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Dr. Joseph T. DiPiro is Dean, Professor and Archie. O. McCalley Chair at the Virginia Commonwealth University School of Pharmacy, Richmond, Virginia. He received his BS in pharmacy (Honors College) from the University of Connecticut and Doctor of Pharmacy from the University of Kentucky. He served a residency at the University of Kentucky Medical Center and a fellowship in Clinical Immunology at Johns Hopkins University. Prior to his service at VCU he was Executive Dean at the South Carolina College of Pharmacy (2005-2014) and Professor and Department Head at the University of Georgia College of Pharmacy (1981-2004).

He is Past-President of the American Association of Colleges of Pharmacy and Past Chair of the Council of Deans. He has served as President of the American College of Clinical Pharmacy. He is a Fellow of the College and has served on the Research Institute Board of Trustees. He has been a member of the American Society of Health-System Pharmacists, having served on the Commission on Therapeutics and the Task Force on Science. In 2002, the American Association of Colleges of Pharmacy selected Dr. DiPiro for the Robert K. Chalmers Distinguished Educator Award. He has also received the Russell R. Miller Literature Award and the Education Award from the American College of Clinical Pharmacy, the Award for Sustained Contributions to the Literature from the American Society of Health-System Pharmacists, and was named in 2013 as the national Rho Chi Distinguished Lecturer. Dr. DiPiro was elected a Fellow in the American Association for the Advancement of Science.

Dr. DiPiro served as Editor of *The American Journal of Pharmaceutical Education* for 12 years. He is an editor for *Pharmacotherapy: A Pathophysiologic Approach*, now in its 11th edition. He is also the author of *Concepts in Clinical Pharmacokinetics* and Editor of the *Encyclopedia of Clinical Pharmacy*. He has published over 200 journal papers, books, book chapters, and editorials in academic and professional journals, mainly related to antibiotics, drug use in surgery, and pharmacy education.

Prof. Dr. V. Gopal
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Prof Dr. Venkatachalam Gopal is the Registrar Academic of Mother Theresa PG&R Institute of Health Sciences, A Government of Puducherry Institution, Accredited by NAAC with 'A' grade. He is also the Principal of the College of Pharmacy, Head of the Department and Professor of Pharmacognosy. He is in-charge of the AYUSH Drug Testing Lab. He has been handling classes for the past twenty-seven years at all levels of Pharmacy education viz. D. Pharm., Diploma in Ayurveda Pharmacy, Diploma in Siddha Pharmacy, Diploma in Homoeopathy Pharmacy, B.Pharm., M.Pharm., M.B.A., and Ph.D. Twenty-sixPh. D scholars have been awarded Ph. D under his guidance. He has published 142 research papers. He is a central council member of the Pharmacy Council of India and member of the All India Board of Pharmaceutical Education of the All India Council of Technical Education. He is Chairman of the Board of Studies of Pharmacy of Pondicherry University. He is the designated officer of the monitoring cell, Dept. of Drugs control, Govt. of Puducherry. He is also the chairman of the Indian Red Cross Society, Puducherry branch. He served as a member of the Siddha Pharmacopoeial committee and member of the Siddha Advisory Board. He has received 15 awards.

Prof Dr. Mohamed Azmi Bin Ahmad Hassali
School of Pharmaceutical Sciences, Universiti Sains Malaysia

Professor Dr. Mohamed Azmi Ahmad Hassali graduated with a bachelor's degree in pharmacy from Universiti Sains Malaysia and a master's degree in the field Clinical Pharmacy from the same university. In the year 2002, he was selected to receive the 'Universiti Sains Malaysia Academic Staff Training Fellowship (ASTS)' to pursue his PhD studies in the field of pharmacy practice in Australia. He was successfully awarded with a PhD degree by the Victorian College of Pharmacy, Monash University, Melbourne, Australia in the year 2006. Professor Azmi is one of the key academicians involved in the setting up of the Discipline of Social and Administrative Pharmacy at the School of Pharmaceutical Sciences, Universiti Sains Malaysia which is the only discipline in South East Asia and caters for graduate research training in the field social and administrative pharmacy for post graduate students of more than 14 countries.As a renowned researcher in the field of pharmacy practice, social pharmacy and quality use of medicines from Asia, he holds membership and council post in many international health based organization namely Health Action International-Asia Pacific (HAI-AP), Action on Antibiotic Resistant (ReAct) and International Network For Rational Use of Drugs (INRUD). Since July 2011, he had been selected to head the Malaysian country group for advocating rational use of medicines in population by INRUD. He had been identified by World Health Organization Western Pacific Office (WHO WPRO) and WHO Geneva as one of the leading researchers and resource person in the field of generic medicines policy analysis and patient medication safety. At national level, he had been appointed by the Malaysian Minister of Health as one of the board members for the Pharmacy Board of Malaysia for a period of 5 years that is from 2011 till 2016. He had been appointed as the council member for the Malaysian Academy of Pharmacy since 2012 till now and had served as a council member for the Malaysian Pharmaceutical Society from 2011 till 2015. As an avid researcher and writer, Professor Azmi had published more than 500 full research journal articles in international peer reviewed journals and had authored more than 150 conference presentations especially in the areas related to quality use of medicines and pharmacy practice research in low and middle income countries. Due to his excellent contribution to pharmacy research and innovative community empowerment projects on rational use of medicines, he had been awarded with ''Distinguished Conduct Medal (*Due to his excellent contributions towards social and administrative pharmacy research field in* PKT)'' by the State the State Governor of Penang *in the year 2014.In October 2015, he had been selected by Academy o*f Sciences Malaysia (ASM) to be one of the recipients for their prestigious "Top Research Scientists Malaysia (TRSM)" award. In the same year, he has been listed in the Malaysian Book of Records as the research scientist with the highest number of publications in Malaysia.

Prof Dr. Wong Tin Wui, PhD
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Universiti Teknologi MARA, Selangor, Malaysia

Professor Dr Wong Tin Wui obtained his PhD degree from the National University of Singapore in 1999. He is presently the lecturer and researcher at the Non-Destructive Biomedical and Pharmaceutical Research Centre, iPROMISE, Universiti Teknologi MARA. His research areas are primarily focused on particle/scaffold design for oral, transdermal, and pulmonary drug delivery, development of novel non-destructive pharmaceutical analyzers, as

well as, design of pharmaceutical processors for innovative dosage form manufacture. He has published over 110 peer reviewed articles. He is the editorial board member of Asian Journal of Pharmaceutical Sciences, associate editor of Drug Development and Industrial Pharmacy, and Drug Design, Development and Therapy, regional editor of Current Drug Delivery, co-Editor-in-Chief of Recent Patents on Drug Delivery and Formulation, and has served as the reviewer for more than 90 international journals (eg. International Journal of Pharmaceutics, ACS Applied Materials and Interfaces, Expert Opinion on Drug Delivery, Pharmaceutical Research, Nanoscale).

Professor Wong is the advisory board member/outstanding scientists jury/lead judge for several international awards (eg. Maurice-Marie Janot Award and Lecture, Tefarco Innova-PharmaTech Scientist Award, Malaysia Technology Expo Award). He serves as the visiting professor of UCSI University and National University of Singapore, and lecture professor of Yangzhou University, China.

Prof. Dr. Khozirah Binti Shaari
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Khozirah Shaari is a Professor in Organic Chemistry at the Department of Chemistry, Faculty of Science, Universiti Putra Malaysia. She received her BSc (Hons.) in Chemistry in 1984 from University of Swansea, Wales, and further obtained a PhD in Phytochemistry from University of Strathclyde, Glasgow, Scotland in 1994. She presently heads the Laboratory of Natural Products at the Institute of Bioscience (IBS), UPM. Prof. Khozirah is an expert in Natural Products Chemistry, with research interests in the chemistry and biology of bioactive natural products, herbal standardization and quality control, metabolite profiling and global molecular networks, organic synthesis and structure-activity-relationship studies, and metabolomics. Prof Khozirah has authored and co-authored more than 200 research publications and holds 4 patents. She is an Associate Editor for *Journal of Ethnopharmacology* (Elsevier), editorial board member for *Phytochemical Analysis* (Wiley), and regularly reviews papers in chemistry- and biomedical-related journals. She is a an Associate Member of IUPAC Committee on Chemistry and Industry (COCI), board member of the Asian Network of Research on Anti-diabetic Plants (ANRAP), member of the Advisory committee for Phytochemical Society of Asia (PSA), and committee member of the Malaysian Natural Products Society (MNPS).

Prof Dr Nor Hadiani Ismail
Atta-ur-Rahman institute for natural products discovery
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Prof Dr Nor Hadiani Ismail obtained her B Sc (Honours Chemistry) from University of Waterloo, Canada, in 1986. She received her Doctor of Philosophy in Natural Product Chemistry from University Putra Malaysia in 1999. She is a faculty member and professor of chemistry at the Faculty of Applied Sciences, Universiti Teknologi MARA, Malaysia. She is a fellow of the Malaysian Institute and Chemistry and was recognized as Top Research Scientist Malaysia by the Academy of Sciences in 2017. Prof Dr Nor Hadiani has vast experience in teaching and research in the area of organic chemistry and natural products Chemistry. She is

currently the vice president of Malaysian Natural Products Chemistry. Exploring the vast biodiversity resources of Malaysia forms the main research theme with chemical composition investigation of medicinal plants and herbs being the core of many projects in search for bioactive compounds that may serve as leads for the development of new pharmaceuticals. Chemical constituents responsible for biological effects are identified, isolated, and purified, while multicomponent herbal extracts are characterized and standardized using cutting edge chemical instrumentations to enable usage as new botanical drugs. Current projects include search of bioactive compounds with anti-plasmodial activities, anti-diabetic properties, antioxidants for cancer prevention and cytotoxic activities. Synthesis of analogues based on structures of the natural compounds, for structure activity relationship (SAR) studies and molecular modelling enables rationalization of the observed pharmacological properties.

Prof Dr Shamsul Azhar Shah
Faculty of Medicine, Universiti Kebangsaan Malaysia

Prof Shamsul Azhar Shah is a Professor of Epidemiology and Statistics and a Faculty member at the Department of Community Health, Faculty of Medicine, UKM. He obtained a bachelor medical degree (MD) and a master's degree in public health subspecialty in epidemiology and statistics from UKM. He received JSPS Ronpaku scholarship to continue with a Ph.D. study in Japan. With more than 150 publications in international and local journals, his research interests are in the fields of non-communicable diseases and infectious diseases. His special interests are but not limited to mental health, cancer, paralympic sports, dengue, and tuberculosis. Prof Shamsul is a member of the Public Health Specialist Association of Malaysia (PPPKAM), International Epidemiological Association (IEA) and the Malaysian Society for Environmental Epidemiology (MySEE). He serves on the Editorial Board of several local and international journals.

Assoc. Prof. Dr. Mohd Makmor Bakry
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Mohd bin Makmor Bakry obtained his BPharm (Hons) in 1999 from Universiti Kebangsaan Malaysia (UKM) and PhD (Medicines and Therapeutics) in 2007 from the University of Glasgow, Scotland. He was the recipient of the University Excellent Teaching Award 2013 by UKM, the Malaysian Higher Education Rethinking and Redesigning Award 2017 and the 11th Anugerah Akademik Negara (National Academic Award) in 2017. Currently, he is the member for several significant National committees such as Malaysia Drug Control Authority, Subject Matter Expert Search Committee and Technical Committee for Malaysian Board of Pharmacy. He has published many papers and delivered many talks related to clinical pharmacy practice. His current research interests include clinical pharmacy practice and pharmacotherapy in Neuromedical illnesses.

Prof. Dr. Chung Lip Yong
Faculty of Pharmacy, University of Malaya

Lip Yong Chung is Professor of Pharmaceutical Sciences and Founding Member of the Department of Pharmacy at the University of Malaya, Malaysia. He received his doctorate in pharmacy from the University of Cardiff, UK in 1990 and joined Cardiff University as a research associate focusing on SERC and industry sponsored research. Since joining the University of Malaya in 1995, he contributed to pharmacy undergraduate teaching, postgraduate training, and research and development, and has supervised more than 35 postgraduates including both MSc and PhDs. His recent work focuses on the discovery of bioactive compounds from natural products, the design of bioactive molecules of pharmacological interest, the study of targeting biological systems and nanotechnology-based drug delivery systems.

Mr Amrahi Buang
President, Malaysian Pharmaceutical Society (MPS)

Mr. Amrahi Buang has obtained his B-Pharmacy (Hons) degree from Universiti Sains Malaysia (USM). He is currently serving as the president of Malaysian Pharmaceutical Society (MPS) since 2016 and previously served as deputy president of Malaysian Pharmaceutical Society from 2014 to 2016. He has 35 years of service at University Malaya Medical Centre, Kuala Lumpur. He has vast experience in hospital pharmacy practice, quality management system, patient safety, medication safety and in the field of halal pharmaceuticals. He is also serving as technical committee member of National Medicines Policy since 2006 till now. He is member of Pharmacy Board Malaysia since 2014 until 2021. He is technical committee since 2013 to 2019 for Halal Pharmaceuticals and published General Guidelines as well as currently working as technical working group for Halal Pharmaceuticals – General Requirements and technical committee for Halal Cosmetics, Pharmaceuticals and Medical Devices in 2020. He was deputy management representative for MS 9001 – 2008 Quality Management System in UMMC from 2000 – 2015. Currently he is also member of various committees and boards including National Patient Safety Council, Poisons Board, National Antimicrobial Resistance Committee, National Tobacco Control Committee, mQuit services committee and National Continuous Professional Development committee.

Mr. Rommel Irwan
General Manager, Tigas Alliance
mMPS, mMPCG, CSCSP (MQUIT) and Certified PRP Preceptor

Rommel Irwan is a Registered Pharmacist with 15 years of key management experience in the Retail Pharmacy industry. He is currently the General Manager of Tigas Alliance. His key role is to oversee strategic planning of business expansion, Store and HQ Operations, Sales, Marketing, Merchandising, Purchasing & Logistics as well as chain supply management in the South East Asian region. He oversees and manages 13 pharmacy outlets across 6 states in Malaysia under Berjaya Pharmacy Sdn Bhd which is a subsidiary company under Berjaya Corporation Bhd. He is also entrusted to grow the banner of MyTigas partnerships into 500+ pharmacies in South East Asia region with local distribution channels Tigas Alliance was awarded as CMO Asia Best Health Care Brand in 2018 & 2019. His past notable experiences

and achievements are:Nationwide Top 3 performer with Cosway Pharmacy consecutively for 2 years (2015 & 2016), Director of Spring Care Pharmacy Sdn Bhd (2009 to 2013), Head of Pharmacist & Director of Sales/Operations with Lifespring Pharmacy (Beacons Pharmaceutical Pte. Ltd in Singapore) He is passionate about building more leaders across the Retail Pharmacy Industry and identifying more #rockstarpharmaciststo be better pharmacists to build healthier & happier communities.

Opportunities and Challenges for Clinical Pharmacy in the 21st Century

Joseph T. DiPiro*
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Pharmacists in many countries have made significant progress over the past few decades in providing patient-oriented (or clinical) services. From a societal health perspective there is a great need for pharmacists to manage medicines and address medication problems. However, the future roles for pharmacists are not clear. As clinical pharmacy services have developed the key questions now are how pharmacists will work in healthcare teams, how they will be paid for clinical services, and how pharmacists will develop a consistent practice model. While past practice models have mainly been as a consultant the future is in collaborative team practice with accountability to patients. A consistent practice model must be accepted to assure that patients receive the highest quality of care. Pharmacy educators should plan for "Next Generation Pharmacists", health care providers and change agents on the interprofessional health care team, personalizing medication use, managing safe and effective medication systems, and creating healthier communities. To achieve this vision requires major changes in pharmacy education and training. Specific competencies and job tasks (as entrustable professional activities) should be defined. To develop team care models, pharmacists will need to be trained in interprofessional practice. Pharmacists have the responsibility and opportunity to play much more important roles in societal health.

Propagating and protecting the native herbal medicine- lessons learnt.

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Background: The use of medicinal plants in India especially in Tamil nadu is considered as a living tradition. The traditional system of medicine, "Siddha" has originated and is practiced in Tamil Nadu. The Siddha System of Medicine mainly depends on natural resources, of which, plants form the bulk. In Siddha, medicinal plants have been used for prophylaxis, cure, nutrition, and cosmetics. For example: Rauwolfia has been used in India for the past 2000 years to treat mental illness and only in 1952 the modern drug, Reserpine was isolated from this plant. Because of modernisation, these medicinal plants are facing habitat destruction. Over 95% of the medicinal plants used by the Indian Pharmaceutical Industry are collected from the wild. Most of the plants are collected by destructive harvesting, leading to depletion of genetic stocks and diversity of medicinal plants. With the rapid spreading of urbanisation, indigenous communities are lost. Promotion of allopathic primary health care has eroded the traditional knowledge on medicinal plants. As of now, there is no consolidated strategy to keep this living tradition alive. The need of the hour is to conserve medicinal plants, protect ethno medical knowledge and propagate native herbal medicine. The existing native herbal medicine

was reviewed. Most importantly, a SWOT analysis was carried out on the consolidated steps taken, to encourage their strengthening and standardisation. Awareness was created, on the scientific basis of the native herbal medicine, among all the stake holders. A case study of the use of selected formulations of the native herbal medicine by the community, after creating awareness on its scientific basis, was carried out. Incentives, by way of recognition, were provided for encouraging the above conservation actions. The acceptance level of the native herbal medicine improved. Due to commercialisation and lack of legal control, quality was found to deteriorate. The indigenous communities were found to be in an urgent need of more technical resources to improve and understand their time tested and trusted native medical practices. Mobilizing public and private resources to meet these needs in co-operation with indigenous communities is an appropriate task to address. The needs and expectations of the holders of traditional knowledge on native herbal medicine were identified and the modalities for the protection of such knowledge was recognised and encouraged.

Innovative approaches in oral, inhalational, and transdermal drug delivery for cancer and diabetes treatment

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Oral, pulmonary, and transdermal drug delivery is challenged by the anatomical and physiological attributes of the respective route of administration. The oral drug delivery is met with harsh gastrointestinal environment which renders therapeutic degradation and reduced ease of drug targeting. The pulmonary drug delivery, specifically via nanoparticulate carriers, is susceptible to drug exhalation due to breathing process and complex lung structure disfavouring deep or peripheral lung deposition of drug. Transdermal drug delivery is hindered by stratum corneum, the outermost skin layer, thus limiting transmucosal drug diffusion even though the drug is processed in the form of nanoparticles. This presentation aims to highlight innovative approaches that have been explored for oral, pulmonary, and transdermal delivery of cancer therapeutics and anti-diabetic protein drug. The cancer therapeutics are characterized by adverse effects over the normal cell populations. The protein drug, such as insulin, is prone to biodegradation and has a relatively low bioavailability with respect to non-injection mode of administration. The consideration of the nature of therapeutics and their routes of administration is imperative to succeed drug delivery.

Evidence-based Phytomedicines: Opportunities in the Metabolomics Era

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Natural products are prolific sources of chemo diversity, a valuable feedstock to drug discovery research and development. The fact that 64% of New Chemical Entities (NCEs) are based on natural products is testimonial to this importance. Despite a declining interest in the screening of natural products by the pharmaceutical industry, natural products have continued to be significant source of drugs and leads. Extracts and infusions containing natural products from plants are also major sources of phytomedicines, often comprising of mixtures of bioactive compounds and imparting complex synergistic effects. In the last century, the recognition that biodiversity resources are rapidly diminishing, is fuelling a renewal of interest in natural

products research. New and innovative approaches are being introduced to increase success rate of discoveries and to improve understanding of the mode and mechanisms of action associated with pharmacological properties. Recently, in addition to using conventional approach, we have also adopted a metabolomics approach in the quest of a more holistic understanding of the biological properties of a medicinal plant or herb. Both approaches have their merits but sample size and loss of biological activity due to the reductionist approach of disturbing the inherent synergism of a plant metabolome, has always been a delimiting factor in the traditional approach. In this regard, systems biology, in metabolomics, is touted for its greater promise in obtaining better insights of a plant metabolome and its effect(s) on a disease state or specific biological perturbations. Metabolomics is a rapidly growing technology that provides a global view of molecular organization at the metabolite level. Complementary to proteomics and genomics; metabolomics reflects the function of organisms from terminal symptoms of metabolic networks and provides a holistic view of the alterations in metabolic pathways caused by disease, environmental exposures, or nutrition. In this presentation selected examples of our findings in using metabolomics approaches in understanding the medicinal properties of several medicinal plants in Malaysia will be discussed.

Phytochemical Exploration of Malaysian Medicinal Plants

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Despite the rapid advancement in modern medicine, the use of herbs to remedy illnesses is still practiced throughout the world. In fact, the trend in using natural and herbal remedy for treatment and health supplement is growing, may be due to the rising cost of conventional drugs and treatment, presence of severe side effects, and the emergence of new diseases. The favourable reputation of herbal and natural remedies attracted strong interest for their development into modern medicine and health products. Thus, prospecting for bioactive constituents from plants with interesting and novel action mechanism, has become one of the most actively pursued activities in drug discovery programs. These constituents are "lead compounds" to be used as templates for more potent, selective, and safe drugs. Tropical plants of Malaysia, with diverse chemical complexities are undoubtedly an important natural resource in the search for bioactive compounds. Herein, phytochemical investigation of two Malaysian medicinal plants is presented. *Renellia elliptica* is used for general health improvements and dubbed as Malaysian Ginseng. Its medicinal uses were documented as treatment of body aches, after-birth tonic, and aphrodisiac. The root extract of *R. elliptica* was found to have anti plasmodial activity. Phytochemical studies of the roots resulted in discovery of a new anthraquinone 1,2- dimethoxy-6-methyl-9,10-anthraquinone, along with ten known ones. *Goniothalamus lanceolatus* Miq. is an endemic plant from the rainforest of Sarawak, Malaysia, used by the indigenous population as an alternative medicine to treat cancer? Eight new bis-styryllactones, goniolanceolatins A−H, possessing a rare α, β-unsaturated δ-lactone moiety with a (6S)-configuration, were isolated from the CH_2Cl_2 extract of the stembark and roots. All the isolates were evaluated for their cytotoxicity against human lung and colorectal cancer cell lines. Goniolancealatin B and D showed cytotoxicity, with IC_{50}values ranging from 2.3 to 4.2 μM, and were inactive toward human noncancerous lung and colorectal cells. Docking studies of these compounds showed that they bind with EGFR tyrosine kinase and cyclin-dependent kinase 2 through hydrogen bonding interactions with the important amino acids, including Lys721, Met769, Asn818, Arg157, Ile10, and Glu12.

Epidemiology Tools in Pharmaceutical Research: Past, Present and Future

Prof Dr. Shamsul Azhar Shah*
Faculty of Medicine, Universiti Kebangsaan Malaysia

In a nutshell, epidemiology is concerned with distribution of disease and its determining factors. It has evolved since more than 100 years ago and help with the understanding of the natural history of the disease and improving human health. From a simple disease distribution to the complex understanding of its causal and treatment outcome, epidemiological concepts have been widely used by all health practitioners including clinicians, public health specialists, health managers, and pharmacists. Many new epidemiological tools have emerged in recent years. Some have proven to be better than the others. The big data epidemiology and the new statistical analysis together with artificial intelligence (AI) programs may improve our understanding of the disease but it may also cause a lot of uncertainty. GIS is also another powerful tool that has been used by many epidemiologists. This presentation will discuss the past, current, and future epidemiological tools that could be used in pharmaceutical research.

Precision Medicine: Future Direction in Pharmacotherapeutics

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Precision medicine is an emerging approach for disease treatment and prevention that considers individual variability in genes, environment, and lifestyle for each person. This approach will allow health professionals and researchers to predict more accurately treatment and prevention strategies for a disease experienced by a specific person. Parts of this approach will significantly change the practice of clinical pharmacy. Many studies currently undergoing to explore the relationship between pharmacokinetics and pharmacodynamics, pharmacogenomics and pharmacokinetics, and pharmacogenomics and pharmacodynamics. More contemporary research such as metabolomics and epigenomics are conducted to further understand the uniqueness of treatment response. This new knowledge will be important for future management of pharmacotherapy that is highly effective and free from adverse effects. Although several examples have been established in several areas of medicine and pharmacy, the role of precision medicine in healthcare practice is relatively limited due to some barriers such as financial and easy to be used assay. Advances in the 'omics' sciences, and the growing availability of health data, present an opportunity to make precise personalized patient care a clinical reality.

Tribenzyltin Carboxylates as Anticancer Agents: From Molecules to Animal Models

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Organotin complexes with carboxylates as ligands are a class of potential metal-based anticancer agents. Previous investigations on organotin complexes mainly focused on preliminary structure-activity relationship of tributyltin and triphenyltin species. In this study, we investigated the in vitro and in vivo antitumour properties of two series of novel tribenzyltin carboxylates. Cytotoxicity of tribenzyltin carboxylates in breast cancer cell lines was evaluated using MTT assay. Modes and pathways of cell death induced by the complexes were

determined using several methods such as flow cytometry aided cell-cycle and phosphatidylserine externalisation analysis, caspase luminescent assay and Western blot analysis. Migration and invasion assays were carried out using transwell inserts, while in vivo antitumour efficacy was evaluated following intravenous injection of the complexes to 4T1-tumour bearing mice. Fluorine-substituted tribenzyltin with *N,N*-diisopropylcarbamothioylsulfanylacetate ligand (C1) and nonhalogenated tribenzyltin containing isonicotinate ligand (C9) demonstrated higher potency than cisplatin in breast cancer cell lines. C1 and C9 also exhibited greater selectivity for breast cancer cells (MCF-7 and MDA-MB231) by ~20- and ~70-fold, respectively, than the normal breast epithelial cells (MCF-10A). Moreover, these complexes induce cell death via apoptosis by modulating intrinsic and extrinsic pathways. Cell cycle arrest at G2/M and S phases, and inhibition of cancer cell motility, migration and invasion were also observed. Additionally, these tin complexes demonstrated improved in vivo antitumour efficacy by approximately 25% compared to cisplatin at the same dose (4 μmole/kg), in a metastatic 4T1 murine model. Entrapment of C1 and C9 in PLGA nanoparticles further improved their in vivo antitumor and antimetastatic efficacy. Several potential upstream therapeutic targets and modes of interaction were identified using in silico techniques. Tribenzyltin carboxylates can be explored as a promising anticancer drug candidate for the treatment of breast cancer.

Health Economic Evaluation of HPV Vaccination policy: A comprehensive example from Indonesia

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Cervical cancer becomes serious burden in developing countries including Indonesia. several strategies have been recommended by WHO to reduce the burden. however, the required budget for prevention program implementation is considerably high. This study will explore the importance of health economic evaluation and its implementation on considering the implementation of cervical cancer prevention policy in Indonesia.A cohort markov model was updated to describe the cost-effectiveness analysis of cervical cancer prevention using HPV vaccine and cervical screening. in addition, this model also provides the clinical, both incidence and mortality-related cervical cancer, impact of the prevention strategies. The Incremental Cost-Effectiveness Ratio (ICER) were compared to a threshold, 3 times Gross Domestic Product of Indonesia, to decide which strategy is cost-effective. additionally, the budget impact analysis was performed to evaluate the impact of the prevention policy to national account. Five out of 6 strategies on cervical cancer prevention using HPV vaccines are cost-effective and one strategy using Bivalent vaccine which is procured using GAVI/UNICEF scheme is considerably cost saving. Finally, the national coverage of the cervical cancer prevention policy requires about US$22 million using GVAI/UNICEF price. most of the cervical cancer prevention scenario is cost-effective strategy in Indonesia and most importantly this strategy is affordable for Indonesian government.

IRCPAS/2020/OP-105

Growth status of infants in relation to age and infection in some part of Sokoto State, Nigeria

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The Geohelminths infecting human are *Ascaris lumbricoides*; *Trichuris trichiura*; Hookworm (*Ancylostoma duodenale* and *Necator americanus*) and *Strogyloides stercoralis*. Before now infantile geohelminths was not considered of importance. This has since changed in the light of reports showing high prevalences of the disease among infants in some part, in and outside Nigeria. Many children showed clinical manifestations and eggs in their faeces from days 1–26 after birth, and more cases occurred within 3 months of birth. This study therefore was to provide data on infantile geohelminths to the pool of information needed to define the status of the disease and for planning intervention strategies in parts of Nigeria. In this review we establish the prevalence of soil transmitted helminths in infants in Sokoto State;the growth status of infants in relation to age and infection and the risk factors associated with infection was also determined. Stool samples were collected and analysed using the formal-ether concentration technique. The weight of each child was measured using a weighing balance. Growth status of each infant was determined by using the weight-for-age percentiles charts from which the physical growth of each infant was classified as obese, overweight, healthy weight or underweight. The infant's demographic data and mothers'behavioral characteristics were recorded. Mothers are most likely responsible for transmitting geohelminths infection to their infants at aged 1-6 months. The level of infection with geohelminths is not significantly associated with growth status of the infants. Those infants, whose mothers use well water, were more likely to be infected than those that use tap water. Mother's education and occupation associate with the prevalence of the parasite infection among the infant. Control measures suggested including good sanitation, enlightenment campaigns and construction of more taps, wells and boreholes as a means of providing safe drinking water to the populace. The low infection of geohelminths in the infants suggests that on-going deworming Programme in the study area is effective in arresting the spread of the disease and should therefore be sustained.

Keywords: Geohelminth;infants; growth status; age; infection; deworming; risk factors

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Examination of Blood for Hepatitis B Virus (HBV) and possible Transmission by Mosquito (*Aedesaegypti*)

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Hepatitis is an inflammation of the liver tissue and its presentations range from complete asymptomatic to severe liver failure. Among others, the causes include viruses and parasites. In 2005, chronic hepatitis B infected 343 million people worldwide. Blood tests and clinical picture are sufficient for diagnosis. Seroepidemiological survey of volunteers was conducted. Three volunteers out of 100 were found infected. *Aedesaegypti* were artificially fed with positive blood samples and could bite rabbits. The rabbits presented geophagy, loss of appetite, thinning, loss of fur and inflammation of liver. This study provides an evidence for transmission of HBV through mosquito. However, this study is limited to transmission to rabbit. A study of transmission to human is recommended.

Keywords: Blood; examination; Hepatitis B; transmission; virus

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Assessment of Bacterial Profilefrom Hydrocarbon Contaminated Soil

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Some species of bacteria are known to have the ability to degrade hydrocarbon compounds. Thus, the identification of such bacteria could be step forward in actualization of bioremediation of toxic compounds in our natural environment. This study analysis 20 samples collected from engine oil contaminated soil in Jega Mechanic workshop in Kebbi state of Nigeria. Total Heterotrophic Bacterial Count (THBC) was carried out and counts ranged from $4.5x10^4$ to $6.2x\ 10^6$cfu/g. Hydrocarbon utilizing bacterial count was also determined and counts ranged from $1.2x10^3$ to $5.0x10^3$cfu/g. The bacterial species identified includes *Micrococcus* spp*, Pseudomonas aeruginosa, Flavobacterium, Alcaligenes, Bacillus* sppand *Staphylococcusaureus.* The study therefore reveals that these indigenous bacterial populations could be capable of mineralizing these pollutants in the environment to safe and acceptable levels if properly harness.

Keywords: Hydrocarbon; degrading; bacteria; mineralizing; bioremediation

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Design, Characterization and Wound Healing Activity of Dual Loaded Flavono Polymeric Nanoparticulate System

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Quercetin-Rutin-Silibinin, three flavonoids are the major flavonolstaken as a function diet. Among these, Quercetin has many benefits, but it is its poor aqueous solubility, lower skin permeation which leads to lower therapeutic efficacy and epithelisation. Recent drug delivery systems such as dualloaded flavono nanoparticulate system (DLFNPs) have not yet been studied with Quercetin and bio enhancers(Rutin and Silibinin).The primary objectives of the research is to prepare, Characterize, wound healing activity of Quercetin-Rutin (Qu-Ru NPs) and Quercetin-Silibinin (Qu-Si NPs) dual loaded flavono polymeric nanoparticles to overcome the limitations of Quercetin. Dual loaded flavono polymeric nanoparticles were prepared by nanoprecipitation technique. Prepared nanoformulation was characterised by various techniques like zeta analysis, surface morphology, drug release, entrapment efficacy and to evaluate the wound healing activityusing excision wound model of prepared Plain (P NPs), Quercetin (Qu NPs), Rutin (Ru NPs), Silibinin (Si NPs), Quercetin-Rutin (Qu-Ru NPs) and Quercetin-Silibinin (Qu-Si NPs) polymeric nanoparticles in comparison with pure corresponding phytochemicals. Result revealed that the prepared nanoparticulate system was spherical in shape. Hence, Quercetin and bio-enhancers like Rutin and Silibinin encapsulated in the polymer matrix will alike in round-morphed and anticipated to enhance the indispensable task of Quercetin and bio-enhancers, release of Quercetin and bio-enhancers from the polymer matrix and dual loaded flavono polymeric nanoparticlessuch as Qu-Ru NPs

and Qu-Si NPs achieved significant wound healing (in mm^2) on 7th day (66.31±1.99****)and (55.86±2.66****) respectively and complete wound healing on 14th daywhen compared to the pure compound Qu(207.86±2.06*) (150.28±2.43**), Ru(209.16±2.43*)(155.90±2.53*) ,Si(139.81±2.21**) (76.07±1.77****),and single loaded Qu-NPs (213.55±2.44*) (157.90±2.53*), Ru- NPs (144.93±2.57**) (79.43±2.79***),Si-NPs (139.81±2.21*) (76.07±1.77****) on 7th and 14th dayrespectively.The animals treated with the Quercetin-Rutin (Qu-Ru NPs), Quercetin-Silibinin (Qu-Si NPs) dual loaded polymeric nanoparticles showed faster wound contraction and re-epithelialization whereas pure compound and Quercetin, Rutin and Silibinin loaded polymeric nanoparticles treated group shown slower wound contraction and epithelialization. Hence this research work concluded that bio enhancer (Rutin, Silibinin) synergistically augment to the therapeutic activity of Quercetin.

Keywords: Dual loaded flavono nanoparticulate system (DLFNPs); Quercetin; wound healing activity.

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Extraction of Phenolic Content and Antioxidant Activities from Selected Medicinal Plants: Effect of Solvents with Different Polarity

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Medicinal plants rich in antioxidants such as polyphenols are potential alternatives to synthetic antioxidants. The choice of extraction solvent is critical for the recovery of plant polyphenols due to their varying polarities and chemical characteristics. Thus far, there is no recommendation of specific extraction solvent for the optimal recovery of plant polyphenols. It is important to establish effective extraction methods to evaluate the antioxidant activity from medicinally or economically viable plant materials. This study aimed to evaluate the effect of four solvents with different polarity on the extraction of phenolic content and antioxidant activity of *Centella asiatica*, *Musa acuminate* flower, *Peperomia pellucida* and *Psophocarpus tetragonolobus.*The edible portions of each plant were shade dried, macerated with four solvents (water, methanol, ethyl acetate and n-hexane) and evaporated under reduced pressure at 40°C. The crude extracts were evaluated with 6 *in vitro* mechanism based assays, namely ferric reducing antioxidant power (FRAP), trolox equivalent antioxidant capacity (TEAC), 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity, TPC, tannin and flavonoids. The yield of crude extract increased with increasing solvent polarity. FRAP values of organic solvent extracts were significantly ($p<0.05$) lower than that of water extract. Extraction solvent had stronger impact on DPPH radical scavenging potential than TEAC of the plant extracts. Total flavonoid was positively correlated with FRAP ($r = 0.583$, $p<0.05$) but inversely associated with IC_{50} of TEAC ($r = -0.740$, $p<0.01$) and DPPH ($r = -0.582$, $p<0.05$). The strong linear correlation found between solvent polarity index and TPC ($r = 0.959$, $p<0.05$), tannin ($r = 0.841$, $p<0.05$), FRAP ($r = 0.972$, $p<0.05$) and IC_{50} of DPPH ($r = -0.978$, $p<0.05$) may suggest that solvents with increasing polarity could enhance the extraction of plant antioxidants. Principle component analysis showed that plant species and extraction solvents contributed to 51.7% and 21.8% of total activity variation, respectively. Water was identified as the ideal extraction solvent for *C. asiatica, M. acuminata* flower and *P. pellucida* while methanol was preferred for *P. tetragonolobus*. The proposed antioxidant index could serve as a novel tool to rank and identify the ideal extraction solvents for medicinal plants.

Keywords: antioxidant; flavonoids; medicinal plants; phenolics; solvent; tannin

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B-Endorphin Attenuates Airway Inflammation inMurine Model of Asthma

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β-Endorphin(END), a neuropeptide primarily synthesized in the hypothalamus, anterior and intermediate lobes of the pituitary and released from central and peripheral nervoussystem has been reported as a powerful immunoregulator. Possessing morphine like effects, it ismainly involved in pain management via binding specifically to mu-opioid (μ-opioid). However, recent studies report thepresence of mRNA transcripts for POMC in immune cells which suggest the capability of these cellsin synthesizing β-END apart from the nerve cells. Neural regulation of airways is under the close control of the pulmonary autonomic nervous system(PNS) which upon stimulation by several factors including stress, cytokine leads to neuronalexcitability. β-END has been reported to act as strong immunomodulatorregulating inflammation by epigenetic modification in many inflammatory disorders but study stilllacks its exploration in regulating airway disease including asthma. The proposed workaims to study the role of β-endorphins in regulating inflammation.Balb/c mice (7-9 weeks; 20-23 gms) were sensitized with 1% TDI intranasally on day 0, 7, and 14 and further challenged thrice a week with 2.5% TDI from day 21-51. Naltrexone (1mg/kgbw) and β-Endorphinwas administered one hour prior to challenge.24 hours after the last exposure mice were sacrificed and BALF, serum and lungs were stored for further studies. Total cell count, differential cell count, eosinophil peroxidase, mylenoperoxide, neutrophil elastase and histology were performed to study inflammation. Naltrexone elevated the inflammatory parameters as total and differential cell count, EPO, MPO and NE when compared with asthmatic mice whileβ-Endorphin reduced all the inflammatory parameters. Architectural changes observed in the alveolar spaces and bronchioles of lungs exhibits inflammation in lungs in asthma induced and naltrexone treated mice which were recovered with β-Endorphintreatment. Naltrexone being an antagonist inhibits the binding of β-Endorphin with μ-opioid receptor and hence inflammation was elevated. Improving ourunderstanding of endogenous opioid mechanisms may provide insight towards thedevelopment of novel treatments against inflammatory disorders.

Keywords:β-Endorphin; immunomodulator; inflammation; airways

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The Anticarcinogenic Effects of *Plectranthus ambonicus*, (Lour.) Spreng. Ethyl Acetate Extract on The Benzo(α)pyrene-Induced Female Mice

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Cancers happen because of the uncontrolled and aimless proliferation. The balance disorders between apoptosis and proliferation are the defining factor of tumorigenicity and tumor

progression. *Plectranthus amboinicus* (Lour.) Spreng. contain flavonoids, terpenoids, saponins, steroids, tannins, which is have strong inhibitors towards lipid peroxidation, scavenger of reactive oxygen and nitrogen species, and also the inhibitor of lipooxygenase. The antioxidants have positive correlation with the cancer prevention. To examine the acute toxicity and to determine the antimutagenic and antiproliferative effects of ethyl acetate extracts from *Plectranthus ambonicus*, (Lour.) Spreng. to the breast cancer cells on mice with the induction of Benzo(α)pyrene. The simplicia was macerated by the ethyl acetate solvents. The acute toxicity test was conducted on male and female mice. The in vivo test of anti-cancer effects on mice was conducted in micronucleus and immunohistochemistry methods. The animals used were mice, weighing 20 -30g each and divided into 5 groups, with each group consists of 5 mice. Cancer induction was given by Benzo(α)pyrene at 15 mg/kg bb for 4 weeksThe *Plectranthus ambonicus*ethyl acetate extract on dose 250; 500; and 750 mg/kg bw enables to inhibit the chromosome damage and the cancer cell proliferation on mice induced by Benzo(α)pyrene with the same dose.The oral single dose of The *Plectranthus ambonicus*ethyl acetate extract given at 10; 100; 1000; and 10000 mg/kg did not affect the behaviors of both male and female mice compared to the control group. The *Plectranthus ambonicus*ethyl acetate extract has the potential effect to be the anti-cancer by preventing chromosome damage by cyclophosphamide induction which inhibits the micronucleus cell formation. there is decrease in proliferation activity on breast cells mice. The highest antiproliferation activity was at 500 mg/kg bw. The *Plectranthus ambonicus*ethyl acetate extract has the antimutagenic and antiproliferative effects on the cancer cells of mice.

Keywords:Anticarcinogenic; *Plectranthus ambonicus*; Benzo(α)pyrene; proliferation

IRCPAS/2020/OP-119

Flaxseed (*Linum usitatissimum*) extract potential effect on human gingival fibroblasts (HGF) cell line

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Herbal medication and natural products have been successfully demonstrated to have general health beneficial effects. The bioactivities of flaxseed (*Linum usitatissimum*)extract have been reported, as previous studies have shown that *l. usitatissimum* extract has various health and beneficial effectssuch as antimicrobial, anti-oxidant and anti-inflammatory effect has skin wound healing activity in addition to that it has good effect on the oral cavity in treating ulcers and general oral health benefits. *L. usitatissimum* is extracted using absolute ethanol and ethanol in water via soxhlet method, gas chromatography mass spectrum (GC-MS) is used to illustrate the components of *l. usitatissimum* extract. 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide(MTT) assay is used to assess the human gingival fibroblasts (HGF) cell viability in different timelines. Scratch assay is used to monitor the healing activity of flaxseed extract against HGF cells. The results illustrate the major component present in *l. usitatissimum* extract and the effect of the ethanol and ethanol in water *l. usitatissimum* extract on HGF cell line, 70% ethanol followed by 100% ethanol extract show high proliferating effect at 24 hours at longer time 48 and 72 hours 100% ethanol extract showed higher proliferating activities. For the wound healing assay the wound started healing as soon as 18 hours' post HGF treatment with *l. usitatissimum* extract. *L. usitatissimum* extract have proliferating effect on HGF cell line therefore it can be a potential promising oral wound healing agent.

Key words: *L. usitatissimum*; GC-MS; MTT; HGF

IRCPAS/2020/OP-120

Antioxidant and sunscreen activity of kencur rhizome*(Kaempferia galanga)*extractand tea leaves*(Camellia sinensis* L.*)*extract cream formula

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Tea (*Camellia sinensis* L.) contains polyphenol chemical compounds that can inhibit oxidation reactions (antioxidants) and free radicals scavenging.Kencur (*Kaempferia galanga* L.) contains Ethyl p-methoxycinamat (EPMS) which functions as a sunscreen by absorbing sunlight energy. This study aims to compare the effect of the combination of tea leaves (*Camellia sinensis* L.) and kencur rhizome extract (*Kaempferia galanga* L.) on antioxidant and sunscreen activity before and after formulation into cream preparations using the Simplex Lattice Design (SLD) model.The study was conducted with a Simplex Lattice Design (SLD) model with 2 components, namely kencur extract (A) and tea extract (B), so that 3 formula designs were obtained namely FI (100% A), F2 (100% B), F3 (50% A and 50% B). The parameters of the physical properties of the cream tested were viscosity, dispersibility and pH. The antioxidant activity test was carried out by the DPPH method, while the sunscreen test was carried out by the UV spectrophotometric method.Based on the SLD model, the optimum formula is obtained by comparing the concentration of kencur extract and tea extract 80%: 20% (formula A) and 70%: 30% (formula B).The results indicate that IC_{50} of tea extract, kencur extract, formula A and formula B were7,863 ppm, 164,414 ppm, 80,429 ppm and 74,166 ppm respectively. IC_{50} value of BHT as a positive control was 22,334 ppm and vitamin E was 8,786 ppm. The SPF values of kencur extract, formula A and formula B were6,368, 5,104and 4,689 respectively. The antioxidant activity of cream preparations was lower than the extract form and positive control. The sunscreen activity of cream combination of tea and kencur extract was increasing after formulation.

KEYWORDS : Kencur; tea; antioxidant; sunscreen; simplex lattice design

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In vitro Cellular Reprogramming and Antioxidant potential of Herbal drug: *Fumaria officinalis*

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Fumaria officinalis (common fumitory, drug fumitory or earth smoke) of family Papaveraceae is the most common species of Western and Central Europe. Extracts of *Fumaria officinalis* have been traditionally used for treatment of some skin diseases (rashes or conjunctivitis), rheumatism, stomachache, abdominal cramps, fever, diarrhea, syphilis and leprosy. The study aimed to investigate the *in-vitro* antioxidant and antiaging potential of MEOH extract of *Fumaria officinalis* by various enzymatic models. Powdered crude drug 100 g were extracted in a soxhlet apparatus with petroleum ether (60-80ºC), chloroform and methanol. After successive solvents extraction, methanolic extract was used for testing of antioxidant potential using DPPH assay. Further, antiaging potential of extract was investigated by inhibitory effect

of various enzymatic estimations i.e. Col-I, Ela-I and Hya-I inhibitory assays on early aging human skin fibroblasts. Phytochemical analysis showed the presence of glycosids, alkaloids flavonoids, and triterpenoids and phenolic compounds in higher concentration. Extract showed inhibitory concentration (IC_{50} = 20.10) and ascorbic acid the standard showed inhibitory concentration (IC_{50} = 35.33). In enzymatic estimations assay, the Col-I, Ela-I and Hya-I of extract were assessed showing inhibitory concentration as Col-I (IC50: 41.25), Ela-I (IC_{50}:35.05) and Hya-I (IC_{50}: 30.55) respectively. Thus, MEOH extract of *Fumaria officinalis* able to inhibit 50% of the activity of aging related enzymes Col-I, Ela-I and Hya-I. This study concluded that MEOH extract of *Fumaria officinalis* has confirmed the high antioxidant potential and *in vitro* inhibitory potential of antiaging enzymes assessed, thus they could be used for further development of anti-aging products and nutraceuticals.

Keywords:*Fumaria officinalis;* soxhlet apparatus; flavonoids; in vitro anti-aging assays; antioxidant activity; elastase inhibitory activity

IRCPAS/2020/OP-123

The Influence of Herb Supplement(A*ndrographis paniculata, Phyllanthus niruri l, Boesenbergia pandurata, Syzygium polyanthum*to Metformin Pharmacokinetic Profile in Wistar Rats

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Diabetes mellitus is one of the causes of top 10 deaths in the world. Sambiloto herb (*Andrographis paniculata*), meniran (*Phyllanthus niruri l*), temu kunci (*Boesenbergia pandurata*), and Bay leaves (*Syzygium polyanthum*) are efficacious as anti-diabetic plants. Conventionally, metformin is also selected as the first aid in diabetes mellitus, but there has not been any study of the combination of it. This study aimed to determine the effect of herbal supplements on changes in metformin pharmacokinetic profiles by using wistar strain rats. This study conducted a simple experimental design with two groups of lab rats, each group consisted of five rats. Group 1 was administered with metformin 320 mg. Group 2 was treated with metformin 320 mg along with herbal supplements. Subsequently, the lab rats were administered at 0.5, 1, 1, 5, 2, 4, 8 and 12 hours; Blood samples were collected to calculate metformin levels in blood plasma by using the HPLC method. The pharmacokinetic profile parameters of metformin such as K, Ka, t½ absorption, t½ elimination, Cpmax, t_{max}, Cl, Vd, and AUC were calculated based on whole metformin levels against time. The pharmacokinetic parameters between treatments were compared statistically using T test with 95% reliability. The results indicated that administration of metformin combined with herbal supplements affected the pharmacokinetic profile in the absorption phase as indicated by changes in the values of Ka, T½ and t_{max} with P value < 0.05, which meant that there were significant differences.

Keywords: pharmacokinetic profile; metformin; herbal medicine

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Optimization and evaluation of hydrodynamically balanced antidiabetic system loaded with Sitagliptin and phytochemical constituents of Triphala churna

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The purpose of this research work is to explore new method for the treatment of constipation or gastrointestinal disorders associated with diabetesmellitus type 2. The main objective is to formulate, optimize and evaluate hydrodynamically balanced system of antidiabetic system incorporated with phytochemical constituents of Triphala extract for the treatment of constipation associated with diabetes. The Triphala churna of two different ratios, 1:1:1 (TC1) and 1:2:4 (TC2) were subjected to hot percolation using Soxhlet apparatus in aqueous and methanol solvents. The obtained extract was subjected to phytochemical screening, physicochemical parameters, acute toxicity studies in mice and laxative activity in rats. The floating matrix tablets of Sitagliptin with methanolic Triphala extract was prepared by wet granulation technique using HPMC K4M as polymer, starch/honey as binder, lactose as diluent and sodium bicarbonate & citric acid as effervescent agents by 2^4 factorial design. The independent variables are drug & Triphala extract ratio (X1), Triphala proportion (X2), binder used for granulation (X3), and amount of effervescent excipients used (X4). The dependent variables are hardness (Y1), buoyancy lag time (Y2), total floating time (Y3), *in-vitro* drug release (Y4), and T50% (Y5). The amount of the phytochemical constituents was found to be more in methanolic extracts when compared to that of the aqueous extract. The acute toxicity studies upto 1000 mg/kg of extract showed no mortality or other negative changes in Swiss Albino mice. The laxative activity showed that increasing amount of extract increased the bowel movements in Wistar Albino rats which in turn increased the total faecal output. The prepared floating tablets were subjected to all post compression parameters. Based on drug content, buoyancy lag time and *in-vitro* drug release the formulations F14 and F16 were selected for *in-vivo* study of the formulation. The *in-vivo* study in Wistar Albino rats revealed that formulation F16 had 83% of laxative activity when compared with that of the standard drug evidencing that Triphala serves as a good laxative in combination with honey.

Keywords: Diabetes, Sitagliptin, Triphala extract, laxative, floating

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The Effect of Negative Pressure of Open Suction Endotracheal 20 and 25 kPA on the Hemodynamics of Patients in Intensive Care Unit (ICU)

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In general, patients treated in the Intensive Care Unit (ICU) require the installation of endotracheal tubes (ETT) and ventilators. Suction is an intervention to prevent airway obstruction and maintain airway patency. ETTsuction caused haemodynamic problems in critically ill patients if the procedure is not appropriate, including pressure regulation and duration of suction. The purpose of this study was to determine the effect of negative suction

pressure on hemodynamic status of patients. This study is a pre-experimental study with a cross-over design involving 40 respondents (n = 40) taken by consecutive sampling technique. Data collection was using patient observation sheets, medical records, and treatment records. Inclusion criteria were adult patients ≥15 years old, using ETT and ventilator and patients with a diagnosis of pneumonia and incomplete observation were study exclusions. Statistical analysis using independent *t*-test with a significance of 5%. The study involved 22 men and 18 women (mean age 46.88 years). Results in suction with a pressure of 20 kPA (mean±*SD*); SpO_2 (97.3±2.6 mmHg), respiratory rate (RR) (19.9 ± 19.9), heart rate (HR) (102.8 ± 18.2 beats/min), and mean arterial pressure (MAP) (98.8±11.6 mmHg), respectively; and in suction with a pressure of 25 kPA (mean±*SD*); SpO_2 (97.0±1.8 mmHg), RR (19.9±19.9 beats/min), HR (105.9±18.5 mmHg), and MAP (107.1±12.5 mmHg), respectively. There was no significant difference between negative pressure suction of 20 kPA and 25 kPA (duration of suction 7 to 10 seconds) on all hemodynamic indicators of patients (SpO_2, RR, HR, and MAP), p >0.05.Both of these pressures are safe to use in open ETT suction, because the increase in hemodynamics is within the safe limits, however suction must be carried out according to procedure and monitored properly.

Kkeywords:Negative pressure, suctionendotracheal tubes (ETT), hemodynamics, intensive Care Unit (ICU**)**

IRCPAS/2020/OP-132

Antidiuretic Activity of the Whole Plant of *Derris brevipes* in Wistar Albino Rats

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The whole plant of *Derris brevipes* has been claimed in Ayurvedic and Unani to possess various medicinal activities including antioxidant, analgesic, and anti-inflammatory, anti-diuretic, and anti-arthritic properties. The aim of the present study was to investigate the antidiuretic activity of the whole plant extracts of *Derris brevipes*. Petroleum ether, chloroform, methanol, and aqueous extracts of *Derris brevipes* were administered to wistar albino rats at a dose of 100mg/kg and 200mg/kg. Furosemide was used to induce diuresis; vasopressin was used as a standard drug. The diuretic effect was evaluated by measuring urine volume and excretion of sodium potassium content. The aqueous extract caused a reduction in urine volume. The reduction in urine volume at doses level of 100 mg/kg body weight and 200 mg/kg body weight were 32 % ($P < 0.01$) and 54 % ($P < 0.001$) respectively compared to the control group. Aqueous extract 200 mg/kg produced significant decrease in electrolytic excretion of Na^+ and K^+, without significant renal excretion of Cl^- when compared to control. The role of vasopressin as the principal factor regulating renal water handling is supported by experience with ADH receptor antagonists. It also indicates the emerging significance of autacoids, and other synergistic factors, to affect ADH receptor/effector mechanisms and to modulate renal ADH responses. Aqueous extract of *Derris brevipes* showed a dose-dependent decrease in urine excretion. The anti-diuretic effects of aqueous extract were indicated by decrease in both water excretion and excretion of sodium and potassium. It can be concluded that aqueous extract of *Derris brevipes* has moderate and safe oral anti diuretic activity.

Key words:*Derris brevipes*; anti-diuretic activity

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Simvastatin, Atorvastatin and Rosuvastatin increase the sensitivity of 5-Fluorouracil to T47D breast cancer cells

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Statins work to inhibit cholesterol synthesis in the mevalonate pathway which results in decreased cholesterol production. Cholesterol is synthesized through the mevalonate pathway which has a correlation with increased regulation of sterol regulatory element-binding proteins (SREBPs) which strongly correlated with cell viability, cell proliferation, cell division and potential metastases. Pre-clinical studies of statin resulted on decreasing of cancer cell proliferation, invasion, and metastasis. 5-Fluorouracil (5-FU) is widely used in the treatment of breast cancer.Clinically, it is used both as a single agent or in combination with other chemotherapies and has been associated with the long-term side effects of cognitive impairment. This study aims to investigate the single and combined cytotoxic and antiproliferative effects of Simvastatin, Atorvastatin and Rosuvastatin with 5-FU on T47D cell growth. The effect of Simvastatin, Atorvastatin, Rosuvastatin and 5-FU single and combination on cell morphology were observed using aninverted microscope. The cytotoxic and proliferation effects of Simvastatin, Atorvastatin, Rosuvastatin and 5-FU single and in combination was observed using the MTT assay. The cytotoxicity potency was determined using IC_{50}values, Combination Index (CI) and inhibition of cell proliferation. Simvastatin, Atorvastatin and Rosuvastatin have a high potencycytotoxicity with IC_{50} value 21, 80 and 26 µg/ml respectively. The potential toxicity of these statin is higher compared with 5-FU (185 µg/ml). The combination of Simvastatin, Atorvastatin or Rosuvastatin with 5-FU shows different results. The combination of Simvastatin and 5-FU produces a moderate-strong synergistic effect (CI <0.5). The combination of Atorvastatin and 5-FU generate a synergistic effect at low concentrations (CI <1) and an additive-antagonist effect at high concentrations. The combination of rosuvastatin and 5FU has a strong synergistic effect (CI <0.3) to a low antagonistic effect (CI> 1.1). Simvastatin, Atorvastatin and Rosuvastatin reduce the breast cancer cell proliferation. The combination of statin and 5-FU inhibitcell proliferation stronger than the single one. Simvastatin, Atorvastatin and Rosuvastatin increase the sensitivity of 5-FU to T47D breast cancer cells. the combination of Simvastatin with 5-FU provides the best sensitivity-enhancing effect so it can be recommended for further investigation to determine its effect in vivo.

Keywords: Simvastatin, Atorvastatin, Rosuvastatin, T47D cells, 5-fluorouracil, toxicity, sensitivity

RCPAS/2020/OP-137

Immunomodulary therapy using phytosomes containing *Nymphaea nouchali* extract complexed with phospholipids

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Immunomodulators are those which modify the immune response or the functioning of the immune system by the stimulation of antibody formation or the inhibition of white blood cell

activity. Immunotherapy, a biologic therapy is designed to treat a disease either by eliciting an immune response i.e. activation immunotherapy or by reducing an immune response i.e. suppression immunotherapy. Phytosomes, an endowed vesicular drug delivery system is formulated to distribute various active phytocomponents at the target site for a wide range of pharmaceutical applications. *Nymphaea nouchali* (Nn), Indian Red Water Lily, belonging to the genus *Nymphaea* and family *Nymphaeaceae* is well thought-out as a medicinal plant under Indian Ayurvedic system of medicine. Nn has been reported to use in treatment of diabetes, tumor, inflammation, liver and urinary disorders, menstruation, and indigestion problems. The aim of the proposed research work is to formulate and evaluate the immunomodulant efficacy of the phytoconstituents present in Nn extract complexed with various phospholipids with the objectives of improving efficacy of the phytoconstituents, bioavailability and thereby treatment strategy. In this proposed research work, phytosomes were obtained by reacting different phospholipids (phosphatidic acid, phosphatidyl choline, phosphatidyl ethanolamine and phosphatidyl serine) in tetrahydrofuran with the selected botanical derivatives in dioxane: methanol solvent system by solvent evaporation technique using rotary flash evaporator by employing different molar ratios of drug and phospholipids. Evaluation includes UV spectrophotometric, FT-IR spectroscopic and DSC studies; surface morphology and particle size distribution; drug content, entrapment efficiency, *in vitro* diffusion and stability studies. *In vivo* immunomodulant activity was measured in terms of delayed type hypersentivity (DTH) reaction and humoral antibody response in mice performed at SASTRA University, Tamil Nadu. As compared to crude drug and other conventional dosage form, the results revealed that the optimized phytosomal carrier exhibited significant effect over the release of loaded Nn phytoconstituents. Thus, the phytosomal carriers could be successfully engineered for Nn bioactives with improved *in vitro* release characteristics and better *in vivo* immunomodulant activity which shows potential for escalating drug delivery.

Keywords: *Nymphaea nouchali;* phosphatidic acid; phosphatidyl choline; phosphatidyl ethanolamine; phosphatidyl serine; phytosomes

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The Effectiveness of *Mirabilis jalapa* Leaf Methanol Extract's Concentration on the Growth of *Streptococcus pyogenes*

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Streptococcus pyogenes is gram-positive bacteria which, if not handledproperly, can cause systemic infections such as necrotizing fasciitis and toxic shocksyndrome. The emergence of antibiotic resistance causes fewer choices for therapy. Researchon herbs that have antibiotic effects could be an option for the development oftherapy. The objective of the study is knowing the effectiveness of the *Mirabilis jalapa's* leaf methanol extracton the growth of *Streptococcus pyogenes*. Laboratory experiments by testing the*M. jalapa's* leaf flower methanol extracts against the growth of *Streptococcuspyogenes* bacteria. The average diameter of inhibition zone of methanol extractof *M. jalapa's* four concentrations of 6.25 mg / ml = 9.97 mm; concentration of 12.5 mg/ ml = 10.20 mm; concentration of 25 mg / ml = 7.75 mm; concentration of 50 mg / ml = 8.08mm; positive control = 25.75 mm; negative control = 6 mm. Phytochemical screening showed that methanol extract containing tannins,saponins, and alkaloids. Tannins extracted from various types of plants have moderateeffectiveness in inhibiting the growth of gram-

negative and gram-positive bacteria.Tanninscan inhibit bacterial enzyme receptors that are important for bacterial metabolism, namely*cellulose, pectinase, xylanase, peroxidase, laccase, glikositransferase*. Methanol extract at*M. jalapa's* leaves with a concentration of 12.5 mg/ml significantly inhibited the growthof *Streptococcus pyogenes* (*p<0,05*).

Keywords*:* Methanol extract; *Mirabilis jalapa's* leaves*; Streptococcus pyogenes*

IRCPAS/2020/OP-139

Gum Arabic Coated Nanoparticles as DNA Plasmids Carrier

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Therapeutic gene editing is becoming a viable biomedical tool with the emergence of the CRISPR/Cas9 system. CRISPR-based technologies have promise as a therapeutic platform for many human genetic diseases previously considered untreatable. However, the successful use of CRISPR/Cas9-based gene editing for therapeutics requires efficient *in vivo* delivery of the CRISPR components, which remains challenging. Thus, for CRISPR-based therapies to be translated to the clinical setting, there is an urgent need to develop optimized carrier for its delivery. This study aimed to characterize gum arabic coated nanoparticles as CRISPR plasmids carrier. CRISPR plasmids encoding gRNA and Cas9 (tagged with red fluorescence protein) were amplified and extracted. CRISPR plasmids were encapsulated into gum arabic coated nanoparticle. The physiochemical properties of nanoparticles (size, zeta potential, encapsulation efficiency, cytotoxicity, and plasmids' stability) were characterized. The nanoparticles were delivered into HepG2 cells for gene transfection. Nanoparticles showed mean size, zeta potential 315 nm and -12 mV, respectively. Over 90% encapsulation efficiency was achieved and mostly supercoil conformation, ensuring DNA stability after encapsulation. MTT results suggest that NPs prepared in this study, were non-toxic and biocompatible. Transfection was confirmed by presence of red colour in the cells after 48 hr. Gum arabic coated nanoparticles appear to be good carrier for delivering plasmids.

Keywords*:* Gum Arabic; nanoparticles; DNA plasmids carrier

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Calcitriol Attenuates Kidney Fibrosis via Decreasing Tubular Injury, M1-M2 Macrophage Ratio, and Myofibroblast

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Chronic Kidney Disease (CKD) is the global health issues with bad prognosis outcome, characterized by kidney fibrosis. Unilateral Ureteral Obstruction (UUO) is the most representative experimental model to obtain kidney fibrosis. Kidney fibrosis observed with progressive injury of parenchymal kidney and extracellular matrix aggregation of type I and III collagen. This study was aimed to explore the effect of calcitriol administration toward the expression of M1-M2 macrophage, tubular injury and myofibroblast in male mice by using UUO. Twenty-five Switzerland furrowed mouse were divided into 5 equals groups: control

group (SO), UUO3, UUO7, UUOD3 and UUOD7. UUO groups received 0.2% ethanol and UUOD groups received 0.5 μg/kg BM calcitriol for 3- and 7-days exposure. Twenty-five paraffin-embedded section of kidney tissue were analysed by Periodic Acid Schiff and immunohistochemical staining against antibody anti-CD68, Arginase I and αSMA. Data were shown in the proportion of tubular injury, M1-M2 macrophage, and myofibroblast accumulation with imageJ software for scoring. Data were analysed using one-way ANOVA using SPSS22 software with $p<0.05$ considered to be statistically significant. UUO treated increased the tubular injury, M1 macrophage and myofibroblast accumulation but decreased M2 macrophage ($p<0.05$) compared to SO group. This study revealed the poorer prognosis in different exposure of treatment ($p<0.05$). But calcitriol (UUOD) administration to the treatment decreased the tubular injury, M1 macrophage and myofibroblast accumulation and increased M2 macrophage ($p<0.05$) compared to UUO groups. Calcitriol decreased chronic kidney disease by reducing tubular injury and M1-M2 macrophage ratio and inducing myofibroblast in mice.

Keywords:Calcitriol; UUO; M1-M2 macrophage ratio; tubular Injury; myofibroblast

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The Effect of Red Watermelon Rind Filtrate on the Increment of Calcium OxalateSolubility *In Vitro*

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More than 80% of kidney stones consist of calcium oxalate orcalcium phosphate. Watermelon is a fruit whose skin is thought to containpotassium of 82 mg and can dissolve kidney stones (urolithikum).Toexamine the effect of onceimmersion of red watermelon rind filtrate during sevendays at 37 °C to the percentage of calcium oxalate solubility. Theresearch method was an experimental research conducted*in vitro.* Theindependent variable was the concentration of red watermelon rind filtrate withthe immersion for seven days; the dependent variable was the solubility ofcalcium oxalate with seven days immersion. Potassium would eliminate calciumin oxalate calcium compound, forming a water-soluble compound. Theresults of *Anova One-Way Test* obtained an F count of 231.793 and $p = 0.000$, $\alpha = 0.05$, $p < \alpha$, so that H0 was rejected, so there was an increase in the solubility of Caoxalate in once-immersion frequency with filtrate of red watermelon rind forseven days with an average of 19.27% b/b. Filtrateof red watermelon rind at a concentration of 75% could increase the solubility ofcalcium oxalate by 19.24% by seven day-immersion, this was estimated to containpotassium. The calcium ($Ca2^{+}$) ion in the CaC2O4 compound is replaced by thepotassium (K^{+}) ion into the K2C2O4 compound that was soluble in water. Theoptimum concentration of red watermelon filtrate was 75% and the optimumweight of Ca oxalate was 0.5000 gram. The average solubility of calcium oxalatein red watermelon rind filtrate for seven days was 2.84, 5.53, 7.88, 12.27, 15.51,16.84, and 19.27% b/b, the greatest increment in the solubility of Calcium oxalatewas found on day 4 which was 4.39% b/b.

Keywords: filtrate of red watermelon rind; solubility of calcium oxalate; frequency of once-immersion

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A Production and Activity Test of Anti-Bacterial Compounds of Endophytic Fungi Br-S1 (A) Isolate Extract in Different General Growth Media

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BR-S1 (A) isolate is an endophytic fungus isolated from themedicinal plant of tea parasite (*Scurrula oortiana*) which is estimated to containanti-bacterial compounds. The research questions are as follow; can an antibacterialcompound be produced in general growth medium? And is it effective ininhibiting or killing methicillin-resistant *Staphylococcus aureus* (MRSA)pathogens? The aim of this research is to find out the types of generalgrowth media that can be used to produce anti-bacterial compounds and todetermine the effectiveness of these compounds in inhibiting/killing pathogenicMRSA bacteria. This research was conducted using laboratoryexperimental methods with the main variables in forms of three different types ofgeneral growth media, namely the *Potato Dextrose Broth* (PDB) medium, the*Czapek Dox Liquid Medium* (CDLM), and the *Malt Extract Broth* (MEB)medium. The results show that the three types of general growth mediawere not able to stimulate the production of anti-bacterial compounds which werecharacterized by the absence of discoloration and medium turbidity and theabsence of thick mycelium growth. The results of anti-bacterial activity tests onpathogenic MRSA bacteria show no inhibition zone formed around the disc paperadded with endophytic fungi extracts, whereas positive controls (*vancomycin*antibiotics) formed inhibitory/clear zones. Theproduction process of BR-S1 (A) isolate anti-bacterial compound was influencedby three factors, namely the composition and chemical properties of the medium,age and number (concentration) of cells, and environmental conditions(temperature and aeration) of the production site. It can be concluded that thethree types of general growth media of PDB, CDLM, and MEB cannot be used asa medium to produce anti-bacterial compounds. The antibacterialcompounds produced by BR-S1 (A) isolates are not effective in inhibitingpathogenic MRSA bacteria.

Keywords: Anti-bacterial compounds; endophytic fungi; general growthmedium

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Anti-Mycobacterium marinum Activity Screening of Some Ethnobotanically Selected Indonesian Plants that are used as Traditional Antituberculosis Drug

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Several plants have been utilised by Indonesian herbalist to treat tuberculosis symptoms such as cough and shortness of breath. However, despite being rich in biodiversity, very little attention has been given to the scientific search of antituberculosis activity of Indonesian medicinal plants. *M. marinum* has close relationship with *M. tuberculosis* and to the similarity of the disease in fish to the disease in humans including the characteristic persistence and granuloma formation. In this research, we screen the anti *M. marinum* activity of some ethnobotanically Indonesian plants that used as traditional antituberculosis drug that is expected to lead the discovery of new candidates of antituberculosis drug. Dried and ground plant material was extracted by maceration method with ethanol 96%. The in vitro activity of these extracts against *M. marinum* was carried out using microplate assay. The lowest

concentration effecting an inhibition of 90% was considered as the MIC. We tested 11 plant species. The result of this study showed that all extracts exhibit in vitro activity against *M. marinum* with varies minimum inhibition concentration (MIC_{90}) values. Ethanolic extract of Maja stem (*Aegle marmelos* Correa) has the most potential activity with MIC at 28.65 µg/ml, while rifampicin gives MIC at 166.35 µg/ml**.** *M. marinum* infected in zebrafish causes formation or organized caseating granulomas much like those found from tuberculosis in humans caused by *M. tuberculosis* infection. That is why many research groups using *M. marinum* as an alternative bacterial to study tuberculosis infection. All plant extracts tested have in vitro activity against *M. marinum* with varies MIC values. *These plants can* be used as promising source of novel antituberculosis substances from natural product.

Keywords: plant; Indonesia; tuberculosis; *Mycobacterium marinum*

IRCPAS/2020/OP-145

Xanthine Oxidase Inhibitor Activity of Dichloromethane Fraction, Ethyl Acetate Fraction of Ethanol Extract of *Nephelium lappaceum* L. Leaves

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The prevalence of hyperuricemia increases from year to year in various countries. In Italy, increased from 85.4/1,000 to 119.3/1,000 in 2005-2009. In Ireland from 2006-2014, increased from 19.7% to 25.0%. Allopurinol as drug that is commonly used causes various side effects, so it needs an alternative treatment based on natural ingredients that have higher safety. This study aims to determine the activity of ethanol extract, dichloromethane fraction, and ethyl acetate fraction *Nephelium lappaceum* L. leaves in inhibiting xanthine oxidase and identification of flavonoid compounds. Testing of xanthine oxidase was carried out using UV-Vis spectrophotometry (λ 295 nm) with allopurinol as a positive control. Flavonoid identification was carried out using TLC (cellulose as stationary phase and HOAc 30% as mobile phase) and spectral observations with NaOH 2M, NaOAc, NaOAc/H_3BO_3, $AlCl_3$, $AlCl_3$/HCl. The results showed that ethanol extract, dichloromethane fraction and ethyl acetate fraction of *N. lappaceum* leaves had activity as xanthine oxidase inhibitors, with the highest activity in ethyl acetate fraction (IC_{50} 17.506 µg/mL), then ethanol extract (IC_{50} 31,148 µg/mL), and dichloromethane fraction (IC_{50} 41,737 µg/mL) with allopurinol (IC_{50} 3,582 µg/mL). From the identification, it was found that ethyl acetate fraction of leaves *N. lappaceum* was a flavonol flavonoid and had a Rf value of 0.14 which same as quercetin.

Keywords: *Nephelium lappaceum* L. leaves; hyperuricemia; xanthine oxidaseinhibitory activity; flavonoids

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Antihypertensive, Antidiabetic and CytotoxicActivities of Indonesian Traditional Medicine

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Indonesian people have long used herbal medicine (jamu) to overcome various diseases, including hypertension and diabetes mellitus diseases. Hypertension and diabetes mellitus are two diseases which are directly related and requires proper and thorough management. The present study investigated the antihypertensive, antidiabetic, and cytotoxic activities ethanol extracts of Indonesian traditional medicine (jamu). Antihypertensive jamu was extracted by maceration using ethanol. Antihypertensive and antidiabetic activity investigated by measurement of ACE inhibitor and alpha-glucosidase inhibitor at a concentration ranging from 125-1000 µg/mL, respectively, by in vitro method. Cytotoxic evaluation of the extract was carried out using Brine Shrimp Lethality Test (BSLT).The results of measurements of ACE inhibitors and alpha-glucosidase inhibitory activity showed that herbal extracts had ACE inhibitors and alpha-glucosidase inhibitors with IC50 values of 292.15 µg/mL and 62.39 µg/mL, respectively. Ethanol extract of herbal medicine (jamu) exerts a cytotoxic effect on larvae of shrimp artemia salina with an IC50 value of 234.52 µg/mL. Hypertension in diabetic patients can increase both microvascular and macrovascular complications. Proper management of hypertension is needed to minimize the occurrence of complications and inhibit disease progression. Barriers to the renin-angiotensin aldosterone system by ACE inhibitors or angiotensin receptor blockers can increase glucose metabolism by preventing the formation of angiotensin II or preventing the activation of Angiotensin II receptors. The herbs used in this study consisted of Simplicia, namely *MorindaeFructus*, *Phyllanthi Herb*, *Centella Herb*, Zingiberis *rhizome*, *Imperata radix*, and *Alyxiae cortex*. These six plants contribute to their antihypertensive and antidiabetic activities.Toxic activity assay is one of the prerequisites for a plant to be developed as a drug, especially as an anticancer. Brine shrimp lethality test (BSLT) is asimple, inexpensive, non-aseptic, and high potentialitycytotoxicity test for bioactive compound andused as a preliminary test fordetermine the activity of a substance ora compound contained in apure extract or plant isolate. Herbal extract has antihypertensive and antidiabetic activity in vitro and cytotoxic effects.

Keywords: jamu; ACE inhibitors; alpha-glucosidase inhibitors; cytotoxic

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Emerging Pollutants in Water: A Threat for Sustainable Water Resource

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Emerging pollutants can be naturally occurring or synthetic that are not regularly monitored but has the potential to harm the environment and human health. These pollutants include and not limited to pesticides, pharmaceuticals, antibiotics, and personal health care. These pollutants are released to the environment at irregularly patterns or continuously in small

quantities that evaluating its accurate contamination is difficult. If measured, concentrations were alarmingly too high or nil. That often made the results inconclusive. This study assessed the emerging organic pollutants using passive samplers in the two river systems with headwaters to Mt. Kitanglad Mountain Range and drains toward Cagayan de Oro River Basin, Philippines. New water sampling technique using samplers were used instead of the traditional grab sampling method. The samplers used were silicone rubber and speedisk to measure respectively the hydrophobic and hydrophilic organic pollutants, Samplers were submerged in river water for uninterruptedly 34 days. Analytical instruments used were HPLC, GC-MS MS and LC-MS MS. The samplers were able to measure 105 organic emerging pollutants, which include mostly of pesticides, caffeine, traces of estrone and sulfamethazine. Trace concentrations of organic compounds that were either banned or restricted by the government for more than a decade ago were also measured. The number of emerging organic pollutants measured showed the effectiveness of the sampling method used. Effective methodology, like the passive sampling method, is necessary to have a reliable water quality assessment. Regular and reliable water quality evaluation is essential to attain a sustainable water resource.

Keywords: Cagayan de Oro River Basin; emerging pollutants; passive samplers; water quality

IRCPAS/2020/OP-155

Evaluation of*In Vivo* Anti-Inflammatory Activity of Biogenic Silver Nanoparticles in Male Sprague Dawley Rats

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In the recent past, plants mediated intra and extracellular synthesis of metal nanoparticles, due to their advantages over chemical and physical approaches, has attained tremendous interest from scientists.Various plants and different parts of plants were exploited in the fabrication of metal nanoparticles.Recent years, many researchers successfully synthesized metal nanoparticles using marine sources. In the present research, biosynthesis of silver nanoparticles (AgNPs)using *Halimeda gracilis* (HG), characterization and evaluation of anti-inflammatory potential of AgNPs were performed.An aqueous extract of HG was prepared and mixed with 1 mM silver nitrate solution. The reddish brown colour indicated the formation of AgNPs and it was characterized by UV-Visible spectrophotometer, Fourier Transform Infrared Spectroscopy (FT-IR), Dynamic light scattering (DLS), Field Emission Scanning Electron Microscopy (FE-SEM), High Resolution Transmission Electron Microscopy (HR-TEM), Energy dispersive X-ray Spectroscopy (EDAX), X-ray diffraction (XRD) and X-ray photoelectron Spectroscopy (XPS).*In vivo* anti-inflammatory activity of the AgNPswas evaluated by carrageenan induced paw edema in male Sprague Dawley rats. In this we have reported the eco-friendly, economic, and green phyco-fabrication of AgNPs from *Halimeda gracilis*. Various characterization studies revealed the size, shape, stability, and crystalline nature of the particles formed. In vivo study results showed that AgNPs were showed comparable anti-inflammatory activity with that of the standard. The outcome of the study indicated that AgNPs synthesized using *Halimeda gracilis* possess significant anti-inflammatory properties and a suitable formulation can be made from AgNPs for the treatment of inflammation.

Keywords: *Halimeda gracilis*; Silver nanoparticles; characterisation; *In vivo* anti-inflammatory activity

IRCPAS/2020/OP-162

Development of plasmids for compatible gene expression in cyanobacteria

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Cyanobacteria is defined as an emerging microbial host that is convenient for eukaryotic transgene expression. However, there are limited studies on expression plasmids for cyanobacteria. Broad host-range (BHR) plasmids have been suggested to be able to replicate and maintain transgenes in multiple microbial hosts.In this study we designed and constructed expression plasmids with Gateway destination cassette specifically for cyanobacteria. *Chalcone synthase* (*CHS*) was cloned into the expression plasmids prior to transformation of *Escherichia coli* (*E. coli*). The positive transformants indicated that the plasmids are functional and stable for downstream transgenic expression. A web service, CYANO-VECTOR Assembly Portal (http://golden.ucsd.edu/CyanoVECTOR/) was used to design the expression plasmids. The parts of SpSm antibiotic resistance marker, Km/Nm antibiotic resistance marker, cPtrc-gateway destination cassette, S7942NS1-RK2BOM integration site and RSF1010 BHR replicon were obtained from pCVD002, pCVD003, pCVD016, pCVD019 and pCVD046 devices, respectively by using either EcoRV-HF or ZraI. The quality of all parts was analysed using agarose gel electrophoresis prior to nucleotide sequencing. The parts were assembled using seamless cloning method and transformed into *E. coli* DH5 Alpha strain. The positive transformants were confirmed using polymerase chain reaction (PCR) colony method and nucleotide sequencing. Four expression plasmids were obtained; pCyano01 and pCyano02 consist of BHR replicon for transgenic replication in cyanobacteria whereas pCyano03 and pCyano04 possess an integration site NS1 for heterologous DNA sequences insertion into neutral site 1 of *Synechococcus elongatus* PCC7942 chromosome. In addition, the expression plasmids also carried either SpSm (spectinomycin and streptomycin) or Km/Nm (kanamycin or neomycin) resistance genes. Four plasmids that are compatible for gene expression in cyanobacteria have been successfully transformed into *E. coli*. Cloning of *chalcone synthase* (*CHS*) gene into the expression plasmids would allow plant transgenic genes to be compatibly expressed in cyanobacteria. This study will provide knowledge on application of BHR plasmids for metabolic engineering of eukaryotic genes into microbial hosts.

Keywords: Cyanobacteria; Broad Host Range (BHR); BHR replicon; Integration site; Expression plasmids; Chalcone synthase (CHS)

IRCPAS/2020/OP-171

Counteraction of Toxicity Prompted by *Vipera russelli* Phospholipase A_2 by crude *Turbinaria ornate,* brown seaweed and purified fucoidan: A comparative study

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Fucoidan is a sulfated polysaccharide found in brown algae and are found in the extracellular matrix of brown algae. Fucoidans have various biological activities, including antiviral,

anticoagulant, anti-inflammatory, immunomodulatory, anti-angiogenic, and anti-adhesive. Natural inhibitors have the potential to neutralize the toxic effects caused by snake venom proteins and enzymes. It has been well recognized for several years that animal sera, some of the plant and marine extracts are the most potent in neutralizing snake venom phospholipase A_2 (svPLA$_2$). There is an overview of the role of PLA$_2$ in inflammation that provides a rationale for seeking inhibitors of PLA$_2$ as anti-inflammatory agents. However, more studies should be considered to evaluate antivenom efficiency of sera and other agents against a variety of snake venoms found in various parts of the world. PLA$_2$IIA was purified from human synovial fluid (HSF) and *Vipera russelii* venom (VRV) The aim of this study is to determine anti-inflammatory effect of crude *Turbinaria ornate*, a brown seaweed and a Sulphated Polysaccharide (Fucoidan). 100 µg of both crude and isolated compound were tested against HSF-PLA$_2$ activity. Both inhibited PLA$_2$ activity; however, inhibition by purified compound was higher than crude. Hence purified compound was further tested with different doses. The dose dependent inhibition study showed that purified compound inhibited HSF-PLA$_2$ activity with an IC_{50} value of ~ 12 µg. Ascorbic acid-6-palmitate (AP), a known inhibitor of PLA$_2$ was taken as positive control. The results revealed that the isolated and purified Fucoidan has potential anti-inflammatory activity than the crude *T.ornate* powder.

Keywords: Brown Seaweed, sulphated polysaccharide, PLA$_2$, *Vipera russelii* venom, anti-inflammation

IRCPAS/2020/OP-181

Anti-Osteoporotic Effects of Alendronate and Sitagliptin in STZ Induced Type 2 Diabetes Mellitus in Ovariectomized Rats

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Diabetes mellitus is a metabolic disorder identifies as hyperglycaemia and osteoporosis is a bone disorder within which quality of bone and bone mineral density decline. Diabetes in osteoporotic patients is remarkably increased risk of bone fracture. Alendronate, a bisphosphonate first line therapy for osteoporosis treatment which prevents bone fracture by inhibiting osteoclast. Sitagliptin, an oral antidiabetic agent used for the treatment of type II diabetes mellitus by inhibiting Dipeptidyl peptidase-4 activity. Sitagliptin may regulate bone homeostasis by inhibiting osteoclast & suppressing osteoclast differentiation. The present study was to investigate the anti-osteoporotic effect of sitagliptin and alendronate on bone mechanical properties in STZ induced diabetes in ovariectomized rats.30 female Wistar rats weighing from 180-250 g were divided into five groups each have 6 rats. Osteoporosis was induced by bilateral ovariectomy. After seven days of surgery the type 2 diabetes mellitus was induced by single intraperitoneal injection of STZ (50 mg/kg) and nicotinamide (110 mg/kg). Groups III, IV and V were treated with alendronate (3 mg/kg), sitagliptin (30 mg/kg) and combination respectively for 42 days.The body weight of sitagliptin (30 mg/kg) and concurrent administration of alendronate (3 mg/kg) and sitagliptin showed significant increased compared to OVX-STZ groups. There is improvement in serum calcium and ALP in sitagliptin, alendronate and combination groups compared to OVX-STZ groups. Alendronate and combination groups showed significant increase in bone weight compared to STZ-OVX group. There is no significant changed in bone length and diameter. The bone mineral mass was increased in alendronate and combination groups compared to OVX-STZ groups. The

concurrent administration of alendronate and sitagliptin showed beneficial effects in STZ induced type II diabetes mellitus in ovariectomized rats.

Keywords:anti-osteoporotic; alendronate; sitagliptin; type 2 diabetes mellitus

IRCPAS/2020/OP-183

Physicochemical Characterization and Application of Betacyanin Pigment from Red Dragon Fruit as Tracking Dye for Gel Electrophoresis

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Red dragon fruit (*Hylocereus lemairel* (Hook.) Britton and Rose) contains betacyanin pigment. Betacyanin (6-O-3-hydroxy-3-methyl-glutaryl)-betanin) is red pigment that has many benefits in biotechnology.The aim of this study was to characterize physiochemically, to measure the stability and to apply betacyanin as a natural dye for tracking dye DNA identification. Extraction was carried out by water using Ultrasonic Assisted Extraction (UAE) method which is sonicated at 50 kHz for 30 minutes at 25°C. Extract was freeze dried for 48 hours. The dried extract was characterized and purified by using preparative TLC and physico-chemically analysed using UV-Vis and FTIR spectrophotometers. This study indicated that betacyanin was found at Rf value of 0.6 same as the betacyanin standard. The maximum wavelength was obtained at 534 nm and the IR spectra showed similarity with betacyanin standard with the same functional groups between 4000-600 cm^{-1}although there was a slight shift in the wavenumber but it still in the range. The stability studies were stable at temperature below 40 °C and at range pH 4-6. The effectiveness of betacyanin as a tracking dye was checked. This study revealed that betacyanin from red dragon fruit can be a potential alternative to synthetic dye used in the loading dye preparation for agarose gel electrophoresis. The method used ultrasonic which can cause a cavitation effect to break down the cell wall and betacyanin is released out easily thus down the cell wall and betacyanin is easily released out thus maximizing extraction results. The drying method used is freeze drying to remove the water solvent. This method aims to maintain sample quality because betacyanin is not stable to heating. From this study, betacyanin was characterized and compared with betacyanin standard. All parameters showed similar type with betacyanin standard. Betacyanin was also used for dying in analysis DNA using Gel electrophoresis and the result showed that betacyanin can be used for tracking in DNA analysis.

Keywords: Betacyanin; dragon fruit; gel electrophoresis; natural dye

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Ruthenium (II) polypyridyl complex, $[Ru(dppz)_2PiP]^{2+}$ synergized with PARP inhibitor for breast cancer treatment

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PARP inhibitor Olaparib has been approved by FDA as chemotherapeutic drug for breast cancers in recent years. Despite its proven therapeutic efficacy, this class of drug is limited to

BRCA1/2-deficient breast cancers and remains ineffective against breast cancers harboring wild type (WT) BRCA genes. Ruthenium(II) polypyridyl complex, $[Ru(dppz)_2PiP]^{2+}$ (DPPZ: dipyridophenazine, and PIP: 2-phenylimidazo[4,5-f][1,10]phenantroline) or Ru-PIP has demonstrated its role in causing DNA damage where upon the addition of Ru-PIP,the DNA replication fork progression in human cancer cellis immediately stalled leading to the activation of DNA damage response (DDR) pathways. This has prompted us in exploring the combination of RPC with PARP inhibitor Olaparib as new promising anticancer strategy. The present study is aimed to investigate the mechanism of drug action from the combination of Ru-PIP and olaparib in BRCA-WT human breast cancer cells.The impact of Ru-PIP with and withoutolaparibon MDA-MB-231 and MCF7 breast cancer cells growth was determined by MTT assay and the combination indices (CI) were calculated using the established method by Chou and Talalay. Breast cancer cellssurvival ability was investigated using clonogenic survival assay and the potential cytotoxicity mechanisms were assessed by cell cycle analysis and Annexin-V-FITC assay. Ru-PIP or Olaparib when used as single agents led to dose- and time-dependent decreases in viability of breast cancer cells. Furthermore, Ru-PIP/olaparib combinations gave synergistic effectsrepresented by CI < 0.8 and almost a total loss in clonogenic potential of breast cancer cells was observed when treated with Ru-PIP/Olaparib combination. Combination treatment also caused an increase in G2/M arrest and apoptotic cell death in comparison to control or when treated with single agents. Importantly, mild impact on the viability of normal human lung fibroblasts NHDF cells was observed for any combination tested.These findings demonstrate new promising therapeutic strategy of drug combination while reducing the adverse effects on healthy cells to overcome the limited clinical option for cancer treatment.

Keywords: *Ruthenium Polypyridyl Complex;* PARP inhibitor; Olaparib;synergistic effect; breast cancer

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Effect of Ethyl Acetate Extract of *Penicillium citrinum* from *Xestopongia testudinaria* on Blood Glucose Level, Insulin Concentration, and Homeostatic Model Assessment of Insulin Resistance (HOMA-IR)

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Insulin resistance is defined as the impaired ability of insulin to promote glucose uptake and exert its metabolic effects in its target tissues (liver, skeletal muscle, and adipose tissue). Insulin resistance plays a major pathophysiological role in type 2 diabetes. In recent years, there has been growing interest in searching for new bioactive compounds to treat type 2 diabetes, including metabolites from endophytic fungi from a marine sponge. The study aims to investigate the effect of ethyl acetate extract of *Penicillium citrinum* from *Xestopongia testudinaria* on blood glucose level, insulin concentration, and homeostatic model assessment of insulin resistance (HOMA-IR). Animals (*Mus musculus*) were divided into 5 groups i.e. normal group, negative control group, and treated groups received ethyl acetate extract of

P.citrinum dose 100, 200 and 400 mg/kg bw. Animals were induced by alloxan 175 mg/kg bw, s.c. After 7 days of experiment and treated with extract, blood glucose level, insulin concentration, and HOMA-IR were measured. Ethyl acetate extract of *P. citrinum* from *Xestopongia testudinaria* showed significant ($p<0.05$) decreased blood glucose level, increased insulin concentration, and decreased HOMA-IR as compared to the normal control group. A decreased blood glucose level, increased insulin concentration and decreased HOMA-IR by the ethyl acetate extract of *P. citrinum* caused by bioactive compounds contained in extracts such as alkaloids, phenolics, terpenoids, and steroids. Based on the research results can be concluded that ethyl acetate extract of *P. citrinum* from *Xestopongia testudinaria* is the potential to developed as an antihyperglycemic agent.

Keywords: *Penicillium citrinum; Xestopongia testudinaria*; blood glucose; insulin; HOMA-IR

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Antioxidant Potential of Fraxetin Ameliorates Lipid Anomalies and Inflammatory Cytokines in High Fat Diet Induced Hypercholesterolemic Rats

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Hypercholesterolemia is closely associated with atherosclerosis, which is the principal cause of mortality in world population. Hypercholesterolemia is characterized by increased serum concentrations of low-density lipoprotein (LDL). Accumulation of oxidized LDL leads to atherosclerotic plaque formation which contributes to stroke, myocardial infarction, and cardiovascular diseases (CVDs). It is well known that the hypocholesterolemic drugs are effective in lowering LDL, but the long-termconsumption causes adverse effects such as liver and muscle injuries, rhabdomyolysis, myopathy and acute renal failure. Thus, the investigation and usage of natural products from plant origin in treating various diseases including CVDs have gained much attention. The present study was aimed to explore the antihyperlipidemic and anti-inflammatory effect of fraxetin on high fat diet fed rats. Hypercholesterolemia was induced by the diet comprising of normal rat chow 84.3%, lard 5%, yolk powder 10%, cholesterol 0.2% and 0.5% bile salt were fed to the rats for the period of 8 weeks. The results showed that abnormally elevated levels of plasma lipid profiles. Three different doses of fraxetin (25, 50 and 100 mg/kg b.w/day) were administered orally to hypercholesterolemia suffering rats for the period of 30 days. Among these three doses offraxetin, the dose 100 mg/kg b.w. was significantly decreased the plasma lipid profiles when compared to other two doses. The effect produced by fraxetin(100 mg/kg b.w) was comparable to that of simvastatin (10mg/kg b.w). Therefore, 100 mg/ kg b.w was fixed as an effective dose and used for further analyses. Fraxetinadministration reinstated the elevated levels of lipid peroxidation markers and decreased levels of enzymic and non-enzymicantioxidants in the cardiac tissue of hypercholesterolemia suffering rats.In addition, fraxetinadministration reinstated the altered levels of inflammatory and anti-inflammatory markers in the plasma and cardiac tissue of hypercholesterolemia suffering rats.These findings suggest that the administration of fraxetinwas potentially ameliorated the lipid anomalies and inflammatory markers through its enhanced antioxidant potential. The result obtained from these studies, it may be concluded that fraxetin can replace the commercially availablestatin- drugswhich could lead to reduction intoxicity and side effect caused by later as well as reduces the secondary complications.

Keywords:Fraxetin; inflammatory markers; hypercholesterolemia; lipid profiles; antioxidants

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Preparation and Evaluation of Membrane Usnic Acid: A Preliminary Study

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Usnic acid, a yellow crystal of secondary metabolite of Usnea sp., has been known for its anti-inflammatory and antibacterial activities. However, the utilization of usnic acid is still limited because of low solubility in water. The purposes of this study were to prepare membrane contained usnic acid and evaluate its effectiveness for burns healing. Usnic acid was modified with PVP K-30 (1:2 w/w) in solid dispersion (SD) system by freeze drying methodto increase its solubility in water. Usnic acid in solid dispersion system at concentration 0.5% (F1), 1% (F2), and 2% (F3) were formulated into membrane using poly-vinyl alcohol (PVA) as the gelling agent, glycerin as the plasticizer and distilled water. The prepared membranes were evaluated for the physical and mechanical properties including appearance, thickness, tensile strength, percentage of elongation, and Modulus Young's. The effectiveness of burn healing activity was conducted by creating superficial burns on male white rabbits,aged 4 -5 months and weight 2–2.5 kg and evaluated for 21 days. The appearance of each membrane was transparent, but the color was quite different due to the amount of usnic acid. The thickness of each membrane was less than 0.1mm. F2 had the highest tensile strength, percent of elongation, and Young's Modulus. Meanwhile, F3 showed a better result in burn healing compared to F2 and F1. Usnic acid solid dispersion membrane had proper physical and mechanical properties. The burn healing experiments showed application of 2%usnic acid solid dispersion in membrane was the most effective among all test groups in burn healing effectiveness ($p<0.05$).

Keywords: usnic acid; solid dispersion; membrane; burn healing

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Study on Wound Healing Activity of *Cynoglossum Zeylanicum*

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Medicinal plants play an important role for the source of novel drug discovery for the treatment of wound. *Cynoglossum Zeylanicum* (CZ) belongs to the family Boraginaceae is an indigenous medicinal plant. The aim of the present study was to evaluate the wound healing activity of different extracts of *Cynoglossum zeylanicum* whole plant using excision wound model in wistar albino rats. The rats were divided in to six groups of six rats each. Group 1 was served as control, group 2 was treated with standard povidone iodine, group 3, 4, 5, and 6 were treated chloroform, ethyl acetate, ethanol and water extract respectively for 20 days. On the 16th post wounding day, the fastest healing of the wound was observed in animals which received ethyl acetate extract of CZ (100% wound contraction) as compared with standard 5% povidone

iodine ointment (91.2% wound contraction on 16th post wounding day). The results found that th e rate of wound contraction in ethyl acetate extract of CZ treated animals shows more potent results compared to standard and other tested extracts of CZ. This study concludes that the *Cynoglossum Zeylanicum* ethyl acetate extract possess potent molecule for would healing activity.

Keywords: *Cynoglossum zeylanicum*; ethyl acetate extract; wound healing study

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Alteration of Mitochondrial Gene Expression in Cervical Cancer Cells by Induction of Oxidative Stress

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Many chemicals in nature can modulate mitochondria gene expression. This event occurs when a biological system alters in response to a change in the level of a reactive oxygen species (ROS). It was believed that many polyphenols induce oxidative stress and alters the expression of the mitochondrial genes. Changing the expression of mitochondrial genes can also activate the intrinsic apoptotic pathway and lead to cell death. In our study, five compounds were isolated by means of activity-guided fractionation from *Symplocos cochinchinensis* (lour.) Moore. Further FTIR, ^{13}CNMR, ^{1}HNMR and LCMS were used to characterize in accordance with three known compounds (Ellagic acid, Damphetamide, Odoroside) and two new compounds (RD1 and RD2). The prooxidant status of RD1 and RD2 were examined using the extent of deoxyribose degradation assay, Cu (II) reduction and treatment of λ phage DNA with compounds and restriction enzyme digestion. RD2 in cervical cancer cells has been improved in terms of production of oxidative stress. Subcellular redox regulations for mitochondria were then screened for RD2. Nuclear apoptosis staining, Nuclear condensation using AO/EtBr, and mitochondrial membrane potential damage study showed the changes in redox regulation signals in cervical cancer cells. The cell death induced through apoptosis was also confirmed by micro fragmented DNA. Finally, the modification of subcellular redox regulations and cell death was validated with semiquantitative RTPCR for various prooxidant biomarkers, and protein expressions with Western blot analysis. The RD2 induced the apoptosis with the close association of down regulating BCL_2, Up regulation of BAX and activation of Caspase3 in Hela cells. These findings offer a fresh compass for future studies on RD2 based drug developments against cervical cancers.

Keywords: Oxidative stress; prooxidants; apoptosis, Redox signalling; *Symplocos cochinchinensis*

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PARP Inhibition Enhances the Anticancer Activity of Ruthenium(II) Polypyridyl Complex, $[Ru(Dppz)_2pip]^{2+}$ in Lung Cancer Cells

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Most ruthenium (II) polypyridyl complexes (RPC) have been designed to mimic platinum-based chemotherapyand in the last decades, several of them have been clinically studied as promising anticancer agents. In our previous study, we have demonstrated the role of $[Ru(dppz)_2PiP]^{2+}$(DPPZ:dipyridophenazine&PIP:2-phenylimidazo[4,5][1,10]phenantroline) or Ru-PIP installing the replication fork progressionleading to DNA double-strand breaks (DSBs). And in response to this DNA replication stress, DNA damage response (DDR) pathways are activated. PARP inhibitors (PARPi) are a part of DDR signalling and several of them have recently passed clinical trials and become FDA-approved chemotherapeutic drugs including olaparib.The present study is aimed to evaluate the use of the RPC; Ru-PIP in combination with olaparib as new therapeutic strategy. The cytotoxic effects of treatment with Ru-PIP and/orolaparibon A549 cells growth was determined by MTT assay and using the established method by Chou and Talalay, the combination indices (CI) were calculated to interpret the interactions between the two agents. A549 cellssurvival ability was investigated using clonogenic survival assay and the potential cytotoxicity mechanisms were assessed by cell cycle analysis and apoptosis assay.Herein, we reported that when used as single agents, both agents caused mild impact on A549 cells even after 72 h treatment. In addition, RuPIP was able to effectively synergize with olaparib in inducing growth inhibitory effect on A549 cells and synergy was further observed where almost a total loss in cells clonogenicity was observed when treated with Ru-PIP/olaparib combination. The selected synergistic combination was found to enhance G1/S cell cycle arrest and result an increase in apoptotic cell death in comparison to single agents alone. These results established that the ruthenium(II) polypyridyl metallo-intercalator, Ru-PIP showed potent synergy with PARP inhibitor olaparib in A549 cells and merit further investigation in pre-clinical and clinical studies with the potential to replace current platinum-chemotherapy for lung cancer treatment.

Keywords: Ruthenium Polypyridyl Complex; PARP inhibitor; Olaparib; synergistic effect; lung cancer

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Oxidation of Propane-1,3-diol (Non-Vicinal) by Potassium Permanganate in Aqueous Medium

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The oxidation of propane-1,3-diol by potassium permanganate in aqueous solution has been studied at λ_{max} 525 nm. The rate of the reaction has been found to increase with the increase in [$KMnO_4$] and [Propane-1,3-diol]. The reaction shows first-order dependence both on [$KMnO_4$] and [Propane-1,3-diol] and independent on the ionic strength of the solution. The $\Delta H^{\#}$(kJ mol^{-}

[1]), $\Delta S^{\#}$(kJK^{-1}mol^{-1}) *and* $\Delta G^{\#}$(kJ mol^{-1}) were 24.98, -0.22 and 90.50 respectively. Negative entropy of activation revealed an ordered transition state for the reaction. Spectroscopic studies showed the product of the reaction to be 3-hydroxyl-propanal. A plausible mechanism in consonance with spectroscopic studies and the kinetic result was proposed.

Keywords: propane-1,3-diol; spectroscopy

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Temperature-Dependent Production and Characterization of Hydroxyapatite (HAP) Sorbent from Periwinkle Shell

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Production of hydroxyapatite from biogenic wastes for the adsorption of Contaminants from wastewater has become attractive in recent times. Hydroxyapatite was produced from periwinkle shell (PSHAP) at different temperatures (400, 500 and 600 °C). The periwinkle shell was washed with distilled-deionized water, sundried for 48 h, soaked in 50 % hydrogen peroxide for 24 h, washed copiously with distilled-deionized water, dried in an oven at 105 °C, calcined at different temperatures (400, 500 and 600°C) for 2 h and ground to granules before soaked with 0.26M diammonium hydrogen phosphate solution for 24 h. Calcined PSHAP was oven-dried at 105 °C for 24 h and sieved to 2 mm mesh sizes. PSHAP was characterized using Fourier Transform Infrared (FTIR), Scanning Electron Microscopy (SEM), Energy Dispersive X-ray (EDX) and X-ray Diffraction (XRD) techniques. The FTIR analysis of the hydroxyapatite was characterized by broadband at 3437 cm-1, representing bonded -OH groups. The medium and sharp peaks at 2513.25 cm-1 for calcined PSHAP at 400, 500 and 600 OC correspond to S-H (stretch). The peaks at 1799.59, 1797.66 and 1797.66 cm-1 respectively correspond to the C=O stretch. Similarly, SEM analysis shows the PSHAP was effective in creating well-developed pores on the surface of the PSHAP, leading to PSHAP with a large surface area and porous structure. EDX spectra of PSHAP indicated that calcium, phosphorus, and oxygen are the significant elements present in the samples. XRD crystal structures of PSHAP had 25, 23 and 24 distinct peaks and percentage crystallinity is 80.4, 77.5 and 81.7 for the PSHAP calcined at for 400, 500 and 600 °C respectively. This indicates that the PSHAP500 °C with lowest percentage crystallinity and intensity had roughest surface suggesting highest adsorptive characteristic.

Keywords: Adsorption; contaminants; Periwinkle shell; production; characterization; Hydroxyapatite

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QBD Based Novel Combinational Nanotransferosomes of Pioglitazone and Eprosartan Mesylate: Pharmacokinetic and Pharmacodynamic Studies

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Diabetes mellitus is a metabolic disorder with multiple etiologies. Type 2 diabetes is also allied with an augmented risk of precocious death due to cardiovascular disease (CVD) where hypertension is a major risk factor. Present medication systems for the treatment of such chronic coexisted diseases are troublesome and inopportune to overcome the side effects by complex therapeutic regimen and to abridge the treatment regimen. Therefore, investigations are desired to deliver Antidiabetics and Antihypertensives using novel delivery approaches followed by their commercialization. The present exploration was aimed to develop and optimize a combinational delivery of Antidiabetics and Antihypertensives Nano transferosomes using Design Expert software Version 11.0 followed by pharmacokinetic and pharmacodynamics studies.In this Nano transferosomes were prepared containing two drugs and optimized using Box-Behnken design by taking ratio of Phospholipon® 90G (X1) and Surfactant (X2), ratio of solvents (X3) and sonication time (X4), each at three levels, were selected as independent variables, while characterization such entrapment efficiency (Y1), (Y2) and flux (Y3), (Y4) of two drugs Pioglitazone and Eprosartan Mesylaterespectively was chosen as dependent variables. The finest formulation was selected by point prediction method. The optimized formulation was further evaluated for SEM, TEM, Zeta sizer, *in vitro* drug release,*in vitro* drug permeation, *in vivo* pharmacokinetic and pharmacodynamic study. Optimized formulation shows entrapment efficiency and flux values which agrees with the predicted values generated by design.The pharmacokinetic study presented that transdermal nano transferosomal gel formulation showed improvement in bioavailability of two drugs with respect to the control formulation. Pharmacodynamic study confirmed the better and prolonged management of diabetes and hypertension after the application of nano transferosomal gel in experimentally induced diabetic and hypertensive Wistar rats as compared with oral control formulation.The inquisitiveness to develop combination of antidiabetic and antihypertensive drugs with special attention to the lipid based nanoparticulate system is to emerge gradually to overcome the problems associated with the multiple treatment regimen and to prevail the confidence of end users towards the higher acceptability. It was concluded that these findings suggested that nano-transfersomal transdermal delivery aimed for both activities have been successfully developed.Thus, these combinations can be explored in future to develop a rational therapy regimen to treat especially hypertensive diabetic patients.

Keyword: Box-Behnken; combination therapy; design expert; flux; hypertensive and diabetic therapy

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Toxicological Evaluation of Column Fractions of Ethanol Leaf Extract of *Ziziphus mauritiana.*

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Different plants may contain varying amount of phytochemical and different degrees of toxicity. This study aimed at evaluating the phytochemical present in the most active column fraction of ethanol leaf extract of *Ziziphus mauritiana* plant and their toxic effects using brine shrimp lethality assay and animal model. The column chromatography experiment exhaustively macerated *Ziziphus mauritiana* leaves revealed 42 fractions pooled into seven fractions. Fraction three (3) was the most toxic with the brine shrimp lethality assay of (31.48 ug/ml) and it toxicological evaluation revealed adverse effect on haematological parameter, biochemical indices and histo-architecture of the liver and kidney of the experimental model studied. The LCMS analysis of the most toxic fraction revealed presence of Antirrhinoside, Lucidumol A, Apigenin 7-glucuronide-4'-(6"-malonylglucoside), Dioscoreside C, Camellioside D and others which have been reported for various pharmacological effects including adverse effects, the mode of toxicity maybe synergistic, individual or antagonistic which may explain moderate toxicity observed in the animal model.

Keywords: Dioscoreside C; chromatographic fractions; *Ziziphus mauritiana*; toxicity

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Phytochemical and Toxicological effect of Alkaloid Fraction of Fresh Ripe Fruit of *Dennettia tripetala* (Pepper fruit) in Albino Rats.

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Toxicological effect of the alkaloid fraction from *Dennettia tripetala* (pepper fruit) was studied using albino rat model. The result of phytochemical study showed the presence of alkaloids (4.8%), saponins (0.8%), flavonoids (3.6%), and tannins (1.2%). The median lethal dose of the alkaloid fraction showed no mortality or sign of toxicity at a dose of 5000 mg/kg body weight in mice. In the sub-acute toxicity, there was significant ($p < 0.05$) increase in percentage body weight of the treated rats compared to that of the control group after 28 days of oral administration of the alkaloid fraction, and there was also significant ($p < 0.05$) difference in relative organs body weight in liver, kidney, heart and spleen of the treated rats. There were elevations in levels of aspartate aminotransaminase, alanine aminotransaminase and alkaline phosphatase activities in groups 4, 5 and 6 animals treated with 100, 150 and 200 mg/kg of the fraction on days 14 and 28 ($p < 0.05$). There were significant ($p < 0.05$) decrease in total protein, albumin and potassium ion, while the creatinine, urea, bilirubin and sodium ion concentrations significantly ($p < 0.05$) increase in animals treated with high concentration of the fraction at both 14 and 28 days compared with the control group. The concentration of malondialdehyde showed significant ($p < 0.05$) increases in groups 4, 5 and 6 rats at days 14 and 28 compared

with the control. There were observed changes in the activities of catalase and superoxide dismutase on days 14 and 28 in all the treated groups. Vitamin C showed decrease on day 14 but showed significant ($p < 0.05$) decrease in groups 3, 4, 5, and 6 animals on day 28. The levels of glutathione, glutathione peroxidase and vitamin E were significantly ($p<0.05$) lowered in groups 3, 4, 5 and 6 on days 14 and 28 compared to the control. There were increases in the serum phospholipid, total cholesterol, triacylglycerol, and low-density lipoproteins concentrations in all the test animals compared to the control after 14 and 28 days. There was also no significant ($p > 0.05$) decrease in the concentration of high-density lipoprotein across the treated groups compared to the control. Tissues of liver and kidney from group 3, 4, 5 and 6 animals showed histological changes consistent with hepatotoxicity and nephrotoxicity. From the results of this study, there is compelling evidence that alkaloid fraction from this fruit might be safe when used as an oral remedy at doses below 100 mg/kg as it has the potentials of being hepatotoxic, nephrotoxic, and might result in death of animals at doses above 200 mg/kg concentrations when administered for longer period.

Keywords: *Dennettia tripetala*; phytochemical; toxicological

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Molecular Docking and QSAR of Pyrrolone Derivatives Against *P. falciparum*

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Malaria being one of the public health problems that afflicts more than 100 countries.The current study aimed at building robust and rational Genetic function approximation (GFA) based on Quantitative Structure-Activity Relationship (QSAR) models and molecular docking studies of pyrrolone derivatives antimalarial agents. In this study, a GFA - QSAR analysis was performed on a data set of forty-nine pyrrolone derivatives antimalarial agents against *P. falciparum*. Forty-two molecules were used as a training set and seven as the test set. The molecules were optimized by Density Functional Theory (DFT) using Becke's three-parameter Lee-Yang-Parr hybrid functional (B3LYP) in combination with 6-31G** basis set. The QSAR models were built using Genetic Function Algorithms (GFAs) method. The model with the best statistical significance ($N = 42$, $R^2_{ext} = 0.700$, $R^2 = 0.933$, $R^2a = 0.916$, $Q^2cv = 0.894$, LOF = 0.417) and Minimum experimental error for non-significant LOF (95%) = 0.250 was selected. The docking experiment was carried out using AutoDock Vina of PyRx and Discovery Studio Visualizer. Docking analysis revealed that three of the studied compounds with binding affinity values of -10.7 kcal/mol, -10.9 kcal/mol and -11.1 kcal/mol possess higher potency than standard antimalarial drugs such as Artemisinin with binding affinity values of -8.2 kcal/mol, -8.1 kcal/mol and -7.9 kcal/mol.It is envisioned that the wealth of information provided by the QSAR and molecular docking results in this study will offer important structural insights for further laboratory experiments in the future design of novel and highly potent antimalarial from pyrrolones.

Keywords: Genetic function approximation; Density Functional Theory; B3LYP; AutoDock Vina

IRCPAS/2020/OP-219

Analysis of Rat Adulteration in Beef Meatball Using Fourier Transform Infrared Spectroscopy and Gas Chromatography-Mass Spectrometry for Halal Authentication

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Beef meatballs are one of the favorite food products that are consumed by many people in Indonesia. Beef is very expensive in the Indonesian market compared to other common meats types such as chicken and lamb. This situation encourages unethical meatball producers to replace or adulterate beef with lower-priced meat like rat meat. This study determines the capability of the FTIR method combined with chemometrics and GCMS method for identification and quantification rat in beef meatball. The lipid fraction of meatball was obtained by extraction technique, namely blight and dyer method. Lipid extracts obtained from meatball were scanned using FTIR spectrophotometer at 4000-650 cm^{-1}. Lipid derivatization to form methyl ester compound was carried out in GCMS method. PCA at combined wavenumber regions of 3010-2850 cm^{-1} and 1250-1100 cm^{-1}could identify rat meat in meatball. These wavenumbers were also used for quantitative analysis rat meat in meatball using PLS model. Rat lipid that analysed by GCMS contains three major fatty acids namely methyl trans-9-octadecenoic**,** hexadecanoic acid, and 9-hexadecenoic acid**.** The small variations among spectra were exploited as a basic tool to differentiate between rat meat and other meat. The differences between animal fat-based from fatty acid constituent, the fatty acid sequence, and the saturation level of fatty acid. We can conclude that FTIR method combines with chemometric and GCMS methods capable to identify rat meat.

Keywords:rat; meatball; Fourier Transform Infra-Red Spectroscopy; as Chromatography-Mass Spectrometry

IRCPAS/2020/OP-221

Sun protection effect of 2-hydroxy-4-(octyloxy)benzophenone in sunscreencreams formulations by a combination of inorganic UV filters

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Overexposure of ultraviolet (UV) radiation, especially UVB (280-320 nm) and UVC (200-280 nm) have a harmful *effect on the skin.* Sunscreen such as derivatives of benzophenone can protect the skin from these detrimental effects. In this research, we evaluated the potency of 2-hydroxy-4-(octyloxy) benzophenone as a sunscreen and improved the ability by combining it with the physical blocker TiO_2 or ZnO in the form of a cream formulation. We use a D-optimal mixture designto obtain the cream formulation with high Sun Protection Factors (SPF) and acceptable characteristics. Several cream formulations containing 2-hydroxy-4-(octyloxy) benzophenone and TiO_2 or 2-hydroxy-4-(octyloxy) benzophenone and ZnO were prepared with concentrations of 5-10%. The creams were tested for the physicochemical parameters such as pH, color, odor, homogeneity, viscosity, and stability. The SPF was observed by spectrophotometry and the value was calculated using the Mansur equation. SPF value of 2-hydroxy-4-(octyloxy) benzophenone, TiO_2, and ZnO were 25.21±0.47; 24.74±0.35; 3.20±0.05, respectively. SPF value of creams combining 2-hydroxy-4-(octyloxy) benzophenone and TiO_2 were in the range of 4.140-6.326. Furthermore, the SPF value of creams combining 2-hydroxy-

4-(octyloxy) benzophenone and ZnO were in the *range of*3.609-8.052. The creams meet the requirement of physicochemical properties with acceptable characteristics. They were *stable* when the creams kept at *room temperature* for one month. In this current study, the formulation of sunscreen creams with high SPF and acceptable characteristics obtained by a combination of 10% 2-hydroxy-4-(octyloxy) benzophenone and 5% titanium dioxide or ZnO.

Keywords:2-hydroxy-4-(octyloxy) benzophenone; sunscreencream; sun protection factor; TiO_2; ZnO

IRCPAS/2020/OP-222

In Silico Studies of Green Tea CatechinsAgainst HER-2 Receptor in Breast Cancer

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Green tea catechins have been widely studied and are known to have anticancer activity, including breast cancer. Breast cancer has the highest prevalence of cancer in Indonesia after cervical cancer. HER-2 (*Human Epidermal Growth Factor Receptor-2*) has an important role in the development of breast cancer, therefore this protein is widely used as a therapeutic target. In this study, we investigated the activity of catechins against the Receptor Tyrosine Kinase (RTK) domain of HER-2 in breast cancer by in silico studies. In this study, four catechin compounds i.e EGCG, EGC, ECG, EC, and one reference were subjected to docking and molecular dynamics simulation studies. Molecular docking was used to study the ligand-protein interactions using AutodockTools. The stability of interacting residue of protein with Catechins was identified by molecular dynamics using GROMACS and free binding energy calculations using MM-PBSA. Of the four Catechins compounds, EGCG has the best RMSD value, this showed that EGCG has the best structural stability. The binding free energy (ΔG) value of the Catechin compounds is greater than the references, this showed that the Catechin compounds have a lower affinity to the HER-2. Based on the results, it is known that the Catechin compounds have a lower activity than the reference. However, the Catechin compounds binding several amino acid residues that are the same as in reference, this indicates that they are binding to the same binding site. Therefore, the Catechin compounds have the potential to be developed as HER-2 inhibitors by designing of Catechin derivatives.

Keywords:Catechin; molecular docking; molecular dynamics; in silico; HER-2, breast cancer

IRCPAS/2020/OP-223

Application Of Extra Virgin Olive Oil As Transformer Oil

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In the 19thcentury since the discovery of transformers, electric power systems using alternating current systems (AC) have become more popular because without transformers, AC systems cannot work. The oil in the transformer is still using oil derived from petroleum, where

petroleum is non-renewable oil, limited in existence, will be depleted, toxic, dangerous for the environment and health, and not environmentally friendly.In this study, researchers wanted to apply extra virgin olive oil to be used as transformer oil. Extra virgin olive oil is the highest quality (Grade A) olive oil and has the best taste because it is produced from the cold pressing process of olive extraction. Extra virgin olive oil, including vegetable oil, has several advantages, namely renewable, unlimited supply, not depleted, non-toxic, harmless to the environment and health, and environmentally friendly.The measured parameter is the breakdown voltage, which is the voltage where the olive oil can no longer function as insulation, where the breakdown voltage standard in testing according to IEC (International Electrotechnical Commission) 158 and 296 is ≥ 30 kV / 2.5 mm with electrodes balls 12.5 mm in diameter.The breakdown voltage value obtained in this study is an average of four experiments.At normal room temperature (28 ^{o}C), the breakdown voltage value is 46.73 kV, while at 90 ^{o}C the breakdown voltage is 55.89 kV.oil is an oil that is rich in benefits, in addition to being consumed by living things for health, and it can also be used for external use (cosmetics, soap), and in fact the olive oil can also be used for transformer oil, especially extra virgin olive oil, which has saturated fatty acid content by 15 percent, better than non-virgin extra valued at 19.07 percent, because there is a negative correlation between the value of the breakdown voltage and saturated fatty acid content.

Keywords:Olive oil;breakdown voltage; transformator oil

IRCPAS/2020/OP-224

Analysis of Prednisone in Indonesian Uric Acid Herbs Using High Performance Liquid Chromatography

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The problem of adulteration of herbal medicines product with active pharmaceutical ingredients (API) has existed for years in Indonesia. According to the government's rules, it is not allowed to add API to traditional herbal medicine. Uric acid herbs are one of the most popular herbal medicine products. Prednisone is one of the corticosteroids that has been reported to be detected in herbal products. The purpose of this study is to identify prednisone in uric acid herbs using HighPerformance Liquid Chromatography (HPLC).The stationary phase used in this study was C18 Puroshper®STAR RP-18e LiChroCART®column(250-4.6 mm; 5µm i.d.), while the mobile phase was methanol: water (60: 40 v/v). The mobile phase flow rate was set at 1 ml/min. Prednisone in the sample was detected at wavelength 243 nm using a UV detector. Validation methods in this study consisted of precision, linearity, the limit of detection (LOD), the limit of quantitation (LOQ), and accuracy. The precision is indicated by the relative standard deviation value of 0.33% (< 2%). The correlation coefficient value (r) of 0.9955 obtained from the prednisone calibration curve shows the linearity of the method. The recovery value of 100.11 ± 0.82 % indicates the accuracy of the method that meets the requirements. The LOD and LOQ values were 2.96 and 9.85 µg/ml, respectively. Method validation parameters have been proven that meet the requirements. The HPLC method can be used to analyze prednisone in uric acid herb samples. The application of the method for analysis of eight herbal products taken from the market shows that prednisone was detected in two products.

Keywords: Prednisone; uric acid herbs; high performance liquid chromatography (HPLC)

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Development and Validation of Attenuated Total Reflectance Fourier Transform Infrared Spectroscopy (ATR-FTIR) Methods for Analysis of Selected Aminoglycosides

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Aminoglycosides (AGAs) are potent broad-spectrum bactericidal agents that have been widely used to treat variety of bacterial infections since decades ago, including the usage of streptomycin in tuberculosis patients to the emergence of kanamycin and gentamicin for the treatment of gram-negative bacillary infections. Huge literature of qualitative and quantitative analysis for the determination of AGAs utilizing spectrophotometric, electrochemical, and chromatographic methods, LC-MS methods are reported. The primary objective of this study was to develop an Attenuated Total Reflectance Fourier Transform Infrared (ATR-FTIR) spectroscopic methods for the analysis of Gentamicin and Tobramycin and their validation as per ICH guidelines. This technique was utilised as a direct, simple, non-destructive, and less time consuming compared to existing methods except simple grinding preparation required prior to read sample on direct diamond sample reader of ATR-FTIR. The calibration curve of gentamicin was constructed in the concentration range from 0.25 – 15.0 (%w/w), on its unique peak band at specific range of 3450-3350cm-1 (secondary amine stretching vibration) with good regression value 1.000 (r2~1.00). As for tobramycin, the linearity range was similar as gentamicin 0.25 – 15 (% w/w) at unique peak wavenumber of 3375-3325cm-1 (primary amine stretching vibration). The linear regression value was found to be 0.9998. Gentamicin was displayed lower limit of detection (LOD) and limit of quantification (LOQ) 0.2006 (%w/w) and 0.6080 (%w/w) respectively. The developed method was estimated to be precise at three points over the range of 0.25 – 15%, with all intra-day and inter-day precision RSD values below 4.00 for both AGAs. The quantification of gentamicin and tobramycin in ophthalmic solution formulation, with their percentage of mean recovery estimated at 100.727 ± 2.3597 with margin of error (±2.65%) at 95% confidence interval and 101.04 ± 1.864 with margin of error (±1.076%) at 95% confidence interval respectively. The proposed ATR-FTIR methods for the analysis of selected AGAs can be used for routine quality control of these active AGAs in pharmaceutical dosage form.

Keywords: Attenuated total reflectance Fourier transform infrared spectroscopy; aminoglycosides

IRCPAS/2020/OP-227

Validated Spectroscopic Methods for the Estimation of Marker Compound Caffeine in *Camellia sinensis L.*

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Plants based medicinal agents are being used across the world for the treatment of myriads of ailments in both humans and animals as well. The pharmaceutical, scientific communities and large numbers of publications have reported the therapeutic uses of natural compounds to validate the claims of their biological activities. In this connection, need of quality and standardization of the herbal medicine starts from the raw material. The aim of the present study is to develop / validate the spectroscopic analytical methods for the assay of marker compounds in the medicinal herbs which are utilized as raw material, crude drug, and herbal drugs for curing the various diseases. We selected commonly utilized plant *Camellia sinensis* L. Extraction of caffeine was done by decoction method. A validated UV Spectrometric method was performed as per ICH guidelines for the estimation of caffeine. The linearity of the compound was in the ranges between 10-60 µg/ml. The average percentage recovery of caffeine from *Camellia sinensis L.* found to be 97.4%. The %RSD of proposed method is accurate, precise, and reproducible. This result shows that it can be adopted for routine analysis of caffeine in various herbal products to develop a common consensus about the quality and efficacy of marketed available caffeine containing herbal formulations.

Keywords: *Camellia sinensis;* UV spectroscopy; caffeine

IRCPAS/2020/OP-230

Synthesis and Evaluation of Prodrugs of some NSAID's

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Pain is an unpleasant sensation experienced by all individuals. Pain is classified as Acute and Chronic. Inflammation is regarded as an injury to cell or tissue, after a trauma or post-operative surgery. Prostaglandins play an important role in the inflammation process. COX enzymes are required for conversion of Arachidonic acid to Prostaglandins. For relief of pain most widely NSAID's are used. The mechanism of action of these NSAID's is they either inhibit COX1 or COX2 or both depending on their individual class of NSAID's. Administration of NSAID's has a major drawback that, formation of ulcers, hence they must be used along with H1 antagonist. The purpose of the study was to synthesize prodrugs of Mefenamic acid, Ibuprofen and Ketoprofen, to avoid use of H1 antagonist by using Polyethylene glycol of different molecular weights as carrier and to evaluate them. Polyethylene glycol 1500 and 6000 molecular weight was selected as carrier molecule and was covalently bound to Mefenamic acid, Ibuprofen and Ketoprofen. Polyethylene glycol 1500, 6000 and spacer Glycine was covalently bound to Mefenamic acid, Ibuprofen and Ketoprofen. The obtained prodrugs were characterized by I.R and N.M.R, and then subjected to *in vitro* drug release studies at pH 1.2 and 7.2. The prodrugs were then evaluated for Analgesic activity, Anti-inflammatory activity and Ulcer protecting activity. I.R and N.M.R results have shown that the drugs were covalently

bound to the polymers PEG 1500, PEG 6000, PEG 1500-Glycine and PEG 6000-Glycine. *In vitro* studies revealed that the prodrugs had shown a higher drug release at pH 7.2 rather than at pH 1.2. *In vivo* evaluation results have shown that the prodrugs had better Analgesic, Antii inflammatory and Ulcer protecting activities than that of parent drugs. The prodrugs of Mefenamic acid, Ibuprofen and Ketoprofen had shown to be better in activities ulcer protecting. Hence based on the results and discussion it can be concluded that these prodrugs had better Analgesic, Anti-inflammatory and Ulcer protecting activites and do not require use of H1 antagonist.

Keywords:Prodrugs; PEG 1500/6000-Mefenamic acid; PEG 1500/6000- Ketoprofen; PEG 1500/6000-Ibuprofen; PEG 1500/6000-Glycine-Mefenamic acid; PEG 1500/6000-Glycine-Ibuprofen; PEG 1500/6000-Glycine-Ketoprofen

IRCPAS/2020/OP-240

HPLC Analysis of Dietary Phytoestrogens from Soy milk

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Phytoestrogens have been reported to be useful in the prevention of menopausal symptoms, skin aging, osteoporosis, cancer, cardiovascular, neurodegenerative, and metabolic diseases. It is imperative to measure the exposure of populations to these compounds by determining their contents in food commonly consumed by the population. The aim of the study was to quantify concentrations of common dietary phytoestrogens (daidzein, genistein and coumestrol) in soy milk using high performance liquid chromatography (HPLC). Freshly prepared soymilk and nineteen commercial soy milk were used in the study. Soy milk samples were extracted using an acid hydrolysis method where samples were mixed with ethanol 96% and HCl and refluxed for 6 hours. The extracted samples were analysed with HPLC. Calibration curves were plotted from different concentrations of daidzein, genistein and coumestrol standards. Fresh soy milk showed the highest content of daidzein (219.08$\pm$53.91 µg/mL) and genistein (176.53$\pm$23.67 µg/mL) while commercial soy milk contained daidzein in the range of 145-14.11 µg/mL and genistein in the range of 49-22 µg/mL. Coumestrol content in fresh soy milk was (11.55$\pm$4.35 µg/mL) and in the range of 10-9.8 µg/mL in the commercial soy milk. It was not detected in some commercial soy milk.The content of daidzein was the highest in all samples while coumestrol was the lowest. Fresh soy milk contained higher dietary phytoestrogens compared with commercial soy milk.

Keywords: Soy milk; phytoestrogen; daidzein; genistein; coumestrol

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Synthesis and Characterization of pH-Responsive Ordered Mesoporous Silica Particles as a Carrier for Biologics

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Strategies to increase the bioavailability of active pharmaceutical ingredients have been constantly researched worldwide. However, formulating oral dosage form of biologics remains as a fascinating challenge due to low bioavailability resulting from their low permeability, instability in gastric acid, enzymatic degeneration, and rapid gastrointestinal clearance. The present study aimed to develop the pH-responsive ordered mesoporous silica particles for loading biologics (insulin), to evaluate the characteristics, drug loading efficiency and *in-vitro* drug release of developed insulin loaded formulations. Two forms of mesoporous silica particles (MPS) were synthesized separately using different surfactant templates (Cetyltrimethylammonium bromide & Pluronic P123) and Tetraethyl orthosilicate precursor by Stober Sol-Gel approach. MPS particles were loaded with insulin and coated with an enteric coating polymer. The developed formulations were analysed in comparison to their morphology (SEM), particle size, surface area (BET), functional groups (ATR/FTIR), and crystallinity (XRD). Then, their drug loading efficiency percentage and *in-vitro* drug release kinetics were evaluated in various gastrointestinal pH conditions. MPS synthesized with CTAB template (MPS_{CTAB}) were short cubic-shaped particles with size <800 nm and BET surface area 858.94±1.57 m^2g^{-1}, while MPS synthesized with P123 template (MPS_{P123}) were long rod-shaped particles with length >1 μm, and BET surface area 631.32±1.88 m^2g^{-1}. The BJH adsorption-desorption pore size and pore volume of MPS_{P123} were higher than MPS_{CTAB}. XRD diffraction patterns described the amorphous nature of silica. Drug loading efficiency of MPS_{P123} was significantly higher than that of MPS_{CTAB}. For both forms of formulations, no drug release was found in gastric pH whereas the significant drug release was observed at intestinal pH.Advantages of MPS_{CTAB} were having smaller particle sizes and larger surface area which led to the faster drug release. In contrast, MPS_{P123} had larger pore volume and pore size which resulted in having better loading efficiency. Polymeric gastric coating of MPS protected insulin during the gastric acid condition and delaying the release until the intestinal conditions were met. In conclusion, gastric coated MPS particles were successfully developed as a promising carrier which significantly enhanced drug release profile for oral delivery of biologics.

Keywords: Mesoporous silica; carrier; synthesis; biologics; insulin; oral dosage form

IRCPAS/2020/OP-303

Sodium Alginate Beads Containing Peppermint Oil: Development and Characterization *In Vitro*, *Ex-Vivo* And *In Vivo*

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Peppermint oil (PO) is frequently used in pharmaceutical formulations. In this sense, PO is attracting deep attention from the scientific community due to its traditional therapeutic claim and pharmacological potential. The main objective of this study is to develop, optimization, *in vitro* characterization, *in vivo* gastrointestinal tract distribution and ex-vivo mucoadhesive properties of PO-loaded alginate beads. The factorial design was conducted to optimize the formulation using Minitab version 17 prepared by high voltage assisted electrospray technique. The average % of yield was 89.46% (n = 3). The optimized beads showed high drug encapsulation efficiency 91.31±3.20% and suitable drug release pattern in gastrointestinal media (98.57±1.78% cumulative drug release after 2 hrs). The mean size and sphericity factor of the beads ranged from 0.75±0.01 to 2.64±0.01 mm and 0.05±0.005 to 0.01±0.00 mm, respectively. On the other hand, 2.39±0.27 to 7.71±0.86% very low release was observed in acidic media (pH 1.2) after 2 hrs. It found to be dominant by first-order kinetic (R^2 = 0.926-0.975) and Hixson-crowell model (R^2 = 0.831-0.983) with a correlation coefficient close to unity over 2 hrs. The beads showed excellent floating behavior, an insight of greater mucoadhesive properties and an almost 100% swelling rate over 2 hrs in buffer media (pH 6.8). GIT distribution properties in *ex-vivo* over 2 & 6 hrs were revealed to a good distribution pattern in various parts of the intestine. The technique for the preparation of sodium alginate beads containing peppermint oil was found to be simple, reproducible, easily controllable, economical, and consistent. Besides, the raw materials used for the formulation in this study such as sodium alginate, lecithin, calcium chloride, and peppermint oil were cheap and easily available. This new approach to sustainable development goal is going to take a step forward, through a wider contribution to the pharmaceutical sector.

KEYWORDS: Peppermint oil; alginate; electrospray; microencapsulation

IRCPAS/2020/OP-304

Formulation and Activity Test of Cinchonine Niosomesas Hair Growth Stimulants

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Cinchonine is practically insoluble in water, slightly soluble in chloroform and alcohol, so that penetration of cinchonine passes through the transfollicular into the dermal papillae is low. Thus, it requires a delivery system that can reduce the hydrophobicity and increase the penetration of cinchonine into the dermal papillae. Niosomes is a vesicle system that able to reduce hydrophobicity and increase cinchonine penetration into the dermal papillae. The aim of this study was formulation and activity test of cinchonine niosomes as hair growth stimulants. Cinchonine niosomes was made by using thin layer hydration method. Formulation

of cinchonine niosomes was Span 60 100 µmol, cinchonine 0,03% w/v and cholesterol 15% *from the total number of surfactants*. *The stability test of* cinchonine niosomes *was* stored at room temperature and 40$_o$C, *humidity level of* 75% for 28 days with *7-day observation interval*. The in vivo test *included the* irritation test using male rabbits of New Zealand strain with observation interval 1, 24, 48 and 72 hours, *and the activity test of the hair growth stimulant* using male guinea pig of Hartley strain for 14 days with *2-day observation interval*. The result of the stability test showed that the cinchonine niosomes had good stability was seen from the vesicle size parameter of 200 – 350 nm, vesicle polydispersity index 0,250 – 0,450 and the entrapment efficiency of cinchonine 83 – 85%. The result of irritation test showed that cinchonine was non-irritant with erythema and edema index of 0. The result of activity test showed that cinchonine had activity as hair growth stimulant was seen from the hair length of test area was 17-43 % lengthier than the control area ($p<0.05$) and the number of hairs was 22-30% more than the control area. Cinchonine stimulates hair growth by telangiectasis through the mechanism of angiogenesis and vasodilation of blood. Cinchonine stimulates the production of Vascular Endothelial Growth Factor (VEGF) cytokines produced by endothelial cells that play a role in the vasodilation process and stimulates new blood formation, thereby increasing the supply of nutrients and oxygen needed for hair growth and regeneration.

Keywords: Cinchonine; noisome; hair growth stimulants

IRCPAS/2020/OP-309

In Vitro Permeation Test of Diclofenac Sodium Nanoemulsion with Combination Tween 80 And Transcutol

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Nanoemulsion developed to increase the bioavailability of the drug in the body. Nanoemulsion made with a combination of oil phase, water phase, surfactant, and cosurfactant. Combination of VCO, Tween 80 and Transcutol can increase the percutaneous absorption through barrier of skin and mucosal. This study aims to determine the combination of Tween 80 and Transcutol in different variations to the permeation rates of diclofenac sodium nanoemulsion. Nanoemulsion was made by spontaneous emulsification method. Ratio of surfactant and cosurfactant were Formula 1 (5:1), Formula 2 (6:1), Formula 3 (7:1), and Formula 4 (1:0). Permeation test is performed using a Franz diffusion cell tool. Diffusion membrane used abdominal membrane wistar male rats aged 2-3 months and body weight 200-300 grams. Permeation rate of Formula 1 was 27.560 ± 5.136 µg.cm-2.h-1; Formula 2 was 22.623 ± 6.738 µg.cm-2.h-1; Formula 3 was 17.347 ± 1.991 µg.cm-2.h-1; Formula 4 was 12.828 ± 3.586 µg.cm-2.h-1. Weights transported of Formula 1 was 317.058 ± 38.170 mg; Formula 2 was 256.853 ± 65.952 mg; Formula 3 was 261.807 ± 82.683 mg; Formula 4 was 138.859 ± 36.218 mg. The results showed that formula 1 was the best of all formulas in penetrating of diclofenac sodium nanoemulsion. Formula 1 had a permeation rates 2 times faster than the formula 4, while the formula 2 and 3 times faster 1 time faster than the formula 4. Variations in concentrations of Tween 80 and Transcutol can affect the permeation velocity and the weight of sodium diclofenac transport in sodium diclofenac nanoemulsion preparations. The more Tween 80 is used the lower the permeation speed and the small transport weight. While the more Transcutol is used, the high permeation speed and large transport weight.

Keywords: Diclofenac sodium nanoemulsion; Tween 80; Transcutol; virgin coconut oil (VCO); permeation test

IRCPAS/2020/OP-310

Optimization of Snakehead *(Ophiocephalus striatus)* Fish Extract Emulgel Formula as Wound Healing using D-Optimal Designs Method

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Snakehead fish extract has a potential to be used as one of the alternative treatments for wound healing because it has Omega 3 and Omega 6, which play a role in the process of wound healing. Mechanism Omega 3 and Omega 6 helps accelerating the healing process by supporting fibroblasts in synthesizing collagen. Amino acid will be hydrolysis in strong acid like gastric acid so it can be formulated as emulgel. The objectives of this research are optimization of HMPC K15M, PEG 400, and Tween 80 concentration by using D-Optimal Designs Method Software Design Expert version 7.1.15. The method of this research divides 4 parts. Part 1 is Emulgel production started by make emulsion production. Tween 80 and PEG 400 are mixed by using magnetic stirrer in 30 minutes at a speed of 250 rpm. Then, it is added with olive oil. Part 2is the gel production. HPMC K-15M is dispersed into Snakehead fish extract which has been dissolved in hot water (70oC). The emulsion and gel phases are mixed until emulgel is formed. Part 2 are Test of Emulgel's Physical Characteristics are pH examination, spreadability test and viscosity test. Part 4, stability test using freze thaw method. The best result shows that composition of sneakhead extract emulgel are HPMC K15M (5,408 %), PEG 400 (22,592 %) and Tween 80 (17,000 %) with spreadability 5,37 ±0,144 cm, viscocity 1534 ± 318,0188 cPs and pH 6,53± 0,024. HPMC K15M is the most influential component than other component of sneakhead fish extract emulgel. Stability test shows that emulgel stable before and after stability test. The component having the greatest effect on the increase of viscosity is HPMC K15M. HPMC K15M is a gelling agent which can form a three-dimensional structure, binding solvent molecules in it so that it can best result to viscosity and spreadabilty. The optimum formula for snake head fish emulgel has been obtained through the D-optimal method. The snake head fish emulgel has a good stability in storage, both at 4°C and 45°C during four cycles.

Keyword:Emulgel; D-optimal design; Snakehead Fish Extract; wound healing

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A Physical Evaluation on Semi-Solid Extemporaneous Compounding in Primary Health Care Centers

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Extemporaneous compounding is a technique to mix, combine or change drugs to produce drugs adjusted to the patients' conditions or special needs. Dosage of extemporaneouscompounding can be in form of oral and topical dosages. One of the topical dosages is semi-solid. Dosage form changes from its initial (previous) forms by forming other dosages can cause stability changes of the drug. This research aims to determine how the physical quality of semi-solid dosage made at the primary health care canters (*Puskesmas*).This research was conducted through an observation on extemporaneous compounding process at

the health centers, then the researchers tested the physical quality of semi-solidextemporaneous compounding obtained from observations at four different primary health care canters. A physical evaluation of the semi-solid compounding was carried out starting from day 0 (Week I) up to the 28th day (Week V) including homogeneity, organoleptic, Scanning Electron Microscope (SEM), pH, viscosity, adhesion, dispersion and yield tests.Based on the results of the physical evaluation, it showed that semi-solid extemporaneous compounding made by *Puskesmas D*was relatively more stable than others where the dosage remained homogeneous until the 14th day (Week III). Organoleptically, the odour occurred on the 21st day (Week IV) , based on SEM test results there was no separation of the prescribed drug, a stable pH during storage with a pH value of 6, had a viscosity value of 5845.60 cps, spreadability of 36.40 cm^2 and adhesion of 0.51 seconds, although there was a weakness in the yield value of 72.90%, where the yield was the lowest among the others.This research showed that the best physical quality of semi-solid extemporaneous compounding made by *Puskesmas D*is the best among others, because it used drugs having the same dosage form in the form of cream dosage and the compounding process was done using mortar and stamper.

Keywords: Physical evaluation; semi-solid extemporaneous compounding; primary health care centers (Puskesmas)

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Development of matrix transdermal patches: Impact of cyclodextrin complexation

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Avoidance of first pass metabolism, ease in termination of dose by just removing the patch from skin and better control of drug delivery over the extended period are some of the advantages of transdermal drug delivery. Cyclodextrins provide better solubilisation and better solution stability for poorly water-soluble drugs by preparing inclusion complexes. The aim of the present study was to investigate and then improve the permeation of duloxetine HCl (DXT) using sulfobutylether-β-cyclodextrin (SBEβCD), a highly soluble β-CD derivative. The dose and dose related side effects can be reduced via transdermal delivery route. The skin safety studies (erythema and edema) for the optimised film was also evaluated using Draize test on wistar rats.1:1 molar inclusioncomplex using spray drying techniques were prepared and after assessing their complexation efficiency and drug content, appropriate amount of complex was incorporated in the medicated films. *Ex vivo* permeation studies indicated enhanced partitioning of drug across the skin barrier. The increase in amount of drug permeated from film after 72 h on addition of permeation enhancer compared to the film having no permeation enhancer was represented in terms of enhancement ratio (ER) and was 3.05 and 1.67 for the film having complexed DXT and spray dried sample of DXT in comparison to neat DXT. The amount of drug retained in skin and in film after 72 h were relatively lower compared to the formulation having neat DXT indicating that more permeation. Enhancement in the transport of drug across skin was observed for the films having complexed form compared to neat drug with low irritancy index. It is concluded that spray dried SBEβCD complexed DXT can serve as potential innovative drug delivery system for sustained delivery of DXT over extended period of time and can provide good alternate for conventional oral route of drug delivery.

Keywords: Matrix transdermal patches; cyclodextrin complexation

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Impact of Logarithmic Transformation on the Restoration of Normality in the Bioequivalence Data of Glimepiride 4 mg tablet

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The Bioequivalence (BE) study data is mostly assumed to be normally distributed but Skewness is also a common feature of it and the Logarithmic Transformation (LT) of such data is widely applied to address this issue. After which the normality is supposedly assumed but this may not be true unless the fundamental assumption of the normal distribution of randomly generated data is verified. In some cases, instead of normality restoration, LT may introduce new problems like inducing skewness and increased variability, which is more difficult to handle. The data of a 2 x 2 BE study of Glimepiride 4 mg Test (T) and Reference (R) tablets, were used. After the drugs administration, blood samples were collected on pre-determined intervals and the plasma drug levels were determined by a validated analytical procedure. The Pharmacokinetic (PK) parameters, AUC0→t, AUC0→∞) and Cmax were derived. The data, was statistically analyzed on the linear and LT scale and a comparison between the two approaches was established, using 90% Confidence Interval (CI), two one-sided testing of hypothesis (TOST), ANOVA, Shapiro-Wilk and Q-Q Plots, using Biostat® software. The results of T and R for Pharmacokinetic metrics, 90% CI and ANOVA on linear and log scale, ANOVA, assessment of Normality restoration by Shapiro-Wilk test and Q-Q plots of the Studentized intra and inter- subject residuals are given in the relevant tables. No significant difference in the results of both approaches was found. The preliminary evidence of the weaknesses and shortcoming of the statistical procedures in normality assumption is identified. It may be concluded that LT is likely to produce inconsistent outcome regarding the restoration of normality. Since there was no significant difference in both approaches, it may be more appropriate to switch to the other distribution-free methods like Wilcoxon-Mann-Whitney two one-sided test (TOST) or the other newer analytic distribution-free methods, like the generalized estimating equations (GEE). An additional exploration is required to strengthen this notion and to identify the circumstances where the deterministic parameters are ascertained to select a suitable model for the data analysis and conclusion.

Keywords: Log transformation; normality assumption; bioequivalence data; parametric; non-parametric; distribution free statistical test

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Formulation Optimization of Nutmeg Oil Nanoemulsion

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The long-term usage of antibiotics for acne treatment can cause resistance, organ damage and immuno hypersensitivity. Hence, the alternatives to cure acne is to use natural ingredients like using nutmeg seed oil, which has antibacterial activity against of *S. aureus.* For application on the face, nanoemulsion provide many advantages: good appearance and less irritating. to obtain the optimum composition of Tween 80 and PEG 400 in nutmeg seed

oil microemulsion as an anti-acne.The optimization used D-Optimal mixture method using Design Expert 11.0.0 software. The independent variable were Tween 80 and PEG, whereas the pH, viscosity, and transmittance as the dependent variables. The optimum formula's stability was tested using *freeze thawcycle* and particle size measurement. The optimum composition was 10 % of nutmeg seed oil, 16.36 % of Tween 80, 13.64 % of PEG 400, and aquadest ad 100 ml. The pH of the optimum formula was 5.29, while the viscosity was 15.77 cps and the transmittance were 99.73 %. The predicted response value from the software was not significantly different with the experimental response value ($p>0.05$), so D-optimal design was verified. The mean of particle size of the optimum formula was 41.4 with the mean of polydispersity index of 0.468. The optimum formula's stability was good during 4 cycles storage. The optimum composition of Tween 80 and PEG 400 in the nutmeg seed oil nanoemulsion can be determined by D-Optimal mixture design. The effects of amount of Tween 80 and PEG 400 to the responses (viscosity, transmittance) can be explained by the equations of the model suggested by D-Optimal design. The predicted responses were not significantly different with the experimental responses, indicated the design was valid. The optimum formula was nanoemulsion. The polydispersity index indicated stability.

Keywords: nutmeg seed oil nanoemulsion; Tween 80 and PEG 400; D-optimal mixture design

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Dissolution Rate Improvement of Ibuprofen by Solid Dispersions in Polyvinylpyrrolidone (PVP) K30 with Melting Method

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Ibuprofen is a non-steroidal anti-inflammatory drug (NSAID) that is widely used as an analgesic and antipyretic. Ibuprofen belongs to class II BCS which has low solubility and permeability. One method to improve drug solubility is the formation of solid dispersions using polyvinylpyrrolidone (PVP) K30. This study aims to determine the effect of PVP K30 as a solid dispersion material on the rate of dissolution of ibuprofen.Solid dispersions are made using the melting method. The composition of the solid dispersion is made with a ratio of ibuprofen: PVP K30 which is 1:0.5; 1:1; 1:3; and 1:5. Physics mixture of Ibuprofen:PVP K30 is made with the same composition. Solid dispersion results were characterized using Differential Scanning Calorimetry (DSC) and Fourier Transform Infra-Red (FTIR). The dissolution test was carried out using a USP type II dissolution test with phosphate buffer pH 7.2. The dissolution profile shows that the ibuprofen solid dispersion system has a higher dissolution rate than the pure physical and ibuprofen mixture. This study proves that the formation of an ibuprofen solid dispersion system with a PVP K30 polymer effectively increases the rate of dissolution of ibuprofen. DSC thermogram of ibuprofen solid dispersion system: PVP K30 shows loss of the exothermic peak of ibuprofen (77.5-93.2 °C). This indicates that the crystalline phase of ibuprofen is homogeneously dispersed in the PVP K30 as hydrophilic polymer matrix and the solid properties change to the amorphous phase. Meanwhile the results of the analysis using FTIR spectroscopy showed a change in the wave number band 1658-1651 cm^{-1} in the ibuprofen-PVP K30 solid dispersion system. This shows the deformation of the carbonyl group (C=O) in ibuprofen due to the formation of hydrogen bonds between ibuprofen and the PVP polymer K30

Keywords: Ibuprofen; dissolution;solid dispersion; PVP K30

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Preparation and Evaluation of Herbal Lipsticks Prepared from *Solanum Lycopersicum L*

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Lipstick is one of the cosmetic products. Lip product is applied on lips to boost attraction and to touch on makeup or to moist the lips. Synthetic lipstick containing heavy metals copper, cobalt, nickel, chromium, and arsenic absorbed by the lip and stomach and causes side effects allergy, skin irritation or rashes on lips. Herbal lipstick was formulated due to its properties of better hydration, antioxidant and able to produce healthy lips and avoid blackening or cracking of the lips and to overcome the issues occurred on synthetic lipstick. The focus of the research was to formulate, evaluate herbal lipstick of *Solanum lycopersicum L* (tomatoes). The herbal lipstick prepared by melting method. The formulated herbal lipsticks evaluated for its quality control tests such as colour, surface texture, fragrance, solubility, weight variation, melting point, breaking load point, spreading ability, pH, perfume stability and aging stability. It showed yellow colour and had a smooth, no defection surface and average weight of 3.53g. The pH of the lipstick was 7. It exhibited the melting point between 60 to 65 °C and breaking load point 8.0 to 15.0 kg/cm2 and good spreading ability. The finding of this study confirmed that all the formulation possessed good physicochemical properties. When applying the lipstick on pH paper no colour changes was observed this indicated that the pH paper remains yellow in colour. This shows that the pH was 7 which is neutral and did not cause any irritation on the lips. The melting point test conferred the lipsticks would not melt in room temperature even when in slightly higher temperature. It confers all the formulations were highly stable during storage. The breaking load point results indicate that the lipstick can withstand a stronger force before it breaks. The perfume stability demonstrated that the fragrance should not be influenced by temperature and should not volatized with time.

Keywords: Herbal lipstick;*Solanum lycopersicum L*; melting; perfume stability;antioxidant; physicochemical tests

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Development of nanoparticulate drug delivery system from marine source against human immunodeficiency virus

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Human immunodeficiencyvirus(HIV) infection causes acquired immuno deficiency syndrome (AIDS) and is a universal public health issue. Anti-HIV therapy involving chemical drugs have expand the life quality of HIV/AIDS patients. Antiviralagents that hinder with HIV at different stages of viral replication have been developed. However, failure in anti-AIDS treatment is witnessed due to the arrival of resistance virus, cross resistance to drugs and celltoxicity.Nanotechnologyis the creation

and exploitation of materials, devices, and systems through the control of matter on the nanometer-length scale, i.e., at the level of atoms, molecules, and supramolecularstructures. The increasing demands on nanoparticles have varied pertinent in almost all the fields. Marine ecosystem has diversity of living resources, which includes prokaryoteslike microorganisms toeukaryotic organisms like higher plants and animals.Allthe existingtreatment modalities against HIV offer a marginal increase in the life expectancy aschitosan was converted to its derivative amino ethyl chitosan bychemicalmethod. Isolation of chitosan from crab shell by chemical method involves two basic steps:Proteinseparation, calcium carbonate separation, deproteinisation, demineralization physicochemical characterization of chitosanpowders.The results revealed the anti-HIV activity of the prepared nanoparticulate system. cytotoxicity assay of the nanoparticulate system was carried out and the CC50 value wasfound to be 38.07±1.42µg/ml indicating that the nanoparticulate system is not cytotoxic. HIV-1 infection inhibition assay was carried out and the nanoparticulate system showed excellentinhibitory activity with an IC50 value of 3.75±0.57µg/ml. It concludes, theCC50andIC50 values, the selectivity index of the nanoparticle was found to be 17.65 compared to thestandard drug nevirapine (82.32),indicating the usefulness of the formulated nanoparticulate system as potential anti-HIVagent.

Keywords:Human immunodeficiency virus (HIV); acquiredimmunodeficiency syndrome (AIDS)

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Fabrication and Characterization of Collagen Film Incorporated with Phenytoin Sodium for Diabetic Foot Ulcer

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Diabetic foot ulcers are major problems among innumerable complications associated with diabetes.Diabetic foot ulcers are associated with slow wound healing and increased susceptibility toinfection. The granulation of wound is a most successive step in the healing. Most of the researchers were focused on the matrix metalloprotease inhibitor, antibodies, and surgery for the effective treatment of wound healing. The current study is focused on the novel approach of phenytoin sodium for diabetic foot ulcer with collagen substitutes. Phenytoin sodium is an anti-epileptic drug, also called an anticonvulsant drug but unknown mechanisms; it was effective treatment for Diabetic foot ulcers. Collagen is the major insoluble fibrous protein in the extracellular matrix and in connective tissue. Chronic wounds like diabetic foot ulcers, matrix metalloproteases depleted collagen on the injury sites. Therefore, collagen film provides supplements of collagen on the injury site and phenytoin sodium enhance formation of granulation tissue, inhibition of Matrix Metalloproteinase, promoting the synthesis of collagen, maintaining the acidic environment on wound site, reducing wound exudate formation. This novel approach of collagen film incorporated with phenytoin sodium was evaluated by tensile strength, water uptake test, scanning electron microscopy, drug content, *in-vitro* drug diffusion release, interaction study (FTIR), *In-vitro* Cell line Study with adipocytes cells and biocompatibility studies (MTT Assay). The project results show enhanced water holding capacity, improved mechanicalstrength, prolonged drug release and good biocompatibility, thus aiding the wound healingprocess.

Keywords: Diabetic foot ulcers; collagen; phenytoin sodium; film

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Bead loading impact of wet milling process on dissolution rate of usnic acid nanocrystal

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Usnic acid is a phenolic compound derived from Usnea sp with an interesting biological and pharmaceutical activity, such as antibacterial, anti-inflammation, anti-fungal, antioxidant and antitumor. Its poor solubility becomes an obstacle in developed its chance as a new pharmaceutical active ingredient. In this object we produce usnic acid nanocrystal with poloxamer 188 as stabilizer by using planetary ball mill. We study the impact of bead loading parameter in wet milling process to its physicochemical characterization and its enhancement of dissolution rate.Usnic acid nanocrystal is prepared in 2 formulas, difference in bead loading each other: 10% and 15% of the milling chamber volume respectively for F1 and F2. Each formula consists usnic acid and poloxamer 188 in ratio 1:1. Primary suspension is prepared by dispersing usnic acid and poloxamer 188 into 10 ml of aquadest. Wet milling process use zirconium beads at 400 rpm for 4 hours. Nanosuspension then dryied by freeze drying method to produce nanocrystal. Nanocrystal then characterized by physicochemical properties using PSA, DSC, XRD and dissolution test. The mean particle size of F1 is 636.35 nm with zeta potential -53.3 mV. The increase of bead loading in F2 showed better result with mean particle size 563.15 nm and zeta potential -56.8. The main absorption band of usnic acid is identified at Diffractogram of usnic acid nanocrystal and bulk usnic acid shows a same pattern among them, that indicate that usnic acid in nanocrystal formulas still in a crystalline state. The milling process only reduce the peak intensity at 10.0725°; 27.1623° and 29.2550° from 22330; 3868.1 and 2549.2 (bulk usnic acid) to 1165.4; 1027.2; 999.54(F1) and 1225.8; 1176.7; 1102.9(F2). Thermogram curve of bulk usnic acid shows a sharp endotermic peak at 203.21°C. That endothermic peak also found in nanocrystal thermogram, with low intensity than bulk usnic acid 186,845°C(F1) and 186,966°C(F2). The reduce of endothermic intensity indicate that nanonization process can cause the reduction of free energy and increase the lattice energy of nanocrystal. Dissolution study in buffer phosphate pH 7.4 investigate that usnic acid in nanocrystal dissolves faster and more than bulk usnic acid. After 60 minutes bulk usnic acid only dissolved 26,3516% ± 0,9458%, while nanocrystal dissolved 88,6871% ± 1,7607% and 95,7870% ± 2,7152% respectively for F1 and F2. Wet milling process using planetary ballmill can produce usnic acid nanocrystal with poloxamer 188 as stabilizer successfully and can reduce particle size of usnic acid below 1000 nm. The characterization of XRD and DSC proved that usnic acid still in crystalline state at nanocrystal formulas. The dissolution rate of usnic acid nanocrystal increase significantly in comparation to bulk usnic acid. Statistical analysis with one-way ANOVA indicate that the ratio of bead volume to the total chamber volume in milling process (beadloading) influence the efficiency of dissolution between nanocrystal formulas significantly.

Keywords: usnic acid; nanocrystal; bead loading; wet milling; poloxamer 188

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Physicochemical Properties and Glucose-Lowering Activity of an Aloe Vera-Insulin Buccal Delivery System

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Subcutaneous injection of insulinis the mainstay therapy for Type I diabetes mellitus, and for Type II diabetes showing insufficient glycaemic control with oral hypoglycemics. However, repeated injections daily may lead to localised swelling and pain. This present a challenge that will influence patient compliance.*Aloe vera*, a perennial plant, are widely used in the development of pharmaceutical products, and has been shown to have glucose lowering activities.The buccal route offers a quick and painless way to deliver insulin, with higher bioavailability compared to routes such as oral delivery. The objective of this study is to develop a formulation intended to be delivered via the buccal cavity as an alternative route.A film formulation containing insulin and *Aloe vera*gelis to be developed, characterised, and finally assessed on its blood glucose lowering activity in an animal model. Film formulation using various film-forming polymers in combination with extract of aloe vera were developed using solvent casting method. Characterisation tests such as physical appearance, thickness and weight variations, rheological measurement, pH value, mechanical properties, folding endurance, moisture content, physicochemical compatibility, mucoadhesion, swelling index, drug content assay, permeation test, and stability test were performed on the most optimal film formulation. Assessment of the hypoglycemic rate and histopathological studies were performed using alloxan-induced diabetic rabbits.Composition of the final film formulation includes 3% w/w SCMC, 40% v/v glycerol, 70% v/v *A. vera*, 0.5% w/v mannitol, 0.125% w/v aspartame, 0.125% v/v Tween 80, and 30.6 mg of insulin. The formulation shows low variation in weight and thickness measurements and acceptable physical appearance, pH value, mechanical properties, and moisture content in addition to sustained drug release for six hours. The film was effective in reducing blood glucose levels compared to the negative control group ($p > 0.05$) in the rabbit model. We have managed to develop a buccal insulin delivery system utilising A. vera. The film had good physical properties and were able to deliver insulin and subsequently lower blood glucose levels in the animal model.

Keywords:Buccal delivery; insulin; aloe vera; film formulation

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A comparative study of Gold nanoparticles with Chitosan/Gold Nanocomposites

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In the present study, we prepared the gold nanoparticles (AgNPs) and chitosan/gold nanocomposites (CAgNPs) with a slight modification of the Turkevich method. The prepared AgNPs and CAgNPs were characters by UV-Vis, IR spectroscopy and Zetasizer. We found that the prepared AgNPs and CAgNPs have λ_{max} of 246 nm and 251 nm, respectively. IR spectroscopic studies revealed that the chitosan was present in the prepared CAgNPs. The zeta potential of AgNPs and CAgNPs sol was found to be -45 and -46 in the sodium citrate solution. Moreover, the size of AgNPs was found to be 20 nm with 0.694 polydispersity index and the

size of CAgNPs was 46 nm with a polydispersity index of 0.432, respectively. Based on these results we conclude that, chitosan was deposited on the surfaces of CAgNPs and the size was increased when compared with AgNPs and the CAgNPs sol color was changed to violet whereas AgNPs was red colour due to shifting in the λ_{max}.

Keywords: Gold nanoparticles, chitosan, IR spectroscopy, UV-Vis spectroscopy, Zetasizer

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Epidemiological Studies of *Schistosomiasis* in Bauchi Central Senatorial Zone, Nigeria

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A twelve-month Epidemiological Studies was conducted in Bauchi Central Senatorial Zone in2016 to determine the prevalence, water contact activities, water quality and vector aspect of schistosomiasis in the study area. Six hundred 600 samples of each urines and stools were collected and examined microscopically for schistosomes eggs. The urine samples were examined using sedimentation method while the stool samples were examined using formal-ether concentration technique. Twelve 12(2%) out of the entire urine samples examined had eggs of *Schistosoma haematobium* and none of the stool samples were positive with the egg of any intestinal shistosomes. Two water bodies were randomly selected from each selected local government for surveyed of the intermediate hosts (snails) of the parasite. The intermediate host were collected and examined for *cercariae* by exposing them to sunshine for 30 minutes in a beaker containing water and water samples were also collected for water quality studies such as Ph, temperature, and dissolved oxygen. Four hundred and twenty-two (422) snails were collected and examined. Out of it, only 21(4.9%) *Bulinus globosus* shed *cercariae* andthe only vector of the parasite found in the area. Six hundred (600) questionnaires were distributed to determine the participants' knowledge and perception about the parasite, sex, age, water source, toilet facilities and their occupations. The infection rates by the parasite in different sexes is not statistically significant ($p>0.05$) while in different age groups, individuals using different water source, individuals using different types of toilet facilities and individual with different occupational groups were all statistically significant ($P<0.05$). The water quality seemed to influence the infectivity of the snail vectors as out of the 422 snails collected and examined only 21(4.9%) snails were infected in the water with low pH value and high dissolved oxygen. From the results obtained, *schistosomiasis* is not endemic in the study area. Health education is recommended to maintain the non-endemic nature of the parasite in the study area.

Keywords : Epidemiology; *Schistosomiasis; Bulinus globosus*; *Cercariae*; Bauchi

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Analysis of the Effectiveness of Drug Awareness Campaigns Using Google Trends

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Globally, drug-related problems have attracted much attention from the public because of the negative health effects and the huge social burden. Therefore, Policymakers, healthcare institutions, and the people are concentrating on improving drug awareness to eradicate the abuse of illicit or prescription drugs for the destiny of a healthier community. They spent a lot to designate drug prevention campaigns as well as programs. However, the previous study has not measured the effectiveness of drug awareness campaigns comprehensively and accurately. The public was also understudied previously where the public learning preference and knowledge loophole of the drug are unclear yet. The foremost objective of this article is to figure out the effectiveness of drug awareness campaigns using Google Trends. It also aims at revealing audiences' preference of search method when they are searching for the related information. This article uses the qualitative method to explore the effectiveness of drug awareness campaign and the preferred search methods of the public to gain information about drugs by analyzing the data on Google Trends which tracks the public interest of "drugs" over time worldwide.The result found that the effect of the global drug awareness campaigns in 2018 is moderate and ephemeral and public prefers using the web search to collect information they want about "drugs". Globally, "pharmaceutical drugs"is the hottest topic related to drugs during the last year. This article finds a generally moderate influence of drug awareness campaigns in 2018. The public prefers to use Web Search to find information about drugs. Moreover, the top 5 countries where the "drugs" gains the highest attention from the public is different when the search method is different.

Keywords: Drug awareness; drug prevention campaigns; effectiveness; drug-related problems; search preference; Google Trends

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Effectiveness of Internal Try Out Toward the Graduation of National OSCE Examination on the Students of Pharmacy Professional Program in University of Muhammaditah Malang

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As a measure of success in the competence, various measuring instruments have been developed to test the success of Pharmacist Professional learning, so currently used methods that can measure the competency of Pharmacist Profession students include the MCQ's (Cognitive Based Test) method which is carried out in the form of CBT (Computer Based-Test) and the OSCE (Objective Structured Clinical Examination) method. Department of Pharmacist Professional Program of the University of Muhammadiyah Malang currently has Pharmacist professional students and has conducted a national try out exam for Batch 2 students, that the student has never received OSCE exam training before. Therefore, it is necessary to study "The Effectiveness of internal tryout on the value of the national OSCE in

Pharmacist students of the University of Muhammadiyah Malang" in Batch 3. Batch 3 has been given an internal tryout, and rehash of the material for OSCE and then continue to the national OSCE formative exam. The results of the national examination from an independent t-test showed p-value 0.03 < 0.05. It means the internal OSCE treatment has a significant effect to increase the number of graduate students in batch 3 compared with batch 2. The average results of the specific competency tests of batch 2 and 3 showed that the highest score was competency 6 (professional attitude and behaviour) and the lowest value was competency 4 (recording and reporting) of a total of 6 competencies tested. Therefore, the results of the average of the anxiety test on the student of batch 2 and 3 that they would face of an OSCE have moderate anxiety.

Keywords: OSCE, University Muhammadiyah Malang; pharmacy professional program

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Evaluation of Self-Medication Practice Among UCSI University Students

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To determine which groups of drugs were most frequently used by students as self-medication and assess the practice and views of self-medication among UCSI University's students using cross-sectional questionnaire-based study. 239 (65.1%) of the respondents practice self-medication in the past one year, among which 101(42.3%) were males and 138 (47.7%) were females. Pharmacy (74.4%) was the main source of self-medication. The most common indication for self-medication was fever (72.8%), followed by cough (67.6%), headache (67.0%), common cold (65.7%) and pain (30.5%). The most common drug classes for self-medication were antipyretics (59.7%) followed by cough syrups (59.1%), vitamins (55.3%), analgesics/anti-inflammatory (45.8%) and cold preparations (36.0%). The practice of self-medication was common among UCSI University students. It was indicated in mild disease conditions such as fever, cough, headache etc.

Keywords: self-medication; UCSI University; students.

IRCPAS/2020/OP-412

Assessment of Knowledge, Attitude and Practice of Malaysian Women Towards Osteoporosis

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Due to increasing proportions of ageing populations in the Asian region, osteoporosis has become more prevalent and increases the health care expenditure in this region. Majority of osteoporotic fractures occur in postmenopausal women. Therefore, it is important to identify women at the highest risk and to prevent further fractures. We aimed to assess knowledge, attitude, and practice of Malaysian women in Klang Valley towards osteoporosis.A cross-sectional study was conducted in 384 Malaysian women aged above 18 years. A researcher-administered questionnaire was used to collect demographic data, knowledge, attitude, and practice towards osteoporosis of participants. The participants were selected conveniently from obstetrics and gynecology (O&G) or orthopedic clinics from 6 districts (the Federal Territory of Kuala Lumpur, Selangor district of Petaling, Klang, Gombak, Hulu Langat, and Kuala Langat) of Klang Valley. Data analysis was done by SPSS version 22, using ANOVA, *t* test, Chi square test and Pearson correlation.The findings show only 2.1% of participants had good score of knowledge towards osteoporosis while most of the participants (77.1%) had poor score. There were significant association between knowledge of osteoporosis and education level, employment status and occupation of participants ($P<0.05$). 75.8% of participants had moderate attitude towards osteoporosis. Age, race, and education level of participants were significantly associated with attitude towards osteoporosis ($P<0.05$). Majority (46.4%) of participants had poor preventive practices against osteoporosis whereas only 19.5% participants had good practices. Prevention practice was significantly associated with races, education level, occupation, and monthly income of participants ($P<0.05$). Both knowledge and attitude towards osteoporosis were correlated with the practices to prevent osteoporosis.The participants had inadequate knowledge, moderate attitude, and low level of practice towards osteoporosis. This could serve as a stimulant for policy makers to increase the education of osteoporosis among younger women. Improving knowledge regarding osteoporosis is important to motivate behavioral change to prevent osteoporosis. Furthermore, the practice against osteoporosis among high-income participants was higher than low-income, even though their knowledge and attitude were same. This indicates that poverty should be addressed in Malaysia.

Keywords: Osteoporosis; public health

IRCPAS/2020/OP-413

Off-Label Drug Use for Pediatric Patients in an Indonesian Hospital

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Off-label drug use remains an important public health issue for pediatric. Off-label drug is often used in the treatment of pediatric patients because this age group is limited to clinical trials of a drug. Overwhelming number of drugs still have no information in the labelling for use in pediatrics. The purpose of off-label use is to benefit the individual patient. Therapeutic decision making must always rely on the best available evidence and the importance of the benefit for the individual patient. This study aimed to evaluate of off-label drugs use in in children aged 0-5 years. This is cross-sectional study with descriptive analysis from prospective data collection. Patient and prescribing data were collected from prescription and medical records during the patient's care period. Drugs were classified as on-label or off-label based on the

Indonesia National Drug Information (IONI). Off label drugs are categorized into off label indications, age range, dosage, contraindications, and route of administration. The study examined 130 patients with a total of 549 drug use during the 4 months of the study, 25% drugs are used off-label especially antibiotics and anticonvulsant. The main type of off-label drug was indication (76%), followed by age range (16%), dose (6%), and contraindication (5%). The study showed that off-label drug use in children is quite high (25%). Off-label drug use in the paediatric can have a risk of lack of efficacy and safety problems. Monitoring regarding the risk of drug use needs to be done. Research on new drugs and off-label drug use is urgently needed to improve the efficacy and safety of drugs used in the pediatric population.

Keywords: drug use; off-label; pediatric

IRCPAS/2020/OP-414

The Knowledge, Attitudes and Characteristics of People with Type II Diabetes Mellitus Patients ofProlanis Members Increased the MedicationObedience in UPTD Primary Care Center CilacapTengah I

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Indonesia is a developing country with a high number of people with diabetes mellitus (DM). Increased prevalence of high DM disease has an impact on the pattern of treatment in DM patients. Obstacles often occur in treatment and these were due to the lack of understanding in taking medicine and the patients' intention including the attitude of the patients. To find out the correlation between knowledge and attitudes of type II DM patients of Prolanis members with medication obedience at the UPTD I Cilacap Tengah Primary Care. The method of this study was observational analytic with a cross-sectional approach and a purposive sampling technique. Respondents in this study were 58 respondents participating in the program at the UPTD I of Cilacap Tengah Primary Care. Medical records and questionnaires were used as the research instruments. Spearman rank test and multivariate logistic regression analysis were used as the data analysis technique. The results showed that level of knowledge of patient with type II DM who followed prolanis in 2018 is mostly in the moderate category (66.5%). This is evidenced by the respondent data that they are mostly 58.7 years old. The attitude of respondents is mostly in category of supporting as many as 12 people (20.7%). The level of respondent's compliance of taking the medicine is mostly in a moderate category of 31 people (53.4%) and none of respondents is in a high level of compliance (0%). As person's age influences one's comprehension and mindset. The result showed that attitude of people with Type II DM who followed prolanis were mostly in the supportive category (79,3%) and few of them are non-supportive category (20.7%). The level of medication copliance in patient with Type II DM as prolanis participants are mostly in the moderate category (53.4%) and there is none in high level of compliance (0%). The knowledge and attitudes of type II DM patients of Prolanis members increased with medication obedience in the UPTD I of Cilacap Tengah Primary Care.

Keywords: knowledge; attitude; medication adherence; diabetes mellitus; Prolanis

IRCPAS/2020/OP-415

The Role of Traditional Birth Attendant in Postpartum Day care for Mothers in Banyumas Regency

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Postpartum is a period immediately after childbirth started from the placenta appears until the uterus returns to a non-pregnant state. Generally, there are three important processes at this period, namely uterus involution, blood viscosity, and lactation or breastfeeding. Many factors affect the process of breastfeeding, such as oxytocin massage. Javanese's habit is to have traditional birth attendant (TBA) care after postpartum. Mother has TBA to massage her and her baby. Their role is very essential to the community, especially in rural areas. TBA massages the postpartum mother expecting to produce smooth breastfeeding milk. The study aims to determine the role of TBA in postpartum daycare for mothers in Banyumas Regency which covers knowledge and motivation toward skill in oxytocin massage in postpartum mothers in Banyumas Regency. This study is observational analytic, where researcher is directed to explain a situation. The time approach used is cross-sectional. An analysis is conducted to test the knowledge and motivation of the TBA in oxytocin massage of postpartum mothers. The data analysis is ordinal scale variable and ordinal is non-parametric statistics. Hypothesis is tested through Chi-square test. The study is implemented in 9 sub-districts in Banyumas regency for 4 months. The respondents are 114 TBAs. The study discovers that 59.6% of TBA obtains good knowledge, 54.4% collects good motivation, and 53.5% presents good skills. A correlation between the knowledge and the skills of TBA in oxytocin massage in postpartum mothers with a p value of 0.031. There is a correlation between the levels of TBAs' motivation and the skills in oxytocin massage in postpartum mothers with p value of 0.029. It can be drawn that there is a correlation between the knowledge of TBA and the skills in oxytocin massage in postpartum mothers and there is a correlation between the motivation of TBA and the skills in oxytocin massage in postpartum mothers.

Keywords: Postpartum; Traditional Birth Attendant (TBA); oxytocin massage

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The Competence of Doctor on Duty Towards Mortality in PKU Muhammadiyah Hospital Gombong

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A doctor will surely be confronted with mortality incident in his profession. From the observations at the hospital, patients characterized by pupillary mydriasis, cessation of heart rate and respiration are considered die. In less than an hour, patients who died either in the Emergency Department, inpatients, ICU are transferred to the mortuary Installation. This sometimes leads to a debate because patient may only come into a near-death experience, while the time required for a definite sign of death is at least one to two hours after clinical death. To assess the competence of doctor on duty towards mortality in hospital. It was a qualitative descriptive study in which the researcher conducted an in-depth study of the doctor's competence in the diagnosis of death according to the Indonesian Doctors Competency Standards in 2012. The population were general doctors at PKU Muhammadiyah Hospital

Gombong. The sample used was someone who had completed general medical education, worked < 10 years both in the Emergency Room and inpatient room at PKU Muhammadiyah Hospital Gombong. The number of samples studied was three, two doctors on duty at the Emergency Department and one doctor on duty in the inpatient room Sampling was obtained by judgmental sampling method by selecting 20% of the total population in PKU Muhammadiyah Hospital Gombong. Two doctors on duty were considered as competent in diagnosing the death because they checked for signs of death. One doctor on duty was considered as incompetent in diagnosing the patient's death because he did not check for signs of death. There are different understandings from the three doctors regarding competence in diagnosing patient deaths. Most doctors have implemented a thanatology and are in accordance with the doctor's competency standards in determining the diagnosis of a patient's death.

Keywords: death diagnosis; competence

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Analysis of the Accuracy of coding Medical Measures in Ropanasuri Surgical Special Hospital Padang Based on ICD-9 CM

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The accuracy of coding in medical procedures and diagnosis is very important for hospital funding. Inaccurate coding will affect the level of funding that has an impact on hospital revenue. In Indonesia, funding for hospitals is the existence of a National Health Insurance (JKN) which aims to ensure that participants receive the benefits of health care and protection in meeting basic health needs. In the implementation of national health insurance (JKN), it has been determined that the pattern of payment to advanced health facilities is through the Indonesian Case Based Groups (INA-CBGs) system that is in accordance with Minister of Health Regulation No. 69 of 2013. Funding with INA CBGs currently there are significant changes, especially related to clinical data coding. The purpose of this study was to analyze the accuracy of the coding of medical measures based on ICD-9 CM by observing the medical record file. This research uses descriptive method with a qualitative approach. The research design used in this study was cross-sectional. Population is all research objects. The population used was the entire surgical patient medical record file in 2018, namely 3.093 medical record files. This sampling procedure is random sampling. Sampling was carried out using a formula according to and 355 medical records were obtained. Data collection technique used is the observation method that is direct observation of the medical record file by analyzing the accuracy of the coding of medical measures based on ICD-9 CM. Based on the analysis results it is known that the accuracy of the coding of medical measures in Ropanasuri Hospital based on ICD-9 CM is 100%. The coding accuracy at Ropanasuri RSKB is influenced by, inter alia, the presence of the anti-fraud team. The most common medical procedures are excision of other soft tissue lesions with code 83.39 and local excision of breast lesions with code 85.21. The formation of an anti-fraud team is one way to minimize coding inaccuracies. At the Ropanasuri Special Surgery Hospital (RSKB) the anti-fraud team helps in checking the accuracy of the coding.

Keywords: Coding Accuracy; ICD-9 CM; anti-fraud

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Mortality Among Chronic Kidney Failure Patients Who Have Died in Last 2 Years and Got Erythropoietin and /or Blood Transfusion as an Anemia Therapy in the Islamic Hospital Jakarta Cempaka Putih

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Anaemia occurs in 80-90% of patients with chronic renal failure. Blood transfusion is one alternative, inexpensive, and effective treatment option for treating anaemia in patients with kidney failure. In addition to blood transfusion, Erythropoietin can also be used as an alternative therapy to treat anaemia in patients with chronic renal failure. The aim of this study was to determine the prevalence, survival analysis, the relationship of therapy to anaemia with hemoglobin levels and the description of the cost of therapy in patients with chronic renal failure who received Erythropoietin therapy, blood transfusion, and a combination of both at the Jakarta Islamic Hospital Cempaka Putih. This study is a cohort study, sampling was done retrospectively with the universal sampling method of medical records of patients with a primary diagnosis of chronic renal failure undergoing hospitalization and having undergone hemodialysis at Jakarta Cempaka Putih Islamic Hospital for the period of January 1, 2016 until December 31, 2017. Analysis The data in this study used data processing software SPPS 22. The results showed that Vulnerable age 46-65 (54%) and male patients (56%) contributed to giving the largest presentation for this study sample. Patients with hypertension (86%) and diabetes mellitus (66%) also contributed the largest presentation in this study sample. Anaemic patients with chronic renal failure who received combination therapy between Erythropoietin and blood transfusions had a higher survival rate compared to patients who received Erythropoietin therapy or blood transfusions. Paired Sample T-Test results showed no significant difference between the first HB level and the last HB checked before patients died in the Erythropoietin therapy group, blood transfusion, and folic acid therapy (P value> 0.05), while in the combined therapy group between Erythropoietin and blood transfusion shows a significant relationship with a P value of 0.030. In this study the largest survival analysis was shown in haemodialysis patients using erythropoietin compared to patients who only used blood transfusions and the highest cost incurred by patients was the combined cost of therapy between erythropoietin and blood transfusion, which was in the range of 1,001,121.03 - 31,120,525.54 rupiahs and with an average cost of 6,890,484.23 rupiahs.

Keywords: Chronic Kidney Failure; blood transfusion; erythropoietin; anaemia

IRCPAS/2020/OP-422

Development and Assessment of Modified Glover Nilsson Vaping Behavioural Questionnaire Among Malaysian Single Electronic Cigarettes Users

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The Glover Nilsson smoking Behavioural Questionnaire (GNSBQ) is the commonly applied scale to assess the behavioural nicotine dependency on conventional tobacco cigarettes (TCG). But the GNSBQ does not evaluate the subject's behavioural dependence to nicotine that administered via electronic cigarette (EC). To develop and assess an equivalent modified Glover Nilsson vaping Behavioural Questionnaire (GNVBQ) scale which measures the nicotine behavioural dependency that administered through EC. The investigator developed the equivalent modified GNVBQ scale which scores identical to original GNSBQ i.e. 0 to 44. The scale scores indicate the nicotine behavioural dependency ranking as slight (1-6), mild (7-11), moderate (12-22), strong (23-33) and very strong (> 33). The developed scale piloted among 15 EC single users. i.e. used only EC. The assessment of the scale was done among 69 EC single users and measured their nicotine behavioural dependency status until one-year period. The modified scale revealed a satisfactory Cronbach's alpha value of 0.74. Further test-retest reliability of the scale showed an acceptable spearman's rank correlation coefficient value of 0.75 ($p > 0.05$). A one-year observation showed that out of 69 single users, 11 single users completely stopped nicotine intake. The EC users who completely stopped nicotine intake after one year had a low nicotine behavioural dependency at the baseline which scores a between 7-11 that was measured by the new modified GNVBQ scale. The modified GNVBQ scale precisely identify the behavioural dependence to nicotine that administered via EC. Therefore, as per the current study results the modified GNVBQ scale can apply in any EC related studies to assess the nicotine behavioural dependency that administered through various electronic cigarette products.

Keywords: Nicotine; vaping; behaviour; dependency; scale

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Healthcare Professionals' Off Label Drug Prescribing Practice in Vulnerable Group of Population

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Despite the pharmaceutical products prescribed for Food and Drug Administration–approved indications, off-label drug uses may have deficiency of rigorous scientific scrutiny. Regardless of concerns about patient care and health care expenditure, little is known about the frequency of off label drug use or the degree of scientific evidence supporting this practice.The current study was conducted to observe the practice of off-label drug prescribing practices in children and neonates in clinical settings of Karachi, Pakistan. A cross sectional study was conducted in different health care setups. Health care practitioners were assessed for prescribing practice

of off-label drugs in children by requesting them to fill and submit the survey form. Multivariate analyses were used to identify drug-specific characteristics predictive of increased unlicensed drug use.The current study revealed that health care practitioners often used the practice of off-label drug prescribing in children. Majority of them considered that off label prescribing safe if used with strong scientific research. Around 68% of physicians and 77% pharmacists revealed that they are lot more concerned about the efficacy of such drugs as compared to that of licensed medicines in children. The most frequent off label categories observed in the study were dose (65.21%) and indication (17.52%). A vast majority (>80%) thought that approving new drugs by regulatory authorities will drop the occurrence of medication errors due to the wrong dose. The present study revealed the common practice of off-label drug prescribing in pediatrics; however, respondents showed their concern towards decreasing such prescribing practice and are likely to welcome initiatives intended to assure the medication safety for children.

Keywords: Unlicensed drug and off label prescribing; physicians; clinical pharmacists; Pakistan

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Medication Adherence Among Schizophrenia Outpatients in National Mental Hospital Indonesia

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Medication adherence is one of the foremost problems affecting antipsychotic efficacy in schizophrenia patients. Medication nonadherence among schizophrenia patients has been often estimated > 50%, leading to higher rates of relapse and hospitalization as well as to decreasing cognitive and functional prognosis. The purpose of the study is to evaluate medication adherence among schizophrenia outpatients. Prospective study with cross sectional design was conducted from October to November 2019. Especially data from schizophrenia outpatients in one of national mental hospital in Indonesia. Nonprobability sampling (purposive sampling) all schizophrenia patients who registered as an outpatient national mental hospital in the chosen sitting and fulfill the inclusion criteria was selected. The participants were 30 patients. schizophrenia outpatients were majority male (60%), the age range from 31-49 years were 70%, most of patients are single (63,33%), 70% have secondary education, 70% of them are from Surabaya area, and half of them their duration of the disease from 1 to 5 years. This study showed that the pattern of prescription of antipsychotics are second generation antipsychotics. Risperidone and clozapine were the most antipsychotics prescribed for schizophrenia outpatients. 40% of patients have good adherence, 40% of patients have partial adherence, and only 20% of patient's poor adherence. Most of schizophrenia outpatients have experience in forget to take his/her medicine and careless at times about taking his/her medicine and less knowledge about schizophrenia. In other hand, 100% patients have agreed by staying on medication, it can prevent getting sick. The mental hospital should utilize educational program to improve patient's awareness about their disorder and their medications to improve their adherence.

Keywords: Schizophrenia; antipsychotics; medication adherence; MARS

IRCPAS/2020/OP-430

Drug Related Problems in Type 2 Diabetes Mellitus with Hypertension at Dr. M.Djamil Hospital Centre Padang Indonesia

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Diabetes is a syndrome that is caused by a relative or an absolute lack of insulin. Impaired insulin secretion and resistance to the action of insulin, rather than an absolute insulin deficiency, characterize patient with type 2 diabetes. Type 2 diabetes is associated with a variety of disorders, including obesity, atherosclerosis, hyperlipidaemia, and hypertension**.** Type 2 Diabetes Mellitus (T2DM) patients with hypertension often receive multiple medications and this can lead to the occurrence of drug-related problems (DRPs). The project aim is to assess and identify the categorize of drug-related problems in type 2 diabetes inpatients care with hypertension at Dr. M.Djamil Hospital Centre Padang. A prospective, crossectional method was conducted in this nonexperimental research. The demographic, disease and treatment data of T2DM patients with hypertension were collected for a period of three months and analysed. The Pharmaceutical Care Network Europe (PCNE) version 5.0 was used to categorize DRPs.Twenty patients with total of 32 DRPs were identified. The assessment of drug-related problems were only two categories which was six primary domains of problems and causes. In this study, there was two primary domains of problems were identified, there was drug choice problems (46%) and dosing problems (54%). The domain causes for DRPs is related with drug or dose selection (59%) and drug use process (41%). The high frequency of drug choice of problems was inappropriate duplication of therapeutic group or active ingredient (28%) and 9% each for problem related with no drug prescribed but clear indication and contraindication. In this study, excessive dosage and subtherapeutic dosage of candesartan and insulin were the second most common DRPs. According to the PCNE classification of DRPs, the primary domain of causes is most related to drug or dose selection focus on inappropriate dosage selection (27%), pharmacokinetic problems (27%) and synergistic/preventive drug required and not given (5%). On the other hand, the less problem was drug use process which caused by drug underused and over administered. Overall, among twenty patients, only 8 patients had DRPs (40%). Hence, assessment and identification of categorize DRPs may heighten the prevention and management DRPs in T2DM patients with hypertension.

Keywords: Drug related problems; diabetes type 2; hypertension; PCNE

IRCPAS/2020/OP-439

Willingness to Donate Kidney Among Malaysians

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Kidney transplantation is a relatively safe procedure and is lifesaving for patients with end-stage renal failure (ESRF). Although the number of patients with ESRF is increasing, kidney donation remains a challenge in Malaysia due to the low number of kidney donors. The reason behind this is unclear. However, work has shown that knowledge and belief are associated with kidney donation. Therefore, the aim of the study is to identify level of knowledge and belief

towards kidney donation as well as factors associated with willingness to donate kidney among the Malaysian population. A cross-sectional study was conducted in Kuala Lumpur, Malaysia. Data was collected using a questionnaire, among Malaysian adults with informed consent. Incomplete questionnaires were excluded. A total number of 391 respondents were included. The majority (n=369, 94.4%) were willing to donate their kidney, while the remaining were not (n=22, 5.6%). The mean total knowledge score was 5.9 ± 2.5 (maximum score of 13). The mean total belief score was 36.3 ± 4.6 (maximum score of 55). It was demonstrated that age, knowledge score and belief score were predictors of willingness to donate kidney. An increase in 1 unit of the knowledge score increased the likelihood of kidney donation by 1.29 times (95% CI = 1.07-1.55, p = 0.006). An increase in 1 unit of belief score increased the likelihood of donation by 1.23 times (95% CI = 1.10-1.37, p < 0.001). A reduction in 1 unit of age, increased the likelihood of kidney donation by 1.06 (95% CI = 0.91-0.98, p = 0.001). The current study indicates Malaysia has a huge population of potential kidney donors. However, there is still a need to improve public knowledge and belief towards kidney donation through health education. Public education on donor registration is also vital to optimize on the number of potential donors.

Keywords: organ donation; kidney donation; public education

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Knowledge, Practice and Perceptions Concerning Sleep from Islamic Perspectives and Modern Sciences Among Undergraduate University Students

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Sleep represents a reversible condition of unresponsiveness to environment. But how quality sleep is conceptualized is often shaped based on the degree to which culture, religion and sciences are practices. According to sciences, sleep hygiene is important for an individual to have a quality sleep, whereby sleep is also important from Islamic perspectives, which, the Quran and Hadith discuss on the types, importance, and quality practices. However, many, including adolescents, regard sleep as unimportant practice in daily life and ignored the consequences due to sleep deprivation like forgetfulness, obesity, and glucose intolerance. Therefore, this study was conducted to identify the knowledge, practice and perceptions among university students concerning sleep from Islamic and modern sciences. This study was conducted cross-sectionally; an online questionnaire consisted of six sections, was distributed via email and social media among university students at universities in Malaysia and Egypt. An agreement to proceed answering the questionnaire was considered as obtained the consent. Data collected that assessed knowledge, practice, and perceptions of the respondents, were analysed using SPSS version.23. A total of 577 students responded to the survey; 97% aged between 18-25 years and 80% were female. About 95% of the respondents have "good" knowledge of sleep, where 53% received information from formal education. Qailullah or midday-nap was known by 96% of the respondents, but only 32% practiced it. Surprisingly, less than 10% admitted as often consumed coffee at night to keep awake. As for perceptions, majority of them (96%) agreed that not getting enough sleep could impair their performance, but only 22% were slept for 7-8 hours/night. Among university students, the perception concerning sleep is proportionate with the level of knowledge, however, to maintain a healthy sleep-wake practice is challenging. It is therefore important to encourage the students to

maintain good sleep hygiene to ensure healthiness and reduce the consequences from sleep deprivation.

Keywords: sleep hygiene; Islamic perspectives; modern sciences

IRCPAS/2020/OP-444

Prevalence of Healthcare Associated Infections (HCAIs) and their Microbial Etiology Among End Stage Renal Disease (ESRD) Patients on Renal Replacement Therapy

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End stage renal disease (ESRD) patients have an increased risk of morbidity and mortality due to infections,as these patients have multiple comorbidities, may suffer frommalnutrition, and have profound alterations in their immune system.To determine the prevalence of healthcare associated infections (HCAIs) and their microbial etiology among ESRD patients undergoing renal replacement therapy (RRT).A multicenter, retrospective study of patients on RRT was conducted from June 2019 to December 2019 at two hospitals, including university of Malaya medical centre (UMMC) and hospital Serdang. ESRD patients with minimum of 6 months on RRT were included in this study, while pregnant patients and patients below the age of 18 years were excluded.To confirm the diagnosis of HCAI, patient had to fulfil at least one of the following criteria: 1) attended a hospital, received intravenous therapy, wound care or specialized nursing care in 30 days before the infection or 2) was hospitalized in an acute care hospital for 2 or more days in the previous 90 days. A data collection form was used to retrieve all sociodemographic and clinical data. To reduce the risk of selection bias, all patients were randomly selected using an online tool; research randomizer, which utilizes total number of patients and the sample size to generate random numbers. The period prevalence showing the proportion of patients that acquired HCAI since the initiation of dialysis till 2019, was calculated using a formula stated by European patients' academy (EUPATI). Total of 670 records were examined, out of which 400 patients were selected based on the inclusion criteria.There were 174 patientswith at least one episode of HCAI,hencethe period prevalence was found to be 43.5%. Catheter related blood stream infection (CRBSI) was the most common infection and occurred in 64 (36.8%) patients, while peritonitis and pneumonia were identified in 44 (25.8%) and 36 (21.2%) patients, respectively. Out of 382 total pathogens identified through microbial culture, 204 (53.4 %) were gram-positive and 162 (42.4%) were gram-negative. Among the gram-positive organisms,*Staphylococcus aureus*was identified in 90 (23.5%) patients, while *Staphylococcus epidermidis* and Streptococcus spp. were seen in 42 (10.9%) and 24 (6.28%) patients, respectively.*Klebsiella pneumonia,* which was found in 36 (9.42%) patients, was most frequently identified gram-negative organism. Moreover, Candida spp. were the only fungal organisms found in this study. Both methicillin sensitive *S. aureus*(MSSA) and methicillin resistant *S. aureus*(MRSA) showed statistically significant associations ($p = <0.05$) with CRBSI, while MSSA and *klebsiella pneumonia* showed similar associations ($p = <0.05$) with pneumonia.High prevalence of HCAIs was found among ESRD patients undergoing RRT, which is quite alarming. Furthermore, broad range of pathogens were also identified to be associated with these infections.

Keywords: Healthcare associated infections; end stage renal disease; renal replacement therapy

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Experience and Expectation of Healthcare Providers Towards Clinical Pharmacy Services

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Clinical pharmacy services have an important role in improving the patients' outcome and minimizing the potential consequences that may occur because of drug-drug interactions or inappropriate choice of treatment. As doctors and nurses are working closely with pharmacists, understanding their experience and expectations towards clinical pharmacy services are essential The purpose of this study is to investigate experience and expectation of doctors and nurses towards clinical pharmacy services in Universiti Kebangsaan Malaysia Medical Centre (UKMMC).: A cross-sectional study was conducted in the general wards of the UKMMC using self-administered and validated questionnaires. A total number of 306 healthcare providers (HCPs) participated in this study with a response rate of 80.5%. Many HCPs (96.4 %) believed that clinical pharmacists play an integral role of the medical team despite only 69% thought that there was an increased interest in the clinical pharmacy services in Malaysia. Based on actual experience of the respondents with clinical pharmacists, 79.1 % (n = 242; p = 0.002) reported that clinical pharmacist always informs them should any clinical problems related to medications were discovered. Moreover, majority of HCPs (93.8 %, n = 287; p = 0.037) expect clinical pharmacists to be knowledgeable and expert in drug therapy while 86.6 % (n = 265; p = 0.044) agreed that clinical pharmacist should be part of the medical team to provide them with drug consultation. Majority of the doctors and nurses believe in the important role of clinical pharmacist and had positive experience towards clinical pharmacy services. They considered clinical pharmacist to be the expert person in medications and part of the medical team to ensure the patient's optimal therapy outcome.

Keywords: Clinical pharmacy services;healthcare providers;experience; expectations

IRCPAS/2020/OP-449

Consumers' Knowledge, Attitudes and Practices towards Medicine Price Transparency at Private Healthcare Setting in Malaysia: A Cross-Sectional Study

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This study aims to evaluate the consumers' knowledge, attitudes, and practices towards medicine price transparency initiative and to investigate potential factors that may influence consumers' good practice related to purchasing medicine in the private health care settings in Malaysia.A cross-sectional survey was conducted among consumers in private healthcare settings in Malaysia.Using a validated self-administered questionnaire, respondents' knowledge, attitudes, and practices towards medicine price transparency were assessed with 26-Likert scale itemswith a mixture of closed-ended question. Binary logistic regression was performed to identify the factors that influence good practice towards medicines price transparency. A total of 679 respondents participated in the study. The mean age of respondents

was 38 ± 13 years with majority were female (420, 61.9%).The mean percentage of respondents' knowledge, attitudes and practices score was 70.49± 18.2, 79.85 ±9.9and 52.50± 13.3, respectively. Majority of respondents 'did not know' or 'not sure' (n = 361, 53%) that they can refer to the Ministry of Health's medicine price guide website. The highest agreement (strongly agreed and agreed) on attitudes was acheived on itemized billing practice that should include detail price of each items (n=623, 91.8%). Nevertheless, in term of practice only 264 (38.9%)'always' or 'often' asked for itemized bill and 77 (11.3%)had negotiated or asked for a discount price when purchasing medicines. Male, Chinese ethnicity, high knowledge and attitudes scores and high cost spent on medicineshad significant influence on good practice of medicine price transparency.The practiceson medicine price transparency initiative was found to be low. Several strategies need to be implemented to increase consumers' empowerement for price transparency initiative in private health care setting in Malaysia such as cosumer education, compulsory itemized bill and medicine price control.

Keywords: Price Transparency;medicine price; consumer.

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Knowledge Attitude and Perception Towards Halal Pharmaceuticals among Chronic Disease Patients

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There is an increasing awareness among the consumers for halal products in Malaysia. Thus, this study aims to assess patients' knowledge, attitude, and perception (KAP) towards halal pharmaceuticals and their belief in medication (BIM). This cross-sectional researcher-assisted survey was conducted among chronic disease patients from February to May 2019. The questionnaire consists of patients' socio-demographic characteristics, patients' belief in medications, knowledge on halal pharmaceuticals, attitude towards halal pharmaceuticals, and perception of halal pharmaceuticals. A total of 403 respondents were included in this study. Majority of the respondents were Malay, aged more than 60 years old and were pensioners. This study found that patients have good knowledge (median=8, IQR=3), attitude (median=27, IQR=9) and perception (median=31, IQR=7) towards halal pharmaceuticals. Generally, patients have higher scores for positive BIM (median=17, IQR=7) than negative BIM (mean=13.44±4.52). Race was associated with good KAP towards halal pharmaceuticals and female scored higher in BIM compared to male. Significant correlation was noted between knowledge-attitude, attitude-perception, and knowledge-perception towards halal pharmaceuticals. It was found that negative BIM was weakly correlated with attitude (r_s=0.12, $p<0.01$). Generally chronic disease patients have good KAP towards halal pharmaceuticals with positive belief in medication.

Keywords: Halal pharmaceutical; KAP; chronic disease patients; belief in medication

IRCPAS/2020/OP-451

Pharmacy Students' Attitudes in Learning Communication Skills and their Readiness for Interprofessional Learning with other Health Care Professionals

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Effective communication can build relationship between pharmacists and patients. This can help improve patients' adherence to achieve better health outcome. Interprofessional Learning (IPL) with other health care professional is crucial to prepare pharmacy students to collaborate with other health care professionals. Students' attitudes towards learning communication skills and readiness for IPL were evaluated in this research. A cross-sectional study consisted of Communication Skills Attitude Scale (CSAS) and Readiness for Interprofessional Learning Scale (RIPLS) was conducted in universities providing pharmacy course in Malaysia from September to November 2019 by distributing questionnaire by hand and online. A total number of 575 pharmacy students were recruited. 37.9% (n = 218) of them were from government universities. The attitudes of students towards learning communication skills and readiness for interprofessional learning were significantly associated with the type of university (government or non-government), ethnicity, students' first language, students' ratings on their own English proficiency and their communication skills, whether they had barriers in learning communication skills and provision of communication skills training and IPL experience in their universities ($p<0.05$). There was significant strong correlation found between CSAS and RIPLS in this research. The Spearman' rho between Positive Attitude Subscale (PAS) and Negative Attitude Subscale (NAS) of CSAS with RIPLS were 0.741 and -0.446 respectively ($p<0.05$). Pharmacy students in this study generally showed favourable attitudes towards learning communication skills and were highly ready for IPL with other health care professionals. Improving students' positive attitudes toward learning communication skills may result in improved readiness for IPL.

Keywords: communication skills learning; interprofessional learning; pharmacy students; Malaysia

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Revisiting the Therapeutic Monitoring of Azathioprine in the Management of Inflammatory Bowel Disease

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The blood levels of two major azathioprine metabolites, namely 6-thioguanine nucleotides (6-TGN) and 6-methylmercaptopurine (6-MMP), have an inconsistent relation with drug efficacy. This is probably because*in vivo* phosphorylation of the metabolites alters their action, and this is not normally measured in clinical studies.We examined the relation between the blood levels of the various phosphorylated forms of 6-TGN and 6-MMP and the outcomes of IBD treatment.We first validated a published HPLC method for measuring the blood levels of the phosphorylated metabolites. Then, we prospectively assembled a small cohort of IBD patients,

who had been treated with azathioprine for≥3 months and obtained blood samples. The patients were classified as responders or non-responders to azathioprine based on their disease activity score (CDAI or Mayo Score).We confirmed that the HPLC method could distinguish the phosphorylated metabolites of azathioprine,namely thioguanosine monophosphate (TGMP), thioguanosine diphosphate (TGDP), thioguanosinetriphosphate (TGTP), and methylthioinosine monophosphate (meTIMP). The method was precise with intraday and interday variation <15% for all the testedmetabolites, and the relative accuracy ranged from 40% to 114%. In 12 responders and6 non-responders to azathioprine,we found TGMP levels to be inversely correlated with CDAI (Spearman's rank correlation coefficient, −0.58042; p=0.0479). However, nostatistically significant relation was found between CDAI or Mayo Score andthe blood levels of TGDP, TGTP, and meTIMP. We noted thatthe responders had a higher median 6-TGN level than the non-responders. However, the difference was not statistically significant (Wilcoxon rank-sum test, 182.29 *vs.* 121.35pmol/30mg Hb,p=0.4627). The higher median 6-TGN level detected in the responders is in keeping with the findings of many prior studies. However, the actual clinical significance of the relation between TGMP levels and drug efficacy is questionable, as TGMP makes up only a small fraction of the azathioprine metabolites. Overall, we could not demonstrate a statistically meaningful relation between the blood levels of azathioprine metabolites and the outcomes of IBD treatment. Future work may focus on optimising the HPLC method further and testing its utility in a larger group of IBD patients.

Keywords: Inflammatory bowel disease; azathioprine; phosphorylation; 6-thioguanine nucleotide; methylmercaptopurine; therapeutic drug monitoring

IRCPAS/2020/OP-453

A Systematic Review of Cost-Effectiveness of Medication Adherence-Enhancing Intervention for Asthma Patients

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Asthma is a noncommunicable disease that had affected three hundred million people worldwide and medication nonadherence leads to many negative health complications and a high economic burden on society. This systematic review aimed to evaluate the evidence on the cost-effectiveness of medication adherence-enhancing intervention, as opposed to usual care or placebo. Search engines such as PubMed, Scopus and EBSCOhost were used to locate all possible studies from the inception of the search engines to 19 October 2018. Drummond checklist was used to appraise the quality of economic evaluation. Data including study characteristics, quality assessment, health outcomes and costs of intervention were narratively summarized. The primary measure is cost-effectiveness (CE) outcome and the secondary outcomes are costs, medication adherence and clinical consequences. A sum of 20 studies was included, where eleven studies were RCTs, six studies were based on comparative studies and three studies adopted Markov models. Fifteen studies evaluated an educational intervention, with 13 of them were cost-effective in improving the health outcomes. An internet-based intervention showed similar CE outcomes between treatment groups. All studies involving a medication regimen simplification and combination of a technology-assisted program and a training lesson had demonstrated the desirable CE outcome. The quality of most studies was fair with four studies showed a high-quality standard. Fundamentally, the medication

adherence-enhancing interventions were cost-effectively showing an increase in medication adherence and positive clinical effectiveness while reducing asthma-associated costs. However, limitations associated with poor methodological conduct must be properly addressed. Further economic evaluations with sound methodological conduct should be encouraged for stronger shreds of evidence in determining the best intervention to improve medication adherence.

Keywords: Pharmacoeconomics; cost-effectiveness analysis; medication adherence; patient compliance; asthma; systematic review

IRCPAS/2020/OP-457

Stigma in Asthmatic Patients: A Cross-Sectional Study to Assess the Psychometric Properties of Bangladeshi Version of Stigma Scale (B-SS)

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Asthma is a serious public health concern in Bangladesh. Asthma-related psychosocial factors including stigma may influence the treatment outcomes and wellbeing of the patients. This study was conducted to establish the reliability and validity of Bangladeshi version of Stigma Scale (B-SS) and assess the levels of stigma among Bangladeshi adults diagnosed with asthma. In this cross-sectional study, 325 adult asthmatics (aged ≥ 18 years old; not diagnosed with other respiratory disease; nil cognitive disability) were recruited from National Asthma Centre, Dhaka, Bangladesh. The researcher-administered questionnaire consisted of three sections: socio-demographic data (6 items); medical data (11 items); and stigma scale (22 items). For stigma scale, the patients' responses were recorded on a 5-point Likert scale, where the response may vary from strongly disagree (score = 1) to strongly agree (score = 5). Higher score reflected higher stigma level. The permission to adapt and translate the questionnaire was obtained from the corresponding author. The finalised questionnaire was translated into Bangla language by forward and backward translation, harmonisation, cognitive debriefing interviews and proof reading. The content and face validations were carried out by three senior clinical experts and five adult asthmatics, respectively. The extracted data from completed questionnaires were analysed for descriptive and inferential statistics. The mean age (±SD) of the respondents was 41.92 (± 15.42) years old and more than half (52.9%) were females. Asthma was not-well controlled (n = 137, 42.2 %), and mostly patients experienced either moderate (n = 215, 66.2 %) or low (n = 93, 28.6 %) levels of stigma. B-SS was found to be a reliable instrument using the measures of internal consistency (Cronbach'α = 0.73) and one-month test-retest reliability (ICC = 0.87). The findings of Chi-Square test suggested that age ($p< 0.01$), number of years since diagnosed as asthmatic ($p< 0.01$), comorbidities ($p< 0.01$) and asthma control ($p< 0.01$) were significantly associated with stigma scores. B-SS appears to be a linguistically reliable and valid instrument. B-SS can be used to evaluate the stigma among adult asthmatics in Bangladeshi healthcare settings.

Keywords: Asthma; asthmatic patients; cross-sectional study; public health; Bangladesh

IRCPAS/2020/OP-511

In vitro anti-allergic activitiy of *Phyllantus amarus* and *Moringa oleifera* extracts

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Phyllantus amarus Schumach. & Thonn. and *Moringa oleifera* Lam. are a well-known plant for its medicinal purpose such as anti-inflammation, antioxidant, antimicrobial and anticancer but none has been reported on its anti-allergic properties. Allergic reaction cases increasing by years and the reactions can be mild as rashes and severe as anaphylaxis that can lead to death. Subsequent exposure of allergen will trigger mast cell degranulation which will release mediators that exhibit allergic symptoms such as bronchoconstriction, vasodilation and increased vascular permeability. This study was aimed to examine the anti-allergic activity of *P. amarus* and *M. oleifera* extracts.*P. amarus* (whole plants) and *M. oleifera* (leaves, seeds and pods) were extracted with 80% of ethanol. The anti-allergic activity of the extracts and ketotifen fumarate as positive control were studied by evaluating their inhibitory activities on β-hexosaminidase and histamine release from RBL-2H3 cells line.The inhibitory activity on mast cell degranulation of *P. amarus* extract on beta-hexosaminidase activity was more significant than its inhibitory activity on beta-hexosaminidase release while the extract *P. amarus* also observed did not inhibit histamine release. Whereas, all three parts of *M. oleifera* inhibited the release of β-hexosaminidase and histamine with *M. oleifera* leaves (IC_{50}:7.17 ± 1.69 µg/mL) exhibited the higher significant activity compared to ketotifen fumarate (IC_{50}:15.86 ± 1.10 µg/mL). The study concludes that *P. amarus* did not inhibit mast cell degranulation but exhibit weak antihistamine activity by binding on the H1 receptor while *M. oleifera* potentially has an anti-allergic activity by binding early phases of allergic reactions.

Keywords: *Phyallantus amarus*;*Moringa oleifera*;anti-allergic; RBL-2H3;histamine; Beta-hexosaminidase.

IRCPAS/2020/OP-512

Interaction of Enalapril with *Moringa oleifera* by affecting Angiotensin Converting Enzyme (ACE) Activities *In Vitro* and *Ex Vivo*

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Enalapril is one of the potent antihypertensive drugs and shows its effect by inhibiting the ACE activities. *Moringa oleifera*, a medicinal plant well-known for its nutritional benefits, especially in lowering blood pressure. Dietary consumption of medical plants is practised in many populations, thus there is an increasing concern regarding concurrent usage of medicinal plants with modern drugs. The aim of the study was to investigate possible interactions between *M. oleifera* and enalapril when used in combination. *In vitro* study determination of IC_{50} values of both compounds at concentration range of 1 to 100 µg/mL was performed. *M. oleifera* and enalapril solotions were prepared and preincubated for 3 minutes with 5mmol/L of Hippuryl-L-histidyl-L-leucine (HHL) at 37°C. Later, 0.1U/mL of ACE was added to the mixture than incubated for 30 minutes. The enzymatic reaction was stopped by adding 0.05mol/L of hydrochloric acid (HCl). The product of the enzymatic reaction, hippuric acid (HA) was extracted with ethyl acetate, centrifuged and its absorbance was determined at 228nm using

UV-visible spectrophotometer. In *ex vivo* study, The ACE activities were analysed in the lung homogenate of Spontaneous Hypertensive Rats (SHR) treated orally with enalapril (1.92mg/kg/day), *M. oleifera*(1g/kg/day) andthe combination of both compounds for 14 days. Using a UV-Vis spectrophotometer, the activity of ACE was determined by quantification of the HA product as described above after reacting with the substrate. The increase in production of HA correlated with the decrease of ACE inhibition activity. Enalapril showed the highest percentage of ACE inhibition activity with 60.57±2.27% ($p<0.001$) and *M. oleifera* with 53.60± 2.60% ($p<0.001$) at a concentration of 60µg/mL compared to negative control. However, when they were used simultaneously, the inhibition effect decreased to 47.57±2.80%. The IC_{50} values of enalapril and *M. oleifera*obtained were 3.91±0.22 µg/mL and 36.70±6.14 µg/mL respectively. *Ex vivo* resultsshowed that enalapril and *M. oleifera* caused a significant ACE inhibition activity with $p<0.01$ and $p<0.05$ respectively while the combination use did not show any significant ($p>0.05$) ACE inhibition activity. *M. oleifera* leaf extract interacted with enalapril which caused reduction in ACE inhibition activity of enalapril.

Keywords: Angiotensin-converting enzyme (ACE) inhibitor; enalapril; *Moringa oleifera*; enalapril-*M. oleifera* interaction; drug-herb interaction

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Relationship Between Knowledge on Diabetes Mellitus, Mental Health Status, and Health-Related Quality of Life Among Patients with Type 2 Diabetes Mellitus

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Diabetes Mellitus (DM) results in elevated blood glucose levels that lead to various macrovascular and microvascular complications if untreated. In Malaysia, an estimated diabetes population of 3,529,804 was reported in 2015. Studies showed that there is a link between diabetes knowledge, depression, and health-related quality of life (HRQoL). Hence, this study aimed to investigate the relationship between diabetes knowledge, depression, and HRQoL among T2DM in a tertiary institution. A cross-sectional survey and clinical data-based study was conducted in Hospital Canselor Tuanku Muhriz (HCTM), Malaysia from September to November 2019. DKQ, PHQ-9, and SF-12 questionnaires were used to determine patient's diabetes knowledge, depression symptoms, and HRQoL respectively. Physical component summary (PCS) and mental component summary (MCS) were the 2 domains for HRQoL. A total of 106 respondents was recruited, and half of the respondents had poor diabetes knowledge. Diabetes knowledge was significantly associated with the academic level and income ($p < 0.05$). The higher the level of diabetes knowledge, the lower the level of depression symptoms. A total of 28.6% of the respondents were found to have depression symptoms. There was a significant association between depression and exercise frequency ($p < 0.05$). For HRQoL, the mean scores of PCS and MCS were 43.34 ± 8.93 and 52.11 ± 7.52 respectively, compared to the standard norm mean of 50 ± 10. The number of comorbidities, medications, and hospitalisations were significantly associated with PCS but not MCS ($p < 0.05$). Depression is a common comorbidity in T2DM patients. Healthcare providers should provide psychological support, pharmacological care and diabetes education programs that are important in self-care management of patients to maintain a good glycemic control, preventing depression, reducing complications, and improving their health-related quality of life.

Keywords: knowledge; depression; quality of life; diabetes mellitus; Malaysia

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Anatomical, Physiological and Pharmacological Examination of Pathways Mediating the Effects of Electrical Stimulation ofthe External Auricle of the Ear on Autonomic Nervous System in Rats.

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The Auricular Branch of the Vagus Nerve (ABVN) is a sensory nerve that innervates select areas of the external auricular dermatome. Electrical stimulation of the auricular region innervated by the ABVN influences the autonomic nervous system, observed by changes in control of the heart in humans and animals. However, the pathways and mechanisms for these effects are unknown. We investigated the pathways mediating the effects of electrical stimulation of the external auricle in rats, comparing an ABVN innervated site of the external ear (the tragus) to an area not reported to receive ABVN innervation, the earlobe. Injection of the neuronal tracer cholera toxin B chain (CTB) into the right tragus (n=4) and right earlobe (n=4) revealed a large degree of similarity in sensory afferent termination sites. Afferent terminals were predominantly labelled ipsilateral to the injection site, with the densest labelling within laminae III-IV of the dorsal horn of the upper cervical spinal cord. Physiological recordings of the responses to ear stimulation were made in an anaesthetic free Working Heart Brainstem Preparation (WHBP) of the rat. Electrical stimulation (100 Hz, 2.5 mA) was delivered for 5 minutes into the auricular stimulation sites in the WHBP. Direct recording from the sympathetic chain revealed a central sympathoinhibition from both tragus and earlobe stimulation. Sectioning of upper cervical afferent nerve roots silenced the sympathoinhibitory effects of tragus stimulation. The sympathoinhibition were further tested in rats with isoprenaline induced myocardial infarction (MI). Langerndorff experimental setup to examine the cardiac haemodynamic property found the electrical ear stimulation significantly improved left ventricular developed pressure in MI rats thus suggests cardio protection from post MI left ventricular remodelling. The cardio protection is however abolished in the presence of atropine, a nicotinic receptor blocker. Considering the predominance of afferent labelling in the cervical spinal cord dorsal horn and that cervical afferent nerve section reduced the sympathoinhibition evoked by tragus stimulation, this suggests that the autonomic effects of auricular stimulation are conveyed through somatosensory afferents rather than the ABVN. However, the Langerndorff examination does suggest the auricular stimulation is mediated by the nicotinic activation associated pathway highlighting the interlinking between these two pathways.

Keywords: Auricular Branch of the Vagus Nerve; electrical stimulation; Langerndorff

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Christia vespertilionis extract inhibits monocyte adherence to endothelial cells through inhibition of pro-atherogenic adhesion molecules expression.

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The progression of atherosclerosis is currently believed to involve the interaction of monocytes with the vascular endothelium. This process is predominantly mediated by cellular adhesion molecules, which are expressed on the vascular endothelium and on circulating leukocytes in response to several inflammatory stimuli. *Christia vespertilionis* (CV) is herbaceous plants that traditionally used for treatment of various inflammation-related ailments. However, there is limited evidence that points to the protective activity of CV against atherosclerosis.This study aimed to investigate the effect and potential mechanism of CV leaves extract on TNF-α-induced adhesion of macrophage to endothelial cells.3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT) assay was used to determine the effect of GP extract on the cell viability of human vein endothelial cells (HUVECs). Monocyte-endothelial adhesion assay was carried out to determine the effect of GP extract on monocyte adhesion. Western blotting was used to determine protein expression of vascular cell adhesion molecule-1 (VCAM-1).Cell viability was maintained at 80% following 24 hours treatment with CV extract at concentration ranging from 5 µg/mL to 60 µg/mL. CV extract (20, 40 and 60 µg/mL) showed significant inhibitory effect on TNF-α-induced monocyte adhesion in HUVECs ($p<0.05$). All selected treatment concentrations of CV extract (20, 40 and 60 µg/mL) also significantly inhibited ($p<0.05$) protein expression of VCAM-1. Protein expression of ICAM-1 was significantly inhibited ($p<0.05$) by 40 and 60 µg/mL CV extract. Results from this study demonstrated CV extract possessed inhibitory effect of on monocyte adhesion to endothelial cells by inhibiting protein expression of VCAM-1 and ICAM-1. This result implicate that CV potentially have beneficial use particularly in vascular inflammation.

Keywords: Atherosclerosis;*Christia vespertilionis*;adhesion molecules

IRCPAS/2020/OP-518

Reinforcing effects of methoxyphenidine and methiopropamine in mice

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New Psychoactive Substances (NPS) or also known as "legal highs" are mainly of synthetic origin and openly sold by online-based vendors as unregulated drug alternatives. Methoxyphenidine and methiopropamine are two of the many NPS with reported cases of intoxication resulting in serious or fatal outcomes. However, preclinical safety assessments of the abuse liability of these NPS are scarce. This study was carried out to determine the reinforcing effects of methoxyphenidineand methiopropamine usingthe conditioned place preference test in mice.A total of 40 male Swiss Albino mice weighing between 25-35 g were randomly divided into five groups (n=8) for each tested substance. Each treatment group

received intraperitoneal injections of either normal saline orvarying dosesof methoxyphenidine (5, 10 and 15 mg/kg) or methiopropamine (1, 2 and 3 mg/kg). The positive control groups for methoxyphenidine and methiopropamine received 6 mg/kg ketamine and 3 mg/kg amphetamine, respectively. The conditioned place preference test was carried out in three phases; pre-conditioning; conditioning and post-conditioning phase. During the conditioning phase, test groups received methoxyphenidine/methiopropamine or the positive control drugs on alternate days in a drug-paired compartment. The time spent in thedrug-paired compartment was scored to calculate the percentage preference towards the drug-paired compartment during post-conditioning phase. Methoxyphenidine at 10 and 15 mg/kg showed a significant difference (P<0.05) in the percentage preference of the drug-paired compartment as compared to the vehicle and positive control groups. Similarly, methiopropamine demonstrated a positive reinforcement at 3 mg/kg but not at lower doses. The reinforcing effects of these substances were comparable to those of their respective positive controls. The reinforcing effects observed with methoxyphenidine and methiopropamine in mice strongly suggests that these substances have potential to cause addiction in humanslike ketamine and amphetamine.Further studies to elucidatethe mechanisms of action involved are also essential for abetter understanding of their effectsin the brain.

Keywords: Methoxyphenidine; Methiopropamine; new psychoactive substances; reinforcing effects; conditioned place preference

IRCPAS/2020/OP-519

In Vitro Content Estimation and Release Profile Evaluation of FGF7 and EV from FGF7: β-CD:EV Complex

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Keratinocyte growth factor (FGF7) mediates its effect through binding specifically to its receptor (FGFR) and promotes receptor dimerization and activation. Activation of FGFR regulates differentiation, proliferation, and migration of cancer cells. Elevated levels of FGF7 are detectable in patients with colorectal cancer. FGF7 is currently the potential targets to develop anticancer drugs through inhibiting the deregulated blood vessel formation in cancer cells. However, the influence of this growth factor on the efficacy of anticancer therapeutics was investigated in our previous work. Our study revealed that the beta-cyclodextrin (β-CD): FGF7 complex has the potential to improve the antiproliferative effect of everolimus (EV) by preventing FGF7 receptor activation and by enhancing EV cellular uptake and intracellular retention. The *in vitro* estimation of FGF7 and EV from FGF7: β-CD:EV complex is challenging. Therefore, in this study we've developed a method to estimate the content and evaluate the release profile of FGF7 and EV from FGF7:β-CD:EV complex by utilizing a size restricted dialysis tubing procedure and measure their concentrations at different time intervals by KGF (FGF7) ELISA kit and HPLC, respectively.In this study, a size-restricted dialysis tubing procedure was employed to measure the content and evaluate the release profile of FGF7 and EV from known amount of FGF7: β-CD:EV complex or pure samples. Briefly, the dialysis bags (MWCO 2000 Da) were filled with samples along with cell culture media and placed in 50 mL of 0.05 M PBS (pH 7.4) at 37°C with slow magnetic stirring (50 rpm) under perfect sink conditions. For the free EV determination, one mL of aliquots was withdrawn from the

external solution and replaced with the same volume of fresh PBS. The free EV concentration was determined by HPLC at 278 nm. Thereafter, the determination of the free FGF7 was performed by analyzing the internal content of the dialysis bag and detected by KGF (FGF7) ELISA kit.The content and the release profile of FGF7 and EV from both pure samples and FGF7: β-CD:EV complex sample were evaluated. The results revealed that after 15 min, around 27% of EV was detected in the receiver compartment from the pure sample. On the other hand, up to 30 min of experiment, there was no detection of free EV from the FGF7:β-CD:EV complex sample in the receiver compartment, and about 35% of free EV was detected after 1 h.Finally, 23% of free FGF7 was detected in the internal content of the dialysis bag after 1 h.In this study, a size-restricted dialysis tubing procedure was employed to measure the content and evaluate the release profile of FGF7 and EV from known amount of FGF7:β-CD:EV complex and measure their concentrations at different time intervals by KGF (FGF7) ELISA kit and HPLC, respectively. Our findings suggested that the complex is stable in cell culture media for at least 30 min and exhibits sustain release profile (up to 4 h).

Keywords: FGF7, everolimus, size-restricted dialysis tubing, ELISA assay, HPLC

IRCPAS/2020/PP-103

Iridoid glycosides from *Scrophularia oxysepala*

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The Scrophulariaceae family consists of 220 genera in which Scrophularia genus is known for its active phytochemical compounds. The Scrophularia genus consists of 60 species in the flora of Iran. The roots and stems of many of them are used as antipyretic, febrifuge and antibacterial in traditional central and west Asian medicine. It is also used as a remedy for erythema, inflammations, ulcerous, and in the treatment of cancer.Previous investigations have been shown that this genus is a rich source of iridoid glucosides and phenylpropanoid glycosides. To isolate and characterize compounds from *S. oxysepala***.** The air-dried and powdered aerial parts of *S. oxysepala* (1800 g) were Soxhlet-extracted with n-hexane, dichloromethane (DCM), and methanol successively (2 L each). All these extracts were separately concentrated using a rotary evaporator at a maximum temperature of 45°C. A portion of the MeOH extract (2 g) was subjected to solid phase extraction (SPE) on a Sep-Pak (10g) C_{18} cartridge using a step gradient of methanol: water mixture (10: 90, 20: 80, 40: 60, 60: 40, 80: 20, 100: 0). The preparative HPLC (Dr. Mainsch GmbH ODS column 20 µm, 250mm × 20 mm); linear gradient 0-45 min 20-90% methanol in water; isocratic gradient 90% methanol in water during 45-50 min; linear gradient 50-52 min 90-100% methanol in water; isocratic gradient 52-55 min 100 methanol; linear gradient 55-58 min 100-20% methanol in water; isocratic gradient 20% methanol in water during 58-65 min; flow rate = 8 ml/min detection at 190-400 nm. We report here isolation and structure determination of chemical compounds of the aerial part of *S. oxysepala.* According to this research, we isolate scrolopioside E (1), gmelinoside M (2), scrophuloside A_9 (3), scrophulosideA_3 (4), loganic acid (5), 7-O-acetyl loganin (6), 8-O-acetyl muralioside (7) and 2- [2-(methoxy methyl) phenyl-β-D-arabinoside (8). All compounds isolated as amorphous powders. The UV spectrum exhibited absorption bands between 215-280 nm characteristic of an iridoidenol ether system and cinnamoyl and cinnamoyl derivatives chromophore. The^{13}C-NMR spectrum showed 6-O-α-L-rhamnopyranosyl-8α-hydroxymethyl-1α, 5β, 6α, 7α, 9β pentahydro-7(8)-epoxy-2oxaind-3-ene1-β-D-glucopyranoside, consisting of

iridoide core, glucose and rhamnose in compounds. Compound 1 called scrolopioside E and Compound 2, gmelinoside M, obtained as amorphous powders, have the molecular compositions $C_{43}H_{50}O_{19}$ and $C_{34}H_{42}O_{17}$ based on high resolution elemental analysis The IR spectrum indicated the presence of a hydroxyl group (3410cm^{-1}), ester group (1698cm^{-1}), double bond (1651 cm^{-1}) and oxirane ring (1193, 840cm^{-1}).

Keywords: *Scrophulariaceae; Scrophularia oxysepala;* Iridoid glycosides; NMR

IRCPAS/2020/PP-106

An *in-vitro* analysis of the staining effect of three different chemical mouthwashes

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Every patient desire to have a radiant white smile without any tooth staining. There is an increasing trend of using mouthwash to maintain oral hygiene and to keep gums healthy. Most popular in the market is Chlorhexidine and Listerine mouthwash. But when used for a long time, they may lead to staining of the teeth. Nowadays chlorhexidine mouthwash with Anti Discoloration Solution (ADS) is available to reduce staining of teeth. Therefore, this study was conducted to evaluate the staining potential of three commercially available types of mouthwashes.To evaluate the staining potential of three commercially available types of mouthwashes.Sixty extracted maxillary central incisor teeth were used for the study. The teeth were randomly divided into three groups (N =20): Group A, B, C [Group A: Listerine (Listerine Mouthwash Original)Group B: Chlorhexidine mouthwash – 0.12% (Hexidine Mouthwash)Group C: Chlorhexidine mouthwash with Anti Discoloration Solution (ADS) (Curasept ADSMouthwash)]. Before the colorimetric measurements, the teeth were cleaned with pumice slurry and rubber prophylactic cups, rinsed with water and then immersed for 24 hours in distilled water at 37°C. The Easyshade spectrophotometer (Vita Zahnfabrik, Germany) was used to assess the color of buccal enamel surface (T1, baseline examination) according to the CIELAB (Commission Internationale de l'Eclairage L*a* and b*) color space system. After baseline color examination, the teeth were immersed in three different mouthwash solutions and stored for 2 months. After the time period of 2 months, the teeth were washed and dried. The colour change (ΔE)was measured with the spectrophotometer after 2 months (T2). The ΔE > 3.5 was considered as a clinically perceptible color change.Statistical analysis was done using ANOVA test and Paired T-test.Greatest staining was caused by Listerine followed by Chlorhexidine mouthwash between the baseline and 2 months' time interval. Least was caused Chlorhexidine mouthwash with ADS, which was statistically significant ($P<0.05$).Chlorhexidine mouthwash with ADS (Anti Discoloration Solution) is a better choice as a mouthwash to minimize tooth staining.

Keywords: Staining; fluoride; chlorhexidine; mouthwash; spectrophotometer.

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Ocimum inhibits airway inflammation in cigarette smoke induced COPD

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Chronic obstructive pulmonary disease (COPD), a major respiratory disorder has been ranked as one of the top four high mortality diseases worldwide. The foremost cause of COPD includes exposure to cigarette smoke and other noxious particles and gases. The pathological condition in COPD finally leads to persistent airway inflammation and deterioration in lung function. The current pharmacological treatment for COPD is symptomatic and based on the use of bronchodilators, as β_2-adrenergic agonists, anticholinergics, theophyline, associated with certain limitations due to which steroids are still preferred over these drugs inspite being associated with number of adverse effects. Hence, there is urgent need for a therapeutic approach to explore novel agents which could act on specific areas of the inflammatory cascade without any serious side effect.To evaluate the therapeutic role of *Ocimum sanctum Linn* (Black tulsi) on the cigarette smoke inducedairwayinflammation**.** Balb/c mice (7-9 weeks; 20-23 gms) were exposed to cigarette smoke in a closed chamber thrice a week. Ethanolic herbal extract of *Ocimum*was administered at a dose of 200 and 400mg/kgbw intraperitonally one hour before smoke induction. 24 hours after the last smoke exposure mice were sacrificed. BALF, serum and lungs were stored to study total cell count, differential cell count, eosinophil peroxidase, myeloperoxidase, neutrophil elastase, and histology for inflammation. Ethanolic extract of *Ocimum* inhibited the total cell count in dose dependent manner which was elevated in COPD mice. Also, the inflammatory markers as eosinophil peroxidase, myeloperoxidase, neutrophil elastase were attenuated in dose dependent manner in *Ocimum* administered mice. Histology of lungs section reveals architectural changes in the alveolar spaces and reduced bronchiole in COPD mice which was also recovered in *Ocimum* administered mice. Medicinal plants are one of the vital resources for alternative medicine and indeed several potent drugs for various human disorders including respiratory diseases have been derived from plant origin. *Ocimum*has therapeutic potential and present study proves its potent efficacy in COPD. It may prove to be a lead molecule for developing potent COPD drug in future with great clinical relevance.

Keywords: Chronic obstructive pulmonary disease; *Ocimum sanctum Linn* (Black tulsi)

IRCPAS/2020/PP-111

Antibacterial activity of Piper betle against Acne causing bacteria

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Acne vulgaris is the most common cutaneous disorder in adolescents and young adult, due to blockage or inflammation of pilosebaceous units caused by bacteria. *Staphylococcus epidermidis* and *Propionibacterium acne* are the major skin bacteria causing inflammation in acne. Many topical antibiotics combined with therapy has become standard for the management of acne, but the gradual development of antibiotic resistance is very hard to prevent and can affect the acne cure. May studies showed the *Piper betle* has antibacterial potential. The aim of this study is to investigate the antibacterial activity of *Piper betle* extract against *Staphylococcus epidermidis* and *Propionibacterium acne. Piper betle* leaves were extracted by

using ethanol. Photochemical group tests were done to detect the photochemical properties of the extract. Disc diffusion method was adopted for evaluation of antibacterial activity of *Piper betle* extract to determine the minimum inhibitory concentration (MIC). Photochemical analysis was found to be positive for flavonoids, tannins and saponins. The result from disc diffusion method showed that *Piper betle* extract could inhibit the growth of *S. epidemidis* and *P. acne*. Hence, *Piper betle* extract has potential to be used as an alternative antibacterial agent for acne causing bacteria, *S. epidermidis* and *P. acne.*

Keywords: *Piper betle*; antibacterial; *Staphylococcus epidermidis; Propionibacterium acne*

IRCPAS/2020/PP-121

Developmental toxicity evaluation of Sub-CO2 extract of *Phaleria macrocarpa* fruit flesh in Danio rerio embryo

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Phaleria macrocarpa, traditionally known as Mahkota dewa, is a therapeutic herb that is being used for anti-obesity, antidiabetic, antioxidant, and anticancer activities. To perform animal study and administration dose measurement, to identify the safety profile level of plant extract is very important. The main aim of this study is to evaluate the toxicity level of the subcritical carbon dioxide (Sub-CO2) extract in Danio-rerio embryo. Fruit was chosen and subsequently extracted using Sub-CO2 technique. The Box-Behnken Design (BBD) was used to optimize and examine the effect of independent variables of Sub-CO2 such as temperature (27-29˚C), pressure (6.8-7.0 MPa) and concentration of co-solvent. The toxicity study of Sub-CO2 extracts was examined by using Danio-rerio embryo at 0 to 96 hpf. The protocol of toxicity study on zebrafish embryo was adapted according to OECD guideline section 212.Sub-CO2 extract of *P. macrocarpa* was applied to screen the toxicity level at the dose of 100 – 1000 µg/ml. The tested report showed considerable (>90%) mortality and delay in hatching at higher concentrations of 1000 µg/ml. Overall, there was a mild toxicity observed in experimental embryo development when compared to control. However, the LC50 value was obtained at the concentration of 841.39 µg/ml. Toxicity study of different ratio of Sub-CO2 extracts from *P. macrocarpaa* showed mild toxic effects on zebrafish embryos, however, the extract comparatively safe for normal embryonic development at the range from 50 to 841.39 µg/ml. This finding could help to predetermine the dosage *P. macrocarpa* fruit for further *in vivo* study.

Keywords:Subcritical CO2 extract; Zebrafish embryo*; Phaleria macrocarpa fruit;* co-solvent; toxicity

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Hematopoieticconstituents from Japanese herbal prescription "Ninjin'yoeito" (Ren-Shen-Yang-Rong-Tang)

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Severeanaemia that is resistant to medicinal treatment is often observed in patients with malignancies and those who are undergoing chemotherapy, and pathogenesis of the anaemia is multifunctional. A low level of circulating hematopoietic growth factor such as erythropoietin (EPO), shortened survival time of circulating red blood cells (RBCs), and a decrease in number of immature erythroid cell in bone marrow probably due to chemotherapy or chronic inflammation have been demonstrated to be the causes of anaemia. To cope with chronic anaemia, transfusion as well as EPO has been used clinically in various situations. Besides hematopoietic growth factors EPO, herbal prescriptions such as Ninjin'yoeito have been used clinically to ameliorate the erythrocytopenia of patients who are undergoing cancer chemotherapy. Herbal prescription,"Ninjin'yoeito (NYT)" that have been clinically used to ameliorate the anaemia and disruption of marrow function were shown to stimulate marrow cell proliferation; however, its active substances were not identified. We tried to explore the active hematopoietic constituents as a marrow cell proliferation-promoting compounds from the NYT by anaemia model *in vivo* and marrow cell *in vitro* culture systems.Hematopoietic efficacy of Ninjin'yoeitowas assessed using anemia model in mice-administered with 5-fluorouracil (5-FU). The active constituent(s) from the extract were isolated in accordance with bioassay-guided fractionation using liquid and colony-forming units assay system in cultured marrow cells*in vitro*. Orally administered NYT inhibited the decrease in peripheral reticulocytes and bone marrow cell counts by 5-FU on day 10 and remarkably hastened the recovery of them on day 20. Erythroid progenitor colonies such as CFU-E and BFU-E formed by marrow cells from mice-treated with 5-FU were significantly increased by oral administration of NYT. *Glycyrrhizae* radix (licorice) and Schisandra fruit strongly enhanced the marrow cell proliferation and the formation of immature marrow cell colonies co-stimulated with interleukin-3and EPO. Flavonoid glycosides, liquiritinand its derivative, were identified as active constituentsof licorice, and hydroxymethylfurfuralwas also purified as active compound from Schisandra fruit. NYT had a potential to protect against hematotoxicity and had a hematopoietic activity through stimulation of immature erythroid progenitor cell differentiation, and liquiritin from licorice and liquiritinderivative from Schisandra fruit could stimulate proliferation of marrow cells suggesting two herbal medicines might be hematopoietic components of NYT.

Keywords:Natural products; bioactive constituents; Kampo prescription;hematopoietic activity; anemia; marrow cell proliferation; liquiritin

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Pharmacognostic and Phytochemical Analysis of Feverfew and Butterbur Plant

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Feverfew (*Tanacetum parthenium* L.) & butterbur (*Petasites hybridus)* are the plants belonging to same family of Asteraceae. These herbs were used as traditional herbs and folk remedies among herbalists. The main active components present in this family include petasin, isopetasin and sesquiterpene lactones. The other chemical constituents present in these herbs include isofraxidin, coumarin, and isofraxidin. Dimentyl ether was present in the root of feverfew. Nearly 90% of volatile oils have been identified in feverfew plant. These active components have been reported to be effective in reducing the severity of migraine. They also have antihistamine and antileukotriene activity. Both the herbs were also used in skin conditions like psoriasis, allergies, contact dermatitis and skin inflammation. Plant materials were extracted. Biological studies and phytochemical analysis were performed.The results showed that the total extract of *Tanacetum parthenium* &*P. hybridus*have antimicrobial and anti-fungal activities. The phytochemistry analysis (TLC) showed a best separation of the extract using different solvent systems. The phytoconstitutents present in these herbs include terpenoids, alkaloids, glycoside, and flavonoids. The present research work revealed that feverfew and butterbur extract have positive biological effects.

Keywords: Feverfew & butterbur extract; TLC analysis; biological studies; antimicrobial; antifungal

IRCPAS/2020/PP-136

Diuretic activity from Nanoparticles of Ekor Naga Leaves (*Rhaphidophora pinnata* (L.f.) Schott)

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Kidney stone disease (nephrolithiasis) is one of the factors causing chronic kidney failure. The decoction of the *Rhaphidophora pinnata*has been used empirically by the community to treat various diseases. However, the diuretic effect has not been conducted yet. The objective of this research is to investigate the diuretic activity of nanoparticle *R. pinnata* nanoparticles. *R. pinnata* dry leaves were made into nanoparticles with high-speed milling. Herbal was then tested the diuretic activity by using Wistar rats. The doses of testing were 100, 150 and 200 mg/kg BW. Furosemide dose of 21.6 mg/kg BW was used as a comparative group. The parameters were urine volume, and electrolytes contents including sodium, potassium, and calcium. Result: The result showed that 24 hours total urine volume from rats were given *R. pinnata* dose of 100, 150 and 200 mg/kg BW respectively as follow: 6.050±1.3301 mL; 10.500±1.9149 mL; 18.500±4.7958 mL, while normal control was 7.475±1.3200 mL and furosemide was 11.750±1.7078 mL. Therefore only *R. pinnata* doses of 150 and 200 mg/kg BW gave the diuretic effect that significantly different to normal control group ($p<0.05$). The results of electrolyte measurements showed an increase in sodium, potassium, chloride as well

as calcium levels ($p<0.05$) at each dose administration of *R. pinnata* nanoparticle. The highest electrolyte yield is shown by a dose of 200 mg/kg BW. *R. pinnata* nanoparticles with the mechanism of action increasing urine flow (diuretics) can be potential as herbal medicines and phytopharmaca to treat nephrolithiasis.

Keywords: Ekor naga leaves; *Rhaphidophora pinnata* (L.f) Schott; diuretic; antinefrolithiasis; nanoparticles

IRCPAS/2020/PP-144

Analgesic and Anti-inflammatory Potential of Four Varieties of Bell Pepper (*Capsicum Annum* L.) in Rodents

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Use of *Capsicum annum* L. for culinary purpose dates to centuries. Its medicinal benefits have also been investigated in past few days. Analgesic and anti-inflammatory activity in 4 different colored (green, yellow, orange, and red) sweet bell peppers (*Capsicum annuum* L.) were investigated in the doses of 200 and 400 mg/kg. Current research is being directed at authenticating if *Capsicum* can be used as an analgesic and anti-inflammatory comparing the effect of most used analgesic aspirin. The effects of ethanol extract of *Capsicum annum* L. were determined for analgesic activity by acetic acid induced writhing, tail immersion and hot plate test. Animals were divided into 10 groups (n=7): (1) Control (2) CAG 200 (3) CAG 400 (4) CAR 200 (5) CAR 400 (6) CAO 200 (7) CAO 400 (8) CAY 200 (9) CAY 400 (10) Standard. All the extracts given intraperitoneally in rat hind paw of seven animals in each of four treatment groups received 200, 400 mg/kg. Acute toxicity was also determined by increasing the dose till 3000 mg/kg, which showed no evidence of mortality. Statistical calculation was done by SPSS software to compare the effects of aspirin and *Capsicum* extract. Positive results were obtained as compared to control group; analgesic effect was statistically significant ($P<0.05$). These observations revealed that the fresh fruits extract of four kinds of Bell pepper at doses of 200 mg/kg and 400 mg/ kg possess anti-inflammatory and pain suppressing activities possibly mediated via PG synthesis inhibition.

Keywords: *Capsicum annum*; analgesic; anti-inflammatory

IRCPAS/2020/PP-148

Comparative studies on *in vitro* cytotoxicity properties of *Areca catechu* nut on human melanoma cell lines

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Nowadays the bioactive product from plants plays an important role in the therapeutic applications. Areca nut (Brown variety and Red variety) is the seed of *Areca catechu* plant widely consumed by all age groups in worldwide. The seed has high content of phenolics, and

flavonoids can be used as antioxidant, antiaging, antihelmintic, antimicrobial, analgesic, and anti-inflammatory in the field of medicine. The aim of this study was to investigate the phytochemical analysis, gas chromatography–mass spectrometry (GC-MS) analysis of Areca nut and their cytotoxicity effects, in human melanoma cell lines (A375). Areca nut (Brown) and Areca nut (red) samples were subjected to Soxhlet extraction method using aqueous ethanol. Phytochemical screening was done by using different biochemical tests. The extracts were further subjected to TLC and GC-MS analysis. *In vitro* anticancer activity on human melanoma cell lines (A375) was evaluated by (3-(4, 5-dimethyl thiazole-2yl)-2, 5-diphenyl tetrazolium bromide) MTT assay. Phytochemical screening confirmed the presence of phytoconstituents like alkaloids, flavonoids, phenols, tannins, glycosides, terpenoids, steroids, proteins, quinones and saponins. Total of five bands were observed Areca nut (Brown) and six bands were observed Areca nut (Red) in TLC fingerprinting. The Areca nut (Brown)extract from GC-MS shows the following active constituents i.e (2S,3S)-(-)-3-Propyloxiranemethanol, n-Propylmalonic acid, 1-acetyloxydodecyl acetate, 2-Nonenoic acid, Carbromal, 3-Nonenoic acid, 2-aminooxypentanoic acid, 2R,3S-9-[[1,3-dihydroxy-4-fluoro-3-butoxy]methyl]guanine. The IC_{50} values of Areca nut (Brown) and Areca nut (Red) extracts in A375 cell lines were found to be 6.28 ± 2.04 µg/ml and 9.62 ± 3.07 µg/ml and 1.8 µM for cisplatin respectively. The result indicates that Areca nut (Brown) showed better anticancer activity against human melanoma (A375) cell lines than Areca nut (Red). Thus, the regular consumption of Areca nut (Brown) can reduce the prevalence of melanoma cancer.

Keywords:*Areca catechu;* melanoma cell lines (A375); phytochemicals; TLC; GC-MS; cytotoxicity

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Antidiabetic Activityof Ethylacetate Fraction of *Orthosiphon glabratus* Benth. Against STZ Induced Diabetic Rats

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Diabetes is a serious public health issues that is approaching epidemic proportions in whole world. In most of the developed countries, diabetes is the 4th leading cause of death. According to IDF (International Diabetes Federation) estimates on comparative prevalence of diabetes during 2007 is 8.0 % and it is likely to increase to 7.3% by 2025. Due to increased efficacy of plant derived drugs and presence of side effects of conventional medicines, there has been increased scientific interest in medicinal plant to treat diabetes. To access the antidiabetic and hepatoprotective activity of *Orthosiphon glabratus* leaves on streptozotocin induced diabetes in wistar rats. Powdered *O.glabratus* of dried leaves was primarily extracted with solvent ethanol and subjected to fraction with ethyl acetate. Further preliminary phytochemical analysis was done with a serious of tests. Diabetes was induced by single dose of intra-peritoneal injection of STZ (45mg/kg). The Diabetes induced animals were divided into 5 groups (Normal, Diabetic control, Diabetic +EAFOG (200mg/kg), Diabetic + EAFOG (400mg/kg) and Glibenclamide). The antidiabetic activity of ethyl acetate fraction of *O.glabratus* was determined using parameters including body weight, blood glucose, glycosylated hemoglobin and plasma insulin. Serum liver parameters were also measured including aminotransferase (AST & ALT) and Phosphatase (ALP). Histopathology of pancreas was also screened. Phytochemical analysis showed the presence of alkaloids, tannins,

flavonoids, phenols, cardiac glycosides, and terpenoids. A dose dependent increase in bodyweight and plasma insulin and significant decrease in blood glucose and glycosylated Hb were observed in diabetic rats when treatment with ethyl acetate fraction of *O.glabratus* leaves. Similarly the treatment with EAFOG showed significant reduction liver marker enzyme AST, ALT & ALP comparable as reference standard glibenclamide. Histological section of pancreas showed regeneration of β cells. Secondary plant metabolites reported to possesses wide range of physiological & pharmacological activity. Hyperglycemia is due to by the cytotoxic effect of STZ on GLUT2 receptor on β cells. The treatment with EAFOG showed reduction in BGL & HbA1c & increased body weight & plasma insulin and restoration β cell architecture which indicates, the management of muscle wasting and improved glycemic control of EAFOG. Decreased level of AST, ALT & ALP on EAFOG treatment showed protective effect of EAFOG on liver against hyperglycemic oxidative stress. From these findings suggest that EAFOG might be considered as potential source of phytoconstituents with potent antidiabetic and hepatoprotective activity.

Keywords: Antidiabetic; hepatoprotective activity;ethylacetate fraction; *Orthosiphon glabratus;* Wistar rats.

IRCPAS/2020/PP-150

Antioxidant and antihyperlipidemic activity of methanolic fraction of *Maytenus heyneana* root

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Oxidative stress is known to be a component of molecular and cellular tissue damage mechanism in a wide spectrum of human disease. Oxidative stress in diabetes is coexists with a reduction in the antioxidant power. Hyperglycemia & hypercholesterolemia were associated with oxidative modification of LDL-C, protein glycation, glucose –autooxidation. The harmful effects of oxidation stress can be rendered by a constant supply of natural products.The present study is aimed to study the antioxidant and antihyperlidimic activity of methanol fraction of *Maytenus heyneana* on STZ induced diabetic rats*Maytenus heyneana* root was extracted with ethanol further it was fractioned with methanol and the obtained samples were subjected to preliminary phytochemical, *in vivo* antioxidant and antihyperlipidemic analysis. Single dose intra peritoneal injection of STZ (45 mg/kg) was used to induce the hyperglycemia. To confirm the hyperglycemia, blood glucose was measured, whereas hyperglycemia induced oxidation was determined by using enzymatic (SOD& CAT), non-enzymatic (GSH) antioxidants and oxidative stress parameter in liver tissue was evaluated by LPO(TBARS). To access the antihyperlipidemic activity, the Total cholesterol, TG, HDL-C and LDL-C were measured. Histology of liver was screened. Phytochemical studies revealed the presence of alkaloids,saponins,flavonoids,phenols,cardiac glycosides and terpenoids. Treatment with the methanolic fraction of *Maytenus heyneana* was effective in reducing the blood glucose level and also found to be potent antioxidant by significantly increase SOD, CAT & GSH and significant decrease in oxidative stress LPO. The dose dependent MFMH on antihyperlipidemic activity was observed by ameliorating the increased level of Total cholesterol, TG,LDL-C and increased the level of HDL-C. Degenerated hepatocytes of STZ diabetic rats were restored to normal morphological features as like reference standard glibenclamide.The phytochemical analysis showed maximum phytochemical present in

MFMH which may be responsible for various physiological activity. Administration of MFMH to diabetic rats resulted in a significant restoration of blood glucose. A dose dependent MFMH & glibenclamide reverses the antioxidant parameters indicating that inhibition of liver tissue chain propagation reaction of LPO, protection of oxidative damage induced diabetic complications in liver tissue & increased plasma antioxidant defense system. A reduction in TG, Total cholesterol, LDL-C with increased HDL-C showed after 14 days treatment indicating the protective effect MFMH on diabetic dyslipidemia. Hence from observation, the MFMH possesses antioxidant as well as antihyperlidimic activity on STZ induced diabetic rats.

Keywords: *Maytenus heyneana* root; antioxidant; Antihyperlipidemic activity; Methanolic fraction; STZ diabetic rats

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Anti-inflammatory Effects of 70% Binahong (*Anredera Cordifolia* (Ten.) Steenis) Ethanol Extract in Rats

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Binahong (*Anredera cordifolia* (Ten.) Steenis) is one of traditional plants of Indonesia which is used as anti-inflammatory agent by ancestors of Indonesia. This study aims to prove the anti-inflammatory effect of 70% Binahong (*Anredera cordifolia* (Ten.) Steenis) ethanol extract on the edema of white mouse paw induced with a 5% egg white solution. The subjects of this study were 25 male Sprague Dawley strain rats which were divided into 5 groups, each of which amounted to 5 individuals. The control group were 0.5% CMC (negative control), Diclofenac Sodium 13.5mg/kg BW (positive control) and the treatment group of 50 mg / kg BW, 75 mg / kg BW, and 100 mg / kg b.w. , given orally. The soles of the mouse paw were injected subplantarly with a 5% egg white solution to trigger edema. Thee dema volume was measured using pletismometer within 30 minutes for 360 minutes. The results showed no significant difference between the administration of 70% Binahong ethanol extract of 75 mg/kg body weight and 100 mg/kg body weight with positive control. It was concluded that the 100 mg/kgBW70%ethanolextractofBinahongis more potential as an anti-inflammatory drug agent compared with the 75 mg/kg BW % ethanol extract of Binahong in terms of the percent inflammation inhibition onaverage.

Keywords: Anti-inflammatory; edema; Binahong Rhizome (*Anredera cordifolia* (Ten.)Steenis)

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Formulation of Merbau (*Intsia bijuga*) Wood Extract and Its Antioxidant Evaluation on H2O2-induced oxidative stress on HaCaT

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Degeneration and the aging process involve complex processes that are influenced by intrinsic and extrinsic factors. Free radicals in the form of reactive oxygen species (ROS) are the main causative factors in the process of skin aging both for intrinsic and extrinsic factors. In addition, excessive production of ROS causes oxidative stress which can contribute to inflammation,

allergic reactions, inhibit the healing process of the skin and trigger skin carcinogenesis. Therefore, antioxidant supplementation is needed on the skin that is able to suppress the cumulative effect of oxidative damage by using cosmetic products that have potent antioxidant activity. The active ingredients for cosmetic may derived from natural resources. Since Indonesia is known for its biodiversity, there should be a rigorous effort to utilize this resource as a way to increase the economic value of Indonesia's natural resources. One plant that has the potential to be developed as a cosmetic ingredient with antioxidant activity is merbau (*Intsia bijuga*) wood. Previous screening study of 35 Indonesia's medicinal plants showed that methanol extract of merbau wood had a potent antioxidant activity with an IC50 of 6.6 µg/mL based on DPPH assay. This research aims to formulate cream of methanol extract of merbau with antioxidant activity. Powdered extract of merbau wood was macerated using methanol and the dried extract was tested for its cytotoxicity and antioxidant activity on human keratinocytes cell line (HaCaT). Different formulations of cream containing non-toxic concentration of merbau extract were made and physical and stability evaluation were performed.The yield of merbau extract was 15%. Cytotoxicity evaluation demonstrated an IC50 of 181.3 µg/mL and antioxidant evaluation of merbau has been performed. Several formulations of merbau extract fulfilled the evaluation parameters. High concentration of merbau resulted significant damage to HaCaT ($p < 0.0001$) and non-toxic concentration should be use as active ingredient for cream. Our study demonstrated that several stable cream formulations of merbau extract was successfully formulated with tested antioxidant activity. Further studies are required to investigate which chemical compounds responsible for the antioxidant activity.

Keywords: *Intsia bijuga*; merbau; antioxidant; oxidative stress

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In vitro haemostatic activities of *Rhodomyrtus tomentosa* leaves and stem methanol extract

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Well known anticoagulant drugs have been reported with highly significant risk of severe or fatal bleeding complications for the patient. Looking forward for the solution to this problem, *Rhodomyrtus tomentosa* has the potential to be developed as the alternative natural-based anticoagulant. *In vitro* haemostatic activity of *Rhodomyrtus tomentosa* methanol leaves and stem extract ranging from 20 – 100 mg/mL were used to determine its effect on routine coagulation testing, which are Prothrombin time (PT), Activated Partial Thromboplastin Time (APTT) and Thrombin time (TT). Total phenolics (TPC) and flavonoids contents (TFC) in the extracts were determined using Folin-Ciocalteu and colorimetric assays with gallic acid and quercetin as standard respectively.The data were analyzed by using One-way Analysis of Variance (ANOVA) followed by post-hoc Dunnet's (1-tailed) and Pearson's correlation. PT revealed significant prolongation of time ($p < 0.05$) at 80 mg/ml to 100 mg/ml for the leaves, but for stem begin at 40 mg/ml, similar results obtained for APTT was at 100 mg/ml for both stem and leaves. As for TT displayed unreliable significant results at 20, 40 and 100 mg/mL concentration. Pearson's correlation test revealed that only APTT and PT tests are significantly correlated to TPC and for TFC showed significantly correlated with PT. In conclusion, *Rhodomyrtus tomentosa* has the profound effect of anticoagulant influenced by the

phytochemical content of phenolic and flavonoid, and it is recommended to identify specific compounds that responsible for anticoagulant effect of *Rhodomyrtus tomentosa.*

Keywords: *Rhodomyrtus tomentosa*, anticoagulant, phytochemicals.

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Antioxidant and Cell Wound Healing Properties of Ethanolic Extract of *Baeckea Frutescens* Leaves

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Baeckea frutescens is an evergreen, heather-like shrub or small tree growing up to 8 meters tall. In Peninsular Malaysia, *B. frutescens* is found on the mountain tops, quartz ridge and sandy coasts. There are numerous pharmacological reports on *B. frutescens* including anti-bacterial, anti-dysentery, anti-pyretic, influenza, coryza, epistaxis, sunstroke, fever, headache, measles, colic, abdominal pain, dyspepsia, jaundice, haemorrhagic dysentery and irregular menstrual cycle. In this study, the antioxidant and wound healing properties of ethanolic extract of *B. frutescens* leaves (BFLE) were studied to postulate the potency of this plant towards the development of wound healing agent. The antioxidant activity of the extract was determined by 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging properties, total phenolics content (TPC) and ferric reducing antioxidant power (FRAP). Cell viability and wound healing property were determined by MTT and cell migration assay respectively. The results of present study showed that DPPH radical scavenging ability of BFLE at 200 µg/ml was very high at 90.02 ± 4.27 % and the median effective concentration (EC_{50}) for DPPH radical scavenging ability was 23.52 ± 4.33 µg/ml. These result was comparable to positive control, green tea extract, which showed 90.84 ± 4.27% of DPPH radical scavenging value and with EC_{50} of 23.58 ± 2.34 µg/ml. Total phenolics content of BFLE was also high with 112.09 ± 3.52 mg GAE/g of extract, just slightly lower than the TPC value of green tea extract, with 177.19 ± 4.14 mg GAE/g of extract. The FRAP value of BFLE was 2.42 ± 0.15 mmol/dm^3 per g of extract, slightly lower than the FRAP value of green tea extract, at 3.19 ± 0.65 mmol/dm^3 per g of extract. Treatment with 25 µg/mL and 50 µg/mL of the extract has shown to increase the spreading of keratinocytes without inhibition on the cell viability.These results demonstrated that BFLE has very good antioxidant and wound healing properties and served as a very good candidate of wound healing agent. However, further studies were required to clarify the mechanism of actions into which pathways involved those contributed for their activities.

Keywords: *Baeckea frutescens;* ethanolic extract; antioxidant; wound healing; keratinocytes

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Identification of compensatory genetic variants associated with metronidazole and levofloxacin resistance in *Helicobacter pylori*

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Helicobacter pylori infections are generally treated with a high dose of single antibiotic or a cocktail of antibiotics together with a proton pump inhibitor. However, treatment failure can occur, most often because of the development of antibiotic resistance in *H. pylori*. Although the acquisition of antibiotic resistance has been postulated to exert a negative impact on bacterial fitness, the association has not been well-established. More importantly, compensatory mechanisms to cope with the cost of resistance in *H. pylori* has not been thoroughly investigated. In this study, we aimed to investigate the compensatory genetic consequences associated with metronidazole and levofloxacin resistance in *H. pylori*. For this, we focused on single nucleotide polymorphisms (SNPs) within the coding regions of the *H. pylori* genomes. Nine pairs of *H. pylori* strains comprising of four naturally occurring metronidazole resistant/sensitive dual population, and five *in vitro* induced levofloxacin-resistant/sensitive pairs, were obtained from the screening of 450 isolates. These paired isolates were verified to be derived from similar parental strains by rapid amplification of polymorphic DNA (RAPD). Mutations in *rdxA*, *frxA, gyrA* and *gyrB* were identified by PCR and Sanger sequencing. The paired strains were cultured, their genomic DNA were extracted and whole genomic sequencing was performed on the Illumina Miseq. The sequences of the resistant strains were assembled with SPAdes, annotated by NCBI, and aligned by mapping their genome with their sensitive wild type pair counterpart as reference using Bowtie2 and SNPs were compared between strains. Statistical significance of SNPs in association with antibiotic resistance was determined using Fisher's exact test and functional interactions of these genes were predicted *in silico* using STRING. From 967 SNPs in the resistant strains, 96 of them in *rfaJ*, *vacA*, *tonB*, *radA*, *lptA*, *hgrA*, *hgrC*, *surA*, *coaX, hsdM*, DUF262 and TIR domain-containing genes, and *ybeY* were found to have occurred significantly and more prevalent in the levofloxacin resistant strains as compared to the metronidazole resistant strains. Among these genes, *vacA*, *radA*, *coaX* and *tonB* were respectively linked to functional networks of energy metabolism, cell motility, lipopolysaccharide biosysnthesis and translation. SNPs were identified within the coding regions of genes that have been identified to be essential for bacterial vitality and might have been selected as a response to the challenge from antibiotics presence or as a survival mechanism. Different compensatory mechanisms for resistance may have developed for the various antibiotics, hence a deeper understanding of their importance will be essential in our battle against emerging antibiotic resistance.

Keywords: *Helicobacter pylori*; antibiotic resistance; SNP; compensatory mechanism

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Effect of Aqueous Extract of *Irvengia gabonensis* on Acetaminophen Induced Renal Toxicity

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The use of medicinal plant to prevent and/or cure kidney problems is a practice not peculiar to developing countries. This research work evaluated the curative ability of aqueous seed extract of *Irvengia gabonensis* on acetaminophen induced renal toxicity. A total of 30 albino rats were grouped into six groups (GI – GVI) of five rats each. GI served as normal control, GII served as positive control, GIII, GIV and GV were induced with kidney damage and administered with the extract at a dose of 50mg/kg, 100mg/kg and 150mg/kg respectively while GVI rats were administered with standard drug at a dose of 10mg/kg. Acute renal failure was induced in groups (II-VI) by single dose of 800mg/kg of acetaminophen. Three rats from group I and II were sacrificed 24 hours after acetaminophen administration to confirm inducement of kidney damage. Groups III, IV, V and VI were administered with the respective doses for two weeks. A significant increase ($p<0.05$) in the mean serum level of Urea, Potassium (K+), Chloride (Cl^-) and Creatinine with a significant decrease ($p<0.05$) in the level of serum Sodium (Na+) and Bicarbonate ($HCO3^-$) was observed in Acetaminophen induced group compared to normal control group. After two weeks of extract administration, significant decrease ($p<0.05$) in mean serum level of serum Urea, Potassium (K+), Chloride (Cl^-) and Creatinine with a concomitant increase in level of Sodium (Na+) and Bicarbonate ($HCO3^-$) was observed compared with positive control group. The curative ability of the plant may be connected to its reported secondary metabolites.

Keywords: Acetaminophen; curative; *Irvengia gabonensis*; renal function indices

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Hypolipidemic Properties of Bioactive Fraction(S) of *Mentha Piperita* Leaves in Poloxamer 407-Induced Hyperlipidemic Rats

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Hyperlipidemia is a condition characterized by high lipid level in the blood, and among the major risk factors of cardiovascular diseases. This study was primarily designed to investigate the hypolipidemic, activities of the extracts and bioactive fraction(s) of *Mentha piperita* leaves in Poloxamer 407-induced hyperlipidemic rats. Hypolipidemic activity of hexane, ethyl acetate and methanolic extracts (100,200 and 300 mg/kg) was evaluated *in vivo* by determining triglycerides, cholesterol, high density, and low-density lipoproteins levels in serum. The most potent hypolipidemic extract (Ethyl acetate at 100mg/kg body weight) was fractionated by column chromatography. Twelve (12) fractions coded F1-F12 were thereafter collected based

on gradient elution system of 100% hexane, followed by different combinations of hexane: ethyl acetate (90:10, 80:20, 70:30, 60:40, 50:50, 40:60, 30:70, 20:80, 10:90, 100% ethyl acetate) and finally washed with 100% methanol. Hypolipidemic activity of the fractions was further evaluated in vivo. The findings indicate that ethyl acetate extract of *Mentha piperita* leaves exhibited higher hypolipidemic effects than the two (2) extracts at a lower dose (100mg/kg body weight). All the fractions exhibited similar pattern of hypolipidemic activity, however fractions 2, 3, 4, 6 and 12, were more hypolipidemic than the other fractions, with Fraction 2 (F2) being the most potent. Overall, the findings suggest that ethyl acetate extract of *Mentha piperita* leaves and its fractions possess hypolipidemic properties and thus may potentially be explored in the management of hyperlipidemia and its related diseases.

Keywords: Hyperlipidaemia; *Mentha piperita* leaves; lipid profile; ethyl acetate extract; fractions

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Anti-Angiogenic Effect of Ethanolic Extract and its Phenolic Rich Fraction of *Filicium Decipiens*in the Chick Embryo Chorioallantoic Membrane Model

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Cancer has been reported to be the 4th most common causes of mortality in Malaysia, in year 2018. Although there are various cancer treatments available, side effects are always be the limitation of these treatments. Various medicinal plants have been studied extensively for their anti-angiogenic activity. Moreover, natural sources are safer and produce lesser side effects. This study aimed to search for alternative cancer treatment from medicinal plant, by examining anti-angiogenic activity of ethanolic extract and its phenolic rich fraction of *Filicium decipiens* (FD)in the chick embryo chorioallantoic membrane (CAM) assay.The plant extract was prepared by maceration in 70%ethanol andits fractions *(*hexane, ethyl acetate and aqueous) were prepared from dry ethanolic extract. Total phenolic content (TPC) of the fraction was assayed by using Folin-Ciocalteu method. CAM *in-ovo* method was used to evaluate the anti-angiogenic activity of ethanolic extract of FD bark (250 µg, 500 µg) and its phenolic rich fraction (50 µg, 100 µg). Prednisone (250 µg) was used as positive control. Qualitative observation of reduction in the thickness of blood vessels and quantitative analysis in the reduction of the number of total blood vessels and percentage of blood vessels inhibition were measured to determine the anti-angiogenic activity of the extract and fraction.Ethyl acetate fraction contained the highest total phenolic content (349.59 mg ± 0.29) than aqueous (123.17 mg ± 0.25), hexane (175.31mg ± 0.18) fractions. Ethanolic extract (250µg, 500µg) and ethyl acetate fraction (50µg, 100µg) showed significant reduction ($P<0.05$) in the total number of blood vessels (43, 14, 46 and 33) compared with negative control (62). Ethanolic extract (250µg and 500µg) and ethyl acetate fraction (50µg and 100µg) showed percentage of blood vessels inhibition of 30.6%, 76.4%, 25.8% and 46.5% respectively. Reduction in the thickness of blood vessels were observed in ethanolic extract (250µg and 500µg) and ethyl acetate fraction (50µg and 100µg).Ethanolic extract and ethyl acetate fraction of FD bark showed anti-angiogenic activity that may have chemotherapeutic potentials.

Keywords: Anti-angiogenic; *Filicium decipiens;* chick embryo chorioallantoic membrane

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Effect of *Spinacia Olerace* onMemory and Learning in Scopolamine induced Cognitive Decline Mice

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The present study was aimed to investigate thenootropic potentialaqueous extract of *Spinacia oleracea*(AESO) in scopolamine induced cognitive decline mice. Memory impairment was produced by administration of Scopolamine (1.4 mg/kg i.p) in albino mice.Nootropicactivity was evaluated following the oral administration of two different doses (200 and400 mg/kg)of aqueous extract of *Spinacia oleracea* Leaves and donepezil (5mg/kg) was used as standard drug. The Effect of extract on learning and memory of mice was evaluated using elevated rectangular maze, pole climbing, morris water maze test and neuroprotective effects were studied by estimating acetyl cholinesterase (AchE), malondialdehyde (MDA), superoxide dismutase (SOD), nitric oxide (NO), catalase (CAT) and glutathione (GSH) levels in the brains of mice.The significant ($P<0.01$)improvement were observed with 200 and400 mg/kg of AESOwhen compared with disease control and significantneuroprotective activitythrough decrease in AchE ($P<0.001$), MDA ($P<0.01$), NO ($P<0.05$) and significantly ($P<0.01$) increased levels of SOD, CAT and GSH with AESO (200 and 400 mg/kg) when compared with disease control.The study has shown that aqueous extract of *Spinacia oleracea* has nootropic potential and has capability of improving the memory of learning and ongoing tasks.

Keywords: *Spinacia oleracea;*nootropic; neuroprotective; scopolamine; donepezil

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Synergistic Antibacterial Potential of Ethanolic Extract of G.glabra with Antimicrobials: An Alternative Effective Multidrug Resistant Anti-Staphylococcal Therapy

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Yashtimadhu (Family: Fabaceae, *Glycyrrhiza glabra* L.) is a traditional ancient medicinal plant which accounts significant pharmacological properties cultivated worldwide.This study demonstrated the antibacterial synergistic, additive and antagonism activity of *G.glabra* ethanolic extract in synergistic combination with different broad spectrum antimicrobials.ATCC MSSA 25293 & MRSA 43300 were used as reference strains, HA-MRSA, VRSA and P.aeruginosa collected from clinical source. Modified paper disk diffusion procedure with 10, 20 and 40 µg/ml of ethanolic fraction with antibiotics combination defined synergism. After synergistic infusion of both compounds against MSSA, MRSA & VRSA recorded in two-time interval zone independently.Concentration dependent killing action of *G.glabra* with respect to time showed more synergistic action with meropenem, moxifloxacin, Polymyxin and Teichoplanin with highest level of integration with meropenem at 40 µg/ml against MSSA ATCC 25293. For MRSA ATCC 43300 showed more synergistic action with meropenem, Polymyxin and Teichoplanin.Concentrated ethanolic extract of licorice of 40 µg/ml elucidate significant co-adjuvant with macrolides with all three synergistic infusions for long duration against VRSA at both intervals.This study concluded that the highest

concentration of *G.glabra* in synergistic dilution of antimicrobials discloses profound significant synergism for 24 hours and 48 hours. This property signifies its therapeutic importance as alternative therapy for prolong duration against multidrug resistant Gram-positive staphylococcus species like Methicillin resistant and Vancomycin resistant enterococcus species.

Keywords: Antibacterial synergistic; *Glycyrrhiza glabra;* HA-MRSA, VRSA and *P. aeruginosa*; traditional ancient medicinal plant

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Evaluation of Antibacterial Activity against Multidrug Resistance (MDR) Bacteria by the Fractions of *Canarium patentinervium* Miq

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Rapid emergence of antimicrobial resistance has become an issue of concern, worldwide. This is due to indiscriminate increase in bacterial adaptation towards conventional antibiotics, which has resulted in the bacteria developing multidrug resistances (MDR). This has led to exploration of bioactive compounds from plants. *Canarium patentinervium* Miq belongs to the family of Burseraceae Kunth and genus *Canarium* L. This plant has been used traditionally in wound healing by indigenous people in Malaysia. This study was aimed to search for alternative antibiotic from medicinal plant and to provide ethnopharmacological evidence to its traditional use. The study aims to fractionate the ethanol extract of the barks of *Canarium patentinervium* Miq. by using three solvents (petroleum ether, chloroform and water) and investigate its antibacterial activity against MDR bacteria.Qualitative phytochemical analysis of the fractions of *Canarium patentinervium* Miq. was examined for the presence of chemical constituents. Antibacterial susceptibility test was evaluated by using disc diffusion method, minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) assay with positive control using 3 reference strains [methicillin sensitive *Staphylococcus aureus* (MSSA) ATCC 29213, *Escherichia coli* (*E.coli*) ATCC 35218, *Klebsiella Pneumoniae* (*K.Pneumoniae*) ATCC 13883] and 3 clinical isolates [methicillin resistant *Staphylococcus aureus* (MRSA) *K.Pneumoniae*, *Acinetobacter Baumannii* (*A.Baumannii*)].Petroleum ether fraction exhibited bactericidal activity against MRSA (MBC= 0.5 mg/ml, MBC/MIC ratio= 4) and *A.Baumannii* (MBC= 2.0 mg/ml, MBC/MIC ratio= 2). Water fraction displayed antibacterial activity against MRSA (MIC= 0.125 mg/ml) and *A.Baumannii* (MIC= 2.0 mg/ml) as compared to positive control respectively (vancomycin, MIC= 0.78 µg/ml and gentamycin, MIC= >25 µg/ml). The antibacterial activity of fractions of ethanol extract of bark of *Canarium patentinervium* Miq. supports the evidence of its traditional use and can be explored for bioactive compounds as antibiotic alternatives.

Keywords: antibacterial; *Canarium patentinervium* Miq.; MDR

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Evaluation of Antibacterial Activity against Multidrug Resistant (MDR) Bacteria by the Fractions of *Artabotrys suaveolens* (Blume)

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Rising of antibiotic resistance is threatening the global health care system andincreasing the worldwide death rate. Therefore, research and discovery of alternativeantimicrobial agents from plant sources is encouraged. The *Artabotrys suaveolens* (Blume) belongs to the Annonaceae family and mainly distributed in tropical and subtropical regions of the world. It was indigenously used to treat postnatal weakness and cholera infection.The study aims to provide evidence of the plant as an alternative source of antibacterial agent based for its folkloric use to treat infection. This study was undertaken to fractionate the chloroform extract of the stem of *Artabotrys suaveolens* (Blume) and to investigate the *in vitro* antimicrobial activities of different solvent fractions against three ATCC and MDR bacteria. Liquid-liquid fractionation was performed resulting with petroleum ether, chloroform and water fraction of the stem of *Artabotrys suaveolens* (Blume). Qualitative phytochemical analysis was conducted for alkaloid, cardiac glycoside, flavonoid, saponin, sterol and tannin. Antibacterial activity was ascertained by disc diffusion assay, minimal inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) against three ATCC strains (MSSA ATCC 29213, *K. pneumoniae* ATCC 13883 and *E. coli* ATCC 35218) and three clinical isolated strains (MRSA, *K. pneumoniae*, *A. baumannii*). GraphPad Prism 8 was used for data analyses. Differences are statistically significant when $p<0.05$. Qualitative phytochemical analysis revealed that both petroleum ether and chloroform fraction contained alkaloid, sterol, and tannin, while water fraction contained cardiac glycoside, saponin, and tannin. Petroleum ether fraction showed notable antibacterial activity against MSSA ATCC 29213 (inhibition zones=10.00±1 mm; MIC=0.5 mg/mL; MBC>2 mg/mL) compared to vancomycin (inhibition zones=10.67±0.58 mm; MIC=0.78 mg/mL; MBC=0.78 mg/mL). It also inhibited MRSA (inhibition zones=11.33±0.58 mm; MIC=0.25 mg/mL; MBC>1 mg/mL) compared to vancomycin (inhibition zones= 11.00±0 mm; MIC=0.78 mg/mL;MBC=0.78mg/mL), followed by chloroform and water fraction. All three fractions werebacteriostatic against MSSA ATCC 29213 and MRSA based on the results.The finding in this study has confirmed *Artabotrys suaveolens* (Blume) stem could be an alternative source of antibacterial agent and has provided evidence to the traditional use of *Artabotrys suaveolens* (Blume) in infection. Future studies on the isolation and characterization of bioactive compounds from the fractions are required to confirm their activity.

Keywords: antibacterial; *Artabotrys suaveolens* (Blume); MDR

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Utilization of Bacterial Isolates from Peatland and Herbivores Manures Samples as a Source of Anticancer Agents

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Indonesia International Institute for Life Sciences

In today's world, chemotherapy remains the standard treatment as anticancer agents. However, due to low specificity, progression of drug resistance, and undesirable side effects produced by

some chemotherapy, the discovery for novel anti-cancer drugs with fewer side effects with greater therapeutic efficiency is still a priority of cancer research. Indonesia is a country rich in biodiversity with a potential to discover natural anticancer agent through the production of a variation of chemical scaffolds on bacteria secondary metabolites. In this study, bacteria associated with peatlands and herbivores manures from Indonesian samples were selected and investigated for their potential to produce secondary metabolites with possible cytotoxic molecules. Four selected genus, Aneurinibacillus, Ochrobactrum, Curtobacterium, Dyella, were cultured and produced a total of five extracts that were tested for cytotoxic activity against HeLa and NIH-3T3 cell lines using MTT Assay. The result of MTT assay showed that Ochrobactrum extracts resulted in a decrease in cell viability percentage on HeLa cells while no decrease on cell viability was observed on NIH-3T3 cells. Aneurinibacillus extracts reduced the cell viability percentage on both cell lines. Curtobacterium and Dyella extracts increased the cell viability percentage of both cell lines, however these extracts demonstrated an observable dead cell under microscope on both cell line treated at higher extracts concentration. The study demonstrated that Ochrobactrum extracts produced the most selective anticancer agents, indicated by a decrease in cell viability percentage on HeLa cells while there are no decreases on NIH-3T3 cells cell viability. In conclusion, Ochrobactrum metabolites was found to be the most potential source of anticancer agents. Optimum metabolites production from Ochrobactrum should be investigated and the resulting metabolites can be used for further testing on different cancer cell lines.

Keywords: anticancer; bacteria; cytotoxic

IRCPAS/2020/PP-169

Normal Human Fibroblast (HFF-1) Cellular Uptake Studies of Human Growth Factor Loaded Chitosan Nanoparticles using Fluorescence Photomicrograph

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For the past decade, protein and peptide-based drugs have been utilised for therapeutic and clinical application because they are essential to treat chronic diseases including cancer. The protein and peptide-based drugs are mostly administered through parenteral route because they may undergo breakdown and absorption reduction in the digestive tract. However, administration by parenteral route may also cause various issues, for instance septic shock, thrombophlebitis and discomfort which may reduce patient's compliance. Thus, administration by mouth which is non-invasive is more beneficial, but several modifications need to be done to synthesised protein-based drugs that are more stable in the gastrointestinal condition. Palifermin or recombinant human keratinocyte growth factor (rHuKGF) is protein-based drug and it has been applied to treat common complications of chemotherapy in patients with tumour growth such as oral mucositis, ileum ulceration and colon inflammation. In this research project, the rHuKGF-loaded chitosan nanoparticles were used to study the fibroblast cells growth rate which the chitosan nanoparticles (CNPs) are being used as the oral drug delivery system. To determine the HFF-1 cellular proliferation rate, a (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) (MTT) assay was applied. Besides, to study the fibroblast cellular uptake of rHuKGF-loaded CNPs and CNPs alone, a fluorescence microscope was used. Both fibroblast cells and CNPs were dyed with fluorochromes before being observed under the fluorescence microscope. The fluorochromes used to stain fibroblast cells was (4',6-diamidino-2-phenylindole) DAPI and Rhodamine 6G was used to stain the CNPs. Key findings: rHuKGF-

loaded CNPs treatment showed a higher growth in fibroblast cellular proliferation than in treatment with rHuKGF alone. Furthermore, there were more uptakes of rHuKGF-loaded CNPs as compared to CNPs by fibroblast cells. rHuKGF-loaded CNPs are capable to increase the proliferation rate of fibroblast cells.

Keywords: Recombinant human keratinocyte growth factor; chitosan nanoparticles; fluorescence microscope; human gastric fibroblast; human foreskin fibroblast cells

IRCPAS/2020/PP-170

Malaysian Medicinal Plants as a Source of Alternative Medicines

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Medicinal plant has been used extensively in Malaysia since a long time ago as a medicine. Malaysia is classified as one of the world's 17 megadiverse countries with high endemism. In Malaysia, approximately 2,000 medicinal plant species are reported to possess medicinal values. Wide ranges of bioactive compounds that present in medicinal plant are able to cure many types of disease. The PRISMA (Preferred Reporting Items for Systematic Review and Meta-analyses) method was used. Published articles were identified through Science Direct, Scopus and Web of Science. In this review, several species of Malaysian medicinal plants including *Clinacanthus nutans, Ficus deltoidea, Stevia rebaudiana* and *Gynura procumbens.* The objective of this review was to compile the information regarding the bioactive compounds in the plant and its biological activities. The review on isolation of bioactive compounds including extraction technique and biological activities for each species were the main focus in this review. All of these medicinal plants are widely being use among local community in Malaysia. Hence, this review will help to add more knowledge on benefits of the *C. nutans, F. deltoidea, S. rebaudiana* and *G. procumbens.*

Keyword: Malaysian medicinal plant; bioactive compounds; biological activities

IRCPAS/2020/PP-172

Antioxidant and antiproliferative activities of 80% methanolic extract of *Cleome gynandra* leaves

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Cleome gynandra (cat whiskers) is a plant known as Maman and Langsana Merah in Malaysia and growing abundantly in tropical and subtropical regions. Beside it is used in traditional culinary, the plant also has traditionally been used for the treatment of headaches, stomach aches, rheumatoid and believed to facilitate childbirth. There are numerous pharmacological reports on *C. gynandra*including anti-inflammatory, radical scavenging, anticancerous, immunomodulatory and antidiabetics agent. The aim of this study was to investigate antioxidant and antiproliferation activities of 80% methanolic extract of *C. gynandra*leaves (CGE). The antioxidant activity of the extract was determined by 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging properties and total phenolics content (TPC)

assaywhile cell proliferation is measured with the colorimetric MTT assay. The results of present study showed that DPPH radical scavenging ability of CGE at 200 µg/ml was at 24.61 ± 2.04 %. Total phenolics content of CGE was 24.61 ± 2.04 mg GAE/g of extract. Cell viability study showed that IC_{50} values of the extract on HepG2 cells at 24, 48 and 72 hourswere 432.15, 331.35, and 256.54 µg/mL, respectively.These results provide evidence that CGE has antioxidant and antiproliferative properties and can be used potentially as ready accessible and valuable bioactive source for isolation of antioxidant and anticancer agents.

Keywords: *Cleome gynandra;* methanolic extract; antioxidant; antiproliferative; HepG2

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Evaluation of Phytochemicals and Antioxidant Activities of Crude and Fractionated Extracts of Selected Medicinal Plants

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The increasing of various metabolic disturbances are important issues in most countries. Metabolic complications and abnormalities increase the risk factors and promote the development of diabetes mellitus, atherogenic dyslipidaemia, stroke and cardiovascular disease, all-cause mortality. Recent studies have suggested that medicinal plants which rich in antioxidant compounds have been used to reduce the development of these complications. The objective of this study was to evaluate the antioxidant potential of *Clinacanthus nutans*, *Plantago asiatica*, *Platycladus orientalis* and *Tradescantia zebrina*.Four local plants were processed and macerated with absolute methanol. The solvent extracts were rotary evaporated, subjected to fractionation with different solvents in the following order: Water, hexane, chloroform, ethyl acetate and n-butanol. Folin-Ciocalteu and aluminium chloride methods were used to quantify the medicinal plants phenolic, flavonoid contents of crude extracts and fractions.The antioxidant activities of extract and fractions were evaluated through FRAP, DPPH and ABTS free radicals scavenging assays. Among the four medicinal plants tested, *Platycladus orientalis* demonstrated considerable high levels of phenolic, flavonoids and antioxidant activities across all extract fractions. The ethyl acetate fraction especially exhibited strongest and highest activities in all assays.In contrast, all *Tradescantia zebrina* extract/ fractions possessed low levels of antioxidant contents and activities as compared to other plants. In general, the phenolic content was positively correlated with flavonoid content ($r = 0.886$, $p < 0.001$) and FRAP ($r = 0.812$, $p < 0.001$). The study showed that the selected medicinal plants, particularly *Platycladus orientalis* possess high antioxidant potential and could be a potential natural source of antioxidant.

Keywords: Medicinal Plants;antioxidant;phenolic;flavonoids

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Antioxidant, Total Phenolic and Flavonoid content of *Mussaenda Erythrophylla* Schum. and Thonn. Stem and Leaf Ethanol Extract

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Mussaenda Erythrophylla Schum. & Thonn.(Rubiaceae)is is commonly known as Red Flag Bush, is an ever green shrub found distributed in Telangana State. The shrub is used traditionally to reduce inflammation, lower fever, to treat viral and bacterial infections. The present study is carried out to explore the total phenolic content, flavonoid content and antioxidant activity of leaf and stem *Mussaenda Erythrophylla*ethanol extract. In this study, the total phenolic content was assessed by using Folin and Ciocalteu reagent method. The Total flavonoid content was assessed with aluminium chloride method and anti-oxidant property was assessed with 2, 2-diphenyl-1-picryl hydrozyl (DPPH) assay and 2,2'-azino-bis3(3-ethylbenzothiazole-6-sulphonic acid) ABTS radical scavenger activity. The Gallic acid, Quercetin and Ascorbic acid were used as standards for phenolic, flavonoid and antioxidant activity respectively. The results of total flavonoid, phenolic content and antioxidant studies were found significant ($P< 0.05$) in leaf when compared to stem ethanol extract.

Keywords: *Mussaenda Erythrophylla Schum; Thonn.* (Rubiaceae); antioxidant; ethanol extract

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Isolation and assessment of novel phytomolecule (BS-3) for Muscle relaxant activity from *Galphimia glauca* Cav.stem methanol extract

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Galphimia glauca Cav is widely distributed in Deccan plateau regions of western and southern India. The *Galphimia glauca Cav.*is traditionally used in conditions of anxiety, phobia, fear, stress and it is as well used to produce a calming effect on the nerves. The current study has been opted to shed light on *G. glauca* stems to isolate, characterize, and explore the muscle relaxant activity using *in vivo* models.This study assesses the muscle relaxant activity of isolated phytomolecule (BS-3) from *G. glauca* stem methanol extract. The BS-3 was isolated from methanol fraction of *G. glauca* stem methanol extract by column chromatography followed by Preparative Thin Layer Chromatography studies. The BS-3 was characterized by Melting point, IR, ^{1}HNMR, ^{13} C NMR and Mass Spectroscopic studies and evaluated for muscle relaxant activity. The BS-3 was administered in Swiss albino mice for one day toassess muscle relaxant activity by Rota rod test and Grip strengthening test.The LD_{50} of the BS-3 was found to be > 2000 mg/kg. Mice treated with BS-3 at 12.5, 25 and 50 mg/kg doses showed significant ($P \leq 0.05$) effects on muscle relaxant activity in Rota rod test and Grip strengthening test in mice ($P \leq 0.05$). The study results conclude that the BS-3has significant muscle relaxant effects.

Keywords: Mice; Rota rod test; grip strengthening test; *Galphimia glauca; c*olumn chromatography

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Aseptic meningitis due to Echovirus 30 (E30) at Aminu Kano Teaching Hospital (AKTH), Kano, Nigeria

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Human echovirus type 30 (E30) is one of the most commonly isolated members of the enterovirus serotypes in multiple aseptic meningitis (AM) outbreaks in adult and children. The highest incidence of meningitis occurs during the neonatal period and infancy.In this study, the role of E30 in neonates AM at Aminu Kano teaching Hospital (AKTH) was investigated in a three-year period from 2017 to 2019. One hundred and ten samples of Cerebrospinal fluid (CSF) were collected from neonates (0-28 days) admitted with symptoms of meningitis at Special care baby unit (SCBU) of AKTH. White blood cells (WBC), glucose and Protein concentrations of the CSF samples were determined. Wellcogen™ Bacterial Antigen kit was used to detect the bacteria present in the samples. Viral RNA was extracted using Life River™ RNA isolation kit. Reverse transcription Real-time Polymerase chain reaction qRT-PCR using Human Echovirus 30 Real- Time RT-PCR kit was used to detect the presence of E30 in the samples. Of the 110 CSF samples collected 54.5% (60/110) were males and 45.5% (50/110) were females, 97.3 %(107/110) of the patients were term neonates'. Glucose and Protein concentration of most of the samples were within normal range. Pleocytosis(neonates≥ 20 white blood count (WBC)/mm3 was seen in 72.7 % (80/110) of the samples. Gram negative and gram-positive bacteria were detected in 4.5% (5/110) of the samples. E30 was detected in 0.9% (1/110) of the neonate patients' negative of bacterial meningitis.The result of our study shows a single case of AM due to E30 that appeared in a 24-hour old female term neonate in the third year of the study. Prevalence of the virus is lower compared to outbreak of the virus reported in China, Korea, Kuwait, Brazil, United States, and European countries. In Africa, a study in Mossel Bay, South Africa reported cases of E30 and other enteroviruses in AM in young children. To the best of our knowledge, our study reports the first case of E30 AM in neonate in Kano State, Nigeria.Early etiologic diagnosis of AM helps to avoid unnecessary antibiotic treatment and additional testing. It is essential to establish an enterovirus molecular surveillance system in the country to prevent mass outbreak of the deadly virus in Nigeria.

Keywords: Echovirus; enterovirus; aseptic meningitis; real-time RT-PCR

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Determination of Antioxidant Activity from Dayak Onion(*Eleutherine bulbosa* Merr) Based on Drying Time

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Dayak onion (*Eleutherine bulbosa* Merr) is a plant that believe to be native to the island of Borneo, especially to Kalimantan, Sarawak, and Sabah. This plant used as folk medicine in Dayak community for heal several types of diseases. This usually used in simplisia (dried) form that brewd with hot water and consume as an antioxidant drink. The process of drying to make simplisia (dried) form of dayak onion may affect the antioxidant activity, because antioxidant is sensitive to light and heat. The objective of this research was to determine the effect of drying time to the antioxidant activity. Sample used on this research was bulb of dayak onion. The bulbs are cut into a small and thin part and dried using oven for 24, 25 and 26 hours at 50^{o}C. Simplisia (dried) form extracted with maceration method with ethanol solvent. Antioxidant activity was test using DPPH method (2,2-diphenyl-1-picrylhydrazyl) and measured with uv-visible spectrophotometer. The result obtained that IC_{50} from three ethanol extract of simplisia (dried) form of Dayak Onion (dried for 24, 25 and 26 hours) are 812.38 ppm, 844.94 ppm and 862.63 ppm. From the result of this study showed that drying time influence antioxidant activity. Short time of drying time had a high antioxidant activity, otherwise long time of drying time had lower of antioxidant activity. In conclusion is antioxidant activity of simplisia (dried) form of dayak onion decrease with increasing drying time.

Keywords: *Eleutherine bulbosa* Merr; dayak onion; antioxidant activity; drying time

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Cytotoxicity, Antioxidant and Antimicrobial Activity of The Leave and Stem Bark Extracts of *Vitellaria Paradoxa*

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The effective use of *Vitellariaparadoxa* in traditional medicine for treatment of various ailments including inflammation, fever, skin irritation, dermatitis, sunburn, rheumatism, diarrhea,stomachache, and ulcers are well established. This study focused on the cytotoxicity of the plant parts using Brine Shrimp Lethality Assay (BSLA), antioxidant activity using 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay and antimicrobial activity against *Bacillus cereus, Staphylococcus aureus*, *Kleibsiella pneumonia* and *Escherichia coli* using disc diffusion and minimum inhibitory concentration methods. Result of cytotoxicity showed that the leaf crude extract (VPL 01) is more cytotoxic with LC_{50} 15.17 µg/mL followed by the methanol fraction of both the leaves (VPL 04) and the stem bark (VPS 04) with LC_{50} values 23.21 and 19.35 µg/mL respectively. The result of the antioxidant assay obeys the Beer-Lambert law over the useful range with both the leave and stem back of *Vitellariaparadoxa* showing great antioxidant potency in all fractions tested. The methanolic fraction standout above other factions with IC_{50} of 9.64 µg/mL and 6.50 µg/mL for the leave and stem bark

fractions respectively followed by the moderately polar ethyl acetate fraction which recorded LC_{50}16.96 µg/mL and 16.38 µg/mL for leave and stem back in comparison with standard ascorbic acid and BHT which recorded 4.77 µg/mL and 9.18 µg/mL respectively. The antimicrobial activity showed that the ethanol leaf extract had the maximum zone of inhibition against the entire test organisms, while the methanol fraction showed maximum inhibition against *B. cereus* (18±0.73 mm), *S. aureus* (19±1.41 mm) and *K. pneumonia* (19±0.03 mm) with MIC values of 62.5µg/mL for all the organisms respectively. These studies indicated that bioactive molecules present in *Vitellariaparadoxa*can be used as a prototype for development of new drugs as pharmaceutical raw materials.

Keywords: *Vitellariaparadoxa*; cytotoxicity; antioxidant; antimicrobial

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Trypanosuppressive Effects of Kolaviron may be Associated with Down Regulation of Trypanothione Reductase in *Trypanosoma Congolense* Infection

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Trypanothione Reductase is a key enzyme in maintaining the redox balance in hemoflagellate protozoan parasites like *T. congolense*. This study aims at unraveling the potency of Kolaviron against trypanothione reductase in *T. congolense* infection using Chrysin as standard. The experiment was performed using three different approaches,*in silico*, *in vitro* and *in vivo* studies. Kolaviron and Chrysin were docked against trypanothione reductase, revealing binding energies (-9.3 and -9.0 kcal/mol) and K_i of 0.211µM and 0.151µM at the active site of trypanothione reductase due to strong hydrophobic/hydrogen bond interactions. Parasitized blood was used for parasite isolation as well as assaying trypanothione activity following a standard protocol. Real-time PCR (qPCR) assay was implored to monitor expression of trypanothione reductase using primers targeting the 177-bp repeat satellite DNA in *T. congolense* with SYBR Green to monitor product accumulation. Kolaviron showed IC_{50} valuesof2.64µg/mlwith % inhibition of 66.78 compared with Chrysin with IC_{50} valuesof1.86µg/ml and % inhibition of 53.80. *In vivo* studies following the administration of these compounds orally after 7 days post inoculation resulted in % inhibition of Chrysin (57.67) and Kolaviron (46.90). Equally, Kolaviron relative to Chrysin was able down regulated the expression trypanothione reductase gene by 1.352 as compared to 3.530 of infected group, in clear agreement with the earlier inhibition observed at the fine type level. Overall, the findings may have unravelled the potency of how Kolaviron against *Trypanosomacongolense*infection.

Keywords: Trypanothione reductase; Kolaviron; Chrysin; inhibition; expression

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In-silico Screening of Selected Flavanone Compounds for HMG Co-A Reductase Inhibitory Activity

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Hypercholesterolemia is one of the potential modifiable risk factors for cardiovascular diseases, the main leading causes of death globally. To keep serum levels of total cholesterol and LDL within the normal limit, statins (HMG CoA reductase inhibitors) are widely prescribed. Statins are generally well tolerated, yet recent studies reported that the use of statins could lead to adverse effects such as elevated hepatic transaminases level, myalgia, and increased risk of diabetes. These adverse effects could reduce patient compliance and results in poor therapeutic outcomes. Various flavanones are shown to possess anti-hypercholesterolemia effect *in vitro*, *in silico* and *in vivo*. The objectives of this study were to estimate the binding energies of the selected flavanone compounds against HMG CoA reductase through *in-silico* screening and to determine the structural activity relationship (SAR) of the selected flavanone compounds *in silico.* The selected flavanones are eriocitrin, eriodictyol, hesperitin, hesperidin, neohesperidin, naringin, naringenin and narirutin. Atorvastatin was used as a positive control to validate the binding and to compare the binding energies of the selected flavanones. The structure of the human HMG CoA reductase (PDB ID: 1DQA) was downloaded from Protein Data Bank, whereas the structures of the flavanones were downloaded from ZINC database. All the compounds were prepared using AutoDock Tools 1.5.6. Then, they were docked against the human HMG CoA reductase using AutoDock Vina 1.1.2 and Accelrys Discovery Studio 4.5. The interactions between flavanones and the protein were analyzed and their structure-activityrelationships were also determined. Results: The binding energy of atorvastatin as a control was -8.0 kcal/mol. Eriocitrin (-10.0 kcal/mol), hesperidin (-9.7 kcal/mol), neohesperidin (-9.5 kcal/mol),narirutin (-9.5 kcal/mol) and naringin (-9.1 kcal/mol) exhibited greater binding affinity towardsHMG CoA reductase, as compared to atorvastatin. They are flavanone glycosides. The aglyconeflavanone compounds, eriodictyol (-7.4 kcal/mol), hesperitin (-7.6 kcal/mol) and naringenin (-7.4kcal/mol) exhibited lower binding energy than atorvastatin. However, they have total polarsurface area (TPSA) lower than 140 Å2 and do not violate the Lipinski's Rule of Five. Discussionand Conclusion: There are studies which showed that the aglycone flavanones (eriodictyol,hesperitin, and naringenin) possess HMG Co-A reductase inhibitory activity. With theircomparable binding energies shown in this study, the eriodictyol, hesperitin and naringenin aresuitable to be used for future drug development against dyslipidemia.

Keywords: Cardiovascular diseases; hypercholesterolemia; in-silico docking; flavanones; atorvastatin; HMGCo-A reductase; HMG Co-A reductase inhibitory activity; binding energy

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Anti-angiogenic effect of ethanolic extract and its phenolic rich fraction of *Acacia auriculiformis* bark in the chick embryo chorioallantoic membrane model

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*Acacia auriculiformis*plant is widely used in traditional medicines for treatment of various diseases. The main objective of this study is to evaluate the anti-angiogenic effect of ethanolic extract and its phenolic rich fraction of *A. auriculiformis* bark in the chick embryo

chorioallantoic membrane model. Dried powdered bark of *A. auriculiformis* was extracted with 70% ethanol and the resultant was partitioned with hexane, ethyl acetate and aqueous.Folin-Ciocalteu assay was used to quantify the phenolic content in fractions of *A. auriculiformis* bark. The anti-angiogenic effect of ethanolic extract and phenolic rich fraction were evaluated by using in-ovo chorioallantoic membrane (CAM) model. The reduction in total blood vessels number in the CAM model was considered as positive indicator of anti-angiogenic effect.Ethyl acetate fraction showed the highest phenolic content which was 621±16.20 mg of gallic acid equivalent per gram of fraction. CAM treated with ethanolic extract (250 µg, 500 µg), ethyl acetate faction (10 µg & 50 µg) and prednisone (250 µg) showed significant reduction ($p<0.05$) in total blood vessel (TBV) 46.4±0.89, 36.4±2.30, 47.6±3.05, 37.6±1.82 & 37.0±2.00 compared with negative control group (61.7±2.52). The anti-angiogenic effect shown by the ethanolic extract and ethyl acetate fraction of *A. auriculiformis* might be due to the presence of phenolic compound. *A. auriculiformis* can be a new source of anti-angiogenic agent in anticancer therapy.

Keywords: Anti-angiogenesis; *A. auriculiformis*; ethanolic extract; phenolic rich fraction; CAM assay

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Effects of Dasatinib with Resveratrol in Toxin-Based Cell Models of Neurodegeneration

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Neurodegenerative disorders like Parkinson's disease are characterized by dysfunction of motor control and cognition due to the gradual loss of dopamine-generating brain cells. Recent research linked kinase activities, its phosphorylation pathways and α-synuclein with mitochondrial dysfunction and neuroinflammation to neurodegeneration. Current treatments provide only symptomatic relief. In the present study, we have demonstrated that a combination of dasatinib and resveratrol has potential to treat neurodegeneration more efficiently. SH-SY5Y (α-synuclein proteinopathy and mitochondrial dysfunction): to study α-synuclein level, autophagy; mitochondrial dysfunction parameters: ROS, complex-1, calcium, PGC-1α activity; dopamine level; beclin1, parkin, SIRT-1 and IMR-32 (neuroinflammation) to study NFkB, TNF α, IL1B gene expression and protein levels; cell line models were used in the study. The drug combination significantly attenuates α-synuclein level, shows 38% increase ($P<0.001$) in mean autophagy intensity (11.53 0.06) compared to rotenone group and an increment of 11% and 19% ($P<0.001$) compared to individual dasatinib and resveratrol treatment groups respectively. ROS fluorescence intensity for the drug combination shows a reduction of 52% ($P< 0.001$) compared to rotenone group and a reduction of 12% ($P<0.01$) and 25% ($P<0.001$) compared to dasatinib and resveratrol groups, respectively. mRNA expression and protein levels for neuroinflammation show reduction in the levels of IL1B, NF B, TNF ($P<0.001$) for the drug combination compared to LPS group. The mechanism of action of the drug combination may possibly due its activity in multiple targets such as α-synucleinopathy, mitochondrial dysfunction and neuroinflammation. The drug combination has, therefore, a better efficacy to treat neurodegeneration.

Keywords: Dasatinib; Resveratrol; neurodegeneration

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Evaluation of the pharmacopoeial quality of oral drug preparations in Katsina State, Nigeria

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Despite the recommended methods of current good manufacturing practices (CGMP) and the efforts of the Government's regulations, there is still suspicion on the circulation of pharmaceutical products with questionable quality primarily in the developing countries. The circulation and use of substandard drugs pose a serious health risk to the patient, therapeutic failure of infectious disease, and the development and spread of antimicrobial resistance. Accordingly, a study on the pharmacopoeial quality was undertaken on oral drug preparations commonly prescribed in public hospitals in Katsina State, Nigeria. A total of 400 oral drug samples were evaluated for microbial and chemical quality using standard procedures as described in the official monograph of the British and United States pharmacopoeia. Microbial limit tests (MLT) was carried out on the sampled pharmaceutical products. Isolated microbial contaminants were identified via conventional microbiological protocols, PCR and 16S rRNAgene sequencing. On the other hand, the chemical quality was evaluated by assessing the presence and the percentage content of the stated active pharmaceutical ingredients using validated HPLC assay and titration methods. The results of the assessment of the microbial quality of the analyzed oral drug samples revealed that 174 comprising 43.5% of the samples had microbial contamination; of these, 82 (20.5%) failed to meet the acceptance criteria set for microbial quality of non-sterile oral dosage forms. The isolated microbial contaminants comprised of 175 (76.8%) bacterial isolates, and 53 (23.8%) fungal isolates. Phylogenetic analysis of the 16S rRNA gene sequences placed the isolates within the genus *Bacillus, Enterobacter,* and *Pseudomonads* and has shown a sequence similarity that ranged from 96 – 100% with other sequences of related bacteria strains from the NCBI data base. Similarly, the result of the chemical quality showed that 37.1% had active ingredient outside the set pharmacopoeial limit and therefore were none compliant to the BP, USP and IP specifications for percentage content. It is recommended that manufacturers of pharmaceutical products should strictly adhere to the current good manufacturing practices (CGMP) at any stage of production, as these may greatly affect the microbiologic and chemical quality of the finished pharmaceutical products.

Keywords: Oral drug formulations; quality; British and United States pharmacopoeia; microbial contaminants; active pharmaceutical ingredients

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16S rRNA Gene Profiling of *Pseudomonas Sp.* Recovered from Contaminated Non-sterile Pharmaceutical Products

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Microbial contamination of non-sterile pharmaceutical products is a serious problem because it can result in the spoilage of the products or pose a serious health risk to the patient. Identification of microbial contaminants is pertinent with the view to help prevent putting patients at risk due to inadequate safety, to track contamination sources and to proffer suitable suggestions that ensure the manufacture and distribution of safe and good quality medicines to the patients. The aim of our study was molecular identification of *Pseudomonas sp.* isolated from non-sterile pharmaceutical formulations. *Pseudomonads* were isolated from non-sterile pharmaceutical liquid formulations on Trypticase soy agar and Cetrimide agar. DNA was extracted from pure cultures using the Boiled lysis method. Amplification of the 16S rRNA gene was carried out using the universal primers 27F and 1492R, and then sequencing of the amplified PCR products was carried out using the Sanger sequencing method. Sequences were aligned by multiple sequence alignment technique using CLUSTAL W and a phylogenetic tree constructed by the neighbor-joining method using MEGA X. Phylogenetic analysis of the 16S rRNA gene sequences of isolates showed a sequence similarity that ranged from 99.85 to 100% with other sequences of *Pseudomonas aeruginosa* from the NCBI data base. On the basis of the phylogenetic analysis and the sequence data submitted to the GenBank, the isolates represent five novel species of the genus *Pseudomonas*, for which the names *Pseudomonas aeruginosa MMM070, MMM221, MMM232, MMM219, MMM234 strain*s with accession numbers MN620435, MN620436, MN620437, MN620438 and MN620439 was assigned respectively. It can be concluded that identification of microbial contaminants based on partial sequencing of the 16*S*rRNA geneis a reliable, accurate and appropriate method which could be used in the quality control of pharmaceutical products.

Keywords: Non-sterile pharmaceutical products; contamination; phylogenetic analysis; *Pseudomonas aeruginosa*

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Evaluation of Ethanolic Extract of *Elettaria cardamomum* Seed for Wound Healing and Analgesic Activity in Sprague Dawley Rats and Albino Mice

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Cardamomum which is known as *Elettaria cardamomum,* has been widely utilizing for thousands of years for various ailments and cooking purpose. This present study was aimed to evaluate the effect *Elettaria cardamomum* seeds on wound healing in Sprague Dawley rats using Excision Wound Model and analgesic activity in Albino mice using Tail Immersion Method. Extracts prepared by cold maceration method. The preliminary phytochemical screening of extract shows the presence of alkaloids, proteins, phenolic compounds, flavonoids, volatile oils and terpenoids. For Excision Wound Model, animals were divided in four groups of six rats each. Group I served as negative control, treated with simple ointment, group II

treated with standard drug, Povidone iodine 10% w/w, group III treated with low dose (5%, w/w) extract and group IV were treated with high dose (10%, w/w) ofextract. All the treatments were done topically and were given once daily. The wound healing effect was observed on 5th, 10th and 15th day. Furthermore, for Tail Immersion Method, mice were divided in four groups with six each. Group I served as normal control, treated with normal saline, group II as standard, and treated with Tramadol (20mg/kg), group III mice treated with ethanolic extract (200mg/kg) as low dose and group IV were treated with extract of (400mg/kg) as high dose. All the extracts and standard drug were given orally, and tail flick response time recorded for 30, 60, 90 and 120 minutes. The highly significant (***P<0.001) *E.cardamomum* ointment was observed in both 5%w/w and 10%w/w on 15th day when compared with negative control. Both 5%w/w and 10% *E.cardamomum* ointment revealed the effectiveness of improved wound healing. Besides that, for analgesic activity, ethanolic extract of *E.cardamom(400*mg/kg), high dose was highly significant (***P<0.001) whereby low dose (200mg/kg) extract showed less significant (*P<0.05) at 120 minutes. This study showed that ethanolic extract of *Elettaria cardamomum* seeds possess wound healing properties and has potential to treat pain, which may be due to presence of alkaloids, proteins, phenolic compounds, flavonoids, volatile oils and terpenoids.

Keywords: *Elettaria cardamomum*; wound healing; analgesic; ethanolic seeds extract; tail immersion method; excision wound model.

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Cytotoxic Effect of Vitexin Compoundon Aβ - Induced BV2 Cells

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Neuroinflammation is a series of neuropathological processes as the result from the interaction with the microenvironmental cues (e.g. lipopolysaccharide (LPS) and amyloid-β (Aβ)) present at the central nervous system (CNS). The neuroinflammation will become chronic when the inflammation persists which lead to the neuronal damage and eventually neurodegeneration development. Thus, the present study aimed to determine the cytotoxic effect of vitexin compound on BV2 cells and Aβ-induced BV2 cells. Cell viability assay was conducted to investigate the toxicity effect of the compound towards both cell cultures. Six concentrations of vitexin were prepared and tested. MTT assay (3-(4, 5-Dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide) was performed to determine the cytotoxicity of the compound. The results from both cultures were expressed as Means ± S.D where $p<0.05$ is considered as significant. The result from this study showed that the vitexin compound possesses cytotoxicity effect in dose-dependent manner upon exposed to both cultures. The IC_{50} from both cell cultures was 50µM. The result suggests that the vitexin compound is safe to be used in both conditions. This preliminary finding allows the future research to be conducted on vitexin in search for the potential treatment for neurodegenerative diseases.

Keywords: Cytotoxicity; Vitexin; Neuroinflammation; Amyloid-β; BV2 cells

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Ficus carica Polyphenolic-rich Extract Facilitate Glucose Uptake, Augment Adiponectin Secretion, and Inhibits Alpha-glucosidase Activity *in vitro*

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To date developing antidiabetic agent devoid of adverse effect remains a challenge to the health care system. *Ficus carica* was known as food and as alternative for the treatment of several ailments for decades. The antidiabetic and antioxidant potentials of *F. carica* leaf were evaluated in vitro using suitable models. Initial comparative study to select the best solvent and technique for the extraction of polyphenolic-rich extract from *F. carica* leaf was conducted. The leaf was extracted using ultrasound assisted bioguided (with hexane, ethyl-acetate, chloroform, methanol and water as solvents) and subcritical water extraction techniques. Subcritical water extraction technique revealed a significantly higher ($P<0.05$) percentage yield (47%) which was twofold more than methanol extract (22%) being the highest in the ultrasound assisted solvent extraction. Total phenolic content varied insignificantly ($P>0.05$) among all extracts except for chloroform extract (97.95±10.56 mg GAE/g extract) that was significantly ($P<0.05$) lower than aqueous extract (160.55±23.05 mg GAE/g extract). However, *F. carica* methanolic leaf extract (FCMLE) revealed the highest total flavonoid content (889.84±7.70 mg RE/g extract) corresponding with its higher ABTS and DPPH radicals scavenging activities, consequently this extract was selected for further in vitro antidiabetic assay. FCMLE showed a dose-dependent increase in adiponectin secretion and 2-NBDglucose uptake by 3T3-L1 cells at basal and upon insulin stimulation and concomitantly antagonize apigenin inhibition of GLUT-1 2NBDG uptake. Similarly, FCMLE showed a dose dependent inhibition of α-glucosidase activity. This study suggests FCMLE as therapeutic candidate in the management of hyperglycemia and hyperglycemia induced oxidative stress.

Keywords: *Ficus carica*; antioxidant; antidiabetic; 2NBDglucose uptake; adiponectinsecretion

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A Systemic Review: Benefits and Harms Using Fondaparinux as Prophylaxis Antithrombotic Therapy in Cesarean Section Recovery Patients

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This study reviews on benefits and harms of using fondaparinux as prophylaxis antithrombotic therapy for the caesarean section recovery patient. Pregnancy is a hypercoagulable state as it develops a mechanism that protects the pregnant mother against excessive bleeding during childbirth. Thus, to prevent that, fondaparinux is used as an antithrombotic prophylaxis over the other low-molecular weight heparin (LMWH). Fondaparinux is efficacious if compared to LMWH or UFH to prevent VTE by treating the deep thrombosis (DVT) and pulmonary

embolism (PE) without any risk of major bleeding. The objective is to calculate the odd ratio of benefits and harms of fondaparinux as a prophylaxis antithrombotic therapy in caesarean section recovery patients by using the Mantel-Haenszel method. This study was conducted based on randomized controlled trials and observational studies. The related articles were searched by using appropriate search engine such as Cochrane Library, Elsevier, PubMed, and Google Scholar. The articles were then selected based on the use of inclusion and exclusion criteria. The types of tools that were used to make the fulfilment of the study are, the appraisal forms like the PRISMA 2009 checklist and flowchart, SPSS to do the calculation on statistics and Microsoft Excel that was formulated. The summary of the forest plot includes all the 11 respective individual studies with OR = 0.74 (95% CI, 0.28 to 2.00). Results of forest plot show that most of the studies shows that fondaparinux has benefits when used as antithrombotic prophylaxis therapy for caesarean section recovery patients. The summary odd ratio shows a positive odd ratio that indicates that fondaparinux is causing more benefits than harms. This review concluded that benefits and harms of using fondaparinux as prophylaxis antithrombotic therapy in caesarean section recovery patients are based on the clinical trials conducted. Fondaparinux can prevent the HIT and hypersensitivity skin reactions that are caused by other LMWH and UFH. Fondaparinux also reduces the occurrence excessive haemorrhage risk in caesarean section recovery patients.

Keywords: Fondaparinux; thromboprophylaxis; cesarean section; venous thromboembolism; pulmonary embolism; deep vein thrombosis

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A Systematic Review: Statin Induced Pain Among Adults

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Statin is among the most extensively prescribed therapies in Malaysia. According to the National Health and Morbidity Surveys (NHMS) the prevalence of the common cardiovascular (CV) risk factors among adults ≥ 18 years had been on an increasing trend. The prevalence of hypercholesterolemia had risen by 46% over the 4 years, 2011 – 2015. Almost 1 in 5 adults in the 18-19-year age group had hypercholesterolemia.The prevalence increased with age, from 22.0% in the 18-19-year age group, reaching a peak of 68.8% among adults aged 55-59 years. Objective was to calculate the odd ratio of statin induce pain by using the Mantel-Haenszel method and determine the summary odd-ratio of studies that evaluate statin induce pain.The results from search engine used Pubmed, Google scholar and Ebscohost are tabulated. Boolean operators were used for appropriate relevant articles. From the search, a total of 31 articles collected from Pubmed, 122 from Google scholar and 15 from Ebscohost were identified. Then, 85 articles were collected after discarding duplicate papers and all the 85 papers are screened. 28 papers were excluded as it does not match the inclusion and exclusion criteria. Respective 40 papers on statin therapy causing pain as adverse effects were collected where 28 papers are randomized controlled trials papers whereas the other 12 papers are incidence papers reporting pain and were excluded due to no odd ratio calculation found in those papers. The outcome is achieved by calculating the odd ratio for each individual study. Forest plot include all the 28 respective individual studies shows OR= 0.08 (95% Cl, 0.04 to 0.12) meta-analysis proved to be positive where statin therapy induces muscle pain. The results showed that statin

can induce pain, but large variations could be the reasons for the summary odds ratio to be rather close to the line of no difference.

Keywords: Statin; pain; hypercholesterolemia;Mantel-Haenszel

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Anti-Cancer Activity of*Hiptage Benghalensis* Bio-Active Fractions Against Cancer Cell Lines

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The objective of the study was to study the cytotoxic effects of effects of different fractions of *Hiptage benghalensis* using various human cancer cell cultures, in vitro by MTT assay, reactive oxygen species (ROS) generation and caspase-3 activities. Human cervical carcinoma (HeLa) cells, human breast cancer (MCF-7) cells and human neuroblastoma (IMR-32) cells were maintained in a 5% CO2 incubator at 37°C. Different concentrations of fractions of *H benghalensis*such as toluene fraction (HT), ethyl acetate fraction (HE), butanone fraction (HB), and aqueous fraction (HAq) in serum-free culture medium were freshly prepared and used for cytotoxic activity by MTT assay, ROS generation and apoptotic effect by caspase-3 activity. Among the four fractions, the HE and HB fractions have revealed that greater percentage inhibition in all types of cancer cells in a dose-dependent manner by MTT assay. The IC50 values of HE fraction were found to be 42.73, 44.61 and 46.94 µg/mL against HeLa, MCF-7, and IMR-32, respectively. The apoptotic activity was evaluated through ROS generation and caspase-3 activities of HE and HAq. The results showed that both fractions have significantly increased the ROS production and caspase-3 levels in all the cell cultures in a dose-dependent manner. The present investigation has shown that the HE and HAq fractions of *H benghalensis* displayed significant cytotoxic activity against all three cancer cells by decreased cell viability, increased generation of ROS and caspase-3 activities.

Keywords: Caspase-3; cell viability;*Hiptage benghalensis*; reactive oxygen species;reactive species

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Determination of Total Phenol Content from Ethanolic Extract of Temu Giring (*Curcuma heyneana*) Using Spectrophotometer

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Temu giring (*Curcuma heyneana*) is a plant from Zingiberaceae and case this plant used traditionally as herbal medicine. Scientific research toward *C. heyneana* remains limited, including determination of total phenol. Phenol content is related to the antioxidant activity which is believed responsible to cure many diseases. The aim of this research was to determine total phenolic contents in ethanolic extract of *C. heyneana* rhizome. The rhizomes of *C. heyneana* were collected from Medan, North Sumatera, Indonesia. The rhizomes were mashed

up then extracted with maceration method. The extract obtained then screened the secondary metabolites and subsequently tested the total phenol using folin-ciocalteau method with gallic acid calibration. Quercetin was used as comparative substances. The maceration process got the result of about 5.6% from *C. heyneana* dry. The phytochemistry screening revealed the extract contains flavonoid, saponin, tannin as well as steroid. The maximum wavelength of gallic acid was 776 nm. The regression equation from calibration of gallic acid was y = 0,0077x + 0,0294 with the correlation coefficient (R2) was 0,994. Ethanol extract of *C. heyneana* has total phenol as 400,37 mg/g gallic acid equivalent, while quercetin was 851,04 mg/g gallic acid equivalent. Total phenol of *C. heyneana* was half compared to the quercetin. Quercetin was the pure compound of phenol while *C. heyneana* contains not only phenol but also various compounds. This research showed that ethanol extract of *C. heyneana* has high total phenol and possibly having antioxidant activity. This extract potential to be developed further as herbal medicine.

Keywords: Temu giring; Curcuma heyneana; total phenol; Folin-ciocalteatu; Gallic acid

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Comparison of Ginger and Papain Assays for Heavy Metals Detection

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Through anthropogenic and industrialization activities, great amounts of trace elements and heavy metals have been excavated and released into the water bodies and subsequently dissipated into the environments. Rapid screening technology for detecting major and trace elements as well as heavy metals in variety of environmental samples is most desired. The need for simple and rapid means of detecting and monitoring these elements and detecting them in real time becomes necessary. In this study, a comparison of heavy-metal assays using ginger and papain proteases is reported. The proteolytic enzymes are assayed using casein as a substrate with Coomassie dye to track the completion of hydrolysis of casein. In the absence of inhibitors, casein is hydrolysed to completion, and the solution is brown. It was found that ginger protease assay requires more casein substrate and longer assay time compared to papain. Ginger proteolytic activity was inhibitedin the presence of metal ions such as Ag^{+}, Hg^{2+} and Cu^{2+}while in the case of papain proteolytic activity was inhibited by Pb^{2+}, Zn^{2+}, Ag^{+} and Hg^{2+}. In both cases, the hydrolysis of casein is inhibited, and the solution remains blue. For papain, the IC_{50} (concentration of toxicant giving 50% inhibition) for $Hg^{2+,}$ $Ag^{2+,}$ Pb^{2+}, Zn^{2+} were 0.39, 0.40, 2.16, 2.11 mg/L, respectively, while that of ginger for Ag^{+}, Hg^{2+} and Cu^{2+} were 0.0194, 0.1980 and 0.2474 mg/L, respectively.. The IC_{50} values of ginger protease assays to heavy metals indicated that it is a more promising proteolytic assay for heavy metals compared to papain. The IC_{50} values for these heavy metals are comparable to several other assays such as coriander, garlic and tomato assays, immobilized urease, 15-min Microtox™, and rainbow trout assays. The potential of this inhibitive assay for monitoring heavy metals in the environment is demonstrated.

Keywords: Coomassie; papain; ginger protease; assay

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Molecular Docking of Papaya Bioactives against Keap1, the Inhibitor of Nrf-2

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Prevention of oxidative stress is accomplished through the Nrf-2 pathway due to its role in regulating multiple biological processes that trigger antioxidative responses. Keap1 binds to Nrf-2 leading the latter to proteasomal-mediated degradation. Hence, inhibition of Keap1 will stabilize Nrf-2 and increases its antioxidative potential. The antioxidants present in the active fraction of papaya leaves obtained in previous study were hexadecanoic acid, linoleic acid, linolenic acid, phytol, α-tocopherol, γ-tocopherol, campesterol and stigmasterol. The objective of this study was to evaluate the binding efficacies of these molecules against Keap1 by virtual screening using Glide and AutoDock molecular docking software. The triterpenoid, gedunin, was used as a positive control. Based on the binding scores from both software, the top four inhibitors were campesterol, stigmasterol, α-tocopherol and γ-tocopherol. All four compounds also fulfilled at least three of Lipinski's rules indicating that they could possess drug-like effects. In view of that, these phytocompounds were possible inhibitors of Keap1 and should be further experimented to validate their inhibitory efficacies.

Keywords: phytochemicals; keap1; nrf2; antioxidant; inhibitor; molecular docking

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Development of solid-phase analytical derivatization for the analysis of drugs in human urine

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Drug analysis of biological samples has been carried out for various purposes, such as identifying a cause of poisoning at a hospital and proving illegal drugs ingestion at a forensic laboratory. Urine is one of the suitable targets because it can be easily collected and often includes enough amount of intact drug or its metabolites. Although various methods for urine analysis have been reported, several them need troublesome multistep operations. Therefore, the whole process tends to be time-consuming. Simple and rapid analytical techniques have been required in various scenes. We developed a pretreatment technique aimed at alleviating the disadvantage. The solid-phase analytical derivatization (SPAD) method was modified for the analysis of psychoactive drugs and stimulants in urine. Nortriptyline (Ntp), desipramine and methamphetamine (MA) were severally dissolved in human urine at appropriate concentrations. The urine samples were loaded to a cation-exchange solid-phase cartridge. After washing with water, the solid phase was dried by passing nitrogen gas. To derivatize the drugs, *N*-methyl-bis(trifluoroacetamide) (MBTFA) was added to the identical cartridge and it was incubated at room temperature for 15 minutes. The derivatized drugs were eluted by ethyl acetate and the eluents were analyzed by gas chromatography/mass spectrometry.Mass spectra of the trifluoroacetyl (TFA)-derivatives were clearly detected from the urine samples at a

concentration of 1 µg/mL. To assess the linearity of calibration curve, various concentrations samples (0.5, 2, 5, 10 µg/mL) were prepared with an internal standard and analyzed. The coefficients of determination (R^2) were 0.9999 (Ntp) and 0.9994 (MA). The drugs at 1 µg/mL in urine samples were detected, thus the present method is applicable to real urine samples collected from drug addicts for identifying drug ingestion. Since the calibration curves of Ntp and MA showed good linearities, quantification of target drugs may be possible using the present method. SPAD does not need complicated processes because extraction and derivatization are carried out on an identical cartridge.The present method should be a useful tool for drug analysis.

Keywords: Biological sample; psychoactive drug; Methamphetamine; solid-phase analytical derivatization

IRCPAS/2020/PP-218

Synthesis and Structural Identification of 5-Amino-4-hydroxyiminopyrazoles and (*E*)-*N*1-Aryl-3-aryl-4- [(substituted pyrazolyl)diazenyl] Pyrazoles from 5-Aminopyrazoles with Ethyl Nitrile

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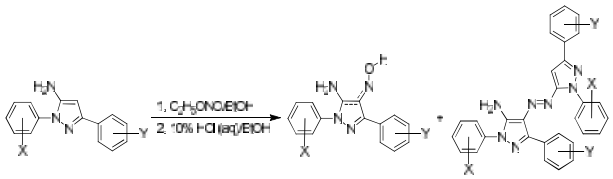
Pyrazoles play an important role among a wide variety of nitrogen heterocycles that have been used for developing useful agrochemicals and pharmacological agents.Furthermore, active nitroso compounds have also played significant roles in the synthesis of many biologically active heterocyclic compounds in organic chemistry and served as important chiral ligands or chiral auxiliaries for asymmetric synthesis.Therefore, 5-amino-4-nitrosopyrazole compounds, which graft nitroso group on the pyrazolic ring, were developed to constitute an efficient synthesis of 5-substituted imidazo[4,5-*c*]pyrazoles as CNS depressants or pyrazolo[3,4,-*b*]pyrazine.We have developed the conveniently one-pot synthesis method to give the 5-amino-4-hydroxyiminopyrazole and (*E*)-*N*1-aryl-3-aryl-4-[(substituted pyrazolyl)diazenyl]pyrazole as the corresponding products by reacting 5-aminopyrazoles with ethyl nitrile (10–20 wt% in EtOH) in presence of 10% $HCl_{(aq)}$.Scheme 1 shows the typical reaction condition of the one-pot synthesis for 5-amino-4-hydroxyiminopyrazole and (*E*)-*N*1-aryl-3-aryl-4-[(substitutedpyrazolyl)diazenyl]pyrazoles. The new procedure involved the treatment of 5-aminopyrazole with ~3.0 equivalent of ethyl nitrile in EtOH solution at room temperature for 10–30 mins. The clearly resulting solution was cooled and added with 10% aqueous hydrochloride acid for stirring within 0.5–1.0 h. While the starting material was consumed, the resulting mixture was concentrated under reduced pressure and worked-up. Consequently, the residue was charged onto the column in a little CH_2Cl_2 and the solvent could percolate down to the surface of silica gel. The column was eluted with EtOAc/*n*-Hexane (2:8). At first diazenylpyrazole band eluted from the column and then 5-amino-4-hydroxyiminopyrazole green band was sequentially eluted and isolated in high purity. The reaction gave a mixture of5-amino-4-hydroxyiminopyrazoleand the significant amount of coupling dimeric diazenylpyrazole, respectively (see Scheme 1).We have successfully developed the one-pot reaction to prepare 5-amino-4-hydroxyiminopyrazole and diazenylpyrazole derivatives by treating 5-aminopyrazoles with ethyl nitrile in presence of 10% $HCl_{(aq)}$. Following the further single-crystal X-ray diffraction study (ORTEP), the 5-amino-4-hydroxyiminopyrazole tautomer structure was first determined and demonstrated. This newly presented 5-amino-4-hydroxyiminopyrazole was opposed to the previous nitroso structure.



Scheme 1.Synthesis of 5-Amino-4-hydroxyiminopyrazoles and (*E*)-*N*1-Aryl-3-aryl-4-[(substituted pyrazolyl)diazenyl] Pyrazoles

Keywords:5-Amino-4-hydroxyiminopyrazole;4-[(Substitutedpyrazolyl)diazenyl] Pyrazoles;Nitrosonation; Pyrazoles;Ethyl nitrile

IRCPAS/2020/PP-220

Targeting Telomeric G-Quadruplex Complex by Perylene Diimides for Anticancer Activity: An *In-Silico* Study

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Telomerase enzyme which is expressed in detectable levels in the cells binds to telomeres and increases their length upon binding. This eventually leads to extension of lifespan of cells and makes telomerase an attractive target for cancer therapy. Perylene diimides bind to duplex genomic DNA of telomerase, and the resulting G-quadruplex ligands are responsible for binding affinity with respective proteins. Based on the IC50 values of perylene diimides, QSAR studies have been performed and the results are elaborated in preliminary research work. From the results of QSAR, the perylene ligands are selected for docking with telomerase as a target/protein. Based on the results of QSAR studies, new compounds are designed and synthesized. Now, the objective of the study was to dock the final synthesized compounds with the telomerase protein to study regarding the pKi value using G-quadruplex ligand database (G4LDB). The docked results are visualized using Discovery Studio Visualizer 4.1. The results are compared with the standard N, N'-bis-(2-(1-piperidino)ethyl)-3,4,9,10-perylene tetracarboxylic acid diimide (PIPER) drug and these compounds will be effective for anticancer therapy. The study was to investigate the docking results of synthesized perylene compounds with the results from G4LDB and visualized by Discovery Studio 4.1 Visualizer. The telomerase proteins selected for the study were extracted from Protein Data Bank, and the proteins selected for the study are (421b). Among the compounds (R1, R2, R3, and R4) screened in G-Quadruplex Ligand Database, compound R3 shows better binding affinity with good pKi value as well the interactions with the protein and ligand show better affinity with the targets and these are compared with the standard drug PIPER drug. Compound R3 possesses the best binding affinity with the target (421b) which shows that the compound will be effective for anticancer therapy

Keywords: AutoDock; G-Quadruplex ligand database; docking; Perylene derivatives

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Synthesis, Characterization, and Invitro Anti-Inflammatory Activity of Methoxydibenzofuran - 1, 3 -Thiazole-Carboxamide Derivatives

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The dibenzofuranthiazole is wellan established group of molecules that has resulted in pharmacologically and biologically active agents these are having a varied type of heterocyclic and straight chain structures. Although many of the existing drugs, for example, Tetomilast, oglemilast and ciliomilast are potent anti-inflammatory agents, they do have serious side effects. These include nausea, emesis, and gastric acid secretion. Many other standard drugs of today also, have other types of unwanted effects one important being drug resistance. Thus, newer agents without such undesirable side effects and better potency are the need of the day. This work was undertaken to study a few new compounds. It has been synthetically prepared in four step procedure by coupling reaction. The purity of all the synthesized derivatives was confirmed by melting point, thin layer chromatography and FTIR spectroscopy mainly. In addition to that ^{1}H NMR, Mass spectra studies were also done with most of the compounds. In continuation of synthesis the anti-inflammatory activities of the synthesized compounds were also studied by protein denaturation assay method. The yields of all the synthesized compounds were between 54-87%. It shows a moderate inhibition effect. Among these compounds 4c, 4d, 4e, 4f, 4g and 4h showed promising activity when compared to STD drug diclofenac sodium at low concentration (100 µg/ml) and the percentage of inhibition are found to be 35.86, 26.70, 27.16, 27.60, 38.54 and 32.57 µg/ml respectively. Where standard drug diclofenac sodium was 25.31 µg/ml. particularly the compound 4d shows very good inhibiting property at all concentrations when compared to the standard drug. This experiment suggests that the anti-inlammatory activity of dibenzofuranthiazole carboxamide derivatives mainly due to the halogenic derivative with para substitution. The fluro, chloro substitution was one of the key groups to enhance greatly the activity with para and ortho substituent, as well as the methoxy derivatives with meta substituent also shows moderate activity.

Keywords: Carboxamide; protein denaturation; anti-inflammatory

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UV-Spectroscopic Method for the Estimation of Fusidic Acid in Bulk and Pharmaceutical Dosage Form

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Fusidic acid is a steroidal antibiotic used to treat infections and acts as a bacterial protein synthesis inhibitor by preventing the turnover of elongation factor G (EF-G) from the ribosome. A simple, precise, and accurate UV Spectrophotometric method was developed for the estimation of Fusidic acid in pure form and in semi-solid dosage form. Cream equivalent to 1.2 gm of Fusidic acid was weighed and dissolved in 6 ml of methanol and centrifuged. Separate the 5 ml of methanol layer containing Fusidic acid and centrifuge transferred in to 100 ml

volumetric flask and sonicated for 15 min and the volume was made up to the mark with methanol to get the final concentration of 40 µg / ml and the absorbance was made at 244nm. The method was validated pertaining to linearity, precision, and accuracy studies, LOD and LOQ consistent with ICH guidelines.Different aliquots of Fusidic acid in methanol were prepared as per the test method in the concentration range of 4-24 µg / ml. The correlation co-efficient, slope, intercept, LOD and LOQ were done statically with of prism software. The correlation co-efficient value for the calibration graph was found to be 0.9996. The amount of Fusidic acid was found to be 100.91 ± 0.4343 Sample solution of Fusidic acid was prepared (12 µg / ml) in methanol for six times. The amount of Fusidic acid was found to be 101.16 ± 0.3182.The rapid UV spectroscopy method developed for quantitative analysis of Fusidic acid cream in pharmaceutical dosage forms is precise, accurate, linear, robust, and specific. This newly developed UV-spectroscopy method for Fusidic acid cream assay determination was found to be capable of giving good resolution these methods was completely showing satisfactory data for all the parameters tested. This method gives excellent performance in terms of sensitivity, speed, and especially concerning in the decrease of solvent consumption when compared to most of the reported Spectrophotometric methods and suitable for rapid analysis of Fusidic acid cream in bulk drug and in dosage forms

Keywords: Fusidic acid; spectrophotometric method; ICH guidelines

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6-Shogaol Attenuates Colonic Tumorigenesis and Oxidoinflammatory Response in Male Balb/C Mice

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Colorectal cancer (CRC) is the fourth leading cause of cancer related mortality worldwide. Several complications such as male reproductive dysfunction have been associated with increased incidence and prevalence of colorectal cancer. Adverse effects have been associated with the treatment of colorectal cancer using the available therapeutic agents. A bioactive component of *Zingiber officinale,* 6-Shogaol(6-S) has been reported to be biologically active in experimental models. However, there are limited information regarding the effect of 6-S on CRC.This study therefore investigated the biological activity of 6-S on CRC. Sixty male BALB/c mice (19±3g) were used for this experiment. Animals were divided into four groups (n=15). Groups 1 and 2 were administered corn oil (2mL/kg) and 6-S (20 mg/kg) orally for 16 weeks. Groups 3 and 4 received a single dose of AOM (10mg/kg, *i*P) and 3 cycles of dextran sulphate sodium (DSS) (2% w/v), singly (group 3) or in combination with 6-S (20 mg/kg) (group 4) for 16 weeks. Biomarkers of CRC such as oxidative stress, inflammation, cell proliferation was assessed colon tissues by microscopy, ELISA and spectrophotometric techniques. Data were analyzed using ANOVA at $P = 0.05$. Tumour incidence, ulcerated adenocarcinoma, tumour necrosis factor alpha, Ki-67 protein, carcinoembrayonic antigen, nitric oxide levels, lipid peroxidation and myeloperoxidase activity were significantly suppressed with pre-treatment with 6-S when compared with the mice treated with AOM/DSS alone. Additionally, glycogen synthase kinase 3β, CAT, SOD, GPx activities and GSH level decreased in mice that received AOM/DSS only. This decrease was conversely prevented in 6-S pre-treated mice. In conclusion, 6-Shogaol showed chemoprotective effect on AOM/DSS induced adenocarcinoma and colorectal cancer in mice through its antioxidant, anti-

proliferative and anti-inflammatory properties. Thus, 6-Shogaol could be a potential phyto-compound for use in the prevention and management of colorectal cancer.

Keywords:6-Shogaol; colorectal cancer; chemoprotective; antioxidant; antiproliferative; anti-inflammatory

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Isolation and Characterisation of *Guiera Senegalensis* Leaves Active Compounds

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Antimalarial plant*Guiera senegalensis* ("Senegal *guiera"* in English; "Sabara" in Hausa) (*Combretaceae*) is used for the management of malaria in Africa and other parts of the world with little or no scientific backing on its active component(s). Thus, the aim of this research was to characterize compounds from *Guiera senegalensis*(GS) ethylacetate extract in an attempt to provide an insight for the identification of lead molecules for drugs development.The study assayed for *in vitro* antiplasmodial activity of different extracts (n-hexane, chloroform, ethylacetate and methanol) of the plant. The most active extract wasfractionated using column chromatography. Further fractionation and isolation (Preparatory Thin Layer Chromatography) were guided by *in vitro* antiplasmodial assay and the characterization/identification of compounds done by LC/MS (QTOF-MS/MS in positive and negative ion modes) and supported by FTIR. The ethylacetate extractwas found to have bestantimalarial activity over the remaining solvent extracts tested (hexane, chloroform, and methanol extracts) and therefore chosen for the study. Fractionation of this ethylacetate extract gave eight (8) fractions and *in vitro* antiplasmodial assay on the fractions revealed GS-8 fraction to have the best % inhibition in parasitaemia. Preparatory-TLC with hexane:ethyleacetate:methanol (3:2:2) revealed 7 compounds. *In vitro* antiplasmodial activity showed sub-fraction GS-8B to have the highest percentage inhibition. Thin layer chromatogram of GS-8B revealed two eluting compounds. The LC-MS (Q-TOF/MS-MS) in the positive and negative ion modes coupled with FTIR revealed that GS-8B compounds are mostly terpenoids and alkaloids. Labdene-13,14,15-triol, Quinoline, Labdane, Quinoxaline, Delta-Valerolactone and Ergostanol were isolated. In conclusion, the constituents of *G. senegalensis* ethylacetate leaf extract (especially the terpenoids and alkaloids) couldbe of great potential for anti-malarial drug developmentand beneficial in the management of malaria as used traditionally.

Keywords: *Guiera senegalensis;* antiplasmodial assay; active compounds

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*In Silico*Evaluation for 8-aminoquinoline Hybrid Compounds as an Antimalarial Agent

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Malaria is a detrimental disease with a history of about 4000 years, is still endemic particularly in developing countries. Disease caused by the *Plasmodium* parasite that transferred in human through the infected female Anopheles mosquito was responsible for an estimated 445000 deaths worldwide in 2016. Few drugs have been well-developed and dispensed for the treatment of malaria such as chloroquine and artemisinin but ever-growing drug resistance against these classes of drugs has rendered them ineffective. Although chloroquine and its derivatives have suffered from the resistance problem, the potential of hybridisation strategy has helped to revive this important class of plasmodial agents by changing the structure of the molecule. In designing new antimalarial hybrid compounds, 8-aminoquinoline and pyranopyrazole compounds are designed together forming a hybrid and evaluated using the in-silico study. This study is utilized to recognize the binding interactions of designed 8-aminoquinoline hybrid compounds with the residues in the active site of plasmodium parasite. Five derivatives of 8-aminoquinoline hybrids were used in this study. The protein crystal structure of the *Plasmodium falciparum* lactate dehydrogenase (*Pf*LDH) enzyme with cofactor was retrieved from the Protein data bank (PDB ID:1CET, resolution: 2.05 Å). Discovery Studio 2016 (Accelrys Inc. San Diego, CA, USA) was used for the above protein and ligand preparation. The PDB files were prepared for docking in Molegro Virtual Docker (MVD) software. The in-silico study revealed that all five newly designed 8-aminoquinoline hybrid compounds exhibited comparable binding affinity as compared to reference established drug chloroquine(-70.4656kcal/mol) with binding affinity between -71.789kcal/mol to -98.8203kcal/mol. All five 8-aminoquinoline hybrid compounds were suitable (RMSD:0.72Å) and successfully docked inside the chloroquine binding site using MVD software. 8-aminoquinoline hybrid compounds exhibited promising binding affinity by forming hydrogen bond with the amino acid residue besides exhibited similar interactions with the amino acid residues as per chloroquine bind to *Pf*LDH enzyme. Those amino acid involve includes VAL26, ALA98, ILE54, GLU122 and ILE119.

Keywords: *Plasmodium* parasite*;* malaria; 8-aminoquinoline; antimalarial hybrid; *in-silico*

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Epitope-based Peptide Vaccine Against Fructose-bisphosphate Aldolase (FBA) of *Madurella mycetomatis* Using Immunoinformatics Approaches

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Mycetoma is a distinct flesh eating and destructive neglected tropical disease. It is endemic inmany tropical and subtropical countries. Mycetoma caused by bacterial infections

(*actinomycetoma*) such as*Streptomycessomaliensis* and *Nocardiae* or true fungi (*eumycetoma*) such as *M.mycetomatis*. To date,treatments fail to cure the infection and the available marketed drugs are expensive and toxic uponprolonged usage. Moreover, no vaccine was prepared yet against mycetoma.The aim of this study is to predict effective epitope-based vaccine against fructose-bisphosphate aldolaseenzymes of *M. mycetomatis* using immunoinformatic approaches.Materials and Methods: Fructose-bisphosphate aldolase of *M.mycetomatis* Sequence was retrieved fromNCBI. Different prediction tools were used to analyze the nominee's epitopes in Immune Epitope Databasefor B-cell, T-cell MHC class II & I. Then the proposed peptides were docked using Autodock 4.0 softwareprogram.Results and Conclusions: The proposed and promising peptides KYLQ show a potent bindingaffinity to Bcell,FEYARKHAF with a very strong binding affinity to MHC1 alleles and FFKEHGVPL thatshow a verystrong binding affinity to MHC11and MHC1 alleles. This indicates a strong potential toformulate a newvaccine, especially with the peptide FFKEHGVPL that is likely to be the first proposed epitope-basedvaccine against Fructose-bisphosphate aldolase of *M.mycetomatis*. This study recommends an in-vivoassessment for the most promising peptides especially FFKEHGVPL.

Keywords: *Madurella mycetomatis*; epitope-based vaccine; docking

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Development and Validation of Spectrophotometric Method for the Determination of Levodopa (L-Dopa) in Pharmaceutical Formulations

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Parkinson's disease is one of the most difficult medical condition. The cause of this disease is a significant depletion of dopamine due to the death of neurons which can produce dopamine in brain. It leads to tremor, muscle stiffness, bradykinesia. Levodopa (L-dopa) is a precursor of dopamine which is an important neurotransmitter which is used for the medication of neural disorders such as Parkinson's disease. After administration, levodopa (L-dopa) is converted into dopamine through enzymatic reaction catalyzed by dopadecarboxylase. (L-3, 4 dihydroxyphenylalanine) is a chemical that is biosynthesized by humans, and some animal from the amino acid L-tyrosine. L-Dopa is used in the treatment of Parkinson's disease and dopamine-responsive dystonia. This study will illustrate a simple rapid spectrophotometric method for determination of L-dopa in its dosage form the method that depends on the reaction between L-dopa and ARS. The experimental conditions (pH, reagent concentration, reaction temperature) were studied and optimized. Following the proposed experimental conditions, the relationship between the absorbance and concentration was quite linear in the concentration ranges. The intercept (a), slope (b), correlation coefficient (r), and molar absorptivity (ε) were determined. **(**ARS) has been used as a color- developing reagent in the spectrophotometric determination of pharmaceutical amines. The reaction of L-dopa with ARS results in the formation of a charge transfer complex of the n–π -type. This compound is an intermediate molecular association complex which dissociates in the corresponding radical anions in ethanolic solvent. The radical anion (absorbing species) absorbs at 588 nm. The described spectrometric method for the determination of L-dopa in pharmaceutical formulation is simple, sensitive, rapid, and accurate. The method is reliable and efficient for routine application in

quality control laboratories for analysis of L-dopa the method was validated using ICH guidelines and the results was satisfactory.

Keywords:Levodopa**;** Alizarin Red S; spectrophotometric; dosage form; absorptivity, validation

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Detection of Mercury Levels in Some Cosmetic Products Marketed in Khartoum, Sudan

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Mercury (Hg) is one of the most hazardous heavy metals that can negatively impact human health. Because of the widespread use of cosmetic products, many studies were reported for the determination of Hg in these products using different methods and instruments. The purpose of this study is to determine the presence of Hg in several cosmetic products marketed in Khartoum, Sudan. Additionally, the study also aims at comparing the Hg level detected in selected samples with the standard limit value set by The United States Food and Drugs Administration (FDA). Samples of the commonly used brands of cosmetic products (n=46) were collected from different markets in Khartoum, Sudan. Samples were categorized under eight main groups, viz: facial powder, mascara, eyeliner, eye shadows, lipsticks, eyebrows powders, foundations, and facial soaps. A weight of 100 mg was tested using a Direct Mercury Analyzer (DMA) device, the integrated sequence of thermal decomposition, catalyst conversion, and amalgamation were applied. Hg concentrations in 61% of the studied samples were found to have Hg level below the limit stated by The United States Food and Drugs Administration (US FDA), which is less than 1 ppm. Where 39% of the samples were having Hg level above 1 ppm. The most significant trend was for (Yoko) soap which has Hg concentration of 10.56 ppm. However, the (Anastasia) eyebrows powder Hg concentration was detected to be (0.07 ppm). Our study showed that Hg was detected in various cosmetic types. The results demonstrate that the concentration of mercury in 39% of products obtained did not meet the FDA standards. Facial soaps along with lipsticks show relatively high levels of Hg. Several brands of eyebrow powders, eyeliners, foundations, mascara, and facial powders had a significant variation at Hg levels. The hazardous nature of these products could result from their route of application. In conclusion, we suggest conducting more studies with larger samples to get a better understanding of the extent of the problem.

Keywords:Cosmetic products; mercury levels; heavy metals; Sudan.

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Prediction and Conservancy Analysis of Multiepitope Based Peptide Vaccine Against Merkel Cell Polyomavirus: An Immunoinformatics Approach

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Merkel cell Polyomavirus is a non-enveloped, dsDNA virus belonging to Polyomaviridae family linked to an uncommon aggressive skin malignancy. The poor prognosis and limited understanding of disease pathogenesis warrants innovative treatment. In this current study we aim to predict T

and B cell immunogenic epitopes from the VP1 protein of all merkel cell polyomavirus strain which will aid in effective epitope-based vaccine design using immuoinformatics approaches. We retrieved 423 full-length VP1 protein sequences of merkel cell polyomaviruse species from the NCBI database. These sequences were analyzed to determine the conserved region and were used to predict the epitopes using the immune epitope data base immunoinformatics algorithms. A total of nine promiscus epitopes were predicted. For B cell, three epitopes were selected as peptide vaccine (QEKTVY, KTVYPK, and QEKTVYP). For T cell the predicted Class-I peptides (SLFSNLMPK, LQMWEAISV and LLVKGGVEV) were found to cover the maximum number of MHC I alleles. The highest scoring Class II MHC binding peptides were (IELYLNPRM, ISSLINVHY and INSLFSNLM). Further in vivo experiments will need to be undertaken to confirm the potential of these predicted epitopes in a future efficacious novel vaccine development.

Keywords: Merkel cell polyomavirus (MCPYV); epitope; peptide vaccine; immune epitope database IEDB

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Design of Epitope Based Peptide Vaccine Against *Plasmodium Falciparum* Translationally Controlled Tumor Protein using Immunoinformatics

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Malaria is a life-threatening disease caused by parasites that are transmitted to people through the bites of infected female Anopheles mosquitoes. *Plasmodium falciparum* is one of four human's parasitic species that belongs to the genus Plasmodium. It is responsible for causing 50% of malaria incidence throughout the world. It is the most lethal and accounts for 98% of all lethal cases. This study is aimed to predict an effective epitope-based vaccine against Translationally controlled tumor protein "TCTP" enzyme of *P.falciparum* using immunoinformatics approaches. *Plasmodium Falciparum* TCTP sequences were retrieved from National Center for Biotechnology Information (NCBI) database. The conserved regions were introduced into Immune Epitope Database (IEDB) analysis resource to predict B-cell, T-cell MHC class I and II. Homology modeling for the protein was obtained using certain homology modeling servers "RaptorX server" and 3D structures of the most promising epitopes were obtained and visualized using visualization programs "UCSF Chimera program". The proposed and promising peptide SYVQQDPFE showed a potent binding affinity to B-cell, MEAGIIYSY with a very strong binding affinity to MHC I alleles, and IYSYYKGEEITPRFV that showed a very strong binding affinity to MHC II alleles. This study is considered as the first study to report the use of TCTP protein as vaccine candidate and support immunization against *P. falciparum* malaria with promising percentage of population coverage in the world and in Sudan. The candidate peptides are selected because they enhance immunity at both B and T-cell levels. The results were promising to formulate a vaccine with more than 93.73 % population coverage worldwide and 82.13 % in Sudan, excluding certain MHC II alleles. This study recommends an in vivo and in vitro assessment for the most promising peptides and considering them as potential candidates for developing a peptide-based vaccine for Malaria.

Keywords:Immunoinformatics; *Plasmodium falciparum*; translationally controlled tumour protein; peptide vaccine; epitope

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In silico Pharmacological Evaluation of a Novel Synthetic Chalcones

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Infectious diseases are major public health problem therefore,development and discovery of new antimicrobials is needed. The computational drugdesign methods: specifically, molecular docking (MD) along with molecular dynamicssimulations (MDS), play an imperative role in understanding the drug-receptormolecular recognition events.To perform an *in-silico*investigations for antimicrobial activity of fournovel chalcone compounds (TChD: 01-04) and to determine the exact possiblemechanism(s) and the site(s) of action.AutoDock Tools 4.0 and Molecular Operating Environment (MOE)programs were used for optimization and validation of the selected targets, thendetermination of the potential binding sites, the four tested ligands were docked intothese pockets. The most promising conformations were further validated usingmolecular dynamics simulations.Compound (TChD-02) docking results demonstrated a higher score in termsof binding free energy with two targets; Dihydrofolate reductase and bacterial DNAgyrase; with binding free energies: -8.21 ±0.09 and -8.89±0.12, respectively. MDsimulations in comparison with the best docked conformers resulted in an enhancedbinding of compound TChD-02 with both targets.MD and MDS were done on four novel chalcone derivatives against sixbacterial targets. Among them, TchD-02, was found to be the most suggestedpromising compound and possibly able to undergo further investigations anddevelopment. Chalcone derivatives have excellent scope for further development as commercial antimicrobial agents.

Keywords: Molecular docking, molecular dynamics simulations, chalconederivatives, antibacterial activity, and DNA gyrase

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The Effectiveness of Automated Tablet Dispensing System In-Patient Pharmacy: A Systematic Review

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In-Patient Pharmacy known as provides services to the wards, clinics, and otherunit's in the hospital through the term called Unit Dose System Supply and top-up medicinestocks. Lately Automated Tablet Dispensing System were introduced at private healthcare in Malaysia. An automated tablet dispensing, and packaging system is generally provided with a tablet packaging portion and a tablet dropping portion placed above the packaging portion.Added advantage for having these Automated Tablet Dispensing System is to minimize thetime of preparing the medication, less medication error and reduction of workload. The focus of the research is to determine the Effectiveness of AutomatedTablet Dispensing System at In-Patient Pharmacy. The effectiveness of Automated Tablet Dispensing System to draw attention to the

pharmacy care providers as it meets patientneeds. We also highlight the importance to minimize the time and workload can be achievedthrough the Automated Tablet Dispensing System.This research was conducted by calculating the average time taken (mins) for preparing the medication through Automated Tablet Dispensing System. The Variables assessed were total number of patient present at each ward and overall time taken (mins). The data was collected for August 2019. In this short period of study, The Effectiveness of Automated Tablet Dispensing System could be determined.Based on the data obtained, total number of patients were 3506 and total time taken was 5430 (mins). Average time taken calculated were 90.5 mins. The result indicates that Automated Tablet Dispensing System is effective in preparing the medication and reducing the workload of the staff.The effectiveness of Automated Tablet Dispensing System proven to be efficient by fulfilling the needs of patient's which can minimize many human related errors.

Keywords:automated tablet dispensing; inpatient pharmacy

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Investigation of thymoquinone stability in black seed oil alginate beads

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Thymoquinone (TQ) is the main active compound in black seed oil (BSO) which has unacceptable taste. Many reports confirmed that TQ has medical uses, but very few reports have documented its quantification or stability after formulation. In addition, stability of pure TQ under different conditions was studied to confirm that TQ was unstable in aqueous solutions, especially under alkaline environment. On the other hand, it is rarely to find reports about stability of TQ in BSO or BSO formulations. However, one of the most strongly used excipient in taste masking and improvement stability of the active compounds is alginate which can fabricated as beads in the presence of calcium ions.This study aimed to encapsulate BSO in alginate beads, and then to study stability of TQ in both forms of BSO and BSO-alginate beads.BSO-alginate beads were fabricated in 1% w/v calcium chloride bath. Concentration of BSO in the beads was 15% w/w. Aqueous solution of TQ, BSO, and BSO-alginate beads were packed in glass containers, well closed and stored at 40°±2°C /75±5% RH for 30 days. After specific time intervals (0, 15, and 30 days), the TQ content was analysed by using HPLC.The results indicated a poor stability of TQ in aqueous solution. Concentration of TQ in the aqueous solution was significantly (p<0.05) decreased from 97.29±0.33 µg/mL at day 0 to 56.80±0.08 µg/mL at day 30. Moreover, concentration of TQ extracted from BSO showed a significant decreasing (p<0.05) from 2.11±0.02% at day 0 to 1.95±0.005% at day 30. On the other hand, the stability of TQ in alginate beads containing BSO was highly improved (p>0.05), (1.65±0.03% at day 0 and 1.58±0.04% at day 30). TQ in BSO may undergo some oxidation reaction during the period of storage in the stability chambers resulting in TQ degradation. On the other hand, alginate beads act a barrier against TQ oxidation, prolonging its shelf-life. This study suggests that BSO-alginate beads are stable in term of active principle and can be used as a source to provide TQ due to the unsuitability of aqueous solutions to formulate TQ.

Keywords: Black seed oil; *Nigella sativa*; thymoquinone; alginate; beads; stability

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Development, Evaluation and Optimization of hydrophilic matrix SR tablet containing paliperidone using3^2fullfactorialdesigns

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Anti- depressantsaretheclassesofdrugswhichcanelevate moodindepressive illness.To maintain the plasma concentration of the drug within the therapeutic window and thereby to increase the patient's compliance, aim to design SR matrix tablets of paliperidonean acceptable pharmaceutical formulation in shortest possibletimeusingminimumnumberofman-hours andrawmaterials.A 3^2 randomized full factorial designs with two factors, each at three levels,utilized in the study. The concentration of HPMC (X_1) and concentration of PEO (X_2) were selected as independent variables. The percentage drug release at 2, 6 and 8^{th} hours were Q2, Q6 and Q8 respectively selected as dependent variables. SR Tablets were prepared by direct compression technique. The powder blends of formulations were evaluated for precompression evaluations. Compatibility studies were carried out prior to the preparation of tablets. The tablet formulations were evaluated for post compression evaluations and subjected for *in vitro*release studies using USP XXIIIapparatus. The samples were analyzed at predetermined interval at 275nm using UV Spectrophotometer. The results of DSC study shown that there was no significant change in the chemical integrity of the drug. The results of precompression evaluation showed acceptable pharmacotechnical properties and complied with the in-house specifications for post compression evaluations. The drug release profiles were characterized by an initial burst effect Q2 i.e. initial 30-35% drug release required in 2 hrs. PEO was responsible for initial burst effect and HPMC was used to sustained drug release. None of the batches gave the release profile as targeted at desired level fixed; however on varying the concentration of HPMC and PEO in various levels, it was found that batch F16 and F17 showed the least release profile and also the release is sustained as the polymer concentration increases, but after reaching certain level, there is no effect on release of drug and F16 in which HPMC and PEO are used at 35% concentration level was selected as optimized batch. The formulations showed good linearity ($R^2$0.963), with slope (n) value 0.369, indicating that diffusion is the dominant mechanism of drug release with these formulations. As the release profile of core matrix tablet could not match the required drug release profile, it is decided to further control the release of drug by functional coating with EC as a polymer using PEG as plasticizer as a future work to match the initial time point of release.

Keywords: Paliperidone; factorialdesign; hydrophilic matrix; SR tablet; HPMC; PEO

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Formulation and optimization of hydralazine HCl sustained release mucoadhesive buccal tablets using 2^3 factorial design

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Hydralazine HClsustained release mucoadhesive buccal tablets usedto treat hypotensive. The objective of the present investigation is to develop unidirectional, bilayered, buccoadhesive tablets of Hydralazine HCl using a buccoadhesive polymers Xanthan gum, HPMC-K4M, Carbopol-974P along with ethyl cellulose and magnesium sterate as an impermeable backing layer. Formulations were prepared using 2^3 full factorial designs to explore the effects of Xanthan gum, HPMC-K4M and carbopol-974 P (as independent variables) on mucoadhesive strength and drug release (as dependent variables). The results of FTIR and DSC analysis indicated that the compatibility of drug with excipients. The buccal tablets were evaluated for theweight variation, content uniformity, surface pH, swelling index, ex vivo mucoadhesive strength, in vitro drug release and ex-vivo permeation studies and release kinetics. The release profile data was subjected to curve fitting analysis to describe the release mechanism from the buccal tablets. The results are within acceptable limit and also the results demonstrated that the mucoadhesive performance of Xanthan gum largely depended on their characteristics, i.e. higher degree of esterification and molecular weight gave a stronger mucoadhesion. Therefore, it can be utilized for the development of mucoadhesive carrier for buccal drug delivery systems utilising Xanthan gum for Hydralazine HCl based on information reported here in.

Keywords:2^3 fullfactorial design;Xanthan gum; hypotensive agent;Hydralazine HCl; bioadhesion.

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Intend of Nanostructured Lipid Carrier Containing O-Padimate - Assessing Quality of Life in Patients with Sunburn

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Sun burn has become a cosmetic and therapeutic concern in day-to-day life, based on most reported outcomes of skin cancer on extensive use of marketed available sunscreen products containing more concentration of sunscreen agents, followed by repeated administration. The above said been intend of study, focused on design of formulation containing 0-padimate Nanostructured Lipid Carrier (NLC) using meta-analysis for rational use of ingredients aiming at improving Trans-epidermal Water Loss (TEWL). The lead formulation passed for safety with tests on epidermal cells, physical behavior by thixotropic analysis and patient compliance as per SKINDEX-16 questionnaire. The SCF-16 formulation with the minimal concentration of 10 mg encapsulation, 97 % drug release for 12 hours and no sensitization reported. The concentration of kernel oil regulates the release and amount of plasticizer regulating membrane permeation proved on analysis of variance by meta-analysis software. (METASOFT-201A). The Nanostructured lipid carrier formulation SCF – 16 with the kernel oil as lipid carrier is a promising formulation with good SPF and TWEL capability.

Keywords:Nanostructure lipid carrier; sunscreen; O-Padimate; trans-epidermal water loss

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iRGD Peptide Mediated Delivery of PLGA Nanoparticles for TargetedDelivery of Garcinol Against Colon Cancer

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Garcinol is a naturally occurring polyisoprenylated benzophenone derivative found to beeffective in experimental cancer models such as colon, tongue, and breast cancer. But due to itshydrophobicity it became difficult to attain significant oral bioavailability while administered*in vivo*. To overcome this problem in the earlier work from our laboratory Gaonkar et alreported encapsulation of garcinol in vitamin E-TPGS emulsified PLGA nanoparticles (GARNPs)and found this nanoparticle to be effective (more cytotoxic) compared to free garcinolagainst many types of cancer cells, as determined by lowering of IC50 values. While evaluating*in vitro* cytotoxicity of garcinol loaded PLGA nanoparticles against human colon carcinomacells; HCT116, low reduction of IC50 value (25.8 µM for GAR and 20 µM for GAR-NPs) drew our attention that there would be some possibilities to increase the selectivity of GARNPtowards colon cancer cells so that we can deliver garcinol loaded nanoparticles towardscolon cancer in a target oriented approach. In this perspective we focused on selectivebiomarkers expressed on colon cancer cell, the integrins; and selected a cyclic peptide (iRGD) containing the RGD motif which is responsible for recognition and binding of peptide to theintegrins (αvβ3, αvβv). In this study we developed a garcinol loaded PLGA nanoparticleconjugated with iRGD peptide (iRGD-GAR-NPs) and demonstrated its *in vitro* and *in vivo*efficacy against colon cancer. HCT116 cells were used for *in vitro* experiments. Dimethylhydrazine induced colon cancer bearing Sprague Dwaley rats were used for *in vivo* assessmentof comparative anticancer efficacy of GAR-NPs and iRGD-GAR-NPs. Our findings suggestedthat in *both in vitro* and *in vivo* experiments peptide conjugated nanoparticles (iRGD-GARNPs)were superior in selectivity and cytotoxicity towards colon cancer compared tounconjugated nanoparticles (GAR-NPs). We are hopeful that this work could be a contributorystep in target-oriented therapy of cancer.

Keywords: Colon cancer; iRGD peptide; Nanoparticles; Garcinol

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Design and *ExVivo* Evaluation of Dantrolene Transdermal Emulgel by Using Natural Penetration Enhancer

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Emulgel is one of the recent technologies in NDDS used for sustaining release of emulsion gel for topical use.Gel formulations generally provide faster drug release compared with conventional ointments and creams. Dantrolene is a well-known therapeutic agent that is used mainly for its skeletal muscle relaxants.The aim and objective of the study is to formulate Dantrolene emulgel for topical application.Emulgel of Dantrolene, consist of Carbopol-940 or HPMC K4 as gelling agents for gel formulation and tween 80, span 20, for emulsion

formulation.The results of the present investigation showed that the formulated Emulgel by emulsion incorporated in gel. Dantrolene loaded emulgel was formulated by using o/w emulsion because of lower solubility in water.Lemon grass oil,Menthol was used as a penetration enhancer in emulgel formulation.Optimized formulation was evaluated for physical examination, swelling index, skin irritation study, extrudability study, drug content determination, spreadability, globule size determination and *invitro* drug release,rheological study. Optimized formulation shown drug release 98.6% for 12^{th} hrs. The stability studies were conducted for optimized formulation and found to be stable during stability studies.The emulgel was exhibited good sustained release characteristics both *in vitro* and *exvivo*.It may be concluded that emulgel using natural penetration enhancers were a suitable candidate for oral sustained delivery of Dantrolene fulfilling one of the major objectives of the investigation.

Keywords:Emulgel; Dantrolene; Carbopol 940 P; Span 20

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Emulgel as Remarkable Drug Delivery System for Topical Preparations: Formulation Development of Emulgel & Quality Control Testing of Chloramphenicol Palmitate

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The emulgels appeared to be the remarkable drug delivery system through topical preparations. The main purpose of these formulations is to provide an alternative drug delivery system for the hydrophobic drugs (like Chloramphenicol) to be in systemic circulation through skin. In current research, Chloramphenicol Palmitate was formulated as topically applied emulgel. Then the prepared emulgel was evaluated for physical appearance, rheological behavior and stability studies. Results reveals that topical emulgel is formulated and developed in good yield and showed acceptable physical properties concerning color, homogeneity, consistency, and pH value. Rheological studies revealed that the developed formulation exhibited a shear thinning behavior with thixotropy. Further these properties were evaluated for 3 months by placing the emulgel in stability chamber and the prepared emulgel is found stable and required parameters were in acceptable limits. Here Chloramphenicol emulgel is formulated for its intended uses with more advantages i.e. non-greasy, spreadable easily, removable easily from the skin, emollient, long shelf life, transparent and bio-friendly with a pleasant appearance and thixotrophic in nature.

Keywords: Chloramphenicol emulgel; long shelf life;quality control testing;rheological studies; topical preparations

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Fabrication and characterization of cationic solid fat nanoemulsions by Quality by design

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Solid Lipid Nanoparticles (SLN) colloidal systems for drug delivery significantly reduces the inconveniences associated with other systems such as liposomes,polymeric nanoparticles, emulsions, etc. The chief advantages including thecapability to integrate both hydrophilic and

hydrophobic drugs as well as the ability to target thecell. The properties of SLN compared to polymeric nanoparticles arebased on their low cytotoxicity, high capacity for transfection, better stability in biologicalsystems. Cationic solid lipid nanoparticles are having the extra properties of making complexwith negatively charged antigens and targeting the antigen to the target cells.Current work is based on thedesigning of cationic solid lipid nanoparticles and its pharmaceutical evaluation will be done. The cationic SLNs were fabricated by Box Behnken design (BBD) and microemulsion (O/W) was prepared using Precirol ATO-5 and stearylamine as the cationic lipid. Precirol ATO-5 is heated to 10ºC above its melting point, and 10 ml of a hot aqueous solution of poloxamer and stearylamine in different proportions is added. The sample is stirred for 30 min at 10,000 rpm in mechanical stirred. The nanoparticles are obtained by dispersing the hot microemulsion in cold water. To separate cationic SLNs, the resultant suspension is centrifuged. Different aliquot of the recombinant Hepatitis B surface antigen (rHBsAg) in PBS is added to different volumes of a suspension of the cationic SLNs by stirring with magnetic stirrer. Particle size and zeta-potential results were obtained (between 339–445 nm and +36–52 mV), it was observed that nanoparticles obtained with stearylamine: poloxamer proportions 1:2, had a better size–charge relationship; smaller cationic SLNs with a higher zeta-potential were obtained. The spherical shape of the complexes was shown by TEM images. Cationic SLNs were obtained with a size of 339 nm and with a surface charge of approximately +41 mV. The method developed by BBD is suitable for obtaining cationic SLNs that can form a complex with rHBsAg.

Keywords: Cationic Solid lipid nanoparticles; rHBsAg; Box Behnken Design

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Some Properties of *Bombax Costatum* Leaf Gum and its Application as Stabiliser in Emulsion

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Gum was isolated from the leaf of *Bombax costatum* plant, characterised and its potential as a stabiliser in emulsions was examined. The yield of *B. costatum* leaf gum was 45.6% and the CHNS analysis showed carbon (40.47%), hydrogen (5.44%), nitrogen (2.29%) and sulphur (0.02%). The levels of Pb, Cd, Cr and Cu in the gum were very low (≤ 0.05 mg/100g). The FT-IR showed characteristic absorption bands at 1700 cm^{-1} (free C=O) and 1609 cm^{-1} (COO^-) indicating presence of uronic acid which was quantified to be 32.0%. The polysaccharide had intrinsic viscosity of 5.8 dL/g and viscosity average molar mass of $5.35x10^5$ g/mol. The polymer concentrations 0.2-2.0% exhibited non-Newtonian properties with Power law index, n, in the range of $0.69 < n < 0.75$. At low concentrations 0.025 – 0.5%, the polysaccharide inhibited creaming of 10% olive oil-in-water emulsion with a third order polynomial fit ($R^2 = 0.9923$), indicating good stability.

Keywords: *Bombax costatum*leaf gum; uronic acid; oil-in-water emulsion; inhibition of creaming; rheology modifier

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Investigating the Effect of Polymer and Growth Factor Complex on Cell Proliferation

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Growth factor is a therapeutic protein used to treat mucositis. The growth factor is available in the form of intravenous bolus injection. In this study, new formulation of growth-factor polymeric complex was developed to improve patient compliance. The aim of this research study was to prepare growth factor complex with polymer to protect the proteolysis of growth factor. Intestinal cell proliferation rate was investigated by using growth factor complexed with polymer. The objective of this research study was to determine the interaction between polymer and growth factor through molecular docking. Furthermore, one of the objectives is to prepare and characterised the complex. Moreover, content analysis of complex was investigated by using ELISA test. Chemical interaction between complex in SDS-PAGE and the effect of complex on cell proliferation rate on intestinal cells was investigated by using MTT Assay were studied in this reseach project.The interaction between the polymer and growth factor complex was studied by molecular docking. Particle size and zeta potential of the complex was investigated by Malvern Zetasizer Nano Zs. Fourier-transform infrared spectroscopy (FTIR) test was performed to characterised the physical mixture of polymer, growth factor and polymer-growth factor complex. Enzyme-linked Immunosorbent Assay (ELISA) test was used for quantitative determination of keratinocyte growth factor. SDS-PAGE was performed to check the interaction between complex and proteases. Cell proliferation of FHs cell was studied by MTT assay by treating with complex.Complex was formed through the hydrogen bonding proven by docking studies. Zeta potential of the complex shown of having good stability. Stable complex was formed at pH 4.5 and being protected from proteolysis shown by SDS PAGE studies. From the MTT assay study, complex was increased the cell proliferation rate of FHs cells.Complex was formed at pH 4.5. Complexation of polymer with growth factor increased the proliferation rate of intestinal cell.

Keywords: Polymer and growth factor complex; Fourier-transform infrared spectroscopy; cell proliferation

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Polymeric Inclusive Complex Protects Growth Factor from Proteolysis: *In-vitro* Characterisation and FHs Cell Proliferation Studies

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To investigate the complexation of polymer with growth factorin protecting growth factor from proteolysis.To prepare and characterise polymer-growth factor complex. To study the effect of proteases on polymer-growth factor complex using Sodium Dodecyl Dulfate–Polyacrylamide Gel Electrophoresis (SDS-PAGE). To study intestinal cells proliferation rate ofpolymer-growth factor complex using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenytetrazolium (MTT) assay.Molecular modelling of growth factor with polymerwas carried out to determine the binding properties between growth factor and polymer. Polymer-growth factor complex wasprepared and characterised by the Fourier-transform infrared spectroscopy (FTIR). Enzyme-linked immunosorbent assay (ELISA) was used for content analysis of polymer-growth factor complex. Various studies conducted to investigate effect of proteases on

polymer-growth factor complex. Lastly, MTT assay was carried out to study the effect of polymer-growth factor complex on intestinal cell proliferation.The complexation between polymer and growth factor are through hydrogen bonding which involved primary amines and secondaryamides of the polymer with the nitrogen and oxygen atoms of amino acid. Our study found that polymer protected the growth factor from proteolysis by forming complex. MTT assay showedpolymer-growth factor complexincrease intestinal cells proliferation.This research study showed that the polymer is a potential nano carrier to protect growth factor from protease enzymes and improve the intestinal cells proliferation by forming a complex with the growth factor.

Keywords: Nanoparticles; polymer;growth factor; proteases enzyme

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Encapsulation of Fenofibrate in Solid Lipid Microparticles by Central Composite Face-Centred Guided by Electrospray

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Electrospray is a method able to produce monodispersed particles using applied electric field to deform the interface of liquid coming out from a capillary. The abstract aims to investigate the influence of electrospray critical process parameters (CPPs), including flow rate, solid concentration, and tip-to-collector distance on the critical quality attributes (CQAs) of electrosprayed fenofibrate microparticles such as: particle size and yield. Central composite face centred (CCF) design was carried out and analysed using partial least square (PLS). The results from the PLS analysis were used to develop models and identify an optimal operating space (OOS). The particle size and the yield models were significant with p-value (<0.05). Flow rate and solid concentration parameters significantly affected the particle size and yield, respectively. A lower flow rate will produce smaller particle size of electrospray particles. Particle size below 2 μm can be obtained when the flow rate is below 13 μL/min for solid concentration between 1-3% (w/w) at 10 cm tip-to-collector distance. To increase yield up to 80%, solid concentration should be around 2.8% electrosprayed at distance of 10.25 cm.Electrospray technique can be used to encapsulate solid lipid fenofibrate microparticles by controlling the parameters including flow rate and solid concentration. The knowledge gained using CCF approach in this study will help in rational experimental design to achieve desired electrospray solid lipid fenofibrate microparticles characteristics.

Keywords: Fenofibrate; solid lipid microparticles; electrospray; central composite face-centered

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Novel Fatty Acid-Based pH-Responsive Nanostructured Lipid Carriers For Enhancing Antibacterial Delivery

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Bacterial infections have been recognized as a major cause of deaths globally due to the limitations of current antibiotic conventional dosage forms. The introduction of nano delivery systems, primarily smart pH-responsive nano antibiotic delivery systems, can potentiate targeted antibiotic release; therefore, combat conventional dosage forms limitations and reduce resistance development. Formulation of pH-responsive nanostructured lipid carrier (NLCs) for targeted delivery of vancomycin (VCM) using synthesized novel fatty acid-based lipids that and undergo surface charge reversal in acidic medium. Two branched lipids [stearic acid derived solid lipid and oleic acid-derived liquid lipid] were synthesized and characterized using FTIR, 1H and 13C NMR. The NLCs were prepared using hot homogenization technique and characterized in terms of size, polydispersity index (PDI), zeta potential (ZP) (Dynamic Light Scattering), surface morphology (TEM), encapsulation efficiency (EE) (HPLC), *in vitro* drug release (Dialysis bag), cell viability study (flow cytometry), *in vitro* (broth dilution) and *in vivo* (mice skin infection model) antibacterial activity. VCM-NLCs were spherically shaped with size, PDI and ZP of 225.9 ± 9.1 nm, 0.258 ± 0.02 and -9.2 ± 2.7 mV respectively. EE (%) was 88.7±13.12. *In vitro* drug release studies revealed that VCM release was faster at pH 6 compared to pH 7.4. Cell viability study showed that NLCs had 2.5-fold better killing percentage than the bare drug at similar concentrations. *Invitro* antibacterial activity against methicillin-susceptible and resistant Staphylococcus aureus proved that at pH 6 NLCs activity was four times and two times better against SA and MRSA, respectively. Interestingly, at pH 6 it was 8 times better than bare VCM against both bacterial strains. *In vivo* study revealed that MRSA CFU load in mice skin treated with VCM-NLCs was 4-fold lower than bare VCM (p 0.0108). This novel pH responsive NLCs can improve bacterial infection treatment by protecting the antibiotics during systemic circulation, improve the targeted antibiotic release and enhance antibiotic localization at the acidic infection sites. Therefore, pH responsive NLCs show potential for efficient antibiotic delivery and serve as a promising nanocarrier for the delivery of poorly soluble antibiotics to enhance the treatment of infections.

Keywords: pH-responsive; Nanostructured Lipid Carriers; vancomycin; Methicillin-Resistant*Staphylococcus aureus* (MRSA)

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Nateglinide Nanocrystals: Development and Investigation of Formulation Variables to Improve Its Solubility

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Nateglinide nanocrystals are produced due to its problem of low solubility and high permeability. It is classified under the Biopharmaceutical Classification System (BCS) Class II. Due to its function as an oral antidiabetic drug, thus its solubility is one of its main factors

owing to its poor bioavailability. The main objectives of this study are to use ultrasonication probe method in the process of the formulation of Nateglinide nanocrystals as well as to find out the parameters that is most optimum during the formulation. The formulation variables considered for the nanocrystal development are the type of surfactants, concentration of surfactants and sonication time. The methodology used for the characterization were Coulter Counter Analysis, Zeta Sizer, Zeta Potential, Differential Scanning Calorimetry, Fourier Transformed Infrared Spectroscopy and Scanning Electron Microscope. There were no interactions between Nateglinide, and the surfactants used which were proven by the FT-IR spectroscopy. Coulter counter analysis serves as a method to choose the best formulation from each surfactant for further analysis by zeta sizer and zeta potential. All formulation showed size < 800 nm except for formulation D4 by the Zeta Sizer whereas Zeta Potential showed that formulation C4 and E2 have good stability. Five formulations A4, B3, C4, D4, and E2 were chosen out of 25 formulations for particle size analysis. Among the five formulations, formulation E2 has been determined as the optimized formulation based on its particle size and zeta potential. Formulation E2 is using polyethyleneimine (PEI) at 0.5% concentration and 15 minutes sonication time. It has particle size <200 nm and it has good stability based on Zeta Sizer and Zeta Potential.

Key words: Nateglinide, Nanocrystals, Surfactant, Ultrasonication, Zeta Sizer, Zeta Potential

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Factors Influencing Self-Medication and its Prevalence Among Adults in the Klang Valley, Malaysia

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Self-medication is an act whereby a person purchases a medication without consulting a healthcare professional. Self-medication can be seen as a form of self-care, whereby an individual treats their symptoms as soon as they recognize them. However, self-medication also brings certain risks such as inappropriate treatment and adverse effects. Currently, the prevalence of self-medication among adults in Klang Valley, Malaysia is unknown.In this study we aim to determine the prevalence of self-medication and the factors that influences the decision of adults in the Klang Valley to self-medicate.This study was a cross-sectional study in which 562 responses were collected from adults in the Klang Valley, Malaysia. Validated, self-administered questionnaires were used in this study. The developed questionnaire consists of three sections. Section A consisted of demographic data while Section B contained two parts: prevalence of self-medication for minor ailments and prevalence of self-medication for chronic conditions. Section C consisted of 20 questions on the perception towards self-medication where the answers were measured using a five-point Likert scale. Descriptive and inferential statistics as well as factor analysis were used to analyze the data. The overall prevalence of self-medication was 63.5%. Self-medication was practiced with no significant differences in populations with regards to age, gender, ethnicity, highest level of education and monthly household income (p> 0.05). Pharmacies are the most preferred place for respondents who self-medicate to obtain medications without consulting a healthcare professional, followed by using leftover medications at home, obtaining medication from friends and family, buying from a clinic without consulting the doctor and buying from a traditional medicines store. Respondents who self-medicate have an overall good perception towards self-medication compared to

respondents who do not self-medicate. The leading factorsinfluencing why respondents self-medicate was a positive perception towards self-medication as well as personal and shared experiences from family and friends in self-medication. The prevalence of self-medication is high among adults in the Klang Valley.Adults who self-medicate view self-medication in a positive light and may underestimate the possible risks that self-medication poses to the consumer.

Keywords: Self-medication; medicine safety; minor ailments

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Evaluation of Effectivity of Antiretroviral (ARV) Drugs Using CD4 Value Before and After ARV Treatment Among HIV Patients in Health Centre, Hospital Grade A and Grade B

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HIV or Human Immunodeficiency Virus is a type of virus that attacks/infects white blood cells that cause a decrease in human immunity Antiretroviral drugs (ARVs) are used in the treatment of HIV cases to improve the quality of life of people living with HIV even though they cannot cure it. CD4 examination is needed in patient monitoring to see the successful of ARVs use. The purpose of this study was to see whether there were significant differences between CD4 values which were indicators of HIV patients before and after using ARVs based on sociodemography and the level of patient compliance in health centre, hospital grade A and grade B. This study was carried out by retrospective cross-sectional study in hospital grade A, grade B and health centre in Jakarta. Total samples were 123 HIV patients whereas all patients who had HIV with grade I and were using ARV more than 6 months and they had complete medical record. The Results of analysis of HIV patients based on sociodemography, patients aged 26-45 years old was 67.47%, 49.59% men, 30.89% had a high study, 52.84% had work and 68.29s% were married. The Wilcoxon test performed showed a significant difference between the initial CD4 cell and the final CD4 cell with a p value of 0,000 ($p < 0.05$). This study showed an increase in CD4 cell count by an average of 129 cells / mm3 with a one-day use rule once a tablet in the form of fixed-dose combination (FDC)/ TDF (300) + 3TC (300) + EFV (600) meanwhile the combination ARV (d4T(30)+3TC(150)+EFV(200) increased CD4 75 cells/mm3. Conclusion of this study was the combination of ARV drugs TDF (300) + 3TC (300) + EFV (600) was the best among others for HIV patients.

Keywords: HIV/AIDS; CD4; ARV

IRCPAS/2020/PP-424

Evaluation of Phototherapy Related Complications in Pediatric Population with Hyperbilirubinaemia - A Pilot Study

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Neonatal jaundice is the yellow coloration of the skin and sclera of newborn babies that result from hyperbilirubinaemia.Recent global estimates suggest that over 1.1 million babies would develop severe hyperbilirubinaemia per each year and the vast majority reside in sub-Saharan Africa and South Asia. Many studies reporting the phototherapy related complications beside the benefits of phototherapy in treating the hyperbilirubinemia in pediatric patients. This study was also aimed to evaluate the phototherapy induced complications in pediatric population with hyperbilirubinaemia in one of the hospitals of Warangal. This is a pilot study conducted in secondary pediatric care hospital in Warangal of Telangana State, India. The study included 99 pediatric patients receiving Phototherapy for hyperbilirubinaemia. The study was conducted for a period of 14 months from November 2017 to January 2018. The recipients of phototherapy were observed for the effectiveness of phototherapy and phototherapy induced complications. Among the 99 pediatric patients with hypebilirbinaemia 62 were males and 37 were females. Phototherapy found to be effective in all recipients, with significant reduction in serum bilirubin levels. Thrombocytopenia was evident in 97 of 99 (97.98%) patients, Hypocalcaemia was seen in 11 of 99 (11.11%) patients and were statistically significant. 19 of 99 patients (19.19%) had elevated body temperature, 6 of 99 (6.06%) were dehydrated and 2 (2.02%) had convulsions. Through the findings of this study, thrombocytopenia was evident in 97 of 99 (97.98%) patients who received phototherapy. A study of Maj Sanjeev Khera et al. revealed thrombocytopenia in 26 (74%) cases during the first 24 hours of phototherapy. Yadav RK et al., Medhat FB et al. and Bahbah et al. observed hypocalcaemia in neonates received phototherapy. A study conducted by Maimburg et al. in Denmark, reported the risk of epilepsy after phototherapy. In this view, the research on Phototherapy complications has to be extended to validate the complications in neonates or children receiving phototherapy must be necessitated to mitigate and prevent the risk related to phototherapy. In-hospital phototherapy practices should prioritize establishing the measures to overcome the phototherapy related complications.

Key Words: Neonatal; phototherapy; hyperbilirubinemia; thrombocytopenia

IRCPAS/2020/PP-425

Assessment of Sleep Apnoea Risk and Quality of Sleep among General Public in Klang Valley

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Obstructive Sleep Apnoea (OSA) is classified as sleep-related breathing disorder that often results in sleep disturbance and poor quality of sleep.It is highly prevalent and often left under-diagnosed.Untreated OSA could lead to several health complications and increase risk of automotive accident. This study aimed to identify the level of risk of OSA, to assess the quality

of sleep among the public in Klang Valley, Malaysia and to find out the association, difference and correlation between them. A descriptive cross-sectional study was carried out and a total of 420 respondents were recruited through convenience sampling from the shopping malls in Klang Valley, Malaysia. Targeted participants with informed consent were invited to complete a pre-validated self-administered questionnaire containing socio-demographics, Berlin questionnaire to classify them into low or high risk of OSA and Pittsburgh Sleep Quality Index (PSQI) questionnaire to categorize them into good or poor sleepers. Mean age of the respondents was 38.52 (±14.19) and 40% of them aged 30 – 49. Majority of the study population were found to have low risk of OSA (81.7%) and were poor sleepers (65.5%). Risk of OSA was significantly associated with quality of sleep (p=0.011, contingency coefficient=0.123) as it was found that poor sleepers (21.8%) were at higher risk of OSA. OSA was significantly correlated with quality of sleep (Φ=0.124, p=0.011). Majority of the study population are at low risk of OSA, even though most of them are poor sleepers. However, high-risk OSA individuals are found to have poor sleep quality. Therefore, OSA may develop in poor sleepers over a period. Study findings will help healthcare providers and policy makers to educate and spread awareness about OSA among the public. This will be beneficial in early diagnosis and treatment of OSA before it complicates to other comorbidities.

Keywords: Obstructive sleep apnea (OSA); quality of sleep; Berlin questionnaire; PSQI questionnaire; Malaysia

IRCPAS/2020/PP-427

Causes of Hepatitis C and the Treatment Pattern and Relation with the Mortality Level Among Hemodialysis Patient in General Hospital, Tangerang, Indonesia

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The mortality of hemodialysis patients due to hepatitis c also continues to increase every year. Room separation between hemodialysis patients infected HCV and hemodialysis patients who are not infected by HCV should be done and routine hepatitis c examination and treatment should also be done regularly.To evaluate the cause of HVC in patients undergoing hemodialysis and the correlation with wortality rate based on the treatment used among these patients. Using a cohort method with prospective and retrospective studies with prospective patients who were undergoing hemodialysis for more than 3 month and retrospective data for patients who in the last 5 years. Hemodialysis patients affected by HCV were caused by the use of same room with HCV infected patients who get infected before starting hemodialysis or when undergoing hemodialysis. There were also many hemodialysis patients with HCV who did not receive treatment to treat hepatitis c, which resulted in patients dying because hepatitis c. The separation of the room and the separation of equipment between hemodialysis patients infected by HCV and not infected by HCV should be done for safety and avoiding the occurrence of HCV cross-infection, and hepatitis c treatment should be given as soon as the patient is known to be positively infected with HCV so as to prolong the patient's life survival.

Keyword: HCV; Indonesia; mortality; separating room

IRCPAS/2020/PP-428

Perception and Knowledge of Herbal Medicine Among Students and Staffs Of KPJUC

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Malaysia is a multiracial country which has different races including Malay, Chinese, Indian and other ethnic groups. Each culture also has their own tradition ways to cure diseases, relieve unpleasant symptom and promote health.Herbal medicine is a blooming healthcare industry and comes with variety of dosage forms and correspondingly, the usage of herbal products is increasing tremendously in Malaysia. A survey was conducted to assess the perception and knowledge of Herbal medicine among students and staffs in KPJUC. The specific objective was to evaluate their knowledge towards the effectiveness and harmful effects of herbal medicine.A cross sectional study was conducted for a total respondent of 400.The convenience sampling was done using a'five Likert Scale' questionnaire. Cronbach Alpha was found to be 0.799. Descriptive analysis and Pearson correlation was done in using SPSS version 20. Most respondents involved in this study were females (n = 346, 86.5%). The usage of herbs for medicinal purpose was reported 65.3% (n = 261). Parent (n = 108, 27.0%) was the major influencer in taking herbs. Lack of scientific evidence and contraindication with other modern medicine were perceived as the main barriers for herbal medicine. The respondents believed that Aloe Vera (45.2%) and Ginger (43.1%) were more effective compare to other herbs that commonly used in Malaysia. About 57.5% of respondents strongly agreed that patients should inform their allopathy doctor about their use of herbal medicine. Race (p value = 0.000) and department (p value = 0.000) have statistically significant relationship with the perception and knowledge of herbal medicine among students and staffs in KPJUC. Students and staffs in KPJUC have positive approach towards herbal medicine and their knowledge is adequate in general. The students and staffs acknowledge the need to be well educated about herbal medicine.

Keywords: Herbal medicine; perception; knowledge; students and staffs

IRCPAS/2020/PP-429

The Influence of Pharmacist Intervention on Health-Related Quality of Life of Diabetic Patients and its Relationshipwith PatientDemographics on EQ-5D Domainsand VAS Score

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Diabetes mellitus is a chronic disease and if it is not treated properly it can cause various complications in patients. To improve the quality of life of the patients with diabetes mellitus needs the appropriate interventions from health care providers. The aim of this study was to measure the effect of pharmacist intervention on patient reported quality of life on all domains of EQ-5D.This randomized controlled study is carried out in different hospitals of Malaysia to evaluate the impact of a pharmacist intervention on patient reported quality of life when there HbA1c is controlled by pharmacist interventions. A generic HRQoL tool EQ-5D was used to report the data.To identifythe problems in individual EQ-5D domains and (VAS) mean scores ANCOVA and Logistic regression was used. The average HbA1c values decreased from

10.15±1.42 to 8.21±0.75 in the intervention group ($p<0.05$). The intervention group showed an improvement in the quality of life domains. Mobility domain decrease from 3.21±0.25 to 2.21±0.45($p<0.013$), Self-care domain decrease from 4.11±0.75 to 3.291±0.85 ($p<0.045$), Usual activity domain decrease from 2.81±0.65 to 2.29±0.85 ($p<0.655$), Pain/ Discomfort domain decrease from 3.19±0.34 to 2.91±0.65 ($p<0.065$), Anxiety/ Depression domain decrease from 3.51±0.25 to 2.21±0.45 ($p<0.015$) and the VAS mean score is improved from 55.59±8 to 65.62±9 ($p<0.005$). Patientsfeatures were significantly related with the HRQoL in type 2 diabetes. The significant and positive improvement was seen among the different domains of EQ-5D with the intervention of pharmacist. With the intervention of pharmacist not only the HbA1c improves but also the all health-related domains also improved in patients.

Keywords: Health-related quality of life; EQ-5D; diabetes mellitus;domains of EQ-5D

IRCPAS/2020/PP-431

Correlation Between Socio-Demographic and Mortality Rate Among Hemodialysis Patients with Hepatitis C at Army Hospital, Jakarta, Indonesia

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Hemodialysis is one of the treatments in replacing kidney function that must be carried out continuously to patients. Therefore, infection is one of the risk factors that may occur among these patients. Hepatitis C (HCV) is a common viral infection. Patients contracting viral infections before or after hemodialysis treatment. The infection may come from a family, from a blood transfusion or ongoing hemodialysis treatment, which is considered a high-risk factor for this infection. To evaluate correlation between socio-demographic with mortality rate among hemodialysis patients with hepatitis C. A prospective and retrospective cohort study was conducted in this study. A prospective cohort study was conducted for 3 months in patients undergoing hemodialysis, whereas a retrospective cohort study by analyzed medical records of hemodialysis patients who died with HCV infection during the past 5 years who were included in the inclusion criteria. Hepatitis C mostly occurred among male and hypertensive patients between 51-60 years old. There is a significant correlation between age and type of infection for all hemodialysis hepatitis patients. Mortality rate among hemodialysis patients with hepatitis C was influenced by practice pattern such us duration of HD and mixing room between Hypertensive with HCV and without HCV

Keywords: Hemodialysis; hepatitis C; socio-demographic; mortality; Indonesia

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Mortality Rate and Survival Analysis Among Hemodialized Patients with Hepatitis C at Cempaka Putih Islamic Hospital, Jakarta, Indonesia

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Hemodialysis (HD) is an invasive action that has a major risk associated with viral infections that can be transmitted through blood, such as hepatitis B, hepatitis C (HVC), Human

Immunodeficiency Virus (HIV), and Human Lymphotropic Virus. The length of time a patient undergoes hemodialysis is a factor related to the prevalence of HVC, this may be related to an increased risk of exposure to HVC virus in the hospital environment such as blood transfusion, repeated vascular access, and hygiene. To evaluate mortality rate and survival analysis among hemodialized patients with hepatitis C. A cohort prospective and retrospective study was done in this research. This study included hemodialysis patients who were followed up for more than 3 months and who died in the last 5 years. Universal sampling was used to select patients based on inclusion criteria. There was a significant correlation between hepatitis C and hypertensive patients and diabetes mellitus (DM) patients. DM in HD patients continue to be very high levels of mortality in hospital.Probability of dying among diabetic and/or hypertensive patients who undergone hemodialysis between both HD centres while the significant relationship is showed in this hospital. Hypertensive and diabetic were one of the risk factors which caused mortality among HD patients. The persentage of survival among hypertensive patients in hemodialysis was lesser than diabetic patients in hemodialysis.

Keywords: Hemodialysis; hypertensive; diabetic; mortality rate; survival analysis; hepatitis C; Indonesia

IRCPAS/2020/PP-433

Evaluation of Antibiotic for Pneumonia in Burns Patients with Nosocomial Pneumonia in RSUD Dr. Soetomo in 2017-2019

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Nosocomial pneumonia is an infection that is often found in burn patients with high levels of morbidity and mortality. Inappropriate use of antibiotics leads to increase risk of side effects, costs, and antibiotics resistance. The main therapy for nosocomial pneumonia is the administration of antibiotics. The purpose of this study was to evaluate the use of antibiotics for pneumonia in burn patients with nosocomial pneumonia qualitatively by the Gyssen method and quantitatively by a defined daily dose (DDD). Data was collected retrospectively by medical records of burn patients with nosocomial pneumonia at RSUD Dr. Soetomo in 2017-2019. The demographic data, laboratory results, antibiotics use, dosage, frequency and days of antibiotics are observed in this study. Seven burn patients with nosocomial pneumonia were obtained in this study. Six patients were classified as hospital acquired pneumonia (HAP), and 1 patient included ventilator associated pneumonia (VAP). The most antibiotics for nosocomial pneumonia are meropenem (34%), levofloxacin and amikacin each by 17%. There were 3 patients (42.9%) included in the IVa category (more effective alternatives), 1 patient (14.2%) included in the IVb category (less toxic alternative), 1 patient (14.2%) included in the IVd category (more spectrum narrow), 1 patient (14.2%) included in category IIa (incorrect dosage) and 1 patient (14.2%) did not belong to category I-IV. The total DDD / 100 patients-days score was 161.09. DDD / 100 antibiotic patients-days in this study are levofloxacin (67.50), amikacin (42.59), meropenem (39.82), and cefoperazon (11.18). The administration of antibiotics to burn patients with nosocomial pneumonia is classified as irrational (85.5%) and 14.5% as rational category. In addition, the selection of antibiotics is still not selective with the high consumption of antibiotics.

Keywords: Burns; nosocomial pneumonia; antibiotics; Gyssen;defined daily dose

IRCPAS/2020/PP-436

Diabetes Knowledge and Medication Adherence Among Patients Receiving Diabetes Management Service in Indonesia

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Diabetes is a chronic disease that cannot be cured but can be managed by controlling the patient's blood sugar, one of which is adhering to antidiabetic drugs. Some factors that can affect medication adherence are the patient's knowledge and characteristics. To evaluate the relationship between diabetes knowledge and patients' characteristics with medication adherence in patients with type II diabetes attending the diabetes management program in Banyumas Regency, Indonesia. A cross-sectional study was conducted with a convenience sample of 210 diabetic patients attending the program in eight community health centers. We used the Diabetes Knowledge Questionnaire (DKQ) to assess diabetes knowledge and the Medication Compliance Questionnaire (MCQ) to assess medication adherence. Patients' characteristics, knowledge and adherence were analyzed descriptively, while the relationship was analyzed using Chi-square. Two hundred and ten patients were analyzed, with a mean age of 60.25 years (SD = 9.38), 77.1% were female, 56.7% graduated from elementary school, and a mean duration of diabetes of 5.44 years (SD = 4.37). Approximately 77.1% of the sample had moderate knowledge with a total mean score of 12.57 (SD = 3.859), while 58.6% were considered adherent with a total mean score of 26.04 (SD = 2.512). Medication adherence associated significantly with diabetes knowledge ($p = 0.049$) and duration of diabetes ($p = 0.034$). Patients' knowledge and duration of diabetes are considered as important factors on medication adherence. Healthcare professionals in the disease management program should beware of this information that patients hold about adherence to medicine regarding diabetes.

Keywords: Diabetes; knowledge; adherence; chronic disease management; Indonesia

IRCPAS/2020/PP-438

Applying a New Indicator Based on the ATC/DDD System for Evaluating the Appropriateness of Clinical Application: Coxibs and Traditional Nonsteroidal Anti-Inflammatory Drugs for Postoperative Orthopaedics Pain Control.

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A recent drug utilization study on antibiotics established a new indicator for evaluating appropriateness of clinical application. The calculation is based on the World Health Organization (WHO) anatomical therapeutic chemical classification system with defined daily doses (ATC/DDD) system. It combines two main indicators for drug i.e. dose and duration of treatment. The aim of current study was to describe the appropriateness of clinical application of coxibs and traditional nonsteroidal anti-inflammatory drugs (tNSAIDs) that indicated for postoperative orthopaedics pain control.A drug utilization review of coxibs and tNSAIDs of an

inpatient orthopaedics department of a private teaching hospital in the city of Seremban, Malaysia was conducted. The NSAIDs were classified and calculated according to the WHO ATC/DDD system. The formula for measuring the appropriateness of clinical application is by calculating the ratio of use density to use rate (UD/UR). A total of 195 patient records who received NSAIDs were randomly selected among 1169 cases. The tNSAIDs were dexketoprofen injection, diclofenac sodium tablet; and the coxibs were celecoxib capsule, etoricoxib tablet and parecoxib injection. The UD/UR for all NSAIDs were less than 100 except for etoricoxib tablet (105.75) and parecoxib injection (108.00). The UD/UR value more than 100, indicating a high possibility of irrational medication and may require of enhanced monitoring.The findings of this DUR study may suggest of applying new indicator for evaluating the appropriateness of clinical application of NSAIDs for postoperative orthopaedics pain control.

Keywords: Coxibs; NSAIDs; orthopedics

IRCPAS/2020/PP-441

Knowledge and Attitude on Sexually Transmitted Diseases (STD) Among KPJUC Students

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Sexually Transmitted Diseases (STDs) are one of the commonest reasons for illness amongst youngsters around the world. It ranges from the least dangerous to the deadliest diseases such as Human Immunodeficiency Virus/Acquired Immunodeficiency Syndrome (HIV/AIDs) and hepatitis B. Fortunately, STDs are preventable and easily avoided with adequate awareness and education to the public. This study aimed to evaluate the students' knowledge and attitude towards STDs. In this cross-sectional study, a validated questionnaire consisting of 20 questions were distributed to 173 students of various programs using simple random sampling method. Out of 173 students, 76.9% were males and 50.9% of them were of Malay ethnicity. 75.7% were pharmacy students. When asked to identify several diseases under STDs, higher proportion of males and females managed to identify HIV/AIDs as an example of STDs (Males: 100%, Females: 95.5%). A high proportion of males and females stated that condoms can prevent STDs (Males: 94.7%, Females: 79.9%). From the aspect of signs and symptoms for STDs, there were significant difference between the genders in identifying swelling in genital area, failure to urinate, genital ulcers and loss of weight. Significant differences exist between genders in terms of identifying complications of STDs such as still birth, miscarriage, and ectopic pregnancy ($p=0.004$, $p=0.027$, $p=0.029$). The findings of this study are congruent to a similar study conducted in Vietnam by Lan, 2009 where students had misconceptions on the causes of STDs. One study conducted by Vardguiden 2011 discovered that a higher proportion of females (71.1%) compared to males stated that condom usage can protect from STDs. Apart from that, both genders seem to agree that young people must have knowledge and must be educated about STDs to prevent STDs. The lack of awareness on STDs amongst university students is alarming and needs immediate attention. Proper sexual education should be provided toinstil such awareness.

Keywords: sexually transmitted diseases; knowledge; awareness

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A Systematic Review of Cost-Effectiveness of Medication Adherence-Enhancing Intervention for Asthma Patients

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Asthma is a noncommunicable disease that had affected three hundred million people worldwide and medication nonadherence leads to many negative health complications and a high economic burden on society. This systematic review aimed to evaluate the evidence on the cost-effectiveness of medication adherence-enhancing intervention, as opposed to usual care or placebo. Search engines such as PubMed, Scopus and EBSCOhost were used to locate all possible studies from the inception of the search engines to 19 October 2018. Drummond checklist was used to appraise the quality of economic evaluation. Data including study characteristics, quality assessment, health outcomes and costs of intervention were narratively summarized. The primary measure is cost-effectiveness (CE) outcome and the secondary outcomes are costs, medication adherence and clinical consequences. A sum of 20 studies was included, where eleven studies were RCTs, six studies were based on comparative studies and three studies adopted Markov models. Fifteen studies evaluated an educational intervention, with 13 of them were cost-effective in improving the health outcomes. An internet-based intervention showed similar CE outcomes between treatment groups. All studies involving a medication regimen simplification and combination of a technology-assisted program and a training lesson had demonstrated the desirable CE outcome. The quality of most studies was fair with four studies showed a high-quality standard.Fundamentally, the medication adherence-enhancing interventions were cost-effectively showing an increase in medication adherence and positive clinical effectiveness while reducing asthma-associated costs. However, limitations associated with poor methodological conduct must be properly addressed. Further economic evaluations with sound methodological conduct should be encouraged for stronger shreds of evidence in determining the best intervention to improve medication adherence.

Keywords: Pharmacoeconomics; cost-effectiveness analysis; medication adherence; patient compliance; asthma; systematic review

IRCPAS/2020/PP-443

Patient knowledge and safety profile in self-medication practice using nonsteroidal anti-inflammatory drugs (NSAIDs)

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Self-medication is a practice of the choice and use of medicines that have not been prescribed by a licensed health care professional for treatment of self-recognized illness or symptom. NSAIDs have a long history of safe and effective use and commonly use in self-medication. In many cases, drug related problems (DRPs) may occur because of side effects of the drug, drug-drug interactions (DDIs) and drug-disease interactions with the concomitant condition of the patient. The purpose of this study was to assess patient's knowledge on NSAIDs and to identify causes for concern of NSAIDs used. This study was conducted in 8 community pharmacies at Ipoh, Malaysia that was selected conveniently. The questionnaires were distributed to suitable

respondents and using purposive sampling method. The descriptive data analysis was implemented. The causes for concern of NSAIDs used were assessed by using the relevant references. A total respondent was 123 patients consist of 67.5% male and 32.5 % female; aged 45-60 years old. The most common used of NSAIDs was ibuprofen (33.3%) followed by diclofenac sodium (26.8%) and mefenamic acid (15.4%). The indication was mostly for joint pain (61.8%) followed by toothache (15.4%) and headache (13.8). The result showed that most of the patient (42.3%) have a poor knowledge towards NSAIDs. Moreover, 50.40% of respondent have a risk of having serious implications on their safety due to DDIs, drug-supplement interactions, and drug-disease interactions with NSAIDs. The risk of DRPs is occurred in self-medication practice with NSAIDs among middle age patients. In certain conditions, it might cause serious implications on patient health condition. Pharmacist as a healthcare provider in community setting is encouraged to assess patient history and prevent DRPs in self-medication practice. Moreover, patient education and drug information also should always be applied in pharmaceutical care practice.

Keywords: Self-medication; non-steroidal anti-inflammatory; NSAIDs; safety profile

IRCPAS/2020/PP-446

Assessing the Treatment Outcome of Sri Lankan Traditional Medicine for Nonunion of Tibia Fractures (A Case Series)

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Background: Sri Lankan Kadum Bidum Vedakama (Sri lankan traditional orthopedic medicine) is a convincing and widely sought alternative medical system in Sri Lanka for more than 4000 years. In this medical system bone fractures, bone dislocations and muscle injuries are treated and healed, successfully. Objectives: Objective of this study was to evaluate the efficacy of Sri Lankan traditional medicine for nonunion fractures after failure with allopathic medicine treatments. Methods: Five cases were selected for this study. The x-ray evidences, sign and symptoms, motor function, sensory function and quality of life were used as the parameters to measure the clinical outcome of the treatments. For the statistical analysis Wilcocxon signed rank test and SAS 9.1 software was used for this study. All cases recruited for this study had been treated by allopathic medicine for more than 6 months and diagnosis was made by the evidence of x-ray. According to Sri Lankan traditional medicine, in first three months all the patients were treated with Seethodaka oil and the next six months treated with Seethodaka oil with Pinda oil application. In first three months all the patients immobilized with bamboo splints and splints put lateral side of the foot and medial side of the foot ankle joint to shaft of the femur. In other period splints were put ankle joint to knee joint. Results: All the patients were completely recovered, and mean valve of the time period was 7 months. Evidence of the x-rays showed the callus formation in the fracture sites of all the patients. Sign and symptoms, motor function, sensory function and quality of life were assessed within two months after starting treatments and all the parameters showed significant ($p<0.05$) improvement. Discussion and Conclusions: According to these results we conclude that procedures and drugs recommended by the Sri Lankan traditional medicine and Ayurveda can be successfully used to treat nonhealing fractures resulted from previously unsuccessful treatment approaches.

Keywords: Sri Lankan traditional medicine; allopathic medicine failure; Nonunion tibia fracture

IRCPAS/2020/PP-448

Drug Prescribing Pattern Among the Patients Visited to the Dental Clinic of a Private Medical University in Kedah State, Malaysia- A Retrospective Study

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The present study was carried out to evaluate the drug prescribing pattern among patients visited to the dental clinic of Faculty of dentistry, AIMST University. A retrospective, cross-sectional study was conducted to analyse the drug prescribing pattern of dentists of AIMST University, Kedah, Malaysia. A total of 875 patients' medical records were scrutinized in stipulated time capsule frame (October 2018 to December 2018) and only 300 patients' medical records were identified to have drug prescriptions. Data pertaining to drug prescribing pattern were collected using pre-designed data collection form. Patient's demographic characteristics and drug prescription data were collected from patient's medical records and drug prescription book. The data were entered into Microsoft Spreadsheet and descriptive statistics were used for quantitative data analysis. Three hundred (n=300) patients' medical records and drug prescription books were analysed. Female patients were greater in number [177(59%)]. The most common dental problem among the patients were gingivitis [145(22.30%)] and dental carries [129(19.85%)] followed by periodontitis [122(18.77%)] and edentulous [101(15.54%)]. A total of 525 drugs were prescribed with an average number of 1.75 drugs per prescription. The most common therapeutic category of drug prescribed was NSAIDs [233(44.38%)] followed by antibiotics [167(31.8%)]. The percentage of encounters with an antibiotic prescription was higher (46.7%) than the WHO standard value (20-27%). Slightly more than half of the drugs [304(58%)] were prescribed with generic name. Majority of the drugs (80.57%) prescribed from essential drug list of Malaysia.Study revealed moderate rational drug prescribing as per the WHO drug prescribing indicators among the patients visited to the dental clinic of AIMST University. However, the appropriate measures can be taken to avoid over prescription of antibiotics and its resistance.

Keywords: Dental;antibiotics;essential drug list; WHO prescribing indicators; NSAIDs; prescribing pattern

IRCPAS/2020/PP-454

Quality of Life of Diabetes Mellitus Type 2 Patients Using the SF-36Questionnaire in Penang, Malaysia.

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Quality of life was an individual's perception of his position to reduce in life, in the context of culture, the value system of its relationship to life goals, expectations, standards, and other related. The purpose of this study was to determine the quality of life related to the health of diabetes mellitus type 2 patients in Penang, Malaysia. This type of research was a prospective study with a cross-sectional approach. Respondents in this study were residents aged ≥18 years. Data analysis was performed with the Kolmogorov-Smirnov normality test, followed by an unpaired t-test for each of the eight domains of life quality SF-36 with health and socioeconomic risk factors. The results of the study were several which showed low results in the Physical Function domain (855.0 ± 157.872), Vitality (301.20 ± 51.494), Mental Health (390.40 ± 57.034), Pain (175.60 ± 29.496) and General Health (372.00 ± 63.011). There are 5 domains of 8 domains which are below the standard value.

Keyword: Quality of Life; SF-36;prospective; cross sectional; diabetes

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Evaluation of the Quality of Life of Hypertensive Patients Using the SF-36 Questionnaire in Penang, Malaysia

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Quality of life was an individual's perception of his position to reduce in life, in the context of culture, the value system of its relationship to life goals, expectations, standards, and other related. The purpose of this study was to determine the quality of life related to the health of hypertensive patients in Penang, Malaysia. This type of research was a prospective study with a cross-sectional approach. Respondents in this study were residents aged ≥18 years. Data analysis was performed with the Kolmogorov-Smirnov normality test, followed by an unpaired t-test for each of the eight domains of life quality SF-36 with health and socioeconomic risk factors. The results of the study were several which showed low results in the Physical Function domain (68.89 ± 29.328), Role of Emotion (54.47 ± 9.387), Mental Health (47.22 ± 48.071), Social Function (44.44 ± 48.222) and General Health (43.69 ± 20.124). There are 5 domains of 8 domains which are below the standard value.

Keyword: Quality of Life; SF-36; prospective; cross sectional; hypertension

IRCPAS/2020/PP-458

Evaluation of Self-Medication Practice among UCSI University Students

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Self-medication is defined as the use of any medication for self-treatment without consulting a healthcare professional. Everyday people throughout the world act on their own for their health through the practice of self-care. Objectives: To determine which groups of drugs were most frequently used by students as self-medication and assess the practice and views of self-medication among UCSI University's students. Methodology: Cross-sectional questionnaire-based study. Major findings: 239 (65.1%) of the respondents practiced self-medication in the past one year, among which 101(42.3%) were males and 138 (57.7%) were females. Pharmacy (74.4%) was the main source of self-medication. The most common indication for self-medication was fever (72.8%), followed by cough (67.6%), headache (67.0%), common cold (65.7%) and pain (30.5%). The most common drug classes for self-medication were antipyretics (59.7%) followed by cough syrups (59.1%), vitamins (55.3%), analgesics/anti-inflammatory (45.8%) and cold preparations (36.0%). 279 (76.0%) of the respondents agreed that all medications including herbals have adverse effects. 354 (96.5%) was aware that increasing drug dose can be dangerous, however, only 153 (41.7%) aware that decreasing drug dose can be dangerous. Majority of the respondents agreed that "Concomitant use of drugs can be dangerous"; "Physician help must be sought in case of adverse effects" and "Using medications with unknown substances in patients with liver and kidney disease is dangerous". Discussion and Conclusion: The practice of self-medication was common among UCSI University students. It was indicated in mild disease condition such as fever, cough, headache and etc. Appropriate use of drugs as self-medication was beneficial to human beings. However, there are many factors that rendered self-medication as the main cause of drug misuse.

Key Words: self-medication; UCSI University; students

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Probiotic Characteristics of Lactic Acid Bacteria Fermented from Food Origin

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Probiotics have become an interest regarding their broad benefits for human health. The most used probiotic strains are the lactic acid bacteria, gram-positive microbes that have been used for centuries in the food production process. The search for probiotic properties of lactic acid bacteria from food is a trend seen in food microbiology research. Microorganisms isolated from foods show better viability in the food environment and guarantee more attractive sensory characteristics in comparison with microorganisms originating from intestines. Some important requirements that must be met for microorganisms to be used as food supplements are safe for humans and arrive in the gut with sufficient living conditions so that able to multiply and temporarily colonize in the intestine.This study aims to examine the characteristics of *Lactobacillusacidophilus* fermented from juice and *Lactobacillusreuteri* from fermented cow's milk as probiotics. In vitro study was used to tested the ability of *Lactobacillusacidophilus* and *Lactobacillusreuteri* as probiotic candidates to survive at pH 2; 2.5; 3.2; and 7.2, resistance to

bile salts and resistance to pathogenic bacteria (*E. coli, S. aureus and E. faecalis*). Probiotics are well known to have broad health benefits for human health, including antimicrobial, anti-inflammatory and immune system modulation. Therefore, it is important to explore various sources including food, to find lactic acid bacteria as a probiotic candidate, for the development of their use. The results showed that the isolates of *Lactobacillusacidophilus* and *Lactobacillusreuteri* were able to survive in their host environment. This isolate also has health benefits, one of which is its antimicrobial properties against pathogenic bacteria in the digestive tract. *Lactobacillusacidophilus* fermented from fruit juices and *Lactobacillusreuteri* from fermented cow's milk showed characteristics as probiotic candidates.

Keywords:*Lactobacillusacidophilus*;*Lactobacillusreuteri*; In Vitro; pH; Oxgall; pathogen

IRCPAS/2020/PP-505

Impact Resistance, Tensile Strength and Water Absorption Properties of Bambara Nut Shell Powder and Eggshell Powder (BNSP/ESP) Filled HDPE Hybrid Composite

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Over the past years, bio-fillers are used in the production of biodegradable polymer composites that can replace the non-biodegradable synthetic polymers that poses serious threat to the environment. In this work, Bambara nut shell powder (BNSP) and eggshell powder (ESP) were incorporated to reinforce HDPE matrix resulting in the formation of a hybrid composite. HDPE was compounded with Bambara nut shell powder and egg shell powder using a two roll mill at different filler loading to obtain the hybrid composites. The tensile strength, the impact strength and water absorption properties of the neat HDPE and that of the HDPE/BNSP/ESP hybrid composite were then investigated. It was observed that the tensile strength decreases throughout with increase in filler content while the impact strength increases with increase in filler loading. Water absorption initially increases with increase in filler loading, then decreases and finally increases until the maximum was attained. The two bio-fillers were successfully used as reinforcement for the HDPE and offers new opportunity in the production of new materials for indoor applications.

Keywords: HDPE; Bambara nut shell powder; eggshell powder; bio-filler; hybrid composite

IRCPAS/2020/PP-506

Haematological Changes in *Eimeria tenella* Infected Broiler Chickens Treated with Kaempferol

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Effect of kaempferol on haematological parameters in two weeks old broiler chickens with experimental *Eimeria tenella* infection was evaluated in this study. Sixty-day old broilers were

randomly allotted into six groups (I-VI) of ten broilers each and brooded for two weeks with commercial broiler feed (vital feed®) and provided water *ad libitum*. At two weeks of age, broilers in group I served as non-infected control. Broilers in groups II- VI were infected with *Eimeria tenella* sporulated oocyst (10^4/ml) via oral inoculation. After infection was established, broilers in groups II-IV were treated orally with 1, 1.5 and 2 mg/kg of kaempferol, respectively. Broilers in group V were treated for five days with amprolium, 1.25 g/Lin drinking water. Broilers in group VI served as infected untreated control. Five days post infection, all broilers were sacrificed by severing their jugular veins. Blood sample from each bird was collected in EDTA container for haematology. Caeca and its contents were harvested and used to determine the lesions score and caecal oocyst count respectively. Data obtained was analyzed using pad prism version 5.0. Mean values of Packed Cell Volume (PCV), haemoglobin (Hb) concentration and Red Blood Cell (RBC) count were significantly ($P<0.05$) increased in groups II, III and IV in a dose dependent manner. Similarly, mean values PCV, Hb concentration and RBC count were significantly ($P<0.05$) increased in groups II, III, and IV when compared to group VI. No significant ($P>0.05$) difference in the mean values of PCV, Hb and RBC count was recorded between groups treated with kaempferol and group V. Caecal oocyst count and lesions score reduced significantly ($P<0.05$) in groups II, III, and IV in a dose dependent manner. It was therefore observed in this study that kaempferol improved haematological parameters and reduced oocyst count as well as the lesion scores in broilers infected with *Eimeria tenella.*

Keywords:Broilers; *Eimeria tenella*; Kaempferol;lesion; scores;oocyst count

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Dengue Virus Entry/Fusion Inhibition by Small Bioactive Molecules

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Dengue virus enters the cell by receptor mediated endocytosis followed by viral E (envelope) protein mediated membrane fusion. Membrane fusion is a central molecular event during viral entry into host cell. E protein is a major component of the virion surface plays an important role in binding to the host receptor and assists virus fusion. Rearrangement and or conformational changes in the hinge region by small molecules may interrupt the fusion process.Among the three domains present in the E protein, Hinge region movement of domain I and II, facilitates the fusion process. Upon lowering the PH, the E protein undergoes major conformational changes in the Hinge Region springing upwards to bring the fusion peptide closer to the host membrane for fusion to occur.A small detergent molecule n-octyl-β-D–glucoside (βOG) occupiesthe hydrophobic pocket which is in the hinge region plays a major role in the rearrangement. It has been clearly reported that mutations within this binding pocket leads to the alterations of pH threshold for fusion. In addition to this the protonation of histidine residues present in the hydrophobic domain would also impart the conformational change of the E protein. The previously reported fusion inhibitors such as peptidic antivirals suffer from poor absorption from the gastrointestinal tract, necessitating intravenous delivery and high manufacturing costs.Keeping these views, it is proposed to design and synthesize a library of novel small bioactive molecules. Inserted at this position may have the ability to interrupt further conformational changes and hence can inhibit the fusion transition.

Keywords: Dengue; Flavivirus; envelop protein; Hinge, βOG, fusion

IRCPAS/2020/PP-508

Predictors of Central Macular Thickness in Diabetic Macular Oedema Patients on Intravitreal Ranibizumab

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Intravitreal Ranibizumab (IVR) is an anti-vascular endothelial growth factor (anti-VEGF) has become the preferred treatment option to improve the vision of diabetic macular oedema (DMO) patients by reducing the fluid build-up particularly in the centre of the macula. Numerous studies have been done to evaluate the intensive therapy with IVR but few evaluate the short-term effects of IVR. Moreover, there were inconsistent and inconclusive findings of the associated factors that may influence the treatment effect in term of changes of the central macular thickness (CMT).The objective of this study was to identify associated factors of changes of the CMT after three injections of IVR in DMO patients.This was a cross-sectional study involved retrospective record review of the 153 DMO patients who received completed three-month treatment of IVR in Hospital Universiti Sains Malaysia from 2016 to 2019. Changes of CMT was calculated based on the difference between baseline CMT and after three months of CMT. The association factors include socio-demographic, comorbidities and ocular factors. Linear regression was applied to analyse the association between associated factors and changes of CMT using STATA SE 14 software.This study included 69 (45.1%) male and 84 (54.9%) female patients with a median (interquartile range (IQR)) age of 59 (11) years and median (IQR) of diabetes duration of 11 (9) years. The median (IQR) of changes of CMT was 123 (191) μm. Factors significantly associated with changes of CMT were baseline CMT [b =0.73; 95% confidence interval (CI): 0.63,0.84; p=<0.001] and presence of subretinal fluid [b= 35.43;95% CI:3.70,67.16; p=0.029]. These factors only explained 58.3% of the variation in changes of CMT. The changes of CMT at month 3 was slightly lower compared with long-term studies of IVR. Patients who presented with subretinal fluid in the eyes and high baseline CMT had greater changes of CMT after receiving three injections of IVR treatments. Consequently, additional injections of IVR need to be considered in patients especially with these factors for better treatment response.

Keywords: Diabetic macular oedema; anti-VEGF; Intravitreal Ranibizumab; central macular thickness

IRCPAS/2020/PP-509

Phytochemical Investigation, Cytotoxicity and Anti-Diabetic Activity of Whole Fresh and Dry Ethanolic Extracts of Sudanese *Portulaca Quadrifida* L

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Natural products have been used since ancient times and in folk medicine for the treatment of many diseases and illnesses. *Portulaca quadrifida* popularly called Chicken weed is an annual mat-forming, succulent species in the family Portulacaceae. It is extending over drought and low fertile soil. *P. quadrifida* distribution is not clearly defined, its concede native to Africa and Western Asia. It is useful traditionally in urinary discharges, inflammations, asthma, cough, and ulcers. In this study, the phytochemical, Cytotoxicity and anti-diabetic activity of whole fresh and dry ethanolic extracts of Sudanese *P. quadrifida* L were investigated.*P. quadrifida* were collected from wild filed in Khartoum, Sudan. Samples were divided into two groups, dry and fresh. Cytotoxicity, anti-diabetic activity, and tannin, ascorbic acid content of the ethanolic extracts of each group were investigated.*P. quadrifida* show high content of tannin 28.07 – 13.68 ppm for fresh and dry sample respectively. Both extracts showed comparable results of vitamin C content (1.16 – 1.76 ppm for fresh and dry sample respectively). In the cytotoxicity test (MTT) assay, the IC50 values were 858 and 155.3092 ppm for dry and fresh *P. quadrifida* sample extracts respectively. The Alpha-amylase inhibition % of both extracts showed high activity with inhibition percentage 95 and 91 % for dry and fresh extracts respectively.Comparing the results from the two extracts, the dry and fresh *P. quadrifida* shows comparable values of vitamin C content and anti-diabetic activity. The dry extract showed a distinct higher value of MTT test, thus lesser cytotoxicity. The fresh extract showed measurable higher tannin content compared to the dry extract. The results of this study augment and improve the importance of the use of natural plant and natural product as a source of medicinal agent

Keywords: *Portulaca quadrifida; c*hicken weed; anti-diabetic activity; cytotoxicity

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Some Aspects of Radiation Safety due to Electromagnetic Field in Normal Human Life

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The exposure to the electromagnetic fields is well known phenomenon. During the 20th century, environmental exposure to man-made electromagnetic fields has been gradually increasing as growing electricity demand. Every advance technology is posing the challenges in social behavior and created more artificial sources of exposure to the electromagnetic fields now a day. People are exposed to a complex mix of weak electric and magnetic fields, at home and at workplacesbyvarious ways, like as generation and transmission of electricity, domestic

appliances, and industrial equipment, to telecommunications and broadcasting. In general, electromagnetic radiations generated as the byproduct of electricity running in electronic devices.An attempt has been made for the study of some valuable bio-effect of electromagnetic radiations with an application of Phantom model.An approach of specific absorption rate (SAR) distribution has been applied in this model. The present results have been complied with the European and USA standards (EN 50361, IEEE 1528) due to an assessment of dosimetric. A gripping device, DUT (Device Under Test) and software Open SAR were used to measure the transmitted radiofrequency (R. F.) or energy in general absorbed by human tissues. The SAR is a function of the electrical conductivity (σ), the induced electric field from the radiated energy (measured in Volt/meter) and the mass density of human tissue.Due to the high sensitivity of the probe, its output voltage is measured without amplification. The electric field probe corresponds to the recommendations of CENELEC (European Committee for the Coordination of Electrical Standards) and ICEEE (the International Committee of Electrical and Electronic Engineers) for measuring electromagnetic fields of cell phones, base stations, and various radiating devices. The DUT handle is constructed of a material with low energy loss and low permittivity. It allows moving axes (X, Y, Z) in all three dimensions (3D) or rotation around the phantom ear for precise positioning of device DUT.The calculated values of specific absorption rate (SAR) distribution were found approximate in order of (15±0.05) grams over per unit grams of human tissue. These results were found within good agreement with others.

Keywords: Health hazard; algorithm calculations; simulations; Specific Absorption Rate (SAR) distribution; bio-effect; Dosimetric elevations; WHO

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Assessment of the Toxicity of Herbal Preparations Which is Commonly Used to Treat Diabetic Mellitus, in a Mice Model

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Diabetes mellitus exerts significant amount of socio-economic burden across the world. In mainstream medical practice there are no drugs or treatment procedures for a complete cure for diabetes. Therefore, assessment of herbal preparations in alternative medicine is vital in controlling diabetes mellitus. However, safety and efficacy of alternative herbal preparations are required to be ensured. The objective of this study was to identify acute and chronic toxicity effects of the commonly used three herbal formulas by Sri Lankan Ayurveda practitioners to treat Diabetes mellitus. The herbal preparation of *Tripala katakana pata* decoction, *Madumehari powder* and *DT6* powder were assessed for the organ toxicity of mice. The herbal Medicines were given orally to 16 mice each in control and treatment groups. Double dosage of human normal dosage (Normal human dose 2 ml/1kg; 1 time per day) was given to the mice for 90 days and assessed the behavioral patterns (movements, eating and sleeping patterns) and body weight of mice in both control and treatment groups. The biochemical parameters of serum creatinine, SGOT, SGPT and Gamma-GT were assessed over the course of the treatment in control and treated groups. Finally, a histopathological study of liver, kidney, heart and

spleen was conducted. There were no significant different in behavioral patterns throughout the 90 days observation period. The body weight of mice in control and treatment groups did not significantly changed (mean values: Before the treatment 30.5 ± 0.5 g; after 90 days of treatment 35.7±0.6 g) The mean value of all the biochemical parameters assessed also were in the normal range in drug treated groups (3 groups for 3 drugs) and the control group. Histopathological examinations revealed normal structures and no significant adverse effects observed in the kidney, heart, liver and spleen tissues. According to these results, we conclude that herbal preparation of *Tripala katakana pata* decoction, *Madumehari powder* and *DT6* powder can be used to treat diabetic mellitus for minimum of 90 days duration without any adverse drug induced reactions. However, case control clinical trials involving human subjects are required in future studies to confirm findings obtained in this mice study.

Keywords: Herbal drugs; toxicity; mice model; histopathology

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Comparative Effects of *Morinda Citrifolia* Linn Fruit Extract and Albendazole Against Earthworm

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Helminth infections are one of the most prevalent infectious disease that impacts on global human's health. World Health Organisation (WHO) has approved a few drugs that are frequently used in the treatment of helminth infestations in human being. Thus, this phenomenon has paved way for researchers to search for potential sources of alternatives including herbs to minimize the problem. *M. citrifolia* Linn, locally known as "noni plant" or "mengkudu", has been reported as traditional remedies for a variety of diseases including its anthelmintic effect.The present study specifically indicated the ethanolic extract of the fruits of *M. citrifolia* Linn exhibited significant anthelmintic activity in terms of time of paralysis and death against the earthworm, *Eudrilus eugeniae* (African Night Crawler). The study was carried out with and without the standard drug albendazole to check the comparative efficacy of *M. citrifolia* Linn on *E. eugeniae*.The fruits of *M. citrifolia* were extracted using absolute alcohol by maceration technique and concentrated via rotatory evaporator. Various concentrations (10, 20, 50, 100, 150, 200 mg/mL) of ethanolic extracts of the fruits were used to determine the *in-vitro* anthelmintic activity by correlation with the ability of the extracts to paralyse and kill the worm. The results showed that the concentration of extract at 200 mg/mL showed the significant decrease in terms of time taken for paralysing and killing the earthworms (9.00± 1.14 min and 25.50± 1.87 min respectively), thus, proved a remarkable anthelmintic potency when compared to the negative control (Dimethyl sulfoxide, DMSO) (>150.00 min) with $P<0.0001$. The extract showed a dose dependant increase in anthelmintic potency up to 50 mg/mL.The ethanolic extract of *M. citrifolia* Linn fruits have showed significant anthelmintic effect in terms of time of paralysis and death on earthworm.

Keywords: *Morinda citrifolia Linn fruit;* anthelmintic*; Eudrilus eugeniae;* albendazole