Xenobiotic Research in India
Second Annual Meeting

Bangalore,
October 25th to 29th 2017

Affiliate of the International Society for the Study of Xenobiotics (ISSX)

SSX-2017

October 25th to 29th 2017
October 25th: Short Course/Workshops
October 26th, 27th and 28th: Main Conference
October 29th :Hands on Mass Spectrometry Training

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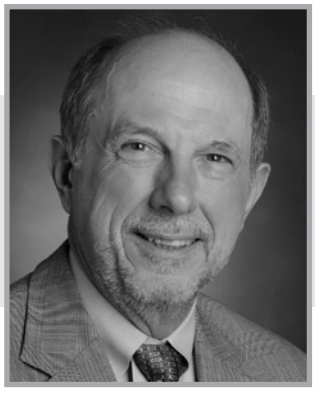
Program at a glance

Day 1: October 26th, 2017		
Time	**Title of Presentation**	***Speakers, Organization***
0845-0850	Opening Remarks	*Griff Humphreys, BMS, USA*
0850-0900	Conference Inauguration	*Murali Subramanian, BBRC, Syngene, India*
Session1: Adverse Drug Reactions		**Chair: F. Peter Guengerich**
0900-0945	Identifying Molecular Basis of Drug Toxicity	*F. Peter Guengerich, Vanderbilt University, USA*
0945-1020	Mechanism of DNA Damage by the Anticancer Drug YM155: Toxicity or Therapeutic Advantage	*Goutam Chowdhury, Shiv Nadar University, India*
1020-1040	Tea Break	
1040-1115	New Methods for the Mechanistic Understanding of DILI: Example with BMS-932481	*Griff Humphreys, BMS, USA*
1115-1135	Small to Large Molecule Metabolite Identification- Sciex Solution for Comprehensive coverage using Novel Workflows	*Vendor Talk; Anoop Kumar, Sciex, India*
1135-1150	Isolation, Characterization and *In vitro* Evaluation of Anti-cataract Potential of Fuciodan from *Sargassum Wightii Greville*	*Young Scientist Presentation by Akshay Shah, M.S.Ramaiah University of Applied Sciences, Bangalore*
1150-1210	Panel Discussion	
1210-1300	**Poster Session**	
1300-1400	**Lunch**	
Session 2: PK/PD Modeling and Simulation, Systems Biology, QSP		**Chair: Vikram Prabhakar Cosponsored by ISoP**
1400-1405	Session Introduction	*Vikram Prabhakar, Vantage, Chennai*
1405-1440	Model-based Drug Discovery and Development	*Sheila Peters, Merck, Germany*
1440-1515	More Power To Permeability Limited PBPK-PD Models	*Matt Harwood, SIMCYP, UK*
1515-1535	Tea Break	
1535-1610	QSP Approaches and Applications in Drug Development	*Rukmini Kumar, Vantage, India*
1610-1645	A Virtual Liver to Predict Hepatotoxicity	*Kas Subramaniam' Syngene Intl, India*
1645-1655	International Society of Pharmacometrics	*Vendor talk, Vikram Prabhakar, Vantage, India*
1655-1720	Panel Discussion	
1720-1735	Interaction Study Between the Hepatic Transport Proteins Organic anion Anion-Transporting Protein 1B1 (OATP1B1) and its Inhibitors via Molecular Docking: An Insight for Drug-Induced Cholestasis	*Young scientist Presentation by Anshul Nigam, Amity University, Mumbai*
1735-1750	Pharmacokinetics and Brain Uptake Study of Novel AMPA Receptor Antagonist using a Validated UHPLC-QTOF-MS method	*Young scientist Presentation by David Paul ,NIPER Hyderabad*
1750-1830	**Visit Vendor Booths**	
1830	**Departure for Reception**	
1900-2130	**Reception: Movenpick Hotel and Spa, Bangalore**	

Day 2: October 27th, 2017		
Session 3:Drug Metabolizing Enzymes and Transporters in Health and Disease Cosponsored by DMDT (IUPHAR) and Solvo Biotech		**Chair: Per Artursson**
Time	**Title of Presentation**	***Speakers, Organization***
0830-0845	Dispersive Liquid-Liquid Microextraction Combined with UPLC-Flourescence as an Efficient and Sensitive Method for Quantification of Benzanthrone in Urine Samples	*Young scientist Presentation by Kajal Karsauliya, IITR, Lucknow*
0845-0930	Intracellular Drug Bioavailability: A New Predictor of Transport and Metabolism Dependent Drug Disposition and Intracellular Target Engagement	*Per Artursson, Uppsala University, Sweden*
0930-1005	Pharmacogenetics of Human CYP2C9 Enzymes	*Allan Rettie, University of Washington, USA*
1005-1025	**Tea Break**	
1025-1100	Investigation of Transporter Mediated Drug-Drug Interactions Using Endogenous Biomarkers	*Hiroyuki Kusuhara, University of Tokyo, Japan*
1100-1135	Prediction of Age and Liver Cirrhosis in Drug Transport and Non-CYP450 Drug Metabolism	*Bhagwat Prasad, University of Washington USA*
1135-1150	Development of a Comprehensive Open-Access Data Module of ADME QPrOmics™ Database Comprising of Abundance an Activity of Drug-Metabolizing Enzymes and Transporters in Humans: Effect Of Age, Gender, Genotype, Ethnicity and Health Condition	*Young scientist Presentation by Mayur Ladumor, NIPER, Mohali*
1150-1210	Panel Discussion	
1210-1300	**Poster Session**	
1300-1400	**Lunch**	
Session 4: DDIs, PK predictions/ DMPK in drug discovery		**Chair: John Miners**
1400-1445	Drug-Drug Interactions Arising from Inhibition of UDP-Glucuronosyltransferase	*John Miners, Flinders University, Australia*
1445-1520	Characterizing *In Vitro* and *In Vivo* Stability of Bispecifics, Fusions, an Immunocytokines in a Drug Discovery Setting	*Larry Weinkers, Amgen, USA*
1520-1540	**Tea Break**	
1540-1615	Renal Drug Metabolism: Implications for Drug and Endobiotic Metabolic Clearance by the Kidney	*Kathie Knights, Flinders Univrsity, Australlia*
1615-1635	Size Will Not Matter	*Venodr Talk,Deepti Bhandarkar, Spinco/ Schimadzu, India*
1635-1650	Perturbation of Arachidonic Acid Metabolism by Dronedarone and Amiodarone: Implications in Cardiac Safety?	
1650-1710	Panel Discussion	
1710-1755	**Special Panel Discussion**	**Moderated by Sandhya Mandlekar**
1755-1900	**High Tea/Networking**	

Day 3: October 28th, 2017		
Session 5: New technologies in DMPK		**Chair: Philip Tagari**
Time	**Title of Presentation**	***Speakers, Organization***
0830-0850	HepaRG and Silensosomes™: *In vitro* Tools to Study Drug Metabolism and Hepatotoxicity for Pharmaceutical Industry	*Vendor talk, Ashwani Sharma, Biopredic, France*
0850-0935	Graphene-based Sensors for Bioanalyte Detection & Quantitation	*Philip Tagari, Amgen, USA*
0935-1010	Exploring Heat Shock Protein 90 as a Target for Anti-infective Drug Development	*Utpal Tatu, IISc, Bangalore*
1010-1030	**Tea Break**	
1030-1105	Advances in Characterization of the Pharmacokinetics and Disposition of Small Molecule Drugs	*Raju Subramanian, Gilead Sciences, USA*
1105-1140	Cellular Simulation for Suspended and Sandwich Hepatocytes Applied to Human Predictions	*Mike Bolger, Simulations Plus, USA*
1140-1155	Studies on Phytoenzymes as Tools for Metabolism Based Phytoremediation	*Young Scientist Presentation by Girish Prakash Gurbani, BCP, Mumbai*
1155-1215	**Panel Discussion**	
1215-1300	**Poster Session**	
1300-1400	**Lunch**	
Session 6: Personalized Medicine/Regulatory Topics		**Chair: Amita Joshi**
1400-1445	Paving the Way for Personalized Medicine: Opportunities for Clinical Pharmacology	*Amita Joshi, Genentech, USA*
1445-1520	Pharmacokinetics and Pharmacodynamics of Biologics: Technical and Regulatory Considerations	*Narendra Chirmule, Biocon, India*
1520-1535	**Tea Break**	
1535-1610	Translational Medicine, from Discovery to First In Human (FIH) and Phase I/II Clinical Studies	*Chandrashekhar Natarajan, Tufts University, USA*
1610-1625	Lipid Carriers Loaded with Antimicrobial Agents for the Prevention of Ventilator Acquired Pneumonia	*Young Scientist Presentation by Vanaja K Satheesh, Visveswarapura Institute of Pharmaceutical Sciences, Bangalore*
1625-1645	Panel Discussion	
1645-1700	Concluding Remarks	*Griff Humphreys, BMS, USA*

Speakers
Bio and Abstracts



Fred Guengerich
Tadashi Inagami Professor,
Biochemistry,
Vanderbilt University School of Medicine, USA

Dr. F. Peter Guengerich received his Ph.D. in Biochemistry from Vanderbilt in 1973 in the area of alkaloid biosynthesis (H. Broquist), was a research fellow at the University of Michigan (M. J. Coon), and joined the faculty at Vanderbilt in 1975 (Professor since 1983). His research deals with the chemical and biological mechanisms by which drugs and cancer-causing chemicals are processed and the relevance to drug development, toxicity, and disease. A major area of interest is the cytochrome P450 enzymes, which are the major catalysts involved in the metabolism of drugs, carcinogens, and steroids. Studies with the recombinant human P450 enzymes involve the molecular basis for substrate and reaction discrimination. He is an author/co-author of 695 original research articles, 220 invited reviews, and 136 proceedings chapters. He is an associate editor of both *The Journal of Biological Chemistry* (also Deputy Editor) and *Chemical Research in Toxicology*. Prof. Guengerich is currently a member of the Expert Panel of the Flavor and Extract Manufacturers' Association. In 1992 he received the B. B. Brodie Award from the American Society for Pharmacology and Experimental Therapeutics and in 2011 he received the Founders' Award from the Chemical Toxicology Division of the American Chemical Society and in 2013 he received the Merit Award of the Society of Toxicology. He has also received the Scott Award from Toxicology Forum and the CIIT Founders' Award. In 2010 he received the ISSX R. T. Williams Distinguished Scientific Achievement Award. Prof. Guengerich has trained 20 graduate students and 134 postdoctoral fellows/visiting scientists and received two Vanderbilt mentoring awards for his work with postdoctoral fellows, one of which is now named for him.

Abstract of the talk

Identifying molecular bases of drug toxicity

Safety issues are a major cause of attrition of drug candidates. Drug-induced liver injury is a very serious clinical challenge in many countries. Understanding the molecular issues is critical in improving screening and regulatory policy, and our knowledge of mechanisms is far from complete. Several mechanisms have been defined and are operative in certain cases, even if they do not account for all aspects of drug toxicity: bioactivation to reactive products, immune hypersensitivity, and changes in some critical proteins. Distinction must be made between intrinsic and extrinsic toxicity, the latter of which is more difficult to predict. Drug metabolism polymorphisms have some roles but have been of limited used in explaining both extrinsic and especially idiosyncratic toxicity. Some new biomarkers for drug-induced liver injury have been developed, and several in vitro strategies are being applied. (Supported in part by U.S. Natl. Inst. Health grant R01 GM118122).

Goutham Chowdhury
Assistant Professor and
Ramalingaswami Fellow of Chemistry,
Shiv Nadar Univeristy, India

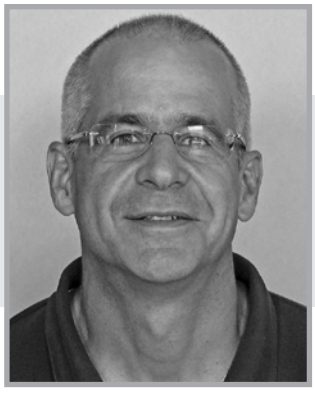
Dr. Goutam Chowdhury did his PhD from Dr. Kent Gates' lab at University of Missouri-Columbia and postdoc from Dr. F. Peter Guengerich's lab in the field of cytochrome P450 and molecular toxicology. He worked as a research instructor at the Vanderbilt University Medical Center before joining Covance Laboratories Inc. in Madison, WI, USA as a staff Scientist. During his career, he has published more than 30 papers in international journals.

Dr. Chowdhury lab specializes in molecular toxicology and drug metabolism. His research spans from the study of DNA and protein damage to P450, drug metabolism, and mass spectrometry. At present, his lab is interested in understanding the mechanism of toxicity of various drugs and molecules including thalidomide, YM155, withanone and 8-nitroguanine. Another area of research in his lab is the development of possible predictive biomarkers of reactive intermediates formed from various drugs.

Abstract of the talk

Mechanism of DNA damage by the anticancer drug YM155: Toxicity or therapeutic advantage

DNA damage, if not repaired leads to cytotoxicity including mutagenesis, DNA aberration and cell death. Yet majority of the chemotherapeutic drugs cause DNA damage. DNA damage provides a therapeutic potential for cancer treatment, although being toxic. Sepantronium bromide (YM155) is a small molecule that selectively inhibits the expression of survivin. Survivin is an “inhibitor of apoptosis” protein, preferentially expressed in transformed cells. It is a nodal protein involved in crosstalk with a number of signaling pathways. YM155 is currently in phase II clinical trials for the treatment of various cancers. Recently, YM155 has been reported to cause DNA damage, although the mechanism is not clear. There are contradictory reports if DNA damage truly is necessary for its mode of action. We show that YM155 causes cytotoxicity and extensive DNA damage particularly in the triple negative breast cancer cells, a subtype that is extremely agrresive with no avaliable targeted therapies. The mechanism of DNA damage involves generation of reactive oxygen species through a reductively activated, radical mediated pathway that is distinct from that of typical quinones. Our data indicate that YM155, although being a targeted anticancer agent, DNA damage may well be its primary mode of action.



Griff Humphreys
Executive Director,
Biotransformation Department,
Bristol-Myers Squibb, USA

Griff Humphreys received his graduate training at the University of Virginia in chemistry and completed a post-doctoral fellowship at Vanderbilt University in the Center in Molecular Toxicology. He then joined Bristol-Myers Squibb and has been with the company for 23 years. He oversees a group responsible for drug metabolism, disposition and metabolite identification studies during the candidate optimization and drug development phases. His interests include the consequences of reactive metabolite formation, development of new analytical methodologies for metabolite detection, reaction phenotyping of CYP and UGT catalyzed biotransformations, predictive metabolism and toxicology models, *in vitro-in vivo* correlations, and strategies for candidate optimization.

Abstract of the talk

New methods for the mechanistic understanding of DILI: Example with BMS-932481

Drug induced liver injury (DILI) is a major cause of attrition in all stages of drug development. The clinical development of BMS-932481, a gamma-secretase inhibitor, was discontinued due to elevated alanine aminotransferase (ALT) levels found in subjects dosed in the multiple ascending dose portion of the first in human protocol. Elevations in ALT levels are generally related to liver injury. This finding was unanticipated as there was no indication of liver related findings in toxicology studies. In order to understand the molecular mechanism of the ALT finding, a series of in vitro experiments in models thought to be predictive of liver toxicity were performed with BMS-932481. The in vitro models employed allowed measurement of oxidative stress, mitochondrial function and inhibition of bile acid transporters. The results of these studies identified bile acid transporter inhibition as a potential mechanism for toxicity. The result was used to gain further insight into the in vivo situation using a PBPK linked systems biology software representation of the liver (DILIsim®). Results from this study as well as future directions will be discussed.

Anoop Kumar
Application Support Manager,
Pharma/Non Pharma Support,
SCIEX, India

Anoop Kumar is working as Application Support Manager in SCIEX, India. Primarily, Anoop is responsible for Pharma/CROs (Qual and Quant) and Non-Pharma (Food & Beverage, Forensic Toxicology, and Clinical Research) application support. Anoop is leading the team of highly enthusiastic mass spectrometric professionals to support the customers and implement the regional and global business strategies. He is responsible for managing the smooth operation of SCIEX demo lab. Anoop did his post-graduation (Organic chemistry) from Gorakhpur University and Ph.D (Analytical Chemistry) from Agra University. Anoop has done his Post-Doctoral Fellowship from Department of Environmental Health Sciences, Tulane University, New Orleans, Louisiana, USA. The post-doctoral research projects includes development of methods for identification and quantification of novel major blood protein adducts as biomarker for human known carcinogens *in vivo* and *in vitro* samples using QTRAP™ LC/MS-MS systems. Anoop worked as Research Associate in Agricultural Chemical, Division of Indian Agricultural Research Institute, New Delhi. Anoop has published more than 35 peer reviewed research papers and technical notes for different application using various analytical tools. He has given several oral and poster presentations in National and International symposium. Currently his major application interest is developing bioanalytical quantitation method for small molecules, peptides, therapeutics drugs and Impurity profiling, genotoxic impurities quantitation and metabolite identification studies using various SCIEX triple quadrupoles, QTRAPs™ and high resolution mass spectrometric system.

Abstract of the talk

Small to Large Molecule Metabolite Identification – Sciex Solution for Comprehensive Coverage using Novel Workflows

High Resolution LC-MS/MS has become an indispensable technique of choice for metabolite identification in recent time. Novel, data-independent acquisition (DIA) strategies such as SWATH Acquisition now make non-targeted analyses a reality in complex biological matrices and provide overall information of low-abundance and major metabolites. The magnificent quantitative capacity of a triple quadrupole and the high-performance accurate mass analyzer of a high-resolution time-of-flight mass spectrometer are united into one innovative, hybrid instrument, the TripleTOF 5600+ and TripleTOF 6600 system – that performs both quantitative and qualitative analyses with just one method. Following topics will be covered during the presentation.

1. MetabolitePilot™ 2.0 Software capabilities for Small and Large molecules data processing
2. Peptide Bioanalysis and Catabolites identification using Triple TOF system.
3. Sequential Windowed Acquisition of All Theoretical MS (SWATH™) approach for Drug Metabolism

As high resolution instruments have become increasingly powerful, manual processing and interpretation of large amounts of data pose a major challenge. Integrated software to detect metabolites, perform structure assignment and MS/MS interpretation, and compare metabolism across multiple samples is essential to realize the benefits of high resolution data. SCIEX TripleTOF® 5600+ and 6600 systems combined with MetabolitePilot™ 2.0 software offer an intuitive fully integrated hardware and software solution that sets the standard in speed, sensitivity, and ease of use.

Sheila Peters
Head, Translational Quantitative Pharmacology, Merck Serono, Germany

Dr. Sheila Annie Peters heads the Translational Quantitative Pharmacology group at Merck Darmstadt. Her areas of expertise in DMPK include Physiologically-based pharmacokinetic (PBPK)-Phamacodynamics (PD) modeling, and PK/PD and drug-drug interactions. Prior to this, she worked at AstraZeneca, Mölndal, where she developed a generic whole-body PBPK model in MATLAB® which she used to support several drug discovery and early development projects across different R&D sites with innovative approaches to identifying potential limitations to drug disposition. She successfully implemented Model-based drug discovery (MBDDx) strategy in Respiratory Inflammation and Autoimmunity iMed through cross-functional collaboration. She won the 2013 IMED (Innovative Medicines) Science Award at AstraZeneca for the "Design and Development of LungSim Simulation tool for Inhalation PK Modelling". She has published several papers in high impact journals as well as a book on PBPK. As part of the IQ Consortium, she co-authored a White Paper on PBPK along with other experts in industry and continues to work with them on various topics of interest in PBPK.

Abstract of the talk

Model-based drug discovery and development

Matthew Harwood
Senior Scientist,
Translational DMPK,
SIMCYP (Certara), UK

Matthew obtained his Bachelor's and Master's degrees in Physiology and Human Nutrition from The University of Sheffield (2000) and PhD (gut transporter proteomics & IVIVE-PBPK) from The University of Manchester (2015). His first PK research involvement was at Hope Hospital, Manchester, sponsored by Pfizer in 2003, using ex vivo intestinal tissue to understand region-specific intestinal drug permeability/transporters. Matthew joined Simcyp Ltd in 2007 and is principally involved (including as project lead) in the development of human & preclinical PBPK models with specialisms in drug transport and metabolism. He has 15 peer reviewed publications and has been a session moderator and invited speaker at the AAPS.

Abstract of the talk

More power to permeability limited PBPK-PD models

Physiologically-Based Pharmacokinetic (PBPK) modelling combined with In Vitro-In Vivo Extrapolation (IVIVE) has become entrenched within pharmaceutical drug development and provides the basis for many regulatory submissions for new drug applications. The ability to model the temporal drug profiles of unbound drug disposition in the blood/plasma and several key organs is attractive from a PK, PD and toxicological perspective. The generation of relevant in silico virtual populations provides the user with the ability to negate or waive clinical trials by employing drug-dependent parameters, i.e. Phys-Chem, enzyme and transporter kinetic properties that drive drug disposition, not only in healthy individuals but those individuals or populations which are deemed to be potential vulnerable to loss of efficacy or toxicity. The focus of this presentation will be to introduce the Simcyp approach to generating virtual populations; case studies demonstrating the utility of PBPK models when investigating the impact of transporters on drug disposition in healthy volunteers but also other sub-populations such as paediatrics are put forward. The final part of the talk will discuss some of the considerations for generating a virtual 'Indian' population and how key demographic and physiological parameters might be different to a healthy North European population and the impact these could have on drug disposition.

Rukmini Kumar
Principal Scientist and Co-founder,
Vantage Research, India

Rukmini Kumar started working on modeling during her Ph.D at University of Pittsburgh, during which she built a systems model of sepsis, collaborating with researchers from University of Pittsburgh Medical Center. Her work contributed to scientific publications and to the initial model developed at Immunetrics. She worked at Entelos, a pioneer in physiological modeling for 8 years, leading several pharma R&D projects in diverse disease areas, as well as engineering efforts in developing new models for the epidermis and liver toxicity. Rukmini then co-founded Vantage Research, a Modeling & Simulation (M&S) company that focuses on Quantitative Systems Pharmacology. Vantage Research works with Pharma R&D clients in therapeutic areas such as diabetes, auto-immune diseases, hypertension and immuno-oncology.

Abstract of the talk

QSP approaches and applications in drug development

Quantitative and Systems Pharmacology (QSP) aims to understand how drugs modulate physiological networks in space and time, in order to predict effect of drug targets and their role in human pathophysiology. The QSP approach relies on developing mathematical models that capture mechanistic details of the physiology of interest, and has a long history in academic research. In QSP, data from multiple scales - from the "top-down" clinical scale to "bottom-up" scales such as protein-receptor interaction, signalling pathways, interaction between multiple organ systems and so on - are used to quantify and constrain the models. Integrating knowledge across these various scales, identifying knowledge gaps and generating plausible hypotheses is the value generated from this approach. As an emerging field, QSP methodologies, standards and tools are continuing to evolve, offering an exciting arena for modelers to push the envelope in the life sciences.

Kas Subramanian
Head of Bioinformatics,
Syngene International,
India

Kalyanasundaram Subramanian (Kas) is the Head of Bioinformatics at Syngene International. For the past two decades he has been working on modeling chemical and biological systems using a variety of techniques ranging from machine learning to dynamical systems analysis. At Syngene, he leads a team that primarily works with biological and chemical data from various sources, integrates them; derive patterns and themes to provide decision support. Prior to Syngene, he was Chief Scientific Officer at Strand Life Sciences Pvt. Ltd where he led the groups involved in the development of the Virtual Liver and the interpretation platforms for NGS-based diagnostics. He headed the Collaborative R&D group for immunology products at Entelos, a systems biology company and has also worked at Genetic Therapy Inc (Novartis) where he helped found a group to perform research in synthetic and hybrid vectors for gene delivery. He has a B.Tech. in chemical engineering from the Indian Institute of Technology, Bombay, India and an MS from SUNY at Buffalo. His Ph.D. is in Biomedical Engineering from the Johns Hopkins University, School of Medicine.

Abstract of the talk

A virtual liver to predict hepatotoxicity

Hepatotoxicity confounds drug development both at preclinical stages as well as during clinical trials. Several methods and systems to predict hepatotoxicity have varying degrees of accuracy and applicability. Our approach is to combine a systems model of liver physiology with an in vitro assay set to predict outcomes in vivo.

To do this, all major forms of liver injury observed in the clinic were studied. These modes of injury were then mapped to the essential biochemical pathways that must be deranged to produce the corresponding clinical manifestations. A mathematical model of the liver was then developed based on the kinetics of the enzymes involved in the essential pathways. This model accurately represents energy and anti-oxidant metabolisms, in that it is able to reproduce liver homeostasis. In addition, it is able to demonstrate liver injury upon simulation of energy or anti-oxidant depleting stresses. It also replicates liver and whole system behavior on induction of stresses that restrict bile flow. In addition, the model evolves to demonstrate fatty liver as a response to mitochondrial dysfunction as well as disturbances in fatty acid uptake. Thus the model is appropriately tuned to address all major forms of liver toxicity, namely, hepatocellular necrosis, cholestasis and steatosis.

In this talk, the process of model building will be described and real-life examples of how this system was used to predict outcomes in pharma R&D will be presented.

Vikram Prabhakar
Co-founder and CEO,
Vantage Research

Vikram Prabhakar is co-founder and CEO at Vantage Research, a Pharma Modeling & Simulation company focused on Quantitative Systems Pharmacology. After an MS in Computer Engineering from Drexel University, Vikram spent 12 years in Networking & Communications industry, first at Marconi and then at Cisco, in roles ranging from engineering to marketing and business development. Since 2013, Vikram has helped build Vantage into an accomplished QSP group, working with multiple global Pharma companies. Vikram has an MBA from Berkeley's Haas School of Business.

Per Artursson
Professor,
Dosage Form Design,
Department of Pharmacy Uppsala University,
Sweden

Per Artursson is a professor in Dosage Form Design at the Department of Pharmacy, Uppsala University, Sweden where he heads the Drug Delivery research team. He is also Director for the Uppsala University drug optimization and pharmaceutical profiling platform within Science for Life Laboratories. His research aims at understanding drug absorption, distribution, metabolism and elimination (ADME) at the molecular and cellular level in order to deliver drugs more effectively via the oral route. He also investigates the effects of drug transporting proteins on drug disposition and drug interactions. He has published over 200 research articles and reviews, is highly cited and has received several international awards for his research.

Abstract of the talk

Intracellular drug bioavailability: A new predictor of transport and metabolism dependent drug disposition and intracellular target engagement.

Intracellular drug exposure is influenced by cell- and tissue-dependent expression of drug-transporting proteins and metabolizing enzymes. Here, we introduce the concept of intracellular bioavailability (Fic) as the fraction of extracellular drug available to bind intracellular targets, and we assess how Fic is affected by cellular drug disposition processes. We first investigated the impact of two essential drug transporters separately, one influx transporter (OATP1B1; SLCO1B1) and one efflux transporter (P-gp; ABCB1), in cells overexpressing these proteins. We showed that OATP1B1 increased Fic of its substrates, while P-gp decreased Fic. We then investigated the impact of the concerted action of multiple transporters and metabolizing enzymes in freshly-isolated human hepatocytes in culture configurations with different levels of expression and activity of these proteins. We observed that Fic was up to 35-fold lower in the configuration with high expression of drug-eliminating transporters and enzymes. We also used Fic to improve the accuracy of in vitro predictions of clinical drug-drug interactions using a series of 10 time dependent inhibitors. Finally, we determined Fic in multiple cellular assays and cell types representing different targets from a number of therapeutic areas, including cancer, inflammation and dementia. We found Fic predicts drug access to intracellular targets and hence pharmacological effect. Further, Fic gives new insights on membrane permeable compounds in terms of cellular potency and intracellular target engagement compared to biochemical potency measurements alone. We conclude that Fic provides a measurement of the net impact of all cellular drug disposition processes on intracellular bioavailable drug levels. Importantly, no prior knowledge of the involved drug distribution pathways is required, allowing for high-throughput determination of drug access to intracellular targets in highly defined cell systems (e.g., single-transporter transfectants) or in complex ones (including primary human cells). Knowledge of the amount of drug that is locally available to bind intracellular targets provides a powerful new tool for compound selection in early drug discovery. Finally, our most recent studies suggest that Fic has the potential to improve the accuracy of in vitro predictions of clinical drug-drug interactions.

Allan Rettie
Professor,
Department of Medicinal Chemistry,
University of Washington, USA

Allan Rettie obtained a Ph.D. in Pharmaceutical Sciences in 1983 from the University of Newcastle-upon-Tyne, England, before moving to Seattle to post-doc with Drs. Mont Juchau and Dr. Bill Trager in the areas of extrahepatic drug metabolism and metabolic drug-drug interactions, respectively. He joined the faculty of the UW School of Pharmacy in 1987 and was Departmental Chair from 2000-14. His primary research areas of interest for almost 30 years have been the elucidation of the chemical, enzymatic and genetic basis for adverse drug reactions, especially those involving CYP2 and CYP4 family enzymes. Dr. Rettie serves or has served on the editorial boards of *Drug Metabolism and Disposition*, *Drug Metabolism Reviews*, *Journal of Pharmacology and Therapeutics*, *Current Drug Metabolism*, *Chemico-Biological Interactions* and *Chemical Research in Toxicology* as well as numerous NIH grant review panels. He is past Chair (2010-14) of the IUPHAR (International Union of Basic and Applied Pharmacology) Section of Drug Metabolism and Transport and past Chair (2013-15) of the Scientific Affairs Committee of International Society for the Study of Xenobiotics (ISSX). Dr. Rettie is a Fellow of the Japanese Society for the Study of Xenobiotics (2016) and received the North American Scientific Achievement Award (2005) from ISSX for his work on elucidating metabolic and genetic mechanisms of adverse drug reactions involving the anticoagulant drug, warfarin.

Abstract of the talk

Pharmacogenomics of human CYP2C enzymes

Precision (or 'personalized') medicine has the goal of improving the quality of patient care by optimizing drug therapy through the application of individualized pharmacogenomic information. This presentation will review polymorphic variability in the human CYP2C sub-family with an emphasis on CYP2C9 and CYP2C19. These two closely related enzymes together metabolize 20-25% of all drugs that are cleared by oxidative metabolism. Updates will be provided on comparative enzyme structure-function relationships, variability in common allele frequencies with an emphasis on Indian populations, clinically relevant drugs that are affected and future prospects for cataloging all functional consequences of coding-region variation at this gene locus.

Hiroyuki Kusuhara
Chair, Laboratory of Molecular Pharmacokinetics, Graduate School of Pharmaceutical Sciences,
University of Tokyo, Japan

Hiroyuki Kusuhara received his BSc, MSc and PhD (Pharmaceutical Sciences) from the University of Tokyo (Japan). Hiroyuki started his carrier as an academic scientist in The University of Tokyo as Assistant Professor of Pharmaceutical Sciences (1998). He was promoted to Associate Professor (2004) and Professor (2012) of Graduate School of Pharmaceutical Sciences, The University of Tokyo. He is currently professor and chair of Laboratory of Molecular Pharmacokinetics at Graduate School of Pharmaceutical Sciences, The University of Tokyo, Tokyo, Japan.

Hiroyuki's major research interest encompass interindividual variability in human drug Disposition, specifically: identification of drug transporters in the tissue distribution, and clearance; pharmacokinetics; modeling and simulation; in vitro-in vivo extrapolation; PK imaging; drug-drug interactions; and metabolomics. He is the author of 180 research papers in these areas.

He is a member of ISSX since 2013. Most recent ISSX activities include Scientific Affairs Committee (2014-2016), and Nominations Committee (2014-2016). Hiroyuki has experience as Council and Director in Japanese societies; JSSX Council (2004-present), Director (2014-2017); APSTJ Council (2010-present), Director (2017-present). *Editorial Board membership:* Drug Metabolism & Disposition; Journal of Pharmaceutical Sciences; Biopharmaceutics and Drug Disposition. *Society membership*: ISSX, ASCPT, and Japanese Societies; JSSX, JSCPT, APSTJ, JPS and JSDDS.

Abstract of the talk

Investigation of transporter mediated drug-drug interactions using endogenous biomarkers

Quantitative assessment of drug-drug interactions is an important issue during development of new investigational drugs. Currently, transporter substrate drugs are administered for assessment of transporter inhibition potency of the new investigational drugs at their clinically relevant doses. We have advocated to use endogenous substrates as surrogate probes for DDI assessment, particularly caused by competitive inhibition. We employed metabolomic analysis to identify appropriate endogenous substrates in human plasma and urine specimens from healthy volunteers given transporter inhibitors for drug transporters such as OATP1B, OAT1, OAT3 and MATEs. Changes of the plasma concentrations of endogenous substrates showed good correlation with those of the typical probe substrates. One of the main advantages of using endogenous probes is that one can circumvent the need to conduct a formal DDI study with probe drugs. Using endogenous probes could allow DDI assessment in the Phase I study, the results of which help designs of Phase II and Phase III studies (exclusion criteria related to concomitant medications), and reduce the costs and time to conduct a DDI study using a probe drug due to false positive DDI prediction. This approach also allows DDI assessment in clinical settings where administration of probe drugs to patients is not ethically allowed.

Bhagwat Prasad
Assistant Professor,
Department of Pharmaceutics,
University of Washington, USA

Dr. Bhagwat Prasad is an assistant professor in the Department of Pharmaceutics, University of Washington (UW), Seattle, WA. He also serves as a co-director of the UW research affiliate program on transporters (UWRAPT). Before joining faculty position, Dr. Prasad worked as a postdoc and a lead scientist, UWRAPT. Dr. Prasad obtained his MS in 2006 and Ph.D. in 2010 in Pharmaceutical Analysis from NIPER, Mohali, India. In his role as a PI or co-inv. in various NIH, US-FDA and industry funded grants, Dr. Prasad has been establishing and applying targeted proteomic assays to quantify population variability in drug transporters and metabolizing enzymes. Dr. Prasad has published >60 peer-reviewed articles and >70 conference abstracts and delivered >35 invited talks.

Abstract of the talk

Prediction of age and liver cirrhosis dependent changes in drug transport and non-CYP450 drug metabolism

Accurate prediction of age- and liver cirrhosis-dependent variability in drug disposition and response is important to ensure safety and efficacy of drugs in these special populations. In the absence of clinical data, physiologically-based pharmacokinetic and pharmacodynamic (PBPK/PD) models are emerging promising. To develop such models, we used quantitative proteomics to determine protein abundance of drug metabolizing enzymes and transporters (DMET) in pediatric, cirrhotic and healthy adult liver tissues. These data were then used to develop scaling factors to predict drug disposition in these populations based on healthy adult pharmacokinetics. Age- and liver cirrhosis-dependent quantitative hepatic transporter and non-CYP DME data will be presented with case studies of zidovudine, oseltamivir and morphine PK prediction using PBPK modeling.

John Miners
Distinguished Professor,
Flinders Univeristy, Australia

John Miners holds BSc, MSc, PhD and DSc degrees. He is a Fellow of the Australian Academy of Science (FAA) and an Honorary Fellow of the Royal Society of New Zealand (HonFRSNZ). He is currently a Matthew Flinders Distinguished Professor in the Flinders University School of Medicine, having served previously as Professor and Head of the Department of Clinical Pharmacology. His broad research interest is drug and chemical metabolism in humans, particularly sources of variability in drug elimination and its pharmacokinetic and therapeutic consequences. Current projects include: (i) structure-function relationships and genetic polymorphisms of human UDP-glucuronosyltransferases and cytochromes P450; (ii) *in vitro* and computational models for the prediction of drug and chemical metabolism parameters *in vivo*; (iii) drug-drug interactions; and (iv) metabolomics. He has published over 300 research papers and reviews across these areas and has an ISI h-index of 62. John Miners has served as President of numerous societies, including the Australasian Society of Experimental Pharmacologists and Toxicologists (ASCEPT), the International Society for the Study of Xenobiotics (ISSX) and the Asia Pacific Federation of Pharmacology, as well as being a member of the Executive Committee of the International Union of Basic and Clinical Pharmacology (IUPHAR). He is a current or past member of the Editorial Boards of ten international journals.

Larry Winkers
Vice President,
Global Head of the Department of
Pharmacokinetics and
Drug Metabolism, Amgen Inc

Larry C. Wienkers received his BS in Chemistry and Biology in 1986 and his MS in Chemistry in 1988 from Western Washington University. He then earned his Ph.D. in Medicinal Chemistry from University of Washington in 1993 under the direction of Dr William F. Trager. He did postdoctoral work in the department of Drug Metabolism at the Upjohn Company and subsequently joined the company as a Research Scientist 1995. In 1998, Larry became Director of Drug Metabolism Enabling Technologies at Pharmacia & Upjohn and in 2002 became Executive Director of Pharmacokinetics, Dynamics and Metabolism at Pfizer. In 2004 Larry moved to Amgen and is currently Vice President of the department of Pharmacokinetics and Drug Metabolism. He an American Association of Pharmaceutical Scientists (AAPS) Fellow and serves as a member of the University Of Washington School Of Pharmacy Corporate Advisory Board. One of Larry's current research interests is focused on exploring bioactivation pathways associated with small molecule drug metabolism with particular focus on the prospective application of this information to predict drug-drug interactions in the clinic. To this end, he and his group apply a multidisciplinary approach using organic chemistry, biochemistry and biophysical techniques to study cytochrome P450 mechanism based inhibition and the characterization of biotransformation pathways of novel therapeutics.

Abstract of the talk

Characterizing *in vitro* and *in vivo* stability of bispecifics, fusions, and immunocytokines in a drug discovery setting

Kathie Knights
Professor Emeritus,
Flinders University,
Australia

Kathie Knights held the position of Professor in Clinical Pharmacology at Flinders University, Adelaide, Australia from 2008-2014 and is currently a Professor Emeritus and a member of the Flinders College of Distinguished Educators. Her main research interests are the metabolism of carboxylic acids, renal drug metabolism and the interplay between renal drug and endobiotic metabolism. She has published >160 research outputs, 6 book chapters and is co-author of the highly successful text *Pharmacology for Health Professionals* (Elsevier). She is currently a member of the editorial board of Drug Metabolism Reviews (2012-2018) and the BJCP International Editorial Board (2012-2019).

In 2007 she was awarded an Australian Carrick Citation for outstanding contribution to student learning and in 2010 the Australasian Society of Clinical and Experimental Pharmacologists (ASCEPT) Teaching Excellence Award. Kathie served as ASCEPT President (2008-2009), ISSX Councillor (2008-2011) and is currently an Internal Auditor of IUPHAR (2015-2018).

Abstract of the talk

Renal drug metabolism: Implications for drug and endobiotic metabolic clearance by the kidney

The kidney eliminates a vast array of drugs, nondrug xenobiotics and endogenous compounds. Renal clearance is normally viewed as the net result of glomerular filtration, tubular secretion and reabsorption with a contribution from basolateral and apical renal transporters. There is now considerable evidence that indicates a role for specific CYP and UGT enzymes in renal drug, endo- and xeno-biotic metabolic clearance. Importantly 'drug' metabolizing enzymes modulate intra-renal exposure to xenobiotics and/or their metabolites as well as regulating the activity of physiological mediators.

The contribution of the kidney to drug metabolic clearance is generally predicted to be less than that of liver given the lower organ weight and microsome yield of kidney. However, there is now a wealth of in vitro and in vivo data that indicates that the human kidney has significant drug metabolizing capacity that may in some instances surpass that of liver. This presentation will review current knowledge of renal drug metabolism focusing on renal UGT enzymes and the relationship between drug and endobiotic metabolic clearance.

Deepti Bhandarkar
Sr. Application Chemist,
Shimadzu Analytical,
India

Ms. Deepti Bhandarkar completed her graduation in B.Sc Microbiology from University of Mumbai and her post-graduation in M.Sc Bio-analytical. She has been working with Shimadzu Analytical India for 5 years, with different responsibilities catering to 2 primary techniques: LC and LCMS. Her working areas include application support in various fields like Biopharma, CRO, Food safety etc. She is keenly interested in biopharma industry and development of methodologies catering their workflow.

Abstract of the talk

Size will not matter!

Triple quadrupole has been long established as gold standard for bioanalytical quantitation. Pharma world is progressing from small molecules to large molecules. But basic modus operandi of small molecule quantitation and large molecule quantitation is different. While small molecules progressed from LC based quantitation to LCMS based quantitation, large molecules, essentially being proteinaceous in nature, are traditionally quantitated using ELISA, which is an established gold standard here.

Quantitation of large molecules, especially monoclonal antibody based drugs needs special pretreatment for selective quantitation. Shimadzu with its novel patented technology-nSMOL, provides quick easy access to mAb quantitation for every lab. We introduce solution to challenging application for large molecules, using LCMS-8060 triple quadrupole mass spectrometer. Precise control over collision energy, and ultrafast measurements, are the key to quantitation of large number of MRM transitions for peptide and protein quantitation.

While discussing solutions for quantitation, it's time to realize the large molecule quantitation can be as efficiently achieved as small molecules.

Ashwani Sharma
Biopredic International,
Parc d'affaires de la Bretèche,
France

Currently, I am working as Manager at Biopredic International and Eurosafe, Rennes.

I have 10 years of working experience in the field of Molecular Modeling, Bioinformatics and using in-silico tools for Biology, Chemistry and Drug Discovery applications.

Earlier, I worked as Invited Senior Researcher at Ecole Polytechnique, Paris (July-August, 2016 and Oct-Nov, 2016). Before this EP position, I have worked as Senior Labex aeem Post doc Fellow at University of Paris Diderot-7 and Equipe de Chimie Theorique et Modelisation (CTM), École national supérieure de chimie de Paris (ENSCP), Chemie Paris Tech, Paris in the field of Electrochemistry and Molecular modeling from 1st July, 2015 to 30th June, 2016.

Before that, i have worked as Invited Researcher at Laboratory of Molecular Chemistry (LCM), Ecole Polytechnique, Paris, France from December, 2013 to Nov, 2014. Here, I worked in the field of Computation Bio-molecular chemistry and Molecular dynamics. Along with this, I worked on Computational Aided Drug Designing project. Earlier, i have worked as Post Doc at Computer-Chemie-Center (CCC), Department of Chemistry and Phramacy, University of Erlangen-Nuremberg, Erlangen, Germany (Nov,2011-Oct,2013) in the field of Protein-Ligand Molecular Docking and Dynamics and Drug Discovery with joint collaboration of Sanofi-Aventis Pharma company and P&G Company. I have also worked at DCMR lab of Ecole Polytechnique, Palaiseau, Paris in the field of Quantum Chemistry and Dynamics (Nov, 2010-Oct, 2011) for my Post Doc job. I have completed my Ph.D research work from Indian Institute of Technology Bombay (IIT Bombay), India in the year of 2011, in the field of Bioinformatics with area of specialization of Molecular Docking and Dynamics.

Abstract of the talk

HepaRG® and Silensomes™: *In vitro* tools to study Drug Metabolism and Hepatotoxicity in the Pharmaceutical Industry

Biopredic International (BPI) is a French biotechnology company founded in 1993 and specialized in the business of toxicology and hepatology. Since 1990, Biopredic has been a strong player in primary hepatocyte cryopreservation and the isolation, production and worldwide distribution of fresh and frozen cell lines and high quality Primary Human Hepatocyte (PHH) (Chesne et al., Hepatology 1990, 1991, Toxicology in vitro 1991, Cryobiology 1998). However, due to difficulty to access the whole liver and shortage of good quality of PHH, there was a need to find alternative in vitro model to study Drug Metabolism and Hepatotoxicity. In 1999, HepaRG®cell line (established from the tumor of a female patient suffering from chronic hepatitis C infection and cholangiocarcinoma) was discovered as a potential solution for these studies. In 2007, Biopredic became the licensee of the patent owner INSERM for HepaRG® cell line and settled both the master and the working banks so crucial for preserving the stability of the line until today. The HepaRG® cell line benefits of a huge number of scientific papers (~450), containing an impressive amount of data proving the performance of these cells in the application of drug metabolism and hepatotoxicity. HepaRG® spontaneously differentiates into a co-culture of adult and fully differentiated hepatocytes and of cholangiocyte -like cells. The phenotype of the hepatocytes is similar to the one of the primary hepatocytes (Antherieu et al, Toxicology in vitro 2012) and HepaRG® emerged as a surrogate to PHHs for pre-clinical hepatotoxicity assays (Grime et al., Current Drug Metabolism 2010). HepaRG® is a highly reproducible cell line, without the donor variability seen in PHHs, and as such ensures a consistent and stable assay system (Gunness et al., Toxicology Science 2013). HepaRG® expresses the liver-specific genes like CYP enzymes, transporters proteins at levels similar to PHH and makes the difference with other liver cell lines (Hart et al., DMD 2010) such as HepG2 and HuH7 (Guo et al., DMD 2011). Differentiated HepaRG® cells are highly polarized with substantial levels of uptake and efflux drug transporters, at the same level as the PHH, and form functional bile canaliculi (Cerec et al., Hepatology 2007, Guillouzo et al., Chem Biol Interact., 2007, Le Vee et al., Eur J Pharmaceut Sci 2006, . HepaRG® are used for clearance prediction and compare well with PHH in matter of prediction of the in-vivo intrinsic clearance (Lübberstedt et al., J Pharmacol Toxicol Methods, 2010; Zanelli et al., DMD, 2012). HepaRG®shows high expression of transcription factors and nuclear receptors responsible for regulating drug metabolism enzymes and transporters (e.g. AhR, CAR, PXR, and PPARa), enabling them to respond to prototypical CYP inducers with maximal induction of CYP mRNAs or activities (Kanebratt and Andersson, DMD, 2008). HepaRG® form the metabolites produced in the PHH (e.g. aflatoxin B_1) (Aninat et al., DMD 2006). Cholestatic drugs can be identified in HepaRG® cells by measuring their effect on bile acid efflux (via direct inhibition of the pumps or by interfering with the bile canaliculi contraction (e.g. chlorpromazine) (Antherieu et al, Hepatology 2013). HepaRG® cells have been applied to a number of in-vitro genotoxicity and carcinogenicity assays (Josse et al., Mutagenesis 2012). Annie Borgne sanchez (CEO, Mitologics) has been extensively utilizing the full mitochondrial functions of the HepaRG® to study mitochondrial toxicity (Borgne et al., Toxicology Science 2012) where HepG2 cells are failing. Recently, HepaRG® cell line has been used to make three-dimensional (3D) HepaRG® spheroid model where spheroid cultures use 50-100× fewer cells than conventional 2D cultures, and enable the identification of metabolically activated toxicants (Ferguson et al., Toxicology Science 2017). Transgenic HepaRG® cells have been created to understand their differentiation into mature hepatocyte-like cells using a dual-color reporter for CYP3A4 and CYP3A7 genes (Ueyama et al., Science Report 2017 and Tsuji et al., Plos One 2014). Recently, HepaRG ® cells have been engineered to form HepaRG knocked out for many genes such as the ABCB11 (BSEP) gene and to study in vitro disposition of bile acids (BAs) as well as hepatic transporter function (Qiu et al., Molecular Pharmaceutics 2016)

Silensomes™ is the other BPI *in vitro* tool for direct and quantitative evaluation of the human CYP enzymes contribution (*fm*) to drug clearance in Drug-Drug interaction analysis (Parmentier *et al.*, *Xenobiotica*, 2017). Silensomes™ are validated human pooled liver microsomes (HLMs) in which a single CYP has been chemically and irreversibly inactivated using mechanism based inhibitors (MBI). It is a simple solution for CYP phenotyping and accurate prediction of *in vivo fm* as required by the FDA DDI guidelines (2012).

Keywords: Drug Induced Liver Injury, Drug Metabolism, Hepatotoxicity, HepaRG cell line, Cholestasis, Silensomes™, Drug-Drug interaction

Reference:

1. In Vitro Platforms for Evaluating Liver Toxicity. Bale *et al., Exp Biol Med (Maywood)*, 2014, 239(9): 1180–1191.
2. The HepaRG cell line: a unique in vitro tool for understanding drug metabolism and toxicology in human. Andersson *et al., Expert Opin Drug Metab Toxicol*, 2012, 8(7):909-20.
3. Direct and quantitative evaluation of the human CYP3A4 contribution (f_m) to drug clearance using the *in vitro* silensomes model. Parmentier *et al., Xenobiotica*, 2017, 47(7) : 562-575.

Philip Tagari
Vice President,
Therapeutic Discovery,
Amgen, USA

Philip Tagari is currently Vice President of Research (Therapeutic Discovery) at Amgen Inc, the world's largest biotechnology company. His laboratories are responsible for biologics discovery, scaffold engineering, optimization and early manufacturability assessment; medicinal, oligonucleotide and peptide chemistry; protein conjugates (ADC, peptibodies) and reagents; assay development, screening, enzymological and pharmacological characterization and profiling (in vitro), as well as structural biology, biophysics, computational and analytical chemistry, materials logistics and automation. Prior to joining Amgen in 1998, Philip was a Research Fellow at Merck Frosst (Canada) Inc, where he contributed to several programs in eicosanoid and inflammatory biology, culminating in the discovery of odanacatib and rofecoxib, as well as the clinically active leukotriene D_4 receptor antagonist MK-571 and the leukotriene biosynthesis inhibitor MK-591. In addition to the elucidation of leukotriene metabolism in several species, Philip established one of the first robotic screening laboratories at Merck, building on work performed at McGill and Oxford Universities on automated image analysis, quantitative immunohistochemistry and neurotransmitter measurements in neurodegeneration and cerebrovascular research. Philip is a graduate of Gonville & Caius College, Cambridge University (UK).

Abstract of the talk

Graphene-based sensors for bioanalyte detection & quantitation

Graphene is a comparatively recently discovered form of carbon consisting of a pure 2-dimensional molecular lattice. As such, it is the thinnest and most conductive material known to man. Graphene can be deposited onto silicon wafers to form molecular-scale transistors that can detect mass and charge of analytes non-plasmonically. Such sensors are thus completely insensitive to the limitations seen with optical sensing technologies and can be manufactured using traditional semiconductor technologies. We demonstrate the use of this technology to measure binding of an antibody and a small molecule to an integral membrane solute transporter, a class of proteins of significant interest for ADME applications and previously intractable to label-free technologies.

Utpal Tatu
Professor,
Indian Institute of Science,
Bangalore, India

Dr. Tatu's research focuses on neglected diseases causing organisms with an emphasis on developing better methods of diagnosis and treatment. Prof. Tatu's research in the fields of genomics, proteomics and Metabolomics is internationally recognized. His research work has made it to the editorials of top scientific magazines including Science and Nature Medicine. Prof. Tatu is a member of the United States Pharmacopoeia Expert panel for Biologics. Prof. Tatu has headed several multi-institutional projects with bilateral collaborations with international research laboratories and industries in UK, Switzerland, Denmark, Brazil, France and USA. Prof. Tatu's lab is funded by national and international funding agencies. He is a recipient of several national and international awards such as Ranbaxy Research Award, Birla Science Prize to mention a few. He is endowed with Adjunct professorship to several institutions and health organizations and is on the advisory board of industries in India and abroad. Prof. Tatu is an elected fellow of the National Academy of Sciences and President elect of the Proteomic Society of India. He serves on the editorial board of Cambridge Press journal 'Parasitology' and is one of the few scientists from India to be invited to present his work on Ted Talk.

Abstract of the talk

Exploring heat shock protein 90 as a target for anti-infective drug development

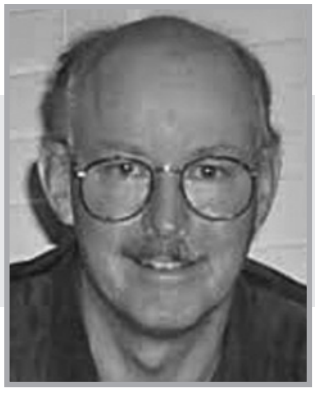
Raju Subramaniam
Director and Head, Pharmacokinetics,
Department of Drug Metabolism and
Pharmacokinetics (DMPK),
Gilead Sciences, USA

Raju Subramanian has over 16 years of experience in DMPK supporting small molecule, peptide and large molecule modalities spanning the discovery and development continuum. He obtained his B. Sc. (Honors), Chemistry, St. Stephen's College, University of Delhi, India 1989; Ph. D., Physical Chemistry, SUNY at Stony Brook, NY 1994; and from 1994-1999 completed post-doctoral training at College of William and Mary (Physics) and University of Illinois at Urbana-Champaign, IL (Electrical and Computer Engineering). Following his post-doctoral training he joined Merck from 1999-2005 where he led a structure elucidation laboratory. He moved to Amgen and over this phase of his career (2005-June 2016), he led a biotransformation/ADME chemistry group and managed broader aspects of ADME science with molecules in discovery through regulatory filing and approval. Externally, Raju has +60 publications, participated in industry consortia and as a speaker, organizer and organizing committees for conferences.

Abstract of the talk

Advances in characterization of the pharmacokinetics and disposition of small molecule drugs

Absorption, distribution, metabolism and excretion (ADME) studies provide a holistic understanding of the pharmacokinetics (PK) and disposition of a therapeutic drug candidate. Regulatory guidance recommend an understanding of the disposition in humans before a large scale trial with emphasis on identification of circulating metabolites and their qualification in nonclinical safety studies. Radiolabel is not required but is beneficial - tools such as nonradioactive liquid chromatography-high resolution mass spectrometric methods are now used to identify circulating metabolites in early clinical studies and prioritize them for subsequent analysis. Advances such as micro-radiotracer dosing with accelerator mass spectrometry enabled detection allows for PK and ADME characterization of drug candidates in settings previously considered difficult or not possible. This presentation will provide an overview of the advances in analytical methods and highlight their use with published case-studies.



Mike Bolger
Chief Scientist,
Simulations Plus,
USA

Professor Bolger is Chief Scientist at Simulations Plus, Inc. in Lancaster, CA. His formal education has been in Biology, Chemistry, Pharmaceutical Sciences, and Pharmacology at UC San Diego and UC San Francisco. He was a professor of Pharmaceutical Sciences for 23 years at the University of Southern California School of Pharmacy and retired from USC in 2004. During this period he developed an interest in the computational aspects of drug design and drug development.

From 1987-1993 he was a founder and Director of Medicinal Chemistry at
CoCensys Inc. There, he studied the chemistry and use of novel neuroactive steroids for treatment of anxiety, epilepsy, and sleep disorders. Drug candidates emanating from seven of Dr. Bolger's patents have been tested in Phase I and II clinical trials for petite mal epilepsy, sleep disorders, and migraine.

Dr. Bolger programmed the first version of GastroPlus in 1997 and for the last 20 years has worked with a team of scientist / programmers at Simulations Plus, Inc. (Lancaster, CA) in the development of software programs for estimation of biopharmaceutical properties and simulations of absorption and bioavailability. He was elected to the rank of Fellow of the American Association for the Advancement of Science in 1996.

Abstract of the talk

Cellular simulation for suspended and sandwich hepatocytes applied to human predictions

Purpose: The purpose of this study was to develop a fully mechanistic simulation of drug transport in both sandwich and suspended hepatocytes using a simulation software program (MembranePlus™, Simulations Plus, Inc.) that allows for simultaneous fitting of Km and Vmax values for metabolic enzymes as well as, Km and Vmax values for active influx/efflux transporters.

Methods: A simulation model for suspended and sandwich culture hepatocytes was developed by utilizing the existing framework in MembranePlus that was established for PAMPA and Caco-2 cellular simulations. The model includes the simulation of diffusion in the unstirred boundary layer, protein binding in culture media and cellular cytosol, drug partitioning into lipid bilayers, active and passive transport across the basolateral membrane and the bile canaliculus, and lysosomal trapping. Several case studies will be presented to demonstrate the simulation of active uptake and biliary excretion of bile salt and statin model compounds.

Results: The model accurately described the mean in vitro concentrations from cell culture experiments for a bile salt and a statin. Using the combination of MembranePlus™ and GastroPlus™ along with the in vitro and in vivo metabolism of a non-linear CYP2D6 substrate (propafenone) the human plasma concentration vs. time was accurately simulated.

Conclusion: This work described the use of mechanistic cellular simulations linked to whole body PBPK simulations for substrates of metabolic enzymes and influx and efflux transporters. The case studies illustrate that these cellular simulations are a valuable automated tool to aid in the extraction of non-linear metabolism and transport kinetics for further usage use in mechanistic oral absorption / physiologically based pharmacokinetic MAM/PBPK modelling.

Amita Joshi
Vice President and Head,
Clinical Pharmacology,
Genentech, USA

Amita Joshi, PhD, is an Executive Leader with 27 years experience in Clinical Drug Development and Clinical Pharmacology. In her role as Vice President and Head of Clinical Pharmacology at Genentech, Amita is responsible for the strategic and scientific direction of Clinical Pharmacology and Pharmacometric aspects of all Genentech programs from Early Clinical Research through post-approval. The scientific staff of Clinical Pharmacology is responsible for ensuring that they identify and deliver the right dose, by the right route and by the right regimen to patients. In her role, Amita has overseen successful development strategies and tactics for first product approvals and/or product extensions for Small molecules, Monoclonal Antibodies and Antibody Drug Conjugates. These include Cotellic, Tecentriq, Kadcyla, Lucentis, Rituxan, Erivedge, Perjeta, Herceptin, Avastin, Xolair and Nutropin. Amita has published more than 40 manuscripts and book chapters and presented at national and international conferences.

Amita is also recognized by her peers in the Biotech industry for her contributions in understanding the Clinical Pharmacology of therapeutic monoclonal antibodies, has been a member of the AAPS and ASCPT scientific leadership and has chaired AAPS's National Biotechnology Conference in 2010. Amita received the Professional Businesswoman "Industry Leader" award in 2015 and received the San Francisco Business Times "100 Most Influential Women in Business" award in 2016.

Abstract of the talk

Paving the way for personalized medicine - opportunities for clinical pharmacology

Narendra Chirmule
Head, R&D,
Biocon, India

Narendra Chirmule*, PhD* is Senior Vice President, and Head of R&D at Biocon Research Labs, Bangalore, India. He has responsible for the development of the pipeline of novel biologics and biosimilars. The role includes oversight of early and late translational sciences, regulatory strategies and intellectual property. He has held senior leadership positions at Amgen (Thousand Oaks, CA) and Merck (Philadelphia, PA) in the departments of Clinical Immunology overseeing drug development in regulated laboratories. His expertise is in the area of immune responses to biologics and vaccines. In Biologics, he has published extensively on the topics of immunogenicity prediction and assessment, predictive toxicology and quality-by-design. In vaccines, his experience spans development of assays for various viruses and bacteria to supporting operations of very large clinical trials. He was involved in the immunogenicity assessment of HPV responses for Gardasil. He has a PhD from University of Mumbai, post-doctoral training at Cornell University Medical College, and teaching and research experience as Assistant Professor in the Human Gene Therapy Group of University of Pennsylvania. He is an advisor to the Filovirus consortium and a reviewer on the HIV vaccine study section for the National Institutes of Health.

In culture, he has a keen interest in Indian Classical music that extends to all genres. In this respect, he has learned and played the tabla from many gurus, including Debasish Chaudhuri of Lucknow Gharana. He has performed with a wide range of amateur and professional artists ranging from Hindustani and Carnatic classical, Bollywood, Jazz, Latin, and Fusion. He is on the board of Rupak School of World Music, Los Angeles. In Bangalore, he volunteers his time towards the promotion of music to underprivileged children.

Abstract of the talk

Pharmacokinetics and pharmacodynamics of biologics: Technical and regulatory considerations.

Nilanjan Sengupta, Anita Rao, Narendra Chirmule, Ramakrishnan Melarkode.

Pharmacokinetics (PK) and pharmacodynamics (PD) aspects of biologic drugs is a critical component of drug development. Novel technologies and modeling methodologies have resulted in a better understanding of the relationship between PK and PD. The nature of non-stoichiometric interactions of ligands with soluble and cell-surface bound receptors, and understanding the characteristics of anti-drug antibodies (ADA), require sophisticated analytical tools that are sensitive, accurate and precise. A detailed work flow for development of these ligand binding assays which captures comprehensively the requirements for detection of drug and ADA in the relevant therapeutic setting will enable a clear interpretation of the PK, PD and immunogenicity results. In this presentation, we will highlight the challenges in i) development, qualification and validation of these assays, ii) understanding the mechanisms PK-PD relationships and iii) developing models for data analysis and interpretation of the results obtained. The analysis of these processes can influence a discussion on the central role PK and PD plays during biologics drug development.

Chandrasekhar Natrajan
Tufts University,
USA

Mr. Chandrasekhar (Nat) Natarajan is the Chief Scientific Officer at ViNa Pharma Consulting, LLC. He is also an Adjunct Faculty at Tufts School of Medicine teaching "Translational Medicine". At Sanofi he was an Associate Vice President in Drug Disposition division and a Strategic Scientific Advisor for Oncology programs providing critical input on translational pharmacology, ADME and PK/PD in discovery and development. Earlier he was the Deputy Global Head of Discovery MPK providing Discovery Support at Sanofi-Aventis overseeing effective chemical optimization and druggability of new compounds for desirable ADME & pharmacokinetic properties.

Nat received his B.Pharm and his MSc in Pharmacology from the University of Madras, India. His MS in Pharmacotherapeutics was from Long Island University, New York and his clinical pharmacy training was at Down State Medical Center in New York.

Nat is an invited speaker at PERI, Harvard, Tufts CSDD, CSIR-Institute of Genomics & Integrative Biology (CSIR-IGIB), and Gulf Medical University. Nat is an active member of AAPS, ACCP, SITC and DIA.

Abstract of the talk

Translational Medicine, from discovery to First in Human (FIH) and Phase I/II clinical studies

This comprehensive presentation covers key processes from drug Discovery to Development, including the progression and translation of scientific information through different development stages and the transition to clinical studies. It also highlights the importance of establishing relevant biomarkers that reflect the pathology and therapeutic effect and PK/PD modelling using exposure parameters. Each participant will have an overall understanding of Translational Pharmacology that is integral to scientific rationale in Drug Research and Development.

- Discovery & Early development Process.
- Establishing Biomarker – Model relevance to Pathology and Therapeutic effect
- Exposure parameters that dictate PK-PD and PK-TD relationship
- Modeling efforts to understand exposure-effect (PK-PD) relationship
- FIH Dose selection
- Therapeutic window – Predicting Clinical Success and/cutting your losses early enough

Michael Sinz
Director,
Biocon-Bristol-Myers Squibb

Michael W. Sinz, Ph.D. is a Director at Biocon-Bristol-Myers Squibb, Department of Metabolism and Pharmacokinetics in Bangalore, India where he manages ADME lead optimization of drug discovery assets (2001-present). Dr. Sinz previously held the position of Section Director-Pharmacokinetics and Drug Metabolism for Parke-Davis/Pfizer Global Research and Development (Ann Arbor, MI, 1991-2001). He received two B.S. degrees, one in Chemistry (ACS) and one in Biology from the University of Wisconsin-Eau Claire, and a Ph.D. in Pharmacognosy/Medicinal Chemistry from the University of Minnesota. Dr. Sinz is an active member of the International Society for the Study of Xenobiotics, IQ Consortium (Drug Metabolism), and American Chemical Society. He is also on the organizing committee for the Land O'Lakes conference on Drug Metabolism and Pharmacokinetics (2004-present), as well as the 2018 Asia Pacific ISSX meeting. He is editor in chief of Current Drug Metabolism, an associate editor of Drug Metabolism Letters, and on the editorial advisory boards of Current Pharmacology Reports and Drug Metabolism and Disposition. His areas of research include in vitro and in vivo drug metabolism, clearance predictions, reaction phenotyping, and predicting drug-drug interactions. Dr. Sinz's resume includes an extensive number of peer reviewed publications, book chapters and external presentations.

Joyce S Macwan
Senior Scientist,
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Dr. Macwan is currently Senior Scientist at Simulations Plus, Inc. in Lancaster, CA. She earned her B.S. and M.S. degrees in Pharmaceutical Sciences at Sardar Patel University, Gujarat, India. She also served as the lecturer for a year at Sat Kaival College of Pharmacy, Gujarat, India.

Dr. Macwan was enrolled in University of Rhode Island, RI, USA as the graduate assistant to pursue her research interest in clinical pharmacokinetics in August 2007. She was honored with graduate student research excellence award in 2012. Dr Macwan earned PhD in pharmaceutical sciences specializing in clinical pharmacokinetic in May 2013. The major focus of her PhD research was to characterize the effect of diseases on pharmacokinetic properties of a lipid-lowering class of drugs through model-based approaches. She used GastroPlusTM software for one of her research projects and during that period she developed the keen interest in physiologically-based pharmacokinetic (PBPK) modeling. Dr Macwan joined Simulations Plus, Inc. as scientist II in July 2013 to further nourish her interest in mechanistic modeling. She has been working on several contract studies in simulation studies team for guiding teams of pharmaceutical scientists to improve decision making-process during drug discovery and various stages of drug development. This work is mainly facilitated by mechanistic absorption and PBPK modeling approach using GastroPlus, which is the company's leading modeling and simulation software.

Dr Macwan demonstrated her proficiency in this field by presenting work at various regional and international conferences either through poster or oral presentations.

Selected Abstracts

Young Scientist Presentations

P-01

Development of a comprehensive open-access data module of ADME QPrOmics™ database comprising of abundance and activity of drug-metabolizing enzymes and transporters in humans: Effect of age, gender, genotype, ethnicity, and health condition

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The disposition of xenobiotics is significantly regulated by several drug metabolizing enzymes and transporter (DMET) proteins. In order to predict the impact of DMET proteins on oral drug absorption, distribution, metabolism and excretion (ADME), quantitative information on expression in different tissues is a necessity. The aim of the project was to create an open access repository of quantitative information on proteins, mRNA, and activity data of drug metabolizing enzymes, transporters and transcriptional factors, which are associated with ADME in human and animal organs. The quantitative data was uploaded as a data module to an online public access ADME QPrOmics™ database (http://qpromics.uw.edu/qpromics/data/), which is being developed by University of Washington, Seattle, USA in collaboration with NIPER, S.A.S. Nagar, Punjab, India. The database is an open access resource for systems pharmacologists and pharmacometricians in the drug development industry, regulatory and academia. The paramount information in data module includes reported quantitative data on abundance, mRNA expression, and activity of DMET proteins in different species (mouse, rat, dog, monkey, and human), tissues (liver, intestine, kidney, brain, and lung) with their associated variability. In addition, the data module provides available information on the impact of demographic variables, like age (neonates to adults), sex, ethnicity, genotype, health condition, smoking, alcohol consumption, and medication on the expression of DMET. These data are critical for developing physiologically based pharmacokinetic and pharmacodynamic (PBPK/PD) models, so that prediction of interindividual variability in drug disposition and response is possible.

P-02

Perturbation of arachidonic acid metabolism by dronedarone and amiodarone: Implications in cardiac safety?

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Objective: Cardiac enzymes such as cytochrome P450 2J2 (CYP2J2) metabolize arachidonic acid (AA) to cardioprotective epoxyeicosatrienoic acids (EETs), which in turn are metabolized by soluble epoxide hydrolase (sEH) to dihydroxyeicosatrienoic acids (DHETs).[1] As EETs and less potent DHETs exhibit cardioprotective and vasoprotective functions, optimum levels of cardiac EETs are paramount in cardiac homeostasis. Dronedarone, a non-iodinated benzofuran analogue of amiodarone causes cardiac failure exacerbation and increases cardiovascular mortality while amiodarone fails to do so.[2] Despite the structural and pharmacological similarities between the two drugs, exact cause for these toxicological differences is currently unknown. We postulate that the modulation of AA metabolism by dronedarone and amiodarone may explain the differences.

Methods: In this project, firstly we examined whether dronedarone, amiodarone and their active metabolites, N-desbutyldronedarone (NDBD) and N-desethylamiodarone (NDEA) respectively, inhibit recombinant CYP2J2 using arachidonic acid as substrate. Secondly, we tested the inhibitory effect on recombinant human sEH (rHsEH) using 14,15-EET as probe substrate. Finally, we developed a sequential metabolism model to predict the effect of sEH and CYP2J2 dual inhibition on the fold-change in 14,15-EET level ().

Results: We found that dronedarone (K_i=3.25 μM), amiodarone (K_i=5.48 μM), NDBD (K_i=1.39 μM) inhibit rCYP2J2-mediated arachidonic acid metabolism. Moreover, we found that dronedarone (K_i=5.10 μM), amiodarone (K_i= 13.08 μM), NDBD (K_i=2.04 μM) and NDEA (K_i=1.88 μM) also inhibit rHsEH. The sequential metabolism model predicted that dronedarone (=0.85), NDBD (=0.75) and amiodarone (=0.48) leads to a decrease in overall 14,15-EET level while NDEA (>35.5) would significantly elevate intracardiac 14,15-EET.

Conclusion: We demonstrate for the first time that dronedarone, amiodarone and NDBD inhibit CYP2J2-mediated metabolism of AA to EETs potently. Additionally, we report the novel inhibition of sEH-mediated metabolism of 14,15-EET to 14,15-DHET by dronedarone, amiodarone, NDBD and NDEA. Based on the dual inhibition of human CYP2J2 and sEH, we predicted a net decrease in the cardiac concentrations of 14,15-EET at steady-state in the presence of dronedarone, amiodarone and NDBD but a net increase in the presence of NDEA.

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P-03

Lipid carriers loaded with antimicrobial agents for the prevention of ventilator acquired pneumonia (vap)

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Ventatlitor acquired pneumonia (VAP) is the 2nd most common nosocomial infection (15%) of all hospital acquired infections. Pneumonia rates are higher in mechanically ventilated patients due to the artificial airway, increasing the opportunity of aspiration and colonization. On-going research is needed to develop a formulation containing an effective anti-microbial agent as part of oral care intervention which can aid in prevention of VAP in patients with mechanical ventilation. Hence, we aimed at development of in-situ mucoadhesive buccal liposome gels loaded with anti-microbial agents (levofloxacin and meropenem) and used gamma irradiation to obtain sterile in-situ liposomal gels to be used in critically ill patients on mechanical ventilator.

Liposomes were formulated adopting thin film hydration method using Phospholipon90H, Cholesterol and Poloxamer, loaded with Levofloxacin(LH) in lipid layer and Meropenem(MP) in aqueous medium. Optimised formula showed 72.06 % (MP) and 63.45% (MP) of drug loading , with mean particle size 224±3.3 nm and PDI <0.3, and zeta potential value > ±30mv. TEM analysis further confirmed the particle size. In-vitro release study using pig buccal membrane showed a controlled of 78.22% cumulative drug release during 24 hours study. Anti-microbial activity of the drugs were retained after loading into liposomes, zone of inhibition studies against P.aeruginosa showed a slight enhancement (10%) of antimicrobial activity compared to the pure drugs. After gamma irradiation, formulations were used re-evaluated for physic-chemical properties and it was seen there was no significant change in the physico-chemical property of formulations. In situ liposomal gels loaded with combination of antimicrobials were successfully formulated and sterilised and will be taken up for clinical evaluation.

P-04

Interaction study between the hepatic transport proteins organic anion-transporting protein 1B1 (OATP1B1) and its Inhibitors via Molecular Docking: an insight for drug-induced cholestasis

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OATP1B1 and 1B3, localized on the sinusoidal membrane of Hepatocytes, are major hepatic uptake transporters that facilitate the efficient cross membrane transport of drugs or Xenobiotics. OATP1B1 can transport a wide variety of drugs, including many statins (e.g., Atorvastatin, Rosuvastatin, Pravastatin and Pitavastatin) and some oral antibiotics (Repaglinide, Troglitazone) (1-3). The substrate specificity of OATP1B3 commonly overlaps that of OATP1B1, but there are some differences as far as substrate recognition and infinity. Clinical evidence has demonstrated that inhibition of OATP mediated uptake can lead to significant alteration of the pharmacokinetic profiles of the "victim" drug, resulting in toxicity; e.g., the immunosuppressive drug, cyclosporine A. Therefore, it is very important to understand the mechanism of inhibition of OATP1B1 and the protein-inhibitor interaction. However, there is no experimental structure for OATP1B1 transporter protein. Therefore due to lack in experimental structure of OATP1B1 transporter protein, we derived 3D structure using the homology modeling method and further studied its interactions with the potential inhibitors as reported in the FDA guidelines.

Here our goal is to understand the inhibitor - OATP1B1 transporter protein interactions which may help better understanding for mechanism of inhibition of OATP1B1 transporter protein during cholestasis disease conditions. The protein sequence of OATP1B1 transporter protein (UniProtKB - Q9Y6L6 (SO1B1_HUMAN)) was submitted to homology modeling via Modbase. The model was generated using the template 1LDT with sequence identity of 42 %. Further, the model was verified by Ramachandran plot as good model with 96 % of the amino acid residues in good regions. Further, the model was minimized by GROMOS force field implemented in Swiss model. Patchdock script was used to perform molecular docking between 3D model of OATP1B1 transporter protein and three FDA approved inhibitors: Cyclosporine A, Rifampicin and Simvastatin. Docking revealed that OATP1B1 has large affinity for Cyclosporine A: 9856 > Rifampicin: 6634 > Simvastatin: 4504 (affinity trends based on docking score). OATP1B1- Cyclosporine A complex analysis shows that inhibitor binds at the binding pocket with making hydrogen bonds with amino acid residues of GLN469, SER468, CYS465 and GLN 513.Therefore, we predicted that these residues play major role in mechanism of inhibition of OATP1B1 transporter protein. Therefore, our work may help to understand the mechanism of OATP1B1 transporter protein inhibition during cholestasis conditions.

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P-05

Pharmacokinetics and brain uptake study of novel AMPA receptor antagonist using a validated UHPLC-QTOF-MS method

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A highly sensitive and rapid UHPLC–QTOF-MS method was established for the determination of a novel AMPA receptor antagonist perampanel (PER) in rat plasma and brain homogenate with alogliptin as internal standard (IS). Chromatographic separation was carried out on an Acquity UPLC HSS Cyano column (100 mm x 2.1 mm, 1.8 µm) using gradient mobile phase system consisting of 0.1% formic acid and acetonitrile with a flow rate of 0. 4 mL/min within a run time of 4 min. Sample preparation was based on simple protein precipitation and dsetection of target ions $[M+H]^+$ at m/z 350.1288 for PER and m/z 340.1779 for IS was performed in positive ionization mode using extracted ion chromatography. The developed analytical method meets the US-FDA and EMA bioanalytical guidelines. The method exhibited linearity over a wide concentration range of 0.4 - 400 ng/mL in both the bio-matrixes and found to be precise, accurate, selective and rugged. The validated method was successfully applied to pharmacokinetics and brain uptake study in SD rats. The study results showed PER has penetrated the blood-brain barrier, brain to plasma ratio (Kp) was found to be 0.62 ± 0.50 and its rapidly eliminated from the brain. Additionally, the developed analytical method can be applied to clinical samples and forensic analysis of PER.

Keywords: Perampanel, Alogliptin, Protein precipitation, UHPLC–QTOF-MS.

P-06

Dispersive liquid-liquid microextraction combined with UPLC-fluorescence as an efficient and sensitive method for quantification of benzanthrone in urine samples

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Benzanthrone (BNZ) is an aromatic hydro carbon derivative used as a dye stuff intermediate for anthraquinones-based dyes. BNZ is believed to be one of the most significant and ubiquitous environmental pollutant produced by natural sources like geochemical formation of fossil fuels such as coal and crude oil deposition as well as anthropogenic sources which include power plants, water, incineration domestic heating and emissions of heavy duty diesel engines, passenger car, wood and coal smoke. Human might be exposed to BNZ through the skin route, inhalation and ingestion absorption. BNZ has been reported to cause itching, burning in the mouth, throat and chest, precocious generalized eczema, photosensitization, pigmentation, liver damage, impairment in the nervous system, upper respiratory tract irritation and increased risk of lung cancer. In the present study, we have developed and validated a very simple, sensitive and high throughput UPLC-fluorescence method for quantification of benzanthrone in urine, using dispersive liquid-liquid microextraction method (DLLME). DLLME have several advantages over liquid-liquid extraction, solid phase extraction , and solid phase microextraction, as it is cost effective, rapid, eco-friendly and economical. DLLME is capable of achieving high recoveries and high enrichment factors which can increase the sensitivity of the method. Some important parameters of DLLME sample preparation technique such as extraction solvents and disperse solvents and volume of them, pH and ionic strength were optimized. At optimum conditions, values of variables set as 150 µL chloroform, 300 µL acetone, pH 6 and no significant effect of ionic strength was reported. Benzanthrone and Napthalene (IS) were separated on a phenomenex C18 column under isocratic condition with mobile phase consisting of acetonitrile and acetic acid buffer (0.2) in the ratio of 60:40(v/v) at a flow rate of 0.6 mL/min. The retention time of analyte and IS were 2.9 min and 3.5 min, respectively. The method was accurate and precise within the linearity range 2.5-500 ng/mL for benzanthrone with a correlation coefficient (r2) of ≥0.997. The intra- and inter- day assay precision ranged from 0.99-13.47 and 1.76-8.74, and intra- and inter-day assay accuracy was between 87.08-111.65 and 88.33-100.00%, respectively, In the conclusion, we have successfully developed and validated the UPLC- fluorescence method for BNZ quantification in urine samples, The developed method can be utilized for toxicokinetic and biomonitoring studies of BNZ.

P-07

Isolation, characterization and *in vitro* evaluation of anti-cataract potential of fucoidan from *Sargassum Wightii Greville*

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The present study was designed to isolate and characterize fucoidan from *Sargassum wightii Greville* and to evaluate its in-vitro anti cataract potential against galactose induced cataract in isolated goat lenses. Fucoidan has recently been reported to possess a wide range of biological activities including antioxidant, antidiabetic, antihyperlipidemic, neuroprotective, anticancer, fibrinolytic, anti-viral and anti-bacterial properties. Studies have also indicated that it is non-toxic, non-allergenic and widely available from marine sources. A substance or a compound having antioxidant property is expected to be useful in the treatment of cataract by combating oxidative stress.

Fucoidan was isolated from *S. Wightii Greville* and its sulfate content was estimated following barium chloride method. HPTLC studies of isolated fucoidan was carried out using D-glucose as standard. The lenses were divided into five groups as follows- Group-I (Normal control- Vehicle treated), Group-II (Disease control- Galactose treated 55mM), Group-III (Galactose 55mM+ Standard-Ascorbic acid 20 µg/ml), Group-IV (Galactose 55 mM+Fucoidan-20 µg/ml) and Group-V (Galactose 55mM+ Fucoidan 40 µg/ml) and incubated for 72hrs respectively and subjected to biochemical estimations. The opacity of lenses was also noted prior to biochemical estimation.

The percentage yield of isolated fucoidan was found to be 0.65%. The extent of sulfation in the isolated fucoidan was found to be as high as 53% and correlated to its potency. Galactose induced cataractous lenses showed significant oxidative stress when compared to normal lenses whereas treatment with fucoidan 20 and 40 µg/ml significantly combated oxidative stress and prevented the development of cataract when compared to cataractous lenses. The results obtained with the treatment of fucoidan was dose dependant and comparable to standard.

P-08

Studies on phytoenzymes as tools for metabolism based phytoremediation

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Plants are known for their ability to produce a variety of secondary metabolites (alkaloids, tannins, terpenes, glycosides, steroids, etc.). In addition, plants also metabolize many exogenous chemicals (xenobiotics) such as environmental pollutants, pesticides including polycyclic chlorinated hydrocarbons, insecticides, etc. By virtue of this ability, plants can act as a global sink for environmental pollutants and have been referred to as 'Green Livers'. We have explored this capacity through studies on the plant's potential to perform phytoremediation via metabolism.

Plant esterases were isolated using differential centrifugation method from 30 different plant samples including seeds, leaves, fruits, rhizome and bark. Protein content of isolated fractions from all the samples was determined by Biuret method. Isolated enzyme fraction was incubated with p-nitrophenyl acetate (at a final concentration of 1 mM) in 0.1 M sodium phosphate buffer, containing 0.1 mM EDTA (pH 6.5) at ambient temperature. The time course of formation of p-nitrophenol was monitored by UV spectrophotometry, by following the increase in absorbance at 400 nm for 10 minutes. The assay was optimized for different pH and volumes of enzyme fraction. Percentage conversion of p-nitrophenyl acetate to p-nitrophenol was calculated for each plant sample (normalized to mg of protein). The esterase activity levels varied by close to 50-fold in 30 different samples. It was highest (3.5% conversion) in *Trigonella foenum graecum* and was lowest (0.06% conversion) in *Cinnamomum verum*. The data suggest that esterase mediated xenobiotic potential is seen in all plants evaluated albeit to different extents. Thus, most plants probably possess an ability for metabolism based phytoremediation of ester xenobiotics.

Keywords: Phytoremediation, esterases, green liver, xenobiotic metabolism.

Poster Abstracts

P-09

A meta-analysis of sex differentiation in drug metabolising enzymes and transporters

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Expression of drug metabolizing enzymes and transporters (DMET) protein varies due to gender differences in human and will show alterations in drug absorption, distribution, metabolism and excretion (ADME), possibly affecting drug safety and efficacy or may lead to adverse reactions. The sexually dimorphic expression of DME and other liver-expressed genes is regulated by the temporal pattern of plasma growth hormone (GH) release by the pituitary gland, which shows significant sex differences. In this study, different enzymes and transporters were taken, the effect of gender on their expression level was evaluated. DMET protein expression data were obtained from data module of ADME QPrOmics™ databse (http://qpromics.uw.edu/qpromics/data/) that are mostly measured by Western blotting or LC-MS/MS based proteomic methodologies. Methodological variability were observed in the DMEPT protein expression levels. To overcome these variabilities and understand the role of gender, a meta-analysis was performed. Also, relative expression level of DMET protein in different tissue was evaluated. The relative expression level of DMET proteins for the male was set as “1,” and the values from female for the same protein were compared. Also corrected expression level of protein was estimated by considering the number of samples and total population samples. Effect of gender dependent protein expression data analyses was evaluated using the Mann-Whitney rank order U test, with P values <0.05 were considered to be significant. The obtained information is expected to be helpful to predict gender dependent PK using PBPK modelling via in vitro to in vivo extrapolation (IVIVE) studies.

P-10

A meta-analysis of ontogeny of drug metabolizing enzymes and transporters

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The pharmacokinetic variability of a drug within and between individuals can be attributed to different factors such as age, genetic polymorphism, epigenetic influence, ethnicity, health conditions, etc. Prediction of consequences of such variations is essential to avoid toxicity, especially in case of narrow therapeutic drugs. In this context, age significantly influences drug metabolizing enzyme and transporter (DMET) proteins. By studying the ontogeny of these DMET proteins, the role of age on drug disposition can be understood and integration of these data in physiologically based pharmacokinetic (PBPK) models can help in prediction of drug disposition. Influence of age on non-cytochrome P450 (CYP) enzymes is scant and thus data of some non-CYP enzymes and transporters were collected for this study. Protein expression of DMET data was obtained from data module of ADME QPrOmics™ database (http://qpromics.uw.edu/qpromics/data/) that were mostly measured by Western blotting and LC-MS/MS-based proteomic methodologies. Methodological variability was observed in the DMET protein expression levels. To overcome these variabilities and understand the role of age, a meta-analysis was performed. The relative expression level of DMET proteins in different tissues for adult (>18 years) was set as "1", and the values from other age tissues like adolescents (12 to 18 years), middle childhood (6 to 12 years), early childhood (1 to 6 years), infants (28 to 364 days), neonates (0 to 27 days), and fetal (before birth) for the same protein were compared. Further, corrected expression level of same protein was estimated. For age dependent (adults to fetal) protein expression, data analysis was done using Kruskal Wallis test followed by Dunn's multiple comparison test, and P values <0.05 were considered to be significant. Information obtained can be further used in PBPK modeling via in vitro to in vivo extrapolation (IVIVE) to predict the disposition of drug in paediatrics.

P-11

A meta-analysis of ethnicity-dependant expression of drug metabolizing enzymes and transporters

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Ethnicity is one factor that may account for the observed differences in both pharmacokinetics (PK) and pharmacodynamics (PD) of drugs, resulting in variability in response to drug therapy. The FDA recognizes that standard methods of defining racial and ethnic subgroups are necessary to ensure consistency in safety and effectiveness. Drug-metabolizing enzymes and transporters (DMET) proteins work in concert to play crucial roles in drug absorption, distribution, and elimination (ADME). It is well recognized that genetic variation in DMET proteins contributes substantially to interindividual differences in drug disposition and response. Ethnicity-related differences in allele frequencies exist in the population and allele frequencies describe the functional activities. Allelic differences caused by functional activities of nonsynonymous variants that differ in their allele frequencies among various ethnic groups. Therefore, the aim of the present study was to calculate the relative expression level of ADME protein in various tissues and for that the value for the Caucasian population was set as "1," and the values for the same protein in other ethnic groups were compared. Mann-Whitney rank order U test was employed to study effect of ethnicity dependent protein expression and values with $P < 0.05$ were considered to be significant. Also corrected expression level of proteins were calculated using number of sample and total population sample. Quantitative protein expression data of DMET in human were collected from data module, which are compiled in a publicly open accessible database (ADME QPrOmics; http://qpromics.uw.edu/qpromics/data/).

P-12

A meta-analysis of genetic polymorphism of drug metabolizing enzymes and transporters

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The disposition of xenobiotics is significantly influenced by several drug metabolizing enzymes and drug transporters (DMET). Drug metabolizing enzymes (DME) are a diverse group of proteins that are responsible for metabolizing drugs and drug transporters (DT) are the proteins that help in movement of drug between different membranes and thereby help in its activity. Genetic polymorphism can either affect protein expression, substrate affinity (Km) or protein localization, thereby affecting the activity of enzyme leading to changes in pharmacokinetic parameters of the drug. In this study, we took different enzymes such as CYP2D6, CYP2C19 and OATP1B1 transporters to evaluate the effect of genetic polymorphism on their expression level. Protein expression data were obtained from the data module of ADME QPrOmics™ (http://qpromics.uw.edu/qpromics/data/) that were mostly measured using Western blotting and LC-MS/MS-based proteomic methodologies. Methodological variability were observed in DMET protein expression. To overcome these variability and to understand role of genotype in disposition of the drug, this meta-analysis study was performed. The relative expression level of protein for the wild type alleles was set as "1," and the values from other mutant type alleles for the same protein was compared. The corrected expression level of protein was also calculated by taking the number of samples and total population into consideration. Effect of genotype dependent protein expression data analyses was evaluated using the Mann-Whitney rank order U test, with P values <0.05 considered to be significant. Information obtained will be discussed, which can be further helpful to predict interindividual variability in response of a drug.

P-13

Expression of hepatic drug-metabolizing enzymes and transporters in healthy and diseased conditions: a meta-analysis

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Inter-individual variability in the expression of drug-metabolizing enzymes and transporters (DMET) in human liver may contribute to inter-individual differences in drug efficacy and adverse reactions. There is growing evidence to suggest that many hepatic, renal and systemic diseases can affect drug metabolism and disposition by regulating the expression and/or activity of DMEs and transporters in the liver. Understanding the disease effect on DMET is clinically important due to the concern of disease-drug interactions. Protein expression of DMET data was obtained from data module of ADME QPrOmics™ database (http://qpromics.uw.edu/qpromics/data/). Variations in the abundance and activity of proteins in diseased conditions with respect to healthy state were seen in literature. To understand the role of health conditions in disposition of drug, a meta-analysis study was performed. The relative expression level of protein for the healthy condition was set as "1," and the values for same protein in the diseased condition was compared. The corrected expression level of protein was also calculated by taking the number of samples and total population into consideration. Effect of health conditions on the protein expression data analyses were evaluated using the Mann-Whitney rank order U test, with P values <0.05 considered to be significant. Information obtained can be further helpful to predict inter-individual variability in response to drug in various health conditions.

P-14

In vitro characterization of stable and reactive metabolites of sunitinib in mouse, rat, dog, monkey and human liver microsomes using high performance liquid chromatography with linear trap quadrupole mass spectrometry

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Sunitinib, N-(2-diethylaminoethyl)-5-[(Z)-(5-fluoro-2-oxo-1H-indol-3-ylidene) methyl]-2,4-dimethyl-1H-pyrrole-3-carboxamide, is a receptor tyrosine kinase inhibitor used to treat malignancies. It has been known to cause idiosyncratic toxicities, which may be attributed to the presence of 5-fluoro-2-oxo-indol-3-ylidene moiety in the structure undergoing bioactivation. Moreover, the structure of sunitinib is very similar to famitinib that has been reported to form reactive metabolites detected by adduct formation with glutathione. The present work focused upon the comprehensive study of sunitinib metabolism. Stable and reactive metabolites were ascertained on examining in vitro metabolism in different liver fractions such as mouse, rat, dog, monkey and human liver microsomes. The contemplated bioactivation potential of sunitinib was examined by use of trapping agent (N-acetylcysteine), in which a reactive metabolite of m/z 558 was detected. The present study provided new insights on the formation of P450 mediated reactive metabolite of sunitinib, and the in vitro metabolic process that had not been identify previous. The proposed pathway for the bioctivation to NAC conjugate was also proposed. Furthermore, chemical inhibition study of CYP3A by this reactive metabolite was carried out and it was found that ketoconazole (CYP3A inhibitor) suppressed the bioactivation of sunitinib, and thus, the CYP3A4 sub family was the primary enzyme responsible for its metabolic activation to form the NAC conjugate. Thus, this study serves as a useful resource to support further research on sunitinib to allow better understanding of the metabolic activation of the drug.

P-15

In silico, *in vitro* and *in vivo* metabolite profiling of labetalol using hyphenated mass tools

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Labetalol, an antihypertensive agent, commonly used to treat pre-eclampsia, is reported to be metabolized into multiple biotransformation products, with a variable profile in mice, rats, dogs, monkeys, and humans. Only some of these have been characterized using age old techniques and others were undefined. Therefore, the motive of the current study was to bridge this space by employing modern in silico tools integrated with advanced liquid chromatography-mass spectrometry (LC-MS) techniques. Initially, a comprehensive mass fragmentation pattern for labetalol was established using the data obtained from multiple-stage MS (MS^n) and QTOF MS experiments. The metabolites were then anticipated in silico by using software such as MetaSite, Xenosite, MetaPrint2D, and SMARTCyp. The in silico results were corroborated through in vitro and in vivo studies. In vitro studies were carried out by incubating the drug with a matrix such as mouse, rat, dog, monkey and human liver microsomes, and rat hepatocytes so as to generate the metabolites, whereas in vivo studies were performed by the administration of 50 mg/kg drug to rats. The blood samples were collected post-dose at different time intervals and subjected to sample preparation involving sequentially protein-precipitation and liquid-freeze separation. The drug and metabolites were separated on an HPLC column followed by LC-MS^n studies. The difference of accurate masses of the drug and metabolites and differences in their mass fragmentation pattern helped to assign structures to the metabolites. In silico detection tools assisted in providing complementary information. Using this strategy, an amalgamation of eleven stable metabolites were ascertained, out of which, nine were Phase I metabolites and two were phase II metabolites. The contemplated bioactivation potential of labetalol was examined by the use of trapping agents (N-acetylcysteine, GSH, and semicarbazide), however, no reactive metabolite was discerned. The present study proffers new insights on the metabolic pathway of labetalol that had not previously been investigated.

P-16

Utility of sandwich cultured rat hepatocytes (SCRH) for metabolite identification studies

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Background/Purpose: A major cause of attrition in preclinical and clinical drug development programs are due to unexpected toxicities and safety issues either with parent drug or its disproportionate drug metabolites. There is increased concern for obtaining metabolite data as early as possible in pre-clinical studies. Variable in-vitro enzyme expression (Cytochrome P450 and Phase II enzymes, drug transporters) and limited culture lifetimes limit the production of all clinically relevant metabolites. Hepatocytes cultured between two layers of collagen (sandwich configuration) improves morphology and viability, and maintains function for longer periods of time in culture. Sandwich-cultured hepatocytes (SCH) are a unique and powerful in-vitro system closely mimicking the in-vivo like situation, which can be utilized to study hepatobiliary drug transport, species differences in drug transport, metabolism, transport protein regulation, drug-drug interactions and hepatotoxicity. The purpose of this study was to assess the utility of long term sandwich culture rat hepatocytes (SCRH) as an alternative hepatocellular model for metabolite identification of low turnover compounds and to find out novel metabolic pathways compared to conventional cultures.

Methods: Freshly prepared rat hepatocytes were seeded in 24 well type-I collagen coated plates at 0.35 x 106 cells/well in 500µL of the DMEM plating medium and cultured at 37°C in a humidified culture chamber with 5% CO2 and 95% relative humidity. At approximately 4 hr post seeding, cells were washed and an overlay with Geltrex™ prepared in incubation media (0.35 mg/mL) was applied and cultured further 16-18 hr prior to use. Post incubation hepatocytes were treated with reference compounds (Tolbutamide, Warfarin, Carbamazepine and Ritonavir, 10µM) prepared in incubation medium and cultured further upto 96 hr with no media change. Samples were collected post 24, 48, 72 and 96 hr of incubation and analyzed for metabolite identification using LTQ Orbitrap Velos. Suspension incubations conducted in parallel upto 4 hr with similar assay conditions for metabolite profile comparisons.

Results: In summary our preliminary results suggest rat hepatocytes with sandwich format were found to be superior to suspension and plated cultures for metabolite identification of low turnover compounds and to find out novel metabolic pathways for reference and project specific compounds compared to conventional cultures. Cryopreserved human hepatocytes with Sandwich format assays to understand the species difference in metabolite formation are ongoing.

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P-17

Solid state manipulation of aripiprazole for enhancement of aqueous solubility

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The current phase of antipsychotic drug development is witnessing an oncoming crisis due to the combined effects of increasing R&D costs, decreasing number of new molecules being launched, blockbuster drugs falling off the patent cliff and a high proportion of advanced drug candidates exhibiting poor aqueous solubility. Among the several methods written in literature for enhancement of aqueous solubility, formulation of polymorphs and co-crystals are of increasing interest because of their ability to alter the solid state for fine tuning the aqueous solubility of the insoluble drugs that are otherwise difficult to deliver. In this work attempts were made to prepare and characterize co-crystals of aripiprazole, BCS class II drug, for enhancement of aqueous solubility for its effective delivery. Co-crystals were prepared by slow evaporation technique at room temperature by selecting coformers from GRAS list of USFDA following proper prediction by means of formula given in literature. The prepared co-crystals were characterized by DBK melting point apparatus, FTIR, DSC, XRD, SEM, aqueous solubility studies and Invitro drug dissolution. Preliminary results revealed that three co-crystals may be formed and the aqueous solubility and dissolution of the co-crystals were more than the commercial sample, which is an encouraging result.

P-18

Metabolite identification of endothelin receptor antagonist, ambresentan by liquid chromatograpy/ tandem mass spectrometry

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Ambrisentan (AMBR) is an endothelin receptor antagonist (ERA), used for the treatment of pulmonary arterial hypertension (PAH). Liver toxicity is the most common reason for drug withdrawals and also ERA as class has been associated with hepatocellular injury. Recent reports on the liver injury due to long uneventful therapy of AMBR, which was earlier reported to be an option for the patients who have discontinued other therapy due to liver function test result failures. This made us to work on to evaluate potential cause of hepatotoxicity by the reactive metabolites of the AMBR preclinically. To identify in vivo metabolites of AMBR, urine, feces and plasma were collected from Sprague-Dawley rats after its oral administration.The samples were prepared by using an optimized sample preparation approach involving protein precipitation followed by solid phase extraction and then subjected to LC-HR-MS/MS analysis. The chromatographic separation of all metabolites was performed on a Waters CSH Phenyl hexyl column (100 x 2.1 mm,1.8µm) using a mobile phase consisting of 0.1% formic acid and acetonitrile in a gradient elution mode at flow rate of 0.3mL/min. A total of 16 metabolites of AMBR have been identified in rat urine and feces which includes epoxide, hydroxylated, O-dealkylated, demethoxylated, decarboxylated and hydrolytic metabolites. In plasma, only hydroxylated metabolites and unchanged AMBR are observed. The suspected epoxide metabolite was one of them. The formation of the same might be an added reason for the hepatotoxicity shown by the drug. The structure elucidation of metabolites was done by fragmentation using MS/MS in combination with HRMS data.

Keywords: Ambrisentan; Metabolite characterization; Liver toxicity; LC-HR-MS/MS

P-19

Elacridar as inhibitor of renal cationic transporters (OCT2, MATE1 and MATE2K)

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Elacridar (GF120918) is a known P-gp and BCRP inhibitor and is extensively used as a P-gp inhibitor both in vitro and in vivo. However, its inhibitory potential towards other hepatic and renal transporters can lead to ambiguous results. Hence it is necessary to exercise caution while using elacridar as an inhibitor. Recently elacridar was shown to inhibit hepatic uptake transporters, such as OATP1B1, OATP2B1 and the efflux transporter MRP2. However, the effect of elacridar on renal transporters is not yet known. The present study focused on exploring the inhibitory profile of elacridar for renal transporters OCT2, MATE1 and MATE2-K. Inhibition experiments were performed in transfected human embryonic kidney (HEK) cells using metformin as a probe substrate. Pyrimethamine was used as a known inhibitor of OCT2, MATE1 and MATE2K. The IC_{50}s of pyrimethamine for OCT2, MATE1 and MATE2-K mediated metformin transport (4.6 μM, 0.2 μM and 0.1 μM, respectively) were similar to literature values. Elacridar inhibited OCT2, MATE1 and MATE2-K with IC_{50} values of 7.6 μM, 0.45 μM and 0.68 μM, respectively. The inhibition of elacridar against these renal transporters are reported for the first time. Further work is in progress to understand the effect of elacridar on other cationic transporters (OCTN1/N2) and OAT1/3. In addition in vivo experiments are planned to evaluate the in vivo relevance of this inhibition.

P-20

A validated UHPLC-QTOF-MS method for quantification of alogliptin in rat plasma: application to pharmacokinetic study

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Alogliptin (ALO) is a potent dipeptidyl peptidase 4 (DPP-4) inhibitor employed for the treatment of type-2 diabetes. Diabetic patients with obesity are vulnerable subjects to cardiovascular complications and its co-administration of Garcinia cambogia (GC) herbal extract offer an alternative approach with antiobesity activity. A sensitive and validated UHPLC-QTOF-MS method was developed for the quantification of ALO in rat plasma using pioglitazone as internal standard (IS). Chromatographic separation were carried out on a Phenomenex Kinetex C-18 (100 mm x 2.1 mm ; 1.7 µm) column using gradient mobile phase system consisting of ammonium formate (10 mM) and methanol at a flow rate of 0.4 mL/min. Detection of ALO (m/z 340.1776) and IS (m/z 357.1265) was performed using high resolution accurate mass spectrometry. The developed method was validated according to US-FDA and EMA bio analytical guidelines. The method LC-MS method meets the guidelines and found to be accurate, precise and selective. The method showed good sensitivity (5 ng/mL) and covered the dynamic range of 5- 5000 ng/mL. The method was successfully applied to pre-clinical monitoring of possible Herbal Drug Interactions (HDI) of GC with ALO in SD rats. Pharmacokinetic parameters of ALO found to be unchanged after co-administration of GC and were found to be safe. The developed method can also be applied to the therapeutic drug monitoring of ALO in clinical samples.

Keywords**:** Garcinia cambogia; Herb drug interaction; UHPLC-QTOF-MS

P-21

Development of porous nano chitin incorporated chitosan composites for potential wound healing application

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Wound healing is a complex process involving an integrated response by many different cell types and growth factors in order to achieve rapid restoration of skin structure and function. The biopolymers, chitin and chitosan have natural biocompatibility and biodegradability with wound healing properties that render them suitable for wound management. The present study evaluated the applicability of nano chitin incorporated lyophilised chitosan membranes for wound healing. DLS of nano chitin indicated an average particle size of 13 μm. The incorporation of nano chitin in the chitosan membranes led to a considerable improvement in tensile strength properties. Lyophilization resulted in softer, thicker and porous film membranes. The porous morphology was clearly indicated in the SEM photomicrographs, which may be favourable for imbibition of wound exudates, while maintaining a moist surface favourable for wound healing. The percentage porosity was found to decrease with increasing concentration of chitosan in the membranes. The water uptake studies showed that the chitosan films absorbed moisture upto 7 hours and retained the shape for more than 3 days. The water vapour permeation of the composite membranes was found to be upto 1119.4 g/m^2.24h. In vitro cytotoxicity on L929 mouse fibroblast cell culture in the presence of Dulbecco's Modified Eagle Medium and 10% Fetal Bovine Serum at 37^0C and 5% CO_2 atmosphere exhibited excellent cytocompatibility and cell viability. These results strongly support the possibility of using this chitin-chitosan composite dressing for improved wound management application.

P-22

Development of antiseptic and antimicrobial wound dressings

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The objective of the work was to develop a delivery system for wound care by incorporating Povidone iodine, Curcumin and silver sulfadiazine. Chitosan, a polymer derived from natural source containing antimicrobial property was incorporated in the formulation. Polyethylene Glycol 6000 was used as film forming polymer, Polyvinylpyrrolidone was used as plasticizer and Gelatin was used to increase the swelling property. The wound dressings were prepared by Dip-coating method. Three different dressing materials such as Absorbent cotton, Gauze bandage and Gauze with cotton were chosen for the study. The developed wound dressings were characterized for viscosity, thickness, scanning electron microscopy, swelling studies, drug content estimation and in vitro drug release studies. The IR spectral analysis showed the expected characteristic absorption peaks for the pure drugs and mixture of drug with the excipients. This confirmed that drugs and the excipients used were found to be compatible. The thickness of the formulations prepared ranged from 0.07mm to 0.22mm. The wound dressings made of gauze with cotton as dressing material showed greater thickness than other materials. Scanning Electron microscopy of the prepared wound dressings revealed that the surface of the dressings contains minute pores. The porous nature of the dressings helps in maintaining the moist environment as well as air permeation. This helps in better and faster healing of the wound. Swelling increased from 200 to 850% in case of Chitosan wound dressing, 125 to 492% in case of Povidone iodine wound dressing, 46 to 400% in case of Curcumin wound dressing and 200 to 784% in case of silver sulfadiazine wound dressing. Drug content of the wound dressings were estimated using UV-Visible spectrophotometer at 231.5nm for povidone iodine, 457nm for Curcumin and 452.5nm for Silver sulfadiazine.

The *in vitro* drug release revealed that the wound dressings provide immediate release of drug. The drug release increased till 4hrs and gradually decreased further. This indicates that there may be an immediate release of the drugs to the surrounding tissue which can provide faster healing rate. The results indicated the acceptability of the prepared wound dressings by the evaluation tests. Based on the in vitro results, three formulations were selected for the in vivo studies and were investigated for wound healing property. The data was analyzed statistically using Dunnett's comparison test. All the formulations showed a better wound healing activity as compared to Povidone-iodine. The accelerated stability studies for the selected formulations were conducted and results showed acceptable range of stability. The results of this research work suggested that the developed wound dressings can provide a better delivery system for wound care.

Keywords**:** wound dressings, Chitosan, Povidone Iodine, Curcumin, Silver sulfadiazine

P-23

Development of taste masked oral suspension using ion exchange resin for paediatric use

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The main objective of the present work is to mask the bitter taste of Cefixime Trihydrate using ion exchange resins and to formulate oral suspension for paediatric use. Different ratios of the ion exchange resin Kyron T-104 were prepared by batch process to obtain the taste masked drug-resin complex. The drug-resin complex was characterised by FTIR, DSC and XRD studies. . Depending upon the drug loading efficiency, the drug-resin complex of ratio 1:3 was selected for the formulation of taste masked suspension. Characterization of the drug resin complex showed complex formation and also compatibility with the other excipients. Reconstitutable suspension was prepared using Xanthan gum as the suspending agent in different concentration (0.2%, 0.4%, 0.6%, 0.8% and 1.0%) and evaluated for parameters like pH, viscosity, sedimentation volume and in vitro drug release. In vivo study was carried using Rat Behavioural Avoidance Model for taste masking. The reconstituted suspension showed easy redispersibility. Formulation F3 showed 90% of drug release within 45 min at pH 1.2. In vivo study revealed that the bitter taste of Cefixime was effectively masked.

Keywords**:** Cefixime, Ion Exchange resin, suspension, taste masking, sedimentation rate, stability

P-24

Development of colloidal silver impregnated biopolymer antimicrobial wafers for wound care

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Wounds are subjected to infection by many micro-organisms and to treat these chronic infected wounds, antimicrobial agents are used. The present study aims to develop antimicrobial wafers by lyophilization technique for potential application on infected chronic wounds. Wafers as drug delivery systems can be applied directly on to the suppurating wounds, which instantaneously adhere to the surface and transforms from porous solids to highly viscous gels maintaining a moist environment essential for wound healing.

Antimicrobial wafers were successfully prepared with natural biopolymers sodium alginate and chitosan with embedded colloidal silver. Wafers were formulated using different polymers and plasticizer concentrations by lyphhilization technique. Viscosity, tensile strength, folding endurance, thickness, swelling study, hydration study, water vapour transmission rate, SEM characterisation and antimicrobial study of the wafer formulations was evaluated in order to select the optimal formulation for excisional wound healing activity on rat models.

The formulation codes A2, AS2, C2 and CS2 containing polymer concentration 3.5% w/v and plasticizer 40% w/w of polymer exhibited the most desirable characteristics as a ideal wound dressing. SEM-EDAX spectroscopic studies showed porous morphological network and spongy like structure with colloidal silver embeddment in the wafer formulation. The colloidal silver incorporated wafer formulations AS2 & CS2 showed synergistic in-vivo wound healing activity on rat models incontrast to A2 & C2 formulations and marketed colloidal silver gel product. The integrity of the wafer product was retained when subjected to accelerated stability studies as per ICH guidelines. Thus lyophilized antimicrobial wound dressing wafers were successfully prepared for combating microbial infections and enhancing the rate of wound healing.

Keywords**:** Antimicrobial wafers, lyophilization, colloidal silver and wound healing

P-25

Tissue distribution study of novel anti-osteoporotic CDRI compound S007-1500 in SD rats using validated LC-ESI-MS/MS method

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Osteoporosis is a common condition characterized by low bone mineral density (BMD) and an increased risk of fragility fractures. S007-1500 is a new chemical entity (NCE) having potent anti-osteoporotic activity and presently under preclinical phase. The preclinical phase plays an important role for identification and optimization of drug like properties. In present study, an LC-ESI-MS/MS method was developed and partially validated for quantification of S007-1500 in rat plasma and various tissues. The simple protein precipitation using acetonitrile (ACN) as precipitating solvent was used to extract S007-1500 from biological matrix. However, chromatographic separation of S007-1500 and internal standard (I.S.) was achieved on C_{18} column (4.6 mm × 50 mm, 5.0 µm) using ACN and 0.01 M ammonium acetate (pH 5.0) in the ratio of 90:10 (v/v) as mobile phase. The parent → product ion transitions (MRM) for analyte and I.S. were 295.2 → 107.2 m/z and 352.3 → 112.2 m/z respectively, and were monitored on a triple quadrupole mass spectrometer, operating in ESI positive ion mode. The method was linear within the concentration range of 5-1000 ng/mL in different biological matrix. The tissue distribution study was performed at 20 mg/kg dose and a level of S007-1500 was detected in various tissue including heart, brain, intestine, liver, spleen, and bone marrow. The maximum concentration of S007-1500 in various tissue was found to be in following order: $C_{intestine}$ (122800.0±148.1ng/g) > C_{liver} (2516.66±98.24 ng/g) > C_{spleen} (986.53±72.3 ng/g) > C_{heart} (401.66±57.00 ng/g) > C_{brain} (189.46±28.25 ng/g) ≈ $C_{bone\ marrow}$ (187.3±26.4 ng/g) > C_{plasma} (72.66±21.44 ng/g). In addition, the above results would be helpful for further preclinical and clinical reference of S007-1500 as a potential candidate drug for the treatment of osteoporosis.

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P-26

A scientific study on potential role of solidified cow urine as a pharmaceutical adjuvant for improved *in vitro* efficacy of poorly bioavailable drugs

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The present investigation deals with the use of solidified cow urine (CU) as a natural pharmaceutical adjuvant in pharmaceutical formulations of poorly soluble and bioavailable drugs. Fresh cow urine was collected and converted into solidified form by drying in a tray dryer and lyophilizer. Aceclofenac (BCS class II) and famotidine (BCS class IV) with low and variable bioavailability issues were the model drugs. Pre-formulation studies were carried out to ascertain the compatibility between the CU powder and model drugs. Tablets were formulated by using powdered CU as a hydrophilic carrier.

Aceclofenac and famotidine tablets were evaluated for post compression parameters, in-vitro dissolution and SEM analysis. FTIR study indicated the compatibility between drugs and solidified CU. SEM studies revealed the occurrence of porous external surface for better wettability and effective drug release. The physicochemical properties of tablets containing cow urine powder were found to be significant and acceptable.

The solubility of drugs from formulations was increased from 2.0 to 3.0 mg/ml compared to pure drugs (0.020-1 mg/ml). The dissolution profile of standardized tablets AF5 and FF5 were found to be 99.34% and 60.92% respectively, indicating better drug release compared to marketed product without CU powder. In-vitro toxicity studies such as hemocompatibility assessment, histocompatibility assay and mutagenicity (AMES test) confirmed the non-toxic nature of the CU powder. Standard formulations demonstrated better physico-chemical properties on accelerated stability studies as per ICH guidelines. Thus, the solidified CU could be a promising natural additive in several pharmaceutical formulations for attaining improved solubility and better bio efficacy.

P-27

CYP inhibition and metabolite identification studies of novel fracture healing CDRI molecule S007-1500

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Osteoporosis is a metabolic bone disorder that is characterized by low bone mass and micro-architectural deterioration of bone tissue which increases bone fragility and the risk of fractures. S007-1500, a novel CDRI molecule having potent fracture healing activity and currently is under preclinical phase. The CYP inhibition assays play an important part in preclinical studies for determining drug interaction potential of the novel compounds. CYP inhibition studies of S007-1500 were performed in all the USFDA recommended CYP isoforms using approved specific inhibitors in rat (RLM) and human (HLM) liver microsomes. All CYP substrates and their metabolites were analyzed using liquid chromatography–tandem mass spectrometry (LC-MS/MS). S007-1500 is metabolized majority via oxidation, dealkylation and dioxidation. Based on IC50 value the compound S007-1500 was found to be moderate inhibitor of CYP1A2 and 2C11 and weak or no inhibition was found for CYP3A2, 2D4 and 2E1 in RLM. Whereas, in case of HLM weak inhibition was observed on CYP1A2 and 3A4 and no inhibition were found for 2E1, 2C9 and 2D6 isozymes [1]. Furthermore, metabolite identification study was performed in RLM and HLM. The information-dependent acquisition (IDA) scan function was utilized for data acquisition using Analyst software, version 1.6 (AB Sciex, Fermingham, MA, USA). A multiple reaction monitoring (MRM) was performed in the IDA experiment to trigger the collection of MS/MS data. Based on the chosen biotransformation, and the parent and product ion m/z of the substrate, the MRM transitions for the metabolites were automatically generated by the IDA function. Data was processed through LightSight software version 2.3 (Applied Biosystems/MDS Sciex, USA) to explore the putative structures of the predicted metabolites. The metabolites were identified based on their protonated molecular ions and fragmentation pattern [2]. Qualitatively similar metabolites were found in both rat and human liver microsomal incubations

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P-28

Development of an uptake assay using LLC-PK1-MDR1 cells to evaluate MDR1 substrates: utilization with verapamil, amiodarone and atenolol

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The conventional trans-well assay using LLC-PK1 cells stably transfected with MDR1 (ABCB1, also referred to as P-gp) has difficulty to accurately assess MDR1-mediated transport for compounds with low permeability or that have nonspecific binding to the outer cell membrane and cell culture plate. To evaluate these types of challenging compounds, LLC-PK1-MDR1 cells were seeded in a monolayer, instead of a trans-well format, and incubations were conducted. The ratio of accumulation in wild type (WT) versus transfected cells (uptake ratio = accumulation in WT cells/ accumulation in MDR1 transfected cells) was used to identify MDR1 substrates. Using this plated format, MDR1 transport of digoxin, verapamil, amidarone, quinidine and atenolol were evaluated. Verapamil accumulation in LLC-PK1-WT cells decreased with increasing concentration of elacridar (a known MDR1 inhibitor). In addition, verapamil uptake in LLC-PK1-WT cells was inhibited by quinidine, another known MDR1 inhibitor that is also a substrate. Therefore, involvement of a potential cationic transporter in the uptake of verapamil in LLC-PK1 cells can be postulated. Work is in progress to understand the cationic uptake transporter(s) of verapamil. In addition, using this format, atenolol was identified as a MDR1 substrate with an uptake ratio of 5 that was reduced to 1.6 in the presence of 5 μM elacridar.

In summary, the plated LLC-PK1-MDR1 format demonstrated that atenolol and verapamil can be detected as possible MDR1 substrates. Possible involvement of uptake transporter of verapamil was also detected in LLC-PK1 cells.

P-29

Effect of *Wattakaka volubilis* root extract in diabetes mellitus associated cataract formation

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Diabetes mellitus is a multifactorial disease and complications arise due to the damage to structural and functional proteins by formation of glycosylation end products. Among all the complications, diabetic cataract may be the most common which is linked to sorbitol formation in lens by the effect of Aldose reductase enzyme. The aim of this study was to determine the Aldose Reductase (AR) inhibition and cataract prevention property of W. volubilis root extract in rats induced with diabetes mellitus. AR activation is the most important mechanisms responsible for the development of diabetic cataract. W. volubilis is traditionally used in Ayurveda as an alternative drug for Murva. Paste of bark and leaves are traditionally used to treat diabetes. In this study, Diabetes was induced by a single i.p., injection of streptozotocin at the dose of 45 mg/kg. Development of hyperglycemia in rats was confirmed by estimation of fasting serum glucose, 72 h post STZ injection. Diabetic animals were treated with the alcohol extract of W. volubilis root at 100 and 200 mg/kg doses for 38 d. At the end of the treatment, animals were sacrificed with excess of ether anesthesia and the lens isolated by posterior approach. Homogenate prepared from pooled lenses was used to estimate Aldose reductase, Sorbitol dehydrogenase, antioxidant parameters, sodium, potassium, calcium and lactate dehydrogenase. The results of this study indicate that alcohol extract of W. volubilis root significantly inhibited aldose reductase activity ($p < 0.001$) and sorbitol formation in the lens with the overall decrease in the severity of cataract progression.

Keywords: Wattakaka volubilis, Diabetes mellitus, Cataract, Aldose reductase, Polyol pathway.

P-30

Using information models for DMPK data and workflow

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This poster demonstrates the advantages of taking a workflow driven approach to data collection in drug discovery and development organisations. The reliance on generic spreadsheets requiring manual data handling steps can lead to inefficiency and human error. Time is lost checking to see if data reaches defined quality criteria and if it is ready for the next step in a process. We will discuss new tools used to reduce manual data manipulation and decrease the need for data checking, therefore increasing efficiency, standardization and quality. In the context of specific assays from DMPK and ADME we will show how combining workflow and data analysis helps to free scientists from the drudgery of daily tasks.

P-31

CYP2D6 polymorphism in eastern Indian population

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Introduction: Almost 25% of prescription drugs are metabolized by CYP2D6. Polymorphism of CYP2D6 is well known and its phenotypic variation in metabolic activity has been studied in various populations around the globe, but it has not been done yet in eastern India population. Here we determined prevalence of CYP2D6 polymorphism phenotypes using dextromethorphan as a probe drug in subjects with at least parental population residing in eastern India.

Method: 60mg dextromethorphan was administered to participants after an overnight fast and 5ml blood was collected 3 hr after dosing. Dextromethorphan and its metabolite dextrorphan concentration was determined in plasma. Metabolic ratio(MR) of dextromethorphan to dextrorphan was calculated for each participant. Histogram of MR suggests bimodal distribution. The polynomial equation derived from probit analysis was solved to obtain antimode of MR. Individuals with log MR higher than antimode were classified as poor metabolizers(PMs) while those with values less than antimode were extensive metabolizers(EMs)

Results: Data from 97 participants were evaluated and the median MR was 0.209 (interquartile range 0.090 – 0.609). The antimode for MR was calculated to be 3.055. From this, it was deduced that three participants were PMs and rest were EMs. No adverse events were encountered. The prevalence of CYP2D6 polymorphism in Eastern Indian population is low (3.09%; 95% confidence interval 0.35 to 6.54%) and is similar to other regions of India.

Conclusions: Variation in CYP2D6 activity can have therapeutic implications particularly in the context of polypharmacy and over-the-counter drug use which are common in India. CYP2D6 phenotyping may help clinicians to avoid adverse drug-drug interactions. However, considering the relatively low frequency of CYP2D6 poor metabolizer phenotype in India, routine screening for this phenotype is likely to be cost-prohibitive and cannot be recommended at this juncture.

P-32

Novel self-emulsifying nano-carrier improves *in vitro* anti-leishmanial activity of amphotericin B

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Visceral leishmaniasis is a dreadful diseases in India and also other parts of the world. Amphotericin B is a gold standard drug used in visceral leishmaniasis, and is available in market as intravenous infusion. Owing to its inherent nature of poor solubility and oral bioavailability it comes in BCS class IV. In present investigation we intended to develop self-emulsifying nano-carrier (SENC) that could enhance oral bioavailability of Amphotericin B. Our preliminary screening revealed maximum solubility of AmB i.e. 2.2±0.5 mg/mL and 1.9±0.8 mg/mL in coconut oil and peanut oil respectively. Further based on solubility of AmB, we selected Labrasol, Lauroglycol 90, Capryol PGMC and Tween 80 as excipient mixture. Taking different centration of excipients SENC was optimized. The optimized formulation exhibited mean droplet diameter; 82.73±10.23 to 217±23.11 nm, PDI; 0.246 to 0.432 and zeta potential; -19.55±2.33 to -24.88±3.18 mV, and self-emulsification time; □ 1 min. Entrapment efficiency and drug content was determined to be 93.25±4.22% and 98.66±1.28% respectively. In vitro anti-leishmanial activity (IC_{50}; 0.149 ± 0.08 µg AmpB/ml) and high cell uptake (20.56-fold) in J774A exhibited improved performance of developed nano-carrier containing AmB. Stability studies assured no significant alterations in the characterized pharmaceutical parameters up to 6 months. In vivo anti-leishmanial activity of the optimized formulation is future prospect of present work.

P-33

Evaluation of the effects of combinations (binary and ternary) of water miscible organic solvents on activity of human plasma esterases.

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Organic solvents are commonly used as co-solvents in in vitro enzyme reaction studies. However, it is known that these organic solvents affect the activities of drug metabolizing enzymes in variable and unpredictable ways. Esterases are a major class of drug metabolizing enzymes especially in the in vivo activation of several ester/amide/phosphate type prodrugs. Thus, the knowledge of effect of organic solvents on esterase activity is important. Although there are reports on the effects of single solvents on esterase activity, little is known about the effect of solvent mixtures on the substrate metabolizing activity of human plasma esterases.

In the present study, we have evaluated the effects of binary and ternary mixtures of different combinations/ratios of three organic solvents acetonitrile, methanol and dimethyl sulfoxide (at 2%v/v total concentration) on the metabolism of p-nitrophenyl acetate by human plasma esterases, using a spectroscopic assay. All the solvents mixtures studied showed concentration dependent inhibition of esterases. Dimethyl sulfoxide was found to be most inhibitory (82% inhibition at 2%v/v concentration) and methanol the least (57% inhibition at 2%v/v concentration). The 2%v/v binary mixture of dimethyl sulfoxide with acetonitrile or methanol, showed greater inhibition than that shown by acetonitrile or methanol alone, and lesser inhibition than that shown by dimethyl sulfoxide alone. Similarly, the 2%v/v binary mixture of acetonitrile and methanol showed greater inhibition than that shown by methanol alone and lesser inhibition than that shown by acetonitrile alone. In the case when 2%v/v ternary mixtures of dimethyl sulfoxide with acetonitrile and methanol, the inhibition observed was greater than that of methanol or acetonitrile alone.

Overall, in mixtures the more inhibitory solvent seemed to have a dominant effect and although the level of inhibition in mixtures was less than that with more inhibitory solvent alone, the effect seemed only marginal. It appears that solvent mixtures do not seem to offer any advantage, as per our preliminary studies, at least in the case of human plasma esterases.

P-34

Dual targeted pyrimidine derivatives in the treatment of inflammation related cancer

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Docking studies were carried out for several standard anticancer drugs in cyclooxygenase 2 (1CX2) and Thymidylate synthase (1HVY) receptors. The binding affinity and interaction with specific amino acid with receptors were studied. On the basis of above facts, several substituted pyrimidine derivatives were designed to bind with specific amino acid within the receptor similar to the standard anticancer drugs. Best hit molecules were selected on basis of their docking score, hydrogen bonding and physicochemical parameters. The best molecules were synthesized and confirmed by IR, H1NMR and Mass spectral data. The synthesized compounds were evaluated for their anti-inflammatory and anticancer activity. In vitro anticancer activities of synthesized compound were carried out using MCF-7 and EAC cell lines. The substituted derivatives, 4-chloro (3e), 3-bromo-4-methoxy (3j) and 3,4-dimethoxy (3l) showed better anti-inflammatory activity and 3j and 3l showed good anticancer activity. Presence of methoxy, chloro and nitro groups as substitution in benzene ring increases the pharmacological activity. Pyrimidine derivative containing indole ring showed excellent anti-inflammatory and anticancer activity.

P-35

Pyrimidine based cycloxygenase-2 and thymidylate synthase inhibitors: Design, synthesis and *in vitro* biological evaluation

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Docking as well as QSPR analysis for 35 standard anticancer drugs were carried out in 1HVY and 1CX2 receptors using Discovery Studio 3.5. Compounds 3c, 3d, 3b, 3l, 3a, 3j, 3e and 3i showed significant docking score in both 1HVY and 1CX2 receptors. Based on docking and QSPR studies, structures were designed to bind with the receptors more effectively. On the basis of docking studies a series of pyrimidine derivatives were designed and synthesized. The structures were confirmed by IR, 1H NMR and mass spectral data. Synthesized compounds were evaluated for their invitro anti-inflammatory and anticancer activities. In vitro anticancer activity was carried out for all the synthesized compounds using MCF-7 and EAC cell lines. Among all the synthesized compounds, 5-bromo-2-methoxy substituted and indole substituted derivatives showed significant anti-inflammatory and anticancer activity

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