Methods:** Bladder cancer 5637 cell line were treated with different concentrations of Cinnamaldehyde. MTT assay was performed to evaluate cell viability at 24, 48 and 72 hours. The concentration of 0.02, 0.04 and 0.08mg/ml of Cinnamaldehyde were selected. Apoptosis was assessed with Annexin V-FITC/PI and Hoechst33258 staining. Cell migration was performed by scratch test. In order to evaluate the effect of Cinnamaldehyde on glycolysis, the gene expression of epidermal growth factor receptor 2 (ErbB2), heat shock protein transcription factor-1 (HSF1) and lactate dehydrogenase A(LDHA) as well as protein levels of HSF1 and LDHA, LDH activity and finally glucose consumption and lactate production were measured. **Results:** Cinnamaldehyde significantly increased the rate of apoptosis in the 5637 cells ( $p<0.05$ ). Also, it significantly reduced the gene expression of ErbB2, HSF1 and LDHA, protein level of HSF1 and LDHA, LDH activity, as well as cell migration, glucose consumption and lactate production ( $p<0.05$ ). These changes were dose-dependent. **Conclusion:** Thus, Cinnamaldehyde induced apoptosis and decreased growth in 5637 cells by reducing ErbB2-HSF1-LDHA pathway.

**Keywords:** Cinnamaldehyde, Bladder Cancer, Cancer Cell Metabolism, LDHA, HSF1, ErbB2

O23

**Effect of Cassiopea Andromeda Venom on P15ink4b, P21 Waf1/Cip1, P53, DNA Methyltransferase 1, and Bcl-2 Genes Expression, Apoptosis Induction, and Cell Growth Inhibition in Acute Promyelocytic Leukemia Nb4 Cell Line**

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**Background:** One of the acute hematologic malignancies is acute promyelocytic leukemia (APL) which resulted in translocation of chromosomes 15 and 17, t (15; 17), and cessation in the maturation of myeloid cell line, and ultimate aggregation of neoplastic promyelocytes. Regarding the appetite of using herbal and marine medicine studies are increasing, and on the other hand, the features of Cassiopea andromeda Venom remained unclear; this study was conducted to determine its effects on NB4 cells as a model for APL. **Methods:** In this experimental study, the cells were treated with C. andromeda Venom concentrations at different periods and times. Growth inhibition and toxic effects of C. andromeda Venom were evaluated through methyl thiazole tetrazolium salt reduction (MTT test). The flow cytometry analysis was carried out using 7AAD and Annexin V stains for evaluating this venom's effect on apoptotic pathways. Besides, a Real-Time polymerase chain reaction was performed to evaluate the relative gene expression. **Results:** C. andromedaVenom inhibited the growth of NB4 cells as correlated with concentration and time. Cell growth was inhibited by 49.1%, after 24 hours of treating NB4 cells with 1000µg/mL C. andromeda Venom. This venom increased the apoptotic process, which was then verified by 7AAD/AnnexinV staining. The fold change of p15INK4b, p21 WAF1/CIP1, P53, DNMT1, and Bcl-2 genes in the NB4 cell line were 144, 2.78, 1.75, 15.24, and 0.33, respectively, which meant that the expression level of p15INK4b, p21 WAF1/CIP1, P53, and DNMT1 were increased by 14400%, 278%, 175%, and 1524%, respectively, and the expression of Bcl-2 was decreased by 67%. **Conclusion:** Considering the inhibitory property of C. andromeda Venom, the authors recommended it as a part of combinational medication for treating APL in animal trials and for other leukemias' in vitro studies.

**Keywords:** Acute Leukemia, Apoptosis, Cassiopea Venom, Epigenesis, Venom



O24

### Genetic Diagnosis in Hereditary Breast and Ovarian Cancer

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It is estimated that 5-10% of cancers have germline mutation in one of cancer susceptibility genes which are known as hereditary cancer syndromes. Usually such syndromes are inherited in autosomal dominant pattern. There are different criteria to select hereditary cases from sporadic through genetic counseling. The diagnosis could be confirmed by using genetic testing for genes which are related to considered syndrome. Based on different syndromes, various groups of genes will be selected for sequencing and analysis. Various genes have different risks and need different approaches for patients and their family members. Risk assessment, family screening and prophylactic procedures are the final steps after genetic diagnosis. Risk Reducing Mastectomy (RRM) and Risk Reducing Salpingo-oophorectomy could be beneficial for patients with mutation in genes with higher risk of cancersusceptibility.



O25

### Updates on Tumor Mutational Burden and the Immunotherapy Biomarker Landscape

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The transmembrane receptor tyrosine kinase HER2 is overexpressed in approximately 15% of breast tumors and correlates with poor clinical prognosis. Several treatments that target HER2 are approved for treatment of HER2-positive metastatic breast cancer. The serum biomarkers most widely used to monitor anti-HER2 therapies in patients with HER2-positive metastatic breast cancer currently are CA15.3 and CEA. Nevertheless, their clinical utility in patients with breast cancer remains a subject of discussion and controversy; thus, additional markers may prove useful in monitoring the therapeutic responses of these patients. The extracellular domain of HER2 can be shed by proteolytic cleavage into the circulation and this shed form, sHER2, is reported to be augmented during metastasis of HER2-positive breast tumors. Here, we studied the clinical usefulness of sHER2, CA15.3, and CEA for monitoring treatment for breast cancer.

O26

## Molecular Profiling for Precision Cancer Therapies

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**Background:** Use of next-generation sequencing (NGS) to identify clinically actionable genomic targets has been incorporated into routine clinical practice in the management of advanced solid tumors. Molecular profiles obtained on tumor DNA and RNA can guide the clinical management of cancer patients. **Method:** For tumor profiling the scientists evaluate the clinical application of capture-based NGS techniques. This test identifies alterations in 300-500 genes, tumour mutational burden and genomic signatures as microsatellite instability. The decision to obtain the NGS assay for a particular patient was done according to investigator's choice **Result:** The actual number of patients starting a targeted agent based on NGS results is low but it provides substantial information in terms of providing additional treatment options, identifying resistance conferring mutations and facilitating clinical trial enrollment. Optimal time of testing, early or late in disease course, financial implications of testing and using targeted therapy and survival benefit of targeted therapy need further studies. **Conclusion:** Molecular tumor profiling by NGS can provide diagnostic or prognostic information, identify a potential treatment regimen or targeted therapy, and determine eligibility for the following: i) a Food and Drug Administration (FDA) approved medication for that tumor type, ii) a medication available as off-label treatment for the specific molecular alteration in a nonapproved tumor type, or iii) a targeted therapy available in clinical trials with investigational agents based on an identified molecular alteration. Considering the lack of large prospective clinical trials that certified its clinical utility, the risks of overdiagnosis and increase costs without survival benefits are real.

**Keywords:** Precision Medicine, Tumor Profiling, Solid Tumors

# COVID-19 Vaccination Updates with Emphasis on Medical Laboratory Aspects

## O27

**Introduction:** Adjuvanted subunit protein vaccines have a reputation for strong safety, immunogenicity and efficacy. SpikoGen® vaccine is a subunit COVID-19 vaccine composed of a recombinant prefusion-stabilized SARS-CoV-2 spike protein extracellular domain antigen combined with Advax-CpG55.2™ adjuvant. The vaccine was safe and well tolerated in Phase 1 trial and protected against COVID-19 infection in relevant animal models.

**Methods:** This randomized, placebo-controlled, double-blind Phase 2 trial was conducted on 400 adult participants randomized 3:1 to receive two doses of 25 µg SpikoGen® vaccine 3 weeks apart or the placebo. The primary safety outcome was incidence of solicited adverse events up to seven days after each dose and unsolicited adverse events up to 28 days after the second vaccination. The primary immunogenicity outcome was seroconversion against S<sub>1</sub> protein and the geometric mean concentration (GMC) of S<sub>1</sub> antibodies.

**Results:** No serious adverse events were recorded with the most common solicited adverse events being injection site pain and fatigue. SpikoGen® vaccine induced robust humoral and T cell responses in the majority of subjects. By day 35 (2 weeks post second dose), 87% of immunized subject sera neutralized SARS-CoV-2 virus at titers > 16 (95% CI: 82.37 – 90.43). Sera from vaccinated individuals were able to cross-neutralize viruses expressing alpha, beta, gamma, delta and lambda spike protein variants.

**Conclusion:** SpikoGen® vaccine had an acceptable safety profile and induced promising humoral and cellular immune responses against SARS-CoV-2 including variants. Based on these results, SpikoGen® was advanced into Phase 3 trials.

**O27****Variants of SARS-CoV-2 and Vaccines Efficacy****Amirmasoud Rayati Damavandi 1, Razieh Dowran 2, Sarah Al Sharif 3, Fatah Kashanchi 3, Reza Jafari 4 \***

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An ongoing pandemic of newly emerged SARS-CoV-2 has puzzled many scientists and health care policymakers around the globe. The appearance of the virus from Wuhan city, China was accompanied by several distinct antigenic changes specifically spike protein which is a key element for host cell entry of virus and major target of currently developing vaccines. Some of these mutations enable the virus to attach to receptors more firmly and easily. Moreover, a growing number of trials are demonstrating higher transmissibility and in some of them, potentially more serious forms of illness related to novel variants. Some of these lineages, especially the Beta variant of concern, were reported to diminish the neutralizing activity of monoclonal and polyclonal antibodies present in both convalescent and vaccine sera. This could imply that these independently emerged variants could make anti-viral strategies prone to serious threats. The rapid changes in the mutational profile of new clades, especially escape mutations, are suggestive of the convergent evolution of the virus due to immune pressure. Nevertheless, great international efforts have been dedicated to producing efficacious vaccines with cutting-edge technologies. Despite the partial decrease in vaccines efficacy against worrisome clades, the majority of current vaccines are still effective at preventing mild to severe forms of disease and also hospital admission or death due to coronavirus disease 2019 (COVID19). Here we summarize existing evidence that is available about newly emerged variants of SARS-CoV-2 and notably, how well vaccines work against targeting new variants in addition to modifications of highly flexible mRNA vaccines which might be required in the future.

**Keywords:** Escape Mutations, Neutralizing Activity, SARS-CoV-2, nCOVID19, Spike Protein, Vaccines Efficacy, Variants of Concern

# Diagnosis of Systemic Mycoses and Quality Control in Mycology Lab

## O28 – O33

Different high-risk individuals and also immunocompromised population groups who are at higher risk for acquiring systemic fungal infections has increased during the past decades. In another side, the variety of opportunistic fungal diseases caused by different emerging and newly recognized fungal species is being to increased.

Early diagnosis and appropriate therapy is very important for patient care in these conditions. In recent years different kinds of laboratory tests were designed and used for correct diagnosis of systemic fungal infections including microscopic morphology-based tests (histopathology, direct microscopy), culture based methods, Immunological and serological and finally molecular biology methods. Although, histopathology is gold standard and has many advantages, but because of some limitations in taking of suitable and enough biopsy materials in complicated conditions i.e thrombocytopenia / neutropenia in severe diseases, we need to perform indirect tests such as biochemical/immunological or molecular tests and each of them has advantages and disadvantages for accurate and precise diagnosis.

In this panel we'll talk about these limitations and how we could performed better management to control the sources of errors in these expensive laboratory tests.

**O28****Laboratory Diagnosis of Invasive Aspergillosis: Quality Control & Sources of Error****Mohammad Taghi Hedayati 1 \***

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Among different fungal genera, *Aspergillus* is the predominant organism causing invasive mold infections. Invasive Aspergillosis (IA) is the most important complication due to *Aspergillus* in aspect of morbidity and mortality. The incidence of IA is still increasing, mainly because of the increasing number of patients at risk for the disease including patients who are receiving prolonged corticosteroid therapy or chemotherapy and due to the increasing number of transplant recipients and more recently patients with COVID 19 and influenza. Early and reliable diagnosis is a significant parameter to start the rapid initiation of appropriate antifungal therapy. One of the main challenges in IA patients is how to get an early diagnosis. Conventional diagnosis approaches including histopathology and culture on the specimens obtained from the normally sterile areas are enough for the definitive diagnosis of IA, in many cases they are not feasible, because of the patients are too ill to undergo invasive procedures to collect of samples. So, the reliance on traditional methods hampers early diagnosis of infection. In addition, culture is slow and often only becoming positive during the later stages of the disease. Non-culture-based method including sero-diagnosis and molecular approach seems to be promising. There are many sources for error which affect on the sensitivity, specificity and quality of non-culture-based methods. In this presentation we will discuss the advantages and drawbacks of conventional and newer diagnostic approaches in IA as well as the recommendations for health care and laboratory specialist to receive an appropriate diagnosis to improve the chance of patients' survival.

**Keywords:** Diagnosis, Non-Culture-Based Methods, Culture-Based Methods, Invasive Aspergillosis, Sources of Error

O29

### **Mucormycosis and the challenges of laboratory diagnosis**

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Mucormycosis is a super-acute and challenging fungal infection. Prompt diagnosis and early treatment before tissue involvement not only saves the patient's life but also eliminates the need for debridement. Mucorals are the causative agents of this disease and the genus *Rhizopus* is the most common cause of the disease in all areas, including hospitals and other environments. Therefore, the cause of the disease is not the entry of open fungi through the body's ducts, but they make the predisposing factors for the disease. It seems that due to the prevalence of underlying factors and diseases, the incidence of this disease is increasing. Blood malignancies are the most common underlying disease in developed countries and uncontrolled diabetes in developing countries. Despite efforts to diagnose and treat invasive mucormycosis early, the mortality rate remains high. Clinical diagnosis is not sensitive enough. Although images of multiple nodules and pleural effusions on radiographs are associated with pulmonary mucormycosis, or a reverse halo sign on CT scan may indicate mucormycosis, none of these diagnostic methods are definitive. Direct experimental methods and culture, as applicable methods with the least facilities, have certain characteristics, while some believe that histopathological tests show definite diagnosis. Molecular assays can also be used to accurately detect or identify mucorals. They can be recommended as valuable additional tools that complement conventional diagnostic methods. In this article, different methods of diagnosing mucormycosis will be explored and the challenges and benefits of each will be discussed based on the conditions and facilities available in health centers

O30

## Rapid, Point-of-Care Antigen and Molecular-Based Tests for Diagnosis of Cryptococcal Meningitis: An Update on Its Limitations and Errors

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**Background:** Cryptococcal meningitis (CM) as an AIDS defining disease remained high in less developed countries. A recent global estimate found about 223,100 cases of cryptococcal meningitis complicating HIV/AIDS. The Indian ink staining can visualize the encapsulated yeast, but its sensitivity is <86%, mainly for low fungal burdens (beginning illness or treated with ART). Culture is the gold standard for diagnosing CM but has several drawbacks, including laboratory facility requirements and false-negative results due to low fungal burden. Latex agglutination test to detect cryptococcal antigens (CrAg) in serum and spinal fluid has demonstrated some false positives. The indirect fluorescent antibody is high sensitive than LA. Enzyme immunoassay (EIA) has been considered the most sensitive and specific for detecting cryptococcal antigen but needs the equipment and trained personnel. CrAg Lateral Flow Assay (LFA) is an immunochromatographic dipstick assay for the qualitative and semi-quantitative detection of CrAg in CSF, serum, or plasma that rapidly detects using monoclonal anti-cryptococcal conjugated antibodies against *C. neoformans* and *C. gatti* complexes. CrAg LFA has revealed equal or higher sensitivity than the enzyme immunoassay (EIA) and latex agglutination (LA) test. **Conclusion** CrAg LFA assay helps screen serum in asymptomatic CM one or two months before progression to the central nervous system. CrAg LFA is the most promising point of care for screening the serum of HIV patients with CD4 < 100 with a sensitivity of 100 % and a specificity of 99.8 %. CrAg LFA is thermostable, affordable, and requires very little training for optimal use, particularly in restricted budget situations.

**Keywords:** Diagnosis, Cryptococcal Meningitis, Antigen, Screening, HIV, Lateral Flow Assay

O31

### Why There is Necessary to Identify Clinically Isolated Yeasts at the Species Level?

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Identification and specification of yeast species is not performed by Clinical laboratories routinely, because there is an idea that many of yeasts are considered as normal inhabitants of body surfaces including skin, mouth and gastrointestinal tracts, and genital mucosa. Because of increasing numbers of non- albicans candida species isolated in clinical specimens and emerging of drug -resistance yeasts, an important challenge for clinicians and microbiologists is appeared, appropriate instruments and materials must be provided, and educated or well-trained personnel should be ready for accurate and precise identification and specification of clinically isolated yeasts at the species level. It is necessary to specification of yeasts in following specimens and conditions: Yeasts isolated from biopsied materials, blood samples, CSF and other body fluids which are sterile in normal situations. Yeasts isolated from clinical materials with large numbers. Yeasts isolated from immunocompromised hosts or drug resistant yeasts isolated from complicated infections. In general, we'll talk about fungal diagnostics, what is currently available. I will only focus here on what's available from the mycology or the microbiology laboratory in this country. So, first of all, we have a number of direct tests or conventional tests. And these are histopathology, direct microscopy, and culture. In this part, I will talk only about microscopic morphology, and finally few words on culture.

**O32**

## **Quality Controls and the Sources of Errors in Diagnosis of Invasive Mycoses**

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The basis of most molecular tests for clinical diagnosis in microbiology are PCR-amplification of DNA or RNA fragments. Given a correct and standard design of the PCR tests used in the clinical laboratory, there is still a possibility of false positives and false negatives results, which not only invalidates the diagnosis but also causes significant financial and human losses for the patient and the laboratory. The most important source of false-positive results is the contamination of the samples with unwanted microorganism cells, and/or their already-amplified molecules. The goals of molecular medical mycology are either to diagnose the disease, or to identify the isolated causative fungus, and rarely other diagnostic applications. As there are significant amounts of cells and then DNA in isolated colonies, and with sufficient experience and knowledge on contamination-prevention protocols, molecular identification of fungal colonies is straightforward without false results. Conversely, in the molecular detection of diseases from clinical samples, there is a high probability of false-positive and negative. In this regard, the most important sources of false-positive are the presence of fungi in the environment (including air, tubes, and laboratory equipment), the presence of contaminating commensal fungi in the clinical samples, and the transfer of pre-existing molecules (eg. during DNA extraction, or electrophoresis) to the patient's test tubes. The sources of false-negative reactions in clinical samples are the low concentration of fungal cells or free DNA, the inadequacy of existing methods of DNA extraction and purification, and the presence of inhibitors in the clinical sample. To investigate the false-positive and negative results, the following points are recommended: 1) Inclusion of at least two negative control tests in the DNA extraction step, 2) Inclusion of at least two negative control tests in the PCR tests, 3) Inclusion of at least one positive control test in the extraction step and one positive control test in the PCR step, 4) Inclusion of the internal control in all clinical specimens, 5) Repeating the test or adding a tube to detect a secondary target, to confirm the results of the tests of those specimens suspected of having systemic infections; and finally, 6) Absolute adherence to standard instructions before, during and after PCR, 7) Regularly and periodically internal and external evaluations of the personnel and the tests to ensure that the quality control criteria are met, especially for quantitative reactions such as real-time PCR.

O33

**A neglected predictive score:**

**Candida colonization index evaluation in pediatric intensive care and bone marrow transplantation units in Children Medical Center, Tehran, Iran**

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**Background:** Understanding a link between Candidemia and Candida colonization is an important issue for clinicians for decades; so, this link has proposed the colonization index as the “missing link”. The aim of this study was to evaluate the Candida colonization index in pediatric patients staying at intensive care units (ICUs) as well as bone-marrow transplantation (BMT) unit and to determine the species distribution and susceptibility pattern of the isolates against several antifungal drugs. **Material and methods:** This study was conducted in the Children’s Medical Center in Tehran-Iran during 6 month. Totally, six hundred and sixty-one samples from 83 patients including oral cavity, skin surrounded catheters; ear, throat, nasal, and urine cultures were collected. Candida colonization index (CI) was calculated according to the previous studies description. The isolates were identified using CHROMagar Candida medium and PCR-based methods to the species-specific complex level. Antifungal Susceptibility test was performed according to the CLSI M27-A2 and M60 documents. **Results:** Of 661 samples, 177 samples from 50 individuals (27%) were considered as positive cultures. Colonization index higher than 0.5 was confirmed in 29 cases (58% of positive samples) in which two child (6.8%) with CI>0.5 developed candidemia. No predisposing factor was significantly affect the CI>0.5 except for acute lymphoblastic leukemia. Candida albicans (n= 53, 49.5%) followed by C. glabrata (n=20, 18.7%), Candida krusei (n=15, 14%), C. parapsilosis (n=12, 11.2%) were the most frequent Candida species in patients with CI>0.5. Among FLZ-resistant strains, 12 isolates (7.01%) were multi-azole resistant which showed high MICs against both ISZ and RVZ



and seven strains (4.09%) were resistant to all echinocandins. Isavuconazole, RVZ and AFG were reported as the most effective antifungals, regarding the values of GM. Conclusion: Patients are at risk of fungal infection in pediatric intensive care units especially candidemia. Here, more than a half of children with positive yeast cultures had  $CI > 0.5$  amongst 6.8% developed candidemia. To the best of our knowledge, there is no study about the evaluation of CI in pediatrics in Iran. More detailed and larger studies should be made to assess the reliability of the score in forecasting invasive candidiasis in children.

**Keywords:** Pediatric, Candida Colonization Index, Candidiasis, Antifungals



# Ensuring Biosafety in The Activities of Clinical Diagnostic Laboratories

**O34 – O40**

O34

### Summermatter Kathrin 1 \*

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Laboratories working with biological agents (e.g. diagnostics, research, etc.) are typically classified in biosafety or containment levels (biosafety level 1, 2, 3 and 4). This classification is not always carried out on the basis of a risk assessment, considering also methods, equipment and staff competency. In addition, despite this common denomination, individual laboratories differ significantly in different countries or even within the same country. The new WHO manual no longer uses biosafety levels for the classification of laboratories. Depending on the risk, appropriate measures can be selected from a set of measures. The risk assessment is considered a process and includes the following steps (risk assessment cycle): gathering of information, evaluation of the risk, development of a risk control strategy, selection and implementation of risk control measures and review of the risk and risk control measures. Have these biosafety levels now become obsolete or will we continue to use them? Will we still be safe in our laboratories or do they now pose a risk for the release of dangerous organisms? These and other issues will be addressed. In presentation, the biosafety levels will be placed in the context of this new approach. The new WHO Laboratory Biosafety Manual distinguishes between core, heightened and maximum requirements.

**Keywords:** Biosafety Level, Risk Assessment Cycle

O35

### Core Requirements

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The new WHO Laboratory Biosafety Manual no longer uses biosafety levels but distinguishes between core, heightened and maximum containment requirements. In the past, for the sake of simplicity, the risk group was equated with a biosafety level, regardless of the activity. The risk-based approach allows to define safety measures tailored to activities. Core requirements is the term used to describe a combination of risk control measures that are both the foundation for, and an integral part of, laboratory biosafety. The core requirements include guidelines for good microbiological practices and procedures, personnel competence and training, facility design, specimen receipt, storage and decontamination and waste management, personal protective equipment, laboratory equipment, emergency response and occupations health. This presentation briefly introduces the different measures with a special emphasis on good microbiological practices and procedures.

**Keywords:** Core Requirements, Risk Control Measures, Good Microbiological Practices and Procedures

**O36****The New WHO Laboratory Biosafety Manual, 4th Edition****Summermatter Kathrin 1 \***

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The 4th edition of the WHO Laboratory Biosafety Manual was published in 2020. The WHO Laboratory Biosafety Manual (LBM) serves as a de facto global standard that presents best practices and sets trends in biosafety. Previous versions of the manual described the classification of biological agents and laboratories in risk groups and biosafety levels. The actual risk of an activity is influenced not only by the agent handled but also by the procedure, the method or the equipment used or the competency of the laboratory staff. Therefore, the recent publication of the fourth edition expands the thorough, evidence-based and transparent assessment of the risks, allowing safety measures to be balanced with the actual risk of working with biological agents on a case-by-case basis. The core document is supplemented by 7 monographs providing more detailed information to help implementing the new approach. The presentation will give an overview about the new laboratory biosafety manual and its associated monographs and how it can be used for our day to day activities.

**Keywords:** WHO Laboratory Biosafety Manual, Risk Based Approach

O37

### Handling of Clinical Specimen in Clinical Microbiology Laboratory

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The Institute for Infectious Diseases (IFIK) in Bern is the only institute in Switzerland comprising all subdivisions of microbiological diagnostics including viral, bacterial, fungal and parasitic infection diagnostics. The coordination of thousands of clinical samples in a large laboratory is a great challenge. Samples have to be quickly aliquoted, efficiently distributed among the subdivisions and correctly stored. In addition, different types of samples require various handling procedures and safety precautions. Risk assessments on these samples depending on clinical information are inevitable. This requires good interaction and communication between clinicians, academic staff and laboratory technicians. In addition, national and international guidelines must always be followed and work must be performed according to good microbiological practice. Each laboratory has different challenges depending on its size, sample types, pathogens to be processed and infrastructure. This presentation will show situations from daily laboratory routine and present our solutions to these challenges.

**Keywords:** Clinical Specimen Handling, Risk Assessment, Routine

**O38****Biosafety Requirements for Laboratories Handling Materials with SARS-CoV-2****Koller Roger 1 \***

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New and emerging pathogens present a special challenge to diagnostic laboratories. Beside the development and establishment of diagnostic tools, safe work in the laboratories must be guaranteed without the risk of transmission or release into the environment. The corona pandemic has kept us busy for almost two years now. Millions of samples are being processed around the world every day, pushing laboratories to their limits with an unprecedented flood of tests. Many laboratories are working 24/7 to identify newly infected people and break chains of infection. Despite the large workload, safe working practices must be ensured in these high-throughput diagnostics settings. This requires coordinated and defined work processes protecting staff from laboratory-transmitted infections. SARS-CoV-2 will be used as an example to show which hurdles had to be overcome to enable safe work with a pathogen that was unknown at the time and how these work processes have changed in the meantime.

**Keywords:** SARS-CoV-2 Handling, Safe Work Processes, High-Throughput Diagnostics

O39

### Transfer and Transportation of Clinical Specimens

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It is often necessary to transport specimens, biological materials or waste that are known or expected to contain biological agents between rooms, laboratories or facilities. In some cases, the material may need to be transported to laboratories in other cities, regions or even countries for further testing, treatment or storage. Transportation of infectious substances may be subject to various national and / or international regulations, depending on the origin, destination and/or the mode of transport being used. Irrespective of the regulations that apply, the aim is always to reduce the likelihood of an exposure to and/or a release of the infectious substance in order to protect personnel, the community and/or the surrounding environment. Transferring or transporting infectious substances within or between laboratories should always be undertaken in a way that minimizes the potential for drop, spillage, collision or similar events. The issues raised in this discussion are outlined below :

- Transfer within the laboratory, within a building, between buildings on the same site
- Regulation of the transport of infectious substances
- Classification of infectious substances
- Triple packaging of infectious substances
- Packaging of biological substances suspicious to dangerous infectious material

**Keywords:** Transport, Category A and B Infectious Substances, Triple Package, Regulation, Exempt Human Specimens

**O40****Risk Assessment in Medical Laboratories****Katayoon Khodaverdian 1 \***

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Risk assessment is a systematic process of gathering information (hazard identification) Evaluating the likelihood and consequence of exposure to staff or the environment using a risk assessment matrix, Determining the appropriate risk control measures to reduce the risk to an acceptable level. It is recommended that each laboratory should conduct a local (that is, institutional) risk assessment to ensure it is competent to safely perform the intended testing with appropriate risk control measures. It is important to note that risk can never be completely eliminated unless the work is not performed at all. Therefore, determining if the initial and/or residual risks are acceptable, controllable or unacceptable is a vital part of the risk evaluation process. Once an acceptable risk has been established, a risk control strategy must be developed to reduce any initial risks to an acceptable risk, then risk control measures must be selected and implemented to allow the work to proceed safely. The issues raised in this discussion are as follows: -Basic concepts in risk assessment -Review five steps in risk assessment process -Risk assessment matrix, how to complete it -Develop risk control strategy -Select and implement risk control measures -Review risks and risk control measures

**Keywords:** Risk, Hazard, Likelihood, Consequence



# Geriatric Medical Laboratory

## O41 – O43

Time modifies many biologic processes. Evaluation of laboratory data in older adults is complicated by a number of factors. However, an analysis in a laboratory is not a descriptive statement based on the subjective experience of the operator, but rather is an objective measurement based on a postulated standard. The reference interval is determined from the range of values in which 95% of the population falls, so 5% of all results from “healthy” people are considered abnormal. A fundamental discussion of the consequences of the current demographic trend in the population has not been conducted in laboratory medicine until now.

**O41****Testing in the Elderly****Reyhaneh Aminalroaya 1 \***

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Laboratory data can play a significant role in the care of older adults and even more so in patients with dementia or altered mental status, where the ability to obtain a history may be limited. Evaluation of laboratory data in older adults is complicated by a number of factors. Starting from the 1960s to 1970s, laboratories shifted away from reporting “normal values” in favor of reporting “reference intervals”. Reference intervals for lab tests are determined by assembling a reference population that typically consist of “healthy” subjects with no chronic diseases and no medications. These “healthy” subjects in geriatric populations are rare, so in practice, most adult reference populations consist of subjects less than 60 years old. The reference interval is determined from the range of values in which 95% of the population falls, so 5% of all results from “healthy” people are considered abnormal. Values falling near the upper or lower limit may be hard to interpret as they may truly be normal results. For parameters which change with age, there is a risk for false negatives and false positive results. An alternative approach is to test patients with and without disease to determine a threshold level for a test beyond which the probability of a specific disease is high enough to prompt further clinical actions. These threshold levels have been called “clinical decision limits.” We discuss about Laboratory tests which change with age, and their clinical implications.

**Keywords:** Older Adults, Laboratory Test, Changes, Reference Intervals

O42

### Pathologic changes in geriatrics

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Time modifies many biologic processes. Aging, characterized by progressive changes associated with increased susceptibility to many diseases, is influenced by multiple factors. We will discuss changes in aging organs. For example functional bone marrow reserves are reduced, reflux esophagitis is common with age.

**Keywords:** Aging, Changes, Pathology

**O43****Laboratories and Geriatrics: More than Reference Intervals****Fatemeh Sadat Mirzadeh 1 \***

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Medications and lifestyle changes have led to an increase in life expectancy. Especially in industrialized countries, the proportion of senior citizens has been steadily increasing, resulting in an inverse demographic pyramid. A fundamental discussion of the consequences of the current demographic trend in the population has not been conducted in laboratory medicine until now, except for an occasional call to adjust reference intervals for specific tests. The physical and/or chemical properties of a biological sample collected from the patient are analyzed, and the results are interpreted in terms of clinical information that indicates the patient's health status. It is possible to express the results quantitatively and describe them as exact numeric values, rather than in words as it is for pathology or radiology, which are other diagnostic disciplines. Consequently, an analysis in a laboratory is not a descriptive statement based on the subjective experience of the operator, but rather is an objective measurement based on a postulated standard. In geriatric laboratory medicine, there is yet another aspect to consider. It is the progressive loss of physiologic functions in an aging human body.

**Keywords:** Aging, Laboratory Test, Changes, Reference Intervals

## Hematologic Neoplasm (ALL, AML)

### O44 – O47

Acute lymphoblastic and Myeloblastic leukemia (ALL and AML) are uncontrolled clonal proliferation of abnormal myeloid and lymphoid progenitor cells in the bone marrow. Morphology, flow cytometry cytogenetics and molecular analysis of fusion genes and WHO 2008/2016 classification are the fundamental aspects in acute leukemia diagnosis. As we know recurrent cytogenetic abnormalities are the driver and initiating events in the leukomogenesis process and have a great impact in pathogenesis and prognosis and treatment of acute leukemia by targeting the essential molecules participating in hematopoietic stem cell differentiation, renewal and tumor suppression. But despite development of risk stratified protocols patients are still suffering from drug toxicities and drug resistance and some of them end up in relapse or even are convicted to death. It seems that the cooperating submicroscopic and genomic aberrations are important players in the prognosis and outcome of disease and can make a specific genomic signature for each patient exclusively and could be used as targets in Pharmacogenomics. For example, target therapy with ABL-TKI in a patient with BCR-ABL like ALL, whom are Philadelphia chromosome negative but have the same genomics signature as Ph.- positive ALL has a great impact in prognosis of this previously poor outcome ALL subgroup.

Advanced technologies are a gateway to identifying the genomic aberrancies like aberrancies in DNA sequence, single nucleotide variants (SNV), small gains and losses (indels), and copy number variation (CNVs), gene expression anomalies, cryptic fusion genes, and epigenetic gene modifiers. These genomic aberrations play an important role in leukomogenesis and drug resistance and blooming of a new leukemic sub clones due to chemotherapy stress and also a great role in prediction of patients outcome and their overall survival.

In this Panel we discuss the different aspect of AML and ALL, diagnosis, follow up, prognosis and risk stratification approaches and the necessity of incorporating new technologies in this field to come up with a better outcome and overall survival in high risk groups and minimize the risk of drug toxicity in low risk group.

O44

## Genomic Risk Stratification in Acute Lymphoblastic and Myeloblastic Leukemia

Soudabeh Hosseini 1 \*

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Acute lymphoblastic and Myeloblastic leukemia (ALL and AML) are uncontrolled clonal proliferation of abnormal myeloid and lymphoid progenitor cells in the bone marrow. According to WHO 2008/2016 classification the recurrent cytogenetic abnormalities are the driver and initiating events in the leukomogenesis process and have a great impact in pathogenesis and prognosis and treatment of acute leukemia by targeting the essential molecules participating in hematopoietic stem cell differentiation, renewal and tumor suppression (Robert et.al 2012 ) But despite development of risk stratified protocols patients are still suffering from drug toxicities and drug resistance and some of them end up in relapse or even are convicted to death. it seems That the cooperating submicroscopic and genomic aberrations are important players in the prognosis and outcome of disease and can make a specific genomic signature for each patient exclusively and could be used as targets in Pharmacogenomics . For example, target therapy with ABL-TKI in a patients with BCR-ABL like ALL (Roberts et al 2014). Schurtz etal, 2014) whom are Philadelphia chromosome negative but have the same genomics signature as Ph- positive ALL has a great impact in prognosis of this previously poor outcome ALL subgroup. Advanced technologies are a gateway to identifying the genomic aberrancies like aberrancies in DNA sequence ,single nucleotide variants(SNV),small gains and losses(indels),and copy number variation(CNVs), gene expression anomalies, cryptic fusion genes ,and epigenetic gene modifiers these genomic aberrations play an important role in leukomogenesis and drug resistance and blooming of a new leukemic sub clones due to chemotherapy stress and also a great role in prediction of patients outcome and their overall survival Identification of Genomic aberrancies and integrated cytogenetic and genomics aberrancies (Anthony v. moorman 2016) and identification of co- mutations and in another words gene –gene interactions (Elli Papaemmanuil et.al 2016) and incorporation of these data in risk stratification protocols will bring about the opportunity of patients target therapy and more precise personalized medicine and less drug toxicity and also less relapse rate.

**Keywords:** Risk Stratification, All, AML, Integrated Cytogenetics and Genomics

O45

### **Multiparameter Flow Cytometry in the Diagnosis and Monitoring of Acute Leukemia in Children**

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Acute leukemias are a heterogeneous group of malignancies with varying clinical, morphologic, immunologic and molecular characteristics. Acute leukemia is the most common group of neoplasms diagnosed in the pediatric population. In particular, acute lymphoblastic leukemia (ALL) of B cell lineage (B-ALL) is both the most common subset of acute leukemia in children and notably more frequent than in the adult population. On the other hand, acute myeloid leukemia (AML) is relatively infrequent in the pediatric population and much more frequent in adults. Distinction between lymphoid and myeloid leukemias is crucially important and most often is made by Flow cytometry. Flow cytometry is routinely used in clinical laboratories for the identification and classification of acute leukemias, as well as assessment of residual disease following therapy. In this presentation, the current status of flow cytometric immunophenotyping as applied to the diagnosis and monitoring of pediatric patients with acute leukemias will be described.

**O46**

## **The Role of The Laboratory in The Diagnosis and Prognosis of Children with Acute Lymphoblastic Leukemia**

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The most common cancer in children and adolescents is acute lymphoblastic leukemia. This disease is usually manifested by nonspecific respiratory infections resistant to common treatments, paleness, and clinical evidence of thrombocytopenia. The reticuloendothelial system is a common site of lymphoblastic infiltration manifested by organomegaly and lymphadenopathy. Leukopenia, anemia and thrombocytopenia along with the above clinical findings strongly suggest this diagnosis. However, the findings of cbc even with the observation of lymphoblasts in the peripheral blood smear make a suggestive ALL and bone marrow aspiration are necessary to confirm the diagnosis. Flow cytometry of bone marrow samples is necessary as a diagnostic aid and at the same time very important in determining the type of treatment regimen. In addition, cytogenetic findings are very important in determining the type and severity of treatment regimen and prognosis of the patient. However, the most important findings in determining the prognosis and treatment policy is the molecular response to treatment or minimal residual disease (MRD).

O47

### Acute Myeloid Leukemia in Children: diagnosis and new approaches to therapy

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Pediatric acute myeloid leukemia (AML) is a heterogeneous disease with generally poor outcomes compared to childhood lymphoid leukemia, children with AML have superior outcomes due to fewer adverse genetic mutations and the ability to tolerate the high-intensity chemotherapy currently necessary for cure. While complete remission (CR) rates are high in pediatric AML at approximately 90%, event-free survival (EFS) and overall survival (OS) remain suboptimal at 45% and 65%, respectively, at 3 years, and nearly half of children will relapse. Even in the low-risk genetic groups, relapse remains common at up to 35%. Unfortunately, children at the highest risk of relapse related to poor genetic features have dismal outcomes altogether and continue to require stem cell transplant (SCT) to achieve cure, with only one in three surviving at 3 years. Recent advances in the genetic characterization of AML have enhanced understanding of individualized patient risk, which has also led to the development of new therapeutic strategies. Recent studies have revealed an increasing number of mutations, including WT1, CBFA2T3-GLIS2, and KAT6A fusions, DEK-NUP214 and NUP98 fusions, and specific KMT2A rearrangements, which are associated with poor outcomes. However, outcomes are starting to improve with the addition of therapies such as gemtuzumab ozogamicin and FLT3 inhibitors, initially developed in adult AML.

# Lab Investigation of Prenatal Genetic Disorders

## O48 – O52

Prenatal tests are screening or diagnostic tests that can help to identify health problems that could affect pregnant women or their unborn babies. Some of these conditions can be treated, so it's important to find them as soon as possible. Therefore during this panel it was aimed to bring the latest advances in prenatal diagnosis field. Our invited speakers will give their talks in both clinical and para-clinical directions in order to make them more applicable in daily practice of specialists in labs and clinics. As you know familiarity with the history of prenatal diagnosis and its development through years, and explaining each test limitations and efficacy could help us to choose the best available tests for individuals and think about the ways that can improve the quality of the services delivered in the hospitals or clinics. The first prenatal screening test was introduced was based on a single maternal serum marker of neural tube defects. Later prenatal screening tests for detecting Down syndrome has been developed. But the sensivity and specificity of the methods was very low.

The introduction of new technologies such as chromosomal microarray analysis, whole-exome sequencing, SNP array, has dramatically changed the current practice of prenatal screening and testing for genetic abnormalities in the fetus. Expanded carrier screening panels and non-invasive cell-free fetal DNA-based screening for aneuploidy and single-gene disorders, and more recently for sub-chromosomal abnormalities, have been introduced into prenatal care. We hope during this panel, all these subjects will be discussed and explained clearly.

O48

### Prenatal Diagnosis: Past, Present and Future

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Prenatal diagnosis is the science of identifying structural and functional abnormalities in the developing fetus. In late 70s, the introduction of ultrasound and the possibility to visualize the fetus in utero was the revolution in prenatal screening. Screening for open neural tube defects using MsAFP levels had been increasingly available since 1973. In 1980 amniocentesis and CVS in advanced maternal age introduced to MFM and in 1990 using the triple/quad screening test for evaluating the 21,18,13 trisomies improved the quality of prenatal care. The 1985 Special Issue was using the PGD for diagnosis the mendelian disorders such as CF. Among the more noteworthy contributions have been reports on the development of aneuploidy detection in interphase by FISH in 1994, DNA probes for the diagnosis of microdeletion syndromes, quantitative fluorescence polymerase chain reaction (QF-PCR) for trisomies in 1997 and array comparative genomic hybridization (CGH) for a wider range of duplication/deletion abnormalities and copy number variation in 2007. Noninvasive prenatal testing (NIPT) is the latest revolutionary development in prenatal screening for trisomy 13, 18, and 21. Each of these methods has its own advantages and limitations. Furthermore, attempts are being made to develop noninvasive screening methods for maternal pregnancy-related disorders, such as maternal plasma RNA screening for preeclampsia. Since it is expected that the scope of prenatal screening will broaden in the near future, from not only medical and ethical perspectives but legal perspective as well, it is important to consider “what to offer” and “how to offer.”

**Keywords:** Prenatal Diagnosis, History

O49

### Application of NIPT in Prenatal Diagnosis

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Noninvasive prenatal testing (NIPT), sometimes called noninvasive prenatal screening (NIPS), is a method of determining the risk that the fetus will be born with certain genetic abnormalities. This testing analyzes small fragments of DNA that are circulating in a pregnant woman's blood. Results: years has been moving towards non-invasive methods to determine the fetal risk for genetic disorders. The rapid advancement of modern high-performance molecular technologies along with the discovery of cell-free fetal DNA (cffDNA) in maternal plasma has led to new methods for the determination of fetal chromosomal aneuploidies. This type of testing is referred to as non-invasive prenatal testing (NIPT). The introduction of Next Generation Sequencing (NGS) into clinical practice has rendered NIPT to have high sensitivity in the screening of aneuploidy. It has also allowed detecting and investigating the fetal genome from maternal plasma Conclusion: Findings suggest that NIPT is a test that can be used to identify genetic abnormalities in pregnancies. It's important to know that NIPT is a screening test — not a diagnostic test. This means that it can't diagnose a genetic condition with certainty. It can, however, predict whether the risk of a genetic condition is high or low.

**Keywords:** NIPT, Prenatal Diagnosis

O50

### Application of Chromosomal Microarray in Prenatal Diagnosis

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**Background:** Chromosomal microarray (CMA) is an emerging molecular genetic detection technology in the field of prenatal diagnosis. This approach can accurately detect the number and structural abnormalities of chromosomal imbalances, and also chromosomal alterations such as microdeletions and microduplications. CMA has the advantages of a high resolution, a short detection cycle, and more objective results. CMA has the ability to disclose a wide range of chromosomal abnormalities with length from 50kb to 100kb, which can produce 100 times better resolution than karyotyping. More and more evidence has shown that CMA improves the diagnostic accuracy by approximately 15% to 20% over that of karyotyping. **Results:** Since 2009, the American Obstetrics and Gynecology Association, the Canadian Obstetrics and Gynecology Association, the European Cytogenetics Association, and China have all published guidelines recommending CMA technology as the first-line prenatal diagnostic detection method after detecting prenatal fetal ultrasound structural malformations. **Conclusion:** chromosomal microarray analysis (CMA) is recommended in prenatal diagnosis for cases with one or more structural abnormality detected by ultrasound. For patients with a structurally normal fetus, invasive testing by either microarray or karyotype is recommended

**Keywords:** Chromosomal Microarray, Prenatal Diagnosis

O51

## Application of Exome Sequencing in Prenatal Diagnosis

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**Background:** Genetic diagnosis provides important information for prenatal decision-making and management. Promising results from exome sequencing (ES) for genetic diagnosis in fetuses with structural anomalies are emerging. It is also often the first step in improving the path towards informed diagnosis and treatment, which is especially important in the era of advancing in utero fetal therapy. The objective of this section is to identify what is known about the use of ES for genetic testing in prenatal cases with known or suspected genetic disease. **Methods:** A rapid scoping review was conducted on English-language peer-reviewed studies related to application of ES in prenatal diagnosis between 2014 and 2020. **Results:** Several studies investigated the utility of ES in prenatal diagnosis. Most commonly reported outcomes were diagnostic yields and prenatal phenotype. Few studies reported clinical outcomes related to impact, decision-making, and clinical utility. Diagnostic rates are variable across studies with improved rates when trio (proband, mother and father) whole exome sequencing is performed. Furthermore, there are many ethical considerations including risks of discrimination that must be considered when whole exome sequencing is performed. **Conclusion:** Findings suggest that prenatal ES is beneficial, but more research is needed to better understand the clinical utility, circumstances for ideal use, feasibility, and costs of offering rapid ES as a routine option for prenatal genetic testing.

**Keywords:** Exome Sequencing, Prenatal Diagnosis

O52

### Application of SNP Array in Prenatal Diagnosis

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**Background:** In the past few years, prenatal diagnosis has expanded from karyotyping and fluorescence in situ hybridization (FISH) to chromosomal microarray analysis (CMA). Recently, CMA has been successfully applied to identify copy number variations (CNVs) that are too small to be seen on a standard G-banded chromosome preparation in postnatal and prenatal subjects, and has led to increase the identified contribution of chromosomal abnormalities. SNP array is a type of CMA which can identify triploidy as well as loss of heterozygosity (LOH) by hybridizing the test sample to the array platform and analyzing the signal intensity of SNP probes. It can be performed on prenatal samples to detect numeric chromosome abnormalities, genome-wide copy number changes, and long contiguous stretches of homozygosity (LCSH) which can reflect uniparental disomy and can help recognizing samples with maternal cell contamination (MCC). **Conclusion:** By using genome-wide high-resolution SNP array, we could obtain high diagnostic rate and uncover additional disease association CNVs including mosaic. Despite the challenges and limitations, SNP array has considerable diagnostic and prognostic values during pregnancy and should therefore be the test of choice.

**Keywords:** Chromosomal Microarray, Copy Number Variations, SNP array

# Laboratory Investigation of Endocrine Disorders in Pregnancy

## O53 – O56

The normal function of hormones is necessary for fertility induction and maintenance in humans. Thyroid hormones have an important role in fertility and lactation. So, the identification of thyroid disorders is important for the prevention of complications.

Prolactin is the other hormone that can be increased to 10-20 times in pregnancy. So, diagnosis of prolactin disturbances is challengeable in pregnancy. On the other hand, the management of the patients with a history of prolactin disorders is challenging during pregnancy.

Also, lipid disorders may be associated with a negative effect on the mother and fetus. So, diagnosis and treatment of these disorders are necessary especially in congenital and severe forms.

Finally, one of the most metabolic disorders in pregnancy is gestational diabetes which can be occurred due to hormonal change and consequent insulin resistance. On the other hand, gestational diabetes can be associated with macrosomia and increased pregnancy and labor complications. So, early diagnosis and treatment of diabetes in pregnancy are necessary.

O53

### Diabetes in pregnancy (GDM)

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It is the most common metabolic disease and it can affect up to 25% of women during pregnancy. Internationally, the prevalence of gestational diabetes mellitus (GDM) varies from 1 to 28%. In Iran, the result of studies showed that the prevalence of GDM is 3.4%. The term “gestational diabetes” has been defined as onset or first recognition of abnormal glucose tolerance during pregnancy. Pregnancy is accompanied by insulin resistance, mediated by placental secretion of diabetogenic hormones including progesterone, prolactin, placental lactogen, growth hormone, and corticotropin-releasing hormones. So, in normal pregnancy the level of serum insulin has been increased to confer with these hormonal changes. The insulin resistance has been occurred in second half of pregnancy especially third trimester. The normal mechanism is disrupted in pregnant women with gestational diabetes. Consequences of GDM include increased risk of maternal and fetal adverse outcomes. There is a risk of preeclampsia, fetal macrosomia (weight more than 4000 gram), dystocia, increased risk of cesarian, and birth injuries. Also, it can be associated with hypoglycemia or hyperbilirubinemia in neonates. Maternal insulin-dependent diabetes has long been associated with increased risk of congenital malformations such as central nervous system, cardiovascular, gastrointestinal and urogenital. In this regard, the World Health Organization (WHO) and International Association of Diabetes and Pregnancy Study Groups (IADPSG), recommended the diagnostic testing by performing a 75 gram, two-hour oral glucose tolerance test (GTT) in all women in pregnancy. The American Diabetes Association’s (ADA’s) Standards of Medical Care in the last publication (2022) recommend the diagnostic testing include fasting blood glucose (FBS) or HbA1c before 15 weeks of gestation at first perinatally visit in all pregnant women especially women with risk factors. Fasting glucose of  $\geq 126$  mg/dl in overt diabetes and HbA1c  $\geq 6.5\%$  in pre-gestational diabetes can be detected. FBS of 110-125 mg/dl or HbA1c of 5.9%-6.4% are abnormal glucose test. In these patients, oral glucose tolerance test with 75 g glucose has been recommended in 24-28 weeks of pregnancy. If at least one test is abnormal, the gestational diabetes can be recognised. abnormal ranges of blood glucose at fasting  $\geq 92$  mg/dl, one hour  $\geq 180$  mg/dl, two hour  $\geq 153$  mg/dl. Women with a history of GDM are at high risk of subsequently developing diabetes. These patients should be screened six to 12 weeks postpartum for persistently abnormal glucose metabolism, and should undergo screening for diabetes every three years thereafter. Metformin and weight reduction should be recommended if the overt diabetes is identified. In women with history of GDM, preventive management and life style modification should be advised in prediabetes or normal tests.

**O54****Thyroid in pregnancy****Hossein Samadanifard 1 \*, Haleh Chehrehgosha 2, Amir Hossein Ghanooni 2**

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Humans need to have a healthy anatomical and physiological element in different organs. In this regard, hormonal balance and secretion of endocrine glands are important elements. In endocrine glands, the thyroid gland has a major effect on ability to fertilization, keep the fertility and breast milking after delivery. In recent years, several studies discussed about thyroid disease in pregnancy and intelligence quotient changes in children born to mothers with thyroid disease. Accurate measurement of hormones and correct decision to treat and reasonable correlation between laboratory results and clinical findings can be effective in reduction of probable side effects.

O55

### **Prolactin Levels and Its Diseases in Pregnancy**

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Pregnant women have high levels of prolactin, which will help them succeed in breastfeeding after pregnancy. Prolactin levels increase 10 to 20 times during pregnancy and remain high after breastfeeding. How much prolactin levels are normal in pregnancy is debatable. Whether measuring its level during pregnancy can be helpful in diagnosing is also controversial. In the presence of diseases associated with increased prolactin secretion, such as pituitary adenoma (prolactinoma), there are considerations for the timing and conditions of pregnancy and the continuation or discontinuation of oral medications. Because during pregnancy, prolactin-secreting adenomas become larger and can compress the optic nerve or worsen their symptoms than before, such as increased headache severity or may experience bleeding and pituitary apoplexy during delivery, therefore, decision making to choose the right treatment has always been a challenge for physicians. In short, knowing the conditions before and after pregnancy and mastering the underlying disease can help patients more and manage their disease.

**O56**

### **Lipid Disorders in Pregnancy**

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In normal people, changes in lipids profile during pregnancy and before pregnancy are usually not so great. However, many studies have been studied to examine the level of blood lipids before pregnancy or in early pregnancy and complications during and after pregnancy, which ultimately leads to recommendations for correcting the profile of blood lipids before pregnancy. On the other hand, the presence of inherited disorders during pregnancy can be more dangerous. Hyperlipidemia during pregnancy can have side effects and risks for the mother and offspring. For example, one of the most common inherited disorders is familial hypertriglyceridemia, which in pregnancy can raise triglyceride levels much higher and create dangerous conditions for both mother and fetus. Identifying these people will help them a lot to prevent dangerous outcomes during pregnancy. In general, knowing the methods of prevention and treatment of lipid disorders during pregnancy is very important.

# Lesson Learnd from COVID 19: from Bench to Bed Side

## O57 – O61

The SARS-COV-2, which causes COVID-19, has infected up to now over 500 million people worldwide, and the number of deaths has totaled nearly 6.2 millions. It is to note that COVID-19 test rates can vary per country. Additionally, big differences show up between countries when combining the number of deaths against confirmed COVID-19 cases.

COVID-19 is not only a respiratory disease but also a multisystem disease and involves pulmonary and extra pulmonary organs.

On the other hand, a subset of patients who sustain an acute SARS-CoV-2 infection are developing a wide range of persistent symptoms over the course of many months, called Long COVID or post-acute sequelae of COVID-19 (PASC).

Ten to twenty percent of COVID-19 adult patients develop symptoms lasting longer than 30 days, and 15% still have at least one symptom after 90 days. The prevalence of long COVID in children is unclear and has been assessed in heterogeneous studies between two to twenty percent.

In our panel we attempt to present interdisciplinary some important aspects of COVID-19 from bench to bedside and to throw light on molecular as well as cellular mechanisms in clinical context behind this disease.



O57

### Management of Long COVID Syndrome

**Amir Sharafkhaneh 1 \***

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COVID-19 pandemic affects a large population all over the world. In addition to significant mortality, the morbidity resulting from this infection continues in many individuals. Long or post-COVID syndrome consists of various symptoms originating from different organ systems including CNS (brain fog and sleep disturbances), respiratory (shortness of breath and cough), cardiovascular (palpitation) and musculoskeletal (fatigue and weakness). The list of subacute and chronic conditions associated with COVID-19 infection is growing. Various treatment strategies are under investigation to define management of this affliction. A comprehensive rehabilitation program addressing all the organ systems involved while personalizing the management for each patient may improve quality of life. However, definitive data is lacking currently. Our team has been involved in Telerehabilitation program for last several years. We have applied our expertise and experience in multidisciplinary Telerehabilitation to treat individuals with significant long COVID syndrome. In this presentation, we will discuss our experience with management of long COVID patients.

O58

### Lesson Learned From COVID-19, from Bench to Bedside

Alireza Ranjbar \*

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The SARS-COV-2, which causes COVID-19, has infected up to now over 500 million people worldwide, and the number of deaths has totaled nearly 6.2 millions. It is to note that COVID-19 test rates can vary per country. Additionally, big differences show up between countries when combining the number of deaths against confirmed COVID-19 cases. COVID-19 is not only a respiratory disease but also a multisystem disease and involves pulmonary and extra pulmonary organs. On the other hand, a subset of patients who sustain an acute SARS-CoV-2 infection are developing a wide range of persistent symptoms over the course of many months, called Long COVID or post-acute sequelae of COVID-19 (PASC). Ten to twenty percent of COVID-19 adult patients develop symptoms lasting longer than 30 days, and 15% still have at least one symptom after 90 days. The prevalence of long COVID in children is unclear and has been assessed in heterogeneous studies between two to twenty percent. In our panel we attempt to present interdisciplinary some important aspects of COVID-19 from bench to bedside and to throw light on molecular as well as cellular mechanisms in clinical context behind this disease.

**O59****Post Covid Pulmonary Fibrosis (PCPF)****Arda Kiani 1 \***

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Fibrosis is not common after a viral disease but it has been observed after severe covid-19 infections. Post covid fibrosis is more prevalent in severe covid patient who were on mechanical ventilation. Severity of the disease, high LDH during the disease and smoking are some of the risk factors of PCPF. In brief, Cytokine storm produce pneumonitis, and aberrant repair leads to PCPF. We try to answer the questions: which, when and how we should follow up the covid patient and is whether there is an effective drug to heal this aberrant lesion.

O60

## Antivirals for COVID-19; an Update

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COVID-19 is caused by the severe acute respiratory syndrome coronavirus 2 (SARSCoV-2). Based on the genome organization of SARS-CoV-2, four enzymes are recognized as attractive drug targets: 3CLpro, PLpro, RNA helicase, RNA-dependent RNA polymerase. Nucleoside analogs represent a well-established class of antiviral agents that inhibit viral replication and Remdesivir was the first approved antiviral drug from this class for COVID-19. The U.S. Food and Drug Administration took two actions to expand the use of remdesivir to certain non-hospitalized adults and pediatric patients for the treatment of mild-to-moderate COVID-19 disease. Molnupiravir is another approved nucleoside analog for COVID-19 which is an oral prodrug of beta-D-N4-hydroxycytidine (NHC). However, as a mutagenic ribonucleoside antiviral agent, there is a theoretical risk that molnupiravir will be metabolized by the human host cell and incorporated into the host DNA, leading to mutations. Ritonavir-Boosted Nirmatrelvir (Paxlovid) is an orally bioavailable protease inhibitor that is active against MPRO, a viral protease that plays an essential role in viral replication by cleaving the 2 viral polyproteins which was approved as another antiviral for COVID-19. There are other approved medications for COVID-19 such as baricitinib which is strongly recommended for patients with severe or critical COVID-19. It is an immunomodulator that could suppress the overstimulation of the immune system. Sotrovimab, as a specific monoclonal antibody for treating mild or moderate COVID-19 in patients who are at high risk of hospitalization is also one of the approved medications for COVID-19.



O61

## The Covid Heart

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Learning objectives: Understanding the involvement of cardiovascular system in Covid -Pathogenesis  
-Clinical manifestations-Cardiovascular side effects of Covid Vaccines -Long lasting consequences

# Microbiological Diagnostic of Tuberculosis and Mycobacteriosis

## O62 – O63

Tuberculosis, despite decades of identification of pathogens and effective drugs, is still the leading cause of death among infectious diseases, so that it is currently the second deadliest infectious disease after Covid-19. In the last two decades, there have been significant advances in the control of tuberculosis in the country, so that the incidence of tuberculosis has decreased from about 25 cases per 100 thousand people in 2000 to 13 cases per 100 thousand people in 2019, which The issue has been provided by active diagnostics and Directly Observed Treatment, Short-course (DOTS) and especially the establishment of a network of capable TB laboratory diagnostics in the country and the use of advanced and new techniques in laboratory diagnosis

In this panel, while reviewing the achievements of the last two decades in the control of tuberculosis in the country, the impact of the Covid-19 pandemic on national programs to combat tuberculosis in the country and the world will be discussed. Also, the decisive role of using new tools and techniques in early detection of tuberculosis, such as genomic sequencing using NGS and LiPA, especially in resistant patients (MDR, XDR) and also the importance of phenotypic identification and drug susceptibility in Mycobacterium. achievements and Innovations.

In this panel, while reviewing the achievements of the last two decades in the control of tuberculosis in the country, the impact of the Covid-19 pandemic on national programs to combat tuberculosis in the country and the world will be discussed. Also, the decisive role of using new tools and techniques in early detection of tuberculosis, such as genomic sequencing using NGS and LiPA, especially in resistant patients (MDR, XDR) and also the importance of phenotypic identification and drug susceptibility in Mycobacterium. Achievements and typologies obtained in this field, such as launching line sensitivity tests for line 1, line 2 and line 3 by conventional relative method, direct method for determining antibiotic resistance for Mycobacterium tuberculosis and determination of pyrazinamide resistance and importance We will address it in a timely manner treatment and the challenges of diagnosing and treating patients.

In the other part of this Topic, we will explain the position and importance of implementing quality control programs and TB proficiency testing in quality assurance of laboratory results and a report on how to implement and results obtained from the three courses of EQAP and PT programs implemented by the Iranian Association of Clinical Laboratory Doctors will be presented. obtained in this field, such as launching line sensitivity tests for line 1, line 2 and line 3 by conventional relative method, Direct Drug susceptibility testing of Mycobacterium Tuberculosis Using the Proportional method and determination of pyrazinamide resistance and importance We will address it in timely treatment and the challenges of diagnosing and treating patients.

In the other part of this Topic, we will explain the position and importance of implementing quality control programs (QC) and TB proficiency testing (PT) in quality assurance of laboratory results and a report on how to implement and results obtained from the three courses of EQAP and PT programs implemented by the Iranian Association of Clinical Laboratory Doctors will be presented.

**O62****Lateral Flow Strip for Detection of Mycobacterium tuberculosis PCR Products****Razieh Nazari Vanani 1 \*, Golamhossein Tondro 2, Rezvan Dehdari Vais 2, Masoud Haghkhah 3, Hossein Heli 2, Naghmeh Sattarahmady 4**

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**Background** Tuberculosis (TB) is one of the first bacterial infectious diseases, and it is estimated that one-third of the world's population is infected by TB. The World Health Organization (WHO) reported in 2015 that there were nearly 10.4 million new TB cases. **Methods** Lateral flow test strips (LFTSs) were fabricated using a new specific probe for Mycobacterium tuberculosis (MTB), based on IS6110 sequence gene, and tailed with poly deoxyadenine (dA). To create test and control zones, streptavidin (STP) and a 150-mer dA were dotted on a nitrocellulose membrane. Gold nanoparticles (GNPs) were conjugated with poly deoxythymidine sequence and placed on the conjugate pad. DNA genome of MTB in clinical samples was amplified with PCR, and then detected by the LFTSs. During the assay, samples were firstly hybridized in two steps and then placed on a conjugate pad in a manner that positive and negative samples provided two and one red lines, respectively, on the detection pad. **Results** The results were observed in figure 1 by the naked eye. Positive samples (7 samples) produced two bands corresponding to the test and the control lines, while negative samples (2 samples) produced a single band corresponding to the control line. **Conclusion** The results indicated that LFTSs could successfully recognize the positive and negative samples. The LFTSs can be redesigned for detection of other photogenics.

**Keywords:** Gold Nanoparticles, Lateral Flow, Test Strip, Detection, Tuberculosis

O63

### Challenges of Diagnosis and Treatment of Mycobacteria

Arash Seifi 1 \*

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Mycobacterial infections remain a public health problem worldwide despite the control efforts. With the advances in molecular identification methods, more than 190 species of mycobacteria have been described. Some mycobacteria such as *Mycobacterium tuberculosis* (MTB), *Mycobacterium leprae*, *Mycobacterium avium* complex (MAC) and other nontuberculous mycobacteria (NTM) are pathogens causing a variety of human diseases. These organisms especially MTB can almost involve all parts of the body e.g. lungs, heart, brain, lymph node, gastrointestinal system (GI), bone, kidney, skin, etc. To show the magnitude of the issue, only *M. tuberculosis* infects one-quarter of the world's population, with more than 10 million new cases reported each year. Another major problem is the intrinsic and acquired drug resistance that's restricted the therapeutic options. Timely and accurate detection and effective management of mycobacterial infections can prevent severe disease and reduce mortality, morbidity and transmission. There are major advances in detection (Computed tomography (CT), Nucleic acid-based tests) which seeks to complement or replace the conventional methods (X-ray, sputum microscopy/culture) in a view to reduction in under-diagnosis and improved management. Despite these advances, diagnosis and treatment of mycobacterial infections still faces numerous challenges. As an example, it's estimated only 7% of multi-drug resistant tuberculosis (MDR-TB) are being detected globally, and even less for extensively drug-resistant cases (XDR-TB) as a result of critical gaps in laboratory capacity. Many times, along with the diagnosis of mycobacterial infections, other differential diagnoses are made. So, besides of clinical and radiological findings, laboratories play a critical role in diagnosis. Therefore, in the 13th International Congress on Quality Improvement in Clinical Laboratories, we discuss on "Challenges of Diagnosis and Treatment of Mycobacteria"

# Modern Technologies in Laboratory Hematology

## O64 – O69

The medical diagnostic laboratory plays a pivotal role in health-system as an exciting profession that remains hidden behind the medical treatment or investigation scene. So that, up to seventy five percent of clinical medicine decision making is predicated upon, or confirmed by, or documented by medical laboratory test results. Members of this challenging profession are responsible for providing accurate, reliable and timely laboratory tests for monitoring health, diagnosing and treating disease, assist in rapid identification of disease, assessment of severity of disease, creation of a therapeutic plan and the more efficient the testing protocol. In other hand, personalized medicine as a new approach to understanding, preventing and treating disease relied on our understanding about data and information on each individual's molecular and genetic profile. Hematologic malignancies include various types of heterogeneous diseases in clinical and biological aspects. The investigation of hematological malignancies requires an integrated diagnostic approach, incorporating a variety of laboratory techniques, including morphology evaluations, immunophenotyping, histopathology, molecular technology using polymerase chain reaction (PCR), karyotype analysis, fluorescence in situ hybridization (FISH), and next-generation sequencing (NGS). For instance, the high risk, TP53 and/or BIRC3 disruption; the intermediate risk, NOTCH1 and/or SF3B1 mutations and/or del11q22-q23 in the absence of TP53 and BIRC3 abnormalities are important in CLL. Hyperdiploidy of Odd chromosomes, abnormalities of 11q23, 6p21, 20q11, 4p16, 17p-, t (4;14) (C), and mutation of Ras FGFR3, CYLD, IAP, P53 genes and c-Myc expression (M) in diagnosis and prognosis as well as monitoring of myeloma are clinically important.

O64

### The Role of Medical Laboratory in Personalized Medicine

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The medical diagnostic laboratory plays a pivotal role in health-system as an exciting profession that remains hidden behind the medical treatment or investigation scene. So that, up to seventy five percent of clinical medicine decision making is predicated upon, or confirmed by, or documented by medical laboratory test results. Members of this challenging profession are responsible for providing accurate, reliable and timely laboratory tests for monitoring health, diagnosing and treating disease, assist in rapid identification of disease, assessment of severity of disease, creation of a therapeutic plan and the more efficient the testing protocol. In other hand, personalized medicine as a new approach to understanding, preventing and treating disease relied on our understanding about data and information on each individual's molecular and genetic profile alongside environmental differences and it is the tailoring of safer and more effective medical treatments based on individual characteristics of each patient leading to significant reducing risk and expenditure associated with selecting an appropriate therapy protocol for patient treatment, appropriate use of medicines/drugs, appropriate use of medical procedures associated with diagnosing or management/monitoring of treatment outcomes. Such approaches are a bright new era of safe and effective treatments for each patient with positive impact for having reasonable cost on healthcare system. Personalized medicine is considered to manage patient care in many diseases specially in cardiovascular diseases or cancer as two main causes of death in many societies.

**Keywords:** Personalized Medicine, Medical Laboratory, Health-Care System

**O65**

## **The Importance of Specialized Laboratory Tests in the Outcome of Hematologic Malignancies Treatment**

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Hematologic malignancies include various types of heterogeneous diseases in clinical and biological aspects. The investigation of hematological malignancies requires an integrated diagnostic approach, incorporating a variety of laboratory techniques, including morphology evaluations, immunophenotyping, histopathology, molecular technology using polymerase chain reaction (PCR), karyotype analysis, fluorescence in situ hybridization (FISH), and next-generation sequencing (NGS). The relative importance of miscellaneous investigations differs by disease entity. In addition to assisting in the diagnosis, each technique can play a role in determining the specific treatment approach for each individual. Immunophenotyping detects cellular antigens, and it is also used for disease staging and monitoring to detect surrogate markers of genetic aberrations, identify potential immunotherapeutic targets, and aid prognostic prediction. Molecular genetic methods facilitate the detection of mutations, rearrangements, or translocations in genes. According to cytogenetic and molecular findings, the clinical hematology guidelines are changed since the patients can be stratified into favorable intermediate or unfavorable categories. Applications in malignant hematology include confirming clonality, detecting disease-associated genotypes, determining prognosis, disease monitoring following therapy, predicting imminent clinical relapse, and in respect to therapies, targeted and immunomodulatory therapies guarantee better results with fewer hematological toxicities. On the other hand, advanced diagnostics enable the clinical team to identify the specifics of each person's disease, divide patients into subgroups that differ in their susceptibility to a condition, prognosis, or probability of responding to a specific therapy, and personalize a course of treatment for the best possible results.

**Keywords:** Hematological Malignancy, Laboratory Tests, Patients' outcome

O66

### **New approaches to acute myeloid therapy; targeting multiple signaling pathways**

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Acute myeloid leukemia (AML) is the most common diagnosed leukemia in adults and the second common leukemia in children. Despite its high prevalence, little improvements are achieved for treating this disease over the last 50 years. The reason is that AML is a heterogeneous disease. AML originates from a dominant mutation, then acquires collaborative transformative mutations leading to myeloid transformation and clinical/biological heterogeneity. This pathophysiology leads to use several targeted therapies simultaneously or beside other standard cytotoxic therapies. Some of the pathways and main goals of the growth and metabolism of AML which can be targeted for treating AML are: 1) Apoptosis pathways 2) Tyrosine kinase receptors 3) Mitochondrial mechanisms 4) Immunotherapy. In a study named “Beat AML Master Trial”, patients with AML disease  $\geq 60$  years were enrolled and divided to two groups. The first group treated with standard therapies (regarding patient condition). The second group were treated with targeted therapies with/ without classic treatments after assessment of special genetic disorder/disorders that is responsible for the disease. This study revealed that patients in the second group (received the targeted therapies) had better results than patients in the first group (received the classic therapies).

**Keywords:** Acute Myeloid Leukemia, Targeted Therapies, Multiple Signals

O67

## Laboratory Advances in Chronic Lymphocytic Leukemia (CLL) Diagnosis and Monitoring

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Chronic lymphocytic leukemia is the most common form of adult leukemia around the world with several atypical morphologies and Immunophenotyping. Some patients may never require treatment, while others may experience poor outcomes. Recent advances in laboratory techniques have significantly revolutionized the consequences of the disease and have focus on accurate diagnosis, risk stratification, finding targets for therapy and patients monitoring. Four CLL subgroups have been categorized based on the mutational and cytogenetic status using FISH, MLPA, Sanger Sequencing and NGS: the high risk, TP53 and/or BIRC3 disruption; the intermediate risk, NOTCH1 and/or SF3B1 mutations and/or del11q22-q23 in the absence of TP53 and BIRC3 abnormalities; the low risk, +12 and wild-type for all genetic abnormalities; the very low risk, del13q14 as the only genetic abnormality. New findings have proved independent risk factor along with above genetic markers for IGHV-UM. These progresses led experts to challenge the role of a “watch and wait” approach in asymptomatic high-risk patients. Using chemoimmunotherapy and more recently novel small molecules into first-line and relapsed patients, high risk patients experience substantial improvement in treatment outcomes. As these patients experience longer progression-free survival and more deep remissions there is an urgent need for sensitive methods such as multi-colour flow cytometry, real-time quantitative polymerase chain reaction (RQ-PCR) and high-throughput sequencing (HTS) for quantification of residual disease after therapy. MRD- guided treatment strategies have strong predictive value in determination of allo-HCT and CAR T cell recipient candidate as well MRD status is in strong relationship with disease biology as well.

**Keywords:** Chronic Lymphocytic Leukemia (CLL), Cytogenetic, Multi-Colour Flow Cytometry (MCF), Real-Time Quantitative Polymerase Chain Reaction (RQ-PCR), Minimal Residual Disease (MRD)

O68

### **Immunophenotype Diagnosis of Leukemia and Lymphoma Based on Computational Thinking: A New Glance in Hematology**

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**Background:** The routine use of flow cytometry in laboratory for clinical cases of lymphoma and leukemias began in 1985. Since then, the flow cytometry has gradually become a routine instrument not only in large medical centers, but also in medium size laboratories. While the personnel in most laboratories are technically capable of handling the flow cytometer, many technicians still lack the expertise to tailor monoclonal antibody panels for various diseases and interpret complicated results. Although there are many authoritative books available on flow cytometry, finding a comprehensive source on in-depth discussion of cases is difficult. On the other hand, it has not been ever any diagnostic and intelligent analytical software for interpreting hematological malignancy based on immunophenotyping data. The aim designing of this software is to meet the needs of the flow cytometry technicians by providing them with standard panels of monoclonal antibody, analysis of results and diagnosis of leukemia and lymphoma. **Designing Method:** firstly, software data base was gathered from more than 180 references including: text book, original articles and also case reports. Eventually diagnostic algorithm was designed by PHP and My SQL in web application platform. **Results:** This software is able to diagnosis of complicated case of leukemia and lymphoma based on immunophenotyping results and eventually final results after adjust with patient clinical and other laboratory findings will be reported. Likewise, the software offers morphologic, cytochemistry, molecular and cytogenetic findings to confirm final diagnosis. **Conclusion:** due to break through in clinical hematology sciences, designing these soft wares which work based on computational thinking are necessary for diagnosis of hematologic disorders.

**Keywords:** Immunophenotype, Leukemia, Lymphoma, Computational Thinking

**O69****MIC-M Based on Multiple Myeloma Diagnosis and Monitoring****Nader Vazifeh Shiran 1 \***

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Since 2008, WHO has established the basis for the diagnosis of malignancies based on the MIC-M criteria. According to the MIC-M criteria, malignancies should be classified based on a panel of microscopic morphology and specific staining (M), immunophenotyping by flow cytometry, immunohistochemistry and immunofixation (I), karyotype, aneuploidy and Qualitative chromosomal changes (C) as well as qualitative and quantitative molecular tests such as PCR and Realtime-PCR (M) were examined and diagnosed, and among the positive and specific cases, recurrent and distinct cases were used for monitoring and MRD of the disease. Accordingly, the morphology of Russell and Dutcher body, Blastic pattern, Multi-nuclearity (M), CD56, CD117, CD79a, CD22, CD10, CD40 and Ki-67 expression, but CD19, CD27 and CD45 no-expression (I), Hyperdiploidy of Odd chromosomes, abnormalities of 11q23, 6p21, 20q11, 4p16, 17p-, t (4;14) (C), and mutation of Ras FGFR3, CYLD, IAP, P53 genes and c-Myc expression (M) in Diagnosis and prognosis as well as monitoring of myeloma are clinically important. As a result, targeted detection and monitoring of specific MIC-M markers can lead to personnel medicine.

**Keywords:** Multiple Myeloma, Diagnosis, amp, Monitoring

# Omics Technologies in Medical Laboratory

## O70 – O73

Recent advances in omics technologies including genomics, transcriptomics, proteomics and metabolomics have enabled personalized medicine to be investigated at an unpredictably detailed molecular level. In the recent years, omics technologies have progressively begun to enter clinical practice. However, each individual technology cannot overcome to the entire biological complication of most human diseases. Combination of multiple omics technologies has emerged as a promising approach to make available a more comprehensive view of biological processes and disease. However, for clinical approval of any technology, high sensitivity and specificity are required for detection and interpretation.

Omics profiling can be a more effective and cheaper tool for detecting large-scale data with more comprehensive results in comparison to performing thousands of individual tests. While challenges persist in establishing clinical guidelines, many of the concepts surrounding the interpretation of genetic variants (particularly rare or novel variants) may relate to a general molecular event (for example a differentially expressed gene, different protein phosphorylation or distinctive metabolome signature) as our understanding of the biology and reference databases developed. At the present time, it has been revealed that at least in some situations, omics technologies (mainly genome sequencing) may have more beneficial performance than traditional clinical tests, however, there are substantial technical and regulatory difficulties to incorporate these technologies into clinical practice. It is likely that combination of these technologies will become common place in future clinical practice to enables a clearer picture of health and disease.

**O70****Role and Application of Genomic Technology in Medical Diagnostic Laboratories****Hamid Reza Khorram Khorshid 1 \*, Emran Esmailzadeh 2**

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The growing application of genomic technologies encompasses all phases of life, from the embryo to the elderly, and even the postmortem phase. Genomic technology is used to diagnose, monitor, treat, predict and prevent disease, as well as promote good health in individuals, across communities and whole populations. Sanger sequencing was developed in the 1970s by Frederick Sanger and was the original method that enables scientists to evaluate the nucleotide sequences of DNA. Technological advances including Next Generation Sequencing (NGS) have allowed for greater integration of genomics into healthcare delivery, from screening and diagnostics, to the accurate detection of pathogens, and the ability to prescribe and monitor the efficacy of more precise therapeutics. NGS allows for the simultaneous sequencing of several DNA fragments, enabling multiple loci to be investigated at one time and therefore, more efficient and cost-effective genomic analysis. Most of the existing and developing usages of genomic healthcare technology comprise analyses for screening, diagnostic or prognostic purposes; testing to guide and evaluate treatment options; and identification and tracking of human disease-causing genes. Additionally, genomic technology is progressively available outside of healthcare settings through personal genomics tests that may be accessed directly by consumers, also known as direct-to-consumer or personal DNA tests. Frequent benefits have been provided using genomic sequencing technology, mainly the significant improvement in the provision rate of molecular diagnoses. Continued developments with massively parallel sequencing include greater sensitivity of detecting previously difficult disease-causing deletions and growing ability to detect copy number variants

**Keywords:** Genomic Technology, Next Generation Sequencing

O71

### **Role and Applications of Transcriptomic Technology in Medical Diagnostic Laboratories**

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Mortality during human life is triggered by three main causes; About 65% are due to multifactorial diseases, about 24% are due to cancer and less than 3% are due to genetic diseases. Definitive diagnosis, timely prevention and effective treatment management are very important. The occurrence of many multifactorial diseases or cancers in different people, despite similar diagnostic and therapeutic approaches, has led to different response, side effects and prognosis. Since the initial map of the human genome was identified in 2000, several projects have been undertaken to update and upgrade the information of this largest project in the history of medicine. Numerous countries around the world tried to prepare their population genomic map, and based on that, design and optimize their health structure. These results in the last two decades have led to the creation of new scientific fields, under the headings of personalized medicine and precision medicine, and new technologies in the field of genomes, including transcriptomics, have been invented as needed. Over the past decade, the growing fields of science and scientists' access to new technologies, in addition to optimally diagnosing and managing disease, have encouraged researchers to consider extending their lifespan, exploring the possibility of space flight and living on the other part of the galaxy. In this opportunity, we try to present and evaluate the role and position of transcriptomic technology in the field of laboratory diagnostics and its role in the optimal management of disease treatment.

**Keywords:** Transcriptomics, Multifactorial Diseases, Cancer, Precision Medicine

O72

## Role and Application of Proteomics Technology in Medical Diagnostic Laboratories

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Proteomics involves the large-scale study of proteins, their structures, functions, and physiological roles. Among other omic technology approaches, proteomics offers unprecedented opportunities to describe individuals' phenotypes as proteins translate the genetic information, respond to environmental perturbations as well as treatments. Clinical proteomics deals with the application of proteomic technologies to help decipher the changes that occur in cells, tissues, and organs under diseased conditions. In this regard, clinical proteomics expands the physicians' toolbox for personalized medicine-based patient care. Although proteomics development offers a great deal of promise in biomarker discovery, few assays or valid protein biomarkers have made their way into the clinical laboratory. The reasons for discrepancy between basic and clinical research in proteomics are in part due to the complexity of the characterization and quantification methods to accomplish it. Mass spectrometry (MS) is the key technology among common techniques for protein characterization in biological samples including western blot, enzyme linked immunosorbent assay (ELISA), two-dimensional gel electrophoresis, and nuclear magnetic resonance (NMR). Due to the recent developments in mass spectrometry, an important role for proteomics in early diagnosis is pending. This topic presents an overview of proteomic approaches in biomarker discovery and describes how proteomic methodology is applied in the medical field with a focus on SELDI-TOF-MS, a novel and powerful technique in the examination of clinical samples. Moreover, it focuses on the potential uses of proteomics in the diagnosis and treatment of multifactorial diseases including cancer, cardiovascular diseases, autoimmune diseases, and neurodegenerative diseases.

**Keywords:** Clinical proteomics, Biomarker, Mass spectrometry, Personalized Medicine, Clinical Laboratory

O73

### Role and Application of Metabolomics Technology in Medical Diagnostic Laboratories

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In recent years, metabolomics has become an inevitable tool in several clinical research fields, helping to discover new diagnostic biomarkers. As metabolites indicate end points of the gene expression, clinical metabolomics can provide a holistic approach for understanding the phenotype and allows us to measure the integrated effects of the environmental exposures, including pharmaceutical and nutritional, on disease onset, diagnosis, and progression. Nevertheless, metabolomics will play a key role in further development of personalized medicine. In addition to basic human biological processes, it is now clear that crosstalk between the microbiome and metabolome has a significant impact on disease onset and progression through a variety of mechanisms. For example, the microbiome gut-brain axis has been suggested to play an important role in Alzheimer's disease pathophysiology. In this regard, large-scale measurement of the metabolome, including basic human metabolites and microbiome metabolites, requires the combination of multiple orthogonal and complementary analytical platforms such as liquid chromatography-mass spectrometry (LC-MS), nuclear magnetic resonance (NMR), gas chromatography-MS (GC-MS), direct injection MS (DIMS), and capillary electrophoresis-MS (CE-MS). With this presentation, a comprehensive compendium of applied clinical metabolomics protocols for diagnostics in clinical laboratories as well as the predictive power of machine learning in data analysis for biomarker-based diagnostics will be described. Moreover, future trends and perspectives in clinical metabolomics will be presented.

**Keywords:** Clinical Metabolomics, Diagnostic Biomarkers, Personalized Medicine, Mass Spectrometry, Machine Learning

# **Sepsis and Systemic Inflammation: Laboratory Verification of Diagnosis**

## **O74 – O76**

Sepsis is a significant public health problem across the world, with more than 31 million cases annually and a 17% mortality. Sepsis is a systemic host response to microbial pathogens that results in significant morbidity and mortality. The concept of the Systemic Inflammatory Response Syndrome (SIRS) was proposed in 1992 and now, major revision has been modified. An accurate and timely diagnosis of sepsis allows prompt and appropriate treatment. This panel discusses laboratory testing for sepsis because differentiating systemic inflammation from infection is challenging. Procalcitonin (PCT) is currently an FDA approved test to aid in the diagnosis of sepsis but with questionable efficacy. However, studies support the use of PCT for antibiotic de-escalation. Serial lactate measurements have been recommended for monitoring treatment efficacy as part of sepsis bundles. The 2016 sepsis consensus definitions include lactate concentrations greater than 2 mmol/L (>18 mg/dL) as part of the definition of septic shock. Also included in the 2016 definitions are measuring bilirubin and creatinine to determine progression of organ failure indicating worse prognosis. Hematologic parameters, including a simple white blood cell count and differential, are frequently part of the initial sepsis diagnostic protocols. Several new biomarkers have been proposed to diagnose sepsis or to predict mortality, but they currently lack sufficient sensitivity and specificity to be considered as stand-alone testing. If sepsis is suspected, new technologies and microbiologic assays allow rapid and specific identification of pathogens.

O74

### Treatment of Septicemia & Immunomodulation

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Sepsis is a life-threatening organ dysfunction that results from the body's response to infection. It requires prompt recognition, appropriate antibiotics, careful hemodynamic support, and control of the source of infection. With the trend in management moving away from protocolized care in favor of appropriate usual care, an understanding of sepsis physiology and best practice guidelines is critical. Delay in giving appropriate antibiotics is associated with a significant increase in mortality rate. Appropriate antimicrobials should be initiated within the first hour of recognizing sepsis, after obtaining relevant samples for culture—provided that doing so does not significantly delay antibiotic administration. The initial antimicrobial drugs should be broad-spectrum, covering all likely pathogens. Multidrug regimens are favored to ensure sufficient coverage, especially in septic shock. The empiric choice of antimicrobials should consider the site of infection, previous antibiotic use, local pathogen susceptibility patterns, immunosuppression, and risk factors for resistant organisms. Double coverage for gram-negative organisms and for methicillin-resistant *Staphylococcus aureus* (MRSA) should be considered for patients with a high likelihood of infection with such pathogens. Double gram-negative coverage may be appropriate when a high degree of suspicion exists for infection with multi-drug-resistant organisms such as *Pseudomonas* or *Acinetobacter*. If a nosocomial source of infection is suspected to be the cause of sepsis, anti-MRSA agents are recommended. The last few decades have seen a 200% rise in the incidence of sepsis due to fungal organisms. Antifungals should be considered for patients at risk, such as those who have had total parenteral nutrition, recent broad-spectrum antibiotic exposure, perforated abdominal viscus, or immunocompromised status, or when clinical suspicion of fungal infection is high. Risk factors for fungal infection in septic shock should trigger the addition of echinocandins or liposomal amphotericin B. Azoles are considered appropriate for hemodynamically stable patients. Sepsis is associated with vasodilation, capillary leak, and decreased effective circulating blood volume, reducing venous return. These hemodynamic effects lead to impaired tissue perfusion and organ dysfunction. The goals of resuscitation in sepsis and septic shock are to restore intravascular volume, increase oxygen delivery to tissues, and reverse organ dysfunction. A crystalloid bolus of 30 mL/kg is recommended within 3 hours of detecting severe sepsis or septic shock. However, only limited data support the benefits of this recommendation, and evidence of harm from sustained positive fluid balance is growing.

**Keywords:** Sepsis, Antimicrobials, Antifungals

**O75****Sepsis in Immunocompromised Patients****Sara Abolghasemi 1 \***

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The classic clinical features of infection and sepsis may be absent in immunocompromised patients owing to their reduced ability to mount an immune response. Although immunocompromised patients are at high risk of sepsis and septic shock, it has been observed that their mortality rate due to sepsis is no greater, and is in some cases less than the rate seen in nonimmunocompromised patients. SEPSIS IN SOT RECIPIENTS: Classic features of sepsis, such as leukocytosis and fever, may be absent, whereas thrombocytopenia and organ failure may be more pronounced. Patients with CMV serologic mismatch have the highest risk for CMV reactivation; because of CMV reactivation, through immunomodulatory mechanisms, the risk of subsequent bacterial and fungal infections is increased. the type of surgical procedure, cold ischemia time, length of surgical procedure, amount of blood loss and transfusions during transplant, the net state of immunosuppression, the use of T-cell-depleting antibodies for induction or rejection, and the use of previous myeloablative regimens are additional risk factors. General management principles for sepsis in SOT recipients do not differ significantly from those in patients who are not transplant recipients.

O76

### The Rule of Procalcitonin in the Diagnosis and Treatment of Patients

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Procalcitonin (PCT) is formed in IL6-mediated, IL8-mediated, and TNF $\alpha$ -mediated systemic inflammation conditions, in multiple organs and structures of the body. In patients with sepsis, significantly increased PCT levels are found. The PCT levels are highly correlated with the severity of the illness, and decreased PCT levels under therapy correlates with a better prognosis. In the differential diagnosis, measuring the PCT level helps differentiate between bacterial and viral infections. Noninfectious inflammatory reactions can, however, show moderately increased PCT levels. Cut-off values depend on renal and hepatic function. A therapeutic algorithm using PCT levels could be used for determining duration of a course of antibiotics, which can reduce antibiotic usage. In this review, the differential diagnostic and differential therapeutic possibilities of PCT levels for critically ill patients are discussed and various modalities for better clinical efficacy of PCT will be presented.

# **Serologic Diagnosis of Infectious Disease (H. Pylori and Brucellosis)**

## **O77 – O79**

Isolation and identification of microorganisms is one of the best methods to diagnose an infectious disease. However, in most cases, isolation of the microorganism is not possible due to the use of antibiotics, presence of fastidious microorganism and delay in the patient's visit to clinician. In such cases, indirect diagnostic methods for detecting of the microorganisms will be the best and fastest mean to diagnose the disease. One of these methods is serological tests, which are based on detecting specific antibodies against the microorganism in the serum of patients. Using serological tests, it is possible to detect changes in the titer of antibodies during the course of the disease and to treat or control the disease. This means that determining the antibody titer in the acute stage of the disease and the convalescence stage can indicate an accurate diagnosis. However, various factors such as the patient's age, duration of illness, patient history, stage of the disease, consumption of drug, vaccination status, repeated exposure to microorganisms, endemicity of the disease in a geographical area and occupational risks should be considered when interpreting serological tests. Of limitations of serological methods in the diagnosis of infectious diseases are low specificity, difficulty in interpreting the results, and distinguishing between active infection and past infection. Despite these limitations, the low cost of serological tests, the feasibility of testing - especially for infectious agents whose diagnosis is based on invasive methods (such as *Helicobacter pylori*) - and the high negative predictive value of these tests, especially for diagnosis of Brucellosis cases should not be ignored.

O77

### **Invasive and Non-Invasive Diagnostic Methods for Helicobacter Pylari**

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There are several methods used for diagnosis of *H. pylori* infections, which are classified into two broad categories: invasive methods, which require endoscopy, and the minimally or non-invasive methods. The invasive methods, in which the biopsy sampling is needed, include growth and culturing of bacteria, Rapid urease test (RUT) or CLO test (Campylobacter-Like Organism test), Polymerase chain Reaction (PCR), fluorescence in situ hybridization (FISH), direct gram staining, histological tests, blue laser imaging (BLI), linked color imaging (LCI), and magnifying endoscopy, while the non-invasive methods include immunological methods, urea breath test (UBT), *H. pylori* stool antigen assay (HpSA) and Stool PCR test. The noninvasive methods are often considered as the first line diagnostic methods, but the sensitivity and specificity of these methods vary according to their entities and host conditions. Artificial intelligence to predict *H. pylori* status based on endoscopic images help clinicians to detect both infection status and histological changes in symptomatic patients. Detection of antibiotic resistance is clinically important for treatment. Although bacterial culture from the gastric biopsy is currently the gold standard and is recommended for antibiotic susceptibility test, increasing attention has been paid to PCR and molecular tests, such as line probe assay, for detection of the resistance genes and nucleotide polymorphisms linked to resistance phenotypes. In future, development of new technologies, such as NGS and transcriptomics, could help us to determine resistance of *H. pylori* strains to diverse antimicrobial agents.

**O78**

### **Does the Results of the Brucella Laboratory Tests Are in Agreement with the Clinical Findings?**

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There are many laboratory requests by the physician for the patients suspected of having brucella infection. The laboratory tests for detection of brucella are wright, 2-ME, anti- brucella IgM/IgG determination and in small cases PCR-specific test for brucella. The overall result from some of medical diagnostic laboratory in Tehran has indicated that the majority of tests were negative. Furthermore, in many of the cases, the results of the above mentioned tests are not comparable with each other which further make it uncertain the initial clinical recognition of brucella in patients.

O79

### Review of Detection of *Brucella* spp. by Molecular Methods in Clinical Laboratory

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Brucellosis is a common disease between humans and animals that causing a high degree of morbidity in humans. Despite the use of culture and serological techniques, definitive diagnosis of brucellosis is still one of the main problems of some countries, especially developing countries. Due to the low sensitivity of the culture method and serological tests (especially in the early stages of the disease where the production of antibodies is low), to create cross-reactions with other microorganisms and false positive and negative results in them, the need for better, more accurate, more sensitive diagnostic methods for brucellosis especially in endemic regions (of which Iran is one of these regions) is strongly felt. For this reason, it is now believed that the key tools for diagnosing brucellosis are molecular techniques that are fast, easy, and reliable, and that these methods can even identify patients without clinical symptoms and in the early stages of the disease and also very appropriate to evaluate the early stages of treatment and follow up after treatment. Molecular methods are very suitable for diagnosing *Brucella* because they have acceptable specificity and sensitivity and pose less risk to laboratory personnel, these methods carry less risk for laboratory personnel. The efficiency of these methods in cases where the culture results are negative and serological tests have false positive or negative results indicates its high sensitivity to these methods. Molecular methods can also be used in cases where the person has taken antibiotics or the amount of laboratory sample is low.

# The Power of Big Data in Laboratory Medicine: Establishing Reference Intervals

## O80 – O84

**Background:** Reference intervals (RI), historically known as ‘normal ranges’ are fundamental for laboratory report interpretation. RIs are ideally established by selecting a statistically sufficient group of apparently healthy individuals. However, defining health is the main challenge in any investigation. Health is a relative term without a well-accepted definition. Health is profoundly influenced by many parameters, including race, age, gender, geographic area of residence, environmental factors and physiological conditions such as puberty, pregnancy, lactation, menopause and menstruation. In this regard, RIs must define a representative window of normality for assessment of health and disease in specific populations. It is practically impossible for any laboratory to establish its own RIs due to many limitations, including high cost and health definition. On the other hand, relying on the RIs provided by the manufacturers might not be appropriate as most are established in a different ethnic population from the target population. **Method:** The Iranian laboratory reference interval project (IRLAR) was designed by Iranian Association of Clinical Laboratory Doctors (IACLD) to estimate RIs using a computerized indirect Truncated Maximum Likelihood (TML) method. Data generated for seventeen analytes including FBS, total cholesterol, LDL cholesterol, HDL cholesterol, triglyceride, SGOT, SGPT, ALKP, urea, creatinin, uric acid, total protein, albumin, total bilirubin, direct bilirubin, LDH and TSH from 2015-2018 was collected retrospectively from more than 50 medical laboratories in 30 provinces of Iran without filtering and exclusion. For each analyte, at least one million data and in some cases more than 10 million data was recorded. RIs are being calculated for each gender and different age ranges and the accuracy of RI estimation is being assessed by comparing the results with published peer-reviewed studies. **Results:** Compatibility of the calculated and published RIs for these analytes will be presented in the IACLD 2022 congress. **Conclusion:** With extensive physiological variations affecting RIs and also difficulties for defining RIs by individual clinical laboratories, the results of this survey will delineate to what extent the indirect TML method could be a basis for accurately establishing RIs for different laboratory analytes.

O80

### **Reference Interval Harmonization in Canada: Harnessing the Power of Big Data Analytics**

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Harmonization in laboratory medicine from specimen collection to result reporting is critical to ensure consistent and accurate clinical decision-making. Harmonized or common RIs refer to using one interpretative recommendation for an analyte across several laboratories, regardless of analytical assay or patient population. Harmonized or common RIs should therefore only be considered for assays that demonstrate minimal bias across considered methodologies. Several national surveys have reported wide variation in reference intervals across healthcare centres in certain regions, even those using the same analytical platform for test measurement. There is a high risk of inappropriate test result interpretation when reference intervals are not appropriately harmonized. The Canadian Society for Clinical Chemistry (CSCC) Working Group on Reference Interval Harmonization was established in 2015 to develop evidence-based harmonized/common reference intervals (hRIs) and support their implementation in laboratories across Canada. After comprehensive review of the literature, our group established a novel approach to reference interval harmonization in adults involving: extraction of data from outpatient community reference laboratories across Canada, 2) assessment of outliers and monthly instability, 3) statistical evaluation of age, sex, and centre-specific differences, 4) derivation of preliminary harmonized reference intervals using a new indirect method (Truncated Maximum Likelihood method), 5) comparison of established harmonized reference intervals to direct a priori data in the healthy Canadian population and 6) verification through a cross-Canada prospective program. Thus far, this approach has led to the development of harmonized reference intervals for 17 biochemical and immunochemical markers. In this presentation, we will discuss the work completed by CSCC hRI WG, challenges encountered, and future plans to support implementation.

O81

## Direct Method for Determination of Reference Interval and Its Challenges

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Interpretation of test results is strongly depend on reference interval with which patient results is compared. It is the responsibility of the laboratory to provide appropriate reference interval for their reported results. There are two direct and indirect methods for establishing reference interval. In direct approach reference interval is defined according to the results of reference individuals which themselves are selected and partitioned according to pre-established inclusion, exclusion and portioning criteria, such as age, sex, drug consumption or food intake. After selection of reference individuals, specimens are collected in a satandardized pre-analytical condition and samples are analyzed by intended method. Results of theses analyses need to be assessed for data distribution and detection of outliers. According to data distribution which may be symmetric (Gaussian) or asymmetric, parametric or nonparametric methods may be used for establishing reference interval, respectively. If distribution is asymmetric and it has skewness or kurtosis, mathematical transformations may change it to a symmetrical distribution. Direct method which is recommended by IFCC and CLSI, has many challenges which make it difficult to be used. The most important of theses challenges are definition and selection of reference individuals, detecting of erroneus and outlier results, and high cost. These practical problems have led to the suggestion of using indirect method which is involves deriving reference limits from using results collected for other purposes. This method is simpler and less expensive, but has its own challenges and should be used with care.

**Keywords:** Reference Interval, Interpretation, Test Result

O82

### **A review of Differentiation between Reference Intervals and Clinical Decision Limits**

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Reference intervals (RIs) and clinical decision limits (CDLs) play critical roles in the interpretation of laboratory data. Although they have different concepts, sometimes these two terms are confused with each other, leading to misinterpretation. Therefore, it seems necessary for all health professionals to know their differences. For the serum level of a metabolite or a health indicator (blood pressure, weight, etc.), the RIs are based on the distribution (typically, the central 95% interval) in an apparently healthy group that should be approximately representative of the general population. It should be noted that values outside of a RI are not necessarily related to an adverse clinical outcome. Furthermore, based on RIs only, individuals should not be tagged as patients or be a candidate for clinical intervention. The CDLs, on the other hand, are ranges that were suggested to have an association with higher risk of clinical adverse outcomes, such as diseases or their complications. The CDLs were mainly assessed from population-based longitudinal studies. Moreover, most of the clinical diagnostic criteria, preventive interventions, and therapeutic criteria are based on CDLs, not RI. The present review aims to define these two terms and their different applications. Finally, considering the importance of the values listed in the reference column of the laboratory results, it will be discussed which of the RI and CDL is the better choice for this column to minimize misinterpretation.

**Keywords:** Reference Interval, Clinical Decision Limits

# Updates on Hepatitis B and C Vaccine and Laboratory Diagnosis

## O85 – O88

Hepatitis B and C infections are considered as a major health problem worldwide. Both infections are transmitted through blood-borne viruses causing life threatening chronic diseases. Early laboratory diagnosis and vaccine development are the main strategic plans implemented by local and international health organizations for the effective control of these diseases. Both molecular and serological assays are available for detection of the viruses and their antigens in blood of infected individuals, however, there is still an urgent need for development of more sensitive assays for early detection of infection and minimizing the window period from the time of infection. Novel diagnostic approaches are being developed, particularly for more sensitive and effective detection of hepatitis C. Considering the influence of mutations and genotype variability on serological detection of hepatitis B and C viruses, selection of more broad cross-variants reactive monoclonal antibodies is essential for more accurate detection of a variety of genotypes and escape mutants of both viruses, particularly hepatitis B virus. These mutations may also affect vaccine effectiveness. Therefore, monitoring these mutations is essential for vaccine development as well as the design of more sensitive and specific diagnostic assays. In this panel of the congress our understanding of the current knowledge and future trends on both topics of vaccine and diagnosis of hepatitis B and C infections will be presented by experts in the fields of Immunology and Virology.

O85

### Hepatitis B Vaccination and Immunoprophylaxis - Key Concepts

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Hepatitis B virus (HBV) infection is a major health problem worldwide. At present, more than 240 million people are chronically infected of whom 1 million die annually. Despite the recent advances in antiviral therapy, vaccination is still the most effective strategy for infection control. The recombinant surface antigen of HBV (HBsAg) is currently employed as an effective vaccine in most countries including Iran. Considering the major transmission route of HBV from mothers to offspring, vaccination with 3 doses of the vaccine starting at birth has been considered for the Expanded Program of Immunization (EPI) worldwide. This immunization protocol induces an active protective immune response. Neonates born from infected mothers need simultaneous administration of active and passive immunizations using hyperimmune HB antibody. Protective antibody response is induced in most vaccinated healthy neonates and adults, but a small proportion of subjects fail to respond. Many parameters contribute to vaccine immunoprotectivity, such as type and dose of vaccine, vaccination schedule and type of adjuvants. In this presentation the national HB vaccination program will be reviewed and recent investigations and achievements in this field will be presented and discussed.

**Keywords:** Hepatitis B, Vaccination, Immunogenicity, Immunoprotection

O86

## Effect of HBs Antigen Mutations and Genotypic Variations on HBV Infection Diagnosis

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Screening for HBV infection is based on the detection of HBsAg by enzyme immunoassay. Confirmation of infection is based on the detection of circulating viral DNA. Although PCR and ELISA-based methods have good specificity, they are not always sensitive enough to detect HBV in patients' samples. Antibody response to HBsAg controls HBV infection. "a" determinant of HBsAg is the most important target for protective antibody response, diagnosis and immunoprophylaxis. Amino acid substitutions in the "a" determinant of HBsAg, whether originating from genetic diversity or from mutations in the HBV strain itself, could affect the sensitivity of diagnostic kits. Methods: We have so far produced and characterized a panel of anti-HBs monoclonal antibodies (mAbs) and evaluated their reactivity with different mutants of HBsAg and also different subgenotypes of HBV. Results: Our results have shown that the most influential mutations in "a" determinant includes T123N and Q129L, located in the proximal loop, and G145R and D144A, located in the distal loop. Reactivity pattern of our anti-HBs mAbs panel with different subgenotypes of HBV also showed that almost half of mAbs showed a moderate to profound loss of reactivity with HBV genotypes/HBsAg subtypes D2/ayw3, E/ayw4, F2/adw4, and H/adw4. Amino acid sequencing of HBsAg showed that substitution at residue 127, located inside "a" determinant, is observed in all of these samples. Conclusion: Diagnostic efficacy of HBsAg detection kits is affected by amino acids changes in "a" determinant. Laboratories should therefore be aware of the analytical sensitivity for HBsAg assays, utilizing mAbs against HBsAg.

**Keywords:** HBsAg, HBsAg Mutations, HBV Genotypic Variations, anti-HBs Monoclonal Antibody, HBsAg Diagnostic Kits

O87

### New Approaches to Simplify HCV Diagnostic Algorithms

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At the present time, the availability of highly effective direct acting antiviral agents (DAAs) not only has revolutionized the treatment of chronic HCV infection but also has changed the paradigm for its management. Since most people infected with HCV are unaware of their infection, despite the advances and opportunities for treatment, they go for therapy when they have advanced liver disease. The key reason for patients' unawareness is the low rate of HCV testing and diagnosis. Method: The first global health sector strategy on hepatitis C virus which was adopted by WHO calls for a 90% diagnosis rate for the year 2030 globally, a goal that is unlikely to be achieved unless the diagnosis algorithms for HCV can be markedly simplified in the near future. In recent years, new approaches have been developed to simplify diagnosis of HCV. They include 1) use of dried blood spot sampling for both serological and NAT technologies for HCV infection 2) use of HCV c Ag test which detects core Ag early and during the course of HCV infection as a surrogate marker of viral replication 3) use of new Point-of-care (POC) tests for HCV RNA detection and RNA copy number determination 4) use of pan genotypic DAA therapy. In this way the test for determination of the viral genotype can be skipped from the HCV diagnosis algorithm. Conclusion: In addition to more identify infected patients, this simplification increases rates of retention and linkage to treatment, reduces the costs of diagnosis, viral transmission and progression of liver disease and hepatitis-related mortality.

**Keywords:** HCV, Diagnosis, HCVcAg, Direct Antiviral Agents

**O88****HCV Vaccine: Updates and Highlights****Nastaran Ansari 1 \***

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The incidence of hepatitis C virus (HCV) infections remains high even more than 10 years after approval of the first direct-acting antivirals for treatment of hepatitis C. In some countries, more people are newly infected with the virus than patients cured by antiviral therapy. The development of a prophylactic vaccine could prevent virus transmission and thereby make a significant contribution to control the global burden of this disease. HCV is a highly diverse and versatile virus that mostly escapes the immune system and establishes chronic infections. However, up to one third of the exposed individuals can spontaneously resolve HCV infections, which indicates that protective immunity can be achieved. Numerous studies on determinants of protective immunity against HCV show an increasingly complete picture of what a vaccine must achieve. It is very likely that both strong neutralizing antibodies and powerful cytotoxic T cells are needed to reliably protect against chronic HCV infection. The key question is which approaches allow maturation of particularly broadly effective antibodies and T cells. This will be necessary to protect against the high number of different HCV variants. Different vaccines have been developed over years including epitope vaccines, vector vaccines, recombinant protein vaccines, and DNA vaccines. The recent successes of mRNA vaccines open new doors for HCV vaccine research and development. Combined with a deeper understanding of the structure and function of the viral proteins, the identification of cross-protective antibody and T-cell epitopes as well as the use of standardized methods to quantify the effectiveness of vaccine candidates, new perspectives arise for the development of a vaccine.

**Keywords:** HCV, Vaccine, Immunity



# New Findings in Laboratory Science (Young Scientists Session)

**O89 – O91**

O89

## Human Cytomegalovirus and Epstein-Barr Virus Infections in Breast Cancer: A Molecular Study on Iranian Women

Hadi Ghafari \*

**Background and Objectives:** The role of human cytomegalovirus (HCMV) and Epstein-Barr virus (EBV) infections in breast cancer pathology is not well understood. Our study aimed to investigate the association of HCMV and EBV infections with breast cancer and distinguish the types of positive EBV and LMP-1 samples in Iranian patients. **METHODS:** Seventy-two formalin-fixed paraffin-embedded (FFPE) breast cancer tissues were analyzed between December 2014 and April 2016. Samples were analyzed for HCMV and EBV using nested-PCR and conventional PCR assays, respectively. Statistical analysis was performed using SPSS software version 18. **RESULTS:** Overall, HCMV and EBV genomes were detected in 6.9% and 16.7% of FFPE breast cancer tissues, respectively. Clinical factors were not statistically associated with the presence of HCMV and EBV. **CONCLUSION:** In this study, we reported EBV and LMP-1 typing in breast carcinoma cases for the first time in Iran. Our findings indicate that HCMV and EBV infections are not associated with the development of breast cancer.

**Keywords:** HCMV, EBV, Breast Cancer, Carcinoma

O90

### Evaluation of Salivary Glands Proteomes from *Phlebotomus Papatasi*-Proven Vector of Zoonotic Cutaneous Leishmaniasis in Iran

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**Background:** Zoonotic Cutaneous Leishmaniasis is increasing in the world and *Phlebotomus papatasi* as a proven vector was considered in different countries especially in middle east. Sandfly salivary glands which provoke host immune system are candidates for developing vaccines. The main purpose of this research was comparing evaluation of salivary glands proteomes from wild *P. papatasi*. Extracting these proteins and purifying of original SP15 as inducer agent in vector salivary glands from endemic leishmaniasis foci were other objectives. **Methods:** Adult sandflies were sampled using aspirators and funnel traps and CDC light trap. Each pair of salivary glands of unfed females was dissected and proteins were extracted using thermal shocking and sonication methods. Purification was performed through RP-HPLC. **Results:** The protein concentration of whole-salivary glands of specimens was determined approximately 1.6  $\mu\text{g}/\mu\text{l}$  (Isfahan) and 1  $\mu\text{g}/\mu\text{l}$  (Varamin and Kashan). SDS-PAGE revealed 10 distinct bands between 10 and 63 kDa. Analysis of proteomes showed some similarities and differences in the chromatograms of different foci. **Conclusions:** Isolation of salivary components of Iranian wild *P. papatasi* is very important for finding potential proteins in vaccine development and measuring control strategy of zoonotic cutaneous leishmaniasis in Iran and elsewhere in the world.

**Keywords:** *Phlebotomus papatasi*, HPLC, PpSP15, Salivary glands, Zoonotic Cutaneous Leishmaniasis

O91

## The Effect of NaCl Concentration on the Activity of a Chimeric Endolysin Against *Staphylococcus Aureus*

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**Background:** *Staphylococcus aureus* is a gram-positive bacterium that causes a wide variety of clinical diseases. The treatment remains challenging due to the emergence of multi-drug resistant strains. Endolysins are essential bacteriophage encoded enzymes in the lytic phage life cycle, which hydrolyze the host cell wall, so they are considered promising antibiotic substituents for fighting against multidrug-resistant bacteria. One of the most essential parameters in enzymatic reactions is the choice of buffer and its composition. In this study, we optimized the NaCl concentration for achieving the highest lytic activity of a recombinant chimeric endolysin against *S. aureus*. **Methods:** To test the effect of salt concentration on the lytic activity of the chimeric protein, two different buffers including monosodium phosphate buffer and Tris (hydroxymethyl) aminomethane buffer at pH 8.5 were used. The enzyme activity was evaluated in buffers containing different NaCl concentrations ranging from 0 to 1000 mM. The lytic activity against a standard *S. aureus* strain was determined with turbidity reduction assay. **Results:** Our findings indicated that the highest lytic activity of the chimeric protein against *S. aureus* was in the absence of NaCl. No significant lytic activity was detected at concentrations of 400 and 500 mM. **Conclusion:** The endolysins of bacteriophages infecting gram-positive bacteria contain both catalytic and binding domains. It seems when the salt concentration is too high, the normal interaction of charged groups in these domains will be blocked and the structure of the protein will be denatured. This structural change decreases the rate of reaction and increases the time required to reach maximal lytic activity.

**Keywords:** Chimeric Protein, *Staphylococcus Aureus*, Endolysin, Multi-Drug Resistant Bacteria

# Lipid Profiling in Clinical Laboratory

## 092 – 093

Nowadays, cardiovascular diseases, obesity, fatty liver and diabetes, as inflammatory diseases, are directly related to diet, lifestyle and disorders of lipid and lipoprotein metabolism. Therefore, lipids and lipoproteins measurement, the related pathophysiology, and metabolism abnormality are very important.

Blood lipids come from both dietary (exogenous) and hepatic (endogenous) sources. Triacylglycerol (TAG) increasing, which mainly comes from dietary origin, causes obesity and fatty liver, therefore its related disorder can be controlled through diet therapy. On the other hand, free cholesterol (FC) essentially originates from liver, is a major cause of cardiovascular disease and its treatment is possible through pharmacological approaches.

Lipids have relatively hydrophobic properties that cannot exist freely in liquid environments such as blood. As a result, in the bloodstream, lipids circulate by specific proteins called apolipoproteins (Apo) in the form of lipoproteins such as chylomicron (Chy), very low density lipoprotein (VLDL), Intermediate-density lipoprotein (IDL), low density lipoprotein (LDL), high density lipoprotein (HDL) and lipoprotein(a) (Lp(a)).

Because of the importance of blood lipids (TC and TAG), their blood measurement is pivotal for diagnosis, treatment and screening of patients. Furthermore, identifying molecular and cellular mechanisms in health and diseases condition seems necessary. In this article, we intended to discuss about blood lipids and lipoproteins in three aspects including; 1) pathophysiology, 2) measurement and interpretations, and 3) disorders in clinical laboratory.

What is very important in this regard is the pathophysiology of lipids and lipoproteins and their related molecular mechanism in non-fasting and fasting state.

O92

## Pathophysiology of Lipid Profile (Lipids and Lipoproteins) in Clinical Laboratory Exogenous (Non-Fasting) and Endogenous (Fasting) Lipid Profile

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Recently, exogenous (Triacylglycerol, TAG<sub>exo</sub>) and endogenous (total cholesterol, TC<sub>endo</sub>) attracts lots of attention; because cardiovascular-diseases especially atherosclerosis, obesity, and fatty liver caused by high fat diet and abnormality in lipids, lipoproteins, and apolipoproteins metabolism. Lipid, lipoprotein, and apolipoprotein profiles obtain from two sources: diet (exogenous) and synthesized in the liver (endogenous). Mainly, the endogenous lipid profile is investigated in clinical laboratory and exogenous lipid profile doesn't take into account. Exogenous lipids mainly consist of TAG<sub>exo</sub>, while endogenous lipids are mainly TC<sub>endo</sub>. Actually, TAG<sub>exo</sub> have a pivotal role in endogenous lipids concentration which should be assessed until the real lipid profile revealed. Recently, lipid profile assessment includes both exogenous (TAG<sub>exo</sub>/diet) and endogenous lipid profile. Pieces of evidences focus on the TAG<sub>exo</sub> role in atherosclerosis pathogenesis. Taken together, today's, lipid profile consists of exogenous and endogenous lipid profile with each other. This article seeks the notion of the concept and value of endogenous and exogenous lipid profile to assess the real lipid profile.

**Keywords:** Fasting Lipid Profile, Non-Fasting Lipid Profile, Atherosclerosis

O93

### Measurement of Lipids and Lipoproteins

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Back ground: The measurement of lipid panel is pivotal in the management of the patient at the risk of cardiovascular disease (CVD). Methods: The measurement of blood lipid profile can be done both in Fasting and in non-Fasting. Cholesterol and Triglyceride assay is done through enzymatic and analysis of Lipoproteins can be done via Techniques such as ultracentrifuge, Precipitation Methods. The reference method for LDL<sub>c</sub> measurement and other Lipoproteins is ultra-centrifugation and chemical precipitation ( $\beta$  Quantification) but is not practical for the clinical Lab, because it is expensive. LP<sub>(a)</sub> are commonly measured by immunoassay. Conclusion: studies show that despite the small uptick in TG and remnants concentration at the postprandial the overall changes in lipids and lipoproteins is negligible. Therefore measuring of lipid profile can be done in fasting and non fasting. Except when TG concentration in non-fasting is above 4.5 mmol/L and that is when the measuring lipid profile in the fasting become necessary. The Fried Wald formula is utilized in calculated of LDL<sub>c</sub> indirectly. But it has certain limitation when the level of TG above 400 mg/dL or when the patient is suspected to have dyslipoproteinemia. The indirect method would not be reliable. Recently the direct method for measuring the LDL<sub>c</sub> and HDL<sub>c</sub> has been adopted by the labs. This method demonstrates satisfactory levels of accuracy and precisions when compared with reference method another biomarker is nonHDL<sub>c</sub> it is stronger predictor of CVD than calculated LDL<sub>c</sub> levels.

**Keywords:** Lipid, Lipoprotein, Risk Assessment



# Sepsis and Systemic Inflammation: Laboratory Verification of Diagnosis

O94

O94

### Disorders of Lipid Profile (Lipids and Lipoproteins) in Clinical Laboratory

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Lipid disorder” imply a range of conditions that can cause abnormal levels of lipids, or fats, in the blood. Dyslipidemias can be classified based on the primary biochemical disturbance, such as high or low plasma levels of low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, triglyceride, or some combination of these. Several monogenic dyslipidemias, such as heterozygous familial hypercholesterolemia, are now defined at the molecular genetic level. Cardiovascular disease and obesity are the major health treating problems for today world health. Despite advances in lipid and lipoproteins managements, cardiovascular risks still unsolved. While most guidelines try to achieving target levels of specific lipids and lipoproteins to reduce these risks, huge body of evidences has shown that molecular modification of these lipoproteins also has a critical impact on their atherogenicity. Post-translational modification by oxidation, carbamylation, glycation, and ditorbation of molecular components can reduce the capacity of high-density lipoprotein (HDL) for reverse cholesterol transport. Elevated levels of triglycerides, apolipoprotein C-III and lipoprotein(a), and a decreased level of apolipoprotein A-I are highly associated with atherosclerotic cardiovascular disease. Medical intervantions aimed to reducing TGs, lipoprotein(a), and apolipoprotein C-III, and enhancing apolipoprotein A-1, and some preliminary data have been shown their success. In this review, aimed to update the evidences in disorders which attributed to changes in lipid and lipoproteins metabolism.

**Keywords:** Dyslipidemia, Obesity, Lipid profile, coronary heart disease



# Posters



# Advances in QC Tools and Techniques in Medical Laboratory

**P1 - P2**

P1

**Designing of Diagnostic API System- Like for Identification of  
Enterobacteriaceae and Non- Fermentative Gram Negative Bacilli (NFB) and  
Comparison with API 20E Kit, Produced by France**

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**Background:** API systems demonstrate diagnostic methods by using a computer- program for identification of isolated bacteria. In this study we designed API system like for diagnosis of two groups of medical gram negative bacteria such as fermentative bacteria (Enterobacteriaceae) and non- fermentative bacilli (NFB). **Methods:** Several gram negative standard and clinical bacteria in mentioned groups were selected, cultured and identified by conventional, API 20E and API system-like methods separately. The API system- like was designed in miniature biochemical tests as prepared media in micro- titer plate with the reagents. The performed tests included fermentation of glucose and lactose, metabolism in MR and VP broth, utilization of citrate, decarboxylation of lysine and ornithine, pattern of growth in oxidation- fermentation media(OF) and ammonia production from urea. Also an Analytical Digital Computed Catalog (ADCC) profile and codes scoring were designed based on reactions of tested bacteria and cultured selective media as API system- like. After evaluating of the bacterial reactions of diagnostic tests, positive and negative reactions were computerized arranged based on codes scoring and ADCC gathering. So, the isolated bacteria were identified and reported. **Results:** All of results were similar in conventional, API 20E and designed API system-like methods. Similar the API kit, it is necessary that oxidase motility, indole and H<sub>2</sub>S production must be done alongside the API system- like. **Conclusion:** The designed API system-like is a rapid and trusted bacterial identification method in comparison of conventional model. Due to the economic sanctions of our country, it seems that designing of such media kit is an opportunistic and affordable situation for improving of the diagnostic methods in clinical laboratories.

**Keywords:** API System-Like, Diagnose

P2

### Prevalence PSM A Gene in Clinical Isolates of Biofilm- Forming Staphylococcus Aureus

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**Background and objectives:** Staphylococcus aureus is a common cause of hospital- and community-associated infections in the world. This organism causes an extensive range of diseases and biofilm formation is as an important mechanism for its virulence. One of the virulence factors of Staphylococcus aureus that are involved in biofilm formation are alpha-toxin and phenol-soluble modulins (PSMs). The aim of this study was to Prevalence PSM A gene among biofilm-forming Staphylococcus aureus isolated from clinical samples of Panje Azar Hospital, Gorgan, Golestan province, Iran. **Material and Methods:** The clinical samples were collected and examined for Staphylococcus aureus by microbiological and biochemical tests. Then, the Prevalence of PSM A gene was determined by PCR. **Results:** A total of 60 strains of Staphylococcus aureus were isolated from the clinical isolates. Of them, 47 strains (78.3%) were positive for biofilm formation. Based on the results, the PSM A gene were present in all the strains. The results of phenotypic and genotypic tests of biofilm were closely related to each other. and the Prevalence of PSM A gene was 80%. It was found that 100% of strains were biofilm producing and PSM A gene was present in 78.3% (47 strains) of them. **Conclusion:** This study showed high prevalence of biofilm formation in the clinical isolates of the Staphylococcus aureus and also presence of PSM A gene among all of the biofilm-forming Staphylococcus aureus. So, it was determined that PSM A gene play a role in biofilm formation and virulence in Staphylococcus aureus.

**Keywords:** Prevalence, PSM A Gene, Clinical Isolates, Staphylococcus Aureus, Biofilm



# Artificial Intelligence, Data Science and Laboratory Medicine

**P3**

P3

## In Silico Design of Suitable Signal Peptides for Secretory Production of Recombinant Glucarpidase

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**Background:** Methotrexate (MTX) is a general chemotherapeutic agent utilized to treat a variety of malignancies, woefully, its high doses can cause nephrotoxicity and subsequent defect in the process of MTX excretion. The recombinant form of glucarpidase is produced by engineered *E. coli* and is a confirmed choice to overcome this problem. **Objective:** In the present study, in silico analyses were performed to select suitable SPs for the secretion of recombinant glucarpidase in *E. coli*. **Methods:** The signal peptide website and UniProt database were employed to collect the SPs and protein sequences. In the next step, SignalP-5.0 helped us to predict the SPs and the position of cleavage sites. Moreover, physicochemical properties and solubility were evaluated using Prot-Param and Protein-sol online software, and finally, ProtCompB was used to predict the final subcellular localization. **Results:** Luckily, all SPs could form soluble fusion proteins. At last, it was found that PPB and TIBA could translocate the glucarpidase into the extracellular compartment. **Conclusion:** This study showed that there are only 2 applicable SPs for the extracellular translocation of glucarpidase. Although the findings were remarkable with high degrees of accuracy and precision based on the utilization of bioinformatics analyses, additional experimental assessments are required to confirm and validate it. Recent patents revealed several inventions related to the clinical aspects of vaccine peptides against human disorders.

**Keywords:** Methotrexate, Bioinformatics, Biopharmaceuticals, Glucarpidase, Malignancies, Recombinant Protein



# Autoimmune Disorders of Coagulation

P4

P4

### **The Important Role of microRNAs in Development, Severity and Complications of the Acquired Hemophilia A: Recent Updates**

**Sheyda Khalilian \***

**Background:** Hemophilia A is a common recessive X-linked bleeding disorder. The Acquired type of HA is a rare disorder of autoimmune nature which presents as sudden bleeding, without any trauma, anticoagulant usage or a history of bleeding. The majority of patients are inherited and caused by F8 gene mutations, but sometimes the genetic defect does not fully explain the severity of the disease. Thus, it increases the likelihood that other molecular pathways could lead to the development of HA or increase the severity of the disease. **Methods:** Pubmed, elsevier, and scopus databases were reviewed to introduce the most recent studies. **Results:** Recent evidences suggest that miRNAs may affect the severity of HA or might develop the disease in the absence of an F8 mutation. miRNAs dysregulation that target mRNAs encoding coagulation factors have been shown to disturb gene expression. Alterations in protein levels involved in the coagulation cascade mediated by miRNAs could cause bleeding disorders. **Conclusion:** Acquired HA is increasingly reported and due to identification of more cases, the need to introduce more miRNA analysis in the context of HA has increased. This review summarizes current evidences on the role of miRNAs in hemophilia A. Identification of miRNA targets, leads to understanding their functions and identifying their role in causing or intensifying the disease. Deep basic research may result in the development of tools for diagnosis, risk evaluation or novel therapeutic approaches.

**Keywords:** Bleeding Disorders, Coagulation Factors, Hemophilia A, microRNAs (miRNAs), Thrombosis

# **Biomarkers of Drug Abuse in Clinical Laboratory (Alcohol, Opioids and Psychotropic Substances)**

**P5 – P7**

P5

### **Evaluating Amphetamine and Methamphetamine Abuser Frequency in Hospitalized Patients in the Qazvin 22 Bahman Psychiatric Hospital**

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**Background:** Amphetamine (AMP) and methamphetamine (MET) are addictive synthetic substances that are in the class of stimulants. Most consumers turn to these substances because of the pleasant effects. Various studies have shown that the prevalence of these substances is increasing and also has a relationship with psychiatric disorders. Therefore, this study aimed to determine the prevalence of amphetamine and methamphetamine abuser in psychiatric patients. **Methods:** In this descriptive cross-sectional study, 2064 patients admitted to the 22 Bahman Medical Center in Qazvin in the year 1400 were studied in terms of amphetamine and methamphetamine abuse. Urine test was used chromatographically to detect amphetamine and methamphetamine. **Results:** The prevalence of amphetamine and methamphetamine abuser in 91 patients (36%) was evaluated. The majority of abusers were male (91% with a mean age of 32 years) and the significant relationship between age, gender and marital status was not found in the prevalence of substance use ( $P > 0.05$ ). The most important psychiatric illness among the abusers of these two substances was bipolar disorder (66%). In this study, no significant correlation was observed between the results of amphetamine and methamphetamine test results ( $P > 0.05$ ). **Conclusion:** The results of this study show that the prevalence of amphetamine and methamphetamine abuse in psychiatric patients is higher than other members of the community and patients with mood disorders are more at risk. This study also shows that the prevalence of substance abuser has increased significantly compared to previous studies.

**Keywords:** Amphetamine, Methamphetamine, Mood Disorders, Patients

P6

## Evaluation of Serum Levels of Liver Enzymes in Heroin Addicts in Qazvin 22 Bahman Hospital

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**Introduction:** Considering the harmful effects of drugs on some organs of the body, it seems necessary to study changes in liver tissue due to heroin use as the most widely used drug in Iran. The aim of this study was to investigate the possible effects of this drug on the levels of liver enzymes aspartate transaminase (AST), alanine transaminase (ALT) and alkaline phosphatase (ALP) in its addicts. **Methods:** The present study is a cross-sectional and case-control study on 92 heroin addicts who first referred to the center in the year 1400 to quit addiction as a case group and 90 healthy individuals as a control group. Enzyme activity levels were measured by calorimetry. The results were evaluated by t- test and SPSS-17 statistical software. **Results:** The results of this study showed that heroin use increases the level of ALT aminotransferase enzyme and there is a significant relationship between heroin use and ALT aminotransferase level in two groups of control and addict ( $P < 0.012$ ). There was also a significant relationship between the duration of heroin use and ALT ( $P < 0.05$ ). **Conclusion:** Due to the changes that heroin causes on liver tissue, it is necessary to introduce this point as another risk of opioid use.

**Keywords:** Heroin, Aspartate Transaminase, Alanine Transaminase, Alkaline Phosphatase

P7

## Frequency of Toxoplasmosis in Addicts Patients in Andimeshk City of Khuzestan Province, 2021

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**Background:** Addiction is known as a mental health problem in societies. Genetic and environmental factors, including infectious agents, are involved in its development. Toxoplasma, with increasing dopamine levels, has a potential role in the development of schizophrenia. **Methods:** The purpose of this study was to determine the prevalence of IgG antibodies against Toxoplasma in addicts patients in Andimeshk county. 350 patient was selected from ten region of Andimeshk county southwest of IRAN. The serum of these patients was assessed by ELISA method. **Results:** The percentage of infections in addicts patients and controls were 38.5 and 22.8, respectively. There was no significant difference between the three groups using chi square analyzing ( $P>0.05$ ). **Conclusion:** However, the percentage of infection in addicts patients is higher than controls groups. Further comprehensive studies should be conducted with more samples to get better results statistically.

**Keywords:** Toxoplasma Gondii, Andimeshk, Addicts Patients



# Challenges in Laboratory Diagnosis of STD

**P8 - P9**

P8

### Challenges of Laboratory Diagnosis of Trichomonas Vaginalis

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**Background:** one of the most momentous aspects of community health is the study of community hygiene. In this regard, one of the most common causes of sexually transmitted diseases is Trichomoniasis. **Methods :** In this study, an attempt has been made to review the diagnostic challenges of Trichomonas vaginalis in laboratories as the main diagnostic element of prevalent infections. **Results :** Based on the fact that sexually transmitted diseases have analogous symptoms with each other, so the only way to diagnose the infection is laboratory methods, which are done on the cervical and posterior vaginal fluid samples of women or prostate secretions in men. **Conclusion** Challenges of laboratory diagnosis include: patient's preparation, contains not using vaginal creams and lotions, correct sampling method, sampling in two separate tubes and isolated from formalin samples, which are mostly for pathology and Pap smear tests, in most laboratories. Clinical studies conducted by researchers in the field are performed by direct expansion test. Since the mentioned protozoan exists only in the form of trophozoites, the movement of trophozoites should be observed microscopically as soon as possible. The sensitivity of this method is about 60% and in this regard, it is highly recommended to use 0.5% glucose in normal saline to observe the movement of the parasite easily and more accurately. The culture method is more sensitive, but it is time consuming and quite costly, although the best molecular method is PCR. Unfortunately, it is not a routine and practical test, so it requires the importance of more attention and attention of esteemed laboratory colleagues in order to accurately and correctly diagnoses the infection. Therefore, it demands the significance of more attention of laboratory experts in order to accurately and correctly diagnose this disease.

**Keywords:** Laboratory Diagnosis, Trichomonas Vaginalis, Trichomoniasis, Trichomonas Vaginitis

P9

## Evaluation of Different Types of Human Papilloma Virus by PCR Method and Determining its Frequency in Pap Smear Samples Referred to Medical Centers in Isfahan and the Association of Cervical Cancer

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**Introduction:** Cervical cancer is the fourth common cancer in the worldwide in developed countries, the incidence of cervical cancer has dropped significantly due to Pap smears and screening programs. Basically, all cervical cancers are caused by HPV infection. This study was performed to determine the prevalence of different types of HPV in women of different ages with normal and abnormal Pap smears. **Materials and Methods:** This descriptive-analytical study was performed on 287 women were referred to specialized gynecological clinics and medical centers in Isfahan in 1400. At first, Pap smear samples of the patients were tested and the results were recorded. Then HPV samples were identified by PCR. Positive samples were tested again to determine genotypes. **Results:** Out of 278 cases, 21 (7.32%) were HPV positive and 266 (92.68%) were negative. In the next stage genotyping was done for, 21 positive cases. 9 cases (42.85%) were type 16, 6 cases (28.57%) were type 18, 3 cases (14.28%) were type 6, 2 cases (9.5%) were identified as type 82 and one case (4.47) was type 11, **Conclusion:** Based on the results of this study, it can be concluded that HPV virus is moderately prevalent in Isfahan, and most of them were high-risk type furthermore, preventive measures for women over 20 years and Married individuals seems necessary.

**Keywords:** HPV, Cervical Cancer, PCR



# Chemical-Microscopic Testing of Biological Liquids

**P10 – P19**

P10

## Long-Term MgSO<sub>4</sub> Treatment Effects on Insulin Resistance and Irisin Levels in Type 2 Diabetic Rats

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**Background:** Magnesium is the second abundant intracellular cation that is involved in the regulation of carbohydrate metabolism. Magnesium deficiency has been associated with the development of type 2 diabetes mellitus (T2DM) and its complications. Irisin improves glucose uptake and hepatic glucose and lipid metabolisms. In the present study, we aimed to evaluate the effects of long-term treatment of MgSO<sub>4</sub> and insulin on insulin resistance, dyslipidemia, serum, and hepatic irisin levels in T2DM rats. **Methods:** Experimental rats were divided into four groups: Control group, diabetic group induced by 3 months high-fat diet+streptozotocin, insulin-treated diabetic group, MgSO<sub>4</sub> treated diabetic group. At the end of the therapies, serum concentrations of FBG, TG, insulin, Ox-LDL, along with serum and hepatic irisin levels were measured. **Results:** MgSO<sub>4</sub> therapy resulted in decreased FBG, TG, Ox-LDL, improved serum insulin and irisin levels compared to the diabetic group. Insulin therapy significantly decreased FBG, Ox-LDL, and serum irisin levels compared with the control group. **Conclusion:** In conclusion, long-term MgSO<sub>4</sub> therapy possibly improves insulin resistance and hyperlipidemia partly through decreasing Ox-LDL and TG and increasing serum irisin levels in T2DM rats. We suggested that MgSO<sub>4</sub> as a suitable adjuvant along with pharmaceutical drugs for better metabolic control in T2DM patients.

**Keywords:** Type 2 Diabetes Mellitus, Irisin, Insulin Resistance, Dyslipidemia

P11

## Comparison of Intracellular and Serum Levels of Zinc, Copper and Magnesium in Men with Type 2 Diabetes with Metabolic Syndrome

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**Background:** Many studies provide evidence of an association among trace elements hemostasis, human body metabolic pathways, antioxidant status and the activity of other enzymes involved in type two diabetes mellitus (T2DM) and the metabolic syndrome (MetS). The present study was designed to investigate the mentioned parameters in patients with T2DM and MetS. **Methods:** Forty men including T2DM and MetS and forty apparently healthy men as the control group participated in this cross-sectional study. After providing a written informed consent, subject's age, anthropometric parameters and blood pressure were measured and recorded. Blood biochemical assays were carried out to determine the parameters related to MetS and T2DM. Copper, magnesium and zinc were analyzed in the plasma and erythrocytes and antioxidant status were determined as well. **Results:** A significant decrease was observed in copper, magnesium and zinc levels in plasma and copper levels in erythrocytes in the patients group compared to the controls. Malondialdehyde was significantly increased in the patients group; but with respect to zinc and magnesium levels in erythrocytes, total antioxidant capacity and the activity of superoxide dismutase enzyme were founded no significant differences between groups. **Conclusion:** Based on the results, notable changes occur in the activity of some enzymes and the levels of some elements which were investigated in the present study among the patients with T2DM and MetS, compared to apparently healthy controls. These alterations may cause a number of consequences on activity of the enzymes involved in glucose metabolism and the progress of diabetes and its long term complications.

**Keywords:** Antioxidant status, Copper, Magnesium, Type 2 Diabetes Mellitus, Zinc

P12

## Long Term GABA Administration Improves Serum Irisin Levels in Chronic Type 2 Diabetic Rats

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**Background:**  $\gamma$ -aminobutyric acid (GABA) is an inhibitory neurotransmitter released from pancreatic alpha cells, stimulates insulin secretion from pancreatic beta cells. Studies showed that GABA has an anti-diabetic effect. In this study, we aimed to investigate whether GABA and insulin regulate irisin production and insulin resistance in type 2 diabetic Mellitus (T2DM) rats. **Methods:** Four groups rats (n=6) were used in this study including: control, T2DM, T2DM+insulin, and T2DM+GABA groups. After T2DM induction for 3 months (high-fat diet+35 mg/kg streptozotocin) and treatment with GABA or insulin, circulating levels of FBG, TG, LDL, Ox-LDL and insulin as well as hepatic and serum irisin levels were measured. **Results:** GABA therapy improved FBG, insulin, LDL, Ox-LDL levels in diabetic rats. Insulin treatment significantly reduced FBG and failed to maintain glucose close to the control level. Insulin therapy significantly decreased the levels of LDL, Ox-LDL, and circulating irisin levels. GABA therapy did not show significant changes in circulating irisin levels compared with the control group. Hepatic irisin levels increased non significantly in the diabetic group. Insulin group show a significant reduction in hepatic irisin levels but GABA did not show a significant change. **Conclusion:** Our findings suggest that the anti-diabetic effects of GABA may be mediated by decrease in Ox-LDL levels and increase in circulating irisin levels in T2DM rats.

**Keywords:** Type 2 Diabetes Mellitus, GABA, Irisin, Insulin Resistance

P13

### Macroscopical and Chemical Analysis for Determination of Physicochemical Properties of Renal Calculus in Urological Diseases

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**Background:** Nephrolithiasis are a prevalent urological disease, which might seriously have an effect on health and quality of life in populations worldwide. Due to the heterogeneity of nephrolithiasis, precise information on their elemental compound is critical for the selection of treatment. Therefore it's necessary to analyze the composition of renal stones in numerous nations to prevent the formation of stones or stop their enlargement. This study was conducted to determine the chemical constituents of renal calculi and aimed to find a relative abundance of urinary calculi by considering their composition. **Subjects and Methods:** In this study which was conducted at Ahvaz Jundishapur University of Medical Sciences, renal calculi from 275 patients were referred to medical laboratories, and after macroscopical characterization, their chemical components were analyzed by using the standard methods. **Results:** Findings show that twenty percent of renal calculi from total patients had a pure chemical component of calcium oxalate and eighty percent of kidney stones had a mixed type. Among the mixed stones studied in all patients, calcium oxalate compounds with 83.6%, calcium phosphate with 43.6%, ammonium urate with 36.8%, and magnesium ammonium phosphate with 30.9% were the most common, and uric acid compounds with 10%, and cysteine with 2.3% had the lowest frequency. Calcium oxalate stones are the most abundant chemical composition of either pure or mixed urinary calculi, which are more common in men, and Struvite stones contain magnesium, ammonium, and phosphate are more common in women. **Conclusion:** Our study indicated that the physicochemical characteristics of kidney stones within the population of the community could be affected by various factors such as age, gender, geographical area, and so on. Knowledge of the chemical composition of a stone is useful in the prevention, treatment, and diagnosis and allows physicians to act specifically in each of these areas.

**Keywords:** Urological Disease, Nephrolithiasis, Analysis of Renal Calculi

P14

## High Prevalence and Worrying of Vitamin D Deficiency in the Community: Curiosity about the Possible Reasons

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**Background:** Vitamin D made in the skin or ingested in the diet is biologically inert and requires two successive hydroxylations in the liver on carbon 25 to form 25-hydroxyvitamin D [25(OH) Vit D], and then in the kidney on carbon 1 to form the biologically active form of vitamin D, 1,25-dihydroxyvitamin D. Throughout the world, measuring serum 25-hydroxyvitamin D is used to assess vitamin D levels. According to this criterion, many epidemiological studies show that vitamin D deficiency is a worldwide health problem and is associated with many chronic diseases. The aim of this study was to evaluate of serum vitamin D levels in Iranian population and remark on the need for its treating. **Methods:** This study was conducted at Ahvaz Jundishapur University of Medical Sciences. This cross-sectional study was carried out on 4574 participants of men and women referred to some diagnostic laboratories. Serum level of 25(OH) vitamin D, as the most important metabolites of vitamin D, was measured by chemiluminescence method and vitamin D deficiency was defined at 25-hydroxyvitamin D level of less than 30 ng / ml. **Results:** Based on the statistical analysis the mean serum level of 25(OH) Vit D was  $21.40 \pm 18.36$  ng/mL 77.8% of participants were assessed as the severe to mild vitamin D deficiency. The vitamin D average for male and female were  $21.45 \pm 16.03$  and  $21.38 \pm 19.00$  ng / ml respectively and it was in insufficiency range for both. **Conclusion:** In this study, it was found that despite the sunny climate of these areas, the serum level of vitamin D is low in study participants and the main reasons for this deficiency can be due to very low intake of vitamin D through foods, fear of exposure to the sun ray, or incorrect assessments of the normal values of vitamin D.

**Keywords:** Vitamin D Deficiency, Serum 25(OH) Vitamin D, Chemiluminescence Method

P15

### Prevalence of A2B Subgroups and Anti-A1 Antibody in Blood Donors in Bushehr, Iran

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**Introduction:** A2B is rare phenotypes of the ABO blood group system. Some individuals with A2B blood group may have anti-A1 antibodies that may be clinically significant or insignificant. The aim of this study was to determine the frequency of A2B phenotypes and anti-A1 antibodies in blood donors in Bushehr city. **Materials and Methods.** Blood samples collected from 17000 blood donors were typed for ABO (cell and serum grouping). Individuals with blood group AB were further subtyped by testing with anti-A1 lectin. In addition to the serum grouping using A2B individuals were screened for the presence of anti-A1 in their sera against A1 red cells at 4°C, 22°C and 37°C to determine the thermal amplitude of the reacting anti-A1 antibody. **Results.** A2B subgroup was found in 0.029% (n=5) cases. No one individual with A2B blood type showed cold reactive anti-A1 antibody. **Conclusion.** A2B is one of the rarest among ABO phenotypes in the studied population. Although rare, anti-A1 antibody is not so uncommon. Care shall be taken during routine ABO grouping especially in cases of mix-field or weak positive reactions in A and AB phenotypes.

**Keywords:** Blood Group, Anti-A1, Blood Donor, Transfusion

P16

## Gating and Immunophenotyping Analysis Scheme for Characterization of Diffuse Large B Cell Lymphoma in Peritoneal Effusion

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**Introduction:** Diffuse large B-cell lymphoma (DLBCL) is an aggressive form of lymphoma which aside from lymph nodes, it can also appear in various extranodal areas. Primary effusion lymphoma (PEL) is an infrequent kind of DLBCL which primarily involves cavities and causes serous effusions without any obvious masses or lymphadenopathy. An accurate and careful immunophenotyping strategy could be helpful in diagnosis of DLBCL in effusions. **Methods and Results (case presentation):** Herein, we describe a case of 74-year-old male presented with anorexia, abdominal pain, excess sweating and massive ascites. Cytological studies revealed a predominant abnormal large lymphoid cells with vesicular nuclei, variable degrees of nucleolar prominence, cellular pleomorphism and nuclear irregularity. Flow cytometry analysis illustrated that almost all the cells express common leukocyte marker (CD45). Gating on the higher SSC population, immunophenotyping showed a small T cell population (about 39% of the cells analyzed) with no aberrant loss or expression of T cell markers and a predominant large B cell population (about 43% of the cells) that was negative for CD5, CD10, and CD23. These B cells were positive for CD20, CD19, and FMC-7. The size of the malignant cells was large based on forward-scatter signal. Small lymphocytes showed immunophenotypic profile consistent with normal T cells. Immunophenotyping and morphological findings were consistent with diffuse large B cell lymphoma. **Conclusion:** This case clarifies the aptitude of flow cytometry and the necessity of careful gating strategy in detection of neoplastic B-lymphocytes in a reactive ascites containing a heterogeneous mixture of cells.

**Keywords:** Diffuse Large B-Cell Lymphoma, Ascites, Immunophenotyping

P17

### Diversity and prevalence of Hemoglobinopathies in the Iranian Population

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**Background:** Hemoglobinopathies are the most common monogenic disorders with remarkable frequency in certain regions of the world, including Iran. At least 5.2% of world population is carrier for a main hemoglobin (Hb) disorder. More than 900 Hemoglobin variants were reported due to globin gene mutations that cause structural changes in globin gene products which ultimately affect the performance and stability of hemoglobin. The main aim of premarriage screening is to reduce the number of affected births and, in the case of sickle cell, reduce childhood morbidity and mortality. The aim of this study was to evaluate the prevalence of hemoglobinopathies in premarriage men referred to the laboratories of Health Centers. **Methods:** Hemoglobin electrophoresis was performed by Sebia capillary electrophoresis and types of hemoglobinopathies were determined. Hematological parameters in blood samples containing EDTA were measured by Sysmex KX-21 hematology analyzer. **Results:** One hundred and eleven of cases (1.25%) from total 8806 cases were has abnormal bands of hemoglobin. From this total, 3 cases (0.03%) were with heterozygous hemoglobin E and 1 case (0.01%) was heterozygous hemoglobin C. Also, 74 cases (0.84%) had hemoglobin D, and 28 patients (0.32%) containing hemoglobin S were with different genotype status; homozygous or heterozygous types. In this study, uncommon Hemoglobin variants (with the prevalence of 0.05%) such as hemoglobin O-Arab and constant spring was observed. HbD with 0.84% and HbC with 0.01% of total have highest and minimum frequency. **Conclusion:** Due to the high prevalence of hemoglobin S among the hemoglobinopathies reported in this study and its association with other types of hemoglobinopathies such as Hb O-Arab, Hb D, and Hb C in the population, there is a possibility of sickle cell disease and increased childhood morbidity or mortality in the Iranian population if not properly screened.

**Keywords:** Hemoglobinopathies, Screening Protocol, Hematological Parameters, Capillary Electrophoresis

P18

## In Vitro Reprotoxicity of Carboxyl-Functionalised Single- and Multi-Walled Carbon Nanotubes on Human Spermatozoa

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Reproductive toxicity of carboxyl-functionalised carbon nanotubes (CNT-COOH), as the most commonly used form of water-soluble CNTs, is not clearly studied. The aim of this study was to investigate in vitro toxicity of carboxylated single-walled and multi-walled CNTs (SWCNT-COOH and MWCNT-COOH) against human spermatozoa. Sperm cells from healthy donors were incubated with 0.1–100 µg/ml of SWCNT-COOH or MWCNT-COOH at 37°C for up to 5 hr. Viability of sperm cells was assessed using MTT test, and sperm motility was evaluated following World Health Organization guideline. Production of reactive oxygen species (ROS) and nitric oxide (NO) in sperm was also assessed. We showed that both MWCNTCOOH and SWCNT-COOH following incubation in vitro with human spermatozoa did not exert negative effect on viability while motility was significantly ( $p < .05$ ) dropped in a dose-dependent manner. Moreover, there was no significant effect of the type, dose and exposure time of the CNT-COOH on NO production. Exposure of sperm cells to both examined types of CNTs at concentrations as low as 0.1 µg/ ml caused a significant increase in ROS levels. In conclusion, carboxylated forms of CNTs seem to be harmful for human spermatozoa. Further studies, especially using in vivo models, are needed to decide about reprotoxicity of carboxylated forms of CNTs.

**Keywords:** Carboxylated Carbon Nanotubes, Human Spermatozoa, Motility, Oxidative Stress, Viability

P19

### Comparison of Changes in Vitamin D and Calcium in Breast Cancer Patients and Normal Persons

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**Introduction:** Breast cancer is one of the most common cancers and the second leading cause of cancer death in women. The protective effects of vitamin D on cancer have been shown and hypercalcemia has been reported in some studies. However, the results are still contradictory. The aim of this study was to compare changes in vitamin D and calcium in breast cancer patients and normal persons. **Method:** Blood samples were obtained from 35 women with breast cancer from the tumor bank of the Cancer Institute. 35 healthy women were selected as the control group. Vitamin D and calcium levels in blood samples of patients and controls were measured. Data were analyzed by statistical tests in SPSS software. A significance level of 0.05 was considered. **Results:** The results of estrogen and progesterone receptor expression tests showed that the receptors for these two hormones were expressed almost equally in breast cancer patients. There was no statistically significant difference in serum vitamin D levels between breast cancer patients and the control group. There was also no association between serum calcium levels and breast cancer. Serum vitamin D levels were significantly higher in patients with a positive PR receptor than in patients with a negative PR receptor ( $P < 0.05$ ). **Discussion and Conclusion:** Unlike previous studies, the data of this study showed that the level of vitamin D and serum calcium was not associated with breast cancer, PR receptor expression could be associated with higher levels of vitamin D.

**Keywords:** Breast Cancer, Vitamin D, Calcium



# Clinical and Fundamental Studies of Tumor Markers

**P20 – P32**

P20

### Development of a Microenvironment Prognostic Model for Acute Myeloid Leukemia

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**Background:** Acute myeloid leukemia (AML) is a rapidly progressing disease with a high mortality rate. Its initiation, progression, drug resistance, and recurrence are all dependent on abnormal molecular and genetic changes within the cell, as well as the Tumor microenvironment (TME) protection. More and more researchers are realizing that current immune therapies and targeted therapies cannot fully address AML's extremely complex heterogeneity, and they are dedicated to discovering new prognostic biomarkers to promote the advancement of AML precision diagnosis and treatment. Despite several different AML prognostic scoring models that have been proposed to assess patient prognosis, accuracy still needs to be improved. Therefore, this study aims to establish a novel microenvironment-related prognostic model for predicting AML prognosis. **Methods:** In this study, the expression profile of AML patients from the Cancer Genome Atlas (TCGA) database was normalized after removing the batch effect. TME cell components were explored through the Estimate algorithm and then hierarchically clustered to establish TME classification. Subsequently, a prognostic model was established by Lasso-Cox. to validate the prognostic performance of the model Multiple Gene Expression Omnibus (GEO) databases and the TCGA dataset were employed. Finally, the prognostic efficacy was evaluated using receiver operating characteristics (ROC). **Results:** We established a TME classification system that divided all patients into three groups with distinct prognostic characteristics. **Conclusion:** Overall, this study provided a new prognostic model to predict the overall survival (OS) of AML patients, which may help in clinical decision-making for AML treatment.

**Keywords:** Acute Myeloid Leukemia, Tumor Microenvironment, Prognostic Model

P21

## Co-administration of miR-193a and 5-FU could modulate gastric cancer cells' migration through targeting MMP16 in-vitro

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The role of microRNAs in modifying gene expression has been shown in several types of cancer. 5-Fluorouracil (5-FU) is a common chemotherapeutic agent used to treat cancer. The present study aimed to evaluate the expression levels of miR-193a and MMP16 in tissues obtained from patients with gastric cancer (GC). GC tissues and adjacent non-cancerous tissue were obtained from 65 patients who had undergone surgery, 15 of which had received 5-FU. Then, qPCR was performed to determine the expression level of miR-193a and MMP16 in GC. In the next step, qRT-PCR and Western blotting were performed to assess the effect of miR-193a and 5-FU on the MMP16 expression. Also, MTT and wound healing assays were performed to determine their role in cell viability and migration. Besides, the relationship between miR-193a and MMP16 with patients' clinical features was investigated. The present study results showed that miR-193a was significantly downregulated, while MMP16 was up-regulated in tumor tissues obtained from patients with GC compared with the adjacent non-tumor healthy controls. In addition, the replacement of miR-193 modified the MMP16, particularly in combination with 5-FU. Besides, the present study showed that miR-193a replacement could suppress MMP16-induced GC cell migration by directly targeting MMP16. Furthermore, there was a significant association between miR-193a and MMP16 with specific clinicopathological characteristics, particularly metastasis-related features. These results suggest that miR-193a has a tumor-suppressive function in GC by alleviating MMP16 in combination with chemotherapeutic drugs.

**Keywords:** HCC, miR-193, MMP16, 5-FU

P22

### Decreased Expression of SHANK3 Is a Potential Biomarker of Human Colon Adenocarcinoma

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**Background:** Human colon adenocarcinoma (COAD) is one of the major causes of tumor-induced death worldwide. A complicated gene network significantly regulates its progression and prognosis. Here, we aimed to investigate the relationship between SH3 and multiple ankyrin repeat domains 3 (SHANK3), a synaptic scaffolding protein, and COAD for the first time. **Methods:** The bioinformatics tools, gene expression profiling interactive analysis 2 (GEPIA2), and DNA Methylation Interactive Visualization Database (DNMIVD) were used to analyze differential expression, prognostic value, and DNA methylation status in COAD patients. Additionally, the functional enrichment analysis was performed for SHANK3 co-expressed genes in COAD using the Enrichr database. **Results:** We found that the mRNA expression of the SHANK3 was down-regulated in COAD. This low expression was not correlated with tumor stage. The results also demonstrated the presence of association between SHANK3 expression and the overall survival of COAD patients. The methylation status of the SHANK3 promoter was significantly increased when compared with normal tissues. The functional enrichment analysis revealed the top five related biological pathways (PI3K-Akt signaling pathway, ECM-receptor interaction, Focal adhesion, MAPK and RAS signaling pathways), and cellular component (G protein-coupled receptor dimeric complex, sorting endosome, integral component of cytoplasmic side of endoplasmic reticulum membrane, connexin complex, integral component of plasma membrane) in GO enrichment analysis. **Conclusion:** The present study indicates that SHANK3 could serve as therapeutic target and prognostic biomarker for COAD. However, further studies are required to confirm these results.

**Keywords:** Colon Adenocarcinoma, Bioinformatics, Prognosis, Methylation

P23

## Thymoquinone Inhibited Vasculogenic Capacity in Human Breast Cancer Stem Cells

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**Background:** Vasculogenic mimicry (VM) is characterized by the formation of tubular structure inside the tumor stroma. It has been shown that a small fraction of cancer cells, namely cancer stem cells (CSCs), could stimulate the development of vascular units in the tumor niche, leading to enhanced metastasis to the remote sites. This study aimed to study the inhibitory effect of phytochemical, Thymoquinone (TQ), on human breast MDA-MB-231 cell line. **Methods:** MDA-MB-231 CSCs were incubated with different concentrations of TQ for 48 h. The viability of CSCs was determined using the MTT assay. By using the Matrigel assay, we measured the tubulogenesis capacity. The percent of CD24<sup>-</sup> CSCs and Rhodamine 123 efflux capacity was studied using flow cytometry analysis. Protein levels endothelialcadherin matrix metalloproteinases-2 and -9 (MMP-2 and -9) were detected by western blotting. **Results:** TQ decreased the viability of CSCs in a dose-dependent manner. The vasculogenic capacity of CSCs was reduced after being-exposed to TQ ( $p < 0.05$ ). Western blotting revealed the decrease of CSCs metastasis by suppressing MMP-2 and -9. TQ had the potential to decrease the number of CD24<sup>-</sup> CSCs and Rhodamine 123 efflux capacity after 48 h. **Conclusion:** TQ could alter the vasculogenic capacity of human breast CSCs. Thus, TQ together with anti-angiogenic therapies may be a novel therapeutic agent in the suppression of VM in breast cancer.

**Keywords:** Thymoquinone, Vasculogenic Mimicry, Breast Cancer Stem Cells

P24

## Thymoquinone Inhibits the Epithelial-Mesenchymal Transition (EMT) of Human Breast Cancer Stem Cells by Down-Regulating VE-Cadherin

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**Background:** Thymoquinone (TQ), an active ingredient of *Nigella sativa*, has been reported to exhibit anti-tumor activities through mechanism(s) that is not fully understood. CSCs are a small population of cells inside a tumor matrix with distinct multipotentiality and self-renewal properties. CSCs could promote the metastasis of remote site via an engaging mechanism entitled epithelial-mesenchymal transition (EMT) process. The present study aimed to evaluate the inhibitory effect of thymoquinone on the epithelial-mesenchymal transition of breast CSC by focusing on the PI3K/Akt and Wnt/ $\beta$ -catenin signaling pathways. **Methods:** Cell survival was assessed by using the MTT assay. The combination of TQ and PI3K and Wnt3a inhibitors was examined in CSCs. We also monitored the stimulatory effects of VEGF, FGF, and EGF in MDA-MB-231 cells pre-treated with TQ using MTT assay. The levels of Akt, p-Akt, Wnt3a, VE-cadherin were measured using western blotting. **Results:** Our results showed that thymoquinone inhibited the viability of CSCs compared to that of control. The combination of TQ with PI3K and Wnt3a inhibitors reduced significantly the survival rate compared to the control group ( $p < 0.05$ ). TQ could blunt the stimulatory effect of VEGF, EGF, FGF on CSCs ( $p < 0.05$ ). In addition, the protein level of VE-cadherin was diminished in TQ-treated CSCs as compared to the control cell ( $p < 0.05$ ). TQ could suppress Wnt3a and PI3K, which coincided with the reduction of the p-Akt/Akt ratio. **Conclusion:** Taken together, our data indicated that TQ treatment can be a promising therapeutic strategy for human malignant breast cancer.

**Keywords:** Thymoquinone, Breast Cancer Stem Cells, Wnt3a/PI3K Signaling Pathways, VE-Cadherin

P25

## The Effect of Silver Nanoparticles Synthesised Using Sargassum on Expression MiR-25 and MiR-143 on Acute Lymphoblastic Leukemic Cells in Jurkat Cell Line

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**Background** Acute lymphoblastic leukemia (ALL) is the most common malignancy among children. One of the side effects of chemotherapy drugs is that in addition to cancer cells, other normal cells are destroyed, and which may follow infection and bleeding occur. The biogenic synthesis of nanoparticles using seaweed is widely used due to its easy availability and effectiveness. The aim of this study was to investigate the effect of Sargassum brown seaweed extract along with silver nanoparticles on the expression of miR-25 and 143 in Jurkat cell line. **Methods** In an interventional study after culturing the Jurkat and obtaining cells with 95% survival, we adjacent concentrations of the drug with maximum cell growth inhibition and IC50 to 20,000 cell after 48 hours of treatment, the RNA was extracted and miR-25 and 143 expression was evaluated by Real Time PCR. Statistical analysis was performed with SPSS software and  $P < 0.05$  was considered as a significant difference. **Result** The expression of miR-25 in the group of treated with maximum dose and IC50 of silver nanoparticles and algae extract was not significantly different from the control, while that for miR-143 both had a remarkable decline. ( $P < 0.0001$ ) **Conclusion** The expression of miR-25 in treated jurkat cells was more decreased than the expression of miR-143 in normal lymphocytes. Due to the oncogenicity of miR-25 in cell line jurkat, it can be said that reducing this microRNA can help kill cancer cells.

**Keywords:** Mir-25, Mir-143, Acute Lymphoblastic Leukemia (ALL), Nano Particles.

P26

## The Investigation of Acetyl-CoA Acetyl Transferase 1 Expression in Oral Squamous Cell Carcinoma

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**Background:** Acetyl CoA acetyltransferase 1 (ACAT1) possesses acetyltransferase activity beyond its thiolase function in ketogenic and ketolysis. Recent reports have demonstrated altered ACAT1 expression in diverse human cancers. However, its expression in OSCC remains elusive. **Material and Methods:** In this study, using specific antibody against ACAT1, western blotting was performed on protein extracted from tumour and adjacent pre-tumour fresh tissues of 5 OSCC patients. **Results:** Our findings indicated the differential expression level of ACAT1 in tumour of OSCC patients suggesting that OSCC tumours are genetically heterogenous. Next, we compared the expression of ACAT1 in tumours and their adjacent pre-tumour tissue. Of 5 OSCC patients, 3 of them showed low level of ACAT1 expression in their tumours compared to their adjacent pre-tumour tissue, in opposite 2 of 5 OSCC patients showed high expression of ACAT1 in tumour in comparison to their adjacent pre-tumour tissue. **Conclusion:** Comparison of ACAT1 expression, one of the key enzymes in the ketone body metabolism pathway, revealed differential expression level of ACAT1 in tumoural and adjacent pre-tumoural tissue. Considering the differential expression level of ACAT1 in OSCC patients, it is possible that OSCC patients with observed low ACAT1 expression in their tumour compared to the pre-tumour tissue respond better to ketogenic diet therapy, although further investigations of other enzymes involved in ketolysis including BDH1, BDH2 and OXCT1 would be needed.

**Keywords:** ACAT1, OSCC, Metabolism

P27

## Evaluation of Bacterial Population (*Streptococcus Gallolyticus*, *Fusobacterium Nucleatum*, *Enterococcus Faecalis*, *Bacteroides Fragilis*) in Paraffin Biopsy Samples of Colorectal Cancer Patients

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**Background:** Colon cancer (CRC) is one of the most common cancers in the world, the main cause of which is epigenetic and environmental factors. Recent studies there is some evidence to associate individual bacteria, with initiation or progression human colorectal cancer (CRC). The aim of this study was to evaluation of bacterial population (*Streptococcus gallolyticus*, *Fusobacterium nucleatum*, *Enterococcus faecalis*, *Bacteroides fragilis*) in biopsy samples of colorectal cancer patients. **Methods:** In this study, we used of 80 paraffin biopsy sample, including 40 sample of patients that suffering from colorectal cancer (CRC) and 40 sample of healthy individuals to extract DNA directly. Real time PCR was performed on the extracted DNA samples to evaluate the frequency of the studied bacteria. **Results:** The prevalence of bacterioids *Fragillis* in cancerous and non-cancerous individuals is 45% and 15%, respectively; The prevalence of *Enterococcus faecalis* in cancer and non-cancer patients was 85% and 82%, respectively. The relative prevalence of *Enterococcus faecalis* and *Bacteroides fragilis* in both cancer and non-cancer groups showed a statistically significant difference ( $P < 0.05$ ). While *Streptococcus gallolithicus* and *Fusobacterium nucleotum* did not show significant differences in both cancer and non-cancer groups ( $P > 0.05$ ). **Conclusions:** Based on the results of the present study, the relative abundance of *Enterococcus faecalis* and *Bacteroides fragillis* in both cancer and non-cancer groups was statistically significant ( $P < 0.05$ ). Also, according to our study, the frequency of the studied bacteria in the late stages of cancer (third and fourth stages) is higher than in the early stages of cancer (first and second stages).

**Keywords:** Colorectal Cancer, *Streptococcus Gallolyticus*, *Bacteroides Fragilis*, *Fusobacterium Nucleatum*, *Enterococcus Faecalis*, Biopsy Samples

P28

## Clinical Application of Nanoparticles in Diagnosis and Treatment of Cancer

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**Background:** Fluorescent nanoparticles known as quantum dots (QDs) can be used for localisation of cancer cells due to their nano size and ability to penetrate individual cancer cells and high resolution imaging. Carbon nanotubes (CNTs) are of interest to the medical community due to their unique properties in which they converting optical energy into thermal energy. Many research laboratories, are investigating the conjugation of QDs to CNTs to allow both localisation of the cancer cells in the patient, by imaging with QDs, and subsequent cell killing, via drug release from the CNT. **Method:** SWCNTs were oxidised and reacted with OctaAmmonium POSS to render them more biocompatible and water dispersible. The functionalised SWCNTs were conjugated with QDs for cancer cell imaging. The composite were characterised by Fourier transform infrared spectroscopy (FTIR), Confocal microscopy, UV-VIS spectrometer, Transmission electron microscopy (TEM). The HT29 (colon) and MCF7 (breast) cancer cell line were used for cancer cell imaging. **Results:** Post functionalization process of SWCNT with OctaAmmonium-POSS, TEM images showed a layer of dots had formed on the surface of the SWCNTs. In the FTIR and UV-Vis spectrometer experiments, result illustrated the presence of the amide bond following the conjugation of SWCNT to QDs in compression to SWCNT and QDs only. Confocal microscopy picture determined the presence of fluorescence in both QDs and QDs conjugated SWCNT. The images obtained by confocal microscopy from HT29 and MCF7 cancer cell line determined the location of SWCNT in different parts of cancer cell at various time intervals. **Conclusion:** Treating pure SWCNTs with OctaAmmonium-POSS is an effective method for functionalization of SWCNTs. Attachment of QDs to SWCNT can be used to track the movement of SWCNT in in vivo studies.

**Keywords:** Clinical Application, Nanoparticles, Diagnosis, Treatment, Cancer

P29

## The mRNA Expression of HDAC9 in Iranian Patients of Oral Squamous Cell Carcinoma

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**Backgrounds:** HDAC9 (histone deacetylase 9) is a member of the class IIa family of histone deacetylases. It can circulate freely between the nucleus and cytoplasm and promotes tissue-specific transcriptional regulation through interaction with histone and nonhistone substrates. HDAC9 plays a critical role in several physiological processes, including cardiac muscle development, bone formation, adipocyte differentiation, and the innate immune system. HDAC9 is involved in various malignant tumors, leading to malignant tumor development and poor prognosis. Recent reports have demonstrated both pro-oncogenic and tumor-suppressive roles for HDAC9 in different cancers; however, its role in OSCC remains elusive. **Methods:** 10 OSCC and 5 normal oral mucosa tissues were collected from clinically and histologically confirmed primary OSCC patients undergoing surgical resection. Total RNA was extracted and RNA purity was evaluated by NanoDrop. 500 ng of total RNA was used to generate cDNA using Takara cDNA synthesis kit and real time analysis was performed on StepOne Plus RT-PCR system using Ampliqon SYBR Green master mix. The data was normalized to GAPDH and expression levels were determined by  $2^{-\Delta\Delta CT}$  method. **Results:** The RT-qPCR analysis did not show significant change in gene expression level of tumoural HDAC9 in comparison with normal oral tissue ( $P=0.29$ ). **Conclusion:** In the results of this study, no particular change was found in the transcriptional level of HDAC9 in Iranian patients of OSCC compared to normal oral tissues.

**Keywords:** HDAC9, Oral Squamous Cell Carcinoma, Epigenetics, Histone

P30

## Effects of Hydroxytyrosol on the Expression of MMP-2, 9 and TIMP-1, 2 Genes in HepG2 Cell Line

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**Background:** Hepatocellular carcinoma (HCC), one of the most common types of cancer in the world, accounts for almost 90% of all primary liver malignancies. Most cancer-related deaths are affected by cell survival and invasion of cancer cells to other organs. Hydroxytyrosol (HT) is a natural polyphenol compound that has numerous activities, like the ability to inhibit metastasis by regulation of the genes and proteases associated with ECM degradation and invasion of the cancer cells. Therefore this study aimed to investigate the effect of HT on the expression of Matrix Metalloproteinase-2, 9 (MMP-2, 9), Tissue Inhibitor of Metalloproteinases-1, 2 (TIMP-1, 2) in HepG2 cells. **Methods:** In the current study, the human hepatocellular carcinoma cell line HepG2 was treated with different concentrations (50, 100, and 150  $\mu$ M) of HT for 24 hours. The expression levels of MMP-2, MMP-9, TIMP-1, and 2 were determined by RT-qPCR. **Results:** The results showed that HT significantly downregulated MMP-2, 9, in different treatment groups compared to the control group. Also, TIMP-1 gene expression was meaningfully decreased at 50 and 150  $\mu$ M of hydroxytyrosol. Furthermore, hydroxytyrosol decreased the expression level of TIMP-2 at concentrations of 50 and 100  $\mu$ M compared to the control. **Conclusion:** Our findings suggested that HT probably plays a critical role in the inhibition of HCC metastasis by downregulating MMP-2, 9, and TIMP-1, 2. so we concluded that hydroxytyrosol can be helpful in preventing the proliferation of cancer cells.

**Keywords:** Hepatocellular Carcinoma, Metastasis, Apoptosis, Hydroxytyrosol, Matrix Metalloproteinase, Tissue Inhibitor of Metalloproteinases

P31

## Evaluation of Cinnamaldehyde and Cinnamon aqueous extract effects on oxidative stress biomarkers and antioxidant enzymes activity in 5637 bladder cancer cells

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**Introduction:** The growing trend of bladder cancer around the world has become a major concern. Recent therapies with side effects are exacerbating the concern. Treatment based on herbal compounds has always been considered for its low side effects and high usefulness. In this study, the effect of Cinnamaldehyde and Cinnamon aqueous extract on oxidative stress biomarkers and antioxidant enzymes activity in bladder cancer carcinoma, 5637 cell line, was investigated. **Methods:** 5637 cell line, were treated with different concentrations of Cinnamaldehyde and Cinnamon aqueous extract. MTT was used to evaluate cell viability at 24 hours. The concentration of 0.02, 0.04 and 0.08mg/ml for Cinnamaldehyde and 1.25, 2.50 and 5mg/ml for aqueous extract of Cinnamon were selected. To Evaluation of oxidative markers, level of reactive oxygen species (ROS), malondialdehyde (MDA), total oxidant status (TOS), total antioxidant capacity (TAC), oxidative stress index (OSI), superoxide dismutase (SOD) inhibition rate, glutathione peroxidase (GPx) activity and catalase (CAT) activity was assayed. **Results:** ROS, MDA, TOS and OSI index were significantly higher in cell treated with both Cinnamaldehyde and Cinnamon aqueous extract than the control group ( $p < 0.05$ ), while in the treated groups TAC and SOD, GPx and CAT were significantly lower than control group ( $p < 0.05$ ). **Conclusion:** According to the results of the present study, it seems that both Cinnamaldehyde and Cinnamon aqueous extract could be effective in cancer therapy by increasing of ROS and reduction of antioxidant enzymes activity in bladder cancer.

**Keywords:** Cinnamaldehyde, Cinnamon Aqueous Extract, Bladder Cancer, ROS, MDA, SOD, GPx, CAT

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## Serum Micrnas: 31-5p and -382-5p as Novel Potential Biomarkers in Acute Lymphoblastic Leukemia

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**Background:** Acute lymphoblastic leukemia (ALL) is the most common malignancy in children and accounts for 20% of acute leukemia in adults. Recently, microRNAs (miRNAs) have been described as important molecules in hematologic malignancies. This study aimed to evaluate the expression status of miR-28-3p, miR-31-5p, miR-378a-3p, and miR-382-5p in patients with ALL. **Methods:** In this study, 21 patients with ALL (before and after treatment) and 21 healthy people were evaluated. After directly synthesizing cDNA from serum, miR-28-3p, miR-31-5p, miR-378a-3p, and miR-382-5p expression were measured using the qRT-PCR method. Data were analyzed using SPSS 20, Genex 6.1, and GraphPad Prism8 software. **Results:** The expression levels of miR-378a-3p and miR-31-5p in patients with newly diagnosed ALL were significantly higher than in healthy individuals ( $P = 0.001$ ,  $P = 0.004$ ), and decreased significantly after treatment. ( $P = 0.04$ ,  $P = 0.03$ ). The expression of miR-28-3p and miR-382-5p in patients with newly diagnosed ALL were significantly lower than healthy individuals ( $P = 0.09$ ,  $P = 0.01$ ) and were increased significantly after treatment ( $P = 0.001$ ,  $P = 0.006$ ). **Conclusion:** Our results indicated that miR-28-3p, miR-31-5p, miR-378a-3p, miR-382-5p may have a potential role in the pathogenesis of the disease; and also, could be considered as a diagnostic marker and therapeutic target in ALL in future studies.

**Keywords:** ALL, miRNA, miR-31-5p, miR-382-5p



# COVID-19 Vaccination Updates with Emphasis on Medical Laboratory Aspects

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P33

## Fabrication and Evaluation of Fibrin Biological Scaffolds from Plasma-Derived Products with Synthetic Teriparatide Peptide

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Fibrin glue is one of the most important biological products that can be extracted from plasma-derived products such as fresh frozen plasma (FFP). In the current study, we fabricated and evaluated the mechanical and biological properties of fibrin glue. In this study, fibrinogen was extracted from FFP by using ethanol. Thrombin was precipitated by ammonium sulfate separation method and then the fibrin scaffold was fabricated by mixing of both fibrinogen and thrombin. Fibrinogen was characterized by FTIR and SDS-PAGE methods. Rheology, porosity, biodegradability, and sterility tests were performed to characterize the fibrin scaffold. In terms of morphology, (SEM) was also taken from the scaffold. Moreover, the rate of teriparatide release within the scaffold was also measured by the spectroscopic method. Viability was assessed by taking advantage of MTT assay concentration of 50 µg/ml, in this study, the gelation time of the fibrin scaffold was 4±0.2 seconds. The highest infrared absorption for fibrinogen was at 1651. For the fibrinogen sample, 3 bands were observed in SDS-PAGE in 46, 52, and 66 KDa. The porosity of the scaffold was 91%. The elasticity, biodegradability, and sterility of the scaffold were confirmed. Plasma-derived fibrin scaffold has suitable mechanical and biological characteristics for applying as a biological scaffold. Teriparatide peptide has acceptable release in fibrin scaffolds. Fibrin scaffolds loaded by teriparatide peptides has high viability for cells survival.

**Keywords:** Fibrin Glue, Teriparatide, Viability

P34

## The Effect of Vaccine on the Level of Spike Antibodies

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**Introduction:** Due to the prevalence of Covid19 pandemic, the effective Prevention tools against SARS-CoV-2, is urgently needed. Spike protein in viruses is one of the suitable items to stimulate the immune system response. So, researchers focused on spike protein for vaccine designing. Research by (Madhumita Shrotri et al) has shown that vaccines designed based Spike protein (like BNT162b2) provide strong and short-term immunogenicity. **Methods:** In this study, people who received complete virus vaccine or vaccine designed based on the Spike protein were evaluated for antibody levels. The level of the spike anti body was evaluated by ELISA method. Also for control, people who had not received any vaccine and had no history of covid19 disease were selected. **Result:** According to the results, 71% of people who received even one dose of the vaccine, had acceptable levels of anti-spike antibodies. Vaccines which were analyzed in the study are complete cells or based spike protein. In this study, people were selected who had been vaccinated for at least 1 month and at most 4 months. And antibodies against spike were measured and were analyzed according the time that the persons vaccinated. **Conclusion:** According to the present study, almost people had suitable immune response against spike protein in 4 months after injection. It was found that the highest level of antibodies against Spike is present in the first two months after injection.

**Keywords:** Spike, Covid19, Vaccine, Immunogenicity

P35

### Specific Regulatory Motifs Network in SARS-CoV-2 Gastrointestinal Infections

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**Background:** The SARS-CoV-2 was primarily noted as a respiratory pathogen spreading via droplets and aerosols. But later clinical reports highlighted extrapulmonary effects and modes of transmission of the virus, particularly by the gastrointestinal (GI) tract. The aim of current study was the prediction of underlying mechanisms associated with the regulatory network motifs, probably responsible for the SARS-CoV-2 effect in the GI tract. **Methods:** The data were obtained from a recently published study on the effect of SARS-CoV-2 on the Caco-2 cell line as a GI tract model. We used transcription factors-miRNA-gene interaction databases to find the key regulatory molecules, then analyzed the data using the FANMOD software for detection of the crucial regulatory motifs. Cytoscape software was then employed to construct and analyze the regulatory network of these motifs. As well, the KEGG pathway and Gene Ontology enrichment analyses were done to predict the probable intermediating biological processes and biochemical pathways. **Results:** Using bioinformatics tools, we demonstrated one 3edge feed-forward loop motifs (FFLs) and recognized the 10 crucial genes in relation with Caco-2 cell infected by SARS-CoV-2, including SP1, TSC22D2, POU2F1, REST, NFIC, CHD7, E2F1, CEBPA, TCF7L2, and TSC22D1, which some of them have a role in the GI cancers. **Conclusion:** Therefore, our results predict that the GI tract infected by SARS-CoV-2 can cause a serious risk in patients with any type of GI cancer. Hence, further studies on these genes and their pathways relevant to SARS-CoV-2 infection, are recommended.

**Keywords:** SARS-CoV-2, Gastrointestinal Tract, Motif, Network, Colon Cancer

P36

## Evaluation of Secondary Fungal Infection {Invasive Aspergillus} in Hospitalized Patients of Covid 19

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**Introduction:** Respiratory viruses are viruses that enter through the respiratory tract and multiply in the lung mucosa. They become infected One of Covid 19's respiratory diseases was {Corona} which has been spread by SaraCovid2 virus since 2019. It has affected the world and caused many deaths. Concurrent with this viral respiratory infection many Patients develop secondary infections, including bacterial and fungal infections. Invasive Aspergillus is one of the most common, it is a fungal infection and is life threatening in patients who usually have immunodeficiency. There are many reasons, including mechanical ventilation and injectable nutrition, and broad-spectrum bacterial therapy and central resident venous Hospitalization can be a secondary cause of invasive fungal infections. **Methods and Findings:** In this laboratory evaluation, molecular kit and PCR method were used to diagnose Covid 19 patients. ASPERGILUS PLATELIA kit and ELISA method were used to diagnose patients with invasive Aspergillus. First, patients with Covid 19 in the last nine months of 2021 are examined for invasive Aspergillus during this study, we found that all of these patients were hospitalized. On the other hand, 300 patients with Aspergillus Was evaluated positively, of which 245 were positively hospitalized. **Conclusion:** During this study, the relationship between 19 patients hospitalized with Covid and concomitant Aspergillus was confirmed. Therefore, Aspergillus can be considered as a Coinfection of Covid19 patients in ICU section.

**Keywords:** Invasive Aspergillus, Secondary fungal infection, Covid19

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## New Strategies for Successful COVID19 Vaccination in Patients with Chronic Lymphoid Leukemia

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Background Among hematological malignancies, chronic lymphocytic leukemia (CLL) has a high risk of hospitalization and death from COVID-19 due to impaired antibody response. Only less than fifty percent of these patients were seropositive after second dose of COVID19 vaccination. This study aims to explain new strategies for successful COVID19 vaccination in patients with CLL. Methods Data gathering related to COVID-19 vaccination published from January 2021 till February 2022 was conducted using MEDLINE and SCOPUS. Results Most studies suggested BNT162b2 mRNA vaccine for CLL patients. Serological response rate (SRR) of patients with CLL varies between 23 and 47% after administration of the second dose and it was more (about 70%) in treatment-naïve patients compared with those who were under treatment within 12 months. Poor SRR of COVID-19 vaccine can be related to dysregulation of their immune system, hypogammaglobulinemia, lacking CD8+/CD4+ T-cell-specific responses, age  $\geq 70$  years old, male gender, recent treatment with Bruton Tyrosine Kinase inhibitors or ongoing treatment of venetoclax  $\pm$  anti-CD20 monoclonal antibodies within 12 months. Conclusion The findings suggest the administration of COVID19 vaccines before the onset of treatments or delaying the use of anti CD20 in patients with lower tumor burden. The administration of booster doses (third and fourth) after six months of active therapy and heterologous vaccination (use of mRNA and adenovirus-based vaccines) is also needed to induce T cell responses. Besides these new strategies, adhering CLL patients to preventive protocols is so important for being safe from severe form of COVID19.

**Keywords:** COVID19, Vaccine, CLL

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## Evaluation of the Astra-Zeneca Vaccine Effectiveness in Omicron Covid 19 Strain in Laboratory Staffs in Tehran

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**Introduction:** Covid-19 disease is caused by SARS-COV2. This acute respiratory illness that started in Wuhan, China, has become the main public health problem and the most important cause of death in many countries of the world. Vaccination is one of the ways to prevent infection or reduce the severity of clinical symptoms after infection. In this study, we investigated the Astra-Zeneca vaccine effectiveness on the prevention of infection and severity of clinical symptoms among laboratory staffs in Tehran. **Method:** 100 staffs from different laboratories in Tehran who received all 3 doses of Astra-Zeneca vaccine were selected as the study group. In the control group, 50 unvaccinated staffs were selected. The incidence of infection (individuals with positive molecular test for Covid-19) and the severity of clinical symptoms were assessed in these two groups. **Results:** The incidence rates in the vaccinated and non-vaccinated groups were 72.6% and 81.9%, respectively. Mild symptoms were observed in 85.3% and 74.7% of unvaccinated and vaccinated individuals, respectively. More severe symptoms were observed in 5.8% and 23.6% of vaccinated and unvaccinated people, respectively. **Discussion and Conclusion:** Astra-Zeneca vaccine has little effect on the prevention of Omicron strain but is effective in reducing the severity of clinical symptoms and duration of the disease. In vaccinated people mild symptoms including dry throat, hoarseness, sneezing and Runny nose is common and lasts a total of 3-5 days. In non-vaccinated people, more severe symptoms including fever and chills, body aches and coughs are more common and last for 5-7 days.

**Keywords:** Astra-Zeneca Vaccine, Omicron Strain, Covid 19, Laboratory



# Diagnosis of Systemic Mycoses and Quality Control in Mycology Lab

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## Molecular Identification of Candida Species Isolated from Candiduria in Hospitalized Patients

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**Background and objectives:** The incidence of candiduria caused by *Candida* spp. has increased in recent years, particularly in hospitalized patients. Candiduria is most commonly caused by *Candida albicans*; however, an increase in the prevalence of non-*albicans* species has been observed during last decades. This study aimed at molecular identification of *Candida* species isolated from candiduria in hospitalized patients. **Methods:** This cross-sectional study was conducted on 530 hospitalized patients in two hospitals in the Mazandaran Province, Iran. Midstream urine specimens were collected and then cultured on CHROMagar *Candida* medium. Molecular identification of common *Candida* species was carried out using the polymerase chain reaction-restriction fragment length polymorphism method after enzymatic digestion with *MspI*. *C. albicans* and *Candida parapsilosis* species complexes were identified by amplification of the HWP1 and intein-containing vacuolar ATPase precursor genes, respectively. **Results:** The frequency of candiduria was estimated at 14% among hospitalized patients. Of 74 samples positive for candiduria, 65 (87.8%) were isolated from females. The most common predisposing factor to candiduria was diabetes (n=36; 48.6%). The most frequent isolates were *C. albicans* complex (n=44; 59.4%), followed by *Candida glabrata* (n= 16; 21.6%), *Candida tropicalis* (n= 10; 13.5%), *Candida Krusei* (n= 3; 4%) and *C. parapsilosis* (n= 1; 1.3%). **Conclusion:** Based on the results, the conventional and molecular methods produced similar results for identification of *Candida* species. However, accurate identification of *Candida* spp. requires the use of molecular techniques such as PCR-RFLP, HWP1, and intein-containing vacuolar ATPase precursor genes. Nevertheless, chromogenic methods such as CHROMagar *Candida* can be used for diagnosis of *Candida* spp. in laboratories with limited resources.

**Keywords:** *Candida*, PCR-RFLP, Candiduria, Hospitalized Patients



# Geriatric Medical Laboratory

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## Prevalence of Iron Deficiency Anemia Based on Laboratory Criteria in the Elderly of Isfahan Province in 1400

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**Introduction:** Iron deficiency anemia (IDA) is one of the most severe and widespread eating disorders in the world. The elderly are the most vulnerable groups of the disease, who often consume less food sources of iron, and on the other hand, due to reduced gastric acid secretion, iron absorption is reduced in their body. Some may also have chronic gastrointestinal bleeding, which can lead to iron deficiency anemia and symptoms such as general weakness, depression, and decreased physical function. Given that the prevalence of anemia varies in different societies, a careful assessment of each community seems necessary. **materials and methods:** This descriptive study was performed on 4670 patients referred to medical centers in Isfahan. These people were divided into different age groups. Their mean age was  $70.88 \pm 15.27$ . After sampling, blood iron, TIBC and ferritin levels were measured using BT4500 device, and cell count was performed using cell counter device. The obtained information was analyzed by SPSS12 software. **findings:** According to the findings, 411 people (8.80%) are iron deficient, of which 370 (90%) were women and 41 (10%) were men. Using the indices of hemoglobin, hematocrit, MCV, TIBC, Ferretin and serum iron, the rate of iron deficiency anemia was 230 (5%) of which 211 (92%) were female and 19 were male (8%). . According to age classification, the highest group with iron deficiency anemia were women  $61.11 \pm 3.21$  with a frequency of 43%. **Conclusion:** The prevalence of iron deficiency anemia is moderate in the population of the center of the country. Due to the complications of iron deficiency disease in the elderly, taking appropriate measures to reduce the existing deficiency and prevent the occurrence of this deficiency is very necessary and recommended.

**Keywords:** Anemia, Iron Deficiency, the Elderly



# Lab Investigation of Prenatal Genetic Disorders

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**Silent  $\beta$ -Thalassemia:  
Transition Mutation of the  $\beta$ -globin Gene (Promoter nt-101 C>T)**

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**Introduction:** Heterozygote  $\beta$ -thalassemia is called carrier or  $\beta$ -thalassemia trait (BTT). Carriers have no clinical symptoms but sometimes have a mild anemia. They can often be identified with  $MCV < 80$  fl,  $MCH < 27$  pg and  $HbA2 > 3.5$  %. However, these tests are not enough to diagnose some unexpected beta-globin mutations in premarital or prenatal screening. **Case Presentation:** A 21-year-old- girl referred to central laboratory (Fars, Iran) for premarital hematological tests in July 2019. CBC showed normal red cell indices ( $MCV$ : 87.4fl,  $MCH$ : 28.4 pg). Since her fiancée was a typical case of  $\beta$ -thalassemia minor ( $RBC$ :  $7.05 \times 106/\mu l$ ,  $MCV$ : 63.3 fl,  $MCH$ : 18.6 pg), both were referred for Hb electrophoresis and re-CBC testing.  $\beta$ -thalassemia minor was diagnosed in the boy with an HBA2 value of about 5%. HBA2 in the girl was borderline (3.6%). As the case was serious, Molecular studies on DNA were prioritized to indicate the unusual  $\beta$ -thalassemia genotype. DNA was extracted from peripheral blood sample to detect the 20 most common Iranian  $\beta$ -thalassemia mutations by ARMS PCR. No mutation was identified. Therefore, we performed beta-globin gene DNA sequencing. Mutational analysis proved to be heterozygous for -101 C>T  $\beta^+$ -thalassemia trait mutation. **Conclusion:** It was the first report from Fars (Iran) and the second one from Iran. The case had normal hematologic indices and borderline hemoglobin A2 values that may be mistakenly interpreted as normal. The presented case showed that electrophoresis and PCR sequencing methods should be applied for screening thalassemia.

**Keywords:** Beta- Thalassemia, Genetic Carrier Screening, Heterozygote, Hematological Diseases, Anemia

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**Undiagnosed Anemia after 9 Years: Diamond-Blackfan  
(Homozygous Variant c.140G>T; p.Gly47Val in ADA2 Gene); A Case Report**

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**Introduction:** Diamond-Blackfan anemia (DBA) was defined as a rare autosomal recessive inheritance erythroblastopenia and pure red cell aplasia. DBA usually raised from an abnormal ribosomal protein gene. Symptoms of anemia appear in the first year of life especially in 2-3 months. Deficiency of ADA2 due to CECR1 -mutations was reported in cytopenias cases. **Case Presentation:** A two-month-old-girl was hospitalized with paleness, reluctance to milk, lethargy and intense anemia. Then, she underwent RBC transfusions several times as an undiagnosed anemia. After 8 years, elevated ferritin, severe neutropenia and drastic infection complicated the situation. At this time, the molecular techniques led to the identification of homozygously variant c.140G>T p.Gly47Val in ADA2 gene that was inherited from heterozygous parents. **conclusion:** The case was first report from Iran that showed screening congenital defects in apparently healthy couples and particularly prenatal diagnosis (PND) with molecular methods and gene sequencing must be considered very seriously.

**Keywords:** Diamond-Blackfan Anemia, Delayed Diagnosis, Adenosin Deaminase 2, CECR1



# Lesson Learnd from COVID 19: from Bench to Bed Side

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### A Review of General and Genetic Risk Factors Affecting Covid-19 Disease

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**Introduction:** Acute Respiratory Severe Coronavirus Syndrome is an international public health issue. After the outbreak, it was observed that the clinical manifestations of Covid-19 disease were different and people showed symptoms from asymptomatic to severe symptoms and death. Therefore, in the present study, we focus on what factors make people different. **Method:** In this study, articles published between 2018 and 2021 and Cochrane library, Psycinfo, CINHALL, Scopus, Medline, Proquesteh databases were used. The search was performed using keywords such as general and genetic factors, Covid-19, pandemic based on MeSH. Finally, 22 of these articles had inclusion criteria. **Results:** Clinical risk factors such as obesity, old age, gender and underlying diseases that can contribute to the complications of this disease such as cardiovascular disorders, diabetes, renal defects and liver and lung injuries and cancer that can lead to disease. Intensify. It has been observed in various studies that polymorphisms in the genes of blood groups, ACE2, different HLAs, TLR7, TMEM189, TMPRSS2 and UBE2V1, as well as switching and isotype switching processes of antibodies can be involved in the severity of the disease. **Discussion & Conclusion:** Based on the findings of this study, there is a wide range of risk factors to predict the severity of Quid 19 disease and its mortality. The near future helped to control and treat the disease by discovering genetic factors influencing the severity of Covid-19 disease.

**Keywords:** Genetic Risk Factors, General Risk Factors, Pandemic, Covid-19

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## Cross Talk Between Inflammation and Blood Coagulation Markers in COVID-19

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**Background:** COVID-19 after entering the human body activate the inflammatory and coagulation cascades. In fact, the cells of adaptive immune systems including B cells and T cells recognize specifically the virus. The repeated virus antigen exposure by immune systems led to hyperactivation of immune systems ended to cytokine storm and exacerbation of the inflammation. Moreover, the other important phenomenon in nearly all hospitalized patients with COVID-19 is hypercoagulability especially the marked increase in D-dimer. The aim of this review is evaluation some important inflammation (IL-33, sST2, IL-8, IL-6, CRP, Albumin, CRP/Albumin, procalcitonin, IL-10, NETosis) and coagulation (D-Dimer, Fibrinogen, Protein C and S, EPCR, thrombomodulin, tissue factor, VWF, adamts13, Plasmin(ogen) System,  $\alpha$ -2 antiplasmin, PAI-1, ) markers in COVID-19 patients to investigate the reciprocal effect between coagulation and inflammation. **Method:** A total of 50 studies related to COVID-19 from 2019 till 2021 was conducted using PubMed, Google Scholar and Scopus. **Result:** Among the markers studied, albumin, ADAMTS13, protein C and S, EPCR, thrombomodulin decreased but the other markers increased. **Conclusion:** This data suggest that the inflammation and coagulation pathways in COVID-19 have mutual effects on each other and the link between these processes is mediated by endothelium, platelets, and the serine proteinases of blood coagulation. Our opinion is knowing the cross- talk between these pathways would be helpful in selecting the appropriate therapy in diseases associated with thrombosis such as COVID-19.

**Keywords:** COVID-19, Inflammation Markers, Blood Coagulation Markers, Cross Talk

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## The Role of CBC and PBS in the Diagnosis, Prognosis and Severity of Covid-19 Disease

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**Introduction:** In December 2019, coronavirus disease-2019 (covid-19) was identified and reported in Wuhan, China and became a global pandemic. There are various challenges to accurately diagnosing this disease. The Hematology Laboratory plays a key role in this crisis to assist in screening, diagnosis and prognosis of patients. **Method:** We read 10 related articles, all related to 2020, and in these studies; CBC and PBS compared people who tested positive for RNAPCR Covid 19 with healthy people. **Results:** Various reports indicate that CBCs in individuals who tested positive for RNAPCR Covid 19 initially show primary neutropenia, lymphocytopenia, monocytopenia, and subsequent monocytosis. The most common hematologic abnormalities of the CBC are anemia with neutrophilia, shift to the left neutrophilia, and lymphopenia. RBCs are normocytic normochromes with shift to the left and a number of NRBCs and Basophilic stippling. Shows morphological changes: Hypo-segmented neutrophils, annular, club shaped, U-shaped and C-shaped, embryonic, named as covid nucleus, long nucleoplasm, Eosinopenia and atypical eosinophils with multiple vacuoles, Monocytes activated with anisocytosis and vacuolated cytoplasm and small number of granules, large nucleus, thin chromatin and intranuclear bubbles, Most lymphocytes are large granular lymphocytes (LGL), round nucleus, dense chromatin, few nucleoli, light blue cytoplasm with azurophilic granules with varying sizes of different forms of NK cell, T cytotoxic, Lymphoplasmacytoid and immunoglobulin-secreting plasma cells and giant platelets and Platelet satellitism. **Discussion:** CBC and PBS can detect the early signs of inflammation and the effect of the virus. The study of morphological changes of neutrophils, lymphocytes, and activated monocytes in peripheral blood smear of patients is easily traceable and repetitive, and helps diagnose and determine prognosis and perform treatment protocols, and is less expensive and safer than other methods. In this review, we discuss the role of CBC and PBS in the diagnosis, prognosis, and severity of covid 19 disease.

**Keywords:** Peripheral Blood Smear (PBS), Complete Blood Count (CBC), Covid-19

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## The Role of NLR and PLR in Predicting COVID-19 Disease and Its Severity

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**Background:** Coronavirus disease 2019 (COVID-19) is caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Recently, neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) have been used as diagnostic biomarkers in various diseases. In this study, we investigated the role of the NLR and PLR in predicting COVID-19. **Methods:** This retrospective cohort study was performed on 218 COVID-19 patients (including 40 non-severe and 24 severe cases on the basis of the WHO guidelines 2020) and 218 healthy individuals as a control group in Persian Cohort Center of Mashhad University of medical sciences. Patients with any underlying disease were excluded. Blood samples were collected from patients (before COVID-19) and the control group for cell blood count (CBC). NLR, PLR and demographic variables were analyzed with SPSS IBM 25.0. Statistical significance was considered as  $P < 0.05$ . **Results:** The COVID-19 patients and control group were similar in many variables such as age, sex, BMI (Body Mass Index) and blood group ( $P > 0.05$ ). There was no significant difference in NLR and PLR between COVID-19 patients and control group ( $1.45 \pm 0.58$  VS  $1.48 \pm 0.54$ ,  $P = 0.4$  and  $96.44 \pm 30.6$  VS  $94.46 \pm 28.5$ ,  $P = 0.2$  respectively). NLR, PLR and the other demographic variables weren't significant difference between severe and non-severe groups ( $P > 0.05$ ); but BMI in the severe group was significantly lower than the non-severe group ( $P = 0.04$ ). **Conclusion:** In our study, it seems that the NLR and PLR don't play a role in predicting COVID-19 and its severity but BMI was lower significantly in severe COVID-19 patients. **Keywords:** COVID-19, Neutrophil-to-lymphocyte ratio, Platelet-to-LYM ratio.

**Keywords:** COVID-19, Neutrophil-to-Lymphocyte Ratio, Platelet-to-Lymphocyte Ratio

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### Application of Hematological Parameters in Differential Diagnosis of Severe COVID-19 from Non-Severe forms of the Disease

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The 2019 novel coronavirus (2019-nCoV) or the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has emerged as a pandemic threat from December 2019. Coronavirus can cause varying degrees of illness that range from mild to severe or fatal disease. The exact mechanism on hematopoiesis induced by this coronavirus is not yet well understood, but scientific evidence indicates that COVID-19 can cause hematological changes in infected patients. The present study summarized pieces of literature regarding hematologic findings of COVID-19 and their correlation with disease severity. Finally, we offered some laboratory abnormalities which help to differentiate severe COVID-19 from non-severe forms of the disease. Among hematological parameters, decreased hemoglobin rather than anemia, leukocytosis, lymphopenia, neutrophilia, and thrombocytopenia have been observed in conducted studies in some patients with COVID-19. Furthermore, as the disease progresses to severe COVID-19, hemoglobin decline, leukocytosis, lymphopenia, neutrophilia, and thrombocytopenia continue to exacerbate. In addition, the neutrophil-to-lymphocyte ratio is also considered as an independent risk factor for severe infection in COVID-19 patients.

**Keywords:** Coronavirus Disease (COVID-19), Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2), Hemoglobin, Lymphopenia, Neutrophilia, Thrombocytopenia

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## The Importance of Hematological Parameters in Covid-19 Patients

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**Background:** SARS-CoV-2 can cause a wide range of respiratory disorders from mild to severe, driving to acute respiratory distress syndrome (ARDS). Common symptoms of Covid-19 comprise fever, dry cough, and fatigue, while it can drive to complications, including cardiac, renal, liver, and gastrointestinal dysfunctions. Disrupting the hematological parameters is one of the SARS-CoV-2 infection complications assessed by our current study. **Methods:** During the current study, hematological parameters, including the levels of platelet, WBC, RBC, neutrophil, lymphocyte, ESR, BS, Hb, HCT, MCV, MCH, and MCHC, have been evaluated and compared in both control and case groups. Moreover, we evaluated the correlations between underlying medical conditions, clinical manifestations and hematological parameters in the case group. **Results:** We have detected that ESR, RBC, and HCT levels demonstrated a significant correlation with novel SARS-CoV-2 infection and these changes face an increase with the disease progression. ESR ( $P= 0.022$ ), BS ( $P= 0.01$ ), and Hb ( $P= 0.032$ ) levels of male individuals infected by this virus demonstrated a significant correlation with the control group. Although, we recorded no significant correlation between the hematological parameters of female individuals in the case and control group. Furthermore, ESR, BS, HCT, Hb and neutrophil indicated significant changes in diabetic patients afflicted by Covid-19. **Conclusions:** The vast decline of the RBC, WBC, platelet, lymphocyte, Hb, and HCT is correlated with a substantially increased mortality rate, while high BS and neutrophil levels are associated with disease worsening. Aware of the SARS-CoV-2 effects on the hematological parameters and their constant monitoring can contribute significantly to choose the proper treatment, take all necessary measures, and supportive therapies to accelerate the treatment process and impede disease worsening.

**Keywords:** SARS-CoV-2, Covid-19, Hematological Parameters

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### Red Cell Distribution as a Prognostic Indicator for Mortality and ICU Admission in Patients with COVID-19

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**Background:** The coronavirus disease 2019 (COVID-19) is an acute respiratory disease with a high rate of hospitalization and risk of mortality. therefore ,prognostic indicators are important for early, aggressive intervention. Red blood cell distribution width (RDW), as a component of complete blood counts that reflects cellular volume variation.it has been shown that RDW is associated with elevated risk of mortality in a wide range of diseases. Therefore, in this study we aimed to determine relationship between RDW and risk of mortality in covid-19 patients. **Methods:** This retrospective study was performed on 592 patients admitted to the hospital between February 2020 and December 2020. Patients were divided into two groups: the group with  $RDW < 14.5$  and the group with  $RDW > 14.5$ . In both groups, the relationship between RDW and mortality, intubation, admission to intensive care unit and the need for oxygen therapy was investigated. **Results:** results of this study showed that the mortality rate was associated to RDW levels, so that the mortality rate in the group  $RDW < 14.5$  was 9.4%, while in the group  $RDW > 14.5$  mortality rate was 20.2 %.)  $P=0.001$ ).also RDW levels was related to ICU admission as ICU admission in ( $RDW < 14.5$ ) was 8% whereas in group with  $RDW > 14.5$  this percent increased to 10%( $p=0.04$ ).The results of the Kaplan Meyer curve showed that the survival rate was higher in the group with  $RDW < 14.5$  than group  $RDW > 14.5$ . Cox results in the crude model showed that the increase in RDW was directly related to the increase in death although it was not significant after adjustment **Conclusion:** The results of our study showed that the increased RDW is associated with an increase in hospitalization in ICU and an increased risk of death and can be used as a accessible indicator to determine the prognosis of covid-19 patients.

**Keywords:** Covid-19, Red Cell Distribution Width, Mortality, Intensive Care Units

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## Evaluation of Inflammatory Factors in Patients with SARS Covid-19

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**Background:** Most patients infected with the novel coronavirus (SARS-CoV-2), as the causative agent of COVID-19 disease, show mild symptoms, but some of them develop severe illness. The purpose of this study was to analyze the blood markers of COVID-19 patients and to investigate the correlation between serum inflammatory cytokines and the disease severity. **Methods:** In this prospective cross-sectional study, 50 patients with COVID-19 and 20 patients without COVID-19 were enrolled. According to ICU admission criteria, patients were divided into two groups of non-severe and severe. Differences in the serum levels of C-reactive protein (CRP), IL-6, and TNF- $\alpha$ , as well as erythrocyte sedimentation rate (ESR), lymphocytes (LYM) count, and neutrophils (NEU) count between the two groups were determined and analyzed. **Results:** Out of the 50 patients with COVID-19, 14 were diagnosed as severe cases. There was no significant difference between the two groups of COVID-19 patients in terms of gender and age. Blood tests of COVID-19 patients showed a significant decrease and increase in NEU and LYM counts, respectively. There were significant differences in the serum levels of IL-6, TNF- $\alpha$ , and CRP between the severe and non-severe groups, which were higher in the severe group. Also, there was a significant correlation between the disease severity and CRP with ESR ( $r = 0.79$ ), CRP with IL-6 ( $r = 0.74$ ), LYM with NEU ( $r = -0.97$ ), and ESR with TNF- $\alpha$  ( $r = 0.7$ ). **Conclusion:** The findings of this study, as the first study in Iran, suggest that the levels of IL-6, TNF- $\alpha$ , ESR, and CRP could be used to predict the severity of COVID-19 disease.

**Keywords:** COVID-19, CRP, Cytokines, IL-6, SARS-CoV-2, TNF- $\alpha$

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**Laboratory Diagnosis Challenges of Covid-19:  
a Real-Time RT-PCR Assaybased Survey**

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**Introduction:** A novel coronavirus has recently been discovered in Wuhan, China in December 2019 named SARSCOV-2. Sensitive molecular assays are now available for detection of SARS-COV-2. The aims of this study were to detect positive subjects of SARS-COV-2 and to survey some challenges that are encountered practically in detection of the virus by Real-time RT-PCR (rRT-PCR) technique. **Materials and Methods:** 271 subjects including 105 women and 166 men were registered by census method from 15 April to 15 Jun of 2020. Nasopharyngeal and oropharyngeal swab samples of suspicious cases were collected for RNA extraction to identify SARS-COV-2 by rRT-PCR amplification. **Results:** 52 women and 82 men were positive for Covid-19. However, technically the results showed that in the time period of 15 April to 15 May of 2020 the contents of received rRT - PCR kits were infected with positive control. 8 % of certain negative control samples showed a peak resembling to that of the positive samples in the rRT - PCR process. In addition, the results showed that in some of the RT -PCR kits the master mix would produce peaks like positive control without adding the sample to it automatically in 7 % of the tests. **Conclusion:** Having different types of negative control for diagnosis of SARS-COV-2 is necessary in extraction and RT- PCR steps to reduce the chance of having false positive results.

**Keywords:** Coronavirus, Polymerase Chain Reaction, Molecular Study, Quality Control

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## Platelet Aggregation and Adhesion Status in Intensive Care Unit (ICU) Admitted COVID-19 Patients: A Case-Control Study

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**Background:** Some patients with severe forms of COVID-19 had experienced higher rates of thrombotic events. In this study, we aimed to study the platelet aggregation, as well as platelet adhesion in hospitalized patients with SARS-CoV-2, compared with healthy individuals. **Materials and Methods:** In this case-control study, 30 ICU admitted COVID-19 patients, and 12 healthy individuals participated, and written consent was obtained. In the case of primary and secondary homeostasis, prothrombin time (PT), partial thromboplastin time (PTT), thrombin time (TT), von Willebrand factor (vWF), protein C, and S, and fibrinogen were determined. The levels of antithrombin (AT), D-Dimer, and fibrin degradable products (FDP) were also measured. Platelet aggregation and platelet adhesion were measured by the turbidometric, and ELISA methods, respectively. **Results:** The mean age of patients and healthy individuals were  $49 \pm 15$ , and  $43 \pm 11$  years, respectively. There were statistically significant differences in the levels of PT, PTT, fibrinogen, D-Dimer, FDP, vWF, and AT between patients and healthy individuals ( $P < 0.01$ ); while TT and protein C levels were not different between both groups ( $p > 0.05$ ). Also, the percentage of platelet aggregation in response to collagen, ADP, and ristocetin agonists was significantly different between study groups ( $p < 0.01$ ). The percentage of platelet adhesion was also significantly higher in the patient group than in the controls ( $p < 0.003$ ). **Conclusion:** The function of the coagulation system in patients with COVID-19 is significantly altered compared to healthy individuals. Measurement of coagulation variables and platelet aggregation can be helpful in determining the prognosis of patients with COVID-19.

**Keywords:** Aggregation, Adhesion, COVID-19, Platelet

P53

## HIV-Positive Patient, with Concurrent COVID-19 and Cryptococcus Infections, a Case Report

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**Background:** HIV positive individuals are more prone to secondary and opportunistic infections. This study investigated a case of HIV-positive diagnosed with COVID-19, who infected with Cryptococcus. **Methods:** The patient was a 53-year-old woman with COVID-19 symptoms. COVID-19 Real-time PCR, hepatitis C virus (HCV) and HIV tests were performed. Lumbar puncture (LP) was done and cerebrospinal fluid (CSF) glucose and protein evaluated. Temporal artery colour-Doppler sonography and Skull XR was performed. **Results:** Severe tension-type -generalized headache, nausea, and photophobia were the main physical symptoms. COVID-19 was found to be positive. Right axis deviation (RAD) and left bundle branch block (LBBB) were diagnosed. HIV-positive and blood impaired circulation were reported. White blood cells on 6 months prior to referral were  $30 \times 10^3$  cells/ $\mu$ l, but during hospitalization it was decreased to  $8.2 \times 10^3$  cells/ $\mu$ l (2021-9-9). Finally, it was increased to  $14.7 \times 10^3$  cells/ $\mu$ l (2021-9-28). There was no change in the amount of C-reactive protein (CRP) (15 mg/dl). CSF test showed that glucose and protein were 15 and 240 g/dL, respectively. Results of direct observation with slide, India ink stain and blood culture confirmed Cryptococcus. Finally, Cryptococcal meningitis was diagnosed. Patient stabilized with amphotericin B, fluorocytosine, fluconazole, acyclovir and vancomycin, co-trimoxazole, dexamethasone, pethidine, liposomal amphotericin, depakine, ondansetron, lorazepam and had been discharged from hospital. **Conclusion:** Fungi are one of the most opportunistic infections that can be cause disease severity in individuals with immunodeficiency disease such as HIV, especially in patients with COVID-19. Prompt diagnosis and treatment should be performed immediately in these patients.

**Keywords:** HIV positive, COVID-19, Cryptococcus, Cryptococcal Meningitis



# Lipid Profiling in Clinical Laboratory

**P54**

P54

**Evaluation of the Effect of Aerobic Exercise with Adenosine Injection on UCP-1 and P38 MAPK Gene Expression in Subcutaneous Adipose Tissue in Male Wistar Rats Fed High-Fat Diet**

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**Background:** Changes in diet to high-fat diets (HFD) along with reduced physical activity increase obesity. **Methods:** In the present experimental study, 40 rats were randomly divided into four groups: 1. normal control, 2. HFD control, 3. HFD + adenosine, and 4. HFD + aerobic exercise + adenosine. After 13 weeks of the HFD, the aerobic exercise group performed the exercise protocol on a treadmill which includes 12 weeks of moderate-intensity endurance training (MIT). UCP-1 mRNA and p38 MAPK levels were measured by RT-PCR. One-way analysis of variance and the Kruskal-Wallis test was used to analyze the data at the level of  $P \leq 0.05$ . **Results:** UCP-1 mRNA and MAPK P38 gene expression showed a significant difference in all four groups ( $P = 0.001$ ). In the HFD + aerobic exercise + adenosine and the HFD + adenosine, a significant increase in UCP-1 mRNA was observed compared to the normal and HFD control groups + placebo. Also, in P38 MAPK, a significant decrease was observed in the HFD + aerobic exercise + adenosine ( $P = 0.002$ ) and HFD + adenosine ( $P = 0.005$ ) compared to the HFD control. **Conclusion:** Aerobic exercise and adenosine can be used as a suitable stimulus in the expression of UCP-1 mRNA and as lipolytic agents in obesity. The p38 MAPK pathway is a double-edged sword that causes increased glucose uptake into insulin and mitochondrial oxidative phosphorylation to occur in a healthy lifestyle through aerobic activity.

**Keywords:** Aerobic Exercise, Adenosine, Uncoupling Protein-1, P38 Mitogen-Activated Protein Kinase, Obesity



# Microbiological Diagnostic of Tuberculosis and Mycobacteriosis

**P55 - P56**

P55

## Evaluation of the Xpert MTB/RIF Test Accuracy for Diagnosis of Pulmonary Tuberculosis in Mazandaran Province

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**Background:** Mycobacterium tuberculosis (MTB) is a causative agent of tuberculosis (TB) which remains as an endemic disease in the North regions of Iran. The sputum smear examination and the culture method were the primary and gold standard methods for TB diagnosis, respectively. Recently, the Xpert MTB/RIF assay (Xpert), was introduced for rapid diagnosis of TB. The aim of this study was to evaluate the performance of the Xpert assay for diagnosis of TB in Mazandaran province. **Methods:** The pulmonary TB suspected cases which referred to Tuberculosis Reference laboratory, Gorgan, Iran, between March 2018 to February 2019 were included to this study. The specimens were decontaminated by Petroff's method then examined using smear microscopy and cultured into Lowenstein-Jensen (LJ) media. The Xpert analysis was performed according to the manufacturer's instructions. Sensitivity and specificity of smear microscopy and Xpert were calculated using a culture method as reference standard. **Results:** Of 42 presumptive TB cases, 31 (73.8%) had culture proven TB. Compared to the culture method, sensitivity and specificity of Xpert was 93.5% (29/31) and 100% (11/11), respectively. Sensitivity and specificity for smear microscopy was 83.9% (26/31) and 100% (11/11), respectively. **Conclusion:** Our findings showed the excellent sensitivity and specificity for the TB detection using Xpert method. In comparison with the culture as a reference standard, the Xpert assay reported two false-negative results, which were smear-negative, too. This could be attributed to low bacillary load in the specimens because the detection limit of the Xpert assay is higher than the culture method. Showing the high sensitivity for Xpert, the smear microscopy could be replaced with Xpert for rapid detection MTB in sputum specimens.

**Keywords:** Mycobacterium Tuberculosis, Xpert MTB/RIF, Lowenstein-Jensen, Smear Microscopy, Tuberculosis

P56

## The Role of Private Sector in the Control and Containment of Tuberculosis

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**Background:** Despite the ceaseless efforts of the government sector, the containment and control of tuberculosis in Iran has been slowed by lack of enough attention to mycobacterial laboratories in the private sector; while about 30% of individuals suspected of tuberculosis refer to laboratories in the private sector. As an example of the role of private mycobacterial laboratories, we present the following statistics. **Results:** In the past two decades only, Zarifi laboratory has performed direct smear and culture tests on 88,303 samples from 58,349 individuals suspected of tuberculosis referred from various hospitals and laboratory centers. Of these samples, 4,842 (8.3%) patients affected with tuberculosis were identified and reported to their treating physicians. Furthermore, of the 58,349 individuals suspected of tuberculosis, 3,844 patients (6.6%) were identified through the direct smear test with an average sensitivity of 78%. In other words, 78% of true positive cases were identified within only 24 hours. Furthermore, 998 patients (1.7%) whose samples contained less than 5,000 acid fast bacilli per milliliter, resulting in a negative smear test, were identified through the culture test in the Löwenstein–Jensen medium. **Conclusion:** Supporting the active mycobacterial laboratories in the private sector and resolving their existing problems and ongoing obstacles as well as enhancing the level of collaboration with these private laboratories by the ministry of health can help expedite the control and containment of tuberculosis in the country.

**Keywords:** Controlling Tuberculosis, Collaboration with the Private Sector



# Modern Technologies in Laboratory Hematology

**P57 – P62**

P57

**Evaluation of Effective Factors in Optimizing Immunization  
(In Vitro Immunization) to Achieve Immortalized Clone Producing Antibody Against  
RhD Antigen in Culture Medium**

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**Background:** Antibodies against RhD antigen are important in transfusion medicine to evaluate the type of RhD and in the clinic to produce rugam ampoule for high-risk Rh-negative mothers. Therefore, in this study, the effective factors in the optimization of immunization to achieve the immortal antibody-producing clone to obtain Anti-D in the culture medium were investigated. **Methods:** Peripheral blood lymphocytes were isolated from O- whole blood by Ficoll method. These lymphocytes were then treated with L-leucyl L-leucine methyl ester (LLME) and cultured with soluble and particulate RhD antigen, interferon-gamma (IFN- $\gamma$ ), interleukin-4 (IL-4), supernatant of Mixed Lymphocyte Reaction (MLR) and CpGODN were exposed to 37 ° C for one week. After this incubation period, in order to evaluate the production of antibodies from lymphocytes on the supernatant obtained from the culture medium, ELISA was performed. After ELISA, antibody-producing lymphocytes were fused with SP20 myeloma cells to achieve a stable hybridoma. **Results:** The use of IFN- $\gamma$ , CpGODN and the combination of IFN- $\gamma$  with IL-4 and IFN- $\gamma$  with CpGODN were identified as the most optimal factors for the production of antibodies from lymphocytes isolated by Ficoll method. The optimizing effect of MLR-derived supernatant on in vitro immunization depends on the type of cytokine and the presence of antibodies in it. Also, lymphocytes immunized with these optimizing agents due to the increase in the number of lymphocytes overcame the low efficiency of the fusion method for the production of monoclonal antibodies. **Conclusion:** The use of immunization optimizing agents has an effective role in achieving immortalized clone producing antibody against RhD antigen.

**Keywords:** Blood Group Antigen, Immunization, In vitro Techniques, Antibodies, Monoclonal

P58

### **Evaluation of Erythrocyte Membrane Protein Defects in Hereditary Hemolytic Anemias using Flowcytometry & Electrophoresis (SDS-PAGE)**

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**Background and Aim:** Anemia is called as low hemoglobin, hematocrit, or red blood cell count lower than normal levels. Hereditary hemolytic anemia may be due to membrane, hemoglobin, or metabolic-enzymatic system of red blood cells. This study evaluated defects of red blood cell membrane proteins in hereditary hemolytic membrane anemia by flow cytometry and electrophoresis method. **Methods and methods:** This study was performed on 70 patients, 34 women (48.6%) and 37 male (51.4%) with a mean age of  $23.25 \pm 16.58$  years with a diagnosis of hemolytic anemia. The diagnostic value of flow cytometry and evaluation of SDS-Page electrophoresis of red blood cell membrane proteins in comparison with routine hematologic and biochemical techniques, for diagnosis of hemolytic anemia was done. **Results:** The values of MCHC, Reticulocyte, RPI, total and direct bilirubin in patients with membrane defect were significantly higher than those without membrane defect and Hyperdense levels were significantly lower than those without membrane defect ( $p: <0.05$ ). MCF levels in osmotic fragility test in patients with hereditary spherocytosis were higher than normal subjects and MFI levels were lower in EMA staining than control group ( $p: <0.05$ ). Electrophoresis evaluation of red blood cell membrane proteins by SDS-Page in patients showed the Spectrin protein deficiency with most frequency (73.5%) and then the Spectrin dimer and band 3 proteins frequency (28.6%) with a sensitivity of 92.86% and 100% specificity respectively. **Conclusion:** The detection of membrane hemolytic anemia at the time of diagnosis in laboratory is difficult and not differentiated from other anemias and additional analyses are needed for the final diagnosis. Before and after osmotic fragility incubation, EMA flow cytometry, OFT-FLOW tests and for definitive diagnosis, protein electrophoresis of red blood cell membrane by SDS-Page is needed.

**Keywords:** Hemolytic Anemia, Red Blood Cell Membrane Defects, Flow Cytometry, Electrophoresis, Osmotic Fragility Test (OFT)

P59

### Prevalence of Heterozygous Hb D Trait in Bushehr

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**Introduction:** Hemoglobin D (Punjab/Los Angeles and Iran) is the second most common hemoglobin variant in South west of Iran. It is derived from a point mutation in the beta-globin gene. Hemoglobin D can be inherited in heterozygosis with hemoglobin A causing no clinical or hematological alterations, or in homozygosis, the rarest form of inheritance, a condition that is commonly not related to clinical symptomatology. In this epidemiological study, the prevalence of HbD among patients with hemoglobinopathy in Bushehr was investigated. **Material and Methods.** This descriptive-analytical cross-sectional study was performed on patients with anemia who referred to Mehr clinical laboratory in Bushehr for hematology tests and hemoglobin electrophoresis. In addition to diagnosing HbD cases, hematologic values and hemoglobin electrophoresis of patients were assessed with SPSS version 22. **Results:** Out of 103 patients with hemoglobinopathy, cases of hemoglobin D (7.76%) were identified. There was a significant direct correlation between HbD and MCV and a significant inverse correlation between HbD and HbA/RDW. **Conclusion:** This study showed that other hemoglobin variants and different beta-gene cluster deletions in the region are considerable and should be screened.

**Keywords:** Hemoglobin D, Hemoglobinopathy, Hemoglobin Variant

P60

### **A Rare Case of IgA/Kappa Monoclonal Plasma Cell Dyscrasia Manifesting with Severe Cold Agglutinin Disease**

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**Introduction:** Cold agglutinin disease (CAD) is an uncommon type of autoimmune disease characterized by autoantibody directed against antigens on the surface of red blood cells. There are compelling evidences that the clonal B lymphocytes in bone marrow are responsible for production of these antibodies. Cold agglutinin disease secondary to lymphoproliferative disorders are usually caused by IgM monoclonal antibodies and the frequency of IgG or IgA type is very rare. **Case presentation (Methods and results):** Herein, we describe a case of severe cold agglutinin disease presenting with a monoclonal immunoglobulin of the IgA class. The initial laboratory analyses showed a negative direct anti-globulin test, and normal levels for LDH, BUN and creatinine. C-reactive protein level was 48 mg/dl and viral studies illustrated negative results for HIV, HBS, and HIV. Further investigations were performed by serum protein electrophoresis, immunofixation, and bone marrow studies. Results demonstrated a monoclonal IgA/kappa band and about 30% monoclonal plasma cells which were positive for CD19, CD56, CD38, CD138 and cytoplasmic kappa light chain restriction. **Conclusion:** So far, a limited numbers of multiple myeloma cases with monoclonal IgA-mediated CAD have been reported. The infrequency of the disease and some misleading lab findings (negative DAT and normal renal panel tests) resulting from the biological characteristics of IgA, may make some diagnostic challenges.

**Keywords:** Monoclonal Gammopathy, Cold Agglutinin Disease

P61

## Investigation on Establishment of Lymphoblastoid Cells Producing KELL Antibody using Alloimmunized Thalassemia Patients Against KELL Antigens

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**Background:** Monoclonal antibody production can be considered as dramatic revolution in the field of treatment, diagnosis and medical research. One of the diagnostic applications of monoclonal antibodies is in the field of immunohematology and blood banking. Determining the compatibility of transfused blood to patients who need frequent blood transfusions such as major thalassemia patients is one of the main steps to prevent alloimmunization of these patients. In an experimental study to produce KELL antibodies of human origin, which is one of the most important antibodies in acute and delayed blood transfusion reactions, we used the blood of patients immunized against KELL antigen and to immortalize the cells we used EBV-Transformation method. **Method:** IN an experimental study, blood was collected from ten thalassemia patients with KELL antibodies. The mononuclear cells were isolated by Ficoll-hypaque density gradient method. The cells were then exposed to EBV virus soup for 2 hours for immortality. The immortalized cells were cultured in RPMI medium containing 1 µg CPG-ODN and 1 µg cyclosporine. **Results:** after 48 hours of culturing immortalized cells, lymphoblastoid cell clones were visible and on the seventh day, antibody production was confirmed by ELISA. **Discussion:** We found that KELL antibodies could be produced from sensitized cells of thalassemia patients against KELL blood group and immortalized by EBV-Transformation method.

**Keywords:** Monoclonal Antibodies, Thalassemia, Alloimmunization, Epstein-Barr Virus

P62

### **Investigation on Establishment of Lymphoblastoid Cells Producing E Antibody using Alloimmunized Thalassemia Patients Against E Antigens**

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**Background:** One of the applications of Epstein-Barr virus in research is the immortalization of B lymphocytes and the lymphoblastoid cell line formation. Thalassemia is one of the most common disorders of hemoglobin and affected people usually need blood transfusions that the main complication of it is alloimmunization. Antibody against RhE antigen cause hemolytic reactions. Due to the high prevalence of anti-E in thalassemia patients and its clinical importance, in this study, using peripheral blood mononuclear cells (PBMC) of alloimmunized thalassemia patients against E-antigen and exposure to EBV, an anti-E-producing lymphoblastoid cell line was established. **Method:** in an experimental study, mononuclear cells were isolated by ficules and exposed to EBV for 2 hours. immortalized cells were cultured in RPMI medium containing 1 µg / ml cyclosporine. After 2-3 weeks, clone formation in the wells was investigated and hemagglutination test was performed for wells containing the clone; The total amount of positive well antibodies was measured by ELISA method. **Results:** Exposure of Patients' mononuclear cells to EBV causes cell transformation and clone formation in some wells. ELISA results showed that the clones are able to produce antibody against RH-E antigen. **Discussion:** We found that E-antibodies could be produced from sensitized cells of thalassemia patients against E blood group and immortalized by EBV-Transformation method.

**Keywords:** Antibody, Thalassemia, Alloimmunization, Rh E Antigen, EBV



# Omics Technologies in Medical Laboratory

P63 – P64

P63

### Association of the E-Selectin Gene Polymorphisms of G98T and C1901T and Serum E-Selectin Level with Risk of Coronary Artery Disease

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**Background:** Genetic factors play an important role in atherosclerosis. Polymorphism in cell adhesion molecules may act as a genetic risk factor for atherosclerosis disease. Relationships among polymorphism in the E-selectin genes and among its soluble levels with atherosclerosis have been observed in different populations. The aim of this study was to investigate these genetic polymorphisms as well as the level of soluble E-selectin in patients with atherosclerosis in comparison with healthy people. **Methods:** 145 normal individuals and 154 patients suffering from atherosclerosis were enrolled into the present study. The presence of disease in the patients was confirmed by the angiographic diagnostic method in the Shahid Madani Hospital. In all samples, the genotypes of Val125Leu and Gly670Arg polymorphisms G98T and C1901T were determined in the E-selectin gene using the RFLP-PCR method, and also the serum level of soluble E-selectin was measured. **Results:** The T allele in C1901T polymorphism was significantly associated with increased risk of atherosclerosis. ( $P = 0.018$ ). Although G98T and C1901T polymorphisms in E-selectin were not significantly associated with increased risk of atherosclerosis. The level of atherogenic lipids was higher in the patient group in comparison with the control group. Moreover, the mean serum level of soluble E-selectin in the patient group was significantly higher than the control group. **Conclusion:** Based on the results of this study, it can be concluded that the allele type in C1901T polymorphism plays a role in increasing the risk of developing atherosclerosis. Furthermore, since serum E-selectin level is associated with the systemic inflammation, it contribute to the increased risk of the disease.

**Keywords:** Cardiovascular Diseases, Atherosclerosis, Adhesion Molecules, Soluble E- Selectin, Genetic Polymorphisms

P64

## The Relationship between FABP2 Ala54Thr and CRP+1059C/G Polymorphisms with Atherosclerosis in the Southwest of Iran

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**Background:** Atherosclerosis is a multifactorial disease influenced by environmental and genetic factors. The polymorphisms of fatty acid-binding protein 2 (FABP2) and C-reactive protein (CRP) might act as genetic risk factors for atherosclerosis. The present study sought to investigate the relationship between FABP2 Ala54Thr (rs1799883) and CRP+1059C/G (rs1800947) polymorphisms and atherosclerosis. **Methods:** In this case-control study, a total of 255 subjects, including 125 controls and 130 patients were included. The DNA was extracted, and then the FABP2 and CRP gene polymorphisms were determined by PCR-restriction fragment length polymorphism and allele-specific PCR methods respectively. **Results:** In this study, there were no significant distinctions between the patient and control groups ( $P>0.05$ ) concerning the genotype and allele frequency of FABP2 Ala54Thr and CRP 1059C/G. **Conclusion:** The present study indicated that FABP2 Ala54Thr and CRP+1059C/G polymorphisms were not associated with any increased risk of atherosclerosis in the population of the Southwest of Iran.

**Keywords:** Atherosclerosis, Fatty Acid-Binding Protein 2, C-Reactive Protein, Polymorphism



# Sepsis and Systemic Inflammation: Laboratory Verification of Diagnosis

**P65**

P65

## Seroepidemiologic Study of Leptospirosis in Slaughter House Personnel Using MAT Technique and Verification of the Positive Samples Using Nested PCR

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Leptospirosis is a common disease between humans and animals. Gilan Province due to its temperate and humid climate, as well as the prevalence of permanent irrigation, traditional livestock breeding in rural homes and also the breeding of some domestic animals and the presence of some wild animals and rodents in the vicinity of villages and farms and finally water abundance. the aim of this study was to investigate the seroepidemiological frequency of leptospirosis in employees of one of the slaughterhouses in Gilan province using MAT method and confirm positive samples by Nested PCR method. In this study, 150 employees of slaughterhouses in Gilan province were sampled after completing a questionnaire with written consent. a serum sample was taken and evaluated by MAT method if the subjects were positive. Urine samples were taken for Nested PCR. According to the results, 98.7% of the participants in this study were women. The mean age of the participants was 35.49 years, of which 44.7% were in the age range of 30 to 39 years, 24.7% were in the age range of less than 30 years and 30.7% were in the age range of more than 40 years. 48% had 1 to 3 years of work experience, 32.7% had 3 to 6 years of work experience and 19.3% had more than 6 years of work experience. The MAT test was positive for 10.7% of participants and negative for 89.3% of them. Nested PCR was negative for MAT positive samples collected from infected patients's urine.

**Keywords:** Leptospirosis, Staff of Slaughterhouses, Gilan Province, MAT Method, Nested PCR



# **Serologic Diagnosis of Infectious Disease (H. Pylori and Brucellosis)**

**P66 – P67**

P66

## Evaluation of Helicobacter Pylori Antigen Frequency in Patients Referred to the Special Gastrointestinal Clinic of Mazandaran University of Medical Sciences in the Years 2020 to 2022

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**Background:** Helicobacter pylori is pathogenic gram negative bacteria that causes gastric chronic inflammation, peptic ulcer and gastric cancer. The most part of patient's do not have symptoms or have a few signs that some of them may have low gastric including vomiting stomach pain and blowing. Therefore, the purpose of this study have done the frequency of helicobacter pylori in stool samples of patients referred to gastrointestinal clinic. **Material and Methods:** In a cross sectional study, from 2020 to 2022, we evaluated 1600 patients that referred to mazums gastrointestinal clinic. Stool sample of patients analyzed for determining of helicobacter pylori antigen. In addition, IgM, IgG and IgA antibodies assessed in the serum of patients by ELISA method. **Results:** The results of this research have shown that 10% of individuals were positive for helicobacter pylori. Furthermore, IGM (2%), IgG (20%) and IgA (12%) was higher than reference value. **Conclusion:** The presence of helicobacter pylori antigens in patients samples with clinical symptoms show a specific important role of this bacteria in pathogenesis. Therefore, more studies is necessary in these individuals for decreasing of disease burden, prevention of more inflammation and also gastric cancer.

**Keywords:** Helicobacter Pylori, Antigen, Antibody

P67

### Mixed Infection of HIV+, Brucellosis and Cutaneous Leishmaniasis: A Case Report

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**Background:** HIV (human immunodeficiency virus) is a virus that damages the cells in human immune system. Brucellosis is a bacterial infection that spreads from animals to people. Most commonly, people are infected by eating raw or unpasteurized dairy products. Leishmaniasis caused by intracellular protozoan parasites (*Leishmania*) and prevalent worldwide, including cutaneous leishmaniasis. **Methods:** In this presentation, a mixed infection of HIV+, brucellosis and disseminated cutaneous leishmaniasis is reported. **Results:** A 30 year old positive HIV man presented with a 3 years history of disseminated cutaneous leishmaniasis lesions on trunk and face and brucellosis infection as well. Amastigots were seen in direct smears stained with wright stain. The causative agent of cutaneous leishmaniasis identified as *Leishmania major*. Clinical symptoms of brucellosis were fever, sweats, joint pain, loose of appetite, weakness and fatigue. The titer of *Bruella* antibody was 1/ 160. **Conclusion:** Immune- compromised patients are susceptible to infection. Brucellosis is under the control. Combination therapy and follow- up appear to be well tolerated, safe, and effective and may be considered as an option for treatment of infection. The cure of lesions is not completed yet.

**Keywords:** HIV+, Cutaneous Leishmaniasis, Brucellosis



# Updates on Hepatitis B and C Vaccine and Laboratory Diagnosis

**P68 – P75**

P68

### **Molecular Investigation of Human Cytomegalovirus and Epstein-Barr virus in Glioblastoma Brain Tumor: A Case-Control Study in Iran**

**Hadi Ghafari \***

**Background:** Glioblastoma multiforme is the most invasive and lethal form of brain cancer with unclear etiology. Our study aimed to investigate the molecular prevalence of HCMV and EBV infections in patients with GBM. **Methods:** This case-control study was conducted on 42 FFPE brain tumor samples from GBM patients and 42 brain autopsies from subjects without neurological disorders. The presence of EBV and HCMV DNA was determined, using PCR and nested-PCR assays, respectively. **Results:** HCMV DNA was detected in 3 out of 42 (7.1%) of GBM samples and was absent from the control group ( $p = 0.07$ ). Importantly, EBV DNA was detected in 9 out of 42 (21.4%) brain tissue specimens of GBM subjects, but again in none of the control group ( $p = 0.001$ ). **Conclusion:** Our findings indicate that infection with EBV is associated with GBM.

**Keywords:** Brain Tumor, Epstein-Barr Virus, Glioblastoma, Human Cytomegalovirus

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### **Inhibition of H1N1 influenza virus infection by zinc oxide nanoparticles: another emerging application of nanomedicine**

**Hadi Ghafari \***

**Background:** Currently available anti-influenza drugs are often associated with limitations such as toxicity and the appearance of drug-resistant strains. Therefore, there is a pressing need for the development of novel, safe and more efficient antiviral agents. In this study, we evaluated the antiviral activity of zinc oxide nanoparticles (ZnO-NPs) and PEGylated zinc oxide nanoparticles against H1N1 influenza virus. **Methods:** The nanoparticles were characterized using the inductively coupled plasma mass spectrometry, x-ray diffraction analysis, and electron microscopy. MTT assay was applied to assess the cytotoxicity of the nanoparticles, and anti-influenza activity was determined by TCID50 and quantitative Real-Time PCR assays. To study the inhibitory impact of nanoparticles on the expression of viral antigens, an indirect immunofluorescence assay was also performed. **Results:** Post-exposure of influenza virus with PEGylated ZnO-NPs and bare ZnO-NPs at the highest non-toxic concentrations could be led to 2.8 and 1.2 log<sub>10</sub> TCID50 reduction in virus titer when compared to the virus control, respectively ( $P < 0.0001$ ). At the highest non-toxic concentrations, the PEGylated and unPEGylated ZnO-NPs led to inhibition rates of 94.6 and 52.2%, respectively, which were calculated based on the viral loads. There was a substantial decrease in fluorescence emission intensity in viral-infected cell treated with PEGylated ZnO-NPs compared to the positive control. **Conclusions:** Taken together, our study indicated that PEGylated ZnO-NPs could be a novel, effective, and promising antiviral agent against H1N1 influenza virus infection, and future studies can be designed to explore the exact antiviral mechanism of these nanoparticles.

**Keywords:** Antiviral Activity, Zinc Oxide Nanoparticle, H1N1 Influenza, Polyethylene Glycol

P70

**Molecular detection of high-risk Human Papillomavirus DNA in oral lichen planus patients: a case-control study**

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**Background:** Oral lichen planus (OLP) is a cell-mediated inflammatory mucosal disorder, classified as an oral potentially malignant disorder. Some research has shown that apoptosis in OLP cells is similar to a viral infection such as human papillomavirus (HPV). This case-control study aims to investigate the association of high-risk HPV with OLP. **Material and methods:** DNA was extracted from Twenty-five OLP and another 25 normal oral tissue as case and control groups, respectively. The presence of high-risk HPV DNA was investigated by PCR. **Results:** Twelve of 25 (48%) of OLP samples were positive for HPV16, compared with six of 25 (24%) of normal tissue ( $P=0.07$ ). in the case of HPV18, three of 25 (12%) of OLP samples were positive compared with one of 25 (4%) of normal tissue ( $P=0.3$ ). However, the total frequency of both high-risk HPV was 14 of 25 (56%) and 7 of 25 (28%) in OLP and normal tissue, respectively, showing a significant association between the high-risk HPV and OLP ( $P=0.04$ ). high-risk HPV was more prevalent in Erosive atrophic (EA) OLP as compared to non-EA, although no significant relationship was found ( $P=0.13$ ). **Conclusion:** The results suggest a possibly important association between High-risk HPV and OLP.

**Keywords:** Human Papillomavirus, Oral Lichen Planus, OLP

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## Persistence and Decline of Anti-SARS-CoV-2 Spike Protein among Iranian Population Based on the Voluntary Blood Donors

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**Introduction** Recent research suggested that SARS-CoV-2 IgG antibodies persisted in patients for months after natural infection. This study aimed to investigate IgG-antibody response to SARS-CoV-2 in Iranian blood donors with positive results in RT-PCR and or chest CT scan testing. **Materials and methods** A population-based serial cross-sectional study was conducted on Iranian blood donors during the third weeks of September 2020 through November 2020 for anti-SARS-COV-2 antibody using EUROIMMUN Anti-SARS-CoV-2 ELISA (IgG). A questionnaire was filled out by the trained physicians to gather the donors, information. Individuals with a positive result in diagnostics tests of RT-PCR and or chest CT scan of the lungs were included in the study. To extrapolate seroprevalence estimation from the blood donation samples to the general population, the weighted seroprevalence adjusted for test performance was estimated. **Results** A total of 344 participants were included. Seroprevalence was detected in 78.18% (70.65-85.70) of the subjects. **Discussion** The results showed that approximately three-quarters of the study population had detectable specific IgG antibodies, and in the rest, specific IgG antibodies were not detected. Our study confirmed that IgG antibodies against SARS-CoV-2 waned overtime after the resolution of infection. Although not producing specific IgG antibodies after exposure to the virus should be considered.

**Keywords:** SARS-CoV-2, COVID-19, Seroprevalence, Blood Donors, Iran

P72

### **Prevalence of Hepatitis B Surface Antigen and Risk Factors Associated with Hepatitis B Infection in Pregnant Women in Yazd City**

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**Introduction:** Hepatitis B virus transmission is possible through various routes, including vertical transmission from mother to fetus. Maternal-to-fetal transmission can lead to chronic hepatitis B infection and eventually adult liver cancer. The aim of this study was to determine the prevalence of hepatitis B surface antigen (HBsAg) in pregnant women in Yazd. **Methods:** The study population consisted of 50 pregnant mothers referred to health centers in Yazd. Blood samples and demographic data were taken from all patients from all cases and analyzed for detection of HBsAg by ELISA. Statistical analysis was performed using SPSS 18 software. **Results:** Study group consisted of 50 pregnant women, among them, one sample (2%) was reactive (HBsAg+), forty-eight samples (96%) were non-reactive (HBsAg-) and one sample (2%) was Border line, and the prevalence of HBsAg positive in pregnant women increases with age increasing. **conclusion:** HBsAg screening test is usually more important in pregnant women, especially in women at risk, and it is recommend retesting of high-risk pregnant women in their 3rd semester whose primary tests are negative.

**Keywords:** HBsAg, Pregnant, Women

P73

## The Frequency of Human Polyomavirus BK in Patients with Systemic Lupus Erythematosus: A Cross-Sectional Case-Control Study

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**Background and Aim:** Systemic lupus erythematosus (SLE) is an autoimmune disease and human polyomavirus BK (BKV) can be reactivated in patients with SLE due to the changes in the immune system and use of immunosuppressive drugs. In this study, we evaluated the prevalence of BKV infection among patients with SLE referred to Golestan hospital in Ahvaz, Iran, April 2013 to June 2016. **Methods:** In this cross-sectional study we studied 75 individuals including 40 patients with SLE and 35 normal individuals. Urine and blood samples were taken and DNA was extracted from urine and plasma. Polymerase Chain Reaction (PCR) test was used to detect the BKV genome and positive samples were sequenced to confirm BKV. BioEdit software and MEGA 6.0 software were used for phylogenetic analysis to assemble the viral genome. A phylogenetic tree was constructed by neighbor-joining analysis with 1,000 replicates of the bootstrap resampling test using Mega 6.0. Statistical analysis was done by SPSS version 22. **Results:** Among the 40 patients, 2 (5%) were men and 38 (95%) were women. The mean age of the patients was  $39 \pm 10$  years. 2.5% of plasma from patients with SLE were positive for BKV but none of the controls were positive in this regard. 0% of control groups ( $p=0.346$ ). Whereas in urine samples, 17.5% and 11.4% ( $p=0.458$ ) of patients and the control group, were positive for BKV, respectively. However, there was no statistically significant difference between the patients and controls. **Conclusion:** BKV reactivation occurs in 17.5% of patients with SLE during immunosuppression therapy. Therefore, more studies on BKV DNA by highly sensitive molecular assays in Patients with SLE seem to be necessary.

**Keywords:** BK Virus, Systemic Lupus Erythematosus, Renal Failure, Polymerase Chain Reaction

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## Potential Total Testing Process Vulnerabilities in Samples Submitted to Molecular Detection Laboratory of Covid-19

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**Background:** In the recent pandemic, the accurate identification of SARS-CoV-2 RNA in naso/oropharyngeal (NOP) swabs by RT-PCR is recommended for diagnosing the COVID-19. Our aim was to determine possible vulnerabilities on the total testing process. **Methods:** From 2020 November 15 to 2021 February 30, 3251 specimens of patients suspected Covid-19 were sent to the Covid-19 laboratory of School of Paramedical Sciences of Tehran University of Medical Sciences from 23 hospital and 8 health centers, were analysed in the specimen delivery room. **Results:** 84%, 7%, 6% and 1% collected specimens were nasopharyngeal, oro-nasopharyngeal, sputum and, nasopharyngeal in order. 27 of the specimens were contaminated with blood due to error in the collection of specimens. majority of hospitals used swabs with polystyrene shafts, however were used 23 swabs with wooden shafts and 2 swab with metal shafts. Moreover, most of the specimens were transferred to a laboratory by insulated cold box but 24 of them were not transferred at cold chain condition. 93% specimens had VTM/UTM, While 4% specimens without VTM and any transmitting medium and 3% specimens with normal saline. The VTM Also, the volume of 190 specimens were lower than 2 ml. **Conclusion:** So total testing process issues especially swabs sampling is a critical step, and especially in case of low viral load, might be a potential source of diagnostic errors.

**Keywords:** Covid-19, Molecular Detection Laboratory, Coronavirus, RT-PCR, Errors

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## Prevalence of hepatitis C in hemodialysis patients of Soodeh Medical Center from 1997 to the end of 1400

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**Background:** Hepatitis C virus transmission through dialysis units is increasing worldwide, ranging from 5% in Western countries to 70% in some developing countries. This disease leads to liver damage and hepatocellular carcinoma; And is one of the leading causes of death in patients undergoing hemodialysis. The aim of this study was to determine the prevalence of hepatitis C virus infection in hemodialysis patients of Soodeh Medical Center as the largest dialysis center in the country. **Materials and Methods:** In a descriptive cross-sectional study, 400 hemodialysis patients in the hemodialysis ward of Soodeh Medical Center were studied by census method. Sample information including name, sex, age were collected. Blood samples were tested for antibodies against hepatitis C virus (Anti HCVAb) C by third generation ELISA. Positive samples of hepatitis C after two tests (ELISA) were examined by PCR. **Results:** 12 patients were HCVAb positive (3%) and 6 patients (1.5%) were PCR positive for HCV. The mean age in HCV positive patients was 53.8 years. 99% of HCV positive patients were male and 1% were female. **Conclusion:** Due to the high prevalence of hepatitis C virus in hemodialysis patients, it is recommended Anti HCV test was performed before admission in hemodialysis units and measures such as careful control of services provided to these patients such as blood transfusions, separation of patients and trained personnel in the prevention of infection seem necessary. **References:** Paper: Prevalence of hepatitis B and C in hemodialysis patients in Rasht Prevalence and risk factors for hepatitis C in hemodialysis patients **Authors:** Judges of Seyedeh Ameneh Hepatitis C in hemodialysis patients Smaragdi Marinaki, John N Boletis, Stratigoula Sakellariou, and Ioanna K Delladetsima

**Keywords:** Hepatitis c, Hemodialysis



# New Findings in Laboratory Science (Young Scientists Session)

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## Dual Anticoagulant-Antioxidant Activities Properties of Exopolysaccharide from Marine Microalgae

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**Background:** There has been rapidly growing interest in developing new natural and safe multitarget bioactive compounds for the pharmaceutical and food industries. Marine organisms are promising sources with a wide variety of bioactivities including antioxidant and anticoagulant properties. Microalgae sulfated polysaccharides as a glycosaminoglycan containing sulfate ester in their various sugar units can be interesting candidates in this respect. **Methods:** The anticoagulant activities of the crude sulfated polysaccharides (sPS) extracted from *Chlorella sorokiniana*, isolated from the Persian Gulf, were evaluated in vitro. The effects of extracts on partial thromboplastin time (aPTT), and prothrombin time (PT) were investigated. The protective properties of sPS on toxic oxygen damage and antioxidant capacity were measured using the DPPH free radical scavenging and ABTS assays. **Results:** The sulfated polysaccharides extracted from *Chlorella sorokiniana* with high hydroxyl content and the sulfate/sugar ratio 0.98 showed a strong ABTS radical scavenging effect (more than 90% at concentration 1mg/mL) as well as anticoagulant activity, though lower than heparin. Therefore, to obtain the same effect as with heparin high concentration of sPS were required. The carbohydrate content in 1 gr of *C. sorokiniana* dry extract was equivalent to 0.7gr heparin sugars content. Also, 1 gr of *C. sorokiniana* dry extract contains sulfate as much as 0.3-gr heparin. **Conclusion:** The results indicated that the sulfated polysaccharides from Iranian habitat microalgae exhibit excellent potential antioxidant activity which could be used as health-improving functional food ingredients. On the other hand, due to the lower bleeding risks of the sulfated polysaccharides extracted of *Chlorella sorokiniana* compared to heparin, at the same therapeutic doses, they could be potential candidates for antithrombotic agents.

**Keywords:** Marine Microalgae, *Chlorella*, Sulfated Exopolysaccharide, Anticoagulant, Antioxidant

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### Prevalence of Hepatitis B in Patients Referred to Golestan Hospital in Ahvaz

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**Introduction:** Hepatitis is an inflammatory disease of the liver parenchyma that causes different metabolic and functional disorders on the liver and body. Numerous factors, including alcohol and some drugs, can cause this disease. The most important cause of hepatitis is the virus. One of the most important viruses is HBV, which despite effective vaccination against it, one million people worldwide die annually due to this disease. Its diagnostic method is HBSAg testing. The aim of this study was to evaluate the frequency of HBSAg in patients referred to Ahvaz Golestan Hospital in 2020-2022. **Method:** Data related to 13371 referred to Golestan Hospital in Ahvaz in 2020-2022, which were examined for HBSAg by ELISA, Aria Mabna kit and Autobioeliza Reader, were collected. Statistical analysis was performed by 20-spss software. **Conclusion:** Out of the total number of clients in this study, 112 (1.1%) were positive for HBSAg. Of these, 112 were 61 males and 51 were females. **Results and Discussion:** Considering the frequency of this disease from 2.8 to 8.7% in the whole country in 2020-2022 and the results obtained from this study in Ahvaz Golestan Hospital, we conclude that we have a low level of this disease. We are in the country and health measures and vaccinations have been carried out in this city in an acceptable manner.

**Keywords:** HBSAg, ELISA, Hepatitis B, Ahvaz

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## Green Synthesis of Silver Nanoparticles Using the Plant Extracts of *Vitex Agnus Castus L* and Evaluating Their Antibacterial Activity

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**Background:** These days, silver nanoparticles (Ag NPs) have been given considerable attention and applied in medical technology due to their great antimicrobial and antioxidant features. In the present study, we aimed to synthesize Ag NPs through reduction of silver nitrate in the presence of *Vitex agnus castus L* fruit extract. **Methods:** After collection of these fruits, their extract was prepared and added to Ag NO<sub>3</sub> to produce Ag NPs. The effect of different parameters like AgNO<sub>3</sub> concentration (0.5, 1, 3, and 5mM), sunlight exposure, and incubation time (10, 20, 30, and 40 min) were investigated in the synthesis of Ag NPs. Their features were characterized using UV-visible spectroscopy, scanning electron microscope (SEM), X-ray diffraction (XRD) analysis, and Dynamic Light Scattering analysis. Moreover, antimicrobial function of Ag NPs was evaluated using *Escherichia coli* and *Bacillus cereus* bacteria species and minimal inhibitory concentration (MIC) of Ag NPs against these two pathogens was measured. **Results:** The results showed that the synthesized nanoparticles had a spherical shape and the range size of 30-60 nm. For the first time, the antimicrobial activity of synthesized Ag NPs of *Vitex agnus castus L* fruit extract was shown. **Conclusion:** It can be stated that the biosynthesis of Ag NPs using fruit extract of this plant is an environmentally friendly, economic and harmless method without any use of poisonous substances and no side effects. These Ag NPs can be considered as suitable antibacterial agents and replacements for antibiotics.

**Keywords:** Silver Nanoparticles, *Vitex Agnus Castus L*, Antibacterial Properties, Green Synthesis, Aqueous Extract

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## In-vitro Effect of Carbapenem, Colistin, and Gentamicin Combination Against Carbapenem-Resistant and Biofilm-forming Enterobacteriaceae

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**Background:** The aim of the present study was to investigate in-vitro antibacterial effect of imipenem, gentamicin and colistin alone and the various combinations against carbapenem-resistant and biofilm-forming Enterobacteriaceae (CRE). **Methods:** Ten CRE isolates from wound were collected. The resistance to carbapenem were determined by the disk diffusion and determination of imipenem minimum inhibitory concentration (MIC) by the broth micro dilution according to the Clinical & Laboratory Standards Institute (CLSI) guideline. To study any inhibitory effect of antimicrobial agents on biofilm, the minimum biofilm inhibitory concentration (MBIC) was determined. The synergetic effect of the antibiotics combinations was studied using the checkerboard assay and the fractional inhibitory concentration (FIC). **Results:** The highest synergetic effect against planktonic form was observed in imipenem/gentamicin (9 of 10 isolates), and the lowest synergetic effect was found in gentamicin/colistin and colistin/gentamicin (3 of 10 isolates). Colistin/imipenem were shown synergetic effect for 6 isolates. The highest synergetic effect against biofilm was observed in colistin/imipenem (6 of 10 isolates) followed by imipenem/gentamicin (4 of 10 isolates) and colistin/gentamicin (3 of 10 isolates). **Conclusion:** The combination of antimicrobial agents had shown the different effects on biofilm and planktonic forms of CRE. Therefore, a separate determination of inhibitory effects of the antibiotic in the combination is necessary for biofilm forms. Gentamicin/imipenem and was more effective against planktonic and colistin/imipenem against biofilm forms of CRE.

**Keywords:** Carbapenem Resistance, Biofilm, Enterobacteriaceae, Synergetic Effect

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## Carbapenems resistant Enterobacteriaceae isolated from wound infections from Tabriz, Iran

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**Background:** The frequency, antibiotic susceptibility pattern and resistance mechanisms of carbapenems resistant Enterobacteriaceae from wound infections were investigated. **Methods:** Forty-one Enterobacteriaceae isolated was obtained from wound infections. The disk diffusion method and minimum inhibitory concentration (MIC) determination were used for testing antibiotics susceptibility. AmpC and efflux pump hyperexpression and carbapenemase production were determined by phenotypic the phenotypic methods. The carbapenemase genes also were detected by the PCR. **Results:** According to the MIC and disk diffusion results, a high level of resistance were found to all group of antimicrobial agents except colistin and amikacin. No efflux pump activity was observed. Among 41 carbapenem resistant Enterobacteriaceae, *Klebsiella pneumoniae* was most common (36/41; 87.8%%) followed by *Escherichia coli* (4/41: 9.75%), and *Enterobacter spp* (1/41; 2.4%). AmpC mediated carbapenem resistance was observed in *Enterobacter spp*. According to the phenotypic test and PCR results, the resistance to carbapenem in *E. coli* and *K. pneumoniae* was observed to be associated with carbapenems production. The most common carbapenemase gene was blaOXA-48-like (56.09%) followed by blaKPC (19.5%) blaNDM (17.07%), and blaVIM (7.31%). **Conclusions:** According to our finding the frequency of carbapenems resistant Enterobacteriaceae is at an alarming level. The most common mechanism of carbapenems resistance among Enterobacteriaceae was carbapenems production. Therefore, we suggest reconsideration in the management program of wound infections by Enterobacteriaceae in our setting. The colistin and amikacin can be considered as a very applicable drug for the treatment of infections caused by the carbapenems resistant Enterobacteriaceae.

**Keywords:** Wound Infections, Carbapenems, Epidemiology, Enterobacteriaceae

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### **In-Vitro Activity of Antimicrobial Agents Combinations Against Carbapenem Non-Susceptible *Pseudomonas Aeruginosa***

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**Aim:** The present study was performed to investigate in-vitro effects of antibiotics combination against planktonic and biofilm forms of carbapenem-resistant *Pseudomonas aeruginosa* (CRPA). **Methods:** Eight CRPA isolates were collected from wound infections. The resistance to meropenem were detected by the phenotypic, PCR, and Real-Time PCR methods. The antimicrobial effect of antimicrobial agents was determined by the broth micro dilution according to the CLSI guideline. To study the inhibitory against the biofilm, the minimum biofilm inhibitory concentration (MBIC) was determined. To determine synergetic effects of the drugs combinations, the checkerboard assay was used for fractional inhibitory concentration (FIC) determination. **Results:** The highest synergic interaction was observed in colistin/fosfomycin and gentamicin/fosfomycin (5 of 8 isolates), and the lowest synergic interaction was observed in gentamicin/imipenem and colistin/gentamicin (1 of 8 isolates). Colistin/fosfomycin, imipenem/fosfomycin, colistin/imipenem, gentamicin/fosfomycin, and gentamicin/imipenem were shown synergic effect for 3, 2, 2, 2 and 1 isolates, respectively. **Conclusion:** Fosfomycin/colistin and fosfomycin/gentamicin were more effective against planktonic form and fosfomycin/colistin against biofilm forms. The combination of antimicrobial agents had unlike activity on the biofilm and planktonic forms of CRPA. Thus, a distinct testing of inhibitory activity of the antimicrobial agent's combination is required.

**Keywords:** Biofilm, Antimicrobial Combination, Carbapenem-Resistant, *Pseudomonas Aeruginosa*, Wound

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## Colistin Susceptibility of Multi- Drug Resistance (MDR) and Carbapenem Resistant Enterobacteriaceae Isolated from Wound Infections, Tabriz, Iran

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**Aim:** The aim of the present study was to assess colistin susceptibility in multi- drug resistance (MDR) and Carbapenem resistant isolates Enterobacteriaceae collected from wound infections. **Methods:** One hundred eighteen clinical isolates of Enterobacteriaceae were collected from wound infection during 2020-2021. The isolates were identified by standard microbiology methods. The disk diffusion method was performed according to the Clinical and Laboratory Standards Institute (CLSI) guidelines. MDR was defined as acquired resistance to at least one agent in three or more antimicrobial categories. The MIC (Minimum Inhibitory Concentration) of colistin was determined by the micro-broth dilution and colistin broth disk elusion methods according to CLSI guidelines. **Results:** Among the 118 Enterobacteriaceae, 98 isolates (83.05%) were MDR and 41 isolates (34.7%) were resistant to carbapenem antibiotics. All carbapenem resistant isolates were MDR. Fifty-seven *Klebsiella pneumoniae* isolates, 28 *Escherichia coli* isolates, 9 *Enterobacter* spp. and 4 *Citrobacter* spp. were distinguished from these 98 isolated. The colistin MIC range was 0.25 to 8 µg/mL. Two *K. pneumoniae* isolates (2.04%) were resistant to colistin antibiotic, one isolate with MIC of 8 µg/mL and one isolate with MIC of 4 µg/mL. MIC<sub>50</sub> and MIC<sub>90</sub> were 0.5 and 1 µg/mL, respectively. The results of MIC obtained by micro-broth dilution and colistin broth disk elusion methods were consistent in 96 isolates. **Conclusion:** Colistin may be an alternative antimicrobial agent for infections due to MDR and Carbapenem resistant Enterobacteriaceae. However, Colistin susceptibility should be studied before its usages in antimicrobial therapy. The MIC of colistin can be determined by the both micro-broth dilution and colistin broth disk elusion methods.

**Keywords:** Carbapenem, Colistin, Enterobacteriaceae, Multi- Drug Resistance

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## Evaluation of Glucantime Activity in Cutaneous Leishmaniasis Lesions Contaminated with Secondary Bacterial Infection Compared with Non Infected Lesions

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**Background:** Glucantime is regarded as the first line in the treatment of cutaneous leishmaniasis, but failure to treatment is a problem in many cases. Comparison of glucantime activity in the cutaneous leishmaniasis complicated with secondary bacterial infection compared with uncomplicated lesions. **Materials & Methods:** This experimental clinical trial Patients enrolled in the study had cutaneous leishmaniasis. The lesions were scraped with sterile swap and cultured into the blood agar medium. All the patients were treated with glucantime for 3 weeks and followed for 3 months. Response to treatment was defined as loss of infiltration, re epithelization and negative smear, and unresponsiveness defined as unchanging the size of the lesions without re epithelization and positive smear at the end of the study. Regarding to results of cultures, the lesions were divided into two groups with positive and negative cultures and glucantime activity was compared between two groups. **Results:** Out of 105 patients enrolled in the study, 79 (75.2%) were Negative and 26 patients (24.8%) were Positive for secondary bacterial infection. In groups with negative bacterial culture response to treatment was 70.6 % (56 Patients) and in the other group was 29.4% (23 patients) and in groups with positive bacterial culture response to treatment was 40 % (10 Patients) and in the other group was 60% (15 patients). (P<0.0001) **Conclusion:** Activity of glucantime decreased in cutaneous leishmaniasis lesions with secondary bacterial infection.

**Keywords:** Bacterial Infection, Glucantime, Cutaneous Leishmaniasis

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## Effects of Newly Synthesized Oxadiazoles on *Candida Albicans* Pathogenic Gene Expression and Evaluation of Their Cell Cytotoxicity

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**Background:** *Candida albicans* is the main human pathogenic fungus. In recent years, drug resistance to candidiasis infections has been increased, thus novel drugs are needed. 1, 3 and 4-oxadiazoles derivatives can be a good alternative candidate against these agents. The aim of this study was to synthesize new oxadiazole derivatives for the first time and to study its effect on the *Candida albicans* samples, the expression of the pathogenic CDR1 gene and also on the K562 cell line. **Methods:** New derivatives of oxadiazoles were synthesized by single-step reaction of N-iso-cyano-imino-3- phenyl-phosphorus, carboxylic acid and 2-pyridine carbaldehyde in acetonitrile. Spectral information of the compounds was obtained using IR (Infrared), C-NMR (Carbon-13 NMR) and H-NMR (hydrogen-1 NMR). Several methods such as, Agar diffusion test, MIC and MFC was used to evaluate the resistance of candida to new synthesized compounds. To check the expression of CDR, Real Time PCR method was performed and MTT method was used to evaluate the toxicity. **Results:** Results showed synthetic compounds containing methoxyphenyl group with a concentration of 5 mg/ml have antifungal properties against *Candida albicans*. Furthermore, these compounds contain proper anti-cancer effects at a concentration of 1.2 mg/ml. No results were obtained regarding to the effect of compounds on CDR1 gene expression. **Conclusion:** In this study, synthetic materials possess cell cytotoxicity in 50% of K562 cell line at a concentration of 1.2 mg/ml, which indicates the anti-cancer ability.

**Keywords:** *Candida Albicans*, Drug Resistance, New Oxadiazole Derivatives

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## Survey of Bacteria Resistant to Multidrug ESBLs Produce in Isolated from Pulmonary Infection in ICU Qazvin Shahid Rajaei Hospital in 2021

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**Introduction:** Nosocomial infection are one of the most important health problems in admission patients in ICU. by extended-spectrum beta lactamase (ESBL)-producing bacteria have become a growing problem worldwide. pneumonia is the most common nosocomial infections in ICUs. Nosocomial infection caused by extended-spectrum beta-lactamase (ESBL)-producing bacteria have become a growing problem worldwide. **Methods:** This study is during 12 months (2021), the 219 samples of trachea tube or pulmonary fluids of patients. Samples were transported into TSB medium, and incubated in for 24 hours. Then subculture on Blood agar, chocolate agar, EMB. After 24h growth. All isolates were identified by routine biochemical methods and antimicrobial susceptibility testing carried out by Kirby-Bauer method. Confirmatory test for production of ESBLs was performed by the combination disk tests. The results were interpreted according to the recommendation of CLSI. **Results:** of the 219 sample, 162(73.9%) gram negative bacilli isolated. ESBL was detected in 6 (3.7%) of isolates. microorganisms ESBL positive in order of frequency included klebsiellasp3 (50%), Pseudomonas aeruginosa2(33.3%), Acinetobacter1 (16.6%), Enterobactersp1 (16.6%). **Conclusion:** The present study shows high broad-spectrum beta-lactamase production in bacterial strains isolated from patients with from pulmonary Infection In patients admitted in ICU. Emergence of multi-drug resistant strains that are resistant to most antibiotic classes is a major public health problem in Iran. To resolve this problem using of practical guidelines is critical. To overcome this problem, it need to develop new antimicrobial agents, limiting the unnecessary use of antimicrobial and increasing compliance with infection control issue.

**Keywords:** Resistant to Multidrug, Respiratory Infection, ICU

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## Molecular Study of *adeIJK* Efflux Pump Genes in *Acinetobacter Baumannii* Isolated from Qom by PCR Method

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**Introduction:** *Acinetobacter baumannii* is widely distributed in nature. The low need of this bacterium for food and its ability to use different sources of carbon, has increased its presence in different parts of the hospital and the incidence of nosocomial infections. Biofilm formation is one of the main virulence factors of this bacterium and has increased the resistance of the bacterium to antimicrobial agents, especially antibiotics. One of the important factors related to biofilm in *Acinetobacter baumannii* is the presence of effusion pumps, especially in the RND family, which confirms the presence of the *adeIJK* gene in the bacterium. **Method:** A total of 108 clinical isolates were prepared from patients isolated in different wards of Qom hospitals from 2012 to 2013. Differential and diagnostic tests were performed to confirm the isolates. Then, the confirmed isolates were examined for the presence of *adeIJK* gene by PCR by designed primers. **Results:** The molecular study of the presence of *adeIJK* genes in *Acinetobacter baumannii* in the present study shows that on average 90% of the samples have the mentioned genes. Separately, the presence of *adeI* gene is equal to 85.1%, *adeJ* gene is equal to 87.9% and *adeK* gene is equal to 98.1%. **Discussion & Conclusion:** The results show that this amount of gene is present.

**Keywords:** *Acinetobacter Baumannii*, Efflux Pump, *Adeiijk* Gene, Antibiotic Resistance

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**Evaluation of Metallo-Beta-Lactamase in Imipenem-Resistant *Pseudomonas Aeruginosa* Isolated from Clinical Specimens of Razi Hospital in Tehran by Combined Disk Diffusion (CDDT)**

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Background: *Pseudomonas aeruginosa* is an opportunistic human pathogen that causes serious challenges, especially in patients who are immunocompromised. Recently, increasing resistance to beta-lactam antibiotics among metallo-beta-lactamase producing bacteria led to several complications in the treatment of infections caused by these bacteria. The aim of this study was to determine the prevalence rate of imipenem resistant *Pseudomonas aeruginosa* carrying metallo- $\beta$ -lactamases (MBLs). Methods: 138 *Pseudomonas aeruginosa* were isolated from two hospitals in Tehran in 2016. These strains which were isolated from different clinical specimens such as: urinary, wound, tracheal samples and etc., were identified using microscopic and biochemical tests such as, catalase, oxidase, growth on Triple Sugar Iron (TSI) agar medium, the reaction in the oxidative fermentative (OF) medium, growth on the cefrimide agar and the ability to grow at 42 °C. Then, the susceptibility antibiotic test was performed by 10 different antibiotics using the disk diffusion method. IMP-EDTA combination disk phenotypic test was also performed based on the CLSI guidelines for detection of MBL producing strains that were resistant to imipenem. Results: The resistance pattern in 236 *Pseudomonas aeruginosa* isolates was: Imipenem (44.9%), meropenem (44.2%), ceftazidime (36.2%), carbenicillin (29.7%), tobramycin (33.3%), amikacin (44.9%), ticarcillin (32.6%), gentamicin (34.05%), cefotaxime (7.2%), ceftizoxime (4.3%). Furthermore, results also showed out of 138 *Pseudomonas aeruginosa* strains, 100 strains (72.4%) were resistant to imipenem which 79.7% were MBLs producing by IMP-EDTA combined disk method. Conclusion: This study displayed an increase in *Pseudomonas aeruginosa* antibiotic resistance due to the production of metallo-beta-lactamase enzyme. Therefore, due to the clinical importance of these resistant strains in under study hospitals, it is necessary to quickly identify the organisms that produce these enzymes and use appropriate infection control tools to prevent further spread of these organisms.

**Keywords:** *Pseudomonas Aeruginosa*, Metallo-B-Lactamases, IMP-EDTA Combination Disk Phenotypic Test

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## Evaluation of the emergence of *Acinetobacter baumannii* isolates containing VIM and SIM and drug resistant MDR genes isolated from surfaces and equipment of Tehran medical centers by PCR

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**Background and Aim:** Excessive use of antimicrobials in hospital and community is one of the most important factors in the development of new resistance of bacteria to antimicrobials. The aim of this study was to determine the frequency of drug-resistant vim and sim genes in *Acinetobacter baumannii* (MDR) isolates isolated from surfaces and equipment using PCR. **Methods:** In this study, 120 samples were collected from the desired levels and sent to the microbiology department of a specialized laboratory in West Tehran. Using culture methods and biochemical tests of strains, *Acinetobacter baumannii* was identified and to determine the degree of antibiotic resistance, disk diffusion and microdialysis broth methods were used according to clsi guidelines. Isolates were examined for the presence of vim and sim genes by molecular PCR. **Results:** The results showed that out of 120 samples, (40) (33.3%) were *Acinetobacter baumannii* (100%) were sensitive to cholestin. Resistance of isolated samples; 40 isolates (100%) to imipenem, meropenem and lincomycin, 39 isolates (97.5%) to ceftazidime, 39 isolates (97.5%) to ciprofloxacin, 36 isolates (90%) to ceftozoxime and oxacillin, 34 isolates (85 %) To gentamicin, 28 isolates (70%) to ampicillin and tetracycline and 27 isolates (67.5%) to cefotaxime and cefxime. MIC calculation for imipenem and meropenem antibiotics was reported in 24 isolates (60%) at 64 ug ug / ml **Conclusion:** Considering the identification of vim and sim genes from *Acinetobacter baumannii* isolates, it is recommended that extensive studies be performed on the patterns of bacterial resistance to antimicrobial agents in different parts of the world, including Iran.

**Keywords:** *Acinetobacter Baumannii*, Beta-Lactamase Genes, Drug Resistance

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## The Frequency of Multidrug- Resistant *Acinetobacter Baumannii* Isolated from Hospitalized Patients with Ventilator Associated Pneumonia in the ICU of Masih Daneshvari Hospital

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**Background:** *Acinetobacter baumannii* is one of the most common causes of ventilator associated pneumonia (VAP) in patients hospitalized in ICU section. Multiple drug resistance leads to excessive use of Colistin antibiotic, which is the latest treatment option for this bacterium. Therefore, the aim of this study was to determine the frequency of multiple resistance among *Acinetobacter baumannii* isolated from patients with VAP and hospitalized in the ICU of Masih Daneshvari hospitals. **Methods:** In this study, 200 isolates of *Acinetobacter baumannii* associated with VAP were collected from ICU section of “Masih Daneshvari” Hospital (during 1397-1391). Samples were bronchoalveolar lavage and tracheal aspiration. Antibacterial susceptibility of isolates for colistin was determined by MIC method and other antibiotics examined by disk diffusion method, according to the CLSI criteria. Multidrug resistance (MDR) and extended –drug resistance (XDR) isolates were determined according to standard definitions of the CLSI. **Results:** All the isolates were susceptible to colistin. However, they were all resistant to piperacillin, piperacillin-tazobactam, ceftazidime, cefotaxime, ceftriaxone, amikacin, gentamycin, levofloxacin, co-trimoxazole, Ciprofloxacin. Antimicrobial resistance for tetracycline and ampicillin sulbactam was 8.5% and 20% respectively. All isolates were MDR, XDR and they were also susceptible to colistin (MIC<sub>50</sub>=1 and MIC<sub>90</sub>=2 µg/ml). **Conclusion:** In this study, *A. baumannii* isolates collected from VAP patients were MDR and XDR. Although all isolates susceptible to colistin and it seems to be the most appropriate antibiotics for VAP treatment, but also colistin resistance can become endemic in the world.

**Keywords:** *Acinetobacter Baumannii*, Ventilator Associated Pneumonia, Multidrug Resistance (MDR), Extended-Drug Resistance (XD)



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