



UNIVERSITI
TEKNOLOGI
MARA

FACULTY of
PHARMACY



5th International Postgraduate Conference on Pharmaceutical Sciences 2017

15th – 18th May 2017

**Faculty of Pharmacy,
Universiti Teknologi MARA, MALAYSIA**



**Concerted Research Towards
Holistic Healthcare through
Pharmaceutical Sciences**



PROGRAMME BOOK

Published by
Asian Journal of Pharmaceutical & Clinical Research
www.ajpcr.com



**WELCOME MESSAGE FROM THE VICE CHANCELLOR OF UNIVERSITI TEKNOLOGI MARA
(UiTM)**



Assalamualaikum wbt

I would like to congratulate the Postgraduate Society of Faculty of Pharmacy, UiTM Puncak Alam for organising the 5th International Postgraduate Conference on Pharmaceutical Sciences 2017 (iPoPS 2017).

This conference has been successfully organised since 2012 by the postgraduate students together with the Faculty of Pharmaceutical Sciences, Tokyo University of Science, Japan without fail. This

symbolises the strong collaboration between UiTM and TUS towards academic excellence, which enhances our academic visibility. UiTM has been ranked top 200 in the QS World University Ranking by subject 2017 for Pharmacy & Pharmacology. This is a big achievement for a young faculty of 14 years.

On that note, contributions from the postgraduates play an important role in the development of human capital headed for a personified nation with humanistic values, forward thinking with an edge in entrepreneurship. Postgraduate students nowadays not only face challenges in completing their studies but also competing with the fast pace of ever changing technology. Therefore, we need to allow their full integration into the global knowledge society together with international collaboration to advance the quality of higher education worldwide.

Participation in event with different cultures and background like iPoPS 2017 helps develop articulate thinking and inspire healthy competition of ideas among the young, budding research generation in Malaysia. Hopefully, by continuing to invite eminent speakers from all over the world, the Conference will gain its recognition in the international research community and make UiTM soar upwards. We trust this Conference will be a success and carry on for years to come.

Congratulations!

EMERITUS PROFESSOR DATO' DR. HASSAN SAID
Vice-Chancellor
Universiti Teknologi MARA (UiTM)

FOREWORD FROM THE DEAN, FACULTY OF PHARMACY, UiTM



Assalamualaikum wbt. Greetings.

It gives me great pleasure to welcome all of you to the 5th International Postgraduate Conference on Pharmaceutical Sciences 2017 (iPoPS 2017). We are proud to host iPoPS 2017 which is organised by postgraduate students from the Faculty of Pharmacy, Universiti Teknologi MARA (UiTM) and the School of Pharmaceutical Sciences, Tokyo University of Science (TUS) on an annual basis.

One of the major thrusts of the Faculty of Pharmacy, UiTM is to develop human capital in core research areas to fulfil the needs of Malaysia in nation development. iPoPS 2017 supports this aspect by enabling young minds

to explore ideas through sharing of thoughts and research findings with other scientists. As part of our efforts in broadening the views of postgraduate students beyond their field of study, we have set the theme of this year's iPoPS as "Research Insights: Holistic Healthcare through Pharmaceutical Sciences".

I trust that all participants of iPoPS 2017 will seize this opportunity to network and initiate collaborative activities. With the participation of local and international postgraduate students, academicians and researchers, I am certain that iPoPS 2017 will serve as an excellent platform for networking.

As the dean of Faculty of Pharmacy, I am very proud of my postgraduate students. It is my hope that they will continue to excel in their research with passion and enthusiasm. I wish to congratulate all committee members, postgraduate students, lecturers, staff, and all those involved for their wonderful effort at making iPoPS 2017 a success. To all conference delegates, may your visit here be fruitful and enjoyable.

Yours truly,

PROF. DR. AISHAH ADAM,
Dean
Faculty of Pharmacy
Universiti Teknologi MARA (UiTM)

FOREWORD FROM CONFERENCE PROGRAM DIRECTOR



Assalamualaikum wbt.

On behalf of the committee members, I would like to sincerely welcome all participants to our 5th International Postgraduate Conference on Pharmaceutical Sciences 2017 (iPoPS 2017), which is held here at the Faculty of Pharmacy, Universiti Teknologi MARA (UiTM), Puncak Alam Campus, from 15th to 18th May 2017. We are delighted to see our annual iPoPS to go from strength to strength under the guidance and leadership of our dean, Prof Dr Aishah Adam. This prestigious event will continue to serve as an excellent platform that showcases

pharmaceutical sciences-related research activities of postgraduate students not only from the UiTM, but also from other institutions of higher learning.

iPoPS 2017 is certainly one of the important highlights of the yearly activities at the Faculty of Pharmacy, UiTM. It creates many wonderful opportunities that allow postgraduate students to enjoy a unique and memorable conference experience. More importantly, iPoPs 2017 facilitates development of young researchers who can think critically and communicate their research findings effectively.

We warmly welcome participants from other institutions of higher learning. Their participation will enrich the diversity of research cultures and also foster linkages between universities. It is our hope that all these interactions will eventually benefit the pharmaceutical industry in this region.

Last but not least, I would like to sincerely express my heartfelt gratitude to all students and staffs of the Faculty of Pharmacy, UiTM, in making iPoPs 2017 a great success. In particular, I would like to thank Prof. Dr. Aishah Adam and Assoc. Prof. Dr. Mizaton Hazizul Hasan for their continuous support and guidance.

I hope that you will have an enjoyable time at this event.

QAMARUL HAFIZ ZAINOL ABIDIN

Program Director

**5th International Postgraduate Conference on Pharmaceutical Sciences
iPoPS 2017**

Day 1: 17th May 2017 (Wednesday)					
Time	Programme				Venue
08:00-09:00	Registration				Pavilion
09:00-09:15	Doa recital & Welcoming speech				DK500
09:15-09:30	Welcoming Address by Dean of Faculty of Pharmacy Prof. Dr. Aishah Adam				DK500
09:30-10:00	Opening Speech by Vice Chancellor of Universiti Teknologi MARA (UiTM) Emeritus Prof. Dato' Dr. Hassan Said				DK500
10:00-10:15	Tea break				Pavilion
10:15-11:15	Keynote Address: Apoptosis, BCL-2 family inhibitors and cancer chemotherapy Prof. Dr. Gerald M. Cohen, University of Liverpool, United Kingdom				DK500
11:15-12:05	Plenary 1: Development of new methods for diagnosis and treatment of cancer in chemistry, biochemistry and material sciences – multidisciplinary approach in TUS Prof. Dr. Shin Aoki, Tokyo University of Science, Japan				DK500
12:05-12:55	Plenary 2: Integrating pharmacogenomics & metabolomics for clinical research Prof. Dr. Teh Lay Kek, Universiti Teknologi MARA, Malaysia				DK500
12:55-14:30	Lunch/Poster Session (Category A and B)				Pavilion
14:30-15:00	Concurrent session 1: Pharmacology	Concurrent session 2: Chemistry	Concurrent session 3: Life Science	Concurrent session 4: Undergraduate	DK500, DK6, DK7 & DK9
	Invited 1: New strategy for cancer immunotherapy targeting myeloid-derived suppressor cells Asst. Prof. Dr. Ichiro Horie TUS, Japan	Invited 2: Design and syntheses of ESI enhancing and deuterium labelling reagents and their applications of LC/EMI-MS/MS analyses for trace bioactive compounds Dr. Shoujiro Ogawa, TUS, Japan	Invited 3: Effects of the anti-convulsants on the neuropathic pain-like state and pain-induced anxiety in mice Dr. Kazumi Yoshizawa, TUS, Japan	Invited 4: Role of pictograms in patient education Dr. Wong Pei Sei, International Medical University, Malaysia	
	OP1	OP1	OP1	OP1	
	OP2	OP2	OP2	OP2	
	OP3	OP3	OP3	OP3	
15:00-16:00	OP4	OP4	OP4	OP4	DK500
	OP5		OP5		
16:00-16:50	Plenary 3: Quality use of medicine research: impact to whom? Assoc. Prof. Dr. Mohd Makmor Bakry, Universiti Kebangsaan Malaysia, Malaysia				DK500
16:50-17:40	Plenary 4: Dihydroartemisinin nanocrystals: characterization and in-vitro anticancer activity Assoc. Prof. Dr. Satit Puttipipatkachorn, Mahidol University, Thailand				DK500
17:40	Tea & End				Pavilion

Day 2: 18th May 2017 (Thursday)					
TIME	Programme				Venue
08:00-09:00	Registration				Pavilion
09:00-09:50	Plenary Speaker 5: Occupational toxicology for understanding environment-induced disorders in humans Prof. Dr. Gaku Ichihara, Tokyo University of Science, Japan				DK500
09:50-10:10	Tea Break				Pavilion
10:10-11:00	Plenary Speaker 6: Application of nanotechnology for future development pharmaceutical and cosmetical products from Indonesian herbal plant Dr Mahdi Jufri, Universitas Indonesia, Indonesia				DK500
11:00 – 11:50	Plenary Speaker 7: Pharmaceutical care: should we be involved in medication administration? Prof. Dr. Mohamed Mansor Manan, KPJ Healthcare University College, Malaysia				DK500
11:50-13:00	Concurrent session 1: Pharmacology	Concurrent session 2: Pharmacy Practice	Concurrent session 3: Pharmaceutics	Concurrent session 4: Life Science/ Chemistry/ Undergraduate	DK500, DK6, DK7 & DK9
	OP1	OP1	OP1	OP1	
	OP2	OP2	OP2	OP2	
	OP3	OP3	OP3	OP3	
	OP4	OP4	OP4	OP4	
	OP5	OP5	OP5	OP5	
	OP6	OP6	OP6	OP6	
13:00-14:30	Lunch/Poster Session (Category C and D)				Pavilion
14:30-15.20	Plenary 8: Computational modelling of host-pathogen interactions: from atoms to systems Dr. Peter J. Bond, Bioinformatics Institute A*STAR, Singapore				DK500
15:20-16:10	Plenary 9: Advancement of pharmacy practice research in Thailand and its impact on national health policy Assoc. Prof. Dr. Surakit Nathisuwan, Mahidol University, Thailand				DK500
16:10-17:00	Plenary 10: Making your research relevant and translatable from bench to market Prof. Dr. Yuen Kah Hay, Universiti Sains Malaysia, Malaysia				DK500
17:00-19:00	Closing Ceremony/ Prize Giving / Grand Hi-Tea and Dismiss				DK500

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Athirah binti Abdul Rahman

SPEAKERS INFORMATION



Prof. Dr. Gerald M. Cohen

University of Liverpool, United Kingdom

Prof. Dr. Gerald M. Cohen's area of specification is Mechanism of Toxicity (Apoptosis). Currently, he concentrates on utilising this knowledge in improving current cancer treatments using chemotherapy. Other than that, his current research involves understanding of both apoptotic and non-apoptotic roles of BCL-2 family proteins and the significance of a novel endoplasmic reticulum stress response was identified recently in his laboratory.



Prof. Dr. Shin Aoki

Tokyo University of Science, Japan

Prof. Dr. Shin Aoki is currently a Professor at the Department of Medical and Life Sciences. He is a recipient of the Award of Japan Society of Coordination Chemistry for Young Scientists (1999), the AJINOMOTO Award in the Synthetic Organic Chemistry, Japan (2001), the Pharmaceutical Society of Japan Award for Young Scientists (2002) and many more.



Prof. Dr. Teh Lay Kek

Universiti Teknologi MARA, Malaysia

Prof. Dr. Teh Lay Kek was registered as a pharmacist in since 1995. Her research areas are Experimental and Clinical Pharmacogenomics and also Clinical and experimental Metabolomics. Currently, she is working as a lecturer at Faculty of Pharmacy and the Deputy Director of Integrative Pharmacogenomics Institute (iPROMISE), one of Centre of Excellence in Universiti Teknologi MARA, Malaysia.



Assoc. Prof. Dr. Mohd Makmor Bakry

Universiti Kebangsaan Malaysia, Malaysia

Assoc. Prof. Dr. Mohd Makmor Bakry is the Deputy Dean Graduate, Research and Innovation. Most of his research interest is regarding Clinical Pharmacy practice and Clinical Pharmacokinetics. He was the recipient of UKM Excellent Teaching Award 2013 (Health Sciences) in 2014.



Assoc. Prof. Dr. Satit Puttipatkhachorn

Faculty of Pharmacy, Mahidol University, Thailand

Dr. Satit Puttipatkhachorn is currently the Head of Department of Manufacturing Pharmacy and associate professor in Pharmaceutics at Faculty of Pharmacy, Mahidol University, Thailand. His research interest is Solid Pharmaceutics, especially physicochemical properties of drug substances and excipients, drug-polymer interaction, and many more. Another area of his research is oral controlled-release drug delivery system, nanoparticulate drug delivery system and new pharmaceutical excipients from polysaccharides. At present, he has published over 100 original articles in international journals.



Prof. Dr. Gaku Ichihara
Tokyo University of Science, Japan

Prof. Dr. Gaku Ichihara is currently a professor at Department of Occupational and Environmental Health, Faculty of Pharmaceutical Sciences, Tokyo University of Science. In 2000, he was the recipient of Incentive Award, 2000, Japan Society for Occupational Health.



Dr. Mahdi Jufri
Universitas Indonesia, Indonesia.

Dr. Mahdi Jufri's latest research involved the creation of phytosome gel from piper betle extract for an anti-acne solution. Dr. Jufri has been active in the academic world and has been invited to present at many events locally and Malaysia. His research papers have been featured in multiple journals such as the *International Journal of PharmTech Research*, *Asia Pacific Journal of Tropical Disease* and many others.



Prof. Dr. Mohamed Mansor Manan
KPJ Healthcare University College, Malaysia

Prof. Dr. Mohamed Mansor Manan. He is an expert in Pharmacokinetics, Pharmacodynamics, Pharmacotherapy, Pharmacoepidemiology and Pharmacoconomics and collaborative study with the PSD on diabetes mellitus drug utilization pattern in Malaysia. He was a pharmacist in the Ministry of Health for 27 years. Currently he is working at KPJ Healthcare University College, Malaysia.



Dr. Peter J Bond
*Bioinformatics Institute (A*STAR), Singapore*

Dr. Peter J Bond is currently appointed as Principal Investigator at Bioinformatics Institute (A*STAR), Singapore. He is experienced in the development and application of multiscale modelling and simulation approaches to understand biomolecular recognition and assembly. His special fields are Bioinformatics, Computational Biology, Molecular Modelling.



Assoc. Prof. Dr. Surakit Nathisuwan
Mahidol University, Thailand

Assoc. Prof. Dr. Surakit Nathisuwan's areas of specialization are efficacy and safety evaluation of drugs on cardiovascular system, evaluation of pharmacogenomics on drug actions pharmaceutical care for patients with cardiovascular diseases, pharmaceutical care for patients in internal medicine ward and medication use evaluation.



Prof. Dr. Yuen Kah Hay
Universiti Sains Malaysia, Malaysia

Prof. Dr. Yuen Kah Hay is an expert in Pharmaceutical Technology. His special field are Biopharmaceutics Studies, Design and Development of novel drug delivery systems. Currently, he is a professor at School of Pharmaceutical Sciences, Universiti Sains Malaysia.



Asst. Prof. Dr. Ichiro Horie
Tokyo University of Science, Japan

Asst. Prof. Dr. Ichiro Horie's research interest is Immunopharmacology, especially to reveal the function or pharmacological regulation of myeloid-derived suppressor cells (MDSC), which plays important roles for immunosuppressive system and involves in the pathophysiology of tumours, autoimmune diseases, or chronic inflammation.



Dr. Shoujiro Ogawa
Tokyo University of Science, Japan

Dr. Shoujiro Ogawa is a lecturer at Faculty of Pharmaceutical Sciences, Tokyo University of Science (Chiba, Japan). He is a recipient of Hoshino Foundation's young scientists awards in BMAS 2016 and Award for young scientists by the division of physical sciences in 2017.



Dr. Kazumi Yoshizawa
Tokyo University of Science, Japan

Dr. Kazumi Yoshizawa's areas of specialization are Pharmacology and Palliative Care. Currently he is a lecturer at Faculty of Pharmaceutical Sciences, Tokyo University of Science. He obtained his Doctor of Philosophy in Pharmaceutical Sciences in September 2011.



Dr. Wong Pei Se
International Medical University, Malaysia

Dr. Wong Pei Se is currently a senior lecturer in the School of Pharmacy, International Medical University. She has over 13 years of teaching experience in the area of Pharmacy Practice. She obtained her PhD in Pharmaceutical Sciences (Pharmaceutical Care) from the University of Strathclyde, United Kingdom. Her research interests include pharmacy education and emerging roles of pharmacists. Her current projects involve better understanding of self-care in chronic disease and developing the evidence to support future roles for pharmacists.

APOPTOSIS, BCL-2 FAMILY INHIBITORS AND CANCER CHEMOTHERAPY

GERALD M COHEN

Department of Molecular and Clinical Cancer Medicine, University of Liverpool, Liverpool, UK

Abstract

Key observations of Kerr, Wyllie and Currie in the 1970s described the importance of programmed cell death and apoptosis in cancer. Subsequent work by Bob Horvitz and colleagues recognised the critical importance of three genes, CED-3, CED-4 and CED-9, in developmental cell death in the nematode, *C. elegans*. This was followed by the identification of the mammalian homologues of these genes. Enormous progress has been made in the last 20 years in understanding the basic mechanisms of apoptosis, in particular, the role of the BCL-2 and the caspase family of proteins as critical regulators or executioners of apoptosis, respectively. Although it is now recognised that deregulation of apoptosis is one of the cardinal features of cancer, there has been limited progress in the translation of this knowledge into clinical application and improved cancer therapy. Apoptosis occurs following either triggering of cell surface death receptors (the extrinsic pathway) or perturbation of mitochondria (the intrinsic pathway). Most drugs, including cancer chemotherapeutic agents, chemicals and irradiation, induce apoptosis by activation of the intrinsic pathway. Members of the BCL-2 family control the integrity of the outer mitochondrial membrane and thus are critical in determining the susceptibility of cells to apoptosis induced by the intrinsic pathway. The BCL-2 family comprises antiapoptotic members, such as BCL-2, MCL-1, BCL-XL and BCL2A1 (BFL-1), multidomain proapoptotic members, such as BAX and BAK, and proapoptotic BH3-only proteins, including BAD, BIM, PUMA, BID, BIK, NOXA and BMF. On receipt of a death signal, BAX and BAK can form oligomers in mitochondrial membranes leading to permeabilisation of the outer mitochondrial membrane, release of cytochrome c and caspase activation, whereas antiapoptotic BCL-2 members prevent this release by blocking activation of BAX and BAK. BH3-only proteins act upstream of BAX and BAK and are critical for cell death initiation. Their activity is tightly controlled by diverse transcriptional and post-translational mechanisms. BH3-only proteins selectively bind into the hydrophobic groove of antiapoptotic BCL-2 family members leading to BAX/BAK activation. Based on the elegant studies by the research group at Abbot (Abbvie) using NMR structure based design, ABT-737, the first selective inhibitor of BCL-2/BCL-XL/ BCL-w was synthesised. ABT-737 binds selectively to the hydrophobic groove, displacing the BH-3 only proteins leading to apoptosis. Recent developments have seen the synthesis of selective inhibitors of the anti-apoptotic BCL-2 family members and their use in treating various cancers will be discussed.

DEVELOPMENT OF NEW METHODS FOR DIAGNOSIS AND TREATMENT OF CANCER IN CHEMISTRY, BIOCHEMISTRY, AND MATERIAL SCIENCES–MULTIDISCIPLINARY APPROACH IN TOKYO UNIVERSITY OF SCIENCE

SHIN AOKI^{1,2,3}

¹Fac. Pharm. Sci., Tokyo Univ. Sci., 2641 Yamazaki, Noda 278-8510, Japan ²Dev. of Medical-Science-Engineering Cooperation, Research Institute for Sci. Tech., Tokyo Univ. Sci., 2641 Yamazaki, Noda 278-8510, Japan ³Imaging Frontier Center, Research Institute for Sci. Tech., Tokyo Univ. Sci., 2641 Yamazaki, Noda 278-8510, Japan

The requirement of new methodologies for diagnosis and treatment (theranostics that is combination of diagnosis and therapeutics) of cancer is rapidly required. Moreover, metastasis is a complex process involving generation and spreading of circulating tumour cells (CTCs) and CTC clusters to form metastatic sites. Therefore, capturing and analysis of CTCs/CTC clusters from cancer patient's blood are clinically important for detailed information on cancer progression and functional characterization of CTCs of individuals. Although recent advances in devices and materials developed to detect, capture and enumerate CTCs from the blood of cancer patients are principally based on their different properties of immunological (e.g.; Cell Search) and physical (size) properties. However, these methods suffer from their tedious manipulation and incomplete separation of CTCs from white blood cells, etc.

In this presentation, we will report on our recent achievement on i) design and synthesis of cyclometalated iridium (III) complexes that are functionalized by specific reactions for detection and cell death induction of cancer cells, ii) convenient marker-free detection of CTCs and CTC clusters based on the size and shape parameters as well as specific interaction between antigens and antibodies, and iii) capture and culture of the cancer cells by glass filters. These are the results of multidisciplinary collaboration in Tokyo University of Science.

INTEGRATING PHARMACOGENETICS AND METABOLOMICS IN CLINICAL RESEACRH

TEH LAY KEK

Integrative Pharmacogenomics Institute (iPROMISE), Universiti Teknologi MARA Selangor, Malaysia

With the advent of biotechnology, more “omics” were introduced with the aim to provide answers to research questions which are ultimately used to help reduce or prevent human diseases. Pharmacogenetics promised to make therapy personalised via detection of the genetic variants affecting the PK-PD while metabolomics which uses technology to profile small molecules at real time helps provide understanding on the effects of genetically determined variation. Here, I illustrate two examples of how metabolomics had enabled a better understanding on the response of colorectal cancer and breast cancer patients treated with 5-fluorouracil (5-FU) and tamoxifen, respectively. In the patients with colorectal cancer, those with DPYD variants contributed to alteration of metabolite profiles and influenced the prognosis CRC patients treated with 5-FU. In addition, eicosapentanoic acid (EPA) and N-acetylsphingosine which have anticancer properties were significantly lower in the patients with metastasis. This suggested their roles in progression of cancer. While, in the breast cancer patients, metabolic datasets were mined using a range of pattern recognition techniques, including hierarchical cluster analysis, principal component analysis, partial least squares and neural networks. Increment of several metabolites in tumour samples has highlighted lipid and eicosanoid metabolic pathways as potential perturbation in the metabolism of tumour cells in comparison to the normal. Further validation on the metabolites is required before they can be used as breast cancer biomarkers. With these 2 examples, our research group believed that integrating pharmacogenetics and metabolomics provide promising information on potential markers to monitor disease progress and drug efficacy. Integration of pharmacogenetics and metabolomics will allow translation of findings obtained from clinical research to personalised patient care.

QUALITY USE OF MEDICINE RESEARCH: IMPACT TO WHOM?

MOHD MAKMOR-BAKRY

Faculty of Pharmacy, Universiti Kebangsaan Malaysia, Malaysia

Drug treatment failure and adverse drug events are commonly associated with poor usage of medicine rather than the drug product itself. Quality Use of Medicines (QUM) involve medicines being used judiciously, appropriately, safely and efficaciously. QUM research aimed to optimise the benefit of drug therapy with appropriate cost and pharmacy services. Stakeholders for QUM research includes consumer or patients, health care professionals and policy makers. QUM research covers various area including pharmacotherapy, psychosocial issue of drug use, pharmacoconomics, pharmacy in health system, pharmacy services development and health professional development and training. Uniqueness of QUM research includes the application of a hybrid method between quantitative and qualitative approaches. Qualitative approaches will further describe the quantitative findings. Collaboration between pharmacy based researchers and other health care professionals increase the value and impact of the research findings. Ultimately, the QUM research outputs will be benefited by the community, pharmacists, policy makers and nation. This contribution is imperative to achieve the global sustainable development goals which include good health and well-being.

DIHYDROARTEMISININ NANOCRYSTALS: CHARACTERIZATION AND *IN VITRO* ANTICANCER ACTIVITY

SATIT PUTTIPIPATKHACHORN^{1*}, PATNAREE SASITHORNWETCHAKUN¹, WAREE LIMWIKRANT¹ AND URACHA RUKTANONCHAI²

¹Department of Manufacturing Pharmacy, Faculty of Pharmacy, Mahidol University, Bangkok 10400, Thailand. ²National Nanotechnology Center, National Science and Technology Development Agency, Pathumthani 12120, Thailand

Dihydroartemisinin (DHA), an artemisinin derivative that is practically used for malaria treatment, has shown to be a potential anticancer drug candidate. Regrettably, its poor water solubility limits its absorption which impacts drug bioavailability and effectiveness. Fabrication of DHA nanocrystals by high pressure homogenization seems to be a promising strategy with a possibility in large-scale manufacturing. The aim of present work was to prepare stable DHA nanocrystals with improved anticancer activity using microfluidizer technology. The effects of stabilizers (sodium deoxycholate, poloxamer 188 and polyvinyl pyrrolidone K30 and their combination on particle size reduction and physical stability were investigated. The average particle size was varied from 155 to 725 nm with the span of 0.360 to 0.484 and zeta potential was ranged from -55.8 to -28.5 mV depending on the stabilizers used. Freeze drying process was applied to enhance the stability of the nanocrystals. The overall results revealed that the nature of stabilizers and their concentration had the effect on particle size reduction, zeta potential, the relative degree of crystallinity, redispersibility, and also the physical and chemical stability of DHA nanocrystals. Field emission scanning electron microscopy and powder X-ray diffraction indicated the presence of DHA in nanocrystalline form. The appropriate combination of these 3 stabilizers could generate DHA nanocrystals that physically and chemically stable at 4 °C for at least 3 months. In addition, DHA nanocrystals showed significantly enhanced DHA saturation solubility in water compare to the physical mixtures. Furthermore, *in vitro* cytotoxicity study provided evidence of the enhanced toxicity to cervical cancer cell lines significantly. Flow cytometry revealed that DHA nanocrystals significantly promote an early apoptosis of HeLa cells in particle size-dependent manner. In conclusion, these results strongly suggest an opportunity to further develop these stable DHA nanocrystals as an effective drug delivery system for cancer treatment.

OCCUPATIONAL TOXICOLOGY FOR UNDERSTANDING ENVIRONMENT-INDUCED DISORDERS IN HUMAN

GAKU ICHIHARA

Tokyo University of Science, Japan

In 1996, reproductive and hematopoietic disorders were found in the workers involved in cleaning electronic parts in Korea. Experimental animal studies revealed that 2-bromopropane, which was introduced as an alternative to Freon, was the causative agent for the disorders. Subsequently the isomer 1-bromopropane has been introduced to the workplace as another alternative to Freon, as it is known to be less mutagenic than 2-bromopropane. Our early animal experiments revealed that 1-bromopropane is more neurotoxic than 2-bromopropane, in contrast to the result of mutagenicity test. These experimental studies played a critical role in diagnosis for the early patients intoxicated with 1-bromopropane. On the other hand, the studies on the human cases intoxicated with 1-bromopropane provided further information that could not be obtained from the animal studies, especially on the symptoms of the disorders related with the central nervous system including cognitive dysfunction or depressive mood. New animal experiments were conducted to test a hypothesis, which was generated from human cases, that exposure to 1-bromopropane induces cognitive dysfunction or depressive mood. Case studies may have limitations in revealing causal relationship as a nature of observational study, but provide significant information that cannot be obtained from experiments. In contrast, experimental studies can reveal causal relationship, but have limitations in terms of species difference. Both of human studies and experimental studies are important and their interactions are encouraged for understanding environment-induced disorders in humans and prevention for them.

APPLICATION OF NANOTECHNOLOGY FOR FUTURE DEVELOPMENT PHARMACEUTICAL AND COSMETICAL PRODUCTS FROM INDONESIAN HERBAL PLANT

MAHDI JUFRI

Faculty of Pharmacy Universitas Indonesia Depok 16424, Indonesia

Indonesia's have natural resources can be used for raw material of medicinal and cosmetic purposes that are beneficial to health. The use of active ingredients from natural are generally considered more secure than the active ingredients of synthetic drugs. Most of the herbal origin drugs possess insoluble character leading to lower bioavailability and increased systemic clearance requiring repeated administration or higher dose, which makes the drug as a poor candidate for therapeutic use. Nanoparticles technology such as nanoemulsion and nanovesicles have been developed to overcome these problems. These researches are conducted to formulated the nanovesicles form like liposome, niosome, ethosome and nanoemulsion with natural materials as active ingredient that have antioxidant activity to maintain healthy skin and antibacterial for the treatment of acne and also to overcome of alopecia problem. The objective of this study was to obtain formulations of nanovesicles which contain extract fraction of mangosteen pericarp and *Centella asiatica* herbs extract, ethosome of *Nothopanax* leaf extract, niosome which contain *Piper betle* leaf extract, and also nanoemulsion gel of curcumin. All the dosages form was developed into topical purposes or cosmetics that are stable and can be well absorbed on the skin so that it can improve the effectiveness of the therapy. All the results from theseresearch using nanotechnology technology method from natural resources can be further developed and manufactured by the pharmaceutical industry into commercial products.

PHARMACEUTICAL CARE: SHOULD WE BE INVOLVED IN MEDICATION ADMINISTRATION?**MOHAMED MANSOR MANAN****School of Pharmacy, KPJ University College, Nilai, Negeri Sembilan**

The Institute of Medicine (IOM) of the National Academic in the United States released a report, *To Err is Human: Building a Safer Health System* which reveals the reality of errors occurring in the healthcare sector. The report urges that everyone should partake in medication and patient safety. Medication error can occur at any stage of the medication process, whether prescribing, compounding, dispensing, distribution, or administration. A UK report showed common error mostly occurs during the administration stage (NPSA, 2009). It was observed that 35,982 cases out of the total 72,482 errors were administration errors. 76% of the reports came from the hospital setting and the rest from community or other healthcare settings. These errors had resulted in 17 death cases and 29 severe harm cases. The NPSA report showed the severity of MAE and MAE was considered as actual medication error and these were the most common errors reported by hospital staff (NPSA, 2009). There are not many studies in Malaysia that explores the occurrence and the contributing factors of medication administration error. Unpublished reports on MAE in an adult medical hospital in Johor found the incidence of 38%. A study by Chua in 2009 discovered the occurrence of medication administration error in hospital wards in Malaysia was at 11.4%. In the same year, an unpublished study conducted at Hospital Melaka on the occurrence of Medicine Administration Errors (MAE) among adult patients in medical wards found MAE is 48% of which the highest error is due to timing error (89.7%). However, these studies were done using the Unit of Use System. In 2013, there was a study at Hospital Selayang regarding drug preparation and administration errors but the scope is limited to intravenous drug (Ong & Subasyini, 2013). Following the above, studies conducted in 2014 and 2015 showed MAE is not an isolated issue but is prevalent in adult, elderly and paediatric wards. Thus, ward pharmacist need to seriously observe and monitor these events as it is a part of their role in practicing pharmaceutical care.

COMPUTATIONAL MODELLING OF HOST-PATHOGEN INTERACTIONS: FROM ATOMS TO SYSTEMS

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Infectious diseases due to bacterial and viral pathogens are a significant threat to human health. The interaction of such pathogens with host cells is critical to infection, the immunological response, and therapeutic interventions. We use multiscale modelling and simulations to investigate these processes in (near)-atomic resolution. I will give an overview of this research, with a particular focus on (1) key aspects of the flavivirus life cycle, and (2) the mammalian immune response to bacterial infection. For the former, we particularly focus on dengue virus, responsible for more than 400 million infections per year, and emerging infectious diseases such as Zika virus. We have recently developed simulation methods to refine the complete "outer shell" envelope structure of these flaviviruses. These "virtual viruses" are now serving as a platform to follow different mechanistic stages along the viral life cycle, including endosomal membrane fusion and infection. For the latter, we have developed complex models of Gram-negative bacterial outer membrane (OM) envelopes, and the receptor cascade of the host immune response, which recognizes OM components called "endotoxin" during infection. Using these models, we are able to trace the complete endotoxin recognition cascade in molecular detail, providing new insights into bacterial infections, barriers to antibiotic therapy, and potential immunomodulatory routes to treatment of endotoxicity and sepsis.

ADVANCEMENT OF PHARMACY PRACTICE RESEARCH IN THAILAND AND ITS IMPACT ON NATIONAL HEALTH POLICY

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Research in the area of pharmacy practice has constantly evolved based on the evolving practice standard and the role of pharmacists in a healthcare system. For Thailand, the country has adopted and adapted the concept of clinical pharmacy, and later, pharmaceutical care to guide the change in both pharmacy education and practice. For education, key changes are the mandate by the Pharmacy Council to change professional degree from a 5-year Bachelor to a 6-year Doctor of Pharmacy degree, expansion of graduate degree in pharmacy practice areas and the introduction of residency/fellowship programs. For practice, starting from sporadic success of practice models in the past, this has currently turned into a national movement. Coalition of leading professional organizations works collaboratively with pharmacy schools and other stakeholders to push this agenda forward. Throughout this practice development, research works conducted to formally evaluate pharmacist interventions in Thailand's context started to flourish. Increasing number of high-quality experimental studies show clear or potential benefit of pharmacist participation in Thai healthcare system. Pharmacy outcome researchers play a prominent role in publishing these studies in collaboration with clinical pharmacists in both local and international journals leading to improved visibility and knowledge sharing. Certain key information generated from these data were later used to support policy movement toward promoting pharmacy agenda at a national level and guiding national drug policy/health policy. In addition, these researchers are now increasing successful in producing research works that are beneficial and relevant to guide drug/health policy on a regional and global scale. With this advancement, pharmacy practice researchers have become important contributors toward national research performance. Their research works can also have direct social impact through changes in drug/health policy at a national and international level.

MAKING YOUR RESEARCH RELEVANT AND TRANSLATABLE FROM BENCH TO MARKET

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A major goal of health sciences research, is to produce meaningful results so that the knowledge gained can be applied in the diagnosis, treatment and management of diseases or for improving human health and wellbeing. To reach clinical application, a drug substance or treatment modality will normally have to successfully go through different phases of development. Hence, research outcomes of each phase must eventually be applicative and translatable, for example from cell to animal and finally to human studies. In many instances, researchers failed to properly address or take cognizance of some important components or elements in their research work that can affect this whole translation process. They include issues such as using unrealistic drug concentrations in cell studies, wrong doses or route of drug administration in animal studies, poor experimental designs and inappropriate outcome measures in the case of clinical studies. Even well-established researchers may sometimes overlook such issues while the less ethical ones may deliberately manipulate the study to yield intended outcomes. If such studies escape the attention of less discerning peer reviewers the work will get published, even in high impact journals. However, the findings from these studies are worthless and will remain just as a publication soon to be forgotten. In this presentation, some of the components or elements of research that can affect the translation process will be discussed and illustrated with specific examples.

NEW STRATEGY FOR CANCER IMMUNOTHERAPY TARGETING MYELOID-DERIVED SUPPRESSOR CELLS

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Myeloid-derived suppressor cells (MDSC) negatively regulate immune responses, and involve in various diseases such as tumours, infection, autoimmune diseases or chronic inflammation. In mice, MDSC are characterized by the dual expression of CD11b and Gr-1. The immunosuppressive activity of MDSC is associated with high levels of arginase-1 (Arg-1), nitric oxide and reactive oxygen species, and MDSC also induce regulatory T cells (Treg), suggesting that MDSC play central roles for immunosuppression system. Especially, in tumours, MDSC increase in tumour microenvironment and contribute to invasion and metastasis by inhibiting tumor immunity. Therefore, although agents to regulate MDSC activity may be new therapeutic drugs for various diseases including tumours, pharmacological regulation of MDSC activity has not been still established. In the present study, to find MDSC regulators, we focused on traditional Kampo medicines, Juzentaihoto (JTT) and Hochuekkito (HET). These drugs have immunomodulatory effects, however, the target or mechanism remain unclear. We examined the effects of these drugs on differentiation and function of MDSC. MDSC were differentiated from mouse bone marrow cells by IL-6 and GM-CSF, and identified as CD11b and Gr-1 double-positive cells using flow cytometry. Interestingly, JTT significantly decreased MDSC population in a concentration-dependent manner. And, JTT also decreased Arg-1 expression in MDSC. In addition, the immunosuppressive activity was attenuated in JTT-treated MDSC, indicating that JTT is a MDSC inhibitor. MDSC consist of two subsets, which are monocytic (M-MDSC) and polymorphonuclear (PMN-MDSC) morphology. JTT selectively decreased M-MDSC population, suggesting that JTT repressed M-MDSC differentiation. Finally, oral administration of JTT also decreased MDSC in tumour-bearing mice *in vivo*. Taken together, we found JTT as a MDSC inhibitor, and the negative regulation of MDSC function by JTT or its active components can be new strategy for cancer immunotherapy.

Keywords: Myeloid-derived suppressor cells (MDSC), tumor immunity, chronic inflammation

DESIGN AND SYNTHESIS OF ESI ENHANCING AND DEUTERIUM LABELLING REAGENTS AND THEIR APPLICATIONS OF LC/ESI-MS/MS FOR BIOACTIVE COMPOUNDS

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Sensitive and specific methods for the detection, characterization and quantification of bioactive compounds in body fluids are necessary for the diagnosis, pathological analysis and treatment of many diseases. Recently, LC/ESI-MS/MS has been widely used for these purposes due to its specificity and versatility. However, there are still two major problems to be solved regarding the biological sample analysis; 1) lack of sensitivity (low ESI sensitivity) and 2) limited availability of stable isotope-labelled internal standards (ISs) for most metabolites. Based on this background, ESI-enhancing and deuterium labelled derivatization (EEDL) reagents can be one of the effective solutions of these problems. Therefore, I have been studying to systematic development of EEDL reagents and their applications to the detection and quantification of metabolites in biological samples. The EEDL derivatization was achieved to improve assay precision without isotope-coded ISs and to enhance the sensitivity together. In this presentation, I will introduce the highlight of my latest research result.

EFFECTS OF THE ANTICONVULSANTS ON THE NEUROPATHIC PAIN-LIKE STATE AND PAIN-INDUCED ANXIETY IN MICE

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The World Health Organization (WHO) developed practical guidelines for pain relief in cancer patients in 1986. However, the cancer pain management is insufficient in Japan. One suggestive indicator of poor pain management in Japan is the small amount of morphine consumed in Japan. This means that, while educational approach has been reinforced in recent years, there is still widespread lack of the latest knowledge about the use of morphine and the cancer pain treatment among the health care workers in Japan. Moreover, it is well known that chronic pain induces depression, anxiety, and a reduced quality of life. Therefore, the present study, we evaluated the anxiolytic or antinociceptive effects of several types of anticonvulsants under a chronic neuropathic pain-like state. The mechanical sensitivity of the mouse hind paw was evaluated using von Frey filaments. Unilateral cuff-implantation induced mechanical allodynia in ipsilateral hind paw. At 4 weeks after cuff-implantation, these mice showed a significant anxiety-related behaviour in the elevated plus-maze test. Under these conditions, acute injection of anticonvulsants, including the carbamazepine, ethosuximide, and gabapentin, significantly attenuated the anxiety-related behaviours induced by chronic neuropathic pain. These anticonvulsants also produced a significant reduction in mechanical allodynia. These findings suggest that the anticonvulsants are effective for treating anxiety associated with chronic neuropathic pain and may be useful for treating neuropathic pain with emotional dysfunction such as anxiety.

ROLE OF PICTOGRAMS IN PATIENT EDUCATION

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Low literacy patients struggle to understand written medication instructions and management points leading to poorer physical condition, little understanding about their conditions and its management, weaker self-management skills and poorer medication adherence rates. Research revealed that humans have a cognitive preference for picture based, rather than text-based information and studies have shown that pictograms are particularly useful in patient education. Pictograms can be useful when delivering information such as dosing schedule, indication of the drug, side effects, instructions of administration and the importance of finishing the medications. Benefits include improve recall, comprehension, and adherence. While pictograms have generally found useful in improving patient comprehension and adherence, not all picture-based interventions have produced positive findings. Some pictograms have been found to be too complex and misleading. When using pictograms in health education materials, systematic evaluation should be considered to understand their effects.

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SYNTHESIS, CHARACTERISATION AND *IN VITRO* TOPICAL DELIVERY OF CAFFEINE-LOADED MULTIRESPONSIVE POLY(*N*-ISOPROPYLACRYLAMIDE)-BASED PARTICLES

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Abstract

This project aimed to investigate the potential of dual-responsive poly(*N*-isopropylacrylamide) (PNI)-based particles as carriers for topical drug application. Firstly, the PNI was copolymerised with acrylic acid (AAc) using the surfactant free emulsion polymerisation technique. They were characterised based on particle size and size distribution, zeta potential, temperature- and pH-sensitivity, phase transition and stability. The chemical structure of the particles was confirmed through Fourier transform infrared (FTIR) spectroscopy and their morphology was studied by using transmission electron microscope (TEM). The particles were loaded with a model permeant, caffeine using the post-fabrication technique. Following this, the particles were subjected to an *in vitro* permeation study across full-thickness rat skin and tape stripping for depth penetration analysis. The resulting mean hydrodynamic diameters of the particles were ranging from 600 to 1300 nm at room temperature with the uniformity values less than 0.3. The average zeta potential for all particles was in the range of moderate stability. All particles de-swelled in response to the increase in temperature ($p < 0.005$). For the pH-responsiveness of the particles, PNI-co-AAc 5% (PN2) and PNI-co-AAc 10% (PN3) particles de-swelled as the pH value decreased ($p < 0.001$) while the size of PNI was not affected by the pH variation ($p = 0.104$). The phase transition for all particles was consistent with the established literature. The TEM images of the particles confirmed that the particles were successfully synthesised as mono-disperse spheres. The *in vitro* permeation data demonstrated that the loaded caffeine have low potentials to rapidly permeate across full-thickness rat skin in comparison to the saturated caffeine control which was supported by the result obtained from *in vitro* penetration study where most of the caffeine-loaded particles remained on the stratum corneum layers. Further studies on *in vitro* permeation studies should be carried out to investigate its potentials for controlled/extended release mode.

Keywords: acrylic acid, caffeine, *N*-isopropylacrylamide, pH-responsive, temperature-responsive, rat skin

FORMULATION AND EVALUATION OF CARVEDILOL BUCCOADHESIVE BILAYERED TABLET: *EX VIVO*- BIOADHESION, RESIDENCE AND PERMEABILITY STUDIES

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Abstract

The present work aimed to develop and evaluate buccal tablet containing carvedilol. The carvedilol, bucco adhesive tablets were developed by direct compression method by employing different ratios of polymers such as sodium alginate, hydroxy propyl cellulose and methyl cellulose. Ethyl cellulose used as backing layer of the tablet. All physical parameters such as thickness, hardness, weight variation and drug uniformity were investigated. The tablet formulations were also subjected to evaluation of drug release in 6.8 phosphate buffer. *Ex vivo* bioadhesion, residence time and permeation through freshly cut porcine buccal mucosa membrane were evaluated. *Ex-vivo* residence time of all the prepared tablets of carvedilol were shows the values between 6.6 ± 0.92 to 7.12 ± 0.03 . Mucoadhesive strength, force of adhesion and bond strengths of the buccal tablets were found to be within the bioadhesive range and was comparable with other GRAS (generally regarded as safe) category polymers studied. A contact time of 4 minutes was paid to achieve optimal bucco adhesive strength. The permeation study shows that drug release more than 80% at the end of 5th hour. The present buccal carvedilol tablet formulations can be suitably developed as an alternate to conventional dosage forms with an added advantage of circumventing the hepatic first pass metabolism.

Keywords: Buccal tablet, carvedilol, ex vivo bioadhesion, ex-vivo permeation, ex-vivo residence.

THE RELEASE OF DIFFERENT TOCOL FROM A SIMPLE MONOLITHIC DRUG IN ADHESIVE (DIA) SYSTEM

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Abstract

The aim of this study was to investigate the feasibility of developing a novel drug in adhesive (DIA) therapeutic system for the purpose of delivering tocol for topical or systemic application. Tocotrienol rich fraction (TRF) extracted and concentrated from palm oil was chosen as a formulation to be incorporated into a patch system because it contains approximately higher amount of tocotrienols and α -tocopherol. Simple monolithic drug in adhesive (DIA) system was prepared using six different pressure sensitive adhesives (PSAs) exhibiting different performances and physicochemical properties. The release rate of tocol from the developed DIA were studied across the silicone membrane and full thickness human skin using jacketed 'Franz' diffusion cells. The prepared tocol formulation differed to a certain extent in their release ability based on the differences observed in their solubility in each PSA. The formulations with low solubility in their corresponding adhesive contributed to higher release rate when tested across the silicone membrane followed by the full thickness human skin. Taken together all these data, the DURO-TAK® 87-2052 and DURO-TAK® 87-2100 seems to be the most appropriate PSAs to be incorporated together with TRF for a transdermal therapeutic system (TTS) development. However, in terms of the amount of individual tocol permeated from the patch formulations, the overall result suggested that the TRF patches were rather promising for topical application indeed of achieving a systemic effect. The suggested results also proved that the preliminary optimisation of PSAs for tocol formulations was important as this ensure the early detection of adhesive failure before an effective TTS can be further developed.

Keywords: Tocol, Pressure sensitive adhesive, Monolithic, 'Franz' diffusion cell

NANOENCAPSULATION OF THIOLATED SODIUM ALGINATE: SYNTHESIS AND DOCETAXEL RELEASE STUDIES FOR COLONIC DRUG DELIVERY

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Abstract

Docetaxel had shown promising therapeutic efficacy towards the treatment of colon cancer. However, oral administration of docetaxel frequently leads to disintegration in the upper gastrointestinal tract and fails to reach the colon. This has caused significant adverse effects. We aim to formulate a colon cancer targeted docetaxel-loaded nanoparticles (DCXNP) through the conjugation of thioglycolic acid with sodium alginate. DCXNP with different formulations were prepared by *in situ* nanoprecipitation method and were characterised in terms of morphology, size distribution, encapsulation efficiency, and *in vitro* release. The nanoencapsulation efficiency of DCXNP was 20.4% and were found to be spherical in shape with mean diameter of 132 nm (Field Emission Scanning Electron Microscope) and 270 nm (Zetasizer), polydispersity index (PDI) recorded below 0.5 and zeta potential at -22 mV. The stability of DCXNP were high with 6 nm increase in size and 0.03 reduction in PDI after storage for a month. In pH sensitivity (pH 2 to 7) studies, the size increased 189.6% showing DCXNP is a pH-dependent swelling transition. Meanwhile, the size of DCXNP increased 58% in reduction-response studies. *In vitro* drug release studies showed that 0.89% of docetaxel released during the first 2.0 h at pH 1.0 glutathione-free media (stomach condition) and 14.8% during the first 5.0 h at pH 7.4 (small intestine condition). The release of docetaxel increased markedly when the DCXNP were exposed to pH 6.0 with glutathione (colon condition) with the cumulative release reached plateau at 16 h with 85.3 %, resulted in enhanced docetaxel release. Overall, these results suggested that the DCXNP described herein could be exploited as a prolonged drug release carrier for colon-targeted drug delivery.

Keywords: Disulfide cross-linked nanoparticles, thiolated sodium alginate, reduction-response, colon-specific drug delivery, docetaxel

VALIDATION OF A SIMPLE HPLC-UV METHOD FOR THE QUANTIFICATION OF ANDROGRAPHOLIDE ISOLATED FROM *ANDROGRAPHIS PANICULATA* NESS IN SELF NANO EMULSIFYING DRUG DELIVERY SYSTEM (SNEDDS) FOR DISSOLUTION STUDY

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Abstract

In this present work, a simple, rapid and accurate HPLC-UV method has been developed for the quantification of andrographolide isolated from *Andrographis paniculata* Ness in Self Nano Emulsifying Drug Delivery System (SNEDDS) formulation for the dissolution test. The assay was performed using a XTerra® MS C18 column (150 mm X 4.6 mm, five μ m) with a mobile phase of methanol and water (70: 30), at 0.8 mL/min flow rate and UV detection at 229 nm. Simulated gastric fluid (SGF) and simulated intestinal fluid (SIF) were prepared as dissolution medium. The validation parameter was conducted including the test on linearity, precision, accuracy, LOD, and LOQ. The result showed an excellent linearity with $r = 0.999$ and good selectivity for both dissolution medium. The method showed sufficient precision, with a relative standard deviation (RSD) smaller than % Horwitz. The accuracy reported as % recovery was found to be 102.61 and 101.17 % in SGF and SIF dissolution medium respectively. LOD and LOQ were found to be 0.46 ppm and 1.40 ppm in SGF medium, 0.87 ppm and 2.64 ppm in SIF medium respectively. In conclusion, the HPLC method developed showed selectivity with linearity in the working range, good precision and accuracy and suitable for quantification andrographolide in dissolution study.

Keywords: Validation, *andrographolide*, SNEDDS, Dissolution, HPLC

SELF-NANOEMULSIFYING DRUG DELIVERY SYSTEM (SNEDDS) OF BAWANG TIWAI (*ELEUTHERINE BULBOSA* MALL. URB.) EXTRACT

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Abstract

Bawang tiwai (*Eleutherine bulbosa* Mall. Urb.) is a herbal medicinal plant that has been widely used as an alternative treatment for diabetes, hypertension, and cancer. One of its major active compound is quinone that has low water solubility. The bioavailability of water-insoluble compounds can be increased by self-nanoemulsifying drug delivery system (SNEDDS). This system comprises of oil, surfactant and co-surfactant. The aim of this study is to determine the influence of oils, tween 20 and PEG 400 to the physical characteristics and physical stability of SNEDDS containing bawang tiwai extract. Oils that were used in the screening including virgin coconut oil, olive oil, oleic acid, and capryol. The ternary diagrams were constructed to indentify the SNEDDS forming region. Particle size, polydispersity index (PI), and zeta potential were determined using particle size analyzer to assess the characteristics of SNEDDS. Result of the oils screening showed that oleic acid has the highest solubilization capacity of bawang tiwai extract. The three best formula consisted of oleic acid as the oil phase, tween 20 as surfactant and PEG 400 as co-surfactant with the ratio of 1:8:1, 1:7:2 and 1:6:3, resulting in SNEDDS having particle size between 15.07 to 103.53 nm, PDI values between 0.285 to 0.471 Φ , and zeta potential between -44.6 to -67.8 mV. Physical stability test of the three best formula had shown good results, there were no visible separation of the SNEDDS components. In conclusion, using 10% of oleic acid, tween 20 approaching 80% and maximum amount of PEG 400 (30%) can produced a good and stable SNEDDS of bawang tiwai. Meanwhile, SNEDDS formula consisted of more than 10% of oleic acid were not stable when diluted with water.

Keywords: SNEDDS, bawang tiwai, oil phase, tween 20, PEG 400

GREEN ENERGY BIOSYNTHESIS FOR PREPARATION AND CHARACTERISATION OF GOLD NANOPARTICLES USING *Leucaena Leucocephala (Lam.)) De Wit*. AQUEOUS EXTRACT

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Abstract

Nanotechnology has become one of the most sophisticated and advanced areas of research in this field. Among nanoparticles, gold nanoparticles demonstrate particular advantages in this field due to their unique properties including small size and high surface area-to-volume ratio. The chemical reduction has been extensively used for the preparation of nanoparticles because these methods are easy and cheap and can be used to prepare large quantities of nanoparticles. Flavonoids one of the compounds has to be used for biosynthesis of nanogold formulation. Flavonoids knew as stabilator and reduction agents in this formulation. The purpose of this study, to get the optimal formulation and characterised the biosynthesis of nanogold in an aqueous extract. This preparation was made by reduction of chlorauric acid in 5% aqueous extract using a bottom-up method. Then the formulations were characterised visually, wavelength value with the spectrophotometer, particle size, and duration when nanogold was made. To know the morphology of nanogold conducted by TEM and SEM testing. This study showed the proportion concentration of chlorauric acid and aqueous extract (2:8) effective to produce nanogold; the solution colour changes from yellow to purple and pink. The morphology of gold nanoparticles is spherical and triangular with the particle size below 50 nm. The aqueous extract contains flavonoids serves as reducing agents for the green and low energy biosynthesis process.

Keyword: Flavonoids, *Leucaena leucocephala (Lam.) de Wit.*, Nanogold, Green Biosynthesis

SNEDDS (SELF-NANOEMULSIFYING DRUG DELIVERY SYSTEM) FORMULATION OF SARANG SEMUT'S (*MYRMECODIA PENDANS*) EXTRACT WITH VARIATION OF CAPRYOL, TWEEN 80, AND PROPYLENE GLYCOL

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Abstract

Sarang semut (*Myrmecodia pendans*) is a traditional medicinal plant that has been widely used as an alternative treatment for diabetes, diarrhoea, and cancer. Flavonoids in *Myrmecodia pendans* extract known as active compounds for pharmacological activity, and it has low solubility in water. The method can be used to improve the availability of water-insoluble compounds for better drugs delivery into the body is SNEDDS (Self-Nano Emulsifying Drug Delivery System). The purpose of this study is to get the formulation and characterised of SNEDDS of ethanolic *Myrmecodia pendans* extract. The oils phase used in the screening including virgin coconut oil, olive oil, and capryol. The parameters used are particle size, polydisperse index (PI), zeta potential, and the ternary diagram which is made to show SNEDDS forming region. Particle size, polydisperse index (PI), and zeta potential analysis using Particle Size Analyzer (PSA), also uniformity analysis for parameter value is using the average value and standard deviation. The result of the formulation. The best formulation capryol as oil vehicle, tween 80 as the surfactant, and propylene glycol as co-surfactant. The amount of extract is 1 mg/mL, the proportion each formulation were 1:8:1 ; 1:7:2 ; 1:6:3, and those formulation resulting particle size between 12.53 nm – 15.03 nm, PI values between 0.276 – 0.479 and the value of the zeta potential -45 mV until -53,86 mV. Physical stability by the three formula had shown good results, without substantial decrease. *Myrmecodia pendans* extract can formulate in SNEDDS formulation as drugs delivery systems with capryol as oil vehicle.

Keywords: *Myrmecodia pendans*, SNEDDS, Capryol, tween 80, Propylene Glycol

EXPLORATION OF ABSORPTION PROPERTIES OF PHARMACEUTICAL GRADE CLAYS AS NAJIS MUGHALLAZDAH CLEANSING AGENT

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Abstract

Dog saliva is categorized as an extreme religiously-prohibited dirt (najis mughalladzah) which the cleansing procedures require the use of soil, sand and clay. The adsorption properties of some clays on several bacteria have been studied, but the study on bacteria from dog's saliva has never been investigated. The purpose of this study was to investigate the adsorption ability of some clays (bentonite and kaolin) on the bacteria isolated from dog's saliva. Adsorption test were performed on *Proteus mirabilis* sp. The bacteria adsorptions were observed after 5 minute treatment with clays using a colony counter. The adsorption ability of bentonite clay on *Proteus mirabilis* sp is higher than kaolin clay. It can be concluded that both bentonite and kaolin have adsorption capability to dog saliva bacteria and can be used as a cleansing agent for najis mughalladzah purification.

Keyword: bentonite, kaolinite, adsorption, bacteria isolated from dog's saliva

SCREENING, OPTIMIZATION AND EVALUATION OILS, SURFACTANTS, CO-SURFACTANTS of NANOEMULSION GREEN BEAN SPROUTS (*Vigna radiata L.*) FORMULATION AS A COSMETIC SERUM

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Abstract

The content of vitamin E in green bean sprouts can be used as an antioxidant. The purpose of this study is to obtain the nanoemulsion formula of green bean sprouts from the results of screening and optimisation. This optimisation of nanoemulsion formulations consists of various oils, surfactant (Tween 20), and co- surfactant (PEG 400). The method uses the solubility screening and optimisation of each green bean sprouts in oil vehicle to formulated in nanoemulsion and evaluated from various parameters such as particle size and PI (polydisperse index), organoleptic test, pH test, and qualitative TLC test. The results of qualitative TLC analysis showed in the extract positive contained vitamin E compare to the reference material. The primary results of screening and optimisation of green bean sprouts, oils which were the best oils (Capryol, Arachidic oil) by formulation comparisons in formulas are particle size between 16.3 nm - 199 nm, and PI 0.242 -0.578. The results showed that the formula using Capryol oil and Arachidic oils as oils vehicle had good chemical, organoleptic, and properties as serum nanoemulsion

Keywords: Green Bean Sprouts, Nanoemulsion, Particle Size Analyzer, Screening, Optimization

IMPACTS OF BINDER AND DISINTEGRANT ON DETAM II SOYBEAN (*GLYCINE MAX* (L.) MERR) TABLET MADE BY DRY GRANULATION

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Abstract

Black soybean (*Glycine max* L. Merr) Detam II Variety has a scavenging activity and total phenolic compound higher than yellow soybean. As a natural compound, the flow properties of black soybean (*Glycine max* (L.) Merr) Detam II powder is poor due to its high content of moisture, so it must be made by dry granulation method. Dry binder and disintegrant have opposite functions on tablets formula where one serves to increase consolidation between particles while other accelerate the disintegration time as the rate limiting step of dissolution. The effects of disintegrant and dry binder variation on the physical characteristics of black soybean tablets have been observed. The variations of polyvinylpyrrolidone (PVP)-K30 combined with Plasdane S-630 were used as binders, while sodium starch glycolate (SSG) was used as a disintegrant. Approximately 4% of PVP-K30 as a single binder in the formula component provides better hardness and friability value of tablets, but the disintegration time was longer than other formulas. The amount of SSG used was increased to 4 and 8%, respectively in the formula component. 4% was the ideal amount of SSG as a disintegrant to get the best physical tablet characteristics and also disintegration time, about 4.45 minutes. Increasing the amount of SSG has no significant effect on the disintegration time even tends to slow down

Keywords: Detam II black soybean (*Glycine max* (L.) Merr), Tablet, Binder, Disintegrant, Disintegration time

CAFFEINE DELIVERY IMPROVMENT WITH CANDLENUT OIL: CHARACTERIZATION AND PENETRATION TEST ON HYDROALCOHOLIC GEL**ANGGIT DIMAS ANGGORO****Abstract**

Caffeine is a chemical agent that we can be easily found and have considerable benefits. However, caffeine is not used frequently. On the other hand, hydroalcoholic gel is an ideal form to deliver caffeine into the skin. The research is aimed to determine the character of hydroalcoholic gel of caffeine and the effect of candlenut oil as an enhancer for caffeine. Four formulas have been used with different candlenut oil level started from 0 ml, 1,5 ml, 3 ml, and 4,5 ml and using HPMC-Carbopol combination as the gel base. The caffeine that penetrated inside the membrane was calculated using Franz diffusion cell over cellophane membrane and the organoleptic, homogeneity, pH, stickiness, viscosity, and spreadability were observed. The results showed that candlenut oil has the best penetration effect at the highest concentration (4,5 ml) which was in the condensed form, white in colour with good homogeneity, pH $\pm$ 6,3, and viscosity $\pm$ 316 dps.

PREPARATION AND CHARACTERIZATION OF GOLD NANOPARTICLES RUTIN TRIHYDRATE 0,1% WITH THE BIOSYNTHESIS PROCESS

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Abstract

Nanotechnology lately has been a lot of interest because is believed to have many uses. Nanoparticle synthesis process can be done by several methods, such as by using a metallic such as gold, silver, zinc, platinum, and others. The synthesis of gold nanoparticles most popular because it can be used as sensors, catalysts, easily synthesized and functionalized has chemical stability, biocompatibility (low toxicity) and optical properties are easily arranged. One compound that can be used to help the process of biosynthesis of gold nanoparticles is rutin. Rutin has physiological actions such as anti-inflammatory, antibacterial, antioxidant, and antitumor. To determine the quality of the gold nanoparticles, it is necessary to do their thorough characterization. Preparation and characterization of gold nanoparticles include observation discoloration, while the formation of gold nanoparticles, particle size, morphology of the gold nanoparticles and gold nanoparticles profile. Observation of the color change is done visually. Time formation of gold nanoparticles is done by looking at the wavelengths used Uv-Vis spectrophotometer. The particle size can be determined by particle size analyzer. The profile of the gold nanoparticles can be determined by using Fourier transform infrared (FTIR). Morphology of the gold nanoparticles can be determined by using SEM and TEM. The results showed that the best formula that can be used to make gold nanoparticles is Formula AJ (regular mix HAuCl₄ trihydrate 1000 l and 900 l) with a time of formation for 24 hours. The particle size of the gold nanoparticles have a size of 112.83 trihydrate routine nm. Morphology AuNPs of routine trihydrate has a non-uniform shape such as triangle, round, pentagon, hexagon and square when viewed from inside the particles and irregularly shaped when viewed from the surface of the particles.

Keywords : Rutin Trihydrate, Gold Nanoparticles, Biosynthesis.

PREPARATION AND CHARACTERISATION GOLD NANOPARTICLE CASSAVA AQUEOUS LEAF EXTRACT (*MANIHOT ESCULENTA CRANTZ*) 1% WITH BIOSYNTHESIS PROCESS

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Abstract

Gold nanoparticles (AuNPs) is a particle formulation dispersed in thousandths of a micron or nanometer size and conjugated with metal ions. AuNPs can be made by using biosynthesis which is need natural reducing and stabilizer agent such as flavonoid. Cassava leaves (*Manihot esculenta Crantz*) is a plant which is has a high content of flavonoid. This study aims to determine the best formulation in nanoparticle formation of gold nanoparticles of cassava leaf aqueous extract with biosynthetic process and characterization of gold nanoparticles. AuNPs made by using chloroauric acid mixed with each of the formulation at room temperature. Characterization of the formation time is done visually and by spectrophotometry, the particle size was measured with a particle size analyzer, the functional group seen by FTIR, and the morphology of nanoparticles were analyzed by SEM and TEM. The result show that the best formulation of AuNPs cassava leaf aqueous extract is formula 40 (1 mL chloroauric acid:1 mL extract). The formation time observation of formula 40 show that it was changed into purple in 10 minutes. The particles size of formula 40 is 55,4 nm. The test results of functional groups by FTIR characterization showed that there are significant differences between the functional groups that exist on a single extract and gold nanoparticles were generated. Based on TEM and SEM tests showed that formula 40 of AuNPs cassava leaf extract have a spherical shape. The best formula of AuNPs is formula 40 which is contain 1 mL chloroauric acid and 1 mL cassava leaf aqueous extract

Keywords: Gold Nanoparticles, *Manihot esculenta Crantz*, Biosynthesis

DEVELOPMENT AND VALIDATION OF A LIQUID CHROMATOGRAPHY-MASS SPECTROMETRIC METHOD FOR QUANTITATIVE ANALYSIS OF MEMANTINE IN PHOSPHATE BUFFER SALINE

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Abstract

A rapid, sensitive and selective analytical method was developed and validated for the determination of memantine, used to alleviate symptoms of Alzheimer's disease. All samples were prepared in phosphate buffer saline, followed by liquid chromatography tandem spectrometric analysis and an electrospray-ionization interface. Memantine and internal standard i.e. amantadine, were analyzed on a Zorbax C18 column (50 mm x 3 mm x 1.7 μ m) using gradient elution with mobile phase 0.1 % formic acid in deionised water and 0.1 % formic acid in methanol at flow rate of 0.3 ml/min and monitored on a triple quadrupole mass spectrometer, operating in the multiple reaction monitoring mode. The method was validated over a concentration range of 0.01-5 μ g/mL. The retention time for memantine and amantadine were 5.27 and 4.84 mins respectively. The assay was linear over the range of calibration standards i.e., a concentration range of 0.1 μ g/mL to 0.62 μ g/mL ($r^2 > 0.997$). The developed method was found to be accurate, reliable and reproducible. Hence it was successfully applied for the quantification of released memantine from nanoparticles in *in vitro* studies and to quantify the amount of memantine in analytes from plasma and brain

Keywords: LCMS; Validation; Alzheimer's disease; Memantine

FORMULATION OF OMEPRAZOLE NANOSUSPENSION USING EVAPORATIVE PRECIPITATION-ULTRASONICATION TECHNIQUE

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Abstract

Formulation of poorly water-soluble drugs remains a challenging problem for experts. Omeprazole is one such agent. This study aimed to formulate and optimize an omeprazole nanosuspension to achieve improved bioavailability and stability. Omeprazole nanosuspensions were prepared using an evaporative precipitation-ultrasonication technique. A series of polymers and surfactants were screened, at pulse and continuous ultrasonication modes for different durations. A total of five minutes of pulse ultrasonication and HMPC with poloxamer 188 were selected for further studies. A Box-Behnken design (BBD) was adopted to optimize the formulation parameters. The nanosuspensions were characterized for particle size, polydispersity index (PDI), and zeta potential. In this study, the optimum values for optimization parameters were 20 mg of omeprazole, 100 mg of HPMC, and 200 mg of poloxamer 188 to produce a nanosuspension of a mean particle size of 295 nm, polydispersity index of 0.41 and zeta potential of -49.3 mV. The feasibility of preparing nanosuspension by evaporative precipitation-ultrasonication technique was shown in our study through the successful formulation of our omeprazole nanosuspension.

Keywords: omeprazole, nanosuspension, evaporative precipitation-ultrasonication technique,

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IDENTIFICATION AND OPTIMISATION OF LIPASE-CATALYSED SYNTHESIS OF BETULINIC ACID AMIDE IN A SOLVENT SYSTEM

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Abstract

Betulinic acid amide was synthesized from the enzymatic reaction of betulinic acid and butylamine catalysed by immobilized lipase, Novozym 435 from *Candida antarctica* lipase. The effect of different reaction parameters, such as effect of reaction time, reaction temperature, amount of enzyme and substrate molar ratio (Betulinic acid: Butylamine) were studied and conventionally optimised. Based on this study, the enzymatic synthesis of betulinic acid amide was found to be 64.6% at the optimum conditions of 24 h, 40 °C, 100 mg enzyme, 1:1 substrate molar ratio (betulinic acid : butylamine) in 9:1 mixture of chloroform and hexane as solvent. The identification of final product was carried out using TLC, melting point, FTIR and NMR showed the presence of betulinic acid amide. Betulinic acid amide showed a better cytotoxicity compared to betulinic acid as the concentration inhibiting 50% of the cell growth (IC₅₀) against MDA-MB-231 cell line (IC₅₀ < 30 µg/ml).

Keywords: Betulinic acid, Butylamine, Novozym 435, Betulinic acid amide

SYNTHESIS OF DISACCHARIDE NUCLEOSIDES AND OLIGOSACCHARIDES WITH THIOLYCOSYL DONORS

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Abstract

Carbohydrates are diverse and abundant molecules which play important roles in biological phenomena.^[1] Since the traditional synthesis of oligosaccharides and glycoconjugates is a time-consuming process, the new and efficient methodologies remain a field of intensive research.^[2] Disaccharide nucleosides and various homologues constitute an important class of natural compounds and they have been found to show antibiotic and other bioactivities as drug candidates.^[3] We have already reported on the synthesis of disaccharide 2'-deoxynucleosides by the chemical *O*-glycosylation of 2'-deoxynucleoside derivatives such as 2'-deoxyadenosine, 2'-deoxyguanosine, 2'-deoxycytidine, and thymidine, with thioglycosyl donors.^[4] As an extension of this research, we examined *O*-glycosylation reactions of ribonucleosides (not 2'-deoxynucleosides), whose 2',3'-diols are temporarily protected with phenylboronic acid.^[5] Moreover, development of oligosaccharide synthesis with template molecules is underway. In this approach, it is expected that the desired oligosaccharide would be obtained by simultaneous activation of all the thioglycosides after they are aligned on the template. *O*-Glycosylation reactions were carried out, resulting in formation of linear and cyclic oligosaccharides.^[6] These results will be reported.

Keywords: Glycosylation, Thioglycoside, Disaccharide Nucleoside, Oligosaccharide

POLYPHENOLIC CONTENT AND ANTIOXIDANT POTENTIAL OF GYNURA PROCUMBENS AND GYNURA CREPIOIDES LEAVES: A COMPARISON STUDY

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Abstract

Antioxidants play a vital role in delaying the progression of chronic diseases such as cancer, diabetes and hypertension. Natural antioxidants from plants are gaining research interest as synthetic antioxidants are carcinogenic. The aim of our present study was to extract phytochemicals from two plants namely *Gynura procumbens* (*G. procumbens*), *Gynura crepioides* (*G. crepioides*) and compare their polyphenolic contents and antioxidant capacity. Optimized ultrasound-assisted extraction (UAE) method with 70 % ethanol for 60 minutes at 75 °C was employed to extract the phytochemicals. The polyphenolic content was investigated by using Folin-Ciocalteu assay, whereas the antioxidant potential was determined by using phosphomolybdenum assay, and 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay. The results showed that phytochemicals were successfully extracted with extraction yield of 19.97 %, 17.70 % to *G. procumbens* and *G. crepioides* respectively. In comparison, *G. procumbens* contained a significantly ($p < 0.05$) higher amount of total phenolic content (TPC) (49.59 ± 0.68 mg GAE/g) than *G. crepioides* (35.90 ± 0.78 mg GAE/g). Besides, *G. procumbens* demonstrated a better total antioxidant capacity (TAC) (77.41 ± 1.49 mg AAE/g) than *G. crepioides* (70.62 ± 0.74 mg AAE/g) ($p < 0.05$). However, *G. crepioides* (101.72 ± 9.09 µg/mL) outshined *G. procumbens* (177.62 ± 1.44 µg/mL) in terms of DPPH radical scavenging activity ($p < 0.05$). Excellent correlation between TPC and TAC was drawn for both *G. procumbens* ($r = 0.987$, and $r^2 = 0.974$) and *G. crepioides* ($r = 0.888$, $r^2 = 0.789$). Strong correlation was also established between TPC and DPPH radical scavenging activity for both *G. procumbens* and *G. crepioides*, with ($r = 0.921$, $r^2 = 0.848$) and ($r = 0.966$, $r^2 = 0.933$) respectively. Overall, *G. procumbens* exhibited better antioxidant potential than *G. crepioides*. However, *G. crepioides* also demonstrated antioxidant activity therefore it could be further explored to determine other beneficial medicinal properties

Keywords: *Gynura procumbens*, *Gynura crepioides*, Folin-Ciocalteu, Phosphomolybdenum assay, DPPH assay

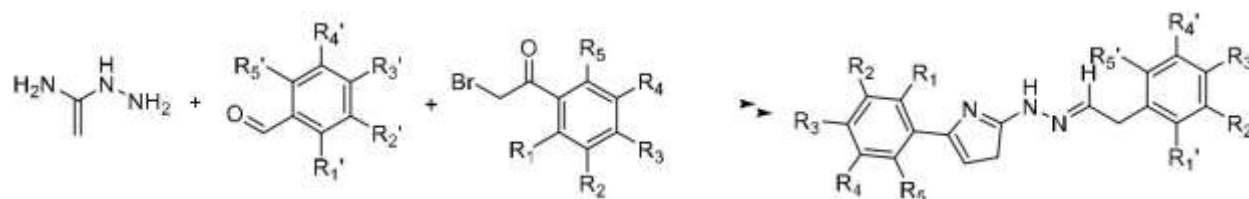
SYNTHESIS OF NOVEL BENZYLIDENEHYDRAZINYLPHENYLTHIAZOLES AND EVALUATION OF THEIR ANTI-INFLAMMATORY ACTIVITY *IN VITRO*

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Abstract

Hydrazones and thiazoles possess a wide spectrum of biological activities including antimicrobial, anticonvulsant, analgesic, anti-inflammatory, antiplatelet, antitubercular and antitumour activities. Acyl hydrazones with an azomethine proton ($-\text{NHN}=\text{CH}-$) possess promising biological activities and existence of thiazole bearing anti-inflammatory drugs such as fanetizole, meloxicam and fentiazac prompted us to design some novel benzylidenehydrazinylphenylthiazoles (BHPT). Novel BHPTs were designed through bioisosteric replacements in anti-inflammatory drugs and addition of azomethine group. BHPTs were synthesised by condensation of thiosemicarbazide and aryl aldehyde followed by cyclisation with 2-bromo-1-phenylethan-1-one. The compounds were characterised using ^{13}C , ^1H , LC-MS, FT-IR, and UV-VIS spectroscopic techniques. Cytotoxic effect and anti-inflammatory activity of tested compounds were evaluated on U937 and RAW 264.7 macrophage cell lines. The differential expressions of cytokines were investigated using multi-analyte ELISA kit. The tested compounds showed a high percentage of cell viability and significantly decreased nitrite accumulation in LPS-stimulated RAW 264.7 cells with IC_{50} values of 20 – 35 μM in inhibiting nitric oxide production.



Keywords: Hydrazinylthiazole, *In vitro* anti-inflammatory, Cytokines.

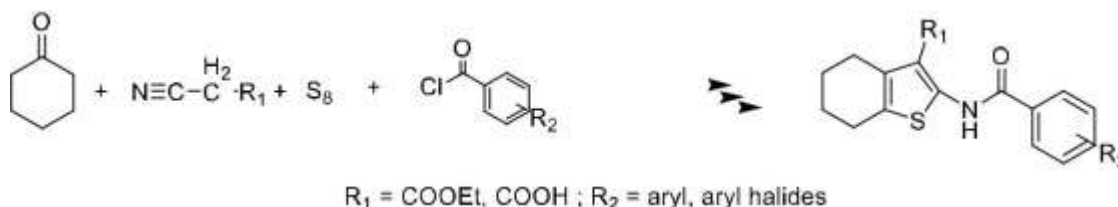
NOVEL 2-AMINO-4,5,6,7- TETRAHYDROBENZO[b]THIOPHENE: MICROWAVE-ASSISTED SYNTHESIS AND *IN VITRO* ANTI-INFLAMMATORY ACTIVITY

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Abstract

As part of our effort to discover novel anti-inflammatory compounds, we synthesized tetrahydrobenzothiophene (THBT) derivatives. In literature, 2-amino-4,5,6,7-tetrahydrobenzo[b]thiophene (2-aminoTHBT) is reported to possess promising antibacterial, anti-inflammatory and antitumour activities. It is synthesised via Gewald reaction followed by acylation. However, this conventional synthetic method poses many limitations such as lower yield and purity, longer reaction time etc. In our lab, we compared the efficacy of microwave-assisted vs conventional synthesis of 2-amino THBT. Microwave assisted synthesis produces higher yields with relatively high purity. The same method was extended to synthesise a library of 2-amino THBT analogs and were characterised using ¹H, ¹³C NMR and FTIR spectroscopic techniques. The compounds' metabolic stability was determined using rat, mice and human liver microsomes and anti-inflammatory activity was determined in vitro using murine macrophages (RAW 264.7 cells) and human macrophages (U937 cells). The compounds were found to be relatively metabolically stable and showed dose-dependent inhibition of nitric oxide production in RAW 264.7 and U937 cells induced by lipopolysaccharide from Escherichia coli (LPSEc). The effect of compounds on expression of various cytokines in RAW 264.7 and U937 cells was also investigated. In conclusion, microwave-assisted synthesis was found to be much superior to conventional synthesis of 2-aminoTHBTs; all the compounds were shown promising anti-inflammatory activity; and metabolically stable.



Keywords: tetrahydrobenzothiophene, microwave-assisted synthesis, anti-inflammatory

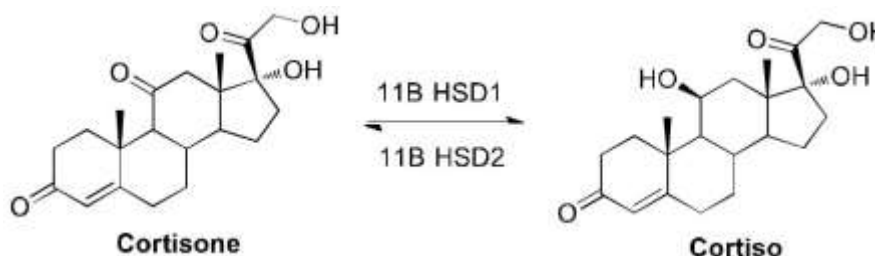
DESIGN AND DISCOVERY OF NOVEL LEAD COMPOUND TETRAHYDROTHIAZOLOPYRIDINE AS 11 β -HYDROXYSTEROID DEHYDROGENASE 1 INHIBITOR AND THEIR EVALUATION OF ANTI-INFLAMMATORY ACTIVITY

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Abstract

Obesity is categorised as an inflammatory disorder as chronic inflammation plays an essential role in the pathogenesis of obesity related disorders. The circulating free fatty acids (FFA) elevates the expression of inflammatory cytokines through nuclear kappa-B (NF-KB) pathway activation. The restricted treatment options for obesity such as diet, lifestyle and a few available anti obesity drugs provide limited success rates. Being a ubiquitous hormone, glucocorticoids play a key role in regulating energy metabolism in which their levels are being dictated by the tissue specific 11 β -hydroxysteroid dehydrogenase 1 (11 β -HSD1). 11 β -HSD1 catalyses the conversion of inert cortisone to active cortisol which consequently leads to obesity due to inclined level of the enzyme in liver and adipose tissues. Thus, a novel pharmacological inhibition of 11 β -HSD1 with anti-inflammatory and anti-adipogenic properties possesses a promising approach. A set of novel Tetrahydrothiazolopyridine derivatives specifically targeting 11 β -HSD1 has been synthesised and their efficacy has been studied using 3T3-L1 adipocytes, cytokines multi-analyte ELISA kit as well as metabolic stability studies. Among the set of studied synthesised compounds, ERGS-TR013A displayed the highest potency, offering a novel therapeutic strategy in ameliorating metabolic alterations found in obesity and diabetes.



Keywords: 11 β -HSD1, anti-inflammatory, Tetrahydrothiazolopyridine, drug discovery, cortisol

ELICITATION OF ASIATIC ACID IN *CENTELLA ASIATICA* USING *KAPPAPHYCUS ALVAREZII*

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Abstract

Centella asiatica (also known as 'Pegaga') is an endangered medicinal herb used in the preparation of herbal drugs mainly. Its usefulness is due to the presence of four pentacyclic triterpene which are asiatic acid, asiaticoside, madecassic acid and madecassoside. It's over exploitation necessitates the development of conservation strategies and enhanced the production of secondary metabolites. In the present study, the effect of various concentrations of seaweed *Kappaphycus alvarezii* elicitor was used to increase the amount of asiatic acid production in *C. asiatica*. Four different concentrations of seaweed elicitor were 0 g/L, 2 g/L, 4 g/L and 8 g/L. The *C. asiatica* plants were harvested at day 0, 7, 14 and 21 and were dried at 40°C for 5 days. They were then soaked with methanol and filtered to obtain brown crude extract. The amount of asiatic acid contained in the samples was analyzed by High Performance Liquid Chromatography and area under the curve of retention peak was calculated. The diameter of leaf, number of new shoots and flowers were measured during successive stages of development. As a result, *K. alvarezii* was proven to increase the production amount of asiatic acid in *C. asiatica* at shorter time, which was at day 14. The concentrations of *K. Alvarezii* at 4g/L and 8 g/L were the best. The additional of certain concentrations of seaweed elicitor also can induce flowering in *C. asiatica*.

Keywords: *Centella asiatica*, asiatic acid, *Kappaphycus alvarezii*, elicitation, secondary metabolites

THE USE OF *CUNNIGHAMELLA ELEGANS* FOR BIOTRANSFORMATION OF LUPEOL

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Abstract

Cunnighamella elegans is a well-known fungus that has been used for biotransformations of organic compounds including coumarins, flavonoids and terpenes. Triterpenes are a versatile group of biologically active ingredients present in several plant extracts. Terpenoids are classified as monoterpenoids (C10), sesquiterpenoids (C15), diterpenoids (C20), triterpenoids (C30), and others. Lupeol is a pentacyclic triterpene and mainly found in medicinal plants. Lupeol was reported to possess beneficial effects as a therapeutic and preventive agent for a range of disorders. Many studies have confirmed that lupeol possesses strong activities such as antioxidant, antiinflammatory, antiarthritic, antimutagenic, and antimalarial, both *in vitro* and *in vivo*, and at its effective therapeutic doses exhibit no toxicity to normal cells and tissues. In our previous preliminary biotransformation screening, *C. elegans* was found to be effective to convert lupeol into polar and non-polar products that's why in current study we aim to carry out fermentation of lupeol with *C. elegans* on large scale in order to isolate and identify transformed products of lupeol using HPLC and spectroscopic techniques

Keywords: Triterpene, biotransformation, *Cunnighamella elegans*

MICROBIAL TRANSFORMATION OF ANDROSTANE STEROIDS BY THREE FUNGAL SPECIES

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Abstract

Microbial transformation of a series of androstane steroids (*cis*-androsterone, -androstane- α -ol-[3,2- androstane-2-hydroxy ethylene- -methyl- -ol-3-one) with three fungal species as biocatalyst was investigated. Sabouraud-4% glucose-agar was used to cultivate the fungal cultures as a solid phase medium. All substrates were incubated with fermentation broth and transformation reaction was carried out on a rotary shaker (220rpm) at 28°C for 12 days. Time course study for biotransformation of all substrates were carried out at different time intervals (4, 8 and 12 days). The fermented flask were subsequently extracted with ethyl acetate for three times and dried in vacuum. The level of transformation of all substrates were monitored by HPLC and TLC methods. Transformation of *cis*-androsterone (**1**) afforded five new products: 3,7,11-trihydroxy-5-androstan-17-one (**2**), 1,3-dihydroxy-5-androstan-17-one (**3**), 3,4,11-trihydroxy-5-androstan-17-one (**4**), 7-dihydroxy-5-androstan-3,17-dione (**5**), and 3,7,12-trihydroxy-5-androstan-17-one (**6**). Their structures were characterized by 2D NMR spectroscopy and mass spectrometry. The breadth of substrate spectrum and the excellent conversion rates achieved with these fungal cultures indicated that these cultures were potential microorganisms for production of valuable pharmaceutical ingredients and precursors.

Keywords: Microbial transformation, Fungal species, Androstane, Steroids, 2D NMR spectroscopy

THREE DECADES OF RESEARCH ON *CINNAMOMUM* SPECIES

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Abstract

A literature search was performed on *Cinnamomum* species from the Lauraceae family. It was found that two recent reviews, almost concurrently, concluded on the phytopharmacology and therapeutic uses of various parts of this plant. The barks are mostly studied, but not ignoring the fruits, leaves, roots, shoots, stems and twigs of this popularly known species of cinnamon, having the natural aromatic compounds. The *Cinnamomum* receives such attention, certainly due to the traditional practices, thus, bearing direct relevance with modern public health. The plant extracts were involved in a wide range of tests, which include antimicrobial, antidiarrheal, antibacterial, anticancer, antioxidant, anti-inflammatory, antipyretic and wound healing studies. Local scientists were heavily engaged in the investigation of cinnamon's chemical constituents and the cytotoxic effects of the essential oils. From the publications, the gas chromatography method was considered appropriate in order to confirm the cinnamon quality from the commercial raw materials. Furthermore, only 13% of the available 300 *Cinnamomum* species were scientifically investigated. Hence, decades' research of *Cinnamomum* and the continuity in studying various capacities of this plant extracts are worthy.

Keywords: chemistry; cinnamon, review

LITERATURE REVIEW OF *MANGIFERA* SPECIES

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Abstract

The *Mangifera* species can be found locally. In Sabah, *Mangifera pajang* (*M. pajang*) or buah bambangan, is considered as an iconic fruit. It is classified in the mango family (Anacardiaceae). The plant is reported as a potential source for functional food and medicine among the indigenous people. From the literatures, the phytochemistry and pharmacology of *M. pajang* were discussed. The compositions of *M. pajang* such as the flavonoids, were identified from the pulp. In addition, the extracts showed antibacterial and anticancer properties. The carotenoids from *M. pajang* and their antioxidant capacity were also evaluated. Latest experiment include the isolation of phenolic compounds from *Mangifera* flowers by using normal phase chromatography. Further study is required to investigate the leaves, stem bark, fruit peel and kernel of the fruit. It is anticipated that the kernel oil could comprise both saturated and unsaturated fatty acids. It is hoped that more research could be carried out on this underutilized plant.

Keywords: chemistry; extraction; literature; *Mangifera*

CURRENT UPDATES ON *CROCUS* PLANTS

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Abstract

This abstract is aimed to highlight the Islamic medicinal herb called Azza'faran or saffron. It is botanically known as *Crocus sativus* and classified in the iris family (Iridaceae). Historically, this plant is used as a food colouring and flavouring agent. It is cultivated in Europe, Turkey, Iran and central of Asia. Local groceries also offer saffron for culinary purposes. It is actually among the most expensive spice in the world. Recently, Malaysian researchers are taking initiatives in cultivating *Crocus* species. This study is performed to review the phytochemicals and the biological properties of *Crocus* species. From the literatures, the extracts were reported to exhibit antioxidant, antimicrobial and antihyperlipidemic activities. The extracts were also analysed in order to detect the potential natural compounds. It was anticipated that major constituents such as the carotenoids could be acquired from this herb. In the research methodology, the *Crocus* extracts could be fractionated by using supercritical carbon dioxide extraction technique. Current research in plants biotechnology would include the identification and quantification of adulterated saffron samples via spectroscopic analysis. Finally, it is hoped that more studies could be conducted on these croci.

Keywords: chemistry, literature, *Crocus*, saffron

RESEARCH HIGHLIGHTS OF *NIGELLA SATIVA***N. F. A. NIK MUSTAFA*, M. MOHAMAD AND I. ABDUL WAHAB**

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Abstract

This abstract is aimed to highlight *Nigella sativa* (*N. sativa*), a miraculous herb from the plant family of Ranunculaceae. It is often being called as the black seed. It is only a tiny seeds, but surprisingly was reported as a very powerful and useful medicinal herb. Since ancient times, people have been using it to cure human diseases and minor ailments. From the literature review, it is described as having thousands of significant therapeutic potentials and has become the centre of attention of many curious scientists and researchers. The investigation of *N. sativa* includes the study of its bioactive components and biological activities. The medicinal properties are antioxidant, antibacterial, antifungal, antiviral, antidiabetic, anticancer, anti-inflammatory, antiparasitic and anti-protozoal. *N. sativa* also showed to have analgesic effect, and protective activities towards cardiovascular, digestive, renal, nervous and hepatic portal system. These properties are due to the presence of its main bioactive component called thymoquinone in nearly 30%-48% in composition. Finally, it is hoped that more scientific studies would be carried out on how it exerts its biological activities and further researches should be focused specifically on other various components of *N. sativa*, which may also possess therapeutic promises.

Keywords: chemistry; black seed, review

ALOE VERA: A TRADITIONAL PLANT WITH PHARMACEUTICAL VALUES

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Abstract

This presentation is to discuss the pharmaceutical uses of aloe vera. It is derived from the Arabic word "alloe", which means clear and bitter substance. The botanical name of aloe vera is *Aloe barbadensis*. It belongs to the Asphodelaceae (Liliaceae) family. Aloe vera has been known and used for centuries for its health, beauty, medicinal and skin care properties. From the literature review, various studies have reported that aloe vera has antiviral and antibacterial properties, and possess the potential in treating different types of diseases such as constipation and diabetes. Two substances from aloe vera, the clear gel and the yellow latex, are used in health products, nowadays. The most important active component in aloe is polymannose. Experts are in agreement that the healing effect of aloe vera is due to the polysaccharide complex. The bioactive compound from aloe vera, which is known as acemannan, is very effective in a lot of treatments, such as burns, allergic reactions, rheumatic fever, ulcers, skin illness, diarrhea and inflammatory conditions of the digestive system. Aloe vera can be taken topically as a skin moisturizer and also orally as a laxative. In short, researchers are continuously investigating aloe vera's capability, in stimulating the effects on the immune system. Hopefully, this plant can be utilised to treat more diseases in the future.

Keywords: aloe, chemistry, review, uses

EVALUATION OF EPIGENETIC MODIFIERS VS MINERALS FOR FUNGAL SECONDARY METABOLITE PRODUCTION

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Abstract

The secondary metabolite production by filamentous fungi was explored in the presence of epigenetic modifiers and minerals. Thirteen fungi were selected from AuRiNS' culture collection. They were isolated from mushrooms, as well as soil samples from Malaysia and Svalbard Island, Norway. Fungi were grown in 96-well microtiter plates with potato dextrose broth (PDB) supplemented with epigenetic modifiers and minerals. The cultures were fermented at 10 C or 28 C and extracted with ethyl acetate (EA). The resulting extracts were subjected to high performance liquid chromatography (HPLC) analysis. Epigenetic modifiers and minerals induced significant changes in the profile of the secondary metabolite. At specific doses, the secondary metabolite production is positively affected. At other dosages or in different combinations, these compounds may partially suppress the production of secondary metabolites. This translates in either the absence of certain peak(s) or a lower amount of the secondary metabolites in the extract. Four compounds were successfully identified from fungi *Penicillium simplicissimum*, *Penicillium* sp., *Pseudallescheria boydii* and *Fusarium solani*.

Keywords: Fungi, secondary metabolites, elicitors, epigenetic modifiers.

OLIGOSTILBENES FROM *DIPTEROCARPUS SEMIVESTITUS*

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Abstract

Dipterocarpus semivestitus is one of the species that belong to Dipterocarpaceae family, a large family of timber plants with about 16 genera and up to 600 species distributed around the world. Oligostilbenes from Dipterocarpaceae are widely studied and can be categorized as dimers, trimers, tetramers, hexamers, heptamers and octamers. Three dimers and a trimer were successfully identified from the acetone extracts of the stem bark of *D.semivestitus*. They were isolated using preparative HPLC and further purified using recycling HPLC. The structures were established by means of spectroscopic methods and comparison with published data.

Keyword: Dipterocarpaceae, *Dipterocarpus semivestitus*, oligostilbene

ISOLATION AND CHARACTERIZATION OF FLAVONES FROM LEAVES EXTRACT OF *MUNTINGIA CALABURA*

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Abstract

Isolation and identification of pure compounds from natural products has contributed to development and designs of new drugs. One of the categories of compound commonly isolated from natural products is flavones which are chemically diversified. Flavones have been known to possess a wide range of therapeutic activities such as anti-inflammatory and antioxidant. In this study, we aim to isolate and characterize flavones in the leave extracts of *M. calabura* (MEMC). Conventional techniques of extraction and chromatography were used for the isolation of flavones. A total of 4.5 kg of *M. calabura* leaves was collected from Puncak Alam, Selangor. The dried, ground leaves were soaked with methanol and the crude extract (MEMC) was obtained by removing the solvent under reduced pressure using rotary evaporator. The MEMC was mixed with petroleum ether, and the petroleum ether partition (PEP) of *M. calabura* was collected. The PEP was fractionated by vacuum liquid chromatography to obtain different fractions which were profiled using thin layer chromatography. Out of the seven fractions that were obtained, fraction C, D and E were further purified using centrifugal thin layer chromatography. Two (2) pure compounds (C1 and C2) were isolated. The percentage of yield for PEP was 10.97% from MEMC. The structures of the compounds isolated were determined using ¹H Nuclear Magnetic Resonance (NMR) at 400 Hz and compared with reported compounds. ¹H NMR signals showed that both of the compounds are known flavones with the presence of mono-substituted benzene ring, methoxy and hydroxyl groups. C1 was identified to be galangin 3,7-dimethyl ether (3,7-dimethoxy-5-hydroxyflavone) and C2 is techtochrysin (5-hydroxy-7-methoxyflavone). The anti-inflammatory properties of these pure compounds will be investigated in the subsequent experimental study.

Keywords: *Muntingia calabura*, plant extract, pure compounds, flavones

DETECTION OF MITRAGYNINE IN *LANSIUM DOMESTICUM* ETHANOLIC EXTRACT

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Abstract

Mitragyna speciosa, also known as 'ketum' in Malay, is an indigenous plant species in Southeast Asia producing mitragynine, an active ingredient proven to have coca- and narcotic-like effects to its users. The widespread use of the *Mitragyna speciosa* extracts for recreational purposes in Malaysia has recently prompted the government to list mitragynine in the Third Schedule of the Poisons (Psychotropic Substances) Regulations of the Poisons Act 1952. Recently, the National Anti-drug Agency (AADK) has reported the use of *Lansium domesticum* by the natives of Perlis as a replacement to *Mitragyna speciosa* as it has become more difficult to obtain the plant. The extract of *Lansium domesticum*, also known as 'dokong' in Malay was claimed to have similar euphoric effects as *Mitragyna speciosa* extract. Therefore, the aim of this study was to detect the presence of mitragynine in leaves extract of *Lansium domesticum*. *Mitragyna speciosa* and *Lansium domesticum* leaves were extracted by soaking the dried leaves powder in 95% ethanol for two days. Chromatographic analysis of extracts was then performed using an LC/MS-QTOF system (LC 1200 Series Agilent Technologies) controlled by Agilent Mass Hunter Workstation Acquisition (B.02.01). The injection volume was 2 µl containing 0.033 µg of extracts and the separation was done at 40 °C using a ZORBAX Eclipse Plus C18 column (2.1 × 100 mm, 1.8 microns) with solvent (A) 0.1% formic acid in water and (B) 100% acetonitrile. The gradient was set up as follows: 0 min, 5% B; 0-36 min, 95% B; 36-41 min, 95% B; 41-41.1 min, 5% B; 41.1-48 min, 5% B and the flow rate was set up at 0.25 mL/min. The chromatogram revealed the presence of mitragynine at 15.399 min of retention time and m/z value of 399.23 in the *Mitragyna speciosa* ethanol extract but not in the *Lansium domesticum* ethanol extract. The results suggest that the euphoric effects of *Lansium domesticum* extracts may be due to other compounds present in the *Lansium domesticum* extracts.

Keywords: Addiction, *Lansium domesticum*, mitragynine, LC-MS

NATURAL INDOLE ALKALOIDS FROM KETUM (*MITRAGYNA SPECIOSA*)

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Abstract

Indole alkaloids have a bicyclic structure that consist of a benzene ring fused to the nitrogen containing pyrrole ring. This pyrrole ring determines the basic properties of indole alkaloids that make them pharmacologically active. From the literature review, the chemistry and pharmacology of the analgesic indole alkaloids from ketum or *Mitragyna speciosa* were published. This Rubiaceae plant is known to contain more than 25 alkaloids among which are the mitrogynine diastereoisomers, speciociliatine, speciogynine and mitraciliatine. These substances only differ in the orientation of the chiral carbons 3, 20 or both with respect to the main alkaloid. The substances show a unique concentration dependent pattern in that it demonstrates cocaine-like stimulating at low doses and opiod-like activity such as sedation when taken at high doses. The leaves of *M. speciosa* are usually used as traditional medicine to treat muscle pain, diarrhea, diabetes and to improve the blood circulation. It also possesses analgesic and anti-depressant effects. *M. speciosa* also has tolerance effect that leads to its abuse as it can cause sedation, pleasure and also has withdrawal effects, such as, hostility, aggression, muscle and bone aches, and jerky limb movements and hallucinations, delusion and confusion, as reported by the Drug Enforcement Administration (DEA). The most effective part of *M. speciosa* is their leaves. It is either prepared as a drink, chewed when freshly picked or smoked when dried. From the publications, these molecules could be identified in commercially available retail products, abroad. The research methodology includes liquid chromatography technique, coupled with ultra violet detection. Meanwhile, local scenario involves the Malaysian Anti-Corruption Commission that monitors ketum producers in Kedah. Earlier in 2015, Malaysian Boarder Security Agency made 406 arrests and raids involving ketum. In short, the public should be continuously educated and well-informed on the abuse of this crop leaves.

Keywords: alkaloids; chemistry; ketum; literature

SECONDARY METABOLITES FROM ENDOPHYTES ASSOCIATED WITH LONGYEARBYEN PLANTS FROM SVALBARD ISLAND, NORWAY

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Abstract

Plant endophytes are an important microorganisms serving as novel resource for many valuable chemical entities with antimicrobial, cytotoxic and anticancer activities. Further, studying microorganisms from a variety of biotopes increases the chances to isolate novel bioactive metabolites. For this reason extremophiles have been deemed as endowed with a greater potential for drug discovery than microorganisms from "normal" ecological niches. In this respect, psychrophiles from the Arctic fulfil the criteria of an interesting source of potential new bioactive chemical entities. In this project, a psychrophilic endophytic fungus isolate identified as *Phaeosphaeria* sp. was obtained from the petals of *Papaver dahlianum* collected in Longyearbyen, Svalbard Island, Norway. An ethyl acetate extract of the culture of this fungus on potato dextrose agar demonstrated good antibacterial activities when using broth microdilution (MTT) against *Staphylococcus aureus*, *Enterococcus faecium*, and *Pseudomonas aeruginosa*. The objective of this study was thus to isolate the secondary metabolites from the culture of the above fungus. The crude extract was analysed and fractionated by high performance liquid chromatography (HPLC). Pure compounds were subjected to standard spectrometric analyses and this led to the isolation of three known compounds, named clearanol C (**1**), annularin D (**2**) and 3,8-dihydroxy-3-hydroxymethyl-8-methoxy-4,5-dimethyl-isochroman-1-one (**3**) as well as one novel likely new compound (**4**) based on an unusual chromone-dihydrospiroisochromene ring system. Only few spiroisochromenes have been reported so far. Additional work is in progress to fully characterize this new novel compound.

Keywords: Arctic, psychrophilic fungi, Antibacterial, HPLC, LC/MS, NMR

UNRAVELING THE ENZYMES INVOLVED IN DIMERIZATION OF RESVERATROL

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Abstract

Within the Dipterocarpaceae, oligomeric stilbenes belong to an important group of polyphenolic secondary metabolites that biosynthetically emerge from the oxidative coupling of resveratrol. However, the enzymatic machinery that responsible for the oligomerization is still unknown. As part of our interest in biocatalysis and, more specifically, in enzyme-catalyzed carbon-carbon bond formation, we started an investigation on the oxidase(s) enzyme involved in dimerization of resveratrol. In order to fulfil the objectives of this work, first we treated the resveratrol with commercially available oxidase enzyme (Laccase) from *Trametes versicolor* fungi. The treatment produced a mixture of oligomers hence confirming the role of oxidase enzyme in the dimerization of resveratrol. Further research is proceeded by extracting proteins from the leaves of *Neobalanocarpus heimii* (Dipterocarpaceae) which locally known as cengal. The plant tissues are ruptured by maceration and the extracted enzymes are purified by various chromatographic techniques. The purified enzymes are then treated with resveratrol to see its oxidative coupling activity for oligomerization.

Keywords: Oligomerization; Oligostilbenes; Oxidase

AMPHOTERICIN-B INDUCED ACUTE KIDNEY INJURY IN A CASE OF ILLEAL HISTOPLASMOSIS

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Abstract

Amphotericin B is a mainstay treatment of disseminated histoplasmosis and the duration of therapy should be continued for up to two weeks or until clinical improvement is observed. Incidence of Amphotericin-B induced acute kidney injury (AKI) was more strongly associated with the absolute daily amphotericin B dose than with the weight-adjusted dose. Indeed, increased weight correlated with a higher, rather than lower, risk of nephrotoxicity for a given dose. The longer the duration of amphotericin B treatment, the greater the cumulative risk of nephrotoxicity; however, the per-day risk of renal toxicity was relatively constant. We present a case of a 32 years-old Chinese gentleman, Mr HCH who is positive for retroviral disease; his most recent CD4 count was 13. His complaints included abdominal pain and diarrhoea for past the one month. He was diagnosed with illeal histoplasmosis based on histopathological examination (HPE) findings. Patient was treated with IV Amphotericin B which after exposure of 1 dose induced acute kidney injury. Amphotericin-B induced nephrotoxicity is usually reversible and several approaches had been studied to minimize its risk of nephrotoxicity. Prehydrating the patient with saline and prolonging the infusion time have proven to be successful measures. Amphotericin B-related nephrotoxicity is an important dose-dependent and duration-dependent toxicity that is accentuated by certain nephrotoxic drugs and patient characteristics. Precautions must be taken when administering the drug to minimize such toxicity.

SYNTHESIS OF THIOLYCOSIDE-BASED MOLECULAR UNITS FOR OLIGOSACCHARIDES SYNTHESIS

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Abstract

Biomacromolecules including oligosaccharides are of growing interest, because of their natural existence, their interesting biological, physicochemical properties, and potential applications in medicine. The total synthetic of complicated oligosaccharides and cyclodextrin-related compounds were reported by Ogawa and co-workers, which required many steps and resulted in low chemical yields. Therefore, new methods are required for that purpose for synthetic. In this presentation, we report on the attempts at the synthesis of molecular units for their purpose. Namely, D-glucosyl thioglycoside analogues were designed and synthesised and their reactivity was examined under reaction conditions for glycosylation.

Keywords: Glycosylation, Thioglycoside, Cyclic oligosaccharides

ENHANCEMENT OF ASIATIC ACID IN *CENTELLA ASIATICA* BY USING PLANT GROWTH REGULATORS

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Abstract:

Centella asiatica L. was commonly known for its medicinal and nutritional values throughout the world. The medicinal values of this plant were mainly attributed to the presence of several triterpenes, namely asiatic acid, madecassic acid, asiaticoside and madecassoside. The aim of this study was to increase the amount of secondary metabolite which was asiatic acid in the plant by manipulating various concentrations of plant growth regulators (PGRs) of Kinetin (Kn) and 6-benzylaminopurine (BAP). *C. asiatica* was treated at four different concentrations of hormones and they were 1.0 mg/mL Kn + 0 mg/mL BAP, 1.0 mg/mL Kn + 1.0 mg/mL BAP, 1.0 mg/mL Kn + 2.0 mg/mL BAP and 1.0 mg/mL Kn + 4.0 mg/mL BAP, respectively. The plant in each concentration was harvested at day 0, 7, 14 and 21. Morphology changes occur on the plant were recorded throughout the days. The amount of asiatic acid content was measured and analyzed by using High Performance Liquid Chromatography (HPLC), with Column-ZORBAX 300SB-C18. *Centella* was found to contain high amount of asiatic acid at day 21 which was 0.104 mg/mL in hormones concentrations at 1.0 mg/mL Kn + 2.0 mg/mL BAP.

Keywords: *Centella asiatica*, asiatic acid, plant growth regulator, secondary metabolites

BIOTRANSFORMATION A TOOL FOR GREEN CHEMISTRY REACTIONS

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Abstract

Isolation and screening of new microbial strains which can carry out specific bioconversions is an important area of research and development. Genetically engineered microorganisms have a diversity of adapted or non-native enzymes which can be used for the manufacturing of novel steroidal metabolites. Conversion of steroids and terpenes by microbial transformation has turned out to be an important milestone in the research and development of steroidal drug industry. Thus, investigations of microorganisms from a variety of biotopes promote the chances to isolate novel biotransformed metabolites. Numerous microbial transformations of steroids and terpenes have been reported. These steroidal compounds have many important specific physiological activities depending on the functional groups, and oxidation state of these rings. The biotransformations of ethynylestradiol, cedryl acetate and funerbene by a series of fungi have been investigated. The HPLC chromatograms of fermented extracts, control cultures, and starting compound were compared in order to observe metabolic changes. The transformations result in foundation of novel and useful products that are complex to be achieved through conservative chemical techniques. It is an alternative tool for the growth of sustainable technologies for the production of chemicals and drugs, which means green chemistry.

Keywords: Green Chemistry, Biotransformation, ethynylestradiol, cedryl acetate, funerbene

ANTIFUNGAL ACTIVITY OF SOOTY MOULDS IN CO-CULTURE

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Abstract

Sooty moulds are darkly pigmented, non parasitic fungi that grow on plant surfaces. They feed on sugary exudates or honeydew secreted by sap feeding insects on plants. With sugar exudates as their nutrient source, there should be a strong competition for nutrient with other types of fungi. However, sooty moulds do not seem to be challenged for niche occupancy. A study on possibilities of sooty moulds producing antimicrobial compounds was conducted by co-culturing sooty moulds with fungi isolated from some sooty mould infested phylloplanes samples. Three sooty mould strains, S01, S04 and DPC052 were observed to have some antimicrobial effects when co-cultured with fungi A015 and B005. The interactions were observed on Malt Extract Agar and categorised as 'distant inhibition', 'contact inhibition' and 'overgrowth interaction'. The co-culture plates were extracted and analysed by HPLC. Co-cultured extracts, however, did not show very significant changes in the metabolic profiles as compared to the control strains. A simple experiment was carried out to examine the possibility of volatile compounds being released and inhibiting co-cultured species by co-incubating challenge culture and sooty moulds in separate 9-cm plates placed in a confined environment (a 20-cm plate). The growth diameters were measured and compared to control strains. However, no significant changes in colony diameters were observed. Two hypotheses can be made. First and most probably, the inhibition activities observed were not caused by volatile compounds or the method may not be suitable to analyse the presence of volatile compounds. Second, the compounds mediating the growth inhibition were not detectable through our standard HPLC-based analytical procedure that is optimised for small UV active organic compounds of intermediate polarities. Future research activities shall go beyond this limitation

Keywords: Sooty moulds, co-culture, antifungal activity.

SECONDARY METABOLISM SHIFT IN FUNGUS *TRICHODERMA VIRENS*

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Abstract

The fungus *Trichoderma virens* F1Td5c1 was isolated from a soil sample collected from a biological reserve forest, at UiTMs' Puncak Alam campus. The fungus has been previously studied in our laboratory and showed to produce two major secondary metabolites from the ethyl acetate (EA) crude extract, namely, viridiol (**1**), a furanosteroid first isolated in 1966; and trichoverinic acid (**2**), a novel sesquiterpenoid. The structures of both compounds had elucidated through extensive spectroscopic analysis. However, limited by the amount of the compound, the absolute configuration of the new compound, **2**, was not established. In an attempt to re-isolate the new compound and establish its absolute configuration *T. virens* F1Td5c1 was regrown in the same conditions as used previously. The second crude EA extract of *T. virens* also showed two major peaks after HPLC analysis and both compounds were successfully isolated. Surprisingly, the two compounds isolated the second time were different from the two compounds isolated previously. The first compound was identified as the daucane CAF-603 (**3**) (isolated in 1990). The second compound, though not fully characterized yet, showed a significantly different UV spectrum than either of the previously isolated compounds, i.e., **1** and **2**. This was further confirmed by the ¹H NMR that confirmed the presence of an ortho-substituted benzene ring, an unprecedented substructure in a *Trichoderma* secondary metabolite. This change in the major secondary metabolite production may be explained by the possibility of a genetic and/or epigenetic drift taking place due to repeated sub-culturing. That is especially true considering the fact that the second investigation is taking place few years after the first one.

Keywords: *Trichoderma virens*, secondary metabolite, metabolic shift.

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IN VITRO CHOLESTEROL ASSIMILATION BY LACTIC ACID BACTERIA: QUANTITATIVE AND QUALITATIVE APPROACHES

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Abstract

Being generally considered as a risk factor for coronary heart disease, hypercholesterolaemia is strongly correlated to high fat/ high cholesterol diet. Given the side effects that arise from the prolonged use of current drug therapy, diet modification is becoming increasingly important in the management of hypercholesterolaemia. Products containing probiotics are recognised for their cholesterol lowering benefits. The probiotic-induced cholesterol lowering effect is, however, strain-dependent. As such, this study assessed 12 lactic acid bacteria (LAB; 5 *Lactobacilli* and 7 *Pediococci*) with probiotic characteristics for their hypocholesterolaemic effect quantitatively and qualitatively. A direct quantitative high performance thin layer chromatography (HPTLC)-based method was developed and utilised for measurement of cholesterol content in supernatant fermented by LAB. Basically, the effect of LAB was explored in media with 100 µg/ mL cholesterol (designated as MRSC) or with cholesterol and 0.3% bile salt (designated as MRSBC). The HPTLC findings were then compared with that of the conventional O-phthalaldehyde method. Transmission Electron Microscopy (TEM) was adopted for qualitative observations of changes in cell wall thickness of LAB exposed to media containing cholesterol. The 12 LAB strains exhibited varying degree of cholesterol assimilation, with LAB12 (*Lactobacillus plantarum*) being the most superior strain in MRSC (cholesterol lowering by 47.57% and 53.12% in o-phthalaldehyde and HPTLC, respectively). In the presence of bile (MRSBC), the *Pediococci* strains, LAB5 and LAB7, exhibited excellent cholesterol-reducing ability. Generally, both o-phthalaldehyde and HPTLC methods showed strong and positive correlation ($r=0.9762$; $p<0.005$). Qualitative TEM study of LAB12 demonstrated an increase in cell wall thickness (MRSC=0.33µm²; MRSBC=0.31µm²) when compared to control (MRS=0.30µm²). HPTLC method may serve as an alternative mean of cholesterol quantification. HPTLC is a rapid and simpler method to screen for cholesterol reducing activity *in vitro* than the conventional o-phthalaldehyde method. The cholesterol reducing ability exhibited by LAB warrants in-depth *in vivo* study.

Keywords: lactic acid bacteria, cholesterol lowering, HPTLC, cell wall, TEM

ADULT ZEBRAFISH-BASED PARKINSON'S DISEASE MODEL WITH FULL REGENERATION OF DIENCEPHALIC DOPAMINERGIC NEURONS 30 DAYS POST 6-OHDA-INDUCED NEUROTOXICITY

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Abstract

Conventional mammalian-based Parkinson's disease (PD) models established for the study of the regeneration potential of endogenous dopaminergic neurons (DN) are often limited by ineffective axonogenesis. The emergence of the non-mammalian zebrafish model with excellent neuroregenerative properties and genomically highly homologous to that of humans may address this limitation. This study investigated a PD-model of the adult zebrafish diencephalic DN regenerative capability and identified the origin of newly generated DN. For this purpose, sexually mature zebrafish (*Danio rerio*) were lesioned by injecting 6-OHDA neurotoxin into the diencephalon region of the zebrafish brain with a microinjector in accordance to a thoroughly optimised protocol. Immunostaining protocols were also optimised for quantification of tyrosine hydroxylase immunoreactive (TH-ir) and BrdU immunoreactive (BrdU-ir) cells in brain regions of interest [olfactory bulb (OB), telencephalon (Tel), preoptic area (POA), posterior tuberculum (PT), and hypothalamus (Hyp)]. Open tank video recordings were conducted to study the locomotor activity of the zebrafish. Successful ablation of >95% of the diencephalic DN (POA, PT, and Hyp) occurred without mortality. Locomotor analysis revealed significant reduction in speed (cm/s) and distance travelled (cm) in lesioned zebrafish. Whilst cell proliferation in the diencephalon remained insignificant, transient increase of cell proliferation in OB and Tel were observed one week following 6-OHDA administrations. Thirty days post neurotoxin ablation, lesioned zebrafish showed no significant difference ($p > 0.05$) in the number of mature diencephalic DN in comparison to unlesioned controls. This study had successfully developed a stable 6-OHDA-induced PD zebrafish model using a reproducible approach and validated the model's exceptional DN regeneration potential. Further investigation using this model will yield opportunities of harnessing highly regenerative prospects of this vertebrate for future application in human DN regeneration strategy

Keywords: Adult zebrafish, Parkinson's disease, neuroregeneration, dopaminergic neurons, 6-OHDA, microinjection, immunostaining

APPLICATION OF A PARTICLE SIZE ANALYZER FOR SIZE-BASED DIFFERENTIATION OF NORMAL AND CANCER CELLS

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Abstract

Effective differentiation between normal and cancer cells can be vital in the field of cancer diagnosis and therapy. Our findings serve to demonstrate the size-based detection of cancer cells and clusters spiked in normal blood using a particle size analyzer. We found that most cancer cells have an inner diameter of over 10 μm , compared to those of normal cells. In addition, for the detection of cancer cells with sizes that are similar to those of white blood cells, we developed a PC software program that permits cancer cells to be detected based on the brightness and smoothness of the cell surface from cell images captured by the particle size analyzer. Furthermore, effect on adherent characteristics and circulating tumor cell (CTC) marker expression of cancer cells was examined after particle size analyzer measurement. These results imply the possible applicability of a particle size analyzer for detection of CTCs and CTC clusters for theranostics

Keywords: Cancer cells, detection, particle size analyzer

PRODUCTION AND CHARACTERIZATION OF THERMOSTABLE ALKALINE PROTEASE FROM *BACILLUS SUBTILIS*

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Abstract

Protease constitute 60-65 percent of global market in various industries of pharmaceutical, detergent, textile, dehairing, food processing, pharmaceutical, leathering, paper and pulp industries [1]. Production of alkaline protease received attention of researcher and industries as alkaliphic protease producing bacteria, *Bacilli* are specific producers of extracellular proteases [2]. The purpose of this study was to produce alkaline thermostable protease and characterize its condition for maximum production of protease. Seven isolated strains from La Hot Spring were screened on skim milk agar overnight which later shows clearing zone around colonies indicating maximum protease activity. A high level of protease production was found at 70°C after 24 hours of incubation at pH 11. In addition, 0.2 M to 1.0 M of NaCl was added to the protease specific medium in order to determine the effect of ionic strength in the production of protease. Production of protease was obviously noted with addition of 1.0 M NaCl. The result significantly proves that *Bacillus subtilis* isolated could produce alkaline protease which highly produced in temperature more than 50. Previous report by Johnvesly and Naik showed the production of thermostable protease from alkaliphic and neutraphilic bacterial [3]. This research is usable and helpful to industries by which it could help to maximise production of protease without trouble to adapt *Bacilli* at high temperature.

Keywords: thermostable protease; *Bacillus subtilis*; protease activity; alkaline protease; protease production

EFFECT OF MISSENSE MUTATIONS IN ORNITHINE TRANSCARBAMYLASE ENZYME (OTCase): MOLECULAR DOCKING AND MOLECULAR DYNAMICS SIMULATION ANALYSIS

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Abstract

Ornithine transcarbamylase enzyme (OTCase) plays a crucial role in urea cycle to synthesize urea from ammonia. Mutations of the enzyme cause various clinical effects which result in neurological symptoms or death. To date, more than 400 mutations are reported, and majority of those are missense mutations. In this study, we investigate the structural effects of OTCase missense mutations that located in the catalytic pocket. *In silico* webserver, molecular docking and molecular dynamics simulation were employed to predict the effect of ligand binding. Results from five *in silico* webserver exhibited that missense mutations were pathogenic. Docking of mutant models with PALO, an analog of N-phosphonacetyl-L-ornithine resulted to minimally displaced the ligand when compared to the native OTC structure. Further studies using molecular dynamics simulations revealed the mutations apparently disrupted interactions between the binding pocket residues and PALO. In conclusion, these findings suggest that the missense mutations located in the catalytic pocket potentially cause the occurrences of OTC deficiency

Keywords: OTCase, mutation, catalytic pocket, docking, molecular dynamics.

MORPHOLOGICAL CHARACTERISTICS OF MARINE DIATOMS ISOLATED FROM PANTAI REMIS

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Abstract

Known as the major component of phytoplankton, diatoms contribute around 40% of the total primary carbon production in the ocean. Diatoms are microalgae that naturally synthesize intricate and unique cell wall made up of silica. This cell wall is called frustules. The unique patterns of frustules allow the classification of diatoms to be made through morphological characterization. This paper documented morphological identifications of diatoms isolated from Pantai Remis, Kuala Selangor. The water samples were collected in February 2015 and July 2016. Multiple series of serial dilutions and agar plating were done to isolate diatoms from the water samples. Isolated diatoms were cultivated in f/2 liquid medium for three weeks. Prior to examination under the scanning electron microscopy, the samples were washed with 1M hydrochloric acid to remove impurities and organic materials off the diatoms frustules. A total of eight species from seven genera were identified from the samples namely, *Amphora*, *Cavinula*, *Navicula*, *Nitzschia*, *Pleurosigma*, *Skeletonema* and *Thalassiosira*. From all the samples, there are six pennate diatoms and two centric diatoms. The most distinct feature of pennate diatoms which distinguish them from centric diatoms is the raphe system. SEM examinations enable morphological characterization of the isolated diatoms thus expanding the knowledge of diatoms diversity in Pantai Remis, Kuala Selangor.

Keywords: Scanning electron microscopy, light microscopy, morphological characteristics, pennate diatoms, centric diatoms

PRODUCTION AND DETERMINATION OF GLUCOSAMINE FROM CHITOSAN OF MUD CRAB SHELL

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Abstract

Glucosamine is a compound naturally found within the cartilage of our joints, made from chains of sugars and proteins bound together. Glucosamine possesses natural anti-inflammatory and anti-aging properties. It is generally produced by the hydrolysis of the exoskeletons of marine animals, one of the types of mud crab (*Scylla serrata* Forskal). In the mud crab shells contain chitin and chitosan compounds that can be derivatives further into glucosamine HCl. This study aimed to isolate and determine the level of glucosamine HCl by hydrolysis of chitosan from mud crab shell. Glucosamine HCl was hydrolyzed chemically using concentrated hydrochloric acid. Characterization glucosamine HCl has done by Fourier Transform Infra Red (FTIR) spectrophotometry, while the determination of levels has done by Uv-Vis spectrophotometry. Both of stage uses Glucosamine HCl standard as a comparison. The results of the qualitative analysis showed a content of glucosamine HCl from sample. It was characterized by the spectrum appears at wave number 1033 cm^{-1} , 1532 cm^{-1} , 3415 cm^{-1} that each showed their vibration ether (-O-), etil (CH₂) and amine group (NH). Match factor (MF) price is 933, which shows that the resulting spectra are identical. The results of the quantitative analysis showed that the levels of glucosamine is 553 mg/g. It can be concluded that the waste of mud crab shell has a potential to produce glucosamine.

Keywords: Glucosamine HCl, mud crab, chitin, FT-IR spectrophotometry, Uv-Vis spectrophotometry

CLASSIFICATION OF TWILIGHT ZONE PROTEINS USING STRUCTURE-BASED PHYLOGENETIC ANALYSIS

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Abstract

Protein classification is important to predict unknown structures, functions and the identification of novel proteins. Conventionally, proteins are classified based on their sequence data. However, the sequence-based approach often fails to detect proteins with low sequence similarity resulting in high fractions of false negatives in the clustering. In contrast, a structural approach is the most suitable method as twilight zone proteins are highly similar in structure. To study the homology between proteins of low sequence similarity, structural alignment tools TM-align and FATCAT were chosen as comparison to evaluate proteins defined in the four main SCOP classes - All- α proteins (Class A), All- β proteins (Class B), All α/β proteins (Class C) and All $\alpha+\beta$ proteins (Class D). The Unweighted Pair Group Method with Arithmetic Mean (UPGMA) approach was used to evaluate structural similarity of the proteins and classify them through the construction of phylogenetic trees. Comparison of protein clusters against the SCOPe classification database shows remarkable results for each of the tools. Besides a visual comparison of the trees, the Adjusted Rand index was applied as a statistical approach for validating the clustering of protein that implies the accuracy of the alignment tools in identifying protein homology. The results shows that protein clusters produced from TM-align alignments confer the best trees for all classes at their superfamily level as compared to FATCAT. Meanwhile, the evaluation of clusters at the family level shows that TM-align offers the best results for classifying proteins in Class A, B and D whereas results from FATCAT flexible alignments are the most prominent for proteins in Class C. Thus, it can be concluded that TM-align can be used for evaluating structures for proteins classified in Class A, B and D whereas, FATCAT flexible alignment can be implemented to examine proteins identified in Class C.

Keywords: Twilight zone proteins, structure-based phylogenetic analysis, structural protein alignment, protein classification

VALIDATION OF POTENTIAL ALZHEIMER'S DISEASE BLOOD-BASED BIOMARKERS

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Abstract

Alzheimer's disease (AD) is expected to increase with the growing ageing population in Malaysia. Biomarkers are needed to improve current diagnosis and monitoring of disease progression. The use of potential AD biomarkers identified from previous studies in clinical settings is, however, prevented by a number of limitations. The acquisition of cerebrospinal fluid as potential source of AD biomarkers is invasive. Neuroimaging biomarkers are expensive and poorly available in hospitals. Blood-based transcription factors, which can be easily accessible, may potentially fulfill the requirement of AD biomarkers. By using transcriptomics, our preliminary study, which involved a test cohort of 180 individuals, had identified sestrin1 (SESN1), CCR4-NOT transcription complex subunit 8 (CNOT8) and mitogen-activated protein kinase 1 (MAP2K1) genes as potential AD biomarkers. Expression of these genes was found to be increased in probable AD patients with gene expression ratio of 3.90 (SESN1), 2.38 (CNOT8) and 2.31 (MAP2K1) when compared to healthy subjects. The present study aimed to further validate these genes in a different cohort of 11 probable AD patients and 9 healthy individuals by qRT-PCR. Total RNA were extracted from whole blood, reverse-transcribed and gene expression was quantified using qRT-PCR. Gene expression of SESN1, CNOT8 and MAP2K1 was increased in probable AD compared to healthy subjects with ratio of 0.63, 0.85 and 3.10, respectively. Amongst the three genes, the gene expression ratio of MAP2K1 was ≥ 2 ($p < 0.05$), which is in agreement with our preliminary results. Both SESN1 and CNOT8 genes, however, yielded expression ratios of ≤ 2 ($p > 0.05$). Small validation cohort and possible degradation of samples due to long term storage may have affected the reproducibility of the gene expressions. It is therefore essential to increase the population size of validation cohort and collect fresh blood samples to ensure reproducible expression of potential AD-related gene biomarkers

Keywords: Alzheimer's disease, biomarker, gene expression, blood, qPCR.

MORPHOLOGICAL AND MOLECULAR IDENTIFICATION OF MALAYSIAN MARINE ENDOPHYTIC FUNGI

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Abstract

Marine endophytic fungi have been proven to be the most promising source of bioactive compounds for drug discovery. Despite the proven significance of active metabolites of marine endophytic fungi, a little is exploited. Therefore considerable study on marine endophytic fungi progress in exploring their diversity, species richness and bioprospecting should be conducted. Marine endophytic fungi isolated from local seaweeds in Teluk Kemang, Negeri Sembilan, Malaysia exhibited anticancer, antibacterial, antifungal, and wound healing properties. In this present study, 18 isolates of marine endophytic fungi derived from eight species of local seaweeds (*Gracilaria arcuata* Zanardini, *Gracilaria coronopifolia* J. Agardh, *Chaetomorpha minima* F.S Collins & Harvey, *Caulerpa sertularioides*, *Acanthophora spicifera* M. Vohl, *Padina minor* Yamada, *Enteromorpha compressa* and *Caulerpa lentifera*) were investigated. The isolated marine endophytic fungi were identified through morphology and molecular technique. The DNAs of the marine endophytic fungi were amplified using ITS 1 and ITS 4 as primers by conventional polymerase chain reaction (PCR) method. Out of 18 marine endophytic fungi, 11 of them were successfully identified. Six marine endophytic fungi (CN, CS1, CS2, ED1, MV and PA1) were dominantly identified as *Aspergillus* sp., whereas another five (UF, ED2, PA2, PA7, CR3) have closest Genbank similarities with *Exophiala dermatitidis*, *Diaporthe pseudomangiferae*, *Psathyrella purpureobadia*, *Arthrimum xenocordella* and *Phanerochaete carnosus* respectively. Results presented from this present study proved that the local seaweeds harbor various species of marine endophytic fungi and might be valuable for the local database on marine endophytic fungi in Malaysia.

Keywords: Marine endophytic fungi, seaweeds, molecular identification, ITS 1, ITS 4

SCREENING OF FERMENTATION PROCESS PARAMETERS AFFECTING BIOMASS PRODUCTION OF *LACTOBACILLUS PLANTARUM* LP5

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Abstract

Lactobacillus plantarum LP5 has a potential to be developed as a commercial probiotic especially for the dairy industry. A fractional factorial design was applied to identify the significant fermentation factors for the biomass production of *Lactobacillus plantarum* LP5, based on two-level statistical approach. Selected factors are the pH (5-6), stirring rate (100-150 rpm) and temperature (35-37°C) were evaluated to maximise the growth of *Lactobacillus plantarum* LP5. The screening results showed that the pH and stirring rate, in the range studied had significant effects ($p < 0.05$) compared to the temperature for higher biomass production. Analysis of variance (ANOVA) derived an established model at the significant value ($p = 0.0466$) and the determination of coefficient ($R^2 = 0.9935$) and indicated that at low level of pH (pH 5) and stirring rate (100 rpm) is the best combination to maximise the growth of *Lactobacillus plantarum* LP5 in batch fermentation culture. The results of this study guide to developing a model to optimise and predict the response.

Keywords: Screening, fractional factorial design, fermentation, *Lactobacillus plantarum*

AGMATINE PROVIDES NEUROPROTECTION IN A 3-NITROPROPIONIC ACID (3-NP)-INDUCED EXPERIMENTAL HUNTINGTON'S DISEASE

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Abstract

Huntington's disease (HD) is an inherited genetic disorder, caused by abnormally expanded and unstable CAG repeat within the coding region of the gene encoding the huntingtin protein. This disease can be characterized by cognitive dysfunction and abnormal body movements called chorea. 3-nitropropionic acid (3-NP) has been widely used as neurotoxin to induce neurodegeneration in the animal model of Huntington's disease (HD). The major deleterious event in 3-NP-induced neuronal apoptosis is caused by the increase of oxidative stress and reduced antioxidant level in brain. The present study aimed to investigate the protective effect of agmatine on 3-NP-induced neurodegeneration through motor coordination activity in Wistar rats and estimate the level of biochemicals in the brain. Agmatine is the metabolite of arginine by arginine decarboxylase. Wistar rats were administered 3-NP (10 mg/kg i.p.) thrice on day 1, 5 and 9. Agmatine (40 and 80 mg/kg) was given intraperitoneally, once a day, from day 9 to day

Body weight and behavioral parameters (grip strength test and rotarod activity) were assessed on 1st, 5th, 9th and 15th-day post-3-NP administration. Animals were sacrificed on day 15, and brains were harvested. The administration of 3-NP showed reduction in body weight, declined motor function and changes in the level of biochemical in 3-NP treated animals. Rats administered with 3-NP showed elevation in the level of lipid peroxidation and nitrite, which significantly decreased upon agmatine treatment. 3-NP-induced rats showed a decrease in the activity of reduced glutathione, which slightly increased upon agmatine treatment. There is also a fall in the level of SOD in the 3-NP induced rat, which rose in the agmatine treatment group. As a conclusion, the present results showed that agmatine could exert protective action over 3-NP and might be potentially used as a neuroprotective agent.

Keywords: Huntington's disease, 3-nitropropionic acid, agmatine

MITOCHONDRIAL DYSFUNCTION IN AGING MODEL (IN VIVO) INDUCED WITH D-GALACTOSE AND ZINC: IMPLICATIONS FOR ALZHEIMER DISEASE AND NEUROPROTECTIVE EFFECT OF CEFTRIAXONE

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Abstract

Zinc (Zn) is an essential trace element present abundantly in the brain. Despite its importance in the normal brain functions, excess of Zn is neurotoxic and may cause neurodegeneration following the transient global ischemia. This indicates that Zn may play a crucial role in the pathogenesis of Alzheimer's disease (AD). Similarly, a surplus of glutamate has been implicated in the development of AD. In this study, we focused on the mitochondrial dysfunction due to excess of Zn in the brain. Mitochondrial dysfunction has been previously implicated as a factor in the pathogenesis of AD. We induced accelerated senescence through subcutaneous injection of d-galactose and oral administration of Zn daily for six weeks. Treatment with the standard AD drug donepezil was compared with ceftriaxone (CTX), a beta lactam antibiotic with neuroprotective activity. Results showed CTX provides protection against d-galactose- and Zn-induced toxicity. The degree of mitochondrial dysfunction in donepezil and CTX treated mice was lower than untreated animals. CTX provides protection against d-galactose and Zn induced toxicity by attenuating mitochondria dysfunction in this model.

Keywords: Zinc, Neurotoxicity, Alzheimer Disease, D-galactose, Ceftriaxone

SCREENING AND REVALIDATION OF *ADENOMATOUS POLYPOSIS COLI* (APC) GENE POLYMORPHISM USING ALLELE-SPECIFIC POLYMERASE CHAIN REACTION (ASPCR) METHOD

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Abstract

Adenomatous polypsis coli (APC) gene polymorphism is a type of hereditary colorectal cancer (CRC) that is associated with familial adenomatous polypsis (FAP). FAP is important in regulating cell growth. Therefore, the mutation of APC gene causes the cell to grow irregularly and consequently prone to the development of CRC. The objective of this research is to screen and revalidate the APC gene polymorphism detection in human subjects by using the allele-specific polymerase chain reaction (ASPCR) method. Ten participants were recruited based on the inclusion and exclusion criteria. The blood obtained was extracted for DNA using NucleoSpin® Blood extraction kit. The DNA was amplified using ASPCR and the gene polymorphism was detected by observation of bands using gel documentation system. Bands appeared at 97 bp indicated the internal control used while other distinct bands at 434 bp determined the samples' variant. None of the samples showed heterozygous variant type. The ASPCR method is suggested as one of the variant detection techniques for APC gene polymorphism. However, DNA sequencing is recommended to further validate the obtained results.

Keywords: Adenomatous polyposis coli (APC), Colorectal cancer (CRC), Familial adenomatous polyposis (FAP), Allele specific polymerase chain reaction (ASPCR)

SCREENING OF MUTL HOMOLOG 1 (MLH1) GENE POLYMORPHISM USING POLYMERASE CHAIN REACTION (PCR) AND DIRECT SEQUENCING METHODS

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Abstract

Colorectal cancer (CRC) has become the third most common cancer in the world including Malaysia. According to Center for Disease Control and Prevention (CDC), the risk of developing CRC is higher in individuals who have a family history of CRC. The most common hereditary CRC syndromes is Lynch syndrome which is recognized and known as inherited germline mutations of DNA mismatch repair (MMR) genes such as *MLH1* and *MSH2*. To screen and detect the *MLH1* gene polymorphism among the high risk participants by using PCR and direct sequencing methods. PCR method was optimised for amplification of the *MLH1* gene upon DNA extraction. The DNA yield are satisfactory for all the samples (40 to 60 ng/μL with purity A260/280 ranges from 1.8 to 1.95). Following the DNA sequencing analysis, a single point mutation G>A was detected on the *MLH1* gene. Preliminary detection of CRC in *MLH1* gene was successfully identified using the PCR and direct sequencing methods.

Keywords: Mutl Homolog 1 (MLH1), Colorectal cancer (CRC), Mismatch repair (MMR), Polymerase chain reaction (PCR)

LACTOBACILLUS PLANTARUM LAB12 MEDIATE CROSSTALK BETWEEN ENS AND CNS THROUGH MODULATION OF GUT MICROBIOTA

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Abstract

Microbiota dysbiosis is closely linked to neuroinflammation via the Gut-Brain Axis. Intake of probiotic lactic acid bacteria (LAB) could potentially correct dysbiosis of microbiota and ameliorate inflammation in the CNS. Our behavioural study found lipopolysaccharide (LPS)-challenged rats, especially those being fed with *Lactobacillus plantarum* LAB12, to exhibit improved memory. This study therefore explored the differential shift of microbiota diversity between LAB12-fed and control LPS-challenged rats. Briefly, caecal content was collected from LPS-challenged (0.25 mg/kg; *i.p* from day 28-31) Sprague Dawley rats (male, 3 months-old; n=6/ group) fed with either commercial feed or that supplemented with LAB12 (10⁹ CFU/ mL; oral gavage) for 31 days. Bacterial 16S rRNA gene of V3 to V4 region was amplified using 515F-806R primers. All PCR reactions were carried out using the Phusion® High-Fidelity PCR Master Mix. Sequencing libraries were generated using TruSeq® DNA PCR-Free Sample Preparation Kit. The library was sequenced on an Illumina HiSeq 2500 platform and 250 bp paired-end reads were generated. Sequences assembled were analysed with Uparse Software for species annotation. In LAB12-fed rats, species abundance heatmap showed two major phyla, *Bacteroidetes* (69%) and *Firmicutes* (14.25%), with small percentages of *Proteobacteria* (5.75%), *Actinobacteria* (0.0825%), *Spirochaetes* (4.425%), *Deferribacteres* (0.4425%) and *Euryarchaota* (4.325%). Control LPS-challenged rats showed varying percentages of phyla, *Bacteroidetes* (47.5%) and *Firmicutes* (41.25%), with small percentages of *Proteobacteria* (5.75%), *Actinobacteria* (0.25%), *Spirochaetes* (2.425%), *Deferribacteres* (0.014%) and *Euryarchaota* (1.475%). LAB12 significantly ($p<0.001$) increased *Bacteroidetes:Firmicutes* ratio when compared to the control rats. Principal Component Analysis plot showed clear separation and less variation of bacteria present in LAB12-fed compared to control rats. The present study indicates that LAB12 is able to alter gut microbiota resulting in improved memory.

Keywords: Gut-Brain Axis, Probiotics, Gut Microbiota, Diversity

EVALUATION OF LUCIFERASE REPORTER VECTOR CONTAINING HYPOTHETICAL MicroRNA BINDING SITES FROM PRIMARY *hsa-miR-195-497* TO ISOLATE NOVEL MicroRNA

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Abstract

MicroRNAs (miRNAs) are small non-coding, endogenous RNAs that are important in being the post-transcriptional regulators of gene expression in many organisms. Complementary binding of a miRNA implies either tumour suppression or oncogenic activation. Recent studies in hepatocellular carcinoma (HCC) show that miRNA-195 and miRNA-497 which are transcribed as one primary transcript were endogenously down-regulated in HCC cell lines. This study focuses on the introduction of reporter vector containing the hypothetical miRNA binding sites from the *hsa-miR-195-497* gene cluster to allow identification of putative microRNAs that are responsible to regulate the primary transcript. The *hsa-miR-195-497* gene sequence was obtained from NCBI database and divided into several parts with approximately 700bp in length each. These sequences were then inserted separately at the downstream of luciferase gene in the reporter vectors. Ligations of inserts were validated by colony PCR and sequencing analysis. The reporter vectors carrying desired regions were then transfected into mature HepG2 cell line with respective controls. A *Dual-Glo® Luciferase Assay System* was used to measure the activity of any particular endogenous miRNAs. Firefly luciferase expression was significantly decreased in region where there are miRNA-195 and miRNA-497 sequences in comparison to the control. Ultimately, the efficiency of antisense oligonucleotide towards the identified putative miRNA can be evaluated to produce oligonucleotide-based drug for combating cancer

Keywords: microRNA, hepatocellular carcinoma (HCC), luciferase assay

PRELIMINARY STUDY ON POTENTIAL NEUROPROTECTIVE EFFECT OF *CENTELLA ASIATICA* EXTRACT ON METHAMPHETAMINE-INDUCED NEUROTOXICITY IN SH-SY5Y CELL LINE

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Abstract

Natural product have been widely used as potential therapeutic agent due to its vast availability, cost-efficient and less adverse effect. *Centella asiatica* or *pegaga* is remarkably known on its antioxidant and neuroprotective properties. Methamphetamine (MA) is illicit psychostimulant drug with neurotoxic effect with persistent dopaminergic neuron which is mimic the pathological of Parkinson's disease. However, neuroprotective effect of *Centella asiatica* extract (CAE) on Methamphetamine-neurotoxicity not well studied. Objective of this preliminary study is to evaluate neuroprotective effect of *Centella asiatica* extract against Methamphetamine- induced neurotoxicity on human neuroblastoma, SH-SY5Y cell line. MA was exhibit IC50 value of 1mg/mL and was used to kill and damaged the cells. Neuroprotection test was conducted using different concentrations of CAE (1 pg/mL, 10 pg/mL, 100 pg/mL, 1 ng/mL, 10 ng/mL, 100 ng/mL, 1 µg/mL, 10 µg/mL, 100 µg/mL, 1 mg/mL) for 24h at 37°C. Cell viability was quantitatively evaluated through MTS assay. From the results obtained, with the increasing of CAE concentrations, there was significant increase in cell viability percentage compared to cell treated with MA only. Thus, we showed that CAE has promising potential against Methamphetamine-neurotoxicity. In conclusion, results from this preliminary study showed that *Centella asiatica* extract acts as potential natural herb extract for treatment of Methamphetamine-neurotoxicity and neurodegenerative disease.

Keywords: *Centella asiatica* extract, Methamphetamine, neuroprotection, SH-SY5Y cell line

EFFECT OF CORTICOSTERONE ON ESTROUS CYCLE IN FEMALE RATS

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Abstract

The onset of puberty in female rats leads to ovarian maturation (1). It will lead to reproductive cyclicity known as estrous cycle. Average estrous cycle length in rats was 4.8 day (2). Normal sequence of estrous cycle in female rat consists of four stages known as proestrous, estrous, diestrous I and diestrous II (3). Different phases in estrous cycle can be detected by examining vaginal cytology (4). The method for determination of the estrous cycle was done through observation of cells obtained from vaginal smearing. Estrous cycle ceases during pregnancy, pseudo-pregnancy and lactation. Corticosterone is a stress-related hormone which released by adrenal gland when animals were subjected to stress. Stress induced activation of the hypothalamus-pituitary-adrenal axis (HPA) which cause corresponding increase in circulating level of glucocorticoids (5) and results in the suppression of the hypothalamus-pituitary-gonadal axis (HPG) which may lead to reproductive dysfunction in female animals such as irregularity of cycle (6). The present study aims to investigate the effect of corticosterone on the estrous cycle of the female rat. Cyclic Sprague Dawley rats were used in this experiment. On the second proestrus, animals were injected with corticosterone 25mg/kg body weight/day for 4 consecutive days. Determination of estrous cycle was continuously done until the next proestrus. Results obtained showed that animals from control group showed four-days cycle whereas animals from corticosterone-treated group showed five to seven-days cycle. In conclusion, injection of corticosterone 25mg/kg body weight/day caused prolonged estrous cycle in female rats.

Keywords: Corticosterone, Estrous Cycle, Female Rats

EFFECTS OF ADIPONECTIN ON IL-6 SECRETION ON VASCULAR CALCIFICATION AND PROLIFERATION

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Abstract

Secretion of interleukin 6 (IL-6), a pro-inflammatory cytokine, is thought to induce vascular calcification during the late stage of atherosclerosis. Adiponectin, an anti-inflammatory adipokine secreted by adipocytes, has been proven to exert many beneficial activities against the progression of atherosclerosis. However, there are some reports that it might increase IL-6 secretion from cells in the arteries. Little is known about the effects of adiponectin on IL-6 expression, vascular calcification and proliferation of human vascular smooth muscle cells. Therefore, this study aims to investigate the effect of trimeric adiponectin on human adult aortic vascular smooth muscle cells (HAoVSM) in terms of IL-6 secretion, cell proliferation and calcification. HAoVSM were incubated with different concentrations of adiponectin (2.5, 5.0, 10.0 and 15.0 µg/mL) for 24 hours, while HAoVSM cells incubated with lipopolysaccharides (LPS) served as a positive control. The secretion of IL-6 and calcification were quantified using eBioscience™ Human IL-6 Platinum ELISA kit and Calcium Colorimetric assay (Sigma) respectively. While cell viability was measured using CellTiter 96® Aqueous One Solution Cell Proliferation Assay (MTS). The incubation of HAoVSM with adiponectin (2.5 - 15.0 µg/mL) showed with >90% no loss of cell viability. HAoVSM incubated with the same range of adiponectin (2.5 - 15.0 µg/mL) however, showed a dose dependent increase of IL-6 protein expression when compared to unstimulated cells where both 10.0 and 15.0 µg/mL concentration show significant result. This increase was accompanied by an increase in calcium release and the proliferation of the adiponectin. However, both calcium release and proliferation rate, were not significant when compared to untreated cells. Although adiponectin lead to an increase of IL-6 secretion by HAoVSM cells, this was not causing vascular calcification and/or vascular smooth muscle cell proliferation. Therefore, we suggest that IL-6 is not acting in an autocrine but rather in a paracrine fashion. However, further studies need to be done to investigate the role of IL-6 in HAoVSM cells.

Keywords: Adiponectin, Interleukin-6 (IL-6), Calcification, MTS

THE IMPACT OF GENETICS AND OTHER FACTORS ON THE INTER-INDIVIDUAL AND INTER-ETHNICITY DIFFERENCES IN CYP2C9-CATALYSED METABOLISM

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Abstract

Wide inter-individual differences were observed in the metabolic ratio of losartan to its metabolite E-3174. This study investigates the molecular reasoning and other potential contributing factors behind inter-individual CYP2C9*1/*1 metabolism using losartan as a probe. Realising that there is also an inter-ethnicity difference in CYP2C9 metabolism, we compared two ethnic groups, Koreans and Swedes. Observing a low metabolic activity in Turkish with Behcet's disease, we also investigated for a link between the disease and CYP2C9 activity. The general aim of this study was to further understand the pharmacokinetic variability of CYP2C9 catalysed drug metabolism. 148 healthy Swedish, 146 healthy Korean, 52 Behcet's diseased patients and 23 healthy Turkish subjects were genotyped for known *CYP2C9* defective variant alleles (*CYP2C9**2, *3). *CYP2C9* phenotyping was done using a single oral dose of 50 mg losartan. Excluding oral contraceptive (OC) users and carriers of *2C9**2 and *3 alleles, 117 Korean and 65 Swedish were genotyped for *POR**5, *13 and *28 using Taqman assays. The urinary metabolic ratio (MR) was calculated with the ratio of losartan to its metabolite E-3174. Intronic variant *CYP2C9*, *IVS8-109A>T* was observed to cause a lower *CYP2C9* activity in Swedes but not Koreans. Besides *CYP2C9* genotype, ethnicity is nominated as a significant factor. Interestingly, Swedish individuals who carry *POR**28 allele were observed to show a 1.40 fold increase of *CYP2C9* activity. In addition, we did not find evidence of *CYP2C9* genotype and medication having any influence on the observed low *CYP2C9* metabolic activity in Behcet's diseased subjects. It is very possible that inflammation response agents caused an inhibitory effect on their *CYP2C9* activity. The contributor to variations of *CYP2C9* activity is mainly genetics. Other factors such as oral contraceptives and anti-fungal medications, and comorbidities are very important to take account for. This study has found that the genotypes of *CYP2C9* and *POR* genes are important to determine the initial metabolic activity of the *CYP2C9* enzyme

Keywords: CYP2C9, POR*28, losartan metabolism, pharmacogenetic, inter-ethnic variation

IDENTIFICATION OF PURINE-RICH SITE FROM UPSTREAM OF miRNA17-92A SEQUENCE FOR A POTENTIAL TRIPLE-HELIX FORMING OLIGONUCLEOTIDES (TFO) TARGET SITE

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Abstract

Most of the choices for cancer treatments recently may affect the surrounding healthy cells. The application of oligonucleotide-based drugs such as triple helix-forming oligonucleotides (TFO) to inhibit the production of oncogenic microRNA (i.e. miR17-92a) may open promising strategies for an accurate treatment to suppress cancer development. The triple helix is sequence-specific and formed by the addition of the third strand (poly-purine DNA strand) to the major groove (purine-rich sequence) of the double helix DNA through reverse-Hoogsteen base pair. Moreover, these antiparallel reverse-Hoogsteen structures are generally independent of pH and it is practical for normal physiological condition. The binding of TFO is expecting to inhibit the transcription and replication processes, thus inhibits the target gene. This preliminary study generally described the process of identifying the purine-rich site in the genomic DNA of HepG2 cancer cell line. 18 bp purine-rich sequences were identified at the upstream of miRNA17-92a from the NCBI database. Appropriate PCR primers were designed using NCBI Primer Designing Tools. The expected size of PCR product was successfully obtained through agarose gel electrophoresis analysis and direct sequencing was performed from two directions using the same forward and reverse PCR primers to verify the sequence of this potential TFO target site. Sequencing results showed that purine-rich site was successfully verified at the upstream of this oncogenic miRNA. With a series of optimizations of TFO binding affinity i.e. denature PAGE analysis and *in vitro* analysis, this potential TFO binding site might be useful therapeutic target site to suppress the synthesis of the unwanted miR17-92a oncogenic miRNA and this finding can also be optimized for any related genetic diseases.

Keywords: triple helix-forming oligonucleotides (TFO), reverse-Hoogsteen base pair, miRNA, PCR, HepG2 cancer cell line

GENE EXPRESSION PROFILING OF PERIPHERAL BLOOD IN MILD COGNITIVE IMPAIRMENT AND ALZHEIMER'S DISEASE

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Abstract

The development of biomarkers for Alzheimer's disease (AD) is hampered by lack of understanding of the early pathogenic mechanism underlying AD. To advance knowledge on potential biomarkers, gene expression data (microarray) from patients with mild cognitive impairment (MCI) and Alzheimer's disease was generated. Peripheral blood samples were collected from patients with MCI and AD, as well as age-matched healthy control subjects. Global cognitive performance was assessed with the Mini-Mental State Examination (MMSE). The total RNA was isolated and microarray experiment was carried out with one array per sample. Validation using quantitative reverse transcriptase-polymerase chain reaction was performed on DDIT4 and SESN1 genes that were previously shown to be dysregulated in AD. The expression pattern of mTOR and p53 signaling genes, namely DDIT4 and SESN1 were found to be upregulated in the peripheral blood of MCI and AD patients. Using both the genes, a decision-tree prediction model was generated for a test cohort of patients yielding an accuracy of 80.0% of the MCI group, while sensitivity of 86.7% was obtained for the AD group. These results provide rationale for further studies on the discovery and validation of candidate mRNAs in peripheral blood and may potentially lead to the development of a non-invasive and sensitive test for AD diagnosis and prognosis.

Keywords: Gene Expression; Alzheimer's disease, Peripheral blood cells, Mini Mental State Examination.

BIOCHEMICAL STUDY OF DIABETIC WOUNDS TREATED WITH MALAYSIAN *GELAM* HONEY

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Abstract

Impaired wound healing is one of the complications associated with diabetes as it increases the susceptibility of patients to infection. Evidence indicates that Malaysian *Gelam* honey exerts wound healing activity, attributed to its antioxidant, anti-inflammatory and antibacterial properties. Therefore, it may be worthwhile to study the efficacy of *Gelam* honey on the healing of diabetic wounds. This study investigates the biochemical effects of topical administration of *Gelam* honey using incision wound model in streptozotocin (STZ)-induced type I diabetic rats. Two full-thickness incisional wounds (3 cm long) were made parallel to and 2 cm from the midline of the dorsum in 48 male Sprague-Dawley rats. The animals were divided into four groups of 12 animals each. Group I was diabetic rats treated with *Gelam* honey, Group II was diabetic rats treated with silver sulfadiazine (SSD), Group III was diabetic rats control group and Group IV served as the non-diabetic control group. On days 1 and 14 post-wounding, six rats from each group were euthanized and the skin samples were taken for biochemical analysis. *Gelam* honey increased cellular proliferation and collagen synthesis at the wound site, as evidence by significant increase in the levels of collagen, hexosamine, protein and uronic acid when compared to other experimental groups. The presence of biologically active components such as flavonoids and phenolic acids may readily account for the observed wound healing activity of *Gelam* honey. The results suggest that topical application of *Gelam* honey promotes tissue repair by accelerating collagen production and promoting overall connective tissue stability in healing wounds of diabetic rats.

Keyword: *Gelam* honey, diabetes rats, wound healing, biochemical, incision wound

FULL-LENGTH CLONING AND STRUCTURAL CHARACTERISATION OF A NOVEL mRNA IN A549 LUNG CANCER CELL LINE BY RACE-PCR

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Abstract

Lung cancer remains as the most common cause of cancer-related death with nearly 1.6 million deaths worldwide in 2012. Meanwhile, in Malaysia, lung cancer had been reported as the commonest cancer in males. To date, there is still no cure for lung cancer disease and the disease is usually detected at the last stage. Most recent treatment for lung cancer is the oncogene-directed targeted therapy which targets specific molecular oncogene driver. In the future, there will be identification of many more of specific molecular targets that are involved in lung cancer development which may lead to the development of effective targeted therapies. Rapid amplification of cDNA ends (RACE) is an inexpensive and powerful tool to amplify the full length cDNA when only partial sequence of the transcript is known. In a previous study, we managed to isolate a few novel transcripts from A549 lung cancer cell lines. Therefore, this study is aim to clone the full-length of the selected transcript by rapid amplification of cDNA ends (RACE) and perform preliminary assessment of gene function by RNA interference gene-knockout experiment. The complete sequence was identified to match to *Homo sapiens* lemur tyrosine kinase 2 (LMTK2), RefSeqGene on chromosome 7, RefSeq_mRNA of LMTK2 and RefSeq LMTK2 Transcript Variant X1 mRNA. Full-length amplification of C13 sequence resulted in 2176 bp with open reading frame of 302 amino acids. Based on the full length sequence of the selected transcript, it could be classified as a novel gene or non coding RNA. A RACE experiment successfully identified the 3'end and 5'end of the selected sequence. Moreover, the sequence obtained was successfully validated using full length PCR on the C13 sequence. Based on the data obtained from the present study, the selected transcript could be a bona fide mRNA, lincRNA or a pseudogene.

Keywords: Lung cancer, A549, RACE, full-length, novel, oncogenic

CONTROL OF RABIES OUTBREAK IN NORTHERN PENINSULAR MALAYSIA

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Abstract

Rabies causes 70,000 deaths annually worldwide. Forty per cent (40%) of people who are bitten by rabid animals are children under the age of eighteen (18) years. Rabies is seen in raccoons, skunks, bats, and foxes in the wild. However in Malaysia, there are no reported cases from the wild and zoo animals. Rabies is caused by Lyssa viruses and spreads when an infected animal scratches or bites another animal or human. Saliva from an infected animal can also transmit rabies when comes into contact with the mouth, nose, or eyes. Overall dogs are the most common animal involved. More than 99% of rabies cases in countries where dogs commonly have the disease are caused by dog bites. The 2015 outbreak in Perlis, Kedah and Penang was alarming to the public as more than 2000 dogs were culled. These states are prone to rabies since the southern Thailand is endemic to rabies. The control of rabies in animal reservoirs depends on control of rabies in dogs. To achieve this, we need co-operation from various agencies. International organisations like WHO, OIE and CDC advocate vaccination. Taking into religious and ethnical consideration, culling is not acceptable. Currently, the strategies employed for the control of rabies in dogs primarily include pre-exposure vaccination and birth control programs to tackle overpopulation of street dogs. This is being accomplished through the involvement of governmental and non-governmental organizations, municipal corporations, and animal welfare organizations. Other approaches are registration of owned pet dogs and their vaccination against rabies annually, compulsory pet licensing, increasing the public awareness (through educational institutions, media, and co-operation) about prevention and control of rabies and the benefits of successful mass anti rabies vaccination. This paper emphasis the 10-day quarantine, 6-month quarantine, diagnosis in dogs and recommended control programmes.

Keywords: Rabies, stray dogs, vaccination, culling, quarantine

ENVIRONMENTAL ENRICHMENT ON MICE AND RATS AT THE LABORATORY ANIMAL FACILITY AND MANAGEMENT (LAFAM), UiTM SELANGOR

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Abstract

Environmental enrichment is a new field in laboratory animals in Malaysia. It is commonly practiced in companion and zoo animals. A suitable enrichment may enhance the well-being of rodents; thereby it helps in refining animal models and improving research data. This study is beneficial for rodents since enrichment may create opportunity and stimulate animal species-specific behaviors that further reduce the development of negative performances such as stress during experimental research. Enrichment tools were introduced into each cage of two strains of mice (ICR, Balb/c) and rats (Sprague-Dawley) in LAFAM. Three rats and five mice were caged in two IVC with controlled temperature, humidity and light cycle (12 hour light/dark cycle). Recyclable objects such as bottles, wooden blocks, boxes, paper egg trays and shredded papers were used as enrichment tools. Behavioral results were tabulated based on observable responses on the enrichment tools. The observations of interaction with cage mates, interaction with the enrichment devices, moving around the cage and fighting were also recorded. After three days of consecutive observation, both sexes of the Sprague-Dawley (SD) rats had shown active behavioral responses towards enrichment tools, but they did not utilize the tools for shelter or nest-building. SD rats were more prone to chewing and gnawing. Balb/c mice displayed active locomotive behaviors in the cage and positive responses towards tools for nest-building. They gathered the tools and pulled them to the edge of the nest. Male ICR mice exhibited normal behavioral responses to tools without showing any maladaptive behavior. Female ICR showed inactive behaviors such as barbering and sleeping. Inability to adapt to the tools given and became threatened to the surroundings are the most common maladaptive responses by rodents. The outcome of this study demonstrated a positive effective state of animals' well-being. The rodents demonstrated a stress free behavioral changes and an active state despite being kept in isolated cages.

Keywords: Environmental enrichment; recyclable objects, rodents, well-being and behavioral

PREVALENCE OF ENDOPARASITES ON LABORATORY RATS AT THE LABORATORY ANIMAL FACILITY AND MANAGEMENT (LAFAM), UiTM SELANGOR

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Abstract

A study on prevalence of endoparasites of laboratory rats was conducted between March and May 2016 at LAFAM. The faecal samples were collected from a total of 187 adult healthy breeding stock rats (3-4 years old) which included 112 Sprague-Dawley and 75 Wistar rats. The faecal samples were examined by direct smear technique as well as faecal floatation technique for parasitic larva, eggs and/or oocysts. Out of the 187 faecal samples examined, 35.83% were found positive for endoparasites. Prevalence of endoparasites was significantly higher in Wistar (54.67%) than in Sprague-Dawley 23.21% (26 of 112) ($P=0.00$). The most prevalent nematode parasites in both species were *Syphacia muris* and *Syphacia obvelata*, with prevalence of 68.66% and 26.87%, respectively. *Aspiculuris tetraptera* was also found in Wistar (7.32%). The prevalence of different species of parasites with their respective hosts were *S. muris* 20 (76.92%) in Sprague-Dawley, 26 (63.41%) in Wistar and prevalence of *S. obvelata* 6 (23.08%) in Sprague-Dawley, 12 (29.27%) in Wistar and prevalence of *A. tetraptera* nil in Sprague-Dawley, 3 (7.32%) in Wistar. This breeding stock indicates the high indication of parasitic load. These infected rodents should not be used for research purpose as this will interfere with the blood parameters.

Keywords: Endoparasite, faecal sample, *Syphacia muris*, *Syphacia obvelata*, *Aspiculuris tetraptera*

PREVALENCE OF ECTOPARASITES ON LABORATORY RATS AT THE LABORATORY ANIMAL FACILITY AND MANAGEMENT (LAFAM), UiTM SELANGOR

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Abstract

Ectoparasites are one of the most common health problems among laboratory rodents. A study on prevalence of ectoparasites of laboratory rats was conducted between March and May 2016 at LAFAM. This study was carried out on 187 laboratory rats (75 Wistar and 112 Sprague-Dawley). The rats' fur were brushed using a tooth brush and the scrapings were examined under a light microscope. The results showed that 38.48% of the rats were positive for ectoparasites. The most prevalent ectoparasites in this breeding colony were *Polyplax spinulosa* and *Radfordia affinis* with prevalence of 77.19% and 22.81%. This study indicates that the animals have never been ruled out for any health problems. All rodents should be screen for ectoparasites during the quarantine period.

Keywords: Ectoparasite, fur scraping, rodents, *Polyplax spinulosa*, *Radfordia affinis*

EXPOSURE OF LABORATORY ANIMAL ALLERGENS AMONG LABORATORY WORKERS AND RESEARCHERS

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Abstract

Laboratory animal allergy is an occupational disease that affects people engaged with the use and care of laboratory animals. Animal urine, saliva, feces, and dander contain proteins that are associated with allergic reaction. The aim of this study was to determine the prevalence and preventive measures taken amongst researchers and laboratory workers exposed to laboratory animal allergens. A questionnaire comprising of four sections (i.e. demographic, occupational history, past health information and present symptoms of laboratory animal allergen) was distributed to 267 respondents (192 female) working at five animal facilities in the UiTM. The majority of the respondent (94.4%) reported to have direct contact with laboratory animals and their excreta within their working environment. Handling soiled bedding (83.5%), stool (70.4%), urine (71.5%), and carcasses (59.9%) were among the duties carried out by the respondents exposing them to animal allergens. Animal dander is small enough to remain in the air for some time and adhere to clothes and skin. Several preventive measures were taken by the laboratory workers to reduce the risk of allergen exposure including using surgical masks, wearing protective glasses, laboratory coats, gloves and cap. The most common allergic reaction experienced by the respondents included itchy eyes (94.0%), frequent runny nose (90.3%), sneezing (89.1%), wheezing (88.8%) and nose congestion (64.8%) with fewer reporting asthma (18.7%) and shortness of breath (10.5%). It is important to reduce allergen exposure by taking appropriate preventive measures. However, prolonged exposure to allergens, lack of preventive awareness and highly sensitive individuals are the common causes of allergic reaction while working with laboratory animals.

Keywords: Allergens, allergic reactions, laboratory animals, excreta, preventive measures.

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ANTIDIABETIC EFFECT OF *MYMECODIA PLATYTYTREA* METHANOLIC TUBER EXTRACT

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Abstract

Tubers of *Myrmecodia platytytrea* (Rubiaceae) has been used traditionally as an alternative therapy for management of cancer. This plant is also believed to have the ability to lower blood glucose level. Nevertheless, no scientific proof is available on its antidiabetic effect. Type 2 diabetes mellitus (T2DM) is one of the main non-communicable chronic diseases. Individuals with T2DM have insulin resistance and usually develop relative insulin deficiency. The purposes of this study were to investigate the antihyperglycemic effect of *M. platytytrea* methanolic tuber extract (MPMTE), in vitro and in vivo. For the in vitro study, expression of genes involved in glucose metabolism namely G6Pase and GLUT4 were determined by quantitative RT-PCR on BRIN-BD11, 3T3 and L6 cell lines treated with MPMTE (125 and 500 µg/ml). Hyperglycemic male Sprague-Dawley rats that were induced with streptozotocin (45 mg/kg) were treated with MPMTE (100, 200 and 400 mg/kg; p.o.) and metformin (positive control, 100 mg/kg; p.o.) daily for 14 days. On day-15, blood was taken to measure fasting blood glucose level and lipid profile, respectively. Results revealed that MPMTE (125 and 500 µg/ml) were able to downregulate expression of G6Pase genes in BRIN-BD11, 3T3 and L6 treated cells, respectively. Upregulation of GLUT4 genes was also observed in BRIN-BD11, 3T3 and L6-treated cells. Furthermore, STZ-induced diabetic rats given MPMTE 200 and 400 mg/kg (p.o.) showed significant decreased ($p < 0.05$) in fasting blood glucose, total cholesterol, triglycerides and LDL levels compared to control rats. However, no changes were seen in HDL levels compared to control rats. In conclusion, MPMTE has the capability to modulate G6Pase and GLUT4 genes in BRIN-BD11, 3T3 and L6 cell lines and remarkably reduced blood glucose, cholesterol and triglyceride levels of STZ-induced diabetic rats, suggesting high potential of MPMTE to be developed as an antihyperglycemic agent.

Keywords: *Myrmecodia platytytrea*, streptozotocin, T2DM, diabetic rats, antihyperglycemic

PPEDIOCOCCUS PENTOSACEUS LAB6 ENHANCED COGNITIVE FUNCTION IN LIPOPOLYSACCHARIDE-INDUCED MEMORY IMPAIRED RAT VIA INHIBITION OF RHOA-AMYLOID- β MEDIATED NEUROINFLAMMATION

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Abstract

The emergence of Gut-Brain Axis concept supports the neuroprotective role of lactic acid bacteria (LAB) in improving cognitive dysfunction in Alzheimer's disease (AD). *Pediococcus pentosaceus* LAB6 are unique LAB isolated from local fermented food. Our previous study of MRS broth fermented with LAB6 had established a correlation between LAB-induced inhibition of RhoA and amyloid beta ($A\beta$) *in vitro*. This study validated the neuroprotective potential of LAB6 *in vivo*. Sprague Dawley rats (male, 3 months-old) were randomly assigned to four groups (n = 6/ group): wild-type, control [lipopolysaccharide (LPS)-induced], vehicle control (LPS-induced-saline) and LPS-induced rats fed with LAB6. The treatment group was being fed with 10^9 CFU/ mL LAB6 for 31 days. LPS (0.25 mg/kg) was injected intraperitoneally for 4 consecutive days (starting from day 28) during which all rats were subjected to the Morris Water Maze Test. Brains were then homogenised and subjected to Western Blot ($A\beta$) and biochemical (Brain-derived neurotrophic factor (BDNF), acetylcholinesterase, nitric oxide and cytokines) analyses. It was found that LPS-challenged rats fed with LAB6 were presented with significantly ($p < 0.05$) reduced escape latency (23.6 %) and escape distance (30.4 %) when compared to control. LAB-fed rats also remained in the platform quadrant a longer time (9.8 ± 0.5 sec) when compared to control (5.1 ± 1.9 sec). The improved memory elicited in LAB6-fed rodents was accompanied with significantly ($p < 0.05$) reduced $A\beta$ (-31.4%), augmented brain homogenate BDNF (+15.4%) and acetylcholine (+19.3%), reduced acetylcholinesterase (-40.1%) and nitric oxide (-30.3%) levels as opposed to control group. LAB-fed rats also showed increased anti-inflammatory cytokine IL-10 (+32.1%) and decreased pro-inflammatory cytokine IL-1 β (-39.5%) when compared to control. The present findings confirmed the neuroprotective effect of LAB6 in LPS-challenged rats mediated via inhibition of RhoA- $A\beta$ generated neuroinflammation

Keywords: Amyloid β , RhoA, lactic acid bacteria, lipopolysaccharide, cognition, neurotransmitters, neuroinflammation

MICROENCAPSULATED *LACTOBACILLUS PLANTARUM* LAB12 SUPPRESSED TUMOUR GROWTH IN ORTHOTOPIC COLORECTAL CANCER MOUSE MODEL

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Abstract

Pre-treatment with *Lactobacillus plantarum* LAB12 (LAB12), a unique lactic acid bacteria (LAB) isolated from local fermented food, reduced formation of aberrant crypt foci (azoxymethane-induced carcinogenesis) and colorectal cancer growth (subcutaneous xenograft mouse model). In spite of their chemopreventive properties, the vulnerability of LAB12 during gastrointestinal transit (pH and enzymatic action) and industrial processing (heat and storage) remains a major concern. This study addressed these issues by immobilising LAB12, by means of microencapsulation, within alginate (Alg)-based polymeric matrix incorporated with vegetable-based protein (VP). The survivability of microencapsulated LAB12 exposed to simulated gastrointestinal fluid, high temperature and various storage conditions were assessed. The chemopreventive properties of microencapsulated LAB12 were validated using an orthotopic xenograft mouse model bearing CT-26 cell transfected with green fluorescent pZsGreen1_N1 plasmid. The LAB feeding trial started 28 days before cell implantation and the feeding was continued until the end of week 7 where the animals were sacrificed and tumour were harvested for semi-quantitative western blot analysis. Alg-VP microcapsules with the mean diameter of $167.7 \pm 31.0 \mu\text{m}$ exhibited excellent tolerance against simulated gastric juice (96.4% survivability), intense heat (80.2% survivability at 100 °C for 30 min), storage ($>7 \log \text{CFU g}^{-1}$ after 8-week storage at 4 and 25 °C), pelletisation (89.4% survivability) and targeted release in simulated intestinal fluid ($>9 \log \text{CFU g}^{-1}$). Interestingly, orthotopic mouse model pre-fed with microencapsulated LAB12 significantly reduced tumour volume (-98.87%) and weight (-89.27%) with no incidence of liver and pulmonary metastases when compared to control. The chemopreventive effect could be attributed to apoptosis and antiangiogenesis mediated, at least in part, through p53 (+32.50%) and caspase-3 (+92.61%), and down-regulation of COX-2 (-63.96%), VEGF (-65.93%) and pcam-1 (-62.72%). Altogether, this study strongly implied the potential use of LAB12-loaded microcapsules in the management of colorectal cancer

Keywords: microencapsulation, lactobacilli, orthotopic, colon cancer, chemoprevention

ANTI-CANCER EFFECT OF *GONIOTHALAMUS LANCEOLATUS* ON MDA-MB-231 AND MDA-MB-468 TRIPLE NEGATIVE BREAST CANCER

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Abstract

Breast cancer is the leading cause of cancer death among women in Malaysia and worldwide. Breast cancer is treated by surgery, chemotherapy, and radiotherapy or a combination of this, but there still remains a high mortality rate. Breast cancer patients also need to bear with the side effects of conventional treatment. Traditional medicine may have reduced of side effects and have lower cost implications as compared to conventional treatment. *Goniothalamus* is an effective medicinal plant in Malaysia for treatment of cancer but little study was conducted on *Goniothalamus lanceolatus*. The present study was designed to investigate the potential role of *Goniothalamus lanceolatus* plant extracts on the chemoprevention and therapy of highly metastatic MDA-MB-231 and MDA-MB-468 triple-negative breast cancer cell lines. To evaluate the cell growth inhibition, cell lines was treated with *Goniothalamus lanceolatus* barks, leaves and roots and was subjected to MTT analysis to assess cell viability and cytotoxic effects. Mode of cell death was evaluated by flow cytometric analysis using Annexin-V/PI. In vitro cytotoxic activity of crude extract showed 50% growth inhibition concentration (IC₅₀) of MDA-MB-231 and MDA-MB-468 after 24 hours of treatment. Lowest IC₅₀ value recorded was 47.95µg/ml in root extract using methanol solvent for MDA-MB-231 and 60.45µg/ml in leaves extract using DCM solvent for MDA-MB-468. *Goniothalamus lanceolatus* markedly induced apoptosis through the investigation of characteristic apoptotic morphological changes, nuclear DNA fragmentation, annexin V-FITC/propidium iodide (PI) double staining. The overall results provide evidence and ethnopharmacological relevance for the potential use of *Goniothalamus lanceolatus* in development of new alternatives anti-cancer agents for the treatment of highly metastatic breast cancer.

Keywords: Breast cancer, MDA-MB-231, MDA-MB-468, *Goniothalamus lanceolatus*, triple negative breast cancer

ANTICANCER AND APOPTOSIS ACTIVITIES OF A CRUDE DICHLOROMETHANE EXTRACT FROM *GONIOTHALAMUS LANCEOLATUS* BARK IN MCF-7 BREAST CANCER CELL LINE

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Abstract

Discovery of cytotoxic activity of *Goniothalamus lanceolatus* presents a potential cure for treatment of breast cancer. In Malaysia, breast cancer is the most common cancer in woman with 1 out of 9 women at risk. *Goniothalamus* sp. has been traditionally used for childbirth, fever and food poisoning. The present study aimed to investigate the anticancer and apoptosis activities of dichloromethane (DCM) crude extracts of *G. lanceolatus* bark against MCF-7 breast cancer cell line. Varying concentrations of *G. lanceolatus* crude extract (100, 200, 400, 600, 800, and 1000 µg/ml) were applied on cultured MCF-7 cell line to determine the cytotoxic effect by measuring the cell proliferation activity after 24 hours of treatment. The dichloromethane crude extract showed cytotoxic activity ($P \leq 0.05$) with the IC₅₀ value of 126.85 µg/ml. Dichloromethane crude extracts significantly suppressed the proliferation of MCF-7 and promoted cell apoptosis in MCF-7 cancer cell line through Annexin V/Propidium Iodide assay. *G. lanceolatus* thus shows promising antiproliferation effect in breast cancer cell.

Keywords: *Goniothalamus lanceolatus*, Cytotoxicity, Apoptosis, Anticancer, MCF-7 Cell Line

COPPER (II) SCHIFF-BASED COMPLEX INDUCED APOPTOSIS AND CELL CYCLE ARREST TOWARDS MDA-MB-231 BREAST CANCER CELL LINE

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Abstract

The concern over acquired drug resistance and serious side-effects of current anticancer drugs in the midst of the rise of cancer, in particular breast cancer, as one of the leading cause of death worldwide, drives the effort to develop better alternatives. Copper complexes have been widely studied for the anti-tumour application as cancer cells are reported to take up greater amounts of copper than normal cells. Therefore, the differential response between normal and cancer cells towards copper are the basis for the development of copper complexes as anticancer agent. Preliminary study revealed that the newly synthesised copper complex [Cu(SBCM)2] displayed marked anti-proliferative towards MDA-MB-231 breast cancer cells. Therefore, Cu(SBCM)2 has great potential to be developed as an agent for the management of breast cancer. The present study was carried out to investigate the mode of cell death induced by Cu(SBCM)2 towards triple-negative MDA-MB-231 breast cancer cells. The inhibitory and morphological changes of MDA-MB-231 cells treated with Cu(SBCM)2 was determined by using MTT assay and inverted light microscope, respectively. Confirmation of apoptosis and cell cycle arrest were determined by flow cytometry analysis. The expression of mutant p53 was detected with western blot analysis. Cu(SBCM)2 significantly inhibited the growth of MDA-MB-231 cells in a dose-dependent manner with GI₅₀ 18.67 ± 3.06 µM. Morphological study revealed that Cu(SBCM)2-treated MDA-MB-231 cells experienced cellular shrinkage, membrane blebbing, chromatin condensation and formation of apoptotic bodies, suggesting that Cu(SBCM)2 induced apoptosis in the cells, which was further confirmed by Annexin-V/PI flow cytometry analysis. It was also found that Cu(SBCM)2 induced G2/M phase cell cycle arrest towards MDA-MB-231 cells. The induction of apoptosis and cell cycle arrest is possibly due to the down-regulation of the mutant p53. In conclusion, Cu(SBCM)2 can be developed as a targeted therapy for the treatment of triple-negative breast cancer.

Keywords: Copper (II) Schiff-base, Breast cancer, Apoptosis, Cell cycle arrest, p53

PG-O 7: IN VIVO MOLECULAR STUDY OF MYRMECODIA PLATYTYREA WATER EXTRACT CHOLESTEROL LOWERING EFFECT ON HYPERCHOLESTEROLEMIC-INDUCED RAT

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Abstract

This study was designed in order to investigate the effect of *Myrmecodia platytyrea* (MyP) extract as an anti-hypercholesterolemic agent. Sprague-Dawley rats (7 weeks old) were randomly assigned into six groups ($n = 6$ for each group). Normal Control group, Negative Control group (NC), Positive Control group (PC), MyP water extract treatment group (MyP 100, MyP 200 and MyP 400). In vivo study results showed that treatment of MyP water extract can significantly reduce ($p < 0.05$) low-density lipoprotein (LDL) and triglyceride (TG) compared to negative control group. The extract significantly increased ($p < 0.05$) high-density lipoproteins (HDL) concentration compared to negative control group. These effects were further examined in molecular study, the result showed that the treatment of MyP water extract down-regulated genes related to atherosclerosis and up-regulated genes related with reverse cholesterol transport (RCT) which decrease the risks of hypercholesterolemic diseases.

Keywords: *Myrmecodia platytyrea* (MyP), anti-hypercholesterolemic, low-density lipoprotein (LDL), high-density lipoproteins (HDL), gene expression.

NEUROPROTECTIVE EFFECT OF *MYRMECODIA PLATYTYREA* AQUEOUS TUBER EXTRACT ON LIPOPOLYSACCHARIDES-INDUCED NEUROINFLAMMATION

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Abstract

Recent studies have demonstrated a close association between neuroinflammatory responses, increased production of inflammatory mediators, and neurodegeneration. Lipopolysaccharide (LPS) was used as neuroinflammatory inducer for neuroinflammatory-mimicking model that could affect performance in learning tasks. *Myrmecodia platytyrea* (MP) tuber extract as a decoction has been claimed to possess cancer-preventive properties and believed to be able to treat inflammatory-related diseases such as rheumatoid arthritis, diabetes and even neuroinflammation. Therefore, this study is designed to determine the potential of MP aqueous extract (MPAE) as an agent for Alzheimer's disease (AD) prevention. Aged male ICR mice (24 weeks old) were administrated with MPAE (100, 200 and 400 mg/kg/bw, p.o) and piracetam (400 mg/kg/bw, p.o.), a nootropic drug, for 7 days. On day 3 of the treatment, mice were given daily injection of LPS (3 mg/kg/b.w., i.p.) for 4 days. The aged mice underwent 3 consecutive days of Morris water maze training before LPS was administrated followed by 3 days of testing, 4 hours after LPS administration and then followed with probe test. Our results showed that intraperitoneal injections of LPS significantly prolonged escape latency, increased distance travelled and decreased their swimming speed discovering the platform. Interestingly, mice treated with piracetam (as reference drug, positive control) and 200 mg/kg MPAE showed significantly decreased escape latency and distance travelled with increased in swimming speed as compared to untreated-LPS group. In conclusion, these finding revealed that neurodegenerative effect of LPS was protected with the administration of MPAE, thus suggesting the potential therapeutic application of MPAE against neuroinflammation.

Keywords: *Myrmecodia platytyrea*, Ant-nest plant, Neuroinflammation, Lipopolysaccharide, Morris water maze, Probe test.

EFFECT OF *PEPEROMIA PELLUCIDA* [L.] KUNTH. HERBS ON BONE QUANTITY BASED ON FEMUR WEIGHT, LENGTH, BONE VOLUME FRACTION, AND CONNECTIVITY DENSITY ON OVARIECTOMIZED RATS

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Abstract

Estrogen has an important role to keep balance the process between bone resorption and formation. In menopausal women, there is a significant decline of estrogen production. Bone resorption rate will be increasing and exceeding its formation. The objective of this study was to investigate the effect of *Peperomia pellucida* (L.) Kunth. herbs juice and ethanol extract on bone quantity on ovariectomized rats. Reduction of estrogen production was induced by ovariectomy with double dorsolateral approach on female Wistar rat. The animals were undergone SHAM (normal group) and ovariectomy surgery. The ovariectomized rats divided into 6 groups treated with CMC Na 0.3% (control group), ethynil estradiol 4.5 µg/kg BW, juice 50 mg/kg BW, juice 100 mg/kg BW, ethanol extract 50 mg/kg BW, and ethanol extract 100 mg/kg BW. The rats were treated for 6 weeks started from one week after operation. At the end of experiment, femur was collected to be analyzed. All of *P. pellucida*-treated groups did not show any improvement in femur weight significantly to control group ($p < 0.05$). Rats treated with ethanol extract at 100 mg/kg BW had femur lengthier significantly compared to control group ($p < 0.05$). This changes did not showed significantly different to normal group ($p < 0.05$). Furthermore, ethanol extract 100 mg/kg BW-treated group showed improvement on bone microarchitecture compared with control group. It counteracted the reduction of bone volume fraction, and connectivity index. As a conclusion, ethanol extract of *P. pellucida* herbs at dose 100 mg/kg BW could improve bone index, length, bone volume fraction, and connectivity index on ovariectomized rats.

Keywords: *Peperomia pellucida* (L.) Kunth., femur, ovariectomy, bone quantity

HYPOGLYCEMIC EFFECT OF ETHANOLIC LEAVES EXTRACT OF *PASSIFLORA FOETIDA* LINN FROM SOUTH SULAWESI INDONESIA IN STREPTOZOTOCIN-INDUCED DIABETIC MICE

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Abstract

Passiflora foetida Linn (permot) is a plant that grows wild in South Sulawesi. People empirically utilise the plants in various diseases. This study aims to determine the effective dose of the leaves extract of permot to provide a hypoglycemic effect for diabetic mice. This research used 15 male mice which divided into 3 groups. Group 1 is a control group, a group that only streptozotocin induced without the leaves extract of permot. Group 2 and 3 are treatment groups, the groups induced by streptozotocin and ethanol leaves extract of permot at a dose of 200 mg/kg and 600 mg/kgbw. Streptozotocin was administered for 5 consecutive days intraperitoneally (i.p). The extracts were administered to the both group 2 and 3 on days 15th - 36th. Measurement of blood glucose levels of mice are done on days 1st, 14th, 22nd, 29th, and 36th. The results were processed statistically using test T-test and one way ANOVA. The Control group showed significantly different ($p < 0.05$) to the treatment groups. This study concludes that ethanol leaves extract of permot at a dose of 200 mg/kgbw and 600 mg/kgbw have an effect on decreasing blood glucose levels in mice.

Keywords: *Passiflora foetida* Linn, hypoglycemic, streptozotocin

ACUTE ORAL TOXICITY OF METHANOL SEED EXTRACT OF *GLYCINE MAX* (L.) MER DETAM I AND II VARIETIES IN MICE

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Abstract

The previous study has shown that soybean seeds (*Glycine max* (L.) Merr) detam I and detam II varieties have antioxidant activity. The purpose of this research was to evaluate the acute oral toxicity of the methanolic extract of *Glycine max* (L.) Merr from both of these varieties. The test was conducted in Swiss Webster mice using the conventional method. Each extract was tested using twenty-five mice and divided into 5 groups, namely control group and four test groups, each given the extract at 78 mg/kg body weight (b.w), 312.5 mg/kg b.w, 1250 mg/kg b.w and 5000 mg/kg b.w, respectively. The mice in all groups were observed for behavioral pattern, physical condition, body weight, organ to body weight ratio, organ histology, and mortality. All of the parameters among experimental groups were comparable. The LD50 of each extract in mice were determined to be greater than 5000 mg/kg b.w, and there were no signs of toxicity and mortality after the administration of each extract for 14 days. All of the test animals did not indicate any change in behavioral pattern and physical conditions. Also, there were no any significant differences ($p > 0.05$) observed in the body weight and organ to body weight ratio. There was no abnormality in histopathological examination on the liver between control and test groups. Results of the present study suggest that the methanolic extract of soybean seeds (*Glycine max* (L.) Merr) detam I and II varieties are safe after single administration at high dose.

Keywords: *Glycine max*, detam I, detam II, methanol extract, acute oral toxicity

THE PROPHYLACTIC EFFECT OF *MYRMECODIA PLATYTYREA* TUBER EXTRACT ON STREPTOZOTOCIN-INDUCED DIABETIC RATS

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Abstract

Type 2 diabetes mellitus (T2DM) is a chronic disease that affects a large number of populations across the world with substantial economic and social burden. Since this disease can only be controlled, it is important to find solutions to prevent the incidence of T2DM. Therefore, this study was designed to explore the potential of the aqueous tuber extract of *Myrmecodia platytyrea* (family Rubiaceae) or locally known as 'sarang semut' as prophylaxis to diabetes mellitus. *M. platytyrea* methanolic tuber extract (MPMTE, 100 mg/kg to 400 mg/kg, p.o) was given to the treatment groups for 14 days followed by injection of streptozotocin (STZ, 45 mg/kg, i.p.). The non-diabetic rats and the untreated group received normal saline (p.o.). Treatment with the extract and normal saline were continued for another 5 days. Fasting blood glucose and body weight were recorded every 7 days until the end of the experiment. On day-19, blood was collected through cardiac puncture to measure blood glucose level, lipid profile and cytokine levels. Pancreas, liver and kidney were excised for histological examination. Results showed that STZ-induced rats receiving 400 mg/kg (p.o.) of extract exhibited significantly ($p < 0.05$) higher HDL and lower LDL, triglycerides and cholesterol levels compared to untreated STZ-induced rats. However, blood sugar levels were not normalized by the administration of MPMTE. Interestingly, TNF- α were reduced and IGF were increased significantly ($p < 0.05$) in STZ-induced rats treated with MPMTE (100-400 mg/kg, p.o.) compared to untreated STZ-induced rats. Furthermore, no histological alteration was observed in STZ-induced rats treated with MPMTE suggesting that sub-acute administration of MPMTE was protective against STZ toxicity. In conclusion, administration of the MPMTE was not able to lower blood glucose level, but improved lipid profile of the STZ-induced rats without any damage to pancreas, liver and kidney.

Keywords: *Myrmecodia platytyrea* methanolic tuber extract; type 2 diabetes mellitus; streptozotocin; prophylactic

THE ANTI-CANCER ACTIVITIES OF ISOLATED COMPOUNDS FROM *PRISMATOMERIS MALAYANA* RIDLEY AGAINST SELECTED CANCER CELL LINES

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Abstract

Prismatomeris malayana is a small shrub distributed in South East Asia with interesting secondary metabolites such as triterpenes, iridoids and anthraquinones. There are many reports on the cytotoxicity effects of isolated compounds from this plant. The objective of this study was to evaluate the antiproliferative effects of the extracts of various part of *Prismatomeris malayana* towards cancerous cell lines; (CCL) tested include MCF7 (breast CCL), CaOV3 (ovarian CCL), SKOV3 (ovarian CCL) and HT-29 (colorectal CCL). The methanol extracts of roots, stems and leaves were fractionated successively by using solvent-solvent partitioning method starting with the non-polar followed by polar solvent to yield petroleum ether, chloroform, ethyl acetate and water fraction. Cytotoxicity test was carried out to determine the IC₅₀ values. This enables the prediction of the potential of an extract having active (hit) compound. Crude extracts that are able to kill 50% of the cancer cells at the concentration of less than 20 µg/ml can be considered to have anti-cancer effect in vitro. However, the extracts of the leaves (PML), roots (PMS) and stems (PMS) together with their fraction were found to be moderately active against these cancer cell lines: (MCF7, CaOV3, SKOV3, HT-29). In conclusion, methanolic extracts of *P. malayana* (leaf, root and stem) may have potential as antiproliferative agent but further studies need to be conducted.

Keywords: Anti-cancer; *Prismatomeris malayana*; isolated compounds; NMR; cancer cell lines

OESTROGENIC ACTIVITY OF *LABISIA PUMILA* VAR. *ALATA* IN IN HUMAN BREAST ADENOCARCINOMA (MCF-7) AND HUMAN BREAST EPITHELIAL (MCF-10A) CELL LINES

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Abstract

Previous study showed that *Labisia pumila* var. *alata* (*Lpva*) is a potential therapeutic alternative to Hormone Replacement Therapy (HRT) due to its estrogenic activity. Thus, this study was undertaken to observe the estrogenic effect of *Lpva* aqueous extract on oestrogen receptor (ER)-positive cell lines. To understand the interaction between *Lpva* and ER, molecular docking study of several phytochemical found in *Lpva* were tested. The binding activity of *Lpva* on oestrogen receptors was examined using a fluorescence polarization-based competitive binding assay. The estrogenic activity of *Lpva* was investigated by using several in-vitro assays. MCF-7 and MCF-10A cell lines were treated with several concentrations of *Lpva* aqueous extract and incubated for 72 hours where they showed estrogenic effect, either with or without ER-antagonist (ICI 182,780). 17 β -estradiol (E2) was used as the positive controls. Molecular docking showed that phytochemicals from *Lpva* can couple into binding site of ER α and ER β . Proliferation assay shows that *Lpva* aqueous extract significantly increases the proliferation of MCF-10A cell at concentration of 25%, 50%, and 100% of 1 μ g/mL ($p < 0.005$). In contrast, *Lpva* significantly exhibit cytotoxic effect to MCF-7 cell at 50% and 100% of 1 μ g/mL ($p < 0.005$) with the IC₅₀ value of 94.42. E2 give significant proliferation to both cell lines. ER-antagonist (ICI 182,780) blocked the proliferation induced by both E2 and *Lpva*, indicating an oestrogen receptor-dependent mechanism for the estrogenic effects on MCF-7 and MCF-10A cell proliferation. *Lpva* aqueous extract gives both efficacy and selectivity effect to MCF-7 cells and MCF-10A cells respectively, which indicates a possible alternative to HRT.

Keywords: *Labisia pumila* var. *alata*, molecular docking, estrogen activity, MCF-7, MCF-10A

THE WOUND HEALING ACTIVITY OF *MIKANIA MICRANTHA* LEAVES EXTRACT

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Abstract

Mikania micrantha Kunth is a perennial creeping vine originated from tropical Central and South America. This plant is commonly known as "selaput tunggal" in Malaysia and "sembung rambat" in Indonesia. *M. micrantha* was claimed to prevent and cure several diseases such as diarrhea, diabetes and stroke. Leaves of *M. micrantha* was used traditionally as poultice for wound and sores. The aim of this study is to investigate the ability of *M. micrantha* ethanol extract in accelerating the wound healing process. Cell viability and scratch assay were carried out on fibroblast cells treated with various concentrations of the extract. Trolox (100 µM) was used as positive control. Results on the MTT assay showed low cytotoxic effect after 24, 48 and 72 h incubation with the extract. Wound healing process was also accelerated significantly (P 0.005) especially after treatment with 7 µg/ml and 15 µg/ml of *M. micrantha* ethanol extract. In conclusion, *M. micrantha* ethanol extract has great potential in accelerating wound healing justifying its traditional use. Further studies will be carried out to determine its molecular mechanism.

Keyword: Mikania micrantha, wound healing, cell proliferation, scratch wound assay

USE OF AEROSOLIZED VANCOMYCIN FOR MRSA PNEUMONIA

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Abstract

Methicillin resistant *Staphylococcus aureus* (MRSA) is a major nosocomial pathogen that causes severe morbidity and mortality. For a systemic infection such as MRSA, the use of oral vancomycin is not recommended because of the low absorption and low serum concentrations. Other alternative for route of administration of vancomycin is through the nebulizer. However only limited studies that provide the evidence to support the use of aerosolized antibiotics to treat multi drug resistant (MDR) pneumonia including MRSA. We present a case of a 43 years-old Malay male with underlying bronchial asthma and ankylosing spondylitis who was referred to a government-funded hospital in Klang Valley for posterior spinal instrumentation. Apart from hospital acquired pneumonia, he presented with multiple bacterial infections such as salmonella sp bacteremia, Ecoli, urinary carbapenem-resistant enterobacteriaceae (CRE) upon transfer from another hospital. In the intensive care unit, he was on a ventilator and plans for the instrumentation procedure was withheld. Sputum, urine and blood cultures revealed isolation of bacteria: *Enterococcus Faecium* Bacteremia (EBB), MRSA pneumonia, CRE and pseudomonas A. Intravenous (IV) vancomycin was given to treat both EBB and MRSA pneumonia. Aerosolized vancomycin was introduced as targeted therapy. Aerosolized vancomycin was used to treat MRSA pneumonia in addition to using IV. There are currently no large-scale randomized trials evaluating the efficacy of its use. A few small studies provided evidence to support the use of aerosolized antibiotics to treat pneumonia in patients infected with multi-drug resistant (MDR) organisms. However this route of administration was especially useful when the minimum inhibitory concentrations for the MDR organism were too high to safely administer intravenous antimicrobial agents.

Keywords: Aerosolized, vancomycin, MRSA, pneumonia

BRUCEA JAVANICA INDUCED APOPTOSIS ON HCT-116 COLON CANCER CELLS

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Abstract

Brucea javanica (L.) Merr., is known as a Chinese medicine. Its fruit and seeds showed impressive efficiency to cure various infectious diseases and recent studies revealed that the herb extract have antiproliferative and pro-apoptotic activities on human cancer cells. Our study on *B. javanica* ethanol extract (BJEE) illustrated potential cytotoxicity effect on colorectal carcinoma cells (HCT-116) with IC₅₀ value of 8.9±1.32 after 24 h treatment. The significant activation of caspase-8 indicated involvement of extrinsic pathway in the apoptosis induction. In addition, activation of caspase-9 and elevation of ROS generation, also revealed mitochondria-apoptotic pathway mediated at induction of apoptosis by BJEE. The results from this study showed BJEE could be a promising agent for colorectal cancer therapy through induction of apoptosis on HCT-116 via extrinsic and intrinsic pathways

Keywords: *Brucea javanica*, apoptosis, colon cancer, HCT-116, caspase

OBESITY AND FOOD INTAKE IN DOWN SYNDROME INDIVIDUALS

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Abstract

Down syndrome (DS) is a common chromosomal abnormality occurring in about 1 in 700 live births. Obesity in DS individuals has been recognized with increasing in age but not in prepubescent. This may be due to neuromotor deficiency in DS, which influences chewing frequency, that leads to limited food choices for DS children. Therefore, information on the food or nutrient intake are vital to ensure healthy growth of children with DS as they are exposed to high risk of obesity and other concomitant diseases. The eating habits, intake of different types of food and intake of complementary or alternative medicine by DS individuals were reported by the parents or caregivers through questionnaires during the interview sessions and were compared to controls. Validated questionnaires included a checklist of the different types of food, portion sizes and frequency intake and also complementary or alternative medicines intake. Measurement of height and weight, and body mass index (BMI) were calculated and compared to age-matched controls. DS persons were significantly shorter than the age-matched controls ($p < 0.05$) who were in age groups of 1 to 7 years, 8 to 12 years and 13 to 20 years. The mean weights of DS persons are similar to controls in all age groups except for age range of 13 to 20 years, where the weights of DS persons are significantly lower than controls ($p < 0.05$). The mean BMI of DS persons range from 17.36 kg/m² to 33.78 kg/m² while for controls the mean BMI range from 15.48 kg/m² to 27.43 kg/m² with no significant difference between groups. Sixteen of the DS subjects (19%) ($n=84$) are obese with BMI values above the recommended level. The overall dietary pattern of DS individuals is similar to that of controls as majority of parents or caregivers (91%) reported that DS children consume similar type of food as their siblings. It is concluded that the findings in this study demonstrate a prevalence of obesity in DS is 19% and similar range of food intake between DS groups and controls. Further study should be conducted to understand the significance of the abnormalities especially the mechanisms of action that contributed to this condition.

Keywords: Down syndrome, obesity, nutrition

HIGH DOSE OF ETHANOLIC EXTRACT OF *CENTELLA ASIATICA* INDUCES CELL PROLIFERATION IN ACUTE LYMPHOID LEUKAEMIA (ALL) CELL LINES

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Abstract

Centella asiatica (Family: Apiaceae) is a tropical medicinal plant with various biological activities. Despite numerous studies evaluating its medicinal effects, little is known about its effect on leukaemia. This study aims to investigate the effect of ethanolic extract of *Centella asiatica* (eCA) on Acute Lymphoid Leukaemia (ALL)-CCRF-CEM cell line. CCRF-CEM leukaemia cells were cultured in RPMI media supplemented with 10% FBS and incubated at 37 °C in 5% CO₂. Cells were exposed to different concentrations of eCA (0-1000 µg/ml) for 24 or 48 hrs. MTT assay was used to measure cell proliferation rate after treatment with eCA. Graphprism software was used to determine IC₅₀ values by non-linear regression method. The rate of all proliferation is similar in both 24 and 48h exposure groups. These data suggest that treatment with high dose of eCA may activate cell proliferation pathway and contribute to cell survival rates. However, further study needs to be done to evaluate the effect of their biologically active components and their mechanisms of action in the cells.

Keywords: *Centella asiatica*, proliferation, leukaemia

APOPTOSIS INDUCTION BY *MYRMECODIA PLATYTYREA* METHANOLIC TUBER EXTRACT IN HUMAN HEPATOCELLULAR CARCINOMA

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Abstract

Hepatocellular carcinoma (HCC) is one of the most prominent cancer of the liver and the most common solid tumour in the world. The only proven cure for HCC at this moment is surgery, nevertheless, development of new tumours has been reported in the remaining liver within 2 years in 80% of cancer patients. Tuber of the *Myrmecodia* spp. (Ant nest plant) is used as decoction, for treatment of mild diseases to a much severe disease such as cancer by the locals in the West Papua. Yet, there are not much study conducted on *Myrmecodia platytyrea* which is locally known as 'sarang semut daging merah'. This study was carried out to investigate the antiproliferative potential of *Myrmecodia platytyrea* methanolic tuber extract (MPMTE) against cancer cells. MTS assay was performed on several carcinoma cell lines such as HepG2 (hepatocellular carcinoma), Chang (chang liver), WRL68 (human embryonic liver), HSC-4 (oral squamous carcinoma), A498 (human kidney carcinoma) and HT29 (human colon adenocarcinoma) cell lines treated with various concentrations of MPMTE (0.1-1000 µg/ml) to determine cell viability. Cell apoptosis was evaluated using Acridine Orange and Propidium Iodide staining observed under fluorescent microscope at 400x magnification whereas apoptosis and cell cycle analysis were conducted using flow cytometry. MPMTE was found to be most potent in inhibiting the growth of HepG2 cells compared to other cancerous cells. Normal liver cell lines (Chang and WRL68) demonstrated low and moderate cytotoxicity, respectively. HepG2 cells treated with MPMTE displayed a distinctive apoptotic characteristic when observed under fluorescent microscope at 400x magnification, dose-dependently. Additionally, HepG2 cell line treated with MPMTE exhibited a significant increase of 32.35% apoptotic death at 6 µg/ml compared to untreated cells. MPMTE facilitate cell cycle arrest at G2/M phase. In conclusion, MPMTE possessed potential anti-proliferative activity and significantly suppressed the growth of HepG2 cells via apoptosis without much affecting the normal cells.

Keywords: Hepatocellular carcinoma (HCC), *Myrmecodia platytyrea*

CYTOTOXICITY EFFECT OF *ASPERGILLUS FUMIGATUS* (MV) FRACTIONS ISOLATED FROM *GRACILARIA ARCUATA ZANARDINI* AGAINST HEPG2 CELLS

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Abstract

Marine natural products are valuable sources that could produce potential chemotherapeutic agents. Red seaweeds were recognized as a rich source of microbial diversity with unique metabolites that can be of pharmaceutical and medicinal importance. Liver cancer is major concern which account for million of deaths annually. It seems to be resistance against many current available synthetic drugs. Our previous study proven, MV extract isolated from *Gracilaria Arcuata Zanardini* exhibited high anticancer activity against many cancerous cell lines. In this present study the cytotoxic effect of MV fractions were assessed. The cytotoxic effect was evaluated using HepG2, human hepatocellular carcinoma cell line and HeLa, normal human epithelial liver cell line. MV isolated from a Malaysian red seaweed was further grown on potatoes dextrose agar (PDA) at two different salinity (1% and 3%) of artificial sea salt (ASS). Both crude extracts of MV 1% and MV 3% induced cytotoxicity in HepG2 cells after 24 hours exposure (IC₅₀; 50.0 ± 8.7 µg/ml and 45.0 ± 5.0 µg/ml respectively). Interestingly non-cytotoxic effect was detected for HeLa cells. MV 3% showed better cytotoxic effect as compared to MV 1%. Therefore, preliminary screening on MV 3% extract through bio-guided assay method found that 12 fractions have been identified to determine cytotoxic effect against HepG2 cell. Amongst the 12 fractions, 20 µg/ml of fraction 3 (F3) showed the most promising result with 55.9 ± 1.5% cell death. This preliminary data suggests that *Gracilaria arcuata zanardini* species of marine endophytic fungi from Malaysian seaweeds have a potential in cytotoxicity against HepG2 without toxic to the HeLa. However, further investigation is needed to explore this potential bioactive fraction as chemotherapeutic agent against HepG2 cell. These results indicate that endophytic fungi *A. fumigatus* isolated from *Gracilaria arcuata zanardini* could be a potential source for bioactive compounds and may find potential use in pharmaceutical industry.

Keywords: cytotoxicity, marine endophytic fungi, Malaysian seaweed, HepG2, HeLa

INHIBITION OF AZOXYMETHANE-INDUCED COLONIC ABERRANT CRYPT FOCI FORMATION BY A NEW SYNTHETIC COMPOUND

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Abstract

This study investigated the effect of a new synthetic compound, abbreviated as DBID on the azoxymethane-induced colonic aberrant crypt foci in rats. Chemopreventive effects of the new benzo indole derivative assessed among twenty-four adult male rats with 15 mg/kg body weight AOM which were injected subcutaneously once a week for two weeks and then were divided randomly into four groups. Group I (Cancer control), Group II (treated with 20 mg/kg of DBID), Group III (treated with 40 mg/kg of DBID) and group IV (35 mg/kg body weight 5-fluorouracil once a week as reference control). On the last day of the experiment, the animals were euthanized using an overdose of ketamine and xylazine and the blood and organs were collected. The acute toxicity test was used to demonstrate the safety usage of the synthetic compound. *In vitro* antioxidant activities of the compound were investigated. The compound showed high antioxidant activity in *in vitro* assays. The acute toxicity study showed no nephrotoxic and no hepatotoxic effects or any Serum biochemical changes in rats that orally administered with 2000 mg/kg body weight of DBID compared to control group. Colon tissues evaluation showed that DBID compound diminished Azoxymethane-induced aberrant crypt foci formation and pathological changes in the colonic mucosal tissues. Following treatment with the compound antioxidant enzyme activity was increased compared to carcinogen groups while the malondialdehyde level was significantly reduced. Furthermore, the downregulation of Bcl-2 and a upregulation of Bax protein was confirmed by western blotting. In Conclusion, this study revealed the inhibition of Azoxymethane-induced Colonic Aberrant Crypt Foci Formation by a new benzo indole derivative compound in rats that might be associated with its potent antioxidant activity and effective activity against free radicals involved in the formation of colonic lesions

Keywords: Synthetic compound; Antioxidant activity; Acute toxicity; Azoxymethane (AOM) -induced colon cancer; Aberrant crypt foci (ACF)

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VANCOMYCIN LOADED BONE CEMENT IN PROSTHETIC JOINT INFECTION

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Abstract

Antibiotic-loaded bone cement (ALBC) has been used more than four decades ago in total joint arthroplasties. ALBC is one way of delivering high drug concentration in situ. An in vitro study was carried out to compare the antibacterial ability of different preparation of vancomycin-loaded bone cement against methicillin-resistant *Staphylococcus aureus* (MRSA) using disc diffusion method. DePuy Smartset Gentamicin 40g bone cement; a polymethyl methacrylate based cement was used in this study. Vancomycin powder added to bone cement powder in a ratio of either 1 in 20 or 1 in 10. Two types of formulations were prepared, powder group (vancomycin powder added to bone cement powder before the addition of monomer) and liquid group (vancomycin powder added after the addition of monomer into the bone cement powder but still in liquid phase). Each preparation (2g powder, 2g liquid, 4g powder, 4g liquid) of vancomycin-loaded bone cement disc were applied on Mueller Hinton agar (MHA) plates lawned with MRSA together with controPP (plain bone cement disc and standard 30mcg vancomycin antibiotic disc). All plates were incubated at 37°C and the diameters of inhibition zone were measured after 24 hours. The same method was aPPo used to test all bone cement discs against MRSA ATCC 43300 as reference. All tests were performed in duplicates with n3. Statistical analyses were performed by using ANOVA test. No differences were observed in the zone of inhibition between the strength of vancomycin added into the bone cement and aPPo in the formulation of bone cement itself ($p > 0.05$). The addition of vancomycin powder before or after the addition of monomer showed no difference in terms of antibacterial activity ($p > 0.05$). Therefore, bone cement disc of 2g vancomycin can be used in managing MRSA infection for prosthetic joint infections.

Keywords: Bone cement, vancomycin, arthroplasty, antibiotic-loaded bone cement

MANAGING TYPE 2 DIABETES MELLITUS (T2DM) WITH PROBIOTICS

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Abstract

Recent evidence suggests that imbalanced gut microbiota could contribute to the onset of pre-diabetes state. Probiotics, which have been extensively documented for their applications in many metabolic diseases, may serve as alternative biotherapeutics against T2DM. Clinical evidence regarding the use of probiotics in T2DM management remains scant, particularly in clinical setting. In fact, findings from the very few clinical trials that have been conducted were inconsistent. As such, this study was geared towards investigating the effects of probiotics on glycaemic control and quality of life (QoL) among T2DM patients. Systematic review and meta-analysis were conducted prior to the commencement of a randomised controlled trial (RCT) that involves 100 T2DM patients who will be assigned to receive daily probiotics or placebo for 24 weeks. Results from the meta-analysis revealed a moderate beneficial hypoglycaemic effect of probiotics, with significant reduction in fasting blood glucose. The effects of probiotics on glycosylated haemoglobin (HbA1c), anti-inflammatory and anti-oxidative markers, however, remained inconsistent. Systematic review found existing clinical trials to be limited by their variation in the duration of intervention, quality and depth of the study design, thus hindering meaningful conclusions that could be made. As QoL assessment provides valuable outcome that supports clinical decision-making, particularly for patients with incurable chronic diseases, a 15-item Diabetes Quality of Life Brief Inventory (DQoL-BCI) was translated into Malay language and validated in the Malaysian setting prior to its use in the RCT. Altogether, the present findings implied that a well-designed clinical trial is essential to confirm the beneficial effects of probiotics as an alternative non-pharmacological approach in managing T2DM. By considering limitations of previous clinical trials, our randomised, double-blinded, placebo controlled trial is expected to elucidate the effects of probiotics on glycaemic control, anti-inflammatory markers, anti-oxidative enzymes, gut hormones regulation and alteration of intestinal microbiota composition in T2DM patients.

Keywords: T2DM, probiotics, systematic review, quality of life, RCT

LABELLING OF DISPENSED MEDICINES ACCORDING TO LEGAL REQUIREMENTS: ANALYSIS AND COMPARISON OF COMMUNITY PHARMACIES AND PRIVATE CLINICS IN SELANGOR, MALAYSIA

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Abstract

Patient misunderstanding and lack of information in dispensed medication labelling may affect the therapeutic goals in medical healthcare system. The labelling of medications encompasses the provision of information and instruction to ensure the safe and effective use of products by patients. The label of a dispensed medication represents one of the most important sources of information available to patients. Lack of medication information may and thus increase the chances of medication error. Nonetheless, medication error that originates from labelling error could be eliminated and prevented from occurring. The objective of this study is to measure and compare the dispensed medication labelling compliance between community pharmacies and private UiTM panel clinics in Selangor. A cross sectional study was conducted from September 2015 to May 2016 which involve the total sample of 131 community pharmacies and 46 UiTM private clinics. Generally, clinic demonstrated higher compliance ($p < 0.001$) in term of providing essentials information such as name of patient, name and address of the clinic, date of supply as well as indication of the medicine. Clinics provide more sufficient information in most aspect of labelling. The government health care regulatory organization need to play a strong and leading role in promoting safe use of medication practice by ensuring the utilization of existing standardized medication labels. This research had thrown up many questions in need of further investigation. Further research need to be done in order to get clearer picture of a healthcare practitioner among pharmacies and clinics as a whole.

Keywords: labelling, dispensed medicine, community pharmacies, private clinics, medication error

RELATIONSHIP OF PATIENT CHARACTERISTICS AND ORGANIZATIONAL FACTORS WITH PATIENT-PROFESSIONAL RELATIONSHIP AMONG PATIENTS WITH TYPE II DIABETES IN PALESTINE

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Abstract

The patient-professional relationship is a major administrative, medical, and social factor that deserves attention by both patients and healthcare professionals and could influence health outcomes. An important factor in the successful implementation of World Health Organization and Palestinian guidelines for management of type II diabetes is the patient-professional relationship. The objectives of this study were to assess the patient-professional relationship and its relationship with patient characteristics and organizational factors in patients with type II diabetes. A cross-sectional study was conducted, adopting the Patient Perception of Diabetes Care Quality Questionnaire to assess organizational factors, and using the Patient Satisfaction with Professional Scale (PSPS) that included physicians, nurses, and dieticians to assess patient-professional relationship. Descriptive and comparative statistics were used to describe patient characteristics. All analyses were performed using SPSS v 16.0. Three hundred thirty patients with type II diabetes were enrolled in the study. The mean age of participants was 60 ± 9.7 years; 51.2% were male. The mean PSPS score was lower than the average score (mean = 38.5 ± 15.9 ; cumulative percentage = 48.1%). There was a significant difference in the median PSPS scores among patients received instructions from healthcare professionals (Kruskal-Wallis test; $p < 0.05$). The Spearman's rank order correlation coefficients showed a significant correlation of patient-professional relationship with preventive monitoring, preventive education, facilitating the patient-professional relationship, and collaboration. Instruction from healthcare professionals ($p < 0.001$), preventive education ($p < 0.001$), facilitating patient-professional relationship ($p = 0.001$), and collaboration ($p < 0.05$) were still statistically significantly related to patient-professional relationship after adjusting for covariates using multiple linear regression. As a conclusion, patient-professional relationship was inappropriate. Instruction and guidance may be an important barrier for achieving appropriate patient-professional relationship. Further improvement of guidance and educational programs that emphasize the role of healthcare providers could be helpful in a patient-professional relationship.

Keywords: Healthcare professionals, Organizational factors, Patient-professional relationship, type II diabetes, PSPS

THE IMPACT OF PHARMACIST COUNSELING ON PATIENT MEDICATION ADHERENCE AMONG JORDANIAN OUTPATIENTS

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Abstract

Pharmacist counselling plays an important role in improving patient's medication adherence that in turn will contribute to enhancing health outcome in patients taken eradication regimen. The objectives of this study were to assess the impact of pharmacist counselling on patient's medication adherence among patients who using *H. pylori* eradication regimen. A prospective Randomized controlled study was conducted, 200 dyspeptic patients, who at endoscopy were found infected with *H. pylori*. Patients were randomly assigned to an intervention group (n= 100) or a control group (n= 100). Intervention patients receive their medications via the hospital pharmacist (who took an education about *H. pylori* infection and eradication regimens) and were counselled (and follow up by the researcher). Control patients receive their medication via the hospital pharmacist (who did not take an education about *H. pylori* infection and eradication regimens) and undergoing the normal procedure without follow up and usual counselling. Two methods were used to investigate the impact of pharmacist counselling on patient's medication adherence, patient non-adherence reasons questionnaire (NARQ) to assess the reasons of patient's poor adherence, and pill count to assess the patient's medication adherence are done two weeks after completing the regimen. All analyses were performed using SPSS v 16.0. Intervention patients exhibited a statistically significant in medication adherence (69% vs 37%; $p < 0.05$, who took all prescribed doses). As a conclusion, structured patient counselling and follow-up can have a significant effect on improving patient's medication adherence and in turn, improving health outcome, and should be a routine part of therapy

Keywords: Pharmacist counselling, *H. pylori*, NARQ, patient medication adherence

GROUP-BASED HYPERTENSION SELF-MANAGEMENT EDUCATION (HSME) PROGRAMME: QUALITATIVE FINDINGS FROM PENANG, MALAYSIA

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Abstract

Hypertension affects approximately one in three Malaysians aged 30-year-old and above. Most patients have difficulties achieving good blood pressure control. A hypertension self-management education (HSME) programme which aimed to empower hypertensive patients with the necessary knowledge and skills needed to manage the disease outside of clinic setting was developed and delivered. However, participants' perception towards the quality and impact of HSME programme remains unknown. Hence, this study was conducted to explore the participants' impressions and experiences regarding HSME programme's structure and its potential effects. A total of three focus group discussions, involving 19 participants who had attended the 4-weekly intensive HSME programme, were conducted using semi-structured moderator guide. Data were audio recorded, transcribed verbatim and analysed using thematic analysis approach. Nvivo10 was used to manage all qualitative data. Most participants agreed that self-management education is crucial in managing hypertension and were satisfied with the HSME programme. They reported positive benefits gained from the programme particularly on healthy diet selection, be physically active and medication taking. Participants reported to have enjoyed the group-based self-management education as support was given among peers and educators of the programme. Furthermore, information on disease management were easy to follow and more detailed compared to their normal routine follow-up with doctor at clinic or hospital. The current study findings suggested that the HSME programme helped motivating participants to better care for their health. A localised, culturally-sensitive structured educational programme should be in place to empower hypertensive patients the knowledge and skills in self-managing hypertension, providing them the confidence to sustain daily self-care activities

Keywords: hypertension, self-efficacy, focus group, perception

ADEQUACY OF EMPIRICAL ANTIBIOTICS: PREDICATORS AND OUTCOMES AMONG SEPSIS ICU ADULT'S PATIENTS

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Abstract

Inadequate antibiotic treatment had an impact on high mortality and poor outcomes. The aim of the study was to determine the adequacy of empirical antibiotics (AEA) & reason of non AEA. The outcomes (survival, Severity APACHE II scores ICU length of stay LOS). A retrospective cohort study conducted in adult's ICU of Hospital Sungi Boluh among sepsis patients. Data were retrieved from patient's files and computer system. Each case has been reviewed for AEA based on ICU guidelines, bacterial sensitivity patterns, dose, frequency, creatinine clearance and time of empirical antibiotics. APACHE score was determined with online clinical calculator. We employed regression modeling to compute adjusted association of sepsis with the risk of receiving AEA/non AEA on ICU-mortality, ICU-LOS, and severity scores index. Among 228 ICU adult's patients the mortality rate was 193(84.6%). It was higher in male group (119) 52.2% than in female 74(32.5%). The mean ICU-LOS was (9.860 ±8.958) days while the mean APACHE II score was (29.596±7.490) points. The AEA was significantly associated with mortality & ICU-LOS ($P<0.005$). In logistic regression, the model of AEA was a predictor for survival (OR=.341, 95% CI .163-.714, $P<0.005$). The odd of survival is increased by 34.1% with AEA. In linear regression, the model of AEA was a predictor of ICU-LOS ($R^2=.055$, 95% CI -7.184--2.114). The patient with AEA was less likely to have ICU-LOS by 4.649 days. The main reason of non AEA was the antibiotics not covered isolated M.O 68(29.8%). While in multiple linear regression model, the predictor dose exceeding dose based on creatinine clearance (CrCL) was more likely to have increment of APACHE score by 5.844 points ($R^2=.074$, 95% CI (2.741-8.946). AEA is predictor for survival and ICU-LOS. The dose exceeding the dosage based on CrCL. was predictor of increased the APACHE II severity score for non AEA

Keywords: Adequacy, Sepsis, Empirical antibiotics, ICU, APACHE, ICU length of stay

ADHERENCE TO INSULIN THERAPY IN TYPE 2 DIABETES MELLITUS PATIENTS TREATED AT THE GOVERNMENT'S PRIMARY HEALTH CARE CENTERS IN KLANG

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Abstract

Insulin therapy is necessary in Type 2 Diabetes Mellitus (T2DM) patients to accomplish targeted glycaemic level and prevent diabetes-related complications. This study aimed to determine the proportion of adherence to insulin therapy in patients who attended Ministry of Health primary care centers and the associated factors leading to the adherence rates. This cross-sectional study was conducted among T2DM patients aged 18 years and above and who were on insulin therapy for at least three months. A systematic sampling method was used. It involved 249 subjects from five government's primary care centers in Klang. Patients were interviewed and records were accessed to collect data on socio-demographic characteristics, disease-related factors, treatment-related factors and clinical parameters. A self-administered validated questionnaire was used. Significant associated factors were identified by using Binary Logistic Regression. The proportion of adherence to insulin therapy was 8.43% (95% CI=0.05, 0.12). After adjusting all factors, three variables were found to be significant. A patient practicing self-monitoring of blood glucose (SMBG) has 5.49 times higher odds to adhere to insulin therapy (Adjusted OR=5.39, 95% CI=1.20, 24.13). A patient who exercises has 3.38 times greater odds to adhere to insulin therapy (Adjusted OR=3.38, 95% CI=1.37, 10.03). There is a 63% increase in the odds of adherence to insulin per 1 unit increase in the frequency of insulin dose per day (Adjusted OR=1.63, 95% CI=1.09, 2.44). Adherence to insulin therapy was poor. A patient who practices SMBG, exercise and more frequent insulin dose a day significantly affected the adherence level.

Keywords: Pharmacy practice, insulin, adherence, diabetes mellitus

PERCEPTION OF ELECTRONIC CIGARETTES AND PHARMACIST'S ROLE IN SMOKING CESSATION

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Abstract

Electronic cigarettes (e-cigarettes) are battery-operated devices designed to deliver nicotine with flavorings and other chemicals to users in vapor instead of smoke. While e-cigarettes are often promoted as safer alternatives to traditional cigarettes, little is actually known about the health risks of using these devices. The aims of this study were to investigate the perception of the public on e-cigarettes and pharmacist's role in smoking cessation. The instrument was adapted from Brown et al. (2014) and J. F. Etter & Bullen (2011) and included new questions relevant to the study consisted of four-parts self-administered 69 items questionnaire covering demographic, usage of conventional and e-cigarettes, perception of e-cigarettes and pharmacist' role in smoking cessation. The instrument was distributed in Batu Pahat Johor. The results were analyzed using Statistical Package for the Social Sciences (SPSS) version 20.0. Majority of the respondents were male (76.8%), 20-30 years old (44.9%) with SPM as the highest level of education (36.2%). Almost a third (29.9%) perceived that e-cigarettes are safer than conventional cigarettes but almost half (49.3%) perceived that both types of cigarettes as harmful on health. Three quarter (75.4%) did not believe e-cigarettes can be used as a smoking cessation tool. Majority of the respondents agreed that pharmacist can advice the public on smoking cessation (82.6%) and on safety and efficacy of e-cigarettes (81.2%), provide smoking cessation counseling (86.9%) and smoking cessation medication (79.7%). In conclusion, respondents agreed that pharmacist play a role in smoking cessation but more effort is needed in disseminating information on e-cigarettes

Keywords: electronic cigarettes, smoking cessation, pharmacist's role, perception

PREGNANT AND POSTPARTUM WOMEN'S PERSPECTIVES TOWARDS TRADITIONAL AND COMPLEMENTARY MEDICINE (TCM) USE

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Abstract

Traditional and complementary medicine (TCM) is flooding the Malaysian market, but reliable information resources are very limited. Therefore, it is important to look into how Malaysian pregnant and postpartum women define and use TCM. This study aims to explore the relationship between women's understanding about the meaning and use of TCM and their TCM use. A cross-sectional study was conducted using self-administered questionnaires. Descriptive statistics are utilized for selected variables. Out of 374 respondents, 285 (76.2%) reported using at least one type of TCM to conceive (n = 94, 25.1%), during pregnancy (n = 183, 48.9%) or during postpartum (n = 237, 63.4%). The majority of respondents indicated that TCM is all about plants or natural products (n = 267, 71.4%, p < 0.001), more than half assumed that TCM refers to home folk medicines (n = 230, 61.5%, p < 0.001) and are only supplements for a healthy lifestyle (n = 205, 54.8%, p = 0.001), while 168 (44.9%, p < 0.001) agreed that TCM includes any treatment that does not require going to hospitals. However, 57.2% (n = 214, p < 0.001) disagreed that TCM is more on believing in traditional healers, not using any medications or therapies. These views have significant associations with the TCM usage. The highest categories of TCM used is biological based therapies (n = 272, 72.7%) while the lowest is energy therapies (n = 8, 2.1%). Traditional Malay massage (n = 170, 45.5%), traditional Malay therapies (n = 147, 39.3%) and vitamins and supplements (n = 131, 35.0%) were the most commonly used T&CM. These findings can contribute to a better understanding of TCM use among Malaysian pregnant and postpartum women and may be useful to policy makers in deciding the scope of awareness needed to be instilled in the public.

Keywords: Traditional and complementary medicine, Quantitative, Pregnancy, Postpartum

PUBLIC PERCEPTION AND SATISFACTION TOWARDS THE ROLE OF COMMUNITY PHARMACIST: A CROSS SECTIONAL STUDY

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Abstract

Community pharmacist is one of the most accessible health professionals to the general public. They play an important role in patient's counselling and providing drug information. Since the role of community pharmacist has become more patient-oriented, there is a need to evaluate the perception and satisfaction towards the role and services provided by community pharmacist. This study was conducted to determine the perception of the public towards the role of community pharmacist and also to evaluate the level of satisfaction on the services provided. This study was conducted in Sungai Petani and Puncak Alam area using a cross-sectional method. Self-administered questionnaire consists of 4 sections which were demographic backgrounds, patient's access to pharmacy, perception on the role and satisfaction level, were distributed. Results were analyzed using 'Statistical Package for the Social Sciences' (SPSS) version 22.0. A total of 142 respondents were obtained in which majority were female (66.9%), Malay (92.3%), aged between 18-23 years (40.8%) and not working or studying in health-related sector (67.6%). The public had a good perception regarding the role of community pharmacist and believed that a pharmacist should answer their drug related questions (94.4%). Majority were also satisfied with the language used by pharmacist (84.5%). There was statistically significant difference in ethnicity and involvement in health sector with the satisfaction level on the services of the community pharmacist. Generally, the public had a good perception on the role of community pharmacist and were satisfied with the services provided to them.

Keywords: community pharmacist, perception, satisfaction level

QUALITY OF LIFE OF UNDERGRADUATE PHARMACY STUDENTS IN UNIVERSITI TEKNOLOGI MARA MALAYSIA

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Abstract

Quality of Life (QoL) is defined by World Health Organization (WHO) as an individual's perception of their position in life in the context of the culture and value systems in which they live related to their goals, expectations, standards and concerns. Therefore, QoL is very subjective as it depends on individual self-evaluation. The QoL of undergraduate pharmacy students in Malaysia remains unknown. Hence, this study was aimed to investigate the QoL of undergraduate pharmacy students in Universiti Teknologi MARA (UiTM), Malaysia. A 26-item WHOQoL-BREF questionnaire, consisted of four domains (i.e. physical health, psychological health, social relationship and environmental), was used to assess the QoL of pharmacy students structure. Data were then analyzed using SPSS version 22.0. Descriptive and inferential analysis were performed. A total of 256 UiTM pharmacy students were surveyed. Findings revealed that UiTM pharmacy students perceived moderate satisfaction in QoL. Among the four domains of QoL, UiTM pharmacy students reported the lowest score in social relationship and the highest in environmental domain. The QoL of students were found to be significantly influenced by the year of study, residency and mode of transportation. The scores in physical and environmental domains among the UiTM pharmacy students were noted to be improved across the year of study. Students who lived with parents showed better physical domain score than those who stayed in college. It is noted that the pharmacy students who came to university by bus had the lowest score in all domains compared to other modes of transportation. Overall, the UiTM pharmacy students reported a moderate score in each QoL domain. Future studies can be done to explore the ways to improve the QoL among UiTM undergraduate pharmacy students.

Keywords: quality of life, perception, pharmacy

A STUDY OF SELF-ESTEEM AND ITS RELATIONSHIP TO ACADEMIC PERFORMANCE AMONG UNDERGRADUATE PHARMACY STUDENTS IN UNIVERSITI TEKNOLOGI MARA MALAYSIA

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Abstract

Self-esteem is defined as positive and negative attitude to own personality. There are various factors involving internal and external criteria that can influence personal self-esteem. Education is one of the aspects that affecting a student's self-esteem. A direct relationship between a student's self-esteem and their academic performance was found, but there was a sensible difference between self-esteem of students across different faculties. As no study has been conducted among undergraduate pharmacy students, this study aimed to determine the level of self-esteem among 239 undergraduate pharmacy students in Universiti Teknologi MARA (UiTM) and to determine the relationship between self-esteem and academic performance. The survey was conducted using a 10-item Rosenberg Self-esteem Scale and cumulative grade point average (CGPA) was collected to evaluate their academic performance. Statistical Package for Social Sciences Program (SPSS) version 22.0 was used to analyse the data. Descriptive statistics (i.e. mean, median, standard deviation and average) were employed. The normality of the distribution for each variable was tested prior to conducting any inferential statistics for continuous data. A total of 256 questionnaires have been distributed among the UiTM undergraduate pharmacy students and only 239 students responded to this survey, giving a response rate of 93.4%. It was found that the level of self-esteem among UiTM undergraduate pharmacy students was moderate. When comparing the self-esteem level across the years of study, no significant difference was found. However, there was a negative correlation between the self-esteem level and academic performance among the UiTM undergraduate pharmacy students. Future studies should evaluate more factors to gain a better perspective on this research area.

Keywords: self-esteem, perception, pharmacy

AN ASSESSMENT OF KNOWLEDGE OF HAND FOOT AND MOUTH DISEASE (HFMD) AMONG CHILD CAREGIVERS AND TEACHERS IN CHILDCARE CENTERS AND PRESCHOOLS IN KLANG VALLEY

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Abstract

Hand foot and mouth disease (HFMD) has become a serious health concern worldwide. In Malaysia, the prevalence of HFMD has reached the outbreak proportions. To date, there is no effective vaccine available for HFMD; therefore, the spread of HFMD can only be controlled by taking the preventive measures. The aim of study was to assess the level of knowledge of HFMD among child caregivers and teachers in childcare centers and preschools in Klang Valley, Malaysia. In this cross-sectional study, a total of 212 child caregivers and teachers from childcare centers and preschools were enrolled using cluster sampling technique. The respondents' responses were recorded on a self-administered pre-validated questionnaire. The study questionnaire consisted of two main parts. Part A of the questionnaire recorded the socio-demographic characteristics of the respondents including age, gender, marital status, education, family income, method to handle HFMD, experience of HFMD outbreak, and the number of children they take care of in their working premises. In part B the knowledge of participants regarding the cause, transmission, symptoms, prevention and treatment of the HFMD was assessed. The results indicated that majority of the respondents (n=144, 67.9%) possessed poor level of knowledge; whereas, 56% (n=26.4%) and 5.7% (n=12) participants showed moderate and good level of knowledge of HFMD, respectively. In addition, majority of the respondents were lacking knowledge in each subscale of knowledge questionnaire: cause, transmission, symptoms, prevention and treatment of the HFMD. As Malaysia is on alert level following an outbreak of the hand, foot and mouth disease (HFMD) nationwide, there is a need to educate the Malaysian child caregivers and teachers in childcare centers and preschools.

Keywords: Knowledge, Hand foot and mouth disease, Malaysia, Students

EFFAVIRENZ INDUCED GYNAECOMASTIA IN MALE WITH HUMAN IMMUNODEFICIENCY VIRUS INFECTION (HIV)

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Abstract

Highly active antiretroviral therapy (HAART) has dramatically reduced HIV-associated morbidity and mortality and has transformed HIV disease into a chronic, manageable condition. Widespread use of potent Anti-Retroviral Therapy has effectively reduced opportunistic infection-related mortality among HIV infected persons, improving quality of life and survival. With the introduction of HAART specifically efavirenz (EFV), however, gynaecomastia is being frequently reported in HIV infected men. We present a case of 52 years-old, malay man positive for human immunodeficiency virus infection and on an efavirenz treatment regime. He presented to the Infectious Disease (ID) clinic at a publicly-funded government hospital with gradual intermittent on the right breast since 2010 and after 2 years similarly on the left side. In April 2016, this condition reportedly worsened and the size of breast was in fact increasing and became more painful. Ultrasound of the breast to confirm the bilateral gynaecomastia, likely secondary to EFV was done. Finally, Efavirenz was discontinued by the doctor and was switched to Nevirapine. Subsequently, his condition improved. Recognition of gynaecomastia in men as a side effect of Efavirenz is important to note as this drug is being used as a preferred component in the first line treatment of HIV infection worldwide.

Keywords: Efavirenz, HIV, Gynaecomastia

CANCER PATIENTS DISCLOSURE ON COMPLEMENTARY AND ALTERNATIVE MEDICINE TO PRESCRIBERS

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Abstract

Managing the side effects of cancer treatment is challenging. Despite chemotherapy side effects were being treated with conventional treatment, some patients still seek treatment by complementary and alternative treatment; and the trend is increasing, hence leaving the cancer patients taking CAM at high risk of adverse drugs reactions due to drug- CAM interactions. The goal of this study is to monitor the prevalence of patients disclosing CAM use for CRSE to their prescribers. This is an interviewer -assisted questionnaire. Data were collected from cancer patients attending to three departments; surgical, medical and gynecology. 273 patients were recruited. 166 used CAM for CRSE management with a prevalence of 60.8%. High percentage of CAM users include female (86.7%), educated (39.8%), employed (79.1%) and married (74.1%). Breast cancer patients were found to be the highest users of CAM (97.4%). 82.5% (n=137%) patients admitted to their prescribers on their CAM use. 21.1% (n=29) doctors agreed or encouraged the use of CAM, 65% (n=89) disagreed and suggested to discontinue CAM while 13.9% (n=19) are unbiased. The prevalence of CAM use in management of CRSE is increasing. This study finds that it is associated to gender, employment, marriage status and education level. There are still prescribers who disagree with combining chemotherapy with complementary medicine, which inhibit the patients to share their use of CAM, hence making patients to use the treatments or products discretely without fully knowing or understanding the danger that might occur due to drug-CAM interactions. More research has to be done to increase understanding and awareness of CAM use and its potential adverse reactions due to interaction with chemotherapy.

Keywords: Complementary and alternative medicine, chemotherapy related side effects, cancer patients, CAM use disclosure to prescribers

DIFFERENTIAL FAECAL MICROBIOTA BETWEEN TYPE 2 DIABETES MELLITUS (T2DM) AND HEALTHY INDIVIDUALS: A SYSTEMATIC REVIEW

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Abstract

There is increasing evidence which supports the association between gut microbiota and diabetes. Dysbiosis of gut microbiota results in the loss of barrier function, allowing translocation of microbial products like lipopolysaccharides (LPS) and lipoteichoic acid (LTA) into the plasma. LPS and LTA promote production of endotoxins, leading to metabolic endotoxemia that contributes to the development of inflammation as seen in T2DM patients. This review highlighted the latest clinical findings with regard to the differential composition of gut microbiota in T2DM patients. Literature search (January 2017 to March 2017) was performed using electronic databases which included PubMed, Embase and Web of Science. Language of articles was restricted to English only. Search terms that were adopted included "type 2 diabetes mellitus", "metagenomics", "gut microbiota", "16S rRNA" and "clinical trial". Additional relevant papers were identified by screening references of the included studies. Relevant studies were selected by independent reviewers according to a set of inclusion and exclusion criteria. Relevant data were extracted and assessed for risk of bias. Seven case-control studies, involving 371 T2DM patients and 371 healthy controls, were included in this review. Whilst four studies were conducted in China, others were in Japan, Europe and Denmark. Majority of the studies were presented with medium risk of bias. Of the seven studies, four showed Firmicutes phylum to dominate the gut of healthy individuals. Overall, T2DM patients exhibited marked changes in intestinal microbiota whereby Firmicutes (74%) dominated the phyla, followed by Bacteroidetes (12%), Proteobacteria (10%) and Actinobacteria (4%). At genus level, five studies demonstrated correlation of *Lactobacillus* with glucose level. Butyrate-producing bacteria were depleted in T2DM patients. This review had identified that gut microbiota is involved in the pathogenesis of T2DM even though some inconsistencies were observed. Dietary consumption, race, geographical locations and experimental methods could be the confounding factors contributed to the inconsistencies.

Keywords: T2DM, intestinal microbiota, 16S rRNA, metagenomics, dysbiosis

ACKNOWLEDGMENT

Special thanks and appreciation are dedicated to Universiti Teknologi MARA (UiTM) Malaysia for making this significant conference possible. Nor forgetting, a big thank you to our organising committee especially the staffs of Faculty of Pharmacy, UiTM for their effort in making this event a successful one. Appreciation also goes to our generous sponsors below.

