KEYNOTE ADDRESS

Keynote Address
A pharmaceutically scientific approach to treating Abeta amyloid as the cause of Alzheimer's disease

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The etiology of Alzheimer's disease (AD) is best understood through the deposition of Aβ-amyloid (Aβ). There are two basic forms of AD. The common (>95%) form is sporadic and is caused by the failure to clear Aβ (mean age at onset 80 years). The rare (< 5%) autosomal dominant familial form is caused by the over-production of $A\beta_{42}$, also on a background of failure to clear (mean age at onset 45 years). In both forms, the kinetics of Aβ accumulation are similar, taking about 30 years to accumulate a total of approximately 7mg of Aβ. Thus, we estimate that sporadic AD starts about the age of 50 years and the autosomal dominant form starts about 15 years of age. The advent of validated biomarkers (PET/CSF Aβ and tau) now provides us with unprecedented opportunities for preclinical diagnosis, enabling the development of primary and secondary prevention strategies. Predictive algorithms utilizing age, biomarkers, polygenic and vascular risk scores are now being developed from longitudinal cohort studies to estimate times of onset and rates of cognitive decline. Applications of biomarker screens (blood, CSF, PET) to subjects who are about to cross the lower cut point threshold will define a population who may be suitable for primary prevention clinical trials.

Therapeutic targeting the Aβ pathway remains the principal strategy for delaying onset of AD. There are many molecular targets in this pathway, and no single one is likely to prove efficacious on its own. Therefore, a combination of strategies needs to be developed and applied.

PLENARY LECTURE

PL1

POTENTIAL THERAPEUTIC TARGETS FOR THE TREATMENT OF TAMOXIFEN-RESISTANT BREAST CANCER

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Breast cancer is the most common malignancy in Western women and grows under hormone-dependent control. Hence, the ability to reduce breast tumor growth through the administration of anti-estrogens has played a key role in the endocrine therapy of breast cancer. The non-steroidal anti-estrogen, tamoxifen (TAM), is the most widely used anti-estrogen in estrogen receptor-positive breast cancer patients. Although most patients are initially responsive, resistance to TAM is a critical problem for anti-estrogen therapy. To mimic this condition, we established an MCF-7 derived TAM-resistant cell line (TAMR-MCF-7 cells) by long-term culture of MCF-7 cells with 4-hydroxytamoxifen in 2007. RNA sequencing analysis using MCF-7 and TAMR-MCF-7 cells showed that many coding and non-coding RNAs regulating both estrogen signaling and epithelial mesenchymal transition were differentially expressed in both the cell types. In this presentation, I will briefly summarize our previous studies identifying potential targets to overcome TAM resistance and the related pharmacological approaches.

Keyword: breast cancer, EMT, tamoxifen-resistance, therapeutic targets

PL2
Vietnamese ginseng – from an ethno-medicine to a national product of Vietnam

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Panax species occur in the northern hemisphere from Central Himalaya to North America through China, Korea and Japan. This genus includes the well-known medicinal plant *Panax ginseng* C.A. Meyer (Korean or Asian ginseng) and its two congeners, *P. notoginseng* (Burk.) F. H. Chen (Sanchi ginseng), and *P. quinquefolium* L. (American ginseng), which have been widely used in many countries of the world and are important plants in terms of therapeutic uses and economic values.

In 1973, a wild *Panax* species was discovered at the elevation of 1,800 m above sea level of Ngoc Linh Mount in Middle Vietnam. The plant used to be a secrete tonic and body-strengthening ethno-medicine of the Sedang ethnic group living in high mountains of the Truong Son Range. In 1985, it was defined as a new *Panax* species with the scientific name *Panax vietnamensis* Ha et Grushv, Araliace family, and is now commonly known as Vietnamese ginseng (VG) which is used for many indications similar to those of *Panax ginseng* (PG), such as enhancement for physical strength, tonic, lowering blood cholesterol, preventing cardiovascular diseases etc.

Since then, scientific studies of VG on botany, cultivation, chemistry, bio-activities, etc., have been done. The result showed that VG contains a characteristic saponin composition, which includes not only known dammarane saponins found in PG such as ginsenoside-Rb_1, -Rb_3, -Rg_1, -Rd, -Re, etc., but also ocotillol saponins in high yield, especially majonoside-R2 (more than 5%). Twenty-five (25) new dammarane saponins named vina-ginesnnosides-R1-R25 from the underground part and 8 named vina-ginsenosides-L1-L8 from the leaves were also isolated and identified. The underground part of VG contains up to 15-20% saponins, which is the highest content compared with that of PG (4-6%) and other *Panax* spp. As for pharmacological activities, VG showed similar effects with those of PG, including tonic, dose-dependent stimulation/depression on CNS, physical strength enhancement, analeptic, antifatigue, adaptogenic, androgenic, anti-tumorigenic etc. VG also showed remarkable physical and psychological anti-stress activities.

Results of scientific studies have proven that VC is a trustful herbal medicine. It has therefore become an important medicinal plant of Vietnam in terms of theuraputic uses and economic value. Recently, the Vietnam government defined VG as an important national product. A national program was set up to protect the wild plant and the biodiversity of its native areas, and to develop the large-scale cultivation of VG. An updated review on VG will be reported to show how the used-to-be ethno-herb VG becomes an national profduct and its impact to the contemporary Vietnam medicine.

Keywords: Vietnamese ginseng, *Panax vietnamensis*, ethno-medicine, Vietnam national product

PL3

Curcumin analogues

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Curcumin is well-proved as an antioxidant, anti-inflammatory, antivirus agent, and anticancer. Curcumin and its derivatives in the ginger plant have antibacterial activities. However, the pre-clinical and clinical studies showed that its bioavailability and pharmacokinetic profile are low.

Design, synthesis and study on antibacterial properties of curcumin analogues by modifying 5-chloro-2-furanyl moiety through aldol condensation reaction has been conducted. Synthesis of monocarbonyl analogue by changing β-diketone into β-monoketone is one of the convenient approaches for structure modification of curcumin. Some synthesized curcumin analogues showed better antibacterial activity than that of curcumin and amoxicillin.

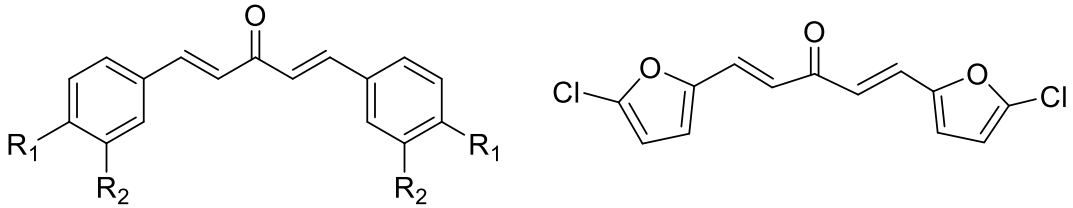

Fig. Structure of curcumin analogues

In addition, a computational study through virtual screening has found some curcumin analogues for some other activities. The results will be discussed.

Keywords: curcumin analogue, synthesis, antibacterial, virtual screening

PL4

Gene-Chemical Interplay in Alzheimer's Disease

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Alzheimer's disease (AD) is a neurodegenerative disease that debilitates numerous human psycho-behavioural functions, notably memory processing. In the developed world AD is considered as one of the major causes of death. In the developing world, the number of people living with AD (PLWAD) is expected to rise significantly in the coming decades. Though AD is more prevalent among the elderlies over 65 years old, cases of early onset AD are also widely known. Both types of AD are linked to genes. Much research is ongoing to elucidate the exact pathophysiology of AD, hence leading to its ultimate cure. Chemicals whether working in synchrony or otherwise, are known to be responsible for the preservation or destruction of the brain function, respectively. Firstly, putative neurotransmitters in normal brain physiology related to AD include acetylcholine, dopamine, serotonin, noradrenaline, aspartate and GABA. Secondly, chemicals that precede the pathology of AD. Among them are the pro inflammatory mediators, the levels of which are constantly checked by anti-inflammatory mediators. Thirdly, the group of chemicals found to play an important role in AD is the pathological proteins. Among them are beta-amyloid, hyperphosphorylated tau and alpha-synuclein. The formation of these proteins leads to the neuronal dysfunction that contributes to the psycho-physical disability of PLWAD. At the heart of the chemical homeostasis or imbalance are the genes. Six genes identified in a Malaysian cohort of PLWAD will be highlighted. The over- or under-expression of these genes tilts the chemical homeostasis, which ultimately promotes the manifestation of symptoms of AD.

INVITED LECTURE

IL1

Discovery of non-pungent TRPV1 agonist as strong topical analgesic

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Background
TRPV1 is a ligand-gated and nonselective cation channel activated by multiple factors such as proton, heat, endogenous ligand and natural vanilloids. Activation of TRPV1 triggers an influx of calcium and sodium, which initiates a cascade of events associated with pain transmission, including membrane depolarization, neuronal firing, and release of pain transmitters. Desensitization by repeated application of an agonist like capsaicin produces the analgesic effect, but the pungency by initial activation is the adverse effect to be overcome. Recently, we discovered MDR-652, a non-pungent agonist with strong analgesic profiles.

Methods
In this study, we performed the optimized scale-up synthesis of MDR-652, the agonistic activity *in vitro* and *in vivo* and, its analgesic activities in acute and neuropathic pain models by topical and intraperitoneal administrations, respectively. In addition, we conducted the pharmacokinetic (PK) and toxicological studies for the topical agent to be developed.

Results
MDR-652 demonstrated high affinity and potent agonism in human and rat TRPV1 *in vitro*. The functional agonism was confirmed *in vivo* through manifestation of hypothermia in body temperature study of mouse subjected to both coadministration with capsaicin and single administration. MDR-652 exhibited excellent analgesic profile, which was superior to capsaicin and gabapentin, in the standard pain models, formalin and spinal nerve ligation model. In addition, it exhibited dose-dependent analgesic profile toward capsaicin-induced allodynia, indicating that it was engaged in TRPV1 for analgesic activity. *In vitro* transdermal delivery test indicated most of MDR-652 remained in the skin and had little systematic absorption. The major of capsaicin, however, was detected in cell acceptor, showing that it has promising PK property as a topical agent. In the toxicology study, MDR-652 not only showed minimal signs of skin irritation and oedema in animal models, much lower LD_{50} when compared to that of capsaicin but also found to be negative in all genotoxicity studies.

Conclusions
MDR-652 is a highly potent and efficacious TRPV1 agonist with promising topical PK profile and no significant toxicity. It is currently under clinical development as a topical agent for neuropathic pain.

Keywords: TRPV1, agonist, capsaicin, analgesic

IL2

Developing proposed national competency framework for pharmacists in Vietnam

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Background
In recent years, the number of pharmaceutical human resource training institutions has been increasing (28 establishments) including public and non-public establishments. However, the training program, facilities, quality of teaching staff, quality of students' inputs and especially the way of implementing training programs, training organization capacity of each institution is different, so the quality of output products, quality of practice is also different. Therefore, it is necessary to have basic competency standards for pharmacists in Vietnam. On the other hand, in the face of extensive regional and international integration needs, managers and employers need to have a set of tools to control, evaluate and standardize the quality of human resources. Recognizing that reality, the Ministry of Health has directed the construction of the Basic Competence Standard for Pharmacists in Vietnam with the participation of all stakeholders including experts in the field of training, employers, Employers, managers, professionals, social organizations. In the process of construction, the Drafting Board has consulted the standard of competencies of pharmacists in the region and the world to adjust to suit the situation in Vietnam. Therefore, this study was carried out to develop a basic competency framework for pharmacist in Vietnam.

Method
The study was conducted by a method of retrospective and cross-sectional descriptions, combining qualitative research (method of in-depth interview; group discussion) and quantitative research based on FIP and Thailand pharmacist competency standards. The data is processed on SPSS software.

Results
A basic competency framework was developed for pharmacist in Vietnam. 98 competencies required for pharmacist, organised into 24 standards, 7 domains: professional and ethical practice, communication and collaboration, organisation and management, quality assurance of pharmaceutical, prepare pharmaceutical products, supply of medicines, safe and rational use of medicines.

Conclusion
The proposed competency framework of pharmacist in Vietnam provides a solid foundation for both pharmacy training and curriculum development and is based on several rounds of scientific research. The proposed competency framework may help understand the pharmacist role and how to best prepare for the Practice of Pharmacy and many added values for stakeholders.

IL3

Tumor microenvironment-modulating anticancer chemo/immunotherapy using nanomaterials

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Nanostructures such as nanosheets, and nanoballs have been studied for delivery of chemical anticancer drugs and oligonucleotides. Although, numerous studies have been done, the translation to commercialized products has not been successful. We aimed to design nanosystems which can be activated in tumor microenvironment for therapeutic and prevention purposes. Graphene-based nanosheets were prepared for tumor microenvironment-responsive anticancer drug delivery. The biofunctionalization of graphene-based nanosheets with melittin peptide derivatives of phospholipids selectively activated the release of melittin in tumor microenvironment. The activation of pore-forming melittin in tumor tissues increased delivery of anticancer drug-loaded GNS to tumor cells. Moreover, the overexpression of matrix metalloproteinase fibroblast-associated protein in tumor microenvironment was used for responsive delivery systems. We designed graphene oxide (GO) nanotheranostics loaded with fibroblast associated protein-activable promellitin derivative. As a tumor microenvironment-activable therapeutic model molecule, pore-forming promellitin chimeric peptide was designed. For comparison, GO loaded with scrambled chimeric peptide was designed. For immunotherapy, adjuvant-loaded nanoparticles were modified with immune checkpoint blockade. In tumor-bearing xenograft, the surface-modified GO provided selective activation by cleavage of fibroblast-associated protein at tumor microenvironment, and greater antitumor effect than other comparison groups. Moreover, we designed an adjuvant-entrapped nanoparticle which can assemble with tumor antigens in situ for effective activation of immune cells. The systemic administration of adjuvant-entrapped nanoparticles with light irradiation increased the activity of immune cell infiltration to the tumor cells, and inhibited tumor growth. These studies provide the potential of tumor microenvironment-responsive delivery of chemicals and adjuvants for next generation nanomedicine products.

IL4

Improvement pharmacological activities of compounds by nanotechnology approach

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The synthesis of nanoparticles has introduced nanotechnology during the last two decades that produced novel compounds applied in various fields. The big issue regarding generate material nanoparticles is increasing surface area of materials and improvement physiochemical properties of substances. In addition, plants are known to be an important component of different ecosystems. Exploiting this beneficial of nanoparticles exhibits new valuable activities of substances especially secondary metabolic of plants which is obtained from plant by extraction. Applied nanotechnology into plant extract generate improvement pharmacological activities of extract in comparison with original extract. Moreover, the extracts nanoparticles provide improvement treatment against of several acute and chronic diseases such as malaria, gout, tuberculosis (TB) and cancer. In others wise plant extracts as biodegradable materials is relative secure for consuming by human.
The objectives of the research were to report improvement pharmacological activities of the plants extract nanoparticles due to increasing various activities such as antioxidant, antimicrobial, anticoagulant and inhibit xanthine oxidase.

Key words: nanoparticles, plant extract, pharmacological activities

IL5

Vietnam pharmaceutical industry: Actual status and perspectives for decade 2020-2030

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The presentation described an overall outlook on the actual status of Vietnam pharmaceutical industry, which is considered as one of the fastest developing sectors among the emerging countries in pharma-industries. The Vietnam general and healthcare indicators were presented and analysed. The presentation also gave a SWOT analysis of the Vietnam pharmaceutical industry. The perspectives of Vietnam pharmaceutical industry for decade 2020-2030 were analysed, based on the policies and strategies determined relevant to the resolutions of the Government of Vietnam especially in New Drug Law 2016. The factors impacting into the process of modernization of Vietnam pharmaceutical industry were discussed and suggested for realization of the objectives of the Vietnam pharmaceutical industry development in the context of deeper participation of the nation in the process of economic globalization in ASEAN and the world.

IL6

Phytochemicals: a new insight into regenerative medicine

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Targetting the impairment of stem cells in disease models and conditions become a primary target of the modern therapeutic approaches. Stem cells sit at the top of the cellular hierarchy, maintain the structure and homeostasis of an organ by uninterrupted tissue-specific cells' supply. Besides, the ageing and diseases processes affect stem cells, many chronic diseases such as cancers, diabetes and other organ-related diseases are consequent of functional impairment of stem cells. Phytochemicals, whose therapeutic activities are not only limited to the somatic cells but showcasing a profound impact on stem cells too. To date, not much research data are available regarding the effect of phytochemicals on stem cells. Amongst, *Moringa oleifera*, a local plant, has exhibited a profound impact on adult mesenchymal stem cells (MSCs). Mesenchymal stem cells are found mainly in the bone marrow, which promote haematopoiesis, alleviate inflammation and mediate tissue repair. In line with this, the ethanol extract of *Moringa oleifera* (MOEE) boosted the proliferation of human MSCs. The enhanced proliferation activity of MSCs was due to an intensification of the cell cycle with reduced apoptosis. The treatment of MOEE altered the cytokine secretory profile of MSCs depicting anti-inflammation with enhanced expression of growth factors that mediate tissue repair. Similarly, various administrations of MOEE in a rat model of immunosuppression showed reconstitution of immune cells by preserving the bone marrow-derived haematopoietic stem cells (HSCs) and MSCs. The phytochemicals from MOEE showed a promising way of recovering immune cells and immunity in degenerative diseases. However, the identification and isolating specific compound/s to accelerate the desired therapeutical properties and challenges of diversified actions in a multiorgan system need to be addressed prior to clinical applications.

IL7

Improving cardiovascular function by targeting endothelial Ca^{2+}-activated K^+ channels

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A hallmark feature of cardiovascular disease is the presence of endothelial dysfunction, an early causative event in the development and progression of cardiovascular complications. Endothelial small and intermediate-conductance, Ca^{2+} -activated K^+ channels play an important role in the regulation of vascular function and systemic blood pressure. Growing evidence reports that they are intimately involved in agonist-evoked vasodilation of small resistance arteries throughout the circulation. Results from our study reveal that SKA-31, a small molecule activator of KCa2.3 and KCa3.1 channels, can acutely inhibit myogenic tone in isolated resistance arteries, induce effective vasodilation in intact vascular beds, enhance coronary circulation, and acutely decrease systemic blood pressure in vivo. Our data also demonstrate that long term administration of SKA-31 in T2DM and ageing conditions can improve cardiovascular function without adversely affecting T cell populations thereby increasing its potential therapeutic benefit in mitigating endothelial dysfunction associated with cardiovascular disease. The blood pressure-lowering effect of SKA-31, and early indications of improved endothelial function suggest that endothelial KCa channel activators could eventually be developed into a new class of endothelium targeted agents to combat hypertension or atherosclerosis.

IL8

Monitoring of adverse drug reactions as a tool to improve patients' compliance

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Adverse drug reactions (ADRs) are one of the leading causes of morbidity and mortality in health care facility. The Institute of Medicine reported in January of 2000 that from 44,000 to 98,000 deaths occur annually from medical errors. Of this total, an estimated 7,000 deaths occur due to ADRs. ADR is a noxious and unintended response to a medicine that occurs at normal therapeutic doses used in humans for prophylaxis, diagnosis, or therapy of disease, or for the modification of physiologic function. ADRs and events constitute a serious problem increasing morbidity and mortality and health care costs worldwide. The information collected during the pre-marketing phase of drug development is inevitably incomplete with regards to possible ADRs. This is mainly because tests in animals are insufficient to predict human safety; patients used in clinical trials are selected and limited in number, the conditions of use differ from those in clinical practice and the duration of trials is limited, by the time of licensing exposure of less than 5000 human subjects to a drug allows only the more common ADR to be detected. Information about rare but serious adverse reactions, chronic toxicity, use in special groups (such as children, the elderly or pregnant women) or drug interactions is often incomplete or not available. Thus, post-marketing surveillance is important to permit detection of less common, but sometimes very serious ADRs. Therefore, health professionals should report on ADRs as it can save their patients' lives and others. Several studies have been carried out in health care facilities, including in hospital, and primary health care in Bandung, Indonesia. Tuberculosis/HIV is a major public health problem in Indonesia. ADRs are a great challenge to national anti-tuberculosis and HIV program. The ADR can negatively affect the compliance, which can result into therapeutic failure and may indirectly contribute to MDR-TB. Antipsychotic medication also causes a wide range of adverse effect, which can be serious and may further harmful both the physical and psychological health of schizophrenic patients.

Keywords: Adverse drug reactions, compliance, tuberculosis, antipsychotic

IL9
Role of Microbial-Catalysed Biotransformation In Sustainable Medicinal Chemistry

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Background
Over the past few years there has been an upsurge interest from medicinal chemistry groups in embracing the philosophy and tools of green chemistry. This philosophy is in part a driver to move towards more sustainable practices, but there is also an interest in using emerging new technologies to speed up the drug discovery process and to discover new and diverse structures as scaffolds and lead compounds. Microbial-catalysed biotransformation plays an important role in the production of commercially valuable steroids and terpenes for therapeutic use by the pharmaceutical industry with the advantage of high stereo- and region-selectivity, which additionally fulfils green chemistry principles.
Methods
Different bioactive natural products have been exposed to the microbial bio-catalysis as an attempt to find further lively and fewer toxic products. Initially screening of selected steroids and terpenes were performed with different fungi. Preparative scale started upon detection of biotransformed products. Resulted metabolites were isolated and elucidated using HPLC, LC-MS, ID and 2D NMR spectroscopic techniques. Resulted metabolites were screened for bioassays including anti-inflammatory, α-glucosidase inhibitory, tyrosinase inhibitory, acetylcholinesterase inhibitory and antiproliferative assays, respectively. The binding interactions of compounds were studied by molecular docking studies.
Results
Novel products were obtained during biotransformation of multifunctional steroid and terpenoid drugs with growing cultures of fungi from different biotopes. Some of the products showed more than or comparable activities to the standard inhibitors.
Conclusions
Hence, the identification of these novel compounds opens the possibility of producing more promising pharmaceutical agents with potential bioactivities with lesser side effects than the existing drugs.

Keywords: Biotransformation, Steroids, Terpenes

IL10

Potential delivery of polymeric-based nanocarrier loading drug or genetic material for the treatment of malaria

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Background

Malaria is an infectious disease in human caused by a parasite of *Plasmodium falciparum*. The main factor that has contributed to the spread of this disease is the increase in the number of drug-resistant parasites. To overcome, we approached two strategies by delivering artemisine drug loaded nanocarrier to the food vacuole and inhibiting specific *Plasmodium falciparum* gene of dhs encapsulated nanocarrier. We attempted to develop an efficient nanocarrier for selective targeting to the *Plasmodium falciparum*.

Methods

Nanocarriers were prepared by ionic gelation or nanoprecipitation method and were consisted of chitosan, poloxamer or polylactic glycolic acid (PLGA). To examine the pharmacokinetics profile of the nanocarriers *in vivo*, they were labeled with inorganic fluorescence nanocrystal of CdSe/ZnS. The nanocarriers were intravenously injected into mice and blood was collected at indicated time. To examine the function of nanocarriers, artemisinin or ODN targeted dhs was encapsulated into the nanocarriers. They were characterized of particle size, entrapment efficiency, release profile, morphological form by Scanning- or Transmission- Electron Microscope (SEM/TEM). To enhance stabilty of nanocarrier loading ODN particularly for shipping and handling, lyophilization process was performed by adding cryoprotectant to the nanocarrier suspension. Lastly, *in vitro* antimalarial assay was performed by observing the inhibition of *Plasmodium falciparum* growth through thick blood smears under microscope for the nanocarrier loading artemisinine and Lactate Dehydrogenase (LDH) assay method for the nanocarrier encapsulating ODN.

Results

The higher poloxamer concentration on the nanocarriers surface, the longer their circulation in the bloodstream. The size of nanocarriers was less than 200 nm with high entrapment efficiency of more than 80%. Additionally, artemisinin was released 80% after 48 h. When the nanocarrier loading artemisinine was used, antimalarial activity was inhibited 70%. The optimum cryoprotectant to protect freezing and drying stress of the nanocarriers was 15% lactose, 10% glucose and 10% mannitol. The use of nanocarrier containing ODN targeted dhs resulted around 50% inhibition of antimalarial activity.

Conclusions

Collectively, artemisinin as well as ODN targeted dhs encapsulated nanocarriers could be developed that targets to its site specific for the treatment of malaria.

Keywords: nanocarrier, artemisinin, oligodeoxynucleotide (ODN), dhs, *Plasmodium falciparum*

IL11

Pharmacogenetics: to do or not to do? Four cases and a discussion

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An obstacle in providing efficient and effective drug treatments in Malaysia are Inter-individual and inter-ethnic variations of drug metabolism. This issue is evident especially for drugs with narrow therapeutic windows. This is an overview contributing to the understanding of the molecular mechanism of these genetically influenced variations.

Case 1 discussed the inter-individual and inter-ethnic differences of drugs metabolised by the CYP2C9 enzyme. We observed a wide inter-individual difference in the metabolic ratio of losartan to E-3174 metabolite among *CYP2C9*1/*1* genotype population group. CYP2C9 metabolic rate differences was observed for the drug losartan between Koreans and Swedes.

Case 2 investigated the possibility of Behcet's disease as a *CYP2C9* metabolism effector. We observe that Behcet's disease patients tend to have a low CYP2C9 metabolic activity. Genetics, medication and inflammation-related biomolecules are suspected to have caused this down-regulation. A typical Behcet's disease medication, colchicine was found to have no influence on the observed low CYP2C9 metabolic activity. It is very possible that inflammation-respond agent caused the inhibitory effect on CYP2C9 activity.

Case 3 observed the effect of age, *CYP2C9* genotype, ethnicity, smoking habit, weight and sex on CYP2C9 metabolic ratio. In this study, ethnicity, which is strongly related to *CYP2C9* genotype was a significant factor influencing between subject-variability in CYP2C9 enzyme activity. The habit of smoking is also a significant contributor to the variation, but only in the Korean population and not the Swedes. The reason behind the smoking effect in Koreans remains unidentified.

Case 4 ventured into the correlation of the P450 oxidoreductase *(POR)*28* variant on the metabolic activity of CYP2C9. Swedish and Koreans subjects with *CYP2C9*1/*1* genotype were screened for *POR* 5, *13* and **28*. While no subject was found to carry **5* or **13*, Swedish subjects who carry *POR*28* allele were observed to display a 1.40-fold increase in CYP2C9 enzyme activity compared to non-carriers. The ultra-rapid metaboliser from case 1 was found to carry this variant.

With the evidence of a significant difference in genetic variant distribution and effects on drug metabolism, this review highlights the importance for researchers to come up with a dataset of pharmacogenetic biomarkers made especially for the Asian population. More studies need to be done in order to fully comprehend the impact of pharmacogenetics and the genetic biomarkers in the healthcare system in the Asian region. With the advancement of new genotyping and genetic screening technologies made available at an affordable cost, it is high time for precision medicine to take the lead in clinically important drug prescriptions.

IL12

Preclinical studies of *Carica papaya* against DEN-2 dengue infection

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Background

Dengue is still a major problem in Malaysia and causing high mortality. There is no specific treatment for dengue and one of the strategy is to study the effect of herbal medication on dengue. The aim is to review the results of the series of preclinical studies that has been conducted for *Carica papaya* in treating dengue fever.

Methods

Several preclinical studies were conducted namely the phytochemical, efficacy and toxicity studies. Phytochemistry studies were conducted on water extract of *C.papaya* with chromatography and spectrometry analysis. The in vitro plaque assay and the in vivo studies on AG129 mice were conducted with non-mouse adapted Malaysian dengue virus type 2 (DEN-2) infection. The mouse model of DENV-infection that closely mimicked the human disease was established and used to study the immunomodulatory activity involving specific cytokines, the endothelial cell biology in dengue infection and the effect of dosing on the day of infection. The genotoxicity and general toxicology studies were also conducted.

Results

The phytochemistry studies allowed confirmation of the herb identity and consistency of the chemical composition for efficacy and toxicity studies. Plaque assay and the in vivo studies have confirmed that the extract of *C. papaya* do not kill the dengue virus. The extract affected the immunomodulatory system and the endothelial cells of the blood vessels. These provide clues to the control of the cytokine 'storm' and the vascular leakage that is the characteristic of dengue haemorrhagic fever. A previous study has confirmed that *C. papaya* juice increases the platelet by inducing the platelet production in the bone marrow. The results of the toxicity studies were also favourable.

Conclusions

The preclinical studies has provided evidence that C. papaya extract worked on different pathogenesis of dengue fever and can be further studied in a clinical trial.

Keywords: preclinical, herbal, *Carica papaya*, dengue

ORAL PRESENTATION

OPT1

The study on ORF239342, a protein isolated from the mushroom *Agaricus bisporus* as a potent pharmaceutical biomolecule

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Background
Drug absorption becomes constrained when the permeability is low leading to limited bioavailability. The use of lectin for glycotargeting is an approach to overcome problems in the delivery of compounds with low permeability. Interaction between lectin with several types of oligosaccharides present in cells on the surface of gastrointestinal wall could facilitate the lectin to be absorbed. Vast glycosylated areas within gastrointestinal tracts can be targeted for this purpose. LSMT (light chain subunit in the tetramer complex of tyrosinase enzyme *Agaricus bisporus*) has the ability to recognize a specific group of sugar moieties, non-toxic, and nonimmunogenic. Formation of LSMT-drug bioconjugate was explored in this study to assess the ability of LSMT as a drug carrier using captopril as a drug model.
Methods
Prior to permeability study, solvent accessibility of cysteine residue (functional target candidate for bioconjugation) using ASAView and NetSurfP programs was conducted. *In vitro* accessibility of cystein was performed to determine free sulfhydryl using DTNB reagent. Conjugation was performed using different conditions of reaction, then characterized.
Results
Lysine is chosen as an active side of the reaction. Conjugate is formed with SMPT as a linker utilizing a reduced disulphide bond to release the drug. Optimum conditions currently found for conjugate formation was at 4°C for 24 hours for protein activation stage with SMPT and 48 hours for captopril binding stage with ratio of protein:SMPT = 1:10 and activated protein:captopril = 1:100. Conjugate substitution obtained under these conditions was between 1-2 mol of captopril per mole of LSMT. Conjugate formed was stable in simulated gastric and intestinal solutions. Furthermore, preliminary *in vitro* permeability study using Caco2 cells and ex vivo with non-everted gut sac method showed intact ability of LSMT to penetrate gastrointestinal wall.
Conclusions
LSMT is a promising biomolecule for a drug carrier to improve per oral boavailability.

Keywords: Light subunit mushroom tyrosinase, recombinant protein, Agaricus bisporus, drug delivery

OPT2

Effects of levodopa loaded chitosan nanoparticles on *in vivo* biodistribution study after intranasal administration

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Background
Levodopa or L-DOPA (L-3,4-dihydroxyphenylalanine) is a drug used to control symptoms of Parkinson's disease (PD) caused by low levels of dopamine in certain parts of the brain. In this study, levodopa loaded chitosan nanoparticle formulation was successfully developed to deliver the drug to the brain via intranasal administration in a rat model with an attempt to bypass the systemic circulation. Furthermore, we also investigated levodopa biodistribution on selected major organs after its intranasal delivery to determine its delivery pathway either directly to the brain or could permeate into other organs.

Methods
Optimization of the formulation parameters for Levodopa-loaded chitosan nanoparticles was conducted by ionic gelation technique. All the *in vivo* studies were performed following the guidelines and approval from the animal ethics committee of Universiti Teknologi MARA (UiTM Care: 42/2014). Dosage of 2.5 mg/kg BW of sample solutions levodopa (control) and levodopa-loaded chitosan nanoparticles) were administered through intranasal route. After four hours of administration, the rats were sacrificed and five organs including heart, lung, liver, kidney and brain were excised, weighed, homogenized and deprotonated by acetonitrile before UPLC analysis.

Results
The concentration of levodopa loaded chitosan nanoparticles was found to be significantly higher ($P<0.05$) in the brain (149.35±10.1 µg/L) followed by lung (53.51±2.8 µg/L) while the concentration of unprocessed levodopa was shown to be lower, which was 79.64 ±10.6 µg/L in the brain followed by lung region (61.86 ±4.8 µg/L). However, the concentration of levodopa was not detected in the heart, liver and kidney regions.

Conclusions
The concentration of levodopa after intranasal delivery of levodopa nanoparticles was significantly higher ($P<0.05$) as compared to the unprocessed levodopa and was prominent in the brain region (149.35 ±10.1 µg/L), considerably lower in the lung and undetected in other organs. Based on our findings, we have shown that significant amount of levodopa could be delivered to brain tissue via intranasal route that offers a promising approach to target the drug to the brain. However, more study should be done to produce information assuring safe administration of this drug to humans.

Keywords: Levodopa nanoparticles, biodistribution, intranasal, Parkinson's disease

OPT3

Formulation design and characterization of self nano emulsifying drug delivery system (SNEDDS) roxitromycin using capryol–90, polysorbat–80 and PEG–400

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Background

Roxithromycin is a macrolide antibiotic included in the biopharmaceutics classification system *(*BCS*)* class II with poor water solubility (0.0189 mg/mL) resulting poor solubility of roxithromycin in gastrointestinal track and decreases bioavailability. Technology development of self nano emulsifying drug delivery system (SNEDDS) to reduce particle size has known effectively for increasing drug solubility.

Methods

An optimum formulation of this reseach were determined by simplex lattice design method in Design Expert®10. Investigated factors were solubility of roxithromycin in capryol-90 and in mixture of polysorbat-80 and PEG-400 also the ternary phase of capryol-90:polysorbat-80:PEG-400. The emulsification system was performed by ultrasonication. The characters of SNEDS were determined by dynamic light scattering and transmission electron microscopy. The thermodynamic stability test was performed by heating-cooling cycle.

Results

Capryol–90 could dissolve roxithromycin properly (2.355±0.040 mg/mL). Polysorbate–80, and PEG–400 also could increase the solubility of roxithromycin in water. Determination of ternary phase diagram to obtain combination proportions formed a spontaneous range of 10 - 60% capryol–90, 20 - 50% polysorbate–80, and 10 - 70% PEG–400. Proportion of optimum roxithromycin SNEDDS formula obtained from simplex lattice® design was resulted 20.00% capryol–90, 60.00% polysorbate–80, and 20.00% PEG–400. Characterization of optimum formula resulted percent of transmittance (80.60 ± 0.35)%, emulsification time (71.70 ± 0.99) second, viscosity (3.76 ± 0.02) cP, pH (7.84 ± 0.07), and robustness to dilution in aquadest, SGF, and SIF (99.16 ± 0.67)%, (93.23 ± 0.14)%, and (98.34 ± 0.34)%. Dissolution test showed that SNEDDS could improve dissolution of roxithromycin in SIF pH 7.4 compared to pure and generic tablet.

Conclusions

Proportion of optimum roxithromycin SNEDDS was resulted 20.00% capryol–90, 60.00% polysorbate–80, and 20.00% PEG–400. Dissolution profile showed that SNEDDS could improve dissolution of roxithromycin in SIF pH 7.4 compared to pure and generic tablet.

Keywords: Formulation-design, roxithromycin, characterization, SNEDDS

OPT4

Hyaluronic Acid Coated Chitosan-Latanoprost-Link Nanoparticle for Prolonged Ocular Drug Delivery

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Background
The major problem with conventional eye drops is the assurance of optimum drug concentration to the target site, due to pre-corneal and nasal drug elimination, as well as barriers of the eye impeding drug access. The use of nanomeric drug delivery systems with mucoadhesive properties may enhance drug residence time to the active site, thus providing better ocular availability, as well as improved tolerability of the formulation. In this study, latanoprost molecules was linked to mucoadhesive nanocarrier, chitosan (CS) and hyaluronic acid (HA) that can control the drug release and prolong residence time in ocular tissues.

Methods
The methods that was used for physical and chemical characterizations were: (1) electron microscopy (2) the dynamic light-scattering method (DLS); (3) Cup and Bob viscometry (4) infrared spectroscopy and (5) high performance liquid chromatography. Draize test was performed to determine the safety of the polymeric nanoparticle.

Results
The optimum CS: TPP ratio had the lowest particle size of 198 nm, with PDI of 0.274, ZP of +27.7mV and an entrapment efficiency (EE) of 62%. It was further coated with HA, where the optimum HA: CS ratio had the lowest particle size of 314 nm, with a PDI of 0.424, ZP of +29.87 mV and an EE of 72%. In the *in vitro* drug release study, the optimum HA coated CS-latanoprost link nanoparticle formulation has 0% drug release in 30 minutes, 29% in 2 hours and 87% in 8 hours as compared with the conventional latanoprost solution that released 28% of the drug in 30 minutes, and 100% in 2 hours. Release mechanism of the drug from the polymeric nanoparticles matrix led to a *zero order* kinetic with a correlation coefficient of 0.9848. Drug release could also be expressed by Higuchi's equation as the plot showed linearity at 0.9492, where the value of diffusion exponent obtained from the Korsemeyer-Peppas model is 1.13. Addition of mucin to the positively charged nanoparticles reduced the ZP to an average of -4.30 mV. The draize test on albino rabbits showed the polymeric nanoparticle were safe for ophthalmic use.

Conclusion
The results of this study could serve as a basis that mucoadhesive HA coated CS-latanoprost-link nanoparticles could provide a prolonged ocular delivery system of latanoprost for better glaucoma treatment.

Keywords: latanoprost, chitosan, hyaluronic acid, nanoparticles, prolonged drug delivery

OPP1

Synthesis, characterization, and biological activities of Schiff bases and their iron and zinc metal complexes

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Background
Schiff bases, being active biological moieties, possess diverse pharmacological activities. Metal ions play vital role in various functions of human body, and diseases may occur due to metal ion deficiencies. The importance of metal complexes of Schiff bases has been acknowledged in the field of biomedical sciences.
Methods
Herein, two Schiff base ligands (L1, L2) underwent metal complex formation, to produce their iron and zinc metal complexes, respectively. Original ligands and their metal complexes were characterized physically as well as by means of spectral characterization techniques such as Infra-red spectroscopy (IR), nuclear magnetic resonance spectroscopy (NMR) and mass spectrometry. Pharmacological perspectives of these Schiff base ligands and their iron and zinc metal complexes such as antibacterial, antifungal and antioxidant assays were assessed.
Results
All compounds exhibited antibacterial and antifungal activities, but the metal complexes showed better activities in comparison to the original ligands, especially all zinc complexes. Zinc complex (L2)2Zn(Ac)2 elicited good antibacterial activity against all gram positive and gram negative bacterial strains and exceptional activity against *Candida albican* strain. Overall, all the compounds showed better antifungal activity against *Candida albican* as compared to *Candida glabrata*. Free ligands illustrated better antioxidant behaviour as compared to the metal complexes.
Conclusions
These results suggest that all the ligands and metal complexes, being active in one way or the other, have the potential to be employed as antibacterial, antifungal and antioxidant agents.

Keywords: Schiff base, Metal complex, Antibacterial, Antifungal, Antioxidant

OPP2

Possible drug-herb interactions between Merunggai (*Moringa oleifera*) and selected antihypertensive drugs

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Background
Herbal medicines have been widely used in Malaysia for cardiovascular pharmacotherapy. This is alarming as little is known about drug-herb interactions of conventional cardiovascular drugs with most Malaysian herbs. *Moringa oleifera* is a medicinal plant with high nutritional values and was reported to possess blood pressure (BP) lowering effect. Hypertension has become a significant health issue globally and is treated with four main classes of drugs namely; angiotensin converting enzyme inhibitors (ACEIs), angiotensin receptor blockers (ARBs), β-blockers and calcium channel blockers. This study aimed to investigate any possible drug-herb interactions between the aqueous leaves extract of *M. oleifera* and selected antihypertensive drugs in normotensive rats (NTs) and spontaneously hypertensive rats (SHRs).

Methods
The study consists of ten groups of SHRs and one group of NTs. The rats were given either drugs alone or drugs in combination with *M. oleifera* extract for 14 days. There were also control groups. Systolic and diastolic blood pressure of the rats were measured on day 1 prior to the treatment and on day 15.

Results
All treatment groups were found to produce significant blood pressure reduction on day 15 when compared with negative control but there was no significance difference when compared with positive controls. Combination of drugs and extract significantly reduced BP but are comparable with the use of drugs alone.

Conclusions
There is a possibility of drug-herb interaction between *M. oleifera* and the selected antihypertensive drugs. Detailed mechanism of actions on how these interactions occur are worth to be investigated further to ensure the safety of *M. oleifera* usage in combination with antihypertensive drugs.

Keywords: *Moringa oleifera*; angiotensin converting enzyme inhibitors; angiotensin receptor blockers; β-blockers; calcium channel blockers

OPP3

Phyllanthus amarus protects against lipopolysaccharide-induced neuroinflammation and memory impairment in rodents

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Background
Phyllanthus amarus (PA) has been widely studied for its anti-inflammatory properties in various peripheral systems. However, the effects of PA in modulating immune responses in the central nervous system leading to protection against its functional changes remain unexplored. Therefore, we examined the protective effects of 80% v/v ethanol extract of PA pre-treatment on lipopolysaccharide (LPS)-induced neuroinflammation with subsequent non-spatial and spatial memory impairment.
Methods
The PA extract was chemically characterised using high-performance liquid chromatography against its selected major phytoconstituents. Rats or mice were treated orally with vehicle (5% Tween 20), PA extract (100, 200, and 400 mg/kg), or ibuprofen (IBF; 40 mg/kg, postitive control) for 14 and 28 days before being subjected to novel object discrimination or 8-radial arm maze tests, respectively. LPS (1 mg/kg) was given intraperitoneally a day prior to the behavioral tests except for the negative control group. At the end of the behavioral tests, the levels of tumor necrosis factor-α (TNF-α), interleukin (IL)-1β, nitric oxide (NO), inducible nitric oxide synthase (iNOS), CD11b/c integrin expression, and synaptophysin immunoreactivity were determined in the brain tissues.
Results
The PA extract was quantitatively profiled for gallic acid, ellagic acid, corilagin, geraniin, niranthin, phyllanthin, hypophyllanthin, phyltetralin, and isonirtetralin content. PA extract administered at 200 and 400 mg/kg for 14 and 28 days effectively protected the rodents from LPS-induced memory impairment. Similar doses significantly ($p < 0.05$) decreased the release of proteins such as TNF-α, IL-1β, and iNOS in the brain tissues. NO levels, CD11b/c integrin expression, and synaptophysin immunoreactivity were also reduced in animals pre-treated with PA or ibuprofen as compared with those that receive LPS only.
Conclusions
Pre-treatment with PA extract or IBF for 14 and 28 days protected the rats from LPS-induced neuroinflammation and memory impairment. Further studies are warranted to identify the bioactive phytochemicals and the precise underlying protective mechanisms.

Keywords: *Phyllanthus amarus*, neuroinflammation, neuroprotection

OPP4

Antioxidant and anti-inflammatory effects of *Erythroxylum cuneatum* leaf extract *in vitro*

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Background
Oxidative stress and inflammation are known to be associated with the pathogenesis of most chronic illnesses. Synthetic drugs are commonly utilized but they are accompanied by adverse effects. *Erythroxylum cuneatum* (EC), which is also known as “Chinta mula” locally, belongs to the family Erythroxylaceae. Our preliminary study found EC acetone and ethanol leaf extracts to be rich in bioactive compounds and with antioxidant properties. Therefore, this research is intended to determine the antioxidant and anti-inflammatory properties of the EC acetone and ethanol leaf extracts *in vitro*.
Methods
The cell viability test of the extracts was conducted using the MTT assay. The antioxidant activity of the extracts against human aortic endothelial cells (HAoEC) stimulated with oxidised low density lipoprotein (oxLDL) was determined using lipid peroxidation, reactive oxygen species (ROS) and nitric oxide (NO) production assays. The anti-inflammatory effects of the extracts was determined using monocyte adhesion and migration assays. Expression of human adhesion molecules was quantified by using ELISA kit.
Results
Both extracts produced significant inhibition of lipid peroxidation at maximum dosage. Moreover, the extract reduced the production of ROS, NO and monocyte adhesion. Acetone and ethanol leaf extracts of EC inhibited the migration of monocyte at 40 µg/mL and 80 µg/mL, respectively. The EC leaf extracts also showed a significant reduction in inhibiting the expression of ICAM-1 and VCAM-1 at maximum dosage.
Conclusions
This study showed that both extracts had antioxidant and anti-inflammatory activities in most of the studied parameters. EC acetone extract showed better bioactivities *in vitro* as compared to ethanol extract.

Keywords: *Erythroxylum cuneatum*; antioxidant activity; anti-inflammatory activity

OPP5

Effect of *Gynura procumbens* and *Christia vespertilionis* extracts on cell adhesion molecules in human umbilical vein endothelial cells

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Background
The initiation of atherosclerotic lesion involves endothelial cell pro-inflammatory state that recruits leukocytes and promotes their movement across endothelium which requiring endothelial expression of cell adhesion molecules. *Gynura procumbens* (GP) and *Christia vespertilionis* (CV) are herbaceous plants that are traditionally used for treatment of various inflammation-related ailments. However, there is limited evidence that points to the protective activity of these plants against inflammation that occurs in atherosclerosis. In this study, we sought to explore the inhibitory effect of GP and CV extracts on TNF-α-induced vascular cell adhesion molecule-1 (VCAM-1) expression and its underlying mechanism.

Methods
Cell viability of HUVEC treated with GP or CV extracts was determined by MTT assay while protein expression of adhesion molecules and cellular signaling molecules were determined by Western blot.

Results
GP or CV extracts at concentration ranging from 5 µg/mL to 60 µg/mL were found to maintain more than 80% cell viability following 24 hours treatment. Selected treatment concentrations (20, 40 and 60 µg/mL) of CV extract showed no effect on TNF-α-induced VCAM-1 expression in HUVEC. On the other hand, pretreatment of 60 µg/mL GP extract demonstrated a significant inhibition on TNF-α-induced VCAM-1 protein expression in HUVEC ($p<0.005$). Pretreatment of 60 µg/mL GP extract also showed a dose-dependent suppression on IKKα/β phosphorylation and significant inhibitory effect ($p<0.05$) on protein expression of phosphorylated NFκB.

Conclusions
Results from this study demonstrated that CV extract may not have inhibitory effect on expression of adhesion molecules but GP extract showed inhibitory effect on VCAM-1 expression by suppressing NFκB signaling pathway. This results implicate that GP extact may have beneficial use particularly in vascular inflammation.

Keywords: *Gynura procumbens*; *Christia vespertilionis*; vascular cell adhesion molecule; NFκB; endothelial cells

OPP6

Optimization of solvent extraction method in recovery of testosterone and 6β-hydroxytestosterone from cell culture media and protein depletion of sample for *in vitro* CYP3A4 mediated 6β-hydroxylation assay

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Keywords: Testosterone, 6β-hydroxytestosterone, UHPLC, metabolism, solvent extraction.

Background
Accurate measurement of testosterone and 6β-hydroxytestosterone is important for *in vitro* CYP3A4 mediated 6β-hydroxylation assay. To increase accuracy of measurement, maximum recovery of analytes from cell culture media must be achieved. Apart from recovery of analyte, protein depletion of cell culture media is also an important step before UHPLC quantification to avoid column clogging. The aim of this study is to investigate optimum solvent extraction method of testosterone and 6β-hydroxytestosterone from culture media and to determine protein depletion efficiency of the solvent extraction method.

Methods
Media collected from WRL68 (normal liver cell line) culture was spiked with 20 µM testosterone and 10 µM 6β-hydroxytestosterone. The analytes were extracted using centrifugation at 15000 rpm for 20 minutes with different solvent including acetone, acetonitrile, methanol, ethyl acetate and dichloromethane and were analysed quantitatively using UHPLC. The protein content in the extracts were determined using Bicinchoninic Acid protein assay. UHPLC analysis method was optimized for analytes quantification.

Results
The methanol extraction method resulted in the highest percentage of recovery (98.3 % for testosterone and 98.4 % for 6β-hydroxytestosterone) with moderate protein depletion (79.95 ± 1.87 %). The ethyl acetate extraction method resulted in the highest protein depletion at 98.8 %, however, the percentage of recovery of analytes is lower than that of methanol extraction method.

Conclusions
As a conclusion, methanol was found to be the most optimum solvent for extraction of testosterone and 6β-hydroxytestosterone from cell culture media compared to other solvents used and the amount of protein left in sample did not interrupt UHPLC analysis.

OPP7

Phytoestrogens induced apoptosis and phagocytosis through modulation of annexin A1 in leukemic cell lines

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Background

Phytoestrogens is a non-steroid plant compound that has structurally similar to estrogen which posses' anti-cancer properties. Phytoestrogens have the ability to induce apoptosis, cell cycle arrest and phagocytosis and reducing Annexin A1 in leukemic cell lines. However, the underlying mechanism of phytoestrogens in inducing cell death is still not fully understood. The present study aimed to investigate the effects of phytoestrogens in inducing of cell death is through decreasing ANXA1 level or independently.

Methods

Leukemic cells and ANXA1-knockdown leukemic cells were incubated with estrogen and phytoestrogens 40 µg/ml for 24 hrs at 37oC. Cells viability were examined by MTT assay and ANXA1 quantification via ELISA Assay. Apoptosis were examined by flow cytometer and phagocytosis were evaluated by haematoxylin-eosin staining. Transfection of ANXA1 siRNA was conducted to down-regulate ANXA1 expression.

Results

In Leukemic cells, coumestrol significantly ($P<0.05$) reduced the total level of ANXA1 in both K562 and U937 cells. Genistein induced a significant ($P<0.05$) reduction in the total level of ANXA1 in K562, Jurkat and U937. Estradiol and daidzein induced similar reduction in U937 and Jurkat cells. Coumestrol and daidzein induced apoptosis in K562 and Jurkat cells, while genistein and estradiol induced apoptosis in all tested cells. Coumestrol, genistein and estradiol induced phagocytosis in all cells but daidzein induced significant ($P<0.05$) phagocytosis in K562 and Jurkat cells only. In ANXA1 knockdown leukemic cells, the expression of ANXA1 was significantly downregulated in all cell lines. Genistein significantly induced apoptosis ($p<0.001$) only in Jurkat cell, contrary coumestrol and deidzein did not induce apoptosis in all the cell lines tested. The percentage of phagocytosis and phagocytosis index increased significantly after treatment with phytoestrogens in all cell lines.

Conclusions

Induction of apoptosis and phagocytosis by phytoestrogens are mediated through decreasing of annexin A1 expression.

Keywords: Phytoestrogens, Annexin A1, Apoptosis, Phagocytosis, Leukemia

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OPL1

Atheroprotection by antilipidaemic *Pediococcus pentosaceus* LAB6- and *Lactobacillus plantarum* LAB12-fermented cell free supernatant *in vitro*

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Background
Current treatments against atherosclerosis rely predominantly on lipid lowering in combination with anti-inflammatory therapies. However, the maximum efficacy of these treatment strategies appears to be rather modest, often compromised by the lack of response by high risk patients and adverse effects. There is a need for alternative approaches that can manage atherosclerosis more effectively. Recent evidence raises the possibility of using antilipidaemic probiotics for atheroprotection. Nevertheless, the beneficial effects of probiotics are believed to be strain-dependent. We had identified unique probiotic lactic acid bacteria (LAB) (i.e. *Pediococcus pentosaceus* LAB6 and *Lactobacillus plantarum* LAB12) with promising cholesterol lowering effects. Capitalising on this beneficial property, the present study aimed to investigate the atheroprotective potential of LAB6 and LAB12 *in vitro.*

Methods
The sub-toxic concentration of 24 h LAB-fermented cell free supernatant (CFS) against RAW264.7 was determined using the sulforhodamine assay. Sub-toxic CFS was added to oxLDL-induced foam cell for 24 h before staining with Oil Red O stain. For semi-quantitative analysis, images captured under light microscopy were analysed for differential intensity using ImageJ. For quantitative analysis, isopropanol was added, and absorbance was measured at 540 nm using a spectrophotometer. The effect of CSF against oxLDL-induced mitochondrial dysfunction was assessed by using the mitochondrial membrane potential assay.

Results
The highest sub-toxic concentration (IC_{15}) of LAB6- and LAB12-derived CFS against RAW264.7 were 7 % and 5.6 %, respectively. Subtoxic LAB6- and LAB12-derived CFS significantly ($p<0.05$) reduced lipid uptake in oxLDL-induced foam cells by at least 47.06% and 47.12%, respectively. LAB-derived CFS also prevented oxLDL-induced mitochondrial dysfunction (early apoptosis) by increasing red (aggregate)/green (monomer) ratio of JC-1 fluorescence by $\leq$ 4.

Conclusion
The present findings strongly implied the atheroprotective potential of LAB6- and LAB12-derived CFS against foam cell formation in the event of atherosclerosis. This in turn warrants further investigations using *in vivo* model.

Keywords: Probiotics, cholesterol lowering, HPTLC, atheroprotective

OPL2

Use of xylazine hydrochloride–ketamine hydrochloride for immobilization of captive large felines in Malaysia: a 15-year retrospective study (1988-2003)

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Background
Chemical immobilisation by anaesthesia with xylazine hydrochloride (XZH) - ketamine hydrochloride (KTH) has been widely used in large felines. This study was aimed to determine the relationship between time of effect and effect of anaesthesia with XZH-KTH.

Methods
Data were retrieved from existing anaesthesia records from different zoos in Malaysia from 1988 to 2003. A total of 66 large felines belonging to 5 different species namely, Malayan Tiger (*Panthera tigris jacksoni)* (*n* = 4), Bengal Tiger (*P. tigris tigris)* (*n* = 10), African Lion (*P. leo)* (*n* = 12), Sumatran Tiger (*P. tigris sumatrae)* (*n* = 17) and Gir Lion (*P. leo persica)* (*n* = 23) were involved in this study. All the large felines were successfully anaesthetised using XZH-KTH. The effects of variables such as body weight, sex, health status, demeanour and fasting time on dose selection were evaluated. The relationship of dose with effect of anaesthesia and time of effect were also studied.

Results
The results showed that the effect of anaesthesia and time of effect had no significant correlation with dose. Among the variables studied, only weight had significant ($p = 0.016$ and $p = 0.002$) effect on dose. When an average dose (KTH = 363.33 mg; XZH = 185.98 mg) was given to the felines, it gave a weak positive correlation with time of effect ($r_{ketamine} = 0.220$; $r_{xylazine} = 0.324$). Similar findings were observed for the effect of anaesthesia ($r_{ketamine} = 0.156$; $r_{xylazine} = 0.227$).

Conclusions
Although the time of effect and effect of anaesthesia were independent of the dose, it is important to determine the weight of the large felines so that the drug administered were sufficient enough to produce the desired anaesthetic effect.

Keywords: ketamine, xylazine, large felines, dose selection.

OPC1

Development of simultaneous analysis method for determining level of losartan potassium and hydroclorotiazid in tablets using high performance liquid chromatography (HPLC)

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Background
Losartan potassium and hydrochlorothiazide are combination of antihypertension drugs from group of angiotensin II receptor blocker (ARB) and diuretic. The assay of both substances needs a method which is able to determine the substances without performing prior separation method. Due to its great sensitivity, Reverse-Phase HPLC with UV detector could be used in simultaneous analysis. This research was conducted to develop the method of losartan potassium and hydrochlorothiazide assay in tablet simultaneously.

Methods
The assay was performed on a system with Inertsil ODS-3 RP-C_{18} 5µm (4,6x50 mm) as column, methanol pro HPLC : H_3PO_4–KH_2PO_4 (55:45) pH 3 as mobile phase, flow rate 1 mL/minute, and detected at 225 nm.

Results
The retention time for losartan potassium and hydrochlorothiazide were 1,842 and 14,473 minutes. The system was linear for losartan potassium 10-60 µg/mL and hydrochlorothiazide 2,5-15 µg/mL with correlation coefficient 0,999. Limits of detection and quantification for losartan potassium and hydrochlorothiazide were 2,001; 6,671 µg/mL and 0,626; 2,087 µg/m, respectively. Relative standard deviation (RSD) of intraday precision for losartan potassium and hydrochlorothiazide were 1,360; 0,959 and 1,455; 1,400 % while the interday precision RSD were 0,333; 0,848 and 0,919; 0,904 %. Percent recovery for losartan potassium and hydrochlorothiazide in simulation were 100,560 ± 1,032 % and 100,356 ± 0,941 %.

Conclusions
Losartan potassium and hydrochlorothiazide content in sample tablet were in range of 97,029 – 99,875 % and 98,054 – 101,506 %. It can be concluded that the developed method is suitable for simultaneous analysis of both active pharmaceutical ingredients.

Keywords: losartan potassium, hydrochlorothiazide, RP-HPLC, simultaneous analysis, validation

OPC2

Synthesis, *in vitro* urease inhibitory potential and molecular docking study of benzimidazole and bi-heterocyclic benzamide analogues

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Background
Urease is a nickel-containing metalloenzyme that widespread in nature among plants, bacteria, fungi, algae and invertebrates. Urease producing *Helicobacter pylori* (*H. pylori*), one of the most successful human bacterial parasites, which colonize more than half of the human population. Urease associated diseases include severe gastroduodenal pathologies, hepatic encephalopathy, urinary catheter encrustation, pyelonephritis and hepatic coma. In this regard, a series of analogues benzimidazole and bi-heterocyclic benzamide were synthesized, characterized and screened for urease inhibitory activity.

Methods
Mixed 1H-benzimidazole-2-thiol with methyl 4- (bromomethyl) benzoate and refluxed for 5 hrs to give methyl 4-(((1H-benzimidazol-2-yl) thio)methyl)benzoate as intermediate product. The intermediate product was finally treated and refluxed with different substituted aldehyde/acetophenone to give the desired benzimidazole and bi-heterocyclic benzamide analogues.

Results
The targeted benzamides and benzimidazole analogues were synthesized in good yields and their structures were confirmed by NMR and elemental analysis. The *in vitro* screening results showed that most of the ligands exhibited good inhibitory potentials against the urease. Molecular docking revealed that fluoro analogue of bi-heterocyclic benzamide exhibited good binding energy value (−8.40 kcal/mol) and was bound within the active region of urease enzyme. Limited SAR suggested that the variations in the inhibitory potentials of the analogues are the result of different substitutions on phenyl ring.

Conclusions
We have synthesized benzamides and benzimidazole analogues and screened against urease inhibitory potential. All analogues revealed more inhibitory potentials than the previously reported analogues for the urease activity on the basis of IC_{50} values, binding interactions of most active compounds & ADMET pharmacokinetics.

Keywords: Bi-heterocycles, Benzamides, Benzimidazole, Urease, Molecular docking

OPC3

Head-to-tail position of two bridged-dimers determines the configuration of oligostilbene structure

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Background
The first report on isolation and characterization of isohopeaphenol A was from *Vatica oblongifolia*. The following year, a compound with the same spectral data was isolated from *Vatica pauciflora*. It was assigned a different stereochemistry and named pauciflorol C. Recently we isolated from *Neobalanocarpus heimii* (Dipterocarpaceae), a compound with the same spectral data. We, therefore, studied the structure in detail. The stereochemistry of the structure was conferred based on NMR spectroscopy and a three-dimensional computer generated structural model.

Methods
The extraction of the plant material was by the classical method of repetitive maceration and lixiviation with methanol. The crude residue was subjected to HPLC for fractionation and isolation processes. The pure compound was isolated as a dark brown amorphous powder. Its structural characterization was performed by means of spectrometric methods, including extensive 2D-NMR. The stereochemistry of the compound was supported by a 3D model, obtained *in silico* with software, Chem 3D Ultra^{TM}.

Results
Preliminary examination of the mass, ^{1}H- and ^{13}C-NMR data suggested a resveratrol tetramer. Thorough analyses of 2D-NMR confirmed the oligomeric degree and elucidated the structure. The compound consists of two similar stilbene dimer plane structures, linked by a bridge. The fact that they are not magnetically equivalent from an NMR perspective suggested stereoisomeric differences for these two dimeric moieties. A NOESY experiment contributed to solve the issue. A 3-dimensional model was performed and it was showed that such correlation was only possible when the second half of the molecule is rotated 180° relative to the first half of the molecule. This information was in agreement with the coupling constant of 11.5 Hz. The absence of cross peak between further supported a *trans* configuration.

Conclusions
The present in-depth analyses of NOE data together with 3D modeling strongly suggest that the initial structure of isohopeaphenol A is correct. It is possible that for pauciflorol C, the author overlooked the possibility for the two halves of the molecule to be positioned in a head-to-tail manner, which is the only way to understand some of the measurements discussed above. As a result, it is concluded that the spectroscopic data is for isohopeaphenol A.

Keywords: isohopeaphenol A, pauciflorol C, oligostilbenes, phytochemistry, spectroscopy.

OPC4

Design and syntheses of *ortho*-, *meta*- and *para*-xylylguanidinium–zn^{2+}–cyclen complexes and their interaction with DNA (cyclen = 1,4,7,10–tetraazacyclododecane)

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Background
Three new zinc ions (Zn^{2+}) complexes, **C^1**, **C^2** and **C^3**, were designed and synthesized by coordination of Zn^{2+} into the integrated 1,4,7,10-tetraazacyclododecane (cyclen) and *ortho*-, *meta*- and *para*-bromoxylylguanidinium pendants group. The aim of synthesizing these Zn^{2+} complexes was to confirm the anticipated interactions of Zn^{2+} complexes towards natural DNA as well as to explore the phosphatase activity of such complexes. A retrosynthetic analysis was carried out to identify and solve problems with regard the selection of organic reactions.
Methods
The syntheses were performed in five steps including of (i) Gabriel and Ing-Manske primary amine synthesis, (ii) S_N2 substitution reaction, (iii) guanylation of primary amine, (iv) deprotection of Boc group, and (v) coordination of Zn^{2+} complex. All the Zn^{2+} complexes structures were characterized by ^{1}H- and ^{13}C-NMR spectroscopy, infrared spectroscopy and mass spectrometry. Ethidium bromide (EB) fluorescence assay and circular dichroism (CD) spectroscopy were used to ascertain the interaction between Zn^{2+} complexes towards natural DNA i.e. calf thymus (ctDNA).
Results
The former assay demonstrated a displacement of EB from its complexes with ctDNA, thus confirming the affinity of these Zn^{2+} complexes towards DNA. CD spectroscopic results also revealed that **C^1** has disturbed both base stacking and right handed helicity properties of ctDNA, but retained the B-form of its structure. By contrast, **C^2** and **C^3** transformed the conformation of ctDNA from B-form into Z-form. This was further supported by thermal denaturation studies showing ΔT_m values of **C^1**, **C^2**, and **C^3** to be +2, +4 and +5, respectively.
Conclusions
The catalytic properties of these complexes for phosphate hydrolysis was evaluated using phosphodiesters bis(*p*-nitrophenyl)phosphate (BNPP) as a model and monitoring by UV spectrometry. The BNPP hydrolysis results (*ca*. 17% after 8 days incubation) suggested that **C^1**, **C^2**, and **C^3** were endowed with still modest yet significant catalytic properties.

Keywords: Zn^{2+} Complex, Guanidinium, DNA Binding, Phosphodiesterase, BNPP Hydrolysis.

OPC5

Persistence of drugs residue in urban river. Case study of Sungai Buloh, Malaysia

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Background
Drugs and their metabolites are continually introduced into the environment and are prevalent at detectable concentrations, which may affect water quality and potentially impact drinking water supplies, ecosystem and human health. The discharge of micropollutants without control can have the adverse health impact and at the same time can disturb the aquatic ecology and systems in a long period of exposure. In addition, the occurrence at trace levels of several drugs in drinking water raises concerns about possible implications for human health. Therefore, it is critically needed to conduct the study on detection of drugs on river water because the data are still insufficient especially in Malaysia. This study is done to trace the residue of drugs in urban surface water which is in Sungai Buloh, Malaysia as a selected urban river.
Methodology
The samples were analysed using liquid chromatography coupled with quadrupole-time-of-flight tandem mass spectrometry (LC-Q-ToF/MS) for compounds tracing purpose.
Results
From the result obtained, several drugs have been traced in river water. All the drugs detected were classified based to their therapeutic usage. The residues detected consist of β-blockers, analgesics and psychoanaleptics.
Conclusion
The river contains drugs that may affect the environment. Further analysis needs to be done to get a more accurate concentration of the drug residue that contaminated the river.

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OPE1

CDIO approach method for supply chain education improve pharmacy students' skills

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Background
Engineering education and real-world demands on engineers have in recent years led engineering schools in the USA and Europe to form the Conceive Design Implement Operate (CDIO) initiative. It is a worldwide collaboration to conceive and develop a new vision of engineering education. The main objective of this research was to determine the implementation of CDIO training methods to pharmacy education (knowledge-skills-attitudes) in the pharmaceutical supply chain can improve the students; skills.
Methods
A cross sectional survey was conducted to assess the current level and level of expectation from stakeholders for the skills of pharmacy students in pharmaceutical industry training universities in Ho Chi Minh City.
Results
Findings from the current study revealed that there are differences between the current level and the level of expectation of the stakeholders on the skills to be trained for students to meet the requirements of employers for work needs.
Conclusions
The findings from this study are the basis of developing pharmaceutical supply chain education and the application of CDIO training methods to the universities in Ho Chi Minh City. This is essential for the students to know and prepare to meet the demand of the workforce and society after graduation.

Keywords: Mapping CDIO skills, CDIO, pharmaceutical supply chain

Poster presentation

PPT1

Evaluation of quality and stability of matrix tablet contained monoammonium glycyrrhizinate

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Background
Monoammonium glycyrrhizinate of Glycyrrhiza root has been used as an expectorant, detoxificator, anti-allergic, and antioxidant. We have isolated monoammonium glycyrrhizinate from Glycyrrhiza root, grown in Mongolia by previous study. The objective of the study was to develop prolonged release matrix tablet with hepatoprotective effect and to evaluate their pharmacotechnical qualities and stability.

Methods
The matrix tablets were prepared by wet granulation method. In order to develop appropriate tablets various excipients such as matrix former, diluents, binder, lubricant and glidiant were added. APIs and matrix former, diluent and binder were mixed properly and were granulated with the 5% solution of PVP K-30 as a binder solution. The wet mass was granulated by wet granulator through the sieve with 2 mm diameter holes and generated wet granules were dried at room temperature. Dry granules were lubricated with talc and magnesium stearate. The matrix tablets were prepared by the compression of the tablet mixture using rotary tablet machine. The quality of the prepared tablets was evaluated according to Mongolian National Pharmacopoeia's methods by criterias such as appearance, average weight, weight variation, hardness, friability, mocrobiological contamination and *in-vitro* dissolution study. Licozinat matrix tablets contained monoammonium glycyrrhizinate 140 mg; glycine 50 mg; LD-methionin 50 mg in each tablet.

Results
Formulations were evaluated and satisfied the quality criteria by Mongolian National Pharmacopoeia methods. The stability of matrix tablet tested by long term method for 12 months and by accelerated method for 6 months according to standard MNS 6439:2014. stability testing results by both long term and accelerated method, Licozinat matrix tablet was stable for 12 months. Stability testing of matrix tablet is continuing by long term method.

Conclusion
Controlled release "Licozinat" matrix tablets were prepared by wet granulation method. Formulation (F5) containing 20% HPMC K4000 releases in the desired manner–and was determined to be the appropriate design. Licozinat matrix tablet was stable for 12 months. Stability testing of matrix tablet is continued by long term method.

Key words: Glycyrrhiza uralensis, monoammonium glycyrrhizinate, matrix tablet, stability testing

PPT2

Formulation and evaluation of *in situ* gelling system for ophthalmic delivery of Erythromycin

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Background
Conventional ophthalmic dosage forms provide low bioavailability and less pre-corneal drug residence time due to nasolacrimal drainage and blinking action of the eyes. The major challenge is to formulate a system to improve the contact time of the drug in the eyes. The present study was aimed to prepare and evaluate in situ gelling system for the effective delivery of Erythromycin to combat ophthalmic infections.

Methods
Development of novel in situ gelling system using Erythromycin was based on the concept of ion triggered in-situ gelation. Sodium Alginate was used as a gelling agent in combination with Hydroxypropyl methylcellulose (HPMC K100) as a viscosity enhancing agent. The prepared formulations were evaluated for physical appearance, pH, gelling capacity, viscosity, stability studies, drug content, in vitro diffusion study and s spreadability test.

Results
All formulations were found to be clear and free from undissolved particles. The pH of the formulations was within the range of 6.8 – 6.92 which is safe for ophthalmic use. Formulation F4 (Sodium Alginate 1.2% and HPMC 0.5%) showed optimum viscosity of 48cps, good spreadability and gelling capacity that will improve residence time of the drug in eyes. All the formulations were found to have drug content uniformity of 98 ±2% p. In vitro, drug release studies showed that the drug was released in the of order F2<F1<F3<F4 over the period of 8 hours. All formulations F1 to F4 followed zero order drug release kinetic with a correlation coefficient of (R2=0.990) followed by the Korsmeyer-Peppas model showed drug released from the system by diffusion mechanism.

Conclusion
The developed in situ gelling systems may provide greater ocular bioavailability and it may be proposed to treat ocular infections by retaining the drug for a prolonged period in the eyes.

Keywords: In situ gel, ophthalmic, Erythromycin, HPMC, Sodium Alginate.

PPT3

Proliposomes for improved oral bioavailability of celecoxib

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Background
Celecoxib (CXB) is a COX-2 inhibitor, and has superior pharmacological effects over non-selective COX inhibitors. However, CXB has poor water-solubility and limited dissolution rate, resulting in low oral bioavailability. Thus, the objective was to formulate and evaluate CXB-loaded proliposomes (PLs) prepared by solid dispersion technique.
Methods
CXB, soy phosphatidylcholine (SPC), sorbitol, and poloxamer 188 were mixed using a water/ethanol binary solvent system, followed by evaporation of ethanol and lyophilization. CXB-loaded proliposomes (CXBPLs) were characterized by evaluating physical state of CXB, size, polydispersity index, zeta potential, morphology, stability study and drug content. Dissolution studies of pure CXB powder, a commercial product of CXB, and CXBPLs were carried out under sink condition. Then, the cytotoxicity and transport ability were evaluated in Caco-2 cell model. In addition, *in vivo* pharmacokinetic studies, histological assay was performed in rats.
Results
The crystalline CXB was transformed into its amorphous state during the preparation process, as evidenced by the solid-state evaluation with powder X-ray diffractometry and differential scanning calorimetry. After the reconstitution by gentle hand-shaking, the generated CXB-loaded liposomes showed nano-sized mean diameter, negative zeta potential, and relatively high entrapment efficiency of 84.7% with vesicular-shaped morphology. CXBPLs exhibited the increased dissolution rate and permeability than the pure CXB without damage on the Caco-2 cell. Moreover, CXBPLs increased the oral bioavailability of CXB and reduced the time to reach the peak plasma concentration in rats. The histological observation with hematoxylin and eosin staining revealed no additional detrimental effect of CXBPLs on the intestinal epithelia of rats compared to that of the pure CXB.
Conclusions
The developed proliposomal system could be a biocompatible platform for enhancing the oral bioavailability of poorly water-soluble CXB.

Keywords: Proliposomes; Dissolution; Oral bioavailability; Pharmacokinetics; Celecoxib

PPT4

Development of glucomannan nano-emulsion formulation as non-steroidal treatment for atopic dermatitis

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Background
Atopic dermatitis is a chronically relapsing pruritic inflammatory disease which affects 15% to 30% of children and 10% of adults in industrialized country. Konjac glucomannan (KGM) isolated from *Amorphophallus konjak* K. Koch exhibit high water solubility, biocompatibility, biodegradability and non-toxic properties. There are vast applications of KGM including biomedical studies such as cholesterol and obesity studies, anti-inflammatory effect, antioxidant study, and wound healing property. In this research, we developed KGM nano-emulsion as drug carrier that acts as hydrogel which stabilized the formulation and moisturize the skin for relieving atopic dermatitis.
Methods
Cream formulations were developed using variable ratios of glucomannan (1%-1.5%), Olive oil (0%-20%) and avocado oil (0%-20%). Oil phase and aqueous phase were mixed under constant stirring using Ika-Werke Eurostar with propeller mixer at 900 rpm for 10 min. The formulations will be tested and measured for particle size and zeta potential using zetasizer (Nano ZS, Malvern Instrument, UK); and Firmness and viscosity using rheometer (Physica MCR 301).
Results
The mean particle size for KGM nano-emulsion ranged from 326.93±11.14 to 586.7±26.48 nm with polydispersity index ranges from 0.41±0.04 to 0.56±0.02. The zeta potentials of KGM nano-emulsion showed low values indicate stable formulations which ranged from -45.83±2.30 to -47.70±354 mV. The firmness of nano-emulsion formulations were lower than control (753.20±7.53 g) which were measured from 477.45±8.52 to 658.84±10.20 g. Finally, viscosity of nano-emulsion also lower than control group (2773±632.64 Pa·S), where the values were ranged from 1393±210.32 to 2033±32.15 Pa·S.
Conclusions
Glucomannan showed promising application in cream development as it exhibits non-toxic and high bioavailability. Development of glucomannan cream using 1.5% glucomannan concentration combination of both avocado oil and olive oil (Formulation C) provides small mean particle size and uniform polydispersity index with good zeta potential. The firmness and work of shear of Formulation C also provides comparable results to control group. Finally, non-Newtonian pseudoplastic properties of these creams provide an even spreadibility on skin.

Keywords: Nanoemulsion, Glucomannan, Atopic dermatitis, Anti-steroidal treatment

PPT5

Preparation and characterisation of fast-dissolving oral films

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Background

Fast-dissolving oral films (ODF) are thin sheets designed to rapidly disintegrate when in contact with saliva to release the incorporated active, without the need for swallowing. Difficulty in swallowing solid dosage forms (e.g. tablets) has been identified as one of the factors affecting the non-compliance of patient populations such as paediatric and geriatric. Thus, ODF may serve as an alternative to existing dosage forms. This study aimed to formulate and characterise a series of ODFs made from hydroxypropyl methylcellulose (HPMC) and carboxymethylcellulose (CMC), plasticised with glycerol and sorbitol.

Methods

Three formulae of each HPMC and CMC were prepared by solvent casting technique. The resulting films were characterised physically (i.e. visual appearance) and mechanically (i.e. mass and thickness variation, folding endurance and tensile strength). Furthermore, the placebo films were also assessed in terms of their disintegration time and contact angle.

Results

The films produced were not sticky, easy to handle and acidic in nature. They had an average mass between 19 to 30 mg and thickness between 41 to 78 µm. Films of CMC were significantly thicker than the HPMCs ($p<0.05$). For CMC films, reduction in CMC and increase in plasticiser contents were found to slightly enhance their tensile strength and elasticity, indicative of weaker and softer films. On the other hand, the HPMC films exhibited greater tensile strength, but lower extensibility than the CMC films. Films dissolved within 180 s and 25 s for CMC and HPMC, respectively. The CMC films took longer time to disintegrate than the HPMC films due to their higher contact angles values with water. The disintegration of all films increased in corresponding to an increase in tensile property.

Conclusion

Formulation HPMC-3 was considered as the best candidate for further optimisation for drug loading as it possessed the ideal balance between toughness and flexibility.

Keywords: hydroxypropyl methylcellulose; carboxymethylcellulose; fast-dissolving; oral film

PPT6

Effects of fermented soybeans on wound healing using diabetic induced rats

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Background
The use of biologic wound dressing derived from plant's active biomolecules provides important insights to the development of biodegradable materials for medical application. Plant-derived proteins such as soybeans have low immunogenicity potential, low molecular weight than animal derived proteins and are biodegradable. In this study, fermented soybeans were used as a biologic wound dressing as they contain isoflavonoid genistein (aglycone) that promote tissue regeneration, support collagen deposition and accelerate wound healing due to the anti-inflammatory and antioxidant properties.

Methods
This study investigated the effects of fermented soybeans (soy sauce) as biologic wound dressing on wound healing using streptozotocin-induced diabetic rats by preparing two different viscosities of fermented soybeans and tested against the healing process. Their wound healing profiles, wound morphology, antimicrobial activities and histology were determined.

Results
The *in vivo* studies performed on Sprague Dawley rats demonstrated that high viscosity fermented soybeans could promote regeneration of skin and accelerate the wound healing process. Nonetheless, fermented soybeans exhibited minimal antibacterial effect due to the absence of phenyl group in genistein resulting in less activity against *Staphylococcus aureus*. The histology analysis showed that the wound closure has achieved 100% re-epithelialization on day 20 in case of rats treated with fermented soybeans of higher viscosity.

Conclusions
These findings indicated that fermented soybeans have the potential to be developed as a biologic wound dressing.

Keywords: Diabetic rats, partial thickness wound, wound healing, fermented soybeans

PPP1

L-Stepholidine (SPD) treatment ameliorates learning and memory deficits in ICR mice

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Background
L-Stepholidine (L-SPD) was recently identified through our virtual screening exercise in search of potential drugs for Alzheimer's disease (AD). L-SPD is an active ingredient of the Chinese herb *Stephania intermedia*. Prior research has reported beneficial effects of L-SPD on dopamine D1- and D2-type receptors, suggesting promising treatment/prevention approach for neurodegenerative diseases. This study evaluated the effect of SPD on spatial learning and memory in lipopolysaccharide (LPS)-induced murine neuroinflammation model.

Methods
ICR male mice (*n*=8/group) were randomly grouped as follows: $control_{saline}$, LPS_{saline}, LPS SPD 3 mg/kg b.w., LPS SPD 5 mg/kg b.w., LPS SPD 10 mg/kg b.w. and positive control: LPS D-Serine (30 mg/kg). The mice were allowed to acclimatize for 3 days prior to treatment with SPD (i.p.) for 5 days (i.e. on day 1, 2, 3, 4, 5). Except for the $control_{saline}$ group, all mice received LPS (1 mg/kg b.w.). Following treatment, the mice were subjected to Morris water maze (MWM) test to evaluate the spatial learning and memory function. Finally, a probe trial was conducted on day 6 to evaluate their memory retention.

Results
SPD treatments at 5 and 10 mg/kg bw displayed earliest measure of learning, with an escape latency of ~18-25 secs compared to ~26-35 secs of control. SPD-treated groups (3, 5 and 10 mg/kg bw; 2.04-2.33) entered the platform zone more frequently compared to positive (1.41) and negative control (1.79). SPD treated mice showed better spatial learning (shorter escape latency and travelled distance) than the LPS control. A high SPD dose (10 mg/kg) showed a significant increase in the number of entries to the platform zone and time spent in the target quadrant.

Conclusions
Based on the swimming time in the target quadrant and the frequency of crossing the platform, SPD treatment may ameliorate cognitive deficits in learning and memory functioning in ICR mice.

Keywords: L-Stepholidine; lipopolysaccharide; neuroinflammation; memory deficits; behavioral test.

PPP2

Synthesis, characterization, and antioxidant potential of biodegradable polyurethane based on polypropylene fumarate as polyol

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Background
Prominent biomaterials for various biomedical applications include natural or synthetic polymers. Among synthetic polymers, polyurethanes (PUs) are unique due to their versatile physiochemical and mechanical properties. Free radicals in body e.g. nitrogen and oxygen are very reactive and cause oxidative damage of cells and tissues, thus affecting normal healing and regeneration processes. There is a need to develop and explore antioxidant potential of ligands, capable of neutralizing reactive free radicals. In the present study, novel biodegradable PU was synthesized, based on polypropylene fumarate diol as polyol, hexamethylene diisocyanate (HDI) and poly-3-hydroxy butyrate as chain extender via two step growth polymerization process.
Methods
The prepared samples were characterized by using Fourier Transform Infrared Spectrophotometer (FTIR), Nuclear Magnetic Resonance (NMR), mass spectrometry and Scanning Electron Microscope (SEM).
Results
The FTIR spectrum of PU prepolymer, exhibiting C=C at 1645 $cm^{-1}$and C=O at 1726 cm^{-1} confirmed the presence of polypropylene fumarate.
Conclusion
The aim of the present study is to exploit antioxidant activity of the synthesized novel polyurethane via DPPH (2,2-diphenyl-1-picryl hydrazyl-hydrate) Assay. The results supported antioxidant potential of the synthesized novel polyurethane, to be employed further in biomedical applications.

Keywords: Polyurethane, Polypropylene Fumarate, Poly-3-hydroxy butyrate, Antioxidant

PPP3

Polyethyleneimine cytotoxicity against human cancer cell lines

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Background
Polyethyleneimine (PEI) is a simple and cost-effective reagent for condensing and linking plasmid DNA to cells for gene delivery. However, its cytotoxicity has not yet to be reported. The aim of this study was to determine the lethal dose (IC_{10}) of PEI against breast cancer (MCF7), lung cancer (A549) and liver cancer (HepG2) cell lines.
Methods
MCF7, A549 and HepG2 cancer cell lines were treated with various concentrations of PEI for 24 hours. The viability of the cells was determined using the MTS assay.
Results
The IC_{10} of PEI for MCF7, A549 and HepG2 cell line were 73.2 µg/mL, 62.0 µg/mL and 70.5 µg/mL, respectively. This indicated that PEI is more cytotoxic towards A549 cancer cell line.
Conclusion
The IC_{10} results obtained from this study is useful to optimise transfection parameters of PEI on A549, MCF7 and HepG2 cell lines.

Keywords: Transfection, polyethyleneimine, cancer

PPP4

Antibacterial activity of selected Cambodia medicinal plants *in vitro*

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Background
Antimicrobial resistance has become a serious problem of public health. It creates a constant need for either new antimicrobial compounds or inhibitors of mechanisms that underlie antibiotic resistance. Cambodia is one of the well-known South-East Asia countries where natural substances are widely used for treatment of many diseases, especially for infectious diseases. As such, the study of antibacterial activity of plants traditionally used by Cambodian traditional healers to treat infectious diseases is important. This study aimed to screen the antimicrobial activity of 138 extracts from 67 plants that are traditionally used by Cambodian traditional healers.
Methods
The plants were collected in eight provinces and cities of Cambodia. The extraction was performed using ethanol:water (50/50 *v/v*) to obtain the majorities of the compounds present in plants. The antibacterial activities of plants extracts were first tested against reference strains, *Staphylococcus aureus* (ATCC 6553; cocci; Gram positive bacteria) and *Pseudomonas aeruginosa* (ATCC 9027; rod; Gram negative bacteria), and then against clinical strains using micro-dilution and macro-dilution tests, respectively.
Results
A total of 138 extracts isolated from 67 species of plants were tested. Most of the extracts were very active against *S. aureus* but less active against *P. aeruginosa*. Only 5 extracts derived from 5 plants were highly active against both standard and isolated strain of *S. aureus*. Three plant extracts were highly active against standard strain of *P. aeruginosa* but weakly active against its isolated strain.
Conclusions
Our results showed a great variability of the bacteriostatic qualities of extracts between isolated and standard strains. These results warrant selection of the most active extracts for development of antimicrobial products based on medicinal plants.

Keywords: Antibacterial activities, *Staphylococcus aureus, Pseudomonas aeruginosa,* Micro-dilution, Macro-dilution

PPP5

Body mass index is not correlated to blood glucose levels in Down Syndrome individuals

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Background

Down syndrome (DS) is a common chromosomal abnormality that occurs in about 1 in 700 live births. Previous studies found that DS is associated with higher obesity rates. There is a wide spectrum of medical complications among individuals with DS, which include diabetes mellitus, that are associated with increased susceptibility to weight gain. Therefore, the present study was carried out to evaluate the association between body mass index (BMI) and fasting blood glucose levels in DS individuals.

Methods

Measurement of height and weight was done, and BMI was calculated. Blood was collected with informed consent from the parents or guardians of DS individuals (n=52) and controls (n=52). Fasting blood glucose level was measured by using the Reflotron® Plus System.

Results

The mean BMI of individuals with DS ranged from 11.1 to 37.2 kg/m^2 with 13.5% (n=7) being overweight and 7.7% (n=4) obese. The mean BMI of controls ranged from 13.8 to 33.3 kg/m2 with 19.2% (n=10) being overweight and 3.8% (n=2) obese with no significant difference (p>0.05) between DS and controls. There was also no significant difference (p>0.05) in the fasting blood glucose levels in DS (mean=5.22 mmol/L) when compared to controls (mean=5.35 mmol/L). With respect to the association of BMI and fasting blood glucose, the present results failed to prove the relationship in both groups (p>0.05).

Conclusions

It can be concluded that there was no significant difference in the levels of fasting blood glucose in DS individuals when compared to controls. This study also found no prove of association between fasting blood glucose levels and BMI in both DS and controls group.

Keywords: Down syndrome, BMI, Glucose

PPP6

Effects of M*yrmecodia platytyrea* methanolic tuber extract on sub-acute cancer-induced severe combined immunodeficiency (SCID) mouse model

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Background
Myrmecodia platytyrea (Ant's plant) is member of Rubiaceae family. Tuber of *M. platytyrea* is used traditionally as decoction to treat various mild and severe diseases including cancer. Other species of *Myrmecodia* including *M. platytyrea* have been reported for their antiproliferative effect against various cancer cells *in vitro*. Hence, this study was carried out to investigate the effect of sub-acute administration of *M. platytyrea* methanolic tuber extract (MPMTE) on hepatocellular carcinoma (HCC)-induced SCID mice.

Method
A total of 36 SCID mice were divided into 6 groups (n=6/group): control 1 (non HCC-induced mice treated with normal saline), control 2 (HCC-induced mice treated with normal saline), control 3 (HCC-induced mice treated with 10 mg/kg doxorubicin) and 3 groups of HCC-induced mice treated with 100, 200 and 400 mg/ kg of MPMTE, respectively. NS and MPMTE were given orally twice daily, for 28 days whereas doxorubin was given intraperitoneally once daily to control 3 at 3-day intervals. Mortality, body weight, food and water intake were recorded throughout the experiment. Physical and behavioural changes were also observed. All mice were sacrificed on day 29. Tumour was excised and weighed.

Result
Control 1, 2 and the HCC-induced mice treated with MPMTE showed no mortality. No significant changes in terms of body weight, food intake and water intake were observed in all groups. However, HCC-induced mice treated with doxorubicin showed symptoms of toxicity and 100% mortality was recorded after 9 days of treatment. Remarkably, sub-acute oral administration of MPMTE (100 and 400 mg/kg, *p.o.*) suppressed tumour development at 13% and 6%, respectively. The tumour volume of mice treated with 200 mg/kg, on the other hand, was found to increase by14%.

Conclusion
SCID mice treated with MPMTE (100 and 400 mg/kg, p.o., bid) for 28 days showed tumour suppression, suggesting potential therapeutic value of the plant.

Keywords: *Myrmecodia platytyrea*; hepatocellular carcinoma; tumour suppression; severe combined immunodeficiency mouse model

PPP7

Antagonistic interactions between C*hromolaena odorata* ethanolic extract and cisplatin against breast cancer cell lines MCF-7 and MDA-MB-231

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Background

In Malaysia, breast cancer, which was ranked as the number one disease among female in 2016, has seen its incidence increased between 2007 to 2011. Cisplatin has been commonly used as the first line treatment against breast cancer. However, the combination uses of alternative medicine (CAM) together with conventional therapy by many cancer patients could possibly lead to unwanted interactions. This study had evaluated *Chromolaena odorata*, locally known as *pokok kapal terbang* for its potential to interact with cisplatin in combination therapy.

Methods

C. odorata was extracted using maceration method with 70% ethanol. Antiproliferative activity of the extract was screened against a panel of cell lines using the MTT assay. For combination study, MCF-7 and MDA-MB-231 breast cancer cell lines were treated with *C. odorata* ethanolic extract in combination with cisplatin. Isobologram and combination index (CI) were derived from the combination treatments.

Results

The yield of extraction was 2.69%. The IC_{50} values of *C. odorata* antiproliferative activity against MCF-7, MDA-MB-231, WRL68 and CRL2522 were 0.15 ± 0.00 mg/mL, 0.43 ± 0.02 mg/mL, 0.31 ± 0.00 mg/mL and 0.63 ± 0.00 mg/mL, respectively. Co-treatment of cisplatin and *C. odorata* ethanolic extract at IC_{10}, IC_{15} and IC_{25} against MCF-7 and MDA-MB-231 resulted in CI greater than one.

Conclusions

Combination treatment of cisplatin and *C. odorata* ethanolic extract leads to antagonistic interactions.

Keywords: *Chromolaena odorata,* cisplatin, MTT, isobologram, combination index

PPP8

In silico prediction of bile pigments binding affinity towards CYP2A6 enzyme

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Background
Binding of cytochrome P450 2A6 (CYP2A6) enzyme to tobacco-specific N-nitrosamine, 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK) results in electrophilic species that would later react with DNA to form DNA adduct. Bile pigments such as bilirubin and biliverdin, which are substrates for CYP2A6 enzyme, may inhibit the binding of NNK to CYP2A6 enzyme. Therefore, the aim of this study was to predict the binding properties and affinity of bile pigments towards CYP2A6 using *in silico* approach.
Methods
Molecular docking using AutoDock software was performed to computationally predict the binding properties and calculate the binding affinity of bile pigment towards the wildtype and CYP2A6 variant proteins. DoGSiteScorer and Allopred programs were used to predict potential drug binding and allosteric pockets on the CYP2A6.
Results
The binding affinity of bilirubin and biliverdin to the active site of CYP2A6 enzyme (with an average of 26.6 kcal/mol and 28.0 kcal/mol, respectively) was lower than that of NNK to CYP2A6 (with an average of -6.77 kcal/mol). Several potential binding pockets were identified on the CYP2A6 enzyme using DoGSiteScorer and Allopred programs. Bilirubin and biliverdin showed high binding affinity to allosteric site as compared to the active site of CYP2A6 enzyme.
Conclusion
High binding affinity of bile pigments indicates their potential to inhibit the binding of NNK to CYP2A6 enzyme. However, this requires further confirmation by enzymology studies.

Keywords: NNK, CYP2A6, Bilirubin, Biliverdin, Molecular docking

PPP9

Mikania micrantha ehanolic leaf extract accelerates wound healing

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Background
Mikania micrantha Kunth is a tropical plant known as *Selaput tunggul* in Malaysia. Leaves of *M. micrantha* are widely used as poultice for wound dressings and healing of sores. Hence, this study was carried out to investigate the ability of *M. micrantha* ethanol extract (MELE) in accelerating wound healing process.
Methods
Excision wound animal model was used in this study. Male Sprague-Dawley rats were topically administered (0.2 ml of treatment twice daily) with MELE (10, 20 and 40 mg/ml), normal saline (vehicle control group) and solcoceryl gel (positive control group) on an excision wound area (2 cm in diameter) at the nape of the dorsal of the rat. The wound closure rate was calculated throughout the study until wounds are completely healed. Then, the rats were sacrificed, and the skin was excised for histological examination.
Results
MELE demonstrated potential wound healing properties compared to solcoseryl gel. Percentage of wound closure of rats treated with MELE was significantly higher than both negative group and positive groups. Rats treated with MELE (40 mg/ml) took only 16 days to completely heal compared to negative group and positive groups, which took 25 days and 18 days, respectively. Furthermore, from the histological examination, wounds on rat treated with MELE had less inflammatory cell, more collagen and angiogenesis tissue.
Conclusion
MELE is effective in accelerating wound healing and further studies will be carried out to identify its molecular mechanism and the responsible bioactive compounds.

Keywords: *Mikania micrantha*, wound healing, cell proliferation, scratch wound assay

PPP10

Study on the effect of some medicinal plants on *in vitro* proliferation of peripheral blood mononuclear cells and their antioxidant activity

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Background
Medicinal plants have been used widely in the treatment of immune-related diseases such as immunodeficiency, hypersensitivity, inflammation, or autoimmune diseases, yet little is known about their mechanisms of action. Therefore, this study was conducted to study the effect of some medicinal plants on in vitro proliferation of peripheral blood mononuclear cells (PBMCs) and their antioxidant activity.
Methods
PBMCs were isolated from whole blood of healthy donors. MTT assay was used to evaluate the effect of 13 extracts in 96% ethanol and 24 fractionated extracts on PBMCs in vitro proliferation. IL–2 concentrations secreted by extracts–treated PBMCs were quantitated using ELISA. The plant extract with the strongest antiproliferative activity was chosen for further evaluation on the apoptosis/necrosis and ratios of TCD3+/CD4+ and TCD3+/CD8+ of PBMCs. Antioxidant activities of 96% ethanol extracts and fractionated extracts were assessed using DPPH assay.
Results
Of the 13 ethanol extracts, 6 extracts inhibited and 2 extracts stimulated the in vitro proliferation of PBMCs. The extracts with inhibitory effects reduced the amount of IL–2, whilst the extracts with stimulatory effects showed no effect on IL–2 expression compared to untreated cells (control). The chloroform extract of *Wedelia chinesis* showed strongest inhibitory activity with an IC50 value of 16.1 ppm, exerting an increase of 19.1% in apoptosis and a decrease of 4.18% in TCD3+/CD4+ ratio compared to untreated cells. The chloroform extract of *Piper betle* showed a strong antioxidant activity with an EC50 of 1.94 ppm, 2.1 times higher than that of vitamin C.
Conclusions
The chloroform extract of *Wedelia chinensis* had a potential of being used in the treatment of autoimmune diseases. Further studies are needed to isolate and identify the compounds responsible for this activity.

Keywords: PBMCs, interleukine–2, TCD3+CD4+/ TCD3+CD8+, cytotoxicity, antioxidant

PPP11

Synthesis, cytotoxic evaluation and docking studies of novel benzimidazoles

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Background
Malignant neoplasm is the second most dreadful disease causing mortality in the world after cardiovascular disorder. Chemotherapy is one of the most effective methods to treat cancer. Therefore, discovery of new anticancer agents with high potency and less toxicity is a major focus in modern research. In present study, a series of novel benzimidazole derivatives were designed, synthesized, characterized and subjected to cytotoxicity and molecular docking studies.
Methods
In this research, we synthesized some new benzimidazole derivatives and N-alkyl/ aryl benzimidazole derivatives. The evaluations of the synthesized compounds for cytotoxicity against two cancerous cell lines (MDA-MB-231, ATCC® CCL-136™) and a normal cell line (LLC-PK1) were carried out by using MTT method. Molecular docking studies of all synthesized compounds into the binding site of tubulin receptor (PDB code: 1SA0) were also performed using Lead IT 2.1.0 software.
Results
Twenty six benzimidazole derivatives and fourteen N-alkyl/ aryl benzimidazole derivatives were obtained, and they were characterized by melting point, thin layer chromatography and structural elucidation by UV, IR and NMR. Cytotoxic activity showed eight compounds were effective against MDA-MB-231 cell line and seventeen compounds were effective against ATCC® CCL-136™ cell line. Among them, 2-(3-methoxyphenyl)-6-methyl-1H benzo[d]imidazole showed highest antitumor effect against MDA-MB-231 cell line (IC50 = 22.7 µM), while 2-(6-methyl-1H-benzo[d]imidazol-2-yl)phenol showed highest antitumor effect against ATCC® CCL-136™ cell line (IC50 = 6.8 µM). Molecular modeling displayed relationship between MDA-MB-231 cell line inhibition and the existence of hydrogen donor of active compounds with Thr-353, between the ATCC® CCL-136™ cell line inhibition and the existence of hydrogen bond with Thr-353 or Ala-354.
Conclusions
Our study discovered new benzimidazole derivatives with good cytotoxicity against breast and skeletal muscle cancers and selectivity on normal kidney cell line. Molecular modeling was performed to gain comprehensive understanding into plausible binding modes and for the purpose of lead optimization.

Keywords: Benzimidazole, cytotoxicity, docking

PPP12

MDA-MB-231 cells are resistant to low concentrations of medroxyprogesterone

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Background

Breast cancer is the most prevalent cancer among women worldwide. Despite treatment options available for breast cancer, the rate of mortality is high with over 500,000 deaths reported annually. The aggressiveness of triple-negative breast cancer (TNBC) makes the treatment challenging and this is especially true in preventing the cells from migrating to other sites in the body. Therefore, identifying compounds that can inhibit TNBC cells from metastasizing to other regions is crucial before it develops a secondary cancer. Medroxyprogesterone (MP) is a synthetic derivative of progesterone and it shares similar pharmacological actions to progestin. The cytotoxic effect of MP has never been reported in MDA-MB-231 cells, a metastatic TNBC cell line. Therefore, in this study, the effect of MP on MDA-MB-231 cells was first determined.

Methods

MDA-MB-231 cells were seeded in a number of 2,000 cells per well in 96-well plates and incubated overnight at 37^{o}C. The cells were then treated with a range of MP concentrations from 0 to 8.5 µM for 24h and cytotoxicity was determined by a colorimetric MTT assay. Absolute DMSO was used to break the formazan crystal formed and absorbance was measured at 550nm using a microplate reader.

Results

The results show that within the concentration range tested, MP did not cause any cytotoxic effect to MDA-MB-231 cells as indicated by a non-significant difference in the percentage of cell viability compared to the control group ($p>0.05$). This indicates that MDA-MB-231 cells are resistant to MP at least at this concentration range and therefore, it is safe to be tested for anti-metastatic activity in the future.

Conclusions

It is confirmed that MDA-MB-231 cells are safe to be treated with MP at a concentration of 0-8.5µM within 24h exposure. This is important to determine the inhibitory effect of MP against the metastatic capability of TNBC cells.

Keywords: Triple-negative breast cancer, Breast cancer metastasis, Medroxyprogesterone

PPP13

Thymoquinone protects against cigarette smoke extract-induced vascular dysfunction through inhibition of the RhoA/Rho-kinase signaling pathway

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Background
There is an ever-growing focus on the role of natural products in modulating reactive oxygen species (ROS) and RhoA/Rho kinase-mediated vascular disease. Thymoquinone (TQ), a constituent of the volatile oil derived from *Nigella sativa* seeds, possesses promising antioxidant and vasodilating properties via its effect on multiple signaling pathways; however, the effect of TQ on the RhoA/Rho-Kinase pathway remains to be investigated. The aims of the present study were to examine whether TQ protects against CSE-induced vascular dysfunction and to identify the underlying mechanisms of TQ on CSE-induced ED in isolated rat aorta.
Methods
Cigarette smoke extract (CSE)-exposed rat aortic rings were mounted onto a wire myograph and subjected to contraction and relaxation. Quantitative assessment of RhoA activation was determined using G-LISA RhoA Activation Assay Kit. Phosphorylation of myosin light chain-20, myosin phosphatase-targeting subunit 1 and protein kinase CPI-17 were determined by Western blot analysis of the whole tissue protein extracts.
Results
TQ protected against CSE-induced impairment of acetylcholine-induced endothelium-dependent relaxation, and decreased CSE-induced ROS generation and glutathione depletion. Preincubation of aortic rings for 20 min with TQ attenuated the CSE-enhanced phenylephrine-induced vascular tension in endothelium-denuded rings. TQ-pretreated rings showed a decrease in CSE-induced RhoA activation and phosphorylation of myosin light chain-20, myosin phosphatase-targeting subunit 1 and protein kinase CPI-17.
Conclusions
These data indicate that TQ inhibited ROS generation-induced RhoA/Rho kinase pathway activation, protecting against CSE-induced vascular dysfunction. This study provides mechanistic insight for understanding the molecular basis and efficacy of TQ on vascular disease management.

Keywords: Thymoquinone, Cigarette smoke, RhoA/Rho kinase.

PPP14

A potential role of norethisterone (ED-4) as an anti-metastatic drug against triple-negative breast cancer cells

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Background
Breast cancer is one of the most common malignant cancer in women. It is a heterogeneous disease that affects one in every eight women worldwide. Breast cancer metastasis is the most life-threatening aspect of breast cancer. It is a multiple step process involving an invasion of a primary tumour cell and followed by a subsequent colonization of the cell at the secondary sites in the body like bone, brain, liver, and lung. It was hypothesized that norethisterone (ED-4) might have the ability to inhibit the migration of metastatic breast cancer cells.
Methods
The MDA-MB-231 cells were treated with a range of ED-4 concentrations, from 0 till 8.5 μM, specifically, the cells were incubated with the drug for 18h at 37℃ at the concentration of 0, 2.5, 3.5, 4.5, 5.5, 6.5 and 8.5μM and their cytotoxicity was performed using a colorimetric MTT assay.
Results
The result showed that ED-4 did not induce cytotoxicity on MDA-MB-231 cells within the concentration range of 1μM up to 8μM ($p>0.05$). Therefore, ED-4 at this concentration range can be used to determine its efficacy as anti-metastasis against triple negative breast cancer cells.
Conclusions
To date, there is no drug available for a prevention of breast cancer metastasis and therefore, norethisterone (ED-4) was proposed as a new drug candidate to inhibit breast cancer metastasis. This potential could have benefits on future studies on the management of breast cancer metastasis among breast cancer patients.

Keywords: Breast cancer, Norethisterone (ED-4), Triple Negative Breast Cancer, Metastasis, MDA-MB-231 cells.

PPP15

Characterization and cytotoxic activity of semi-purified Fucoidan extract from *Sargassum polycystum* C. Agardh (Sargassaceae) against Acute Myelogenous Leukemia (AMLK) cell line using MTT assay

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Background

Leukemia is one of the most prevalent cancer types for Filipinos becoming the 7th leading form of cancer in both sexes. In 2012, the Philippines estimated national standardized mortality of leukemia were of 3.9 per 100,000. The brown macroalgae specifically from the *Sargassum* are considered as rich sources of phytochemicals such as Fucoidan and Fucoxanthin that act on multi-signaling pathways needed to combat cancer.

Methods

Isolation of the semi-purified Fucoidan extract was based on a process developed by Mak. W. (2012), in which pre-treatment of the sample with ethanol, precipitation with Calcium Chloride and ethanol concentrations, with centrifugation steps in between. The study evaluated the physicochemical characteristics including the following: i) organoleptic, ii) solubility, iii) phytochemical assay, iv) fucose, sulfate and glucoronic content using UV-VIS spectroscopy, instrumental analysis using Fourier Transform Infrared Spectroscopy (FTIR) and the cyototoxic activity of semi purified Fucoidan extract against Acute Myelogenous Leukemia (AMLK) cell line using MTT assay using doxorubicin as the positive control.

Results

The obtained percentage yield showed that 440.15 g of the pretreated *Sargassum polycystum* contained 1.13% semi-purified fucoidan extract. Solubility test confirm the solubility of the extract to water and hydrochloric acid. The semi-purified Fucoidan isolate was characterized of its fucose, sulfate, and glucuronic acid content, with results of 26.23%, 23.52%, and 32.71%, respectively. FTIR spectrum confirms the presence functional moieties i.e. isothiocyanate and sulfonyl that are also found on fucoidan standard and sulfated polysaccharides, these functional groups may be attributed to the different biological activities that Fucoidan exhibits. Cytotoxic activity was evaluated using MTT assay, wherein results showed that the semi-purified Fucoidan extract from *S. polycystum C. Agardh* (Sargassaceae) exhibits cytotoxic activity against AMLK cell line, with concentration of 6.25 µg/mL having the highest inhibitory rate of 44.08%. Statistical treatment showed significant difference between the semi-purified Fucoidan extract and the standard drug, doxorubicin.

Conclusions

In conclusion, the semi-purified Fucoidan extract from *S. polycystum C. Agardh* (Sargassaceae) may exhibit anti-proliferative effect against AMLK cell line.

Keywords: Sargassum, Cytotoxicity, Fucoidan, MTT assay, AMLK cell line

PPP16

Formulation of antibacterial ointment from the ethanolic crude extract of ikmo leaves (*Piper betle* Linn. Piperaceae family)

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Background

According to World Health Organization cases of Antimicrobial Resistance (AMR) have exponentially increased yet fewer antibacterial agents are discovered on the past years. AMR hampers the control of infectious diseases resulting to an increase in health care cost and risk of spreading resistant microorganisms in the community, these events is a growing public health challenge and poses a global health crisis if remain uncontrolled. Ikmo leaves on the otherhand has been well studied and has shown abundant and potential source of phytoconstituents that may be developed as antimicrobial agent and incorporate it to an applicable dosage form, therefore to address this concern the researchers formulate a plant-derived antibacterial ointment from the ethanolic crude extract from *Piper betlle* locally known as Ikmo.

Methods

Mature Ikmo leaves were collected, dried and extracted. The extract was then subjected to physicochemical characterization and antibacterial assay by means of agar-plate method. The plant concentration that exhibits the most active effect against *Staphylococcus aureus* and *Pseudomonas aeruginosa* ($p<0.05$) will be used in the formulation of antibacterial ointment. To ensure the safety of the formulated product, initial dermal irritation test was conducted using rabbits.

Results

The yield of ethanolic extract of Ikmo leaves extract is 9.922% and is found to have greenish-black color, creosote-like odor and has syrupy consistency. The ethanolic crude extract was soluble in acetone, ethanol, and ether and insoluble in water. The optimized extract concentration of 60% was further develop to ointment and is the subjected to antibacterial assay against *Staphylococcoccus aureus* and *Pseudomonas aeruginosa* resulting to a zone of inhibition of 23.05±1.35 mm and 26.40±0.89 mm compared to mupirocin (*14.93* ±0.03 mm and 17.55±0.03 mm). Dermal irritation test has also shown that the formulated extract does not show any skin reactions to test animals.

Conclusion

Based on the result of the study, the formulated ointment of the optimized ethanolic crude extract of Ikmo leaves has shown to be a potential agent to be further studied considering its good preliminary antibacterial effect and dermal irritation test.

Keywords: *Piper betle* Linn. Ikmo leaves, *Staphylococcus aureus, Pseudomonas aeruginosa,* antibacterial, ointment

PPP17

Plastics exposure reveals significant metabolic changes *in vivo*

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Background

Plastics debris is well-known for its adverse effects on the ecosystems. However, there has been limited evidence of the exposure of the plastic to the metabolism of the soil-living organism. This study set out to examine the association between plastics exposure and metabolic alterations *in vivo*.

Methods

Firstly, we established an *in vivo* model of soil organism. Next, several bioassays regarding oxidative stress, locomotion, and reproduction were performed to examine the effect of plastic exposure to the living organism. We then developed an analytical method to unbiasedly examine the metabolic alterations of the living organism. Finally, biological interpretation was conducted using bioinformatics software.

Results

Plastics appeared to affect the reproduction, locomotion, and oxidative stress of the exposed living organisms. The effects were found even at the low concentration of plastics. Plastics of smaller size showed more severe consequences than those of bigger size. Our global metabolite profiling experiment also found a significant change regarding the energy-related metabolism and the intermediates of neurotransmitters.

Conclusions

This study provided a proof-of-concept regarding the effect of plastic exposure to various phenotypes of the soil-living organism. More studies are needed to extend our observations.

Keywords: plastics, *in vivo* model, omics, metabolism

PPP18

Antibacterial activity of *Musa paradisiaca* stem extracts against isolated UTI pathogens

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Background

Urinary tract infection (UTI) has become a more serious problem today, due to multidrug resistance of Gram-positive (GP) and Gram-negative (GN) bacteria. *Musa paradisiaca* is used as a medicinal plant in traditional system of healing many infectious diseases. The goal of our research was to evaluate antimicrobial efficiency of *Musa paradisiaca* (banana) stem extracts against isolated UTI pathogens.

Methods

Banana stem extracts were obtained with maceration technique using two solvents separately: distilled water and methanol . Agar well diffusion assay was used for evaluation of antimicrobial properties of stem extracts against isolated UTI pathogens. Minimum inhibitory concentrations and minimum bactericidal concentrations were determined by broth dilution method and agar plate method. The preliminary phytochemical analyses of the plants were carried out using standard procedure.

Results

A total of 5 UTI pathogens were isolated from the UTI patients attending in the hospital such as *Pseudomonas aeruginosa, Klebsiella pneumoniae, E. coli, Enterococcus faecalis* and *Staphylococcus aureus*. Aqueous and ethanol extracts expressed antimicrobial activity against isolated UTI pathogens except *S.aureus* at 500 mg/ml. Zone of inhibition of the extracts were compared with ciprofloxacin (250mg/ml). Ethanolic extracts of *M paradisiaca* inhibited the growth of *P. aeruginosa* and *E. faecalis* at 62.5 mg/ml and *K. pneumoniae* at 125 mg/ml. Aqueous extracts of *M paradisiaca* inhibited the growth of *K. pneumoniae* and *E.coli* at 250mg/ml. Ethanol extracts of *M. paradisiaca* exhibited bactericidal activity against *P. aeruginosa* and *E. faecalis* at 250 mg/ml. Ethanolic extracts exhibited better antibacterial activity against tested strains than water extracts. The antibacterial activity of the *M paradisiaca* was due to the presence of alkaloids, tannins, flavanoids, terpenoids and sugars.

Conclusions

Hence, the plant *M. paradisiaca* stem contains potential antimicrobial compounds against UTI pathogens.Further study is required to identify the bioactive compounds, mode of action and in vivo toxic effect of *M. paradisiaca*.

Keywords: antibacterial activity, *M paradisiaca* stem extracts, minimum inhibitory concentration, phytochemicals

PPL1

TM4SF5 senses lysosomal arginine

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Background
The mechanistic target of rapamycin complex (mTORC1) acts as a signaling hub on the lysosome surface, responding to lysosomal amino acids. Although arginine is metabolically important, the physiological arginine sensor that activates mTOR remains unclear.
Methods
Here, we show that transmembrane 4 L six family member 5 (TM4SF5) translocates from plasma membrane to lysosome upon arginine sufficiency and senses arginine, culminating in mTOR and S6K1 activation.
Results
TM4SF5 bound active mTOR upon arginine sufficiency and constitutively bound amino acid transporter SLC38A9. TM4SF5 binding to the cytosolic arginine sensor Castor1 decreased upon arginine sufficiency, thus allowing TM4SF5-mediated sensing of metabolic amino acids. TM4SF5 directly bound free L-arginine via its extracellular loop possibly for its efflux, which was supported by mutant study and homology and molecular docking modeling.
Conclusions
Therefore, we propose that lysosomal TM4SF5 senses enables arginine efflux for mTORC1/S6K1 activation, and that arginine-auxotroph in hepatocellular carcinoma may be demised by anti-TM4SF5 reagents.

Keywords: arginine, liver cancer, mTOR, structural modeling, TM4SF5

PPL2

Ultrastructure of the liver in rat model of insulin resistance

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Background

Liver sinusoidal endothelial cells (LSECs) are perforated with transcellular fenestrations that provide unimpeded access of substrates between sinusoidal blood and hepatocytes. Defenestration refers to the loss of fenestration number and/or decreasing in fenestration diameter which alters metabolic homeostasis. Insulin resistance has been reported to promote accumulation of fat in the liver leading to fatty liver disease. However, the effect of insulin resistance specifically on fenestrations is yet to be investigated. This study was conducted to observe changes in fenestrations of LSEC in response to insulin resistance.

Methods

Adult male Sprague-Dawley rats were divided into two groups (n=8) where control group received 0.9% NaCl and treatment group received dexamethasone injection (1 mg/kg) i.p once daily for ten days. At day 11, all rats were anaesthetised using ketamine/xylazine followed by cardiac puncture. Rats were dissected and livers were perfusion-fixed for electron microscopy. Fenestrations were examined using Quanta FEG450 Scanning electron microscope at 15000x magnification. Ten random images per sample were taken for analysis of fenestrations diameter and porosity using ImageJ software. Data was analysed using SPSS version 23.0.

Results

Dexamethasone induced insulin resistance as observed by a significant reduction of body weight (D=276.84 ±7.87 vs C=393.84±12.47g; p=0.00), increased fasting blood glucose (D=5.57 ±1.30 vs C=3.97±0.55mg/dl; p=0.02) and higher HOMA-IR value (D=1.37±0.52 vs C=0.85±0.22; p=0.00) in the treatment group as compared to the control. Analysis of the liver images showed that insulin resistance caused defenestration of LSEC with a significant decrease in fenestrations frequency (D=3.202±1.16 vs C=2.656±1.044; p=0.04) and endothelial porosity (D=2.17±0.74 vs C=1.77±0.9; p=0.049) but not fenestration diameter.

Conclusions

In conclusion, this finding shows that insulin resistance can affect the integrity of liver endothelium specifically on fenestration frequency and liver porosity which will consequently lead to serious implications on liver function as the main site for metabolism.

Keywords: Insulin resistance, liver endothelium, fenestrations, electron microscopy

PPL3

Locomotor, exploratory and anxiety-like behavior assessment of aged rats following intrahippocampal injection with streptozotocin: A novel Alzheimer's disease rodent model

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Background
Alzheimer's disease (AD) is a progressive neurodegenerative disorder associated with a series of pathophysiological changes, accumulation of amyloid plaques and tau tangles. The hippocampal region which is responsible for long-term memory and spatial navigation demonstrated neuronal loss in AD patients. Peripheral exposure of streptozotocin (STZ) used in diabetic studies showed AD pathogenesis in 8 months. A reliable AD rat model should resemble the brain metabolic and behavioral disturbances in humans. Thus, the present study was conducted to investigate the effects of intrahippocampal (IH)-STZ administration that directly target the insulin receptors on the locomotor activity and anxiety-like behavior at two time points (3 and 12 weeks) post-STZ injection.
Methods
Forty-male (12 months old) Sprague-Dawley rats (350-450 g) were divided into two groups to monitor the progression of AD at two time points (3 weeks and 12 weeks, n=20 respectively). The rats were further divided to control (no treatment, n=5), sham-operated (received PBS, n=5) and treatment (IH-STZ, n=10). STZ (3 mg/kg; 5 µl) was administered bilaterally as a single injection into the dorsal hippocampus of the rats using a stereotaxic apparatus. The open field test using the open square maze (50 cm x 50 cm) tracked with software (ANY-maze) was used to record the rat's behavior.
Results
There were no significant differences in spontaneous locomotor, exploratory activity and time spent in the central area between the groups. Rats from group 3 and 12 weeks also did not show significant changes in all the parameters when compared between the two time points ($p < 0.05$).
Conclusions
STZ when administered intrahippocampally did not impair the rats' locomotor activity, absent of any signs of anxiety and exhibited normal exploratory behavior. The rodent IH-STZ is a suitable model to study treatment and prevention of AD as the behavior and pathology resembled AD patients.

Keywords: Alzheimer's disease, Intrahippocampal, Rodent model, Streptozotocin, Locomotor

PPL4

Prediction of breast cancer utilizing genomic data: A review

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Background

Breast cancer is one of the most commonly diagnosed cancer with high mortality rate affecting millions of women around the world. In recent years there is an increasing interest to detect and predict breast cancer prognosis and treatment including using genomic data. Consequently, huge number of publications were found in this area. This study is therefore aimed to review relevant articles related to breast cancer prediction that utilized genomic data to determine data that is most effective in predicting breast cancer.

Methods

Keywords used in the article search were “prediction”, “breast cancer”, “genomics” and “biomarkers”. A Boolean search was conducted in four databases and 38 articles were chosen; PubMed (n=20), Science Direct (n=5), Wiley Online Library (n=2) and Scopus (n=11). The inclusion criteria were articles available in full text, written in English language, published from 2013 to 2019 and excluded that of review, meta-analysis and systematic review.

Results

Expression profiles, DNA methylation, copy number alteration, protein expression, interactome networks, somatic mutation and single nucleotide polymorphisms were used in breast cancer prognosis prediction. However, the most relevant data with high prediction value is the DNA expression profile. Also, the combination of different types of omics data in a prediction were revealed to have better performance than using only one single data type.

Conclusion

Genomic data could be an effective prediction tool and biomarker for early detection of breast cancer, its monitoring and prognosis. Hence, utilizing these data might lead to reduction in breast cancer morbidity and mortality rate promoting better healthcare for breast cancer patients.

Keywords: Prediction, prognosis, breast cancer, genomic, biomarkers

PPL5

Machine learning based prediction of potential interaction between leukaemia-related proteins and C*entella asiatica* compounds

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Background
Leukaemia is one of the leading causes of morbidity and mortality in adult and children worldwide. Finding specific targets for anti-leukaemic activity is challenging, due to the limited understanding of target selectivity features and compounds. For this purpose, a one vs one (OvO) classification model was built on bioactivity data of 23 leukaemia-related proteins to assess potential compound-target interaction of 4 main *Centella asiatica* compounds.

Methods
An OvO classification model was trained on bioactivity data containing protein-ligand interactions between 23 leukaemia-related proteins and 17,637 compounds. The data was obtained from ChEMBL (https://www.ebi.ac.uk/chembl/) database. The compounds were converted to ECFP_4 fingerprint and Random Forests was used as the machine learning algorithm to deduce a mathematical correlation between compound structure and protein receptor in the training set. The model was validated using a 5-fold cross validation and potential target interaction of *C. asiatica* compounds; Asiaticoside, Madecassoside, Asiatic acid and Madecassic acid were then identified using the model.

Results
In the internal validation, the OvO model exhibited an average sensitivity of 0.87, specificity of 0.96, q^2 value of 0.57, and root-mean-square error (RMSE) of 0.22. In the prediction of potential protein targets for *C. asiatica* compounds, 3 potential proteins (CHEMBL1997, CHEMBL1825 and CHEMBL2034) may interact with the tested compounds. The next phase of the study will involve testing the 4 compounds against the 3 predicted proteins *in vitro*.

Conclusions
Machine learning based prediction of interaction between protein target and bioactive compounds may serve as a valuable tool in searching for potential lead compounds in leukemic diseases.

Keywords: One vs one classification; target prediction; machine learning; interaction; leukaemia-related protein

PPL6

Centella asiatica extract (CAE) improves motor performance of methamphetamine-treated rats

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Background
Methamphetamine or METH, a psychostimulant with devastating neurotoxic effects on the central nervous system. METH's abuser has been associated with Parkinson's disease (PD)-like motor deficits. *Centella asiatica* or "pegaga" is remarkably known to improve behavioural and motor impairment.Therefore, in this study, narrow beam test was performed to evaluate motor performance of rats following METH and CAE treatments.
Methods
Male Sprague-Dawley rats were assigned into Group I (Control), Group II (50mg/kg METH twice per day for 4 days), Group III (300mg/kg CAE for 21 days), Group IV (500mg/kg CAE for 21 days), Group V (50mg/kg METH + 300mg/kg of CAE for 21 days) and Group VI (50mg/kg METH + 500mg/kg CAE for 21 days). Rats were subjected to narrow beam test on the 1st day after the last treatment. Rats were placed at the initial start of wooden narrow beam with smooth surface of 100cm in length and 6mm in width and 100cm of height. Total time of 120s was set as a maximum limit. Time taken for each rat to cross beam or reach escape box (escape latency) was recorded. Error was recorded as any failed attempt to reach escape box, loss of balance and fall from the beam before 120s of maximum total time. $P<0.05$ was indicated as statistically significant.
Results
No significant change of body weight was observed on each treatment group. All rats were able to perform in the narrow beam test. A longert time taken for escape latency was showed by Group II, V and VI compared to control group. Group III and IV were showed significant shorter time for escape latency as compared to Group II. Meanwhile, Group II was exhibited significant increases in the number of error and difficulties as compared to control group.
Conclusions
Results demonstrated that CAE able to improve motor performance of METH-treated rats.

Keywords: Methamphetamine, *Centella asiatica* extract, motor performance, narrow beam test

PPL7

Intrahippocampal Administration of Streptozotocin Induces Spatial Learning and Memory Impairment in Rats

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Background
Alzheimer's disease (AD) is the most common neurodegenerative disorder characterized by a progressive decline of memory, cognitive impairments, and changes in behaviour and personality. The past decade has seen extensive intervention directed to AD treatment, but with little success to fully cure AD. In the search for new therapeutic interventions and to better understand the disease, various animal models have been developed. The ideal AD model should mimic the pathological aspects of human AD. Intracerebroventricular (ICV) injection of streptozotocin (STZ) has been widely used to induced sporadic AD in the rodent. However, insulin receptors (IR) which are very sensitive to STZ are more abundant in the hippocampal region but there have been no studies on the effect of intrahippocampal (IH) injection of STZ. Therefore, the present study was conducted to investigate the effects of bilateral IH injection of STZ on spatial reference learning and memory in rats.
Methods
Male Sprague Dawley rats (350–4 50g) were administered with a single bilateral intrahippocampal injection of STZ (3 mg/kg) or an equal volume of PBS. Two weeks post-surgery, the spatial learning and memory of the rats were assessed using the Morris Water Maze task.
Results
Rats subjected to bilateral IH-STZ injection took longer latency to locate the hidden platform in acquisition trials and spent less time in the target quadrant than the control group which indicate impaired spatial memory retention.
Conclusions
The present study demonstrates the potential of STZ to promote spatial learning and memory impairment in rats through IH injection that can be used as a reliable rodent model for AD.

Keywords: Alzheimer's Disease, Memory, Rat Model, Streptozotocin, Morris Water Maze

PPL8

Ceftriaxone attenuates oxidative stress and enriched antioxidants in memory dysfunction in aging rodent models

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Background
Excessive zinc (Zn) levels in the brain are known to impair cellular energy production through an inhibitory action on mitochondria. Mitochondrial dysfunction is known to be a factor in the pathogenesis of neurodegenerative disorders, like Alzheimer's disease (AD). Zn is an essential trace element in the brain, however, too much Zn has been associated with the pathogenesis of AD. Similarly, a surplus of glutamate has also been implicated in the development of AD. Ceftriaxone (CTX) is a beta lactam antibiotic with neuroprotective activity. The aim of this study was to investigate the effectiveness of CTX compared to donepezil (Don), an established drug for alleviating AD symptoms, in improving memory impairment induced by an excess of Zn in the senescent mice model.

Method
In this study we applied a behavioural tool, Morris Water Maze (MWM), followed by biochemical assays. The MWM task is a widely accepted method for investigating spatial learning and memory of rodents by measuring escape latency (EL), distance travelled (DT), the distance travelled before reaching the platform; and the time spent in the target quadrant (TQ). Accelerated senescence was induced through subcutaneous injection of d-galactose (D-gal) and oral administration of Zn daily for six weeks, an established model of aging. Mice were divided into 5 groups: the memory intact, untreated wild type (WT); memory-impaired via Zn-D-gal combination (control); and Zn-D-gal induced memory impaired treated with donepezil (Don) or 100 mg/kg (CTX-100) or 200 mg/kg (CTX-200) ceftriaxone. On days 35 to 37 of the treatment, all groups were put through MWM test.

Results
Based on the MWM test, treatment with CTX provides protection against Zn-D-gal induced toxicity. Both CTX-100 and CTX-200 groups appeared to have reversed memory impairment in the Zn-D-gal treated animals, evidenced by the shorter EL and DT, and longer TQ compared to the control group. The Don group also had improved memory. However, these improvements did not exceed the performance of the WT. The levels of lipid peroxidation and nitrite estimation were increased in the disease model while the superoxide dismutase (SOD) and reduced gluthathione were decreased, in these groups compared to groups treated with Don and CTX.

Conclusion
CTX provides protection against Zn-D-gal-induced toxicity, presumably by alleviating mitochondrial dysfunction of this model.

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

PPL9

Tth111i Single nucleotide polymorphism (SNP) among Malay subjects as detected by polymerase chain reaction (PCR) and DNA sequencing methods

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Background
Glucocorticoid receptor (GR) plays an integral role in regulating body functions. Polymorphism of the *Tth111I* theoretically increases the sensitivity of the GR receptor, with prominent evidence at higher HDL-C levels. Thus, this study aims to screen and to find the *Tth111I* SNP association with the HDL-C level by using polymerase chain reaction (PCR) and DNA sequencing methods among Malay subjects.

Methods
DNA was extracted and amplified from blood samples of 24 Malay subjects, which consist of 12 normal lean and 12 obese respondents.

Results
Among the ten sequenced samples however, none was detected as mutant. Since, all the samples were wild-types (WTs), hence, the association between the *Tth111I* SNP with the HDL-C level could not be made.

Conclusions
A larger sample size must be recruited, and further studies need to be conducted to determine the impact of this SNP on the HDL-c level to explain the potential roleof *Tth111I* SNP in preventing cardiovascular disease (CVD) among the Malay Malaysians.

Keywords: glucocorticoid receptor gene, single nucleotide polymorphism, *Tth111I*, HDL-C

PPL10

Assessment of high-throughput extraction methods for metabolomic studies

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Background
Metabolic phenotyping has become an important approach not only in translational experiments but also in clinical and epidemiological research. There have been various extraction methods designed for efficient extraction of small molecules (e.g., < 1500 Da) for high-throughput experiments. However, there has been little effort in summarizing or comparing these methods in practice.
Methods
This study set out to summarize and assess the current practice of metabolite extraction methods. We selectively focused on the methodologies that were developed for high-throughput experiments. Polar metabolites and lipids were investigated separately, since there are considerable differences regarding their chemical properties and methodologies. Some recommendations for the development of high-throughput small molecules extraction methods are proposed.
Results
Various methods have been developed for the extraction of polar metabolites whilst very few for that of lipids. Among the developed methods for the extraction of polar metabolites, the methods that are based on methanol:water with the ratio of 8:2 appeared to be the best method. For lipids, Methyl tert-butyl ether stands out from the rest due to its high-capacity and reduced toxicity.
Conclusions
More efforts should be given regarding the development of new extraction methods, especially for lipids and fragile compounds. Importantly, environment-friendly methods are of high interest.

Keywords: metabolomics, lipidomics, extraction method

PPL11

Ciproxifan protects the effects of D-galactose/aluminium chloride-induced memory impairment in mice through BDNF and neuronal cell marker

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Background
Alzheimer's disease (AD) type of dementia is related to β-amyloid deposition and formation of neurofibrillary tangles leading to the neuronal loss. Currently, only a handful of drugs are used to treat AD such as antagonists of acetylcholinesterase (AChE) and NMDA. Histamine H_3-receptor is an autoreceptor that controls the brain histamine release as well as other neurotransmitters such as acetylcholine and dopamine (heteroreceptor). Ciproxifan is a H_3-receptor antagonist that binds to the receptor and induces more histamine to be released, by blocking the negative feedback inhibition. It has also been shown to enhance cognition and memory.

Methods
Combination of D-galactose (D-gal) and aluminum chloride ($AlCl_3$) was used to induce memory impairment in ICR mice. Groups of mice were injected with ciproxifan (1 & 3 mg/kg, i.p.) for 42 consecutive days and administered with D-galactose by subcutaneous injection (100 mg/kg, s.c.) for 42 days, while $AlCl_3$ (100 mg/kg) was added in the water bottle, except for control. After 42 days, brain samples were harvested to determine the levels of brain-derived neurotrophic factor (BDNF)) and mRNA of PSD-95 and MAP2.

Results
Level of BDNF in the D-gal/$AlCl_3$ group was significantly lower ($P < 0.01$) than the control group. Conversely, BDNF levels in ciproxifan-treated mice (1 mg/kg and 3 mg/kg) were significantly higher ($P < 0.01$ and $P < 0.001$, respectively) than the D-gal/$AlCl_3$ group (negative control). Also, the administration of ciproxifan (3 mg/kg) in D-gal/$AlCl_3$-induced mice significantly upregulated the mRNA expression of PSD-95 gene ($P<0.05$) as compared to the control group. Moreover, the mRNA expression of MAP2 gene was significantly upregulated in ciproxifan-treated mice (1 mg/kg and 3 mg/kg) as compared to the control group ($P <0.05$ and $P<0.01$, respectively).

Conclusions
Ciproxifan may have the potential to increase neuronal activity through enhancement of the BDNF signaling pathway and neuronal cell markers (PSD-95 and MAP2).

Keywords: histamine H_3-receptor antagonist, ciproxifan, memory impairment, Brain-Derived Neurotrophic Factor (BDNF), PSD-95, MAP2

PPL12

Amyloid-beta aggregation inhibitory compounds isolated from fermented tea (*Camellia japonica*)

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Background

Alzheimer's disease (AD) is a progressive neurodegenerative disorder, and is associated with the formation of amyloid-β (Aβ) plaques which are generated from the cleavage of amyloid precursor protein. Thus far, (–)-epigallocatechin gallate(EGCG), curcumin and resveratrol are some of the natural product based compounds that possess inhibitory activities against Aβ aggregation. The current study was desingned to discover Ab aggregation inhibitory compounds from fermented tea (*Camellia japonica*).

Methods

Fermented tea was provided by Amore Pacific Co., and was extracted using acetone and ethanol. The etanol soluble extract was separated by diverse column chromatography methods. Isolated compounds were identifed by interpretation of spectroscopic data including one-, two-dimentional NMR, UV, IR and ESI-Q-TOF-MS. Amyloid-beta aggregation inhibitory activity was evaluated using Thioflavin T beta-amyloid aggreation kit and negative -stained transmission electron microscopy. The protective effect of the compounds was tested in Aβ-treatedSH-SY5Y cells by estimating the viability using the CCK-8 assay kit.

Results

Phytochemical investigation of the fermented tea led to isolation of 31 phenolic compounds including three new flavonoid glycosides. Among the compounds,, (–)-catechin gallate (CG), (–)-epicatechin gallate (ECG), and (–)-epigallocatechin gallate (EGCG) showed strong Aβ aggregation inhibitory effect whilstCG exhibited high protection in SH-SY5Y cells against Aβ-induced cytotoxicity.

Conclusions

CG and ECG showed more potent anti- Aβ aggregation effects than EGCG, a well-known natural Aβ aggregation inhibitor. The current study provides scientific evidences that compounds from fermented tea possess beneficial actions against neurodegeneration *in vitro*.

Keywords: *Camellia japonica*, phenolic compounds, anti-Aβ aggregation

PPL13

Cultivation of *Panax vietnamensis* in Lam Dong Province, Vietnam

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Background
In 1973, *Panax vietnamensis*, namely Vietnamese ginseng (VG) was first found at an altitude of 1,700~1,800 m on 14º north latitude in Mt. Ngoc Linh, Quang Nam province, Vietnam. Since then, VG has been mainly cultivated in Mt. Ngoc Linh area under the natural shade of trees, which is similar to the growing condition of wild VG. Consequently, arable land is limited and cultivation technique is difficult to standardize. Therefore, development of cultivation technique that can overcome such difficulties is necessary.

Methods
From 2014, we started a study on the cultivation of VG at Lam Dong Province, Vietnam with the seeds purchased from the VG farm in Mt. Ngoc Linh. They were sown in a flat piece of land located on 12º north latitude and at elevation of about 1,400 m under the artificial shade.

Results
The VG seeds were germinated very well. We could harvest the seeds from the 3 years old plants in 2017. Harvested seeds were sown again in the same experimental farm and successfully germinated and grown well. We were able to grow large number of VG plants from one to five years old in the same farm.

Conclusions
Our result demonstrates the possibility of large-scale cultivation of VG under the artificial shade in Lam Dong province, which is located on 12º north latitude. It is a record of most southern large-scale cultivation of *Panax* species in the northern hemisphere at lowest latitude. Although some minor amendments in the cultivation method requires optimization , we believe that the developed method is applicable not only in Lam Dong province but also in some other provinces in Vietnam.

Keywords: *Panax vietnamensis*, Vietnamese ginseng, cultivation

PPL14

Agmatine prevents mitochondrial dysfunction in 3-nitropropionic acid-induced experimental Huntington's Disease

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Background
Huntington's disease (HD) is an inherited genetic disorder, caused by the mutation of abnormally expanded and unstable CAG repeat within the coding region of the huntingtin protein gene. At the molecular level, mitochondrial dysfunction plays a significant role in the pathogenesis of HD. 3-nitropropionic acid (3-NP) is a neurotoxin which induces neurodegeneration in the animal model of Huntington's disease (HD). It is an irreversible inhibitor of mitochondrial complex II (SDH) enzyme of the electron transport chain. Agmatine is the metabolite of arginine by arginine decarboxylase and has been suggested to be a neuroprotective agent. The objective of this study was to investigate the protective effect of agmatine on 3-NP-induced neurodegeneration through the estimation of mitochondrial enzymatic profile in Wistar rats.
Methods
The experimental protocol design includes systemic 3-NP (10 mg/kg, i.p.) treatment thrice, i.e. on day 1, 5 and 9. Agmatine (40 and 80 mg/kg) was also given i.p. daily, from day 9 to day 15.
Results
Enzymatic levels in mitochondrial complexes-I, II, III and IV were found to be significantly lowered in the brain of rats treated with 3-NP. Mitochondrial SDH contributes to cell viability reduction, hence, the decrease of cell viability approves irreversible inhibition of SDH by 3-NP. The level of enzymes of all complexes in groups treated with agmatine (40 and 80 mg/kg) was significantly increased.
Conclusion
The present study provides evidence that agmatine exerts protective action over 3-NP-induced neurodegeneration by preventing mitochondrial dysfunction and thus, may be potentially used as a neuroprotective agent.

Keywords: Huntington's disease, 3-NP, agmatine, mitochondrial dysfunction

PPL15

Towards the discovery of novel dengue NS3 antiviral drug: Application of proteochemometric (PCM) modelling and *in vitro* validation in drug repurposing

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Background
The current treatment for DENV infection is only supportive care involving fluid replacement, analgesics and bed rest. Dengvaxia®, a DENV vaccine was recently approved by FDA but its usage is age-limited and only for patients with confirmed previous dengue infection. Antivirals for DENV infection that can reducethe risk of severe cases of patients from any background is crucial. Hence this study aims to screen currently available drugs (a process known as drug repurposing) for potential antiviral activity that targets the NS3 protease of DENV through proteochemometric (PCM) modeling and subsequent *in vitro* validation.

Methods
The PCM model was built on a training set which comprises of 62,746 bioactivity data from ten serine proteases available from public databases. Aitchison-Aitken kernel and sequence identity were used to calculate chemical and biological similarity respectively while Parzen-Rosenblatt Window was used as the classification algorithm. The performance of the model was validated to measure the accuracy of the prediction model. Drugs from the SWEETLEAD database were then screened for potential activity against NS3 protease using the validated model and further tested *in vitro* for their ability to inhibit DENV activity. Molecular docking was performed to model the interaction between drugs and NS3 protease.

Results
The performance of the model was validated internally (goodness of fit RMSE = 0.315, predictive ability Q^2 = 0.567) and externally (RMSE = 0.466, and Q^2 = -1.509). The screening showed that Zileuton and Trimethadione have the potential as antiviral with good binding affinity at the active sites. The *in vitro* assay further validated that Trimethadione possess better anti-DENV activity with 80% foci reduction when tested at 20 mM drug concentration.

Conclusions
Drug repurposing through PCM modelling is a promising technique to accelerate the discovery of novel dengue antiviral drug.

Keywords: Dengue virus, antiviral drug, proteochemometric (PCM) modelling, drug repurposing

PPL16

Application of molecular networking technique to isolate selaginellin derivatives from *Selaginella tamariscina*

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Background
Selaginellins, unique pigments found in the genus *Selaginella*, were reported as potent phosphodiesterase-4 (PDE4) inhibitors in recent studies.
Method
To isolate diverse natural selaginellin derivatives, we applied a MS/MS based molecular networking strategy enhanced by *in silico* structural annotation to the *Selaginella tamariscina* extracts. It led to the prioritization of chromatographic peaks predicted as previously unknown selaginellin derivatives.
Results and conclusion
As a result, we could isolate ten previously unknown compounds containing two unusual selaginellin analogs with 1*H*,3*H*-dibenzo[*de,h*]isochromene skeleton named selariscins A (**1**) and B (**2**) along with eight diarylfluorene derivatives, selaginpulvilins M−T (**3**−**10**). The absolute configurations were elucidated by computational electronic circular dichroism (ECD) spectral calculations. Some isolates showed PDE4 inhibitory activity with IC_{50} values in the range of 2.8−33.8 μM, and their binding modes were suggested using a molecular docking study.

PPL17

Amplification of purine rich site from COL4A3 gene for triple helix study in keratoconus eye disease

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Background

Keratoconus (KC) eye disease is a non-inflammatory disorder characterized by eye bulging due to corneal thinning and results in blurred vision and astigmatism. Several factors lead to KC development include genetic factor and polymorphism of COL4A3. Triple helix is DNA structure where third single strand of DNA fragment bind to the purine rich site of DNA duplex in reverse Hoogsteen hydrogen bonds. This triplex structure is enable to suppress gene expression by inhibiting the initiation of transcription. The objective of the study was to identify and amplify the purine rich site in COL4A3 gene.

Methods

The desired purine rich site of COL4A3 gene was amplified using designed PCR primers based on sequence from NCBI (NG_011591.1). The PCR steps were repeated for 30 cycles by using 54.5 ^{o}C annealing temperature (Tm). The amplicon then subjected for 1% agarose gel electrophoresis for DNA separation and observed under UV light through ethidium bromide staining before advancing for sequencing.

Results

The PCR product bands with size of 429 nucleotides were successfully observed. Based on the sequencing analysis, 88.8% of amplicon aligned with original sequence from NCBI and there was one base deletion from the amplicon. This shows that the purine rich region of COL4A3 gene was successfully amplified.

Conclusions

Results demonstrated that the purine rich region of COL4A3 gene was successfully identified and amplified and it can be used as triple helix forming oligonucleotides binding site.

Keywords: keratoconus eye disease, COL4A3 gene, triple helix

PPL18

Photodamage attenuating effects of marine endophytic fungus (MV) fractions against fibroblast cell line

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Background
Over exposure to sunlight increased UVB radiation, lead to potential photoaging and skin cancer. As these trends are likely to continue for the foreseeable future, the adverse effect of UVB has become a major human health concern.

Methods
Marine endophytic fungus isolated from red seaweed, *Gracilaria arcuata* Zanardini (MV) collected from Port Dickson, Negeri Sembilan, Malaysia was investigated for its potential in attenuating the photodamage effects of UVB against fibroblast (CRL 2522) cell line by MTT assay.

Results
The aim of this study was to investigate the potential of marine endophytic fungus (MV) fractions in stimulating DNA repair of CRL 2522 cells against UVB-induced DNA damage. About 13 of MV fractions showed increased of CRL 2522 cell viability (70-80%) after 30 min exposure to UVB radiation. Five of fractions (MV14, MV35, MV41, MV45 and MV50) significantly increased (p<0.05) cell viability. These data suggest a greater potential of marine endophytic fungus (MV) fractions in stimulating DNA-repair against UVB-induced damaging cells. Further study of these 13 active fractions should be carried out to determine the photodamage attenuating effects of MV fractions against fibroblasts and Hacat cell lines.

Conclusions
These five potential MV fractions might be useful as a starting point for developing dermatological products to prevent oxidative skin damage.

Keywords: seaweed, marine endophytic fungi, photodamage, UVB

PPC1

The study of effect of method and time of extraction on antioxidant, total phenolic content and total flavonoid content of F*icus deltoidea*

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Background
Ficus deltoidea is a well-known medicinal plant which has long been used by the Malay community in treating various health problems. Due to its antioxidant property and the presence of phenolics and flavonoids, the plant contributes the various biological activities. These properties vary significantly with different extraction methods and time. This study aimed to evaluate the effect of method and time of extraction on antioxidant, total phenolic content (TPC) and total flavonoid content (TFC) of *Ficus deltoidea.*
Methods
Different extraction methods like continuous shaking extraction (CSE) with time (30,160 and 360 min), ultrasonic extraction (USE) with time (5, 15 and 30 min) and microwave assisted extraction (MAE) with time (1, 3 and 5 min) were applied to see the effect on total antioxidant activity, TPC and TFC quantitatively. The antioxidant and total phenolic and total flavonoid content of extracts were evaluated by DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging activity, Folin-Ciocalteu and aluminum chloride ($AlCl_3$) tests, respectively.
Results
The microwave extraction method provided good extractive yield, superior scavenging activity and higher yield of TPC and TFC compared to the other two methods used. The outcome of this experiment also indicates that TPC and TFC increases as the increase in extraction time in each different methods of extraction.
Conclusions
MAE showed good results even in shorter time of extraction may be due to the rapid heating mechanism of microwave. The hot solvent produced in MAE penetrated easily into the matrix and extract compounds from the lysed plant cells. Therefore, the MAE method is more efficient in extracting phenolic and flavonoid compounds and showed better antioxidant effect compared to USE and CSE methods.

Keywords: *Ficus deltoidea*, antioxidant, total phenolic content, total flavonoid content, microwave assisted extraction

PPC2

Investigation of some metals in underground part of the *Adenophora stenanthiana (Ledeb) Kitag)*

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Background
Adenophora stenanthiana (Ledeb) Kitag, family of Campanulaceae, is widely grown in China. It is documented in the Chinese Medical Encyclopedia to be beneficial for gout, rheumatism, leprosy, some bacterial infections and cancer. The part grown under ground contained glycosides, terpenes, and small amount of alkaloids. To determine the safety of the raw material of *Adenophora stenanthiana* (Ledeb) Kitag, the content of some heavy metals were determined by atomic absorption spectrometer.

Methods
Eight samples of *Adenophora stenanthiana* were collected from different geographical regions of China, according to the Chinese pharmacopoeia. Heavy metals such as lead, cadmium and copper were measured in the samples with standard atomic absorption spectrometric method (CP- A/59).

Results
The heavy metal content of the 8 samples were determined. According to the Medicinal Plasma Standard (GAPS) the permissible content were as follow: (Cu ≤ 20.0 μg /g, Pb ≤ 5.0 μg/g, Cd ≤ 0.3 μg /g). Sample 6 and 8 were relatively high in lead with 18.85 μg/g and 41.92 μg/g, respectively, However, both samples did not exceed the acceptable concentration for cadmium and copper.

Conclusions
Adenophora stenanthiana grown in different geographical regions presented with different concentrations of heavy metals.

Keywords
Adenophora stenanthiana, copper, cadmium, lead, atomic absorption spectrometer

PPC2

New phenalenone aspergillussanone C and D from tubers of *Pinellia ternate* derived *Aspergillus sp*.

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Background
Phenalenones are a unique class of natural products with three-ring systems of hydroxyl-perinaphthenones, which possess diverse and significant biological activities, such as antimicrobial, antimalarial, cytotoxical, and anti-HIV activities. Fungal metabolites are important sources of phenalenone with various structures originated from heptaketides and hexaketides.
Methods
Optical rotations were measured on a JASCO P-1020 polarimeter.IR spectra were measured on a Bruker Tensor 27 spectrometer using KBr discs. ECD spectra were recorded on a JASCO J-810 spectrometer. The NMR spectra were obtained on a BrukerAvanceIII NMR instrument (^{1}H: 500 MHz, ^{13}C: 125 MHz) at 300 K with tetramethylsilane (TMS) as internal standard, using solvent signals (CD_3OD: δ_H 3.31 and δ_C 49.15) as references. ESI-MS data were acquired on an Agilent 1100 Series LC/MSD ion-trap mass spectrometer and HRESIMS data were recorded on an Agilent 6520B UPLC-Q-TOF mass spectrometer. Preparative HPLC was conducted on a Shimadzu LC-10A series instrument. All the solvents used were of analytical grade.
Results
Aspergillussanone C was isolated as a pale yellow gum. HRESIMS ion at *m*/*z* 647.2826 $[M+Na]^+$ revealed the molecular formula of $C_{35}H_{44}O_{10}$, which shows fourteen indices of hydrogen deficiency. Its NMR spectra gave the characteristic signals of phenalenone moiety, including one aromatic proton of a penta substituted benzene (δ_H 6.72, s, 1H), two ketone carbonyls (δ_C 200.8 and 203.6), two quaternary methyl linked to benzene (δ_H 2.17 and 2.87, 3H each), and ten aromatic carbon signals (δ_C 103.3, 107.5, 108.5, 114.7, 119.1, 137.3, 151.4, 165.3, 165.6, and 167.4). Aspergillussanone D (**2**), $C_{35}H_{44}O_8$, was also established to have the same phenalenone and similar diterpenoid moieties by NMR data analysis.

Conclusions
In conclusion, aspergillussanones C-D, two new phenalenones with diverse frameworks, were obtained from tubers of *Pinellia ternate* derived *Aspergillus sp*. Two isolates determined the same phenalenone skeleton (C-1~ C-9′) but they are unusual acyclic diterpenoid adducts, which are diversely oxidized and partly epoxidized to form different heterocycles (C-9′~C-18′).

Keywords: Phenalenones, *Aspergillus sp*., Antibacterial activities

PPC4

An Efficient Synthesis of Fluoro-neplanocin A Analogs Using Electrophilic Fluorination and Palladium-catalyzed Dehydrosilylation

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Neplanocin A analogues constitute a valuable class of carbocyclic nucleosides in biological and medicinal chemistry studies due to their potent and broad-spectrum antiviral and antitumor activities]. A close relation has been identified between antiviral activity of neplanocin A and inhibition of cellular *S*-adenosylhomocysteine (SAH) hydrolase. Therefore, SAH hydrolase has been recognized as potential pharmacological target for the development of new antiviral agents. Presently, there are very few synthetic methods available for the synthesis of key fluoro-cyclopentenyl moiety and also these methodologies had limitations such as number of steps, harsh reaction conditions, and difficulty in separation of products. Hence, in order to further explore neplanocin A analogs potential as therapeutic agents, an alternative and efficient synthesis of fluoro-neplanocin A analogs has been developed by employing stereoselective electrophilic fluorination and palladium-catalyzed dehydrosilylation as key steps. In comparison to previous syntheses, the present synthetic methodology provides easy access to key intermediates that could contribute to expanding further the structure–activity relationship studies of neplanocin A analogs.

Keywords: Fluoro-neplanocin A nalogs, *S*-adenosylhomocysteine (SAH) hydrolase, Electrophilic Fluorination and Palladium-catalyzed Dehydrosilylation.

PPC5

Analysis of methanolic extract of *Aloe vera* by reverse phase high performance liquid chromatography (RP-HPLC)

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Background

Aloe vera is also known as *Aloe barbadensis* Miller, a plant that has been used for the purpose of medication and cure for health conditions.The *Aloe vera* extract was studied by using reversed phase high performance liquid chromatographic (RP-HPLC) method.

Methods

The RP-HPLC system included a Model 1100 pump supplied with a multi solvent delivery system, an Agilent C18 (5 µm, 4.6 x 250 mm) column and a photodiode array detector.The solvent consisted of acetonitrile (CH_3CN) and water (0.01% formic acid). It was set up to run in a gradient elution as follows: 0 min, 10:90; 3 min, 10:90; 30 min, 90:10; 35 min, 90:10; 36 min, 10:90; and 45 min, 10:90. The flow rate was set as 1 mL/min (temperature of the column = 25^oC) and the UV absorbances were measured at λ = 210, 254 and 280 nm. The peaks in the chromatograms were recorded and reviewed. A triplicate trial was performed for each sample volume = 10 µL, per injection.

Results

The compounds with the highest absorbance values were eluted within nine minutes, whereby the solvent ratio was 30:70 (CH_3CN:H_2O). It is suggested that aloe emodin was separated much earlier, at retention time, R_T = 1.676 minutes. Later, the anthrone *C*-glycosides [aloin A (barbaloin) and aloin B (isobarbaloin) could be eluted, respectively at R_T = 8.171 and 8.721 minutes.

Conclusions

The *Aloe* compounds could be identified by comparing their retention times with the monograph. Some unresolved, minor peaks, that were not well isolated (R_T = 2.2 and 8.3 minutes) could be attributed to the less polar metabolites of aloins, for example, the aloe emodin anthraquinone and rhein. The RP-HPLC technique appears to be adequate for routine analysis of the *Aloe* extract.

Keywords: *Aloe*, chromatography, extraction, separation

PPC6

Antiglycation and antioxidant potential of novel imidazo[4,5-b]pyridine benzohydrazones

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Background
Glycation occurs due to reaction which occurs slowly between reducing sugars and free amino acids. The reaction process which is also known as Maillard reaction involves a series of complex non-enzymatic reactions. An intermediate known as Amadori product is produced in early stages of glycation process. Further, rearrangement and reduction of this intermediate resulted in the production of various advanced glycation endproducts (AGEs). Glycation, which occurs in the body could modify biomolecules such as lens crystallins, collagen, hemoglobin (Hb), albumin, immunoglobulins, LDL and DNA that leads to impaired protein function. This can contribute to microvascular disease which slows down wound healing for diabetic patients. Thus, agents having antioxidative effect against production of Amadori product and capable of inhibiting the production of AGEs would potentially be used to treat patients with age-related diseases and diabetes.
Methods
Synthesis of novel imidazo[4,5-b]pyridine benzohydrazones **5** and its 30 derivatives were carried out. NMR experiments were performed on UltraShield Bruker FT NMR 500 MHz; CHN analysis was performed on a Carlo Erba Strumentazion-Mod-1106, Italy. Electron impact mass spectra (EI-MS), Germany. Chromatograms were visualized by UV at 254 and 365 nm. The antioxidant capacities were measured by 1,1- diphenyl-2-picrylhydrazil (DPPH) and ferric reducing antioxidant power (FRAP) assays. The percent inhibition of AGE formation was also measured.
Results
The result for antiglycation shows that IC_{50} values of the derivatives ranges between 140.16 and 420.12 µM .Among the compounds which show potent activity are compounds 5, 6, 8, 10, 11, 14, 15, 25 and 33. The results of synthesized compounds on DPPH antioxidant indicated that compounds 10, 11, 14, 15, 25, 31, 33 and 34 exhibited better radical scavenging ability compared to gallic acid (EC_{50} = 40.0 ± 0.12 µM). Compound 25 which displayed the best antiglycation activity was among the compound which showed potent radical scavenging activity with an EC_{50} value of 26.12 ± 0.15 µM. It was found that compounds having more than one hydroxyl group have higher ability to scavenge radical.
Conclusion
In conclusion, the synthesized imidazo[4,5-b]pyridine benzohydrazones derivatives having hydroxyl group are showing potent antiglycation and antioxidative activity. The studies also showed that the ability of some compounds to inhibit glycation is indirectly dependent upon their antioxidative capabilities.

Keywords: Antioxidative potential, antiglycation, benzohydrazide.

PPC7

Phytochemical investigation of *Combretum indicum* leaves extracts

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Background

Combretum indicum is also known as Akar Dani or Rangoon Creeper and can be found throughout Asia and tropical Africa. *C. indicum* is a plant species belonging to the *Combretum* genus and the family of Combretaceae. Numerous studies proved the therapeutic effects of this plant including anti-obesity, anti-inflammatory, antioxidant, insecticidal, antimicrobial, cytotoxic and immunomodulatory properties. The previous phytochemical studies of *Combretum indicum* has revealed the presence of tetracyclic triterpenes, trigonelline (alkaloid), rutin (flavonoid), tannins, L-proline (α-amino acid), L-asparagine (α-amino acid) and quisqualic acid. In addition, isoenzyme A and isoenzyme B (Enzyme), the two forms of the cysteine synthase are also present in *C. indicum*.

Methods

The chemical constituents of leaves of *C. indicum* were extracted using organic solvents. The TLC profile of chloroform extract of *C. indicum* was established and chemical constituents were purified by PTLC.

Results

Two long chain fatty acids derivatives were successfully isolated from the crude chloroform extract and the structures were confirmed by using NMR analysis.

Conclusions

The phytochemical study on Malaysian *C. indicum* confirmed the presence of terpenes in chloroform extract.

Keywords: *Combretum indicum,* Combretaceae*,* Phytochemical study

PPC8

Analysis of saponins in Vietnamese ginseng cultivated in Lam Dong Province, Vietnam by HPLC-PDA/CAD

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Background
Panax vietnamens is, namely Vietnamese ginseng (VG), was discovered under the canopy of Ngoc Linh mountain in Kon Tum and Quang Nam provinces in 1973. Since then, VG had been believed to be an herbaceous perennial plant native to this area only until recently it was acclimatized to Lam Dong province and cultivated successfully. In this comparative study, we analyzed the saponin composition in both VG cultivated in Lam Dong (VG-LD) and VG cultivated in Ngoc Linh area (VG-NL).
Methods
Saponins in the underground part of VG-LD from 2-4 years old were analyzed in comparison with with those of VG-NL. Separation, qualitative and quantitative analysis of twelve main VG saponins including N-R1, M-R1, G-Rg1, G-Re, M-R2, P-RT4, V-R11, V-R2, G-Rh1, G-Rb1, G-Rc, and G-Rd were obtained by HPLC coupled with diode array (PDA) and charge aerosol (CAD) detectors.
Results
VG-LD not only yielded the same chemical composition but also exhibited the considerably higher total saponin content than that of VG-NL at all ages. For instance, total saponin contents of 2-4 years old VG-LD roots, on average, were 9.95%, 11.73%, and 12.84%, respectively, whereas those of VG-NL, on average, were 2.91%, 4.18%, and 10.31%, respectively. Similarly, total saponin contents of 2-4 years old VG-LD rhizomes, on average, were 11.39%, 17.21%, and 14.96%, respectively, whereas those of VG-NL were 3.94%, 5.77%, and 9.59%, respectively.
Conclusions
The result indicates that, regarding the saponin composition, the cultivation of VG in Lam Dong province is successful and, therefore, deserves the support from both central and local governments to nurture and develop the achievement. Further comparative study on the saponin composition of VG-NL and VG-LD at different ages is now in progress to observe the accumulation of saponins over years.

Keywords: Vietnemese ginseng, Panax vietnamensis, HPLC-PDA/CAD, analysis of saponin composition

PPC9

Development and validation of a HPLC method for the determination of hippadine in the bulbs of *Crinum latifolium* L.

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Background
Hippadine is a biologically active alkaloid isolated from *Crinum latifolium* L. It has been shown to decrease the heart rate and blood pressure due to α_1 and β_1 adrenoceptor inhibition.
Methods
An isocratic HPLC method was developed to determine hippadine in the bulbs of *Crinum latifolium* L. The chromatographic separation was achieved using a mobile phase consisted of acetonitrile – phosphoric acid pH 3 (46:54 v/v) on a C_{18} column (100 x 4.6 mm, 3.5 µm) and detection was carried out at 299 nm. The injection volume was 10 µL, the flow rate was 1 mL/min and column temperature was set at 30 °C. The method was validated with respect to system suitability, specificity, linearity, accuracy and precision.
Results
The content of hippadine in the bulbs of *Crinum latifolium* L. collected from Binh Dinh province (Viet Nam) was found as 315.8 mcg/g (0.0316%). The method was precised with an intra-day RSD = 0.6% and inter-day RSD = 1.16%. The detector's response was linear (R^2> 0.999) and reliable for hippadine quantitation from 1 up to 20 ppm. Through recovery studies, accuracy of the method was averagely estimated to be 98.06 – 99.65%.
Conclusions
The HPLC method was proved to determine hippadine in the bulbs of *Crinum latifolium* L. with sufficient accuracy and precision.

Keywords: HPLC, hippadine, *Crinum latifolium* L.

PPC10

Synthesis and antibacterial activity of some new chlorobenzothiazole derivatives

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Background

Benzothiazole derivatives are well known as antibacterial agents. In the present study, a series of novel amides containing chlorobenzothiazole were synthesized, characterized and evaluated for their antibacterial properties.

Methods

In this research, we synthesized some new 2-acetamido-chlorobenzothiazole derivatives from 2-chloroanilin and 2,4,5-trichloroanilin through four reactions. The evaluation of the synthesized compounds for antibacterial activities were carried out by using agar diffusion method.

Results

Seven 2-acetamido-chlorobenzothiazole derivatives were obtained. All of the newly synthesized compounds were characterized by melting point, thin layer chromatography, structural elucidation by UV, IR, ^{1}H-NMR, ^{13}C-NMR and MS. This research also presents the result of the investigation antibacterial activities of the 2-acetamido-clorobenzothiazole derivatives on the *Escherichia coli* ATCC 25922, *Staphylococcus aureus* (MRSA) ATCC 43300, *Pseudomonas aeruginosa* ATCC 27853; *Streptococcus faecalis* ATCC 29212. Tests on biological activity showed that 5 of the synthesized derivatives (3b, 3c, 4g, 4h, 4r) were effective against two referenced strains of bacteria. 4g and 4r showed higher antibacterial effect against the MRSA (MIC_{4g} = MIC_{4r} = 16 mg/ml).

Conclusions

We have discovered some new chlorobenzothiazole derivatives and bioassay results showed that some of these synthesized derivatives displayed medium antibacterial activities against various bacterial species. These results are the basis for synthesis of new antimicrobial drugs which can be suitable for this current.

Keywords: Benzothiazole, 2-chloroanilin, 2,4,5-trichloroanilin, Antibacterial activity.

PPC11

Development and validation of HPLC bioanalytical method for the determination of tinidazole in rabbit serum

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Background
A simple, rapid, sensitive and isocratic RP-HPLC-UV method was developed and validated according to the FDA guidelines for the quantification of tinidazole (TD) in rabbit serum.
Methods
Sample preparation was accomplished through protein precipitation method and metronidazole (MD) was used as an internal standard. Following the extraction procedure, the chromatographic separation was carried out on a ZORBAX Eclipse XDB-C18 (4.6 × 150 mm, 5 µm). The mobile phase mixture consisting of 0.005 M phosphate buffer (82%), acetonitrile (9%) and methanol (9%) was used at a flow rate of 1mL/min. The detection wavelength was set at 317 nm.
Results
The calibration curve was linear over a concentration range of 0.5 - 100 µg/ml (r^2 = 0.9991) with a limit of quantification, 0.5 µg/ml. No interference was observed at the retention times of analyte (6.55 mins) and internal standard (3.73 mins). The intra-day and inter-day precision and accuracy results were in between 1.47 and 7.83% and 1.23 and 5.46%, respectively. The mean recoveries of TD and MD were above 94%. Serum samples containing analyte were found to be stable at -70°C for 30 days.
Conclusion
The developed and validated HPLC-UV method is simple, rapid and specific, and it can be used for the pharmacokinetic sample analysis of tinidazole in rabbit serum.

Keywords: Tinidazole, HPLC-UV, rabbit serum, protein precipitation and validation

PPC13

Study and establish of process of synthesis some 2-hydrazinylthiazolopyridine derivatives

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Background
The derivatives contain thiazolopyridine ring show that many potential biological activities have been reported in literature as antitumoral activity, anticonvulsant, cytotoxicity, antibacterial activity and antifungal.Therefore, the aim of this study was to establish an effective process for synthesis some thiazolopyridine derivatives.
Methods
Similar benzothiazole ring, thiazolopyridine heterocycle can be synthesized with various methods such as Hugerchoff's cyclization reaction with bromine agent or Jacobsen reaction with $K_3[Fe(CN)_6]$ agent. In this research, we synthesized 2-hydrazinylthiazolopyridine derivatives by thiazolopyridine cyclization reaction in dimethyl sufoxide with sodium methoxide agent.
Results
The derivatives containing thiazolopyridine ring are synthesized through 5 stages. A compound N-(pyridylcarbamothioyl)benzamide was obtained from the reaction between 4-methyl-2-chloro-3-aminopyridine and benzoyl isothiocyanate. N-(pyridylcarbamothioyl) benzamide was cyclized by CH_3ONa agent in DMSO to form the structure of thiazolopyridine. Hydrolyzing benzamide in sulfuric acid 70% agent lead to formation of 2-aminothiazolopyridine. Then condensate the obtained derivative with hydrazine sulfate to obtain 2-hydrazinylthiazolopyridine. Finally reacted with different aldehydes and obtained 6 new derivatives. All of the synthesized compounds were characterized by melting point, thin layer chromatography, structural elucidation by UV, IR, 1H-NMR, 13C-NMR and MS.
Conclusions
This study have established procedure to synthesize 2-hydrazinylthiazolopyridine derivatives from the initial material 4-methyl-2-chloro-3-aminopyridine with good performance. The intermediate and final derivatives are precisely defined chemical structures with the expected formula. Therefore, we propose to synthesize thiazolopyridine derivatives according to the process that has been investigated and tested for biological activity to receive compounds with good biological activity.

Keywords: Thiazolopyridine, 2-hydrazinylthiazolopyridine, thiazole

PPC14

Determination of amygdalin in "Xuefu Zhuyu" capsules by HPLC - PDA method for use in the drug quality control

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Background
Xuefu Zhuyu is a famous traditional Chinese medicine widely used in the treatment of cardiovascular diseases such as thrombosis, angina pectoris, heart attack, strokes. Xuefu Zhuyu comprises of 11 herbs: *Semen Persicae, Flos Carthami tinctorii, Radix Angelicae sinensis, Rhizoma Ligustici wallichii, Radix Paeoniae, Radix Bupleuri, Radix Rehmanniae glutinosae, Fructus Aurantii, Radix Platycodi grandiflori, Radix Achyranthis bidentatae, Radix Glycyrrhizae.* The determination of amygdalin, a bioactive component in Xuefu Zhuyu capsules was developed by using liquid chromatography with photo diode array detector (HPLC - PDA). This HPLC method was validated and applied to the quality assessment of Xuefu Zhuyu capsules.
Methods
The sample preparation method and the chromatographic conditions were optimized to quantify amygdalin in the Xuefu Zhuyu capsules by HPLC. The optimization was obtained when the peak area of amygdalin in the chromatogram of sample solution was maximum and chromatographic parameters met requirements such as theoretical plate number (N > 5000), resolution (Rs > 1.5), asymmetry (0.8 - 1.5) and peak purity (purity factor > 999.000). This method was subsequently validated according to the ICH guideline Q2 (R1) (ICH 2005) with respect to system suitability, specificity, linearity, repeatability, intermediate precision, accuracy and range of analytical procedures.
Results
The optimal sample preparation method was found. The powder of Xuefu Zhuyu was extracted with methanol in ultrasonic bath for 15 min. The chromatographic conditions were as follows: Mobile phase methanol - water (21.5 : 78.5), column Phenomenex Gemini C_{18} (250 × 4.6 mm; 5 μm), column temperature 25 ^{o}C, photo diode array detector set at 210 nm, flow rate 1.0 mL/min, injective volume 20 μL. The developed method showed system suitability, specificity, linearity within 2.4 - 48.0 μg/mL ($\hat{y}$ = 18.104x, R^2 = 1), repeatability (RSD = 0.3%), intermediate precision (RSD = 0.6%), accuracy with recovery rate 98.1 - 101.1% and the range 9.6 - 38.4 μg/mL.
Conclusions
In the present study, a simple, accurate and reliable analytical method for determination of amygdalin in Xuefu Zhuyu capsules was developed by using HPLC - PDA. The result of this study would be helpful to build the quality control standard of Xuefu Zhuyu capsules.

Keywords: Amygdalin, Xuefu Zhuyu, determination, traditional Chinese medicine, HPLC.

PPR1

Perception of problem-based learning among academic staff of Faculty of Pharmacy, UiTM

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Background
Problem-based learning is an approach that is focused on self-directed learning and small group discussion whereupon students work through a given case to acquire knowledge. It is a learning strategy that is commonly adopted in higher education institutions, including pharmacy schools throughout the world. Perception, whether good or bad, can have an impact on the effectiveness of PBL implementation. This study aimed to determine the perception of PBL among academic staff at the Faculty of Pharmacy, UiTM Puncak Alam Campus.

Methods
Cross sectional study was conducted from March to May 2018. Data was collected through a 27 item, self-administered questionnaire. Descriptive analyses were performed using frequency counts, percentages, means and standard deviations.

Results
A total of 74 questionnaires were distributed to academic staff involved in the Bachelor of Pharmacy (B. Pharm) curriculum, with a 56.94% response rate. The majority of respondents agree that PBL is an effective learning strategy, with several advantages, among others, a more thorough knowledge gain and enhancement of public speaking skills.

Conclusions
PBL is viewed by the academic staff as an effective teaching and learning approach. Nonetheless, it is important to ensure careful planning of PBL and adequate training of faculty members to ensure its successful implementation in the UiTM B.Pharm programme.

Keywords: Perception, Problem-based learning, Teaching-learning, Pharmacy

PPR2

Preliminary study on medicinal plants used in the treatment of arthritis among medicinal plant practitioners in Kampot Province, Cambodia

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Background

This research is based on an ethnobotanical investigation which focused on medicinal plants used to treat arthritis by traditional healers as well as local people in Kampot province. According to the WHO, about 80% of the world population including Cambodia uses medicinal plants for treatment since the ancient time. This survey was conducted on the uses of medicinal plants for arthritic treatment. Tis study was aimed to document all of this indigenous knowledge to for sustainability and improvement of usage as arthritis was seen commonly occurring in the society of Cambodia.

Methods

The data collection was conducted in Kampot province among traditional healers and local people. Five traditional healers and 45 local people who are also medicinal plant practitioners responded to the following the semi-structured interviews.

Results

Twenty eight medicinal plants were listed with information on local, scientific and family name, plant parts used, mode of preparation and administration. Leea rubra Blume., *Achyranthes aspera* Linn., *Morus alba* Linn., and *Zingiber officinale* Rose. were the most identified and mentioned by various sources (book, international papers, survey). Leaves were the most common to use for the treatment of arthritis, which represents 20% among the other plant parts. The frequent preparation method and administration was drying and decoction, taken orally which represented 64% of all methods used.

Conclusions

Throughout this research, it illustrates the diversity of plants which have been used among traditional healers and local people who were mentioned differently in therapeutic practices. Interestingly, 5 medicinal plants have been identified and *Leea rubra* Blume. was considered as one of the most potential plants which should be focused for further investigation.

Keywords: Ethnobotanical investigation, traditional healers, indigenous knowledge

PPR3

Exploration of pharmacology facilitators' satisfaction level in problem-based learning at Pharmacy School

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Background

Problem-based learning (PBL) is a constructivist teaching-learning method by which students are to construct idea based on their existing knowledge. Facilitators on the other hand plays a less active role by facilitating students learning. Because of this, the characteristics and skills of effective facilitators have received relatively more attention. Therefore, this study was conducted to investigate facilitators' satisfaction of PBL and to determine correlation between the facilitators' position and their satisfaction level on PBL.

Methods

Subjects of this study were lecturers who have been involved in facilitating PBL in Pharmacology subjects. A total of 14 subjects were asked to assess their satisfaction on PBL using a self-administered questionnaire. The questionnaire consisted of 21 items group as seven factors related to student's role, tutor's role, designated problems, environment of classroom, allotted time, evaluation process and overall satisfaction.

Results

The result indicate that facilitators were moderately satisfied with the PBL method. Interestingly, their position i.e. senior lecturer or professor determines their satisfaction level.

Conclusions

In conclusion, the role of the facilitator is of pivotal importance, providing students with proper guidance during the PBL process. As such, a PBL training program is desirable to prepare tutors for facilitation of PBL.

Keywords: satisfaction, problem-based learning, facilitator, Pharmacology, Pharmacy

PPR4

A case study research prevalence of Alcohol Consumption among women in Phnom Penh Capital City and Kampong Cham Province 2018

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Background
Alcoholic drinks have been a part of the community life and up until now societies have always found it difficult to understand or restrain their use. The aim of this study was to find out the prevalence of alcohol consumption among women in community of Phnom Penh and Kampong Cham.
Methods
The review was conducted at two sites (Kampong Cham and Phnom Penh). These questionnaires were pretested and proved to be well understood by responders. Data analyses are achieved using SPSS version 18. Results was counted in number (n) and percentage (%).
Results
The study reveals that among the 384 respondents, prevalence of consuming alcohol among women was 65.4%. About 69.3% and 61.5% of women in Phnom Penh and Kampong Cham, respectively consumed alcohol beverages. The factors that prompted these women to take alcohol were due to the influence of society and family members, self-medication, and during postpartum.
Conclusion
In conclusion, there are more women in Phnom Penh that consumed alcohol than women who lives in Kampong Cham.

Keyword: Alcohol, Prevalence, Women, Cambodia

PPR5
Multidisciplinary education approach to optimize phosphate control among hemodialysis patients (MEPS-HD)

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Background
Hyperphosphatemia is a common consequence in end stage renal disease. It is associated with increased cardiovascular risk and mortality, also development of hyperparathyroidism and mineral bone disease. A patient educational program (PEP) involving physician, pharmacist and dietician was developed to manage hyperphosphatemia among hemodialysis patients.
Methods
This was a non-randomized, single-arm community trial running for a period of 6 months. The PEP consisted of a small group seminar and individual counseling sessions. Two individual counseling sessions were conducted for each patient, focusing on diet and medication adherence, by an accredited dietician and pharmacist respectively. The group seminar was delivered by a multidisciplinary team involving a physician, pharmacist and dietician. Topics included basic knowledge of hyperphosphatemia, phosphate binder and dietary phosphate control. Eligible and consented patients had knowledge and medication adherence assessment, measurement of pre-dialysis serum calcium, albumin, phosphate, haemoglobin and alkaline phosphatase before and after the PEP.
Results
Fifty seven patients completed the PEP and were included into final data analysis. The median (IQR) phosphate level (mmol/L) was 1.86 (1.45-2.24) before and decreased to 1.47 (1.211.91) and 1.49 (1.28-1.81) 3 months and 6 months after PEP ($p<0.001$). The percentage of patients with uncontrolled phosphate level was reduced from 59.3% to 35.6% and 42.1% after the PEP ($p=0.003$). The mean knowledge score almost doubled after the intervention, with a mean pre-score of 8.61 (95% CI 7.85-9.37) to mean post-score of 15.31 (95% CI 14.85-15.76). The adherence to phosphate binder also improved from 17.2% to 41.4% after PEP ($p=0.007$).
Conclusions
Multidisciplinary PEP is an effective approach to manage hyperphosphatemia among hemodialysis patients.

Keywords: Education, hemodialysis, mineral bone disease, multidisciplinary, phosphate

PPR6

A preliminary study on ethnobotanical survey of medicinal plants used by traditional healers to treat toothaches

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Background
Medicinal plants have potential in treating different kinds of diseases since many centuries. They have been the primary solution to care for people's oral health due to the inexpensive costs and potentials. The aim of this study was to document the types of medicinal plants and therapeutic methods used by traditional healers to relieve toothaches and also the use of these plants to produce an effective modern medicine.

Methods
The study was conducted from 25th September to 30th October in 2017 using questionnaires, following the WHO guideline. The information was collected from three key traditional healers, who are from National Center of Traditional Medicine and Faculty of Pharmacy, University of Health Sciences, due to their vast knowledge on medicinal plants.

Results
A total 37 medicinal plants were identified for the use in treating toothaches, which were collected from three references. Among these plants, 3 medicinal plants (*Spilanthes acmella*, *Syzygium aromaticum* and *Piper lolot*) were commonly used for the toothache treatment. This study shows that leaves and barks were the most frequently used parts of the plants, followed by resin, flower, root and stem. These plants were applied directly to the infected area of the tooth in different ways such as decoction, maceration, pounding or chewing.

Conclusions
This study shows that the diversity of each plants has different effect as remedies to treat toothaches. Three medicinal plants have been recognised as potential cure. Moreover, leaves were the most common plants part used which were generally prepared through decoction.

Keywords: Toothache, Medicinal plant, Traditional healer, Ethnobotany

PPR7

A case study research of self-medication as a daily living behavior of people in community of Kampong Cham province and Phnom Penh capital

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Background
According to WHO (2000), self-medication is the use of medicinal products to treat self-recognized disorder or symptoms. There are several circumstances arising from self-medication, including irrational use of drug, mistreatment and risk of drug abuse. However, no research has addressed the extent of self-medication practice in local community of Cambodia.
Methods
A quantitative cross-sectional study was conducted with a total of 312 samples, 156 samples in Phnom Penh and 156 in Kampong Cham, were selected randomly among patients who purchased medicines from pharmacies. The study used questionnaires as the study tool for face-to-face semi-direct interview. Data analysis is achieved through using SPSS version 18. Results were presented as count (n) and percentage (%).
Results
In Phnom Penh, 91% of 156 respondents, self-medicate, where the prevalence rate in Kampong Cham was 70.2% of 156 respondents. Cost efficiency was the main reason of self-medication in both areas. The most common illness was flu (15.3% of all reported cases) and the most prescribed medicine was analgesics (24.9%). Respondents from both areas prefer pharmacy as the first choice for healthcare service. Practice of patient counselling was very low in Phnom Penh (1.3%) compared to Kampong Cham (79.5%).
Conclusions
Prevalence rate of self-medication was higher in Phnom Penh compared to Kampong Cham (22.52% difference). Although pharmacy is the first choice of healthcare service, awareness should be raised as serious illnesses require proper diagnosis and treatment. Dependency may develop due to the misuse of analgesics. Good pharmacy practice should be applied to ensure safety, efficacy and efficiency use of medication.

Keywords: Self-medication; Pharmacy practice; Dependency

PPR8

Cross-sectional study of fever management of Influenza among undergraduate student in Phnom Penh, Cambodia: Preliminary result in 2018

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Background
In low income country, fever is common among adolescents and adults seeking for healthcare. Fever is one of the symptoms of influenza (flu) which happened every year in Cambodia. This research aims to expose the experiences and attitudes of undergraduate students from different universities on fever and flu medication and care.
Methods A cross-sectional survey of 453 undergraduate students was conducted in Phnom Penh using semi-structured questionnaire which was prepared and validated. Epidata was used to insert data before analyzed by STATA version 12.
Results
A total number of 453 undergraduate students from 30 universities in Phnom Penh successfully consented and completed the questionnaire. Alternative methods were mostly used to confirm the body temperature; however, 61.22% used thermometer. 58.72% of participants relied on both medication and selfcare while flu medicines (55.06%) was commonly used following by unknown medication, antipyretic and antibiotic that were used for less than 3 days (45.19%). Besides, resting (68.56%), hot bath, wiping, home remedies, coining and exercise were preferable practices for selfcare. Although the respondents preferred both methods, they felt more confident to use medication than self-care.
Conclusions
Medication and self-care are widely practiced among undergraduate students to manage fever and flu. Education about fever management relying on medication and self-care should be promoted in Cambodia.

Keywords: Cross-sectional study, Fever management, Undergraduate students