Indian Journal of Clinical Biochemistry

Official Journal of the Association of Clinical Biochemists of India

Aims and Scope

The primary mission of the journal is to promote improvement in the health and well being of community through the development and practice of clinical biochemistry and dissemination of knowledge and recent advances in this discipline among professionals, diagnostics industry, government and non-government organizations. Indian Journal of Clinical Biochemistry (IJCB) publishes peer reviewed articles that contribute to the existing knowledge in all fields of Clinical biochemistry, either experimental or theoretical, particularly deal with the applications of biochemistry, molecular biology, genetics, biotechnology, and immunology to the diagnosis, treatment, monitoring and prevention of human diseases. The articles published also include those covering the analytical and molecular diagnostic techniques, instrumentation, data processing, quality assurance and accreditation aspects of the clinical investigations in which chemistry has played a major role, or laboratory animal studies with biochemical and clinical relevance.

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www.springer.com/12291
Electronic edition: link.springer.com/journal/12291

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Indian Journal of Clinical Biochemistry is quarterly (January, April, July and October) publication of Association of Clinical Biochemists of India (ACBI). Subscriptions, address changes and claims should be sent to Springer Nature Customer Service Center: customerservice@springernature.com, Springer Nature Customer Service Center GmbH, Haberstraße 7, 69126 Heidelberg, Germany or Springer Nature Journal Fulfillment, 233 Spring Street, New York, NY 10013-1578, USA

ISSN: 0970-1915 print
ISSN: 0974-0422 electronic

Subscription Price

Subscription Rates: For India Rs. 6800 + 150 (Postage) and US$ 305 (International).

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Office of Publication

Springer (India) Private Ltd.,
7th Floor, Vijaya Building,
17, Barakhamba Road,
New Delhi 110 001, India

Publisher

Association of Clinical Biochemists of India (ACBI), Biochem Lab, East Boring Canal Road, Patna-800001 (India), E-mail: kpsacbi@yahoo.co.in, Phone & Fax: +91- 612-2531212, Website: www.acbindia.org

Distributor

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ISSN 0970-1915

IJCB

Volume 34 • Issue Supp. 1 • November 2019

Indian Journal of Clinical Biochemistry

12291

Official Journal of the Association of Clinical Biochemists of India

Proceedings: 15th Asia-Pacific Federation For Clinical Biochemistry and Laboratory Medicine (APFCB) Congress 2019 from 17-20 November, 2019, Jaipur, India

PL001

Ammonia in Acute Liver Failure: It's influence on pathogenesis, Prognosis and Management

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ALF is defined by : (1) rapid development of hepatocellular dysfunction(jaundice, coaugulopathy) (2) encephalopathy, (3) absence of a prior history of liver disease .Raised arterial ammonia is implicated in pathogenesis of encephalopathy and consequent complications .

In animal models and in ALF patients swelling of astrocytes and alteration in astrocytic proteins have been reported . During ALF, the ability of liver to metabolise ammonia is compromised causing hyperammonemic neurotoxicity which includes: astrocyte volume dysregulation, cerebral energy homeostasis, alteration in neuronal protein expression, with oxidative and nitrosative stress.

Arterial ammonia measured within 24 hours of patient presentation has been asssociated with patient outcome in ALF unlike in cirrhosis . We reported that non-survivors had significantly higher ammonia levels than survivors (174.7 v 105.0 mmol/l; p,0.001). Arterial ammonia level > 124 mmol/l could predict mortality . Patients with higher ammonia levels developed more complications. However, ALF is a dynamic process where variables determining prognosis at admission change over time. Therefore dynamic changes in arterial ammonia and its influence on prognosis was assessed with a stastical model (ALF Early dynamic model-ALF-ED). 380 ALF (derivation cohort n=244,validation cohort n=136) participated in a prospective study. ALFED model had four variables: arterial ammonia, serum bilirubin, INR and encephalopathy >grade II. The model was validated. ALFED had excellent discrimination with an AUROC of 0.91 and 0.92 in the derivation and validation cohort respectively. ALF-ED was well calibrated in both cohorts and showed increase in mortality with increasing risk scores from 0 to 6. ALFED model was superior to KCH and MELD score. An ALFED score of >4 had 85% and 87% PPV and NPV in the validation cohort.

Decrease in arterial ammonia and absence of it was associated with improved and decreased survival respectively. Arterial ammonia lowering therapy is the future promise in treatment of ALF.

PL002

Expanding Space for Next Generation Sequencing Diagnostics Applications

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Vita-Salute San Raffaele University, Italy

The recent advances in the genomic field and the development of new technologies for DNA testing started the revolution of the diagnostic laboratory. For the diagnosis DNA-based diagnostics provide a sensitive alternative to protein-based diagnostics and the mutation detection is one of the most important areas of molecular diagnostics today.

Advances in DNA analysis to develop methods, which are increasingly specific, sensitive, fast, simple, automatable, and cost-effective, are considered paramount. These demands are currently driving the rapid evolution of a diverse range of newer technologies. Researchers have discovered hundreds of genes that harbour variations contributing to human illness, identified genetic variability in patients' responses to dozens of treatments, and begun to target the molecular causes of some diseases. In addition, scientists are developing and using diagnostic tests based on genetics or other molecular mechanisms to better predict patients' responses to targeted therapy.

For the future of genomics is demanding the rapid evolution of high-throughput genotyping technologies (next generation sequencing) toward increased speed and reduced cost. The speed, accuracy, efficiency, and cost-effectiveness of DNA sequencing have been improving continuously since the initial derivation of the technique. With the advent of massively parallel sequencing technologies, DNA sequencing costs have been dramatically reduced. The recent introduction of instruments capable of producing millions of DNA sequence reads in a single run is rapidly changing the landscape of genetic diagnostics, providing the ability to answer questions with heretofore unimaginable speed.

PL003

Alzheimer's a Cerebrovascular Disease?

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Changes in the brain pathology and functions, related to Alzheimer's disease (AD), begin years before the symptoms appear. Numerous studies reported a relationship between genes and single nucleotide polymorphisms (SNPs) and the risk for AD. This includes, among others, ApoE ε4 allele, rs688 (LDLR and LRP-apoE receptor), and rs17571 (lysosomal protease cathepsinD), proteins that are involved in lipid transport and lysosomal degradation. Many studies have implicated altered lipid metabolism in Alzheimer's and other forms of dementia, not surprising considering sphingolipids and cholesterol are major components of myelin. Our long term interest has been on cardiovascular diseases, particularly on atherosclerosis. We use mice models for atherosclerosis research. We recently observed that ApoE-Paraoxonase 1 (PON1) double knockout (DKO) mice over 18 months of age developed increased and severe calcified atherosclerosis of the arteries that supply blood to the brain, even on normal diet. Furthermore, these mice suffered from seizures and other neurological symptoms, such as paw and neck edema and gait imbalance. Besides, the gene expression and amount of the calcium binding S100 protein were increased in these mice while that of LRP (a receptor for apoE) was suppressed. Blood-brain barrier function was compromised in the older mice as compared to younger mice. Based on these, we propose the hypothesis that combined deficiencies in ApoE and in antioxidant defense (exasperated by the lack of PON 1 and ageing), together with the development of carotid atherosclerosis, would promote vascular dementia and particularly would mimic human Alzheimer's pathology. We further propose, alterations in LRP, a protein that recognizes apoE might be a key factor in defective remyelination and Alzheimer pathology.

PL004

The Opioid Crisis in North America

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The Opioid Crisis in North America has been termed an epidemic by some and a public health crisis by others. It is a significant medical, social and political problem. In 2017, more than 47,000 Americans died as a result of an opioid overdose, including prescription opioids, heroin, and illicitly manufactured fentanyl and other synthetic opioids. More than 10,300 Canadians died as a result of an apparent opioid-related overdose between January 2016 and September 2018, and over 600 individuals died in the province of Alberta, Canada. The cost to healthcare systems is in the millions of dollars, and has prompted emotional responses from the public and health care providers, leading both the Federal government and provincial governments in Canada to take action. The causes of this event are multiple and controversial, including the liberal opioid prescribing behaviors of physicians, prompted at least in part by unfounded claims from pharmaceutical companies, and the lack of and disparity among drug treatment programs. Others cite the lack of social support programs for the poor and persons at risk, and the biased, punishment based treatment of persons with addictions, while others blame the lack of respect for laws and lack of consequences in today's society as fueling the problem. Significant contributors are certainly the sophistication of illegal "chemists", the availability of legal internationally imported of drugs, and the ease of online drug purchase and delivery. The laboratory has traditionally played a role in the investigation and management of overdose patients, as well as in supporting patients in drug treatment programs. Changes in clinical management of these patients are changing the role of the laboratory. The presentation will discuss the purported causes of the opioid crises and some of the strategies that have been employed to address the problem. The role of the laboratory in supporting medical and drug treatment programs will also be discussed.

S001

Clinical Proteomics: Promise and Pitfalls

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Although the term proteomics was first coined in 1994, laboratory medicine has its foundation in the measurement of proteins in order to help in the diagnosis of disease. Tests for proteinuria in cases of suspected renal disease date back to the early 19th Century. Currently, measurement of proteins and peptides in human plasma represent a significant portion of testing performed by medical laboratories. Analytical and technological innovations in the past 20 years have greatly expanded the number of proteins which can reliably be measured and considered as potential diagnostic markers. Although estimates vary widely by study human plasma likely contains > 10,000 different proteins with approximately 1,500 identified so far. These span a wide range of concentrations (>10 logs), from ca 50 g/L (e.g. albumin) to <50 ng/L (e.g. parathyrin). While the genetic code for humans has been largely completed through the human genome project, proteomics will be a much longer, more intensive, and time-consuming study. Discovery techniques used in proteomics are generally capable of multi-analyte profiling and include two-dimensional gel electrophoresis, multiplex immunoassay panels, protein microarrays, and MALDI-TOF tandem mass spectrometry. With use of high-throughput technologies, and large volumes of proteomics data generated, bioinformatics is an essential component of proteomics. Several proteomic panels (often resulting in a single reportable result or 'score' from multivariate analysis of concentration patterns in plasma) have been suggested for diagnosis and risk stratification in numerous conditions. Clinical examples will highlight advances made recently in areas of liver disease (fibrosis in nonalcoholic fatty liver disease), oncology, and multifactorial conditions such as autism spectrum disorders. However, reference intervals and inter-individual variations for new protein markers have yet to be established and, although markers discovered through proteomics may have clinical validity, their clinical utility has yet to be verified.

S002

Clinical Proteomics in the Trenches: Practical Aspects of Targeted Mass Spec in Clinical Labs

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Clinical Proteomics through targeted mass spectrometry (MS) approaches for peptide and protein analysis are quickly becoming an essential part of laboratory medicine due to their tremendous potential in clinical application and clinical proteomics. Adoption of targeted MS to study clinical questions is well underway as these assays provide higher specificity, sensitivity, short development time, and allows for multiplexing analytes as compared to conventional platforms, further providing potential savings in a large volume environment.

However, establishing and validating the performance of these tests to make informed clinical and public health decisions pose significant clinical and scientific challenges. Most clinical laboratory are not familiar with the mass spectrometry platform. For those who have had experience in this area, they have been focusing on small molecular testing. Protein or peptide based targeted mass spectrometry assays requires different workflow and operational process. These skills are not readily available to the current laboratories. This presentation will discuss these challenges, the pitfalls and recommendations to implement the targeted mass spectrometry assays in a routine clinical laboratory to provide value to healthcare system and patient services.

S003

Cardiovascular Precision Medicine: The Transformative Impact of Metabolomics/ Proteomics

Sridevi Devaraj

Texas Children's Hospital; Baylor College of Medicine, USA

Atherosclerotic cardiovascular disease (ASCVD) is a leading cause of morbidity and mortality worldwide. Over the last several decades, our understanding of the risk factors and ways to manage modifiable risks through lifestyle changes or therapeutic interventions has significantly improved the mortality rate. Furthermore, evidence-based clinical practice guidelines surrounding the management of ASCVD have provided a framework for clinical decisions and supporting best practices. In November 2018, the American College of Cardiology (ACC) and American Heart Association (AHA), along with many additional collaborating organizations, published new practice guidelines for

managing blood cholesterol. This symposium will explore how the guidelines can be incorporated into the current testing modalities for cardiovascular risk and prevention. The role of precision medicine in the use of non-fasting lipids, new calculations of LDL-C, and recommendations on assessing risk-enhancing factors in certain patient populations to help clinicians decide on statin and non-statin therapy will be discussed. The new guidelines incorporate non-traditional risk factors such as chronic inflammatory conditions, chronic kidney disease, metabolic syndrome, preeclampsia, and persistently elevated LDL-C, among others. How precision medicine can target a complex multifactorial disease such as atherothrombosis, and how precision medicine offers a new strategy for the care and management of patients that takes into account individual differences in genetics, environment, and lifestyle will be reviewed. Finally, the laboratory can play an integral role in delivering individualized risk assessment and treatment options. The utility of metabolomics and proteomics in population health and cardiovascular risk and utility of an advanced EMR system that can combine these elements and help clinicians not only in diagnosis but also in making decisions on lifestyle coaching and therapeutic intervention.

S004

Innovations in Real-time Diabetes Biomarker Tracking

Sunil Sethi

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Diabetes is the most significant healthcare issue in terms of resource requirements. Diabetes is becoming more common and the overall global prevalence of diabetes in adults is 8.5% of total population. Diabetes is a major cause of blindness, renal failure, heart disease, stroke and non-traumatic limb amputation. Healthcare providers in many third-world countries simply do not have the resources to adequately manage the multitude of complications. Because of the large numbers of patients, it is estimated that doctor-patient interactions are limited to less than 5 minutes per outpatient visit. In this short period, numerous biomarkers need to be assessed for adequacy of control. It is imperative that healthcare professionals utilize newer technologies to diagnose and manage such patients. This presentation will cover prevalence of diabetes in Asia and the solutions for laboratory management of the chronic disorder currently and potentially available in the city state of Singapore.

S005

Role of Laboratory in Management of Diabetes

Leslie Charles Lai

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Type 2 diabetes (T2D) is increasing in prevalence worldwide at an alarming rate. People with diabetes have a two to four-fold risk of acute myocardial infarction and stroke: Diabetes is the commonest cause of blindness, chronic kidney disease Stage 5 and amputations in the world. Hence, it is crucial for people with diabetes to have good glucose control and optimal control of other cardiovascular risk factors. HbA1c is now used for both the diagnosis of diabetes and in assessing glycaemic control. On 21st June 2018, the FDA cleared the Afinion HbA1c Dx assay as the first ever point of care HbA1c assay for the diagnosis of diabetes and assessment of patients' risk of developing diabetes. Since HbA1c reflects glucose control over the past two to three months it should not be used to diagnose type 1 diabetes (T1D) or gestational diabetes where diabetes can develop rapidly over a short period of time. It is important to ensure that lipid levels are optimised to reduce cardiovascular risk and to monitor patients' renal function (serum creatinine, eGFR calculated using the CKD-EPI formula and urine albumin to creatinine ratio on the first void urine in the morning). Insulin and/or C-peptide measurements may be useful in ascertaining the body's ability to produce insulin and can help differentiate between T1D and T2D although insulin and C-peptide levels may be low in the presence of glucose toxicity in patients with T2D and should be measured when glucose control has been good for several weeks. Islet Cell Cytoplasmic Autoantibodies (ICA); Insulin Autoantibodies (IAA); Glutamic Acid Decarboxylase-65 Autoantibodies (GAD65); Insulinoma-Associated-2 Autoantibodies (IA-2A); Zinc Transporter-8 Autoantibodies (ZnT8A) may be present in T1D. T1D may be part of a pluriglandular autoimmune syndrome that includes hypothyroidism/ hyperthyroidism, Addison's disease and/or vitiligo.

S006

Diabetes Testing and Reporting Practices in the Asia Pacific Region

Sam Vasikaran

Royal Perth Hospital, Perth, Australia

Diabetes mellitus is a disease of carbohydrate metabolism, due to absolute or relative insulin deficiency. Type 2 diabetes is by far the most common form of the disease. Nearly one in ten adults worldwide now has diabetes mellitus. Asia is a major focus of the rapidly emerging T2DM global epidemic. Undiagnosed diabetes accounts for about half the cases. Diabetes mellitus is a

major cause of blindness, kidney failure, heart attacks, stroke and lower limb amputation. The clinical laboratory has a major role in the diagnosis and management of the disease. The role of the laboratory may be broken down into the following steps: Screening, diagnosis, monitoring glycaemic control, assessment of risk factors and detection and management of chronic complications and acute metabolic complications. In addition to offering appropriate tests and ensuring the results are accurate and precise, the laboratory has a major role in education clinicians and patients in choosing the right tests for the purpose and the correct interpretation of results in ensuring the optimum and timely care of patients. The laboratory profession has a responsibility in offering patients appropriate and timely access to tests. The profession has an important role in quality assurance of the tests that they offer as well as the interpretation of the results. Results of surveys of laboratory testing and reporting practices in the Asia Pacific Region will be presented and discussed.

S007

Trends in Lab Testing in Diabetes: An Indian Perspective

Mithu Banerjee

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India is the diabetes capital of the world. The number of diabetics in India is approximately 51 million. The diagnosis & management of diabetes is dependant to a very large extent on laboratory parameters. Hence, it is imperative that trends in lab testing be studied so that policy formulations can be undertaken which will ensure correct diagnosis and management of diabetes universally.

A survey of 890 practising biochemists/pathologists was conducted through Survey Monkey between July to October 2018 to examine lab testing practices for diabetes.

A total of 313 (35%) responded. Of these, 39%, 25%, 22%, 11%, 2%, and 1% participants worked in academic institution, public hospital laboratory, private hospital laboratory, private stand-alone laboratory, research laboratory and public stand-alone laboratory respectively. HbA1c was approved for diagnosis in 82% of laboratories but not in 18% of laboratories/hospitals. HbA1c method was NGSP (National Glycohaemoglobin Standardisation Programme) certified in 70% of laboratories but not certified in 30%. OGTT was recommended for diagnosis of gestational diabetes (GDM) in 56% cases but not in a whopping 44% institutions. Sixty percent respondents preferred an early morning urine sample for microalbuminuria testing whilst 39% and 2% opted for 24 hr urine and timed overnight sample respectively. Twelve percent and 4% respondents respectively participated in proficiency testing (PT) for glucose only and HbA1c only. Sixty six percent participated in PT for both glucose and HbA1c. 9% participated in PT for neither. Based on the above survey we recommend that scientific bodies/ Associations should educate biochemists to adopt NGSP certified methods for HbA1c testing. Awareness among obstetricians & Biochemists should be increased for recommendation for OGTT for timely diagnosis of GDM. Also, standard guidelines, either DIPSI/ACOG/WHO for diagnosis of GDM, should be followed by all institutes. Morning spot sample be adopted universally for microalbuminuria testing.

S008

e-Health Tools for the Medical Lab for Better Outcomes

Bernard Gouget

Public Health FHF, France

The advent of smartphones in 2007/8 has changed many aspects of our everyday lives. This technologically convergent device serves as a telephone, still/video camera, MP3 player, a source of news and weather, and provides electronic storage and real-time access to the wealth of information available via the internet. In addition, the capabilities of a smartphone can be expanded via an ever-growing number of easily downloaded applications (apps).

The potential of a smartphone for healthcare applications was quickly realized and it has been used in a number of different ways. Medical apps for fitness and health have been very popular and are expected to grow almost exponentially in the next future. A device that plugged into a smartphone to create a medical test device was the next phase of development and current capabilities range from glucose testing to ultrasound scanning. A further development has been medical test devices that connect wirelessly to a smartphone (e.g., Bluetooth connected pregnancy test; Clearblue Connected Ovulation test).

Another use of a smartphone in POC testing exploits the built-in camera for urinalysis. An app guides the user (aided by a chatbot nurse, named Emily) through the Dip.io home-based urinalysis testing process. The camera scans a conventional dipstick placed on a color chart (colorboard). It then uses color recognition, computer vision, and AI to ensure accurate testing under differing conditions and with different smartphones. Results, after classification in the cloud, are automatically sent to the patient's electronic medical record. The menu of tests based on this imaging technique is expected to expand to include urinary albumin:creatinine ratios.

S009

Digital Tools for Researchers in Laboratory Medicine

Alexander Haliassos

ESEAP-Proficiency Testing Scheme for Clinical Laboratories, Greece

Information exchange and communication channels are critical elements of a sound biomedical education and research. High performance computing and wide area fast networks provide the possibility of examining and simulating systems and processes at unprecedented levels of detail and accuracy. The combination of processing power with large-scale databases enables analysis of big volumes of data from today's experiments and simulations. When these technologies are coupled with new capabilities in mobile communications using web enabled smart personal devices an opportunity is created that can revolutionize the scope and the process of scientific investigation. Collaboration tools can help scientists work in harmony and learn together from a distance. These tools span a wide variety of applications, from simple text-based e-mail clients to complex online meeting tools. The term collaboratory or virtual laboratory is used to refer to a set of technologies leading to the generation of new opportunities to create and sustain active scientific communities. The Internet Collaboratories can support expensive equipment to address complex problems, and accelerate discovery and innovation in biomedical research. Having the right tools and technology is a necessary foundation, and building a community needs conscious effort among website designers, community promoters, and leaders. Moreover, web-based applications can provide novel teaching materials in an interactive form. e-Learning can be defined as any use of the Internet and web technologies to help create teaching and learning experiences. Web-based training is individualized, group tutoring or mentoring, delivered over computer networks. Unfortunately, the digital divide, a term describing the differences between those who have access to the Internet and those who do not because of economic reasons, the lack of computer competency or self-efficacy, or the lack of communications infrastructure, negatively affects their use.

S010

Machine Learning in Laboratory Medicine

Sergio Bernardini

University of Tor Vergata, Rome-Dept of Experimental Medicine, Italy

Artificial intelligence (AI) is the simulation of intelligent behavior by machines (e.g., robot, computer) and it encompasses deep learning, machine learning, expert systems and neural networks. In recent years there has been rapid growth in AI and current applications include, for example: speech recognition (conversational systems)(e.g., Apple Siri, Amazon Alexa), face recognition (e.g., Facebook), robotic vacuum cleaners, driverless cars, automated online assistants (chatbots), mobile check deposits, and voice-to-text.

In the clinical laboratory, AI-based technologies that may have a future role include: diagnostic image analysis, digital twins, conversational systems, intelligent things, and augmented reality.

In the routine evaluation of peripheral blood and urine, automated instrumentation has been introduced. A digital twin is a virtual version of a system where it is possible to test the impact of change prior to implementation of the change in the real world. This AI-based technology is finding application in hospital planning and management, and in the future, a digital twin of a clinical laboratory may be an important management tool. The future role of conversational systems in the clinical laboratory is difficult to predict. Current medical uses include Alexa-based advice apps, e.g., the Boston Children's Hospital KidsMD skill provides health information for common illnesses and medication dosing, and the Mayo Clinic First Aid provides answers to everyday health issues or self-care instructions. Certainly, the hands-free aspect for issuing instructions and requesting information can be advantageous, and as the technology becomes more established, some significant roles in clinical laboratory may emerge in the next future.

S011

A Holistic Approach to Risk Management

Wong Moh Sim

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Healthcare organisations have a responsibility to ensure the health and safety of their staff and their patients.

Clinical risk management is implemented to mitigate adverse events in patients. Clinical risk management is defined as improving the quality and safety of healthcare services by identifying the circumstances and opportunities that put patients at risk of harm and then acting to prevent or control those risks. An adverse event refers to an an unintended injury or complication which results in temporary or permanent disability or death, which is caused by healthcare management rather than the disease process. Adverse clinical events include healthcare associated infections, medication errors, patient falls and pressure injuries.

Risk assessments should be done before new processes or activities are introduced and before changes are introduced to existing processes or activities. A risk assessment involves risk identification, risk analysis and evaluation and risk control. Once the risks have been identified, the organisation must implement control processes as well as continuously monitor and modify them, to ensure that risk is maintained at a clinically acceptable level.

S012

A Blizzard of Hazards: Managing Risk in the Clinical Laboratory

Leslie Lam

Department of Laboratory Medicine, Ng Teng Fong General Hospital, Singapore

In the provision of quality patient care, the clinical laboratory is well known to perform broad, efficient, and detailed quality control giving healthcare professionals assurance that test results obtained on patient specimens are constantly accurate and reliable. Unfortunately, errors can occur at any point in the preanalytical, analytical and postanalytical phases of testing. Risk management involves the anticipation of what and where errors could occur, estimation of likelihood and frequency of errors, the consequences or the gravity of the effects caused by it, and ultimately to determine what can be done to reduce the risk to an acceptable clinical level. This talk will make reference to EP23-A, a guideline from CLSI that introduces risk management principles to the clinical laboratory. This guideline derives ideas from manufacturing industry and promotes laboratories to create risk management plans that address the risks intrinsic to each lab. The application of EP23-A should not be problematic for laboratories since they already carry out activities that could be considered risk management, including evaluating the performance of new instruments and assays before testing patient samples, performing regular maintenance and quality control, replying to physician complaints, and troubleshooting errors.

Also included in this talk will be the development of a quality control plan (QCP) to help detect weaknesses in the 3 phases of testing process and outlines specific actions to discover, avoid and control errors that can result in patient harm.

S013

Brisk Tips to Manage Laboratory Risk

Sharon Saw

Department of Laboratory Medicine, National University Hospital, Singapore

The clinical laboratory is an operational area within the healthcare environment in which the potential for errors is high due to high workloads. What may be considered a small error may have a high cost in terms of missed diagnosis, medical mismanagement or increase in procedures and length of stay for a patient.

The laboratory does not have a "perfect" instrument, otherwise we would all be using it. Any instrument can and will fail under the right conditions. Our discussions of risk must begin with what can go wrong with a test (all errors). Lab tests are not fool-proof! This talk will review multiple aspects of laboratory operations and mitigating potential risks.

S014

Family Studies Identify Novel Pre-dispositions to Dyslexia

Subrata Sinha

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Dyslexia is a specific learning disability which manifests as difficulty in reading in spite of adequate opportunity and intelligence. It affects about 10% of the population with varying severity and has a strong familial association. Many genetic loci have been shown to be associated with dyslexia, however there is no predominance of any susceptibility locus across different populations. We have studied three large multi-generational families from different endogamous groups by behavioral studies and high throughput sequencing, followed by molecular characterization. The three families had a distinct pattern of inheritance, being autosomal dominant in one, autosomal recessive in the other and co-morbid with Attention Deficit Hyperactivity Disorder in the third. The susceptibility associated loci also differed in the three families. We observed that the associated molecular associations were localized to the Protocadherin locus in a family with a dominant pattern of inheritance. Interestingly, while seven single nucleotide polymorphisms in the risk variants in this family were present in Neanderthal and the Denisovan genomes and also conserved in non-human primates, the non-risk variant was preponderant in modern humans (Homo sapiens).

A long noncoding (lnc) RNA involved in stem cell differentiation and a FAM family gene which was a regulator of GABAergic pathways comprised the other associated loci. This substantiates the viewpoint that dyslexia comprises of a number of endophenotypes and is hence is a basket of distinct molecular diseases, leading to a similar endpoint. Such studies in endogamous groups enable the identification of distinct neurodevelopmental pathways required for proficient reading.

Bharat Prajapati , Teesta Naskar and Shubha Devasenapati majorly contributed to the work, in collaboration with the laboratories of Drs. Nandini C Singh, Pankaj Seth, Mitali Mukerji, M Faruq, Deepti Jain and S Sengupta.

S015

Population Diversity in India: Implications for Health

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With the ultimate goal of providing a rich resource for precision medicine, we have systematically characterized genome variation among Asian populations. We have focused on population isolates to capture a broad swath of genetic diversity across the continent. We have characterized the genomes of 1,739 individuals, including 1,236 newly sequenced genomes, representing 64 countries and more than 200 ethnic groups. We identified 63 million SNPs, 29 million of which have not been previously described, and nearly 4 million indels. Analysis of these data revealed correlates of extinct hominid admixture with present day social structure in South Asia. We also identified ad-mixture events in Southeast Asia that shed light on where modern humans interacted with deniso-vans during their migrations into the area. To evaluate the eventual value of a population-scale catalogue of Asian variants to precision medicine and molecular diagnostics, we generated pharmacogenomics predictions, identified Asian specific or enriched disease alleles and demonstrated the value of using Asian allele frequency filters in disease gene discovery.
In collaboration with Andrew Peterson, Jeff Wall, Sekar Seshagiri, Sam Santhosh, Stephan Schuster, Analabha Basu, Nidhan Biswas et al.

S016

Ayur Genomics: Tradition and Modernity

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A paradigm shift from a reactive healthcare to a holistic system is being proposed for managing wellness and quality of life in diseased conditions. This has been propelled by advancements in genomics technologies along with deep-phenotyping and AI based analytics. However, innovative strategies are needed for this to be implemented and affordable across diverse populations and in resource constraint settings. This is especially important for delivering affordable health care solutions in the Indian populations. A prerequisite is to understand human genome structure and variations and contextualize it in the context of the population and individual's background. This is a challenge as we witness an unprecedented extent of variability housed in an individual. We have used an integrative genomics approach that combines various aspects of genomics with the traditional system of medicine, Ayurveda to understand the genomic basis of human individuality. This Ayurgenomics framework has provided us a novel framework for precision medicine. The conceptual basis, its validation and contemporariness in global settings would be presented.

S017

Iron - Why Too Little or Too Much is Bad for You

Helen Martin

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Iron is an essential nutrient for almost all living cells, however too little or too much iron can cause significant health problems. Iron deficiency, caused mainly by dietary deficiency or blood loss is common worldwide and the World Health Organisation estimates that more than a billion people, including 40-50% of all children are iron deficient. Iron overload is also common although the causes are more varied. Iron status is assessed by a combination of biochemical and haematological markers that traditionally include iron, transferrin, transferrin saturation, ferritin, haemoglobin, mean corpuscular volume and mean corpuscular haemoglobin. While some patterns of these test results, such as those in classical iron deficiency or primary iron overload of long standing, are easy to interpret, many are difficult. This is particularly true when iron deficiency co-exists with significant inflammatory illness; in such settings additional tests such as soluble transferrin receptor or hepcidin, markers of inflammation such as C-reactive protein or erythrocyte sedimentation rate, or haemoglobin response to a clinical trial of iron therapy may be helpful.

S018

Vitamin D Requirement for Optimal Health Outcomes

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The well characterised endocrine pathway of vitamin D metabolism and its activities are solely responsible for vitamin D regulation of plasma calcium and phosphate homeostasis under control of serum 1,25-dihydroxyvitamin D, the biologically active metabolite of vitamin D. This pathway protects against the metabolic bone disease of rickets in children or osteomalacia in adults. The critical level for serum 25-hydroxyvitamin D to maintain adequate serum 1,25-dihydroxyvitamin D is 20 nmol/L (8 ng/ml) and is synthesised by the kidney. In contrast adequate serum 25-hydroxyvitamin D protects against osteoporosis and reduces risk of fracture at a higher level because this activity depends on 1,25-dihydroxyvitamin D being synthesized by bone cells. Similar metabolism of vitamin D protects against premature mortality,

cancer, infectious disease and adverse outcomes of pregnancy. Metabolism of 25-hydroxyvitamin D to 1,25-dihydroxyvitamin D by macrophages has been well described to enhance killing of microorganisms. The critical level for serum 25-hydroxyvitamin D for metabolism by non-renal cells is 50 to 75 nmol/L (20 to 30 ng/ml). Such autocrine actions of 1,25-dihydroxyvitamin D have now been demonstrated in skin, prostate, breast and colonic tissues, in these latter tissues to protect against cancer. In these tissues activities of vitamin D include reduction of cell proliferation and stimulation of cell maturation, activities which reduce the risk of cancer. The critical level of serum 25-hydroxyvitamin D for optimal health of these tissues is dependent on the level of the enzyme which converts 25-hydroxyvitamin D to 1,25-dihydroxyvitamin D (CYP27B1) and therefore may vary for different tissues.

S019

Laboratory Investigation of Trace Element Deficiencies

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Trace elements are present in human tissue at concentrations of μg/kg or less. Those elements for which a specific deficiency syndrome exists are known as the 'essential' trace elements. Deficiencies may result from inadequate intake, disorders of absorption and/or excessive loss. Iron is the most commonly identified trace element associated with nutritional deficiency and has been discussed separately. In this presentation, I will review the role that the biochemistry laboratory has in investigating nutritional deficiencies for the next most commonly identified trace elements iodine, zinc, copper and selenium. Clinical aspects of trace element deficiencies and the analytical techniques used to quantitatively define an individual's trace element status will be discussed.

S020

Can Atherosclerosis be Prevented- Beyond Nutrition and Exercise?

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Primary (disease prevention), secondary (slow down the progression of disease), and tertiary (disease treatment) are fundamental goals of disease management. Atherosclerosis is a major form of cardiovascular disease (CVD). Cholesterol, associated with low density lipoprotein, as well as chronic inflammation, are suggested to contribute to the development of the disease. While lowering cholesterol has been proven beyond doubt to lower the incidence of coronary diseases, the question whether attenuation of inflammation would affect atherosclerosis has become important. Physical activity and diet have been recognized as deterrents of CVD. During the past decade, we provided evidence that treatment of atherosclerosis susceptible mice with the Sesame oil or an aqueous extract prepared from Sesame oil, reduced the development of atherosclerosis and treatment after the establishment of atherosclerosis enhanced its regression. More importantly, pre-treatment of atherosclerosis susceptible mice with the oil or the extract reduced the subsequent development of atherosclerosis without affecting lipid levels.

Our data indicated that attenuation of monocyte/macrophage cell functions and inflammation, independent of lipid lowering might be beneficial and important. If tested and effective, the anti-inflammatory agents could serve as a valuable therapy for reducing uncontrolled inflammation in many diseases, including CVD. While, drugs, such as statins have proven valuable in the treatment of atherosclerosis, diet and nutrition-based prevention strategies might become valuable in preventing inflammatory diseases even at a younger age.

S021

Cardiac Biomarkers: Diving in Data and Multidisciplinary Discussions to Find New Targets

Damien Gruson

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Cardiovascular diseases represent a global burden because of related high morbidity, high mortality and a huge impact on health economies. The fight against cardiovascular diseases is crucial and laboratory tests are important to assist physicians for prevention, diagnosis and prognosis of cardiovascular diseases. Many new biomarkers are emerging from a better understanding of pathophysiological pathways and translational research. Biomarkers related to cardiorenal function and cardiac remodeling are good examples and may provide additional information to natriuretic peptides testing and help to develop more tailor based strategies for treatment. Innovations are also coming from the field of data mining and integration as it will allow to combine clinical and biological features for a more accurate management of patients and will facilitate the identification of clusters of patients at higher risk or more potent to be selected for clinical trials. Beside the shift of paradigm for biomarkers testing and date, the recent progresses of the science of mobile Health (mHealth) are also spectacular and mHealth tools and applications can contribute to precision care and development of new services for patients management. However, to reach the prime time, innovative biomarkers and emerging technologies will require a multidisciplinary assessment of technical, clinical and economical outcomes, meaning that the communication between specialists in laboratory medicine

and other healthcare professional will be needed for an efficient translation into daily practices.

S022

Implementing a New Cardiac Biomarker: Keys for Successes

Evgenija Homsak

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Cardiac biomarkers are important tools in a combination with other clinical or imaging methods for managing several cardiovascular diseases (CVD) that concern and involve heart overload, heart failure (HF), myocardial infarction, stroke or death. The importance of using them is very wide and diverse. By knowing pathophysiological mechanisms in which they are involved, their structures, sources of synthesis and production, metabolism, and other important characteristics, we can help to explain, to diagnose and prognosis the disease development. Therefore, if they are applicable with a quality satisfied measurement method and met the criteria of a good biomarker, with good reproducibility, sensitivity and specificity they can be used as an additional diagnostic or prognostic markers, for follow up the disease or monitoring treatment.

Today we have several already established cardiac biomarkers on the menu, that are on disposal for the routine use. Natriruetic peptides are important markers for the diagnosis, management and prognosis of HF. Troponin T or I and especially the high sensitivity tests for their determination are important as diagnostic markers and for follow up myocardial infarction. Lately, there were introduced also other biomarkers (Galectin-3, GDF-15 and sST2), that are becoming important, as a sole and especially as additional biomarkers in combination. According to different pathophysiological mechanisms, in which they are involved, giving the added value and rising up the strength and importance of combine biomarkers use. Galectin-3 and sST2 are markers of fibrosis and cardiac re-modelling and are important for CVD and HF prognosis assessment. According to important characteristics of sST2 its use is also important for follow up patients and treatment decision. Last studies have confirmed also its important role in the management of Chronic Kidney Disease, especially in risk assessment of the patients with End Stage Renal Disease.

S023

How Education can Facilitate the Use of New Biomarkers: Technologies and Future Directions

Pradeep Dabla

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Biomarkers play a vital role in disease detection and treatment follow-up. It is important to note that the diseases in the early stage are typically treated with the greatest probability of success. Therefore, the early detection of biomarkers is very important in the case of cancer, cardiovascular disorders, and other pathological conditions Biomarkers may includes antigens, DNA, mRNA, and enzymes. Various highly specific recognition biomarkers have been developed based on enzyme-linked immunosorbent assay (ELISA), gel electrophoresis, surface plasmon resonance (SPR), Mass-sensing BioCD protein array, surface enhanced Raman spectroscopy (SERS), colorimetric assay, electrochemical assay and fluorescence methods. The ultimate goal behind the use of biomarkers for diagnosis is to develop reliable and cost-effective powerful detection tools for early diagnosis of diseases and disorders. A serial combination testing of a sensitive early marker (e.g., H-FABP, myoglobin, or CK-MB isoforms) and one of the cardiac-specific troponins (cTnT or cTnI) offers the best approach. Two serial testings within a minimum of 12 hours (e.g., at 0, 4-6, or 12 hours) after symptom onset provide reliable sensitivity and specificity for detecting cardiac ischemia and myocardial infarction. Trends in the clinical diagnostics indicate the necessity of a diagnostic test to be performed near the patient sites. Thus, biomarker detection platforms must therefore be adapted for a rapid and sensitive point-of-care testing.

S024

CTC Analysis: An Overview of CTC Technologies and Clinical Significance

Evi Lianidou

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Liquid biopsy provides a valuable source of biomarkers through simple and minimally invasive serial blood draws and represents a highly dynamic diagnostic, prognostic and theranostic tool for the management of cancer patients. Circulating tumor cells (CTCs) are major players in liquid biopsy and their presence has been linked to worse prognosis and early relapse in numerous clinical studies. CTC molecular characterization offers an exciting approach to monitor the efficacy of systemic therapies in real-time, unravel the biology of cancer cell dissemination, understand

resistance to established therapies and identify gene targets and signaling pathways relevant to therapeutic interventions. Single-cell CTC analysis is a powerful tool to understand tumor heterogeneity and the mechanisms involved in cancer progression with potential implications for improving treatment strategies. However, there are still some barriers to the establishment of CTCs in routine clinical use: 1. The numerous technologies available for their detection and characterization should be standardized and clinically validated, 2. The number of biomarkers for evaluation in CTCs is constantly increasing while the amount of sample is limited; 3. pre-analytical phases must be standardized in order to obtain robust and reproducible results; 4. the turnaround time of CTC analysis is currently slow. This overview is focused on the latest developments in the detection and molecular characterization of CTCs, and their clinical applications in many types of cancer.

S025

Circulating Tumor DNA (ctDNA): Detection Systems and Clinical Significance in Cancer

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In the last several years ctDNA as a molecular blood biopsy has made significant progress in diagnosis of cancer status and outcome in solid tumors. In the USA there are multiple assays approved by CLIA and FDA, and growing in numbers every year. This demonstrates how the field of ctDNA has moved significantly in diagnosis and follow up of cancer patients in multiple cancer types. The ctDNA assays have been effectively used in entering patients in modern day respective targeted therapies. More recently our group and others have shown ctDNA can be used in assessing immunotherapies particularly checkpoint inhibitor immunotherapies (CII). In the complexity of combined CII and targeted therapies realtime monitoring of patients through ctDNA has become increasing more important for patient management decisions. Multigene approach of ctDNA has significant advantages in realtime patient diagnosis and follow up particularily in tumors that are evolving and metastasizing. Temporal tumor genomic heterogeneity can be monitored by ctDNA. ctDNA can be in multiple forms that include mutations, amplifications and methylation of genes. Recently we have shown that multipanel 70 gene mutation ctDNA can be effective in monitoring patients realtime changes during follow up of progressive disease. This supports through a blood biopsy the tumor mutation burden (TMB) as in tumor tissue analysis can be important in treatment. Recently, we have shown a novel gene region amplication at 1q ctDNA biomarker in solid tumors can be helpful in chemotherapy and CII monitoring patients responses. Another type of blood cell free DNA is ctmiRNA . Using a 2100 miR next generation sequence based assay we can detect primary gliomas and brain metastasis. The ctmiRNA panel offers a highly sensitive assay for monitoring patients in addition to ctDNA. As the field grows the molecular blood nucleic acid assays are becoming more sensitive and monitoring realtime cancer patient status efficiently.

S026

Liquid Biopsy: the New Frontier in the War Against Cancer

Harriet Winkman

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Liquid biopsy research has received increasing attention over the past years due to their many potential clinical applications in the individual management of cancer patients. In the metastatic setting, when an operation is no more beneficial, if biopsies are taken, they are usually obtained from single sites. These biopsies are not always sufficient to recapitulate the vast intra-patient heterogeneity of metastatic lesions, and therefore some clinically relevant alterations can be overlooked. CTCs and ctDNA are released from all sites, thus reflecting the heterogeneity of the different tumor cells. A great benefit of liquid biopsy is the easy access of samples allowing a repeated assessment of patient during the whole course of the disease.

The different liquid Biopsy analytes have shown to recapitulate the tumor response during treatment and the elimination or decrease of CTCs and ctDNA following treatment is associated with improved clinical outcomes. Liquid Biopsy can also reveal novel targetable alterations, such as expression of HER2, not seen in the primary tumor of the same patient, as well as markers of therapy resistance such as the androgen receptor variant v7 or T790M mutation in prostate and lung cancer. CTCs and ctDNA thus represent two complementary liquid biopsy approaches with great potential to track molecular tumor evolution in individual cancer patients. This overview will focus on the use of liquid biopsy to understand tumor heterogeneity and the mechanisms leading to cancer metastasis.

S027

Role of Laboratory in Assessing Athletic Performance

Roberto Verna

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One of the most important achievements of medicine over the past two decades is the role of exercise in maintaining and improving health. The current situation, in fact, confirms that the main risk for health is sedentary, that causes 2 million deaths per

year in the world. Physical inactivity is responsible or at least favors several pathologies and regular physical activity is essential for prevention.
In 2018, the estimated Italian Public Health spending will reach around 115 billion euro, 2.0% increase if compared to 2015. In 2012 it was 111 billion, equivalent to 7% of GDP (about 1,867 euro per year per inhabitant). Health expenditure is growing but, news of these days, in Italy the average duration of life, even if only slightly, is coming down.
Does it mean that, despite the huge professional and financial commitment, the resources are badly or not adequately employed, or is it just the result of the increase in costs? In any case, although much lower than that of other major European countries, it is necessary to find new ways to reduce health care spending.
The linear cuts made until now were strongly, and rightly, disputed because they do not guarantee an improvement of health; indeed, they are likely to worsen the resulting further increase in costs. It is, in fact, necessary to start a path to appropriateness, but this cannot be just a term; we need a strategy shared with health stakeholders. Prevention is certainly the best way to rationalize spending and thus reduce it; but we all know that proper prevention can give long-term results, although it certainly leads to a substantial savings, at present risks only to be a cost; and resources seem exhausted.
It is, therefore, necessary to decide whether to run after the disease trying to patch the most damage created by it, spending as little as possible or, rather, do some investment, perhaps in new technologies, especially diagnostics, to reduce the occurrence of disease. Or, more simply, to try to get less sick.
Physical inactivity is a health risk, because it produces 2 million deaths / year worldwide. In particular, physical inactivity favors the 10-16% of cases of breast cancer, colon cancer and diabetes and 22% of heart attacks. Regular physical activity is thus critical for prevention.
The health benefits brought by the change in lifestyle habits are proven by a 25-year study in which it was shown that the change in lifestyle has reduced deaths from cardiovascular disease (-68%), stroke (-73%), cancer (-44%).
A more active lifestyle would lead to the prevention of at least 2 million premature deaths and 20 million DALYs (Disability-Adjusted Life Year) in the world.

S028

Epigenetic Inheritance and Therapeutic Strategies Underlying Gender Medicine. A New Challenge for Women Health

Cinzia Marchese

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Epigenetics refers to structural modifications to genes that do not change the nucleotide sequence itself but instead control and regulate gene expression. New data suggest that epigenetic modifications might be involved in the pathophysiology of adult disorders such as cancer, diabetes, obesity, and cardiovascular diseases.
A growing body of evidence suggests that epigenetic markers in cardiovascular diseases (CVDs) are becoming useful for clinical diagnosis of heart diseases and therapeutic approaches. Findings from clinical studies suggest that premenopausal women are relatively protected from the incidence of CVDs and resultant morbidity and mortality compared to age-matched men. In this field, gender-specific medicine is useful to understand how biological sex and gender affect disease progression. The current hypothesis is that estrogen levels might contribute to the sex-based difference by providing vascular protection, this suggesting the use the sex hormone E2 as a preventive pharmacological approach for CVD in women. However, recent clinical trials revealed conflicting data about the effect of E2 on CVD. This paradox might be explained by epigenetic changes involving estrogen-related pathways. Moreover, gender- based differences do not rely only on hormonal status, but also on environmental factors, such as nutrition, chemicals and social behaviors, which can further influence epigenetic marks in a sex-specific manner.
Nowadays, epigenomic analysis on a large scale provides a useful tool for the discovery of novel epigenetic marks regulation gene expression of molecules involved in estrogen pathways linked to CVD pathogenesis.
In conclusion, the identification of sex-specific epigenetic patterns is strongly recommended to predict the different cardiovascular susceptibility between male and female individuals, and to develop pharmacological approaches based on selective modulation of the epigenetic landscape.

S029

Lapland Extreme 900 km: An Outstanding Stress Model or a Fabulous Positive State?

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Lapland Extreme Challenge (LEC) is a 900 km ultra-event through the Finnish Lapland wilderness. The athletes need to transport with them everything necessary to self-sufficiency. Stressors as strenuous exercise, freezing temperatures, sleep restriction, isolation/solitude - were all present simultaneously. We monitored clinical and biochemical parameters (BP) of one of the athletes. On the tenth day, after 450 km, he gave up due to frostbite (thumb and part of the left foot, not permanently damaging). Our main results were: a significant increase of creatine kinase (CK), lactate dehydrogenase (LDH), myoglobin (MYO), potassium (K) after LEC, a significant decrease of Calcium (Ca), osteocalcin (OSTEO), bone alkaline phosphatase (BALP) and cortisol (CORT). The drop of CORT was unexpected. A possible explanation could be that for this élite athlete walking alone, in isolation, far from

any noise, living with nature's rhythm, in silence, represented not stress, but a positive state, like Shinrin-yoku (taking in the forest atmosphere or forest bathing), that led him to psychological well-being associated with less cortisol secretion, independently of the extreme load and risk associated with extreme environmental conditions. UREA, uric acid (UA) and creatinine (CREA) were basically unvaried. All BP showed no significant changes during training. Biochemical variations observed during the competition were resolved during the restoring period. In summary, apart from frostbite, this athlete adapted psychologically/physiologically to LEC.

S030

Quantifying Critical States of Complex Diseases Using a Phase-transition Model of Network Dynamics

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Considerable evidence suggests that during the progression of complex diseases, the deteriorations are not necessarily smooth but are abrupt, and may cause a critical transition from one state to another at a tipping point. Some key parameters undergo wide fluctuations at these bifurcation points, driving thus the system toward different outcomes. By identifying those criticalities, we could be able to recognize relevant factors involved in the pathogenesis of the disease, and finally the threshold value below/above which the disease process begins. We develop an experimental model to detect early-warning signals of such critical transitions, even with only a small number of samples. Specifically, we theoretically derive an index based on a dynamical network biomarker, which would act as a general early-warning signal indicating an imminent bifurcation or sudden deterioration before the critical transition occurs. Based on theoretical analyses derived from cell and animal studies, we show that predicting a sudden transition is achievable provided that there are a large number of measurements for each sample, e.g., high-throughput data. Yet, the relevance of such approach in both diagnosis and therapeutic monitoring needs to be validated by clinical trials.

S031

Serum Proteome Based Diagnostic Biomarkers of Antitubercular Drug Induced Liver Injury

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Drug toxicity is an unfortunate outcome of currently used tuberculosis treatment. Adverse effects of TB therapy experienced by the patient results in discontinuation of the drug treatment thus facilitating emergence of M.tb resistant strains. Currently available diagnostic markers for DILI have serious limitations as they have specificity beyond liver. To help identify and accurately diagnose the onset of DILI, there is an urgent need for discovery of more sensitive and specific biomarker(s) that could aid patient diagnosis and treatment. Currently, no single biomarker is suitable for the diagnosis of DILI, due to the multifactorial character of its pathophysiology. We evaluated serum samples from TB patients having antitubercular DILI, healthy controls , Naïve TB patients & Non-toxic patients to search for novel potential proteomic and genomic biomarkers which can help in early detection and monitoring of drug induced liver injury associated pathology. Differential proteome obtained using in gel trypsin digestion of peptides and bioinformatics search for annotating the functional categories and pathway similarities revealed 5 proteins to be differentially expressed. These proteins have major role in various liver functions viz. protein binding, lipid metabolism, transport of free hemoglobin back to liver for recycling, antioxidant etc. Further genomic approach elucidated role of predictive genomic biomarkers i.e. liver specific microRNAs due to their organ specific location and their pathway similarity with identified proteins from proteomic profiling . Study elucidated significant role of miRNAs (mir122 and mir192) in antitubercular DILI as they play crucial role in liver cell development, differentiation & homeostasis.These observations may open a new window for development of potential circulating biomarkers to improve the surveillance mechanism for detection of antitubercular drug induced liver injury.

S032

Transcriptomic Signatures as Diagnostic Biomarkers for Pulmonary and Extra-Pulmonary Tuberculosis

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To achieve the ambitious target of eliminating tuberculosis (TB) by 2025, novel diagnostics are required particularly to fill the gaps in the current diagnostic setup. There is need for the identification of biomarkers that are specifically expressed by pathogen/host abundantly during the disease condition. Work from our lab has led to identification of Mycobacterium tuberculosis (M. tb) genes expressed at the site of infection both during pulmonary TB (PTB) and extra-pulmonary TB (EPTB). Whole genome microarray of the mycobacterial transcriptome in biological specimens from PTB and Pleural TB demonstrated a set of genes that are upregulated only in TB patients and not in the disease control. Based on these transcripts, a RNA based molecular assay led to detection of sputum smear positive and sputum smear negative patients with sensitivities ranging from 87-100% and 50-67% respectively with high specificities. In pleural TB patients, transcripts identified from mycobacterial transcriptome analysis in the pleural biopsies were further validated in the pleural fluid specimens thus confirming the potential of in vivo expressed mycobacterial transcripts as diagnostic biomarkers. Studies are also going on to identify the host transcriptomic signatures for their utility as an adjunct to pathogen associated biomarkers particularly for pleural TB which is still difficult to diagnose. Further, using a novel technique of peptide array, we have identified the immunodominant epitopes of the mycobacterial proteins encoded by in vivo expressed transcripts. Multi-well arrays have been designed with the identified immunodominant epitopes for their evaluation in large number of PTB and pleural TB patients. Our study signifies the importance of biomarkers being expressed under in vivo conditions in clinical specimens of TB patients and such carefully identified biomarkers can be utilized for the development of both molecular as well as immunological tests for the diagnosis of TB.

S033

Role of Biomarkers in Risk Prediction and Progression of HBV Related Hepatocellular Carcinoma

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Hepatocellular carcinoma (HCC), the predominant type of primary liver cancer, is one of the most serious life-threatening malignancies, worldwide. In majority of the cases, HCC develops after prolonged and persistent chronic liver disease. Hepatitis B virus (HBV) or HCV infection is prominent etiological factors, attributing to this condition. It has been well documented that HBV, being the inducer of chronic inflammation, is the main causative agent in causing HCC, particularly in Asian countries. The HBV infection leads to a wide range of clinical symptoms from carrier state to malignancy. Cytokines being immune-modulatory molecules, are the key mediators in the defence mechanism against viral infection. SNPs in the key cytokine genes determine the variability in the clinical manifestations in an HBV-afflicted individual, which might finally, culminates into HCC. Since cytokine production is regulated genetically, the cytokine promoter region single-nucleotide polymorphisms induced changes, greatly affects the cytokine production, thus resulting into differential outcome of immune balance. We determine the association between various SNPs in Th1 and Th2 cytokines genes and the risk of HBV disease chronicity to develop HCC.The current biomarkers for the diagnosis of chronic HBV-related HCC is limited. Circulating microRNAs (miRNAs) were first proposed as potential cancer biomarkers in 2008 as these are reported to be stably present in different body fluids including plasma and serum. miRNAs regulate expression of their target mRNAs mainly through their degradation, cleavage, or translational repression. Among various miRNA that have been deregulated in HCC, miR-183-96-182 cluster has been found to be up regulated. We evaluated the biomarker potential of these miRNAs during the progression of HBV disease to HCC. The plasma levels of hsa-miR-182 and hsa-miR-96 were found to distinguish between HBV-HCC and normal subjects whereas levels of miR-96 could distihere

S034

Ischemia Modified Albumin, a Possible Marker of Long-term Hypoxia in Respiratory Disease

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Partial pressure of oxygen in arterial blood (PaO2) and oxygen saturation (SaO2) have been used as conventional markers of hypoxia. However, since they represent degree of hypoxia only at the moment, physicians cannot evaluate severity of hypoxia correctly while patients are outside of hospital. We focused ischemia-modified albumin (IMA) could be an indicator of long-term hypoxia. When pH drops, eight amino acids from N-terminal of albumin reduce their ability to bind transition metals. Applying this phenomenon, we hypothesized IMA may contribute as a long-term hypoxia marker reflecting hypoxemia in recent weeks.

Nineteen respiratory disease patients, 14 males, 5 females, Ages were enrolled. They were 11 COPD, 3 pneumonia, and 4 other diseases. As normal control, sera from 24 healthy adult volunteers (12 males) were collected. Subjects with ischemic heart, cerebrovascular diseases were excluded. IMA was assayed by the albumin-cobalt-binding test (Clinical Chemistry 49:581,2003) and expressed as absorbance arbitrary units (AU) using Versa Max microplate reader.

Average concentration of IMA in respiratory disease patients was 0.48AU, which was significantly higher than that of healthy adults (0.36AU, p=0.000138). IMA had mild regression with SaO2 (r=0.337), and PaO2 (r=0.343). Concentration of IMA decreased after successful treatment of hypoxia such as oxygen inhalation and anti-microbial administration.

We previously reported increase of IMA in neonates' sera with fetal distress (Gugliucci A. Clin Chim Acta.2005). Serum IMA concentration is increased in hypoxia patients. After successful treatment, decrease of IMA was observed. Low correlation to PaO2 as well as SaO2 may reflect IMA as an independent marker of long-term hypoxia. Although sample number is limited, our data suggest that IMA could be an indicator of long-term hypoxia. More study is needed to confirm the result. (Kimura S, et al. Clin Chem Lab Med 2018)

S035

Presepsin: A Novel Biomarker for Sepsis

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Sepsis is a serious medical condition and a major challenge in hospitals, where it's one of the leading causes of death. In 2016, sepsis is defined as "life-threatening organ dysfunction caused by a dysregulated host response to infection". Following this new definition (Sepsis-3), Surviving Sepsis Campaign: International Guidelines for Management of Sepsis and Septic Shock: 2016 (SSCG2016) and the Japanese clinical practice guidelines for management of sepsis and septic shock 2016 (J-SSCG 2016) were published. For the diagnosis of sepsis, blood culture test is thought as a gold standard of infections, but it is low sensitive and time-consuming. So, there is a demand for high-sensitive biomarker for early diagnosis of sepsis. Presepsin, a truncated form of soluble CD14, is a novel sepsis marker found in Japan which concentration in plasma is increased specifically in sepsis patients. Clinical studies revealed the usefulness of Presepsin measurement compared to C-reactive protein (CRP) and procalcitonin mainly as follows; Presepsin is 1) increased earlier after the onset 2) less affected by the other inflammation such as trauma, 3) reflected severity and prognosis of illness. Now, it is recommended to measure Presepsin for severe patients as an aid of diagnosis of sepsis (Strength of recommendation: 2B) in the J-SSCG 2016. We have developed the Presepsin reagent for the PATHFAST, a compact automated instrument. This technology achieved presepsin measurement with high sensitivity in a very short time (within 17 min). PATHFAST Pre-sepsin kit was approved in Japan with a reimbursement in 2014, and is now also used in the countries over the world, including some Asian countries. In this section, the features of Presepsin from aspects of biochemistry and clinical utility will be discussed.

S036

Tryptophan Metabolites as a Potential Tumor Marker

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The kynurenine pathway of tryptophan metabolism converts the essential amino acid L-tryptophan into a number of biologically active metabolites such as kynurenine (Kyn), kynurenic acid, 3-hydroxykynurenine, and quinolinic acid. Indoleamine 2,3-dioxygenases (IDO1 and IDO2) and tryptophan 2,3-dioxygease (TDO) mediate the central route of tryptophan metabolism, which is related to the degradation of tryptophan and the accumulation of kynurenine pathway metabolites in the microenvironment. IDO1 is overexpressed in many cancers. IDO1 is thought to represent a major mechanism of the escape of tumor from the host immune system. Several strategies for targeting IDO1 is currently being assessed in multiple clinical trials. In addition to IDO1, we demonstrated that IDO2 is also an important immune regulator in the tumor microenvironment. In this presentation, we will address the important role of tryptophan-metabolizing enzymes in tumor microenvironment, and serum Kyn can be used as a biomarker for the prognosis and the rate of progression of several cancers. We demonstrated that serum Kyn is correlated with poor prognosis of several cancers such

as leukemia/lymphoma. We will also address that quantification of tryptophan metabolites by our recently developed liquid chromatography-mass spectrometry (LC-MS) method can be applied to analyze changes in tryptophan metabolism in serum. Imbalances in tryptophan metabolism is implicated in diseases ranging from cancer to neuropsychiatric diseases. We believe that this method could offers a novel approach to develop potential biomarkers for diagnosis, assessment and prognosis of the diseases.

S037

Implementation of Front-End Automation System in Clinical Chemistry Laboratory

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Laboratory test turnaround time and errors are largely affected by pre-analytical phase, front end processing makes a major contribution to improve the turnaround time and mitigate these errors. Several barriers hamper the implementation of the front end automation system. These are largely attributed to lack of desire and understanding of improvement of workflow, the process, the costs, safety issue, and the customer expectations; unclear the functionality of front end system; less flexibility of connection and interface with complicated analytical systems; inadequate technical support including IT; failure to optimize current processes prior to automation. To overcome these, possible solutions were raised and identified to meet the needs during the process of selection and implementation of front end automation system to ensure a smooth "go-live". This session will share how we successfully implemented the Roche cobas 8100 automated system and the improvement of efficiency. In particular, case-based specific scenarios and solutions such as tube size, sorting, centrifugation, aliquoting, barcode reading, specimen integrity inspection, LIS interfacing, and LIS functional testing before go-live will be discussed. After the session, participants will then understand the process, solutions, and benefits of implementation of front end automation system to maximize laboratory efficiency.

S038

Automated Lab Results Reporting for Improved Patient Care

Lakshmi V Ramanathan

Memorial Sloan Kettering Cancer Center, New York, USA

The challenge facing clinical laboratories is a reduction in skilled and trained manpower coupled with a growing demand to improve turnaround time. This has led to an explosion of automation systems of varying complexity. In this session, we will focus on the relationship between the automation and the laboratory information system that interact with each other seamlessly.

We will highlight the role of middleware that is information software installed between the LIS and insturments connected to the automation system. Emphasis will be placed on the types of rules can be built into the middleware that impact verification of test results. These include autoverification, specimen integrity checks, quality control flags and moving averages. The process of verifying and periodically checking rules that are built in middleware will be discussed.

In addition to the analytical systems in the laboratory, the impact of rules for test ordering and specimen collection will be examined. At the conclusion of this session, participants will understand different programs that can be built in the middleware for accurate and rapid reporting of test results for improved patient care.

S039

Automation in Clinical Microbiology: Challenges and Opportunities

Yi-Wei Tang

Memorial Sloan Kettering Cancer Center, New York, USA

The trend toward automation in clinical pathology laboratories has largely bypassed the clinical microbiology practice till recent years; the one exception has been the partially automated, continuously monitored blood culture systems. The relatively recent use of automated devices for front-end processing as well as workup of specimens submitted to a laboratory for analysis has initiated a revolution in the clinical microbiology services resulting in both challenges and opportunities. The potential value of these automated devices includes (1) the possibility of substantial operation cost savings, (2) standardization of specimen processing decreasing errors, (3) more rapid and consistent provision of both identification and antimicrobial susceptibility test results, (4) enhanced recovery of fastidious microorganisms, and (5) diminished risk for laboratory-acquired infections. In this presentation, I will review the historical impediments to automation in the microbiology laboratory and introduce the currently available specimen-processing devices as well as the total laboratory automation solutions. Lastly, I will outline the types of studies that will need to be performed to fully assess the benefits of automation in microbiology laboratories.

S040

Approaches and Challenges of Automation & Interferences in Clinical Laboratories

Amitava Dasgupta

University of Texas McGovern Medical School at Houston, USA

Immunoassays are widely used in clinical laboratory due to ease of automation, running specimens in batch and generating results in a timely fashion so that clinical laboratory can meet requirement of turnaround time. However, depending on specific immunoassays, interferences may be observed in 0.4 to up to 4% specimens. When test results are falsely elevated and do not correlate with clinical picture clinicians call the lab for and express their concern that values may be falsely elevated giving an opportunity for laboratory professionals to investigate the problem but negative interference where expected abnormal value is within reference range, it may escape scrutiny of a busy clinicians. Death of a person due to digoxin overdose as a result of negative interference of canrenone in MEIA digoxin assay has been reported. Similarly death has been reported from myocardial infarction due to negative interference in troponin I measurement. Moreover, autoantibody, heterophilic antibody and paraprotein may cause both positive and negative interference on the same analyte depending on assay design. Negative interference in certain vancomycin immunoassay by paraproteins may confuse a clinician resulting in unnecessary increase in dosage causing severe vancomycin toxicity. Negative interference due to hook (prozone) effect is also problematic resulting in wrong diagnosis. In some immunoassays, interference is bidirectional where interference is negative if concentration of the interfering compound is moderate but interference is positive with high concentration of interfering substance. Such bidirectional interference is more problematic than either positive or negative interference. In this lecture all these issue will be discussed with real life case reports from our hospital and also interesting cases reported in the literature.

S041

Pathology Supported Genetic Testing-Experience from Africa

Rajiv Timothy Erasmus

Stellenbosch University, South Africa

The diagnostic process begins with clinical observations and the need for accurate pathological assessments to arrive at a specific diagnosis. Pathology results, similar to genetic test results, are interpreted within a clinical context as an accurate diagnosis depends on careful clinic-pathological correlation. Through clinic-pathological correlation, all laboratory results including those obtained through molecular genetic applications, are finally interpreted and integrated with the patient's clinical findings and data from other special investigations. Identification of genetic subgroups at risk of drug side effects or with different treatment or dietary requirements provides a scientific basis for targeted intervention as opposed to a one-size-fits-all approach. Pathology supported genetic testing (PSGT) involves five steps: (a) Document family history and evaluate the patient's current health status (b) Choose appropriate genetic test(s) based on the medical history and lifestyle risk factors (c) Combine information obtained in 1 and 2 into an informative test report, providing risk implications and health guidelines based on gene expression, if any (d) Apply test information to rectify gene-environment mismatches that may be reflected as biochemical abnormalities or clinical symptoms.

Although targeted sequencing panels are currently the method of choice in breast cancer diagnostics, we favour the use of whole exome sequencing (WES) preceded by PSGT to facilitate patient selection , clinical interpretation and personalised therapeutic intervention. The PSGT strategy, which incorporates lifestyle factors with pathology and genetic tests strives to facilitate patient selection and clinical interpretation. In this presentation our experience in the identification of patients with an increased risk of breast cancer through the use of PSGT and those benefitting or not benefitting from recommended treatment regimens in an African context will be discussed.

S042

Viral Load Scale Up in Zambia: Challenges and Successes in Central, Copper-belt and North-Western Provinces

Hilary Lumano

John Snow Incorporated SAFE Project, Zambia

Zambia through the Ministry of Health has been implementing strategies to enhance UNAIDS 90-90-90 Global AIDS Strategy. Scaling up viral load (VL) testing and determining suppression rates has been a major preoccupation of the John Snow Incorporated (JSI) Supporting an AIDS Free ERA Project (SAFE). The SAFE Project through United States Government funding supports the Zambian Ministry of Health with critical VL scale up activities. SAFE is currently supporting a total of about 230,000 clients on antiretroviral treatment (ART) in three (3) out of ten (10) Zambian provinces namely Copperbelt, Central and North Western Provinces. SAFE is supporting 270 facilities in the three supported provinces and has provided 56 motorcycles to improve on VL sample movements from collection points to sample processing labs and ultimately to hubs. Sample movements from hubs to PCR labs is facilitated by vehicles which SAFE has strategically positioned across the provinces. Copperbelt province is the most urbanized province and has the highest number of clients on ART averaging 140,000 while Central and North Western Provinces have 80,000

and 16,000 respectively. Three PCR Labs are providing testing services for the Copperbelt, one for Central and one for North Western province. SAFE is using a cross cutting approach to improve on VL coverage including community outreach, identification of clients due via manual and electronic means, pharmacy interception and by using the appointment system.
To facilitate sample collections, processing, packaging and transportation SAFE has recruited PCR support scientists in the high volume district hubs, this has enhanced VL sample collections and has further enhanced the joint process of identifying clients due for VL testing. SAFE has also been working with facility leadership and district and provincial leadership on using criteria isolating who is due and who is not. The Zambia consolidated guidelines provide clear guidance even though this appears to be challenged by different interpretations. Turnaround (TAT) of VL results across all three provinces has ranged between 4-12 weeks and was clearly problematic; the introduction of an electronic system that improves and monitors the sample chain of custody and resulting via electronic gadgets has so far reduced TAT to between 1-3 weeks (e_Labs). Improved TAT allows for prompt community and clinical interventions.

S043

Management of Lysosomal Storage Diseases in Developing Countries- An African Experience

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University Hospital of Rabat, Morocco

Many inherited diseases of metabolism are due to the deficiency of a lysosomal enzyme, which leads to the accumulation of macromolecules (mucopolysaccharides, sphyngolipids etc.) at the origin of the disease. Mucopolysaccharidosis and sphyngolipidosis are the most interesting because many of them can be treated effectively by enzyme replacement therapy (ERT). The center we set up in Rabat since 1991 Uses basic unsophisticated techniques that allows early diagnosis and treatment of several patients (MPS I, Gaucher, Fabry and Pomp Diseases). Mucopolysaccharidoses (MPS) are diagnosed by DMB colorimetric assay followed by Glycosaminoglycannes (GAG) electrophoresis and enzymatic activities measurement (fluorometry and colorimetry). The diagnosis of sphyngolipidosis is guided by clinical signs and confirmed by the measurement of enzymatic activities. In our Moroccan center, more than 400 patients have been diagnosed, 60% of whom are carriers of MPS I. Followed by Gaucher disease, then Sanfilippo, Morquio, Hunter, metachromatic leukodystrophy, Niemann-Pick, Fabry and Pompe Diseases. Several MPS I, Gaucher and Fabry patients are on ERT. Followed by Gaucher disease, then Sanfilippo, Morquio, Hunter, metachromatic leukodystrophy, Niemann-Pick, Fabry and Pompe Diseases. Several MPS I, Gaucher and Fabry patients are on ERT.

We also demonstrated the causal mutations in about 80 MPS I patients and in several MPS II, MPS III, MPS IV patients, as well as for Gaucher, Fabry and Metachromatic Leukodystrophy patients. In some other African countries such as Tunisia, Algeria, Egypt, even though there is no specialized center for the study of lysosomal storage diseases, several studies are published concerning studies of causal mutations in several diseases. There is certainly a lot of work to be done to generalize the diagnosis and treatment of lysosomal storage diseases in African countries where, because of inbreeding, these diseases are certainly more frequent than elsewhere.

S044

National Survey of eGFR and Urine Albumin Reporting

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The prerequisite for screening policies for chronic kidney disease (CKD) is standardisation of measurement of serum creatinine and automatic reporting of estimated glomerular filtration rate (eGFR) as a marker of kidney function, and uniform assessment of albuminuria particularly the type of sample used and reporting units. A survey was performed to assess the current practice among Malaysian laboratories in CKD evaluation. An e-mail invitation to participate in an online survey was sent to about 140 medical laboratories. Questions regarding methods for measuring creatinine, equations for calculating eGFR, whether eGFR was reported, the terminology used in reporting urine albumin, types of samples used for the measurment and the cut-off values for urine albumin. A total of 42/140 (30%) laboratories answered the questionnaire. The most prevalent method used for serum creatinine measurement was the Jaffe method (88.1%) traceable to Isotope Dilution Mass Spectrometry method (IDMS). Five labs used a correction factor to enable reporting IDMS traceable results for the Jaffe methods. Sixteen labs did not routinely report eGFR with serum creatinine results. However, 14 of the labs reported eGFR on request by the clinician. The formula used by these labs for eGFR reporting was MDRD (64.3%) and all of them except three reported eGFR values more than 90ml/min/1.73 m^2. Regarding urine albumin measurement, nearly 83.3% of labs still used the terminology microalbumin and the sample collected varied. There is a large heterogeneity among the labs regarding type of sample recommended for measuring urine albumin, cut-off value and reporting units. It is evident from the survey results that laboratory diagnostics of chronic kidney disease in Malaysia is not standardised. It is essential to provide a national framework for standardised reporting of eGFR and urine albumin.

S045

Recommendations of the MACB CKD Task Force on Laboratory Reporting of eGFR and Urine Albumin

Leslie Charles Lai

Gleneagles Hospital, Kuala Lumpur, Malaysia

The Malaysian Association of Clinical Biochemists Chronic Kidney Disease Task Force (MACB-CKD-TF) has produced guidelines on the laboratory reporting of eGFR and urine albumin based on the KDIGO and NICE guidelines and in consultation with nephrologists and MACB members. Summary of the guidelines: 1. The Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation shall be used in the calculation of eGFR and not the Modification of Diet in Renal Disease (MDRD) equation. 2. eGFR values <90 ml/min/1.73m^2 shall be reported as whole numbers and eGFR values >90 ml/min/1.73m^2 shall be reported as >90 ml/min/1.73m^2. 3. eGFR shall be calculated for all creatinine measurements except for people <18 years of age and pregnant women. 4. Creatinine measurement shall be traceable to isotope dilution mass spectrometry (IDMS). 5. Where a correction factor has been provided by a manufacturer to enable reporting of IDMS-traceable results for the Jaffe methods this correction factor shall be used. 6. Apply a correction factor to eGFR values for people of African-Caribbean or African family origin. 7. Laboratories using methods for creatinine measurement that are not IDMS-aligned shall not calculate eGFR. 8. All laboratories that measure creatinine must participate in a national or an international external quality assessment scheme for creatinine. 9. Albuminuria shall replace the term microalbuminuria. 10. Albumin to Creatinine Ratio (ACR) shall replace Protein to Creatinine Ratio (PCR) in the initial screening for proteinuria. 11. The first void urine in the morning shall be used for the measurement of ACR. 12. ACR is recommended for screening and monitoring people with diabetes. 13. In the initial detection of proteinuria, if the initial urine ACR is between 3 and 70 mg/mmol this should be confirmed by measuring ACR on another early morning urine sample within 3 months. 14. Where the ACR is >70 mg/mmol PCR may be used for monitoring.

S046

The Implementation of eGFR Reporting in the Ministry of Health Hospitals in Malaysia

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Hospital Putrajaya, Malaysia

Malaysia is relatively a small country comprising 13 states and 3 federal territories. The health system in Malaysia can be divided into 2 categories, which are services provided by the government and services provided by the private hospitals. There are 137 listed government hospitals and 150 private hospitals. Following the publication of the Malaysian Clinical Practice Guidelines (CPG) on Management of Chronic Kidney Disease (CKD) in Adults in June 2011, the National Chemical Pathology Committee on CKD was established to standardize the reporting for estimated glomerular filtration rate (eGFR) in government hospitals. The recommended formula to calculate eGFR in the first edition was the Modification of Diet in Renal Disease (MDRD) equation. The second edition of the CPG on Management of CKD in Adults was published in 2018 and the recommended formula to calculate eGFR is the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation. The National Chemical Pathology Committee must now update all hospital laboratories under the government of the current change in the reporting of eGFR.

S047

Algorithmic Assessment of Bleeding Disorders

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Continental Hospital, Hyderabad, India

Bleeding disorders are of two types: 1) primary - dealing with platelets and vasculature and 2) secondary - dealing with clotting factors. Superficial bleeds are mucosal and include recurrent episodes of epistaxis with or without apparent cause, minor injuries and usually associated with primary, of which von Willebrand, Glanzmann thrombasthenia and Bernard Soulier are common. The secondary type usually presents as prolonged bleeding caused spontaneously or by injury, surgery, trauma, menstruation and many a times is unidentifiable. The frequent sites of deep bleeds are joints, large muscles such as iliopsoas, intracranial bleeds and menorrhagia. The most common bleeding disorders are Haemophilia A, Haemophilia B and Von Willebrand disease. Less commom factor deficiencies are that of factors I, V, VII, X, XI and XII.

The first line investigation is that of peripheral smear study and bleeding time which usually identify primary type. The secondary includes screen profile of Prothrombin Time (PT), Activated Partial Thromboplastin Time (APTT), Fibrinogen, Thrombin Time (TT) and Factor XIII, (the latter now being said to be missed on the qualitative analysis), followed by mixing studies and specific assays. Factor inhibitor assays are done in both congenital and acquired deficiencies when there is no response to replacement therapy. Automation in terms of aggregometry and advanced coagulation analyzers have eased the laboratory protocols. The most essential is, clinical and family history, in terms of type of bleed, frequency and site for an appropriate workflow.

S048

Laboratory Evaluation of Hereditary and Acquired Hypercoagulability

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Tapadia Diagnostic and Research Centre, Hyderabad, India

During the last few years, thrombophilia has contributed substantially to increased pressure on clinical laboratories and demand for testing has increased dramatically. Laboratory investigations of thrombophilia were until recently, based on investigation of the plasmatic anticoagulant pathways to detect antithrombin, protein C, and protein S deficiencies, dysfibrinogenemia and anti-phospholipid antibodies/lupus anticoagulants. They have been expanded to include activated protein C (APC) resistance, Factor V Leiden mutation testing, hyperprothrombinemia (prothrombin gene mutation G20210A) and hyperhomocysteinemia (enzymatic and/or vitamin deficiencies leading to impairment of the relevant metabolic pathway). Since patients to be investigated are not well defined, this places laboratories to spend considerable amounts of time performing investigations that are not always justified. Each laboratory has to review and update the situation with respect to the various aspects associated with thrombophilia, with focus on the following like (a) the paediatric and adult biological reference intervals, (b) who, why and when to be tested (c) which assay is most appropriate, (d) timing of the sample and (e) correlation with medication which throw challenges (especially the latter two) at providing clarity of results. Our experience highlights rarity of Prothrombin G20210A gene mutation, large number of heterozygous Factor V Leiden and Homocysteinemia both homozygous and heterozygous are identified.

S049

Factor Deficiency, Inhibitors, Therapeutic Drug Monitoring

Shanthi Naidu Kamatham

CARE Hospital, Banjara Hills, Hyderabad, India

The case scenarios of bleeding and thrombosis are wide and varied at our center, being a tertiary care hospital.

Over the past five years there has been a tremendous increase of laboratory work on coagulation studies leading to identification of the deficiency. Coagulation in neonates and children below two years of age is a challenge to the laboratory. A spectrum of case reports from Hemolytic Disease of Newborn, Vitamin K and Factor deficiencies are most common. In adolescence, the clinical presentation is a guideline to the laboratory protocol to be followed. Menorrhagia is a common referral to the laboratory.

Hemophilia A (Factor VIII deficiency) and Hemophilia B (Factor IX deficiency), are the most common along with von Willebrand disease. Due to increasing facilities of testing, rare deficiencies of other factors such as Factor VII, Factor X and Factor V are being seen in large numbers. Prophylaxis treatment is being considered in our area and therefore work up for factor inhibitors is on the rise. Interestingly a significant number of acquired inhibitors in the elderly age group is being observed. Incidence of bleeding disorders without any family history are also being documented.

Thrombotic diagnosis is yet another challenge and increased incidence of Anti Phospholipid Antibody syndrome(APLA) is being seen in all age groups, both the primary and secondary type.

Therapeutic monitoring of anticoagulants, Heparin Induced Thrombocytopenia are also frequent referral to the laboratory. Understanding and standarisation of newer anticoagulants is the present herculean task in the hemostasis laboratory.

S050

Immunoassay Interferences and their Impact on Patient Care

Carmen Wiley

VERAVAS, Inc., USA

Immunoassays are the workhorses of clinical laboratories. Using antibodies for diagnostic measurements provides high specificity and sensitivity. Despite all the successful uses of immunoassays, there are still problems interferences. In this talk I review how the common immunoassay formats work. I cover the double antibody sandwich assays, competitive inhibition assays, and delayed capture assays. I then review how the different types of interference mechanisms cause falsely elevated and suppressed results for each assay format (i.e. steric hindrance, bridging, etc.). Finally, I review current strategies to troubleshoot interference.

Learning objectives: After completing this activity, the learner will be able to:

1. list the common interferences that impact immunoassay results
2. describe how interferences impact patient results
3. create a plan for mitigating these interferences

S051

Standardizing LC-MS/MS Assays for Hormone Analysis and the Positive Impact this has on Patient Care

Ravinder Singh

Mayo Clinic, USA

Until recently most of the phenotypic information on congenital endocrine disorders have relied on biochemical testing of steroids, biogenic amines and peptides but is now being combined with the molecular testing. In spite of the mutational analysis of endocrine disorders the correlation of the phenotype relies more on biochemical testing than the molecular testing. Immunoassays have been the methodology of choice for the analysis of steroids and amines in making diagnosis of patients affected with Cushing's, Pheochromocytoma and Congenital Adrenal Hyperplasia (CAH). But very often the results from the endocrine laboratory had to be repeated with the HPLC-extraction assays to rule out the possible cross reactivities with the glucocorticoids, steroid metabolites and drugs with the antibody detecting the analyte. In the past the use of gold standard MS technology in the clinical diagnostic labs have been limited because of labor intensive extraction, sample preparations and chromatographic separations. Recently the use of MS/MS (tandem MS) technology in liquid and gas chromatography has revolutionized the application of MS technology in clinical laboratories. This is due to reduction in effort for extraction and chromatography and as a result has a scope for expediting the analysis of steroids, biogenic amines and peptides for the diagnosis of various endocrine disorders. We at Mayo Clinic have implemented this technology for the routine analysis of steroids, biogenic amines and peptides. These methods not only provide reliable results for endocrine disorders but also can be used as reference methods by other laboratories and accreditation agencies.

S052

Challenges in Harmonization of Pediatric Laboratory Testing

Khushbu Patel

UT Southwestern Medical Center, USA

Harmonization in laboratory medicine requires attention to the total testing process from pre-analytical to post-analytical factors. Failures in any stage of the testing process can negatively impact the quality of test results and lead to adverse patient outcomes. Pediatric laboratory medicine has its own unique challenges when it comes to harmonization. Unique pre-analtyical factors in this patient population can influence both the anlaytical and post-analytical stages of testing. Furthermore, accurate interpreation of laboratory data requires age- and sex- specific reference intervals. This session will discuss the pre-analytical challenges associated with low samples volumes. Examples of strategies used to harmonize laboratory test results across a pediatric hospital system will be provided in this presentation. Lastly, this session will also focus on informatics approaches and other resources used in implementing pediatric reference intervals.
Learning Objectives:

1. List preanalytical and analytical factors unique to pediatric populations and their impact on laboratory testing.
2. Discuss how to overcome operational challenges in harmonizing patient laboratory data across a pediatric health system
3. Identify approaches to establishing and verifying pediatric reference intervals

S053

Calcium and Metabolic Bone Disorders

Sam Vasikaran

Department of Clinical Biochemistry, PathWest-Fiona Stanley Hospital, Murdoch,Western Australia

Parathyroid overactivity due to autonomous parathyroid hormone (PTH) secretion (primary hyperparathyroidism) can cause hypercalcaemia, and parathyroid underactivity, hypocalcaemia. Secondary hyperparathyroidism is seen in response to hypocalcaemia due to any cause, commonly chronic renal failure and vitamin D deficiency. Long-standing secondary hyperparathyroidism can, over time, lead to autonomous overproduction of PTH: tertiary hyperparathyroidism. Investigation for the diagnosis of any of the above conditions should initially entail the measurement of calcium and PTH in the same venous draw. Current generation immunoassays measure the intact PTH molecule although third generation 'bio-intact' assays are also now available. Ionised calcium is more sensitive than total (albumin adjusted) calcium for the diagnosis of hypercalcaemia. Urine calcium and phosphate handling are affected by PTH action and are a useful adjunct in the diagnostic work up. The above measurements may also point to the rarer genetic conditions such as familial hypocalciuric hypercalcaemia, peudohypoparathyroidism etc, for which genetic testing is available, as for the familial forms of primary hyperparathyroidism, either on its own or as part of multiple endocrine neoplasia syndromes.
Biochemical markers of bone turnover (BTM) are released during bone remodeling and can be measured in blood or urine as non-invasive surrogate markers for the bone remodeling rate, and are useful in investigation and management of metabolic bone disease. Method specific differences for some BTM have led to initiatives to standardize or harmonize commercial assays. BTMs increase around menopause and a raised BTM is a risk factor for fracture independent of bone mineral densi-ty. The changes seen in BTMs

with anti-resorptive therapies have been well characterized and this has led to their widespread use for monitoring therapy in osteoporosis. BTMs also show promise in the management of metastatic bone disease.

S054

Diabetes Mellitus

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Diabetes mellitus is a group of metabolic diseases characterised by hyperglycaemia, caused by defects in insulin secretion, action or both. The global prevalence of diabetes mellitus, especially Type 2 diabetes, is increasing rapidly. Diabetes is associated with reduced life expectancy and considerable morbidity. Glycosylated Haemoglobin (HbA1c), a measure of the average blood glucose concentrations in an individual over the preceding 2-3 months, is used to monitor long-term glycaemic control and predict outcome risk in patients with diabetes. Both the Diabetes Control and Complications Trial (DCCT) and UK Prospective Diabetes Study (UKPDS) trials have demonstrated that lowering HbA1c can reduce the risk of microvascular complications such as retinopathy, nephropathy and neuropathy. HbA1c can be measured by several methods, with each method having its specific advantages and limitations. Diabetic patients are also increasingly expected to participate in self-monitoring of blood glucose. In this session, a series of cases is presented to illustrate challenges in glycaemic control monitoring in diabetic patients.

S055

Disorders of the Adrenal, Pituitary and Thyroid Glands

Leslie Charles Lai

Gleneagles Hospital, Kuala Lumpur, Kuala Lumpur, Malaysia

Interesting cases will be presented focusing on adrenal, thyroid and pituitary disorders. The clinical conditions that will be presented and discussed include Graves disease, Hashimoto's thyroiditis, multinodular goitre, Conn syndrome, Cushing syndrome, hypopituitarism, Syndrome of Inappropriate Antidiuretic Hormone (SIADH) secretion, diabetes insipidus and some of the complexities encountered in clinical practice. The emphasis will be on the clinical and biochemical findings in these conditions.

S056

Traceability and the Role it Plays in Standardization

Graham Jones

SydPath, St Vincent's Hospital, Sydney, Australia

Results of laboratory tests are used for medical decision-making with the aim of improving patient health. A fundamental part of the role of the routine laboratory is getting the right result for a laboratory test. This means having assay with good precision, low bias and freedom from interferences. Bias in particular can affect all results for a test leading to incorrect medical decisions. As we now compare laboratory results with decision points and information in the medical literature from all parts of the world, in order to achieve safe, evidence-based laboratory medicine we need to ensure that all results for a test are comparable across the globe. This can be achieved through the process of metrological traceability.

A key component of traceability is selecting the international reference materials and methods that manufacturers should use for setting calibrator values. The Joint Committee for Traceability in Laboratory Medicine (JCTLM) has been formed by the major international organisations in measurement science, laboratory medicine and laboratory accreditation to help with this task. Manufacturers should use the best available materials and methods to assign values to calibrators and laboratories should select traceable methods and verify accuracy to ensure the best outcome for patients.

Assay results are derived by comparing values in patient samples with values in the assay calibrators. The values for concentrations in calibrators are set by comparison with other calibrators and reference materials. As well has selecting the best reference materials, it is necessary to transfer of those values to routine patient samples with a low uncertainty. In order to achieve accurate results for patient care, good practice is required from reference laboratories, manufacturers, routine laboratories, regulators and accreditors.

S057

Moving Average Techniques

Tony Badrick

RCPAQAP, Australia

IQC has evolved over time and practice to become an essential component of laboratory process control. Stop/go decisions are made based on the results of well trialled rules using highly defined material. However, patient based real time quality control (PBRTQC) models have been around for many years and have

recently been investigated as alternatives because:

- Sometimes other controls are unavailable or impractical.
- Patient results might detect an issue that other forms of QC cannot because of commutability issues.
- The state of the testing process can be assessed between the times of routine control-based QC, which may be run infrequently.
- There is little cost.

The development of better techniques and middleware to allow the practical application of PBRTQC has led to a rethinking of conventional QC, and the relationship between IQC and EQA.

In this presentation we will consider PBRTQC and how it could be used in conjunction with IQC or indeed instead of it. The information that can be gathered from some forms of EQA will also be described in terms of understanding of assay stability and capability. A model of integrating IQC/PBRTQC and EQA will also be described.

S058

EQA in Developing Countries Giving Zambia and Nepal as Example

Renze Bais

IFCC, Queensland Australia

As part of its commitment to improving laboratory medicine in emerging countries, the IFCC supports a number of EQA programs. Programs in Zambia and Nepal, using EQA material supplied by RCPAQAP Pty Ltd at a reduced cost, highlight very different challenges needing to be addressed.

The program in Zambia has been running for 4 years with 32 laboratories currently enrolled most of which have a limited biochemistry menu. Laboratories submitted results 94% of the time with reasons for not returning results including instrument down, internet issues, EQA sample delivery or no explanation.

Data was analysed using computer programs written by Renze Bais and laboratories received two types of reports on a monthly basis, one comparing their results with the other Zambian laboratories while the other compared their results with those from the World-wide results. Based on imprecision, the analytical performance of laboratories in Zambia compared to the World-wide performance for creatinine, AST, conjugated bilirubin, cholesterol and triglyceride. They perform poorly for sodium and potassium, protein and albumin

However, much more concerning has been that very few times laboratories were able to return a complete set of results for the tests they perform, the main reason being a lack of reagents. On average, laboratories returned only 50 - 60% of their results, the low figure almost always due to lack of reagents. Throughout the 4 years of the program, it is clear that the supply of reagents is the biggest issue facing laboratories in Zambia.

The program in Nepal has demonstrated that laboratories run comprehensive menus on a variety of instruments. Analytical performance is poor for some analytes compared to World-wide peer laboratories. One issue, that could easily be addressed, is that results throughout the country are reported in different units including non-SI, SI and laboratory specific ones.

We are now developing educational material for developing countries.

S059

Overview and Effectiveness of Strategies towards Appropriate Utilization of Laboratory Services

Praveen Sharma

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Appropriate utilization of Laboratory Services - refers to the entire process of external and internal aspects of laboratory including pre-analytical, analytical, and post analytical stages. A medical laboratory can induce harm to serving patients in three main ways - ordering the wrong test, failing to retrieve a test, and misinterpreting a test result. Various strategies exist to evaluate appropriate utilization of Laboratory services. Laboratory performs tests, mainly on physician request, regardless of their clinical indication or frequency of request. This may lead to inappropriate consumption of resources thereby increasing total health care costs, especially if the test is unnecessary. Thus, educating physicians about costs and giving them access to cost information is one vital strategy towards appropriate utilization of laboratory services. Introducing a new laboratory test requires evaluation of its efficacy for its appropriate use in medical testing. For these, performing retrospective audits are essential. Development of expert systems that may help in prioritize and interpret laboratory tests is another such strategy. An effective Laboratory Information System (LIS) is a valuable resource of an efficient laboratory that may aid in measurement of effective utilization. The laboratory administration has the responsibility for evaluating public health programs and developing quality policies and disseminating them to laboratory managers who must ensure their proper implementation. Laboratory professionals must work together in developing more strategies towards better utilization of services.

S060

Developing a Roadmap for Laboratory Utilization Management Program (IFCC - Abbott VLP)

Sedef Yenice

Group Florence Nightingale Hospitals, Department of Central Clinical Laboratory Services, Istanbul

Utilization management has been a traditional approach to control costs in clinical laboratory services for several decades. Following utilization management, best practices results in the highest quality care at the lowest cost, supports Lean and Six Sigma initiatives, and saves significant time and money. In fact, appropriate utilization reduces patient risk and empowers organizations to provide the highest quality of care. While it is good to have an understanding of utilization management, IFCC Committee on Clinical Laboratory Management has recently conducted an international survey to investigate what does this mean for the laboratory leaders and examined the state of medical laboratory test utilization management and relevant practices which are country-specific from a laboratory leader perspective. The findings of this survey revealed that the recognition of test utilization management, current practices, and maturation of those programs are significantly diverse among countries. It is relatively well established in most developed nations. However, the findings have confirmed that the need to develop a roadmap and to construct essential strategies for managing laboratory test utilization is a common interest. With this regard, it is of importance to select the right management tool to implement an optimal laboratory test utilization. This presentation will address the following key points for implementing utilization management initiatives:

- Structure of effective communication
- Infrastructure to assist implementation
- Establishing a laboratory formulary
- Gatekeeping mechanisms
- Clinical decision support
- Benchmarking and management metrics
- Consultative and Interpretive services

S061

Promotion of Appropriate Laboratory Test Utilization: From financial Accountability to Appropriateness and Quality of Care

Edward Randell

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The policy development concerning the wise use of scarce healthcare dollars is part of good management practice for clinical laboratory services, whether working in a government and publically funded laboratory, or in a private laboratory setting. Improving utilization of laboratory services involves putting in place measures to address misuse of specific tests and services, but also developing and implementing broader scope strategies to address the core problems leading to misuse and in a systematic manner. The speaker in this session will review initiatives conducted over the past 10 years involving his laboratory, and discuss how these initiatives were developed, implemented, and expanded; and how they are maintained over time. Moreover, the impact of these initiatives on test usage and cost avoidance will be explored. All change comes with its barriers and challenges. These will be addressed during discussion of specific examples of initiatives. Organization is key to sustainability. The session will provide examples of several organizational frameworks used to maintain momentum on utilization management activities.

During this session participants will be provided with:

1. A description of successfully used strategies that improve the chances of success of an initiative to modify inappropriate test usage.
2. An overview of successful utilization management initiatives that have curtailed inappropriate use of specific tests and are based on evidence.
3. A description of which stakeholder groups should be involved to facilitate improvement of test usage.
4. A description of the organizational structures required to sustain a broad scope utilization management program, such as a laboratory test formulary, and how the development of such a program can present the infrastructure for sustainable utilization management activities for any sized organizations.

S062

Pediatric Laboratory Medicine: Why is it Different?

Sridevi Devaraj

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The requirements when providing pediatric laboratory medicine differ in many aspects to the requirements for an adult laboratory service. Children, in particular, neonates are much smaller than most and have smaller blood volumes creating a need for separate mode of analysis than in adult practice. Frequently, the platforms that are available for adult measurement and automation are not capable of handling small samples from children so separate analytical platforms are required. Sometimes the only available approach for handling small samples is manual and therefore labor-intensive analysis. Sick neonates are additionally compromised as they also have immature organ systems, even smaller blood volumes and a greater need for more frequent laboratory analysis as physiological and pathological changes occur more rapidly. Older children demonstrate issues related to development, including those associated with puberty. Developmental aspects of organ systems in neonates, later infancy and normal physiological sexual development will be discussed in this presentation. An additional confounding concern in pediatric laboratory medicine is a lack of universal reference interval for many biomarkers.

S063

Autoimmune & Neuroimmune Markers in Pediatric Cases

Barnali Das

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Autoimmune diseases are a family of more than 80 chronic, and often disabling, illnesses that develop when underlying defects in the immune system lead the body to attack its own organs, tissues, and cells. While many of these diseases are rare, collectively they affect 14.7 to 23.5 million people in this country, and - for reasons unknown - their prevalence is rising. They often endure debilitating symptoms, loss of organ function, reduced productivity at work, and high medical expenses. This talk focuses on the detection of Autoimmune markers in pediatric patients with different diseases and finding the clinical significance of the particular disease and the autoimmune markers found in them during clinical diagnosis. The epilepsies, encephalopathies and movement disorders are associated with autoantibodies against neuronal proteins (LGI1, CASPR2 & VGKC-Complex antibody, NMDA Receptor antibody); antibody against Aquaporin-4/ NMO-IgG and paraneoplastic |profile auto antibodies (anti Hu, CRMP5, Yo, Ri, Ma, CV2 &ampiphysin).

The method used here is indirect immunfluorecence assay which is considered a gold standard for detection of autoimmune markers. The test detects the presence of ANA in the blood of the patient which adhere to reagent test cells (substrate), forming distinct fluorescence patterns that are associated with certain autoimmune diseases. Other markers detected by immunfluorecence assay and immunoblotare ANCA, dsDNA and ASMA, LGI1/ CASPR2/ VGKC-Complex antibody, NMDA Receptor antibody, antibody against Aquaporin-4/ NMO-IgG and paraneoplastic profile auto antibodies (anti Hu, CRMP5, Yo, Ri, Ma, CV2 & ampiphysin).

I will discuss basics and case studies segregated according to the autoimmune markers. I will discuss few case studies of all the autoimmune markers and Neuro immune marker in Pediatric Population.

S064

Newborn Screening for Inborn Errors of Metabolism - Current Status & Challenges

Kannan Vaidyanathan

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Newborn Screening (NBS) for Inborn Errors of Metabolism (IEM) is an accepted practice in developed countries. In developing countries including India it is at best partially available. Our own results suggest that lack of screening has led to permanent sequel in the child and burden to the society. The global status of newborn screening with a special emphasis on India will be made during this presentation. Interestingly, there is a difference in the pattern of incidence of IEM in India compared with the rest of the world. We shall also address possible strategies for successful implementation of NBS.

S065

Electronic Apps and Medical Diagnostics Data Management

Khosrow Adeli

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Laboratory medicine is a domain which offers a unique opportunity to analyze objective patient laboratory data and enable ready communication to both healthcare workers as well as patients. In recent years, an increasing number of web-based and mobile applications has been developed to improve access to

laboratory test information and test result interpretation. They range from simple apps that provide reference lab value information to complex medical diagnostics data management. As examples, the "eLab" developed by Tru-Solutions Inc. is a comprehensive medical diagnostic center and lab management software that provides a user friendly interface and access control. It is linked iMedDx.com to allow flexible patient search and selection and includes an eLab Dashboard on mobile/tablet, allowing patients and labs/hospitals access to lab reports online. The Davis's Laboratory & Diagnostic Tests medical app provides another useful app with a wide-breadth of tests, as well as guidance on how to counsel and collect tests. The app is available on multiple platforms including the iPhone/iPad, Android and Blackberry. The "LabGear" is a medical lab reference app providing a pocket tool for medical laboratory test and is integrated with MedCalc with normal lab value reference information for over 200+ lab tests. There are several other medical apps that provide reference lab values including CALIPER, MedRef, Normal Lab Values, and Lab Tests. The CALIPER App has been developed in our laboratory for paediatricians, family physicians, and other healthcare workers worldwide. It is a user friendly and easy tool to assess a child's laboratory test results using the latest reference value database developed based on a study of thousands of healthy children and adolescents. The CALIPER apps allow pediatricians & family physicians to interpret laboratory test results for over 170 medical laboratory tests in children and adolescents using a comprehensive database of pediatric data.

S066

Communication Strategies in Education and Distance Learning

Eduardo Luis Freggiaro

Argentine Biochemical Foundation, Argentina

Education and communications has been shaped by the Internet since it arose. Today there are no limits for contents distribution and scientific information dissemination. Lab professionals can build its own network of contacts on social media, create online contents and access to a huge amount of educational materials. By this way, any professional can build a PLE (Personal Learning Environment), a system of contacts and contents that helps to manage their own learning.

Using new communication technologies IFCC developed "eAcademy", an open educational resource containing distance learning material created and/or reviewed by IFCC experts for the continuous professional development of the members of IFCC affiliated organizations. This is a key project in the IFCC mission. The talk will explain how it was developed from the academic and technical perspectives. All the eAcademy contents are structured according to the Curriculum that was specially created for the project in order to organize the materials offered online.

During the presentation, the eAcademy will be showcased in order to demonstrate how it works for lab professionals continue education.

Also IFCC has a permanent presence on Social Media to spread their activities and increase the participation according with the P4 Medicine concept.

For the future, virtual congresses promise to be a new strategy in scientific data dissemination. The Argentine Biochemical Foundation held its first virtual congress on biochemistry in 2015 and gained lot of experience on this kind of events. In this presentation, some examples and experiences about this new idea will be shared.

S067

Online Resources for Patients and Healthcare Professionals

Tahir Pillay

University of Pretoria & National Health Laboratory Service, South Africa

The internet provides vast resources for patients and healthcare professionals to access healthcare information. Websites offering disease-specific information are increasing and are contributing immensely to the "participatory" aspect of P4 medicine. Patients can also access professional information from open-access publications. Online resources also offer opportunity for communication between patients and between patients and health care providers. Such resources can also be useful for disease management and this is noteworthy in diabetes and asthma where frequent monitoring can lead to early detection of impending crises. There are also online resources for patients with extremely rare conditions eg. inborn errors of metabolism where experiences can be shared with other patients or healthcare providers across the world. Lab tests online, Labs Are Vital and Know Pathology, Know Healthcare are three prominent resources that patients can access to gain specific information about clinical laboratory tests. Lab Tests Online helps patients and caregivers understand the many lab tests that are a vital part of medical care and understand the meaning and implications of results. Labs Are Vital is an online community to support pathologists and laboratory professionals worldwide to elevate the role and reputation of pathology and laboratory medicine in health care. Know Pathology, Know Healthcare is an initiative of Pathology Awareness Australia.

Social media platforms are also important portals accessible to both patients and health-care professions alike. Patients and healthcare professionals can obtain realtime updates on major microblogging platforms such as Twitter and Facebook. Healthcare professionals can engage in case discussions, research collaborations, medical education and crowdsourcing/crowdfunding and likewise with patient groups. As the era of AI evolves, a number of AI portals are emerging to assist patients manage clinical and laboratory data.

S068

Regional Experience in the Implementation of Laboratory Accreditation

Rosa Sierra - Amor

Laboratorio LAQUIMS, S.C., Mexico

Laboratory accreditation based on ISO 15189, landed in Latin America in 2003. Spanish translations of this standard became national but voluntary standards. Later on, accreditation bodies created working groups in laboratory accreditation, which are now committees. Professionals with expertise in different fields of laboratory sciences became technical assessors for the accreditation bodies in Latin America. The Inter-American Accreditation Committee (IAAC) has full and associated members from several countries in the region, and it has been in charge to develop memorandum of understanding (MOU) to recognize the work done in laboratory accreditation regionwise. In 2014, the Chilean Society of Clinical Chemistry initiated a collaboration among professional societies in clinical biochemistry and laboratory sciences, accreditation bodies and metrology institutes in the region, with the aim to let everyone know the specifics of the ISO 15189 standard, and its interpretation. A group of peer review experts were invited to participate, to explain some of the requirements of the standard, including topics such as management, quality assurance, pre and post analytical phase, trazability, certified reference materials, among others. The success of this collaboration allowed all three organizations first of all to meet, to learn from each other, to question the standard, to suggest changes to the standard, and to motivate all those involved to coordinate efforts for the implementation of ISO 1589 in the region. Surveys were sent, questions were reviewed, a better understanding of the state-of-the-art standard was achieved by 2018. This effort has become an initiative of the Latin American Confederation of Clinical Biochemistry (COLABLIOCLI) with the auspices of the International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) and for the benefit of the patient.

S069

Knowledge of Management In Clinical Biochemistry

Hernan Fares-Taie

Laboratorio FARES TAIE, Argentina

The most important objective of Evidence-Base Medicine (EBM) is to improve and optimize clinical decision making by using the best evidence available. Even though medical knowledge is primarily based on science it has some empirical based too. By using data and information from well-designed clinical trials, and by classifying evidence according to its strength, EBM goes further. The evolution of laboratory tests from few to thousands paired with the physician's limited time with the patient can make accurate diagnosis and treatment challenging. The application of EBM to laboratory medicine ensures that the best evidence on testing is available to the clinician when taking care of individual patients leading to improvement on healthcare outcomes. EBM is used to develop laboratory orders as Electronic Health Record (EHR), there is great potential for quality improvement, reduction of errors, reduction of clinical variability and even improved patient and physician satisfaction. When the right test is performed to the right patient at the right time producing the right result. Today, EBM is considered the gold standard of clinical practice; however, EBM has limitations and short comings. Many clinical decisions are influenced by laboratory results, and laboratory professionals have valuable knowledge about the utility and performance of various tests; therefore, it is vital for laboratorians to be involved in developing evidence-base clinical practice at their institutions. Laboratorian's main goal should be the correct utilization of laboratory tests for diagnosis, monitoring, and/or prognosis of patient disease using the best EBM available to them and use the proper implementation tools such as EHR. Participation by laboratory professionals in EBM is sensible and enhances the value of laboratory professionals.

S070

Automated Microbiology Diagnosis

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Universidad Nacional de Asuncion, Paraguay

Clinical laboratories have started more than a century ago, but it was not until the middle of this period that they had a revolution. From the first manual methods to the industrial production of reagents, which ensured quality and reproducibility until reaching the growth in demand for tests, which has pushed automatization to become part of routine purposes. This emergence of adapting technological devices in medical laboratories has increased productivity, reduced costs and improved efficiency. Nowadays, the situation of the Clinical Laboratory requires more reliable, efficient and error-free analytical tests. Nevertheless, in the field of microbiology, it is only until a few years ago that the automation had arisen. This is due to the continuous necessity of improving the time of diagnosis, treatment of patients, reliable antimicrobial susceptibility tests, and reproducible results in less time. Not only has the identification of etiological agents of infectious diseases from cultures and sensitivity studies been improved, but there is also an improvement in the sensitivity of blood cultures and other sterile fluids. Several automated platforms for the identification of micro-organisms are currently used in Latin America. Using these methods depends on the level of medical care, the type of patients, and the characteristics of the hospital. In

our region, not all countries have available technology because of its high requirement of technical support. This ends up resulting in higher costs when comparing to traditional manual methods. Therefore, it is indispensable to study cost-benefits, evaluate the clinical impact, the use and rationalization of the resources offered in each country. Hence the importance of education, communication and sharing experiences between colleagues in our region and the rest of the world.

S071

Liquid Profiling of Circulating Nucleic Acids and Its Role for Laboratory Medicine

Michael Neumaier

Medical Faculty Mannheim of Heidelberg University, Institute for Clinical Chemistry, Germany

The detection of malignant disease is a daunting challenge in diagnostics. Recent advances in bio-analytical technologies and our increasing understanding of tumour biology now allow to decipher the complexity of malignant growth both at the genotype and phenotype level.
The classical serum tumour-associated antigens were insufficiently specific surrogate markers of malignant growth with limited clinical utility. The advent of "circulating nucleic acids in serum and plasma" (CNAPS) and in other bodily fluids reflecting tumour-specific genotypes now allows to detect - by Liquid Profiling - individual molecular defects including those representing druggable targets. Liquid Profiling (aka liquid biopsy) of tumour mutations qualifies as actionable health information with high relevance for the choice of anti-tumour drugs. Probably most, if not all malignant tumours change under the selection pressure of cancer treatment thus requiring close monitoring of minimal residual disease to detect therapy resistance early for continuous clinical decision-making. Medical laboratories need to meet the analytical and diagnostic challenges of this modern oncology. This includes continuous further development and validation of highly sensitive and specific methods for detection, rigorous quality assessment in both Preanalytics and Analytics as well as willingness and competence to enter into the medical dialogue to counsel with the physician at the bedside. Very much comparable to the antibiogram of the microbiologist in the "rat race" to treat microbial infections, Laboratory Medicine should perceive CNAPS as a new (still largely un-known) universe of biomarkers offering related clinical opportunities in systemic disease. Also, recent evidence suggests that the combination of genotype from CNAPS with phenotypic data traditionally obtained in the laboratory may be of value for an earlier detection of malignant disease. The implications of recent findings will be discussed in details.

S072

Generation and Utility of Biological Variation Data with Emphasis on Personalized Reference Intervals and Reference Change Values?

Sverre Sandberg

Norwegian Quality Improvement of Laboratory Examinations (Noklus), Norway

After the strategic conference of EFLM in 2014, a WG and a Task Group (TGs) worked with the following topics: a) to develop a critical appraisal check list to evaluate literature on biological variation, b) to evaluate the literature on biological variation and extract essential information from the papers as well as summarise the information and c) to generate a new biological variation database (BVD). The EFLM BVD should contain essential information about the biological variation and derived performance specifications for different measurands as well as the evidence behind. Different groups are established for different measurands. The groups have used a critical appraisal list categorised papers as A, B, C and D depending on their methodological quality, with category A papers indicating high-quality and D poor quality. The BVD is now established on the EFLM web-site. The critical appraisal check list can also be followed when you are setting up new studies on biological variation. The groups are now also working to establish new methods for generating biological variation data as extraction information from Big Data and by using a Bayes approach to calculate biological variation. Information from these different approaches can be used to generate data useful establishing analytical performance specifications, reference change values, index of individuality and personalized reference intervals.

S073

Patient Focused Optimisation of Laboratory Services

Ian Watson

University Hospital Aintree, Liverpool, United Kingdom

Increasingly patients are engaging with their healthcare, often encouraged by their healthcare providers and governments, as an approach to managing health resource utilization, intimate to such access are results of laboratory medicine investigations; while having access to results satisfies engagement, comprehension of the significance of the results in their personal context is challenging and the evidence indicates that patients have poor understanding of their results; even when explained they mis-recall the significance and risk associated with them, this undermines the expected benefit of access and positive engagement.

To address this difficulty the patient requires knowledge and comprehension support; Patient Focused Laboratory Medicine (PFLM) envisages specialists in laboratory medicine providing direct access for patients to their expertise (see CCLM 2019; 57: 383). Modes of provision and methods of delivery are being explored through the EFLM Working Group on PFLM to encourage widespread adoption of such an approach. This includes proposals for providing personalised comments, within agreed clinical practice guidelines, exploring the presentation of numeric information in non-numeric formats and readily understood physio-pathological background to organ function, metabolism and treatment. Guidance for those developing such an approach are also considered.

S074

Tumor Markers, Challenges in Standardization and Assays

Lakshmi V Ramanathan

Memorial Sloan Kettering Cancer Centre, New York, USA

The quest for the ideal tumor marker that is 100% specific, highly sensitive, easy to measure and affordable still continues. In spite of continuing technological improvements in the currently available assays, there is considerable variability among assays making it very challenging for physicians to interpret results.
In this session, we will review with clinical cases, current challenges in traditional as well as new tumor marker assays due to assay limitations as well as lack of standardizaion. Possible harmonization of these assays will be discussed that can serve as a practical solution that would allow clinical comparison of the different methodologies.
With potentially new tumor markers on the horizon, this session will assist you in understanding standardization of assays in detail. This should be encouraged because it makes inter-laboratory testing clinically useful, clinical collaboration feasible and clinicians will more readily accept the data.

S075

The Significance of Aberrant Routine Laboratory Test Results in Oncology

Qing Meng

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Cancer biology involves cancer specific genetic mutations, tumor biology, and pathology. Thus, there are characteristic and unique changes including tumor markers and genetic mutations. While we always focus on these specific biological and pathological changes, cancer is also very heterogeneous and undergoes general biochemical changes. These changes reflect the abnormalities of routine laboratory results. These routine laboratory tests can be used as a part of a routine checkup to provide information about a patient's general health; be in aid of making the diagnosis; help physicians manage or adjust treatment plan; and predict patient outcome. This session will review those general biochemical changes and discuss how those abnormal laboratory results can be used in cancer patient management. In some types of cancer, those routine biochemical changes can be even indicative and very valuable for cancer diagnosis and management. Participants will be able to understand and interpret the abnormal changes of general biochemical test results in cancer patients. Moreover, this session will help you to develop investigation algorithms of these abnormal results for better patient management in oncology.

S076

Novel Microbial Biomarkers in Cancer Diagnosis and Monitoring

Yi-Wei Tang

Memorial Sloan Kettering Cancer Center, New York, USA

Widespread adoption of rapid and accurate biomarker tests for cancer detection and diagnosis as well as therapy selection and monitoring may lead to a paradigm shift in the way that clinical medicine is practiced. Detection and characterization of oncogenic microbial organisms have been used in cancer diagnosis and monitoring. Recent technological advances, especially in the fields of genomics, have made it easier to identify microbial biomarkers in relation to cancer pathogenesis. These biomarkers could be either specific pathogen elements or host microbiome profiles that associated with cancer development and advancement. In this presentation, I will first describe an isothermal, multiplex amplification assay for detection and genotyping of human papillomaviruses (HPV) in formalin-fixed paraffin-embedded tissues. I will then to describe detection of high-risk HPV in non-invasive specimens such as mouth washes as a screening tool for HPV-associated oropharyngeal carcinoma. I will finally provide take home messages on specific pathogen elements or host microbiome profiles as potential biomarkers for cancer diagnosis and monitoring.

S077

RNA Epitranscriptome Landscape of Glioma Stem-like Cells: Essential Role for m6A Modification in Gene Expression

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Glioma stem-like cells (GSCs) represent a primitive form in the hierarchy of tumor cells, which can initiate and populate the tumor. GSCs are highly tumorigenic and contribute to poor therapy response of glioblastoma (GBM) patients. Dissecting the cellular mechanisms that sustain GSCs growth is essential for identifying therapeutic targets. GSC is a flexible cellular system that undergoes differentiation (differentiated glioma cell/DGC) in the presence of serum and can revert to stem-like cells through reprogramming/dedifferentiation. The m6A epitranscriptome regulates the expression of a plethora of genes by altering various steps of RNA metabolism, such as mRNA stability, RNA export, alternative polyadenylation, splicing and translatability. The m6A modification is facilitated by two methylases - METTL3 and METTL14 - while demethylation is mediated by ALKBH5 and FTO. Despite recent advances in m6A biology, the regulation of crucial RNA processing steps by the RNA methylase METTL3 in glioma stem-like cells (GSCs) remains obscure. In this study, we investigated the METTL3-mediated m6A regulome in GSCs by m6A RNA immunoprecipitation (RIP) sequencing combined with transcriptome analysis after METTL3 silencing. We present here the impact of METTL3 silencing on global m6A modification, transcriptome, various RNA processing steps and important functional pathways. Collectively, our study uncovered the crucial collaborative functions of METTL3-dependent m6A modification in RNA metabolism including RNA stability specific for GSCs.

S078

Cathepsin B: A potential Therapeutic Target in Paediatric AML

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Cathepsin B (CTSB) has been used as a biomarker for predicting survival outcome in several solid tumours. Inhibition of its activity and/or down regulation of its expression has also been reported to decrease proliferation and survival of these tumour cells. To explore the clinical utility and role of this protease in paediatric AML, we measured its enzymatic activity and mRNA expression in mononuclear cells isolated from peripheral blood (PBMCs) as well as bone marrow aspirate (BMMCs) of paediatric AML patients and PBMCs of healthy controls. Our results revealed elevated CTSB activity ($p<0.01$) and over expression of its mRNA ($p<0.01$) in AML patients. Increased CTSB levels in patients were associated with inferior survival outcomes. Moreover, chemical inhibition of CTSB activity in leukemic cells, reduced their survival and induced apoptosis. We further demonstrate inhibition of CTSB activity by chemotherapeutic agent doxorubicin in these cells. Docking and simulation study confirmed binding of doxorubicin to CTSB with higher affinity than its inhibitor CA-074Me, thereby indicating that cell death induced by this drug may at least partly be me-diated by CTSB inhibition. CTSB, therefore, may serve as a marker of disease prognosis and an attractive chemotherapeutic target in paediatric AML.

S079

Cancer Immunotherapy: Opportunities and Challenges using Unmodified Gamma Delta T Cells

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Cathepsin B (CTSB) has been used as a biomarker for predicting survival outcome in several solid During the past decade anti-cancer immunotherapy has evolved from a promising therapeutic option to robust reality that has changed treatment outcomes. Gamma delta T (γδ T) cells constitute a separate lineage of T lymphocytes which differ from conventional Alpha beta T cells with regard to T cell receptor repertoire, antigens recognition, tissue localization and effector functions. γδ Tells infiltrate tumours and exhibit potent antitumor activity, hence are becoming attractive candidates for cancer immunotherapy. Human Vg9Vd2 T cells recognize metabolites that are overproduced by transformed cells, specifically the pyrophosphate intermediates, such as isopentenyl pyrophosphate (IPP), of the mevalonate pathway involved in cholesterol synthesis and protein prenylation. Human Vg9Vd2 T cells recognize these intermediates in tumor cells and mount potent cytotoxic responses against tumors.. In the present study we have investigated the effect of hypoxia and HDAC inhibitors on the effector functions of human γδ T cells. HDAC inhibitors inhibited the antigen specific proliferative responses of γδ T cells and cell cycle progression. In antigen activated γδ T cells, the expression of transcription factors (Eomes and Tbet) and effector molecules (perforin and granzyme B) was decreased upon treatment with HDAC inhibitors. The anti-tumor cytotoxic potential of γδ T cells was attenuated which correlated with enhanced expression of immune checkpoints PD-1 and PD-L1 in γδ T cells. Interestingly, PD-1 blockade improved the anti-tumor effector functions of HDAC inhibitor treated γδ T cells, which reflected in the increased expression of Granzyme B and Lamp-1. In summary, these unique features make γδ T cells attractive candidates for adoptive cancer immunotherapy. Combination of HDAC inhibitors and immune

checkpoint blockade along with γδ T cells can further increase the efficacy of this therapeutic modality.

S080

Changing Lab Environment- Perspective from Young Scientist

Marie Lenski

Faculty of Pharmacy, University of Lille, France

Laboratory medicine is a specialty that is burgeoning. New biomarkers and technologies that are more accurate are in continuous development. However, the new technologies also bring new challenges. For example, in molecular genetics, the next-generation sequencing (NGS) has been introduced about 10 years ago and has revolutionized the diagnosis of hereditary diseases. The progress of NGS is leading to the increase of discovery of number of genes associated to human inherited disorders and to the elucidation of molecular basis of complex disease. Moreover, since on NGS platforms it is possible to perform a parallel sequencing of different target regions, the use of NGS in clinical laboratories has became increasingly widespread, used in diagnostics of infectious diseases, immune disorders, human hereditary disorders and in non-invasive prenatal diagnosis, and in the therapeutic decision making for somatic cancers. New professional figure of bioinformatics has emerged and the role of specialist in laboratory medicine is evolving. In France, national genomics networks have emerged to meet the needs of clinicians in the management of patients. In this talk, we will deal with the changing patterns of laboratory medicine using the example of molecular genomics that now require a team-based approach, including clinicians, specialist in molecular genetics, in cytogenetics and in bioinformatics.

S081

Are we Prepared? How can we do it?- Step to the Future

Santiago Fares Taie

Fares Taie Instituto de Análisis, Argentina

Laboratory medicine is a fast-changing profession with dynamic roles and new technologies appearing every year. Progress in science has changed the scope of laboratory medicine in the last 100 years, and is challenging to change even faster in the next decades. Some examples are point of care, personalized and predictive medicine, nanotechnology, biochemistry 2.0, Big data and Artificial Intelligence, among others.

Young Scientists have the ability to adapt fast and be creative. It is a special moment in science to make significant improvements in laboratory medicine with the use of new technologies, networking and professionals prepared to face the new advances.

This session will present the Argentinean YS situation and how we prepare for the future laboratory medicine.

S082

Core Laboratory Concept

Giulia Sancesario

Tor Vergata University of Rome, Italy

Recent advances in analytical technology have led to a new scenario in Clinical Laboratory Science, which aims at the total automation of routine laboratory activities. This new concept for clinical laboratories allows integrating the need to optimize costs for laboratory tests together with the need to reduce medical errors and reduce the biological risk associated with exposure to biological material that could be dangerous for technicians.

The Core Laboratory Concept summarizes the modern idea of a clinical laboratory with the complete automation of the analytical phase. This new concept has revolutionized the organization of clinical laboratory, with an exponential growth of diagnostic tests and the identification of new markers.

In the era of precision medicine, the value of laboratory test is fundamental for driving clinical decision and therapies. Finally, the laboratory medicine is an open Clinical Laboratory Science, and constitute the link between patient and clinic in the diagnostic process.

S083

Bringing Your Lab to the Operating Room

Joe El-Khoury

Yale University in New Haven, CT, USA

This session will describe how Yale University optimized intraoperative parathyroid hormone testing to provide a 12-minute turn-around-time (TAT) for our endocrine surgeons versus the 30-45 minute TAT that most labs provide via central lab testing. Testing procedures, workflows, personnel scheduling, and an overview of our facilities redesign will be presented.

Learning Objective 1: Discuss the clinical utility of rapid intraoperative parathyroid hormone testing

Learning Objective 2: Describe how intraoperative parathyroid hormone testing is performed at Yale

Learning Objective 3: Develop a plan to optimize intraoperative parathyroid hormone testing at your institution

This would be focused on how we implemented intraoperative parathyroid hormone testing near the operating room (lab connected by a window, see attached pictures). This reduced our turn-around-

time to 12 minutes and provided an estimated cost savings of $2,700/patient by improving efficiency of the operating room and faster patient recovery.

S084

Circulating Tumor Biomarker and Its Clinical Application

Wei Cui

National Cancer Center, Cancer Hospital, Chinese Academy of Medical Sciences, China

In recent years, with the clinical application of targeted drug therapy for cancer, circulating tumor cells (CTC) and circulating tumor nucleic acid (ctDNA), as liquid biopsy parameters, have received much attention in clinical application because of their non-invasiveness, dynamics, and feasibility. However, exosomes are still in the stage of clinical transformation research. Because the contents of ctDNA, CTC and exosomes in the peripheral circulation are very small, and the heterogeneity of tumor exists, it is very difficult for conventional methods to achieve high sensitivity and specificity in detecting these parameters. A sensitive and specific liquid biopsy technique is very important to meet clinical needs. The clinical application and research of liquid biopsy technology include the following aspects: 1) guiding clinical medication, real-time monitoring of sensitive genes and drug-resistant clones has become a major advantage of liquid biopsy; 2) efficacy monitoring: evaluation of tumor load; 3) prognosis judgment; 4) early screening. Although liquid biopsy technology has gradually used in clinical practice, the overall sensitivity is not high. Laboratory operation processes should be standardized. We need to pay more attention to the interpretation of results and strengthen the communication between clinical and laboratory.

S085

Application of Exosomes in the Diagnosis of Prostate Cancer

Zhuo Li

Xijing Hospital, China

Prostate cancer(PCa) is the most common malignant tumors in male urinary system. Novel and non-invasive biomarkers with higher sensitivity and specificity for the diagnosis of PCa are urgently needed. Exosomes are widely recognized as a good source of tumor biomarkers. In the present study, exosomes were isolated from the serum of patients with PCa, benign prostate hyperpla-sia (BPH) and healthy controls (HC). Then, quantitative reverse transcription-polymerase chain reac-tion (qRT-PCR) test showed that the expression of miR-141 was higher in exosomes than in the whole serum ($P<0.05$). Exosomal miR-141 in serum was significantly higher in the patients with PCa compared with BPH (3.85-fold, $P<0.05$) and the HC (4.06-fold, $P<0.05$), and also significantly higher in metastatic PCa than the localized PCa ($P<0.05$). These results demonstrated that, compar-ing with the whole serum, circulating exosomes could be a more suitable potential non-invasive biomarker for the detection of PCa. Based on this, a new technology "Tethered Cationic Lipoplex Nanoparticle" (TCLN) chip was explored. Three mRNAs and four miRNAs in serum were selected as candidate markers and then detected by TCLN chip. The results showed that exosomal miR-141 could distinguish PCa from HC (AUC=0.739) and also from BPH (AUC=0.721). Four candidate biomarkers could distinguish PCa from HC ($P<0.05$), including PCA3 (AUC=0.806), Let-7c (AUC=0.711), PSA (AUC=0.725), and miR-375 (AUC=0.648). In combination of these four bio-markers, the diagnosis model has the diagnostic performance of AUC=0.843/0.747 of PCa vs HC and PCa vs BPH, respectively. These results indicated that the circulating exosomal RNAs can serve as a novel and non-invasive biomarker in the diagnosis of prostate cancer. TCLN chip can realize "in situ capture" of exosomes, with no need of RNA extraction and amplification, which helps to achieve fast analysis of the circulating exosomal RNAs biomarkers.

S086

Laboratory Diagnosis of Mitochondrial Genetic Disorders

Hezhi Fang

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Mitochondrial genetic disorder is one of the most common metabolic genetic disease with an estimated prevalence of 1 in 5,000 live births. Both mitochondrial DNA and nuclear DNA mutations that lead to mitochondrial malfunction are responsible for mitochodnrial genetic disorders. Clinical and laboratory diagnosis of mitochondrial genetic disorders is difficult worldwide due to the disease heterogeneity. Although thousands of mutations in 100-200 mitochondrial disease causing genes were identified, mutation spectrums in regard of Chinese population are lacking, which may limit the precision diagnosis of mitochondrial genetic disorders in China. To perform molecular and genetic diagnosis of mitochondrial disorders, we applied a combined strategy of whole exome sequencing technology and mitocondrial pathology analysis in our lab. By using this strategy, multiple novel mitochondrial disease causing genes such as TIMMDC1 and TOM70A, were identified and functional confirmed.

S087

Accuracy Monitoring Program for HbA1c Reagents

Kyunghoon LEE

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The hemoglobin A1c (HbA1c) level is widely used to diagnose diabetes mellitus and monitor glycemic control in diabetes mellitus patients, and various methods are used for its determination. According to the criteria for the diagnosis of diabetes at American Diabetes Association, HbA1c test should be performed in a laboratory using a method that is NGSP certified and standardized to the DCCT assay. To achieve a uniform international standardization, the IFCC established a working group on HbA1c standardization in 1995. The IFCC and NGSP have being making efforts to standardize HbA1c test and has certified the methods and reagents for HbA1c, respectively. Since 2011, KCDC (Korea Centers for Disease Control & Prevention) and KSLM (Korean Society for Laboratory Medicine) have been working to establish a standardization system for the laboratory medicine in Korea. As a result, KCDC has the national laboratory for standardization, IFCC HbA1c Network Laboratories certified by IFCC. And, the accuracy of the HbA1c in the clinical laboratories has been improved through the project of the External Quality Assurance (EQA). However, it is not enough for the standardization of HbA1c in Korea to manage this EQA project. Therefore, our society need to assess the performance of HbA1c analyzers and certify each lot of reagent and calibrator for HbA1c standardization like IFCC and NGSP. Last year, the twenty-four commutable frozen whole blood specimens were sent to the selected five manufactures and all specimens' reference values were assigned by KCDC national laboratory for standardization. According to the IFCC certification criteria, the observed bias, precision, and linearity were assessed. In this year, more than twenty manufactures participate in the certification of the performance of HbA1c analyzer. We will share the evaluation results of a variety of HbA1c analyzers, reagents and calibrators.

S088

Accuracy Monitoring Program for Creatinine Reagents

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The diagnosis and treatment of chronic kidney disease (CKD) is critical to ongoing global public health. The effect of a variation in creatinine results on the estimation of glomerular filtration rate (eGFR) which can impact on classification of patients into CKD stage. The creatinine assays providing the more accurate results for GFR estimation is important. The creatinine certification program is part of the creatinine standardization program by KCDC and KSLM to improve the accuracy and consistency of creatinine measurements. This study is a pilot project to assess the performance of assays and certify each lot of reagents and calibrators. The commutable frozen serum samples were sent to the manufacturers and the observed bias and linearity were compared to predefined error limits. Twenty four samples, with 1.0 mL each, with creatinine concentrations <5.0 mg/dL (range 0.56-4.86) were sent to the manufacturers followed by analyzing the specimens one run per day. Reference values were assigned to the survey specimens by the reference laboratory, which uses isotope dilution gas chromatography/mass spectrometry methods. The criteria for acceptable bias performance was the minimum performance goal for total allowable error from the NKDEP's Laboratory Working Group. When 22 or more of the total 24 samples met the criteria, the overall performance for bias was considered passing. We estimated the linearity or nonlinearity using College of American Pathologists linearity evaluation criteria. The one-fourth the goal for total error of 25% was used to specify limits. We evaluated about 50 creatinine assays from 14 manufacturers based on the combination of instruments and different lots of reagents and calibrators. The requirement that clinical laboratories should use creatinine assays certified by KCDC and KSLM has been added to the checklists of laboratory accreditation program since 2019.

S089

Accuracy Monitoring Program for Lipids Reagents

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The accuracy of the test results must be maintained so that reliable policies can be established and the health of the people can be improved. The project of the External Quality Assurance (EQA) is the primary method for improving the accuracy, but it may not perfect method to ensure accuracy since most participating laboratories may show similar errors together. Therefore, efforts are being made to improve the accuracy of the manufacturer's product. Sometimes the reagents or calibrators manufactured and delivered by the manufacturer do not fully satisfy the performance criteria. Therefore, assessment program for the manufacturer's product are required to verify them. In Korea, various combinations of calibrators, reagents and equipment are used for lipid measurement. In the case of domestic manufacturing reagents, it is difficult to collect samples needed for international certification of CRMLN, and it is often difficult to obtain CRMLN certification due to high cost. Therefore, it is necessary to develop domestic quality assessment program that will evaluate the accuracy of various combinations of calibrators, reagents and equipment. This

study is a pilot project to assess the performance of assays and certify each lot of reagents and calibrators of lipid test such as total choesterol, HDL-, LDL-cholesetrol, and triglyceride. The ten commutable frozen serum samples were sent to the manufacturers and the observed bias, precision and total were compared to predefined error limits. Reference values were assigned to the survey specimens by the reference laboratory, which uses isotope dilution gas chromatography/mass spectrometry methods. The criteria for acceptable performance was the NCEP recommendations. In the pilot study, we evaluated only 6 combinations of calibrator lots, reagent lots and equipment. By operating a quality assessment project for lipid products, the company can improve the accuracy of reagent and encourage the standardization of lipid testing.

S090

Alcohol Related Diseases and Biological Markers

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Alcohol is a very dangerous cytotoxic substance damaging the organism both acutely and chronically. The direct effect manifests itself mainly in changes to biological membranes and influencing their fluidity, potentially also by intercellular interactions with the possible alcohol-induced malnutrition. Alcohol consumption is associated with more than 200 diseases, including tumors, hypertension, liver cirrhosis, brain damage and diabetes and injuries. Drinking alcohol during pregnancy may cause fetal alcohol syndrome with an incidence of 3.7 per 1000 live births in Europe. Alcohol is most commonly associated with liver damage - steatosis, alcoholic hepatitis, cirrhosis, and hepatocellular carcinoma. The influence of ethanol on the cardiovascular system is very much discussed in terms of its cardioprotective effects at very small doses (20 g/day). The protective mechanism is enabled by an increase in HDL, ApoAI, paraoxonase activity and adiponectin and lowering LDL; by an antithrombotic effect; and by an increase in insulin receptor sensitivity. The recent Wood et. al study (2018) processed data from 83 studies on 599,912 participants shows the decrease only in myocardial infarction and consuming higher doses then 100-200g of alcohol/week means a higher risk of heart attack, atrial fibrillation. Chronic alcohol consumption is associated with 10 % of tumors in males and 3 % in females. Alcohol is considered as a risk factor in upper gastrointestinal tract tumors (UADT-25-68 % of them are associated with alcohol), hepatocellular carcinoma, colorectal carcinoma and breast cancer. There are a number of mechanisms - acetaldehyde, oxidative stress, activation of procancerogens, folic acid deficiency, decreased levels of retinoic acid, allelic variants of ADH and AlDH genes, etc. Laboratory markers of alcohol consumpition include classical as ALT, MCV, GGT but also new markers as CDT, ethyl glucuronide. Combination of GGT and CDT or CDT and ethyl glucuronide.

S091

Perceived Response to Alcohol as a Phenotype for Risk of Alcohol Related Problems in Young Japanese Adults.

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A Level of Response (LR) to alcohol is genetically influenced and low LR is a possible predictor of alcohol use disorder (AUD). Further, alcohol's stimulating and sedating responses have been found to predict increased binge drinking frequency and AUD risk among social drinkers. However, this relationship has not been examined in Asian samples. Further, variations in the alcohol dehydrogenase (ADH1B) and aldehyde dehydrogenase (ALDH2) genes may influence the LR to alcohol by increasing levels of acetaldehyde during alcohol metabolism. This presentation will examine: 1) the relationship between the genetic variation of ADH1B and ALDH2 and subjective response to alcohol, 2) the association between blood acetaldehyde levels and subjective response to alcohol and 3) the association between the subjective response to alcohol and AUD risk in Japanese young adults studied prospectively.

Subjects included 424 healthy Japanese college students aged 20-23 years old. Using the alcohol clamp method, we infused diluted alcohol (6%) to maintain a blood alcohol concentration (BAC) at 50 ± 5 mg% for 3 hours. The LR to alcohol was assessed using the Biphasic Alcohol Effects Scale before infusion began and every 30 minutes during the infusion procedure. Subjects have been followed using the Alcohol Use Disorder Identification Test (AUDIT). Subjects with inactive ALDH2 showed stronger sedative subjective responses to alcohol than those with active ALDH2. The sedative effects of alcohol were significantly correlated with blood acetaldehyde levels. Examining the relationship between subjective response to alcohol and AUDIT scores at follow up revealed that subjects with higher AUDIT scores showed weaker sedative responses to alcohol than those with lower AUDIT scores.

Our findings suggest that blood acetaldehyde levels influence sedative LR to alcohol and acetaldehyde might play a major role in protective effect from alcohol problems.

S-092

Alcohol and Adducts: Not Just Bad News for the Liver!

Simon Worrall

The University of Queensland, Australia

The long-term drinking of excessive amounts of alcohol is commonly known to be associated with injury to the liver, and brain, but it is less well known that it also injures other tissues such as cardiac and skeletal muscle, the pancreas and stomach. Despite alcohol being associated with negative effects for a long time researchers have only partially delineated the mechanisms involved in its toxic actions. One of the phenomena associated with the consumption of alcohol is the modification of cellular components such as proteins by reactive substances produced directly or indirectly during its metabolism. The main site of alcohol metabolism is the liver and the primary metabolite produced is acetaldehyde, together with free radicals. Indirect metabolites such as malondialdehyde and 4-hydroxynonenal are also produced through the attack of these free radicals on unsaturated fatty acids. All of these compounds are highly reactive and can form reversible and/or stable modifications on proteins and other molecules. These modifications can lead to loss of cellular function and the formation of neoantigens. Many researchers have shown the presence of these modifications in the liver, particularly those formed by the reactions of acetaldehyde, together with reactive antibodies in the circulation. Furthermore, animal models have shown that the induction of high titres of these antibodies in alcohol-fed rats can result in liver injury in these animals but not in alcohol naïve ones. More recent studies have now also identified similar modified proteins in extrahepatic tissues such as muscle and brain which have much lower capacities for alcohol metabolism. Whether these modified proteins play a role in injury to these tissues remains to be resolved. It has also been suggested that modified proteins and their reactive antibodies could be used as biomarkers of alcohol consumption in a manner analogous to glycated haemoglobin in diabetes.

S093

CAR T-cell Therapy: Is it a Real Opportunity to Treat Hematological Malignancy?

Sadhna Sharma

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Understanding the intricate relationship between the immune system and the tumor cells have accelerated many opportunities of cancer treatment. Turning on immune cell against the cancer cell either through antibodies mediated therapeutics or re-infusion of trained autologous immune cells into the cancer patient are excellent examples of cancer immunotherapy. Among all the cell based therapies, the genetically engineered Chimeric Antigen Receptor (CAR) bearing T-cells (CAR T cell) for targeting cancer cells have been emerged as the successful therapy in haematological malignancy. In 2017, two anti-CD19 CAR T-cell therapies have been approved by the US Food and Drug Administration (FDA)-Tisagenlecleucel (Kymriah, Novartis) for the treatment of acute lymphoblastic leukemia (ALL) in children and young adults and Axicabtagene ciloleucel (Yescarta, Kite Pharma) for adult patients with relapsed/refractory large B-cell lymphoma, both using CD19 antigen overexpressed on malignant B-cells. After that, a burst in the experiments have been reported to test this magical therapy in various cancer types including solid tumors. In general, CAR T-cells when infused in patient, act as a live drug which proliferate and destroy tumour cells while sparing normal cells. However, recent follow up of clinical trials are showing that the tumor cells are evolving with tricky mechanisms to get survival advantage against CAR T cell. To improve the efficacy, safety and specificity of CAR T cell therapy further research is needed in context of tumor antigen selection, cancer types and personalized immunity of individual cancer patient. Having high cost of treatment in other countries and increasing number of cancer patients in India, we set out to develop CAR T-cell therapy for B-cell Chronic lymphocytic leukemia (CLL). Consequently, this will establish a paradigm infrastructures of CAR T cell therapy for better affordability to economically poor patient.

S094

Micro RNAs in Multiple Myeloma: From Diagnosis to Therapeutics

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Multiple Myeloma (MM) is second most common hematological malignancy characterized by uncontrolled proliferation of abnormal plasma cells in bone marrow (BM). Myeloma cells require BM niche consisting of proteoglycans, cytokines & growth factors for their growth. Earlier, we reported diagnostic potential of a chondroitin sulfate proteoglycan, Versican (VCAN) in MM. Hence, we aim to identify the theranostic potential of microRNAs (miR-144, miR-199 & miR-203) associated with VCAN for which no report is available in MM. 30 patients & 20 controls were recruited. The microRNAs and VCAN expression were examined in serum, BM supernatant fluid (BMSF) and BM mononuclear cells (BMMNCs). ROC curve was plotted to determine diagnostic potential of microRNAs. In addition, miRNA inhibitors were used in MM cell lines for their therapeutic role. The cancer associated hallmarks and the downstream signaling cascade affected by miR inhibitors were examined. microRNAs associated with VCAN were significantly downregulated in both BM and blood of MM patients while VCAN levels were

significantly higher and negatively correlated with microRNAs. The expression of microRNAs showed negative trend while VCAN levels showed positive trend with severity of disease. miR-203 showed significant correlation with myeloma-associated parameters and also showed optimum sensitivity and specificity for diagnosis of MM in serum. The inhibitors for miR-144 & miR-199 in MM had shown upregulation of VCAN in myeloma cells followed by alteration in cancer hallmarks in favour of myeloma progression and activation of FAK/STAT3 signaling pathways. The correlation of cell-free miR-203 with myeloma clinical parameters along with optimum sensitivity and specificity affirms its non-invasive diagnostic potential in MM. The microRNA inhibitors (miR-144 and miR-199) resulted in myeloma progression via upregulation of VCAN, hence, can be of therapeutic utility and can be a novel approach for better management of MM.

S095

Micro RNAs at Cross-road of Innate and Adaptive Immunity in Solid Tumour

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A major mechanism of tumor development and progression is silencing of the patient's immune response to cancer-specific antigens. Defects in the so-called cancer immunity cycle may occur at any stage of tumor development. Within the tumor microenvironment, aberrant expression of immune checkpoint molecules with activating or inhibitory effects on T lymphocytes induces immune tolerance and cellular immune escape. Innate and adaptive immune cells not only infiltrate tumors themselves, but also the tumor milieu. miRNAs are expressed at different levels in multiple cell and tissue types and are also involved in tumorigenesis and the progression of aggressive cancer phenotypes. These molecules have been observed to affect both innate as well as adaptive immune systems check-points. In tumourgenesis, these molecules are associated with cell growth, proliferation, migration, invasion, and apoptosis; and can even alter tumour immune cells development. RNA sequencing has confirmed that miRNA profiles can serve as highly sensitive and specific diagnostic and prognostic biomarkers. Because these molecules can be detected in diverse tumor tissues compared to normal samples and are associated with different clinicopathologic characteristics, differentially expressed miRNAs can be employed to assess the pathogenesis of number of cancers including solid tumours like breast cancer or oral squamous cell carcinoma and also help assess clinical prognosis. Recently there has been a lot of interest in cancer immune-therapy and miRNA being regulators of important immune-check points, are also being explored as therapeutic targets, with some in phase one trial already. Thus have indicated their latent therapeutic value for successful clinical translation. The current talk would be focussing on role of micro RNAs as regulators of innate and adaptive immunity in solid tumours, in diagnosis, prognosis and their future as therapeutic targets.

S096

Pitfalls in Interpretation of Laboratory Test Results

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The disease diagnosis is done based on preliminary data, listing of problems, diagnostic hypothesis, laboratory tests and subsequent confirmation or review of the diagnosis. In this process of disease diagnosis, it is expected that the laboratory test results should agree with the clinical picture and each other. If not, the accuracy of the test results are questioned by the clinicians. There are various factors that cause unexpected or unusual test results. These factors may be pre-analytical, analytical or post-analytical. Patient factors like age, gender, physiological changes, intercurrent illnesses, drugs and nutritional status and specimen factors like sample collection and handling and sample storage are some examples for factors which influence test results. In this presentation several clinical cases with abnormal test results are discussed in the aim of avoiding fit falls in the diagnosis.

S097

Adventures of Chyle

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Dietary triglycerides (TG) are emulsified in bile and hydrolyzed by intestinal lipase to form a mixture of free fatty acids (FFA) and monoglycerides. These are re esterified in the enterocytes after absorption and incorporated with phospholipids, cholesterol esters, and apolipoprotein B-48 to form a nascent chylomicron. Nascent chylomicrons secreted from the enterocytes into the lymphatic vessels originating in the villi of the small intestine, and are then secreted into the bloodstream via the thoracic duct. Soon after entering the circulation these particles acquire Apo C and Apo E from circulating HDL. Lipoprotein lipase (LPL) of endothelial tissue activated by Apo CII on the surface of chylomicron to rapidly hydrolyze triglycerides which is the main constituent of chylomicrons. Remnant Chylomicrons send back to the liver and taken up by Apo E receptor.

Any defect in the above pathway will result in accumulation of chylomicrons leading to hypertriglyceridemia or elevated triglycerides in body fluids giving milky appearance. Hypertriglyceridemia, chyluria, chylothorax, chyloperitonium are various presentations of defects of above path giving a clue to locate

the problem.
Marked hypertriglyceridemia results in genetic mutations of LPL or Apo E receptor. Blockage to lymphatic drainage at various places of the mesenteric circulation leads to chyloperitonium or chyluria giving a clue to diagnose an important underlying disorder. Chylothorax result in damage to thoracic duct due to various reasons. Even though elevated TG is a challenge to measurement of other analytes in serum and body fluids, systematic approach will open up the path to resolve the hidden puzzle.

S098

Challenges in Electrophoresis Interpretation

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Serum protein electrophoresis is the main investigation used in the diagnosis and follow up of patients with plasma cell myeloma in the National Cancer Instittue Maharagama. During the last 12 years the incidence of multiple myeloma has increased significantly with resulting increase of sample numbers. As this hospital is the tertiary care hospital in the cancer management in Sri Lanka we get the bulk of plasma cell disorders from all over the country with some unusual presentations.
This presentation mainly focus on unusual cases received over a period of one year including several cases of biclonal and triclonal gammapathies.
After introduction of immune-fixation electrophoresis and urine protein electrophoresis/immune-fixation the diagnostic power improved and the presentation will show some cases of interest.

S099

Preparation of Sample for Mass Spectrometry

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LC-MS/MS is a well-established tool for the identification and quantification of small molecules, as well as macro molecules, in both research and industry field. Thanks to technology breakthroughs, it has recently moved into the healthcare arena and become the technique choice for many small molecule testing in clinical application. This technique, historically, implementation the challenge for clinical research laboratories has been the development of reliable, reproducible and cost-effective sample preparation methods. In clinical, molecule analysis can be performed on a variety of sample types in a clinical research setting-including whole blood, serum, plasma, urine and CSF. To avoid clogging the chromatography column, the most straightforward of sample preparation is to remove proteins and other constituents that may precipitate when injected into the LC mobile phase. Besides, due to profound effect of pH, buffer and aqueous composition of the sample on chromatography peak shapes, peak separation and retention times, without samples preparation processing prior to LC-MS/MS analysis these factors can influence the sensitivity, selectivity and robustness of assay. To overcome these issues, complex bio-fluids often need to be exchanged for an injection solution compatible with the LC method prior to injection. The most important issue for all the manager of institution are instrument maintenance. Effective pre-analytical sample clean-up can also make the need for servicing more predictable, which ideally limiting servicing to scheduled six-month preventive maintenance visits. Compared with a sudden, unexpected loss of sensitivity due to insufficient sample preparation, this avoids batch failures and unplanned downtime, which have the knock-on effects of more sample repeats, turnaround time delays and higher production costs.

S100

The Medical Surveillance of Emergency Poisoning Related with Drug Abuse in Taiwan - Development of a Screening Analysis by LC-QTOF MS for Abuse Substances

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New psychoactive substances (NPS) pose a challenge for forensic and clinical toxicologists, as well as for legislators. Although the exact prevalence of NPS abuse in Taiwan remains ill-defined, the trend of combining NPS with the domestic popular illicit drugs is becoming inevitable in Taiwan. With the increasing prevalence of NPS, poisonings or accidents related to drug abuse are increasing as well. It is crucial to develop a testing protocol in practical to be able to pick up a wide range of drugs to assist the medical diagnosis under emergency situation and drug abuse prevention in Taiwan.
The Taipei Veterans General Hospital was very honor to involved "The Medical Surveillance System of New Psychoactive Substance (NPS) Abuse in emergency department (ED) patients in Taiwan" project granted by FDA in 2017 and 2019. We supported this project by providing wide spectrum of drug screen up to 100 NPS and 14 classical abuse substances also made effort in educating of how to managing the NPS intoxicated patients in EDs.
This project will setup the novel method protocol of Liquid Chromatography Tandem mass spectrometry (LC/MS/MS) and Liquid Chromatography/Time of flight mass spectrometry (LC/Q-TOF HRMS) method to examine the urine specimen collected from any suspect of NPS intoxication cases among the emergency rooms in Taiwan. Therefore, this project could collect high resolution of mass spectra data and build its library, and each compound's

retention time. Besides classical abuse substances such as opiates, cannabis, benzodiazepines, Z-type hypnotics and amphetamines, The NPS substances are substituted phenethylamines, synthetic cathinones, hallucinogenic tryptamines, piperidines, opioid related substances, ketamine and related substances (in total of 114 substances).

S101

Clinical Application of Lab Developed Test in Cancer Precision Medicine

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Lung cancer is a prominent example of precision medicine among solid tumor malignancies. With the understanding of molecular medicine and the development of target therapy, the molecular testing becomes increasingly important. In the mention of target therapy efficacy, identification of "targets" and development of "darts" are two prior issues in the clinical practice. Therefore, customized lab developed test (LDT) is the most efficient strategy to satisfy clinical unmet needs. Stand on the clinical laboratory medicine views, providing accurate testing results for actionable mutation identification is the most important issue. Here I would like to share the experience in lung cancer precision medicine in Taiwan. Firstly, we developed a highly sensitive platform for EGFR mutation diagnostics in lung cancer. Based on clinical needs, we revised the testing panel based on lung cancer mutation profiling by adding EGFR, KRAS, BRAF, HER2 mutation hot spots. In the past 8 years, the service provided over 2500 cases/year to help patients to apply health insurances. Recently, with the discovery of novel fusion mutation and the utilization corresponding drugs, we further developed customized fusion mutation detection panel from formalin-fixed paraffin embedded RNA extracts. On the other hand, in the concern of the availability of surgical biopsy and the monitor of disease progression as well as the prediction of drug responsiveness, we also tried to develop non-invasive molecular diagnostics in cell-free DNA from peripheral blood. This progress not only enhances the detection sensitivity by modifying the previous platform but also identifies testing threshold for predicting cancer progressive disease and target therapy effectiveness. Taken together, laboratory R&D can satisfy the clinical unmet needs and we should keep in mind that the analytical and clinical reliability, feasibility, and practicability have to be always concerned during each step.

S102

Heparan Sulfate Proteoglycans Vascular Endothelial as an Early Marker of Plasma Leakage in Dengue Infection

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Plasma leakage is the main complication of dengue infection. Currently plasma leakage is monitored by hematocrit or ultrasonography (USG). Proteoglycan is a thin layer which cover luminal face of vascular endothelial cells. Heparan sulfate is the major component of proteoglycan. The aims of this study were to know whether heparan sulfate proteoglycan (HSPG) can be used as an early marker of plasma leakage, and to search any correlation between HSPG with tumor necrosis factor α (TNFα). This prospective cohort study was a part of the Community Based Dengue Study which have been approved by the ethics committee of the Faculty of Medicine, Universitas Indonesia. Forty subjects with fever less than 48 hours and positive NS1 determined by NS1 antigen Rapid Test, as inclusion criteria, who agree to participate by signing informed consent were enrolled for this study. Samples collection were done by consecutive sampling from February 2010 until January 2011. Dengue infection was confirmed by polymerase chain reaction (PCR). Subjects divided into two groups i.e. plasma leakage and without plasma leakage. Plasma leakage was determined by USG, or hemoconcentration, or hypoalbuminemia. Measurement of HSPG and TNFα were performed by ELISA on plasma which were collected from the first day until the third day of admission. Data distribution was analyzed by using Shapiro Wilk. Difference between group was analyzed by using Mann Whitney and correlation was analyzed using Spearman. The results showed that HSPG in plasma leakage group was significantly higher than that in group without plasma leakage ($p=0.001$). The HSPG cut-off value was 2179.73 pg/mL, sensitivity 89.4%, specificity 79.3%, positive predictive value 73.9% and negative predictive value 92.0%. There was no correlation between HSPG and TNFα levels ($p>0.05$). It was concluded that HSPG can be used as an early marker of plasma leakage in dengue infection.

S103

Prothrombin Fragments 1.2 in Relation with Plasma Leakage and Thrombocytopenia in Dengue Infection

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Clinical manifestations of dengue hemorrhagic fever are plasma leakage and thrombocytopenia. Both manifestations are thought to be caused by increased thrombin generation as a result of coagulation activation. During thrombin generation, prothrombin fragment 1.2 (F1.2) was cleavaged from prothrombin molecule. The aims of this study were to look for any association between F1.2 level with plasma leakage, to know whether F1.2 level in critical phase was higher than that in convalescent phase, and to search any correlation between F1.2 level and platelet count in dengue infected patients. This cross sectional study have been approved by the ethics committee of Medical Faculty, Universitas Indonesia. Twenty subjects consisted of 10 subjects with plasma leakage and 10 subjects without plasma leakage, who agree to participate by signing the informed consent were enrolled in this study. Plasma leakage was defined by at least one of these things : increased hematocrit ≥ 20% from reference value or a decrease in hematocrit ≥ 20% after fluid replacement therapy, or evidence of pleural effusion and/or ascites, and/or by albumin >3.5 g/dL. Six pairs samples were used for comparison of F1.2 level in critical phase and convalescent phase, and 26 samples were used for correlation between F1.2 and platelet count. Recruitment of subjects were done from January until September 2013. F1.2 level was determined by ELISA. The results showed that F1.2 level in patients with plasma leakage (147.4±105.82 pg/mL) was significantly higher than that in patient without plasma leakage (51.3±39.92pg/mL). F1.2 level in critical phase (186.3 pg/mL) was significantly higher than that in convalescent phase (46.5 pg/mL). There was a negative correlation between F1.2 level and platelet count (r=0.609). The results of this study suggested that at critical phase of dengue infection, activation of coagulation occurred which was associated with plasma leakage and thrombocytopenia.

S104

The Proportion and Factors Associated with Clopidogrel Resistance in Patients with Acute Coronary Syndrome

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The role of platelets in the pathogenesis of cardiovascular events has been recognized for a long time. Therefore clopidogrel as inhibitor for platelet aggregation has been used as standard therapy for patients with acute coronary syndrome and post percutaneous coronary intervention. Unfortunately not all patients responsive to inhibitory effect of clopidogrel. The aims of this study is to ob-tain the proportion of clopidogrel resistance and to find out factors associated, in patients with acute coronary syndrome and/or post coronary intervention. One hundred patients with acute coronary syndrome and/or post coronary intervention were enrolled in this study. Platelet aggregation test were performed based on light transmittance changes using AggramÒ aggregation remote analyzer. Clopidogrel resistance was defined if maximum aggregation > 59% with agonist ADP 20 μM. Genetic polymorphism of CYP2C19*2 and *3 were performed by Polymerase Chain Reaction (PCR). Clinical data were obtained from patient's medical record. The results indicated that maximum aggregation >59% was found in 36 out of 100 patients. In the calculation to get final predictive model for clopidogrel resistance, there was 4 factors that associated with clopidogrel resistance, i.e.: CYP2C19*2 (p=0.05;OR 4.0), CYP2C19*3 (p=0.03;OR 2.8), non smoking (p=0.061;OR 2.4), and diabetes mellitus (p=0.062;OR 2.4). It was concluded that the proportion of clopidogrel resistance was 36% and factors associated with clopidogrel resistance were CYP2C29*2 and *3.

S105

Quality Assurance in Health Laboratory Services: Continuous Process Improvement

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Continuous process improvement is the movement from the current state to future state towards providing improved laboratory services.

It is a quality management process that encourages all team members to continuously ask the questions:

- How are we doing?
- Can we do it better?
- Can we do it more efficiently?

- Can we do it more effective?
- Can we do it faster?
- Can we do it in a more timely manner?

At the end of this lecture, participants should be able to 1) Relate the historical perspective of continuous quality improvement 2) Describe the importance of continuous process improvement in maintaining quality 3) Explain the need for tools to monitor laboratory processes.

S106

Strengthening Health Laboratory Network Services in the Country

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Pursuant to the achievement of quality service and accessible health facilities which is one of the Strategies of Universal Health Care, the Department of Health, through the Health Facility Development Bureau, established the "National Framework of the National Health Laboratory Network" and the "Strategic Plan for the National Health Laboratory Network in the Philippines". The vision of the Strategic Plan is the development of a National Health Laboratory Network (NHLN) that provides quality, reliable, affordable and accessible laboratory information for the appropriate management of patients and prevention/control of diseases. The reference in the formation of the Laboratory Network (LabNet) is the WHO's Asia Pacific Strategy for the Strengthening of Health Laboratories.

The journey in the implementation of the strategic plan at the national level, regional level and laboratory will be discussed including the milestones along the journey and its challenges.

S107

Biosafety, Biosecurity and Biorisk Management in the Clinical Laboratory

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One of the major challenges regarding laboratory biosafety and biosecurity is the increasing availability and accessibility of potentially harmful technology. Biomedical advances and the globalization of scientific and technical expertise have made it possible to greatly improve public health. However, there is also the risk that advances in this field can lead to detrimental outcomes. The proliferation of high biosafety level laboratories in the clinical, academic and industry settings has many experts worried about availability of targets for those that might be interested in stealing dangerous pathogens. This concern is highly magnified in the Asia Pacific Region especially for developing countries with limited resources. Emerging and re-emerging disease is also a serious biosecurity concern. In particular, the clinical laboratory is a central locus to this concern. Several incidents had already occurred in the past that gives evidence to the potential of this threat. In this regard, there is a need to provide support to the development and use of appropriate training and competency development programs and associated materials to practitioners and their respective institutions to understand, adopt and implement biorisk management strategies and thus reinforce their respective capacities to effectively reduce biorisk in clinical laboratory environments. The objectives of this lecture are: (a) to enhance the current state of knowledge regarding laboratory biosafety and biosecurity among laboratory professionals; (b) to determine relevant local and international legislation on, in references to, biosecurity, biosafety, bioethics, dual-use issues, international prohibition regimes against biological and toxin weapons, and ethical guidelines or code of conduct in addressing dual-use issues, and; (c) introduce concepts of biorisk management including risk identification and management.

S108

Prevalence Of Micronutrient Deficiencies - Do We Need Food Fortification?

Seema Bhargava

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Despite epidemiological transition, India has a continued burden of infectious diseases and nutritional deficiencies. In addition, there are several reports claiming a high prevalence of vitamin D deficiency in Indians. Population data on these vitamin deficiencies, however, are meagre.

3 years retrospective data included 52,267 subjects. Data was subject to statistical analysis for assessment of age and sex-wise prevalence of deficiencies of iron, vitamin B12, folate, and 25-hydroxy-vitamin D.

Mean iron, B12 and 25-hydroxy-vitamin D were significantly ($p=0.031$, 0.002, 0.048, respectively) lower in females as compared to males. Mean iron was lower in the >60years age group ($p=0.001$), whereas B12 was significantly ($p<0.0001$) lower in the 18-40 years age group. Overall percent pre-valence of deficiency was 66.73, 44.09, 5.08 and 79.05 for iron, B12, folate and vitamin D, respectively. In patients with anemia, prevalent deficiencies were of iron alone(66.73%), B12 alone (36.53%), combined iron and B12 (18.88%), folate alone (2.92%), and combined iron and folate (1.77%).

The high prevalence of micronutrient deficiencies (iron, vitamins B12 and D) in India, indicates a requirement of population-wide mitigative measures. The most plausible programmes may be

- education in diet diversification, and

- fortification of food grains (rice and wheat flour) with iron and B12, and
- of milk with vitamin D,
- continuation of the NNACP (National Nutritional Anemia Control Program).

S109

Folic Acid Supplementation - is there Enough Evidence to Justify it?

Annalise E Zemlin

Stellenbosch University and National Health Laboratory Service (NHLS), South Africa

This talk will cover the basic biochemistry of folate as well as the pathophysiology and clinical implications of folate deficiency. After recognizing that folate deficiency is the most common vitamin deficiency in developed countries and was implicated in not only megaloblastic anaemia but also other disorders such as neural tube defects, Down's syndrome, cardiovascular disease and Alzheimer's disease, folate supplementation of foods was introduced. Howver, is this always beneficial? What about those for whom this supplementation was not targeted such as the elderly population or those with possible undetected malignancy? The evidence will be discussed pros and cons of folate supplementation argued.

S110

Vitamin D - is there Enough Evidence?

Rajiv T Erasmus

Stellenbosch University, Cape Town, South Africa

Historically, vitamin D deficiency was associated with rickets or osteomalacia and its effect on calciumetabolism was well-described. However, recent literature has highlighted other effects of vitamin D and its association with cancer, cardiovascular disease, auto-immune disorders and other conditions have been described. This talk will examine some of these and the possible underlying pathophysiology. However, the evidence will be examined. Many of these cut-offs were determined on assays no longer is use. Can we still accept these cut-offs and the study results used with old assays?

S111

BioSpaceForming, Chronic Hypoxia, and the future of Humans in Space

Gustavo Zubieta-Calleja

High Altitude Pulmonary and Pathology Institute, Bolivia

Continuous process improvement is the movement from the current state to future state towards providing improved laboratory services.
It is a quality management process that encourages all team members to continuously ask the questions:
- How are we doing?
- Can we do it better?
- Can we do it more efficiently?
- Can we do it more effective?
- Can we do it faster?
- Can we do it in a more timely manner?

At the end of this lecture, participants should be able to 1) Relate the historical perspective of continuous quality improvement 2) Describe the importance of continuous process improvement in maintaining quality 3) Explain the need for tools to monitor laboratory processes.

S112

Heart, Breath and Brain: the Cellular Determinants of the Hypoxic Response

Jan Marino Ramirez

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All mammals developed effective strategies to cope with reduced oxygen availability or other metabolic, environmental and behavioral challenges. An important prerequisite for survival is the necessity to be flexible and adaptive, while maintaining functional integrity during times of extreme challenges. This is particularly important for the neuronal networks that control breathing and the heart beat. These networks are amenable to a rigorous cellular and subcellular analysis. Using modern transgenic, optogenetic and molecular techniques we identify the critical microcircuits for breathing and demonstrate that neuromodulators imbue this network with the dynamic ability to reconfigure and alter the distribution of respiratory activity within the ventral respiratory column in the brainstem. This network reconfiguration involves the differential activation and inhibition of identified excitatory and inhibitory respiratory neurons as well as glia. We also show that breathing can occur as a 1-, 2-, or 3-phase rhythm, and that every breath is assembled stochastically, with each phase being generated

independently by a dedicated excitatory microcircuit. The ability of these microcircuits to reconfigure may allow breathing to remain robust, yet plastic enough to adapt not only to metabolic challenges, but also to conform to non-ventilatory behaviors such as vocalization, swallowing and coughing. The respiratory network also controls the neurons involved in regulating the heartbeat. Here we show that inspiratory preBötC neurons phasically inhibit neurons within the Nucleus ambiguus, these neurons include the parasympathetic cardiac vagal neurons. This interaction seems to generate the respiratory sinus arrhythmia. Lessons learned from the respiratory network may translate to other highly dynamic and integrated rhythmic systems.

S113

Polyerythrocythemia and Cerebro-Vascular Accidents at Different Altitudes

Natalia Zubieta DeUrioste

High Altitude Pulmonary and Pathology Institute, Bolivia

When sea level humans go to high altitude, they have to increase their red blood cells in order to compensate for the environment hypobaric hypoxia. What are the normal levels of hemoglobin, hematocrit, and red blood cells for each altitude? What are the specific variations in each civilization? We have over 49 years of work at the high Altitude Pulmonary and Pathology Institute, on this subject. We established the normal values for the city of La Paz, Bolivia at 3500m, in 1979 but it was poorly understood and other centers applied it without considering the variations of altitude. One can take different parameters to evaluate where the top cutoff limit for polyerythrocythemia is and likewise where anemia is present at high altitude. We proposed likewise a classification: Mild, Moderate and Severe, depending on the amount of red blood cells present. The limits where one can speak of polyerythrocythemia, depend on where the symptomatology (if present) appears. But it can also be the standard deviation. The Gauss curve of distribution can also be visually analyzed to set the limits.

The complexity of different altitudes, different nutrition, different genetic buildup, altitude changes, is evident in setting the normal values. This is why there is so much confusion. We propose a generalization strategy based on the average red blood cell counts for different altitudes, where we can set estimated limits to be considered above that of the normal residents. The importance of this proposal is that many people receive treatments of bleeding unnecessarily, due to this lack of standards. These parameters will now be applied to a multicenter study being performed in South America to evaluate if Cerebro-Vascular Accidents are related to polyerythrocythemia. In a preliminary study in a hospital in La Paz, 10% of the CVA's had polyerythrocythemia. However, of that 10% (n=10), all of them had a concurrent diagnosis such as Arterial Hypertension (9/10), diabetes (2/10) and dehydration (1/10) which are all well known causes and possibly rule out polyerythrocythemia as the fundamental ethiopathogeny. Therefore this suggests that AVC should not be seen as an expectable complication of polyerythrocythemia at high altitude.

S114

Lead and Hypoxia

Thuppil Venkatesh

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Lead is number one environmental poison to all forms of life even for high altitude dwellings. Lead is also used in paints, pigments and in traditional medicines apart the production of various cosmetics used by high landers who were using natural dyes earlier. Of late one of the major source of lead to high landers is from UPS batteries. Lead is known to have deleterious effects on antioxidant enzymes. Work carried out by the authors on the blood lead levels have indicated the elevated blood lead levels in many high landers. Elevated BLL is also found to have an impact on the Intelligent Quotient (IQ). Author has found that most of the vegetable dyes used for dyeing and for of age old Thanka paintings by Tibetan high landers are being replaced by synthetic dyes resulting in high exposure to lead. Though many studies indicate that younger children involved in Thanka paintings have disturbed cognitive functions resulting in imperfect art work. And lack of attention.

BLL is directly associated with the antioxidant status seen at sea level, at high altitude condition lead seems to have many effect. Lead is known to inhibit ferro-chellatase and thereby causing anaemia while low oxygen availability causes polycythemia. Under hypoxic condition elevated blood lead level will be deleterious to life, Lead poisoning in hypoxic condition will result in more serious conditions. Some of the intricate details will be discussed by the author who has travelled across Himalayan high altitude habitat and also currently is the President of the International Chronic Hypoxia Society at IPPA.

O-001

Chemokines in Liver Diseases: New Insight in Iron Trafficking Proteins and Prokineticin in Metabolic Syndrome and Non-alcoholic Fatty Liver Disease (NAFLD)

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Non-alcoholic fatty liver disease is increasing worldwide, imitating the current epidemics of lifestyle diseases. This is escorted by dyslipidemia, inadequate exercise, oxidative stress and leading to NAFLD/NASH. Unhealthy diet, genetics, environmental exposures and lifestyle changes may lead to cirrhosis and HCC. Expansion of adipocytes, recruitment of macrophages and monocytes releases prokineticins, iron trafficking proteins and growth differential factor -15. Iron trafficking proteins and Prokineticins have potential role and interlinked with excessive oxidative stress, mediators of inflammatory molecules in disease conditions. As NAFLD is progressed by multiple parallel hit mechanisms thus we explored whether GDF-15, iron trafficking proteins and Prokineticins affect the severity of NAFLD. Blood samples were collected from the OPD of Gastroenterology, and department of Medicine, AIIMS, New Delhi. Biopsy samples were collected only from NAFLD patients. Each was screened for serology and were categorized into sub-groups, for NAFLD (n=30), Hepatitis (disease control n=38), diabetes mellitus without metabolic syndrome (DM without Mets) (n=20), Metabolic syndrome without diabetes mellitus (Mets without DM) (n=25), and Diabetes mellitus with Metabolic syndrome (DM with Mets) (n=30). Healthy subjects (n=25) were also screened for serology. Parameters such as Adiponectin, leptin, Insulin Resistance, BMI, pro-inflammatory and anti-inflammatory cytokine, oxidative markers, Prokineticins and GDF-15 were done in all groups. GDF15 levels, iron trafficking proteins and Prokineticin expressions were higher in NAFLD subjects, DM with MetS, and MetS without DM than in healthy controls. GDF-15 levels have some hidden relations towards changes in body weight, waist hip ratio, BMI and HOMA IR. Prokineticins expressions were found higher in NAFLD, HCC patients, and DM with Metabolic syndrome than diseases controls (HBV & HCV) and healthy controls. Furthermore, increasing pattern of viral load of HBV and HCV infections (>104 IU/ml) have suggested that the expression of Prokineticins are noticed more than 3-4 fold change in patients with cirrhosis and liver cancer (HCC). Our finding suggests that high circulating levels of GDF-15, iron trafficking protein (Zip8) and overexpression of Prokineticins could be potential biomarker of NAFLD, and decreased levels of antioxidants could be linked in development of future diabetes, CVD and metabolic syndrome. Thus, Prokineticins and iron trafficking proteins can be novel emerging biomarkers to treat NAFLD and diabetes.

O-002

Klotho Protein as a Marker and Protective Agent During Ischemia/Reperfusion Injury of the Heart

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Hypoxic injury of the heart is one of the most frequent complications following ischemia. The injury of myocardium during ischemia/reperfusion (I/R) is a complex and multifactorial process involving metabolic, morphological and contractile disorders. Klotho is a membrane-bound or soluble antiaging protein with antioxidative and antiapoptotic activity. The aim of the study was to determine expression of Klotho protein in the cardiac cells during I/R, as well as to evaluate the potential cardioprotective role of Klotho. Isolated Wistar rat hearts perfused with Langendorff method and human cardiac myocytes (HCM) were subjected to I/R injury. Hemodynamic parameters of heart function, markers of I/R injury, gene and protein expression of Klotho were measured. HCM were incubated in the presence of recombinant Klotho protein and the viability of cells was measured. There was a higher expression of Klotho gene ($p<0.05$) and protein synthesis ($p<0.05$) in the cardiomyocytes subjected to I/R, as well as the compensatory production and release of Klotho protein from cardiac tissue into extracellular space during I/R ($p<0.05$). Klotho level in coronary effluents positively correlated with the level of injury marker ($p<0.05$, $r = 0.6$) and negatively correlated with cardiac mechanical function ($p<0.05$, $r = -0,6$). The administration of Klotho protein resulted in increased viability ($p<0.05$) and metabolic activity ($p<0.05$) of HCM subjected to I/R. An increased cardiac expression of Klotho during I/R and its release into extracellular space suggest a potential compensatory role and use of Klotho as a marker of cardiac damage. Klotho may serve as a potential preventive/protective agent during ischemic injury of the heart. This work was supported by the National Science Centre [grant number UMO-2016/23/B/NZ3/03151].

O-003

Role of Vitamin D in Innate Immunity Modulation Based Therapeutic Targets in Pulmonary Tuberculosis

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Vitamin D, its transport protein vitamin D binding protein (VDBP) and vitamin D receptor (VDR), may play a role in altering the host defense against Mtb via production of cathelicidin (antimicrobial peptide) and regulating the expression of inducible nitric oxide synthase (iNOS) required for production of bactericidal Nitric oxide (NO). In the present study, levels of Vitamin D, NO and their associated molecules were evaluated in 100 active tuberculosis (TB) patients, 75 household contacts and 70 healthy controls. VDR and iNOS mRNA levels were found to be lower in active TB group compared to household contacts and healthy controls (P=0.0001 and 0.005 respectively). Though insignificant, expression of VDBP mRNA was lower in active TB group as compared to household contact and control groups. The serum levels of Vitamin D were also found to be lower in active TB group compared to healthy controls (P =0.001). Levels of cathelicidin and NO were higher in patient group as compared to other groups (p=0.01 and 0.5 respectively). However, the expression of VDR and iNOS and levels of vitamin D was significantly (P< 0.05) higher in household contacts compared to both active TB and healthy control groups. Our observations suggest that vitamin D might have a protective role against TB which prevents activation of the disease. Decreased vitamin D levels could be implicated in disease progression. Supplementation of vitamin D in household contacts of TB and as an adjuvant therapy along with ATT in active TB should be evaluated to assess therapeutic potential.

O-004

Anti-Nuclear Antibody Pattern Distribution and Clinical Profile in Patients Suspected of Auto-Immune Disorders

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Autoimmune responses can occur against various proteins like enzymes, receptors, glycoproteins, phospholipids and nucleic acids. Antinuclear antibodies (ANA), are a response by the body to its own cell nuclei. Autoimmune disease may present in varied manner with various clinical signs and symptoms. These disorders can be systemic autoimmune response, or organ specific response. IIF (Indirect Immunoflorescence) is the gold standard method for screening antinuclear antibodies. This study aims to investigate the ANA pattern distribution and clinical profile of patients suspected of autoimmune disorders. ANA-IIF reports of patients, referred to the Department of Biochemistry, AIIMS Raipur, over a period of one year, was analyzed in a retrospective cross sectional study. Confirmation of anti ds DNA by ELISA and anti extractable nuclear antigen(anti-ENA) was done by Dot Immunoblot. High prevalence of positive ANA patterns in suspected cases of CTD and other auto-immune conditions suggests that the parameter would be a successful screener for them and also play an adjuvant to the diagnosis. An early knowledge about future autoimmunity will earn better prognostic achievements.

O-005

Association Amongst Leukocytic Telomere Length, Inflammatory Markers and Mitochondrial DNA Copy Number in Pregnant Women: A Pilot Study

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Both short telomere length, increase of inflammatory markers and mitochondrial dysfunction have been associated with pregnancy complications such as pre-eclampsia and intrauterine growth restriction (IUGR). However, the relationship between these three biomarkers of oxidative stress, during pregnancy, is unknown or not documented. This study investigated the association of leukocyte telomere length with mitochondrial DNA (mt DNA) copy number and inflammatory markers IL6, IL10, TNF-α as indicators of inflammation, of mitochondrial density and possible mitochondrial dysfunction, using maternal blood samples collected from women with pregnancies uncomplicated by gestational diabetes or hypertensive disorders or a bad obstetric history. Leukocyte telomere length and mtDNA copy number were determined in 100 pregnant women registered in and around NCR, using quantitative real-time PCR (BioRad). IL-6, 10 and TNF α were assayed on ELISA Station (BioRad). Bivariate and multivariable linear regression procedures were used to evaluate associations of these three categories of biomarkers. Leukocyte mtDNA copy number (natural logarithm) was positively associated with telomere length (Pearson correlation coefficient = 0.30, p-value = 0.009). After adjusting for maternal age and plasma vitamin B12, natural-log mtDNA copy number increased by 0.80 (f = 0.80; 95% CI 0.25 - 1.34, p-value = 0.005) for every 1 unit increase of telomere length. Approximately 11% of the variation in natural-long mt DNA copy number was explained by the model (adjusted R2 = 0.11). The value of IL 6 and 10 and TNFα correlated fairly

well and showed negative association (Pearson correlation coefficient= -0.38, p-value =0.00012). This cross sectional data suggests an association of mtDNA copy number with telomere length, two emergent biological markers of potential importance in portal of perinatal health research. There is a state of microinflammation in the arterioles which is proven by raised IL6, IL10 and TNF α. The consequences of oxidative stress, cellular senescence (as reflected by relatively shorter telomere length) and mitochondrial dysfunction, on the course and outcomes of pregnancy remain to be elucidated in larger prospective studies that include these biological markers.

O-006

Serum SYPL1 is a Promising Diagnostic Biomarker for Colorectal Cancer

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At present, the overall sensitivity and specificity of blood biomarkers is hard to meet the diagnosis of CRC. Via the analysis of the genes with cancer positive rate ranking at the top 10% in both GEO datasets, SYPL1 was singled out from the 8 membrane proteins of secretory vesicle. The confirmation tests showed SYPL1 was upregulated in colon cancer cell lines and CRC tissues at both mRNA and protein levels, compared to the normal controls. Consistently, the serum SYPL1 (sSYPL1) was significantly higher in CRC patients (n=151) than both healthy controls (n=89) and adenoma patients (n=73) (P<0.0001), and associated with lymph node invasion (P<0.05). While, the sSYPL1 was declined after radical operation (P<0.0001). Further, ROC curve showed that the performance of sSYPL1 was prominent in distinguishing CRC patients from healthy controls (AUC: 0.9481, sensitivity: 86.09% and specificity: 91.01%) or adenoma (AUC: 0.8631, sensitivity: 98.68% and specificity: 78.08%). This was much better than the performance of carcinoembryonic antigen (CEA) or Carbohydrate antigen 19-9 (CA19-9) or their combination. Even for the patients in the gray zones of CEA under 2.2 ng/mL or 5 ng/mL, SYPL1 still kept the same high performance for the identification of CRC. Conclusion: serum SYPL1 is an outstanding marker to establish simple, fast, and robust approaches for CRC diagnosis, especially for those patients with low CEA.

O-007

Pro-inflammatory Cytokines in Occupationally Lead Exposed Individuals: A Pilot Study

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Lead (Pb) is a heavy metal which has been widely used in various industrial and domestic settings due to its physical and chemical properties. In developing countries, many workers continue to be exposed to toxic effects of Pb due to lack of knowledge on its safe handling. Lead is also able to induce inflammatory processes by modulation and activation of intracellular signaling pathways. We aimed at estimating pro-inflammatory cytokines IL-6 and TNF alpha in Pb exposed individuals. 40 chronically Pb exposed welders and 29 healthy controls consented for the study. Venous blood samples were collected taking due aspectic precautions. Blood Pb levels (BLL) were analyzed using Dual Atomic Absorption Spectrophotometer (ICE 3500 Thermofischer). Commercial reference materials were obtained from Bio-Rad (Lyphochek® Whole Blood Metals Control) for the internal quality assurance and control program. Cytokines IL-6 and TNF alpha were estimated with commercially available ELISA kits. Parameters were not normally distributed. Data was expressed as Median (IQR) and Mann Whitney U test was used for comparison. Median BLL in cases was 4.42 μg/dL (Range: 0.68-31.76,95% CI: 5.019-9.019). The median BLL in controls was 2.9 μg/dL (Range:1- 4.5, 95% CI: 2.2-2.95). The median TNF alpha in cases was 43.36 pg/mL (Range: 4.93-971.8, 95% CI: 162.2-383.2) and in controls (Median: 10.02 pg/mL, Range: 0.90-1082, 95% CI: 37.99 - 243.4). The IL-6 values in cases were 4.59 pg/mL (Range: 0.97-109.5, 95% CI: 24.58-45.04) where as in controls it was 2.26 pg/mL (Range:1.34-3.23, 95% CI:2.19-2.54).The pro inflammatory cytokines IL-6 & TNF alpha were significantly higher in occupationally lead exposed individuals in comparison to controls (p<0.05). This study concludes that occupational Pb exposure promotes inflammatory processes via induction of pro-inflammatory cytokines.

O-008

Decoding the Psoriasis and Cardiovascular Disease Link Conundrum?

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Psoriasis is a chronic immune-mediated skin disease. The concept of "psoriatic march" implicates systemic inflammation, leading to oxidative stress and insulin resistance, resulting in endothelial dysfunction, the major causative factor in increased cardiovascular co-morbidity in psoriatic patients. This study was undertaken to study the psoriasis-cardiovascular disease link and to assess the effect of methotrexate-monotherapy on markers of systemic inflammation, adipokines, markers of insulin resistance, oxidative stress, endothelial dysfunction, atherothrombosis in patients with psoriasis.This study involved 87 patients with severe psoriasis and 87 age and gender-matched controls. All 87 cases were followed up at 12 weeks of methotrexate-monotherapy. In all cases of psoriasis, disease severity was assessed by Psoriasis Area Severity Index score (PASI). Non-invasive assessment of endothelial dysfunction and the biochemical markers were estimated at baseline in all subjects and after 12 weeks of methotrexate-monotherapy in cases. The markers of systemic inflammation, insulin resistance, oxidative stress, endothelial dysfunction and atherothrombosis were significantly elevated along with non-invasive markers of cardiovascular risk at baseline in patients with psoriasis, correlating significantly with PASI and showed a significant decline in their levels, after 12 weeks of methotrexate-monotherapy. Psoriasis remained independently associated with flow-mediated-dilatation of brachial artery (FMD) even after adjusting for confounders. This study suggest that psoriasis is an independent risk factor for cardiovascular disease and methotrexate-monotherapy significantly ameliorates systemic inflammation, insulin resistance, oxidative stress, endothelial dysfunction and atherothrombosis which might reduce the cardiovascular co-morbidity in psoriasis. This emphasizes the need to timely initiation as well as proper compliance with treatment in patients with psoriasis so as to reduce morbidity and improve their quality of life.

O-009

Root Bark Extract of Calliandra Portoricensis (Jacq.) Benth. Chemoprevents N-methyl-N-nitrosourea-induced Mammary Gland Toxicity in Rats

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Calliandra portoricensis (CP) is a herb widely used in Nigeria for the treatment of breast engorgement. However, the scientific evidence of this use and its mechanisms of action are not clearly understood. We assessed the chemo preventive effects of methanol extract of CP on *N*-methyl-*N*-nitrosourea (NMU)-induced mammary gland toxicity in rats. Fingerprinting of methanol extract of CP by Gas Chromatography-Mass Spectrometry (GC-MS) was done. Female Wistar rats were assigned into eight groups: Group 1 (control), group 2 received NMU only, groups 3, 4 and 5 received NMU and CP at doses of 100, 200 and 300 mg/kg, respectively. Group 6 received CP (300mg/kg), group 7 received NMU and vincristine, while group 8 received vincristine. From these findings, the weight-gain by rats decreased in all groups that received NMU. Administration of NMU significantly increased organo-somatic weight of mammary gland by 52%. The NMU increased serum nitric oxide, total bilirubin, mammary myeloperoxidase and lipid peroxidation by 76%, 87%, 130% and 21%, respectively, as well as activities of serum aspartate aminotransferase and lactate dehydrogenase. Also, NMU-treated rats had decreased total sulphydryl, reduced glutathione and catalase. Histology showed moderate increase in periductal fibrous tissues and benign fibroadenomas in NMU-treated rats relative to control. However, groups treated with CP and vincristine had normal stroma with slight increase in periductal fibrous tissues without benign fibroadenomas. Immunohistochemistry revealed strong expression of estrogen, progesterone and EGFR-2 proteins in NMU-treated rats. Treatment with CP (200 and 300 mg/kg) attenuated NMU-induced inflammation and oxidative stress. CP ameliorated NMU-induced mammary gland toxicity by modulating different cellular targets.

O-010

Assessment of Circulating miR-20b, miR-221 and miR-155 in Occupationally Lead Exposed Workers of Jodhpur

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Lead (Pb), a toxic heavy metal, is capable of inducing several adverse health effects following its accumulation in the body. Lead is a potential carcinogen, capable of causing multi-system alterations. The molecular mechanisms by which lead exerts systemic damage in human body is under exploration. Recent reports identify small regulatory RNA molecules - miRNAs, which show differential expression in individuals exposed to similar levels of lead. These miRNAs can become potential molecular biomarkers of lead toxicity in the future and may unravel the possible molecular pathways through which this metal may exert its toxic manifestations. The present study had an aim to assess the circulating levels of miRNA-20b, 221 and 155 in occupationally lead exposed workers and correlate them with blood lead levels. 87 participants working in handicraft and wielding factory of Jodhpur and equal number of participants not occupationally exposed to lead were recruited after obtaining due informed consent. Blood lead level was estimated by graphite furnace-atomic absorption spectrophotometry (GF-AAS). Circulating miRNAs were isolated from serum by Qiagen miRNA isolation kit and converted to cDNA by commercial kit. Expression profiles of miR-20b, miR-221 and miR-155 was performed in RT-PCR using Qiagen miRNA PCR assays. The blood lead level (mean± SEM) of occupationally lead exposed subjects was 6.94± 1.40 µg/dl while that of non-exposed was 1.01±0.1 µg/dl. All the three miRNAs were downregulated in the participants. When compared between lead exposed and non-exposed, miR-20b, miR-221 and miR-155 showed a 0.7, 16.67 and 0.66 fold change respectively. Functional analysis revealed that these miRNAs have potential to trigger various cellular genes and pathways. Findings of our study highlight the importance of miRNA dysregulation in lead exposed individuals that may contribute to systemic effects of lead toxicity.

O-011

Development of Validation Design of HbA1c Control

Shyamali Pal

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Diabetic profile is a common health check-up profile and laboratory services all over India aspires to keep under the scope of accreditation. The diabetic profile control from BIORAD is expensive and after reconstitution stability is 7 days only as per manufacturer's insert. If laboratories follow the 7 days regime the daily quality control cost would be very high resulting either restricting test performance once/ twice in a week affecting regular services or to keep HbA1c out of scope of accreditation affecting patient confidence. A search was done to find out actual stability of the controls. The validation data of such stability period may be utilised by the laboratories aspire to keep HbA1c under the scope of accreditation subject to findings of the laboratory results in accordance with the study. The diabetic profile Level 1 & 2 controls of three different lot numbers are reconstituted and preserved as 10 µL aliquots at 2°C-8°C. One aliquot is used for one-time test performance only. The Level 1 & 2 controls are used on alternate day making use of one lot for 90 days. Three different lots were analysed in two laboratories to obtain inter and intra assay deviations. Tests performed in BIORAD D1O. Mean, SD, CV%, Total Allowable Error (TAE), %Bias, z-score and sigma calculations were done. The medical method decision charts were created for all lots based upon normalized operational specifications which showed excellent precision (Sigma >6 in inter and intra assay results) in both control levels. Number of rejections in the study was nil. Thus, the extended use of controls are validated.

O-012

Validation and Verification of Body Fluids for Chemistry Parameters on Automated Integrated System

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Majority of automated instruments are validated and verified for serum and urine chemistry parameters. However, manufacturers do not provide performance verification for body fluids. Since body fluids such as ascitic, pleural, CSF, dialysate and pericardial fluid specimens form a part of testing panel for variety of analytes, it becomes important to verify/validate the performance of these fluids. The accreditation bodies that certify and accredit labs follow ISO 15189:2012 that states validation

should be carried out whenever there is a deviation from stated procedure. The aim of this study was to establish performance characteristics for commonly requested analytes in body fluids. Left over samples were utilised for validation/verification of ten common analytes specific to each of the above-mentioned body fluids on Abbott Architect ci8200 integrated system. Performance characteristics that were verified are as follows: Imprecision, accuracy, reportable range, stability and interference. Assay imprecision for body fluid assays were comparable to imprecision obtained for serum. Accuracy was evaluated with recovery experiment after spiking the sample with a known concentration of substance. Reportable range was found to be comparable to serum parameters. Chemistry analytes for body fluid analytes was found to be stable for a period of 20 days at -20° C. It is mandatory for lab to establish validation/verification of body fluid assays for commonly requested analytes. This would improve the reliability of the testing system.

O-013

Utility of Serum free Light Chains Assay in Assessing Patients Presenting with Polyneuropathy

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American Academy of Neurology 2009 guidelines recommend immunofixation electrophoresis (IFE) as an initial screening test, along with plasma glucose and vitamin B12 for polyneuropathy workup. The guidelines are based on small studies that used outdated methods and do not include the current screening modalities such as the serum free light chains (sFLC) assay. Our objectives were to evaluate the prevalence of monoclonal immunoglobulins (M-components) in our polyneuropathy population using contemporary methods, including capillary zone serum protein electrophoresis (SPEP), IFE, and sFLCs. We also compared the diagnostic accuracy of the sFLC assay to IFE in this population. Serum samples from neurology patients were obtained. SPEP, IFE, and sFLC were performed. A total of 657 unique patient samples were included in this study. At least one M-component was detected in 103 patients (15%); of these, 42% were unquantifiable and the remaining 60 patients had a median M-component concentration (95%-confidence interval, CI) of 0.28 (0.24-0.32) g/dL. The median (95%-CI) kappa/lambda ratio was 1.29 (1.26-1.32). The kappa/lambda ratio was abnormally elevated (>1.65) and abnormally decreased (<0.26) in 141 (21%) and 1 (<1%) of all specimens, respectively. The sensitivity and specificity of the sFLC to detect an M-component were 40% and 82%, respectively. We observed a slightly higher prevalence of M-components in our polyneuropathy population compared to earlier studies. This may be attributed to modern SPEP/IFE methods used in our study. The concentrations of M-components were quite low in all patients. The low concentrations may, in part, explain the poor sensitivity of the sFLC ratio to detect the presence of M-components. Correlation of these findings with subsequent workup (PET-scans, bone marrow biopsies) and clinical outcomes (malignancy diagnoses, initiation of therapy) will be necessary to determine the impact of using the sFLC and current electrophoresis methods in the polyneuropathy diagnostic evaluation.

O-014

Preemptive CYP4F2 Genotyping for Warfarin Dosing Using a Novel Multiplex Assay

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The large inter-individual variability and narrow therapeutic index of Warfarin has made dose management challenging. Recent literature recommends genetic testing for CYP4F2 genetic variant along with VKORC1 and CYP2C9 variants for effective warfarin dose management. Therefore, the present study has been aimed to develop a novel multiplex assay that can effectively screen 04 genetic variants in VKORC1, CYP2C9 and CYP4F2 gene as well as determine the impact of newer CYP4F2 genetic variant on warfarin dose management. The present ongoing study has led to recruitment of 87 patients on warfarin therapy. Patients mean daily warfarin dose, international normalized ratio (INR) and demographics were recorded. Development of the novel multiplex allele-specific assay was carried out using the positive DNA controls for VKORC1 [c.1173C>T], CYP2C9 [*2,*3], CYP4F2 [C.1297G>A] variants and the results were validated by Sanger sequencing. CYP4F2 genotype frequencies were tested for Hardy-Weinberg equilibrium. Krushal-Wallis and Chi-square test were performed for comparison of warfarin dose and INR with CYP4F2 mutants using GraphPad Prism (v7.02). The mutant allele frequencies for VKORC1 [c.1173C>T], CYP2C9 [*2,*3], CYP4F2 [C.1297G>A] variants were found to be 0.14, 0.05, 0.13 and 0.41 respectively. The mean warfarin doses as well as the mean INR were not statistically significant with CYP4F2 genotypes. Importantly, only 47% of patients with supra [>3.0] and sub-therapeutic [<2.0] INR were explained by VKORC1 and CYP2C9 genotyping, which was significantly increased to 79% with inclusion of *CYP4F2* genotyping. Further in the above subgroup, 63% of patients with CYP2C9+VKORC1 wild-type showed the presence of CYP4F2 variant. Similar association between CYP4F2 variant and supra and sub-therapeutic warfarin dose was observed. The novel multiplex assay developed is sensitive, rapid and cost-effective genetic screening tool for warfarin dosing. Addition of CYP4F2 genotyping has significant impact on warfarin dosing thereby strongly suggesting pre-emptive CYP4F2 genotyping for patient on warfarin therapy.

O-015

A Comparative Evaluation of Four Phenotypic Methods for Detection of Class A and B Carbapenemase Producing Enterobacteriaceae in China

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Early identification of carbapenemase-producing Enterobacteriaceae (CPE) is of paramount importance to prevent their dissemination within health care settings.
We performed a comprehensive method comparison study to assess the accuracy of four screening methods, modified Hodge test (MHT), Carba NP test, meropenem hydrolysis assay (MHA) with 1/2 h incubation and modified carbapenem inactivation method (mCIM) with meropenem (MEM), imipenem (IMP), and ertapenem(ETP), in carbapenemases detection on a total of 342 carbapenem-resistant Enterobacteriaceae (CRE) referred by 34 teaching hospitals including 244 CPE and 98 non-CPE using genotypic assay as a gold standard. The 2h-incubation MHA had the highest performance observed among all assays, followed by Carba NP with a sensitivity of 99.6% while the 1h-incubation MHA performed much worse with an overall sensitivity of 71.3%, mainly due to failure in KPC detection. Comparison between different carbapenem disks using mCIM showed that MEM performed best among the three. The MHT performed worst among all assays which was mainly manifested in specificity (88.8%). Comparison between specific carbapenemase classes showed that all assays, except for the 1h-incubation MHA, had a >98% sensitivity in detecting the 172 KPCs, which failed to recognize 68 KPC-2 carbapenemase. Likewise, all assays demonstrated a >95% sensitivity in detecting the 70 Class B carbapenemases except for MHT (82.9%) due to invalid results for one IMP and 11 VIM producers. As for different species, almost all the assays had sensitivities and specificities over 90% for the ten species with the notable exceptions in sensitivities for Klebsiella pneumoniae (56.2%) and Serratia marcescens (75.0%) by 1h-incubation MHA. Our findings suggested that the MHA was the most practical assay to select for carbapenemase detection. For those who can't afford the equipment at the moment, both Carba NP and mCIM were good alternatives.

O-016

Ethics in Laboratory Medicine: Indian Scenario

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Ethics has been defined as 'a set of principles of right conduct' by various bodies. As per common understanding, it is the science of discerning right from wrong, good from bad or sometimes beneficial from harmful. IFCC has taken up the issue of laboratory ethics actively as it believes "decisions about diagnosis, prognosis and treatment are frequently based on results and interpretations of laboratory tests and irreversible harm may be caused by erroneous tests". Although guidelines for ethical standards vary widely between countries, cultures, and geographies, overall, the ethical principles remain the same: 1) Respect for persons: Acknowledgement of autonomy and protection of those with diminished autonomy. 2) Beneficence: The duty to act in the best interests of patients or research subjects with the goal of maximizing benefits and minimizing harm (nonmaleficence) and 3) Justice: The duty or obligation to treat patients equally, and to distribute, by allocating fairly, what is rightly due in terms of benefits, risks and cost. As of date, many countries and professional societies in the world have developed policies and guidance materials on ethical issues related to laboratory medicine, however, India needs to develop a consensus document. Even IFCC has recently constituted a task force for ethics (TF-E) with the goal of facilitating the same. The TF-E has streamlined documents available worldwide (https://www.ifcc.org/taskforceethics/). International Organization for Standardization (ISO) has created ISO 15189:2012 Medical laboratories: Requirements for quality and competence (section 4.1.1.3) summarizes the ethical conduct expected in laboratories, including topics like confidentiality, conflict of interest, undue pressures and influences and legal requirements. The ethical standards require understanding of ethical conduct in terms of pre-analytical, analytical and post-analytical phases of lab work. The roles of all members of the laboratory needs to be defined with an understanding of collective responsibility.

O-017

Iron Profile and Parkinson's Disease Risk: Evidence from Discriminant Analysis

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Parkinson's disease is a second most common of neurodegenerative disorder, affecting 2% of the population over the age of 65 years. Neurochemical studies have implicated metals in pathogenesis of PD, including Copper & Iron. To examine the

association of serum iron, transferrin, ferritin, transferrin saturation & UIBC in PD patients. Also to derive the Discrimination Function, with its score, of independent biochemical marker variables under study to correctly classify PD cases and healthy controls. In the present study, identification of biomarker pool in case-control study involving 79 PD cases and 80 healthy controls were performed to examine association among multiple biomarkers. The results of independent t-test analysis showed that PD cases presented significantly higher ($P< 0.01$) level of transferrin, TIBC, UIBC and urea than controls. Discriminant analysis was performed to determine the factors that best discriminates between the categories of an outcome variables (Disease status = PD & Control) and total of five biochemical independent variables (UIBC, transferrin, serum iron, transferrin saturation, and copper) were taken into consideration. In present study, UIBC (μmol/L)has emerged out to be highest discriminating powerful independent variable among considered independent variables. After development of Discriminant Function (Z) and also calculation of discriminant function cut points, a cross-validation analysis of PD cases and controls were also conducted. By the present discriminant function model, only one PD case (0.013 %) was wrongly classified as control whereas only 13 controls (0.16%), out of 80, were wrongly classified as PD cases. This discriminant function model appears to be slightly more sensitive than specific, the sensitivity of the developed model was 98.73% and specificity was observed 83.75%. Prospective validation of Discriminant model in large cohort and in other ethnic populations is warranted in future studies.

O-018

Digital Dysphoria in Biochemistry Reports

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The reports of clinical biochemistry are expressed in digits only. Biochemical tests in blood are done to confirm or rule out a suspected clinical diagnosis, follow up during treatment and as a part of routine medical check-up based on infrastructural facilities including cost-effectiveness by our organization. The concerned doctors who advise anticipate some results and interpret the same with their wisdom. Whenever, the reports do not match with their expectations, dysphoria starts regarding credibility which results in repetition by our laboratory or usually by another laboratory to honour the wishes of the patients. The patients who or patients' samples which were sent by the concerned doctors including those patients whose reports were suspected after analyses by us for some reasons were taken into this study. After interacting with the patients mainly, clarifications were given, or fresh samples were taken for repetition of the investigations to rule out any mistake. In almost all cases, apart from preanalytical errors committed usually unknowingly, no mistake was found. Proper communication to the doctors or clarifications directly to the patients solved all the problems practically. Associated obsession due to pre-leant knowledge against display of figures for the concerned patient or individual led to 'digital dysphoria'. Both the patients and the doctors suffered from confusion with the perception of wrong report by one laboratory without any analytical fault in the samples. The paucity of information was the main cause to explain in a rational way in each individual patient always. Any persistent normal or abnormal report was also not ignored. Normal figures offered at least 'digital pleasure' to all. Mismatch was almost a rule during early including recovery phase of any acute illness and cunning noncompliance by the chronic patients.

O-019

Minimizing errors in POCT: A Risk Assessment Analysis

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Complex regulatory requirements, training of testing personnel and implementation of uniform quality policy in varied locations across the hospital, pose the greatest challenge in achieving the desired results in point-of-care-testing. Over seventy POCT instruments are presently being used across our hospital (a six-fifty bedded tertiary care hospital in Delhi, India). CLIA '88 states that all testing is site neutral and therefore the same regulations apply regardless of where the test is performed. JCI requires that POCT be brought under the direct supervision of the central laboratory. In 2017 during the process of preparing for accreditation by the Joint Commission International (JCI), we were faced with the real challenges of implementing QA practices in point-of-care-testing across the hospital. Initial internal audits revealed absence of quality assurance policies, inadequate documentation and lack of uniformity in POCT practices across the hospital. A POCT committee was formed and SOPs were documented. Training of testing personnel was conducted. IQC and PT programs were designed and implemented. A list of critical/ alert values for all POCT devices was prepared by the laboratory in consultation with the attending physicians. The biggest challenge however, was to ensure compliance to established policies. The major obstacles being change of testing personnel, continued training and increase in workload. A risk assessment analysis was done using FMEA as the tool to identify areas vulnerable to errors. Criticality (Severity x Detectability) according to CLSI guidelines was assigned to each source of error. Criticality of 9 and above (identified by FMEA) were addressed immediately. Measures for the reduction of risk were undertaken. Compliance audits identified improvement in areas like: knowledge of POCT (nursing staff), on-time information of critical alerts ,quality assurance practices (IQC&PT). Intensive efforts are in process to further reduce risk in identified areas.

O-020

Sigma Calculation Harmonization: Why a Necessity

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Processes harmonization aims to implement process standards, same as process standardization. However, standardization strives for uniformity of processes, while harmonization allows for more variation to ensure harmonious acceptance of the standard Patient centred care, the main target of medical laboratories, depends on the key concepts of internal quality control (IQC) and external quality assurance (EQA) programs, which established in the late nineties as a complementary pillar to IQC, provide a tool of peer comparison. It is now necessary to determine how the performance of a measurement procedure relates to the medical requirements for interpreting results in order to determine the frequency to measure and evaluate quality control (QC) samples and results (7,8). Sigma metrics (SM) have been used to assess quality in a quantitative manner. Lack of a harmonized approach for sigma calculation is considered an obstacle in the objective comparability of analytical performance among laboratories adopting sigma metrics. It is urgently needed that all laboratory professionals interested in the analytical quality to work hard towards harmonization protocol for sigma calculation in order to properly select their analytical goals.

O-021

Role of Functional Foods in Prevention of Oxidative Stress in Various Diseases

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In the last decade, preventive medicine has undergone a great advance, especially in developed countries. Research has demonstrated that nutrition plays a crucial role in the prevention of chronic diseases, as most of them can be related to diet. Functional food enters the concept of considering food not only necessary for living but also as a source of mental and physical well-being, contributing to the prevention and reduction of risk factors for several diseases or enhancing certain physiological functions. Natural phytochemicals from edible and medicinal plants are of importance because they may be useful for disease prevention and have no undesirable xenobiotic effects on living organisms. Natural antioxidants may lessen or hinder the harmful prospective of xenobiotic. Regular consumption of plant food can provide ways for preventing oxidative stress and mutations in cells that conceivably result in various diseases. Even for populations which use functional foods traditionally, encouraging the use of spices with chemo-preventive actions could be helpful as part of life expectancy for two main reasons, firstly their costs are significantly low, secondly functional foods have no toxicity during long-term oral administration and are relatively available at large scale. A wide variety of substances obtained from plants in our foods (vegetables, fruits and spices) have been shown to possess anti-oxidant effects. Especially, the functional foods such as garlic, wheatgrass and carotenoids contain bioactive phenolic substances with potent anti-oxidative and chemo-preventive properties. In the present lecture, preventive role of functional foods like garlic, wheatgrass, and carotenoids against oxidative stress in different diseases will be discussed.

O-022

Identification of a Suitable Milk Product the Consumption of which could Improve Vitamin B12 Deficiency in Indian Vegetarians (IMPROVIT)

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The proportions of humans adapting to vegetarianism is on the rise worldwide with vitamin B12 being the only micronutrient absent. Hence, humans adhering to vegetarian diet can obtain B12 from milk, foods fortified with B12 and B12 pills. Milk and milk products are efficient carriers of highly bioavailable B12. Cow and buffalo milk (HO-B12) contain approximately the same amount of B12, 3-4 μg/L. The aim of the study is to identify a suitable milk fraction the consumption of which could improve vitamin B12 deficiency in vegetarians (Indo-Danish project). The following groups with 20 subjects each were taken - a) Various doses of Ho-B12 and CN-B12 (1.5, 2 and 6 μg/day) were administered and CobaSorb test was performed in B12 replete and deficient subjects. b) 3 μg of CN-B12/HO-B12/placebo was administered daily for 8 weeks to B12 deficient vegetarians and B12 biomarkers were analysed. c) 2X200 mL of cow/buffalo and equimolar concentration of CN-B12 was supplemented for 8 weeks to B12 deficient vegetarians d) LR-82 (Milk extracted B12) / CN-B12 (5.6 μg/day) was supplemented to B12 deficient vegetarians. CN-B12 treated groups have higher circulating vitamin B12 and holotranscobalamin concentrations and absorptive capacity was reached only by doses above 3 μg of cobalamin.3 μg/day for 8 weeks did not improve the B12 status. Consumption of cow milk was able to reduce plasma homocysteine concentration. 5.6 μg of LR-82 could improve and sustain B12 status. The study concludes that Daily recommended

dose of vitamin B12 in lactovegetarians needs to be amended. Milk extracted vitamin B12 (HO-B12) has better health beneficial effects as compared to CN-B12 tablets.

O-023

Radioprotective Immunomodulatory Activity of Grape (Vitis vinifera) Extracts in Human Lymphocyte

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Ionizing radiation (IR) causes oxidative stress through the overwhelming generation of reactive oxygen species (ROS) in the living cells leading further to the oxidative damage to biomolecules. Grapes *(Vitis vinifera)* contain several bioactive phytochemicals and are the richest source of antioxidant. In this study, we investigated the levels of phytochemicals, profile of enzymes, ROS- and oxidant-scavenging activities, and the radioprotective activities in human lymphocytes of the grape extracts of four different cultivars, including the Thompson seedless, Flame seedless, Kishmish chorni and Red globe. The activities of ascorbic acid oxidase and catalase significantly ($p<0.01$) differed among extracts within the same cultivar, while that of peroxidase and polyphenol oxidase did not alter significantly among extracts of any cultivar. The superoxide radical-scavenging activity (SRSA), ferric-reducing antioxidant power (FRAP) assay and ABTS were higher in the seed as compared to the skin or pulp of the same cultivar. Pre-treatment with grape extracts attenuates oxidative stress and DNA damage induced by 4 Gy -radiation in human lymphocytes in vitro; and also maintains balance between pro- and anti-apoptotic markers. The single cell gel electrophoresis revealed that pretreatment of grape extracts significantly reduced the increase in the tail length in gamma irradiated lymphocytes. Further, -radiation induced increase in caspase 3/7 activity and mRNA expression of p53 and Bax were significantly attenuated by grape extracts. These results suggest that grape extract serve as a potential source of natural antioxidants against the IR-induced oxidative stress, inhibit apoptosis and significantly prevent radiation-induced cellular DNA damage. Furthermore, the protective action of grape depends on the source of extract (seed, skin or pulp) and type of the cultivars.

O-024

Macronutrient and Micronutrient Status in Pulmonary Tuberculosis and their Correlation with Disease Severity and Inflammation.

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Tuberculosis is a socio-economic and a highly contagious disaster that spread through cough, sneeze, spit of infected persons and caused by Mycobacterium Tuberculosis. Malnutrition is the most prevalent risk factor of TB associated with minor and major complications, influence disease outcome, reactivation of latent infection and mortality. Role of macro and micronutrients and their correlation to disease severity and inflammation was evaluated. The study was conducted on 60 newly diagnosed and 60 relapsed Pulmonary Tuberculosis (PTB) patients recruited from the "Institute of Respiratory Disease, SMS Medical College Jaipur". Sixty healthy individuals were used as controls. Sputum positivity was obtained by direct smear microscopy and Bandim TB score was used for severity of disease. Anthropometric measurements, routine and specific biochemical investigations (such as micronutrients-Vitamin D, β-carotene, Cu, Zn, Fe and Prealbumin, Transferrin, Ferritin, Cerruloplasmin), CRP and ADA were assessed and statistically evaluated. This study reported that BMI, MUAC, Total cholesterol, Triglyceride, HDL-cholesterol, Protein and Albumin were significantly reduced in PTB than controls. None of the macronutrients showed significant correlation with disease severity and sputum positivity. However, calcium corrected for hypoalbuminemia was high in newly diagnosed and relapsed PTB and showed weak positive correlation to disease severity and sputum positivity. Vitamin D correlated negatively to disease severity and sputum positivity. Among other micronutrients, Cu was raised in PTB patients. Zn and β-carotene levels were reduced. β-carotene correlated negatively to CRP, ADA and to sputum positivity. Altered iron profile with hypoferremia, hypotransferrrinemia and hyperferritinemia was observed in our PTB cases. Negative association of transferrin to disease severity and ADA was observed indicating its role in monitoring treatment response. Prealbumin was significantly reduced in PTB than controls. Our study showed essential linkage between disease severity and macro-micronutrient status. Study of micronutrient metabolism could also help in understanding their role in pathophysiology of PTB.

O-025

Dysregulated Iron Homeostasis in Ulcerative Colitis

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Hepcidin is the central hormone involved in systemic iron homeostasis. It is known to be regulated by a number of factors. For example, anemia and erythroid factors (such as erythroferrone [ERFE] and growth differentiation factor 15, both of which are released from the bone marrow during erythropoiesis) down-regulate it. On the other hand, inflammation up-regulates it. So, when anemia and inflammation are both present, which of these opposing factors has a predominant influence on hepcidin? To attempt to answer this question, we studied patients with ulcerative colitis (UC), an inflammatory bowel disease, in which anemia often co-exists with inflammation. Hemoglobin and serum iron levels were significantly lower and C-reactive protein levels (a marker of inflammation) significantly higher in patients with UC, than in those without. Despite the presence of systemic inflammation, however, serum hepcidin concentrations were found to be significantly lower in these patients; they also had significantly higher levels of serum ERFE, when compared to control subjects. When patients with UC were stratified into those with and without anemia, it was found that anemic patients had significantly lower hepcidin and higher erythroferrone levels than non-anemic subjects. These results indicate that, when anemia and inflammation co-exist, the effect of anemia on hepcidin appears to dominate over that of inflammation. This effect is possibly mediated through increased ERFE levels.

O-026

Prevalence of Food Specific IgG Antibody Levels in a North Indian Population

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Food intolerance, mediated by food specific IgG antibodies, have been implicated in a variety of disorders like inflammatory bowel disease, Crohn's disease, migraine and more recently, atherosclerosis and asthma. Its prevalence is 5-20% and is associated with many specific and non-specific symptoms. Food intolerance, unfortunately, mostly goes undiagnosed and is often confused with IgE mediated food allergy. To investigate the prevalence of food specific IgG antibodies in patients presenting clinically with allergic symptoms but no laboratory diagnosis of allergy. 150 patients were screened for food specific IgG antibodies to 56 foods by semi-quantitative ELISA (Food Detective IS Professional, Omega Diagnostics) for mild, moderate and severe intolerance. The most frequently occurring food specific IgG antibodies were against the foods commonly consumed in the Indian diet i.e. cow's milk (70%) followed by mushroom/yeast (42%), semolina (36.7%), gluten (32%), legume mix (30%), cashew (28%), almond and chana dal (24.7% each), whole egg (24%), wheat (22%) and moong dal (21.3%). Though 55.3% males and 44.7% females showed presence of food specific IgG antibodies, females showed intolerance to more types of foods. Significantly higher intolerance was observed in males only for cow's milk (p=0.008), and in females for legume mix (p=0.038). Incidence of intolerance was highest in the 0-10 years group (40%) followed by the 26-40 years group (22.7%) and 41-55 years group (18%). Significant association of food specific IgG was observed for semolina (p=0.042), gluten (p=0.000) and cow's milk (p=0.003) in 0-10 years and for flax seeds (p= 0.002), almonds (p= 0.015) and mushrooms/yeast (p=0.000) between 41-55 years. To conclude, IgG mediated food intolerance causes significant preventable morbidity. Hence, food elimination, based on detection of food specific antibodies, would be effective in reducing, not only the symptoms, but also the associated financial burden of treatment.

O-027

Status of Serum Vitamin D in a Rural and Urban Population from the NCR Region, India and Comparison of Levels based on Two Estimation Methods

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Vitamin D deficiency appears to be rampant amongst Indians despite India being a sunlight sufficient country. Vitamin D levels were estimated using a Diasorin Chemiluminescent Immunoassay in a sub-set of rural and urban samples (n=1403) that were collected as part of a previous ICMR sponsored study. The mean Vitamin D levels in samples from the rural population was 17.6 ng/ml and in those from urban population was 8.8 ng/ml. The mean Vitamin D levels in males and females were 14.0 and 12.5 ng/ml respectively. Among the urban group, 70% had severe vitamin D deficiency, 26.7% had levels between 10-20ng/ml and 1.9% between 20-30 ng/ml. Just 1% of the urban population had levels above 30 ng/ml. The respective percentages for severe deficiency, deficiency, insufficiency and adequacy were 19.7, 46.6, 25.2 and 8.6 % respectively in the rural group. Mean Vitamin D levels were 14.2 and 11.4 ng/ml respectively in groups with primary/high school level educational and higher secondary/graduate level education ($p < 0.01$). There was an inverse correlation between

BMI and Vitamin D levels, with people with BMI < 20, between 20-25, between 25-30, and those above 30, having mean levels of 14.9 (9.3), 13.7 (8.4), 12.3 (7.9), and 11.2 (7.3) ng/ml respectively. The Bland-Altman test was used to test for the agreement between the two methods using the values obtained from CLIA and LC-MS for the rural and urban serum samples (n=157). There was poor agreement between the Vitamin D values obtained by CLIA and the sub-set of samples analyzed by LC-MS. In general, the CLIA method appeared to underestimate the Vitamin D levels to varying degrees with differences greater at higher levels of Vitamin D. There is a need to implement strategies to improve Vitamin D levels. However, considerations on optimal cut-offs based on estimation methods are also required.

O-028

Iron Homeostasis is Dysregulated, but the Iron-hepcidin Axis is Functional, in Chronic Liver Disease

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Perturbations in iron homeostasis have been reported to be associated with irreversible liver injury in chronic liver disease (CLD). However, it is not clear whether liver dysfunction per se underlies such dysregulation or whether other factors also contribute to it. This study attempted to examine the issues involved. Patients, diagnosed to have chronic liver disease (n=63), who underwent a medically-indicated upper gastrointestinal endoscopy, were the subjects of this study. Patients with dyspepsia, who underwent such a procedure, and were found to have no endoscopic abnormalities, were used as control subjects (n=49). Duodenal mucosal samples were obtained to study mRNA and protein levels of duodenal proteins involved in iron absorption. A blood sample was also obtained for estimation of hematological, iron-related, inflammatory and liver function-related parameters. Patients, with CLD had impaired liver function, anemia of inflammation and lower serum levels of hepcidin than control subjects. Gene (mRNA) expression levels of duodenal ferroportin and duodenal cytochrome b (proteins involved in iron absorption) were decreased, while that of divalent metal transporter-1 (DMT-1) was unchanged. Protein expression of DMT-1 was, however, decreased while that of ferroportin was unchanged. CLD patients with serum ferritin greater than 300 µg/dL had significantly greater liver dysfunction (as indicated by significantly higher serum concentrations of bilirubin, AST and ALT, and MELD scores), higher serum concentrations of CRP and hepcidin, and lower ferroportin protein expression, than those with serum ferritin >300 µg/dL. In conclusion, anemia of inflammation and low serum hepcidin levels were found to paradoxically co-exist in patients with CLD. Expression of duodenal proteins involved in iron absorption were either decreased or unaltered in these patients. The hepcidin response to higher body iron levels and/or inflammation appeared to be functional in these patients, despite the presence of liver disease.

O-029

Prevalence of Biotin Supplement Ingestion in Outpatients and Emergency Department

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Biotin interferences have been reported in immunoassay methods. Immunoassays have been used in several laboratory tests such as thyroid function, drugs, hormones, tumor markers, and cardiac markers. The aim of this study was to determine the prevalence of biotin supplement ingestion in outpatients and emergency department. This was a cross-sectional study conducted at Siriraj hospital, a tertiary-care university hospital in Thailand, during June-July 2019. Data were collected through interview by a questionnaire inquiring about demographic data, departments visited, types of clinic, underlying diseases and supplements taken. Three hundred and thirty-eight patients (259 female, 79 male) with mean (SD) age of 54.9 (16.8) were enrolled. Overall prevalence of biotin supplement ingestion was 6.8% (23/338). All of the patients received biotin from vitamin B complex which contained 150 mg of biotin. Percentages of biotin taken were not different when compared between outpatients and patients in emergency department (6.4% vs 18.2%, P =0.128), and between patients attended regular clinics and after-hour clinics (6.8% vs 6.8%, P = 0.993). Among 23 patients who took biotin, 4 had either thyroid or cardiac diseases. From our survey results, the prevalence of biotin used was common, but the dosage of biotin was low. There were no differences of biotin used among different departments and clinics.

O-030

Vitamin D Insufficiency Correlates with Decreased Levels of Active Folate in Postpartum Women in Spite of Folate Supplementation

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Vitamin D is known to influence folate levels through improved functioning of the intestinal microbiome and/or absorption by upregulation of proton coupled folate transporter at intestine in animal models. As this relationship is unknown in humans, the study aims to analyze the relationship between vitamin D status and the active form of folate, 5-methyl tetra hydro folic acid (5-methyl THFA) in postpartum women on folate supplementation. At 6 weeks postdelivery, 434 women were studied for plasma levels of 25-OH vitamin D and 5-methyl THF by ELISA. As the data was non-normal in distribution, log-transformed values were used for statistical analysis. According to Mayo classification of vitamin D insufficiency (Mayo Clin Proc. 2010;85(8):752-758), the cohort was divided into three groups as optimal (25-80 ng/ml), mild to moderate deficiency (10-24 ng/ml), and severe deficiency (<10 ng/ml). The study observed a significant low levels of 5-methyl THF levels in mild-moderate vitamin D deficiency women in comparison to women with optimal vitamin D levels using Kruskal Wallis test. Linear regression analysis revealed a positive association between 25-OH vitamin D and 5-methyl THF in whole cohort ($r=0.109$; $p = 0.002$). Therefore, the study concludes the possibility of low folate levels, in spite of folate supplementation in women if associated with vitamin D insufficiency.

O-031

Neonatal Screening for Congenital Hypothyroidism and G6PD Deficiency in a Tertiary Care Hospital in Chhattisgarh

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The fundamental pre-requisite of a screening program is the accessible epidemiological data regarding disease burden. This is the foremost study in this state to estimate the burden of congenital hypothyroidism and glucose-6-phosphate dehydrogenase enzyme deficiency in infants born or attending a tertiary care hospital. The screening was conducted on 1282 babies, 48 hours subsequent to birth and upto 8 weeks of age. The dried blood spot specimens collected were analysed for thyroid stimulating hormone level and glucose-6-phosphate dehydrogenase enzyme activity by fluoro-immunoassay method. The proficiency of the program for all live birth babies delivered in the institute was 94.6%. In consequence to improper sampling, 4.1% samples could not be analysed. The screening tests reported elevated TSH values in 1.6% of total population and glucose-6-phosphate dehydrogenase enzyme deficiency in 2.6% of all babies enrolled. Confirmatory tests revealed that 4 (3.1/1000) babies were declared positive for congenital hypothyroidism and 8 (6.2/1000) were announced to be G6PD deficient. The new prevalence data urges mandatory and immediate need for development of neonatal screening program in the state. The recommendations for improvisation and up-gradation of regional diagnostic centre with facilities for primary and secondary level testing would be more cost effective in contrast to private partnership.

O-032

Serum Cortisol and Thyroid Profile in Diagnosis of Hyponatremia

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Hyponatremia is the most prevalent electrolyte derangement with multifactorial origin. Euvolemic hyponatremia is associated with underlying Syndrome of inappropriate antidiuretic hormone secretion (SIADH), hypothyroidism and adrenal insufficiency, which are often overlooked in clinical diagnosis. Hence this study was conducted with an objective to evaluate the proportion of thyroid and adrenal derangement in patients with hyponatremia and to correlate this with severity of Hyponatremia and Plasma osmolarity. A Hospital based cross sectional study was conducted with 60 clinically diagnosed euvolemic hyponatremia patients without co-morbidities like Diabetes Mellitus, congestive Cardiac failure, Nephrotic Syndrome, severe diarrhea with vomiting as well as patients with Thiazide diuretics . Thyroid profile i.e. Free T3 (FT3), Free T4 (FT4) and TSH and cortisol levels were estimated along with fasting Blood Sugar, Urea, Potassium Uric acid and Creatinine. A significant difference in the osmolarity and sodium levels was found in cases as compared to control. Nineteen patients had thyroid derangement with 17/19 cases with hypothyroidism, more frequently (08/17) seen in severe hyponatremia. The alteration in serum cortisol levels was found in 24/60 patients with low levels (28ng/ml) in 17/60 cases which was more predominant in Moderate hyponatremia (08/29). Correlation study showed a negative association between sodium and parameters like TSH, Cortisol, uric acid and a positive association with total protein. However none of the associations were statistically significant.With these findings we conclude that patients

with euvolemic hyponatremia should be thoroughly investigated for adrenal and thyroid hormone status to reach to an etiological diagnosis and these parameters would help in treating the underlying disorders associated with in case of hyponatremia .

O-033

Vitamin D, Vitamin D Binding Proteins and VDR Polymorphisms in Individuals with Hyperglycaemia

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Vitamin D (25(OH)D) deficiency and polymorphisms in the vitamin D receptor gene (VDR) are associated with increased risk for diabetes, however findings are inconsistent and inconclusive. We aimed to investigate the association between vitamin D, vitamin D binding proteins (VDBP) and vitamin D receptor (VDR) polymorphisms in individuals with prediabetes and type 2 diabetes mellitus (T2DM) from South Africa. The current study comprised of 1603 participants (387 males). Vitamin D levels were measured using the paramagnetic particle chemiluminescence assay. ELISA was used to measure VDBP and VDR single nucleotide polymorphisms, Fok1 (rs2228570), Apa1 (rs7975232) and Taq1 (rs731236) were genotyped using the TaqMan assays and confirmed by direct sequencing. Vitamin D statuses was classified as follows: deficient levels <20 ng/mL, insufficient levels 20-29 ng/mL and optimal levels 30 to 100 ng/mL. The 25(OH)D deficiency (44%) and insufficiency (42.6%) were highly prevalent whilst optimal 25(OH)D levels were found in 13%. Vitamin D and VDBP varied according to gender with males having higher 25(OH)D levels than females, 23.6±7 vs 21.5±7.5ng/mL, p=0.0006, respectively, whilst females had significantly higher serum VDBP levels 299.1±71.2 vs 315.9±76.1 µg/mL, p<0.0001. Although no significant differences were observed in the expression of VDBP, 25(OH)D levels were significantly decreased in subjects with T2DM compared to controls, respectively 17.0±6.1vs 24.2±8.2, p<0.0214. In multiple linear regression analysis, low 25(OH)D was associated with increased LDL-C and PTH in both males and females irrespective of glycaemic status. The allele and genotype distribution were not significantly different across glucose tolerance statuses. This study showed high occurrence of vitamin D deficiency/insufficiency in this South African population. Furthermore, vitamin D levels are decreased in individuals with hyperglycaemia and this is not linked to VDBP or polymorphisms in the VDR gene. The mechanisms effecting lower vitamin D levels in individuals with hyperglycaemia needs further investigation.

O-034

Molecular Basis of Insulin Signaling Pathway in Pathogenesis of Polycystic Ovary Syndrome (PCOS).

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Polycystic ovary syndrome is a heterogeneous disorder affecting millions of women and most common cause of anovulatory infertility. Insulin resistance plays key role in its pathogenesis & reproductive abnormalities. Post binding defects in insulin receptor signaling is now considered major molecular basis of insulin resistance in PCOS. Single nucleotide polymorphism in tyrosine kinase domain of insulin receptor gene has been shown to be significantly associated with development of PCOS. Insulin receptor substrate proteins are also critical for insulin mediated signal transduction. Besides, genetic polymorphism is distinct in specific population, ethnicity, geographical region due to exposure to different environmental factors. Keeping in view present study was conducted to explore genes involved in insulin signaling and risk of PCOS in north Indian females. A case control study was conducted in department of biochemistry & department of obstetrics & gynecology at VMMC & Safdarjung hospital, New Delhi. 100 cases of PCOS & 100 controls were included in this study. The DNA extraction was done using KRISHGEN DNA kit & genotyping was done by PCR-RFLP. Subjects with CT genotypes had 2.47 times higher risk of developing PCOS as compared to CC genotype (OR=2.47, CI=1.3-4.6, p-value= 0.006). Subjects with TT genotype have 5.6 times higher risk of developing PCOS as compared to CC genotype (OR=5.6, CI=2.41-13, p-value≤0.001). Both CT & TT genotype of insulin receptor (INSR) gene SNP (rs1799817 C->T) is associated with hyperinsulinemia, insulin resistance & increased susceptibility to PCOS. Insulin receptor gene may be considered as a candidate gene for predisposition to PCOS & its metabolic dysfunction. Both CT &TT genotype of INSR gene SNPs (rs1799817) constitutes a significant proportion of general population. Hence, it could be screened & therapeutic approach can be designed accordingly to lessen co-morbidities associated with PCOS.

O-035

Randomized Control Study to Assess the Effect of Aqueous Extract of Trigonella Foenum Graceum (Fenugreek) Seeds on Glycaemic Control by Measuring Insulin Resistance (HOMA-2) in Normal, Metabolic Syndrome and Type 2 Diabetic Patients

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Insulin resistance is important metabolic abnormality associated with metabolic syndrome and type II diabetes. Fenugreek seeds have been reported to have a multiple benefits to control diabetes. It is part of Indian cuisine. Open-label, randomized, controlled, prospective study of 3 months duration included 253 subjects. They were divided into subgroups- healthy (Control 30, test 31), metabolic syndrome (Control 30, test 30), diabetes group (Control 30, test - newly diagnosed for monotherapy 68, and fully developed cases as adjunct therapy 30). Control group did not receive medicine. Test group was administered 4 tablets of Fenugreek / day (1.32 gm extract equivalent to 13.2 gm Fenugreek seed powder). Follow up was done by HOMA II (computerized model with S. Insulin, Fasting blood sugar) and HbA1c. Study was carried out after ethical committee clearance under supervision and guidance of physician and in real life situation. In test group: base to 3 months mean ± SD FBS (mg%): Healthy 85.19±5.42 to 75.71±5.57; metabolic syndrome 105.21±3.98 to 76.94±4.08; Monotherapy 157.91±11.24 to 118.06±5.80; Adjunct 257.00±42.19 to 197.00±38.83. HbA1c (%): Healthy 4.59±0.63 to 4.59±0.63, metabolic syndrome 5.68±0.59 to 5.03±0.67; Monotherapy 7.38±0.48 to 7.07±0.36; Adjunct 11.63±1.61 to 10.35±1.04.S. Insulin (μIU/ml): Healthy 5.89±1.09 to 5.65±1.05; metabolic syndrome 21.41±17.68 to 14.72±6.03; Mono therapy 15.42±4.60 to 12.46±3.63; Adjunct 18.60±6.18 to 14.96±4.13. HOMA IR II (%): Healthy 0.73±0.15 to 0.70 ± 0.15, metabolic syndrome 2.45±0.92 to 1.80±0.73; Mono therapy 2.27±0.71 to 1.69±0.52; Adjunct 3.36±1.35 to 2.32±0.69. p value<0.05 for FBS, HbA1c, HOMA in metabolic syndrome and diabetic group. Values were not significantly altered in healthy subjects. In control group (healthy, metabolic syndrome and diabetic group) base to 3 months no significant changes. Safety parameters unaltered, no serious side effects. Aqueous extract of Fenugreek in tablet form is effective to control metabolic disturbances- hyperglycemia and insulin resistance in metabolic syndrome and type 2 diabetes.

O-036

Effect of Tulsi (Ocimum Sanctum Linn.) Supplementation on Metabolic parameters in Young Overweight and Obese Subjects

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Ocimum sanctum (also known as Tulsi) is a sacred Indian plant, the beneficial role of which, in obesity and diabetes is described traditionally. This is a randomized, parallel group, open label pilot study to investigate the effect of Ocimum sanctum on metabolic and biochemical parameters in thirty overweight/obese subjects, divided into two groups A and B. Group A (n=16) received one 250 mg capsule of Tulsi (Ocimum sanctum) extract twice daily in empty stomach for 8 weeks and group B (n=14) received no intervention. Statistically significant improvements in the values of serum triglycerides (p=0.019); low density lipoprotein (p=0.001); high density lipoprotein (p=0.001); very low density lipoprotein (p=0.019); Body Mass Index, BMI (p=0.005); plasma insulin (p=0.021) and insulin resistance (p=0.049) were observed after 8 weeks in the Ocimum sanctum intervention group. The improvement in HDL-C in the intervention group when compared to the control group was also statistically significant (p=0.037). There was no significant alteration of the liver enzymes SGOT and SGPT in both the intervention (p=0.141; p=0.074) and control arms (p=0.102; p=0.055) respectively. These observations clearly indicate the beneficial effects of Ocimum Sanctum on various biochemical parameters in young overweight/obese subjects.

O-037

Serum Testosterone Level and Erectile Dysfunction among Type 2 Diabetes Mellitus.

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Diabetes is ranked the top five non communicable disease of the world including the southeast Asia like India and Nepal. Besides the notorious complications like renal failure and acute coronary syndrome, conditions having the psychosocial impact like erectile dysfunction is also worth looking into. Lower Serum Testosterone (LST) level and Erectile dysfunction (ED) are the distressing complications of diabetes contributed by impaired vasodilatory signalling, smooth muscle cell hyper contractility, microvascular changes, neuropathy and endothelial dysfunction due

to hyperglycemia and hypogonadism. Male patients visiting medicine department of Tribhuvan University Teaching Hospital with Type 2 Diabetes Mellitus (T2DM) were enrolled in this cross-sectional study. A pre-structured questionnaire was used for demographic data. A validated questionnaire, an abridged 5-item version of International Index of Erectile Function (IIEF-5), was used to assess ED where ED was established when IIEF score came. Blood samples were taken and assayed for Serum Total Testosterone (STT) and Glycated Hemoglobin (HbA1c). A total of 65 patients were enrolled in which the mean age of patients was 47 years and 75.38 % of them showed various degree of ED. A total 55.38 % patients had STT level > 300 ng/dl whereas 44.62 % had lower level. The occurrence of ED among T2DM patients with hypertension was much higher as compared with T2DM only (p=0.05). However, there was no significant difference in the level of STT between T2DM males with ED and T2DM without ED[331.33±138.36 ng/dl vs359.0±148.8 ng/dl, p=0.49].A significant (p=0.001) negative correlation was seen between IIEF score and HbA1c level [r=-0.617]. An increase in Body Mass Index (BMI) is also significantly associated with decrease in STT level (p=0.003). The higher prevalence of diabetic ED was shown in the study which was also attributed with higher HbA1c level, BMI and hypertension.

O-038

Comparison of Formulae for Calculated Low-Density Lipoprotein Cholesterol with Direct Assay in Patients With Hypothyroidism

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Low-density lipoprotein cholesterol (LDL-C) is an established cardiovascular risk factor. As LDL-C is associated with cardiovascular risk stratification, formula for calculation of LDL-C should have good accuracy with minimal bias. The aim of present study was to compare nine different formulae for Calculated LDL-C with LDL-C by direst assay in patients with subclinical and overt hypothyroidism. In this analytical cross-sectional study, a total of 105 patients with laboratory evidence of subclinical and overt hypothyroidism presenting in AIIMS Jodhpur were recruited and blood samples were subjected for lipid profile analysis at Clinical Biochemistry Lab. The results of triglycerides, total cholesterol, HDL-cholesterol and LDL-cholesterol by direct assay were analyzed. Calculated LDL-C was assessed by different formulae. The study observed that calculated LDL-C by all 9 different formulae significantly differed from Direct LDL ($p< 0.05$). The study findings revealed that LDL_Friedewald, LDL_Cordova, LDL_Anandaraja, LDL_Hattori and LDL_Chen have bias less than ±5 when compared with Direct LDL and LDL_Anandaraja having the lowest bias (2.744) among them. Out of all different formulae, LDL_Cordova and LDL_Hattori had shown negative bias with comparison to Direct LDL. On intra class analysis based on triglyceride levels, LDL_Friedewald and LDL_Hattori had bias less than ±5 except when Triglyceride is more than 300 mg/dL (Bias -10.87 and -18.20 respectively). LDL_Anandaraja had bias less than ±5 within the Triglyceride values of 100-300 mg/dL. LDL_DeLong, LDL_Rao and LDL_Teerankanchana were found to consistently give overestimated LDL values when compared with Direct LDL. In conclusion, the Friedewald and Anandaraja formulae outperformed other for estimating LDL-C against a direct measurement. Among the formulae LDL_Anandaraj had a lesser bias. The Friedewald and Anandaraj formula could be used as an alternative cost-effective tool to measure LDL-C when the direct measurement cannot be afforded in hypothyroid patients. Key words: LDL-cholesterol, hypothyroidism, Friedewald-formula, Anandaraj-formula, lipid-profile.

O-039

Antidiabetic Activity of Azadirachta Indica: Comparison of Young and Matured Leaves

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Azadirachta indica (Neem) is used for its anti-diabetic properties in India, Pakistan, and Nigeria; and is also recognized in Ayurveda. The present study was undertaken to investigate the anti-diabetic capacity of the ethanolic extract of the matured and young leaves of Azadirachta indica in streptozotocin induced diabetic rats. Azadirachta indica leaf extract (AIOLE) was orally administered at 400mg/kg body weight (BW) dose to Streptozotocin induced diabetic rats. After 28 days consecutive treatment, various diabetic parameters were studied and compared with the untreated rats. Furthermore, Laser Induced Breakdown Spectroscopy (LIBS) and Fourier Transformed Infra-red Spectroscopy (FTIR) were used to determine the elemental analysis and functional groups respectively. The matured and young leaf extract demonstrated antihyperglycemic activity by reducing 75.4% blood glucose level after 28 days treatment. Oral glucose tolerance test (OGTT) revealed increasing glucose tolerance as shown by a 72% decrease in blood glucose in 3 hours post treatment. Treatment of the diabetic rats with ethanolic extract of leaves of matured and young A. indica at a dose of 400 mg/kg body weight for 28 days resulted in gradual but significant normalization of plasma sialic acid (N-acetyl neuraminic acid -NANA) level, erythrocyte reduced glutathione (GSH), erythrocyte malondialdehyde (MDA) content, plasma antioxidant activity by FRAP method, erythrocyte plasma membrane redox system (PMRS) activity. Different elemental compositions were identified with LIBS and the FTIR revealed the different functional groups present in the matured and young

leaf. The present investigation revealed that Azadirachta indica possesses potent anti-diabetic activity as claimed in different ethno pharmacological practices.

O-040

Type 2 Diabetes Mellitus - Use of eGFR and Microalbumin in Concurrence to Detect and Prevent CKD

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According to World Health Organization, Type 2 diabetes mellitus (DM) has now become a global health problem due to rapidly increasing urbanization and high prevalence of obesity and physical inactivity. As per estimates nearly 98 million people in India may have type 2 diabetes by 2030. DM is the leading cause of chronic kidney disease (CKD) and is defined by elevated urine albumin excretion or reduced glomerular filtration rate (GFR) or both and occurs in 20% to 40% of all diabetics. Aim of this study was to evaluate diabetic patients in routine for the Microalbumin and eGFR to detect and monitor CKD. We conducted this study on 500 patients with type 2 DM. The parameters studied included diabetic profile (glycemia), urinary microalbumin, renal function test with eGFR (using the Cockcroft Gault formula).A significant ($p<0.0001$) correlation between eGFR and microalbuminuria was observed. It was found that 63.2% patients had high microalbumin and normal eGFR and 28.2% patients had normal microalbumin and normal eGFR and 7.6% had normal microalbumin and low eGFR. It was found that with increase in blood glucose and microalbumin level, the eGFR decreases i.e. increase the risk factor to CKD. Screening of all individuals with diabetes is recommended to detect changing levels of albuminuria and renal function (i.e., eGFR). Our results indicate that these two parameters provide a complimentary benefit in diagnosis and management of diabetic patients with CKD. Hence, we recommend that laboratories should calculate and report an eGFR with every request for renal function tests in adults to determine the presence of renal disease, stage of CKD and to monitor response to treatment. This will allow reducing the risks of kidney failure progression in DM patients without any additional cost to the patient.

P-001

Involvement of Xanthine Oxidase-derived Reactive Oxygen Species in Cardiac Oxidative Stress in Rats Fed a High-fructose Diet

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Oxidative stress is induced via reactive oxygen species (ROS) in the living body. It has been reported that cardiac oxidative stress occurs in rats with high fructose-induced metabolic syndrome (MetS). However, the mechanism by which cardiac oxidative stress is induced in rats with high fructose-induced MetS is still unknown. Therefore, we examined whether ROS are involved in cardiac oxidative stress in rats with high fructose-induced MetS diet by treating with allopurinol (AP), which works as both a xanthine oxidase (XOD) inhibitor and a free radical scavenger. Male Wistar rats were pair-fed either a diet containing 60% fructose (HFD) or a diet containing 60% dextrose used as the control diet for 4 or 6 weeks. AP dissolved in drinking water, at a dose of 20 mg/kg body weight/day was administered to rats with HFD feeding daily for 2 weeks, starting at 4-week HFD feeding. Rats fasted for 15 h were killed under anaesthesia. Serum was used for assays of glucose, triglyceride, uric acid, free fatty acids, total-cholesterol, and lipid peroxide (LPO). Hearts isolated from rats were used for assays of LPO, ascorbic acid, reduced glutathione (GSH), NOx (NO_2^-/NO_3^-), XOD, and ROS. Rats fed HFD for 4 and 6 weeks had increased serum triglyceride, uric acid, free fatty acids, total-cholesterol, and LPO levels. AP administration attenuated these increases found at 6-week HFD feeding. At 4- and 6-week HFD feeding, cardiac LPO and ROS levels and XOD activity increased and cardiac ascorbic acid level decreased. At 6-week HFD feeding, cardiac NOx level increased and cardiac GSH level decreased. AP administration attenuated the changes in these cardiac oxidative stress parameters found at 6-week HFD feeding. These results indicate that ROS derived from XOD are involved in cardiac oxidative stress in rats with high fructose-induced MetS.

P-002

Discovering Association between Serum Liver Enzymes and Metabolic Syndrome in Urban Thai Population: A Data Mining Approach

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Liver enzymes included ALT, AST and ALP were used to investigate liver abnormality. The aims of this study to determine association between liver enzymes and metabolic syndrome (MS), and find cutoff point of the liver enzymes to identify MS as well as classification of MS using data mining, a decision tree analysis. A data set composed of 14,355 individuals who received annual checkup from the Mobile Health Unit, Faculty of Medical Technology, Mahidol University during January - December 2015. The MS was identified using the International Diabetes Federation criteria. The optimal cutoff point of each liver enzyme was performed using receiver operating characteristic curve and a decision tree analysis was used to classify individuals with and without MS based on MS components and liver enzymes. The overall prevalence of MS was 46.83% which are 52.23% and 42.71% in males and in females, respectively. The elevated serum liver enzyme displayed associated with increased number of MS components. Furthermore, optimal cutoff point of serum liver enzymes exhibited 22.50, 21.50, 68.50 U/L for ALT, AST, and ALP, respectively. In addition, MS and non-MS was classified using decision tree analysis which gave accuracy of 96.89% based on MS components and serum liver enzyme while accuracy of 59.81% based on only serum liver enzyme. Interestingly, triglyceride and ALT enzyme was identified as the most important parameters for classifying MS. Elevated levels of serum liver enzymes were associated with MS, and optimal cutoff points as well as decision tree models were employed for identification of MS. These finding could be potential to use early diagnosis of MS, and prevent fat accumulation in liver lead to non-alcoholic fatty liver disease (NAFLD).

P-003

Structure Activity Relationship Studies for HDAC Subtype Selective Drug Designing

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Histone deacetylase (HDAC) subtypes play an important role in post-translational modification of various proteins involved in cancer origin and development, so are used as cancer drug target. There are four classes of HDACs, class I, II and IV have 11 subtypes having Zn binding motif and class III has 7 sirtuins based subtypes which differentially express in cancer types. These subtypes are involved in multiple signaling pathways at different molecular levels. These Zn binding HDACs share significant structural similarity; they are different in sequence length but active sites lie at a similar segment and also have 6 patches of 2-3 amino acid conserved sequences along the subtypes. Therefore, pan HDAC inhibitors such as Vorinostat, Trichostatin etc. face selectivity issue. Designing subtype specific inhibitors may resolve the problem of HDAC selectivity. In this study we attempted to comparatively analyze the ligand binding features in the binding sites of different subtypes taking reported active and inactive compounds for each subtype. A total of about 3000 active molecules with IC50 values ranging from 0.002 to 1000 nM and about 1000 inactive molecules with IC50 more than 5000 nM reported for 11 subtypes of HDACs were collected from ChEMBL database. Glide docking was performed using respective crystal structures and interactions of active and inactive molecules with the corresponding crystal structures were analyzed. Ligand interaction analyses show D269, D176, E98, for HDAC1, K25, R301, R265, R275, for HDAC3, H122, G331 for HDAC4, S568, N494 for HDAC6 and K33, H142 for HDAC8 are the unique residues that can be targeted for selective drug design. QSAR modelling taking active and inactive molecules is in progress to get further insights into the chemotype selectivity's of different HDACs.

P-004

Computer Application in a Clinical Laboratory: Report of an Experience in a Developing Country

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Nowadays, computer technology has become a necessity in the medical field in general and in clinical laboratories in particular. In fact, the use of computer software in clinical laboratories has revolutionized the management of the various medical analysis. We report our experience in the clinical laboratory in the regional hospital of Ben Arous, a governorate in a developing country (Tunisia).The hospital provides inpatient and outpatient care. Our laboratory is a multi-purpose laboratory (biochemistry, hematology, bacteriology and parasitology) in which we carry out an average of 350 analysis requests a day with a 24 hours per day, 7 days a week activity. The use of a computer application called "SANTE LAB" started since February 2018. This application was developed by the computer center of Tunisian ministry of health. It starts from the registration of analysis requests to the validation of results. It allowed the centralized registration of analysis requests, their transmission to the instruments and the collection of results from the instruments to the computer. In addition, the computer application is connected to care services in the hospital which allowed the computerized transmission of results. It also improved the different stages of analytical process, minimised the risk of errors, ensured traceability and let possible follow up by showing antriorities. Moreover, it allowed the management of emergency analysis requests for a rapid transmission of results. Despite the progress made from one version to another, this application still has some limits and needs more improvement.

P-005

Significance of Serum Albumin as a Prognostic Factor before Treatment in Multiple Myeloma

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Multiple myeloma (MM) is a malignant plasma cell disorder that accounts for approximately 10% of all hematologic cancers. It usually evolves from an asymptomatic premalignant stage of clonal plasma cell proliferation termed "monoclonal gammopathy of undetermined significance" (MGUS). Serum albumin level, in association with serum beta2 microglobulin level, is a significant prognostic factor in multiple myeloma patients. The aim of this study was to investigate whether serum albumin has utility as a prognostic indicator of cancer survival in Multiple myeloma. We retrospectively reviewed the records of 60 patients diagnosed with multiple myeloma at RGCI & RC. Serum Albumin level was measured by spectrophotometric assay based on BCG on Beckman AU 480. Serum Albumin values are expressed as mean ± 2 SD. Chi-square test is used to find an association between two groups according to serum albumin level (above or below 3.5 g/dL 95% CI). Significance is considered at p-value < 0.05. Patients were divided into two groups according to serum albumin level (above or below 3.5 g/dL, the prognostic cutoff value). We aimed to identify any parameters associated with low serum albumin levels. The group with serum albumin <3.5 g/dL showed lower hemoglobin level, and higher serum β^2-microglobulin levels, however, it is not found to be clinically significant. The P values obtained are 0.087 and 0.982 respectively. Lower levels of serum

albumin are indicative of an ongoing systemic response that causes the loss of these proteins. Serum albumin level is not only a window into the patient's nutritional status but also a useful factor for predicting patient prognosis. The potential advantage of serum albumin level as a pretreatment prognostic factor in cancer patients is that it is inexpensive, reproducible and powerful. However, its efficacy should be evaluated by conducting further large prospective studies.

P-006

Intestinal Oxidative and Inflammatory Markers in Etoposide Induced Mucositis in Albino-Wistar Rats on Daily Basis

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Mucositis is one of the most debilitating outcomes of chemotherapy where the mucosal lining of the GI tract, causing painful ulceration. Etoposide is commonly used chemotherapeutic agent. During the course of mucositis, there are a number of oxidative and inflammatory changes taking place within the tissue due to the possible DNA damage causing the production of numerous cytokines as explained by Sonis. In this study albino-wister rats were administered intraperitoneally with a single dose of 65mg/kg body weight of etoposide to develop mucositis. 30 albino-wister rats were divided into 5 groups of 6 animals each. Group 1 was the normal control. Group 2, 3, 4 and 5 had the rats which were sacrificed after 24, 48, 72 and 96 hours after etoposide administration respectively. Intestinal tissue homogenate was used for the estimation of oxidative markers like Glutathione (GSH) and Total antioxidant activity (TAO), inflammatory markers namely Myeloperoxidase (MPO) and Nitric Oxide (NO) and Na^+- K^+ ATPase studied using manual methods. Data for the above-mentioned parameters was expressed as Mean ± SEM and median with 95% confidence interval. Parametric ANOVA and non- parametric Kruskal Wallis tests were used to find the differences between the means/medians; p value < 0.05 was considered significant. All the 5 parameters, TAO, GSH, NO, MPO and Na^+- K^+ ATPase revealed a significant difference among all the 5 groups from that of Normal control group. TAO & GSH decreased with time. NO, marker of inflammation and oxidative stress was found to be increased. MPO and Na^+- K^+ ATPase, though were found to be decrease soon after etoposide administration, later on started elevating suggestive of etoposide toxicity MPO in a long run. The results when extrapolated can implicate with the effectiveness of chemotherapy.

P-007

Correlation of Immunofixation with Protein Electrophoresis and Serum Free Light Chain Assays in a Tertiary Cancer Care Center

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Patients suspected of Multiple Myeloma (MM) are initially tested for paraproteins using serum protein electrophoresis (SPE), serum immunofixation electrophoresis (IFE), and Serum Free light chain (sFLC) measurements. Non-correlation between the tests result may, sometimes, occur which pose a diagnostic challenge. 142 cases of MM were identified from the hospital database over a period of 16 months. Serial data (1-2 weeks apart) from 106 patients were included in the study where serum tests result for SPE, IFE, and sFLC were available. 48 patients were new diagnoses and 58 were on follow-up. Treatment modalities included stem cell transplantation and standard chemotherapy regimens. 32% cases showed non-correlating results between the three techniques. 20% of the patients were the new cases and 41% in the follow-up group. It must be noted that 68% of cases were consistent in all the three tests. The discrepancies were: IFE showing less restriction bands as compared to SPE (8.6% in the follow up cases); an abnormal sFLC ratio with SPE or IFE negative (12.5% in new cases and 13.7% in follow-ups); and finally patients (8% in new cases, 22% in follow-ups) with normal sFLC ratio but with paraproteins in the other techniques. The combined use of SPE, IFE, sFLC measurements, and clinical assessment help in efficient diagnosis and monitoring of MM. However, the variable and discordant results may lead to difficulties in clinical decision making. Our study indicates that results of IFE may not always correlate with either of the SPE or sFLC results. This may be due to different sensitivities, antigen antibody interactions, or the effect of drug treatment. We found that SPE plus sFLC may make a more worthwhile contribution to the diagnosis, particularly the patients on follow-up.

P-008

Metachronous Onset of Breast and Bladder Cancer Associated with the BRCA2 c.8162delG Founder Mutation Detected in a Patient with a Low MammaPrint Recurrence Risk

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Restricting genetic screening programs to cancer patients fulfilling high-risk criteria may limit the identification of genes with pleiotropic function underpinning different cancer types. Metachronous onset of bladder cancer in an apparently sporadic breast cancer patient previously classified with a MammaPrint low-risk profile, prompted a mixed methods study design aimed at improvement of the selection criteria used for this assay. In the first phase of this study, we reviewed routine histological characteristics of tissue morphology and immunohistochemistry (IHC) reports to rule out metastasis of breast cancer. Use of a commercially-available pharmacogenomics assay (OncoDNA, Belgium) integrating IHC and next generation sequencing (NGS) of tumor DNA, led to a change of established clinical protocols by administering Doxorubicin (60 mg) and Etoposide (150 mg). Next generation sequencing (NGS) of the tumor DNA identified a BRCA2 mutation (c.8162delG), which was not suitable as a gene target for PARB inhibitors in this patient without metastasis. Prolonged survival of 12 months supports the benefit of precision medicine in this patient with a poor prognosis at referral. We recommend the inclusion of germline variants in the pre-screen algorithm previously developed for cost-effective implementation of the MammaPrint test in South Africa. BRCA2 provides a genetic link between breast and bladder cancer, which is of particular relevance to at-risk family members of the index case.

P-009

Reuptake of Membrane Bound Carbonic Anhydrase IX (CA-IX) is Higher in Invasive Glioblastoma Cell Line

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Glioblastoma multiforme is known to be highly malignant and most aggressive intracranial tumor. CA-IX controls the pH in solid tumors with the aim of maintaining alkaline intracellular pH and generating acidic extracellular tumor microenvironment. Recently CA-IX has been validated as a biomarker of poor prognosis in various cancers including astrocytoma. CA-IX has been also shown to be internalized more in hypoxic tumor cell lines. In this work, we have studied the reuptake profile of CA-IX in invasive and noninvasive cells under normoxia. U87MG cells were separated into migratory and non-migratory subpopulation using transwell migration assay and then were further grown on separate gelatin precoated coverslips. Live cells on coverslips were stained for membrane bound CA-IX for five minutes and then were incubated at 37°C for an hour to allow active internalization. After fixation, staining was done by tyramide super boost kit to enhance CA-IX fluorescent signal. Internalized CA-IX protein was quantified by confocal microscopy and image J software. We provide evidence that cell membrane bound CA-IX was internalized significantly more in the cytoplasm in cancer cells which have migratory potential as compared to those cancer cells which do not have such potential. There was no significant passive CA-IX internalization in cancer cells whose active intracellular membrane transport processes has been blocked by exposing them to metabolic inhibitor. Enhanced cell surface bound CA-IX internalization into cytoplasm of migratory glioma cells can be exploited for targeted killing of chemo resistant metastasizing glioblastoma tumors in patients.

P-010

Vitamin D and Total Calcium Level in Lymph Node Positive and Negative Patients of Carcinoma Breast

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Breast cancer is a malignant proliferation of epithelial lining of the ducts or lobules of the breast. It accounts for about one third cancer in females worldwide. Breast cancer is a hormone dependent disease which occurs as a result of hyperestrogenic state.

The highest incidence is above the age of 40 years. Receptors for Vitamin D (VDR) are present in a variety of cells including breast and the VDR is also known to be involved in cell proliferation and differentiation apart from its classical role in mineral metabolism. Due to the important role of Vitamin D in regulation of cellular growth and differentiation in normal and malignant cells, the present study was planned to study the level of Vitamin D and total calcium in carcinoma breast patients. The present study was conducted in the department of Biochemistry in collaboration with the department of Surgery. 30 newly diagnosed carcinoma breast patients confirmed by biopsy/histopathology were taken as cases and 30 age and sex matched healthy controls were included in the study. Clinical assessment of lymph node status was done in cases and later classified as clinical lymph node positive and negative. Vitamin D was estimated by RIA method and Total Calcium by Arsenazo method on autoanalyzer in the serum sample of all the cases and controls. 25(OH) D level was significantly low in cases as compared to controls (P=.0001). 20 cases were clinically lymph node positive but the vitamin D was comparable between lymph node positive and negative cases (P=0.934). Data of the study supports the possible protective role Vitamin D in breast cancer. However, no association was found between the lymph node status and serum level of Vitamin D. The mean value of serum calcium level in cases was low as compared to controls(P=0.066) though not significant statistically. However serum calcium level was high in lymph node positive cases as compared to lymph node negative cases.

P-011

Expression of CYP1B1 and Assay of Antioxidant Status and Phase II Detoxification Enzyme in Patients with Oral Squamous Cell Cancer.

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Oral squamous cell carcinoma spreads worldwide and a fairly onerous prognosis, encouraging further research on factors that might modify disease outcome. Cancer etiology is very complex and is related to the type of carcinogen, its dose, its frequency and its applications. Tobacco, whether smoked or chewed are carcinogens and it is implicated in 90% of OSCC. The aim of the study was to assay the expression of CYP1B1 gene in OSCC patients along with its relation with anti-oxidant status and phase-2 detoxification enzyme status. In the present study, CYP1B1 genotypic analysis was carried out along with total anti-oxidant power by FRAP assay and serum glutathione-S-Transferase (GST) activity in 20 OSCC patients and statistically compared with that of age matched 20 controls by using student's t-test. It was observed that 85% of histopathologically diagnosed OSCC patients had CYP1B1 expression with significantly decreased levels of serum total anti-oxidant activity ($p<0.05$) and serum GST levels ($p<0.05$) as compared to the healthy controls. On the basis of the present study we conclude that the expression of CYP1B1 is an important determinant of carcinogenesis and has significant association with decreased levels of serum GST and total anti-oxidant levels in OSCC patients.

P-012

Correlation of CA15-3 and CEA in Different Molecular Subtypes of Metastatic Breast Cancer

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Breast cancer is the most common cancer occurring in woman worldwide. Nearly 25% of all cancers is breast cancer with 1.67 million new cases diagnosed in 2012. In recent years, in patients of breast cancer, prognostic value of preoperative CEA and CA15-3 levels has gained much attention. Study has revealed that preoperative plasma level of CEA combined with CA15-3 may provide useful information for diagnosis and treatment of breast cancer. The aim of this study was to correlate CA15-3 and CEA level with clinicopathological parameters for early diagnosis and prognosis of metastatic breast cancer. This prospective study was done in department of biochemistry, IGIMS Patna. 75 diagnosed metastatic breast cancer patients of age group 30-70 years were included in this study. Correlation of CA15-3 and CEA levels with clinicopathological parameters were analyzed. In 75 patients of metastatic breast cancer, elevated CA 15-3 and CEA levels were seen in 44 (58.6%) and 26 (34.7%) patients, respectively. Level of CA15-3 and CEA were significantly associated with different molecular subtypes ($p=0.025$ and $p=0.043$, respectively). CA15-3 level was more elevated in Luminal A, Luminal B, and HER2 positive cases whereas CEA level was more elevated in HER2 positive cases. Elevation of CA 15-3 was significantly more common in multiple site metastasis of breast cancer compared with a single site metastasis ($p<0.0001$). However, elevated CEA levels was not significant between patients with a single and those with multiple site metastasis. Elevation of CA15-3 and CEA levels were found to be significantly associated with different molecular subtypes of metastatic breast cancer and CA15-3 level was also significantly associated with multiple site metastasis of malignant breast cancer.

P-013

Role of Microsatellite Instability in Colorectal Cancer and its Clinicopathological Correlation

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Microsatellite instability (MSI) and Immunohistochemistry (IHC) are generally used to identify patients having family history of cancer with a possible DNA mismatch repair defect (hereditary nonpolyposis colorectal carcinoma i.e HNPCC). A prospective study was conducted on histologically proven patients of CRC in a tertiary care hospital in North India between 2014 - 2018. Mismatch repair (MMR) protein loss was determined by using Immunohistochemistry for MLH1, MSH2, PMS2 and MSH6 and Microsatellite analysis for NCI Panel markers (BAT25, BAT26, D2S123, D5S346, and D17S250). SPSS version 16.0 was used for statistical analysis. 77 patients (49 males and 28 females) underwent resection for colorectal cancer with the median age of 52 years (16-81 years). 44% of the patients were younger than 50 years of the age. 6 patients had associated history of malignancy in the family. These 6 patients' tumor showed normal expression of the MMR proteins by IHC analysis whereas 3 patients showed Microsatellite instable High status and 3 patients with Microsatellite instable low status analyzed by PCR Fragment analysis. 42 (55%) patients had right colon cancer, 15 (19%) left colon cancer and 20 (26%) rectal cancer. Histology revealed well differentiated tumor in 26, moderately in 15 and poorly differentiated tumor in 36 patients. MMR protein loss was seen in 23 (30%) patients. Seven (46%) of these patients were less than 50 years of age. Combined loss of MSH2 and MSH6 was seen in 6 patients. 18 (78%) patients with MMR protein loss had tumor located proximal to the splenic flexure compared to 5 (22%) located distal to the splenic flexure. The current work revealed that there were less than 30% MMR protein loss in colorectal cancer patients in north Indian population. The loss was most commonly seen in right sided colon cancer than left. Young patients had lower incidence of right sided colon cancer and a higher incidence of rectal cancer than older ones.

P-014

Assessment of Serum HE4 and CA15-3 in Breast Tumors

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Despite advances in biology of cancer, there is lack of serum biomarkers which are specific and sensitive for detection and disease evolution of breast cancer. The most generally used serum tumor marker is cancer antigen 15-3 (CA15-3); however, its sensitivity and specificity are inadequate. Expression of Human epididymal protein 4 (HE4) has been demonstrated in ductal carcinoma of the breast tissue. The aim of the present study was to evaluate serum HE4 levels as diagnostic marker in breast cancer patients and to comparatively assess serum HE4 and CA15-3 in breast tumors. The cases selected were females with a breast lump. After confirmation with biopsy, cases were divided into 2 groups - malignant (n=30) and benign tumors (n=15). Age matched control samples were drawn from healthy volunteers (n=28). HE4 and CA15-3 were assayed by ECLIA method. A significant difference in the median (IQR) of HE4 (pmol/l) was identified among breast cancer, benign tumor patients and healthy volunteers {62.47(52.68-73.75) vs 52.3(43.6-62.0) vs 52.47(50.3-63.4) P=0.034}. The cutoff value for prediction of breast cancer was determined at >54.47 pmol/l for HE4, with a sensitivity of 73.33%, specificity of 58.14 %, accuracy of 67.4% whereas cutoff value of CA15-3 is >21.24 U/L with a sensitivity of 56.67%, specificity of 72.09% and accuracy of 63.8%. With a cutoff of 35 U/L (currently used reference range) the accuracy was found to be 50% only. A significant elevation of serum HE4 levels in patients with breast cancer compared with that in benign breast tumors and healthy controls was identified. HE4 may serve as an additional biomarker for the diagnosis of breast cancer. Changing the normal reference range of CA15-3 from the current 35 U/L to 21.24 U/L will improve its diagnostic accuracy from 50% to 63.8%.

P-015

Nutritional Status in Patients with Carcinoma Stomach

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Gastric cancers are the fifth most common cancers worldwide based on GLOBOCAN 2018 estimates. Evidence is accumulating regarding the role of vitamin B12 and micronutrients in folate metabolism in cancer risk. Hence we evaluated the nutritional status in patients with gastric carcinoma. A case-control study done on 30 cases (M=19, F=11) diagnosed with carcinoma stomach. 30 age and gender matched controls without any comorbidities were included. Patients with other malignancies were excluded. Serum Vitamin B12, Folate, Plasma Homocysteine (chemiluminescence immunoassay), Serum Calcium(O-Cresolpthalein), Phosphorous (ammonium molybdate) and Iron (Ferrozine) were estimated. Statistics were done using SPSS. Median(IQR) of Serum Folate levels in cases vs controls [5.52(4-7.43) vs 5.78(3.88-13.65) p = 0.02]ng/ml, Plasma Homocysteine [10.82(8.8-14.48) vs 22.65(9.4-33.7) p = 0.014]µmol/L, Serum Calcium[8.2(7.77-8.72) vs 8.9(7.97-9.52) p = 0.002]mg/dl, Phosphorous[3.5(3-3.9) vs 4.55(3.55-6.57) p = 0.028]mg/dl, Iron[34.5(28.25-63.25) vs 70.5(55.75-105.25); p = 0.001]µg/dl, TIBC[282(195.2-339.5) vs 332.5(304-361.5) p = 0.00074]µg/dl, Magnesium[0.77(0.68-0.85) vs 0.83(0.75-0.90) p = 0.023]mmol/L were significantly low in cases compared to controls. Vitamin B12 levels were raised in cases compared to controls but not statistically significant [472(317-789) vs 314(231-534) p = 0.16] pg/ml. Folate depletion can result in global DNA hypomethylation, DNA damage, impaired DNA repair and altered proto-oncogene expressions. Pernicious anemia represents another potential link between vitamin B12 malabsorption and increased risk of gastric cancer, but elevated plasma B12 levels have been observed in our cases, similar to few other studies. The underlying mechanisms are poorly elucidated, though the increased release of haptocorrin (HC; vitamin B12-binding protein) or a direct increase in plasma vitamin B12 by liberation from an internal reservoir into the circulation might be a plausible explanation. Our study found the deficiencies of micronutrients in the cases. A further prospective cohort study is required to know whether they are a cause or result of Gastric cancers.

P-016

Does Chromogranin A an Indicative Biomarker of Gastric Cancer?

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Chromogranin A (CgA), a hydrophilic glycoprotein present in large dense core vesicles of neuroendocrine cells, and occurs throughout the length of entire gut on about two-third of them are gastroenteropancreatic NET. Due to its primarily expression of Chromogranin A is widely accepted potential biomarker of neuroendocrine tumors (NET) which has been recently implicated in benign and malignant status. Therefore, the present study has planned to know does Chromogranin A play potential role as a biomarker in a wide spectrum of benign and malignant disease. Samples were collected from the department of Gastroenterology and B.R.A IRCH, AIIMS, New Delhi. Clinical, biochemical, radiological or imaging investigations were done in all subjects. The subjects were categorized into 4 groups (i) Ca stomach (n=12) (ii) Ca Pancreas (n=16) (iii) Liver cirrhosis or HCC (n=24) (iv) Healthy control (n=40). Group (i), Group (ii) and group (iii) patients were biopsy proven cancer. Clinical, biochemical, hematological investigations were done for healthy control (HC). Chromogranin A ELISA kit was procured from Demeditec Diagnostics GmbH (Germany) and all samples were processed in duplicates and quantification of chromogranin A was done as per manufacturer's instructions. Statistical analysis (mean ± SD) was done for comparative analysis of samples. It was observed that chromogranin A level was significantly higher in Ca Pancreas as compared with liver cirrhosis and also in Ca stomach from healthy subjects ($p<0.001$). Evaluation of Chromogranin A could be more reliable particularly in response to therapy or in disease progression for an early diagnosis or recurrence. Furthermore, Chromogranin A can be a strong predictor of neuroendocrine differentiation as well as in adenocarcinoma.

P-017

To Evaluate Effect of Imatinib Therapy on Pituitary Hormones (Serum LH, FSH and Prolactin) in Newly Diagnosed CML Patients

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Chronic myelogenous leukemia is characterized by the fusion gene BCR-ABL, encoding a constitutively active tyrosine kinase and treated by tyrosine kinase inhibitor imatinib. Some case reports in literature suggest imatinib therapy may alter the pituitary hormones LH, FSH and Prolactin levels affecting various functions of reproductive system. The study was designed to prospectively study serum LH, FSH and Prolactin levels at baseline and at 6 months of imatinib treatment in 30 newly diagnosed BCR-ABL positive CML patients. The hormones were measured by highly accurate two-site sandwich immunoassay using chemiluminescence method on ADVIA Centaur system from Siemens. Of the 30 CML patients (17M & 13F), 2 patients presented in accelerated phase and 26 achieved haematological remission by 6 months. Baseline levels of LH, FSH and prolactin were similar in patients and controls. S. LH levels increased significantly after 6 months of imatinib therapy in male patients (13.82±4.25 mIU/mL vs. 7.32±3.84 mIU/mL, p=0.001) and in female patients (12.67±4.74 mIU/mL vs.7.16±3.94 mIU/mL, p=0.002). S. FSH levels increased significantly after 6 months of imatinib therapy in male patients (22.30±8.74 mIU/mL vs. 11.77±7.23 mIU/mL, p=0.003) and also in female patients (12.57±4.94 mIU/mL vs. 5.51±1.82 mIU/mL, p=0.005). Serum prolactin levels decreased significantly after 6 months of imatinib therapy in male patients (10.93±7.77 ng/mL vs. 14.21±12.68 ng/mL, p=0.039). Baseline levels of Serum LH, FSH and Prolactin were similar in patients and controls. The findings of significantly increased LH and FSH levels after imatinib therapy, present study is consistent with the previous reports that document the effect of imatinib on testicular function in adult male patients. The findings of significant increase in LH and FSH levels in pre-menopausal females in present study suggests that further studies on lager number of female patients are required to fully assess the effects of imatinib on multiple female reproductive hormones and their clinical significance.

P-018

A Pilot Study Exploring role of Growth Differentiation Factor 15 as a Cause of Drug Resistance in Breast Cancer

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Breast cancer (BC) is a complex and heterogeneous disease and it is one of the leading causes of cancer-related deaths among women. Triple-negative breast cancer (TNBC) represents a most lethal and aggressive subgroup of breast tumors and it is often associated with a worse prognosis. Elevated plasma levels of GDF-15 have been seen in patients with malignancy associated with poor prognosis. There are no studies which explore the correlation between GDF-15 and drug resistance through ABCC5 in BC. The aim of current study is to analyze the role of GDF-15 in drug resistance in BC. In the current study we have included 19 breast tumor tissues, out of which 15 were non-TNBC and 4 were TNBC, and 8 surrounding control breast tissue, from patients undergoing surgery at a tertiary care centre of Western Rajasthan. RNA was isolated from the tissue and expression level of GDF-15 and ABCC5 were analyzed using RT-PCR. The data were analyzed using SPSS22 software and $p<0.05$ was considered to be statistically significant. We demonstrated that, fold change expression of GDF-15 in TNBC (5.6 folds) and non-TNBC (4.5 folds) patient tissue was found to be higher as compared to surrounding control tissue. Similarly, fold change expression of ABCC5 is higher in TNBC (4.7 folds) and non-TNBC tissue (4.9 folds) as compare to surrounding control tissue. Further, there is a significant positive correlation between GDF-15 and ABCC5 ($r=0.761$, $p=0.000$) in BC tissue samples. Thus, current study suggests that expression of GDF-15 and ABCC-5 is higher in BC and there is a significant positive correlation between GDF-15 and drug resistance in BC, suggesting GDF-15 to be a possible cause of drug resistance and a potential therapeutic target.

P-019

Role of Free / Total PSA ratio to Differentiate BPH and Prostate Cancer

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Prostate cancer is the most common cause of death from cancer in men over 75 yrs. Screening is focused only on total prostate-specific antigen (PSA) levels which lead to many false-positive results, with unnecessary biopsies and emotional distress. There is considerable overlap in PSA concentration in men with benign prostate disease. This study was undertaken to determine Free to Total PSA ratio to differentiate between Benign Prostate Hyperplasia (BPH) and Prostate Cancer (PCa) and to improve the accuracy of the PSA test and the specificity of PCa detection particularly when PSA levels fall between 4-10 ng/ml levels. The study group consisted of a total of 124 subjects. Free PSA and Total PSA were determined using assay, which is based on Chemiluminescent Microparticle Immunoassay (CMIA) technology. The Statistical test used were Students' t test for difference of Mean and Pearson Correlation test, using SPSS and MS Excel Statistical packages. Free/Total PSA ratios in the prostate cancer group were significantly lower than those in the BPH group. A cut-off of F/T PSA ratio% at 18% improves diagnostic sensitivity and specificity for prostate cancer.

P-020

Diagnostic and Prognostic Value of the Cancer-Testis antigen LDH-C4 in Breast Cancer

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Lactate dehydrogenase C4 (LDH-C4) is a cancer/testis antigen (CTA) that is expressed abnormally in certain malignant tumors. However, the expression and clinical significance of LDH-C4 in breast cancer (BC) have not been determined. Quantitative reverse transcription-polymerase chain reaction (RT-PCR) was adopted to investigate the expression of LDHC mRNA in the serum or serum-derived exosomes of patients with BC. The expression of LDH-C4 protein in BC tissues was also evaluated by using high-throughput tissue microarray analysis and immunohistochemistry. The results showed that LDHC mRNA was highly expressed in the serum and serum-derived exosomes of BC patients. The area under the curve (AUC) of serum and exosome LDHC in distinguishing BC from healthy individuals was estimated to be 0.9587 and 0.9464, respectively. The LDHC level in the serum-derived exosomes of patients with BC was significantly associated with tumor size and positively correlated with the expression of HER2 and Ki-67 (all with $P<0.05$). Serum and exosome LDHC levels in BC patients were negatively correlated with medical treatment and positively correlated with recurrence. Survival analysis showed that LDH-C4 expression was negatively associated with BC prognosis. Serum and serum-derived exosome LDHC may be an effective indicator for diagnosis and therapeutic efficacy, as well as a predictor for the recurrence of BC. LDH-C4 may be a biomarker to determine the prognosis of BC.

P-021

Effect of Treatment on Renal Functions in Lung Cancer Patients

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Lung carcinoma is the major cause of premature and avoidable mortality worldwide. Renal Function Tests are found to be deranged in many of the cancers. Metastasis rate is very high in lung carcinoma. Kidneys are very common sites for the spread. So, the study was planned to find out whether Kidney Function Tests are compromised in these patients or not. The aim was to find out renal status in patients of lung cancer before and after treatment and to compare the results with age and sex matched healthy controls. Twenty-five cases of lung carcinoma irrespective of age, sex and stage were selected. Diagnosis was established with help of detailed history, clinical examination, radiological and histopathological examination. The patients of lung cancers were compared with the healthy controls (group Ia), before treatment (group IIa), after 4 weeks of chemo radiation (group IIb). Renal function tests were analyzed in patients before and one month after treatment. The serum urea levels were statistically significant between group I and IIa with $p=0.01$. The serum uric acid levels were statistically significant between groups I and IIb ($p=0.005$). All the other parameters were statistically insignificant. Though the results were not statistically significant in patients before and after treatment, but we may conclude that Renal Function Tests are compromised in patients of lung cancer.

P-022

Correlation of Serum Lactate Dehydrogenase and Alkaline Phosphatase in Different Histological Grades of Head and Neck Squamous Cell Carcinoma and Premalignant Lesions in a Tertiary Care Hospital.

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Cancer is the second most common disease in India responsible for about 0.3 million deaths per year. Squamous cell carcinoma of the head and neck (HNSCC) is considered as fourth most common malignant disease in the world. Over 200,000 new cases of head and neck cancers are registered every year in India. Increased serum Lactate Dehydrogenase (LDH) activity is considered as a marker of cellular necrosis while serum Alkaline Phosphatase (ALP) is recognized as an important marker of induction of tumor cell differentiation. Considering the importance and need of biomarker in HNCA and there is very less literature available on tumor markers from this highly prevalent region (Western Rajasthan), It was intended to examine the role of serum LDH and ALP in head and neck squamous cell carcinoma (HNSCC) and epithelial precursor lesions (EPL). The aims of the study were to evaluate serum LDH and ALP level in head and neck squamous cell carcinoma (HNSCC) and epithelial precursor lesions (EPL), and assess the relationship between serum LDH and ALP with the histological grades of HNSCC and EPL. This is a prospective observational study on 25 patients of either sex with HNSCC and precancerous lesions attending OPD, Department of Radiation oncology, MDM Hospital, Jodhpur during Jan-19 to June-19. Serum LDH and ALP was measured using fully automated analyzer. Results were compared with healthy controls. Results were subjected to suitable statistical analysis using student 't' test and p-value. Serum LDH level was significantly increased in while serum Alkaline phosphatase was insignificant when results were compared with healthy controls suggesting possibly LDH as a marker of HNSCC. Estimation of serum LDH is simple, reliable, economic and sensitive and it can be used as an early marker for screening/ diagnosis of malignant and premalignant conditions of the head and neck cancers to prevent serious complications and sequel of disease.

P-023

Significance of VMA, NSE, LDH, Ferritin and MIBG Scan in Neuroblastoma

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The study was designed to investigate urinary Vanillyl Mandelic Acid (VMA), serum Neuron-specific enolase (NSE), Ferritin, lactate dehydrogenase (LDH) and MIBG scan in Neuroblastoma (NB) and to evaluate the utility of these markers in Indian population. Blood samples and 24 hour urine samples were collected from 40 healthy subjects and 91 untreated patients with histologically proven Stage III & IV NB cases referred to our institute. Method used for NSE was Enzyme Immunoassay (Elisa), MIA for serum Ferritin, LDH by photometry and VMA by Column Chromatography. Amongst the parameters studied, VMA showed highest sensitivity (91%), specificity (94.4%), positive predictive value (97.8%) and negative predictive value (85%) [cut off levels of 7 mg/ml of creatinine was taken] as compared to other studied parameters. This study suggested that the detection of VMA in combination with routine histological examination, MIBG scan, serum NSE and LDH may improve the diagnosis of Neuroblastoma.

P-024

Role of Serum FLC Measurement, Ratio of Total to Free Light Chains and BJP in Differential Diagnosis of Multiple Myeloma.

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Diagnosis and monitoring of MM and related conditions are usually carried out by means of serum and urine protein electrophoresis and immunofixation. Several reports have demonstrated the usefulness of the free light chains assay for the diagnosis and monitoring of MM. In patients with no measurable M protein in serum or urine using SPE, serum FLC assays are helpful. Aim of this study was to evaluate the role of serum FLC measurement in differential diagnosis of MM and clinical significance of total light chain & free light chain ratio and Bence Jones Proteins in the diagnosis of multiple myeloma. The material for the study comprised of 120 newly diagnosed (with serum protein electrophoresis) multiple myeloma cases in our institute. Analysis of serum Free light chain Kappa and Lambda and B2M was done by turbidimetry on Beckman coulter AU 2700 autoanalyzer. Total Kappa light chain, Lambda light chains and urinary Bence Jones Protein were estimated by nephelometry on Beckman coulter Image

800 Immunoassay analyzer. Ratio of FLC kappa to Lambda showed the highest sensitivity (85%), specificity (95%), Positive predictive value (95%) and Negative predictive value (84%). When we compared ratio of FLCs to β_2 microglobulin and creatinine values, the result were significant at $p < 0.05$. Even 4% of cases did not show any M spike but ratio of sFLC Kappa/Lambda was abnormal. Hence, we conclude that serum FLC assay and ratio can be considered reliable for the diagnosis, monitoring, and prognosis of different plasma cell disorders.

P-025

Bio-evaluation of ^{68}Ga-DOTATATE Formulation for invivo Diagnosis of Neuroendocrine Tumors: Board of Radiation and Isotope Technology, India Perspective

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^{68}Ga-DOTATATE is a radiolabeled somatostatin analogue for in vivo imaging of neuroendocrine tumors using Positron Emission Tomography (PET). The objective of the current work was to perform the pre-clinical evaluation of this product produced in-house for clinical usage. Two different production batches of ^{68}Ga-DOTATATE formulations prepared in- house were evaluated for their physicochemical and pharmacokinetic parameters employing Wistar rat model. Each batch formulation underwent a physiological distribution study by scintigraphic assessment at 30min, 1, and 2 h post- injection of the tracer. Briefly, four Wistar rats per time point were intravenously administered with about 0.1 mL of ^{68}Ga-DOTATOC (~740 kBq). The animals were sacrificed at respective time points after which the relevant organs were excised for determination of the associated radioactivity using a flat-bed NaI (Tl) scintillation counter with optimal energy window for ^{68}Ga. Consistently high radiochemical yields (>95%) were obtained on radiolabeling with ^{68}Ga. The radiolabeled peptides exhibited excellent in vitro stability. ^{68}Ga-DOTATATE uptake was noted in the SSTR2-expressing organs such as salivary glands, pancreas adrenals, stomach, kidneys, liver with excreted activity in the bowel kidneys and urinary bladder. Of the major organs, the highest uptake at 30min, 1, and 2 h after injection was observed in pancreas, followed by intestines. The most important feature is the absence of bone uptake, suggesting the absence of free ^{68}Ga- in the synthesized ^{68}Ga-DOTATATE formulation. Uptake in the kidneys is for the most part due to re-absorption of the radiolabeled peptide in the renal tubular cells after glomerular filtration. The objective of formulating a "Ready-to-Use" preparation of ^{68}Ga-DOTATATE, a long felt need of the nuclear medicine community of INDIA commensurate with clinical requirements, has been achieved. These formulations provide a high degree of convenient and reliable method for facile preparation of ^{68}Ga-DOTATATE.

P-026

Role of CA-125 and HE4 in Diagnosing Patients with Ovarian Malignancy

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Ovarian cancer is 3rd most common type of gynaecological malignancy and is leading cause of death in women. It is often detected at an advanced stage and results in poor prognosis. Its accurate diagnosis at an early stage is required. CA-125 is commonly used as tumor marker for diagnosis of ovarian cancer. Now a days, HE4 (Human Epididymus protein 4) has emerged as an important biomarker for diagnosing of ovarian cancer. It is also known as WAP-type four disulphide core 2 (WFDC2). It primarily express in reproductive and respiratory tract but overexpress in ovarian cancer. It has been reported that HE4 is more specific than CA-125 in distinguishing benign from malignant gynaecological conditions. It can benefit us as a screening tool for detection of early stage of ovarian cancer. In this study, we evaluated diagnostic performance of both HE4 and CA-125 in discriminating ovarian cancer from other benign gynaecological conditions and controls. It was a hospital-based study. 40 female patients of age 20-80 were selected with recently diagnosed ovarian cancer and 20 females with benign gynaecological diseases were also included. 20 healthy age matched females were taken as controls. The samples were taken from Department of Obstetrics and Gynecology at Guru Gobind Singh Medical College and Hospital, Faridkot. Patients with chronic heart failure, chronic liver or renal disorders and pregnancy were excluded. The samples were tested for CA-125 using Analyzer access-2, HE4 will be tested by an ELISA kit method according to manufacturer's instructions. There was significant difference in CA-125 and HE4 values between ovarian cancer group than benign gynaecological conditions ($p=0.001$) and control group p (<0.001). A positive correlation was seen between levels of serum HE4 and CA-125 in females with EOC, benign gynaecological diseases group and healthy control group.

P-027

Phytochemical Analysis and Dose Dependent Effect of Enicostemma Axillare on Breast Cancer Cell Line

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Breast cancer is the second most common cancer in female population worldwide. For which various treatment like chemotherapy and radiotherapy are available. These lines of treatment do not decrease mortality but to a certain extent reduce morbidity. Medicinal plants have been used in traditional treatments for various human diseases since ancient times. Enicostemma axillare (E.axillare) found in India, exhibit anticancer effects against various cancers. It has been long since used in folk medicine to treat spectrum of disease like diabetes mellitus, rheumatism, abdominal ulcer, hernia, swelling, itching and insect poisoning. In these recent years, using advanced technologies, the therapeutic benefits of medicinal plants are generally traced to specific compounds; specifically the active constituents of the plants. Thus, this study was focused on the phytochemical analysis and standardization of E.axillare on MCF-7 cell line. The whole plant were collected from Chengalpet, Kanchipuram District, Tamil Nadu, India. The whole plant materials were shade dried and powdered. The plant extract was prepared by soxhlet apparatus using ethanol. With that extract phytochemical screening was done for alkaloids, flavonoids, steroids, carbohydrates, glycosides, amino acid, tannins, terpenoids and saponins. Standardization of E.axillare was done by MTT on MCF-7 cell line. Statistical analyses were conducted by one-way ANOVA in (SPSS) software version 16.0. The phytochemical analysis of E.axillare extract shows the presence of alkaloids, flavonoids, carbohydrates, glycosides, amino acid, tannins, terpenoids and saponins. MTT results that the IC50 value for E.axillare is 12.5 µg/ml. The present study revealed that ethanol extract of E.axillare was rich in phytochemical constituents. Standardization of the drug may helpful to develop molecular studies, which might be of great contribution for the society. Thus, E.Axillare could be a potential natural therapeutic agent in future.

P-028

To Investigate the Probability of Serum Amylase, Total Calcium and Phosphorus as Prognostic Marker in Patients of Head and Neck Cancer

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Head and neck cancer is the sixth among the cancer burden of the world and so an important health problem in the community. In India, it is the most common cancer in men and also in the both sexes. For early stages, surgery or radiotherapy may be the choice but for locally advanced cases, chemotherapy is administered as concomitant or adjuvant to radiotherapy depending on the aim of the therapy. The levels of serum amylase, serum total calcium and phosphorus as the markers of response to treatment in the head and neck cancer were analyzed. The levels of serum amylase, serum total calcium and serum phosphate in patients of head and neck cancer before and after treatment and also in controls (group-A, B & C respectively) were analyzed by standard methods on Randox autoanalyzer and the values were compared and analyzed by appropriate statistical methods. The amylase levels between group A and C were significant (p=0.013). The levels of serum total calcium were statistically significant among all three groups (p value between group A and B = 0.0001, between A & C = 0.006 and between B and C $\leq$ 0.0001).The phosphorus levels were also statistically significant among all the three groups (p value between group A and B = 0.003, between A and C = 0.060 and between B and C $\leq$ 0.0001). From the above finding we can come to a conclusion that serum amylase, total calcium and phosphorus are potential prognostic markers in the patients of head and neck cancer indicating the response of the patients to the treatment.

P-029

Basophil Lymphocyte Ratio are Different Among Menstrual Status and Subtype in Indonesian Breast Cancer Patient

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Breast cancer is a complex of diseases with various clinical outcomes despite the advancement of the treatments. Recent evidences showed that inflammation has crucial role both in cancer progression and also as biomarker to determine treatment outcome, particularly the basophil to lymphocyte ratio (BLR). However, studies regarding the role of BLR in breast cancer yield various result and considered as inconsistent. Therefore, we evaluated the BLR status in breast cancer patients in relation to menstrual status and subtype. A total of 105 breast cancer patients were evaluated retrospectively from medical records between 2013-2015 period. Patients were classified into two subtypes: luminal (Luminal A and Luminal B) and non-luminal (Her2 and Triple negative [TNBC]); menstrual status were classified as pre-menopause and post-menopause. The basophil and lymphocyte count were obtained from medical records and the ratio was subsequently calculated. The BLR cut-off point was statistically calculated and found to be 0.039 (AUC: 0.507). Luminal type consists of 66.7% of the subjects and 43.8% samples were pre-menopause. BLR was found to be significantly higher in patients with pre-menopause (mean: 1.61±0.493 vs 1.41±0.495; p=0.041), higher nodal status (N2-N3) (mean: 1.72±0.461 vs 1.46±0.502; p=0.048) and non-luminal (mean: 1.66±0.482 vs 1.41±0.496; p=0.020). No significant association was found with age, tumor, metastasis, stadium, and receptors expression (ER, PR, and Her2). From this preliminary study, we can conclude that BLR was significantly associated with pre-menopause, higher nodal status and tendency toward non-luminal type. However, larger and more comprehensive studies are needed to validate this results and to find the clinical relevance of BLR for breast cancer patients.

P-030

Clinical Utility of Polygenic Risk Score in Determining Cancer Risk

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Non-communicable diseases (NCD) have been a top contributor of mortality in Indonesia. Nevertheless, preventive measures can be taken to reduce the morbidity and mortality of NCD. Combination of factors such as genetics, lifestyle, food, and environmental conditions might reduce or increase risk of diseases. In clinical practices, various prediction models can be used to assess those risk factors and estimate an absolute disease risk for the individual. Accurate prediction of an individual's risk of certain disease allows the individual to select the best preventive strategy. This study aimed to evaluate the utility of polygenic risk score (PRS) in determining risk for cancer. Genotype data from 263 subjects was obtained by using Infinium Asian Screening Array-24 v1.0 (Illumina, USA). Among these, 103 subjects have family history of cancer. PRS is calculated for all of the subjects based on 135 single nucleotide polymorphisms (SNPs) associated with six cancer types (breast, cervical, colorectal, lung, pancreatic, and non-Hodgkin lymphoma). Subjects with PRS above the 99.7% is classify as high risk. Six out of seven subjects which are classified as high risk for certain cancer have family history of cancer. Two subjects which classified as potential or high risk of colorectal cancer have a family member with nasopharyngeal cancer. The result of this study indicates that despite constituted by low-penetrance variants, PRS can differentiate people at risk of familial cancer. Variants used in determining the risk for colorectal cancer might be overlap with risk of nasopharyngeal cancer. However further study with larger number of subjects will be needed.

P-031

Correlation of Parity and BMI on Level of Serum CA125 in Ovarian Tumor.

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Ovarian cancer is the 3rd most common gynaecological cancer in India. Incidence of ovarian cancer is increasing day by day. Age standardized incidence rate (ASR) of 2001-2006 increases 0.9 to 8.4/100,000 population to 6.1/100,000 population (2011). This study aim is to show impact of BMI and parity on level of serum CA125 in ovarian tumor patient. The study was carried in the Biochemistry Department of Darbhanga medical college, Bihar. For this study, 100 women were taken, of which 50 women were

control, 25 women having benign and 25 women having malignant ovarian tumor. This study shows positive impact of BMI on serum CA125 level, as BMI increases level of serum CA125 also increases. In benign study group with BMI >30, CA125 levels were found to be 259.41±82.56 and 1099.61±273.79 in malignant study group with BMI >30. In parity, as parity decreases level of serum CA125 decreases. Nulliparous women in benign study group had levels 282.21±94.89 and in malignant ovarian tumor study group it was 1025.43±243.23 which is significantly high. The study shows positive and direct relation with parity and BMI on serum CA125 level in ovarian tumor.

P-032

Time Dependent Effects of Polyamide NV1078 in decreasing Human Papilloma Virus DNA in Cells

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Human Papilloma Virus (HPV) infection is a common sexually transmitted disease. Persistent infection by HPV is associated with development of cervical, oropharyngeal and anogenital cancers. HPV-16 is most commonly linked with cancer, out of more than 30 known types of HPV. Vaccines are available for patients who are 26 years or younger, but protects only 2-7 of 15 high risk HPV types. Owing to the unavailability of anti-viral agents, drugs that selectively target HPV but do not damage the host cell are needed. Polyamides are asymmetric hair-pin polyamides with tetramethyl guanidine capped N terminal end. We conducted antiviral assays with the polyamide NV1078 to determine the concentration that reduces HPV 16 episomes by 50% (pseudo-IC50) and 90% (pseudo-IC90) as a function of time. W12E cells harboring HPV-16 episomes are cultured on Mitomycin-C treated 3T3-J2 fibroblast cells. Polyamide concentrations between 0.001μM - 10μM with a final DMSO concentration of 0.1% in E-complete media were added to the culturing cells and incubated at specific time frames. Cells were harvested and total DNA (genomic DNA and viral DNA) was collected. Concentration of DNA was quantified with a nanodrop and normalized to a specific DNA concentration. Episomal copy number was determined using a standard curve of known HPV episomal copy numbers and samples were amplified and quantified by SYBR Green qPCR within the HPV-L1 gene. A melt curve was used to ensure only the product was amplified. Episomal decrease was determined by the relative percentage of control DNA from untreated cells. IC50 and IC90 were calculated in KaleidaGraph and other software. Cell Viability assay was also conducted by using Reliablue cell viability reagent and measured the fluorescence. NV1078 was found to decrease episome levels in a dose-dependent manner. Cell viability was not affected even at 10 μM concentration of NV1078.

P-033

Alpha Fetoprotein, AFP-L3 and PIVKA-II in Patients with Hepatic Cirrhosis and Hepatocellular Carcinoma

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Hepatocellular carcinoma (HCC) is a common complication in patients with Hepatic Cirrhosis. Detection of HCC at an early stage is critical for a favourable clinical outcome. The study aim was to: (i) compare the levels of α-fetoprotein (AFP), AFP-L3 and Protein Induced by Vitamin K Absence II in HCC patients and in hepatitis cirrhosis patients without HCC; (ii) to define the level of each tumor marker with the best sensitivity and specificity for HCC diagnosis; and (iii) to correlate the levels of these markers with respect to disease severity. The study group consisted of 60 patients - 30 HCC, 30 HC; and 30 healthy subjects. All evaluated for AFP-L3, PIVKA-II and Total AFP (T.AFP). Levels of DCP, AFP and AFP L-3 were significantly higher in patients with HCC than in those without HCC ($P < 0.0001$). Receiver-operating curves (ROC) indicated that the cut-off value with the best sensitivity and specificity was 6.715 ng/ml for PIVKA-II, 12.2 ng/mL for T.AFP and 1.05 ng/ml for AFP-L3. The sensitivity and specificity for AFP-L3 was 90% and 86%; PIVKA-II 85.71% and 95%; and for T.AFP 70% and 94% respectively. The AUROC was 0.910, 0.870 and 0.820 for AFP-L3, PIVKA-II and T.AFP respectively. When all 3 markers combined AUROC increased to 0.943. Also, there was strong positive correlation of AFP-L3 and PIVKA-II levels with tumour size (P = 0.0001, P = 0.043), while no such significant association was found with T.AFP. AFP-L3 had the highest sensitivity and AUC for HCC diagnosis, had a direct correlation with tumor size. Hence AFP-L3 should be used as the main serum test for HCC detection.

P-034

Pretreatment Platelet-to-Lymphocyte Ratio (PLR) as a Predictive Value of Hematological Markers in Breast Cancer

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Clinicopathological of breast cancer have different treatment responses and clinical outcomes. Several studies had evaluated the role of peripheral blood test as a diagnostic and prognostic tool in cancer assessment. The platelet lymphocyte ratio (PLR) is an emerging indicator for inflammation. The aim of the study was to investigate whether the pretreatment platelet-to-lymphocyte ratio (PLR) could be used as in predicting the stage of breast cancer. A retrospective study recruiting 105 patients with breast cancer during a five-year period, 2013-2017, at the Sanglah General Hospital. The histopathological records and complete blood counts (CBC) of the patients were collected and analyzed using SPSS ver. 25 software for Windows. Stage I-II and III-IV were classified as early and advance stage respectively. Risk analysis model, receiver operator characteristics (ROC) analysis, and correlation test were used to evaluate the association of PLR with stage. A positive correlation was found between the staging of breast cancer with PLR ($r=0.277$, $p<0.05$). PLR has a statistically significant area under the curve (AUC) (0.638; 95% CI 0.523-0.753; P=0.021). The cut-off, sensitivity, and specificity value of PLR was 137.8 (71%; 58%). Advanced stage of breast cancer was found in high PLR (adjusted OR: 0.650; 95% CI=0.464-0.911; p=0.004) and early stage in low PLR (adjusted OR: 3.33; 95% CI = 1.429-7.774; p=0.006). From this preliminary study, pretreatment PLR values might be a useful information in predicting the staging of breast cancer.

P-035

Novel PALB2 Deleterious Mutations in Breast Cancer Patients from South Indian Population

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Breast cancer is the most common cancer in women across the world. Partner and localizer of BRCA2 (PALB2) were recently identified as a breast cancer predisposition gene. A mutation in this gene in familial breast cancer is about 1%-2% from different population reported. However, there are no extensive reports from the Indian population. The ultimate goal of this study is to understand the status of PALB2 mutations among the breast cancer subjects from the Indian population. We have evaluated the PALB2 gene mutation in 200 breast cancer patients and 200 controls that tested negative for BRCA1/2. Single Stranded Conformation Polymorphism (SSCP) assay was performed for screening the variants on amplified regions followed by direct sequencing for conforming mutations. Among the 200 breast cancer subjects analyzed, 128 patients were of inherited familial history and the rest 72 patients were of sporadic cases. Two novel deleterious mutations and one novel missense mutation were identified from this study, with the prevalence of about 3.5% (7/200). Among the deleterious mutations in familial breast cancer, PALB2 c.780delG was identified with the prevalence of 1.5% (3/200) and c.725delT was identified with the prevalence of 1% (2/200) in the fourth exonic region. One missense mutation of PALB2 c.404C>T was found in familial and one in sporadic breast cancer. In this study, the prevalence of PALB2 gene mutation in familial breast cancer is about 4.6% (6/128). Our study demonstrates that the PALB2 gene mutations prevail among familial breast cancer patients who had attended the tertiary care center at Madurai, India. Clinically, these data may be helpful in the genetic counseling for breast cancer patients with PALB2 germline mutation.

P-036

Significance of Serum Levels of LDH, ALP and GGT in Breast Cancer

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Breast cancer is the second most common cancer in women next only to cervical cancer. Multiple factors are associated with an increased risk of developing breast cancer. Unfortunately, most

cancers do not produce any symptoms until the tumors are either too large to be removed surgically or metastasis has taken place. Therefore, there is need for simple biochemical investigations, which can be easily assayed, less expensive and can be used for the early detection of carcinoma of breast and prognosis. To analyze serum Lactate Dehydrogenase (LDH), Gamma Glutamyl Transpeptidase (GGT) and alkaline phosphatase (ALP) levels in diagnosis of carcinoma breast patients and compared with controls. And evaluate the relationship of serum LDH, GGT and ALP levels in carcinoma breast patients of pre and post-menopausal age group of women. The study was conducted in king George hospital, Visakhapatnam. Serum biochemical markers were estimated in 60 (30-premenopausal women, 30-post menopausal women) clinically and histopathological confirmed patients with carcinoma breast and 40 number of healthy age- matched individuals served as control. Lactate dehydrogenase (LDH), Serum Gamma Glutamyl Transpeptidase (GGT) and alkaline phosphatase (ALP) levels were analyzed and estimated. The mean serum LDH, GGT, and ALP activities in patients with carcinoma breast were significantly ($p<0.0001$) increased as compared to controls, and a steady increase was observed in their levels from premenopausal to post-menopausal women. In this study, though less sensitive than imaging procedures, it is to suggest that the estimation of LDH, GGT, and ALP can be useful in smaller laboratories as routine screening tests in all suspected carcinoma breast patients and for general population and can also be used to detect metastasis.

P-037

Diagnostic Performance of ROMA in Epithelial Ovarian Cancer: a Systematic Review

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Epithelial ovarian cancer has become the most frequent cause of deaths from among gynecologic malignancies. Our study elucidates the diagnostic performance of ROMA, HE4 and CA-125. Objective of the study was to compare the diagnostic accuracy of ROMA, HE4 and CA-125 in the early diagnosis and screening of Epithelial Ovarian Cancer. Search strategy-Literature search in electronic databases. Selection criteria-Studies that evaluated the diagnostic measures of ROMA, HE4 and CA-125 by using CLIA or ECLIA as index tests. Data collection and analysis-24 studies (6,704 women) were included in our meta-analysis .We calculated AUC by SROC, pooled estimated like sensitivity, specificity, likelihood ratio, diagnostic odds ratio, Tau square, Cochran Q through random effect analysis and meta-regression. Data was retrieved from 24 studies. The pooled estimates of the three markers were sensitivity : ROMA(0.86, 95% CI 0.84-0.88)> CA-125(0.84, 95% CI 0.82-0.85)> HE4 (0.73, 95% CI 0.82-0.85) specificity: HE4 (0.91, 95% CI 0.90-0.91) > ROMA (0.76, 95% CI 0.75-0.77) > CA125 (0.71, 95%CI 0.69-0.72) AUC(SE): ROMA (0.913(0.016)) > HE4 (0.906(0.017)) > CA125 (0.846(0.019) through bivariate random effects model considering the heterogeneity. Our study found ROMA as the best marker to differentiate EOC from benign pelvic masses with high diagnostic accuracy (AUC- 0.913) as compared to HE4 (0.906) and CA-125 (0.846) with highest pooled sensitivity as compared to other markers. Specificity for ROMA (0.76) was also higher than CA-125. The AUC (0.93) and sensitivity (0.88) for ROMA was highest for postmenopausal women suggesting it as promising predictor of Epithelial ovarian cancer in this group however its utilization requires further exploration.

P-038

Association of Vitamin D with CRP Levels in Patients with Prostate Cancer

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Prostate cancer is implicated as an important cause of morbidity and mortality in men worldwide. Inflammation plays a significant role in cancer. CRP is a well-known inflammatory marker and role of vitamin D in regressing inflammation and its anti-cancerous effect has long been known. Hence, we sought to study the association of vitamin D levels with CRP levels in prostate cancer patients. The study was conducted in Department of biochemistry in association with Department of urology, VMMC and SJH, New Delhi. The case control study included forty newly diagnosed cases of prostate cancer histologically confirmed by trans rectal needle biopsy with elevated PSA level>4ng/ml and forty age matched healthy control. Vitamin D, CRP and PSA levels were estimated and its association seen. Vitamin D was significantly low ($p<0.05$) and CRP significantly high ($p<0.001$) in prostate cancer patients compared to control. A significant negative association was observed between Vitamin D and CRP in cancer patients ($p=0.0004$) which was not there in case of controls ($p=0.4418$). Moreover a significant negative association of Vitamin D with PSA (0.0042) and positive association of CRP with PSA ($p<0.0001$) was also seen. A marked increase in CRP and decrease in vitamin D supports the inflammatory etiology of Prostate cancer. It has been known that vitamin D receptors and enzymes are also expressed in human prostate epithelial cells and vitamin D has potential immunomodulatory role in reducing inflammation which could be the possible reason of a strong negative association with CRP levels as we observe in our study. Therefore it can be suggested that therapeutic supplementation of vitamin D can be recommended for chemoprevention since it can be easily supplemented as vitamins and fortified foods.

P-039

Association between Obesity and Antimüllerian Hormone (AMH) in Breast Carcinoma

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Obesity suppresses ovarian function, leading to fewer ovulatory menstrual cycles and altered circulating levels of hormones in adolescent and premenopausal women. Anti-müllerian hormone (AMH) has become an increasingly useful marker for assessing ovarian reserve. AMH plays an important role in the recruitment and growth of follicles, and in the regulation of normal breast development and involution. The aim of present study was to determine whether there is an association between obesity and AMH in breast cancer patients and to compare them with age and sex matched healthy controls. The present study was conducted in the Department of Biochemistry in collaboration with Department of Radiotherapy, Pt. B.D. Sharma, University of Health Sciences, Rohtak. The subjects were divided into two groups: Control (Group I): Healthy age matched female volunteers with no prior history of breast cancer (n=30). Group II: Newly diagnosed confirmed cases of breast cancer (n=30). Antimüllerian hormone (AMH) estimation done by a sandwich enzyme Linked-Immuno-Sorbent Assay (ELISA). The present study showed higher mean serum AMH levels in Group I (1.98 ± 0.41 ng/mL) as compared to Group II (1.67 ± 0.49 ng/mL) and this difference was statistically significant (p=0.006). Also, there was significant negative correlation between BMI and serum AMH levels in both cases and controls (r= -0.372 and r= -0.434 respectively & p=0.043 and p=0.017 respectively). Findings of our study states that AMH levels are negatively associated with obesity in both controls and breast cancer patients. Thus indicating poor ovarian reserve in obese female as compared to non- obese in both the groups. Further studies of AMH throughout the reproductive years and elucidation of its role in follicular function and follicular depletion are needed to clarify the effect of obesity on AMH levels and reproductive function.

P-040

Correlation of Levels of Proteolysis inducing Factor, C-reactive Protein, and Albumin with Nutritional Status of Patients with Gynecological Malignancies

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Response to treatment and Quality of life of cancer patients is limited by poor nutrition encountered in these patients. Most commonly implicated cause of malnutrition in cancer patients is the activation of pro-inflammatory and neuroendocrine responses to tumor elaborated mediators. Activation of the ATP-ubiquitin-dependent pathway has been found to play a major role in inducing proteolysis in cancer patients, resulting in severe malnutrition. This in part is induced by the glycoprotein, Proteolysis Inducing Factor (PIF). Literature reveals that no study has looked at role of proteolysis in malnutrition related to gynaecological malignancies. We undertook this study to explore the role of catabolic marker PIF, inflammatory marker CRP and albumin in patients with gynaecological malignancies and to look for correlation if any between the levels of these markers and nutritional status of these patients. This prospective study, on 60 patients with histopathological diagnosis of gynaecological malignancies, was undertaken in the Department of Biochemistry and Radiation Oncology at Christian Medical College & Hospital, Ludhiana. The levels of Albumin, CRP and PIF were evaluated and correlated with the nutritional status of the patients as assessed by PG-SGA. There was a significant difference in the value of PIF ($p<0.0001$) of the patients in three PG-SGA groups. The PIF levels in severely malnourished patients were significantly higher than in moderately malnourished and well-nourished patients (p=0.000). At levels of PIF above a cut off value of 5.18 ng/ml was able to predict malnutrition with 65% sensitivity and 71% specificity. However, Albumin and CRP did not have a significant correlation with the PG-SGA scores. The prevalence of malnutrition in patients with gynaecological malignancies warrants nutritional assessment and intervention to be an integral part of the treatment.

P-041

Significance of Breast Cancer Stem Cell Marker and Tumor Suppressor miRNAs (miR 200a, miR 200b, miR205 and miR 145) in Breast Carcinoma: A Step Toward Precision Medicine

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Breast cancer is a complex disease with heterogeneity and several studies have been conducted to explore differentially expressed miRNA in carcinogenesis. Tumor suppressor miRNAs (miR 200a, miR 200b, miR205 and miR 145) are involved in various signalling pathways and promote carcinogenesis and Cancer stem cells (CSCs) noticed as the driving force of tumorigenesis and metastases. Thus, my objective was to explore relationship of expressed miRNAs and Cancer Stem Cells in breast cancer patients before and after chemotherapy. 39 Breast Cancer samples were recruited after pathological approval and ethical clarification. miRNA was quantified on real-time PCR by using exiqon cDNA and Sybr green kit. CSCs (CD44+/CD24-) were characterized by using CD44 and CD24 antibodies on BD flow cytometer. Breast Cancer Stem Cell marker CD44+/CD24- were significantly reduced after three cycle of chemotherapy (Average% & Mean counts: 7.60% & 590 Vs 3.22% & 291). However, the highest frequency of cells with expression of CD44-/CD24+ were observed and remain almost unchanged after 3 cycle of chemotherapy (Average % & Mean counts: 33.68% & 23,953 Vs 32.63% & 21,648). The Breast cancer patients showed significant ($p<0.5$) down-regulated expression of miR 21 (Mean Cq 27.95±1.63 Vs 26.51±1.00) after 3 cycle of standard chemotherapy out of four tumor suppressor mir-200a, mir-200b, mir-205 and mir-145). This study had shown the all tumor suppressor miRNAs 205 showed higher expression with decrease in mean count of CSCs (CD44+/CD24-) in patients positively responding therapy. So, this and similar type of study may help in guiding more precise treatment of chemotherapy with gene therapy in near future.

P-042

Characterization of FAT1 Processing and Its Interaction with β-catenin in Glioma

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Glioblastoma multiforme patients have a very poor prognosis and average survival rate is less than 18 months, so there is a need to identify new molecular therapeutic. FAT1 is a 506 kDa transmembrane adhesion molecule protein. Studying the role of FAT1 and its interaction with beta catenin in glioma will increase our knowledge and enable to develop a treatment option with this molecule. To study the oncogenic role of FAT1 in hypoxia in glioma. Experiment were carried out in U87MG glioma cells maintained under normoxia ($20\%O_2$) and severe hypoxia ($0.2\%O_2$). Immunocytochemistry, migration/invasion, western blotting and real-time gene expression study was carried out after FAT1 modulation (FAT1 over expressed/knockdown). Furin inhibitor treatment was done to analysis FAT1 processing in glioma. CO-Ip experiment was done to study the interaction of FAT1 and β-catenin in different subcellular fraction. We observed along with full-length FAT1 protein (508 kDa), multiple smaller bands in the range of 171 to 117 kDa in different cell lines. On Furin inhibitor treatment, there was decrease in the intensity of cleaved protein bands (171 to 117 kDa) and increase in the full length FAT1 protein (1.53-fold). On exposure to hypoxia there was 1.98 folds increase in FAT1 protein and also increased nuclear localisation. Nuclear localisation of FAT1 was reduced on treatment with Furin Inhibitors. FAT1 mediated CO-IP pull down showed interaction with β-catenin in nuclear lysate. FAT1 knockdown decreases β-catenin ICC signals in nucleus both under hypoxia and normoxia. On FAT1 over expression under hypoxia, we observed, increase in the levels of β-catenin target proteins (c-JUN 1.63 fold, c-MYC 1.48-fold, VEGF 1.38-fold and Cyclin D 1.17-fold) and decreased on FAT1 knockdown samples. FAT1 may act as oncogene by aiding transportation of known oncogene β-catenin to nucleus under hypoxia condition in GBM.

P-043

Diagnostic & Prognostic Utility of IGF-1, IGFBP-3 and CEA of Squamous Cell Carcinoma in Oral Cavity

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Oral cancer represents the majority of head and neck cancer with more than half a million patients being affected each year worldwide. The high death rate of oral cancer is not because it is hard to discover or diagnose, but due to the cancer being neglected during patient's daily life. Although, early diagnosis of cancer is the best defense against this devastating disease. The IGF signaling pathway has a pathogenic role in cancer. IGF-1 in contributing to tumor angiogenesis has been suggested because IGF-I induces vascular endothelial growth factor. In addition to its role in normal growth and differentiation processes, the IGF system is involved in several pathological oral processes. IGFBP-3 exerts anti-proliferative effects in many cell type by blocking the ability of IGF-1 and IGF-2 to activate the IGF1R (which stimulates cell proliferation). Contrasting with the typical growth-inhibitory effects of IGFBP-3, stimulation of cell proliferation by IGFBP-3 has also been observed. This can occur either by enhancing IGF-stimulated proliferation. The circulating IGFBP-3 level showed a modest association with increased risk for a number of cancers, but the results vary according to site. CEA describe a set of highly related glycoproteins involved in cell adhesion. Serum level can also be elevated in heavy smokers. CEA is elevated more in tumors with lymph node and distant metastasis than in organ-confined tumors and varies directly with tumor stage. Recent evidences suggest that people today have significantly higher risk for many forms of cancer. However, the number of studies investigating the association between Oral squamous cell carcinomas with these Biochemical tests are limited.

P-044

Does Circulating miRNA-126 and -122 Plays any Role in Development of Coronary Artery Disease?

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Circulating microRNAs (miRNA) have a potential in identifying cardiac dysfunctions and shown a major interest as a biomarker for diagnosis of cardiovascular disease. The current study was designed to find the circulating miR-126 and -122 as an independent risk predictor of Indian coronary artery disease cases. Serum sample was collected from coronary artery disease (CAD) patients (n=100) and non CAD patients (n=100). Total RNA was isolated from serum by Trizol method. Circulating miRNA levels were measured by quantitative real-time polymerase chain reaction. MiR-126 significantly down-regulated in CAD patients compared to non CAD patients (80.0% vs. 39.0%, χ^2=14.95, p<0.001). The miR-122 was significantly higher in CAD patient patients compared to non CAD patients ((14.0% vs. 63.0%, χ^2=21.23, p<0.001). Multivariate analysis found chest pain (OR=37.07, 95% CI=3.21-169.04, p=0.017) and miR-126 (OR=0.01, 95% CI=0.00-0.63, p=0.030) as an independent risk predictor of CAD. Our results suggest that circulating miR-126 might be novel, non-invasive biomarkers in risk prediction for CAD. Further exposition of the role of miR-122 & 126 in the pathogenesis and progression of CAD will add to our understanding of the disease process leading to a new diagnostic approach.

P-045

Association of Serum Tenascin-C and Growth Differentiation Factor-15 Levels with the Risk of Acute Coronary Syndrome in Patients with Type 2 Diabetes Mellitus

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Increase prevalence of type 2 diabetes mellitus (T2DM) is associated with increase prevalence of acute coronary syndrome (ACS). Inflammation is one of the important contributors in the pathogenesis and complications of coronary atherosclerotic plaque. Growth Differentiation Factor-15 (GDF-15) and Tenascin-C (TNC) play important role in initiation of atherosclerotic plaque as well as its rupture. The objective of the study was to evaluate the association between serum GDF-15, TNC levels and risk of ACS among T2DM patients. A cross-sectional hospital based-study has been conducted on 42 T2DM patients with ACS and 42 T2DM patients during time period of July 2017 and December 2018. Anthropometric parameters, routine biochemical investigations like liver and kidney function tests, lipid profile and Creatine Kinase-Total (CK-T), Creatine Kinase-MB (CK-MB) were measured on Automated Clinical Analyzer. Serum GDF-15 and TNC levels were estimated by Human Sandwich-ELISA kits. Serum GDF-15 (1155.45 ± 321.01 vs 841.63 ± 477.04 pg/mL, P=0.011) and TNC (13.06 ± 9.80 vs 6.16 ± 3.58 ng/mL, P<0.001) levels were

significantly higher in T2DM patients with ACS as compared to T2DM patients. Serum GDF-15 level was significantly correlated with waist circumference, diastolic blood pressure, pulse, serum CK-T (r=0.329, P=0.031) and CK-MB (r=0.436, P=0.014) levels. Serum TNC level was significantly correlated with pulse, serum CK-T (r=0.320, P=0.021), CK-MB (r=0.560, P=0.011), high density lipoprotein-cholesterol and blood urea nitrogen. Multivariate linear regression analysis revealed that larger waist circumference to be positively associated with serum GDF-15 levels [Adjusted odd ratio (95% CI) = 4.72(2.40-21.18), P=0.014]. Serum GDF-15 and TNC levels may be considered as one of the parameters for predicting, diagnosis of acute coronary event in patients with T2DM.

P-046

Study of Lipid Profile and hsCRP Levels in Obesity

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Numerous biomarkers involved in inflammation have been associated with cardiovascular events, out of which high sensitivity C reactive protein (hsCRP), an acute phase protein, appears to be the most promising. This study was designed to find the statistical relation between hsCRP and lipid profile with obesity. The present study was carried out in the Department of Biochemistry, Kalpana Chawla Government Medical College, Karnal (Haryana) on 100 apparently healthy volunteers attending the OPD/ward as attendants of patients, who were divided on the basis of body mass index (BMI) according to Asian guidelines into three groups; Healthy (Normal): 18.5-22.9 kg/m^2, Overweight: 23-24.9 kg/m^2, and Obese>25 kg/m^2. Anthropometric measurements and biochemical investigations were carried out in all the study subjects. The median levels of HDL were the lowest in the obese group (37.9 mg/dl) and a statistically significant difference was observed in HDL levels between healthy and obese group (z=3.190, p=0.001) and between overweight and obese group (z=2.760, p=0.006). The median hsCRP levels were highest in the obese group (2.5 mg/L) followed by overweight (2.0 mg/L) and the lowest in the healthy group (1.1 mg/L). A statistically significant difference in the levels of hsCRP was observed between healthy and overweight group (z=2.009, p=0.044) and between healthy and obese group (z=2.849, p=0.004). A significant positive correlation was observed between BMI and hsCRP levels (r=0.302, p=0.002). It was further observed that seventeen subjects of obese group had hsCRP levels greater than 3 mg/L as compared to eight of healthy group and nine of overweight group. According to the recommended cut off point for cardiovascular disease (CVD) assessment, it can be safely concluded that the subjects of the obese group are at the highest risk of CVD.

P-047

Association between Pro-inflammatory Markers and Risk Factors of Cardiovascular Disease in Night Shift Health Care Workers

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Chronic disruption of circadian rhythm in night shift workers can lead to various immunological responses ranging from low grade systemic inflammation to endocrinal rhythm disruptions leading to increased risk of CVD and other metabolic disorders. Aim was to find out the levels of pro-inflammatory markers (hsCRP) in health care workers those doing night shift and find its association with the risk factors (Total cholesterol, TG, HDL, BMI) of CVD and metabolic disorders. Health care workers of age group 20-60 years were recruited for the study segregated into two groups, those doing night shift (n=105) and only regular time work (n=105). Groups were age and sex matched. Participants with known inflammatory disorders, DM, Hypertension, CVD were excluded from the study. 5ml of venous blood sample was collected for analysis of Fasting blood sugar (FBS), TC, TG, HDL, LDL and hsCRP. Statistical analysis was done by Epi info software (7.1.2.0). Comparison between two group was done by independent t- test. Chi-square test was used for analysis of categorical variables. The mean value of hsCRP, BMI, TC and LDL were higher among night shift worker. 65.52% of night shift and 34.48% of day shift worker had TC>200 mg/dl (p=0.08). 60.94% of night shift and 39.06 % of day shift worker had HDL<40 mg/dl (p=0.03). Night shift worker had high LDL cholesterol, which is found to be highly significant (p=0.008). TG value was not significant between the group.11.9% had raised hsCRP. Among them 80% were of those in night shift & 20% worked in day shift. These differences were statistically significant (P<0.003). Hence hsCRP can be used for early detection of long-term health side effects of night shift work. Health education can be provided related to the CVD and co- morbidities associated with shift work.

P-048

Effect of Smoking on Matrix Metalloproteinase-9 in Young Smokers in the Acceleration of Coronary Heart Disease

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Smoking causes up to one fifth of total global cardiovascular deaths. Smoking has numerous effects that may promote atherosclerosis through vascular inflammation and oxidative stress, but the pathogenesis of smoking related cardiovascular disease remains unclear. Matrix metalloproteinases (MMP) are a family of Zinc-containing zymogen endoproteinases, which has a role in the normal and injury-induced turnover of the extracellular matrix. MMP-9 degrades interstitial collagens, proteoglycan core protein, elastin and type IV collagen in the basement membrane. It has been proposed that localized inflammation may promote production and activation of MMP-9 that brings about de-stabilizing structural changes resulting in formation of vulnerable atherosclerotic plaques. This study was planned to determine the circulating levels of serum MMP-9 in subjects with and without the habit of smoking. This cross-sectional study comprised of 200 smokers and 200 controls in the age group of 20-45 years. MMP-9 levels were measured using sandwich enzyme immunoassay method. Plasma MMP-9 levels were found to be significantly increased in smokers compared to non- smokers. MMP-9 is a useful biomarker found to be increased in young smokers compared to non -smokers and also useful in predicting the future CHD risk.

P-049

Biochemical Markers of Endothelial Dysfunction and their Association with Disease Severity in Pre-dialysis Non-diabetic Chronic Kidney Disease.

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Chronic kidney disease (CKD) is associated with increased cardiovascular (CVD) morbidity and mortality. Hence, this study was carried out to assess the biomarkers of endothelial dysfunction and inflammation as predictors of CVD risk in patients with CKD. In this cross-sectional study, we recruited 43 patients with pre-dialysis non-diabetic CKD and 43 age and gender-matched healthy controls. Serum levels of endothelial dysfunction markers (ADMA, ANGPTL2, MMP-9) and systemic inflammation (hsCRP) were assayed in all study participants. All study participants underwent flow mediated vasodilation (FMD) of the brachial artery, which is a non-invasive marker of endothelial function. CKD patients showed markedly elevated levels of ADMA, ANGPTL2, MMP-9, and hsCRP, whilst FMD and eGFR (disease severity) were significantly decreased in cases, as compared to the controls. ADMA, ANGPTL2, MMP-9 and hsCRP showed significant positive correlation with one another and significant negative correlation with FMD and CKD severity. We also observed a significant negative correlation of FMD with CKD severity and duration. In CKD patients, there is significantly increased endothelial dysfunction and systemic inflammation, which showed positive correlation with disease severity. Thus, the biochemical markers of endothelial dysfunction and systemic inflammation can be used to assess the CVD risk in CKD.

P-050

Association between Carbonyl Stress Markers and Risk of Acute Coronary Syndrome in Patients with Type 2 Diabetes Mellitus-A Pilot Study

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Carbonyl stress is either due to increased production or inadequate detoxification of reactive carbonyl compounds (methylglyoxal and glyoxal). Reactive carbonyl compounds are responsible for synthesis of advanced glycation end products (AGEs). AGEs, mainly pentosidine, carboxymethyllysine (CML), methylglyoxal-derived hydroimidazolone-1 (MG-H1), are one of the causes for pathogenesis and complications of type 2 diabetes mellitus (T2DM). Diabetic subjects have 2 to 4 folds higher risk of coronary artery disease. Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) and triose phosphate isomerase (TPI) are involved in synthesis while glyoxalase 1 (GLO1) and aldose reductase (ALR) are responsible for detoxification of reactive carbonyl compounds. Hence the objectives of this pilot study were to measure carbonyl stress markers and to determine their association with risk of acute coronary syndrome (ACS) among T2DM patients. Forty T2DM patients with ACS as cases and 40 T2DM patients as controls were recruited. Anthropometric parameters and routine biochemical blood investigations were

estimated. Serum carbonyl stress markers (methylglyoxal, pentosidine, CML, MG-H1, GAPDH, TPI, GLO1, ADR) were estimated by Human Sandwich-ELISA kits. Fasting plasma glucose and serum methylglyoxal were significantly higher in cases as compared to controls. Serum MG-H1, CML were also significantly higher in cases as compared to controls. Serum GAPDH and GLO1 levels were significantly lower in cases as compared to controls. Fasting blood glucose was significantly positively correlated with serum methylglyoxal ($r = 0.441$, $P = 0.001$), CML ($r = 0.649$, $P < 0.001$), MG-H1 (0.725, $P < 0.001$) and negatively correlated with serum GAPDH ($r = -0.268$, $P = 0.012$) and GLO1 ($r = -0.634$, $P < 0.001$). In conclusion, reduced GAPDH levels increase the availability of substrate and reduced GLO1 levels result in less detoxification of carbonyl compounds, which lead to more production of AGEs that may predispose T2DM patients for higher risk of ACS.

P-051

Serum Esterified Cholesterol Subfractions as a Risk Marker for Coronary Artery Disease

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Coronary artery disease (CAD) a modern epidemic is caused by atherosclerosis, which involves formation of plaques where cholesterol uptake takes place from the remnant lipoprotein (RLP) and the small dense low-density lipoprotein (LDL) particles. The remnant lipoproteins and the LDL cholesterol are relatively cholesterol ester rich. Hence cholesterol ester, which is the main component of these lipoproteins, is atherogenic and hence total esterified cholesterol and esterified cholesterol fraction of non HDL cholesterol were measured to assess as a better and more specific risk marker for coronary artery disease. 56 Patients of coronary artery disease proven by angiography with significant coronary artery stenosis were taken. The biochemical parameters estimated were - the classical lipid profile including total cholesterol, LDL cholesterol, HDL cholesterol and triglycerides; serum non-HDL Cholesterol; serum esterified cholesterol and free cholesterol; and serum non-HDL esterified cholesterol values. The control group consisted of forty health individuals based on routine annual medical checkup, and were negative for IHD on ECG. They were also subjected to a similar measurement of biochemical parameters. Odd's ratio and p value was evaluated for each risk factor. Presence of diabetes mellitus and smoking were statistically significant. All parameters namely total cholesterol, LDL-C, HDL-C, Non HDL-C, triglycerides, total free cholesterol, esterified and free fraction of Non HDL-C and total esterified cholesterol were compared using Z statistical analysis test. Total esterified cholesterol was not found to be statistically significant. Non-HDL esterified cholesterol was found to be statistically significant and hence its cut off value was established by using Receiver Operator Characteristics (ROC) Curve. The cut off value was found to be 107 mg/dl. Hence Non HDL esterified cholesterol can be used as a risk marker for assessment of CAD, though further studies are needed.

P-052

Association between Oxidized LDL and LDL in the Development of Coronary Heart Disease

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Oxidative stress plays a critical role in the initiation and progression of atherosclerosis. An accumulation of LDL in the arterial intima is an early step in atherosclerosis the elevated concentration of Low Density Lipoprotein (LDL) are found to accelerate atherogenesis by enhancing oxidative modification of LDL to form oxidized LDL. Oxidized LDL is a marker of oxidative stress Oxidized LDL (OxLDL) promotes the immune and inflammatory reactions that characterize atherosclerosis. The study was carried-out to evaluate the association between Oxidized LDL and LDL in the development of Coronary Heart Disease. This Cross-Sectional study was conducted on 100 Coronary Heart Disease patients (study group) attending the cardiology OP at SRM Medical College Hospital, whereas age and sex matched healthy 100 people were selected as controls. Blood Samples collected after 12 hours fasting. Total Cholesterol using Cholesterol Oxidase, Triglycerides using Glycerol Peroxidase, Lipid Profiles Using Direct Antibody Inhibition, Oxidized LDL were estimated using ELISA kit. Data were be analysed using statistical package for social service (SPSS 22.0). Student 't' - test were performed to compare the different measured variables & Pearson's correlation analysis used to study the correlation between Case and Control. Patients with Coronary Heart Disease showed significant elevation of LDL (121.23±39.87) when compared to controls (97.43±24.79) ($p<0.005$). Oxidized LDL also showed significant increase (123.4±38.9) in study group when compared to controls (84.57±23.68) ($p<0.005$). Oxidative modification of LDL plays a pivotal role in atherosclerosis and LDL oxidation is a direct contributor to atherogenesis. This study concludes that the serum Ox-LDL increased with the development of disease and more attention should be focused on marker of oxidative stress in the management of CHD.

P-053

Increased Risk of Subclinical Atherosclerosis in Patients with Diabetic Neuropathy

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The macroangiopathy complications of type 2 diabetes like retinopathy and nephropathy has been associated with atherosclerosis but association of peripheral neuropathy is not widely reported. In this study the risk factors of atherosclerosis NO, hsCRP, PAI-1 (Plasminogen activator inhibitor), APC (activated protein C) levels were analysed in patients with type 2 diabetes mellitus with and without neuropathy. In this study we screened 252 patients and enrolled 32 diabetic patients with neuropathy without evidence of nephropathy/retinopathy/ macroangiopathy and 32 patients with type 2 Diabetes without any complications were selected. Blood glucose and HbA1c were estimated by commercially available kit by autoanalyser. Nitric oxide (NO) was estimated by modified Griess method. PAI-1, APC, hs-CRP levels were estimated by ELISA using commercially available kits. Statistical analyses were performed using SPSS version 19.0. The mean age of diabetics with neuropathy was statistically high. PAI-1, APC, hs-CRP levels were significantly high in patients with neuropathy ($p<0.05$). Regression analysis was done and unstandarized coefficient (B) was calculated to predict the dependent variable from the independent variable, values were 0.189, 0.16, 0.085, 0.957 for PAI-1, NO, APC, hs-CRP resp. All values were significantly high. Area under receiver operating characteristic curve was maximum for hs-CRP. Our findings suggest that diabetic neuropathy is an independent risk factor for subclinical atherosclerosis. We suggest that careful cardiovascular assessment in patients with diabetic neuropathy at the time of diagnosis might decrease the risk of future cardiovascular events but the sample size in our study was small so further long-term prospective research is necessary to confirm our findings.

P-054

Investigation of Correlation between Glycemic Control and Plasma Homocysteine, Lipoprotein a, hsCRP and Atherogenic Index to Assess Cardiovascular Risk

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Diabetes mellitus is an important risk factor for cardiovascular disease, which is a major cause for morbidity and mortality in the world. The aim of our study was to determine the association between different grades of HbA1c and markers of inflammation i.e. plasma homocysteine, hsCRP, lipoprotein A, atherogenic index and lipid profile to assess cardiovascular risk. 60 cases of type 2 Diabetes mellitus were selected from patients attending OPD in NIMS Hospital. Fasting samples were collected and were analyzed for glycosylated hemoglobin, homocysteine, lipid profile, serum creatinine, lipoprotein A, fasting glucose and hsCRP levels in the autoanalyser. Atherogenic index of plasma (log TG/HDL) was calculated. Patients were grouped depending on the level of glycemic control into three groups- HbA1c <7% (good control), HbA1c 7-9% (poor control) and HbA1c >9% (uncontrolled). The mean HbA1C level in each group was 5.8, 7.9 and 10.6 respectively. Kruskal-Wallis test was applied across the three categories to compare the variables due to non-normal distribution. Significant difference is seen in hsCRP, triglycerides, HDL and atherogenic index with H value of 16.1, 7.63, 11.5 and 10.37 respectively while LPA and homocysteine showed H value of 1.22 and 1.49 which were not significant. Spearman rank correlation coefficient showed positive association between HbA1c and hsCRP, Lipoprotein A, atherogenic index, triglycerides, while an inverse association was noted between HbA1c and homocysteine and HDL. Based on this study we have concluded that poorly controlled diabetes mellitus is associated with increased levels of markers of inflammation i.e. hsCRP, AIP and lipoprotein A and decreased levels of HDL. These markers may be contributing factors for atherosclerosis in diabetic patients. We recommend that subjects with uncontrolled diabetes need to be evaluated by a multi-marker approach for possible risk for atherogenesis.

P-055

Evaluation of Serum Cardiac Biomarkers in Acute Myocardial Infarction

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Acute Myocardial Infarction (AMI) is the leading cause of mortality and morbidity, and early diagnosis is key to successful management of these patients. Commonly used biomarkers of AMI are; Creatine Kinase MB isoenzyme (CK-MB) Cardiac- troponin I (cTnI), Lactate Dehydrogenase isoenzyme (LDH), myoglobin etc. In our institute CKMB and cTnI are routinely performed in patients presenting with complaints suggestive of MI, hence this study was planned to find the sensitivity and specificity of these markers. Secondary data analysis was carried out for finding the sensitivity and specificity of cardiac biomarker. During the period of December 2018 to April 2019, total 528 patients presented with complaints of chest pain, and were managed as per standard protocol, out of these we included 228 patients in whom CKMB and cTnI were performed within 24 hours of onset of chest pain. CKMB was performed by Immuno-Inhibition by blend of monoclonal antibody and cTnI was performed by chromatographic immunoassay rapid card test. Depending upon the final diagnosis, they were divided into two groups, Group I patients with conformed diagnosis of MI (n=73) and Group II patients without MI (n=155). Data was tabulated into groups; CKMB ≥24, CKMB >24, cTnI +ve, cTnI -ve. On the basis of these data the sensitivity and specificity of CKMB, cTnI and combined CKMB with cTnI were calculated. Within 24 hours of onset of chest pain the sensitivity and specificity for MI, CKMB were 82.19% and 65.8%, and for cTnI were 57.53% and 92.25% respectively, whereas the combined sensitivity and specificity were 86.3% and 63.87% respectively. From this study we conclude that CKMB is more sensitive and cTnI is more specific for diagnosis of MI. When both tests are combined the sensitivity is further increased. Therefore, it is advisable to perfom cTnI rapid card test in combination with CKMB in MI suspected patients.

P-056

Detection of Possible Determinants for Prediction of Risk for Cardiovascular Disease in the Rural Population of Central India: A Cross-Sectional Study

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Estimation of risk for the coronary heart disease (CHD) in population involves identification of those persons at high risk. This provides a cost-effective prevention strategy against CHD-associated health burden in the resource-poor countries including awareness and lifestyle changes. Towards this end, a cross-sectional study including 572 subjects was carried out in rural population of Wardha during 2017-19 to detect at-risk individuals and its underlying determinants. The study population was then stratified in following categories of CHD risk group by definitions used from Framingham's risk score online calculator; low risk <10%, moderately high risk 10-20% and high risk >20%. Of total 572 subjects, 47.7% population were female and 52.3% were male. Majority of population is involved in physical work except 1.4% of population belonging to service class. As per definitions of HOMA indexing, 92% (517) of study subjects did not have insulin resistance while only 8% (45) had it. Based risk categories for CHD, 12.8% (72) were at moderate risk and 2.0% (11) were at high risk for cardiovascular events in next 10 years. Logistic regression analysis revealed that female sex (95% CI 8 to 147.16; $p<0.0001$), in-service persons within high income tertile (95% CI 1.365 to 25.92; $p<0.01$) and 30-50 years of age (95% CI 8 to 65.56; $p<0.0001$) are risk determinants of CVD. Study had concluded that risk of CHD events seems to be rising in female population of rural Wardha without frank insulin resistance, a popularly known predictor of CHD. This study definitely highlighted the importance of considering other pathophysiological factors as predictors of CHD. These findings have significant implications for prediction of CHD risk in young rural women and in service class with higher income. Hence emphasis on detection of factors involved in up-surge of CHD and its preventive management is imminent.

P-057

Novel Cardiac Biomarkers

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Measurement of biomarkers has revolutionized the work-up of patients with suspected cardiovascular disease. The most widely used contemporary cardiovascular biomarkers are the natriuretic peptides in the diagnosis and prognosis of heart failure and cardiac troponins in the diagnosis of acute myocardial infarction. Numerous other biomarkers pertaining to diagnosis, prognosis, and risk prediction have been identified, but few have made their way to clinical practice. This review describes the fundamental approach to evaluate a novel biomarker. Before implementation of a biomarker into clinical practice, several stringent criteria related to its clinical utility are required. Biomarkers are categorized according to main groups of cardiovascular pathology: 1. Myocardial injury (Cardiac troponins); 2. Myocardial stress (B-type natriuretic peptides and N-terminal B-type natriuretic peptides); 3. Inflammation (C-reactive protein); 4. Risk stratification of heart failure (suppression of tumorigenicity-2 ST-2); 5. New biomarkers for risk stratification of developing heart disease in individuals who are otherwise apparently healthy (NT-proBNP). This review highlights the importance of relating cardiac biochemical markers with specific time points along the cardiovascular continuum, especially during the early transient phase of pathology progression.

P-058

Variability in Coronary Artery Disease Risk Assessment Score Assessed by American Diabetes Association and Indian Diabetes Risk Score in First Degree Relatives with a Family History of Coronary Artery Disease

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Coronary artery disease (CAD) risk assessment among asymptomatic individuals is inconsistent in the context that individuals with no predisposing factors or one risk factor might experience cardiac attack. Furthermore, the factors included by various risk engines for risk score calculation influence the final score. Hence the present study was designed to compare the variability in coronary artery disease risk score quantified by American Diabetes Association and Indian Diabetes Risk Score in first degree relatives. This cross sectional study was conducted among first degree relatives (n = 113) with a family history of CAD. The risk was quantified by using the algorithms of American Diabetes Association (ADA) and Indian diabetes risk score (IDRS). The total score was compared among the groups. ADA risk score was significantly ($p<0.01$) high (6.75 ± 1.96) in first degree relatives of established CAD subjects (n = 73) as compared to the individuals with no family history (n = 40) of CAD (5.72±1.79). In IDRS risk score first degree relatives of both parents with CAD history (n = 14) had highest score (86±13) followed by the relatives of one parent CAD positive (n = 51) history (66±19) and no family history (n = 48) (59±19) and the difference was statistically significant ($p<0.01$). Reflecting on both the common factors like race, ethnicity, gender and the prevalence of specific predisposing risk factors in a particular geographic location, use of population specific risk assessment tools should be considered for CAD risk quantification.

P-059

The Assessment of the Important Biochemical Parameters in the Urban Hypertensive-Adolescents

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The prevalence of Hypertension (HT) in the young population(Age group 12-20 years) is increasing. The early diagnosis of HT in adolescent population, the detection of metabolic defects of HT, and the management of the same are crucial points in the disorder. The causes and the metabolic changes in the disorders are obscure. The comparative aspects of various biochemical parameters (Fasting glucose, LFT, RFT, Electrolytes, Lipid profile, Hs-CRP and Homocysteine) in HT- Adolescents (HT-A) and Control Adolescents (C-A) were studied. The results were statistically analysed (N=10). The study revealed decrease (10-20%) in the biochemical parameters viz. ALP, BUN, UA, Calcium, and the Electrolytes of confirmed HT-adolescent subjects (Age 12-20 years), while the elevations were observed in Hs-CRP (41.3%) and Homocysteine (75.6%) levels. It exposed early alterations in the HT-adolescents.

P-060

Can Diuretics Modulate the Gut Microbiome to Show Superiority Over Other Class of Antihypertensive Medication?

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Hypertension is a silent killer of mankind and thiazide, as well as some loop diuretics, is shown to be superior for control of hypertension among other class of medicines used to treat hypertension. The mechanism of such action is currently unknown to mankind. In this context, we have observed that diuretics that offer protection have sulphonamide backbone and binds with bacterial enzymes of folate metabolism. Secondly, it is observed that it shows antimicrobial properties to the component of the gut microbiome that contains enzymes of folate metabolism. As per literature, some butyrate-producing taxa of the gut microbiome which do not contain DHFR are also present. So, diuretics administration akin to Metformin can proliferate the butyrate-producing taxa of the gut microbiome and butyrate being a vasodilator can offer extra protection in hypertension.

P-061

Long Term Renal Outcomes in Children Post Cardiac Surgery AKI- Results of ISN Clinical Research Program Grant

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Acute kidney injury (AKI) is associated with poor short-term outcomes such as mortality, duration of ventilation etc. This study aims to study the long-term renal outcomes and markers of kidney injury in paediatric patients with and without AKI post cardiopulmonary bypass (CPB) surgery. In a prospective case-control observational study, all children who underwent CPB surgery (December 2010-2017) were evaluated. Cases were defined as patients who developed AKI and were matched to consecutive controls who did not develop AKI post operatively. During the study period 2,035 patients underwent CPB of whom 9.8% (200/2,035) developed AKI post operatively. However, only 44 patients who had postoperative AKI, had a long term follow up, and met our inclusion criteria and therefore they were matched to 49 controls. There were no significant differences between cases and controls at the time of surgery, except that patients who developed AKI had a higher baseline serum creatinine, a higher postoperative serum creatine, a longer ICU length of stay, and a trend towards a larger weight, a longer CPB time, and a higher rate of sepsis than their controls . At follow-up, patients with post-operative AKI had a higher serum creatinine level, and a trend towards higher urinary KIM-1 levels, and a lower estimated GFR. CPB time was the only risk factor associated with a lower GFR at follow up CPB time, baseline serum creatinine and AKI remained the only risk factors associated with KIM-1. In children with congenital heart disease, CPB time is significantly associated with a decrease in GFR and a rise in kidney injury biomarker KIM-1 level several months post operatively independent of postoperative AKI.

P-062

Serum sFlt-1: PlGF Ratio in Complicated and Non-complicated Preeclampsia

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Women with preeclampsia are at an increased risk for life-threatening obstetric or medical complications. Various recent studies have revealed that the sFlt-1:PlGF ratio (serum soluble Fms like tyrosine kinase -1: Placental growth factor ratio) in the Preeclamptic women is significantly higher compared to controls. The purpose of this study is to follow the cases of Preeclampsia till delivery to correlate the serum sFLT1:PLGF ratio with possible Preeclamptic complications in these follow up cases. Singleton live pregnancy in ≥ 20 years of maternal age with weeks of gestation from 20 till delivery presenting with features of Preeclampsia were recruited for this study. Total forty-four cases of Preeclampsia were selected according to the criteria defined by the American College of Obstetricians and Gynecologists. Blood pressure, urinary protein, serum sFlt-1, serum PlGF and sFlt-1:PlGF ratio was recorded in each case at the time of presentation. Concentration of sFlt-1 and PlGF were measured with commercially available ELISA kits. The cases were followed up till delivery to observe for the possible maternal and fetal complication 27 (61.36%) cases of preeclampsia developed complication. Eclampsia, persistent thrombocytopenia and elevated liver enzymes were observed as maternal complication. Likewise, low birth weight, intrauterine fetal death, immaturity, intrauterine growth retardation and birth asphyxia were seen as fetal complication.

P-063

Appraisal of Diabetic Nephropathy through GFR Estimation in View of Various Cystatin C Equations A Relative Investigation

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Diabetes mellitus is a chronic polygenic syndrome which impairs carbohydrate metabolism. The causes for impaired carbohydrate metabolism are deficiency or decreased effectiveness of hormone insulin known as peripheral insulin resistance or due to decreased ratio between insulin and anti-insulin hormones. Commonest complication Diabetic nephropathy results due to the glycation of the basement membrane protein which occurs due to longstanding hyperglycemia. Estimation of GFR is the most accurate method of detecting the initial renal impairment for which the most commonly used parameter is serum creatinine regardless of its demerits. An alternative marker is serum cystatin c which is found more advantageous. To estimate the level of cystatin c in patients with long standing type II diabetes mellitus. To compare the serum levels of cystatin c in type II diabetic patients and controls. To compare the serum levels of creatinine in type II diabetic patients and controls. Estimation of Glomerular Filtration Rate by various equations using the parameters cystatin c and creatinine. Study was conducted both as a cross-sectional study involving 60 type II diabetic patients (based on elevated HbA1c levels) and a case-control study which included 60 non-diabetic controls and 60 type II diabetic patients. Serum level of both cystatin c and creatinine were measured in both groups. Serum creatinine estimated by Jaffe's kinetic method and serum cystatin c by immunoturbidimetric method. Serum levels of both cystatin c and creatinine were significantly increased in cases as compared to the non- diabetic controls. The most significant equation for estimating GFR was CKD-EPI (cystatin c based). Cystatin c is an alternative to creatinine. Cystatin c based CKD- EPI formula may be considered for estimating GFR than creatinine based MDRD formula.

P-064

Microalbuminuria in Chronic Obstructive Pulmonary Disease Patients

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To determine urine microalbumin in COPD patients and asymptomatic smokers. This is a case control study involving two hundred and forty nine patients diagnosed with COPD, and ninety eight asymptomatic controls (smokers) who were matched according to age and sex. They were selected from outpatient department (OPD) and In patients Department (IPD) of Respiratory and Critical care unit in Tribhuvan University Teaching Hospital (TUTH). Urine microalbumin, Arterial Blood Gas Analysis and Pulmonary Function Test were assessed in both groups. For comparison, COPD patients were divided into four different subgroups based on the duration of COPD. SPSS ver.21.0 was used to analyze the data. Mean comparison was done by t-test. Group comparison was done by Chi-square test. Pearson's correlation was used to establish the correlation. Majority of patients with microalbuminuria (MAB) in this study were in the GOLD stage of I and II and rarely of stage III but stage IV was absent. Patients with COPD had significantly higher levels of microalbuminuria than control subject ; higher UACR ; lower FEV1 ; lower FVC and lower FEV1/FVC ratio with $p \leq 0.001$, higher PCO2 ($p=0.010$) and lower PH ($p=0.028$) respectively. Out of two hundred and forty eight COPD patients, two hundred ten were with MAB and higher UACR ($p \leq 0.001$), thirty eight patients were without MAB. COPD patients with MAB were more hypoxic and more hypercapnic compared to COPD patients without MAB but was statistically non-significant. Microalbuminuria is found increased in patients with COPD compared to healthy smokers. Its level is also increased as the disease progresses in terms of duration. Microalbuminuria can be a clinically relevant tool identifying COPD patients with poor prognosis and their monitoring.

P-065

Estimation of Serum Parathyroid Hormone (PTH) in Different Stages of Chronic Kidney Disease and its Relationship with the Severity of the Disease

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Despite the importance of PTH measurements in CKD still there remains a dilemma regarding clinical relevance and modes of action of PTH. Besides this even though various recognized therapies like vitamin D analogs, management of serum mineral concentration with dietary regulations, binders and dialysis still, many patients eventually develop refractory metabolic abnormalities of severe hyperparathyroidism. The present study is an attempt to measure the levels of PTH at different stages of CKD and find out the factors responsible for elevated PTH. 146 CKD patients were recruited for the study within a span of 2 years. They were further subdivided into different stages based upon eGFR according to the MDRD formula. 5 ml of venous blood samples were collected for analysis of serum creatinine, calcium and phosphorus by an automated analyzer and, vitamin D and PTH by electrochemiluminescence method. All the variables were

distributed stage-wise expressed as mean ± standard deviation. Univariate and multivariate analysis was done to analyze the independent predictor of elevated PTH with the severity of CKD. Univariate analysis showed higher creatinine (Odds ratio=1.30) and phosphate (OR=1.25) and lower eGFR (OR=0.94) are associated with the severity of CKD (p<0.001). There is a significant increase in PTH with the increase in staging (p<0.001). Lower values of vitamin D and eGFR are related to higher PTH (p<0.001). Serum creatinine, calcium and vitamin D are independent predictors of elevated PTH. This study suggests the importance of monitoring the levels of PTH regularly in CKD so that it may prevent further complications like bone mineral disorders and cardiovascular mortality related to hyperparathyroidism.

P-066

Association of Inflammatory Markers and Negative Symptoms in Patients with Schizophrenia in a Tertiary Care Hospital, West Bengal

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The influence of the immune system deregulation on the risk of schizophrenia is increasingly recognized. Several proinflammatory and anti-inflammatory cytokines have been studied in drug-naive, first-episode, and/or chronic schizophrenia patients. Peripheral and CNS-localized inflammatory processes are hypothesized to contribute to the complex pathophysiology of schizophrenia. We thus hypothesized that inflammatory markers would predict development of psychotic symptoms in patients with schizophrenia. The aim of the study was to assess the association of serum interleukin-6 (IL-6) level, high sensitivity C-reactive protein (hsCRP) levels with positive and negative symptoms of schizophrenia. We recruited 90 clinically stable patients with schizophrenia and 80 healthy control subjects from Nil Ratan Sircar Medical College, Kolkata, West Bengal. Psychopathology was evaluated using Positive and Negative Syndrome Scale (PANSS) and Wisconsin Card Sorting Test to screen the neurocognitive Function. Serum IL-6 and hsCRP levels were assessed. A positive correlation was observed between CRP and PANSS negative symptoms (r=0.6, p=0.03), between IL-6 and PANSS negative symptoms (r=0.56, p=0.001) and There was no correlation between CRP, IL-6 with positive symptoms of schizophrenia (p=0.3 and p=0.21). Serum IL-6 was significantly higher in Patients in comparison with healthy controls. The correlation between CRP, IL-6 and negative PANSS of patients supports a role for inflammation in the pathophysiology of schizophrenia. Till now no specific treatment can be recommend for patients with more negative symptoms. This is particularly problematic for individuals burdened with negative symptoms in the face of mild or absent positive symptoms. These patients are at clinical high-risk and associated with worse functional outcomes. Though limited by a relatively small sample size, our findings demonstrate that inflammatory cytokines may underlie the development of negative symptoms in individuals with schizophrenia. Identification of biomarkers may help to improve treatment efficacy as well as outcomes.

P-067

An Audit of Patients with Low Serum Total Alkaline Phosphatase in an Australian Hospital Population

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Hypophosphatasia (HPP) is an inborn error of metabolism. Adult onset HPP is usually asymptomatic, of little clinical consequence and unknown prevalence. We surveyed clinical records of hospital patients with a very low total alkaline phosphatase (ALP). All ALP ≤ 20 U/L in hospital patients >18 years age during 2016 and 2017 were extracted from the laboratory information system. Their clinical notes were reviewed on the hospital electronic medical record. A checklist of documented clinical features, medications, laboratory and radiological features of HPP was completed for each patient. A total of 101 patients had an ALP ≥20 U/L out of 284,341 reported values. Eight patients with a history of massive transfusion were excluded and the rest (33 females and 60 males, age 19-91 years) were studied. ALP ranged from 5-20 U/L (median, 18 U/L). Thirty eight patients (41%) were on medication known to cause a low ALP. Fourteen of these and another six patients (altogether 22%) had magnesium deficiency. Eight of these were given magnesium replacement and ALP normalised in five. Eleven patients (12%) had a past history of fracture; six had a single fracture and five had multiple fractures. Twenty-one (23%) gave a history of muscle weakness, 44 (47%) had a history of skeletal or joint pain, two (2%) had lost secondary dentition and one (1%) had short stature. The low ALP was specifically recorded in the clinical notes in three (3.2%) patients only; one of these patients had been diagnosed with congenital HPP and managed accordingly. Most patients attending hospital with a very low ALP do not have their ALP status recorded in their notes suggesting the low ALP may not be recognized nor investigated for the possibility of HPP. Recognition of a low ALP and further investigation where warranted may help identify patients with adult onset HPP.

P-068

Validation of HbA1c Assay from Whole Blood Samples Collected in Gray top BD Vacutainer® for Glucose Measurement

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Diabetes mellitus is a condition characterized by hyperglycemia resulting from the body's inability to use glucose for energy. Whole blood sample collected in lavender top EDTA vacutainer is a recommended specimen for HbA1c assay. The present gray top BD vacutainer® for glucose measurement contains Na Fluoride and Na_2 EDTA as anticoagulant. The aim of the present study is to validate HbA1c assay from whole blood sample collected in grey top BD vacutainer® for glucose measurement. We have recruited 125 patients who are advised to get both glucose and HbA1c tests. Patients were segregated into 5 groups based on their HbA1c levels (4-16%). Simultaneously, HbA1c levels were also measured from samples collected in gray top vacutainers. HbA1c levels were estimated by using Bio-Rad D-10TM using HPLC technique. Spearman rank correlation was performed to establish the correlation between HbA1c values obtained in two different vacutainers. Bland and Altman analysis were done to elucidate the mean difference and bias between two approaches. The correlation coefficients between HbA1c values on parallel samples collected in EDTA and Na fluoride/Na EDTA were 0.97 and 0.99 in non-diabetic (HbA1c: 4-6%) and diabetic patients (HbA1c>6.0% respectively. Cumulatively, the correlation coefficient for entire range of HbA1c was 0.99) suggesting that both the vacutainers yielded similar performance. Bland and Altman analysis revealed 0.04% as the mean difference between the HbA1c values between both the vacutainers. The bias was estimated to be ±0.2%. The coefficient of variation (%CV) on storage for 72 hrs was 0.94 in Na fluoride/Na EDTA sample while it was 0.81 in EDTA sample. In conclusion, Our results clearly demonstrated that there is a good correlation between HbA1c levels in EDTA anticoagulant and Sodium fluoride/Na EDTA samples. Measurement of Glucose and HbA1c from single BD glucose vacutainer® is cost effective and can be used safely.

P-069

Perturbations in Iron Metabolism in Uncontrolled Diabetes Mellitus

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The relation between iron metabolism and diabetes is bi-directional, iron affects glucose metabolism and glucose metabolism impinge on several iron metabolic pathways. In the steady state, circulating iron is bound to transferrin and is taken up from the blood by a high-affinity specific transferrin receptor. The transferrin-receptor complex is internalized by endocytosis and released into a nonacidic cellular compartment, where it can be used in the synthesis of essential cellular components. Several pathways of iron metabolism are modified according to systemic glucose levels. The rationale of the current study is to investigate the influence of uncontrolled diabetes on iron metabolism. A total of 130 samples from 3 groups of HbA1c levels (>6 nondiabetic, 6-8 diabetic, >8-uncontrolled DM) were studied for the correlation pattern of iron with other variables. ANOVA and student's t-test has been performed to reveal the correlation between serum free iron levels and other variables with DM. Serum iron has shown to be depleted significantly (F=6.92, P=0.001) along with percentage saturation (F=11.7, P=0.0001) with increase in diabetic severity. Serum ferritin levels were depleted as expected. However, there was no significant decrease in controlled DM and uncontrolled samples (t=1.93, P=0.05). Total Iron Binding Capacity was found to be significantly increased in uncontrolled DM samples. Abnormal transferrin glycosylation was observed in subjects very high TIBC levels. To conclude, uncontrolled diabetes affects the glycosylation of transferring also thus perturbating iron metabolism. This study emphasizes the need to minitor iron, Ferritin and TIBC levels in subjects with uncontrolled diabetes.

P-070

Therapeutic Effect of Yoga on Improving Wellbeing and Quality of Life in Type 2 Diabetics

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Yoga is a mind-body practice that positively impacts health. Chronic illness has several consequences that can affect wellbeing and quality of life. Stress associated disorders affecting the working skills and lifestyle management. Patients positive outlook improve their recovery. Study aim is to compare the benefits of yoga on Quality of life, Psychosocial factors in patients of type 2 Diabetes mellitus with non yoga diabetic subjects. A hospital based prospective randomized trial was conducted on 60 patients of confirmed cases of type 2 Diabetes mellitus (age group 30-65 of either gender). Subjects were randomized in two groups viz. Group 1- Patients of type 2 Diabetes mellitus without intervention of Yoga (n=30) which acted as control and Group 2- Patients of type 2 Diabetes mellitus with intervention of Yoga (n=30). (Approx.40 min yoga pattern, minimum five times a week over a period of six months). Anthropometric parameters were noted and serum glucose (Fating & Post Prandial), HbA1c were measured on Fully automated analyzer. Questionnaire for Psychosocial risk factors (Inter heart study) and Quality of life (SF-36) was filled by participants of the study. Psychosocial stress was assessed by questionnaire about stress at work and at home, financial stress and major life events in the past years. Statistical Analysis was done using student "t"-test. Yoga experienced group had significantly increased mental wellbeing, personal control, psychological and environmental quality of life compared with non-yoga group ($p<.05$). A significant decrease in blood glucose, HbA1c levels were observed in diabetics of yoga group as compared to diabetics of non-yoga group ($p<.001$). Results indicate that Yoga can be potentially beneficial for improves glycemic control, preventing and treating anxiety, insomnia, depression and improving the quality of life in subjects of T2DM.

P-071

To Study the Association of Tumour Necrosis Factor-α and Anthropometric Marker in Cigarette Smokers

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Cigarette smoking (CS) and obesity are major public health challenges and the prevalence of both is increasing globally. Smoking increases the risk of cancer, respiratory and cardiovascular diseases, and is the leading preventable cause of death in developed & developing countries. Obesity is the fifth leading cause of death, globally, and accounts for 23% of ischemic heart disease. Body Mass Index (BMI) & Waist-Hip Ratio (WHR) is positively associated with the number of pack years of smoking, and there is a dose-response relation between BMI, WHR and the number of cigarettes smoked. Tumor necrosis factor α (TNF-α), a powerful pro-inflammatory cytokine primarily produced by activated macrophages, is thought to play a critical role in the pathogenesis of proinflammatory mediators which lead to tissue damage and remodeling. The present study was carried out in the Department of Biochemistry, Government Medical College, Haldwani & Santosh Medical College & Hospital Ghaziabad. 284 Healthy Cigarette smokers in the age group of 18-60 years were included in the study. The analysis was carried out using the SPSS 19.0.2 program for windows. Unpaired "t" test was used to analyze all the data for statistical significance. Mean TNF-α, BMI & WHR level in cigarette smokers were (69.49±4.12, 24.07±1.62 & 0.926±.029) as compared to those nonsmokers (22.21±1.31, 22.99±2.09 & 0.859±.027). The mean TNF-α, BMI & WHR level were markedly raised in cigarette smokers as compared to those nonsmokers. These differences were found to be markedly significant ($p < 0.05$). In conclusion, we found that heavy smoking could induce a significant increase in BMI, WHR & serum TNF-α levels, suggesting the imbalance between the proinflammatory and anti-inflammatory factors.

P-072

Role of Serum beta 2 Micro Globulin and Urinary Albumin Creatinine Ratio in Early Stages of Chronic Kidney Disease

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Chronic diseases are a leading cause of morbidity and mortality in India and other low and middle-income countries. The Chronic Kidney Disease (CKD) burden is increasing rapidly worldwide. CKD typically evolves over many years, with a long latent period when the disease is clinically silent. Most of the commonly used parameters of renal function are able to detect kidney disease when GFR has already fallen substantially and so early detection of patients of CKD is still a challenge. This study was therefore planned to assess the utility of serum β_2Microglobulin (β_2M) and Urinary Albumin Creatinine Ratio (UACR) for early detection of patients of CKD. Objectives of the study was to measure serum β_2M and UACR in patients at high risk of CKD and to correlate serum ?2M and UACR with eGFR in all study subjects. A cross sectional observational study was carried out in clinical chemistry laboratory of S.S.G Hospital & Medical College Vadodara, from January to June 2017. In this period 90 patients were included in the study. In all these cases, serum β_2M, albumin and creatinine were measured. UACR and eGFR were calculated. On the basis of UACR and eGFR study participants were divided into 3 major groups (No CKD, Early CKD and Advanced CKD).The median levels of serum β_2M and UACR in early and advanced stage of CKD were 3.98 µg/ml, 7.49 µg/ml and 47.41, 71.9 respectively. Although in No CKD group creatinine was within the normal range (0.43-0.88 mg/dl), serum β_2M was on the higher side (1.36-2.78 µg/ml). The correlation of eGFR with serum β_2M (r=-0.762) was significantly better than with UACR (r= -0.578) in early stages of CKD. To conclude, Serum β_2M may be considered as a better predictor and promising marker for assessing glomerular function in adults even in no CKD group.

P-073

Assessment of Effect of Levothyroxine Replacement Therapy on Lipid Profile Parameters in Patients with Subclinical Hypothyroidism, A Follow up Study

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Subclinical hypothyroidism (SCH) represents a condition of mild to moderate thyroid failure characterized by normal levels of thyroxine (T4) and triiodothyronine (T3), with mildly elevated concentration of thyroid stimulating hormone (TSH) with or without clinical symptoms. Various studies have shown inconsistent results concerning the derangements of lipid profile parameters in SCH , also there is no consensus on whether substitution of levothyroxine (L-t4) has beneficial effects on serum lipid profile. This study was therefore planned to evaluate the effects of hypothyroidism on lipid profile and to study the effects of L-t4 treatment on thyroid and lipid parameters in area around kumaon region. This study included a group of 100 newly diagnosed SCH patients and 75 age & sex matched euthyroid controls. Thyroid function tests (T3, T4, TSH) and lipid profile parameters (Total cholesterol (TC), Low density lipoprotein cholesterol (LDLc), Triglyceride, High density lipoprotein cholesterol (HDLc) were estimated in all subjects after an overnight fast of 12 hours. Out of 100 SCH patients 58 selected patients were given L-t4 treatment and were followed up after 3 months with repeat thyroid and lipid profile tests. Our results showed significantly raised levels of TC, LDLc, TG in SCH patients when compared with euthyroid controls. HDLc levels were also raised in SCH patients but insignificant statistically. After thyroxine replacement therapy for 3 months , there was a significant decrease in levels of TC and LDLc. The mean levels of TG and HDLc were also reduced after L-t4 therapy but insignificant statistically. This study demonstrated the subclinical hypothyroidism has adverse effects on lipid profile parameters. The restoration of euthyroidism with L-thyroxine therapy effectively reduces reduces atherogenic lipids and thus may reduce the risk of cardiovascular complications.

P-074

Analysis of Laboratory Sample Rejections in the Pre-analytical Stage at an Oncology Centre

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Clinical laboratories play a crucial role in the diagnosis and management of patients. There are some of the key indicators of errors that can help & identify potential improvements in patient safety during pre-analytical phase in clinical laboratories. Clot was found to be the major cause of rejection of samples, followed by insufficient sample volume, patients clinical history was not provided, improperly labelled samples, samples collected in expired vacutainers, samples received without requisition, unlabelled samples & haemolysed samples. Errors in clinical laboratories have a great impact on safety and care of patients. The pre-analytical phase is responsible for about 70% of errors. Quality indicators in the clinical laboratory provide a useful tool for continuous improvement of laboratory services. In this study, we aimed to evaluate the sample rejection ratios according to the types of pre-analytical errors. A retrospective, intervention and prospective

analysis of the samples rejected from the total samples received in our laboratories, during a period from January 2017 to June 2019 was undertaken. Out of 216,631 samples received during January 2017-June 2019, 318 samples (0.15%) were rejected. The most common reasons for rejection is clotted blood samples (57.14%), followed by improperly labelled samples (14.28%), haemolysed samples (11.42%), insufficient sample volume (8.57%), Samples without requisition (5.71%) & Samples in expired vacutainers (2.85%). This study has shown that the most frequent causes of pre-analytical errors are clotted samples, improperly labelled samples, haemolysed samples & samples with insufficient volume.

P-075

Elevated Matrixmetalloproteinase-9 is Associated with Maniac Symptoms in Bipolar Disorder

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Bipolar disorder is a highly prevalent psychiatry disorder associated with suicidal complications. Among various mechanisms that contribute to complications in BD patients, synaptic plasticity plays a key role. Matrix metalloproteinases (MMPs) are extracellular proteases that play a role in hippocampal synaptic physiology, long term potentiation and memory. The objective of the study was to investigate whether MMP-9 is associated with severity of bipolar disorder. 80 bipolar disorder patients on treatment who were presented maniac or depressive symptoms and 80 age and gender matched controls were enrolled in the study. MMP-9 was estimated in both the groups. The patients will be assessed for presence of depressive symptoms using Hamilton Depression Rating Scale (HDRS) and manic symptoms using Young's Mania Rating Scale (YMRS) scales. MMP-9 was significantly increased ($p < 0.05$) in bipolar disorder patients compared to controls. Also we found significant association of MMP-9 with YMRS scale ($r = 0.253$, $p = 0.024$) in patients with bipolar disorder. There was no significant association of MMP-9 with HDRS scale, age and duration of illness. We conclude that MMP-9 is associated with maniac symptoms in bipolar disorder.

P-076

Association of Serum Adiponectin Level with Atherosclerotic Risk Factors in Obese Individuals

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Adiponectin secreted from adipocytes plays important role in energy homeostasis and lipid metabolism, whose level reduces in obesity and its deficiency results in higher incidence of atherosclerosis. It has been proposed as a marker of coronary heart diseases in obese adults. Adiponectin is shown to have insulin sensitizing, anti-atherogenic and anti-inflammatory properties. Obesity classified in terms of BMI as normal weight (18.5-22.9 kg/m^2), overweight (23-24.9 kg/m^2) and obese (>25 kg/m^2). Aim of the study is to compare the level of adiponectin in obese individuals with non-obese controls and to find out the correlation of adiponectin with BMI, WHR, Lipid Profile and Fasting Glucose and insulin level. Present study included 60 subjects. Equal number of obese individuals and age and sex matched controls in the age group of 20-45 years were studied. Subjects having chronic diseases, on any medications, alcoholics and smokers are excluded. Fasting blood sugar, Serum Lipid Profile, Urea, Creatinine are quantitated in TOSHIBA TBA 120 FR. Fasting Insulin is measured in ROCHE Cobas e411. Serum adiponectin is assayed by Systronic ELISA Reader 641. It is a case-control study. Statistical analysis was done using SPSS version 16 and correlation by Pearson correlation test. Serum adiponectin in cases (2.34±0.79) found to be significantly lower than controls (8.28±3.04). BMI, WHR, Insulin level, FBS, TCHO, TG, LDL are significantly higher and HDL is lower in cases than non-obese controls. We found statistically significant negative correlation of serum adiponectin with lipid Profile [i.e. TCHO ($r=-0.9377$, $p<0.00001$), TG ($r = -0.966$, $p<0.00001$) LDL ($r=-0.877$, $p<0.00001$)], FBS ($r=-0.7533$, $p<0.00001$), BMI ($r=-0.8126$, $p<0.00001$) and WHR ($r=-0.8806$, $p<0.00001$), fasting insulin ($r=-0.9103$, $p<0.0001$) and positive correlation with HDL ($r=0.9565$, $p<0.00001$). Adiponectin level reduced in obese people and its association with atherosclerotic risk factors increases the risk of coronary heart disease.

P-077

Effectiveness of CSF ADA as A Marker in Diagnosis of Tubercular Meningitis (TBM)

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Among the patients with meningitis, tubercular meningitis is an important cause of morbidity and mortality in India and other developing countries. The diagnosis of TBM highly depends upon microscopic methods and CSF culture for mycobacterium tuberculosis which takes about 6-8 weeks. A simple and cost-effective test for the diagnosis of tuberculous meningitis patients would help to make diagnosis easier. CSF ADA levels measurement can be utilized as rapid and inexpensive test to differentiate TBM from other causes of meningitis. The present study is conducted to check the usefulness of the current cut off point of 8 U/L of CSF ADA as a marker of TDM. 50 patients between the age of 6-60 years attended hospital with symptoms and sign of meningitis are selected. They are divided into two groups; Patient with Tubercular meningitis and patients with Non-tubercular meningitis base on the clinical findings. CSF specimens of each of the patient obtained by lumbar puncture performed by a trained medical officer. ADA Assay will be carried out in the biochemistry laboratory by NonGiustic Enzymatic method (kinetic). Study consists of total 50 subjects. Out of which male comprise of 33 (66%) and 17 (34%) female. In this maximum numbers of subjects 20 were in age group 46-60. Out of the total 50 patients, 19 have tubercular meningitis (TBM) and 31 have Non-tubercular meningitis. Of this, 19 TBM patients, 14 had CSF ADA activity at or above 8 U/L while 5 had below. Out of 31 Non-tubercular patients only 4 have ADA level above 8 U/L and 27 had below. We concluded that CSF-ADA activity was higher in patients with tubercular meningitis as compare to pyogenic and viral meningitis. So, it can be used as simple rapid inexpensive test for differentiating tubercular from pyogenic and viral meningitis.

P-078

Candida Parapsilosis Meningitis in A Patient with AIDS. Report of A Case and Review of the Literature

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We report on a 35-year-old male that was admitted due to headache and recurrent high fever status, associated with oropharyngeal candidiasis as the first manifestation of AIDS. On admission, imaging studies showed no evidence of fungal origin, and the first diagnostic hypotheses were cryptococcal meningitis or tuberculosis of the central nervous system because of the prevalence of these diseases. Microscopic examination of the first 6 daily cerebrospinal fluid (CSF) cultures on Sabouraud medium revealed creamy and smooth white yeast colonies, and confirmation of Candida parapsilosis was based on filamentation test, chlamydoconidia production and biochemical test (auxacolor). The treatment was initiated with fluconazole but the patient died while receiving treatment. This case underscores the need for suspicion of Candida parapsilosis as a cause of meningitis in HIV patients, on the other hand, speed to diagnosis is a key risk factor in patient outcomes.

P-079

To Establish Serum Gamma Glutamate Transferase and Serum Ferritin as a Risk Marker for Metabolic Syndrome

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The metabolic syndrome is a constellation consists of an atherogenic dyslipidemia (i.e. elevated triglycerides, low high-density lipoprotein cholesterol (HDL-C)), the elevation of blood pressure and glucose, is a prothrombotic and proinflammatory state. Gamma-glutamyl transferase (GGT) and ferritin participate in common pathophysiological processes, including oxidative stress and lipid peroxidation, which are important to the pathogenesis and development of insulin resistance and Metabolic syndrome. The actions of GGT and Ferritin can help in predicting MS in on risk people. Besides these are commonly done in routine laboratories which will make these parameters a cheaper alternative. This study was aimed to determine the relationship between GGT and ferritin and the risk of developing Metabolic syndrome. A prospective study with 60 cases of Metabolic Syndrome defined according to NCEP/ATP III guidelines and 60 clinically healthy controls, was conducted. Serum GGT was estimated with Mod IFCC method and serum Ferritin was estimated with CLIA. The median and interquartile range for Ferritin and GGT were 81 (45-133) ng/dL and 26 (20- 41) IU/L respectively in cases whereas in control group these values were 23 (17-48.4) ng/dL and 16 (11-19) IU/L. A positive and significant correlation with r-value of 0.271 (p value-0.038) was found between Ferritin and Triglycerides. Positive correlation was seen between Gamma-Glutamatetransferase and Ferritin in cases with p-value of 0.057. Intergroup comparison gave a significant association for Gamma-Glutamatetransferase and Ferritin with a p-value of ≤.001. The median±interquartile range observed in cases was higher than the controls suggesting inflammatory load. The positive correlation between Ferritin and Triglyceride- a diagnostic marker of Metabolic Syndrome, indicates a possible association. Further progress is going on this study, which will yield a more conclusive picture.

P-080

Evaluation of Serum Uric Acid levels in Diabetic Individuals

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Serum uric acid is the metabolic end product of purine. Various studies have shown hyperuricemia as a risk factor of metabolic syndrome, cardiovascular diseases, and diabetes. Studies evaluating relationship between uric acid and glucose had given the inconsistent results. This study thus aims to assess the value of Uric acid in Diabetes Mellitus patients and verify an existing relationship. A retrospective analysis of reports of 55 patients were reviewed and analyzed. Among these 31 were healthy controls and 24 were cases of Diabetes Mellitus. Glucose and Uric acid of these patients had been estimated by Glucose-Oxidase-Peroxidase and Uricase methods respectively. Mean±SD in diabetic and non-diabetic were 5.17±1.32 mg/dL and 5.32±1.84 mg/dL respectively showing no significant difference. A similar mean in both the study groups suggest no significant relationship between uric acid and FBS in diabetic individuals. However, a wider sample selection will give a better understanding and more comprehensive results.

P-081

Monitoring of Thiopurine Metabolite Levels based on Biochemical and Haematological Indices in Inflammatory Bowel Disease (IBD)

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IBD is characterised by chronic inflammation of the digestive tract and extended in terms of ulcerative colitis and Crohn's disease. Thiopurine drug is used for remission of IBD with the combination of immunosuppressant drugs. The treatment outcome of thiopurine metabolites are essentially required to measure in RBCs as therapeutic drug monitoring of thiopurine erythrocyte levels is not available in country. The aim of this study was to determine 6-thioguanine and 6-methylmercaptopurine levels in IBD patients to predict non-adherence in patients treated with azathioprine. 111 IBD patients were recruited from the IBD clinic of Gastroenterology AIIMS, New Delhi those on Azathioprine therapy and samples were collected at baseline, 3 months and 6 months. Out of 111 subjects only 38 patients were matched with all stages (at baseline, 3 months and 6 months) and rest collected samples were not matched due to dropout by several factors. The diagnosis of IBD was made on presence of clinical manifestations, endoscopic features and histologic features. 6-TG and 6-MMP levels, hematologic indices, Vitamin B12, Ferritin, Iron, Folate and clinical biochemistry were done by HPLC, haematology, chemiluminescence and multichannel chemistry analyser respectively. Hct, MCV, MCH, MPV and HB were in increased from baseline to 6 months while MCHC, RDW were decreased. Uric acid was significantly higher ($p<0.001$). Folate levels were significantly lowered while vitamin B12, Ferritin and Iron levels were suboptimal. 6-TG and 6-MMP levels in RBCs were significantly higher and the ratio was more than 1.0. This study demonstrated that patients with elevated ratio of 6TG and 6MMP could be deleterious to mucosal ulceration, loss of colonic architecture, fibrinous exudate. Decreased levels of Folate, increased levels of uric acid and defect in haematological indices could be suggestive intracellular defect and leading to myelotoxicity, leucocytopenia, thrombocytopenia. Therefore 6-TG and 6-MMP could be an important tool for therapy.

P-082

An Atypical Case of Osteoporotic Compression Fracture Mimicking Multiple Myeloma

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Threatened multiple myeloma (MM) presents with varied phenotypes, multiple spinal osteoporotic fractures with constitutional features suggest malignancy. Apparent IgG, IgA & IgM myeloma in serum electrophoresis along with the presence light & heavy chains tracks the diagnosis towards MM. Prevalence of Kappa & Lambda light chains or any of the other types of heavy chains is not uncommon. An insidious case presenting with multiple spinal fractures along with very high level of beta 2-microglobulin, to the tune of 5450 ng/ml with higher Kappa & Lambda light chains in serum along with classical clinical features warrants for myeloma but it makes the diagnosis frippery and creates a clinical dilemma when the case goes for dramatic recovery. A 62 year old, chronic type 2 diabetic, male patient presented with complaints of backache since 6 months. He developed pain in the back, insidious and gradually progressive. After primary investigations he was diagnosed with partial collapse of D5, D6, D12, L1, L4 vertebrae. T score -3.5, absence of M-Spike, high serum IgG, high urinary Kappa & Lambda light chains, high beta 2-microglobulin. Bone marrow aspiration from right Posterior Superior Iliac Spine (PSIS) showed normal cytology/non-specific, CT guided biopsy from D12 shows no evidence of any granulomatous/neoplastic lesion. Oftentimes, osteoporotic fracture presents with severe pain, no pain resolution with empirical treatment necessitates the point of care

investigations and comprehensive work-up of the patient. This patient was evaluated for suspicion of vertebral collapse due to malignancy but all relevant investigations turned up negative and there was no evidence of any malignancy. Patient was diagnosed as a case of Osteoporosis and treatment was started, conservative, physiotherapy and mobilization. This case became curious because of its non-sine qua non findings and dramatic resolution advocating the diagnosis for uncommon variant of Beta 2-Microglobinemia with Light Chain Gammopathy.

P-083

Association of Serum Osteocalcin in Metabolic Syndrome and Insulin Resistance

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Metabolic Syndrome is an important public health burden associated with increased visceral fat which leads to insulin resistance and five-fold higher risk of developing type2 DM. Recent studies described that osteoblasts produce osteocalcin which increases insulin secretion and adiponectin production resulting in insulin sensitivity. To evaluate whether serum osteocalcin is associated with insulin resistance and metabolic syndrome. The ongoing case control study was carried out in the department of biochemistry at MKCG Medical College, Berhampur. Thirty cases between the age group of 20 - 45 years meeting the criteria of metabolic syndrome and equal no of age and sex matched healthy controls were included in the study. Individuals with any systemic illness or on any kind of medications were excluded from the study. Informed and written consents were obtained from each individual. Ethical clearance was taken. Fasting blood sugar, lipid profile was measured by standard procedures. Serum osteocalcin was estimated by sandwich ELISA and serum insulin was estimated in Roche eCobas 311. Insulin resistance was calculated by Homeostasis Model Assessment of Insulin Resistance. Serum osteocalcin was found to be lower in cases as compared to controls with (mean±SD) (7.4013±5.5001 ng/mL and 18.4663±6.9483 ng/mL respectively). Osteocalcin was also found to be significantly negatively correlated with HOMA-IR, waist circumference, triglyceride and fasting blood sugar in cases with ($r=-0.363$, $p<0.05$), ($r=-0.822$, $p<0.05$), |($r=-0.367$, $p<0.05$), ($r=-0.506$, $p<0.01$) respectively and also in controls with ($r=-0.387$, $p<0.05$), ($r=-0.219$, $p=0.245$), ($r=-0.562$, $p<0.01$), ($r=-0.455$, $p<0.05$) respectively. Serum osteocalcin being negatively correlated with insulin resistance can be used as a biochemical marker for metabolic syndrome.

P-084

Follow up Study of Renal Function of Term Neonates Surviving Acute Kidney Injury

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It had been a consensus for a long period that in paediatric patients Acute Kidney Injury (AKI) is followed by complete renal recovery. Better health care facilities had improved neonatal survival rate from two important risk factors of AKI i.e. Birth Asphyxia and Neonatal Sepsis. This in turn opens a bigger window to look into patients of AKI and follow up at tertiary centre. We selected full term newborn admitted in NICU, having high baseline S. creatinine after 48 hrs of birth. We further studied for serial Urinary Output & S. Creatinine. Patients were classified in *oliguric* (UO <0.7 mL/kg/hr for 24 hrs) & *non-oliguric*. 61 Term Sick Neonates were diagnosed of developing AKI as per neonatal-RIFLE criteria. FENa of every patient was calculated to define pre-renal (<2.5) or intrinsic cause (>2.5) of renal pathology. USG KUB ruled out any congenital pathology. Discharged patients were followed up on 14th week and 6 month of age for growth parameters, BP, Urinalysis, Spot protein/Cr. ratio in urine, Venus Blood Gas Analysis, KFT & USG KUB.AKI developed as a complication of primary disorder as Neonatal Sepsis (54.1%), Birth Asphyxia (34.4%), Nephrotoxic Drug (4.9%) & combined (6.5%). All patients had pre-renal insult, out of which 77.1% patients were oliguric in early stage. However in subgroup of patients with neonatal sepsis, significantly 97% patients were oliguric. Mean S.Creatinine level was 2.7 mg/dL (1.8-4.6). Deranged KFT were present in cases of birth asphyxia (61.9%) and Neonatal sepsis (51.5%). On follow up at 14 weeks, 3.26% of these neonates show growth retardation, which was not present at 6 month of age. Sample size is small, however significant proportion of patients of neonatal sepsis remains at high risk of developing Chronic Kidney Disease. Long term follow up for survivors of AKI is recommended.

P-085

Altered Glycans in Hepatitis Related Liver Fibrosis

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The serum proteins are synthesized in the liver and undergo posttranslational modification, glycosylation. Glycosylation is important for proper protein folding, host-pathogen interaction, cell

communication and signal transduction. During the progression of liver disease, the sugars such as fucose, galactose and sialic acid are altered which in turn leads to altered glycosylation of proteins. This pilot study included healthy controls (n=20) and biopsy proven patients with liver fibrosis and cirrhosis related to hepatitis virus (n=35). Serum and urine samples were used for the colorimetric estimation of fucose, galactose, sialic acid and galactosamine. The liver fibrosis group showed a significant decrease in serum free fucose, serum free galactose, total sialic acid and total galactosamine compared to the control group. There was no significant difference noted in urine galactosamine and urine hexose sugars fucose and galactose between the control and the fibrosis group. However, a significant increase in urine sialic acid content was noted in fibrosis group compared to the controls. Our study shows that in chronic viral hepatitis changes in the concentration of hexose sugars and sialic acid can lead to disturbance in the glycosylation of serum proteins.

P-086

Critical Values: Report and Evaluation

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Critical values, misnamed panic values, are results that express a medical situation that may endanger the patient's life if not properly and timely intervened, hence the need to be informed immediately after being detected to the doctor who requested them. The laboratory accrediting agencies have made critical value reporting part of the requirements for accreditation. Furthermore, the immediate notification of a critical value as a special requisite has been recognized and implemented worldwide through the International Organization for Standardization (ISO) 15189:2012, and has been adopted as a standard of Good Laboratory Practice. The aim of the study is to determine the situation of our laboratory for the notification of critical values. Descriptive study based on critical values in our laboratory during a period of one year. Data were collected using LIS Modulab (Werfen®) software. In our laboratory; an action plan was designed in the presence of critical values. This plan is based on the elaboration of a list with critical values in consensus with clinicians, verification and identification of the result and a rapid notification to the requesting doctor or the person responsible for the patient. During the period from January to December 2018, our clinical laboratory performed approximately 108.447 emergency tests and the number of critical values detected were 1.276 (1.2%). The parameter with the highest percentage of all critical values was hemoglobin (32%), followed by potassium ion (31%), glucose (13%), sodium ion (12%), chlorine ion (6%), calcium (II) (4%), phosphorus (1%) and magnesium (II) (1%). Although these data demonstrate that the protocol is working, it is necessary to optimize the detection of critical values.

P-087

Serum Albumin as a Prognostic Marker of Clinical Outcome in Critically Ill Patients.

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Albumin is a major component of plasma protein and is required to maintain the oncotic pressure, microvascular permeability, acid base balance, prevention of platelet aggregation, and acts as a ligand binding agent. It is a useful marker to assess the nutritional status of an individual. Critically ill patients are at an increased risk of hypoalbuminemia due to compromised nutrition superimposed on the underlying pathology. Intensivists often feel the need of a reliable and cost effective biochemical marker which could predict the outcome in critically ill patients. This prospective observational study was aimed to explore the utility of estimating serum albumin levels in critically ill patients as a marker of clinical outcome in a tertiary care hospital of North India. Serum albumin levels were measured in 93 critically ill patients admitted in intensive care unit. The patients were categorised into hypoalbuminemic and normoalbuminemic group depending on serum albumin levels. The survival rate in the study group was found to be 66.6%. Hypoalbuminemia was found to be present in 62.4% of patients. There was no statistically significant difference in mean albumin levels between survivors and nonsurvivors. The mean duration of hospital stay was also not found to be dependent on albumin levels in survivors. The use of inotropes was significantly higher in hypoalbuminemic patients as compared to normoalbuminemic patients. The need for mechanical ventilation was also found to be higher in hypoalbuminemic patients as compared to normoalbuminemic, although, the difference was not statistically significant. Measurement of serum albumin at the time of admission might help in identification of patients requiring use of inotropes and mechanical ventilation. Clinical randomized trials and evidence based guidelines are required regarding the supplementation regimen.

P-088

The RAGE Gene Polymorphism Modulate Anti-proteinuric Response of ACEI Therapy in Diabetic Nephropathy Patients: One Year Follow up Study

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Advanced glycation end product (AGE) and receptor for AGE (RAGE) axis and renin-angiotensin-aldosterone system (RAAS) play an important role in the development of diabetic nephropathy (DN). Angiotensin converting enzyme inhibitors (ACEIs) are commonly used for the reno-protection in DN patients. These drugs also reduces the progression of diabetic nephropathy by inhibition of AGE formation and activation of RAGE gene. However, the impact of polymorphisms of RAGE gene on the anti-proteinuric action of these drugs in diabetic nephropathy still remains obscure. The aim of this study is to evaluate the role of RAGE gene polymorphisms such as; -374T/A, -429T/C and Gly82Ser on the anti-proteinuric response of ACEI therapy in DN patients. Total 203 type 2 diabetes mellitus patients with nephropathy were enrolled and treated with ACEI (ramipril; 5 mg/day) and followed-up for 12 months. Serum and urine creatinine were measured by alkaline picrate Jaffee´s kinetic method. Urine albumin was estimated by turbidometric method by using nephelometer. Albumin/creatinine ratio (ACR) was calculated by using urine albumin and creatinine. The clinical response was defined as decrease in urinary ACR ≥30% from the baseline values after treatement with ACE inhibitor. Genotyping of RAGE gene polymorphisms were performed by PCR-RFLP method. Significant reduction in urinary ACR was observed after twelve months treatment with ACEI irrespective of whether DN patients were micro-albuminuric or macro-albuminuric at the time of enrolment. However, macro-albuminuric patients (75%) showed better response to ACEI therapy. Reduction in urinary ACR level was found independent of genotypes of RAGE gene polymorphism. In conclusion, ACEI therapy reduced ≥30% urinary ACR in 73% of DN patients (responders) after 12 months follow up. Polymorphisms of RAGE gene; -374T/A and -429T/C did not alter the ACE inhibitor-mediated anti-proteinuric efficacy in diabetic nephropathy patients; however, patients having SS genotype of Gly82Ser SNP were poorly responsive to ACEI therapy

P-089

Study of Changes in Circulating Adipo-Cytokines Due to Bariatric Surgery Induced Weight Loss and their Association with Co-morbidities in Obesity

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Obesity and associated metabolic pathologies are most common and detrimental metabolic diseases of modern world. Obesity has been shown to be associated with a chronic inflammation and bariatric surgery has come up as one of the most effective weight loss procedure. We aim to characterize the effect of surgically induced weight loss in terms of adipo-cytokines and inflammatory status of the obese individuals. To estimate and compare the serum levels of Adiponectin, Resistin, Visfatin, IL-6, IL-1β, TNF-α, MCP-1, CRP, IL-8 and IL-10, in both obese and non-obese individuals and to assess the changes in the serum levels of the same markers pre- and post-operatively in the obese group of individuals who have undergone bariatric surgery. Obese individuals (BMI ≥ 30; n=30) aged 18-60 years and their age matched controls (BMI < 30; n=21) were included. The serum cytokine analysis in both groups was done using Luminex multiplex bead array system with xMAP technology. Same parameters were again assessed post-operatively in obese after bariatric surgery (6-12 month follow up). The results were analyzed statistically by Student's t-test. These results were also correlated with HOMA-IR. Levels of Adiponectin, Visfatin and IL-6 were lower in the obese group as compared to controls, out of which Visfatin levels were significantly decreased ($P = 0.04$). Other inflammatory markers were raised in obese group but were not significant. IL-1β was significantly raised ($P = 0.0281$) in follow up analysis. A very significant improvement was seen in the follow up HOMA-IR ($P = 0.0002$) and was found to be correlated with improved Adiponectin levels ($r = 0.8035$; $P = 0.0295$), which were also significantly correlated with the improved fasting insulin levels ($r = 0.8454$; $P = 0.0166$). Low grade adipose tissue inflammation was improved by surgically induced weight loss and is also reflected in improved insulin sensitivity.

P-090

A Study to Assess the Usefulness of Serum Cystatin C as an Early Marker for Acute Kidney Injury in ICU Patients

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Acute renal failure is a very common complication occurring in the medical intensive care, causing significant morbidity and mortality. Still serum Creatinine is the marker of choice despite having several short comings. In this study we tried to assess the usefulness of serum Cystatin C as an early marker of acute kidney injury. We conducted a clinical cross sectional on 30 patients admitted in the medical ICU of Bowring & Lady Curzon Hospital & Victoria hospital, BMCRI Bangalore. We observed that Cystatin C in cases was estimated to be 744.58± 321.00 ng/ml. Data showed that 50% cases had abnormal values for serum Cystatin C. ROC analysis with AUC showed that Cystatin C with a cut off > 40 had sensitivity and specificity of 100% and AUC 1.000 ($p<0.001$). The creatinine clearance as estimated by both MDRD and CKD EPI formulae were found to be normal in cases. In conclusion, higher serum Cystatin C values observed in cases in spite of normal creatinine levels indicate that the elevation of serum Cystatin C precedes the elevation of creatinine. The higher sensitivity and specificity of serum Cystatin C supports the assumption that serum Cystatin C can be used as a reliable marker for early identification of patients at risk of AKI.

P-091

A Cross-sectional Study of Epidemiology and Biochemical Profile of Primary Hypertension Among Young Adults

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Hypertension is the most common non-communicable disease of pandemic trend. Primary hypertension accounts for 90% cases of all hypertensions. It is a well-known risk factor for various cardiovascular and renal events resulting from interaction of several genetic and environmental factors. A rising trend in young adults more so with family history calls for early intervention to reduce morbidity and mortality. The main objective was to find out the prevalence of hypertension having family history and assess their biochemical profile to evaluate renal, lipid and electrolyte parameters to create awareness regarding cause and consequences of hypertension. In this case control study including 300 young students of medical college in the age group of 18-25 years of either sex was enrolled and divided into 2 groups based on presence or absence of family history of hypertension in their first-degree relatives. Those with hypertension as per WHO guidelines were included along with their demographic variables and serum was obtained for estimation of sugar, Na^+, K^+, lipid profile, urea, creatinine and uric acid using standard procedures in Toshiba TBA 120 FR. 32.3% of students are having a family history of hypertension, out of which 41.2% are having hypertension. A significantly higher level of BMI, SBP, DBP, urea, creatinine, uric acid, K^+ were found compared to healthy controls without any family history of hypertension. BMI, lipid profile, uric acid & creatinine are significantly correlated with BP. This study shows altered renal, lipid and electrolyte status compared to healthy controls. So, raising awareness for early intervention for life style modification can reduce the morbidity and mortality due to hypertension.

P-092

A Study of Insulin Resistance, B -Cell Function and Lipid Profile in Type II Diabetes Mellitus

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Type 2 diabetes is not a disease but rather a heterogenous group of multifactorial syndromes characterized by elevated fasting blood glucose caused by relative or absolute deficiency of insulin. It is associated with a cluster of interrelated plasma glucose, lipid and lipoprotein abnormalities, including a predominance of total cholesterol, elevated triglycerides and reduced HDL. This study was conducted to evaluate the effect of altered lipid profiles of the established cases of diabetes mellitus patients on their insulin resistance states as depicted by their HOMA-IR and HOMA-β scores. This study is a cross-sectional analysis of the samples collected at New Medical College, Kota. One hundred and four subjects ≥40 years were selected from the elderly population, which were established cases of type 2 DM. The same number of healthy non-diabetic subjects were evaluated. When the diabetic group and the control group were compared, significant statistical difference was observed in fasting serum glucose, cholesterol, triglycerides, VLDL, LDL HbA1c, serum insulin, HOMA-IR and HOMA-β scores.Above findings reflect that there is a positive correlation between diabetes and altered lipid profile, serum insulin, Hb1Ac and HOMA-IR.There is a high prevalence of dyslipidemia, which might be playing a major role in the development of cardiovascular diseases and cerebrovascular accidents among diabetic patients. Efforts to achieve lifestyle changes, such as weight reduction, physical exercise and smoking cessation should be encouraged and initiated first and then followed

by medication with lipid-lowering drugs prescribed in evidence-based necessary conditions.

P-093

A Clinical Study of Serum hs-CRP and Uric Acid in Patients with Psoriasis in a Tertiary Care Hospital

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Psoriasis is a chronic skin disease. CRP has special importance for psoriasis due to its relation with cytokines. Present study determines the CRP by using highly sensitive method (hs-CRP) and its correlation with severity of disease, to assess the relationship between hyperuricemia and psoriasis. This case control study has been conducted in the dermatology and biochemistry lab of Osmania general hospital, Hyderabad. 50 patients of 20-50 years of age group diagnosed with psoriasis from skin and VD department form the test group. 50 age, sex matched healthy volunteers forms the control group. Cases were graded according to PASI into three categories - mild, moderate and severe. 5 ml of blood sample was collected with consent under aseptic precautions in a plain vacutainer. Estimation of hs-CRP was done by latex turbidimetry method using kit from ERBA. Serum uric acid is measured by uricase method on fully automated analyzer. Serum urea creatinine and SGPT is also measured to rule out involvement of liver and kidney disease. Data were expressed as mean ± SD. The statistical analysis was done using graph pad prism version 7.0. The mean value of uric acid in cases was (5.46 ± 1.5) (5.42 ± 2.2) and in controls (5.7 ± 0.57 mg/dl) (P value > 0.05) Mean value of hs-CRP in cases (13.6 ± 8.5) (75.19 ± 37.9) and in controls (1.44 ± 0.25) (P < 0.05). t-test and correlation coefficient done. P < 0.05 was considered significant. Serum uric acid level does not correlate with PASI but psoriasis patients with (PASI > 10) have higher mean serum hs-CRP than patients with (PASI < 10) and controls. Serum hs-CRP level correlate significantly with PASI and can be used as marker for assessing severity of disease.

P-094

Establishing Indian Population-Specific Vitamin D Thresholds Associated with Secondary Hyperparathyroidism Using Machine Learning Algorithm

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Parathyroid hormone (PTH) controls calcium and phosphate levels in blood and calcium in bones. Secondary hyperparathyroidism is usually due to insufficient amounts of calcium in blood, which may occur during Vitamin D deficiency, which lead to increased PTH production. The incidence of vitamin D deficiency is high in India despite the fact that it is a tropical country. In view of high prevalence of vitamin D deficiency in Indian population, we have aimed to establish cutoff value of Vitamin D where PTH levels start raising (secondary hyperparathyroidism) in healthy Indian population. We have enrolled a total of 1026 subjects (678 men and 348 women) in the age group of 50.7±16.5 years whose vitamin D levels were < 30 ng/ml. Intact parathyroid hormone (PTH) and Vitamin D total assays are performed on ADVIA Centaur® XP Immunoassay system using kits manufactured by Siemens Healthcare Diagnostics Inc., U.S.A. A total of 542 subjects (52.8%) had normal PTH levels despite vitamin D deficiency. Machine learning tools were used to deduce the threshold of Vitamin D associated with secondary hypothyroidism. This analysis revealed that the cut-off value of vitamin D is <6.3 ng/ml, which is associated with secondary hyperparathyroidism (PTH>155.4 ng/L). This study emphasizes the need to evaluate factors that contribute to the absence of PTH response in more than 50% of our population with either Vit D deficiency or insufficiency. As per the existing data, vitamin D <6.3 ng/ml might be associated with significant influence on bone markers due to secondary hyperparathyroidism.

P-095

Evaluation of Bone and Mineral Metabolites Following Renal Transplantation

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Renal replacement or kidney transplantation is the most recommended therapy for patients with end stage renal disease (ESRD). Advances in technology, surgical techniques and pharmacotherapy have increased the success rate of renal transplantation. However, various long- term side effects are reported post transplantation. A commonly reported complication is mineral and bone disorder (MBD). Conditions such as persistent hyperparathyroidism and the use of certain immunosuppressive agents have a deleterious effect on the post renal transplant bone. The present study aimed at evaluating various parameters of bone metabolism pre and post six months of renal transplant. Fifty patients diagnosed with ESRD opting for renal transplantation were selected for the study. Their blood sample were collected and analyzed for serum calcium, phosphorus, alkaline phosphatase (ALP), Parathyroid hormone (PTH) and 25 (OH) Vitamin D. All parameters were again evaluated six months post renal transplant. Results obtained were compared by applying paired t-test. Serum Vitamin D and calcium levels were observed to increase following renal transplant ($P < 0.0001$) whereas serum PTH, Phosphorus and ALP exhibited a significant fall following renal transplant. MBD following kidney transplantation is common and characterized by loss of bone volume and mineralization abnormalities often leading to low turnover bone disease. Although there are no well-established therapeutic approaches for management of MBD in renal transplant recipients, clinicians should continue individualizing therapy as needed. Abnormal bone and mineral metabolism is common in patients with kidney failure and often persists after successful kidney transplant. Parathyroid hormone levels decreased significantly during the first 6 months after transplant but not stabilized. Calcium tended to increase after transplant and then stabilize at the higher end of the normal range Phosphorus decreased rapidly to within or below normal levels after surgery.

P-096

Comparison of Iron Indices in Liver Disease According to Child Pugh Score

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Chronic liver disease (CLD) is a disorder involving impairment of liver function due to progressive destruction and simultaneous regeneration of the liver parenchyma. This leads to fibrosis and finally cirrhosis of liver. Various etiological factors are associated with Chronic liver disease such as Alcohol, Portal Hypertension, NASH, NAFLD, Autoimmune, Hepatitis B, C and others. To assess the severity of liver damage various scores are available. One such score is Child Turcotte-Pugh (CTP) score. Liver plays an important role in iron homeostasis. Iron indices are expected to be deranged during CLD. The present study aimed at comparing iron indices with CTP score and to explore the correlation of CTP score with iron indices. 300 patients diagnosed with CLD were enrolled in the study based on predefined inclusion and exclusion criteria. The selected patients were grouped as A, B & C based on their CTP score in relation to minimal (n=13), moderate (n=136), or worse (n=151) prognosis respectively. Blood samples were collected and analyzed for serum iron, TIBC, ferritin and transferrin saturation. It was observed that serum iron did not show a significant variation with progression of CTP score. However, serum TIBC, ferritin and transferrin saturation exhibited significant variation with progression of CTP score. Serum ferritin levels showed an increase with increased CTP score (P=0.000). However, serum TIBC exhibited a gradual fall (P=0.000). Evaluation of iron indices, specially ferritin and TIBC can be helpful in assessing the degree of liver tissue damage. Early detection of such prognostic factors may be helpful in timely and suitable management of the patient.

P-097

Association of Homocysteine Levels with Lipid Risk Factor in Patients with Vitiligo

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Vitiligo is an acquired, depigmenting skin disease with still unclear, multifactorial etiopathogenesis which not only affects the skin but it may also be connected with metabolic abnormalities, including glucose intolerance and lipid abnormalities, confirming to the systemic nature of the disease. Nutritional deficiencies as vitamin B12 and folate have been implicated in vitiligo that can

result in an increase in homocysteine levels. Further, an association between hyperlipidemia and hyper homocysteinemia has been suggested which is clinically important in management of vascular risk factors especially in diseases with metabolic abnormalities as in vitiligo. The present study was thus aimed to assess Hcy levels and lipid risk factors in vitiligo patients and to study their interrelationship to predict the cardiometabolic risk in vitiligo and its management. 54 were patients with generalized vitiligo and 54 were age and sex-matched healthy controls were enrolled. Patients were assessed for disease severity (VASI Score) and were evaluated for the lipid profile and serum Hcy levels. Significantly higher LDL-C levels (p=0.010), significantly lower HDL-C (p=0.003) and significantly higher LDL/HDL ratio (p=0.001) were observed in patients with vitiligo in comparison with the control group. The mean serum Hcy levels in vitiligo patients (18.76±10.02 mol/L) were significantly higher than of controls (10.04±5.34 μmol/L) (P=0.000). Serum Hcy levels showed a positive correlation with VASI score (p=0.000). No significant correlation was observed between serum Hcy levels and lipid profile. Higher Hcy levels in vitiligo patients may be a precipitating factor in the pathogenesis of vitiligo in predisposed individuals. The results of our study are also indicative of lipid disturbances in vitiligo reflecting some ongoing abnormal metabolic processes in patients with vitiligo. Therefore, we recommend routine estimation of homocysteine and lipid profile in vitiligo patients both of which should be regarded as significant contributing factors worth considering in the management of these patients.

P-098

A Study on Relationship of Serum GGT and Magnesium Level in Alcohol Dependence Syndrome

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A study on changes in Serum GGT and Magnesium level in Alcohol Dependence Syndrome. Serum GGT and Serum Mg^{++} were estimated with the help of commercially available kit in patients of Alcohol dependence syndrome (n=50) and Normal Individuals (n=50) on fully automated biochemistry analyzer. Serum GGT level was found significantly higher (P<0.01) in Alcoholic patients as compared to healthy non-alcoholics. Moreover, Serum Mg^{++} was found significantly lower (P<0.01) in Alcoholic Liver Disease as compare to normal Individuals. In addition to that there is significant inverse correlation (r=-0.553) between serum GGT and Mg^{++} in study group. None of the individual tests of conventional liver function tests are of much importance in diagnosis of liver disease; however, when many of the liver function tests are abnormal at the same time, liver disease is the most probable diagnosis. Data of the present study clearly conclude that serum GGT activity along with serum Mg^{++} status can be useful marker for alcoholic liver disease.

P-099

To Study the Patterns of Analytes of Bilirubin and Heme Metabolism in Patients of Non-alcoholic Fatty Liver Disease

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The liver has a central role in metabolism of bilirubin and lipids, and these and related parameters are frequently affected in diseases of the liver, including non-alcoholic fatty liver disease (NAFLD. The rate limiting enzyme, lipoprotein lipase (LPL) hydrolyses triglycerides (TG) present in chylomicrons and VLDL to liberate free fatty acids (FFA) and glycerol. Heme, the breakdown product of hemoglobin (Hb) gets converted to biliverdin (which subsequently is converted to bilirubin), carbon monoxide (CO) and iron under the influence of the enzyme, heme oxygenase-1 (HO-1). HO-1 and CO have been shown in few studies to be cytoprotective. There are hardly any reports that mention the changes in the patterns of related biochemistry parameters in NAFLD patients compared to control subjects. A total of 40 patients with NAFLD and 40 control subjects in the age group of 20 to 60 years of both genders were taken for the study. Anthropometric parameters were taken and body mass index (BMI) was calculated. LPL, glycerol, FFA, carboxyhemoglobin percentage (COHb%, for the activity of HO-1) were performed manually by chemical methods. All other parameters like, lipid profile, liver function tests, glucose, lipase, were estimated in auto-analyzers. Data were analyzed by SPSS for Windows. There were significant increases in the levels of glucose, D.bilirubin, AST, ALT, ALP and TG and significant decreases in albumin and HDL in NAFLD patients compared to control subjects. BMI and waist circumference (WC) were also significantly higher in NAFLD patients. It assumes significance as prediction equations can be prepared with routine biochemistry and anthropometric parameters when large numbers are studied and patients are properly segregated. Though insulin was not measured, fasting and postprandial glucose values were not deranged in patients. However, taking BMI and WC into consideration, development of insulin resistance cannot be ruled out.

P-100

Effect of Yoga Therapy on State and Trait Anxiety in Perimenopausal Women: A Non-Randomized Controlled Study

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To determine the efficacy of 12-week Hatha yoga on state and trait anxiety score, serum cortisol and protein thiols in perimenopausal women and to compare it with physical exercise (control) group. In this non- randomized controlled trial, 247 women aged between 40 and 60 years experiencing perimenopausal symptoms were recruited at the beginning of the study. After 12.59% loss to follow up in yoga group and 12.5% loss to follow up in control group, the remaining 216 women participated either in a yoga group (n=111) or a control group (physical exercise) (n=105). The intervention in the yoga or control group consisted of a lecture, demonstration and practice session of 45 minutes each day approximately for 12 weeks. In the yoga group, practice session consisted of asanas (postures), pranayamas (breathing exercise) and dhyana (meditation) conducted by a yoga practitioner. In the control group (physical exercise) the practice session consisted of loosening exercises for 10-15 minutes and strengthening exercises for 30-35 minutes. State and trait anxiety were assessed using STAI inventory. Outcomes included a change in participants' state and trait anxiety score and change in serum cortisol and protein thiol levels after 12 weeks of intervention. State and trait anxiety were reduced significantly ($p<0.001$) in the yoga group but not in the control group. Serum cortisol and protein thiols increased significantly ($p<0.05$) in the control group but unchanged in the yoga group. Yoga intervention is effective in reducing state and trait anxiety than the physical exercise in women experiencing perimenopausal symptoms.

P-101

The Value of Arterial Blood Gas (ABG) Parameters as a Predictor of 48-hours Mortality Risk in Pediatric Patients with Sepsis

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Sepsis is a complex condition characterized by the simultaneous activation of inflammation and coagulation in response to microbial insult. The first 48-hours during admission is a critically period in predicting mortality risk among patients with sepsis, particularly in paediatrics. Arterial blood gas (ABG) parameters have been proposed to enable quick evaluation of acid-base balance associated with sepsis-related mortality. This study aims to determine the role of ABG parameter in assessing the 48-hours mortality rate in paediatric patients with sepsis during admission at Sanglah General Hospital, Bali, Indonesia. A retrospective cross-sectional study has been conducted to 73 sepsis-related mortality in paediatric patients during January-December 2018 at Sanglah General Hospital, Bali, using a consecutive technique sampling. Those cases were divided into 2 groups: ≤ 48 hours and > 48 hours of mortality risk. ABG assessments were included anion gap (AG), base excess (BE), acid-base concentration (pH), CO_2-partial pressure (pCO_2), bicarbonate ions (HCO_3^-), and electrolyte (Na^+, Cl^-, and K^+) using Siemens RAPIDLab 348 EX-Blood Gas System. Data were analysed using SPSS version 25 for Windows. Most cases were males (50.7%). There was no significant relationship between gender, ages, anion gap, electrolytes, haemoglobin, and RDW levels ($P>0.05$) in both groups. However, a statistically significant difference was found in pH, pCO_2 saturation, BE, and HCO_3^- ion ($P<0.05$) between groups. Based on receiver operating curve (ROC) analysis, several parameters have a specific significant cut-off to predict 48-hours mortality risk in children with sepsis such as pH 7.24 (AUC:74.8%; 95%CI: 63.2-86.3%; P=0.000), BE -16.45 (AUC:77.7%; 95%CI: 66.1-89.2%; P=0.000), and AG 29.35 (AUC:64.8%; 95%CI: 51.6-77.9%; P=0.031). Several parameters of ABG such as pH, BE, and AG using a specific cut-off can be used as a predictive value to assess 48-hours mortality rates among paediatric patients with sepsis.

P-102

New DiaSys Enzymatic Method for Determination of Total Bile Acids in Serum on a BioMajesty® JCA 6010/C Clinical Chemistry Analyzer

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Total bile acid (TBA) content is a sensitive marker of liver function for diagnosis and monitoring of various liver diseases. Increased TBA levels are associated with acute and chronic hepatitis, intrahepatic cholestasis of pregnancy (ICP), liver sclerosis, cirrhosis and cancer. Commercially available assays show limitations regarding the detection of clinically relevant primary and secondary bile acids. DiaSys introduces a new liquid-stable, ready-to-use reagent for assessment of all relevant bile acids in a sample offering the possibility to precisely cover all stages of liver diseases. The enzymatic Total bile acids 21 FS test is based on a specific 3-α-hydroxysteroid dehydrogenase cycling reaction converting Thio-NAD to Thio-NADH. In a second reaction step, oxidized bile acids are reduced by the same enzyme with subsequent reduction of NADH to NAD. The rate of Thio-NADH formation is determined by a change of absorbance at 410/596 nm, which is directly proportional to the concentration of bile acids in the sample material. Recovery studies on various primary and secondary bile acids were performed using 50 µM aqueous bile acids solutions. In-series precision studies have been performed according to CLSI protocol (EP5-A3). Comparative studies were performed with 100 serum samples. Data have been evaluated by using regression analysis according to Passing and Bablok. Linearity of the new Total bile acids 21 FS test is up to 220 µmol/L. Total bile acids 21 FS shows a very good in-series precision with a CV of ≤ 0.83% (at 10 µmol/L). Furthermore, method comparison of Total bile acids 21 FS with 100 native samples against a competitor test demonstrated excellent correlation [r = 0.9963; Passing/Bablok: y = 1.026 x + 0.211 µmol/L]. Calibration stability of up to 6 weeks has been achieved for the new test on BioMajesty® JCA 6010/C which is a decisive advantage over competition.

P-103

Inter-Instrumental Comparison of Serum Electrolytes in Admitted Patients in Haryana Population

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Electrolyte imbalance can cause significant risk to life. So, timely and accurate analysis of electrolytes is of critical importance. In our institute, electrolytes are analyzed by Ion selective electrode (ISE) and autoanalyzer and the aim of present study is to compare the measurement of electrolytes by these two different methodologies with the possibility of using both interchangeably. This cross sectional study was done at Department of Biochemistry, Pt B D Sharma, PGIMS, Rohtak on routine biochemistry samples over a period of two months. A total of 100 samples were selected for this study. We used Roche Cobas C auto analyzer and Eschweiler Combiline electrolyte analyzer for measurement of serum electrolytes. Patients from all age groups and both genders with any severity of injury were included. Samples which were lipemic, hyperbilirubinemia and hemolyzed were excluded from the study. The mean ± S.D. of Na in serum samples using autoanalyzer and ISE were 140.7 ± 5.26 mmol/L and 142.4 ± 7.58 mmol/L respectively. Similarly, mean ± S.D. of K in serum samples using autoanalyzer and ISE were 4.09 ± 0.76 mmol/L and 4.36 ± 0.66 mmol/L respectively. Both the values were statistically significant with p value <0.001 for both sodium and potassium samples. The Bland-Altman analysis with the 95% limit of agreement between methods were -15.67 to 12.33 mmol/L for sodium and -2.27 to 1.73 mmol/L for potassium and the mean difference was 1.7 mmol/L and 0.27 mmol/L for sodium and potassium respectively. According to gold standard measure of the standard calibration solution, The United States Clinical Laboratory Improvement Amendments (US CLIA) accepts a difference of 4 mmol/L and 0.5 mmol/L in measured sodium and potassium levels respectively. As the differences observed were within acceptable limits without much clinical significance, thus the two methods are equivalent/comparable and can be used interchangeably.

P-104

Correct Blood Sampling for Blood Gas Analysis

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ABG analysis is an essential part of management of critically ill patients admitted in ICU/ICCU. The clinical biochemists have to face a very common complaint that ABG results from their laboratories are not satisfactory. But ABG measurements are

particularly vulnerable to many pre-analytical errors. The most common sources of error encountered are improper concentration of anti-coagulant in the sample, air bubbles in the sample and delayed transport of non-cooled sample. For ABG, heparinized arterial blood sample is used. The correct heparin to blood ratio is very important to obtain accurate results and to prevent blood coagulation. According to International Federation of Clinical Chemistry (IFCC), final Heparin to blood ratio should be 50U heparin per ml arterial blood for accurate ABG results. So, syringe should be heparinized with 1000 U/ml of heparin vial, and then volume of blood equal to 20 times the dead space should be added. Dead space of a syringe varies according to the size of syringe and needle, in which liquid heparin remains even after complete flushing. Underfilling of syringe causes dilutional effects of heparin, which is the most common cause of error. Plasma electrolytes (particularly Sodium), pCO_2 and Glucose values decrease linearly with dilution of plasma. Hence this may result in false low values of these parameters. Commercial heparin is also available in 5000 U/ml and 10,000U/ml concentrations. So, using correct strength of heparin vial is equally important while preparing ABG syringe. Another very common error is due to air bubbles in ABG sample. A single air bubble may seriously affect pO_2 value. Last but not least is delayed transport of non-cooled sample, in which we get erroneous results due to cell metabolisms. It is concluded that for ABG samples, preanalytical errors must be minimized so that critically ill patients are given accurate quality reports.

P-105

Comparison of Methods for the Determination of Sodium and Potassium Concentration in Chronic Kidney Diseases

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Acid-base and electrolyte alterations are common in patients with Chronic kidney disease (CKD). As the kidneys play a central role in the regulation of body fluids, electrolytes, and acid-base balance, CKD predictably results in multiple derangements including hyponatremia, hyperkalemia, and other metabolic derangements which, in turn, are intricately linked to morbidity and mortality. For a renal patient, electrolytes and arterial blood gas analysis are crucial and are monitored frequently. Hence, accuracy in their results is at paramount importance. The widely used method for electrolytes measurement is ion-selective electrode: direct potentiometry (direct ISE) or indirect potentiometry (indirect ISE). Studies have been done to compare the two methods especially in critically ill patients but none from an exclusive renal center, hence the objective of the present study was to investigate whether results from ABG analyzer and autoanalyzer (AAs) are comparable when used to assess electrolyte levels (sodium and potassium) from patients with CKD especially in routine day-to-day scenario. A total of 242 samples were included in the study, where the venous and arterial electrolytes were measured by indirect ISE in Abbott ci4100 autoanalyzer and by direct ISE, Cobas b121 from Roche Diagnostics respectively. The mean sodium level measured by AA was 132.62±5.70 mmol/L compared to 129.68±7.89 mmol/L in ABG. Pearson's correlation coefficient was 0.548 ($p<0.0001$). The mean potassium level measured by AA was 4.45±1.12 mmol/L compared to 4.22±1.02 mmol/L in ABG with associated Pearson's correlation coefficient of 0.849 ($p<0.0001$). In conclusion, our study findings are in agreement with other studies which have shown that sodium results from AA and ABG are not equivalent and cannot be used interchangeably, whereas potassium results were in good agreement. But it does not overrule the importance of a simultaneous follow-up sample especially in severe and critical disorders where efficient electrolyte status determination, is vital.

P-106

Serum Sodium and Potassium: Comparison of Analytical Methods and Factors Affecting them

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Electrolytes play a major role in the physiological, biochemical, metabolic and neurohormonal functions in the human body. Direct and Indirect Ion selective electrodes (ISE) are two techniques commonly used for estimation of serum electrolytes. While direct ISE technology is predominantly used in the point-of-care testing (POCT) devices, most of the high throughput autoanalysers used in the clinical chemistry laboratories use the indirect ISE technology. Any discrepancy between the values obtained from POCT devices and autoanalysers may mislead the clinicians. Knowledge about the reliability of the values obtained and the factors affecting them is extremely important for the appropriate management of patients. We compared the values of serum sodium and potassium measured using direct and indirect ISE techniques and investigated the relationship between the differences observed with the total protein, albumin and lipid profile levels of 300 samples received from patients in the age group 19-64 years, at the Clinical Biochemistry Unit, NIMHANS. Haemolysed samples were excluded. Serum sodium and potassium levels were estimated using direct ISE in ILYTE equipment and indirect ISE in Beckman coulter AU 680. Serum total protein, albumin, total cholesterol, HDL cholesterol and triglycerides were measured in AU 680. Mean value of sodium measured using direct ISE was found to be significantly higher than that measured using indirect ISE while no significant difference was found between the potassium values. Significant positive correlation was observed between differences in the sodium values with total protein and albumin levels. No such correlation was observed with total cholesterol, triglyceride and HDL- cholesterol. On further subdividing the samples into hypo, normo and

hyperproteinaemic groups, indirect ISE sodium values were found to be significantly lower in the hyperproteinaemic group. Our data provides evidence of indirect ISE sodium underestimation due to hyperproteinemia. Physicians must be made aware of potential therapeutic implications of the same.

P-107

Relationship Between Serum Electrolyte Levels with Severity of Hypothyroidism: A Retrospective Study in a Tertiary Care Hospital in West Bengal

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Research has been focused in recent years on outcomes of patients with electrolyte disorders, mainly hypo- and hypernatremia, which were found to be associated with increased mortality. Thyroid hormone has an influence on renal hemodynamics, glomerular filtration, as well as the renin angiotensin aldosterone system and renal electrolyte handling. In many standard textbooks and reviews different electrolyte disorders were associated with thyroid dysfunction. Surprisingly, very few original investigations could be found on the prevalence of electrolyte disorders in patients with mild forms of hypo- or hyperthyroidism. Moreover, there is considerable confusion regarding any association between the said parameters. We therefore wanted to investigate the effects of thyroid stimulating hormone (TSH) on serum electrolytes in a broad spectrum of patients who came to outdoor of tertiary care hospital of West Bengal. Retrospective analysis of laboratory records of 150 individuals with TSH level above the normal served as cases and 50 individuals with normal TSH served as controls. These 150 people were again divided into 3 groups of mild, moderate and severe hypothyroid based on serum TSH levels. Serum TSH levels were measured by sustained chemiluniscence method and the electrolytes were measured by direct ISE method. The mean and SD of the three hypothyroid groups was calculated for all the three electrolytes and since the data is not distributed normally, non-parametric Kruskal Wallis test and one-way ANOVA were done to test the difference in significance of means of these three parameters. It was found that only serum sodium was significantly different in the three groups. Correlation and regression analysis showed that though serum sodium was associated with serum hypothyroid levels but this association is only of statistical significance but not of clinical significance. The percentage of people with actual hyponatremia was only 15%. A systemic review with meta-analysis is essential to draw conclusions.

P-108

Study on Serum Electrolytes in Sickle Cell Disease Patients on Hydroxyurea Therapy & Non-hydroxyurea Therapy

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Sickle disease affects millions of people worldwide. Where as in India, prevalence is high in Orissa, Madhya Pradesh, Maharashtra, western regions of Gujarat and Chhattisgarh. The morbidity and mortality associated with the disease, complications and sickle cell crisis is a significant health issue. Hydroxyurea is an anticancer agent, which proved to be the wonder drug in lower doses in sickle cell disease. The main aim of the study is to evaluate the serum electrolytes level in sickle disease patients and find out the effect of hydroxyurea. Sickle cell disease patients and 20 normal individuals as controls are included in the study. 11 patients on hydroxyurea therapy and 19 are non-hydroxyurea therapy. The serum levels of sodium, potassium, chloride, calcium, magnesium and phosphate were estimated in all the study subjects. There is significant difference observed among Total Bilirubin, Urea, and Creatinine and chloride levels among Sickle disease patients compared with normal individuals. No much significant results were observed among patients on hydroxyurea therapy and non-hydroxy urea therapy except for Total bilirubin and Sodium levels. Electrolytes plays crucial role in the pathophysiology of sickle cell disease, but hydroxyurea therapy does not seem to alter the electrolyte levels in patients.

P-109

Serum Creatinine Measurement: Do We Need to Change to an Enzymatic Assay?

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The 2 assays available for measurement of creatinine include picric acid based Jaffe method and enzymatic method. Jaffe method is susceptible to interference by non-creatinine chromogens such as protein, glucose, ascorbic acid, cephalosporin, keto acids. Although, enzymatic method is less prone to interferences, it is considerably more expensive. In this study, assay performance of Jaffe and enzymatic methods for serum creatinine measurement were compared using routine 493 samples at a tertiary care hospital in Sri Lanka. Serum creatinine level was measured by both Jaffe and enzymatic assays while total protein, bilirubin and glucose

levels of each sample were also measured. Creatinine concentration of routine specimens ranged from 30-1017 μmol/L. The correlation coefficient (R2) for serum creatinine between the 2 methods was 0.9529. The Jaffe method gave higher creatinine results than the enzymatic method with a mean bias of 5.9 μmol/L. The difference between the 2 assay methods was significant in higher creatinine concentrations according to the Bland-Altman plot with a more positive bias in Jaffe method compared to enzymatic assay. The average total protein, bilirubin and glucose concentrations in the routine samples were 72.8 g/L, 12.46 μmol/L, and 111.28 mg/dL respectively. According to the bias plots, both positive and negative biases were seen with lower glucose values (<100 mg/dL) while mainly positive biases were seen with higher glucose values (>200 mg/dL). The biases were evenly distributed among different levels of protein and bilirubin in the routine samples. However, all values had clinically acceptable percentage bias (<3.96%) with an average of 0.18%. The results of the above comparison study indicate that Jaffe method can produce comparable results to enzymatic method with clinically insignificant level of bias. Therefore, the decision to changing into an enzymatic method from Jaffe method requires detailed risk-benefit assessment and investigation of commonly encountered interreferences and reporting range of the catering population.

P-110

Role of Inflammatory Cytokines as Predictive Markers in Type 2 Diabetes Mellitus Patients of Kashmir, India.

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Type II Diabetes mellitus (T2DM) is a multifactorial disease and a leading cause of premature deaths. Inflammatory cytokines are reported that they have potential to enhance insulin resistance and hence T2DM. The current research was taken to study the role of inflammatory markers in T2DM along with insulin sensitivity, biochemical and hematological parameters in mountainous valley of Kashmiri population. Till date, no such kind of research has been carried out in the Kashmiri ethnicity. Therefore, we designed this beneficial study to evaluate role of inflammatory markers in T2DM in this population. A total of 340 subjects were selected for the study among them 160 were T2DM cases and 180 were healthy controls. Serum expression of markers (TNF-α and IL-6) were quantified by ELISA technique, WBC count was measured on Sysmax (Germany) hematology analyzer, biochemical and Immunoassay parameters were done on Abbott c4000 (USA) and Abbott C1000 (USA) fully automatic analyzer. The expression of candidate cytokines (TNF-α, IL-6, CRP and WBC) were highly significant ($p<0.001$) in T2DM. Among candidate cytokine markers of study only TNF-α shows positive correlation ($p<0.001$) with glycemic profile and insulin sensitivity in T2DM cases in comparison with healthy. Biochemical (fasting sugar, HbA1c, insulin resistance, lipid profile) and anthropometric (BMI) parameter was highly significant ($p<0.001$) in T2DM cases as compared to non-diabetic normal. Our findings confirm that candidate marker, TNF-α shows an important role in the pathogenesis of T2DM in the Kashmiri ethnic population and can act as early prediction biomarkers which can prevent T2DM. Further studies on the wider range of pro and anti- inflammatory markers in association with other biochemical, immunoassay and hematological parameters are needed to establish role of inflammatory markers as early prediction biomarkers which can prevent T2DM in this population.

P-111

The Contribution of Bisphenol-A to Pituitary-Gonadal Hormones Disruption and Oxidative Stress in Exposed Male Plastic Industry Workers

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Controversy exists in disruption of the pituitary-gonadal axis by Bisphenol-A (BPA), an endocrine-disrupting chemical contained in plastics. The contribution of BPA to pituitary-gonadal hormones disruption and oxidative stress in exposed male plastic industry workers was investigated. Eighty apparently healthy male individuals aged 18-62 years (male plastic industry workers (PW) age matched with 40 non-plastic industry workers (NPW)) were enrolled in this cross-sectional study. Socio-demography, reproductive history, occupational exposure duration, blood pressure and anthropometric measures were obtained by standard methods. Blood (10 mL) was collected from participants for biochemical analyses-hormones (oestradiol, testosterone, Follicle Stimulating Hormone (FSH), Luteinising Hormone (LH) and prolactin) and oxidative stress biomarkers (catalase, total antioxidant capacity, malondialdehyde and Total Plasma Peroxide (TPP)) were estimated by ELISA and spectrophotometrically respectively. Bisphenol-A was estimated by HPLC while Oxidative Stress Index (OSI) and oestradiol: testosterone ratio were calculated. Data analysed using appropriate statistical tools were significant at $p<0.05$. Among the PW, 77.5%, 12.5% and 10% participants had eugonadism, compensated hypogonadism and sertoli cell dysfunction respectively while 90%, 7.5% and 2.5% of the NPW had eugonadism, compensated hypogonadism and sertoli cell

dysfunction respectively. Diastolic blood pressure, weight, body mass index, waist circumference, hip circumference, catalase, TPP, OSI and BPA were raised while FSH was reduced in NPW compared with PW ($p<0.05$). Bisphenol-A showed a negative relationship with testosterone in NPW but direct relationships with FSH, oestradiol, TPP and OSI in PW. Bisphenol-A altered the pituitary-gonadal status and induced oxidative stress in both occupationally exposed plastic industry workers and unexposed males.

P-112

Evaluation of Adipocytokine Profile in Android and Gynoid Obese Females with Unexplained Infertility

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Adipocytokines play an important role of immune-endocrine cross-talk that contributes to disturbed reproductive processes. The purpose of the present study was to evaluate Adiponectin, Leptin and Resistin levels in android and gynoid obese females as contributing factor that leads to unexplained infertility and compare between physically active and inactive infertile females. The cross-sectional study was conducted in Infertility clinic. Participants were recruited based on the inclusion criteria of FSH < 10 μIU/ml, normal AMH, BMI (23-30) and antral follicle count with minimum of 7 to 9; and divided as Group A n= 64 infertile unexplained females and Group B n = 60 fertile females. Participants were further categorized as Group (I) Android group n= 24 with WC more than 88 cm and WHR ≥ 0.85 and Group (II) Gynoid group n = 40 based on WC less than 88 cm and WHR < 0.85. Serum Adiponectin, Leptin and Resistin were analyzed with standard ELISA kits in BIORAD ELISA instrument. Based on Global Physical Activity Questionnaire the participants were sub-grouped as physically active and inactive. Mean Adiponectin levels decreased, and leptin levels increased in unexplained infertility females. Leptin levels increased in gynoid obese females and observed linear positive correlation with BMI. Serum Adiponectin levels decreased in android obese infertile females and observed linear negative correlation with WC. Resistin levels did not show statistically significant difference. Adiponectin levels (5.13 ± 0.85 μg/ml) were significantly decreased in physically inactive Android individuals, ($p\leq0.0008$) than physically active. Leptin levels (31.96 ± 6.42 ng/ml) were significantly increased in physically inactive Gynoid individuals ($p\leq0.0001$). Decreased Adiponectin and increased Leptin levels reflect the altered adipocytokine effects in unexplained infertility. Lifestyle modifications such as moderate physical activity do create a favorable environment for the regular reproductive processes.

P-113

Effect of Thyroid Status on Reproductive Hormones and Biochemical Parameters in Postmenopausal Women

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Menopause and hypothyroidism, both individually, affect the reproductive hormone profile as well as body metabolism which is reflected in the form of deranged biochemical profile. This study was conducted on 30 postmenopausal women with newly diagnosed primary hypothyroidism and 30 euthyroid menopausal females as controls. Serum samples of all the subjects were analyzed for complete thyroid profile including total T3 (TT3), total T4 (TT4), free T3 (FT3), free T4 (FT4), thyroid stimulating hormone (TSH), estradiol, progesterone, fasting glucose, renal function tests, liver function tests and lipid profile. Data of both the groups was compared using student t-test. There was no statistically significant difference observed between the fasting glucose levels and renal and liver function tests in both the groups ($p>0.05$). Serum triglycerides, total cholesterol, low density lipoprotein cholesterol (LDL-C) and very low density lipoprotein cholesterol (VLDL-C) were found to be significantly increased ($p<0.05$) while high density lipoprotein cholesterol (HDL-C), estradiol and progesterone were found to be significantly decreased ($p<0.05$) in menopausal hypothyroid women as compared to their euthyroid counterparts. Therefore, it may be concluded that an association of both menopause and hypothyroidism may lead to accentuation of effect of each on biochemical and reproductive hormone profile.

P-114

Biochemical Markers of Bone Turnover in Hyperthyroid Patients Before and During Treatment

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Hyperthyroidism is known to accelerate bone resorption and biochemical markers of bone turnover: alkaline phosphatase (ALP), Total Procollagen Type I Intact N-Terminal Propeptide (TP1NP), C Terminal Telopeptide Type I Collagen (B-CTx) increase signifying an increase in osteoclastic and osteoblastic activity. Aim of the study was to assess selected biochemical markers of bone turnover in Hyperthyroid patients before and during treatment in comparison to euthyroid patients. Newly diagnosed 102 hyperthyroid subjects randomly selected - 35 male and 67 female

in pre and postmenopausal, age group (46±10 yrs.). Age-matched 76 controls- 30 male and 46 females (mean age 47±12). Twenty hyperthyroid subjects became euthyroid following anti-thyroid treatment of 1-3 months. Blood samples for markers of bone turnover were assessed. Vitamin D, TP1NP, B CTx were analysed using Cobas e602 electrochemiluminescence immunoassay. Thyroid function tests were assayed using the chemiluminescene immunoassay centaur XPi from Siemens and serum creatinine for kidney function. Hyperthyroid subjects had shown a statistically significant increase in ALP, TP1NP and B CTx compared to euthyroid subjects (p value <0.0001). Calcium levels and Vitamin D levels were found to be significantly higher among hyperthyroid subjects than euthyroid patients (p value <0.01). 20 Hyperthyroid subjects became euthyroid after anti-thyroid treatment of 1-3 months. Decrease in FT4 and TT4 levels was statistically significant (p value <0.0001). On the other hand, the increase in TSH was also statically significant (p value <0.05). The Hyperthyroid patients had shown significant (p value<0.001) decrease in B CTx levels but no changes in serum calcium or Vitamin D were found. In conclusion, Hyperthyroid subjects have increased biochemical markers of bone turnover which indicates significant bone loss. Furthermore, following antithyroid treatment of 1-3 months they were euthyroid with unchanged bone formation markers except for bone resorption marker.

P-115

Evaluation of Serum Ferritin Concentration in Hypothyroidism and Relation with Anti-thyroid Peroxidase Antibody Titer

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Ferritin is an iron storage protein found in the body tissues. Serum ferritin levels have been reported to be altered in patients with thyroid disease. Thus, changes in the serum concentrations of ferritin may reflect thyroid function. Furthermore, administration of T3 to hypothyroid individuals produced a significant increase in the serum ferritin level. Thyroid peroxidase (TPO) is a membrane-bound glycosylated haemoprotein that plays a key role in the biosynthesis of thyroid hormones. Anti-thyroid peroxidase (anti-TPO) antibodies are specific for the autoantigen TPO. We have undertaken the study to evaluate the serum concentration of ferritin in newly diagnosed hypothyroid patients. The titre of Anti TPO is also analysed in these patients and correlation of ferritin concentration is studied between antibody positive and antibody negative patients. It is a hospital based cross sectional study done in Department of Biochemistry, AIIMS Bhubaneswar. All patients were above 18 years of age with newly diagnosed Hypothyroidism (Clinical or Subclinical). The control group consists of patients with normal thyroid profile status. The study is going on and till now 43 patient samples were analysed along with 20 controls for thyroid-stimulating hormone (TSH), fT3, fT4 and Anti TPO on Siemens Advia Centaur XP chemiluminescence analyser and ferritin concentration by ELISA kit. Statistical analysis is being done using Microsoft Excel software. Serum ferritin levels were found to be significantly reduced in patients with hypothyroidism compared to normal subjects ($p < 0.05$). There is a significant difference observed in ferritin concentration amongst cases with high Anti TPO antibody in comparison to hypothyroid cases with normal Anti TPO antibody titre. Although the study is in initial stages, still it can be concluded that estimation of serum ferritin may be of significance in patients with hypothyroidism as it may affect the treatment protocol and also aid in monitoring the disease.

P-116

Assessment of Oxidative Stress and Inflammation in Adequately Corrected Hypothyroidism

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Hypothyroidism is among the most common endocrine disorders in India, associated with chronic complications such as secondary obesity and atherosclerotic coronary artery disease. Prolonged oxidative stress (OS) is associated with risk factors for premature atherosclerotic complications. Thyroid hormone replacement (Levo-thyroxine) is the treatment of choice. With adequate correction of thyroid profile (TSH below 10I U/mL and FT3 and FT4 within normal range) patient is considered healthy and thyroid hormone supplementation continues lifelong. However, it is not known what happens to their oxidative stress and cardiovascular risk level despite correction of thyroid profile. Therefore, in the present study we wanted to compare the OS and inflammation in adequately corrected hypothyroid patients with euthyroid controls. In a cross-sectional (analytical) design, from endocrinology OPD we recruited 40 hypothyroid patients, aged 30 to 60 years and BMI within 18.5 to 35 kg/m^2, whose thyroid profile has long been adequately corrected. Then we recruited age and gender matched healthy euthyroid volunteers as control and compared the levels lipid profile, lipid risk factors, oxidative stress and inflammation. We found no significant difference in BMI, waist hip ratio and lipid profile and lipid risk factors and blood-pressure parameters and total antioxidant status of adequately corrected hypothyroid patients. However, there was a significant increase in the levels of inflammatory marker hsCRP (3.41±2.84 versus 5.82±4.77 mg/L; $p < 0.05$), OS marker Malondialdehyde (3.79±2.78 versus 7.70±5.32 m/L; $p < 0.001$) in spite of normalized thyroid

hormone levels in hypothyroid participants. As lack of endogenous capacity to maintain FT4 in hypothyroidism is a lifelong feature, persistent elevation of OS and inflammation for such a long-time can increase the risk for future cardiovascular diseases. Therefore, we conclude that the persistent higher level of OS and inflammation has to be addressed by the treating physicians. Antioxidant therapy may be considered with caution while treating hypothyroidism.

P-117

Prevalence of Complications in Type 2 Diabetics of North West Punjabi Population and Their Relation to Duration of Diabetes

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Diabetes is the most widespread affection of mankind. As per WHO, number of type 2 diabetes patients is expected to rise globally by more than a fifth from 406 million in 2018 to 511 million in 2030. More than 50% of them will be residing in three countries; China (130 million) has got dubious distinction of being diabetic capital, India (98 million) and US (32 million). Current prevalence of diabetes in India is 8.7%. Persistent hyperglycemia in diabetes is responsible for initiation and progression of microvascular and macrovascular complications of diabetes. Microvascular complications affect retina, nervous system, and kidneys leading to retinopathy, neuropathy and nephropathy. Macrovascular complications lead to coronary artery disease (CAD), cerebrovascular diseases (CVD) and peripheral disease (PAD). These complications are leading causes of morbidity and mortality in patients with diabetes. Study was conducted in department of biochemistry GMC Amritsar in collaboration with department of Biochemistry and department of medicine ASCOMS Jammu. Individuals suffering from type 2 diabetes were included and investigated for various biochemical parameters. All the individuals were divided into groups depending on duration of diabetes and type of complications. Keeping in view the prevalence of diabetic complications present study was planned to investigate the prevalence of complications in type 2 diabetics of North West Punjabi Population and their relation to duration of diabetes. It was observed, that with increasing duration of diabetes prevalence of complications also increased. Prevalence of retinopathy increased from 2.5% to 7.5%. neuropathy from 0.5% to 11%, nephropathy from 2% to 17.5%, CAD from 1.5% to11%, PAD from 1% to 4.5% and CVD from 2 to 6%.

P-118

Optimal HbA1c Level for Screening of Prediabetes and Diabetes in a Multiethnic Singaporean Population

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Many organizations including American Diabetes Association (ADA) recommend HbA1c of ≥6.5% as an appropriate cutoff to diagnose diabetes mellitus. Until recently, in Singapore the use of HbA1c has not been accepted due to apprehensions around the influence of ethnicity, hemoglobinopathies and other disorders that alter red cell turnover. The aim of this study are to evaluate the use of HbA1c for diabetes screening, and to determine the optimal HbA1c cutoff for screening of prediabetes and diabetes in Singapore. We designed a prospective study, whereby subjects were recruited from multiple community health screening events from Mar 2017 - Mar 2018. The inclusion criteria include both sexes, age (20 - 80 years old) and ethnicity (Chinese, Malay or Indian). Laboratory tests performed were HbA1c and fasting plasma glucose (FPG). The exclusion criteria were as follows: those who were pregnant, ever diagnosed with diabetes, those with chronic kidney disease, and those with hemoglobinopathies. 214 subjects were included in the final analysis. In our study, the ADA recommended HbA1c level of 5.7% had a high sensitivity (90.0%) and low specificity (60.3%) when used as a threshold for the identification of prediabetes. In contrast, an HbA1c cutoff of 6.2% predicts prediabetes and diabetes with a sensitivity (75.0%) and specificity (95.4%) and the AUC was 0.884. Our study findings were consistent with the recommendations of the Singapore Ministry of Health March 2019 health screening guidelines for diabetes. Our proposed HbA1c cutoff at 6.2% has similar sensitivity and specificity in screening for prediabetes and diabetes compared to the recommended 6.1%.

P-119

Association of Serum TSH and Anti-thyroid Peroxidase Antibodies in Subclinical Hypothyroidism

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Thyroid disorder is common endocrine illness in India. Prevalence of Subclinical hypothyroidism is more than clinical hypothyroidism. Subclinical hypothyroidism is defined as laboratory higher value of serum thyroid stimulating hormone (TSH) > 5 μIU/

ml and free thyroxine (FT4) within reference range. Aim of present study was to determine association of serum TSH and Anti-TPO antibodies in subclinical hypothyroidism. Present study was a cross-section observational study conducted from January 2019 to June 2019. Sixty (60) subclinical hypothyroid patients were selected including 51 females and 9 males between age 20-40 years and grouped as group A (study group); group B comprising of age -and sex-matched sixty euthyroid control. Study samples were drawn and serum TSH, FreeT4, anti-TPO antibodies was analyzed by sandwich enzyme linked immune-sorbent assay. Mean age of both groups were 29.0 years. free T4 of both groups were within reference range. There was significant higher serum TSH in group A (7.61±1.39 μIU/ml) as compared to group B (2.34±0.40 μIU/ml). Mean serum anti-TPO antibodies in group A (62.7±78.1 IU/ml) was significantly higher than group B (9.04 ±3.59 IU/ml). Statistically significant positive correlation was found between serum TSH and anti-TPO antibodies in group A with P value<0.01, but not statistically significant correlation was found in group B. The finding of present study supports the positive association of serum TSH and anti-TPO antibodies in subclinical hypothyroidism suggestive of autoimmune etiology and usefulness of this test in early diagnosis and treatment to prevent further progression of illness.

P-120

Evaluation of Serum Electrolyte Pattern and LDH in Hypothyroidism and their Correlation

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Thyroid hormone is a central regulator of body hemodynamics, thermoregulation and metabolic functions and in the disorders of thyroid gland signs are often nonspecific. Both subclinical hypothyroidism and hyperthyroidism are associated with an increased risk of disease, as well as alteration in biochemical and physiologic measures in patients with overt thyroid disease. In severe hypothyroidism and myxedema hyponatremia was described to be a consequence of enhanced renal water retention mediated by vasopressin. Lactate dehydrogenase (LDH) is an enzyme found in nearly all living cells. It catalyzes the conversion of lactate to pyruvate and back, as it converts NAD^+ to NADH. However, only a few studies have investigated serum lactate dehydrogenase activity in patients with thyroid dysfunction. To estimate the levels of serum electrolytes like sodium, potassium and magnesium along with LDH in hypothyroid patients and to correlate them. The study population was serum specimens from 50 patients with hypothyroidism (as per the reports available) with age ranging from 25-65 years. The samples were collected from the Thyroid laboratory, Department of Biochemistry, PGIMS, Rohtak. The serum electrolytes like sodium and potassium were estimated by the electrolyte analyzer whereas the concentrations of magnesium and LDH were determined on the autoanalyzer. The mean values for Na, K, Mg, and LDH were found to be 132.82±22.10, 6.06±2.85, 2.2±0.5 and 910±1771. There was statistically insignificant positive correlation between Na and Mg, K and Mg, Mg and LDH while there was statistically insignificant negative correlation between Na and LDH, K and LDH. Our findings support the fact that hypothyroid patients are having subtle disturbances of electrolytes and LDH levels.

P-121

A Case Control Study on Insulin Resistance, Metabolic Syndrome in North-Indian Females with PCOS

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Polycystic Ovary syndrome (PCOS) is one of the most common endocrinopathies among women of reproductive age and a leading cause of infertility. Insulin resistance is known to be intrinsic to this disorder & contributes a major role in its pathogenesis. Insulin resistance also increases risk of development of glucose intolerance, type 2 diabetes mellitus, hypertension, dyslipidemia & cardiovascular abnormalities & metabolic syndrome in these patients. Keeping this in view, present study was conducted to study Insulin resistance, metabolic syndrome & risk of PCOS in North-Indian females. A case-control study was conducted in department of Biochemistry & department of Obstetrics & Gynaecology at VMMC & Safdarjung hospital, New Delhi. 100 cases of PCOS & 100 controls were included in this study. Serum lipid profile and fasting plasma sugar levels were estimated by using automated Clinical Chemistry Analyser Siemens ADVIA 2400. Plasma Insulin estimation was done by commercially available ELISA kit. Mean Plasma sugar was significantly higher (p-value 0.001) in cases (96.5mg/dl) than controls (79.9 mg/dl). Mean fasting Insulin levels were also significantly higher (p-value=0.001) in cases (20 μIU/ml) than controls (11.6 μIU/ml). HOMA-IR index was higher (p-value<0.001) in cases (5) than controls (2.3). Serum HDL were lower (p-value<0.001) in cases (45.6 mg/dl) than controls (55.4 mg/dl). Mean Waist circumference was higher (p-value=0.04) in cases (76.4 cm) than controls (75.1 cm) and both systolic (p-value=0.001) and diastolic (p-value=0.006) BP were higher among cases than controls. Women with PCOS show higher metabolic abnormalities including higher BP, higher waist circumference and lower HDL with insulin resistance. Early screening of these features may prove advantageous in designing therapeutic approach to PCOS & also to prevent or slow the progression to complications like diabetes and cardiovascular disorders in these women.

P-122

Evaluation of Serum Ferritin as an Inflammatory Marker in Comparison to TNF-α in Obese Individuals

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Recent studies have implicated the role of adipose tissue (AT) as a modulator of inflammation. Pro inflammatory cytokines such as TNF-α are secreted from adipocytes. Serum ferritin is associated with inflammation but whether it is involved in the inflammatory cycle or not is yet to be elucidated. Hence, this study aims to evaluate and compare the serum ferritin levels as an inflammatory marker with TNF-α in obese individuals. The study included 64 obese individuals. The average age of the study population was 32±10 years of age. Obese individuals were classified on the basis of Body Mass Index (BMI) taking Asia-Pacific Body mass index classification. Serum ferritin was estimated by Chemiluminescence in COBAS-e411, Roche and Serum TNF-α by ELISA kits adapted to EVOLIS TWIN PLUS, Bio-Rad. All data is represented as Mean ± Standard Deviation. These data were compared by Pearson's correlation test using SPSS version 24. We found a significant positive correlation between serum ferritin, TNF-α and obesity ($P<0.05$, $r= 0.869$). Serum ferritin correlates positively and can be used as an inflammatory marker in comparison to TNF-α in obese individuals. Hence, serum ferritin can be used as a marker for inflammation, cellular stress and damage.

P-123

Association of Periodic Weight and Waist Circumference Change and Risk of Future Type 2 DM in Obese and Normal Weight in Adults

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Metabolic syndrome and obesity are often suggested as the precursor stage for developing Type 2 Diabetes Mellitus (T2DM) and cardiometabolic disease. Recent advances in weight loss techniques and awareness of lifestyle diseases has encouraged the young adults to achieve weight and waist circumference variations. But there is lack of research to compare responses to weight and waist circumference change in metabolically healthy obese individuals (MHO) and metabolically healthy normal weight young adults (MHNW). Hence, in this study we compared the impact of weight change, change in waist circumference with Glycated Hb and cardiometabolic risk factors. This retrospective study included the data from December 2015 to December 2018. Multiple observations on 1507 individuals were included. The definition for metabolically healthy included the absence of all characteristics of Metabolic syndrome. The 5 risk factors of cardiometabolic diseases include increased BP, insulin resistance (HOMA-IR), waist circumference, high serum TG and low serum HDL. BMI was classified using Asia Pacific Classification. Fasting samples were used to estimate blood sugar, lipid profile, HbA1c by autoanalyzer. Fasting Insulin was estimated by chemiluminescence and insulin resistance was calculated by HOMA-IR. Weight loss was not associated with any significant change in Glycated Hb or cardiometabolic risk factors in MHO and MHNW individuals. But we observed a association of weight gain with an increase in both diastolic and systolic BP, serum TG levels, Fasting Blood Sugar and Glycated Hb in MHO adults as compared to MHNW adults. MHO weight losers had a favorable change in the 5 cardiometabolic risk factors and glycated Hb ($p < 0.05$). The study elucidates the variations in MHO and MHNW adults and recommends weight loss in MHO adults to avoid increase in risk factors for development of cardiometabolic diseases and T2DM.

P-124

Serum Leptin Level and Its Correlation with Insulin Resistance in North Indian Females with Polycystic Ovarian Syndrome

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Polycystic ovarian syndrome is a common endocrinological disorder, among women of reproductive age with global prevalence up to 5- 7%, which is frequently associated with chronic anovulation, hyperandrogenemia, insulin resistance and obesity. Leptin, hormone product of obesity (ob) gene, synthesized exclusively in adipose tissue. Recently Leptin resistance has been reported to have key role in development of obesity also accompanied by insulin resistance (IR), compensatory hyperinsulinemia suggesting the possibility of interaction between insulin and leptin. However, the relationship between Leptin and Insulin resistance in Polycystic ovarian syndrome is still controversial. Keeping in view present study was conducted to evaluate the serum Leptin level and its correlation with body mass index and insulin level in PCOS in North Indian population. A case control study was conducted in department of biochemistry and department of obstetrics and gynecology at VMMC & Safdarjung hospital, New Delhi. 50 cases diagnosed with PCOS satisfying the Rotterdam criteria were enrolled in the study. 50 age and sex matched controls were taken excluding patients with any

endocrinological disorder or taking hormonal supplementation. Plasma Insulin and Serum Leptin levels were done by commercially available ELISA kit. The mean leptin level was 27.86±1.33 ng/ml and 12.26±1.13 ng/ml was observed in PCOS patients and controls. Positive correlation was observed in serum leptin level and BMI (r=0.90 p<0.0001) Mean serum Insulin level was 12.26 mIU/L and 8.26 mIU/L. Obese women with PCOS have significantly higher level of serum Leptin. Correlation between serum leptin level and fasting insulin was insignificant(p>0.05) .Leptin resistance impairs peripheral glucose homeostasis by decreasing hypothalamic response to insulin and may lead to type 2 diabetes mellitus. Thus, Serum fasting Insulin and Serum Leptin levels could serve as an early biomarkers for PCOS complications hence, early intervention could prevent progression of disease to Diabetes and metabolic syndrome.

P-125

Role of MicroRNA-181b and Growth Differentiation Factor-15 on Insulin Resistance in Type II Diabetes

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Animal studies demonstrate that microRNA-181b affects glucose homeostasis. Recent observations on cell lines have shown it as a downstream effector of transforming growth factor-β (TGF-β) signaling. Moreover, Growth Differentiation Factor-15 (GDF-15), belonging to TGF-β superfamily, is implicated as a possible biomarker for diabetes risk. This pilot experiment investigates and assesses the expression of hsa-miR181b and serum GDF-15 levels in type II diabetes. Over a one-year period, 20 diagnosed cases and 20 age and sex-matched healthy controls were recruited after taking informed consent. Clinical history and anthropometric measurements were taken and basic biochemical profile was carried out; hsa-miR181b expression and serum GDF-15 levels were evaluated by RT-qPCR and sandwich ELISA respectively. All analysis were done using R software (version 3.5.3). We observed a significant increase in Body mass index (BMI) [p=0.006], Waist-to-hip ratio (WHR) [p=0.007], Fasting blood glucose (FBS) [p=0.000], HbA1c [p=0.000], and GDF-15 [p=0.011] in diabetic group as compared to control group, while HOMA-β was significantly decreased [p=0.000]. GDF-15 showed a significant positive correlation with FBS (Spearman's rank correlation coefficient ρ=0.45, p=0.044) and HbA1c (ρ=0.51, p=0.022), and a negative correlation with HOMA-β (ρ=-0.58, p=0.009). We found hsa-miR181b expression to be significantly upregulated (p=0.007, 95% CI 0.72-4.29) in patients compared to control subjects, with a 5.66 fold change. Significant positive correlations between hsa-miR181b and HbA1c (ρ=0.45, p=0.045) as well as with HOMA-IR (ρ=0.46, p=0.040) were seen. Thus, our study reports a significantly high circulating hsa-miR181b and serum GDF-15 levels among type II diabetic patients. The association data suggest that miR181b has an inductive effect in type II diabetics by causing insulin resistance; also, increased GDF-15 levels hints at a possible role of TGF-β signaling and miR181b upregulation in the progression of diabetes. But further studies with large population are necessary to evince this claim.

P-126

Serum Adiponectin Levels in Women with Polycystic Ovarian Syndrome (PCOS) and its Association with Adiposity and Insulin Resistance

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Polycystic ovarian syndrome (PCOS) is characterized by chronic anovulation and hyperandrogenism and compensatory hyperinsulinemia. Both lean and obese women with PCOS show reduced insulin sensitivity and hyperinsulinemia to some extent but insulin resistance is exacerbated with obesity. The aim of present study to measure serum adiponectin, BMI and its correlation with insulin resistance. Adiponectin action in increasing fatty acid oxidation and insulin sensitivity explains its crucial role in the metabolism. Studies have demonstrated hypo-adiponectinemia and its association with increased insulin resistance and subsequent development of Diabetes. A case-control study was conducted in the department of biochemistry and department of obstetrics and gynecology at VMMC and Safdarjung hospital, New Delhi. 50 cases diagnosed with PCOS satisfying the Rotterdam criteria were enrolled in the study. 50 Age and sex matched controls were taken excluding patients with any endocrinological disorder or taking hormonal supplementation. Plasma Insulin and Serum adiponectin levels were done by commercially available ELISA kit. The mean adiponectin level was 0.46±0.30 ng/ml and 21.90±1.11 ng/ml was observed in PCOS cases and controls found to be statistically significant (p<0.05). Mean serum Insulin level was 18.68±11.04 and 9.41±4.31 in cases and controls which was also statistically significant. Correlation was observed in serum adiponectin level with BMI (r=0.14 p>0.05 (0.30), serum glucose level (r=0.05 p>0.05 (0.68) and serum fasting insulin (r=0.076 p>0.05 (0.59) but was statistically insignificant. The serum adiponectin level has inverse relationship with Body mass index and Serum Glucose level. Decreased Serum adiponectin level is related to increased insulin resistance and subsequent development of complications of PCOS. The role of adiponectin supplements could play a significant role in the control of metabolism and decease morbidity associated with PCOS.

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Assessment of Oxidative Stress Index in Sub-clinical Hypothyroidism

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Oxidative stress caused by reactive oxygen species (ROS) causes an imbalance in the antioxidant/oxidant status of an individual .It is well recognized in hyperthyroidism & overt hypothyroidism but not in subclinical hypothyroidism. SCH has recently attracted much attention as it exhibits the same cardiovascular consequences as overt hypothyroidism. The main objective of the study was to evaluate the oxidative stress index (OSI) and correlation of OSI with thyroid profile and Paraoxonase-1 (PON-1) the antioxidant enzyme associated with HDL in subjects with subclinical hypothyroidism. OSI was calculated after measuring the total oxidant status (TOS) and total antioxidant status (TAS). This cross-sectional study included 64 subjects with sub-clinical hypothyroidism, and 128 subjects with normal thyroid profile between 25 and 75 yrs of age. Differences between the variables in the two groups were analyzed by the unpaired t test. Correlation analysis was done by Pearson's test. Subjects with SCH showed significant increase in oxidant status and oxidative stress index. There was no significant change in the antioxidant status as well the PON-1 activity in subjects with SCH. OSI can be considered as an efficient marker of oxidative stress. Its diagnostic ability was assessed by ROC Curve which gave a significant sensitivity and specificity.

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Reproductive Dysfunction & Role of Anticonvulsant Drugs in Epileptic Women

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The aim of the present study is conducted to assess the relationship between reproductive hormonal dysfunction, ovarian echo graphic abnormalities and clinical presentation with anti-epileptic drugs in female patients. Females of age more than 12 years, attending the Epilepsy Clinic, Neurology OPD or admitted to the neurology ward of GIPMER were included in the study. Serum samples were analyzed for levels of antiepileptic drugs and LH, FSH, Estradiol, Prolactin, SHBG hormones levels. Women with epilepsy who had ovarian echographic abnormalities were compared with epileptic females with normal ovarian ultrasound. 38.5% of the participants on polytherapy developed abnormal ovarian morphology whereas only 26.3% on monotherapy developed the same. The difference was statistically significant. LH levels were significantly higher in the epileptic patients with abnormal ovarian morphology ($p = 0.001$). LH and SHBG levels were significantly elevated in the patients on polytherapy with p value = 0.000 and $p = 0.002$ respectively. A significant correlation was observed between LH and clinical parameters such as age of patient ($p = 0.011$), age of onset of disease ($p = 0.015$) and duration of disease ($p = 0.023$). Our study indicates that AED therapy has an adverse effect on reproductive health in women with epilepsy. Complete endocrine evaluation is needed to be considered before administering antiepileptic drug treatment.

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Role of Interferon Gamma in Gestational Diabetes Mellitus

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Gestational Diabetes Mellitus (GDM) exposes women to a higher risk for development of type 2 diabetes mellitus in later years of life. Newborns of mothers with GDM are at increased risk for acute perinatal complications including hypoglycemia, jaundice and being large for gestational age. The inflammatory system in the pathogenesis of T2DM and GDM has been increasingly investigated, one of which includes interferon gamma; in this paper we analyze the role of this mediator in women with GDM. To study the levels of interferon gamma in Gestational Diabetes Mellitus cases and controls. Correlate the levels of interferon gamma with fasting plasma glucose in cases and controls. This case control study was performed in 90 females (45 GDM patients and 45 control subjects without GDM) in the age group of 20-40 years referred to a tertiary care hospital. Blood samples were analyzed for fasting plasma glucose & Interferon gamma was analyzed using ELISA technique. Interferon gamma concentrations were lower in women with gestational diabetes mellitus (28.98±10.31 pg/ml), as compared to those having a normal pregnancy (38.62±16.47 pg/ml). The change in interferon gamma was highly significant statistically ($p = 0.000$). There was statistically significant negative correlation between fasting plasma glucose and Interferon gamma ($p = 0.002$, $r = -0.345$). GDM may be associated with a state of chronic, low-grade inflammation termed "metainflammation", as opposed to an acute inflammatory response. An amplification of the low-grade inflammation already existing in normal pregnancy may lead to decrease in interferon

gamma which may play a role in development of gestational diabetes mellitus. Further studies are required for establishing it as a marker for gestational diabetes mellitus.

P-130

Evaluation of IL-10R and IL-18R Expression in Type 2 Diabetes Mellitus: A Pilot Study

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Type 2 Diabetes Mellitus (T2DM) is a multifactorial disease in which genetic, lifestyle, and environmental and inflammatory factors combine to produce insulin resistance in target tissues, resulting in hyperglycemia. Various cytokines have been associated with Type 2 Diabetes but there is insufficient data on the alteration of Interleukin receptor expression in this disease. Database analysis have shown association of IL-10R & IL-18R in Type 2 Diabetes but their roles remain unclear. The aim of this study was to compare IL-10R and IL-18R expression in Type 2 Diabetes Mellitus patients and healthy controls. 22 diabetic cases and 22 healthy controls were recruited after obtaining due informed consent. Venous blood was obtained under aseptic conditions. Biochemical parameters were analysed using autoanalyzer. RNA was isolated from whole blood by Trizol LS followed by cDNA synthesis using commercial kit. Expression of IL-10R and IL-18R was done using RT-PCR by Taqman Gene Expression assay. GAPDH was used as housekeeping gene. The mean ± SD of fasting blood glucose levels in cases was 130.43 ± 35.97 and in controls was 89.40 ± 9.5. The median 2^-Ct value of IL-10R expression was 0.16 and 0.716 among diabetes and healthy control respectively. The median 2^-Ct value of IL-18R expression was 0.0027 and 0.933 among diabetes and healthy control respectively. The differences of IL-10R and IL-18R expression was found to be statistically significant ($p<0.05$) among diabetes and healthy controls. IL-10R expression showed a 4.56-fold repression while IL-18R showed a 14.9-fold repression among diabetic patients in compare to healthy controls. Findings of our study suggests that expression of IL-10 and IL-18 receptors are dysregulated in Type 2 Diabetes Mellitus.

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Correlation between Inflammation, endothelial dysfunction, and Platelet Activation in Central Obesity Hypertensive Subjects: A study on C-Reactive Protein, P-Selectin, and CD40 Ligand

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Central obesity and hypertension are risk factors for cardio-metabolic disease. Vascular inflammation, an underlying mechanism in those conditions also related to platelet activation, which increases the risk of atherothrombosis event. The aimed of this study was to investigate the correlation between inflammation which assessed by high sensitivity C-Reactive Protein (hs-CRP), endothelial dysfunction (P-Selectin) and platelet activation (CD40L) in central obesity hypertensive subjects.

This was a cross-sectional study involved 53 eligible subjects. Central obesity determined by waist circumference .90 cm for male and .80 cm for female, while naive hypertensive subjects have systolic blood pressure .140 mmHg and/or diastolic blood pressure .90 mmHg and not taking anti-hypertensive medication; Subjects were men or women aged 30-65 with normoglycemia (FPG <126 mg/dL and or OGTT <200 mg/dL) and normal kidney function (eGFR .60 mL/minutes). hsCRP .10 mg/L showed no acute inflammation or unspecific infection and subjects not in anti-inflammation medication. Spearman correlation and Mann Whitney were used for data analysis.

This study showed no significant correlation between hs-CRP with P-selectin (p=0.785) and CD40L (p=0.520) in central obesity hypertensive subject but there was significant correlation between P-Selectin and CD40L (p=0.0001). hs-CRP (2.636 vs 1.024 mg/L, p=0.007) and CD40L (6,460 vs 4,871 pg/mL, p=0.243) levels were higher in central obesity hypertensive subjects compare to non-obes hypertensive subjects while there•fs no difference on P-Selectin level (p=0.990)

This study concluded that obesity in hypertensive subjects would augment pro-inflammatory status and platelet activation while endothelial dysfunction closely related to increased platelet activation.

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Unusual Presentation of Liddle's Syndrome in a Neonate

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Liddle syndrome is an autosomal dominant disorder leading to refractory hypertension, hypokalemia and metabolic alkalosis, hyporeninemia and hypoaldosteronism. The molecular basis of Liddle syndrome resides in the mutations of the SCNN1A, SCNN1B and SCNN1G genes, which encode for the alpha, beta and gamma epithelial sodium channel (ENAC) respectively. We present a 6 year old male who at the age of 10 days was admitted to the neonatal ICU with severe dehydration and acute renal failure. The renal impairment resolved following treatment and he was discharged. He subsequently developed uncontrolled hypertension requiring readmission. To our knowledge this is the youngest patient diagnosed with Liddle syndrome. The patient is managed on multiple antihypertensive drugs and potassium supplements. Electrolytes were analyzed using ion selective electrodes (ISEs) on the Abbott Architect ci8200. Initial genetic testing was limited to c1815G>A (pR563Q) of the SCNN1B gene. Electrolyte measurements revealed potassium values ranging between 2.0 to 3.0 (3.7-5.9 mmol/L), sodium levels 159 to 171 (136-145 mmol/L). Metabolic alkalosis was present. In addition, it was found that both aldosterone and renin were suppressed aldosterone <27 (49-643pmol/L) and renin 0.5 (6.5-36.2 miUL). As the c1815G>A (pR563Q) mutation was not detected, further testing involved sequencing of the distal part of exon 13 of SCNN1B gene. No mutations were detected. Further investigations are underway to evaluate other subunits of the ENAC gene. The clinical and biochemical features are in keeping with Liddle syndrome, a rare disease that can be easily missed or overlooked in pediatric patients. It should be considered in patients presenting with hypokalemia, hypertension in the presence of low renin and aldosterone levels.

P-133

Thyroid Dysfunction in Polycystic Ovarian Syndrome

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Thyroid disorders and Polycystic Ovarian Syndrome are the two most common endocrine disorders in the general population. Polycystic Ovarian Syndrome is seen in women with the reproductive age group, seems to be adversely affected by associated thyroid dysfunction. Both thyroid disorders and polycystic ovarian syndrome are independent risks for Ovarian failure, infertility and pregnancy related complications. The aim of the study was to evaluate thyroid dysfunction in patients of Polycystic Ovarian Syndrome. The study was conducted on 80 ultrasonography diagnosed cases of Polycystic Ovarian Syndrome, who attended the OP of Obstetrics and Gynecology department, Government General Hospital, Kurnool Andhra Pradesh. In cases of Polycystic Ovarian Syndrome, the blood samples were collected to investigate Fasting Blood glucose, Post Prandial Blood glucose, Serum Thyroid Stimulating Hormone Levels, Free T3, Free T4, Serum Insulin Levels, Serum Follicular Stimulating Hormone Levels, Serum Luteinizing hormone Levels. There was significant increase in Fasting Blood Glucose Level, Post Prandial Blood Glucose Level, increase in Thyroid Stimulating Hormone Levels, decrease in Free T3 and Free T4 Levels and there is increase in Follicular Stimulating Hormone and Luteinizing Hormone Levels. The findings of the study show that Polycystic Ovarian Syndrome is associated with hypothyroidism and Insulin resistance.

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Status of Serum Plasminogen Activator Inhibitor-1 in Metabolic Healthy and Unhealthy Obesity as Compare to Healthy Control: A Cross-sectional Study

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Metabolic Unhealthy Obese (MUO) are at increased risk of thrombotic episodes. Metabolic Healthy Obese (MHO), a controversial concept has been considered physiological normal due to absence of any metabolic abnormality. The present study

aims to study the status of pro thrombotic mediator and adipokine - Plasminogen Activator Inhibitor-1 (PAI-1) in MHO, MUO as compare to healthy control. PAI-1 has been consider a biomarker for developing thrombotic episodes. The study group consisted of 100 subjects which was divided into two groups. Group 1 consist of 50 obese subjects with and without metabolic abnormalities. Group 1 further subdivided into two groups. Group1A consisted of 25 MHO and Group2A consisted of 25 MUO. Group 2 consisted of 50 non obese healthy controls. Clinical data was collected and routine investigation was done. Serum PAI-1 levels were measured by ELISA. The mean serum PAI-1 levels were higher in MHO and MUO as compare to healthy control (262±28 ng/ml vs. 286±348 ng/ml, P = 0.341). More over the mean level of serum PAI-1 were higher in MHO as compare to healthy controls (217±100 ng/ml). Increased PAI-1 levels even in MHO point towards that they have subtle levels of alteration from normal physiological adipokines homeostasis. Even MHO might be at increased risk of adverse thrombotic outcomes. PAI-1 may also be considered as biomarker in assessing thrombotic outcomes in MHO.

P-135

Il-2 Levels in Type-2 Diabetes Mellitus: A Pilot Study

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Diabetes Mellitus represents a heterogeneous group of metabolic abnormalities with hyperglycaemia due to either absolute insulin deficiency or a reduction of insulin biological function. Increased production of inflammatory cytokines and certain adipokines also contributes to the development of insulin resistance suggesting that diabetes is associated with enhanced cytokine production, raising the possibility that metabolic abnormalities in diabetes may originate from or exacerbated by alterations in cytokine signalling. One of a kind is interleukin-2, which is a pro-inflammatory cytokine. It has been extensively studied in Type 1 Diabetes Mellitus but little knowledge remains regarding its role in Type 2 Diabetes Mellitus. The aim of this study was to compare the levels of IL-2 in cases of type-2 diabetes mellitus and healthy controls. 21 cases of type-2 diabetes mellitus and 21 healthy controls were recruited after obtaining due informed consent. Biochemical parameters were estimated in auto analyser. Serum Interleukin-2 levels were estimated by ELISA using commercial kit. The mean±SE of IL-2 in case and controls were 139.7±37.15 pg/ml and 12.69±0.87 pg/ml respectively. The differences in the levels of IL-2 among diabetic cases and healthy controls were statistically significant ($p<0.05$). Findings of our study highlights the importance of IL-2 dysregulation in Type-2 Diabetes Mellitus.

P-136

Susceptibility Genes in Autoimmune Thyroiditis

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Autoimmune thyroiditis (AITD) results from dysregulation of the immune system leading to an immune attack on the thyroid gland. AITD is T cell-mediated organ-specific autoimmune disorders and it is seen mostly in women between 30-50 years of age. Autoimmune thyroiditis can cause several forms of thyroiditis ranging from hypothyroidism to hyperthyroidism. The two major clinical presentations are Grave's disease and Hashimoto's thyroiditis. AITD caused by an interaction between susceptibility genes and environmental triggers. AITD susceptibility genes can be categorized as either thyroid-specific (Tg, TSHR) or immune-modulating (FOXP3, CD25, CD40, CTLA-4, HLA), with HLA-DR3 carrying the highest risk. Of the AITD susceptibility genes, FOXP3 and CD25 play critical roles in the establishment of peripheral tolerance while CD40, CTLA-4, and the HLA genes are pivotal for T lymphocyte activation and antigen presentation. The development of antibodies to thyroid peroxidase (TPO) thyroglobulin (TG) and thyroid-stimulating hormone receptor (TSH R) is the main hallmark of AITD. Circulating T Lymphocytes are increased in AITD and the thyroid gland is infiltrated with CD4+ and CD8+ T cells. Polymorphisms in these immune-modulating genes, in particular, significantly contribute to the predisposition for GD, HT and, unsurprisingly, other autoimmune diseases Recent studies have shown the importance cytokines and chemokines in the pathogenesis of AITD. In thyroid tissue, recruited T helper 1 (Th1) lymphocytes may be responsible for enhanced IFN-γ and TNF-α production, which in turn stimulates CXCL10 (the prototype of the IFN-y-inducible Th1 chemokines) secretion from the thyroid cells, therefore creating an amplification feedback loop initiating and perpetuating the autoimmune process. Further knowledge of the precise mechanisms of interaction between environmental factors and genes in inducing thyroid autoimmunity could result in the development of new strategies for diagnosis and treatment.

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Effect of Lithium Therapy on Thyroid Function Tests in Patients with Bipolar Disorder

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Lithium is the most effective long-term therapy for bipolar disorder, protecting against both depression and mania and reducing the risk of suicide and short term mortality. The antithyroid effect of lithium is one of the most common side effects. Follow-up studies of lithium treated patients have revealed the presence of varying degrees of hypothyroidism, ranging from 1% to 50%. This cross-sectional study was conducted in TU Teaching Hospital during the period of October 2014 to November 2015 (14 months). 75 bipolar disorder patients treated with lithium and 75 age sex matched bipolar disorder patients without any psychotic drug treatment as controls were enrolled. Bipolar disorder was defined as per ICD-10-DCR guidelines by consultant Psychiatrist. Thyroid function test (fT3, fT4 and TSH level) was estimated by CLIA method. The prevalence of subclinical hypothyroidism was found slightly higher in female than that of male group (17.6% vs. 16.6%). The percentage of hypothyroidism and subclinical hypothyroidism increases significantly ($p<0.001$) as the duration of lithium therapy increases. Lithium treated group has lower level of mean fT3 than that of control group (5.61±1.35 vs. 6.02) but this was statistically not significant ($p=0.51$). Similarly, serum fT4 level was significantly ($p=0.019$) decreased in lithium treated group as compared to control (17.57±6.35 vs. 19.71±4.56). Serum TSH level was found significantly ($P<0.001$) higher in lithium treated group than that of control (9.67±12.47 vs. 3.41±3.69). Lithium treatment in bipolar disorder patients is associated with higher degree of hypothyroidism and subclinical hypothyroidism as compared with that of non treated patients.

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IGF-1 Levels in COPD Patients during Stable and Acute Exacerbation Phases And its Relation to the Severity of Disease

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Central to the somatotropic axis are GH and IGF-1, GH mediates its major metabolic effects through activation of somatomedins, predominantly Insulin-like growth factor 1 (IGF-1). GH/IGF-1 axis is often considered as a major metabolic regulator of muscle mass. IGF-1 stimulates muscle protein synthesis and hypertrophy, and inhibits protein catabolism via the phosphotidylinositol 3- Kinase (PI3/Akt). Evaluation of IGF-1 levels in patients with COPD is significant as in patients with COPD decreased muscle mass reduces respiratory muscle function, limb muscle function, exercise capacity and life expectancy. The present study aimed to evaluate the serum levels of IGF -1 in patients with COPD during stable and Acute exacerbation phases and relationship was concluded with the severity of disease. In this case control type of cross sectional analytical study, a total of 146 (70 stable and 76 AE COPD) male patients and 79 age matched healthy control subjects were admitted. Pulmonary function tests, ABG analysis and serum IGF-1 levels were measured and compared. IGF-1 levels were significantly lower in AE group of COPD (120.34±10.95 ng/ml) in comparison with the stable COPD (222.07±32.23 ng/ml) and control group (134.84±9.14 ng/ml) (post hoc-Tukey's HSD test in ANOVA analysis). The significant difference was observed in levels of IGF-1 between different stages of obstruction in COPD patients. IGF-1 was significantly ($p<0.05$) lower in COPD patients with severe and very severe obstruction as compared to patients with mild and moderate obstruction. However, no significant difference ($p>0.05$) was found between patients with mild and moderate obstruction as well as severe and very severe type of obstruction. The present study suggests that IGF -1 levels in COPD patients tend to be low consistent with the impression that the growth hormone axis is suppressed by chronic disease. As the disease severity increases, the local inflammation is more pronounced and the proinflammatory cytokines increases, so the more decrease of serum IGF-1 levels which is due to increase of IGF binding proteins.

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Association of Serum Growth Differentiation Factor-15 (GDF-15) with Insulin and HOMA-Beta in Type 2 Diabetes Mellitus

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India is the diabetic capital of the world with an estimated 69.2 million adults diagnosed with diabetes. Insulin resistance (IR) is characteristic of type 2 diabetic mellitus (T2DM) which causes insulin insufficiency in the body. Growth differentiation factor15 (GDF15) has been suggested as a novel marker of IR. However there is a lack of correlation studies of GDF-15, with routine diagnostic parameters in T2DM patients, for western-Indian population. Therefore, in a pilot study, we recruited twenty (20) age and gender matched study subjects (ten diagnosed with T2DM and ten healthy controls) from a tertiary care center of Western Rajasthan. Serum fasting blood sugar (FBS), glycated hemoglobin

(HbA1c) and fasting insulin levels were measured. Serum GDF-15 was evaluated using Sandwich ELISA and insulin resistance was ascertained through calculation of HOMA-IR and HOMA-β. The data was statistically analyzed using SPSS21 by students T test and Pearson's correlation analysis. This case control study reported that, among the total recruited samples, Hb1Ac and FBS were higher in T2DM patients (8.96±3.11 and 186.2±80.17 respectively), as compared to control HbA1c (5.42±0.23) and FBS (93.4±9.35) levels. Serum GDF-15 was significantly (p=0.003) higher in diabetic patients (mean±SD: 1225.17±405.48 ng/mL), when compared to healthy controls (mean SD 703.22±258.69 ng/mL). Further, the serum GDF-15 values were found to be positively correlated with FBS (r=0.51, p=0.023) and HbA1c (r=0.59, p=0.007) values, whereas the GDF-15 values showed negative correlation with HOMA-beta (r=-0.47, p=0.038) and insulin (r=-0.41, p=0.074) values. This study concludes that increased expression of GDF-15 in T2DM patients is directly associated with FBS and HbA1c levels and inversely associated with beta-cell function. Hence, this correlation furthers the acceptability of GDF-15 as a putative biomarker for T2DM and larger population studies may help establish its significance as a potential biomarker.

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An Empirical Study of Circulating miR-21 as a Potential Biomarker in Type-2 Diabetes Mellitus

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Diabetes has become a major global health problem worldwide. Currently, glycated hemoglobin (HbA1c) and fasting blood glucose (FBS) are used as diagnostic markers for type 2 diabetes mellitus (T2DM), however, based on FBS and HbA1c levels many cases go undiagnosed. Therefore, a novel biomarker with sensitivity at par with HbA1c is need of the hour for an early detection of the disease. There are no research studies to our knowledge which explores the association of circulating hsa-miR-21 and T2DM in western Rajasthan. Therefore, the aim of current research study is to analyze hsa-miR-21 as a novel diagnostic marker for T2DM. In this study we have recruited 60 participants (30 T2DM and 30 healthy controls) which were age and sex matched, from a tertiary care center of Western Rajasthan. Anthropometric measurements were taken which included Body-Mass Index (BMI) and Waist-to-Hip Ratio (WHR). Biochemical profile including FBS and serum insulin levels were measured. HOMA-IR and HOMA-β were calculated as indicators of insulin resistance and insulin sensitivity. Micro RNA expression was done on Bio-Rad CFX96 using SYBR green chemistry. The ΔCq was calculated using GAPDH as internal control and ROC curve was plotted using SPSS23. Further statistical analysis was done using SPSS23 software for continuous variables and $p<0.05$ was considered to be statistically significant. The study reports significant difference in the scores for ΔCt of hsa-miR-21 T2DM -6.471 ± 0.471 and ΔCt of hsa-miR-21 T2DM Control -3.708 ± 0.649, $p < 0.001$. The ROC curve analysis for hsa-miR-21 in T2DM showed an area under curve (AUC) of 0.732 (95% CI=0.604-0.860). Thus the current study suggests that circulating hsa-miR-21 may be a strong potential diagnostic marker for T2DM.

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Evaluation of Serum Telomerase Level, Oxidative Stress, Inflammation as Markers of Senescence in Type 2 Diabetes Mellitus Patients with Nephropathy

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The prevalence of Type 2 Diabetes Mellitus (T2DM) in India is 8.7% (International Diabetes Federation data 2017). Complications of T2DM affects the health economy adversely. The multifactorial pathophysiology still eludes complete understanding. Recent studies have implicated the association of hyperglycemia, inflammation, oxidative stress in disease progression. Serum Telomerase is a marker of cellular ageing and is currently being explored as a therapeutic agent. Hence, this study aims to evaluate Serum Telomerase level, oxidative stress, inflammation as markers of senescence in T2DM patients with nephropathy. This case control study included 138 age and gender matched individuals. The average age of the participants was 35 ± 12.6 years of age. All the participants were categorized into three groups. Group 1 T2DM patients without any complications, Group 2 T2DM patients with microalbuminuria as markers of Diabetic Nephropathy (DN), Group 3 Healthy volunteers. Serum Telomerase and TNF-α (markers of inflammation) was estimated by ELISA kits; Serum Insulin by Chemiluminescence and Fasting Blood Glucose (FBG), Lipid profile, Urea and Creatinine, urinary microalbumin was estimated by autoanalyzer. Oxidative stress (OS) was evaluated by FOX2 (Ferrous oxidation xylenol orange) assay and total antioxidant capacity by FRAP (Ferric reducing antioxidant power) assay. The data is analyzed by One-way ANOVA, Pearson's correlation test and Logistic regression. We observed a significant decrease in the Serum Telomerase level, an increase in OS among the Type 2 DM patients. An inverse relationship was observed between serum telomerase and serum TNF-α, FBG, Insulin resistance and urinary microalbuminuria. Serum Telomerase can be an early marker in detecting impending DN in T2DM patients and prognostic marker DN. Since the study population was limited, we suggest a

multicentric study with larger study population which shall implicate the therapeutic role of telomerase.

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Prevalence of Anti-thyroid Peroxidase Antibodies in Patients with Thyroid Dysfunctions Attending Chitwan Medical College Teaching Hospital

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Thyroid dysfunctions are common endocrine disorders and autoimmune thyroid disorders (AITDs) are most common organ specific autoimmune disorders throughout the world. The global prevalence of AITD shows an increasing trend in both developed and under developed countries. Anti-Thyroid peroxidase (anti-TPO) antibody is an important tool in the evaluation of autoimmune thyroid disorders. The study was aimed at determining the prevalence of thyroid hormones dysfunction and positive anti-TPO antibody titer in patients with thyroid disorders. A prospective cross-sectional observational study was conducted from February to September 2017 in Department of Laboratory Medicine, CMC-TH. Serum anti-TPO and TFT (TSH, fT3 and fT4) levels were estimated in 162 selected individuals using chemi-luminescence immunoassay (CLIA) on Siemens ADVIA Centaur XP. Of 162 selected cases, majority (54.3%) belonged to 20-39 years of age. Females represented 86.4% of study population and the rest 13.6% were males. Hypothyroidism was prevalent in 47.5% of individuals, while hyperthyroidism accounted for 21.6% of the cases. 50% of the study population demonstrated positive anti-TPO titer. Females corresponded to 96% of the positive cases. Individuals in between 20-39 years demonstrated highest frequency of thyroid disorders and highest cases of positive anti-TPO titer. Serum anti-TPO titer demonstrated statistically significant association with TSH ($p=0.009$), fT3 ($p=0.037$) and sex of individuals ($p<0.001$). An insignificant association of serum anti-TPO titer existed with fT4 ($p=0.071$) and age of respondents ($p=0.582$). Hypothyroidism was most prevalent; age group of 20-39 years was most affected and demonstrated highest number of anti-TPO positive cases. Both incidence of thyroid disorders and prevalence of anti-TPO antibodies showed a marked female preponderance. A significant association existed in between anti-TPO and TSH, fT3 and sex of respondents whereas serum fT4 and age of individuals did not reveal a significant association. Thus, evaluation of serum anti-TPO titer is vital for establishing diagnosis of autoimmune thyroid disorders.

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Role of Thyroid Profile in Assessing Severity of Pre-Eclampsia

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Hypertensive disorders of pregnancy complicate about 5-10 % of all pregnancies. The majority of deaths due to pre-eclampsia and eclampsia are avoidable through the provision of timely and effective care to the women presenting with these complications. Optimizing health care to prevent and treat women with hypertensive disorders is a necessary step towards achieving goals. The present study is aimed to evaluate thyroid function in pre-eclampsia and to compare maternal outcome in cases of pre-eclampsia. A case-control study was conducted at King George Hospital, Visakhapatnam from September 2017 to July 2019. 50 pregnant women meeting the criteria of pre-eclampsia presenting to the antenatal ward were selected and compared to 50 normotensive pregnant women beyond 20 weeks of pregnancy. TSH levels were significantly raised in pre-eclampsia cases compared to controls. There is no significant difference in T3 and T4 levels between pre-eclamptic women and normotensive women. Pre-eclamptic women with TSH >4mIU/L had more complications compared to pre-eclamptic women with TSH <4mIU/L. Pre-eclampsia is associated with hypothyroidism. Pre-eclamptic patients with raised TSH levels had poor maternal outcome compared to those with normal levels. Thyroid function tests must be done in all pre-eclampsia cases. Therefore, identification of thyroid abnormalities and appropriate measures might affect the occurrence and severity of the morbidity and mortality associated with pre-eclampsia.

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Role of Vitamin D Receptor in Prediabetes

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Recent epidemiological evidence points to a potential association of vitamin D insufficiency with adverse metabolic risk and in the pathogenesis of cancer, cardiovascular diseases, type 2 diabetes and other diseases. Vitamin D exerts its action in a variety of cell types through vitamin D receptor. No reports are available in the literature regarding vitamin D and vitamin D receptor status in prediabetics. The present study was conducted in 80 persons who were divided into two groups, Case group (n= 40)- diagnosed cases of prediabetes, and Control group (n=40)- healthy normoglycemic controls. Serum 25-hydroxy vitamin D [25(OH)D] was analysed

by radioimmunoassay (RIA). Serum vitamin D receptor (VDR) protein was analysed by sandwich enzyme immunoassay (ELISA). Serum 25(OH) vitamin D levels were significantly decreased in prediabetic cases as compared to normoglycemic controls [p<0.001]. Serum Vitamin D receptor protein levels were significantly decreased in prediabetic cases as compared to normoglycemic controls [p=0.00]. Serum 25(OH)D levels showed a highly significant positive correlation with serum VDR levels in both the groups [p<0.001 at both levels]. The findings of the present study indicate that vitamin D and VDR can serve as a possible screening marker and target for modulation for management and alleviating the progress and complications of diabetes. In future, vitamin D supplementation could be of help in managing prediabetes.

P-145

Study of Correlation Between HbA1C Level and Dyslipidemia in Patients of Type 2 Diabetes Mellitus in North Indian Urban Population

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In Type 2 diabetes mellitus, dyslipidemia is one of the major risk factors for the development of macrovascular complications which may lead to CVD. The aim of this study was to correlate the association between HbA1C level and pattern of dyslipidemia among the Type 2 diabetic patients for early prevention of CVD. A retrospective cross-sectional study was carried out in ESIC Hospital Rohini, New Delhi, India, on 110 diagnosed Type 2 diabetic patients (60 males and 50 females). HbA1C, fasting blood sugar (FBS), total cholesterol, triglycerides (TG), high-density lipoprotein (HDL) cholesterol, and low-density lipoprotein (LDL) cholesterol were done on fasting venous blood samples. Data were collected and analysed through Epi - info 7. Variation in both HbA1C and FBS level show direct correlations with serum cholesterol, TG, and LDL level while an inverse correlation with serum HDL level. Patients with worse glycemic control as compared to good glycemic control show a significantly higher level of serum cholesterol and TG and a significant lower level of serum HDL. In type 2 diabetes mellitus, HbA1C is routinely used as a biomarker of long-term glycemic control and apart from this it can also be used as a predictor of dyslipidemia.

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Correlation between HbA1c and Fructosamine in Determining Glycemic Control in Thalassemic Population with Type II Diabetes in Sri Lanka

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HbA1c is considered not reliable in diagnosing and monitoring diabetes in thalassemic patients due to assay interferences and shortened life span of red cell making diagnosis and monitoring of glycemic control a challenge. Our aim was to evaluate the correlation between HbA1c and fructosamine in determining glycemic control in thalassemic patients with type II diabetes in Sri Lanka. A prospective study was carried out among 33 thalassemic (Beta thalassemia major, E-beta thalassemia, thalassemia intermedia) patients with diabetes. Glucose monitoring by two fasting and one postprandial capillary blood glucose measurements per week were done using glucometers for a period of three months. Blood was drawn from participants every 20 days for HbA1c and fructosamine. HbA1c was measured by both HPLC and capillary electrophoresis. The median for fasting, postprandial glucose and fructosamine among the thalassemic patients were 135.88 mg/dL (IQR:111.78-179.30), 177.80 mg/dL (IQR:156.40-220.25) and 420.30 µmol/L (IQR:365.55-523.65). The median HbA1c values from capillary and HPLC methods were 9.3% (IQR:7.7-10.9) and 9.1% (IQR:7.5-11.2) respectively. There was good correlation between the HbA1c values (capillary electrophoresis) and fructosamine levels (r^2=0.653, p<0.001), mean fasting glucose (r=0.526, p=0.002), and mean postprandial glucose (0.489, p=0.004) values. There was also good correlation between the HbA1c values assessed by HPLC method and fructosamine (r2=0.505, p=0.003), fasting glucose (r=0.441, p=0.01), postprandial glucose (0.537, p=0.001) values. The two methods to assess HbA1c correlated significantly with each other (r=0.816, p<0.001). When the cut off value of HbA1c (capillary) is 7.0% for the population, the sensitivity and specificity for control of diabetes (defined as fructosamine <330 µmol/L) are 90.9% and 37.5%, respectively (area under the curve: 0.800).

P-147

Study of Pro Inflammatory and Anti Inflammatory Gene Polymorphism with Biochemical Changes in Type 2 Diabetes

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Diabetes mellitus has been described as silent epidemic. A subclinical inflammatory reaction has been shown to precede the onset of Type 2 Diabetes Mellitus (T2DM). Various groups all over the world are relentlessly working out the possible role of vast number of genes associated with T2DM. Inflammation is an important outcome of any kind of imbalance in the body and is therefore an indicator of several diseases including T2DM. Various ethnic populations around the world show different levels of Single Nucleotide Polymorphism. The present study was done to explore association of cytokine gene polymorphism (Adiponectin and IL-6) and their serum levels with T2DM in population of Bhopal region. This will lead to the understanding of the roles of cytokines genes and their products in T2DM risk and development. A total of 500 participants were enrolled in the study. After following proper inclusion and exclusion criteria age-sex matched 250 controls were compared to 250 cases. Adiponectin (ADIPOQ 45 T/G (rs2241766) polymorphism in exon and IL-6 (-174 G/C rs1800795) promoter polymorphism were analyzed in DNA isolated from venous blood sample of individuals by PCR-RFLP followed by DNA Sequencing. Serum levels of Adiponectin and IL-6 was analyzed by ELISA. Serum levels of FBS, PPBS, HbA1C and CRP were estimated by fully automated analyzers. The genetic and biochemical parameters of T2DM cases and controls showed significant difference when compared. Serum level of adiponectin was significantly lower in cases than controls and other parameters were significantly higher in cases than controls. The SNP of adiponectin and IL-6 gene also showed significant difference when their allelic frequency were compared. Our findings suggests that ADIPOQ and IL-6 gene polymorphism are involved in complication and co-morbidities in T2DM patients.

P-148

Correlation of Serum Leptin, Adiponectin Levels and Leptin: Adiponectin Ratio with Carotid Intima Media Thickness in Chronic Kidney Disease Patients

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Patients with Chronic Kidney Disease (CKD) are associated with greater risk of atherosclerosis as compared to the general population. In CKD, leptin and adiponectin accumulate in serum due to reduced renal clearance and may be associated with cardiovascular damage. This study was done to find the association of Serum Leptin, Adiponectin Levels and leptin: adiponectin ratio with Carotid Intima Media Thickness in various stages of CKD patients. A retrospective case control study was carried out on total 130 patients (65 cases, 65 controls) with chronic kidney disease as defined by KDIGO guidelines with estimated GFR<60ml/min/1.73m2 for more than 3 months. Staging of the patients was done by using the MDRD formula (Modification of diet in renal disease). The CIMT was measured using B-mode ultrasound and a 7.5 MHz transducer. Serum adiponectin and leptin levels were measured using commercially available ELISA kit Methods. The serum leptin, adiponectin level and leptin: adiponectin ratio were significantly high in cases as compared to controls with P value <0.0001, 0.002 and <.0001 respectively. The correlation coefficient between stages of kidney and serum leptin, adiponectin and leptin/adiponectin ratio (LAR) was 0.772 (P value= 0.0001), 0.509 (P value <0.0001) and 0.74 (P value <0.0001) suggesting strong correlation with severity of CKD. The correlation of serum leptin and leptin: adiponectin ratio with average CIMT was found to be stronger (r=0.733 and 0.669 respectively; p<.0001) as compared to adiponectin with average CIMT (r=0.49; p<.0001). Higher levels of serum adiponectin, leptin, and LAR were positively associated with CKD and all three were good predictors of CIMT in CKD patients; leptin having the highest level of correlation. Measures to reduce adipokines like leptin and adiponectin can potentially reduce cardiovascular events and their levels may help in assessment of the stages of CKD and management of CKD patients.

P-149

Evaluation of Single Point Insulin Sensitivity Estimator (SPISE) as an Index for Insulin Sensitivity in Comparison to Triglycerides/ HDL-C Ratio

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Various surrogate markers for estimating insulin sensitivity have been devised, based on glucose tolerance test and fasting insulin levels. However, they are laborious, time?consuming, and costly. Triglycerides to high?density lipoproteins cholesterol ratio (TG/HDL-C) was introduced as an affordable tool, however it has shown to lack specificity and showed variability with regard to different populations. Recently, Paulmichl et al. devised Single Point Insulin Sensitivity Estimator (SPISE) index, based on TG, HDL, and BMI, to be comparable to M-clamp (gold standard) test and insulin dependent indices. So the purpose of our study was to investigate the usefulness of SPISE in comparison to TG/HDL-C ratio, as an insulin resistance (IR) marker for patients with prediabetes and diabetes mellitus. IR is hallmark of metabolic syndrome so, 41 patients with prediabetes and 42 patients with diabetes mellitus were divided into IR and non-IR, using South Asian Modified National Cholesterol Education Program criteria for metabolic syndrome. TG/HDL-C ratio and SPISE index was calculated for all the subjects. Receiver operating characteristic (ROC) curve was plotted to assess discriminatory ability of TG/ HDL-C ratio and SPISE to differentiate between IR and non-IR cases. Area under the curve was found to be 0.88 for SPISE ($p<0.001$, 95% CI=0.81-0.96) and 0.65 for TG/HDL-C ($p=0.03$, 95% CI=0.53-0.77). This shows, SPISE has good predictive ability to discriminate IR from non-IR cases which is much better than that of TG/HDL-C ratio. Therefore, SPISE could be a useful potential low-cost indicator with high sensitivity and specificity for predicting IR.

P-150

Effect of Subclinical Hypothyroidism and Thyroid Peroxidase Antibody (TPOAb) on Outcome of Pregnancy

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Thyroid dysfunction is the second most common endocrine disease in females of reproductive age. Although the extent of thyroid dysfunction causes both hypothyroidism and hyperthyroidism in subclinical and overt form, hypothyroidism is more common. Although it is well accepted that overt hypothyroidism and hyperthyroidism have a deleterious impact on pregnancy, studies are now focusing on the potential impact of subclinical hypothyroidism and hyperthyroidism on maternal and fetal health. Therefore this observational study was done to find the frequency of subclinical hypothyroidism and presence of TPOAb in pregnant females of our population and its effect on outcome of pregnancy. One hundred twenty pregnant females (70 subclinical hypothyroid and 50 euthyroid) having singleton pregnancy were enrolled. In the study. blood sample was analysed to estimate FT3, FT4, TSH and TPOAb. L-Thyroxine (LT4) supplementation was given to subclinical hypothyroid TPOAb positive patients. Pregnancy outcome was analysed in terms of abortion, intrauterine growth retardation (IUGR), preterm delivery, APGAR score of the newborn with SPSS. Pregnancy induced hypertension (PIH) and preterm deliveries and IUGR were found to be more in treated subclinically hypothyroid, TPOAb positive patients than euthyroid patients, it was not significantly higher. TPO positive euthyroid patients also had significantly higher risk of PIH. This study indicates that TPOAb with FT4 and TSH should be included in routine screening of pregnant females and TPOAb positive patients should be supplemented with L-Thyroxine to improve the outcome of pregnancy.

P-151

Validation of Reference Intervals Using a 'Healthy' Population - What is Considered 'Healthy'?

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Khoo Teck Puat Hospital (KTPH) is a 761-bed acute care hospital serving more than 800,000 people living in the north of Singapore. The Department of Laboratory Medicine, KTPH has

been running a study to validate the reference intervals (RIs) of core laboratory tests since October 2013. The aim of the study was to validate in-use RIs for the multi-ethnic population seen in KTPH. This study was conducted with the approval of the ethics committee. The design of the study was based on CLSI guideline on Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory (EP28-A3C). Every year, 200 volunteers were recruited at annual staff health screening events organised by KTPH. Blood specimens collected at the health screening sites were delivered to the laboratory for analysis within time frames specified by laboratory procedures. Data was analysed using SPSS statistical software and reference limits were established. RIs established from data collected over the past 6 years were similar to reference intervals already in use for most analytes. However, differences were observed between established and in-use RIs for vitamin B12, ferritin, lactate dehydrogenase (LDH), aspartate transaminase (AST) and creatine kinase (CK). When specimens were re-collected from subjects with high LDH, AST and CK, the results are usually normal (LDH 88.2%; AST 83.3% and CK 74.3%). RIs for analytes that vary significantly with lifestyle changes are complex to establish or validate. Elevated LDH, AST and CK are common in a subset of 'healthy' populations who engage in high-intensity exercise. A priori selection of reference individuals from apparently 'healthy' populations of healthcare workers working in a health-promoting workplace is inadequate to validate RIs for the general population. More stringent screening, a posteriori data exclusion and indirect sampling techniques should also be used when validating RIs of these analytes.

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Validation Study of In-vitro Stability of Analytes in Gel-separated Plasma Collection Tubes

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The Department of Laboratory Medicine of Khoo Teck Puat Hospital (KTPH) receives approximately 100,000 biochemistry and immunology test requests monthly, with add-on test requests taking up 2.5% of these requests. In January 2017, plasma separator tubes (PST) was introduced as the recommended blood collection tubes for biochemistry and immunology laboratory tests to reduce turnaround time for urgent requests. The objectives for this study are to assess the in-vitro stability of 28 analytes in PST and to establish appropriate cut-off timing for add-on test requests. Anonymized blood specimens collected in PST (n=31) were processed and analysed on cobas 8100 and cobas 8000 (Roche Diagnostics, Switzerland) respectively. Analyses were performed within 30 minutes of collection, and 2, 8, 12 and 24 hours after collection. Specimens were stored in p501 post-analytical storage system (Roche Diagnostics, Switzerland) at 2-8ºC between analyses. The mean change in analytes concentrations was plotted against time elapsed using EP Evaluator software (Data Innovations, USA). All analytes were stable in PST for up to 24 hours of refrigerated storage with exceptions to aspartate aminotransferase (AST), bicarbonate and phosphate, which were stable for 5.1 hours, 10.5 hours and 12.1 hours respectively. PST was demonstrated to not have adverse effect on the stability of most analytes when stored at 2-8°C up to 24 hours with exceptions for AST, bicarbonate and phosphate. Allowable add test time frame for bicarbonate, AST and phosphate shall be changed to 4 hours, 10 hours and 12 hours respectively. Add test time frame for all other analytes evaluated in this study is retained at 24 hours. The study has also provided evidence that plasma must be removed from PST in times of analyser downtime when sample testing is delayed for more than 4 hours.

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Difference of Free Fatty Acids Concentration in Fasting and Postprandial Status with Influence of Sample Preparation Time

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Free fatty acids (FFA) are known to be associated with metabolic status of persons with endocrinopathies like metabolic syndrome. Since FFA are a byproduct of lipid metabolism from triglycerides (TG), high level of TG after food intake can be one of the reason of increased FFA concentration, and also longer preparation time after blood collection may affect to FFA concentration, but there is no sufficient evidence of relationship between fasting status, centrifuge, measurement time and serum FFA concentration yet. Two pilot samples of fasting and postprandial status were collected from 2 male adult, then centrifuged after 30 and 60 minutes each after venipuncture. We measured serum FFA, TG, ketone, β-hydroxybutyrate concentration in directly, after 1, 2, 4 hours, and 1 day. We also collected remaining fasting and post-prandial blood samples from 40 adults, which paired for two-hour postprandial glucose test. Samples were centrifuged and frozen in less than two hours after venipuncture, then we measured serum FFA, ketone, β-hydroxybutyrate concentrations. Mean serum FFA concentration of fasting pilot samples show significantly increased level than postprandial pilot samples (548.5 μEq/L vs 235.4 μEq/L). Late centrifuged samples show slightly higher serum FFA concentration than early centrifuged samples (391.9 μEq/L vs 410.9 μEq/L, P=0.0026). Late-measured sample after centrifuge show higher serum FFA concentration, while later-measured TG, ketone, β-hydroxybutyrate concentration show no significant difference. Mean fasting and postprandial FFA of 40 adults was 491.7 μEq/L vs 177.6 μEq/L (P<0.0001), and ketone

and β-hydroxybutyrate show also significant difference (90.7 vs 34.6 μmol/L, P=0.001, 72.0 vs 22.3 mmol/L, P=0.0006). Since fasting and postprandial serum FFA concentration show significant difference and increasing serum FFA concentration in late centrifugation and measurement, serum FFA measurement should be done in fasting status with rapid sample preparation and measurement.

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Pre-analytical Errors in Clinical Chemistry Laboratory of a Tertiary Care Hospital

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Pre-analytical errors influence the total error thus hindering TQM in laboratory, consequently decreasing the accuracy and reliability of the results generated. This study was conducted with the aim to determine nature and frequency of the occurrence of pre-analytical errors. These errors were identified and corrective measures were suggested to minimise them. To determine the frequency of occurrence of pre-analytical errors and to take corrective measures to prevent the occurrence of such errors in future. This study was conducted on 13,892 (OPD & IPD) samples and pre-analytical causes for sample rejection were noted and the data was analysed. Pre-analytical errors were responsible for 2.91% (404) of samples to be rejected over a period of 3 months. The majority of the rejected samples were hemolyzed (ie, 129 out of 404 rejected samples). Substantial number of samples undergoes repeated testing because of rejection owing to pre-analytical errors. The efforts should be aimed to reduce the rates of rejected samples can provide to improve the quality of laboratory based health care processes.

P-155

Evaluation of Performance and Application of Two Nucleic acid Extraction Methods for Quantification of Plasma Epstein-Barr Virus (EBV) DNA

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To evaluate and compare the analytical performances and clinical application values of two nucleic acid extraction methods for the detection of plasma EBV-DNA. Nucleic acid was extracted parallel by silicon membrane centrifugal column method or automatic magnetic bead adsorption method, and EBV-DNA was determined by real-time fluorescence quantitative PCR. Using the third-party fixed value reference material, the detection performance of the two methods was compared, and the plasma of 100 patients with NPC and 100 healthy subjects were measured to evaluate the clinical value of the two methods. The accuracy and imprecision of the two methods for the extraction and detection of EBV-DNA met the requirements, and the results of clinical samples were linearly correlated. However, the repeatability of magnetic bead method was smaller and more stable than that of centrifugal column method (all < 3%), the minimum detection limit (about 3.334×101 IU/ml) was slightly more sensitive than that of centrifugal column method (4.159×101 IU/ml), and the positive rate and average viral load of NPC samples (95%, 8.342×103 IU/ml) were significantly higher than those of centrifugal column method (84%, 4.707×103 IU/ml) (P< 0.05).The automatic magnetic bead adsorption method for the extraction and detection of plasma EBV-DNA can achieve better detection performance and has higher clinical application value.

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Two Methods for Potassium Correction in Haemolysed Samples and their Effectiveness

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Pseudohyperkaleamia due to haemolysis is frequently encountered by laboratories. The degree of hemolysis can be spectrophotometrically measured by modern chemistry analysers and quantified as an Haemolysis Index (HI) [preferably in mg/dL or μmol/L]. We seek to develop two methods for haemolysis-corrected potassium and test their effectiveness in our laboratory. Method 1: We derived a correction equation based on first principles. The increase in potassium concentration due to lysed RBCs is directly proportional to the product of HI and RBC intracellular potassium (RBC[K]), and inversely proportional to the mean corpuscular hemoglobin concentration (MCHC): Increase in plasma [K] = (Hemolysis Index*RBC[K])/MCHC. Now, assuming RBC[K] = 150mmol/L and MCHC measured in g/dL, the correction formula will be: subtract (15/MCHC) mmol/L potassium per 100mg/dL hemolysis. Method 2: A regression line can be obtained by plotting historical serum potassium (Y-axis) against HI (X-axis). From the gradient, we developed a correction formula: subtract 0.58 mmol/L potassium per 100 mg/dL of hemolysis. We measured MCHC on the Sysmex XN-9000 analyser while HI and potassium were measured on the Roche Cobas 8000 analyser. Testing phase: From the Laboratory Information System, 51 patients who had a repeated potassium collected within 2.5 hours of a haemolysed potassium sample were selected. The repeated potassium results were compared with the corrected potassium based on the two

formulas. The mean difference between actual and corrected potassium is 0.64mmol/L(SD 0.73) and 0.29mmol/L(SD 0.70) for Methods 1 and 2 respectively. The number of patients who had a corrected potassium within 0.5 mmol/L (CLIA' 88 Requirement) of actual is 23(45.1%) and 32(62.7%) for Method 1 and 2 respectively. Eleven (21.6%) and twelve (23.5%) subjects have a difference of less than 5.3% (Lab RCV) between actual and corrected potassium for Methods 1 and 2 respectively. Caution must be advised when using corrected formulas as large biases may exist.

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Pre-analytical Errors, Reality or Myth?

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To err is to human, to not is robots. The recent advancements in the lab technologies and the techniques has given us assistance in surpassing human errors in the analytical phase of biochemical analysis of clinical parameters and achieve accurate results but the preanalytical errors bascially resulting due to human flaws and imperfection are still creating problems in laboratories and affecting post analytical patient management. Most of the samples received in the biochemistry lab have following pre-analytical errors- 1.QNS (Quantity not sufficient), 2.Wrong pateint identification (e.g mislabelled vials) 3.Incorrect vial 4.Incorrect method of sampling, 5.Delay in transport of samples to the lab,etc. QNS- It is mostly found in samples received from neonatology unit. Most of the tests requested are named panel tests, repetition of which causes not only the over burden on paramedics but also leads to unnecessary repeated invasive procedures in patients. The requesting doctors should abondon this practice and accquire a more rational, logical and specific approach before requesting the tests. Errors of vials and other problems- These are very unique in nature and very random, these can be minimised but cannot be eliminated.

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Reference Range - Award Rewarding Challenge or a Nightmare

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We have observed over time that consistently the total protein and albumin ranges of proteins at U.P.U.M.S. Saifai, have shown an inclination towards the lower limit of the normal provided reference range. We have spent time and brains over this issue and found that reference ranges should be taken out, documented and set with the change in landmarks, as reference range depends upon the race, geographical location and dietary habits. Hence we would like to justify our results, and establish a clinical correlation with varying ranges of proteins and albumin. The reference ranges provided by the kit manufacturers and textbooks are totally foreign to the local population here on the grounds of eating habits, physique, climatic conditions and economic status of the people.

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Blood Calcium Levels in Stable Haemodialysis Patients Which Calcium to Measure?

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In the presence of hypoalbuminemia, there is an increase in ICa level relative to tCa level; thus, serum tCa measurement may underestimate the ICa level. The Kidney Disease Improving Global Outcomes (KDIGO) foundation recommends the measurement of ICa levels if possible. We have evaluated the use of ionized calcium as opposed to total calcium in an unselected group of patients on maintenance hemodialysis (MHD). Blood samples were obtained before the start of dialysis, two hours after starting the dialysis and after the completion of dialysis. All statistical analyses were performed using GraphPad Prism version 5.00 and a P value<0.05 was considered significant. On carrying repeated measures ANOVA on total calcium data in all the three groups p=0.46 and same in ionised calcium data was p=0.3912. Using the normal range for total calcium, 2.12 - 2.62 mmol/L, ten patients (62.5%) during pre-dialysis and post-dialysis, and nine patients (56.25%) during dialysis would have been classified as normocalcaemic respectively. Six patients (37.5%) during pre-dialysis and post-dialysis, and seven patients (43.75%) during dialysis would have been classified as hypocalcemic. None of the patients was hypercalcemic. Using normal ionized calcium reference of 1.13 - 1.31 mmol/L, only one patient (6.25%) fell within the normal range for the laboratory during pre-dialysis session. None of the patients were classified normocalcaemic during dialysis and post-dialysis session. As a result nine patients (56.25%) and ten patients (62.5%) were misclassified as normocalcaemic during dialysis and post-dialysis session respectively by total calcium compared to ionized calcium concentrations. As is clear from the results prevalence of hypocalcaemia is underestimated by the use of total calcium level and prevalence of normocalcemia is overestimated as compared to use of ionized calcium. As a result we suggest that calcium homeostasis in the hemodialysis patients is most accurately assessed by ionized calcium levels.

P-160

Analytical Verification of the Tosoh HLC-723 G11 HbA1c Analyser and Comparison with Bio-Rad Variant Turbo 2.0

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Haemoglobin A1c (HbA1c) is used for the diagnosis and monitoring of glycaemic control in diabetes mellitus. The different analytical methods that are currently used by clinical laboratories are ion-exchange chromatography, affinity chromatography, capillary electrophoresis, immunoassay and enzymatic methods. We performed method verification of HLC-723 G11 (Tosoh Corporation, Tokyo, Japan) and compared with Bio-Rad variant II turbo 2.0 (Bio-Rad Laboratories, USA). Linearity, total analytical imprecision and carryover were verified for Tosoh HLC-723G11. Method comparison was performed between Tosoh and Bio-Rad variant II Turbo 2.0 using 100 diabetic and non-diabetic samples. To determine the influence of HbE trait on HbA1c analysis, 77 samples were simultaneously measured by Tosoh HLC-723G11 (statistical software version 3.02) as well as by Bio-Rad Variant II Turbo 2.0. We found good linearity for HbA1c values ranging from 3.9% to 15% with a correlation coefficient of R^2=0.99. The imprecision met the manufacturer's claim and the total imprecision was less than 2%. There was no carryover between high and low samples. Passing-Bablok analysis showed a good correlation between Bio-Rad variant II turbo 2.0 and Tosho G11for homozygous HbAA samples (regression equation y=0.000x + 1.000). Bland Altman plot showed a mean difference of - 0.2% between Tosoh G11 and Bio-Rad for the samples ranging from 3.8 to 16.5%. However, we noticed a mean difference of 4.2% (-1.1% to 9.6%) for HbE heterozygous samples between Tosoh G11 and Bio-Rad variant II turbo 2.0 although it has been acknowledged that there is no interference by HbAE trait when compared to boronate affinity method. Tosoh HLC-723 G11 showed good linearity, accuracy and imprecision for routine use in clinical laboratories. Laboratories should be careful about reporting results when the presence of a variant is detected. There might be interference in the HbA1c measurement in the presence of HbE heterozygotes.

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Sodium Fluoride-oxalate Vacutainer can be used as an Alternative to EDTA Vacutainer for Measurement of Co-requested HbA1c with Blood Glucose

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Glycated haemoglobin (HbA1c) gives an integrated index of glycaemia over the entire 120-day lifespan of the red blood cell, showing the cumulative effect over the past 2-3 months. HbA1c is increasingly being used as diagnostic marker in assessment of diabetes mellitus patients. Mostly HbA1c estimation require sample to be collected in EDTA (ethylenediaminetetraacetic acid) anticoagulant, which requires additional sample collection for blood glucose estimation in fluoride/potassium oxalate vial. Blood sugar vacutainers contain sodium fluoride and potassium oxalate as anticoagulants which, incidentally, can also be used in preparation of hemolysate.Thus, this study was undertaken to determine the effect of EDTA and fluoride/potassium oxalate anticoagulants on HbA1c estimation and also to observe the variation in results of HbA1c after one-week storage at -20°C. Blood samples were collected in both EDTA and fluoride/potassium oxalate vacutainers from 280 randomly selected patients of either sex. The estimation of HbA1c was done using latex agglutination inhibition method. The results show no significant changes in HbA1c values between EDTA and fluoride/oxalate vacutainers estimated on same day and after seven days of sample storage at -20°C. The two methods, using different anticoagulants, were found to be comparable on Bland Altman plot comparison. In conclusion, the fluoride/oxalate vacutainer, used for estimation of blood glucose, can also be used for HbA1c estimation particularly when these tests are co-requested. Using a single vacutainer would not only require lesser amount of blood but also be convenient for the patient, which in turn, will definitely improve patient compliance and reduce the cost of resources.

P-162

Effect of Dilution and Deproteinization in Serum Urea Estimation by DAM Method

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Determination of serum urea nitrogen is the most widely used screening test for renal function. In the rural area of country serum urea is usually measured by manual DAM method. It is done

by either by deproteinization or dilution of the serum samples. This study aimed to find out the effect of dilution and deproteinization in serum urea estimation as compared to the result of automated analyzer. A methodology based Comparative based Cross-sectional study was done in the Department of Biochemistry, BPKIHS. Blood samples of patients for their blood urea measurement were included in the study. A total of 103 was divided into three groups based on serum urea concentration measured by autoanlyzer (Cobas C311, Roche); Serum Urea ≤25mg/dL, 25-45 mg/dL and >45 mg/dL. Serum urea was estimated by DAM method by dilution and deprotenization of serum. Data was expressed in Mean and SD, Median, IQR, based on the nature of the data. Pair t test and Wilcoxon Signed Ranks Test were applied .for the comparison of serum urea concentration. The serum urea by dilution method is more closer than deproteinization in all groups with reference to autoanalyzer method (Group I dilution vs deprotenization (24.58±4.65 mg/dL vs (28.29±4.72 mg/dL), group II (dilution vs deprotenization (40.85±5.09 mg/dL vs 43.80±6.45 mg/dL) and and group III (dilution vs deproteinization (109.50 (62.25, 188.16) mg/dL vs 127.86 (65.57, 193.68) mg/dL), which are significantly different ($p<0.001$). The dilution method is better than deprotenization method for serum urea estimation by DAM method.

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Comparison of the Effects of Alpha-thalassemia 1 and Alpha-thalassemia 2 to HbA1c Level Using Eight Methods of Measurements

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The accuracy of HbA1c methods can be adversely affected by hemoglobin (Hb) variants. α-thalassemia1 and α-thalassemia2 are Hb variants commonly found in Southeast Asia. The sample was collected from the Central Laboratory, Department of Clinical Pathology. We evaluated 8 methods, including Roche Cobas c513, Bio-Rad D100, Tosoh G11, Sebia Capillary3, Mindray BS-240, Mindray BS-800, Sysmex JCA-BM6010/c, and Roche Cobas b101. An overall test of coincidence of 2 least-squares linear regression lines was used to determine the presence of α-thalassemia1 and α-thalassemia2 caused a statistically significant difference in results relative to the comparison method (Roche Cobas c513). Deming regression was used to determine whether α-thalassemia1 and α-thalassemia2 produced a clinically significant effect on HbA1c value. A total of 1958 cases comprised normal hemoglobin (n=1753), α-thalassemia1(n=97) and α-thalassemia2 (n=108). Overall median (IQR) HbA1c values were 6.3 (5.8-7.8), 6.2 (5.7-7.5), 6.3 (5.7-7.7), 6.3(5.8-7.8), 6.1 (5.3-7.2), 5.9 (5.2-7.2), 6.6 (6.0-8.1) and 6.6 (6.0-8.0), in Roche Cobas c513, Bio-Rad D100, Tosoh G11, Sebia Capillary3, Mindray BS-240, Mindray BS-800, Sysmex JCA-BM6010/C and Roche Cobas b101, respectively. In α-thalassemia1, HbA1c analysed by Sebia Capillary3 was statistically significant differences from those analysed by Roche Cobas c513 ($P<0.001$) while 6 methods were not statistically significant differences from Roche Cobas c513 ($P>0.05$). For α-thalassemia2, HbA1c analysed by Bio-Rad D100 ($P<0.001$), Tosoh G11 ($P<0.001$), Mindray BS-800 ($P<0.005$) and Roche Cobas b101 ($P<0.001$) were statistically significant differences from those analysed by Roche Cobas c513. HbA1c analysed using Mindray BS-800 was clinically significant differences from the those using Roche Cobas c513 in normal hemoglobin and Sysmex JCA-BM6010/C was clinically significant differences from the Roche Cobas c513 in α-thalassemia1. Normal hemoglobin analyzed by Mindray BS-800 showed clinically significant differences from those analyzed by c513, whereas HbA1c results from α-thalassemia1, Sysmex JCA-BM6010/C showed clinically significant differences from those measured by the c513.

P-164

Evaluation of Sigma-metric and Application of Quality Tools in Clinical Laboratory of a Tertiary Care Hospital

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Variability in analytical performance of some analyte indicated the need of evaluation of quality plan of our laboratory. We tried to put the same degree of effort into our quality metrics as we put into the laboratory processes themselves. Application of six sigma methodologies improves the quality by focusing on the root causes of the problems in performance and analyzing by flowcharts, fishbone diagrams and other quality tools. Sigma-metric were calculated for routine biochemical parameters and immunological assays (hormones) for a period of six months from January 2019 to June 2019. The major parameters with unsatisfactory performance in our laboratory were free thyroxine (fT4), Sodium, Magnesium, and Calcium. A road map was designed with Define-Measure-Analyse-Improve-Control (DMAIC) model to solve the issue. Possible causes for low analytical performance were depicted in Fishbone diagram. As a part of control phase of DMAIC Sigma-metric were assessed once again after correction of concerned issue. Sigma-metric of four analytes namely free thyroxine (FT4), Sodium, Magnesium and Calcium were below 3. The Fishbone analysis identified the most frequent causes of poor performance.

Performance issues with Sodium, Magnesium and Calcium were related to water quality and for thyroxine it was temperature. Identification of problems led to reduction in non value added work leading to adequate resource utilization by addressing the priority issue. In control phase repeated sigma-metric showed marked improvement in all parameters, thereby reinforcing the quality in the laboratory. Six sigma application for quality improvement in clinical laboratory is extremely valuable. Our fish bone analysis will be quite helpful for trobleshootings in performance of laboratory parameter.

P-165

Sample Rejection Rate as a Quality Indicator: Comparative Findings from Clinical Biochemistry Laboratory of Two Tertiary Hospitals from Southern India

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In a biochemistry laboratory, there are many determinants responsible for sample rejection, the majority of them falling into pre-analytical error.1 With advances in Laboratory & Clinical Medicine, clinical diagnosis is largely based upon biochemical investigations. Collecting & analysing data consistently are necessary tasks for assessing quality, monitoring standardized processes, improving performance and patient safety in clinical laboratories. This study was based upon retrospective data analysis of all the biochemistry laboratory samples received and total number of samples rejected in 1 year from two hospitals, JSS Hospital, (JSS) and Kasturba Hospital, Mangalore (KMC), based upon the analysis of different rejection rate, types of unintelligible approach and level of inappropriateness. We have used predefined criteria for sample rejection to assess the level of appropriateness, namely haemolysis, insufficient volume, clotted, wrong vacutainer, mismatch, venous blood & test raised by mistake. Annual sample rejection rate was 0.997% (JSS) and 1.695% (KMC). From JSS clinical biochemistry laboratory, total tests done were 697908 from 162079 vacutainers. From KMC total 1022188 tests were done from 157259 vacutainers. Haemolysis (50%) was the most common reason and mismatch (0.61%) was the least common reason. Among the inpatient departments, medicine had the highest percentage (31%) and paediatrics had the lowest percentage (0.86%). Beside common criteria, other causes like venous sample for arterial blood gas analysis with a rejection rate of 12% was the second most common cause. We need a median rejection rate to compare various rejection rates. Variation in rejection rates is dependent upon different criteria adopted by different laboratories. Clinical diagnosis pertaining to the services rendered by laboratories cannot afford to increase the Turn around Time and compromise on Total Testing Process. Further comparative studies involving multiple laboratories should be done to design a manifesto to detect and correct varied determinants of sample rejection.

P-166

Evaluation of Sigma Metrics and Measurement of Uncertainty in a Clinical Biochemistry Lab

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Majority of important clinical decisions related to diagnosis and treatment of patients are based on laboratory test results. It is thus of utmost importance that labs adopt stringent quality control measures to ensure accuracy and precision. Random and systematic errors can produce an error of measurement and generate a doubt about the true value of the measured quantity. The assessment of performance can be done using six sigma metrics, while, measurement uncertainty can be calculated to relate to the margin of doubt that exists for the result of any measurement. The aim of the study was to assess performance and calculate measurement uncertainty of the routine biochemical parameters in order to identify gaps and therefore areas of improvement. Internal quality control data was analyzed retrospectively over a period of six months to calculate lab mean, standard deviation and coefficient of variation. Validation of quality control was done by calculating the bias, from the external quality assurance scheme data. Sigma was calculated for both the levels of internal quality control. Acceptable sigma metrics of more than 3 was obtained for majority of analytes. Although day to day quality control for urea, creatinine, sodium and potassium was always found to within limits, but was not found satisfactory on sigma scale. The results emphasize that in spite of acceptable conventional quality control tools, application of sigma metrics can identify deficits in analysis and areas that need improvement in clinical labs. Measurement uncertainty was calculated for each parameter using average coefficient of variation and could be used to find out if the difference between two results could be ignored due to uncertainty or considered significant due to a genuine change in the condition of the patient. Together these parameters can help meet the ultimate goal of reducing diagnostic uncertainty and improving patient care.

P-167

Comparison of Three Different Methods of Genomic DNA Extraction and the Effect of Storage Temperature on Its Yield

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Good quality of extracted DNA is the primary step for any research in the field of molecular biology. Its extraction techniques have been improvised over the years in terms of quality and time. In present study, three different methods of DNA extraction were compared, namely DNAzol, TRI reagent and Spin column. DNA extraction was done by all three methods, in each of 10 whole blood samples taken for the study. The extracted DNA was then divided into three aliquots and stored at different temperatures (4^{o}C, -20^{o}C, -80^{o}C). To study the effect of storage temperature, extracted DNA was subsequently measured on day 5, 10, 15 and compared with baseline. DNA quantification (yield) and quality check (Absorbance ratio at 260/280) was done using Nanodrop OneC. At baseline, the average yield (Mean±SD in ug/ml of whole blood) of DNA extracted by DNAzol, TRI and Spin column methods were 10.07±3.62, 30.26±6.04 and 29.42±10.72 respectively. The means of the absorbance ratio obtained for above methods were 1.99±0.06, 1.37±0.06 and 1.84±0.06 respectively. Measurement of values over succeeding days, at different temperatures, depicted no significant changes at 4^{o}C and -80^{o}C. A gradual increase in yield at -20^{o}C was observed for DNAzol and TRI methods but did not show any significant variation in Spin column. This can be attributed to better solubilization of aggregates over time, that were formed in DNAzol and TRI but obviated from Spin column. To conclude, Spin column is the best method yielding high concentration of DNA per unit of whole blood with good absorbance ratio (slightly less only to DNAzol) and least variation with temperature. DNAzol method had the best absorbance ratio but the yield was significantly lower ($p<0.05$) and TRI reagent was least precise with unacceptable absorbance ratios despite giving high yields that were comparable with Spin column method.

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Pre-analytical Variables in Laboratory Testing

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The aim and objective of the present study was to enumerate and evaluate different types of pre-analytical errors in the clinical biochemistry laboratory and to compare the frequency of errors in the pre-analytical phase of testing before and after training the technical staff posted in the clinical biochemistry laboratory. This study was done at Government General Hospital a tertiary care teaching hospital in Kurnool, Andhra Pradesh, for a period of three months from MARCH 2019 to MAY 2019. During this period different types of pre-analytical errors were monitored. Of the 26732 Samples received during the study period 580 Samples were found to be unsuitable for testing, accounting for 2.16% of rejection. All these samples were rejected due to different types of pre-analytical errors that are due to wrong timing of sample collection (0.72%), inadequate sample (0.69%), wrong vacutainer (0.21%), missing sample (0.17%), sample drawn from IV Site (0.15%), hemolysed sample (0.15%), wrong identification (0.08%). Of all samples received in clinical biochemistry laboratory the overall percentage of rejection is 2.16%. We also found that there was reduction in the frequency of errors before and after training the staff.

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Awareness about Fasting-Related-Preanalytic-Factors for Glucose and Lipid Profile Testing Among Patients Visiting a Government-College-Hospital Versus Educated Community from Pali Rajasthan

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Fasting for 8-14 hours are required for fasting-blood-glucose (FBG) as well as lipid profile as per various guidelines of various countries. Preanalytic factors related to fasting become extremely important factors affecting the test result significantly. We compared the level of awareness with local educated community, two surveys were undertaken. A face-to-face survey was done on outpatients. Educated community from same region (at least 10th pass, ~68% pursuing Bachelor or higher degrees) was surveyed through Surveymonkey. Exclusion criteria were fully trained health professionals e.g. doctor, technician, and nurse. Information collected included demographics; perception of hours-of-fasting required; whether water-intake, beverage, snacks, Religious food-Prasad/Sehri, smoking, drinking, medication, exercise are allowed, availability-source-nature of instructions, hours fasted, and compliance. 45 patients and 156 educated controls responded to the study. Patient fasting duration varied from 1 hour to 16 hours. Even among people with educated background surveyed, only 25% could guess the correct range for fasting duration. 60% of patients perceived that nobody explained to them anything about nature of fasting. 35% of educated survey respondents felt that they never received information from any source. Among patients: Of those instructed, 83% were aware and 72% were compliant about light snacks Regarding compliance about

tea/coffea was 67% for instructed vs 30% for the uninstructed. Even 33% of instructed thought religious food could be taken in morning, and 11% actually took them whereas 55% of the uninstructed group thought it was allowable and 19% took them. Even among educated 30% thought tea/coffea are allowed and 10% thought snacks are allowed and 3% thought religious food was allowed. Doctors were overburdened with patients and yet they were usual source and communication was not in print. It could be improved by introducing distribution of printed leaflets, posters and also training the nurses, phlebotomists and social workers.

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Big Data Analysis Reveals the Existence of Seasonal Pseudohyperkalaemia Even in Temperate Climates

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Seasonal pseudohyperkalaemia has been described in colder northern hemisphere countries. The lower temperatures may inhibit red cell Na-K-ATPase allowing the efflux of potassium and higher measured levels. It has not been described in warmer subtropical climates. The aim was to determine if seasonal variation in serum potassium occurred in a temperate climate. We conducted a retrospective review of serum potassium results over two years in two South African provinces with different microclimates and seasonal temperatures. The study included patient samples from surrounding clinics and hospitals in Pretoria, Gauteng province, and in Durban, KwaZulu-Natal province, South Africa. Average temperature ranges were obtained from the South African weather service from the same period (June 2015-June 2017). A total of 91 420 results were analysed and we found a statistically significant difference between the January (summer) and June (winter) serum potassium levels ($p<0.0001$). These results demonstrate that the winter months in South Africa are associated with significantly higher measured potassium results. Seasonal pseudohyperkalaemia may be more widespread than realized and can occur in more temperate climates and laboratories should take the appropriate action when transporting samples as this could influence interpretation and clinical management.

P-171

Evaluation of Sigma Metrics of Clinical Chemistry Assays: Importance of the Allowable Total Error (TEa) Target

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Analytical quality is a prerequisite for the clinical laboratory, but it can be difficult to assess it. Sigma metrics is an objective way to measure and quantify quality. It combines total allowable error (TEa), bias and precision. TEa for an analyte is obtained from literature and can vary based on the source of data used such as Biological Variation data or Clinical Laboratory Improvement Amendments (CLIA) guidelines. Hence, we conducted this study to highlight the importance of TEa goals. The objective of our study was to calculate and compare sigma metrics of 16 clinical chemistry assays using TEa data from various sources. Precision is expressed as coefficient of variation (%CV) and Bias was calculated from target mean provided by the manufacturer and lab mean. Sources of TEa used are Biological Variability (Desirable, Optimal & Minimum) and CLIA (Old Guidelines & New proposed guidelines 2019). Sigma metric was calculated by formula "Sigma metric= (TEa-Bias)/Precision". Triglyceride both the levels showed sigma>6, with TEa biological variability desirable and old CLIA guidelines while Amylase showed sigma >3 with Biological variability minimum and old CLIA guidelines whereas, it showed sigma <2 with the Biological variability optimal & New CLIA guidelines. Sigma metrics as a quality assurance tool should be periodically used to monitor changes in assay quality. Laboratories need to improve their performance to reach the desired quality goals. Inconsistent TEa targets from different independent sources can create a dilemma and should be chosen based on assay performance. We found Biological Variability TEa values to be too demanding for routine performance whereas; old CLIA can be considered lenient.

P-172

Serum Separation: An Interesting Challenge

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Routine estimation of biochemical parameters of a blood sample in a tertiary-level cancer hospital is generally one of the most common and easy investigations to be performed. However, at times a simple procedure like this may become challenging due to various

pre-analytical problems in a laboratory. We had an interesting case where we were unable to separate serum from a blood sample since the blood sample acquired gel-like consistency soon after its receiving. We aimed to troubleshoot this pre-analytical phenomenon in order to get an accurate patient result. The standing time of the blood sample was increased in order to separate the serum but it was in vain. Then, the sample was incubated at 37°C in an incubator followed by centrifugation. This gave us a very scanty amount of serum. So, the cycle of incubation at 37°C and centrifugation was repeated a few times in order to get a sufficient amount of serum to perform the test. After repeated incubation and centrifugation, we were successfully able to acquire the desired amount of serum to perform the test. This process of repeated incubation and centrifugation may be utilized to separate the serum in blood samples having such pre-analytical phenomena. This may help us to process the sample and get the desired result rather than rejecting the sample or using expensive or laborious serum separation methods.

P-173

Study of Bilirubin Interference in Common Clinical Biochemistry Assays

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Bilirubin is an oxidative end product of heme catabolism. Bilirubin occurs in serum as relatively insoluble free bilirubin, as water soluble conjugate (mono- and di-glucuronides), and covalently bound to albumin. Interference from icterus (elevated levels of bilirubin) is so common with routine chemistry assays, that validation by manufacturers always includes interference testing for this substances. Today, when clinical laboratory is equipped with most advanced instrumentation; bilirubin interference makes clinical biochemist unanswered to clinician seeking useful diagnostic information. Aims and objectives of present work is to study bilirubin interference in biochemical assays, compare available methods to remove bilirubin interference and find out possible way to avoid bilirubin interference completely in biochemical assays. Study was conducted on 200 left out laboratory serum/plasma samples with high bilirubin (5 to 25 mg/dl). Samples were excluded having possible error causing substances such as plasma expanders, preservatives, stabilizers and presence of lipaemia & haemolysis. To study bilirubin interference in biochemical assays concentration of analytes, conjugated and unconjugated serum bilirubin was measured by standard methods, also after employing two methods used to remove bilirubin interference from serum/plasma 1. Addition of bilirubin oxidase and 2. Alkali pre-treatment of sample. The CLSI guidelines and recovery experiments described by Westgard were used to calculate interference caused by bilirubin in different assays. Bilirubin oxidase and alkali pretreatment both reduces bilirubin interference in the Jaffe's creatinine Assay. However, elimination of this interference in other biochemistry assays has not been satisfactorily achieved. We propose a chemical procedure of binding bilirubin to serum proteins and followed by precipitation of bilirubin-protein conjugate to get colourless bilirubin free supernatant where creatinine was measured. Creatinine measured by this method, showed a good correlation with standard method. The method of bilirubin removal from serum found to be applicable to assays of non-protein analytes where bilirubin interferes.

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Drug Interference in HbA1c Analysis

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The primary goal of diabetic management is to maintain the blood glucose concentration within or near the non diabetic range. Hemoglobin A1c (HbA1c) is a biochemical marker widely used in patient's with Diabetes mellitus (DM) to monitor the adequacy of long-term glycemic control. Diabetes control and Complications Trials (DCCT) and United Kingdom Prospective Diabetes study (UKPDS) both these studies showed a direct relationship between the Glycated hemoglobin (GHb) concentration and the risk of complications.ADA has also recommended stand-alone use of the HbA1c test to diagnose diabetes (with a threshold level of ≥6.5%) in addition to the current use for monitoring the effectiveness of glycemic control. GHb values are free of the influence of day to day glucose fluctuations and are unaffected by recent exercise or food ingestion. Levels of HbA1c reflect average blood glucose levels over the previous 2 to 3 months. However when patient blood glucose levels are not correlating with the measured HbA1C levels, we should investigate for the possible causes. Hemoglobinopathies and factors that affect the red blood cell life span must be considered as the possible causes. If the red blood cell life span is decreased the hemoglobin will have less time to become glycosylated and the glycosylated hemoglobin level will be lower. We present 2 cases, case-1 A 40 yrs Female patient diagnosed with Hansen's disease and second case 60 yrs known diabetic patient diagnosed with dermatitis herpetiformis were treated with dapsone presented with reduced HbA1c levels. Dapsone cause hemolysis there by reducing the life span of RBC and it also cause oxidation of hemoglobin to meth hemoglobin and interferes with glycation of hemoglobin resulting in low HbA1c values. Hence we should identify the possible factors interfering with the test result before reporting HbA1c result.

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Ischemia Modified Albumin (IMA) Estimation and Utility of Different Albumin Adjustments as Markers to Predict Early Kidney Damage in Diabetes

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UACR has been the standard for detecting albuminuria in diabetics but the presence of "non-albuminuric renal impairment" encouraged the search for novel markers. Increased serum IMA levels have recently been implicated in diabetic nephropathy cases but its estimation by conventional albumin cobalt assay is confounded by serum albumin levels. Hence, this study was conducted to estimate IMA and albumin adjusted IMA's to assess their utility in diagnosing early the renal damage in diabetes. This cross sectional study in 30 controls and 60 diabetes patients (classified as normo-, micro- and macro-albuminuric groups having UACR of <30, 30-300 and >300 mg/g of creatinine respectively) estimated serum IMA using ELISA. Albumin adjusted IMA's were calculated, namely, Adjusted IMA {(individual serum albumin concentration/median albumin concentration of population) ×IMA value}; IMA index {serum albumin×23+IMA-100} and IMA ratio {IMA/serum albumin}. SPSS ver. 20 employed ANOVA for intergroup comparison of means, Pearsonn's correlation coefficient for correlation analysis and ROC curve to assess the diagnostic performance. IMA, adjusted IMA, IMA index and IMA ratio significantly increased in diabetics showing an increasing trend across the normo-, micro- and macro-albuminuric groups implicating no confounding effect of serum albumin on IMA estimation within the normal reference range of serum albumin. IMA correlated inversely with serum albumin in micro- and macro-albuminurics whereas IMA ratio correlated strongly in all the groups. ROC curve showed highest sensitivity and specificity for IMA (97.5% and 78% at 99ng/ml cut-off) and IMA ratio (97.5% and 76% at 24.5 cut-off); AUC 0.93 for both. Similar increasing trend of IMA and albumin adjusted IMA's across various stages of albuminuria is suggestive of no effect of serum albumin concentration on IMA estimation within the normal reference range of serum albumin. Albumin adjusted IMA ratio showed greatest diagnostic performance for early detection of renal damage in diabetes.

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Comparative Evaluation of Different Vacutainers for Plasma Glucose Estimation

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The importance attached to NaF/Potassium oxalate tubes has come under scrutiny recently. To find if red (clot activator tubes) and lavender top (EDTA) tubes can be used to estimate plasma glucose, and if so till what hour? Blood was collected in grey (NaF/Potassium oxalate), red (plain) and lavender top (EDTA) tubes of BD company from 30 patients visiting out-patient department in May 2019. Following immediate centrifugation, each sample was tested thrice for glucose within 4 hours, 6 hours and 24 hours. Samples were stored at 2-8°C. Glucose was estimated by Hexokinase method on Beckman AU-5800. Grey tube glucose values correlated with red and lavender tube values ($r=0.99$, $p<0.001$) when processed within 4 hours. Median glucose (mg/dl) in grey, red, lavender tubes were 97, 96, 88 respectively. Kruskal Wallis test found no significant difference between grey vs red, grey vs lavender. Bland-Altman analysis gave a non-significant constant bias signifying good agreement. Analysis at 6 hrs revealed a % decrease of 2.06, 2.8, 2.2 in grey, red and lavender tubes respectively, which is not statistically significant. Though close correlation was found, the values after 6 hours differed significantly. After 24 hours, glucose decreased by 9% in both red and lavender tubes. No difference in glucose values was noticed among 3 tubes within 4 hours as NaF inhibition starts acting on enolase only after 4 hrs. So grey tubes offer no advantage in this era of sample transporters and pneumatic tubes. Red tubes serve as universal tubes for most of the investigations. As NABL laboratories seeking to lower the turn-around time are able to complete the tests in a shorter span, estimating glucose in them will cost-effectively, reduce the number of vacutainer tubes and blood drawn red and lavender top tubes can be used to estimate glucose upto 6 hours at room temperature.

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Establishment Of Reference Intervals For Closure Time PFA-100 In Algerian Adult According To The IFCC Method

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Platelet Function Analyzer-100 (PFA-100, Dade-Behring, Germany) is an instrument that simulates in vivo hemostatic plug formation under high shear flow by measuring the time required to occlude aperture. In Algeria, the majority of medical laboratories adopt intervals recommended by manufacturers or by scientific literature due to lack of local reference range. Furthermore, the reference values of PFA 100 TM used in laboratories have been established in the western population but these values can be challenged because of ethnic, genetic, lifestyle and diet differences. The introduction of this new analyzer in our laboratory was the opportunity to make a priori correct determination of the reference values of CT PFA 100 TM in healthy Algerien adults. A total of 248 healthy individuals were enrolled. Closure times (CT) with the collagen/epinephrine (COL/EPI) and the collagen/ADP (COL/ADP) cartridges were measured. The reference values obtained in our study and more specifically the Upper limit values are different from those of the Western and Asian population ($0.001<p<0.0001$). Thus, the results of our study indicate that many healthy Algerian people (more than 25%) would be incorrectly identified as having a primary hemostasis abnormality by the reference values of the manufacturer. The results of the present study provide reference interval for CT PFA 100 TM in healthy Algerian Adult. These results will improve accuracy of diagnosis and patient care in Algeria.

P-178

Association of Iron Deficiency Anemia with Depression in Women

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The most common form of anemia affecting 3.5 billion people in developing countries is Iron Deficiency anemia (IDA). Women of reproductive age group (15-49 years) are most commonly affected. The consequences of IDA are decreased productivity, disinterest in activities, and decreased immunity and mood disorders such as depression. The study included 92 female patients with depression in the age group of 21-45 years. The study was conducted during January 2019 to June 2019 with collaboration of the Department of Psychiatry. The Hamilton depression rating score (HDRS) was used to assess the severity of depression. Fasting whole blood was collected for analyzing Complete blood count (CBC), Hemoglobin level, Plasma ferritin, Serum Iron level, and Total iron binding capacity (TIBC). Hemoglobin level of less than 11gm/dl was considered anemia. National guidelines for Prevention and control of IDA in India were followed for detection of grading of anemia.The mean age of the study population 32±11.45 years. The Hamilton depression scores of patients were 33.1±3.49. We observed 58% of the women had IDA; out of which 9% had severe anaemia, 21% had moderate anemia and 28% of the patients had mild anemia. The HDRS score had a significant POSITIVE correlation with the severity of anemia ($p<0.05$; $r=0.868$). HDRS score had a significant NEGATIVE correlation with level of hemoglobin and plasma iron ($p<0.05$; $r=-0.902$). We observed a significant positive association of IDA with severity of depression among women of the reproductive age group. Hence, we suggest early evaluation of anemia and treatment of anemia should be done in patients with depression.

P-179

Genetic Variants in HBS1L-MYB Intergenic HMIP2 Block: Implication in Fetal Hemoglobin Regulation

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Increase fetal hemoglobin (HbF) level in beta-hemoglobinopathies (sickle cell anemia and beta-thalassemia) is known to have a significant impact on the progress of the diseases. In both beta-hemoglobinopathies, the level of HbF varies extensively. Research has indicated that the intergenic region between HBS1L (GTP-binding elongation factor) and MYB (myeloblastosis oncogene) contributes to nearly 20% of the overall HbF level. This intergenic region contains three linkage disequilibrium blocks- HMIP1, HMIP2, and HMIP3. Out of the three blocks, genetic variations in the HMIP2 block have been found to be significantly associated with increased HbF level. Surprisingly, the mechanism of their association with HbF level has not been investigated at length. The purpose of this study was to identify the functional annotation of the HMIP2 variants which may affect HbF level by using a computational approach. To achieve this purpose, the sequence of the HBS1L-MYB intergenic region was downloaded from NCBI. Variants in HMIP block 2 were retrieved from RegulomeDB database resource. Functional annotation of HMIP2 variants was analyzed using HaploReg and RegulomeDB. A total of 83 SNPs were reported in the HMIP2 region. Among the reported SNPs, rs9376092 and rs4895441 are likely to affect transcription factor binding. These variants are also associated with gamma-globin

genes (HBG1 and HBG2) expression based on expression quantitative trait loci (eQTL) data. As identified by HaploReg, HMIP2 variants affect the binding of transcription factors such as GATA1, GATA2, and MEF2 which are involved in gamma-globin activation. Therefore, this study provides novel insights into the association of HBS1L-MYB intergenic HMIP2 variants with HbF regulation.

P-180

Analytical Performance of Eight New Coagulation Assays on the Cobas t 411 Analyser

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Careful assessment of new laboratory methods for measurement of haemostatic function is required before routine use. We evaluated the analytical performance of eight new coagulation assays (Roche Diagnostics) on the cobas t 411 analyser, which has been developed for use in low-to-medium-throughput laboratories (100tests/hour) and can be used to perform screening and specialised coagulation tests. Repeatability, intermediate precision and total reproducibility were assessed for the following assays on the cobas t 411 analyser: antithrombin, aPTT HighS, aPTT MedS, aPTT LowS, D-Dimer Gen 2, fibrinogen, PT Screen, and thrombin time. Experiments were conducted under routine conditions at three sites (UK, Germany, and the Netherlands) following ethical approval, using anonymised residual plasma samples or commercially purchased samples. Repeatability was determined using two or three control samples and five human plasma pool (HPP) samples covering the relevant measurement range (single run; 21 replicates). Intermediate precision and total reproducibility were evaluated over five days using two or three control samples and three HPP samples (one aliquot of each sample per day). Coefficients of variation (CV) were calculated. For control and HPP samples, the range of CVs across all sites for each parameter (repeatability; intermediate precision; total reproducibility, respectively) were: antithrombin (1.5-6.1%; 2.3-9.7%; 3.6-8.1%); aPTT HighS (0.4-3.1%; 1.1-4.8%; 4.2-13.1%); aPTT MedS (0.4-2.2%; 0.5-2.9%; 0.7-3.4%); aPTT LowS (0.5-4.0%; 0.5-4.9%; 2.9-5.3%); D-Dimer Gen 2 (0.8-2.9%; 0.7-4.0%; 1.5-5.8%); fibrinogen (0.8-13.3%; 1.7-10.7%; 4.2-9.3%); PT Screen (0.4-3.9%; 0.7-2.8%; 2.0-4.9%); thrombin time (1.2-14.6%; 2.6-3.3%; 3.4-4.1%). All coagulation assays demonstrated a robust analytical performance on the cobas t 411 analyser under routine conditions.

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Method Comparison of Eight New Coagulation Assays on the Cobas t 411 Analyser

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New laboratory methods to measure haemostatic function require careful assessment before routine use. We performed a method comparison of eight new coagulation assays (Roche Diagnostics) on the automated cobas t 411 analyser, developed for use in low-to-medium throughput laboratories. Method comparisons of Roche coagulation assays on the cobas t 411 analyser versus a commercially available cobas 8000 analyser or Siemens assays (Siemens BCS, CA-7000, Sysmex CS-2100 or CS-5100i analysers) were conducted: antithrombin and D-Dimer Gen 2 cobas t 411 versus cobas 8000 analyser; aPTT HighS versus Siemens Actin FSL; aPTT MedS versus Pathromtin SL; aPTT LowS versus Siemens Actin FS; fibrinogen versus Siemens Dade Thrombin; PT Screen versus Siemens Innovin; Thrombin time versus Siemens BC/Test Thrombin. Experiments were performed under routine conditions according to CLSI EP09-A3 guidelines, at three sites (UK, Germany and the Netherlands; two reagent lots per site). Comparisons used ≥120 residual anonymised human plasma samples representing the measuring range of each analyte. Passing-Bablok analyses were performed and Pearson's correlation coefficients were estimated. Good agreement was observed versus reference methods for the antithrombin (slope, 0.98-1.13; bias at 50 IU/dL activity range, -7.2 to -2.4), aPTT HighS (slope, 0.92-1.33; intercept, -11.4 to -1.7), aPTT MedS (slope, 0.92-1.08; intercept, -3.5-0.3), aPTT LowS (slope, 1.01-1.27; intercept, -7.3 to -2.0), D-Dimer Gen 2 (slope, 0.99; intercept, 0.08-0.09), fibrinogen (slope, 0.96; intercept, 20.0) and PT screen (slope, 0.98-1.10; bias at INR 1.0, -0.05-0.08) assays; Pearson's r=0.917 (across assays). Thrombin time assay performance was as expected (slope, 0.41-1.20; intercept, -4.37-15.3), with slightly greater variation due to differences in reagent composition. On the cobas t 411 analyser, antithrombin, aPTT (high, medium and low lupus/heparin sensitivity), D-Dimer, fibrinogen and PT screen assays showed good agreement with commercially available reference methods.

P-182

Diagnosis of Iron Deficiency Anaemia Using the Reticulocyte Haemoglobin in Children Suffering from Leukemia

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Anemia is a major cause of morbidity in patients with cancer. There are multiple causative factors, including absolute iron deficiency due to blood loss and/or nutritional deficiencies, anemia of chronic disease, myelosuppressive effects of chemotherapy, metastatic infiltration of the bone marrow. The identification of iron deficiency in patients with cancer is important and difficult. Conventional tests S. Ferritin, S. Transferrin are costlier; difficult to do, affected by infection and inflammation while Ret-He is easily available and reproducible on automated cell counters. This study examined use of the hemoglobin content of reticulocytes (RET-He) to find out iron deficiency as defined by serum iron studies (transferrin saturation <20%, serum iron <40 µg/ dL, and ferritin <100 ng/mL), in paediatric leukemia patients. The goal of the present study was to establish a RET- He cutoff that could rapidly rule out iron deficiency. 60 consecutive Children uptill 16 years receiving chemotherapy in the haematoncology ward, Dept. of Paediatrics, J.K. Lon, Hospital, S.M.S. Medical College, Jaipur were investigated for serum iron, serum T.I.B.C, serum Ferritin, Total reticulocyte profile to assess iron deficiency anaemia. Morning samples were collected in plain and E.D.T.A. vials, serum analysed on Beckman Coulter AU680 and Siemens ADVIA Centaur Autoanalysers whereas reticulocyte profile and CBC were analysed on Siemens ADVIA 2120i. The data suggests that RET- He, at a threshold of 29.3 pg/cell, may be an important discriminator to rule out iron deficiency anaemia in patients with cancer.

P-183

Quantification of M-protein: Comparison between Perpendicular Drop and Tangent Skimming Method on Capillary Zone Electrophoresis

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Quantification of monoclonal protein (M-protein) is frequently determined by integration of the M-protein peak in the electropherogram. Three methods can be used to quantify the M-protein; perpendicular drop (PD), corrected perpendicular drop or tangent skimming (TS). Most laboratories opted for PD; however it tends to overestimate M-proteins at low concentrations (<10g/L). This study compared the two integration methods for the quantification of M-proteins. A total of 95 samples from patients with monoclonal gammopathy were analysed Capillarys 2 capillary zone electrophoresis system (Sebia, Issy-les-Moulinaux, France). M-protein concentrations were determined using PD and TS for each sample. Regression analysis and Bland-Altman plot were performed to evaluate the degree of agreement between PD and TS in determining M-protein concentration. Passing Bablok regression analysis showed good correlation between PD and TS methods. However, the PD showed a positive bias of 3.3 g/L when compared with TS (M protein perp. = 1.08 x M-protein tangent + 3.28). Bland Altman analysis revealed that the bias between both the methods is high especially at the lower concentration (<15g/L) of monoclonal protein. Accurate quantification of M-proteins are crucial for the follow-up of patients with monoclonal gammopathy. Perpendicular drop method tends to overestimate M-protein especially at a lower concentration probably due to background normal immunoglobulin. There is no consensus on the method to quantify M-protein by the international guidelines. Hence, it is important that the laboratory follows only one method to quantify M-protein.

P-184

Behaviour of RBC Treated with Aldehydes in Different Osmotic Conditions; Can Use for Preparation of MCV Controls

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Human RBC are highly vulnerable to different osmotic conditions. Normal plasma osmolality is 275-300 mOsm/kg, which should be strictly maintained to ensure the size and shape of RBC inside the body. Treating with aldehydes is a common method to fix and stabilise the RBC for long time storage. Aldehydes strengthen the cell wall of RBC and protect them from decaying. In normal case any cell suspended in hyperosmotic fluid will decrease its size and in hypoosmotic fluid will increase its size. But our observation is that the RBC treated with aldehydes when suspended in hyperosmotic medium increasing its size instead of decreasing and their size decreasing when suspended in hypoosmotic fluid. This property can useful to prepare RBC of desired size by suspending them in different osmotic medium and used as MCV controls. Blood drown by venepuncture in anticoagulated bottle. Red blood cells are separated by centrifugation and washing in 0.9% NaCl. Formaldehyde-Glutaraldehyde cell fixing solution added to RBC suspension and keep at 2-8°C for 12 hours. Phosphate buffer saline (PBS) with different osmolality are prepared by decreasing or increasing its salt concentration. PBS with 3 osmolality are prepared (270,335,400 mOsm/kg) and equal quantity of RBC are suspended in each. Kept

the mediums overnight at 2-8°C. MCV of each fluid are measured by using AGAPPE Hematology analyser. Red blood cells treated with aldehydes when suspended in hyperosmotic medium increases its volume and in hypoosmotic medium decreases its volume. Thus Red blood cells with desired MCV can obtain by either increasing or decreasing osmolality of suspension medium. This can be useful for preparation of MCV controls by fixing the RBC in aldehydes and suspending them in different osmotic medium.

P-185

Analytical Performance of the Lupus Screen and Lupus Confirm Assays on the Cobas t 711 Analyser

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Lupus anticoagulant (LA) is a biomarker for autoimmune disease and a risk factor for thrombosis and miscarriage, which can guide appropriate anticoagulant use. In vitro assays to screen for and confirm (Lupus Screen/Lupus Confirm [Roche Diagnostics]) the presence of LA could aid in the diagnosis of coagulopathies. We evaluate the analytical performance of the Lupus Screen/Lupus Confirm assays on the cobas t 711 analyser. Ethically-approved, residual anonymised human citrated plasma samples were evaluated using the Lupus Screen/Lupus Confirm assays (based on dilute Russell's viper venom time; cobas t 711 analyser) at three sites under routine conditions. Within-run precision (21 replicates/sample) and 5-day reproducibility were evaluated using five human plasma pools and two controls covering the measuring range. Lot-to-lot comparisons (three lots; ≥100 positive, ≥100 negative) and method comparisons (three lots; two sites; Stago STA Staclot DRVV Screen/Confirm assays on Stago STA-R Evolution analysers [Diagnostica Stago]) were conducted in a 4-field plot analysis. Total agreement was calculated; results were compared against predefined acceptance criteria. Final data were expressed as normalised ratio (NR), calculated using mean reference interval for each lot and site. For human plasma pools with mean LA NR of 1.09-2.75, ranges of coefficients of variation (CV) across all three sites for within-run precision, intermediate precision, lot-to-lot, site-to-site and total reproducibility were: 0.9-1.5%, 1.2-2.3%, 0.0-0.9%, 2.0-5.2% and 2.4-5.5%, respectively. CVs met the predefined acceptance criteria at all sites. Good correlation was demonstrated between Lupus Screen/Lupus Confirm assays and the reference method: concordance values were 90.60% (n=351), 90.21% (n=235), and 91.03% (n=223). For lot-to-lot comparability, concordance values were 98.31% (n=354), 99.58% (n=238), and 98.22% (n=225). The Lupus Screen/Lupus Confirm assays demonstrated excellent analytical performance under routine conditions, and compared favourably with commercially available assays/platforms, demonstrating suitability for use in coagulation laboratories.

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Analytical Performance of the Free Protein S Assay on the Cobas t 711 Analyser

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Protein S is a vitamin K-dependent glycoprotein cofactor necessary for the anticoagulant activity of Protein C, and plays a vital role in inhibiting excessive blood clotting. Protein S deficiency is associated with increased risk of venous thromboembolism; therefore, reliable diagnostic methods are required to measure Protein S in plasma. The Free Protein S assay (Roche Diagnostics) could facilitate accurate diagnosis of Protein S deficiency. We evaluate the analytical performance of the Free Protein S assay (cobas t 711 analyser) under routine conditions. Within-run precision (21 replicates/sample) and 5-day reproducibility were evaluated for the Free Protein S assay (cobas t 711 analyser) using anonymised human citrated plasma samples (two controls; five human plasma pools). Method comparison was performed with the Siemens Innovation Free PS Ag assay (Siemens Sysmex CS-5100 analyser; n=120 samples covering the measuring range), using Deming regression analyses. Lot-to-lot comparability was determined according to Passing-Bablok regression; Pearson's correlation coefficients were estimated. Results were compared against predefined acceptance criteria. For human plasma pools with mean free Protein S concentration 11.5-128.0 IU/dL, ranges of coefficients of variation (CV) across all three sites for within-run precision, intermediate precision, lot-to-lot, site-to-site and total reproducibility were: 0.4-1.0%, 0.9-2.1%, 0.4-2.0%, 0.3-3.0% and 1.0-3.4%, respectively; all were within accepted ranges. Good agreement was observed between the Free Protein S assay and the reference method; Pearson's r values were 0.998 (n=145), 0.998 (n=124), and 0.997 (n=179). Lot-to-lot comparability measurements were also within accepted ranges; Pearson's r values were 0.999 (n=145), 0.998 (n=124), and 0.999 (n=179). These results demonstrate the excellent analytical performance of the Free Protein S assay, supporting its use as an accurate and reliable diagnostic tool. Good correlation with a commercially available assay further demonstrates the suitability of this assay for use in clinical practice.

P-187

Proposal of a Fibrinogen Demand Management in Our Emergency Laboratory

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The increase in the demand of analytical tests beside health care costs associated has revealed the need to improve their management in order to reduce the number of unnecessary tests. In our emergency laboratory, a protocol has been proposed to remove coagulative fibrinogen determination of the "urgent coagulation profile" when it is unnecessary unless it could be requested by the physician. The aim of the study is to determine the number of pathological fibrinogen results that would justify the realization of this demand management. Retrospective study based on the determination of fibrinogen test (coagulative and calculated, Siemens) and C-Reactive Protein (CRP) (Beckman-Coulter) during a period of six months. Data were obtained using LIS Modulab (Werfen®). Data analysis were performed using regression analysis (Microsoft Excel). Of a total of 21.950 fibrinogen results, 18.235 (83,1%) were processed urgently. 13.345 (73,2%) determinations presented a pathological result (<200 mg/dL or >350 mg/dL): 4,3% due to a fibrinogen deficiency and 68,9% because of high results. Because one cause of the increase of fibrinogen levels is due to acute phase reactant, the results of the CRP were analyzed, so that 89,7% of high fibrinogen results coincide with values of pathological CRP (>5 mg/L). A good concordance of both coagulative and calculated fibrinogen results was obtained, being the linear regression coefficient R = 0.9324. A high percentage of elevated fibrinogen results is associated with high CRP values, showing that fibrinogen is an acute phase reactant. These results could allow us to exclude the coagulative fibrinogen of the "urgent coagulation profile", decreasing signficantly the demand for this test besides a drastic reduction in the laboratory's costs. With the good concordance between fibrinogen methods; coagulative assay could be replaced by calculated fibrinogen in the "urgent coagulation profile", without losing analytical information for physicians.

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The Role of Platelet-to-Mean Platelet Volume Ratio (PMPV-R) as Hematological Markers in Predicting Early Mortality Among Children with Sepsis

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Early recognition and treatment are playing a pivotal role in preventing the progression of sepsis among children. Complete Blood Count (CBC) test are the initial preferred tests usually use prior to doing the particular assessment for sepsis. This study aims to investigate the predictive value of platelet-to-mean platelet volume ratio (PMPV-R) as hematological markers to the risk of early mortality among children with sepsis. A consecutive technique sampling was carried out among 74 children with sepsis using retrospective cross-sectional study approach during 2018 period at Sanglah General Hospital, Bali, Indonesia. CBC tests were evaluated to determine the association of PMPV-R with the early mortality risk using Haematology Analyzer SYSMEX XT-4000i prior to appropriate medication provided. The death cases were divided into early (<48 hours) and late (>48 hours) mortality. Data regarding age, gender, and CBC assessment were analyzed in mean, standard deviation, median, confidence interval, as well as ROC and logistic regression using SPSS version 25 for Windows. There is no significant difference with age, pH, and most white blood cells (WBC) parameters between groups (P>0.05). Several parameters such as neutrophil, mean platelet volume (MPV), PMPV-R, and platelet (PLT) show a significant difference in both groups (P>0.05). PMPV-R has a statistically significant area under the curve (AUC) (0.804; 95% CI 0.690-0.917; P=0.000), followed by PLT (0.760; 95% CI: 0.639-0.881; P=0.001) and MPV (0.761; 95% CI: 0.649-0.873; P=0.001) to the risk of early mortality in sepsis. Logistic regression test using particular cut-off for PMPV-R (32.1; 78.9% Sensitivity and 70.9% Specificity) show a statistically significant results (Adj.OR: 9.141; 95% CI: 2.627-31.809; P=0.001) compared with others. High level of PMPV-R might have a role as a simple hematological marker in predicting early mortality among children with sepsis.

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Ethnically Specific Genetic Variations in Indonesia

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Genetic mismatching between donor and recipient is always a concern during blood transfusion. Many studies showed that other important antigens besides ABO and Rhesus blood groups also play role in compatibility for transfusion procedures, such as human platelet antigens, human leukocyte antigens (HLAs), and other blood group antigens (Kell, Kidd, Duffy, and MNS). These antigens allow transfusion to suceed, especially for patients with multiple antibodies due to prior transfusion or patients with highly active immune system. Furthermore, these antigens are highy polymorphic and able to define population origin as well. Therefore, genotyping these clinically relevant blood group (Kell, Kidd, Duffy, and MNS) in Indonesian population will be beneficial to determine the specific genetic variations in the population and might support safer and personalized transfusion in the future. Genotype data from 1278 subjects of Indonesian population was obtained by using Infinium Asian Screening Array-24 v1.0 (Illumina, USA). The data was clustered using filter of single nucleotide polymorphisms (SNP) specific for Kell, Kidd, Duffy, and MNS blood group antigens. The prevalence of each blood group antigens in Indonesian population are determined. 99,84% (1274/1276) of Indonesian population harbor genetic variants for k/k and Kp(a-/b+) antigens of Kell group, 84,27% (1077/1278) for Fyx-, Fy(a+b-), and Fyx- antigens of Duffy group, 100%(1278/1278) for s/s antigens of MNS group, and 44,2% (565/1278) for Jk(a+/b+) antigens of Kidd group. Indonesian population has specific genetic variants of blood group antigens. Therefore, blood group genotyping can serve as personalized compatibility testing for transfusion in Indonesia.

P-190

Evaluation of Serum Adropin Levels in Type 2 Diabetic Patients and its Correlation with Insulin Resistance

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Adropin is a newly identified regulatory protein encoded by Enho gene (energy homeostasis associated) in the brain and liver and has a role in the energy homeostasis and insulin resistance. To evaluate serum adropin level in type 2 diabetic patients and its correlation with diabetic related parameters. Blood samples were collected from 50 type 2 diabetic patients and 50 healthy individuals, age and sex matched matched with patients, as controls. Adropin level were found to be significantly lower in type 2 diabetic patients compared to healthy subjects. Adropin was inversily correlated with Fasting blood sugar in type 2 diabetic patients and was also negatively correlated with HOMA-IR. Type 2 diabetic patients have lower adropin levels and serum adropin is inversely correlated with insulin resistance; this indicates a close association between adropin and type 2 diabetes mellitus.

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An Unusual Case of Monoclonal Gammopathy in a 12 Year Old Male

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Plasmablastic Lymphoma (PBL) is a rare and aggressive haematological malignancy usually associated with HIV infection. This malignancy is immunophenotypically related to Multiple Myeloma. It is rare in paediatric patients with only a few cases having been described. We present a 12-year-old newly diagnosed HIV-positive male on treatment, who presented with a pathological fracture of the femur. On physical examination the patient was found to have soft tissue tumour on the chin. Laboratory investigations showed a microcytic hypochromic anaemia with a haemoglobin of 9.6 (reference range 10.3-15.5 g/dL), β-2 microglobulin of 4 (reference range 1.1-2.5 mg/L) and raised inflammatory markers. An SPE and immunofixation showed an unquantifiable monoclonal band that was typed as IgA Lambda. A BMAT revealed depleted iron stores and 8 % plasma cells that

appeared dysplastic and reactive. Radiological investigations demonstrated the presence of a right proximal tibial and femoral mass located where the pathological fracture occurred. Histology of the biopsies taken of the lesions demonstrated large cells, most of which had a plasmacytoid appearance with multiple prominent basophilic nucleoli and eccentric nuclei, prominent apoptotic bodies and brisk mitotic activity, features consistent with PBL. The tumour cells were positive for CD138, MUM1 and CD56 by immunohistochemistry. The proliferation marker (Ki-67) approached 100%. The CD20, CD79a, Bcl2 and Bcl6 proved negative. EBER-ISH (Epstein-Barr virus-encoded RNA in situ hybridisation) yielded a positive result. Plasmablastic lymphoma should be suspected in any paediatric patient with pathological fractures in the background of an immunocompromised state. At present, evidence of a monoclonal band is not part of the diagnostic criteria for PBL. This study has demonstrated the potential value of SPE and immunofixation in children who present with pathological fractures.

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Study of the Sensitization to Pneumoallergens in the Southern Area of Spain

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The determination of specific IgE with immunoassays is a highly sensitive and specific method that is used as a screening method in patients with suspected sensitization to pneumoallergens. The objective of our study is to evaluate the incidence of sensitization to pneumoallergens through the screening test and the percentage of positive results for each of the panels and individual allergens carried out in our geographical area (Granada - Spain). Retrospective study based on the determinations of pneumoallergens in the Allergy Laboratory of our hospital during the first quarter of the year 2019. The samples were analyzed in the Immulite 2000XPi (Siemens) autoanalyzer in which allergen-specific IgE assays are based on a chemiluminescent detection. During the period study; 6093 determinations of specific IgE to pneumoallergens were performed, which was positive in 2314 patients (38%). In these patients, panels and individual positive allergens were: Timothy grass (g6): 57% (1123 of 1967 determinations), Parietaria (w19): 37% (543 of 1972 determinations), Alternaria alternata (m6): 17% (358 of 2097 determinations), Dermatophagoides pteronyssinus (d1): 35% (707 of 2000 determinations),Cat dander (e1): 52% (1022 of 1963 determinations),Dog dander (e5): 32% (635 of 1955 determinations),Olive (t9): 85% (1952 of 2314 determinations), Platanus acerifolia (t11): 19% (430 of 2314 determinations),Cypress (t23): 58% (1351 of 2314 determinations) .In our geographical area we highlight the high incidence of allergy to olive (85%), cypress (58%) and grasses (57%). The lowest percentages of sensitivity are found for alternaria alternata (17%) and platanus acerifolia (19%).

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Association of TLR-2 Polymorphism with Ankylosing Spondylitis: A Hospital Based Case-Control Study

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Ankylosing spondylitis (AS) is an inflammatory disorder where innate immunity has an important role in pathogenesis. Recently, a 23 bp ins/del polymorphism at 5'UTR of TLR gene has been documented, which is associated with high TNF-alpha levels (a major inflammatory marker in pathogenesis of AS). In this study we investigated the association of TLR-2 (23 bp ins/del) polymorphism and clinical severity in AS patients from Odisha, India. AS patients (n=100) fulfilling ASAS criteria for axial and peripheral spondyloarthritis were enrolled along with 100 healthy age matched controls from similar geographical areas. BASDAI and BASFAI were recorded. TLR-2 (23 bp ins/del: rs111200466) polymorphism was studied by DNA extraction and PCR, followed by agarose gel electrophoresis and visualisation by Gel documentation system.All patients enrolled in the present study were males. The mean age of AS patients and healthy controls was 31.21±11.43 and 28.28±9.62 years, respectively. At the time of enrolment, mean disease duration of patients was 2.07±1.13 years. BASDAI and BASFI scores were above 5. Distribution of TLR2 (23 bp ins/del) polymorphism was in accordance with Hardy-Weinberg Equilibrium. Prevalence of del/del genotype was significantly higher in AS patients compared to healthy controls (P=0.01, OR=5.65), indicating a possible contributory role of TLR2 on predisposition to AS. Distribution of heterozygous genotype (ins/del) and minor allele (del) were comparable among different clinical categories. Furthermore, no significant association of TLR-2 polymorphism was observed with disease severity. TLR2 5'UTR homozygous mutants (23 bp deletion) were significantly associated with patients of AS but not with disease severity. Larger sample size in multicentre will lend validity to the observation.

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Comparison of Immunoglobulins Study from Turbidometric and Nephelometric Methods

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The principle used in this investigation is to study the formation of Ag-Ab complexes by nephelometric (light scattering) and turbidometric (light absorbance) measurements and to assess the two methods. In nephelometry the scattered light is measured which leaves the solution at an angle other than that of the incident beam. Turbidimetry refers to the measurement of the transmitted light at the same wavelength and direction as the incident beam. We wanted to study the correlation between the results of the two tests and assess the suitability of using the method interchangeably. We used Siemens Dimension RxL analyser for turbidimetric studies at ACTREC and Beckman Coulter Immage for nephelometric studies TMH. The results were analysed for correlation using Passing-Bablock mehod and for bias using Bland Altman Plots. 'R' value correlation between nephelometry & turbidimetry for IgG, IgM, IgA were 0.985, 0.994 & 0.994 respectively. Bias was observed between the two methods as 9.5 units,-23 units and 9 units for IgM, IgG and IgA respectively. The results of this investigation reinforce the findings that both nephelometry and turbidimetry are suitable techniques for the assay of immunoglobulins. However, the bias showed in the results of two methods should be evaluated and correlated clinically.

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(K+l) Index as a New Measure of Disease Status in Rheumatoid Arthritis

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Rheumatoid Arthritis (RA) is a systemic inflammatory autoimmune disease characterized by chronic polyarthritis and bone destruction. B cells are implicated in the inflammatory events of RA by producing antibodies and a polyclonal excess of serum free light chains kappa (K) and lambda (L). Thus, the sum of the serum levels of K and L reflects B cells activation and the inflammatory activity of the RA. This study aims to compare the (K+L) index between healthy controls and RA patients, to compare between seropositive (SP) and seronegative (SN) RA patients and also to correlate (K+L) index with ACR2010 score (criteria for earlier diagnosis of RA) and inflammatory marker hsCRP. Study is based on 68 subjects (24 SN, 19 SP and 25 healthy controls) with a mean age of 44.2±11 years (F:M=57:11). Disease activity of RA was evaluated using ACR 2010 score. The cases included were having a score of ≥6 indicating active disease. Serum free light chains Kappa and Lambda were analyzed by using mmunoturbidimetry method (Binding site kits, UK) and hsCRP was analyzed also by immunoturbidimetry method. Serum levels of (K+L) index in patients with RA were significantly higher than healthy controls, 66.0 (59.0 - 69.39) vs 36.28 (33.07 - 40.85) mg/L respectively ($p<0.0001$). The area under curve was 0.94 (95% CI: 0.872 to 1.00). However there is no difference between SP and SN cases. A good linear correlation was found for (K+L) index with ACR 2010 SCORE ($r=0.63$; $p<0.0001$) and hsCRP ($r=0.469$; $p<0.0004$). The levels of serum "(K+L) index" could be useful to differentiate RA cases from healthy controls. However it is not useful to discriminate between SN and SP. The (K+L) index was found to be associated with disease severity as well as inflammation.

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High IgE Level and Eosinophil Count: Clinical Correlation

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IgE is a class of antibody that plays an important role in allergy and is especially associated with type I hypersensitivity. It also plays a central role in the pathophysiology of chronic inflammatory allergic diseases. In genetically susceptible individuals, exposure to specific allergens results in an increase of specific IgE. These can bind onto effector cells through a high affinity receptor known as FcεRI expressed in mast cells and basophils. IgE determination is valuable in the diagnostic assessment of patients with established or suspected allergic diseases such as asthma, allergic rhinitis, eczema, urticaria, atopic dermatitis and some parasitic infections. This study was undertaken to evaluate the clinical profile of patients with increased Ig E level and find its association with eosinophil count. This descriptive study was conducted in the department of Biochemistry, IGIMS, Patna over a period of six months. 100 samples with high IgE value were selected at random. Patients clinical profile and other relevant investigations were searched out and evaluated. IgE estimation was done on AU 5800 taking care of all the quality control measures. Results are expressed as mean ± 1SD. The mean age of the patients with high Ig E level were 39.8 ± 16.8 years. 54% were males and 46% were females. 90% of the patients were of >18 years of age. Most common clinical diagnosis of the patients with high IgE level were Atopy with or without asthma. The eosinophil percentage of patients with extremely high IgE values (>30000 IU/ml) were found to be greater

than 50 percent. Among the various causes for high IgE level in the adult age group, atopy is the most common with diverse clinical presentation.

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Elevated Circulating Th17 Count Points Towards Th17 Dependent Neuro-inflammation in MDD

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Recent advances in neuro-immunology has led to compelling evidence that immuno-inflammatory pathways may also be involved in the etiology and pathophysiology of Major depressive disorder (MDD), a chronic neuropsychiatric disorder with a high lifetime prevalence all over the world. Common depression models in mice showed elevated levels of Th17 and Th1 cells in the mouse brain. However, the clinical studies investigating Th17 cells in MDD have not reached consensus in their results. With this background, the current study was carried out to assay the circulating Th17 cell count in major depressive disorder patients. The study population included 53 patients of first episode, drug naïve MDD patients recruited from the out-patient department of Psychiatry. The diagnosis and classification of patients were done as per the ICD-10 classification of MDD and Hamilton Depression Rating Scale. 53 non psychiatric volunteers satisfying the exclusion criteria were taken as healthy controls in the study. General Health Questionaire-12 (GHQ-12) was used to assess suitability of controls with a score of < 2 taken as the cutoff for inclusion. Th17, Th1 and Treg cell counts were assessed by flowcytometry. The mean percentage of Th17 cells in cases (1.87 ± 1.03) was significantly higher than the mean in controls (1.13 ± 0.94) (p value < 0.001). There was no statistically significant difference in the Treg counts between the cases (2.12 ± 2.0) and controls (2.27 ± 0.95). But mean count was higher in the controls compared to the cases. The Th17: Treg ratio was significantly higher in cases (5.41 ± 10.84) compared to the controls (0.69 ± 0.89) (p value < 0.05). In conclusion, as per the hypothesis of the study, it was seen that circulating Th17 count and Th17: Treg ratio were elevated in MDD cases compared to controls.

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Effect of Lead on Immunoregulatory Cytokines in Occupationally Exposed Workers

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Occupational and environmental exposure to metals can adversely affect human health. Lead is a common and abundant toxic heavy metal present in the inert environment and in all biological system. Exposure to lead have several detrimental effects on nervous, cardiovascular, skeletal, renal and hemopoietic system of body. However, little is known about the influence of lead on immune system in human population. The aim of this study was to examine blood lead levels in occupationally exposed individual and to correlate with serum IL-2, IL-4, IL-17 levels. The study comprises of 50 male individuals (aged 30-61 years) working in factories (handicraft, welding) with occupational exposure for at least 2 years and 30 apparently healthy subjects with no occupational exposure were recruited in the study. A written consent was taken from each subject in the study. Blood lead level (BLL) served as biomarker for lead exposure and was determined using Dual Atomic Absorption Spectrophotometer (ICE 3500 Thermofisher). Serum cytokine levels (IL-2, IL-17, IL-4) were determined using commercially available ELISA kits. A statistically significant higher blood lead levels were observed in the exposed group when compared to the non-exposed group ($p<0.05$). Among the immunoregulatory cytokines a statistically significant negative correlation was found between the blood lead levels and IL-2 ($r=-0.250$, $p=.011$) in all the exposed subjects when compared with the non-exposed subjects. On the other hand, serum IL-17, IL-4 did not show any statistically significant difference in the levels when compared between the two study groups. Our results, suggests that occupational exposure to lead may affect the immune system of host by altering the levels of various immunoregulatory cytokines in the body.

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Serum Immunoglobin E (IgE) Estimation as a Prognostic Marker of Allergic Rhinitis by ARIA Classification

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Allergic rhinitis represents a global health problem which is defined clinically by the symptoms caused by immunologically mediated inflammation after the exposure of the nasal mucous membranes. The recommendations of Allergic Rhinitis and its Impact on Asthma (ARIA) classification provide evidence-based guidelines for management of allergic rhinitis worldwide. The development of allergy involves production of specific antibody called Immunoglobin E (IgE). Hence, serum IgE estimation is routinely utilized as biochemical marker in allergic rhinitis management. However, the utility of serum IgE in updated ARIA classification protocol is still a matter of debate. This study was undertaken to assess this relationship of serum IgE levels with symptom severity assessed by ARIA classification. Patient clinically diagnosed to be suffering from allergic rhinitis were enrolled in the study and their symptom profile was categorised as per ARIA classification. Serum IgE levels were measured in these patients and statistically interpreted in relation to severity of symptoms. We noted a statistically significant correlation between elevated levels of serum IgE and increased severity and duration of allergic rhinitis. We thus conclude that serum IgE can serve as a low cost, effective and clinically relevant investigative tool to inform and guide clinical decision makers in accordance with ARIA guidelines.

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Rates of Positive Oligoclonal Band in Western Singapore

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Cerebrospinal fluid (CSF) Oligoclonal bands (OCB) are used for diagnosis of Multiple Sclerosis (MS). There are limited data about presence of CSF OCB in Asians with MS and how it affects diagnosis, treatment and follow up. We reviewed prevalence of CSF OCB detected in western Singapore in a cross-sectional study. CSF OCB requests to our laboratory in 2018 were reviewed, with only the first test result included in analysis. CSF OCB were reported as positive, negative, identical CSF OCB (to that in serum) or monoclonal OCB. CSF OCB were performed using Sebia Isoelectric focusing electrophoresis. There were 225 CSF OCB requests in 2018, mean age(±2SD) was 43(±38) years old with Female:Male ratios of 1.18. The percentage of Chinese: Malay:Indian:Other ethnic groups was 61%:8%:9%:22%. There were 28% identical CSF OCB with mean age of 48 years old, Female:Male ratio of 0.68 and ethnic distribution of 59%:6%:11%:24%. There were 55% negative CSF OCB with mean age of 42 years old and Female:Male ratio 1.20. The ethnic distribution was 62%:11%:9%:18%. There was no monoclonal OCB reported. There were 17% CSF OCB positive results. Mean age for CSF OCB positivity was 39 years old, with Female:Male ratio of 3.22 and ethnic distribution of 61%:2%:8%:29%. Upon dividing the age distribution into tertiles, the peak age group for positive CSF OCB was between 21-30 years old. There were 71% locals and their mean age was 40 years old, compared to 36 in non-locals.CSF OCB positivity peaked between 21-30 years old and this matches international epidemiology data for MS. Patients without CSF OCB were older, had lower proportion of females and other ethnic groups and higher percentage of Indians.Further studies looking at correlation between CSF OCB positivity and diagnosis of MS and case control review of ethnic differences may be helpful.

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Acetylcholine Receptor Antibody Testing in Singapore

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Anti-Acetylcholine Receptor Antibodies (ACAB) is used for diagnosis of Myasthenia Gravis (MG). Seropositivity in Asians are unknown and affects final diagnosis, treatment and prognostication. We reviewed ACAB results in western population of Singapore cross-sectionally in 2013 and 2018. All ACAB requests to our laboratory in 2013 and 2018 were reviewed together with demographic details. For patients with more than one visit in the same year, only initial result was included. ACAB results were divided into positive, borderline positive and negative results. ACAB was performed using radioimmunoassay on IBL International. There were 731 and 1131 ACAB requests in 2013 and 2018 respectively. Mean age (±2SD) were 55 (±36) and 59 (±34) years old in 2013 and 2018 respectively, with Female:Male ratios of 1.07 and 0.86, and distribution of Chinese: Malay:Indian:Other ethnic groups at 78%:6%:7%:9% and 78%:7%:6%:9% respectively. Comparing 2013 and 2018, there were 18% and 12% ACAB positive results, and 1% and 1% borderline positive results respectively. Mean age for ACAB positivity were 56 and 61 years old and for ACAB borderline positivity were 46 and 64 years old. Female:Male ratio were 0.90 and 1.09 for seropositive and 0.33 and 0.60 for borderline results respectively. Ethnic distribution was 80%:6%:5%:9% and 80%:7%:3%:10% for seropositive and 50%:0%:38%:12% and

90%:12%:0%:0% for borderline results. The percentage of seropositive results requested from outpatient eye and neurology clinics were 18% and 13% compared to 20% and 14% in other locations in 2013 and 2018 respectively. In ACAB seropositive patients, mean antibody concentrations were 49.7 and 35.3 nmol/L for females and males in 2013 and 57.3 and 45.0 nmol/L in 2018.There was no difference in age, gender or ethnic distribution in seropositive ACAB in 2013 and 2018. Females have higher antibody readings than males and further studies are needed to review ACAB results against clinical diagnosis.

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Prevalence of HLA B27: A Case Study Among the Spa Samples from North Kerala

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Human Leukocyte Antigen B27 (HLAB27), an allomorph of MHC class 1B molecule is known to be strongly associated with the development of a group of inflammatory rheumatic diseases known as spndyloarthritides (SpA). The present study aims to inspect the HLAB27 status of samples collected from clinically suspected SpA patients at AZA diagnostic centre clinical laboratory, calicut. Real Time PCR (RT PCR) for HLAB27 gene was performed on SpA patients of 100 whole blood samples. Only seven of the samples between age groups 34-62 came out to be positive for HLAB27 gene. Further, only the samples collected from male patients with ankylosing spondylitis (AS) were positive for HLAB27. Our findings confirm the earlier reports on male preponderance to HLAB27 associated AS.

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Association of Elevated Matrix Metalloproteinase-9 and Interleukin-17 with Psychological Stress in Men with Abnormal Semen Parameters

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To assess the association between psychological stress with MMP-9 and interleukin-17 (IL-17) in men with abnormal semen parameters. 78 men who attended infertility clinic along with their spouses were included in the study. Based on semen analysis, they were divided into two groups (normal and abnormal semen parameters). MMP-9 and interleukin-17 were estimated in all the subjects. Psychological stress was assessed using perceived stress scale. MMP-9 and IL-17 were significantly increased in men with abnormal semen parameters compared to those with normal semen parameters. MMP-9 was positively correlated with IL-17 and perceived stress scale MMP-9 is elevated in men with abnormal semen parameters and is associated with inflammation and psychological stress.

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Evaluation of C3 and C4 Complement Factors in Children with β-Thalassemia Major in East Delhi, India

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Thalassemia is the most common single gene disorder in India. β thalassemia major is one of the chronic haemolytic anaemias resulting from defect in β globulin chain. It requires frequent blood transfusions plus other treatment modalities. These treatment modalities are associated with various immunological modulations. The present study was therefore aimed to assess the complement status of children with β thalassemia major. 40 β thalassemia patients admitted to the thalassemia ward of CNBC Hospital, New Delhi were included as cases and 40 healthy age and sex matched children were taken as control. CBC plus serum levels of ferritin along with complement factors C3 and C4 levels were measured. The mean age of the patients was 8.06 ± 2.4 years with a male female ratio of 3:2. The average age of onset of disease was 1.5 ± 0.3 years. The study showed a significant reduction in serum levels of C3 and C4 in patients compared to controls (p value < 0.001). When C3 and C4 levels were compared with respect to duration of blood transfusion, levels were found to be significantly decreased in children receiving blood transfusion for more than 5 years in comparison to those receiving blood transfusion for a shorter period. On comparing levels of C3 and C4 with serum ferritin a significant decrease in both levels of complement was observed ($p < 0.001$ and $p<0.05$ respectively) for children with high serum ferritin levels (>2500 ng/mL) in comparison to those with low ferritin levels < 1500 ng/ml. Decreased synthesis or increased consumption of complement factors in patients receiving blood transfusion might lead to continuous contact between the immune system and various antigens. This may result in nonstop use of complement factors, recurrent infections, alterations in the parameters of the immune system due to iron overload through repeated blood transfusions.

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Aerobic Bacterial Profile and Antibiotic Resistance During Tropical Transition Weather in Ibnu Sina Hospital Gresik Indonesia

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Indonesia lies right on the equator with sunshine throughout the year and the air temperature ranges between 25-36oC. This study aimed to determine the aerobic bacterial profile and antibiotic resistance during weather transition from rainy to dry season.The study was conducted during February 2019. Data were obtained from a number of culture results in Department of Clinical Pathology Ibnu Sina Hospital Gresik Indonesia. Bacteria were isolated from blood, urine, abcess, steril body site, and respiratory samples. Identification and antibiotic susceptibility pattern were determined using BD Phoenix automated microbiology system (BD Diagnostic Systems, Sparks, MD).The culture results were positive in 19 (17.6%) of 108 specimens. The positive culture results specimen were 8 abscess, 5 blood, 3 respiratory, 2 urine, and 1 sterile body site. The predominant bacterial isolates were Escherichia coli (36.7%), followed by Staphylococcus haemolyticus (21.0%). Other bacterial isolates were Enterobacter aerogenes (5.3%), Enterobacter cloacae (5.3%), Staphylococcus epidermidis (5.3%), Streptococcus acidominimus (5.3%), Morganella morganii (5.3%), Rothia mucilaginosa (5.3%), and unidentified organism (10.5%). Multidrug resistance was detected in 10 (52.6%) bacterial isolates. Beta-lactamase was detected in 50% of Staphylococcus haemolyticus. Methicillin-resistant Staphylococcus species (MRSS) was detected in 66.7% of Staphylococcus haemolyticus and 33.3% of Staphylococcus epidermidis. Macrolide-Lincosamide-Streptogramin B (MLSB) resistance was detected in 100% of Staphylococcus haemolyticus. Extended Spectrum Beta-Lactamase (ESBL) was detected in 100% of Escherichia coli. The present study confirmed the high prevalence of multidrug-resistant pathogens during tropical transition weather. It is important for physicians practicing in temperate climates to evaluate the different microorganisms infecting in hospitalized patients and to know the antibiotic susceptibility patterns of the isolates.

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Multidrug Resistant Bacteria in Hospital Wards of Tertiary Care Hospital and the Antibiogram of the Isolates

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Multidrug resistant (MDR) is antimicrobial resistance shown by a species of microorganism to multiple antimicrobial drugs. These MDR bacteria are frequently associated with infections in the patients admitted to critical units of hospitals. Infections caused by gram-negative bacteria have features that are of particular concern. These organisms are highly efficient at up-regulating or acquiring genes that code for mechanisms of antibiotic drug resistance, especially in the presence of antibiotic selection pressure. This cross sectional study was conducted in the microbiology department of Manmohan Cardiothoracic Vascular and Transplant Center (MCVTC), a pioneer public hospital in cardiac and vascular surgery, of Nepal. The total of 848 blood samples for culture was received from 15th June to 15th December. The samples were collected and kept in bottle containing brain heart infusion broth in the ratio of 1:9 in sterile condition and incubated at 37°C and sub-cultured in blood agar and macConkey agar plates the following days until 96 hours in conventional way. Antibiotic sensitivity testing was performed by disc diffusion method and antibiogram was reported based on Clinical and Laboratory Standards Institute (CLSI) susceptibility criteria. Out of total 848 blood samples, 78 samples (9.1%) showed growth of organisms. Among various organisms were isolated, B. cepacia complex and k.pneumoniae were the major isolates from ICU, CCU and GWD wards following the others. Majority of these isolates (56%) were MDR strains. Most of the MDR bacteria showed 100% sensitive to Polymyxin B and colistin sulphate antibiotics while Imipenem and meropenem were moderately sensitive. B.cepacia complex was 100% sensitive to Tegicycline and 88% to cotrimoxazole. Thus this study concluded that Gram negative bacteria are the major pathogens in blood culture and MDR strains are isolated from hospital wards with limited antibiotic choice.

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Diagnostic Evaluation of Presepsin and Procalcitonin in Sepsis Patients

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Sepsis and septic shock represent major challenges of modern intensive care medicine. Early diagnosis and treatment is still the best method. However, there is a great lack of evidence for biomarkers to reliably diagnose and predict the future course of patients suffering from sepsis. Objectives of this study are to evaluate the change in levels of Presepsin (PSEP) and Procalcitonin (PCT) with treatment and to compare the diagnostic performance of PSEP and PCT in Sepsis cases. This study includes consecutive patients admitted to Acute Medical Care Unit with clinical criteria of Sepsis from September 2017 to February 2018. The study protocol was approved by the institutional ethics committee. Blood samples were collected on Day 1 and Day 5 of admission. PCT was estimated by ECLIA and PSEP by sandwich ELISA Kit. SOFA score was calculated on Day 1 and Day 5.Cases were divided into culture positive and culture negative patients based on the blood culture report. Age and Gender matched apparently healthy subjects were taken as control group- 30 controls. The statistical analysis was done by Med calc software. Results showed that there were 14 culture positive cases and 36 culture negative cases. Levels of PSEP and PCT are significantly higher in culture positive cases on both DAY 1 and DAY 5. There is decrease of levels of Presepsin from DAY 1 to DAY 5 in both the groups. PSEP has higher AUC - 0.843, with a sensitivity of 71.43% and specificity of 91.67% when compared to PCT - AUC-0.651, sensitivity - 64.29% and specificity of 75%. In conclusion higher levels of Presepsin are found in culture positive sepsis patients both on days 1 and 5. PSEP showed better diagnostic performance compared to PCT. PSEP levels decreased with treatment in both culture positive and culture negative cases of sepsis.

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Role of Regulatory Proteins Involved in Iron Homeostasis in Pulmonary Tuberculosis and Household Contacts Compared to Healthy Controls

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Early identification of active tuberculosis (TB) among high risk household contacts could limit new transmission and better clinical outcome thus decreasing TB burden. As iron is critical for M.tb growth in macrophages, proteins involved in iron homeostasis could form important biomarkers as risk factors for development of TB. In the present study, iron homeostasis (hemoglobin, iron, ferritin, hepcidin, ferroportin and transferrin) was assessed in blood serum samples & PBMCs of 50 active TB cases, 50 household contacts and compared with 50 healthy controls. The hemoglobin was assessed by using automated cell cytometer, hepcidin & transferrin was assessed using ELISA, iron levels were measured using kit based method, ferritin was estimated using chemiluminescence based assay, the expression of ferroportin was assessed using western blot. Iron homeostasis differed between active TB and household contacts with significantly lower hemoglobin levels despite optimum serum iron levels in active TB compared to household contacts and healthy controls pointing towards anemia of inflammation. Significantly higher serum hepcidin and ferritin concentration along with lower monocyte ferroportin expression was observed in active TB compared to both household contacts and healthy controls due to IL-6 induced hepcidin production in TB. Transferrin levels were found to be significantly lower in active TB patients and household contacts as compared to healthy controls owing to higher ferritin levels (pseudo iron overload) in active TB group. In conclusion, upon infection, regulation of iron absorption is disturbed via increased hepcidin levels which leads to ferroportin internalization and thus inhibition of iron export from enterocytes and macrophages. Sequestered iron in macrophages in turn plays important role in TB pathogenesis. Hepcidin along with ferritin and transferrin could be used as marker for activation of TB in household contacts. However follow up studies are required in household contacts to determine clinical utility of monitoring iron homeostasis markers.

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Candida Isolates Causing Refractory or Recurrent Oropharyngeal Candidiasis in 11 Hospitals in China

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We studied the species distribution and antifungal susceptibilities of Candida isolates causing refractory or recurrent oropharyngeal candidiasis (OPC) in a multicenter study in China (2013-2016). Species identification was performed using the Bruker Biotyper (Bruker Daltoniks, Germany) matrix-assisted laser desorption/ionization time of flight mass spectrometry (MALDI-TOF MS) system supplemented by internal transcribed spacer (ITS)sequencing as required. Antifungal susceptibilities were determined by Clinical and Laboratory Standards Institute document (CLSI) M27-A3 broth micro dilution methodology. A total of 558 non-duplicate Candida isolates comprising 10 species were obtained from 535 patients. Candida albicans was the most common species (89.6%), followed by C. glabrata (5.2%), C. tropicalis (2.9%) and C. parapsilosis (0.7%). Azoles were active against C. albicans with susceptibility rates of 96% and 95.8% for fluconazole and voriconazole, respectively. MIC50values of C. albicans to fluconazole, voriconazole, itraconazole and miconazole were 1 μg/ml, 0.03 μg/ml, 0.25 μg/ml and 0.12 μg/ml, respectively, higher than those in previous studies of which OPC patients (corresponding MIC50 values of 0.25 μg/ml, 0.015 μg/ml, 0.06 μg/ml and 0.03 μg/ml). Except for itraconazole, the MIC50 and MIC90 values of 58 non-C. albicans to other azoles were 2 to 3-fold higher than C. albicans. Miconazole, amphotericin B, nystatin and 5-flucytosinehad good in vitro antifungal activity for all isolates. The study provides valuable data on the species distribution and antifungal susceptibility of oropharyngeal Candida isolates from geographically-diverse areas of China. C. albicans remains the most common species but with increasing rates of azoles resistance.

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Epidemiolozgy and Antifungal Susceptibility Patterns of Invasive Fungal Infections from 2012 to 2014 in a Teaching Hospital in Central China

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As a participant organization of the national China Hospital Invasive Fungal Surveillance Net (CHIF-NET) program, the present study sought to describe the epidemiology and antifungal susceptibility patterns of yeast isolates obtained from invasive fungal infection patients presented to the First Affiliated Hospital of Zhengzhou University in central China. This study includes a total of 434 yeast isolates recovered from blood and other sterile body ?uids were defined by the matrix-assisted laser desorption ionization-time of flight mass spectrometry supplemented by DNA sequencing as required. Antifungal susceptibilities were determined by Sensititre Yeast OneTM YO10 methodology. This study reported that C. albicans remained the most common species (33.9%) but with significantly decreased frequency from 37.2% to 27.7% C. tropicalis as the second pathogen, its overall isolation rate reached up to 25.1%, followed by the C. parapsilosis complex (17.3%), C. glabrata (9%) and C. pelliculosa (6.7%), with other species comprising 8% of isolates. All three echino candins exhibited potent in vitro activities against the vast majority of Candida isolates. Azoles demonstrated potential in vitro activities against C. albicans (>95% susceptibility rate) and C. parapsilosis complex (>95% susceptibility rate), while serious azole resistance mainly observed in C. tropicalis and C. glabrata with resistance rate to fluconazole and voriconazole of 11.9%, 9.1% and 7.7%, 28.2%. Note worthily, C.pelliculosa had extremely high incidence rate in newborns and dramatically rates of resistance to fluconazole and voriconazole of 55.2% and 41.4%. Thus this present study provided valuable local surveillance data on the epidemiology and antifungal susceptibilities of invasive yeast species isolated from the First Affiliated Hospital of Zhengzhou University, which was essential for guiding the selection of adequate antifungal therapy.

P-211

Clinical, Phenotypic and Molecular Characteristics of Invasive Pulmonary Aspergillosis Caused by Aspergillus Lentulus in China

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We aimed to investigate the clinical, phenotypic and molecular characteristics of proven or probable Invasive Aspergillosis (IA) caused by A. lentulus in immunocompromised patients in China. This study includes a panel of six non-duplicate A. lentulus isolates recovered from respiratory tract of patients with proven or probable IA during August 2016 to July 2017.Accurate identification and phylogenetic analysis of all the isolates were based on MLST of five genes. Seven microsatellite markers employed for genotyping. In vitro susceptibility to nine antifungal drugs was determined by CLSI M27-A3 broth microdilution methodology. All the patients had severe immune disease who have been treated by immune suppressive drugs and four of them were known that received prior antifungal therapy. All seven of the isolates slow sporulating grew at 28°C and 35°C, but not grew at all at 48°C on Sabouraud dextrose agar. All the atypical isolates grew as fluffy white colonies mainly consisting of hyphae interspersed with sporadic gray-green spores after 7 days of incubation. Microscopic examination on day 3 to day 21 show stipes, head and conidia is nearly identical to A. fumigatus sensu stricto. All isolates clustered together in a clade distinct from other members of the A. fumigatus. MC1, MC3, MC5 and MC6a might be employed for A. lentulus genotyping. Cyp51A and cyp51B genes of A. lentulus were identified. VitekMS systems identifed the six isolates to species level while Bruker Biotyper had no identification. Echinocandins exhibited high in vitro activities against all A. lentulus isolates. While all isolates had high MIC value to azoles and polyenes, especially to voriconazole with MIC from 8 to 16 μg/ml. In conclusion this is the first investigation about clinical, phenotypic and molecular characteristics of proven or probable IA caused by A. lentulusin immunocompromised patients in China.

P-212

Clinical and Molecular Characteristics of Tsukamurella Tyrosinosolvens Causing an Infection Misdiagnosed as Mycobacteria Tuberculosis, and its Antimicrobial Susceptibilities

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Tsukamurella species are aerobic gram-positive organisms from the order Actinomycetales which are generally a weakly acid-fast, nonsporeforming rod and shares many features with Mycobacterium. This study aims to investigate one T. tyrosinosolvens isolate from patients with lung disease who was misdiagnosed as tuberculosis. Species identification was performed by sequencing of the five gene targets including 16S rRNA, ssrA, secA, rpoB, and groEL. Antifungal susceptibilities were determined by Clinical and Laboratory Standards Institute document (CLSI) M100 broth microdilution methodology. The T. tyrosinosolvens isolate obtained from a 51-year-old immunocompetent woman who was misdiagnosed as pulmonary tuberculosis and treated for nearly 4 years but showed no improvement in symptoms. The correct diagnosis was suspected only when the results of mycobacteria nucleic acid amplification tests were negative. The isolates grows best on blood agar as white, dry and rough colonies, with irregular spreading margins after 24 h of incubation at 37. The performance of groEL gene sequencing for species-level identification of T sukamurella was better than other four genes. The T. tyrosinosolvens isolate is resistant to rifampicin (>2 μg/ml), ceftazidime (>32 μg/ml), ampicillin (16 μg/ml), aztreonam (>128 μg/ml), erythromycin (>8 μg/ml), vancomycin (8 μg/ml), daptomycin (4 μg/ml) and colistin (>8 μg/ml). While it is susceptible to ceftaroline (0.25 μg/ml), ceftriaxone (1 μg/ml), imipenem (0.25 μg/ml), meropenem (1 μg/ml), linezolid (1 μg/ml), levofloxacin (0.5 μg/ml), moxifloxacin (0.25 μg/ml) and tigecycline (0.25 μg/ml). Based on our and previous study, fluoroquinolones is considered as an excellent antibiotic to treat this uncommon micro-organism. Thus this study points to a possible emergence of T. tyrosinosolvens as a significant pathogen cause lung disease which was prone to misdiagnose as pulmonary tuberculosis in the immunocompetent patients. Molecular methods are needed for accurate identifcation and further molecular characterization of this species. Fluoroquinolone may be a successful oral antibiotic regimen.

P-213

Invasive Infections Due to Trichosporon: Species Distribution, Genotyping, and Antifungal Susceptibilities from a Multicenter Study in China

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A total of 133 clinical Trichosporon isolates were collected in the National China Hospital Invasive Fungal Surveillance Net (CHIF-NET) program in 2009 to 2016. Accurate identification was performed by sequencing of the intergenic spacer 1 (IGS1) region. Among these isolates, Trichosporon asahii (108 isolates [81.2%]) was the leading species, followed by Trichosporon dermatis (7 isolates [5.3%]), Trichosporon asteroides (5 isolates [3.8%]), Trichosporon inkin (5 isolates [3.8%]), Trichosporon dohaense (3 isolates [2.3%]), and 1 isolate (0.7%) each of Trichosporon faecale, Trichosporon jirovecii, Trichosporon mucoides, Trichosporon coremiiforme, and Trichosporon montevideense. Both the Vitek mass spectrometry (MS) (bioMérieux, Marcy l'Etoile, France) and Bruker Biotyper MS (Bruker Daltonics GmbH, Germany) platforms gave high levels (>97.5%) of correct identification when the species were present in the database. The geometric mean (GM) of amphotericin B MICs for T. asahii was 2-fold higher than that for non-asahii Trichosporon. High fluconazole MICs (≥8 μg/ml) were observed for 25% of T. asahii isolates (27/108 isolates) and 16% of non-asahii Trichosporon (4/25 isolates) isolates. Itraconazole MICs were ≤0.5 μg/ml for 89.5% of the isolates. Voriconazole was the most potent antifungal agent in vitro, with a GM of 0.09?μg/ml. Genotyping of the isolates using IGS1 sequence alignment revealed that genotype 1 was most common (41.7%), followed by genotype 4 (31.5%), genotype 3 (23.1%), genotype 5 (0.9%), genotype 6 (0.9%), and genotype 7 (1.8%). Our data on species distribution, genotypes, and antifungal susceptibilities may contribute to a better understanding of the epidemiology of invasive Trichosporon infections throughout China.

P-214

In Search of Novel Drug Targets for Control of Mycobacterium Tuberculosis

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Tuberculosis is an infectious disease which is not at all controlled. More than one third of global population is infected with Mycobacterium tuberculosis. Recently multiple drug resistant and extensive drug resistant strains of the bacteria has emerged and causing increasing number of tuberculosis patient throughout the Globe. Therefore, new antitubercular drug development is an emerging field of research at the present moment. In this context we have identified proteins of Mycobacterium tuberculosis with known three dimensional structures that are important for pathogenesis of tuberculosis and studied interaction with existing anti tubercular drugs and novel small molecules using tools of in silico biology. The results have predicted that existing antitubercular drugs and selected small molecules (with known and unknown targets) has the potential to bind with new target protein of Mycobacterium tuberculosis. We feel that wet lab experiments are required to validate the results.

P-215

Active Surveillance of Carbapenemase-Producing Organisms (CPO) Colonization with Xpert Carba-r Assay Plus Positive Patient Isolation

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Rapid screening of patients for carbapenemase-producing organisms (CPO) colonization, coupled with implementation of infection prevention strategies, has the potential to contain the spread of CPO. We firstly evaluated the performance of Xpert Carba-R assay, in comparison with other phenotypic methods, for carbapenemase detection using clinical isolates and then used it to determine intestinal CPO colonization in hospitalized patients, and coupled it with patient isolation in a medical intensive care unit ward. The Xpert Carba-R assay required the least processing time to results and showed a 100% sensitivity and specificity in carbapenemase detection, except for IMP-8 (n =4). During the six-month period, 134 patients in one ward were studied. Fifteen patients (11.2%) were CPO colonized, as detected by Xpert Carba-

R assay, including three NDM, three IMP and nine KPC strains. The overall colonization and CPO infection rates were both 11.2% each. A significant decreasing trend was noted in both colonization (from 28.6% to 5.6%) and infection rates (from 35.7% to 2.8%) during the study period ($p<0.05$). This study concluded that Active surveillance of CPO, utilizing the Xpert Carba-R assay supplemented with immediate patient isolation, proved to be an effective measure to limit the spread of CPO in a health care setting.

P-216

Real-World Evaluation of Nine Infectious Disease Assays on Roche Cobas E 801 Versus Abbott ARCHITECT Or Diasorin Liaison XL Platforms

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This study aimed to compare the performance of nine infectious disease assays on different high-throughput platforms. Samples for fertility/pregnancy and trisomy screening were assessed for HBsAg, anti-HBc, anti-HCV, HIV (4th generation assay), rubella IgM and IgG, CMV IgM and IgG and CMV avidity at a single centre, using the cobas e 801 platform and either the ARCHITECT, Liaison XL, or VIDAS platforms. Indeterminate/discrepant samples were confirmed using bioMérieux VIDAS and/or immunoblot testing, or LiPA (for hepatitis). For HBsAg, specificity in the two sample groups was 100% for the cobas e 801 platform and 99.71% for the ARCHITECT platform (N=1052 fertility/pregnancy samples), and was 100% for both platforms (N=200 trisomy-testing samples). For anti-HBc, specificity was 100% for both the cobas e 801 and ARCHITECT platforms (N=1051 fertility/pregnancy samples and N=200 trisomy-testing samples). For anti-HCV, specificity was 100% for the cobas e 801 platform and 99.5% for the ARCHITECT platform (N=200 trisomy-testing samples), and 99.81% for both platforms (N=1051 fertility/pregnancy samples). For HIV, specificity was 99.82% for the cobas e 801 platform and 99.73% the ARCHITECT platform (N=1102). For rubella IgG (N=429), specificity was 100% for the cobas e 801 and Liaison XL platforms. For rubella IgM (N=92), specificity was 99% for both the cobas e 801 and Liaison XL platforms. For CMV IgG (N=687), specificity was 98.94% for the cobas e 801 platform and 97.18% for the ARCHITECT platform. For CMV IgM (N=680), specificity was 97.16% for both platforms. For CMV avidity low (N=25) and high (N=29), 96% and 93% correlation was observed between cobas e 801 and VIDAS platforms. This study concluded that in routine samples, performance of the cobas e 801 platform was superior or equal to performance of the ARCHITECT, Liaison XL, or VIDAS platforms.

P-217

Association of Plasma Matrix Metalloproteinase 9 and Cholinesterases with Zinc in Vivax Malaria Patients

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Zinc is a micronutrient whose deficiency may increase the risk of plasmodium infection in humans. Cholinesterases are low grade inflammatory markers that have zinc dependent carboxypeptidase activity .Matrix metalloproteinases (MMP), are zinc containing endopeptidases which participate in inflammatory processes and contribute to tissue remodeling and repair . The present study attempts to establish an association of zinc with these enzymes in vivax malaria. Plasma zinc, butyrylcholinesterase (BChE) and erythrocyte acetylcholinesterase (AChE) were estimated spectrophotometrically in 100 malaria patients and 50 normal subjects. MMP9 was estimated using ELISA. Plasma zinc was markedly lower in malaria patients compared to healthy controls. Both BChE and AChE decreased significantly in malaria ($p<0.00$) compared to normal. MMP9 increased in malaria compared to control group, although the increase was statistically insignificant. Cholinesterases correlated positively with zinc in controls but not in malaria. The metalloenzyme MMP9 showed a significant positive correlation with zinc in malaria patients ($r=0.405$, $p=0.05$). The present study justifies the contribution of zinc in maintenance of enzyme activities and also establishes its role the pathology of malaria.

P-218

Association of Matrix Metalloproteinase 9, a Marker of Inflammatory Tissue Damage with Serum Albumin and Iron in Pulmonary Tuberculosis Patients Prior to and During Treatment

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Matrix metalloproteinases (MMPs) are calcium dependant zinc containing endopeptidases capable of degrading extra cellular matrix. MMP9 is expressed by pulmonary epithelial cells, which

play a role in extensive lung matrix destruction leading to cavitation in pulmonary tuberculosis (PTB) patients. Malnutrition can lead to secondary immunodeficiency which in turn gives an easy pathway for the causative agent to infect the host, Conversely, inflammation could also serve as a risk factor for malnutrition. Treatment with anti tuberculosis drugs should generally improve the immune status and associated anemia in addition to killing the pathogen. Therefore, this work focuses on estimating the MMP 9 levels in serum and correlate the same with nutritional status using serum albumin and iron levels as indices in PTB patients before and after 2 months of Directly Observed Treatment Short Course (DOTS) treatment and determine if the drug treatment has any effect on lung damage and associated nutritional deficiencies. Twenty-five newly diagnosed PTB patients of both sexes in the age group 18 to 65 years were analysed for serum MMP9, iron, total protein and albumin levels. Results showed a significant increase in serum iron ($p< 0.05$) and serum total protein ($p<0.05$) post 2 months of DOTS treatment. Increase in serum albumin observed after treatment was only apparent. In conclusion, DOTS treatment is found to improve the nutritional status of PTB patients along with alleviating the symptoms of PTB.

P-219

Fungal Contamination of Orthodontic Appliances in "Hassani Abdelkader" Hospital, Algeria

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Orthodontic treatment makes necessary to use a longtime fixed-band appliance, it offers suitable conditions for fungal growth. The aim of our study is the identification of the species colonizing surface of orthodontic appliances, the cross-sectional study was carried out on 60 patients wearing dental appliances, during 5 months, sterile swabs were used and inoculated into Sabouraud's dextrose agar sterile tubes. Yeast identification has been based on germ tube test, chlamydoconidia production and biochemical tests (Auxacolor, Api20). The overall prevalence of fungal contamination of dental orthodontic appliance was 35%, the prevalence rate was 40% among patients who had dental appliances for 1 to 2 months, and 45.5% for patients brushing their teeths at least twice daily. The fungal species most recovered were Candida parapsilosis 21.47%, Candida zeylanoides 21.47%, Candida albicans 17.38% and Cryptococcus terreus 13.04%. This study concluded that Mycological monitoring of dental prothesis is crucial to prevent possible fungal's adverse health effects.

P-220

Analysis of Oxidative DNA damage in Pulmonary Tuberculosis

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Tuberculosis is one of the major health problems and one of the leading causes of death from infectious diseases. Oxidative damage results from biochemical interactions between reactive oxygen species (ROS) and target biomolecules. ROS can damage nucleic acids, lipids, proteins which figures prominently in progression of carcinogenesis. M.tuberculosis cells are exposed to DNA damaging agents such as reactive oxygen intermediates (ROI) and reactive nitrogen intermediates (RNI) generated by host macrophages. In nuclear and mitochondrial DNA, 8-hydroxy-2-deoxyguanosine (8-OHdG) or 8-oxo-7,8-dihydro-2-deoxyguanosine (8-oxodG) is one of the predominant forms of free radical-induced oxidative lesions and has therefore been widely used as a biomarker for oxidative stress and carcinogenesis. In view of this present study was undertaken to evaluate the oxidative damage on deoxy nucleic acid in pulmonary tuberculosis. Present study included 50 patients of Pulmonary Tuberculosis on Anti-Tubercular treatment and 50 age and sex matched healthy control subjects without Pulmonary Tuberculosis. Oxidative DNA damage was assessed by using 8-hydroxy-2-deoxy guanosineas biomarker with highly sensitive enzyme linked immunosorbent assay (ELISA) kit.8-hydroxy-2'-deoxy guanosine level was significantly higher in pulmonary TB patients as compared to controls ($p<0.0001$) as a marker of oxidative DNA damage, increase in the level of 8-OH-dG

P-221

The Seroprevalence of Anti-Hepatitis A IgG in the Korean Population

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Hepatitis A infection caused by the hepatitis A virus (HAV) is a global health concern, with an estimated 1.5 million people infected annually with HAV. In this study, we aimed to investigate the seroprevalence of anti-HAV IgG in the South Korean population. That seroprevalence data will be the baseline for the establishment of HAV prevention strategy. Seroprevalence data of anti-HAV IgG from January 2015 to December 2017 were obtained from the laboratory information system of Green Cross Laboratories, one of the largest referral laboratories in South Korea. The subjects

were categorized into age groups by decade and sex, and all data were anonymized before analysis. Seroprevalence of anti-HAV IgG antibody was also evaluated by geographic region in Korea. During the three-year study period, we obtained 353,099 test results of anti-HAV IgG from 346,041 individuals (124,581 males and 221,460 females) from 1,503 hospitals and/or local clinics throughout South Korea. The median (range) age was 37.8 (18.0-101.9) years. The annual seroprevalence of anti-HAV IgG was 53.1% in 2015, 54.2% in 2016, and 55.3% in 2017. There was a statistically significant difference in seroprevalence by sex (males 61.0% vs. females 50.4%, P<0.001). The age-related seroprevalence (age) of anti-HAV IgG was 30.2% (10s), 24.2% (20s), 32.9% (30s), 73.6% (40s), 96.8% (50s), 99.7% (60s), 99.7% (70s), 99.5% (80s), and 98.9% (90s and 100s). Men living in Sejong city and women living in Jeju were the most likely to get HAV infection. Present study concluded that young adults (20s-40s) are especially at-risk population for an HAV epidemic, and this population subgroup should be identified and vaccinated. This study provides valuable information for establishing a catch-up vaccination program in South Korea.

P-222

Correlation of Serum Fibronectin with Indirect Markers of Liver Fibrosis in HCV Patients

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Hepatitis C virus (HCV) infection is the most common problem these days which can lead to chronic infections causing liver fibrosis, cirrhosis, and hepatocellular carcinoma. More than 170 million people worldwide are affected with HCV. Liver fibrosis involves a disorganized accumulation of extra cellular matrix (ECM) components leading to loss of normal cell functions. The assessment of liver fibrosis in HCV patients is considered a key for decision making and patient care. The main invasive method for the same is liver biopsy but it has certain limitations. Hence, the use of non-invasive biomarkers is suggested. The APRI, i.e., AST to platelet ratio index is the most useful score to predict fibrosis. Moreover, the Serum Fibronectin (FN) is another non-invasive biomarker which can be considered. FN is a glycoprotein of ECM which is produced by hepatocytes. The study aims to investigate the role of serum FN levels to assess liver fibrosis for HCV patients. Further, FN levels were correlated with APRI and FIB-4. A total number of 40 HCV Patients attending Department of Medicine, Guru Gobind Singh Medical College and Hospital Faridkot were considered in our study. Informed written consent was taken. 40 healthy individuals (age and sex matched) were taken as controls. Investigations such as Viral markers, Complete Blood Count, Blood Glucose, Renal Function Tests, Liver Function tests (AST, ALT, ALP, GGT), and Serum Fibronectin were performed. By using indirect markers, ratios like APRI, and FIB-4 were calculated. Serum FN levels were significantly lower in HCV Patients (137.8±42.4μg/ml) when compared with controls (250.8±36.8μg/ml) Moreover, a negative correlation was found between serum FN and AST (P≤0.005), ALT (P≤0.005), and GGT levels (P≤0.001). An inverse relationship is found between serum Fibronectin levels and APRI, and FIB-4. Thus, decrease in FN may indicate hepatitis severity along with APRI, and FIB-4.

P-223

Effective Infectious Disease Management for Transfusion by Flexible Multiplex Screening Technology

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The SG CapTM Technology provided by PCL Inc. represents a powerful approach where the sol gel matrix constrains the motion of the encapsulated biomolecules without physical adsorption or any modification. Discrete three-dimensional sol-gel spots, each containing a different immobilized target, can be spotted within a single well, allowing multiple markers to be detected simultaneously. Here we introduce the Hi4-TP combo kit based on the SG CapTM Technology, which is designed to detect antibodies against HCV protein (Core, NS3, NS4, NS5), HIV 1/2/O type protein and TP (p15, p17, p47) and antigen against HIV p24 and HBsAg protein, respectively. In the Clinical trial at the Seoul St. Mary's Hospital in Korea, the results showed that the sensitivity of Hi4-TP combo kit was 100% (n=600; including 100 HIV Korean positive specimens, 100 HIV 1, 49 HIV2, 1 HIV1O-subtypes, 50 HIV p24 antigen positive and 300 not classified HIV European positive specimens, n=600; HCV Korean and European positive specimens, n=400; TP Korean and European positive specimens and n=400; HBsAg positive specimens), and the specificity for HIV Ab-Ag, HCV Ab, TP Ab and HBsAg were 99.90%, 99.95%, 99.83% and 99.98%, respectively (n=6,000 negative specimens). Finally, we suggest that SG CapTM Technology holds promising potential as a novel and versatile multiplex blood screening platform that can be used for effective infectious disease management

P-224

Plasma Leakage in Patients with Dengue Infection and Possible Role of Innate and Adaptive Immune Receptors on Platelet Surface

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Platelet activation and cells expressing DC-SIGN and Fc GammaR2A receptors have been reported to play a major role in dengue infection. The present study assesses the expression of the DC-SIGN and FcGammaR2A receptor on platelet surface in dengue patients and its association plasma leakage by hematocrit. This was an analytical cross-sectional study carried out in JIPMER hospital, Puducherry. 35 patients with dengue infection (cases) and 36 patients with non dengue acute other febrile illness (controls) were recruited. Hematological parameters on admission and Platelet rich plasma was assessed for DC-SIGN and FcGammaR2A using BD FACS CaliburTM on: admission, day 3 & discharge. Subjects were divided into groups based on the gender specific mean value of hematocrit. We observed a decreased expression of DC-SIGN in cases compared to controls on admission in both groups. An increasing trend, though not significant, was observed in expression of DC-SIGN in both groups in cases and in patients with higher hematocrit, DC-SIGN expression remain suppressed along the course of disease. There was significant difference in expression of FcGammaR2A along the course of disease in both groups in dengue cases. In patients with hematocrit more than the mean value, FcGammaR2A expression on surface of platelets remain suppressed even on Day of discharge. Our results suggest that DC-SIGN, which is a receptor for viral capture and FcGammaR2A which is a receptor for IgG complexes might be prolongedly suppressed in dengue patients with plasma leakage. FcGammaR2A receptor has been found to play a role in platelet activation, immune mediated clearance and Antibody dependent enhancement of infection. Dengue induced antibodies are also found to cause endothelial damage and vascular leakage. This could be part of protective response from the host to prevent platelet activation and subsequent endothelial damage.

P-225

Novel Function-Spacer-Lipid (FSL) Trypanosoma Cruzi Constructs as Sensitive and Specific FSL Kodecytes Assay

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Trypanosoma cruzi is a protozoan parasite that causes Chagas disease or American trypanosomiasis in humans affecting 8 million populations and 10,000 deaths per year worldwide (CDC, WHO 2019). This disease has three clinical phases in humans - acute, chronic-intermediate and chronic. Diagnosis of chagas disease is very challenging as in case of low infection it is very hard to detect the parasites and PCR fail to detect 50% positive chronic cases. Consequently, diagnosis is possible by serological methods. To design a new serological test method, choosing the pertinent antigen and technological platform can maximize the test efficiency. Kodecytes function-spacer-lipid (FSL) constructs technology has already been successfully used to detect syphilis antibodies (Venkata Sarvani Komarraju 2010). In this study using a peptide selection algorithm a series of function-spacer-lipid (FSL) constructs were created and attached to human red cells creating T. Cruzi kodecytes. These kodecytes were then tested against human serum from panels and blood donors of known chagas status. Human serums positive and negative to chagas infection were obtained from Brazil and tested at 20μg/ml concentration in standard tube serology. Of 118 positive and 116 negative serums tested, FSL CHA-2 was 96% sensitive and 81.2% specific, while FSL CHA-4 is 97.5% sensitive and 100% specific. Thus, it was found that T. cruzi kodecytes reported here are capable of same sensitivity and improved specificity as validated assays (ORTHO, 2006).

P-226

HIV Infection and Liver Disease: A Tertiary Care Centre Study

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Diseases of hepato-biliary system is a major problem in patients with HIV infection. It has been estimated that approximately one third of the death of patients with HIV infection are in some way related to liver disease. Liver disease in HIV patient is

attributed by co-infection with Hepatitis Bor C,drug hepatotoxicity, fatty liver and alcohol abuse. It is also a reflection of the hepatic injury in the form of hepatic steatosis that can be due to anti-retroviral therapy. There had been little work done on liver function tests in HIV patients without pre-existing liver disease. So, this study was designed to assess the pattern of liver function tests derangement in HIV patients. To study different patterns of hepato-biliary involvement in HIV positive patients, and to assess its severity, we study included 100 HIV positive patients coming to SMS hospital and Medical College, Jaipur, in medicine and HIV clinic of skin and VD department. Subjects having HIV test positive by ELISA, are included in this study. Other systemic causes of pre-existing liver disease were excluded from the study. Out of 100 cases, 83 were male and 17 were female with mean age 38.12±32 years. Out of these, 17 patients had coexisting HBV infection and 4 patients had HCV infection.73 cases had abnormal liver function tests in the form of raised AST, ALT, total bilirubin in 54, 68 and 37 patients respectively. Cholesteric pattern of liver injury was found in 41 patients with raised serum Alkaline phosphatase. Evidence of stones in biliary system was found in 13 patients. Thus, almost all types of liver disease were found in HIV patients. The pattern of hepato-biliary involvement varied from asymptomatic elevation of liver function tests to clinically significant liver disease in HIV patients. Prompt diagnosis and treatment required to decrease morbidity.

P-227

Evaluation of Adenosine Deaminase (ADA) Values for Detection of Extra-Pulmonary Tuberculosis

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There is need of precise and faster diagnosis for extra-pulmonary tuberculosis (EPTB). Accordingly, there develops a considerable interest in the development of tests based on biochemical response of body towards tuberculosis infection (i.e. estimation of serum adenosine deaminase). These test appear to be a promising approach for the diagnosis of pulmonary as well as extra pulmonary TB. Total 100 individuals recruited for the study, out of which 50 cases EPTB, 50 ages and sex match controls were included, out of which 15 were healthy controls and 35 were disease controls. Serum adenosine deaminase (ADA) and its isoenzymes were estimated in serum, plural fluid, CSF by using commercial ADA-MTB kit. ADA concentration in patient with EPTB was found to be significantly higher ($p<0.05$) as compare to disease and healthy controls. In detecting extra pulmonary tuberculosis, the sensitivity and specificity, positive and negative predictive values of serum ADA were 94% for each. Our observation suggest that serum & fluid adenosine deaminase (ADA) and its isoenzymes has a good detection potential for EPTB.

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Role of Nitric Oxide Donor L-Arginine and Ciprofloxacin Against Typhoid

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Typhoid caused by Salmonella typhi remains a major health concern worldwide. The emergence of multidrug-resistant (MDR) strains of Salmonella with increased virulence leading to increased morbidity and mortality has further complicated its management. Human typhoid is similar to the infection caused by Salmonella typhimurium in mice. Most of the antibiotic are resistant and vaccines have less than desired efficacy and certain unacceptable side effects, making it pertinent to search for new suitable formulation. Nitric oxide (NO) is a gaseous free radical molecule; produced in biological systems. During enzymatic conversion of L-arginine to L-citrulline by NO synthase (NOS) nitric oxide is produced. Ciprofloxacin one such fluoroquinolones have been shown to achieve high intracellular concentrations and least resistant antibiotic used against typhoid. Exogenous administration of L-arginine results in increased NO production, indicating that endogenous substrate is insufficient for maximal NO production. By considering these facts, it was thought to see the effect of oral administration of NO donor i.e. L arginine along with the low doses of antibiotic (ciprofloxacin). NO estimation was done by the fluorometric method (detection limit in nM range). Hepatic nitrite level in mice infected with 0.6xLD50 of S. typhimurium was 8.33%, higher than control animals (treated with saline) at day 8,and in a different groups B+Arg, B+Cip & B+1/2Arg+1/2Cip were 16.66%, and 12.5% & 10.25% respectively as compared to only S. typhimurium infected mice. Formulation of low doses of L-arginine and ciprofloxacin shows better therapeutic induction against typhoid, so that it can use for future treatment. This study concluded that increase of nitrite level (metabolites of nitric oxide) is may be due to enhanced pro-inflammatory IFN-γ, TNF-α cytokine expression.

P-229

Atherosclerosis

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Atherosclerosis is a disease state of heart which act as the base of other disease of heart . So my research work is to dissolve this plaque more efficiently with less side effect and more effectively .This plaque is majorly formed by cholesterol (LDL) so to dissolve this first we need to stop the formation this LDL. So there is composition of chemical which lead to decrease or stop the formation of LDL. In second step the plaque already form in the heart blood vessel we need to dissolve them So for this there is composition of chemical which will lead to dissolve this plaque and thrombus. The plaque is dissolved with very less side effect like urine colour will change into yellow if given then prescribed dose constipation can occur . Dizziness may occur. Cannot given to a patient suffering from asthma and liver cirrhosis. If given in prescribed amount then it will reduce plaque as it is a good lipid lowering agent So my research can be used in clinical practice.

P-230

Study on ADA and Lipid Profile Levels in Alcoholic Liver Disease

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Alcohol consumption is associated with a number of changes in hepatic cell functions as liver is the major site for its metabolism. The liver plays a crucial role in the synthesis, secretion, catabolism and storage of lipids. The aim of this study was to evaluate the effect of alcoholic liver disease on the serum level of lipid profile and ADA. 50 clinically diagnosed cases of Alcoholic Liver disease (ALD) supported with serological test, ultra-sonogram in the age group of 25-60 years were enrolled for study. They were matched with 50 healthy controls. Total duration of study was two years. Lipid profile measured by enzymatic method. Serum LDL cholesterol Derived by Friedwald's equation Method. Adenosine deaminase was estimated by the method of Giusti and Galanti. Mean triglyceride and VLDL level showed statistically no significant difference between patients and healthy controls($p>0.05$) but cholesterol, HDL and LDL were found to be significantly low in patients as compared to controls. In our study, mean cholesterol was 119.32±39.37 mg/dL in cases and 168.92±27.94 mg/dL in control. Similarly, HDL was found to be 28.76±10.55 mg/dL in cases and 43.52±7.53 mg/dL in control and LDL was 70±25.82 mg/dL and 103.16±25.94 mg/dL in cases and control respectively ($p<0.001$). Mean of ADA was 66.55±30.89 U/L in cases and 6.14±2.56 U/L in control. On statistical comparison; we found significant difference between both the groups ($p<0.001$). The levels of lipids progressively diminished with the deterioration of liver function in patients with Alcoholic liver disease, ADA levels also significantly increased. Screening for the same is important for prognosis and treatment.

P-231

Association Between Carotid-Intima Media Thickness and Triglyceride Glucose Index in Hypertensive and Normotensive Individuals

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Hypertension is an important risk factor for development of cardiovascular disease. High blood pressure is implicated for development of myocardial infarction and stroke. Carotid intima media thickness (c-IMT) is a common tool for risk stratification for cardiovascular event due to atherosclerosis. Insulin resistance is another mechanism underlying atherosclerosis in the absence of diabetes mellitus and hyperglycemia. The present study was thus aimed to determine the effect of hypertension on IMT of the common carotid artery and evaluate the relationship between triglyceride glucose index (TyG) and atherosclerosis. Data from 178 hypertensive and 66 normotensive subjects were analysed in the present study. The IMT of the common carotid artery and TyG Index were higher in the hypertensive individuals as compared to control ($p = 0.002$ respectively). Age, hypertension and TyG Index were important risk factors for increased c-IMT in CCA [odds ratio 1.10 (1.06-1.13); 1.24 (0.69-1.20) and 1.54 (0.70-2.41) respectively]. Age, hypertension and Triglyceride glucose Index were found to be important determinants of increased c-IMT. It is henceforth crucial to manage traditional risk factors in order to decelerate increase in cardiovascular disease incidence.

P-232

Eminent Need of Newer Validated Formula for Low-Density Lipoprotein Cholesterol Estimation for Best Therapeutic Intervention

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Coronary Artery Disease (CAD) is one of the leading causes of morbidity and mortality with increasing incidence in India. As currently Low-density lipoprotein cholesterol (LDL-C) is the primary target for CAD risk prediction, there is eminent need of new validated formula for accurate and precise LDL-C estimation for best therapeutic intervention. A cross sectional study was conducted in Department of Biochemistry, VMMC and SJH, New Delhi. The aim of this study was to compare the results obtained by direct homogenous assay for LDL-C to those obtained by different formulas. Lipid profile reports of 500 patients above 18 years (TG<300 mg/dL) were analyzed. LDL-C estimation was done by homogenous assay and also calculated using the Friedewald's, Anandaraja, Vujovic, Chen, De Cordova and Hattori formula. In study population having Triglyceride(TG) concentration less than 100mg/dL, LDL-C values calculated by Friedwald formula, Anandaraja, Vujovic, Chen, Cordova, Hattori formula were lower by 5.7%, 3.23%, 3.61%, 10.22%, 18.13%, 13.28% respectively as compared to direct homogenous method of LDL-C values. Similarly, in study group having triglyceride concentration between 100-300 mg/dL LDL cholesterol values were underestimated by 9.9%, 11.2%, 6.3%, 10.7%, 15.5%, 19.4% by Friedwald formula, Anandaraja, Vujovic, Chen, de Cordova, Hattori formula respectively as compared to direct homogenous method. LDL cholesterol estimation is crucial for monitoring the effect of lipid-modifying therapies since high LDL cholesterol is central to coronary artery disease. However, different formulas have been used and verified in different populations. Therefore, it is important to devise and validate a new modified formula for its correct estimation and interpretation.

P-233

Can Calculated SdLDL Serve as a Substitute for Estimated SdLDL?

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SdLDL is the actual atherogenic component of LDL. Though it is recognized as a predictive biomarker for coronary artery disease, lack of standardisation and complexity and cost of the analytical techniques has prevented sdLDL from being routinely estimated in clinical practice. Methods available for analysis include Gradient Gel Electrophoresis, Ultracentrifugation and NMR. Hirano et al separated and measured sdLDL from lbLDL using detergent and sphingomyelinase treatment. Srisawasdi et al. developed a formula for sdLDL estimation using common lipid parameters; sdLDL (mg/dl) = 0.580 (non HDL- Cholesterol) + 0.407(direct LDL- Cholesterol) - 0.719 (calculated LDL- Cholesterol) - 12.05. Mohan et al. proposed that Triglyceride/HDL ratio of 3.0 had optimum sensitivity for predicting elevated sdLDL levels. We did a retrospective study done on 374 samples for which total cholesterol, HDL, LDL and triglycerides had been estimated using routine methods and sdLDL using the enzymatic method. SdLDL was calculated using Srisawasdi's formula and correlation was determined between estimated and calculated sdLDL. We also determined the correlation between the estimated sdLDL and TG/HDL ratio, and with Non HDL. A highly significant and positive correlation was found to exist between estimated and calculated sdLDL (r=0.74, p=0.00), and between estimated sdLDL and non HDL (r=0.721, p=0.00). The correlation between SdLDL and TG/HDL ratio was positive but poor (r=0.353). We concluded that calculated sdLDL may be used as a substitute for estimated sdLDL. Further studies on a larger population are required before use of calculated sdLDL can implemented in routine clinical practice.

P-234

Association of 25-(OH)-Vitamin D with Lipid Profile Among Poor Glycaemic Control Type II Diabetes Patient

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25-(OH)- vitamin D has a significant role to minimize chronic metabolic syndromes and cardiovascular disease in type 2 diabetes mellitus patient. 25-(OH)-D down-regulates the serum lipid by lipogenesis. However, studies between relationships between 25-(OH)-D are inadequate. We aimed to evaluate the association between 25-(OH)-D and serum lipid in poor glycaemic control type 2 diabetic Nepalese populations. This cross-sectional study was carried out among 247 poor glycaemic control diabetes patients attended in Modern Diagnostic and Research Center. Socio-demographic data and anthropometric measurements were recorded using a standard questionnaire. Fasting plasma glucose, HbA1c and Vitamin D3 & Vitamin B12 were estimated by Dimension RxL Max Chemistry Analyser, Lefetronic-H9 Hemoglobin Analyser, and Advia Centaur XP Immunoassay. Student's t-test, One-way Anova test, Mann-Whitney U test, and Kruskal Wallis test were used for comparison between different groups and the correlation was established by Spearman's correlation. The median serum 25-(OH)-D level was 17.89 ng/ml, and the prevalence of hypovitaminosis was 83%. Serum 25 (OH)D deficient patients had a significantly higher level of Fasting blood sugar, Total cholesterol (TC), Triglyceride (TG), and Non-HDL-C ($p < 0.05$). Atherogenic variables such as Cardiac risk ratio, Atherogenic coefficient, Atherogenic index plasma and TC, TG and non-HDL-C shows significantly negative correlation with serum vitamin D level. Serum Vitamin D level shows a negative correlation with lipid profile. Vitamin D supplementation may reduce the risk of cardiovascular complication among type 2 diabetic population.

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Anti-Hyperlipidaemic and Antioxidant Activity of Ethyl Acetate Fractions of Khaya Senegalensis Stem Bark on Diet Induced Hyperlipidaemic Rats

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Khaya senegalensis A. Juss (Meliaceae), is regarded as the most popular medicinal meliaceous plant family. The present study was conducted to evaluate anti-hyperlipidaemic and antioxidant potentials of column chromatography fractions from ethyl acetate extract of Khayasenegalensisstem bark. The chromatography was carried out using Silica gel [(60-120 mesh) (500g)] in slurry of n-hexane. The column was eluted using solvents and solvent mixtures of increasing polarity. The fractions obtained were monitored using analytical TLC technique on different solvent system, fractions with similar Rf value were pooled together and a total of seven fractions (FI-FVII) were obtained. Fifty rats (50) were grouped into ten groups of five rats each, rats from groups III-X were induced with hyperlipidaemia using high fat diet. Group I served as Normal control, Group II served as hyperlipidaemic control, Group III were administered with standard drug (Atorvastatin: 10mg/kg body weight) while Group IV-X were administered with 50mg/kg body weight of fraction I, II, III, IV, V, VI, and VII respectively. At the end of the experimental period (two weeks), the rats were euthanized and blood samples collected into a labelled centrifuge tube, centrifuged and serum obtained was used for analysis of Lipid profile, Antioxidant enzymes, Lactate dehydrogenase, Creatine kinase and HMG CoA reductase activities.A significant ($p<0.05$) decrease in serum Total Cholesterol, Triglyceride and LDL-cholesterol was observed in hyperlipidaemic rats administered with fractions VI compared to other fractions and hyperlipidaemic control. Fraction VI also decreases the levels of serum Lactate dehydrogenase and Creatine kinase activity as well as increase in levels of heart tissue antioxidant enzymes (superoxide dismutase, catalase and glutathione peroxidase) with a concomitant decrease in levels of Thiobarbituric reactive substances compared to hyperlipidaemic control rats. The study concludes that; the anti-hyperlipidaemic property of Khayasenegalensis stem bark may be mediated through its antioxidant properties.

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HbA1c Levels and Atherogenic Index in Non-Obese Diabetics

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Diabetes Mellitus is a major global health problem with 425 million diabetics world-wide. India is one amongst the 6 countries of IDF SEA region with a prevalence of 8.8%. Diabetes Mellitus refers to a group of common metabolic disorders that share the phenotype of hyperglycemia. The disease is characterized by a large number of atherosclerotic complications due to dyslipidemia leading to morbidity and premature mortality. Appropriate management and regular screening is of paramount importance in reducing morbidity and mortality due to its complications. This dyslipedemia can be indicated by atherogenic index and can be correlated with the HbA1c levels. Hence present study was planned with an aim to evaluate the HbA1c levels and atherogenic index in non-obese diabetics and compare it with the healthy controls. Present study included 100 non-obese diabetic patients from R.C.S.M Government Medical College, Kolhapur and compared with 100 age and sex matched healthy controls. 12 hours fasting non-hemolysed serum was used for estimation of Triglycerides and HDL. Hemolysate was used for estimation of HbA1c. The levels of Triglycerides, HDL and HbA1c were estimated by GPO-POD, CHOD-POD and end point immunoturbidimetric method respectively. Atherogenic index is calculated by using the above parameters. HDL levels were significantly ($p < 0.05$) decreased along with a significant increase in the levels of triglycerides and total cholesterol. A negative correlation was observed between HbA1c levels and atherogenic index in non-obese diabetics when compared with age and sex matched healthy controls. Theatherogenic index was increased in non-obese diabetics which may be due to increased HbA1c and reduced HDL levels. This reduced HDL may be associated with a reduction of reverse cholesterol transport. This might be a major risk factor for the development of cardiovascular disease in non-obese diabetics.

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Assessment of Dyslipidemia and Its Associated Factors Among Diabetic Patients at Felegehiwot Referral Hospital, Bahir Dar, Ethiopia

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Dyslipidemia predisposes atherosclerotic cardiovascular disease which remains the principal cause of death and disability among patients with Diabetes Mellitus. Early detection and treatment of dyslipidemia in diabetes mellitus can prevent risk for atherogenic cardiovascular disorder. However, there is limited data on the magnitude and risk factors associated with dyslipidemia among diabetic patients in Bahir Dar, Ethiopia. The purpose of this study is to assess dyslipidemia and its associated factors among Diabetic patients in FelegeHiwot Referral Hospital, Bahir Dar, Ethiopia. A Cross-sectional study was conducted from June 8, 2018, to August 7, 2018. A total of 112 diabetic patients were selected for the study using a simple random sampling technique. Body Mass Index, Fasting blood sugar and lipid profile tests were measured. The prevalence of dyslipidemia was 89.1% of which TC > 200 mg/dL, TG > 150 mg/dL, LDL-C > 130 mg/dL, and low HDL-C < 40mg/dL were observed in 25.9%, 43.8%, 18.8%, and 69.6% Diabetic patients, respectively. Abnormal BMI showed positive correlation with raised TC (P-value = 0.003, AOR = 5.02, 95% CI (1.75, 14.39), raised LDL (p-value = 0.001, AOR = 8.64, 95% CI (2.67, 27.94) and raised TG (P-value =0.001, AOR = 5.9, 95% CI (2.34, 15.09). In addition, raised TG were also associated with daily physical inactivity (p-value < 0.026, AOR=2.8, 95% CI (1.13, 7.06)) and poorly glycemic level FBS >130 and FBS <70 (p-value < 0.002, AOR = 7.3, 95% CI (2.12, 25.32). Moreover, decreased HDL was associated with diabetic patients who had a habit of whole fat daily product intake (p-value < 0.037, AOR= 3.33, 95% CI (1.08, 10.32). Therefore, dietary modification with a decreased daily intake of whole-fat food, control of body weight and glycemic level, and regular daily physical activities will help to reduce the risk of dyslipidemia of Diabetic patients.

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Is Functional Quality of High Density Lipoprotein Compromised in Metabolic Syndrome?

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Metabolic syndrome represents a cluster of metabolic abnormalities and is strongly associated with an increased risk for developing diabetes and atherosclerotic cardiovascular disease. High density lipoprotein particles have extensive athero protective properties however higher HDL levels may not always be protective as it can become dysfunctional.In cases of systemic oxidative stress such as diabetes mellitus and MetS, HDL particles may progressively lose its normal biological activities and acquire altered properties due to modifications of enzymes and apoproteins, and hence it cannot promote cholesterol efflux and transforms into a pro inflammatory molecule. The following study has been undertaken to assess the functionality of HDL and oxidative stress in patients with MetS, a risk factor for CVD, in Indian subjects. To analyze the functional quality of HDL & oxidative stress in patients with Metabolic Syndrome and its impact on CVD. A total of 308 samples were collected and classified according to the NCEP ATP III criteria for MetS. Various biochemical markers were analyzed & HDL functional properties were estimated by fluorometric cell-free assay that measures HDL lipid peroxidation based on the oxidation of dihydrorhodamine123. The results showed that atherogenic risk factors like TC, TG, LDL, VLDL, Apo A1, Apo B, Non HDL, TG/HDL ratio, inflammatory marker (CRP) and oxidative stress marker (MDA) were significantly high in subjects with metabolic syndrome whereas Total antioxidant capacity is significantly low in test population. This study clearly indicates that HDL lipid peroxidation was significantly high in subjects with MetS. Also ApoA1, MDA and TAOC were showing significant positive correlation with HDL antioxidant function.Patients with systemic oxidative stress such as diabetes mellitus and MetS, HDL loses its normal biological activities and acquire altered properties and becomes dysfunctional which can leads to CVD.

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The Effects of Chronic Fructose Intake on Lipogenesis and Glucose Metabolism in Mice with and without Bitter Melon Supplementation

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High-fructose diet is known to give rise to metabolic pathologies which in turn, increase the risk for diabetes mellitus and cardiovascular diseases. We aim to determine the effects of chronic glucose and fructose intake on lipogenesis and glucose metabolism in mice with and without Bitter melon supplementation.Bitter melon Extracts were prepared using 50% ethanol as solvent, biochemical assays like Estimation for phenolic compounds, total carbohydrate, reducing sugar, GOD-POD, FRAP, DPPH, Paper chromatography were done. Male Swiss albino mice were divided into Seven groups(n=6), Control, Glucose, Glucose + BME(GB), Sucrose, Sucrose + BME(SB), Fructose, Fructose + BME(FB) groups. Each group was administered with respective sugars (30%) for 8 weeks, and BME supplementation (300 mg/kg body weight) groups were administered with the extract along with sugars after 4 weeks. Blood glucose level and Body weight measurement were performed every week till the end of the study period. Animals were dissected and organs were collected along with blood for lipid profile by cardiac puncture. Retroperitoneal adipose tissue (RPAT) were isolated stained with H&E and microscopic examination of adipocytes for cell count/size were performed. Body weight increased gradually before treatment with BME in all groups. Upon supplementation with BME significant decrease in the body weight was observed. Blood glucose levels increased in all the groups before treatment, later significant reduction was seen in all the treated groups except FB group. Cholesterol and triglycerides were significantly decreased in all group treated with BME, except for SB group where the triglyceride levels were increased.We conclude that Fructose rich diet has adverse effects on health by inducing lipogenesis and BME didn't reveal significant reversal to lipogenesis at given dose of 4 weeks' supplementation. Further study might be needed to conclude the appropriate effect of the extract when supplemented at different doses and time interval.

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Perfusion of Isolated Goat Liver with 28-Homobrassinolide Evokes Hepatic Marker Expression Corroborating LXR Signaling as Basis for Marker Gene Activation

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Although infectious vectors and cellular derangements have been known to cause alterations in marker status, dietary factors have been recently implicated to drive such changes, though not studied in specific detail. The aim of this study was to analyse changes in hepatic marker levels on perfusing isolated goat liver with subliminal amount of a dietary plant oxysterol - 28-homobrassinolide (28-HB). Ex vivo perfusion of goat liver was performed using 28-HB (333 g/kg wet weight) in Krebs-Henseleit buffer for 2h and 4h. Changes in marker levels, both in the liver homogenate and perfusate, were analyzed. Hexokinase activity was assayed and glucokinase (GCK), ABCA1 and SREBF1 expression was assessed using real time PCR. While tissue glucose and cholesterol levels decreased in 28-HB perfused livers in 2 h compared to control liver, level of cholesterol increased in the perfusate by 54% ($p < 0.05$). Tissue hexokinase activity increased 23% ($p < 0.05$). GCK, ABCA1 and SREBF1 mRNA levels increased 2.6 fold, 5.37 fold and 2.85 fold respectively in 28-HB groups compared to control. Since oxysterols mostly target liver X receptor (LXR) that function as a transcription factor in mammalian cells, changes in marker levels noted in this organ is considered as a result of coordinated regulation of hepatic marker gene expression through LXR by 28-HB.

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Calculated Values of Serum LDL-Cholesterol (LDL-C) - for Better or Worse?

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The use of Friedewald's formula to calculate values for serum low-density lipoprotein cholesterol (LDL-C) has its limitations. A modification of it, in 2013, has been proposed to be superior. This study aimed to test this. LDL-C values were calculated from estimated lipid profiles, using Friedewald's formula and its modification. Kappa statistics and intra-class correlation coefficient (ICC) were used to determine degree of agreement between estimated and calculated values. Bias and percentage total error of the values. LDL-C concentrations calculated by the modified formula showed a greater degree of agreement with estimated values (kappa = 0.761, 0.84, 0.84 and 0.793 for LDL-C cut-offs of 70 mg/dL, 100 mg/dL, 130 mg/dL and 160 mg/dL, respectively) than those calculated by Friedewald's formula (kappa = 0.64, 0.77, 0.49 and 0.74 for LDL-C cut-offs of 70 mg/dL, 100 mg/dL , 130 mg/dL and 160 mg/dL, respectively). ICC of calculated LDL-C values by the modified formula showed moderate to strong agreement with estimated LDL-C, while that by Friedewald's formula showed only fair to moderate agreement. Use of both the formulae produced negative biases. Total percentage errors of the values were greater than recommended limits, across a range of LDL-C and triglyceride concentrations. Calculated LDL-C values, using the modified formula, were in better agreement with estimated values than those obtained by Friedewald's formula. However, both formulae produced negatively biased results, with the percentage total error being higher than the limit recommended for LDL-C.

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Correlation of the Steady State Biomarker Indices for Glycosylated Hemoglobin, Lipids and Anthropometry in Diabetes and Metabolic Syndrome

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Type 2 diabetes mellitus is the most prevalent disorder that increasing health burden in both developed and underdeveloped countries. Insulin resistance is core global issue of metabolic syndrome while it seems to omnipresent in every corner of society. The significant factors are the sedentary lifestyle changes, rapid urbanization & adoption of industrialized food culture habits. The

most determined parameters and clinically occupied role of lipids in relationship to HbA1c and Insulin resistance may need of definitive translation value. Therefore, this study was planned to correlate steady state indices in relation to glycosylated haemoglobin (HbA1c), IR indices with added Triglyceride (HOMA TG-index) and Blood pressures in patients with diabetes and metabolic syndrome. Ninety subjects were recruited from the obesity and metabolic clinic of AIIMS, New Delhi. All subjects were screened as per criteria of ADA and ATPIII guideline. The subjects were categorized into four groups i.e. Group I (healthy control), Group II (diabetes without metabolic syndrome), Group III (diabetes with metabolic syndrome) and Group IV (metabolic syndrome without diabetes). IR Index, Lipid indices was done in chemistry and immunoassay analyzer. HbA1c was done in HPLC (HbA1c) analyzer (Trinity). Anthropometry was done using Tanita body analyzer. Insulin level was raised in group III (10.54±3.8 mUl/mL), HOMA IR was 2.46± 0.45 mmol/l in diabetes with metabolic syndrome group. BMI was higher in group IV among other groups of the patients. Fasting insulin resistance index (FIRI) was raised in group II patients as compared to healthy control (11.84± 0.82 mmol/l).While; HOMA TG was raised in diabetes with metabolic syndrome (12.48±3.2 mmol/l). Moreover Glycated haemoglobin was positively correlated with high triglyceride in subjects with Diabetes with Metabolic syndrome. Thus, assessment of HOMA-IR and HOMA-TG indices could be potential tool in correlation with anthropometry to provide better diagnostic efficiency for diagnosing diabetes and metabolic syndrome.

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Study of Lipid Profile and Apo B in Patients with Hypothyroidism

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Hypothyroidism is a condition with decreased levels of thyroid hormones and elevated levels of Thyroid stimulating Hormone. Hypothyroidism is one of the most common causes secondary dyslipidemia. Thyroid function significantly effects lipoprotein metabolism therefore cardiovascular risk. cross-sectional study was carried out in the Department of Biochemistry, Andhra Medical College, Visakhapatnam, with approval from the Institutional Scientific and Ethics Committee from December 2017 to July 2019.A total of 50 patients of hypothyroidism and 50 normal subjects (age 20 to 60 years) attending OPD of Endocrinology Department, KGH were studied. T3, T4 and TSH were quantitatively estimated by chemiluminescence (CLIA) method. Serum was estimated on the fully automated biochemistry Beckman-AU 480 analyzer. Total cholesterol, Triglycerides and HDL cholesterol are estimated. LDL cholesterol was calculated by using Friedewald's formula. Apo B levels are estimated by immunoturbidometry method. Out of 50 patients, 30 patients belong to subclinical hypothyroidism and 20 belong to overt hypothyroidism. Females had higher prevalence consisting of 85% of total cases. In control patients, the mean level of serum cholesterol, HDL cholesterol, LDL cholesterol and Triglycerides were found to be 172.67 ±14.76, 42.37±6.49, 112.89 ±18.46, 124.89 ± 26.05 respectively. In subclinical and overt hypothyroid patients, mean level of s. cholesterol, HDL cholesterol, LDL cholesterol and triglycerides were found to be 275.44 ±14.49, 39.83 ± 6.76, 152.32 ± 15.29, 187.88 ± 16.69 mg/dl, respectively, and 301.55 ± 20.53, 30.55 ± 7.29, 181.87 ± 20.11, 216.49 ± 20.72, respectively. ApoB in cases was 124.8 ±19.6 and in controls it was 102.9 ±16.4 It can be concluded that hypothyroid patients show dyslipidemic profile and Apo-B is significantly high and they are improved markers for prediction of cardiovascular risk in hypothyroidism.

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Molecular Species of Triglyceride and It Hydroperoxide in Single Lipid Droplets from Human Macrophages

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Uptake of oxidized low-density lipoproteins (LDL) by macrophages results in the formation of lipid droplets (LDs)-laden macrophages or foam cells. The role for the lipid species of LDs in the development of atherosclerosis remains obscure. Monocytes-derived macrophages were loaded with fatty acids (FAs), or native/oxidized lipoproteins. Single LDs were aspirated from the living cells using 3D mobile manipulator visualized under a bright-field microscope, and analyzed in the LTQ Orbitrap equipped with nano-electrospray ionization source1.FAs and native/oxidized lipoproteins induced LDs in the macrophages. In the LDs, we identified several lipid species of triglycerides (TG) and TG hydroperoxides (TGOOH). TGOOH increased by addition of oxidized lipoproteins. Molecular species of phosphatidylcholine (PC) and TG changed according to the structures of added FAs.This study revealed the presence of several molecular species of TG, PC, and more importantly, their hydroperoxides in the LDs. The lipid hydroperoxides in the LDs can initiate the propagation of reactive oxygen species (ROS) in the cells, which might suggest a central role for the LDs in macrophages in the development of atherosclerosis.

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The Correlation Between Direct and Calculated LDL-C Among Patients Referred for Lipid Profile at a Tertiary Care Hospital - Sri Lanka

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The validity of Friedewald formula (FF) in the presence of >400 mg/dL of triglyceride is considered low. However, patients with metabolic syndrome have altered lipoprotein metabolism that would affect the usual ratios between different lipid fractions; especially triglycerides. Despite these pitfalls FF is widely used in the clinical laboratory to cater all patients in general regardless of their disease conditions. The aim of this study was to describe the correlation between direct and calculated LDL-c among patients regardless of their diagnoses as referred to a laboratory. A cross sectional comparison study was conducted among 291 patients referred for lipid profile to the laboratory at a tertiary care hospital - Sri Lanka during a period of 1 month. Direct LDL-c and conventional lipid profile with calculated LDL-c by FF were measured. The mean fasting triglyceride level among the general patient population catered by the laboratory was 138.6 mg/dL (SD:64.9) with a range from 45-464 mg/dL. The mean calculated LDL-c was 103.6 mg/dL (SD:38.4, range:14.3-233 mg/dL) while mean direct LDL-c measurement was 252.2 mg/dL (SD:39.6, range:11.2-252.2 mg/dL). The calculated and the direct LDL-c measurements had good correlation at different levels of triglycerides (r^2=0.9319 at Triglyceride <100 mg/dL, r^2=0.9188 at Triglyceride 101-150 mg/dL, r^2=0.9542 at Triglyceride 151-200 mg/dL, r^2=0.9121 at Triglyceride >200 mg/dL). The use of calculated LDL-c caused a mean negative bias of 7.38 mg/dL and according to the Bland-Altman plot lower concentrations were more affected. The percentage bias when using calculated LDL-c was >5.46% (the desirable specification for bias) in 63.9% of the patients while only 1/3 of them had a triglyceride level >150 mg/dL and the majority (117; 63%) had normal triglyceride levels. Calculated LDL-c by FF may result in clinically significant bias even in patients with normal triglyceride levels. Therefore, normal triglyceride levels do not warrant accurate LDL-c results by FF especially if the metabolic disease status of the patient is unknown.

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Evaluation of Preanalytical Quality Indicators by Six Sigma Methodology and Pareto's Principle at S.H.K.M Govt. Medical College Nuh, Haryana, India

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About 70% of medical diagnostic decisions depend on the accuracy of laboratory tests. Preanalytical errors predominate in the laboratory and constitute approximately 60% of total laboratory errors. Six sigma methodologies can be used by medical laboratories to detect errors in their quality system and enhance the total quality management. This observational study was conducted at Department of Biochemistry, SHKM Govt. Medical College Nuh from September 2018 to February 2019. A total of 13,717 samples and 10,478 test requisition forms for 40,946 routine biochemistry OPD tests were screened for preanalytical errors. All the data was tabulated and sigma values were analysed for each preanalytical error using Westgard online formula. Cumulative percentage of preanalytical errors was calculated by using Microsoft Excel 2010. Sigma value for hemolysed sample, sample not adequate, lipemic sample, missing samples, and collection in wrong vacutainer were 4.3, 4.5, 4.7, 4.7 and 4.8 respectively. It indicates 'good performance' of phlebotomy unit because sigma value greater than or equal to 4 is consider as well controlled process. Sigma values of missing information in test requisition forms like OPD number, date, signature of doctor, name of investigation, diagnosis, age and gender of patients were 4.6, 4.2, 4.1, 4.5, 3.5, 3.9 and 4.0 respectively. Sigma value between 3 and 4 is consider as 'minimal performance' and need improvements. Cumulative percentage of 6 preanalytical errors (missing information about diagnosis, age, gender, OPD number of patients, signature of doctor and hemolysed sample) were about 80% on Parato's chart and need immediate improvements so that overall quality of lab reports can improve. Sigma value indicates the efficacy of laboratory in managing the quality processes and corrective measures should be taken to improve the total quality management in the laboratory.

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Single Platform Vs Multiple Platform: Which One Better for Recources Effectiveness

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In the era of universal health coverage, laboratory is now cost centre rather than become revenue centre. Laboratory must find a way to reduce cost but maintain its quality. Many laboratories using multiple platform for 1 parameter because of many reasons such as using cheaper equipment as back up instrument, for comparing the test result or afraid of reagent supply continuity. We are changing our multiple platform into single platform in 2017. Before 2017, we are using 3 different platform in chemistry analyzer, 2 different platform in hemostasis analyzer, and 2 platform in hematology analyzer. For analyst who responsible for the test we were providing 4 analysts in clinical chemistry, 2 analysts in hematology and 2 analysts in hemostasis. So, we have 8 analyst for taking care of the instrument. In 2017 we were using 1 platform in clinical chemistry, 1 platform in hematology analyzer and 1 platform in hemostasis analyzer. For the analyst we were reducing our recources, only 2 analysts in clinical chemistry, 1 analyst in hematology and 2 analyst in hemostasis. So now we only have 5 analyst for taking care of the instruments and tests, reducing cost in human recources. For cost pertest, in clinical chemistry we can reduce from Rp 7,384.28 pertest to Rp 7,334.42 pertest. In hematology, a slight increment for cost pertest from Rp 20,737.87 to Rp 21,223.55 because we set up to many rerun rules. In hemostasis analyzer, we can cut the cost from Rp 34,544.65 per test to Rp 21,481.44. For 2017 - 2019 we don't have reagent supply problem, instrument downtime no longer than 2 hours. Single platform are more effective in reducing recources than multiple platform.

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External Quality Assessment (EQA) Program on Urinalysis in Thailand

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Thailand Association of Clinical Biochemist (TACB) was introduced External Quality Assurance schemes (EQAs) for urinalysis (UA) using urine strip in 2018. The few available External Quality Assessment (EQA) programs on urinary microalbumin rarely include an evaluation of clinical cases. The present study provides a descriptive analysis of biochemical urinalysis included urine microalbumin in the Thailand laboratory practice. From January 2018 to December 2018, four surveys were organized. Eight EQA urine samples were distributed to the participants by mail. The participants measured the UA of 2 samples quarterly and returned the results together with the information about their instruments and suggestion for the performance of the laboratory report quarterly and summary of situation of each laboratory by online system. Fifty-eight laboratories participated in the survey. The EQA panels included positive and negative samples. The overall accuracy, specificity, and sensitivity were 92.6%, 85.7%, and 75,4%, respectively. The major issues were observed: the low sensitivity for the detection of low concentration samples and the incapacity of several methods to detect the positive sample. The reassuring is needed to continuously evaluate the improvement proficiency for laboratory in Thailand.

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Comparison of Pooled Human Sera with the Commercial Control Sera for the Daily Internal Quality Control Run for Biochemistry Parameters

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Internal quality control is in integral part of the clinical Biochemistry laboratory as it validates and assures the reliability and reproducibility of the results obtained and reported from the clinical specimens submitted for analysis. However the cost of the internal quality control has been a real challenge for the laboratories in the developing countries to incorpororate the IQC in daily basis. Study has been done to look for the possibility of replacing the commercial control sera with the low cost pooled human sera. A prospective study was designed to compare the low cost pooled human sera with the commercial control sera in the clinical Biochemistry laboratory of Tribhuvan University Teaching Hospital. Pooled human sera was obtained from 20 healthy volunteers (10 male and 10 female) and aliquots prepared. These pooled human control were run In parallel to the commercial control sera in the Automated Biochemisty analyser (BT 300) and results obtained for 20 routine Biochemistry parameters for 20 days. The CV mean and SD of the common Biochemistry parameters were compared between commercial and pooled human This study found a similar CV for the pooled sera and commercial control sera for all the Biochemistry parameters analyzed which showed the good stability and acceptable vial to vial variation of the pooled human sera. Our study clearly showed that the in house pooled human sera could be

a good option to replace the expensive control sera. And this would make the IQC programs expensive and accessible to all the laboratories.

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Public and Private

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The past decade has seen an increasing involvement of private for-profit medical laboratories in national healthcare provision. But the majority of patients still use services of public hospital laboratories. We assessed the status of private and public medical laboratories based on ISO 15189 requirements. The overall capacity of nine private and ten public laboratories was assessed using a questionnaire based on ten main requirements of ISO 15189. In general, the public laboratories scored better that the private laboratories but the difference was not significant (79 points for public laboratories vs. 72 points for private laboratories; p=0.12>0.05). The main differences were in the areas of equipment management (82 points for private vs. 92 points for public; p=0.03) and testing performances (77 points for private vs. 91 points for public; p=0.01). Out of the remaining eight assessment sections, the private laboratories scored better, but not significantly, in the areas of laboratory management and laboratory information system.

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Establishment of Reference Range in Clinical Laboratory

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The important role of the laboratory in field of diagnosis is to guide the clinician in interpreting observed values by providing relevant reference values which help them in assessing physiological function of various organ, for diagnosis of disease and it's their Clinical interpretation and in prognosis of therapy. Reference value is a value obtained by observation or measurement of a particular type quantity on a reference individual Observed value is a value of a particular type of quantity, obtained by observation or measurement and produced to make a medical decision. Observed values can be compared with reference values, reference distributions, reference limits, or reference intervals. Reference limit is a descriptive of the reference distribution they tell us something about the observed variation value in the selected subset of reference individuals Reference values have impact of various parameters such as differences in genetic load, sex, age, lifestyle, diet, pre analytical aspect, analytical aspect and method of reference interval calculation Most of laboratories adopt reference intervals provided from manufacturers without on-site testing of healthy individuals. According to WHO Reference individual that is healthy individual selected for comparison using defined criteria with healthy state that is a state of complete physical, mental, social well being and not merely absence of disease selection of reference individuals is direct that the individuals are selected from a parent population using defined criteria and indirect that the individuals are not considered, but certain statistical methods an' applied to analytical values in a laboratory database to obtain estimates with specified characteristics. This proves the importance of establishing reference range in Indian population using the international federation of clinical chemistry IFCC and clinical and laboratory standards institute (CLSI) guidelines in clinical laboratory.

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The Performance Characteristics of the Diagnostic Laboratory- Biochemistry

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System can be defined in such a way by which it can fulfill the requirement of the clients to maintain the all quality of the service, education and research. Part wise it can be stated as 1. Pre-pre analytical part. 2 Pre analytical parts. 3. Analytical part. 4. Post analytical part. 5. Beyond the post analytical part. Componentwise it can be mentioned as 1. Human automated, robotic, highly skilled. 2. Fully automated machine. 3. Semi automated machine. 4. Human non automated, semi skilled. 5. Method, A) Follows fully automated procedural sheet. B) Semi automated & partly done by skilled person. C) Manual Procedure follows. 6. Data feeder automated/ non-automated. 7. Data interpreter automated/ non-automated. 8. Data distributer automated/ non-automated. 9. Application / properly utilization of generated data. 10. Monitoring of all component. Patient test results are meaningless without QC. Darts are the QC results obtained. Dart distribution is indicator of precision (repeatability). Frequency of total darts on the board from darts thrown measures the test system reliability. Quality control does not prevent pre examination or post examination errors. QC is not equals to QA. QA is equals to QC + Process measurement and monitoring + Pre examination processes + Analytic processes + Post examination processes as well as Evaluation & Audit and Inter laboratory comparison. QMS is integrated processes. QMS is based on processes 1) Management responsibility, 2) Resource management, 3) Laboratory user requirement & satisfaction (i.e. a customer focus), 4) Workflow process sequence (the same for any type, size, scope, discipline, location of laboratory, pre examination, examination, post examination processes), 5) Measurement and analysis and 6)

Continual improvement. Key elements of quality policy are the commitment to good professional practices and commitment to ethical practices compliance requirements. Goals and objectives of quality policy are 1) Improve customer service, 2) Improve report quality and 3) Decrease failure cost.

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Use of Six-Sigma for Quality Control for Biochemistry Parameters

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The most important duty of the clinical laboratory is to provide the accurate test results to patient on which decisions of the physicians rely. Hence, the clinical laboratory must undergo performance testing regularly. One of the methods of performance testing adopted nowadays is six-sigma. TQM of the clinical laboratory can also be achieved by use of six-sigma. The biochemistry analyzers used were BT 1500 and BT 3500 in TUTH Biochemistry Laboratory. 12 clinical analytes were measured using both analyzers for a period of one month, April 2019. IQC performed routinely for both levels were noted from both analyzers and used for calculation of coefficient of variation (CV%). Bias was estimated based on the difference of the average obtained for each analyte from the target values provided. Values for Total Allowable Errors (TEa), were taken from Clinical laboratories Improvement Act guidelines for various clinical analytes. Sigma values were calculated using- CV%, percentage bias and TEa. Sigma values greater than 6 were found for Aspartate aminotransferase (L2), Alanine aminotransferase which shows less strict QC rules are needed to be followed for high error detection and low false rejection. Sigma-values between 3 and 6 are found for uric acid for control levels (L1 and L2), aspartate amino transferase for control level (L1) in both analyzers. Glucose (L1), Total protein (L2) in BT1500, Triglyceride for both level of controls (L1 and L2) in BT3500 - demanding more QC rules to be implicated. Less than 3 sigma values were obtained for parameters- Urea, Creatinine, Albumin, Triglyceride, Total Cholesterol, Alkaline phosphatase, Magnesium for both level of controls (L1 and L2) in both the analyzers indicating the need towards the improvement in these methods. Incorporation of Six-sigma rule would be useful for evaluation of performance testing of clinical laboratories.

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Establishment of Reference Interval for Liver Biochemical Parameters in Apparently Healthy Nepalese Adults

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Reference interval (RI) is a set of values important for decision making support tool used for interpretation of numerical laboratory reports. The quality of reference interval and test result is equally important for result interpretation. Mostly, published reference interval not represents specific reference population. There is no information of reference interval for liver specific biochemical parameters in Nepalese population. So, we aim to establish the reference interval for liver parameters in Nepalese people. In this study total of 617 apparently healthy Nepalese volunteers of age 18-65 years were selected randomly from five major cities of Nepal, using IFCC/C-RIDL guideline (C28-A3). Volunteers were requested to avoid excessive physical exertion/exercise/excessive eating, drinking and fast overnight for 10-12 hour. Blood samples were collected from 120±10 subjects from each five centers of the country between 7:00-10:00 am, serum were separated and refrigerated at-20 in cryo-vials. Liver biochemical parameters were measured by fully automated biochemistry analyzer, Beckman Coulter (BC480). Eighty five results were excluded, applying Latent Abnormal Value Exclusion (LAVE). Reference interval is derived by parametric method. The lower limit and upper of RI derived for Total protein (67.0-82.0 g/L), Albumin (41.0-52.0 g/L), Total Bilirubin (2.70-21.90 μmol/L), Aspartate amino transferase (AST) (7.0-37.0 U/L), Alanine amino transferase (ALT) (3.0-44.0 U/L), Alkaline phosphatase (ALP) (131.0-399.0 U/L) and Gamma glutamyl transferase (GGT) (10.0-81.0 U/L) Nepalese health care providers/ clinicians can use this reference interval of liver parameters for diagnosis, treatment and monitoring of disease.

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A Study of Preanalytical Errors and it's Impact on Turnaround Time (TAT) of Biochemistry Laboratory in a Tertiary Health Centre

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Pre analytical phase is one of the most important and critical step in a diagnostic laboratory. Each laboratory should have documented procedure for preanalytical activities to ensure the validity of the testing and improving the turnaround time (TAT). Aim of this study was to identify and evaluate the preanalytical errors and its impact on TAT. Study was carried out in samples received by biochemistry laboratory, Silchar Medical College and hospital from January 2019 to June 2019, TAT was also evaluated during this period. This study included 36,900 samples. 20,850 samples were received from outdoor department and 16,050 samples were collected from indoor wards. 5% of samples (1,845 out of 36,900 samples) were found as insufficient volume, 2% of samples (738 out of 36,900) were rejected as hemolysed. 1% of sample (369 out of 36,900) were found as mis-labelled, incomplete requisition form and sample in wrong vial. Preanalytical TAT was found 58.06%, analytical TAT was 25.8% and post analytical TAT was 16.3% of total TAT of laboratory respectively. 22.2% of preanalytical TAT was observed in the step of data entry to sample collection, 50% in the sample collection to reception step and 27.7% in the step of sample reception to sample processing for testing. 60.7% of reports exceeding the predefined TAT (120 minutes) of laboratory due to delay in pre analytical phase. Our study revealed that insufficient volume and hemolysed sample was the two major preanalytical errors. Delay in sample collected and reception step of preanalytical phase increased the total TAT of laboratory. This study was to outline suggestive measures how to reduce preanalytical errors and improve turnaround time (TAT).

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To Study the Perception of Post Graduate Students of Pathology Towards Introduction of Applied Biochemistry Teaching and its Effectiveness

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Biochemistry is one of the most important fundamental sciences in medical curriculum and its knowledge is mandatory in clinical practice or in a diagnostic laboratory. It is taught in the first professional year of MBBS, hence recalling the subject in later years of clinical study becomes difficult for students. The present study was planned with an objective of exploring the students' perception about need and application of Biochemistry in the Post Graduate years of studying Pathology and to evaluate its effectiveness. Twenty-one post graduate students of Pathology were subjected to pre-test for testing knowledge of the topic of Biochemistry (Inborn errors of Metabolism) learnt during the first year of MBBS. The topic was then again taught to them in two sessions by interactive small group discussion. After the sessions post-test was conducted and feedback was obtained in the form of questionnaire. The assessments of both pre-test and post-test as well as feedback analysis using Likert scale were carried out. The mean of pre-test score and the post test score was calculated and learning gain was found to be 57% for entire class. The feedback analysis of students showed that 86% students strongly agree and remaining 14% agree that this learning experience increased their understanding and their existing knowledge and more than 75% students strongly agree and 25% agree that teaching applied Biochemistry should be incorporated during post graduate teaching. Their perception revealed that Applied Biochemistry should be incorporated during Post graduate teaching in Pathology. The feedback analysis showed reintroducing Biochemistry teaching in their post graduate course may help in improving understanding and existing knowledge of the subject. The learning experience can be improved significantly by integration of basic subjects at postgraduate level.

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Perception of Medical Undergraduate Students in Biochemistry Subject Towards Advance Planned Tutorials

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Tutorial is a method of transferring knowledge and may be used as a part of a learning process. In present scenario, tutorial lacks uniformity and do not encourage participation of all the students. The principles used in active learning are said to promote student interactivity as well as encourage them to become self learners. Tutorials help to develop, test and clarify their ideas which are taught in lecture. Aim of proposed study is to evaluate effectiveness of advance planned tutorial through pre-test and post-test score of first MBBS students in Biochemistry. The study was carried out in the department of Biochemistry at Jawaharlal Nehru Medical College, Datta Meghe Institute of Medical Sciences, Sawangi (M), Wardha. A total of 70 students were randomised into four groups (17-18 students in each groups). Frequency of meeting-once in a week. Duration of session-one hour to two hours. Proper defining of learning objectives and communicating them to students

well in advance. Providing tutor guide to faculty to facilitate the proper, structured and uniform discussion in all batches. Ensuring the availability all resources required for conduction of tutorial. Mean difference score of UG Students by traditional method was 2.55±1.30 and using advance planned tutorials it was 4.28±1.55 by using Mann Whitney U Test Statistical significant found in mean difference in perception score by two methods (Z=6.40, p=0.0001). The study depicted advance planned tutorial more effective than the traditional tutorial as shown by improvement in students performance.

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7 Cases of Formiminoglutamic Aciduria Detected Through Newborn Screening

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Formiminoglutamic aciduria is the second most common inherited disorders of folate metabolism. The deficiency is due to mutations in the FTCD-encoded bifunctional protein, which is required for histidine and folate metabolism. It was first discovered in a Japanese family with a severe intellectual disability, megaloblastic anemia and high excretion of formiminoglutamate (FIGLU) cases. Through the newborn screening we have detected elevation of FIGLU in children with no symptoms that suggest a mild phenotype. This is a retrospective analysis of the heel's dry blood done inside the newborn screening program. Aminoacids and acylcarnitines profiles were analysed with a commercial kit from MassChrom (Chromosystem®) by tandem mass spectrometry, API3200 (Sciex®). The study included newborns with birth date between April 2010 and December 2018. 362,152 newborn were analysed, being detected 7 cases with elevated FIGLU concentration in dry blood. The positive results were biochemically confirmed in plasma in all of them. In 4 cases, a histidine increased were detected, although none evidence of folate alteration were seen in any of the cases. 3 newborn were found to have elevation of methylmalonate and in one of them a B12 deficiency. In 2 children elevated excretion of FIGLU were seen, while in the other 5 cases a high excretion of hydantoin-5-propionate (FIGLU's product of degradation) were evidenced. After the revision of their medical records, we did not find any references to neurological development delay in the children. The results of these 7 asymptomatic cases detected by newborn screening raise doubts about how we should manage and follow these patients. We consider that a mention in their medical records could be useful to future revisions and we think it is necessary to make genetic studies of FTCD gene in all the cases in order to stablish the pathogenicity of the founded variants.

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Utility of Untargeted Lipidomics in Prediction of Pre-eclampsia

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Objective Identify biomarkers for the early prediction of preeclampsia by of non-targeted lipidomics and enhance the recognition of the pathogenesis of preeclampsia. Pregnant women identified with high risk preeclampsia factors were enrolled and divided into two groups according to whether or not they developed preeclampsia in subsequent follow-up. Total 66 serum samples from two group were analyzed for lipid metabolite profiling. Univariate statistical analysis with fold change value and Wilcoxon rank sum test of nonparametric test, pattern recognition methods such as principal component analysis (PCA) and partial least squares discriminant (PLSDA) were used for screening metabolites. Differential metabolites were then analyzed for metabolic pathways and the degree of enrichment. Total 42 metabolites were identified, and three metabolic pathways were recognized including sphingolipid metabolism, glycerophospholipid metabolism and primary bile acid biosynthesis. Differential metabolites are mainly involved in alpha-linolenic acid and linolenic acid metabolism, plasmalogen synthesis, mitochondrial oxidation of long-chain saturated fatty acids, sphingolipid metabolism and bile acid biosynthesis. Lipid metabolomics can effectively distinguish the preeclampsia group from the control group. We found that glycerophospholipid metabolism, primary bile acid metabolism, alpha-linolenic acid and linolenic acid metabolism, and choline metabolism may contribute to preeclampsia pathogenesis.

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Assessment of Organophosphate Pesticides Exposure in Male Patients with Idiopathic Abnormal Semen Analysis- A Pilot Study

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Organophosphate (OP) pesticides are major environmental contaminants in the developing countries because of its widespread use in agriculture. OP pesticides and their residues are known to alter sperm concentration, quality, vitality, motility, impair spermatogenesis by affecting the Sertoli and Leydig cells and alter the hypothalamic-pituitary axis by acting as endocrine disrupting chemicals. As the prevalence of idiopathic male infertility is rising, it is important to search environmental causes of infertility. Therefore, the objective of this pilot study was to assess the OP pesticide exposure in men with idiopathic abnormal semen analysis. All men attending the infertility Clinic, JIPMER, Pondicherry for inability of their spouses to conceive after at least one year of unprotected intercourse were included in the study. Fifty men with abnormal semen analysis without any identifiable underlying pathology were recruited as cases and fifty men with normal semen analysis were recruited as controls. Details history, general and systemic examination was carried out. OP pesticides exposure among study population were determined by plasma cholinesterase and acetylcholinesterase levels (spectrophotometric kits) according to the Proudfoot classification and by measuring urinary OP metabolites i.e. dimethyl phosphate, diethyl thiophosphate, diethyl dithiophosphate by gas chromatography mass spectrometry. Plasma cholinesterase and acetylcholinesterase levels were significantly lower in cases as compared to controls. Cases (20%) had significantly higher OP exposure as compared to controls (4%). Cases with OP exposure (20%) showed no significant difference with respect to socioeconomic status, type of diets, source of water, smoking and alcoholics as compared to non-OP exposed cases (80%). Farmers staying in rural area had significantly lower incidence of OP exposure. In conclusion, men with abnormal semen analysis had significantly higher OP exposure as compared to men with normal semen analysis.

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DNA Methylation in Promoter Region of P15 Gene in Patients of Acute Myeloid Leukaemia and Myelodysplastic Syndrome

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Acute Myeloid Leukemia (AML) & Myelodysplastic syndrome (MDS) are hematological malignancies with ineffective hematopoiesis. Epigenetic modifications like DNA methylation and histone modifications are changes in DNA structure that do not involve sequence changes but are stably inherited. DNA methylation in promoter region results in transcriptional inactivation of tumor suppressor genes. Silencing of P15 gene can provide a selective advantage for tumors cells. Demethylating agents as anti-cancer drugs, have potential to revert the expression of P15 gene. Role of promoter methylation in p15 gene among AML & MDS and use of demethylating agents for treatment of AML & MDS are recent advances to be considered. Promoter Methylation in P15 gene in these malignancies varies worldwide due to environmental and ethnic variations. Due to paucity of Indian data and considering the therapeutic potential which methylation pattern offers, it is imperative to establish the methylation pattern among Indian patients. To study the DNA Methylation status in the promoter region of P15 gene in forty blood sample of AML & MDS cases and twenty five controls. DNA was extracted from forty one cases of AML & MDS patient's blood sample and controls. Extracted DNA was Bisulphite converted followed by MS-PCR. Amplified products were run on gel electrophoresis and analyzed under UV transilluminator. DNA methylation in promoter region of P15 gene in AML and MDS patients was found 72.7% and 62.5% respectively. In combined data analysis (AML+MDS) positivity was 70.7%, whilst all controls were negative. On statistical analysis study outcome were significant. Promoter P15 DNA methylation pattern in this study represent a novel additional tool to define the epigenetic subset in AML and MDS patients who might be benefitted from demethylating agents, thus providing the molecular basis for targeted therapeutic approaches, monitoring its efficacy and prognostication.

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First Trimester Down's Syndrome Screening: A Novel Approach

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In the past decade, one of the major developments in prenatal screening has been two new maternal serum markers of Down syndrome, placental growth factor (PlGF) and α-fetoprotein (AFP), which become available in the first trimester. A first-trimester serum-only screening test based on existing markers, PAPP-A, free β-hCG, together with PlGF and AFP, might have a discriminatory power equivalent to a second-trimester Quad test. The aim of this study was to assess the screening performance for Down syndrome using 1st trimester screening (FTS) and two additional markers, serum placental growth factor (PlGF) and α-fetoprotein (AFP). A pilot retrospective case-control study of 1013 unaffected pregnancies and 67 pregnancies affected by Down syndrome was conducted. Serum samples were tested for pregnancy-associated plasma protein A (PAPPA), free-β human chorionic gonadotrophin, placental growth factor (PLGF), and α-fetoprotein (AFP), and results were expressed as multiples of median (MoM). Multivariate Gaussian modelling was used to calculate risks for different combinations of markers and to predict the detection rate (DR) and false positive rate (FPR). The predicted performance of enhanced FTS (FTS plus PlGF and AFP) was compared with usual FTS; the performance with and without nuchal translucency (1st trimester quad) was also assessed. For the pregnancies affected with Down's syndrome, the median PlGF level was 0.622 MoM and median AFP 0.764 MoM. Adding PlGF and AFP improved the screening performance. At 5% FPR, DR increased by 3.8% from 89.6% to 93.4% using enhanced FTS combined with nuchal translucency. When assessed without the nuchal translucency, at 5% FPR the DR using enhanced FTS was 81.8%. The performance of FTS is enhanced by adding PlGF and AFP both with and without nuchal translucency.

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Sickle Cell Trait Presenting as Chronic Calcific Pancreatitis with Pseudocyst

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Pancreatic pseudocyst in pediatric population is quite rare, which occurs because of pancreatic insult with ductal disruption and leakage of pancreatic enzymes into surrounding soft tissues. Sickle cell disease may cause acute pancreatitis either due to gall stones obstructing the pancreatic duct or by vaso occlusive mechanism. However, chronic pancreatitis is a very rare complication in sickle cell anemia. We report a case of a child presenting with acute pancreatitis with history of similar episodes repeating at every 3-4 months for 1 year. He was a diagnosed case of sickle cell trait with HbS-25.6%. A localized tender swelling over left subcostal area was found, which on deep palpation revealed smooth, cystic lump. Initial investigations revealed increased serum Lipase (1875 U/L) and amylase (495 U/L). Chronic pancreatitis with pancreatic pseudocyst was kept as provisional diagnosis. USG Abdomen and CT abdomen confirmed the diagnosis. Etiological work up for the possible pathogenesis of chronic pancreatitis including Calcium, PTH, Lipid profile, and ANA did not reveal anything except sickle cell trait. HPLC showed HbA-58.5%, HbA2-3.4%, HbS-27.2% HbF-2.4%, in consistence with the diagnosis of sickle cell trait. Although sickle cell trait is benign carrier state, it is associated with rare but fatal renal medullary cancer, exercise- related deaths, splenic infarction, hematuria, hyposthenuria, venous thromboembolism, complicated hyphema, and foetal loss. People with sickle cell trait often experience subclinical tissue infarction from microvascular obstruction by rigid erythrocytes. This case represents a rare association between chronic pancreatitis and sickle cell trait.

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Serial Nutritional Assessment of Breast Milk in Mothers of Very Preterm (<32 Weeks) Neonates

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Mother's milk is the recommended food for all preterm neonates. Apart from individual variations, human breast milk also shows considerable differences based on gestational age and post-partum day. Giving expressed breast milk only, to these preterm infants does not meet the recommended daily intakes and hence is often supplemented with human milk fortifiers. For optimal nutrition of preterm neonates, it is desirable to know the nutritional composition of mother's milk on different postpartum days, in order to achieve desirable levels of supplementation. So, the present study was conducted to serially evaluate the nutritional composition of breast milk in mothers of very preterm (<32 weeks) neonates. 3-5ml of breast milk was collected by machine or manual expression on post-partum day 7, day 14 and day 21 from mothers (N=35) of very preterm neonates (birth before 32 weeks gestation). Milk was stored at -20°C until analysis. Lactose (the predominant carbohydrate in breast milk) was measured using commercially available kit (Sigma-Aldrich, St. Louis, USA) using colorimetric method. Total proteins, triacylglycerol (the major fat in breast milk), total calcium and phosphate were measured on chemistry

autoanalyzer using photometric methods. Total calories were calculated based on the following conversion factors: Lactose-3.95 kcal/g, Protein-5.65 kcal/g, fat- 9.25 kcal/g as reported by Anderson et al. (1981). On statistical analysis, lactose was found to be significantly increased (3.10±0.84, 3.49±0.77, 3.70±0.60 in g/dL) on day 7, day 14 and day 21 post-partum respectively; whereas, protein (1.11±0.27, 0.91±0.17, 0.85±0.21 in g/dL), total calcium (26.60±7.27, 23.69±6.32, 23.31±5.65 in mg/dL) and phosphate (7.49±2.16, 6.51±1.74, 6.51±1.99 in mg/dL) were found to be significantly decreased. Similar significant difference was not observed with triacylglycerol and total calories. Besides, values of protein, lactose, phosphate and calories in breast milk of mothers delivering prematurely were found to be lower than currently accepted nutrient levels (Boyce et al. 2016).

P-265

Pregnancy Associated Plasma Protein A (PAPP-A) not a Reliable Criteria for Risk Assessment of Aneuploidy in Low Maternal Weight Pregnancies

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Prenatal diagnosis for aneuploidies by Combined Dual Marker Test using free Beta Human Chorionic Gonadotropin (β-HCG), Pregnancy associated plasma protein-A (PAPP-A) and Nuchal Translucency (NT) are paraphernalia for risk assessment of Down syndrome (trisomy 21), Edwards syndrome (trisomy 18) and Patau syndrome (Trisomy 13). PAPP-A is a protease acting on Insulin like growth factor binding proteins and seen only in pregnancy and absent in non-pregnant women, men or cord blood. Levels of PAPP-A in pregnancy are decreased in trisomy 21, 13 and 18, similar trends are seen in Monosomy X and Digynic Triploidy, Pre-eclampsia and Small for Gestational Age (SGA) babies. Low maternal weight is defined as pre-gravid weight less than 45.35 kg or Body Mass Index (BMI) of ≤ 19.8 kg/m^2. A total of 341 samples were analysed and maternal serum PAPP-A levels were measured during gestational period of 11 weeks to 13 weeks and 6 days. Females below 45.35 kg were taken as cases (27) and above 45.35 kg as controls (314). Twin pregnancies and those with other complications like GDM, PIH were excluded from the study. The study was conducted using Cobas e411 system (Roche Diagnostics, Penzberg, Germany). T 21, 13 and 18 Risk were assessed using Ssdw software based on Bayes theorem. Values for PAPP-A for the control group showed normal Gaussian distribution whereas for cases it was skewed based on Shapiro-Wilk test with an alpha level of 0.05. For group I the mean was 14118.37 and standard deviation was 17566.61 and for group II the mean was 5473.05 with a standard deviation of 3581.65. Student t test showed statistical significance with p value <0.0000001. The present study has demonstrated that PAPP-A value is unreliable in pregnancies with low maternal weight because of skewed distribution and abnormal production and requires further investigations.

P-266

Does Body Mass Index (BMI) Correlate with Incidence of Gestational Diabetes Mellitus (GDM) in North Indian Pregnant Mothers?

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Gestational Diabetes Mellitus (GDM) is a well studied entity. It has proven association with significant insulin resistance (IR), feto-maternal complications and is more prevalent in Asian population. Body Mass Index (BMI) cut off has been lowered among Asians thanks to higher prevalence of abdominal (central) obesity. BMI has proven association with cardiovascular diseases and IR. To find the association of BMI with GDM in pregnant mothers compared to their normal counterparts, 179 pregnant women were included in this study according to inclusion/ exclusion criteria. Study participants were classified by 2-hour post glucose load blood glucose (PGBS) level (DIPSI guideline) irrespective of period of gestation (POG). Participants with PGBS values ≥140 mg/dl were diagnosed as GDM. PGBS, glycated hemoglobin (HbA1c) and BMI of pregnant mothers were recorded. POG at the time point of interaction with study participants in this cross-sectional study was noted. Both HbA1c and PGBS had significant positive correlation with BMI among all participants. PGBS among normoglycemic and HbA1c among GDM mothers exhibited significant difference among participants falling into various BMI categories (South Asian Classification). Similarly HbA1c in first and PGBS in second trimester mothers showed significant difference among participants falling into different BMI categories. Again PGBS was significantly different during early second trimester (POG-II), but not during late second trimester (POG-III). Body mass index (BMI) failed to exhibit any association with GDM even in high risk North Indian population. HbA1c cannot be used as diagnostic marker for GDM because of lower levels during all three trimesters of pregnancy.

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Implications of Non-Correlation Between Serum TSH Level and Serum Calcium Level in Pre-eclampsia

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Pre-eclampsia is characterised by high blood pressure and proteinuria (>300mg/24 hrs) developing after 20 weeks of gestation. Despite evidences of thyroid hormones influencing serum calcium levels in hypothyroid patients, and alteration of thyroid profile and serum calcium levels in preeclampsia, no study regarding the interrelationship of these parameters during pre-eclamptic conditions is done till date. The study was therefore conducted to estimate and analyse serum calcium level, level of hormones related to thyroid profile, their inter-correlation and their correlation with occurrence of preeclampsia. A case-control study was carried out in this regard that comprised of 80 pre-eclamptic cases and 90 normotensive controls. Their blood samples were analysed for significant differences ($p<0.05$) in total serum calcium level and thyroid profile among cases and controls. Mean total serum calcium levels in cases were lower than in controls. Mean serum TSH levels in cases were higher than that in controls. No significant differences were observed in fT3 and fT4 levels among cases and controls. Results show serum calcium level <8.2 mg/dl had a higher odds ratio of 10.8 as compared to serum TSH levels >5.7μIU/ml with an odds ratio of 4.98. Interestingly, we observed that an anticipated inverse TSH-Calcium correlation was absent in both cases and controls. Serum calcium level is a more significant predisposing risk factor for pre-eclamptic occurrence than serum TSH level. A non-correlation signifies that subclinical hypothyroidism (TSH levels >5.7μIU/ml, normal fT3 and normal fT4 levels) and hypocalcaemia (serum calcium level <8.2 mg/dl) may associate independently to occurrence of preeclampsia. Serum TSH levels and serum calcium level therefore may have other factors to account for their variability during pregnancy and preeclampsia. Screening and analysis of thyroid profile and serum calcium levels during pregnancy is highly recommended in order to minimise risk of pre-eclamptic occurrence.

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Novel Accurate LC-MS/MS Method for Quantitative Determination of Z-lumirubin

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Phototherapy (PT) represents a standard treatment for neonatal jaundice. However, no validated clinical method for determination of bilirubin photo oxidation products is available. Thus, the aim of our study was to establish such method for clinical use. A LC-MS/MS assay for simultaneous determination of Z-lumirubin (LR, the major bilirubin photo oxidation product) and unconjugated bilirubin (UCB) was conducted, using mesobilirubin as an internal standard. LR was prepared by photo-irradiation of UCB and purified by TLC. The assay was tested on human sera from neonates treated with standard PT. Samples were separated on HPLC system with a Poroshell 120 EC-C18 column using a binary mobile phase (NH4F in water/methanol) and the analytes were detected in triple quadrupole mass spectrometer operating in a positive SRM mode. The method was linear up to 100 and 400 μmol/L for LR and UCB, respectively, with submicromolar limits of detection and validity parameters relevant for use in clinical chemistry. Exposure of newborns to PT raised serum LR concentrations three-fold ($p<0.01$), but the absolute concentrations were surprisingly low (6.4±2.9 μmol/L), despite dramatic decrease of serum UCB concentrations (232.6±41.2 vs. 176.0±58.1 μmol/L, $p<0.01$) suggesting formation of additional bilirubin photo oxidation products and presumably increased urinary and biliary secretion of these polar products. In conclusions, we established and validated for clinical use a LC-MS/MS method for the simultaneous determination of LR and UCB in human serum. This method should help to monitor neonates on PT, as well as to improve our understanding of kinetics and biology of bilirubin photo oxidation products.

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Dried Blood Spots -Total Antioxidant Capacity Status as Early Biochemical Marker: In Inborn Errors of Metabolism

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The present study is to investigate the total antioxidant activity (TAC) in suspected neonates of inborn errors of metabolism with high risk, moderate risk and low risk. Dried blood spots have

potential use in remote health applications for individual and population diagnosis and can enable epidemiological surveillance for known and unknown diseases. Dried blood spot sampling is most common method for collection, storing, transporting and analyzing a variety of human body fluids. While newborn screening is done in early period, the balance residual samples were used for specific secondary research studies with patient consent. Here we are using the secondary studies of DBS for total antioxidant activity as one of the biomarkers to find out the disease condition. TAC was evaluated in 449 neonates including high, moderate and low risk neonates. The risk variables included were birth weight, apgar score, gestational age and previous complications of pregnancy. In DBS -TAC method, the sample is eluted by using phosphate buffered saline. After sample preparation, TAC by using FRAP assay (ferric reducing antioxidant power assay) were estimated among three groups with risk indicators. Statistical comparisons and correlations at 5% level of significance were determined. The mean TAC concentration was significantly elevated in high risk group. The mean TAC for moderate and high-risk group (Mean ± SD values were 700.4 ± 100.6 μmol/L and 510.4 ± 110 μmol/L respectively) patients were significantly reduced compared with control group (Mean ± S.D. valued was 954.8 ± 132.2 μmol/L) ($p < 0.01$). This result clearly shows that antioxidant capacity in high risk neonates in comparison with controls increases significantly with progression of disease i.e., High risk > Moderate risk > Low risk. DBS-TAC can be used as an early biochemical marker for oxidative stress in high risk for IEM (Inborn errors of metabolism) which may result in reduced tissue damage by free radicals and help to monitor and optimize antioxidant therapy in such high risk neonates.

P-270

Osteoprotegerin Gene Polymorphism in Gestational Diabetes Mellitus in South Indian Population

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Gestational diabetes mellitus (GDM) is associated with future risk of type 2 diabetes mellitus (DM), cardiovascular disease (CVD) etc. Osteoprotegerin (OPG) is one of the factors secreted from the skeletal myocytes and is involved in the pathogenesis of insulin resistance, and it is emerging as an independent biomarker for atherosclerosis. Even at the genetic level, some SNPs in the OPG gene, which alter the expression of OPG, have shown to be related to the development of CVD. Since the risk of developing T2DM and CVD is very high in GDM patients, it is relevant to study new biomarkers for early diagnosis of complications from GDM. The anthropometry, OGTT, lipid profile, osteoprotegerin, insulin, TNF-α, genotyping for OPG gene T950C polymorphism (rs2073617) were measured in 73 GDM patients and compared with age, gestational age matched normal antenatal women. The median insulin level, HOMA IR was significantly higher in the GDM group when compared to the controls ($p<0.01$) whereas QUICKI ($p<0.01$) and HOMA-β ($p=0.047$) were significantly lower. Proatherogenic indices and TNFα were significantly higher in the GDM group when compared with the control group. The allele frequency of the T950C polymorphism was similar between the groups and was not in Hardy Weinberg equilibrium. However, when the GDM patients were sub grouped based on their alleles, OPG was significantly higher in the heterozygous CT group ($P=0.042$). Furthermore, correlation between OPG levels and other study parameters in the TT vs CT group showed significant positive correlation with insulin resistance index HOMA-IR ($r=0.314$, $p=0.016$) and negative correlation with insulin sensitivity index QUICKI ($r= -0.314$, $p=0.016$). This suggests that the ladies having the CT allele have a greater scope of developing insulin resistance and cardiovascular risk in their future. It may serve as a biomarker for predicting the outcomes and future complications in the postnatal period.

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Cord Blood Lipid Profile in Term Neonates and their Correlation to Birth Weight

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Human fetuses are known to permanently change their physiology and metabolism to adapt to limited supply of nutrients in utero. These programmed changes can later be the cause for the origin of diseases like coronary artery disease, diabetes mellitus and hypertension. Fetal maturation appears to be associated with lifelong changes in metabolic functions. Both genetic and environmental factors can influence the cord blood lipid profile. Prevention of CAD must be initiated at younger ages, because there is an opportunity to begin preventive interventions for cardiovascular risk factors; furthermore, the study of CAD risk factors is greatly recommended during this period. This cross-sectional observational study was done in the department of Biochemistry and Obs.- Gyn., IGIMS, Patna over a period of six months. A total of 100 cord blood samples were collected for lipid profile analysis. There were 56 male and 44 female newborns in this study. The number of normal vaginal deliveries were 52 while 42 babies were delivered by LSCS. The mean values of different lipid profile parameters were -Total Cholesterol - 59.6±21.3 mg/dl, HDL - 24.4±9.0 mg/dl , LDL-33.1±15.3 mg/dl and TG-33.6±13.8 mg/dl respectively. The mean body weight of newborns were 2.96±0.54 kg. A negative correlation was found between total cholesterol level and birth weight. All the lipid profile parameters were found to be

significantly higher in small for gestational age babies as compared to normal weight babies. There is no significant difference in the cord blood lipid profile parameters of male and female neonates. Low birth weight babies are at risk of atherosclerosis and CAD.

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A Rare Case Report of Malonic Aciduria Diagnosed by Newborn Screening In Maharashtra

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Malonic aciduria is a rare autosomal recessive organic acid disorder. It is caused by malonyl-CoA decarboxylase (MCD) deficiency. Malonyl-CoA decarboxylase is expressed by the MLYCD gene on chromosome 16 and catalyzes the decarboxylation of malonyl-CoA to acetyl-CoA. Until now, very few cases have been reported in the literature. With the widespread use of tandem mass spectrometry methods for amino acid/acylcarnitine (AA/AC) screening on dried blood spots (DBS), this condition can be readily diagnosed and can be included in the organic acid screen in NBS programs. In Maharashtra, we report the first case of an asymptomatic baby screened and diagnosed with malonic aciduria through NBS. It is challenging to diagnose such rare inborn errors of metabolism in NBS programs. Access to confirmatory tests for plasma acylcarnitines, urine organic acids and molecular genetics is the key in the differentiation and confirmation of these disorders .The clinical phenotype of malonic aciduria is variable and the pathophysiology is not fully understood. There is no established guidance or recommendations regarding the appropriate treatment regimen, dietary therapy or regular follow-up of these patients. Most available evidence for treatment is based on a single study or case report.

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Role of Macronutrients in Prevention of Pre-Eclampsia

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Hypertensive disorders of pregnancy are the most significant and intriguing unsolved problems in obstetrics. These disorders complicate 5 to 10 percent of all pregnancies, and together, they are one of the deadly triads along with hemorrhage and infection that contributes significantly to maternal morbidity and mortality rates. Alterations of phosphate and most notably calcium excretion are characteristic findings of hypertension. Serum calcium, magnesium, serum phosphorus decrease in preeclampsia. The present study on these parameters is essential in clinically diagnosed patients of hypertensive disorders to show their role in pathogenesis and also to ascertain their role as biochemical markers of the disease for prevention, early diagnosis, and to monitor the prognosis of hypertensive disorders of pregnancy. A cross-sectional study was carried out in the Department of Biochemistry, Andhra Medical College, Visakhapatnam, with approval from the Institutional Scientific and Ethics Committee from December 2017 to July 2019. 42 pregnant women meeting the criteria for pre-eclampsia attending department of obstetrics in the third trimester were selected and compared to 42 normotensive pregnant women in the third trimester. The [mean ± SD] serum calcium level in cases was 8.29±0.69 mg/ dl and in controls, 8.94±0.26 mg/dl. The [mean ± SD] serum magnesium level in cases was 1.61 ±0.33 mg/ dl and in controls 2.05±0.28 mg/dl. The [mean ± SD] serum phosphorus levels in cases was 3.68±0.42 mg/dl and in controls 4.24±0.53 mg/dl. The levels of calcium, magnesium, and phosphorus are significantly reduced in pregnant women with pre-eclampsia. Preeclamptic patients with decreased calcium, magnesium and phosphorus have poor maternal and fetal outcomes. Nutritional intervention along with the assessment of their blood levels helps in early detection, prevention of pre-eclampsia and progression to eclampsia.

P-274

Utility of HbA1c as a Tool for Diagnosis of Gestational Diabetes Mellitus and Correlation Between HbA1c and OGTT in Patients Attending Clinics of Tertiary Care Settings in Sri Lanka

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Prevalence of gestational diabetes mellitus (GDM) in Sri Lanka is 13.9% at present and it has been on the rise since 1998, all over the world. As the early diagnosis is directly related to reduction of maternal and fetal morbidity and mortality, many studies have been done on early and correct identification of this disease. Currently a time consuming test, OGTT, is used as the main tool of diagnosis. This study aimed at using HbA1c as an alternative to OGTT in diagnosis of GDM. Hospital based cross sectional study was conducted among 154 GDM positive and negative pregnant women at POA between 24-28 weeks. Attendees with diagnosed Type 2 diabetes mellitus, multiple pregnancies, previous GDM, renal pathologies, hemoglobinopathies and anemia (Hb < 10.5 g/ dL) were excluded. HbA1c of the sample was measured by Sebia 2 flex capillary electrophoresis analyser. Independent t test, correlation coefficient and Receiver Operating Characteristics

(ROC) curve were done using SPSS 21. Mean HbA1c for GDM positive women was significantly higher than that of GDM negative women ($P < 0.05$). Pearson correlation between HbA1c and OGTT were 0.604, 0.683 and 0.66 at 0, 1 and 2 hour respectively. The area under the curve at cutoff value of 5.45%, was 0.845 and sensitivity and specificity at the cutoff were 80% and 82% respectively. HbA1c can't replace OGTT for the diagnosis of GDM. Although the sample is small, findings can be used as a pilot study to establish cutoff values of HbA1c for the diagnosis of GDM.

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Prevalence of Di-George Syndrome with Orofacial Cleft Patients in Indian Population

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Orofacial clefts, also categorised as either Cleft Lip/Palate or Cleft Palate, are the second most common congenital deformity. Worldwide birth prevalence for orofacial clefts is 2.62 per 1000 live births. It can occur as part of mendelian syndromes or isolated/non-syndromic clefts. It is a heterogeneous group of disorders, and varies with ethnicity, gender and cleft type. It can arise through single gene mutations, chromosomal abnormalities and effect of teratogens. The present study was conducted on 25 syndromic orofacial cleft patients present with cardiac anomalies, typical facies, learning disabilities and other common microdeletion syndromes. Genomic DNA was extracted from peripheral blood and multiplex ligation dependent probe amplification was performed for three probe sets, for common microdeletions, Di-George syndrome and subtelomeres. Sequence type electrophoresis was performed using ABI prism 310 genetic analyser. Later, fragment and comparative analysis part was carried out by coffalyser software. We detected 3 patients were with 22q11.2 deletion and shown the association of Di-George syndrome. No cases were detected for the other common microdeletion syndromes. Our work demonstrated that Di-George Syndrome (22q11.2 deletion) is most common syndrome associated with orofacial cleft patients and it can be used as primary screening through the MLPA in orofacial cleft patients.

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Spot Urine Protein Creatinine Ratio as A Predictor of Severity of Nephrotic Syndrome

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Nephrotic syndrome is a nonspecific disorder in which the kidneys are damaged, causing them to leak large amounts of protein (proteinuria at least 3.5 grams per day per 1.73 m^2 body surface area) from the blood into the urine. Aim of present study was to determine Spot PCR as a strong predictor in Nephrotic Syndrome. The present study was conducted at King George Hospital, Visakhapatnam. 25 Patients attending the department of pediatrics were assessed and 25 healthy age matched children were used as control. Urine protein was measured using dye based method, urine Creatinine was measured by Jaffe's method and then ratio was calculated. High level of Spot Protein Creatinine Ratio is observed at acute phase of Nephrotic syndrome and was a risk factor for relapse. Spot protein creatinine level for more severe cases was 20.97±10.44 mg/mg and for less severe cases was 14.94±8.74 mg/mg. Severity of Renal involvement in Nephrotic syndrome was determined by level of proteinuria and spot protein creatinine ratio. Spot protein creatinine ratio is simple to perform and as efficient as 24 hours protein in urine.

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Respiratory Distress in a Preterm Neonate Due to a Milky Pleural Effusion

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Chylothorax is a rare presentation defined as presence of chyle in the pleural space due to leakage from thoracic duct or its tributaries following damage or blockage. It contains cholesterol, triglycerides, chylomicrons and lymphocytes in large amounts. Main clinical manifestations depend on the rate of chyle leakage resulting in respiratory compromise. A neonate born at 25-weeks of gestation (665g) was ventilated due to respiratory distress at birth and surfactant deficient lung disease. She was started on treatment for presumed sepsis. Her chest X-rays revealed obliterated right costophrenic angle indicating a pleural effusion which prompted thoracentesis due to frequent desaturations. Interestingly, pleural fluid was milky in appearance and biochemical and cytological analysis revealed high triglyceride level (57mg/dL), low cholesterol level (<50mg/dL) with lymphocyte predominance on microscopy. The pleural fluid remained uniform during centrifugation and fluid to serum triglyceride ratio was 1.9 and cholesterol ratio was <1

confirming the diagnosis of chylothorax. The baby was kept nil-by-mouth and started on parenteral nutrition following clinical diagnosis of chylothorax and an intercostal tube was inserted to drain the chylothorax. However baby expired on day 12 due to extreme prematurity, sepsis although right side chylothorax had resolved by then. The established cutoff level for triglyceride in pleural fluid for chylothorax is 110 mg/dL. However, this limit could be too high for premature neonates as their intake is low. In such instances the recommendation is to demonstrate the presence of chylomicrons by lipoprotein electrophoresis (gold standard) which may not be freely available. Therefore, fluid to serum triglyceride (>1) and cholesterol (<1) ratios can be conveniently used as the cutoff limits for the diagnosis of chylothorax. Although rare, chylothorax should be considered during differential diagnosis in the presence of milky pleural effusion in neonates which could be due to congenital anomalies.

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Comparison of Serum Lipid Profile between Healthy Pregnant Women and Pregnancy Induced Hypertensive Women

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Hypertension during Pregnancy is one of the leading causes of maternal and perinatal morbidity and mortality. Serum lipid profile plays a very important role in the regulation of blood pressure during pregnancy. The aim of this study was to Compare lipid profile in patients of Pregnancy induced hypertension with that of a healthy pregnant woman. This study was done at Government General Hospital, a tertiary care teaching hospital in Kurnool, Andhra Pradesh. We have taken Pregnant women in the age group of 21-35 years, with a gestational age group of 20-35 weeks, Primigravida/Multigravida with a singleton Pregnancy. This includes 2 groups - 50 Subjects of healthy pregnant women and 50 Pregnancy induced hypertension patients, in whom fasting blood samples were sent for the estimation of serum lipid profile. There was a significant rise in Triglycerides, Total cholesterol, LDL Cholesterol, VLDL Cholesterol & fall in HDL Cholesterol in Pregnancy induced Hypertensive Patients as compared to that of healthy pregnant women. The altered values of lipid profile are associated with pathological process of Pregnancy induced hypertension. It is therefore essential to measure Lipid Profile in pregnant women during whole Pregnancy since it may be useful in early diagnosis, treatment of PIH and for prevention of obstetric complications such as Preeclampsia and eclampsia.

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Profile of Vitamin D and Vitamin D Receptor Polymorphism BsmI and FokI in End Stage Renal Disease Nepalese Patients

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Vitamin D receptor (VDR) genotypes differentiation may cause modification of VDR structural, ultimately leading to changed receptor function, which may alter VDR protein gene expression thereby causing end-stage of renal disease (ESRD). VDR genes polymorphism (BsmI and FokI) are known as reliable markers of abnormal vitamin D signaling pathway. VDR polymorphism Bsml and Fokl may be associated with ESRD. The aim was to find out the status of Vitamin D and to assess the relationship of VDR gene polymorphism in ESRD patients on maintenance hemodialysis. A cross-sectional study involving 207 participants, having 138 ESRD patients from hospitals maintenance hemodialysis and 69 healthy participants from July 2016 to September 2018. For molecular studies, the two major VDR Gene polymorphism (BsmI and FokI) were genotyped by adopting PCR and RFLP techniques. Enrolled ESRD patients were significantly ($P<0.05$) low serum 25 (OH) vitamin D level (13.76±6.59) as compared to healthy control having serum 25 (OH) vitamin D level (32±10.27). BsmI genotyped frequencies as BB (67), Bb (57) and bb (14) in ESRD patients group whereas BB (36), Bb (25) and bb (8) in the healthy control group ($P= 0.775$). FokI genotyped frequencies as FF (74), Ff (56) and ff (8) in ESRD patients group and FF(34), Ff (32) and ff (3) in healthy controls group ($p=0.700$). No statistically significant difference in Vitamin D polymorphism(BsmI and FokI) genotypes frequency could be observed between hemodialysis ESRD patients and healthy controls, which suggesting that pathogenesis of ESRD had no association with Vitamin D receptor BsmI and FokI gene polymorphism. High prevalence of hypovitaminosis D in ESRD patient as compared to healthy control. Vitamin D polymorphism (BsmI and FokI) were not associated with ESRD among Nepalese Population.

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Correlation of Oxidative Stress with Global DNA Methylation in Type 2 DM

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The incidence and prevalence of Type 2 Diabetes Mellitus (T2DM) is increasing. T2DM is a multifactorial, polygenetic disease due to impaired insulin secretion and insulin resistance. Single nucleotide polymorphisms explain less than 20% of disease progression and heritability. Various studies have associated oxidative stress and environmental influence on development of T2DM. Hence, this study was designed with the objective of exploring the association of global DNA methylation and oxidative stress with glycemic control in T2DM patient. The study included 42 T2DM patients (35 to 70 yrs of age) without any complications and an equal number of matched healthy volunteers as controls. An equal number of matched T2DM patients with complication were also included in the study population. A twelve hour overnight fasting blood sample was used to determine HOMA-IR (marker for insulin resistance), Glycated Hb (marker for glycemic control), oxidant load (Ferrous oxidation xylenol orange - FOX2 assay), Total antioxidant capacity (Ferric reducing antioxidant power - FRAP assay) to assertion oxidative stress. Global DNA methylation (epigenetic changes) was estimated by ELISA kits after extraction of DNA by Magnapure (Roche diagnostics). An increased HOMA-IR, oxidant load and DNA methylation was observed in T2DM patients with complications as compared to T2DM patients without any complications. Similar trend was observed among T2DM patients and controls. DNA methylation correlated with oxidative stress and HOMA-IR positively. The study reveals the positive association of DNA methylation with HOMA-IR and oxidative stress, implicating a key role of epigenetics in disease progression and development of complication in T2DM.

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Study of Serum Vitamin B12, Vitamin D3 Levels, Serum Telomerase Changes in Patients with Depression

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Depression affects approximately 121 million people worldwide and is the leading cause of disability. WHO data predicts that depression shall be the second cause of global disease burden by the year 2020. Depression is associated with selected nutritional deficiency such as Vitamin B12, Vitamin D. The pathophysiology of disease development and response to treatment is unclear. Research is being aimed at revealing its pathophysiology at a cellular and molecular level. An important locus of cellular damage and ageing is "Leucocyte telomere length". Telomere shortening is implicated in cellular ageing. Telomerase, a ribonucleoprotein enzyme is responsible for repairing, protecting and maintaining telomere length. Hence, this study aims (i) at evaluating serum Vitamin B12, Vitamin D3 and serum telomerase level in patients with depression; (ii) association of serum telomerase level with severity of depression. The study included 48 patients with depression and equal number of matched healthy volunteers. The severity of depression was assessed by HAMILTON Depression Score. Serum Vitamin B12 and Vitamin D3 were measured by Chemiluminescence kits adapted to Cobas e411, Roche diagnostics. Serum Telomerase was estimated by ELISA kits adapted to automated ELISA system, Biorad. The data was analyzed by the statistical software SPSS version 24.We observed significant lower Vitamin B12, Vitamin D3 and serum Telomerase activity among the patients with depression, as compared to healthy individuals. A significant inverse correlation was observed between serum Telomerase activity and severity of depression. The study implicates the role of serum Telomerase as a novel prognostic marker for depression. Further, and increased study population can elucidate the optimum role of Telomerase in the pathophysiology of depression. We also suggest that status of Vitamin B12 and D3 should be evaluated and nutritional supplement should be given.

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Global DNA Methylation as a Biomarker for Early Detection of Chronic Kidney Disease

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Chronic kidney disease (CKD) is a global health problem affecting health economics. The disease is often associated with increasing age, co-morbidities such as Diabetes Mellitus, Hypertension. The conventional markers used for early detection of CKD are serum urea, creatinine, estimated Glomerular Filtration Rate (eGFR), serum cystatin, Tissue inhibitor of metalloproteinase (TIMP), Monocyte chemoattractant protein (MCP). However, these biomarkers have limitations and cannot predict early or impending CKD, which would aid diagnosis and prognosis and help therapeutic management. Hence, this study is designed to assess the changes in global DNA methylation (a marker of gene expression and epigenetic changes) for early detection of CKD. The study comprises of 204 (51 numbers in each category) individuals in the age group of 35 to 65yrs, in 4 categories -Healthy volunteers, Type 2 Diabetes Mellitus (T2DM) patients without any complications, T2DM patients with nephropathy, CKD patients without T2DM. Renal function test, Fasting Blood Sugar (FBS), Glycated Hb, estimated avg blood glucose, eGFR, Urinary microalbumin was measured by commercial kits adapted to auto analyzer. Estimated glomerular filtration rate was calculated by CG formula. Global DNA methylation was estimated by ELISA kits. A detoriation of renal function tests, eGFR was associated with increase in FBS and HbA1c. The global DNA methylation was also significantly associated with reduced renal function. A multivariate analysis showed global DNA methylation can significantly predict reduced eGFR after adjusting age, HbA1c.We observed DNA methylation to be significantly increased in patients with CKD and was associated with reduced eGFR. Hence, alterations in global DNA methylation can be used as indicator of deteriorating renal function. The limitations of this study are limited sample size.

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MTHFR Gene Polymorphism C677T and DNA Hypermethylation in the Promoter Region of the MTHFR Gene in Coronary Artery Disease Patients (CAD)

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Methylene tetra hydro folate reductase (MTHFR) is a key enzyme to catalyze methylation of Homocysteine (Hcy) to Methionine. High levels of Hcy (hyperhomocysteinemia) have been identified as a risk factor for CAD. Elevations in Hcy might be due to MTHFR (C677T) gene polymorphism (genetic),DNA methylation (epigenetic change).The aim of this study was to assess the correlation of MTHFR gene polymorphismC677TandDNA Hypermethylation in the promoter region of the MTHFR gene with Hyperhomocysteinemia in CAD. The correlation may provide an epigenetic and genetic basis for the prevention, early diagnosis, and treatment of the disorder. It was a retrospective study on 200 - age and sex matched Cases and Controls. SNP(C677T) was done by PCR-RFLP. DNA methylation study was done by Bisulfite conversion method. Hcy was assayed by ELISA. As per our findings the OR for C vs T allele showed that 'T' allele of MTHFR gene genotype was adding 2.98 folds (p = 0.003) risk for the development of CAD, and TT genotype increases 4.5 fold risk of Hyperhomocysteinemia in CAD(OR=4.5, CI=2.7-9.7, P value= 0.02) in case. In MTHFR Promoter region it has been observed that methylation indices were 39% and 18% in the cases and control groups, adding 2.91 folds (p = 0.001) risk for the development of CAD and 87% of cases and 41% controls with methylation had Hyperhomocysteinemia indicating that Hypermethylation add 3.05 times risk of causing Hyperhomocysteinemia (OR=3.05, CI=1.7-5.6, P value≤0.01). MTHFR gene polymorphism causes dysfunctional thermolabile MTHFR enzyme and Hypermethylation in MTHFR gene promoter region leads to reduction in MTHFR enzyme production, which eventually reduces Hcy metabolism leading to Hyperhomocysteinemia, which indicates MTHFR gene C677T polymorphism and MTHFR gene promoter methylation are independent risk factor for CAD, but further more sample size study is required to corroborate the findings.

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The Effect of ANTI-MIRNA 144 on the Expression of a Globin Chain in PBMC (Peripheral Blood Mononuclear Cell) of Major β thalassemia Patients

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The main pathophysiology basis of β-thalassemia is excess unbound α-globin chain due to reduction α-globin expression. Reduction of α-globin chain can lead clinical improvement. Reduction of α-globin chain is an alternative therapy in β major thalassemia patients. microRNA (miRNA) can regulate α-globin chain through targeting transcription factor. Klfd and GATA-1 erythroid transcription factor regulated by miRNA-144. This study was an experimental study using PBMC of major β thalassemia patient. PBMC divided into two groups that were not transfected and transfected anti-miRNA 144. The expression of miRNA-144 were detected using Exiqon's miRCURY LNATM universal RT microRNA PCR. The expression of α-globin chain protein were detected using immunobloting technique.There are differences in the expression of miRNA-144 who have carried out anti-miRNA 144 transfections with those that have not been conducted transfections in PBMC major β thalassemia patients. There are no differences in the expression of α globin chain who have carried out anti-miRNA 144 transfections with those that have not been conducted transfections in PBMC major β thalassemia patients. Based on this study, the administration of anti-miRNA 144 cannot decrease the expression α globin chain expression in PBMC major β thalassemia patients.

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Polymorphism of Vitamin D Receptor Gene Variants in East Indian Women with Polycystic Ovary Syndrome

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Polycystic ovarian syndrome (PCOS), the most common gynecological endocrinopathy. It affects 4-12% of women of reproductive age worldwide. Women with PCOS frequently suffer from metabolic disturbances including insulin resistance (IR), diabetes, obesity, hypertension and dyslipidemia. Evidences suggests that vitamin D deficiency might be a causal factor in the pathogenesis of IR and metabolic syndrome in PCOS women. The aim of this study was to investigate the association of VDR variants (Cdx2 and DHCR7) genes with metabolic and endocrine parameters including 25(OH)D levels in PCOS women. Moreover we examined whether there are associations with PCOS susceptibility. Metabolic, endocrine and anthropometric measurements were performed in 100 PCOS patients and 100 control women. Genotyping of vitamin D receptor (VDR variant) Cdx2 and DHCR7 gene was performed between the groups. Mean serum hydroxy vitamin D [25(OH)D] were significantly lower in PCOS patients compared to controls .In PCOS women ,the VDR Cdx2 "AA" genotype was associated with lower fasting insulin and HOMA-IR. Also the DHCR7 "GG" genotype had a significantly higher risk for 25(OH)D levels <20ng/ml when compared to other genotypes. Data from this study indicate that vitamin D deficiency is more frequent in PCOS patients than in controls. The present findings also suggest VDR gene (Cdx2) and vitamin D level related variant (DHCR7) are associated with metabolic and endocrine parameters including 25(OH)D levels in PCOS women. Thus, vitamin D supplementation have a favorable effect on glucose metabolism as well as overall morbidity in PCOS women.

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Association of MTHFR Gene C677T Polymorphism and Breast Cancer Risk in Population of Bihar

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Cancer is a multifactorial disease that starts when cell grows out of control or cell performs uncontrolled division and crowd out normal cell (American cancer society). Breast cancer (BC) is the leading cause of death among women worldwide and it is a major public health problem. Disturbance in folate metabolism may be involved in predisposition to BC and a specific gene is responsible in folate metabolism. Methylene tetra hydro folate reductase gene (MTHFR) is an important enzyme that is involved in folate metabolism. There are two polymorphism associated with this gene(C677T and A1298C) (4).The functional polymorphism C677T may lead to decreased enzyme activity and affect chemo sensitivity of tumor cell. The aim of the study is to elucidate the association of MTHFR C677T polymorphism and breast cancer risk. MTHFR C677T polymorphism in breast cancer patients (N=60) and control group (N=50) (without any type of cancer)was investigated. Inclusion criteria were diagnosed breast cancer patient (operative& inoperative cases). Exclusion criteria were control

group should not have family history of any type of cancer or other serious disease such as asthma, diabetes etc. Duration of study was 6 months. Genotyping for C677T was performed by PCR-RFLP method in case and control group. The genotype frequencies i.e. CC, CT and TT among the case (N=60) was 45%, 35% and 20% respectively while in control (N=50) CC, CT and TT frequencies was 57%, 43% and 0% respectively. Chi square test (χ^2) was used for difference in genotype distribution among case and control. p value <0.05 were considered statistically significant. The study supports that there is a significant association of genetic polymorphism C677T and breast cancer risk. Finding also revealed that CT genotype and T- allele of MTHFR C677T gene have increased genetic risk for breast cancer among the population of Bihar.

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Association of Serum Brain Derived Neurotrophic Factor (BDNF) levels with *BDNF* (rs6265) Polymorphism in Schizophrenia

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Schizophrenia is a severe neuropsychiatric disorder affecting 1% of the population. Disturbances in neuronal development and synaptic connections are important factors in the pathogenesis of schizophrenia. BDNF, a member of the neurotrophin family, plays a critical role in development of neurons. Among several polymorphisms reported in BDNF, the rs6265 polymorphism is known to be associated with many neuropsychiatric diseases. So, it was hypothesized that this SNP may have an association with schizophrenia. This study was aimed to determine the frequency of BDNF (rs6265) polymorphism and to check for plausible association of this polymorphism with serum BDNF levels in schizophrenia. In total, 50 schizophrenia patients and 50 controls were recruited after obtaining written informed consent. Cases were diagnosed as per the ICD-10 criteria and the severity was assessed using GAF and PANSS score. Serum BDNF levels were estimated using ELISA.BDNF polymorphism was genotyped using T-ARMS PCR. Mean ±SD of Serum BDNF levels of the cases and controls were 65.78 ± 21.67ng/dl (range 30-124 ng/dl) and 89.72 ± 24.43ng/dl (range 44-140 ng/dl). In controls, the distribution of genotypes was in the order of GG (70%), AG (24%), and AA (6%) whereas in cases the distribution was GG (58%), AG (36%),and AA (6%).The frequency distribution of SNP rs6265 was in Hardy Weinberg equilibrium. Allelic frequency of the A and G allele in controls was 82% and 18%, whereas in cases it was 76% and 24% respectively. Allele "A" could be a risk factor for schizophrenia as it is significantly higher in cases when compared to controls. Serum BDNF levels were significantly decreased in patients of schizophrenia and the BDNF levels of the genotype GG and AG were significantly lowered between cases and controls indicating that rs6265 polymorphism have an effect on the serum BDNF levels in schizophrenia.

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Differential Expression of SLC25A38 Gene in Patients of Acute Lymphoblastic Leukemia

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Acute lymphoblastic leukemia is a malignancy of hematopoietic stem cell origin and it is characterized by a group of molecular and cytogenetic abnormality. It is the most frequent malignancy of childhood and has peak prevalence for children 2-5 years of age and those older than 50 years .Five year survival rate for the patient who take proper treatment ranges between 76-86%. SLC25A38 is a mitochondrial solute carrier family protein. It is encoded by nuclear gene, which after synthesis in cytosol translocate into inner mitochondrial membrane. This protein is responsible for transporting glycine/ALA across the mitochondrial inner membrane, which is a rate-limiting step for heam synthesis. This step is essential for various biological processes such as detoxification, respiration and signal transduction. It has been shown that increased expression of SLC25A38 protein is over expressed in patients of acute lymphoblastic leukemia. Aim of my study is to investigate whether over expression of this gene could have a role in the diagnosis of ALL. So that it can be used as a biomarker for diagnosis of disease in early stage of disease. Study was conducted in the department of biochemistry, IGIMS, Patna. 30 newly diagnosed cases of acute lymphoblastic leukemia were taken as cases and 30 healthy volunteers of same age and sex group were taken as control. we screened SLC25A38 gene over expression by using RT PCR in cases and control. SLC25A38 positivity were found in 7/19 (36.8%) in children and 5/11 (45.45%) in adult patients. Increased expression were high in adult. Positive SLC gene expression was significantly higher in ALL group (P<0.001). Our result suggest that SLC25A38 gene was over expressed in patients of ALL. So it can be used as a useful target for diagnosing the disease in early stage

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Alliance of Single Nucleotide Polymorphisms (rs1501299 and rs2241766) of Adiponectin with Metabolic Syndrome

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Murine, and clinical studies imply that adiponectin plays an arbitrating role in obesity allied complications, and although less clearly established in humans, this suggests that molecular variants of adiponectin as a lucid candidate for unraveling the genetic basis of metabolic syndrome. To assess the pattern and alliance of metabolic syndrome with two single nucleotide polymorphism (SNP) variants, namely +276G>T (rs1501299) and + 45 T>G (rs2241766) of adiponectin in Gujarati population. Genotyping was performed by PCR-RFLP method for characterization of adiponectin gene (+276G>T and +45 T>G) polymorphism in a total of 113 cases with metabolic syndrome and 125 control participants. No departure from Hardy-Weinberg equilibrium was observed in either cases or controls for rs1501299 as well as for 2241766. An increased frequency of mutant allele of rs1501299 (p:0084) and of rs2241766 (p:0.0313) was observed among patients affected with metabolic syndrome, which resulted in significant difference in genotypic ($p<0.027$) and allelic ($p<0.005$) distribution between cases and controls. A significant association was found for G/G variant (p:0.0143) of rs2241766 and T/T variant (p:0.0054) of rs1501299, thus demonstrating the adiponectin as a pleiotropic locus for metabolic syndrome and its components. The logistic regression analysis adjusted by gender and age showed a ostensibly significant alliance for rs2241766 (OR:2.14; CI:1.36-2.62; p:0.04)and rs1501299 (OR:2.25; CI:1.42-2.77; p:0.02). Genetic variants of adiponectin have functional consequences and were selectively and specifically associated with the concomitant presence of metabolic syndrome and its components, suggesting impending disposition with metabolic syndrome.

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Association between Vitamin B12 Metabolism Related Indicators and Polymorphism of TCN-2 Gene in Postpartum Women with Postpartum Depression

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Vitamin B12 deficiency has been linked to the susceptibility of developing depression. Transcobalamin-2(TCN-2), is a glycoprotein essential for the intracellular transport of vitamin B12. A functional polymorphism in TCN-2 gene (rs1801198; G776C) influence the binding capacity of TCN-2 and may lead to intracellular deficiency of vitamin B12. We observed the association of this polymorphism with postpartum depression, and its effect on the circulating markers of vitamin B12 deficiency. Women were screened at 6 weeks postpartum for the probability of depression by EPDS score, and a score of >10 was taken as a cut-off (n=434; 217 in each group). Plasma was used for the estimation of homocysteine, methyl malonic acid (MMA), S-adenosyl methionine (SAM) and holotranscobalamin by commercially available ELISA kits and whole blood was used for extraction of DNA, which was further genotyped by real time PCR using Taqman probes. Of all the markers of vitamin B12 deficiency, total vitamin B12 was significantly lower ($p\text{-value}\leq 0.001$) and methylmalonic acid (MMA) was significantly higher in women with probable depression (p-value= 0.002). Plasma homocysteine levels although higher in depressed group, were not statistically significant (p-value=0.057). No difference was observed between the distribution of genotypes (p-value-0.619) and alleles (p-value-0.398) between women with and without probable depression. On comparing the effect of various genotypes on circulating levels of holotranscobalamin , homocysteine and MMA, we observed GG genotype to be associated with higher MMA levels in women with probable depression. In conclusion, Genotype GG of TCN-2(rs1801198) is associated with higher MMA levels in women with probable depression, indicating decreased availability and cellular deficiency of vitamin B12.

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Serum Folate and Vitamin B12 Levels in Acute MI Patients-a Case Control Study: Indian Population

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The incidence of coronary artery disease (CAD) is increasing at an alarming rate, especially in developing countries, such as India. It is often advocated that a vegetarian lifestyle could reduce the burden of CAD. However, in spite of a majority of Indians being vegetarians, the incidence of CAD is still very high in this population. In this study we estimated levels of vitamin B12 and Folate in patients of acute MI and compared them with controls. Individuals presenting to medicine emergency with clinical features suggestive of myocardial infarction, after detailed clinical examination, were screened with help of electrocardiographic and biochemical markers (increase in cardiac enzymes). 52 patients with evidence of fresh myocardial infarction, after satisfying the inclusion and exclusion criteria, were taken up for the study. 73 controls with no past or present history of myocardial infarction were also included in the study. Their serum samples (cases and controls) were analyzed for levels of Folate and vitamin B12 by ELISA. serum Folate levels in Cases (6.45 ng/ml) was significantly lower as compared to controls (14.78 ng/ml) with p value <0.001. Also, vitamin B12 levels were (0.54 ng/ml) was significantly lower in cases compared to controls (2.42 ng/ml) with p value <0. 001. So, we conclude that reduced levels of Folate and vitamin B12 can be considered as high risk factors for occurrence of MI, by virtue of their direct and indirect effects. However, more studies with larger samples are needed to confirm risk associated with reduced levels of these vitamins with MI.

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Vitamin D Status and Its Determinants Amongst Young Unmarried Educated Adult Females in North East India

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Widespread undiagnosed and untreated Vitamin D Deficiency is now becoming a serious concern in India. It has been observed that prevalence and severity of vitamin D deficiency is more in Indian females compared to males may be due to several socio economic factors. Data related vitamin D status specifically amongst young unmarried educated adult Indian females is not available. In our study, 198 young adult female Nursing students who were attending the health clinic of Gynaecology OPD of a tertiary care hospital in North East India were taken into the consideration. They were asked to respond to questions about vitamin D deficiency and related socio cultural practices. Amongst them, those 126 participants who have volunteered, levels of 25-OH Vitamin-D total were evaluated by Enzyme-linked Immunofluorescent Assay (ELFA) method along with other blood parameters in them irrespective of the symptom. In our study, amongst 198 study participants attending the health clinic 75.3 % had average <1 hour daily exposure to sun though 93.4% had the knowledge that sun exposure is the best source of Vitamin D. All the 08 participants who did not have the idea of sun exposure as source of Vitamin D were found to be deficient. It was found that out of studied 126 females 50% had vitamin D level <8.1 ng/ml (severely deficient) and 45.2% had Vitamin D level <20 ng/ml (deficient). 3.2% i.e. 04 participants showed Vitamin D insufficiency (Vitamin D level of 20-29 ng/ml). Vitamin D deficiency/ insufficiency is highly prevalent in studied population similar to other study population in other parts of India. Unmarried young adult females face considerable barriers related to socio-cultural and gender norms. Our study recommends specific intervention programs and efforts to prevent Vitamin D deficiency-linked health outcomes particularly in young adult females.

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Is Overweight and Obesity a Risk Factor for Iron Deficiency in an Urban Kenyan Population

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Overweight and obesity is a global epidemic and among the leading drivers of non-communicable diseases like heart disease, diabetes and cancer worldwide. Current trends predict a worsening profile due to the unhealthy lifestyles of the modern world.

Iron deficiency is the world's commonest nutritional deficiency and also a major cause of morbidity and mortality especially in sub-Saharan Africa. Chronic inflammatory states both infectious and non-infectious have been shown to cause a state of functional and/ or actual iron deficiency. We previously demonstrated that unlike normal weight individuals, those who were overweight or obese had signs of background inflammation including raised ferritin levels which is an acute phase reactant. We set out to determine if overweight and obesity was associated with a state of iron deficiency using transferrin saturation (Tsat) as a marker of tissue iron delivery in an urban population in Kenya. We used participant data collected as part of a global study by the International Federation of Clinical Chemistry to determine adult reference intervals for common laboratory tests. We recruited adults aged

18-65. Serum iron and Total iron binding capacity were determined using a Beckman Coulter DXI analyzer and Tsat was derived. BMI was determined by measuring weight (kg) and height (m), then derived. Mann-Whitney U test was used to compare Tsat and BMI categories i.e. <25, and ≥ 25 and also between males and females. We reviewed data from 528 participants, 254 males and 274 females. The median Tsat for males was 28 and females 20 percent respectively. Comparison indicated that Tsat was lower in females than males p≤.001 but similarity was found between Tsat against BMI categories in males (p= .317), and females (p= .085) We thus found no association between overweight and obesity and serum transferrin saturation as a marker of tissue iron delivery.

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Comparative Bioavailability of Synthetic and Dietary Vitamin B12 Present in Cow and Buffalo Milk: A Prospective Study in Lactovegetarian Indians

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We assessed improvements in the vitamin B12 status of Indian lactovegetarians receiving four weeks supplementation with natural B12 in milk versus cyano-B12 in capsules. Three groups (n = 22, 23, 22) received daily oral doses of cyano-B12 (2 × 0.76 μg) or milk (2 × 200 mL) from a cow or buffalo (amounting to B12 ≈ 2 × 0.76 μg). Their blood was examined at baseline and each following week. The baselines (median (min/max)) indicated a low B12 status: plasma B12 (116(51/314)) pmol/L, holotranscobalamin (holoTC) (30(7/119)) pmol/L, total homocysteine (Hcy) (24(10/118)) μmol/L, methylmalonic acid (MMA) (0.58(0.15/2.2)) μmol/L and combined B12 index (cB12) (-1.32 - (-3.12/+0.29)). Shifts from the baselines (B12, holoTC, cB12) and ratios to the baselines (Hcy, MMA) were analyzed over time. The cyano-B12 treatment gave more total B12 in plasma at week one (+29 pmol/L, p = 0.004) but showed no further increase. Other biomarkers changed more comparably between the three groups ($p \geq 0.05$): holoTC showed a transient spike that leveled off, Hcy finally decreased to 0.8 × baseline, while MMA showed marginal changes. The combined indexes improved comparably (p = 0.6) in all groups (+0.2(–0.3/+0.9), $p \leq 0.002$). In conclusion, the tested formulations similarly improved B12 status, but did not normalize it.

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Validation of Cobas 8000 E801 Module for The Quantitative Analysis of Total Vitamin D in Serum and Plasma

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The new Cobas 8000 e801 module has a high-volume capacity, big reagent pack size (Vitamin D total II) and wide analytical measurement range (AMR) compared to Cobas 8000 e602 module that uses the (Vitamin D total I) reagent. The Aim of this study was to compare total Vitamin-D values measured on Cobas 8000 e801 with those measured on Cobas 8000 e602. Method validation was performed using serum and plasma samples according to the Clinical and Laboratory Standards Institute (CLSI) guidelines. Between-days precision study was performed using 50 quality control samples of two different concentrations (low and high) for a period of 5 days. Mean, SD and CV% were calculated and compared to the manufacturer recommendation. Method comparison study was done using 25 serum and 20 plasma samples using Vitamin-D Roche Reagents on Cobas 8000 e602 and e801 modules with an acceptable criterion of slope between 0.9-1.1, correlation coefficient (r)>0.975, and % Bias < 17.5 % [Vitamin-D total allowable error (TEa)]. EP evaluator was used for data analysis. Linearity study was done using 8 different concentrations of serum samples that are spanning the AMR from 11.6-250 nmol/L. Sensitivity (limit of detection (LLOD)) test was performed using 10 universal diluent samples. The mean±2SD was calculated and compared to the manufacturer claim (7.5 nmol/L). The results between days precision for low and high concentrations CV%, were 2.7 and 3.2 respectively. The method was found linear over the AMR of 7.5-250 nmol/L. Observed LLOD was 8.3±1.6 nmol/L. Method comparison passed for serum, with slope=0.941, (r)=0.976 and %Bias=4.5%, and failed for plasma with slope=1.021, (r)=0.0.8784 and % Bias=25%. In conclusion, Overall performance of Cobas 8000 e801 module for vitamin-D measurement is acceptable and provides reliable results for serum samples, but not for plasma samples. Therefore, plasma should not be used for this test on e801 module.

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Association of Vitamin B12 and Vitamin D3 Deficiency with Severity of Depression: Cause or Consequence

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Depression is a disease with social importance and leads to everyday life infringements and disability. Depression is also one of the main causes of suicide and eating disorders. Vitamin B12 and Vitamin D3 are often associated with depression by various studies. Hence, in this study we aim to evaluate and compare the levels of serum vitamin B_{12} and D_3 in patients with severity of depression. We evaluated the serum vitamin B12 and vit D_3 levels in 74 patients with depression and an equal number of matched healthy volunteers. Serum vitamin B_{12} and vit D_3 levels were estimated by Chemiluminescence. The normal range of serum vitamin B_{12} is 200 to 900 pg/ml and D_3 is 20 to 50 ng/ml. The severity of depression was assessed by Hamilton depression score. In our study 52% of the depression patients had vitamin B_{12} levels below 200 pg/ml and 58% have vitamin D_3 level below 20 ng/ml. 18% of the patients had vitamin D_3 level lower than 10 ng/ml and 22% had vitamin B12 lower than 150 pg/ml. The vitamin deficiency was associated significantly with severity of depression. Our study reveals a correlation of vitamin deficiency with severity of depression. Since vitamin deficiency is a treatable state and can be avoided with balanced diet, we suggest evaluation of nutrition deficiency in all patients of depression which shall help further untoward consequences.

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Association of Trace Element with Bone Metabolism in Inflammatory Bowel Disease Patients

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Bone demineralization is a significant problem in Inflammatory Bowel Disease. Contributing factors including inadequate nutrition, corticosteroid, and decreased physical activity .Trace elements play an important role in the growth development and maintenance of bones .The aim of our study was to assess the relationship between the serum Zinc level and the bone Mineral indexes in Inflammatory Bowel Disease patients. Forty-two newly diagnosed patients of Inflammatory Bowel Disease and forty healthy Controls of both gender ranging in age from 19-50 years were included in the study. Fasting blood samples were processed for following biochemical parameters- Serum Calcium, Phosphorus, Vitamin D, Parathyroid Hormone and Zinc. The subjects were evaluated for Bone Mineral Density (g/cm^2) using Dual Energy X-ray Absorptiometry scan and T score was calculated to assess Osteoporosis. Student's unpaired t-test, one way ANOVA and Pearson correlation tests were used for statistical analysis . Inflammatory Bowel Disease patients had significantly lower Bone Mineral Density than the Controls. Bone Mineral Density values were not different between the subtypes Crohn's Disease and Ulcerative Colitis. Though Ulcerative Colitis and Crohn's Disease patients had significantly lower Bone Mineral Density than the Controls. Low Zinc level was observed in 50% of Osteopenic and 80% of Osteoporotic subjects. Zinc level was positively correlated with Bone Mineral Density($r=0.24$) and Vitamin D ($r=0.25$). In conclusion, Patients with Inflammatory Bowel Disease are more prone to develop metabolic bone disease. Along with other nutrients supplement Zinc should be added to prevent bone loss.

P-298

Assessment of Urinary Iodine Concentration Status Among the Hypothyroid Patients

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Nepal is considered as endemic zone of iodine deficiency. Hypothyroidism is a clinical state resulting from insufficient secretion of thyroid hormone from thyroid gland due to structural or/and functional impairments in thyroid hormone production. Iodine deficiency is one of the cause of hypothyroidism leads to insufficient production of thyroid hormones. This study aimed to measure the urinary iodine concentration among the hypothyroid patients attending immunoassay lab in BPKIHS, Dharan. A cross section study was conducted in immunoassay lab in BPKIHS, Dharan and diagnosed cases of subclinical and overt hypothyroidism were enrolled by purposive sampling technique. Ten milliliters causal urine samples were collected in a clean, tightly screw capped plastic container after informed consent from the patients and stored at 4°C until analysis. Urinary Iodine Concentration (UIC) was measured by ammonium persulphate digestion microplate (APDM) method. Data were expressed in median (IQR) and Mann-Whitney U test were applied to test the significance considering $p<0.05$ at 95% confidence interval. A total of 30 diagnosed cases of subclinical and overt hypothyroidism were

enrolled in this study out of them nine 9 male and 21were female. Seven and 23 patients were suffering from overt and subclinical hypothyroidism. Median (IQR) UIC of overt and subclinical hypothyroidism patients were 362.94 (271.40, 556.01) μg/L and 296.81 (117.01, 491.42) μg/L respectively and statistically not significant (p=0.311). Median UIC shows the optimal iodine nutrition in both overt and sub clinical hypothyroidism patients.

P-299

Comparison of Student based Teaching Method and Teacher Based Teaching Method Among Medical Students in Biochemistry Classes in Different Government Medical Colleges of Odisha

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Recent times have witnessed a vast change in the undergraduate medical curriculum and teaching methods. Various types of teaching methodology have been introduced. This study aims at evaluating undergraduate medical students regarding the better teaching modality: student-based teaching methods and teacher-based teaching methods in biochemistry class. We included 450 medical undergraduate students of 3 govt medical colleges of Odisha. The Vitamins chapter of the Biochemistry syllabus was included. 50% of the chapter is presented in didactic lecture in teacher based teaching method and remaining 50% of the chapter was presented as Problem Based Learning (PBL) by student-based teaching method. The feedback from the students was analyzed by statistical software SPSS version 24. A more positive response was observed in the feedback form about student-based teaching by PBL as compared to the didactic lecture in teacher-based learning but the teacher-based learning provided newer and additional information. On assessment the mean score of medical undergraduate students in teacher-based learning was 14.2±5.6 and in PBL, it was 24.8±4.8 (significantly higher; $p<0.05$). The problem-based learning in student-based teaching method is more acceptable and satisfying for the students, in comparison to teacher-based learning. The students appreciated the additional information in teacher-based method. Therefore, a balance of both methods has to be implemented in the curriculum.

P-300

Vitamins A and E in Pregnancy: Establishment of Trimester-Specific Reference Intervals and Associations with Complete Blood Count

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Profound physiological changes during pregnancy may affect the requirement of vitamin A and vitamin E which are essential micronutrients to maternal health and fetal development. However, the current reference intervals (RIs) of vitamins A and E are based on non-pregnant population. In this study, we established trimester-specific RIs of vitamins A and E with 31301 paired screening results of outpatients participating the prenatal vitamins A/E evaluation program at our hospital using the Hoffmann method, which is a simple indirect RI estimation-establishing method that does not require recruiting healthy subjects. Meanwhile, a serum vitamins A and E quantitation method by LC-MS/MS was established and validated. To explore the associations between vitamin A, E levels and complete blood count (CBC) metrics, the results of a total of 1977 pregnant outpatients who were tested within 7 days for vitamins A, E and CBC in the third trimester were analyzed. Although no essential changes were noticed for vitamin A level throughout the entire pregnancy, vitamin E level was significantly elevated as the increase of the gestational age. The association between vitamin A status and anemia was confirmed. Furthermore, increased vitamin A and decreased vitamin E were associated with higher platelet counts.

P-301

Protein Energy Wasting in Chronic Kidney Disease Patients and Effect of Dietary Counselling

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Protein Energy Wasting (PEW) is a state of decreased body stores of protein energy in the most advanced stages of chronic kidney disease. The main objective of this study is to assess the PEW in chronic kidney disease patient (CKD) and to reduce the mortality rate and to improve the patient quality of life by providing dietary counselling through information leaflets. It is a hospital based

concurrent interventional study, was conducted for a period of six months on 100 patients having PEW in chronic renal failure condition. Inclusion criteria was diagnosed CKD patients coming to nephrology OPD. Patients of age groups between 18-60years of age. Exclusion criteria were patients with any inflammatory illness, AIDS, Active hepatitis B and C, malignancy, liver cirrhosis. Patients of acute renal failure were excluded from the study. Patient demographics and laboratory data like Albumin, Cholesterol, Muscle mass was periodically collected and reviewed. Patient information leaflets, diet chart was provided along with counselling at baseline and during follow-ups. These study results were analyzed to find the significance of study parameters at baseline and follow-ups. At 1st visit albumin level (<3.8 gm/dl) was found in 69% patient that decreases to 52% after 1st follow up (nutritional counseling) Similarly albumin level (>3.8 gm/dl) was 31% and 48%, Cholesterol (<100 gm/dl) in 63% and 70% and >100 gm/dl in 37% and 30%. Weight loss (>10%>6 months) found in 43% and 56% Weight loss (<10%>6 months) found in 57% and 44%. Body fat % loss (>10%>6 months) was found in 7% and 0% Body fat % loss (<10%>6 months) was 93% and 100%. Dietary intake of calories (<25 kcal/kg/day) & (protein <0.6 gm/kg/d) found in 72% and 82 % respectively. After 6 months calories (<25 kcal/kg/d) & protein (<0.6 gm/kg/d) found in 28% and 18% respectively. In conclusion, early detection and conducting effective dietary counselling was found to be more effective and beneficial for CKD patients in reducing the risk of PEW.

P-302

Evaluation of Lead, Aluminum and Zinc in Occupationally Heavy Metal Exposed Population

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Occupational exposure to heavy metals causes a wide range of biological effects, depending upon the metal level and duration of exposure. Lead is widely used in industries and its toxicity is a major public health problem. It causes a number of adverse effects on different body systems. Aluminum is widely used in industries, utensils, packing and mainly accompanied by the emission of dust and gases at the occupational settings which is very harmful. Zinc is an essential trace mineral required for growth, development, DNA synthesis, immunity, and many other critical biological processes. The aim of this study was to assess the levels of lead, aluminum and Zinc in occupationally exposed population. We studied a population of 88 male workers exposed to heavy metal in handicraft and welding industries in Jodhpur, Rajasthan. Blood lead and aluminum levels were estimated by Graphite furnace atomic absorption spectrophotometer and Zinc levels were estimated by flame atomic absorption spectrometry (ICE 3000, Thermo Fisher Scientific). The mean ± SD of Lead, aluminum and zinc was 5.43 ± 7.47, 46.87±37.76, 35.24±19.46 in handicraft workers and 10.03±18.70, 57.43±2.40, 36.68±19.06 was in welding workers. 29% of workers were having high lead level (>5 μg/dL) in handicraft group whereas 50% had high lead level (>5 μg/dL) in welding workers. Zinc levels were found deficient in both the handicraft and welding groups. Occupational exposure of heavy metal causes adverse health effects. Welding workers are more prone to have high blood lead levels compared to handicraft workers.

P-303

Impact of Lead Exposure on Neurobehavioral Development in School Going Children

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Lead is a versatile heavy metal and a common environmental toxicant known to affect multiorgan systems. Early exposure to lead is known to cause adverse health effects including intellectual and behavioral deficits in children, deficits in motor function and hand eye coordination, hyperactivity, and lower performance on intelligence tests. Childhood Psychopathology Measurement Schedule (CPMS) is an Indian adaptation of Achenback's child behavior check list (CBCL) and has been found to be valid and reliable tool for screening psychiatric morbidity among Indian children. The aim of this study was to assess the impact of lead on neurobehavioral changes in children by means of CPMS score. The study was conducted in school going children under the age of 15 years (N=68). BLL were assessed by Atomic Absorption Spectrophotometry. Neurobehavioral assessment was carried out in all children through screening for psychiatric symptoms by interviewing the parents / teacher or the key informant of the children using Hindi version of CPMS. The mean ±SD of BLLs were 6.00±5.47μg/dl. Statistical analysis of BLL and CPMS score showed significant positive correlation ($r=0.4042$, $p<0.001$). They were categorized into two groups, BLL < 5 μg/dl (N=44) and BLL > 5 μg/dl (N=24) respectively. The difference in CPMS scores of the two groups were found to be statistically significant ($p<0.01$). Children with high BLL had high CPMS scores indicating possible neurobehavioral abnormality. Based on our study findings we could predict that lead has an impact on CPMS scores and thus on neurobehavioral development in children. The underlying mechanisms are unclear and requires further studies.

P-304

Levels of Essential Trace Metals in Occupationally Lead Exposed Individuals: A Pilot Study

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Lead (Pb), a toxic heavy metal extensively used in industries is capable of inducing several adverse health effects in the workers exposed to this metal. Lead interacts with some of the essential metals like selenium, which plays a major role in overcoming oxidative stress induced by lead toxicity. We aimed at estimating levels of selenium in occupationally heavy metal exposed individuals. We have included 40 heavy metal exposed individuals for the study after obtaining informed consent. Venous blood samples were collected taking due aseptic precautions. Blood Pb levels (BLL) and Serum Se levels were analyzed using Dual Atomic Absorption Spectrophotometer (ICE 3500 Thermofischer) .Commercial reference materials were obtained from Bio-Rad (Lyphochek ® Whole Blood & Serum Metals Control) for the internal quality assurance and control program. The Mean ± SD of BLL and selenium were 6.93 ± 6.04 ug/dL and 97.2 ± 28.04 ng/ml respectively. Selenium had a significant negative correlation with BLL ($r = -0.342$, $p < 0.05$), Further the levels of selenium were compared in workers with BLL < 5 ug/dL & BLL > 5 ug/dL were also found to be statistically significant with p value 0.0039. The selenium levels are decreased in workers with high BLL, so the individuals who are exposed to heavy metals may require selenium supplementation to fight against lead induced oxidative stress.

P-305

Effect of Artificial Sweeteners on Glucose Uptake in 3T3 Adipocytes

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Escalating rates of obesity and public health messages to reduce excessive sugar intake have fuelled the consumption of artificial sweeteners (AS) in a wide range of products from breakfast cereals to snacks and beverages. Several studies indicate an association of AS consumption with increased appetite, food intake, weight gain and glucose intolerance. Multiple studies have studied the effect of AS on glucose absorption in the gut, there are very few probing the same in adipocytes. Therefore, we want to study the effect of different AS on glucose uptake in adipocytes. The aim is to study effect of saccharin, sucralose and cyclamate on glucose uptake in 3T3 cell lines. 3T3 cells (mouse origin) from ATCC (American Type Culture Collection) were grown in Dulbecco's Modified Eagle's Medium (DMEM). After differentiation, they were incubated overnight with the AS (0, 1 nmol, 1 mmol and 1 mmol of the 3 AS) and glucose uptake assay was performed the next day for basal and insulin stimulated uptake. All experiments were performed in triplicates. 2way Anova was done. Almost all of them showed significant increase in uptake in stimulated condition compared to basal. Cyclamate and saccharin showed an increase in uptake but sucralose shows a significant decrease in uptake with increase in concentration. AS are usually consumed by people with insulin resistance. All AS are not having similar effect on glucose uptake by cells. Sucralose is decreasing the glucose uptake by cells and may not be the AS of choice in people who already have decreased glucose uptake. Whereas, Saccharin and cyclamate are increasing glucose uptake and could be beneficial in individuals with insulin resistance. AS are not metabolically inert and it will be necessary to choose the right AS based on the metabolic status of the individual.

P-306

Vitamin D Deficiency Among Patients Attending BPKIHS, A Tertiary Care Centre of Eastern Nepal

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Vitamin D is an essential fat-soluble vitamin for calcium homeostasis, bone health. It has also been related to hypertension, diabetes, metabolic syndrome, cancer, autoimmune and chronic diseases. These conditions are major public health problems worldwide. The deficiency of vitamin D is widespread in individuals, irrespective of age, gender, race and geography. Since many countries have a relatively low supply of foods rich in vitamin D and inadequate exposure to natural ultraviolet B (UVB) radiation from sunlight and therefore a significant proportion of the global population is at risk of vitamin D deficiency. The prevalence of vitamin D deficiency and insufficiency was observed 74.1% in Nepal by S Regmi et al in 2017. There is limited information of the vitamin D deficiency in people of eastern Nepal. Therefore, we assessed serum 25(OH) Hydroxyvitamin D3 [25(OH) D)] level among patient who has attended at BPKIHS for assessment of vitamin D level between February to March 2019 after ethical clearance from IRC. Serum 25 (OH) D was measured in 3250 patients sample by CLIA in Maglumi 2000 immunoassay analyzer. Out of total participants, 2399 male and 851 female were enrolled in the study among which 66, 133 and 3250 were children, adolescent and adult respectively. The study revealed, 61.2%, 27.6% and 11.2% patients having deficient, insufficient and sufficient level

of serum vitamin D respectively. Serum vitamin D level in children, adolescent and adult were found 21.51 (15.87, 26.83) ng/dL, 16.06 (13.14, 20.48) ng/dL and 17.71 (13.66, 23.70) ng/dL respectively which is statistically significant. The study showed a higher prevalence of vitamin D deficiency in all age group and adolescents were observed relatively low serum vitamin D level as compared to adult and children.

P-307

A Hospital Based Evaluation of Micronutrient Status in Central India

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Micronutrient deficiency is a major health issue in the developing countries including India. The dependence of all the major functions of the human body on these elements make them indispensable and unavoidable for healthy living at all stages of life. Deficiency of the nutrients cause different pathological conditions, manifestations ranging from minor to very severe. However, overuse of these micronutrients, in response to better screening, increased awareness and over the counter availability, is now leading to toxicity and unnecessary monetary burden on the patients. Hence, the study aimed to evaluate the current status of serum vitamin B12 in patients of a tertiary care centre of Chhattisgarh, along with other nutritional parameters. A retrospective data analysis in patients of both genders and various age groups found the prevalence of vitamin B12 deficiency to be 11.5% while a higher percentage of the patients, 20.1%, showed hypervitaminosis B12. Male patients showed almost double the prevalence of deficiency than that in females (14.2% in males and 8.7% in females). The age group, 21-30 years had the maximum patients with deficiency (13.9%) followed closely by the 41-50 years group with 13.09% prevalence. Another 20.9% patients showed borderline values suggesting further evaluation and follow up. Vitamin D deficiency was observed in a massive 55.5% of patients while 13.1% showed values of hypervitaminosis D. A high prevalence of vitamin deficiency and a higher prevalence of hypervitaminosis not only necessitates more efficient screening and correction measures but also a rational and monitored dosing of drug in clinics and community.

P-308

Can Hypomagnesemia be the Reason for Increased Risk of Bone Fractures? : A Cross-Sectional Study

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Magnesium is an essential trace element that plays a key role in several cellular processes. It is a major component of bone, which is 67% of total body magnesium. As the hydrogen/potassium-ATPase pump in the cells of periosteum and endosteum are magnesium dependent, the pH of the bone extracellular fluid may fall in magnesium deficiency, resulting in demineralisation of bone. However, its relationship with risk of major bone fractures is uncertain. Here we aimed to find out the association of baseline serum magnesium level with risk of fractures. A cross-sectional study was carried out on 140 male participants having age between 40 to 60 years. 70 patients having long bone fracture were considered in case group while 70 age and sex matched healthy control were taken as control group. Serum magnesium estimation was carried out by Calmagite method for all samples. Results were noted in mean±SD and p value was calculated for the case and control group for finding the proper significant difference. The normal serum magnesium level is 1.6-2.6 mg/dl. The observed mean concentrations of case group was 1.56±0.21 mg/dl and 1.86±0.19 mg/dl for control group. P value was found statistically significant ($P<0.001$). It indicates that low serum magnesium levels are associated with the increased risk of bone fractures. Further study is required by optimizing magnesium levels by diet or medication and observing its effect on incident of bone fractures.

P-309

Vitamin D and Vitamin B12 Status in Post-Menopausal Nepalese Women: An Unseen Truth

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Menopause is a universal phenomenon in the women above the age of 50-55 years, accounting for one third of the rest of their life. The end of reproductive life possesses various complications along with numerous nutritional deficiencies including low Vitamin D. The aim of the study was to assess the occurrence of vitamins D and B12 deficiency in Nepalese post-menopausal women (PMW) along with some specific biochemical parameters including Calcium, Phosphorus, Hemoglobin, Iron and lipid profile and to establish the reference interval of vitamin D

and vitamin B12 level in PMW. This was a community based descriptive cross-sectional study conducted in 150 apparently healthy PMW attending BPKIHS using a structured proforma after ethical approval from IRC. Biochemical parameters were analyzed in Roche cobas c311; vitamin D and B12 were analyzed in Maglumi 1000 by CLIA. A p value < 0.05 was considered to be statistically significant. The mean age of the study population was 53.01± 5.32. BMI and waist circumference were higher in postmenopausal women. High prevalence of vitamin D and vitamin B12 deficiency was found (81.33% and 19.33% respectively) among the studied subjects. The median of vitamin D and B12 were 20 (5, 27.25) ng/ml and 472 (250, 582.05) pg/ml respectively. In addition, majority of the study population had the following status of biochemical parameters: hypocalcaemia (57.4%), hyperphosphotemia (78%), sufficient iron level (93.3%), normal hemoglobin (70.7%), hypercholesterolemia (64.7%), hypertriglyceridemia (70.7%), normal level of HDL-C (56%) and LDL-C (63.3%). Considering the participants to be apparently healthy were healthy, the reference intervals estimated for vitamin D and vitamin B12 for the post-menopausal women were 10-42 ng/mL and 108-1072 pg/mL respectively. Low vitamin D and B12 concentration was found among the postmenopausal Nepalese women.

P-310

A Study to Investigate the Correlation Between Selected Micronutrient (Iron, Copper and Zinc) Status in the Maternal Blood and the Cord Blood of the Newborn in a Tertiary Care Hospital in Eastern India-A Brief Report

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During pregnancy, the metabolic demand of the body increases considerably because of increased cell growth and differentiation. This increase in demand includes micronutrients also, namely vitamins and minerals which are deficient in supply even during the non-pregnant state. The commonest cause of anemia during pregnancy is due to iron deficiency not always corrected by iron supplementation or changes in diet. This may be due to an underlying deficiency of zinc which is also required in iron metabolism. Copper acts as a cofactor for the numerous cupro-enzymes, and also plays an important role in iron metabolism. The objective of the present cross sectional and observational study was to assess the serum copper, zinc, ferritin and iron status in the maternal and cord blood of the newborn by validated kits using two levels of control and to investigate any underlying correlation between them. The present study is being conducted in the Department of Gynecology and Obstetrics and the Department of Biochemistry, College of Medicine & Sagore Dutta Hospital. The target sample size is 336 and so far 86 pregnant women and their newborns have been covered. So far, the mean concentration for copper was 132.91±47.9 μg/dl, zinc was 46.19±18.69μg/dl, iron was 144.37±66.44 μg/dl, and ferritin was 70.22±59.50 μg/dl in the maternal serum. In the cord blood, the figures were51.5±16.85 μg/dl,64.52±24.17 μg/dl, 147.26±65.64 μg/dl, and 199.31±94.24 μg/dl respectively. The correlation between maternal and fetal concentrations of copper was found to be y = -0.11073X+64.8873, for zinc y=0.23631x+47.31368, for iron y=0.31721x+149.98268 and ferritin y=-0.161154x+159.09583.

P-311

Association of Vitamin B12 with Coronary Artery Disease in Indian Population

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Coronary Artery Disease (CAD) is the most common type of cardiac disease and the leading cause of all deaths globally. The prevalence of CAD and its mortality is increasing rapidly over the past two decades in India. There is a significant association between hyperhomocysteinemia and cardiovascular disease, its complications like heart attacks and strokes. Hyperhomocysteinemia damages the endothelium and vascular smooth muscle cells of arteries. Vitamin B12 deficiency causes hyperhomocysteinemia as it is required as coenzyme by methionine synthase for the conversion of homocysteine to methionine. In this cross-sectional study, we tried to find out the association between vitamin B12 and CAD by estimating its levels in the peripheral blood of 30 CAD patients and 30 normal healthy controls in the Department of Biochemistry, VMMC & Safdarjung Hospital, New Delhi. The mean age of CAD patients was 57±11 years. After collection of peripheral blood from CAD patients and controls, serum was separated by centrifugation at 3000 rpm for 10 minutes. Serum vitamin B12 levels were then measured by ELISA. Statistical analysis was done to compare the vitamin B12 levels in cases and controls by using T-test. The mean of serum vitamin B12 levels in cases and controls were 328 pg/ml(range 228-440 pg/ml) and 370 pg/ml (range 215-562 pg/ml) respectively. Our results show that CAD patients have significantly lower vitamin B12 levels than controls in this study group (p = 0.04). This study shows a positive disease association with low vitamin B12 levels and CAD. We also consider, vitamin B12 supplementation as a treatment option for CAD would offer some protection in our study group. Also, vitamin

B12 supplementation can be tried to prevent CAD for high risk cases. Decreased vitamin B12 levels lead to elevated homocysteine, which has been implicated as an independent risk factor for CAD.

P-312

Assessment of Serum Zinc Status in Carcinoma of Lung and Esophagus Patients in North West Rajasthan

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Some minerals & Trace-heavy elements play a significant role in human health and disease. Trace elements at optimum levels are required for numerous metabolic and physiological processes in the human body. The association of serum trace element like Zinc has been found in different types of cancer. This study was conducted to see the serum level of trace element (Zn) in carcinoma of lung and esophagus patients. Study group consisted of 50 clinically diagnosed subjects (Biopsy confirmed 25 cases with Lung carcinoma and 25 cases with Esophageal carcinoma). The control group consisted of 50 Healthy subjects were included in the study. Both study and control group patients were of same socio-economic status and dietary habits. Venous blood samples of each lung and esophagus cancer were obtained and serum Zn level was analyzed by Atomic Absorption Spectrophotometer (AAS) measurements. The serum Zn level was significantly lower in serum of lung and esophagus cancer group than controls (P<0.0001). The mean serum zinc level was found to be decreased significantly in lung and esophageal cancer patients as compared to that control group. To conclude, Serum trace element like zinc, might be play a role in the patients of cancers. Zn may protective as potent lung cancer. In addition, it is suggested that low levels of zinc can induce the pathogenesis of cancer.

P-313

Reference Value for Serum Zinc Level of Adult Population in Bangladesh

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Zinc is an essential trace element that has an enormous role in regulation of physiological processes whose deviant value leads to malfunction in the body. So, establishing a country specific reference value is needed to serve as a standard for the interpretation of laboratory results during clinical decision making. The objective of this study was to determine the reference value of serum zinc level of adult population in Bangladesh. The overnight fasting blood was collected from 154 apparently healthy individuals aged 18 to 65 years, from a rural community after considering several criteria. Graphite furnace atomic absorption spectrophotometry (GFAAS) method was used for serum zinc analysis. The 2.5th and 97.5th percentiles of zinc value were calculated for the reference value according to the recommendations of the International Federation of Clinical Chemistry. The estimated reference range of serum zinc level in sample population was 60-120 μg/dl, where the range was 59-125 μg/dl for male and 50-103 μg/dl for female. Significant differences of serum zinc level between male and female ($p < 0.001$) was observed. However, there was no significant correlation between age of the respondents and serum zinc level ($r=0.110$, $p>0.05$). The estimated reference range for serum zinc level in adult population of Bangladesh can serve as a useful indicator for clinical decision making.

P-314

A Study on Vitamin D and Vitamin B12 Levels in Patients of Tertiary Care Hospital- An Observational Study

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Vitamin B12 and 25-hydroxy Vitamin D [25(OH) Vit D] deficiency is a serious public health problem, particularly in the Indian sub-continent. The objective of the present study was to study the prevalence of 25(OH) Vit D and Vitamin B12 in different age groups in our hospital. Batra Hospital & Medical Research Centre is tertiary care 450 bedded multispecialty hospital catering to different population groups. 25(OH) Vit D and Vitamin B12 levels of 587 and 402 ostensibly healthy individuals visiting Batra Hospital & Medical Research Centre, Delhi, was studied over a period of 6 months respectively. Analysis was performed on fully automated Abbot Architect analyzer in Clinical Biochemistry section, department of Lab Medicine. 25(OH) Vit D deficiency was defined as 25(OH) Vit D < 30 ng/ml, insufficiency as 25(OH) Vit D between 30 and 40 ng/ml and 25(OH) Vit D sufficiency as 25(OH) Vit D 40 ng/mL and the vitamin B12 deficiency < 187 pg/mL, sufficiency as Vitamin B12 187-883 pg/ml. 25 (OH) Vit D deficiency in 78% and Vitamin B12 deficiency in 33% of the subject population was observed. Maximum number of the subjects belonged to the age group of 40-60 years. 43% had frank 25 (OH) Vit D deficiencies when cut off level was 20 ng/ml. 25 (OH) Vit D levels were found significantly low in male subjects when compared with females. Our study demonstrates a high prevalence of 25(OH) Vit D deficiency and Vitamin B12 deficiency in random healthy population.

P-315

Serum Trace Elements and their Association with Type 2 Diabetes Mellitus

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Insulin action on reducing blood glucose can be enhanced by some trace elements such as chromium, magnesium, zinc, manganese, and selenium. It is also reported that the metabolism of several trace elements alters type 2 diabetes mellitus (T2DM) and these elements might have specific roles in the pathogenesis and progress of diabetes. However, it is unclear whether DM and hyperglycemia affect trace element status or DM sets in due to the deficiency of trace elements. The aim of the present study was to evaluate in changes in trace elements such as copper, zinc, magnesium, selenium and chromium in type 2 diabetes mellitus patients (T2DM) and their correlation with metabolic parameters such as duration of diabetes, BMI, HbA1C, triglycerides, HDL and LDL. One hundred subjects with T2DM and equal number of age and sex matched controls were also included for the study. Trace elements were studied using inductively coupled plasma mass spectrometry (ICP-MS). Correlation analyses of trace elements with metabolic parameters were analyzed using Pearson correlation coefficient. Significantly higher levels of FBS, HbA1C, BMI and triglycerides whereas significantly lower levels of serum Zn, Se, Cr, and Fe were found in T2DM as compared to controls ($p<0.05$). A positive correlation was noticed between Cu and duration of the diabetes ($p<0.05$). Similarly Zn also showed a positive correlation with HbA1C (<0.05) whereas negative correlation was found between Se and FBS, HDL cholesterol (<0.05). In addition, highly significant negative correlation was observed between serum Mg and HbA1C ($p<0.05$). Significantly lower levels of Zn, Mg, Cr, Se, Fe and higher levels of Cu in the present study signifies their association with T2DM. Lower Mg and Zn levels have been linked to poor glycemic control in T2DM; therefore, screening for Mg and Zn deficiency should be corrected in T2DM patients along with oral antihyperglycemic agents.

P-316

Role of Vitamin B12 and Folic Acid Supplementation on Serum Homocysteine Levels in Deep Vein Thrombosis

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Deep vein thrombosis is a common vascular disorder and association of homocysteine with DVT is known. Evidence of decrease B12 and folic acid levels in DVT with Hyperhomocysteinemia has also been reported. Scanty data is available regarding role of folate and vitamin B12 on Homocysteine levels in DVT patients. All the patients of study group were given vitamin B12 and folic acid treatment along with standard treatment of DVT. Serum Homocysteine, folate, vitamin B12 were measured in all the patients in both the groups and estimated by chemiluminescence technology. After 12 weeks of vitamin B12 and folic acid therapy, serum Homocysteine levels, folic acid levels and vitamin B12 levels of study group were again estimated. The present study was undertaken in 100 patients. Study group comprised of 50 patients who had clinical features suggestive of DVT and 50 patients, who did not have clinical features suggestive of any venous or arterial disorder, were labelled as control group. The collected records and data was analysed statistically by Student t-test and Chi-square test. Mean Homocysteine level in study group was significantly higher as compared to control group. Mean Homocysteine levels before treatment and after treatment were

significantly reduced. Hyperhomocysteinemia is a risk factor for deep vein thrombosis. Folic acid and B12 therapy reduced the level of Homocysteine suggesting that this may decrease the chances of recurrence of DVT.

P-317

Study of Trace Elements in Liver Cirrhosis Patients and their Role in Prognosis of Disease

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The objectives of this study are to evaluate trace elements in patients with liver cirrhosis and to assess their association with severity of the disease. One hundred fifty cirrhotic subjects of either sex ranging in age from 20-70 years were included in the study, and the results were compared with 50 age- and sex-matched healthy control subjects. All cirrhotic subjects were assessed for severity of disease as mild (Child A), moderate (Child B), and severe (Child C) as per Child-Pugh classification. Routine investigations were done and trace elements (Cu, Zn, Se, and Mg) were analyzed on atomic absorption spectrophotometer. Serum level of copper was found significantly increased in patients with liver cirrhosis as compared to control group. Whereas serum zinc, selenium, and magnesium levels were significantly decreased in cirrhotic subjects as compared to controls. Trace elements were com-pared with severity of liver cirrhosis. Serum copper concentration was slightly increased in patients with more severe clinical state of liver cirrhosis; however, mean level difference of copper among the Child-Pugh groups were statistically not significant. Moreover, there was no significant correlation between copper and Child-Pugh Score. However, copper showed a significant negative correlation with zinc. Serum zinc, magnesium, and selenium levels were significantly decreased with advancement of liver disease as compared to early stage of liver cirrhosis and showed a significant negative correlation with Child-Pugh Score. Trace element abnormalities may reflect the condition of liver dysfunction. These results suggest that liver dysfunction may alter the metabolism of trace elements. Our study shows that micronutrients status in liver cirrhosis correlates well with severity of liver cirrhosis. Micronutrients supplementation in liver cirrhotic patients may prevent progression of disease and development of complications; however, further research needs to be done.

P-318

Nutritional Status of Children's of Western Odisha Tribal Population

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Many anemia patients are coming to the hospital belongs to the western Odisha tribal population. Children's below 10 years are screened for hemoglobin status. Those having Hb less than normal level are subjected to a questionnaire. Blood samples were tested for sugar lipid calcium magnesium iron G6PD vitamin B12 and sickling. Same age and sex matched controls were also included in this study with informed consent from parents. It was found that 51 % children's having sickling positive, 30 % have iron deficiencies, 13% having B12 deficiencies and rest due to chronic illness.

P-319

Prospective Case-control Study of Clinical Profile and Serum Electrolytes in Children with Febrile Convulsion

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Febrile convulsions, is one of the most common disorder seen during childhood which is benign in nature, generally having an excellent prognosis. During an acute febrile disease, mild disturbance of water and electrolyte balance occurs frequently. It has been suggested that changes in serum electrolyte levels, might enhance the susceptibility to seizure and its recurrence during a febrile disease in childhood. The purpose of this study was to assess the prevalence of electrolytes disturbance in children with simple and recurrent febrile convulsion. This study was performed at the Department of Pediatrics and biochemistry. Patients diagnosed as febrile seizures as per the criteria. Venous blood samples were obtained at the time of initial evaluation and analysis of serum sodium, potassium, calcium and magnesium done by electrolyte analyzer machine at Department of Clinical Biochemistry. In this study the majority of cases were seen in the age group of 13-24 months. Febrile convulsions were noticed in 47 males (55.29%) and in 38 females (44.70%). Hyponatremia is associated in 19 cases of febrile convulsion (22.35%) with P value 0.0001. It indicates hyponatremia was associated in the majority of febrile convulsion patient. Incidence of febrile seizures is more in children before 24 months of age and among the male children. Mainly hyponatremia was associated in the majority of febrile convulsion patient. There

is no association between levels of serum potassium, calcium & magnesium with febrile convulsion. So the laboratory investigations should be directed towards identification of the source of fever after a detailed history and clinical examination.

P-320

Assessment of Serum Trace Element (Copper) Level, as Associated Risk Factor in Patients with Chronic Kidney Disease

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Chronic Kidney Disease (CKD) is a progressive loss of kidney function and is a worldwide public health problem both for the number of patients and for the cost of treatment. Trace element such as copper (Cu) is altered in CKD. We assessed 100 subjects of both sex with different age groups, among them 50 are normal healthy controls (group 1) and 50 are CKD patients (group 2) from dialysis ward (medicine) S.R.G. Hospital, Jhalawar Medical College, Jhalawar (Raj.) Serum copper was estimated using Flame Atomic Absorption Spectrophotometer (AAS). Serum copper level was significantly low in CKD patients, mean ± SD, (1.2041 ± 0.46360) ($p<0.05$) when compared with healthy control group (1.3823 ± 0.25250) ($p<0.05$). Gender had no significant effect on serum copper level, in males, (1.2483 ± 0.36215) and in females (1.3578 ± 0.40478) ($p>0.05$). There was no significant correlation between age and overall distribution of serum copper level, (1.2932 ± 0.38206) and age (41.0700 ± 15.48362) ($p>0.05$). Conclusion of the study was that in CKD patients, trace element derangement is important in primary diagnosis of trace element dysfunction and medical management of CKD.

P-321

Improvement in Cardiovascular Status by Addition of Small Amounts of Raw Vegetables and Fruits in the Diet

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Cardiovascular diseases (CVD) are on the rise. In India the rates of coronary disease have increased from 4% to 11% among urban populations. CVD affects not only the older but also the younger population and hence is responsible for reducing the productivity. Changing food habits due to modernization may be responsible for low intake of fruits and vegetables in daily diet. This factor can contribute to increase of CVD. The aim of our study is to note whether a small and sustainable increase in fruits, sprouts and raw vegetables in the daily diet can help to lower weight, blood pressure, improve exercise capacity, cholesterol, triglycerides and blood sugar. 30 Teaching and Non- teaching faculty of our college were included in the study. The age group was between 25 and 60 yrs of age with no metabolic disorders. After 10 minutes of rest the blood pressure was recorded. Their Height and weight were measured and BMI was calculated. The subjects underwent six minutes' walk test in the hallway and the distance in meters covered was noted. Fatigue and dyspnoea according to BORG's scale was assessed. Biochemical parameters were measured which included Lipid profile and Blood sugar. All the above parameters were again measured after the dietary intervention which included a slight modification in the diet by addition of minimal amounts of raw vegetables fruits and sprouts for a period of 90 days. A significant decrease in BMI, Systolic and Diastolic pressure and an increase in the distance covered in six minutes. A decrease in Serum Cholesterol, S.LDL and S Triglycerides was observed.

P-322

Emerging Biomarkers and Biosensors In Point- of-Care Diagnostic Devices For Cancer Detection

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Screening and early diagnosis of cancer increases the likelihood of survival, lowers morbidity and cost of treatment. Building diagnostic capacity by means of ultrasensitive point of care (POC) diagnostic devices integrated with emerging biomarkers and biosensors can overcome common barriers to timely diagnosis. Here attempt has been taken to document key developments in POC diagnostic devices with notion to bring the test conveniently and immediately to the cancer patients even in limited health infrastructure. Nanomaterial, microfluidics and cell phone based POC diagnostic devices for cancers are being developed that fulfills the ASSURED criteria of being affordable, sensitive, specific, user friendly, rapid & robust, equipment free and delivered to end users with ease. Exosomes being present in all biological fluid contains protein, lipids, different RNAs and their molecular and surface constituents have been found to be associated with progression and metastasis of various cancers. Similarly tumor antigens which can be a mutated oncogenes, aberrantly expressed cellular proteins, antigens produced by oncogenic viruses, oncofetal antigens and altered cell surface glycoprotein and glycolipid all can evoke an immune response and creates cancer specific autoantibodies. These autoantibodies are highly stable in serum and signal the presence of cancer at an early stage prior to appearance of symptoms and also the appearance of tumor associated antigens in serum. So these

can be excellent biomarker candidates for POC devices. Early and accurate diagnosis of cancer plays a decisive role for its effective treatment. Diagnosis based on single biomarker is not adequate. Keeping this in mind advanced POC diagnostic devices with multiplex analysis on a single platform are in demand as they require low sample volume, can be used in resource poor setting and have high sensitivity and specificity.

P-323

Evaluation of the i-STAT Alinity Point-of-Care Analyzer

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The objective of this study was to evaluate the analytical performance of CG4+ and CHEM8+ cartridges on the i-STAT Alinity analyzer prior to use in patient testing. We also evaluated the ease of use, design, and safety features to determine its suitability for use by the clinicians in our hospital. The Abbott i-STAT System Performance Verification Protocol was observed for the imprecision study and was performed over the course of 2 days using 2 levels of control material (Abbott i-STAT Tri Control Level 1 and Level 3). The CLSI-EP6-A guideline was used to verify the assay reportable range performance using 5 levels of linearity material (Abbott i-Stat Tri Control Calibration Verification Set). The method comparison study was performed using up to 60 leftover anonymized heparinized whole-blood samples and serum samples against existing laboratory instruments (Siemens Rapid point 500, Abbott Architect C16000, and Sysmex XN9000). Precision was good (coefficient of variation <2%) for electrolytes, glucose, lactate, and pH, and satisfactory (coefficient of variation <5.2%) for blood gases, urea, creatinine, and hematocrit. Linearity concentrations spanning the analytical measuring ranges were demonstrated for all analytes. Method comparison studies revealed that agreement between the i-STAT Alinity analyzer and the central laboratory analyzers was good and clinically acceptable. The i-STAT Alinity analyzer has good analytical performance, and we established the analyzer meets our safety and regulatory requirements and therefore suitable for use in our hospital as a point-of-care testing device.

P-324

Primary Trauma Care Set up Need Point-of-Care Head Injury Biomarkers

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Globally head injury is a major public health problem; yet remains as under-diagnosed and underreported events closely associated with all forms of trauma of army personal, contacts sports participants, and victims of road crashes with colossal fatalities and disabilities. Infrastructure poor primary healthcare set up in our country require dedicated point-of-care biomarkers for early diagnosis, prompt intervention and optimum decision for timely referral. The aim and objective of the study is to explore user-friendly sensitive and specific panel of point-of-care biomarkers of head injury useful both for pre-hospital injury care and dedicated intervention centers. Literature search has enriched us with the information that research groups are working in laboratory set-up to find chemical biomarkers of neuronal injury in cerebrospinal fluid and serum. Most important neurochemicals earmarked as point-of-care candidate head injury biomarkers are Glial specific neurochemicals viz. Glial fibrillary acidic protein (GFAP), Ubiquitin Carboxy-terminal Hydrolase-L1 (UCH-L1), S100 Calcium-binding β protein (S100β) which can be exploited as the panel for early non-invasive diagnostic investigation in the algorithm to help resolve dilemma of neurological interventions from injury site to outcomes analysis. These lab based research findings on post-injury biomarkers need further translational research from bench-to-bedside to identify cost-effective neuro-chemicals to predict optimum outcome of interventions at the point-of-care. In downstream activities this innovative idea will help us develop diagnostic algorithm to stratify injury severity and early triage followed by targeted interventions in the primary care clinical practice guidelines and standard operative procedures of referral to trauma care centers.

P-325

Performance of Point-of-Care HbA1c Device

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Point-of-care testing (POCT) devices, due to the ease of use, minimal sample volume and fast turnaround time is now commonly used in many healthcare settings. POCT HbA1c testing is important because it is used for diabetic monitoring and diagnosis. In National University Hospital (NUH), POCT HbA1c testing is used for outpatients diabetic monitoring. NUH is currently using

the Cobas b101 analyser. This evaluation study aims to determine the accuracy and reliability of our POCT device against the lab reference method before the analyser is deployed for use. The correlation study was performed using 50 patient venous samples collected in EDTA tube with lab HbA1c test request. The whole blood were first tested on lab reference method, Bio-Rad HPLC. HbA1c samples were chosen based on the HPLC results, across Cobas b101 measuring range. The blood samples were tested within the same day. The concentration evaluated in this study ranged from 4.0 to 12.8 %. Evaluation criteria was based on CAP criteria of ± 7%. Pooled whole blood precision at low and medium concentration were tested over 5 days. Interferences study were also performed for the following: Hb variant, High urea concentration of >20 mmol/L, Low Hb of <7.5g/dL and Pregnancy The Cobas b101 showed good correlation with lab reference method, within CAP criteria of ± 7%. Interferences study results for Cobas b101 correlates with Bio-Rad HPLC except for low Hb, with 90% results giving a negative bias. WB total imprecision at 5.2% and 9.7% concentration were 1.6 and 1.3 %CV respectively. Cobas b101 has good correlation with lab reference method and low CV, making it an important POCT device in NUH outpatient clinics for diabetic monitoring.

P-326

Evaluation of a New Point-Of-Care Testing Device (Pixotest) for Lipid Panel Analysis.

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The PixoTest (iXensor, Taiwan) has been introduced as a point-of-care testing (POCT) device for triglycerides (TG), total cholesterol (TC) and HDL cholesterol (HDL-C). It also reports a calculated LDL-cholesterol based on the Friedewald equation. We evaluated the PixoTest's performance against our laboratory Cobas (Cobas c701, Roche Diagnostics, Singapore) analyser.The performance evaluation included assay linearity (using pooled serum samples), Bland-Altman analysis, analytical precision (using either pooled serum samples and/or Roche control materials), and regression analysis. The PixoTest utilizes enzymatic reactions (TC: Cholesterol esterase/Cholesterol oxidase; TG: Lipoprotein lipase/Glycerol kinase/Glycerophosphate oxidase; HDL-C: detergent, Cholesterol esterase/Cholesterol oxidase) to generate hydrogen peroxide which reacts with a coupler and Peroxidase to form a coloured dye (Quinoneimine) that is read by reflectance photometry. The PixoTest generates results in 3 minutes using either capillary (finger prick) or whole blood (EDTA) samples. The Cobas utilizes enzymatic colorimetric TC/TG/HDL-C assays. Statistical analyses were performed on MedCalc software v18.11.3 (MedCalc, Ostend, Belgium). The PixoTest assays were linear for TC 2.6-9.86 mmol/L, TG 0.76-6.87 mmol/L and HDL 0.98-2.25mmol/L (Table 1). Bland-Altman analysis showed no significant difference between Roche and PixoTest TG (mean difference = -0.05mmol/L, $p = 0.0898$) and HDL-C (mean difference = -0.02mmol/L, $p = 0.3522$) assays, but a significant difference in the TC assay (mean difference = -0.3mmol/L, $p < 0.0001$). Regression analyses showed PixoTest TC = 0.168 + 1.030Cobas, PixoTest TG = -0.0356 + 1.051Cobas and PixoTest HDL-C = 0.154 + 0.894Cobas. The correlation coefficients were close - TC r = 0.91, TG r = 0.97 and HDL-C r = 0.87. Inter-assay (n=20) precision was satisfactory. The performance of TC, TG and HDL-C assays on the PixoTest is good, within the manufacturer's claims, comparable to the Roche assays and fit for operational use, especially in busy outpatient lipid clinics.

P-327

Artificial Intelligence in Clinical Laboratory Medicine

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Artificial intelligence (AI) is rapidly developing in healthcare, as is their translation into laboratory medicine. The computational analysis applies to data analysis, machine learning and computational modelling to make the lab output both more useful and convenient for clinical decision-making and point of care testing (POCT) and diagnostics. AI assists physicians to make better and efficient clinical decisions and or even replace human judgement in the functional areas of healthcare. Powerful AI techniques can unlock clinically relevant information hidden in the massive amount of data, which in turn can assist clinical decision making. Experts have been selected to explain how AI system extracts useful information from a large patient population and assist in making real-time inferences for health risk alert and health outcome prediction. This paper deals with artificial intelligence and its advantages in improving prediction performance and support in the clinical decision-making process. This paper also highlights the AI paradigm shift to healthcare, powered by increasing availability of healthcare data and rapid progress of analytics techniques. A brief discussion on the insights, recent developments in clinical laboratories, along with potential ethical challenges related to artificial intelligence, is discussed.

P-328

Pseudoesterase Activity of Albumin: A Novel Method for Detection of Microalbuminuria

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Microalbuminuria is recognized as a diagnostic biomarker for diabetic nephropathy. The available methods for its detection are dye-based and immunochemical-based. However, they are not free from limitations. The immunochemical-based methods are less sensitive, while the dye-based methods are less specific. So, there is recent interest regarding the development of the method for the detection of microalbuminuria. In this context, we have explored the pseudoesterase activity of albumin for the development of the method for the detection of microalbuminuria. We have observed that several ester substrates when docked in an in-silico system fit at the known active site for the pseudoesterase activity. Following the in-silico study in-vitro validation of the predictions are done. In the in-vitro study, it is observed that human serum albumin when incubated with ester substrate cleaved it into a product which can be detected by coupling it to suitable dye or by fluorescence study. Based on the result obtained, we can say that our method is novel and promising one for the detection of albumin in biological samples.

P-329

Quick Visualization of Proteins on Gel

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Various dyes are known to bind with human serum proteins, which render significant importance in quick visualization of serum proteins. This became the most popular lab practices among clinical and research industries to deal with patient samples. In this context, the effort is to study or investigate the interactions of these dyes with the plasma sample and to detect the presence of serum proteins in a very short period. The interaction of the two dyes named amido black and coomassie brilliant blue (CBB) with human plasma sample was observed by agarose thick layer gel electrophoresis. Amido Black is found to produce protein bands in 30 minutes of destaining after one minute of staining. The bands were more conspicuous in case of CBB (coomassie brilliant blue) in stain only after 30 minutes without any destaining. So, CBB staining is a quick way to produce the results for protein visualization. Our protocol avoids overnight staining followed by destaining, which is generally followed. So CBB can be used rapidly to demonstrate protein bands on the gel.

P-330

Evaluation of Salivary Alpha Amylase (SAA) as a Biomarker for Pre and Intra-operative Stress in Patients Undergoing Infra-umbilical Elective Surgery Under Spinal Anaesthesia.

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Stress is a part of systemic reaction to injury, adverse or very demanding circumstances encompassing a wide range of biochemical, endocrinal, immunological and hematological effects. Subjective assessment tools based on patients' questionnaires e.g. STAI-score, visual analogue score, and Modified Ramsay Sedation Score have limited and disputed value over biomarkers as objective assessment tools. Salivary biomarkers, sampled non-invasively, have been evaluated as stress biomarkers showing a significant increase in response to acute stress. We aimed to evaluate SAA (Salivary alpha amylase) as a biomarker for pre and intra-operative stress in patients undergoing infra-umbilical elective surgery under spinal anesthesia. The study was conducted on 40 patients each in test groups A and B. In total, four saliva samples were collected-first in the evening prior to surgery and second in preoperative room. In group A, third sample was collected 5 min after administering I/V normal saline and fourth sample 15 min after spinal anesthesia followed by I/V Midazolam. In group B, third sample was taken 5 min after administering I/V Midazolam and fourth sample was collected15 min after spinal anesthesia. SAA level was estimated in all samples. P value of SAA levels in first & second samples in group A and B was statistically in significant at 0.973 and 0.285 respectively showing that secretion of SAA in both groups is similar under stress. However, P value=0.001 of SAA levels in both third & fourth samples in group A and B was statistically significant showing the stress lowering effect of Midazolam in group B. Study demonstrates that SAA level is directly related to stress and increases with it. Thus, it can be concluded that SAA can be used objectively for pre and intra operative assessment of stress in patients.

P-331

Effect of Storage on the Stability of Enzyme Activities in Pooled Serum

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To assess the stability of enzymatic parameters in home-made Frozen Human Serum on storage and compare it with commercially prepared lyophilized human sera already being used in our laboratory at IGIMS, Patna. The home made QC serum was prepared from twenty healthy volunteers and was screened for HIV and HBV, pooled together and stabilized with 0.1%Sodium Azide. Preliminary control limits (i.e Mean, SD) was calculated from 20 runs of first month for three enzymatic parameters and results were compared with those of commercially available lyophilized human sera. ALP had narrower coefficients of variation in the home made serum making it a stable control material in comparison to the commercial ones whereas other enzymatic parameters showed unsteadiness over the 6 months storage period. We conclude that the activity of ALP in pooled sera was stable when stored frozen at -20°C over the 6 months storage period.

P-332

Markers in Estimation of Glomerular Filtration Rate in Early Kidney Disease- A Comparative Study

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Biochemical markers have long been the corner stone of diagnosis in kidney disease. The purpose of this study is to compare serum BTP (Beta-trace protein) and serum creatinine in estimation of GFR among the patients with early stage kidney disease. Total of 72 individuals with marginally raised serum creatinine irrespective of their gender between 20 to 65 yrs was included in the study. Renal dysfunction was suspected in all of them. It's a prospective cross-sectional study conducted between June 2018 to December 2018.Serum creatinine by Jaffe's method in Beckmann coulter AU480; BTP by Heterogeneous Sandwich ELISA. Calculation of GFR:-Creatinine based MDRD. BTP based Poge formula were applied. In this study total 72 subjects were included among them the GFR (creatinine based) ranged from 41-150 ml/min/1.73 with mean of 80.94±24.16 SD. The GFR (BTP based) with a mean of 67.26±20.54 SD. GFR estimated by creatinine showed that 59 among the 72 of subjects had GFR > 60 and 13 were < 60, where as GFR estimated by BTP only 44 subjects had GFR > 60 and rest 28 had < than 60, this clearly tells that BTP is able to detect renal impairment better than serum creatinine with a P value of < 0.001 which is highly significant and helps in diagnosing early stages of kidney disease. The need for this study is to find a better marker to evaluate the early functional deterioration of kidney, which would aid in diagnosis and early prevention of devastating kidney damage that may end up with renal transplantation.

P-333

Comparison of Pre and Post Hemodialysis Serum Aminotransferases and their Correlation with Estimated Glomerular Filtration Rate

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Previous studies have consistently indicated low levels of serum aminotransferases in chronic kidney disease (CKD) stage 5, however the cause remains inconclusive. Our study aimed to compare pre and post dialysis values of serum aminotransferases to elucidate the role of dialysis in their alteration. It also aimed to correlate pre-dialysis aspartate aminotransferase (AST), alanine aminotransferase (ALT) and their ratio with pre-dialysis urea, creatinine and eGFR and to detect a relation between weight loss in dialysis and serum aminotransferase levels. A cross-sectional study was conducted on 160 ESRD patients undergoing regular hemodialysis. Pre and post dialysis values were compared using paired t test and Wilcoxon signed rank test. Correlations between the study parameters were assessed by Spearman's correlation test. Body weight, serum urea and AST/ALT ratio showed a highly significant ($p < 0.001$) decrease after dialysis whereas serum AST and ALT concentrations were significantly higher ($p<0.001$). AST showed a significant negative correlation with serum urea ($r=-0.313$, $p=0.015$) and also with serum creatinine ($r=-0.304$, $p=0.018$). A statistically significant correlation was found between weight loss and increase in AST in dialysis ($r=0.3$, $p=0.02$). Our findings support the hypothesis of hemodilution being linked with low serum aminotransferases in End Stage Renal Disease (ESRD). It also emphasizes on the importance of taking into account pre and post dialysis alterations while interpreting the serum aminotransferase values in ESRD for better diagnosis and monitoring of hepatic diseases in ESRD patients.

P-334

Differential Proteomics of Peripheral Blood Monocytes (PBMCs) from Patients with Antitubercular Drug Induced Liver Injury

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Antitubercular Drug Treatment is most commonly associated with hepatocellular type of liver injury; it can be either steatosis or necrosis. Steatosis has been associated with first line antitubercular drug-isoniazid and its metabolites like hydrazine in animal models. To determine steatosis in antitubercular therapy (ATT) induced liver injury we analyzed and compared the proteomic profile of PBMCs among healthy, naïve untreated tuberculosis, tuberculosis without ATT hepatotoxicity (non-toxic) and ATT induced liver injury (toxic) tuberculosis patients. Proteins extracted from PBMCs were separated by two dimensional-polyacrylamide gel electrophoresis (2D-PAGE) and gels were silver stained. Gel images were compared with ImageMaster Platinum 6.0 software. Match sets containing three groups (healthy vs toxic, naive untreated tuberculosis vs toxic, non-toxic vs toxic) were created. Total number of spots and %spot intensities were compared among the groups. The number of spots identified in healthy, untreated naïve TB, non-toxic TB and toxic TB groups were 31, 44, 26 and 31 respectively. Between "toxic v/s healthy" groups, there were 18 matched and 13 mismatched protein spots and of the matched proteins, 3 were upregulated and 3 were down regulated in toxic group. In "toxic v/s untreated" there were 22 matched and 9 mismatched spots and of 22 matched, 10 were upregulated and 2 were down regulated in toxic group. In "toxic v/s non-toxic" there were 19 matched and 12 mismatched protein spots and of 19 matched proteins, 2 were upregulated and 7 were down regulated in toxic group. Thus, the proteomic profile of ATT induced liver injury patients was found to be quite different from that of healthy, untreated naïve tuberculosis and non-toxic TB patients. Further identification of these proteins by LCMS along with pathway analysis can determine the signature proteins and their mechanistic role in ATT induced steatosis.

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Liver Enzymes in Alcohol Dependence Syndrome Patients

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Alcohol dependence syndrome (ADS) has become a global public health challenge because of its high prevalence and the concomitant increase in risk of liver disease, cardiovascular disease and premature death. Influence of alcohol use on liver metabolism is well recognized. This study was aimed at examining the association of liver markers like bilirubin, serum albumin, ??glutamyltransferase (GGT) and aminotransferase, with alcohol dependence syndrome patients. This cross-sectional study was conducted in TU Teaching Hospital. ADS patients were screened by the consultant psychiatrist using the Alcohol Use Disorder Identification Test (AUDIT) questionnaire. A total of 89 patients scored positive on the AUDIT as having alcohol-related problems and were included in the study. Blood Pressure and other anthropometric parameters were measured while blood samples were analyzed for liver markers and other blood parameters. Mean age of cases and controls was 35.42 ± 5.6 & 34.53 ± 3.5 years respectively. The mean values of Gamma GT (181.02 ± 78.16), Alkaline Phosphatase (219.93 ± 76.87), albumin (36.61 ± 5.5). The mean values for serum bilirubin (total as well as direct), SGOT and SGPT were elevated significantly in cases as compared to the controls ($p<0.001$). Among the ADS cases serum GGT level was elevated in 97% patients. The SGOT/SGPT ratio was also significantly higher in cases (2.02 ± 0.39) and control (1.45±0.62). It was found that 15.1% cases had low serum protein level and 32.9% cases were low serum albumin level. Albumin to globulin ratio was also significantly decreased in cases (1.16±0.29). These findings support the hypothesis that, alcohol may affect the pattern of liver markers and also damage the liver cells. Decrease in serum albumin and elevation of SGOT to SGPT ratio more than two is suggestive of development of liver cirrhosis in alcohol dependence patients.

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Usefulness of Serum Globulin Levels for the Discerning Patients with Monoclonal Gammopathies/ Paraproteinemias

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The confirmatory step in diagnosis of monoclonal gammopathies, is bone marrow biopsy and presence of M-protein in serum protein electrophoresis. These tests are relatively expensive & invasive for screening and unavailable in low resource settings. An increased total protein and globulin are clues to the diagnosis of monoclonal gammopathy. We explored their utility for discerning monoclonal gammopathies. The aim of this study was to assess the relevance of serum globulin in the discriminating between patients with & without monoclonal gammopathies/ paraproteinemia. Results of serum protein electrophoresis, and related tests, usually done for investigation of suspected monoclonal gammopathy, were reviewed retrospectively. Reports with an M-band were considered as paraproteinemias, and those without as controls. Serum globulin was calculated as difference of total protein & albumin. Of the 63 paraproteinemias cases, 39 were males & 24 females. Among 84 controls, 41 were males & 43 females. Median serum globulin values in cases were 4.4 (3.5-6.3) g/dL in males and 3.65 (3.33-5.0) g/dL in females. They were significantly higher than those with normal SPE pattern, i.e. 2.9 (2.6-3.1) g/dL in males and 3.1 (2.8-3.2) g/dL in females, with a $p < 0.001$. A cut-off value of 3.25 g/dL of globulin could distinguish between paraproteinemias and normal patients with a sensitivity of 82.1% and specificity of 85.4% in males; a sensitivity of 79.2%, a specificity of 76.7% for females. At another cutoff value of 3.4 g/dL, sensitivity was 77% and specificity 92.7% for males; sensitivity was 75% and specificity 83.7% for females. Alternatively, a cut-off value of 0.458 of globulin/total protein ratio could distinguish at a best sensitivity & specificity of 80% and 89% in males; 83.3% and 83.7% in females. Serum globulin values and globulin/total protein ratio can reliably differentiate between patients with & without paraproteinemias.

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Study of Serum Paraoxonase2 Levels and Carbamylated Total Protein in Chronic Kidney Disease

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There are several diseases which contribute to high prevalence of CKD in India. Prevalence of CKD in India observed to be 17.2% with 6% patients having stage 3 or worse. Carbamylation is posttranslational modification of proteins. Paraoxonase has anti-atherogenic and anti-inflammatory properties. Present study correlates arylesterase activity of PON-2 enzyme with carbamylation of total serum protein in CKD. In this case control study, 30 CKD patients who were not on dialysis were compared with 30 healthy age and sex matched controls. Lactonase activity of PON-2 in monocyte was measured and serum carbamylated total protein was measured by Balion C M et al method. Paraoxonase 2 Lactonase activity in cases (n=30) 1.5002 (±0.791) and controls 1.943 (±0.7469) $p < 0.005$. Carbamylated protein levels in serum OD/mg of protein in cases 0.1306(±0.0619) and controls 0.076 (±0.034) with $p<0.0001$. There is inverse correlation between PON2 Lactonase and carbamylated protein. Spearman $r = - 0.2318$ but it is not statistically significant as $p= 0.07$ Present study shows that CKD is associated with decrease in PON2 Lactonase activity and increased carbamylated proteins. Decreased activities of PON-2 enzymes in CKD may be due to uremic milieu and increased oxidative stress. Increased carbamylated plasma proteins are due to increased cynate formed from chronically raised urea. Carbamylation may leads to decreased activity of protective PON-2 enzyme.

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C-reactive Protein as Risk Factors for Peripheral Vascular Disease in Diabetes Mellitus

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Peripheral arterial disease (PAD) is a condition characterized by atherosclerotic occlusive disease of the lower extremities. While PAD is a major risk factor for lower-extremity amputation, it is also accompanied by a high likelihood for symptomatic cardiovascular and cerebrovascular disease. C-reactive protein (CRP) as a marker of inflammation plays an important role in PAD and now CRP level has emerged as an interesting novel and clinically useful marker for increased cardiovascular risk. The study was aimed to compare CRP as an inflammatory marker and

traditional risk factors in predicting peripheral vascular disease (PVD) in patients with diabetes mellitus. It was a prospective case control study, conducted at Acharya Vinoba Bhave Rural Hospital, Jawaharlal Nehru Medical College, Sawangi (Meghe).The levels of CRP were more in nondiabetics as compared to diabetics. A significant positive correlation was found between CRP and number of vessel involved in disease process. Total Cholesterol, Low density lipoproteins, Very Low density lipoproteins and Triglycerides were positively correlated with the number of blood vessels involved whereas High density lipoproteins were significantly negatively correlated. A non significant positive correlation was found between CRP and Ankle Brachial Index. The Mean ABI in Diabetic Population was less than Non-Diabetic Population suggesting .more occlusive pathology in diabetics than nondiabetics. So, it is concluded that, CRP is an inflammatory predictor of peripheral vascular disease (PVD). CRP is a marker of peripheral vascular disease in predicting the number of vessels involved or the severity of the disease process. The CRP increases with increase in comorbidities along with diabetes and dyslipidemia is a strong predictor of peripheral arterial disease.

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Urinary Enzymes LDH and GGT as Early Diagnostic Marker for Sickle Cell Disease Associated Nephropathy-a Cross Sectional Study.

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Sickle cell nephropathy is a major complication of sickle cell disease. It manifests in different forms, including glomerulopathy, proteinuria, hematuria, and tubular defects, and frequently results in end-stage renal disease. Markers of renal injury, such as proteinuria and tubular dysfunction, have been associated with outcomes among patients with sickle cell nephropathy and provide means for early detection of nephropathy and screening prior to progression to renal failure. Nevertheless, despite the availability of diagnostic and therapeutic strategies, sickle cell nephropathy remains a challenging and under-recognized complication for patients with sickle cell disease. To estimate, assess and compare urinary levels of Lactate Dehydrogenase, Gamma glutamyl transferase, microalbumin in normal healthy subjects, Sickle cell disease with and without nephropathy. Study was conducted at tertiary health care centre in patients of sickle cell disease and normal patients as control. 90 patients ≤17 years were enrolled for the study divided into 3 groups -Group 1: 30 subjects with SCD and microalbuminuria ,Group 2: 30 subjects with sickle cell disease without microalbuminuria, Group 3: Minimum 30 normal healthy subjects .5 ml of spot midstream clean catch urine sample was obtained in a closed container from participants. Urinary enzymes [GGT and LDH] microalbumin and creatinine was estimated , 5ml of blood were collected via venepuncture in a 5 ml syringe and divided into 3ml (plain) and 2ml (EDTA) vacutainers. A positive correlation was found in LDH and GGT level with microalbuminuria in SCD patients with and without microlbuminuria. The study also showed the positive correlation between serum creatinine, LDH, GGT in SCD patients with renal failure. Urinary level of LDH, GGT may be auxillary marker of tubular dysfunction in Sickle cell nephropathy

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Primary Plasma Cell Leukaemia Presenting with a Tri-clonal Gammopathy

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Primary plasma cell leukaemia (PCL) is a rare aggressive form of plasma cell dyscrasia characterized by presence of circulating plasma cells in the peripheral blood. It is diagnosed by either the presence of >20% or absolute count >2x109/L of circulating plasma cells. PCL typically presents with a single monoclonal protein but may rarely be non-secretary and its association to multi-clonal gammopathy is rare. We present a rare case of PCL presenting with a tri-clonal gammopathy. A 43-year-old male was found to have leukocytosis (17.55x109/L) with anemia (Hb 8.2 g/dL) and thrombocytopaenia (55x109/L) during basic investigations following minor injury. There were 21% circulating atypical plasma cells with an absolute plasma cell count of 3.69x109/L on the peripheral blood film. The bone marrow aspirate and the trephine biopsy showed >50% abnormal plasma cells confirming the diagnosis of primary PCL. Interestingly, we discovered 3 discrete monoclonal protein bands (one Ig G lambda and two other free-lambda light chains) in the serum by agarose gel electrophoresis and immunofixation. However, capillary electrophoresis and immuno-subtraction reveled only Ig G lambda band. Serum free-light-chain assays showed increased free-lambda (727.7 mg/L; normal 4.23-27.69) with elevated involved/uninvolved ratio (124). His urine was positive for Bence Johns proteins revealing 2 distinct bands in the gamma region on urine protein electrophoresis. He was a diagnosed patient with liver cirrhosis since 2-years back and during the hospital stay developed features of decompensation and expired prior to commencement of specific treatment for PCL. Multi-clonal gammopathies may be transitory and can be observed at presentation or any time during the course of the disease. Association of PCL to a tri-clonal gammopathy is a rare presentation and the correct diagnosis requires use of appropriate analytical technique as capillary electrophoresis may not detect free-light-chains.

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The Effect of Gym Training and Cycling on Albuminuria among Gym Trainees and Professional Cyclists - A Study from Sri Lanka

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Albuminuria is a sign of defective glomerular filtration membrane. It can be benign and reversible in physical exercise or pathological as in nephrotic syndrome and diabetic nephropathy. The prime aim of this study was to describe effects exercise on urine albumin excretion. A quasi experimental research design was used for the study with an interventional approach. Thirty gym trainees and twelve cyclists were selected using proportionate stratified random sampling and total population sampling respectively from centers in Gampaha district. Urine Albumin to Creatinine Ratio (ACR) was used to assess albuminuria of both groups before and after the standardized training sessions. Paired t-test was used to test the effect of gym training and cycling on albuminuria. Mann Whitney test was performed to compare the post session ACR of gym training and cyclists. The pre and post-session ACR were 1.147 mg/mmol and 3.293 mg/mmol for gym trainees and 1.144 mg/mmol and 1.305 mg/mmol for cyclists respectively. There was a significant difference between pre and post session ACR for both cyclists and gym trainees (P=0.003). There was a positive correlation between the ACR difference and the intensity of exercise (p=0.004, r=0.519), (p=0.002, r=0.793) for gym trainees and cyclists respectively. The gym trainees showed a higher elevation of post session ACR reaching the cut-off limit for micro-albuminuria according to the mean ACR while the mean ACR of cyclists remained normal throughout. The albuminuria is directly proportional to the intensity of the exercise. More research need to be done in order to state that the demonstration of recovery of albuminuria can be beneficial in athletes engaged in severe exercise to ensure absence of negative effect of training on glomerular function. There is also possibility that the post-exercise urinary albumin measurement can be employed in diabetic and hypertensive patients for the early detection of nephropathy prior to development of microalbuminuria.

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Performance Evaluation of ARCHITECT i2000 for the Determination of Whole Blood Cyclosporin A and Tacrolimus

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Regular monitoring of immunosuppressant concentrations in organ recipients is required to maintain their concentrations within the therapeutic range. The blood concentrations of immunosuppressants are routinely measured using one of several automated immunoassays, such as chemiluminescence immunoassays (CLIAs) and liquid chromatography-tandem mass spectrometry (LC-TMS). The ARCHITECT i2000 immunoassay analyzer (Abbott Diagnostics, USA) was developed as an automated CLIA analyzer for the measurement of cyclosporin A and tacrolimus in whole blood. Here, the precision and linearity of the ARCHITECT i2000 analyzer for the detection of cyclosporin A and tacrolimus in whole blood were evaluated according to Clinical and Laboratory Standards Institute guidelines and were compared with those of an LC-TMS detection method. The total coefficient of variation for the two drugs was less than 10%, and they showed linearity values of 0.97 or more, which was within the manufacturer's range. The measurements of both immunosuppressants by the ARCHITECT i2000 were closely correlated with measurements determined by LC-TMS. However, most measurements were lower with LC-TMS than with the ARCHITECT i2000. Measurement of cyclosporin A and tacrolimus in whole blood using the ARCHITECT i2000 showed very satisfactory performance in terms of precision and linearity as well as good correlation with the comparative method.

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An In-silico Approach to Study Lysis Potential of Cholinesterase Substrate in the Presence of Non-oxime Cholinesterase Reactivators

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The problem of acute organophosphorus poisoning is far from control. Erythrocyte cholinesterase (AChE) is considered as a gold standard biomarker of such poisoning. However, activity measurement of AChE is of limited use as the enzyme activity exhibits intra and inters individual variations. For all practical

purpose, the baseline value of the enzyme is not known. Therefore, development of a baseline independent assay of the enzyme activity is of recent concern. In this context, we have observed that reactivation based method development is possible, which will provide baseline independent, personalized assessment of the enzyme activity. We have observed using tools of computational biology that the interaction of the established non-oxime reactivators (NOR)with the classical enzyme substrate is more than oximate ion. This leads us to predict that the lysis of the classical enzyme substrate will be more in the presence of NORs in comparison to oximolysis. Keeping the importance of the baseline independent assay development in mind, we feel that our data is going to help the researchers in the field.

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Drug Screening Test Using LC/MS for Old Patient with Uncertain Medication History

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Many drugs can cause individual adverse effects or can have interactions with other drugs. Therefore, knowledge of current medications is important for accurate diagnosis and appropriate treatment. However, this comes mainly from reviewing medical records and by noting patient history, which are frequently inaccurate. We presently evaluated the performance of liquid chromatography/mass spectrometry (LC/MS) using the 6490 triple quadrupole system (Agilent Technologies, USA) for drug screening of old patients. One hundred serum samples from 98 patients older than 70 years, were collected during outpatient visits. The patients were randomly selected regardless of the disease or medications, without record review, for blind selection. Serum from a healthy volunteer was used as negative control. More than 50 types of drugs (Sigma-Aldrich, USA) were spiked in blank serum (100 ng/mL for each drug) that served as the positive control. The tests were calibrated using blank serum and spiked serum, followed by five spiked sera as quality control and serum from 20 patients each day for 5 days. Samples with a drug concentration of over 1 ng/mL in the quantitative analysis were considered 'positive' and the patients' prior 6-month history was reviewed. A total of 38 samples from 36 patients were positive for a total of 73 items. These sample was positive for approximately two drugs. Only 22 items from 15 patients were confirmed by medical records, whereas 51 items were not recognized. These missed items were predominantly non-prescription drugs, such as anti-histamines and mucolytics. However, prescription drugs likely prescribed by other hospitals were also missed. Although history taking can provide additional information missed from medical records, LC/MS drug screening can be helpful to understand medication history in a more comprehensive manner, particularly in old patients who commonly take multiple drugs. The LC/MS screening detected many drugs not recognized by medical record review.

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Therapeutic Drug Monitoring of Moxifloxacin in Indian Mdr-Tb Patients

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Moxifloxacin (Mfx) is a broad spectrum fluoroquinolone antibiotic used in treatment of multidrug resistant tuberculosis (MDR-TB). Mfx is well absorbed from the gastrointestinal tract however it exhibits wide inter individual variability and drug-drug interactions. There is limited literature on pharmacokinetics of Mfx especially in MDR TB patients where treatment monitoring & adherence play a vital role. The present study aimed to quantitate Mfx levels in patients on ongoing therapy. Mfx quantification was standardized and validated using liquid chromatography mass spectrometry. Plasma levels(pre-dose and 2 hours post dose) were quantitated from 57 treatment-naive MDR-TB patients being followed up at months 1(M1), 2(M2),6(M6) and 12(M12) of treatment. Mfx levels were assessed in 57 patients with a median age of 24 years (range 15-77), 37% men and 63%women with a median weight of 54.8 kg (range 35-91) . At M1, 21% of the study group had sub-therapeutic Mfx levels which reduced to 11% by M2 of treatment. However, 49% patients had toxic levels at M1 which continued at 42% by M2 of treatment. A wide inter as well as intra-individual variability was observed with 4 patients having sub-therapeutic levels at M1 had toxic levels at M2 while 2 patients with toxic levels at M1 showed sub-therapeutic levels at M2 of treatments despite being on same dosage regimens. Mfx levels of M6 & M12 are in process. Median Mfx levels observed at M1 and M2 were 4.72 (0.498-8.7 mg/l) and 4.67 mg/l (0.183-8.78 mg/l) respectively. Significant difference in pre-dose and 2 hour levels were observed (p=0.001) at both M1 & M2. On stepwise multiple

regression analysis, males were found to be statistically significant influencing Mfx levels both at M1 (p=0.02) & M2 (p=0.03). Wide inter-individual & intra-individual variability is observed between MFX levels at month 1 & month 2 of treatment. Monitoring drug levels will be essential to bring the patient in most advantageous position possible.

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Implementation of Rapid Liquid Chromatography-Tandem Mass Spectrometry Assays to Determine Plasma Atorvastatin and Rosuvastatin Concentrations

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The aim of this study was to implement and validate the analytical performance of rapid liquid chromatography-tandem mass spectrometry (LC-MS/MS) assays to determine plasma atorvastatin (AT) and rosuvastatin (RST) concentrations for the pharmacokinetic study in Korean population. We developed two LC-MS/MS methods based on the analysis of 10 μl of buffered human plasma with atorvastatin-d5 (1 μg/mL in 50% acetonitrile) and with rosuvastatin-d6 (5 ng/mL in 50% methanol) as internal standards (IS). Sample preparation of the both assays were based on liquid-liquid extraction with tert-butyl methyl ether (MTBE), and followed by drying, reconstitution, and followed by LC-MS/MS analysis in electrospray ionization positive mode. Mass spectrometry was performed in multiple reaction monitoring mode. Linearity, lower limit of quantitation, accuracy, imprecision, sample stability, carry over, and matrix effect were evaluated for the performance validation of the method. The separation of all compounds was achieved in less than 5 min. The LC-MS/MS method for atorvastatin (AT) and rosuvastatin (RST) showed a good linearity (R2=0.9999) from 0.5 to 20.0 ng/mL and from 0.75 to 15.0 ng/mL, respectively. The lower limit of quantitation (LLOQ) were 0.050 ng/mL for all the analytes. Intra- and inter-run mean percent accuracy were within 94.7-103.1 % and percent imprecision was ≤6%. Stability studies after preparation revealed that all the analytes were stable on 4°C auto-sampler (at 30 and 75 h for AT, and at 24 and 48 h for RST), and also stable at each end of three times freeze and thaw cycles. Carry-over was found to be less than 0.04% for all the analytes. Ion suppression or enhancement were not observed in blank and 6 patient samples. The implemented LC-MS/MS assays to determine atorvastatin and rosuvastatin concentrations in human plasma showed a good accuracy, precision, sensitivity and linearity. The LC-MS/MS assays could be used in various clinical pharmacokinetic studies for atorvastatin and rosuvastatin.

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Utility of Filter Paper Spots in Drug Treatment Laboratories: Experience from AIIMS

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Use of filter paper for bio-sampling was identified way back in 1960s. Dr. Robert Guthrie first used filter paper to screen new born metabolic disorders. After this landmark there was no looking back for the adoption of assays on filter paper. Recently world health organization (WHO) and national institute of health (NIH) has established guidelines for collection and transport of biological samples on filter paper. The use of filter paper spots (urine and blood) for the testing of drug use has grown recently. With advances in detection techniques dried spots has emerged as a potential method for drug testing. Over conventional sampling dried spot sampling is less invasive, simple to store, infection safe, easy to transport and cost effective. Many drugs are reported to be stabilized over dried matrix. This talk would cover the recent advances in dried spots testing for drugs with an emphasis on our own experience of using filter paper as a matrix to collect biological (urine and serum) samples. The dried spots were then analyzed to test drug use at a national level drug treatment center. The clinical validation of the filter paper assay was carried out using samples collected from community clinics run by the center. Filter paper has the potential to be use in drug screening programs with as much accuracy and reproducibility as the conventional methods.

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Improving Operational Efficiency with Siemens Healthineers Atellica Solution

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Operational efficiency is a factor to further improve laboratory performance. This is a study to verify hands-on time for system maintenance and to assess the handling time of paediatric samples on the newly-launched Atellica Solution immunoassay and clinical chemistry analyzer. The operational hands on time for Atellica Solution Daily Maintenance and Weekly Maintenance activities had been verified as 228 seconds and 126 seconds, respectively. This shows a major reduction in hands on time when compared to current instrument in the laboratory with maintenance hands on time of

2062 seconds and 1704 seconds for Daily Maintenance and Weekly Maintenance respectively. The reduction is contributed by the on-the-fly reagent loading capability and auto QC loading feature as well as technical advancement minimizing operator driven maintenance. The ability to schedule the automated maintenance task at off-peak hours improved user efficiency and reduced competency training time. In handling paediatric samples, the time required to process them was also compared to current instruments in the laboratory in three different scenarios: 1) Sample with Chemistry Assays Only, 2) Sample with Immunoassays Only 3) both Chemistry and Immunoassays. The handling time required with Atellica Solution was determined to be 2.67 minutes for all three scenarios while for current instruments, the handling time required were 4.36 minutes, 3.7 minutes and 6.7 minutes respectively. The time saving was due to the feature in Atellica Solution that helped to eradicate the need of pouring sample into a sample cup, followed by the loading of paediatric samples continuously without pausing the system and having one loading point for both the chemistry and immunoassays. The result of this study has shown that the new features in Atellica Solution will help in improving the operational efficiency in the laboratory.

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Immobilization of DNA on Polycarbonate Track-etched Membranes - A Novel Support for DNA Microarrays.

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Fabrication of DNA microarray demands that between ten (diagnostic microarrays) to thousands of probes (research or screening microarrays) are efficiently immobilized to a suitable surface using a suitable chemistry. DNA microarray performance is measured by spot morphology and intensity, background, specificity and sensitivity. To fabricate DNA microarray using polycarbonate track-etched membranes (PC-TEM) as a novel solid support and its comparison to glass and microporous membranes (nylon, nitrocellulose and PVDF).

Optimization of immobilization parameters on PC-TEM. Single stranded DNA specific to rpoB region of Mycobacterium Tuberculosis was used for immobilization studies. DNA was immobilized on glutaraldehyde activated PC-TEMs, silanized and glutaraldehyde activated glass discs, other microporous membranes using 5' amino modified oligonucleotide. Immobilized DNA was hybridized with 5' biotin labelled complementary target DNA and subsequently reacted with 125I labelled streptavidin. Signal intensity and spot morphology was viewed using X-ray film. Various parameters viz. glutaraldehyde, probe DNA, target DNA, and 125I-stpetavidin concentration of as well as optimum time of glutaraldehyde activation, immobilization, hybridization and 125I-strptavidin incubation were optimized. Hydrophobic PC-TEM gave the highest signal to noise ratio and best spot morphology of all the supports tested. PC-TEM activated with 5% glutaraldehyde for two hours and immobilized with 3 μM of probe oligos for overnight gave maximum spot intensity. Optimal target DNA concentration was 1 nM hybridized on chip for 1 hour. Results indicate PC-TEM is a suitable support for the development of DNA microarrays and provides better immobilization than glass. PC-TEM can be used to prepare DNA microarray to study gene specific mutation conferring antibiotic resistance in Mycobacterium Tuberculosis.

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Evaluation of BD Vacutainer Barricor Blood Collection Tubes for Selected Routine Chemistry Testing on a Roche CobasÂ® 8000 Analyzer

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Barricor vacutainers are novel non-gel mechanical separator blood collection tubes. These tubes enable faster pre-analytical processing which could reduce turnaround time, eliminate gel-related problems and reduces cellular contamination effects. Our aim was to evaluate the bias, integrity and analytical performance of these tubes compared to Plasma Separator Tubes (PST) and to non-gel lithium heparin tubes (LHT) for 15 routine chemistry analytes on Roche Cobas® 8000 analyzer. Blood samples were collected in the three vacutainers from each of the 25 participating blood donors. Barricor vacutainers were centrifuged for 3 min at 4000g and PST and LHT for 10 min at 1300g within two hours of collection. Plasma samples (n=75) were then analysed for albumin, alanine aminotransferase, alkaline phosphatase, total bilirubin, calcium, sodium, chloride, potassium, Lactate Dehydrogenase (LDH), phosphorus, NT-Pro-BNP, free-thyroxin, thyroid stimulating hormone, creatinine, and urea. Bias and correlation parameters were determined between tubes. Physical performance evaluation, including tube barrier integrity, fitness to pre-analytical machines, vacuum evaluation, and Plasma appearance, was carried out for the Barricor tubes. All 15 analytes, except LDH, demonstrated comparable results across the reference range (average absolute %bias; linear regression slopes; correlation coefficients) between Barricor and PST (0.06%-5.59%; 0.876-1.019; ≥0.973) and between Barricor and LHT (0.09%-5.75%; 0.936-1.050; ≥0.955). Values for LDH were 10.02%; 1.039; 0.899 between Barricor and PST and 9.42%; 1.043; 0.872 between Barricor and LHT tubes. Lower LDH results were observed with Barricor, which are possibly due to the decreased cellular contamination expected with these tubes. Barricor tubes passed all parameters of the physical performance evaluation. In conclusion, Barricor Tubes demonstrated technically acceptable and analytically equivalent performance for the selected routine chemistry analytes evaluated in this study when compared with PST and LHT tubes, which makes them acceptable alternatives

while offering the added benefit of decreased pre-analytical processing time and less cellular contamination.

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Sialochemistry as a Diagnostic Tool-Establishing Reference Values for Various Biochemical Parameters

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Salivary diagnostics offers an easy, inexpensive, painless, and stress free approach to disease detection. The many advantages of saliva as a clinical tool over serum and tissues are non-invasive collection of samples, good cooperation with patients, cost effectiveness, easy storage and transportation, greater sensitivity, and correlation with levels in blood. Promising new technologies have unveiled large numbers of medically valuable salivary biomarkers for different disease conditions including cancer, autoimmune, viral, bacterial, cardiovascular, and metabolic diseases. Advances in saliva based systems biology has also contributed towards identification of several biomarkers, and other sensitive analytical techniques. However, there is lacuna in relation to standardization of pre-analytical and analytical variables, such as collection and storage methods, circadian variation, sample recovery, accurate choice of collection methods (stimulated or unstimulated), prevention of sample contamination and analytical procedures. Stimulated saliva may be collected through gustatory stimulation, mastication or citric acid use. Unstimulated Saliva can be collected by passive drool directly into plastic tubes. Passive collection is the most recommended method for most analytes. Several physiological factors (e.g. mastication, psychological stress, physical exercise etc.) affect various salivary parameters. Fasting saliva sample is generally preferred as some of its components are influenced by circadian rhythm. Saliva is a hypotonic fluid compared to plasma, some components are found in lower (sodium, magnesium, chloride), higher (potassium, calcium, phosphate) and similar concentrations. Salivary flow rate, composition and protein concentration varies amongst individuals depending on factors such as age, sample processing, preservation. The present study was designed in GGS Medical College Faridkot, a tertiary care hospital to make protocols for analysing routine biochemical parameters in saliva to enhance the utility of this biological matrix. Two hundred healthy participants were enrolled for the study. Both stimulated and unstimulated saliva samples were assessed to define analytical procedure and reference values of various biochemical parameters.

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Thanato-biochemistry in Medico-legal practice: Current Updates

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Thanato-biochemistry has evolved as a specialized branch in the practice of forensic medicine in recent years. Biochemical analysis of postmortem body fluids helps the investigators in cases of natural deaths like diabetic ketoacidosis and metabolic derangements. Additionally, such studies can also aid in medico-legal deaths related to submersion, anaphylaxis, fatal asphyxia, hyperthermia and deaths related to inflammatory insults to the body tissues like early stages of myocardial infarctions and sepsis. The qualitative interpretation of endogenous biochemical substances can become more challenging in forensic autopsies where there is involvement of xenobiotics like sodium chloride, insulin and miscellaneous poisons which may mimic the chemical constitution of endogenous substances. The postmortem evaluation of these substances has to be done considering their postmortem fluctuations in body fluids and changes of decomposition. Of special interest are the studies conducted on vitreous humor, owing to the protected environment around it. Vitreous humor has been used in qualitative and quantitative estimation of D-glucose, creatinine, urea and potassium for various medico-legal deaths. Similarly, various studies have used blood for estimation of acetone beta-hydroxybutyrate in deaths related to alcohol abuse and carbohydrate metabolism dysfunction. Other body fluids like CSF, pericardial fluid, aqueous and synovial fluid have also been studied in forensic autopsies. In spite of these the utility of post mortem biochemistry can be a difficult task owing to the difficulty in assessing the antemortem standardized reference rates, scarcity of data regarding distribution of certain substrates in the body fluids and lack of understanding about the effect of changes of decomposition on the various substrates. Further, future studies are required to explore more in the field of thanato-biochemistry.

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Prevalence of Micronutrient Deficiencies â•'' Do We Need Food Fortification?

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Despite epidemiological transition, India has a continued burden of infectious diseases and nutritional deficiencies. NNACP(National Nutritional Anemia Control Program) was implemented for women in the reproductive age group on basis of 1998 WHO data(>40% prevalence of anemia; DALY 445 per 100,000). Yet, the 2007 WHO database also showed ~40% prevalence of anemia. This may be because NNACP only addresses iron deficiency anemia and does not cater to Vitamin B12 or folate deficiencies. In addition, there are several reports claiming a high prevalence of vitamin D deficiency in Indians. Population data on these deficiencies (vitamins B12, D and folate), however, are meagre. 3 years' retrospective data included 52,267 subjects. Data was subject to statistical analysis for assessment of age and sex-wise prevalence of deficiencies of iron, vitamin B12, folate, and 25-hydroxy-vitamin D

Parameter estimated	Vitamin B12	Iron	Folate	Vitamin D
No. of subjects [Total n=52267]	48307	12140	12601	23005

Mean iron, B12 and 25-hydroxy-vitamin D were significantly(p=0.031, 0.002, 0.048, respectively) lower in females as compared to males. Mean iron was lower in the >60years age group(p=0.001), whereas B12 was significantly(p<0.0001) lower in the 18-40years age group. Overall percent prevalence of deficiency was 66.73, 44.09, 5.08 and 79.05 for iron, B12, folate and vitamin D, respectively. In patients with anemia, prevalent deficiencies were of iron alone(66.73%), B12 alone(36.53%), combined iron and B12(18.88%), folate alone(2.92%), and combined iron and folate(1.77%). The high prevalence of micronutrient deficiencies (iron, vitamins B12 and D) in India, indicates a requirement of population-wide mitigative measures. The most plausible programmes may be

• education in diet diversification, and
— fortification
– of food grains (rice and wheat flour) with iron and B12, and
– of milk with vitamin D,
• continuation of the NNACP.

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Detection of Hemoglobin Variants During Analysis of HbA1c Using HPLC : Time for Focusing on Standardized Report Format

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Hemoglobin A1c (HbA1c) is defined by the International Federation of Clinical Chemistry working group (IFCC) as hemoglobin that is irreversibly glycated at one or both N-terminal valines of the beta chains. It is formed from irreversible, slow, non-enzymatic addition of a sugar residue to the hemoglobin, and the rate of production is directly proportional to the ambient glucose concentration. The long lifespan of erythrocytes (mean 120 days) enables HbA1c to be used as an index of glycemic control over the preceding two to three months and as the adequacy of treatment in diabetic patients. Various factors may affect the accuracy of HbA1c measurements according to the assay method used, of which hemoglobin variants are one of them. Therefore, a falsely high or low HbA1c value caused by the presence of a clinically silent hemoglobin variant may lead to over- or under-treatment of diabetic patients. Cation-exchange high performance liquid chromatography (HPLC) is one of the methods that are vulnerable to the effect of hemoglobin variants on HbA1c measurements, as has been reported previously. In the present study we performed a retrospective analysis of laboratory database on HbA1c reports, performed on Bio-Rad variant HbA1c analyser for the last one year (June 2018 to June 2019). Of the referred cases 63 were identified to be hemoglobin variants, among them 26 cases of Beta thalassemia, 15 of HbE Heterozygous, 13 of HbE Homozygous and 9 of possibilities of iron deficiency, sideroblastic and alpha chain variants were reported. Hb variants may invalidate the results of HbA1c analysis and could result in mismanagement of a patient with diabetes mellitus. It is, therefore, imperative that a comment alerting the requesting clinician to the presence of the Hb variant is appended to the HbA1c results.

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Blood Lead Levels in Antenatal Women with Iron Deficiency Anaemia and Adverse Pregnancy Outcomes: A Pilot Study

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It has been known that serum iron deficiency is associated with increased serum lead levels as Lead is a particularly pernicious element to iron metabolism Exposure to lead is inadvertent as it is widely used in the construction, plumbing, batteries and cosmetics. With increasing industrialization in western Rajasthan, the present study was planned to determine lead levels in pregnant women so as to identify antenatal complications associated with it and initiate changes in the environmental policies and public health care programmes directed towards reduction in lead exposure in the high risk obstetric population. A total of 140 pregnant women were enrolled in our study between 2018-19. Antenatal women was assessed for risk of lead toxicity. Blood lead concentrations were measured in anaemic mothers during their antenatal visit after confirming Iron deficiency anaemia and excluding patients with either non-iron deficiency anaemia or mothers with identified obstetric complications. Neonatal and pregnancy outcomes were assessed in all mothers included in the study. Out of 140 women, anaemic cases were 99 (70.7%) and 41 patients were non-anaemic controls. Age and parity were comparable between anaemic and non-anaemic women. Lead levels were significantly higher in anaemic cases 11 out of 99(10.1%) than in normal patients 1 out of 41(2.4%). There was no dose effect relationship between lead levels and anaemia. Out of 11 patients, 27% (3) women had FGR pregnancy, 9.1%(one) had PROM ,9.1% (one) had preterm delivery and 9.1% (one) baby had congenital anomaly. Women with high blood lead levels are found to have increased incidence of iron deficiency anaemia, FGR and preterm deliveries. Thus it is necessary to make changes in environmental policies towards reduction in lead exposure in a community along with screening mothers with iron deficiency anaemia for lead levels.

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Management of Lower Ureteral Stones Using Tamsulosin - A Prospective Study

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It has been scientifically established that ?1-adrenergic antagonists cause inhibition of the basal tone, peristaltic frequency, and contractions in the lower ureter. Earlier studies have shown promising results with Tamsulosin (?1-adrenergic antagonist) helping in spontaneous passage of Ureteral stones. Hence this prospective randomized double-blind placebo-controlled study is taken up to establish the real impact of Tamsulosin a specific alpha blocker (?1-adrenergic receptors) in expelling distal ureteral stones. This study aimed to include 80 patients with ureteral stones of less than 5 mm and more than 5 mm in size located in the distal ureter. Patients will be randomized to two groups to receive Tamsulosin and placebo along with analgesics whenever required. Patients will be followed up for a period of one month to study the stone expulsion rate, drug side effects and pain episodes. All 80 patients complied with prescribed treatment schedules except 4 patients in placebo group and 2 in study group who were lost to follow up. At the end of 4 weeks, stone expulsion was seen in 30 out of 38 (79% patients in study group and 20 patients out of 36 (56%) in placebo group. The stone expulsion time was shorter in the study group (6.2±3.2 days) and in 9.67±5.4 days for placebo group. No significant impact on the expulsion rate was seen in relation with age, gender and ureteric stones present either in right side or left side. The frequency of pain episodes was almost same and mild in both groups. Tamsulosin is safe and effective drug to enhance spontaneous passage of smaller stones present in distal ureter.

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Involvement of Vitamin D Supplements in Pregnancy with their Infancy

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Fat-soluble Vitamin D (VD) is synthesized in UV-B exposed human skin. It regulates calcium and phosphorus homeostasis and plays an important role in bone mineralization with control of cellular growth and adaptive immune system. The requirement for the nutrients is high during pregnancy and infancy which may increase VD deficiency with risk in developing chronic illnesses and affecting musculoskeletal health. In order to understand the VD status ew associated 25 OHD levels at the time of pregnancy and in infancy along with one year toddler in relation with supplementation in our center. Cross-sectional study was done on 100 pregnant females along with toddler presenting at our center over a period of two years. All were assumed by a questionnaire for regular adequately supplemented VD as recommended by consultants. 25 OHD was investigated in serum by ELISA for qualitative determination. The mean 25 OHD levels in neonatal serum were >25ng/dl. After one year the toddler VD levels were deficient in 55% of cases while severe deficiency was in 35% cases and rests were mild deficient. When considering the maternal VD intake during pregnancy all mother demonstrated normal or high 25 OHD levels till delivery time but after delivery, 75% mother showed a severe deficiency. Only 65% of mothers were aware of the recommendation to give daily VD supplements to their toddles until they were one year of age while a majority of mothers were spending more than 30 minutes a day outside that to completely covering the body (100%) and sunscreen was used by 50%. The study demonstrated a high prevalence of VD status with significant differences due to the existence of supplementation. To improve this situation, pregnant mothers should be educated about the importance of VD deficiency at and after birth for the development of r

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Evaluation of Results Obtained from Patient Analysis with ADPKD by Next Generation Sequencing

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Validation of the new gene panel for PKD1 and PKD2 for the study of Autosomal dominant polycystic kidney disease (ADPKD). DNA extraction was performed using QIAcube and Quiagen kits, and generation of libraries using the Sophia genetics Nephropathies solution Kit. Sequencing was performed with MiSeq (Illumina) using capture amplification. All variants of interest were confirmed by Sanger in external laboratories. Variant interpretation was performed with SOPHIA DDM. A total of 40 variants were detected in the 66 patients analyzed, 28 in PKD1, 9 in PKD2, 1 in PKHD1 in heterozygosis and 1 double mutation in PKD1 and GANAB. Of these 28 variants in PKD1, 13 were known and 15 were not described in the databases consulted. We found 9 variants in PKD2, only 3 of which were known. In this study, 34 (40%) variants were identified as pathogenic (21.2%) or probably pathogenic (18.8%), 11 were considered known mutations, while the other 18 were considered new mutations. We located 8 variants of uncertain significance, 6 in PKD1, 1 in PKHD1 in heterozygosis and 1 in GANAB together with another pathogenic mutation in PKD1, which rules it out as a possible cause of disease. Of the 6 VUS located in PKD1, 3 were not described and we have no family segregation information. The other 3 VUS were described in databases and there was a family history. We performed the segregation study and observed that the variant segregated with disease. The study using NGS has allowed us to find new variants not described previously and that segregate with the pathology, which will allow us to understand ADPKD better. The performance of segregation studies allows us to reclassify the variants which have diagnostic, prognostic and reproductive implications in polycystic patients.

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Serum Gamma Glutamyl Transferase in Cigarette Smokers in South - South, Nigeria

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Gamma glutamyl transferase (GGT) activity is the most useful in the diagnosis of cholestasis caused by the ingestion of substances. The enzyme only reacts on a peptide like compound containing a terminal glutamate residue joined to the remainder of the compound through the terminal (5 or gamma) carboxyl. GGT is present in the kidney cells, pancreas, thymus, prostate and all other tissue except those of the muscle.

In the liver, however, GGT is predominantly present in the cell membranes of the hepatocytes with a small portion found in the cytosol. GGT activity is elevated in any and all forms of liver disease, hence a sensitive indicator of liver disease. Cigarette smoking is a significant factor in the predisposition of individuals to hepatic damage. Substances like Nicotine and Carbon monoxide in Cigarette when inhaled have adverse effect on various delicate organs in the body including the lungs, brain and liver.

The aim of this study is to investigate the influence of cigarette smoking on Serum GGT in cigarette smokers with a view of formulating policies of diagnosis and treatment in this group of patients.

A total of one hundred (100) subjects comprising of fifty (50) male smokers as test and fifty (50) male non-smokers as controls were used for the study. A questionnaire was administered to obtain information such as age, sex, cigarette sticks smoked per day, how often, whether or not they take alcohol alongside smoking. GGT activity and levels was assessed spectrophotometrically using kinetic method from the serum of blood sample collected from the subjects.

Results showed that the serum GGT levels of smokers vary considerably from non-smokers. Serum GGT activity (mean± SD) in smokers and non-smokers were 49 ± 16.26U/L and 15± 7.04U/L respectively. This result shows that serum GGT activity in smokers were significantly higher when compared with control groups. The serum GGT activity in smokers who takes alcohol alongside were 73± 4.69U/L and 43±11.89 U/L respectively and were significantly higher when compared with the controls.

This study shows that smoking and alcohol intake have a synergistic effect in elevating serum GGT activity. Therefore, in the assay of serum GGT in the laboratory for the investigation of liver diseases, smoking and drinking (alcohol intake) habit of the patient should be taken into consideration as this could lead to spurious increase in its activity.

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Study of Vitamin-D Receptor Gene Polymorphism in Association with Serum 25-OH Vitamin-D Levels and Inflammatory Cytokines in Essential Hypertension

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Essential hypertension is a typical example of a complex, multifactorial and polygenic trait. Sandberg K, et al1 Essential hypertension is also called primary hypertension or idiopathic hypertension (Gold black hypertension). A comparative case control study was conducted. All samples were processed and examined according to principles of good laboratory practice at central laboratory Narayana Medical College and Hospital, Nellore. and CLRD. Both male and female hypertensive patients aged between 25-60 years (SBP ?140mmHg and/ or DBP ? 90mmHg) were included. In this study, however, the mean 25(OH) D levels for Essential hypertensive were found to be 24.04 ± 8.62 ng/ml, while in the normal controls, it was 50.46 ± 15.46 ng/ml and p-value (p = < 0.0001 S) was statically significant. Higher hs-CRP levels significantly correlated with higher grades of hypertension and inversely correlated to Vitamin D. In this study IL-17 levels in cases mean ± SD (71.01 ± 13.65 pg/ml) were increased in comparison to controls mean ± SD (19.61 ± 6.08 pg/ml) and p value (p = < 0.0001 S) was statically significant. Our data suggest that VDR gene Fok I polymorphism is associated with the risk of developing essential hypertension. The clarification of essential hypertension etiologies at the molecular level and the identification of genetic variation that confer disease susceptibility are likely to contribute both to disease presentation and development of new medicine.

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Evaluation of Oxidative Dna Damage And Its Correlation With Type 2 Diabetic Patients

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Diabetes is associated with excessive production of reactive oxygen species, which can damage cellular macromolecules, leading to DNA and protein modification and lipid peroxidation. OBJECTIVE: To detect oxidative DNA damage in type 2 diabetic patients alkaline single cell gel electrophoresis (comet assay) and to study the correlation between DNA damage and hyperglycemia, lipid profile, malondialdehyde in type 2 diabetic patients. Blood samples were collected from 50 type 2 diabetic patients and 50 healthy individuals, age and sex matched matched with patients, as controls. Fasting blood sugar and glycosylated hemoglobin were assessed in diabetic patients and healthy controls. Comet assay was used to detect DNA damage. The percent of DNA damage of peripheral blood mononuclear cells was higher in type 2 diabetic patients (34.18±2.84) compared to healthy controls (3.5±0.99) ($P<0.0001$). Pearson correlation analysis showed a significant positive correlation of DNA damage with fasting blood sugar and glycated hemoglobin, but not with serum total chlesterol, triglycerides, high density lipoprotein cholesterol & low density lipoprotein cholesterol and malondialdehyde. Type 2 diabetic patients have more oxidative DNA damage than healthy controls and poor glycemic comtrol may aggravate this damage. Dislipidemia and malondialdehyde is not a contributing factor for DNA damage in type 2 diabetes melllitus.

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Role of Platelets in patients with Acute on chronic liver failure (ACLF)

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Platelet functions have recently been linked in Acute on Chronic Liver Failure (ACLF) patients with reduction in blood counts. However, in context to ACLF patients, involvements of platelet indices are having short term mortality. Platelet is the fragmented part of megakaryocytes and linked to inflammation. Due to high mortality rate in ACLF, the patients are required to hospitalize for immediate management. Though the ammonia levels are raised and predicted to brain herniation in ACLF patients. Since, no study has been established on platelets functions in ACLF so far, therefore we determined the platelet function and correlation of ammonia if any in ACLF patients. Total 25 patients of ACLF were recruited from AB2 ward of department of Gastroenterology, AIIMS NEW Delhi. Healthy subjects (n=15) were also included in the study. Eight ml blood samples were collected from all the subjects and their clinical, radiological, biochemical, hematological investigation were done in all subjects. Platelets indices, ammonia levels, PT and INR were done by standard methods. Increased levels of ammonia were found in ACLF patients as compared to healthy subjects. The platelets counts were also significantly decreased in ACLF ($p < 0.05$) as compared with healthy subjects. Significant increased of ammonia and increased levels of PT/INR in ACLF, it suggest that some hidden mechanism are being involved in progression of ACLF and even platelet counts were also decreases and other platelets indices were changed in ACLF patients

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Investigating the Prognostic Value of Calcitonin and CEA as Biomarkers in Medullary Thyroid Carcinoma

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Medullary thyroid cancer (MTC) are the third most common of all thyroid cancers and characterised with high mortality rate on invasion. MTC is a neuroendocrine tumor effecting the parafollicular cells, also known as C- cells in the thyroid gland. Epidemiology studies demonstrated germ line mutation or family history serve as a background for MTC in patients. Since MTC tends to grow slowly, it usually not characterises with symptoms in patients and therefore, difficult to diagnose in early stage. However, when symptoms do appear, they consist of painless lump in the front of the neck or throat in patients. Therefore, characterising potential biomarkers may facilitate in early detection of the diseaseand serve as a prognostic marker in MTC patients. In normal human physiology, C cells regulate the calcium level through secreting calcitonin. Therefore, calcitonin was chosen as a target biomarker for MTC-cases and a pilot study was conducted using the serological data of MTC patients from last 3 years at Tata memorial cancer research centre. Interestingly, along with calcitonin, the level of carcinoembryonic antigen (CEA) increases in MTC cases. Hence, further comparative study in the pre and post treated cases of same patients was performed to validate calcitonin and CEA as MTC markers in patients. A significant alteration in the calcitonin and CEA level in the preoperative and

postoperative cases further confirms their role as diagnostic and prognostic biomarkers in MTC cases.

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Increased D-dimers in Cerebral Sinovenous Thrombosis with Cardiac Myxoma

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Numerous causes have been provided for the development of Cerebral Sinovenous Thrombosis (CSVT), which can be broadly divided into two categories-infectious, and non-infectious. Formation of thrombi in venous channels draining the brain is a consequence of the characteristic risk factors under the heading of Virchow's triad, which includes local trauma to vessel wall, stasis and a hypercoagulable state, the latter being the most important factor in development of CSVT. The diagnosis of CSVT is based on the presence of risk factors for venous thrombosis, to exclude a diagnosis using D-dimer, and brain imaging using MRI / MRV as gold standard. Myxoma in the heart causes the triad complications, which are the blockage associated with the size and location of the tumor, stasis, pulmonary and systemic embolism as well as the constitutional symptoms due to the release of pro-inflammatory cytokines due to hypercoagulability.

83-year-old woman with low consciousness, spasms and weakness of the extremities, with an increase in D-dimer levels of 2230 ng / mL, based on brain MRI/MRV thrombosis was found in the superior sagittal sinus and myxoma in the left atrium by Echocardiography. The diagnosis of CSVT as a cause of clinical manifestations in patients is established based on brain MRI / MRV, supported by an increase D-dimer and from clinical data was found that there were risk factors that supporting the stasis and hypercoagulability, an elderly with low consciousness and weakness of extremities which were immobilized and also atrial left myxoma. However, it seems that this risk factor is not related to CSVT.

Left atrial myoma as one of the risk factors for CSVT that still needs further investigation to confirm. Clinical communication, history tracking, laboratory tests could support or eliminate risk factors for CSVT.

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The Elusive Diagnosis of Non-Secretory Multiple Myeloma

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Non-secretory multiple myeloma (NSMM) is an uncommon variant of myeloma occurring in less than 10% of myeloma patients. NSMM may arise from plasma cells which are unable to secrete paraprotein or from true absence of paraprotein production owing to defective immunoglobulin synthesis. NSMM is diagnosed by the absence of M-protein in serum and urine with immunofixation, bone marrow clonal plasmacytosis =10% or plasmacytoma and related organ or tissue impairment. Approximately 60% of patients with NSMM produce abnormal serum monoclonal free light chains and are classified non-measurable free light chain-only myeloma in contrast to true NSMM.

A 58 year old female presented to the academic hospital with a three month history of lower back pain, fever and significant weight loss. She was controlled on medical therapy for hypertension, diabetes mellitus and osteoarthritis. Her physical examination revealed pallor and lower back tenderness. Radiographs of her thoracolumbar spine revealed lytic bone lesions. Her haematological investigations identified a normocytic hypochromic anaemia, elevated ESR, bone marrow plasmacytosis of >10% and an abnormal serum free kappa:lambda ratio of 0.15 (reference range 0.26 - 1.65). Her biochemical tests identified normal liver and renal functions, a normal globulin gap and hypercalcaemia. Her urine and serum protein electrophoresis and immunofixation studies did not identify free nor intact immunoglobulins. A diagnosis of NSMM was made and she was referred to oncology for further management. The diagnosis of NSMM in this patient is made from organ impairment attributed to plasma cell proliferation evidenced by lytic bone lesions, hypercalcaemia and anaemia. Furthermore, bone marrow plasmacytosis and abnormal serum free light chains confirmed this diagnosis

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Quantitation of Uracil-DNA Glycosylase Activity by MALDI-TOF Mass Spectrometry Analysis

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Uracil DNA glycosylase (UDG) is a highly conserved DNA damage repair enzyme, acts as a key component in base excision repair (BER) pathway to repair hydrolytic deamination of cytosine in DNA. Thus it is very important in maintaining genome integrity in living organisms. We report here a non-labeled and non-radio-isotopic and very specific method to measure UDG activity. Oligodeoxyribonucleotides containing a site specific uracil is annealed to template DNA forming a defined G-U mismatch and is hydrolyzed by UDG. Resulting product containing an apurinic/apyrimidinic (AP) site is subjected to Matrix Assisted Laser Desorption/Ionization-time of flight mass spectrometry (MALDI-TOF MS) analysis. The cleavage of uracil is identified by the mass change from uracil substrate to AP product. The high resolution of MS results clearly separate product signal (mz = 5445.6) from substrate signal (mz = 5541.6) by the difference of hydrolysis a uracil from DNA (?mz = 96 = 112-16). From UDG kinetic analysis, the Km is 50 nM and kcat is 2.9 s-1 respectively. The method is applied to test uracil glycosylase inhibitor with the IC50 value of 550 nM. We examined UDG activity with single strand and double strand DNAs containing single uracil, the initial rate for removing uracil in single strand substrate was 1.6 faster than in double strand DNA. We also tested uracil at various positions of the oligonucleotide substrates. All the uracil substrates can be hydrolyzed by UDG with different efficiencies. However uracil near the extreme positions of 3' or 5' ends render it difficult to be hydrolyzed by UDG. Taken together, the simple, rapid, sensitive and versatile method has potential to be the reference method for UDG measurement, and can also be used as a tool for UDG inhibitors screening.