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INSIGHTS INTO THE HALAL STATUS OF RESPIRATORY, IMMUNOLOGICAL PRODUCTS AND VACCINES IN MALAYSIA

A. R. SURAIYA[1]*, W. R. W. ROSALINA[1], M. T. A. RASHIDI[1], M. I. S. N. AMIRAH[1], A. MOHAMED[1], B. AMRAHI[2], B. A HALIM[3], A. M ADLAN[4], S. SYAZFEEZA[4], I. ZHARI[5]

[1]Faculty of Pharmacy, Department of Pharmaceutical Technology and Industry, Cyberjaya University College of Medical Sciences, Malaysia, [2]Dept of Pharmacy, University Malaya Medical Centre, Malaysia, [3]Pharmacy Branch, Health Services Division, Malaysian Armed Forces HQ, [4]Dept of Pharmacy, Tuanku Mizan Armed Forces Hospital, Malaysia, [5]School of Pharmaceutical Sciences, Department of Pharmaceutical Chemistry, University of Science Malaysia, Penang, Malaysia
Email: suraiya@cybermed.edu.my

ABSTRACT

Malaysia has launched the world's first Halal pharmaceutical standards known as MS 2424:2012. The biggest challenge faced by pharmaceutical companies to comply is determining ingredient origins and halal status. This exploratory study aims to determine the halal status of respiratory, immunological products and vaccines by analyzing the origin of their active pharmaceutical ingredients (API) and excipients in a series of evaluations on medicines in formularies from two Malaysian government hospital divided by pharmacological categories. Products were assessed from information obtained in product package leaflets, manufacturer and standard pharmaceutical references. The ingredients were categorized as halal, musbooh or haram based upon their content and definition set in the MS 2424:2012. There were 123 medications included in the study where 91 of them were respiratory products while 32 were immunological products and vaccines. For the respiratory products, 54.9% (50) were halal, 45.1% (41) mushbooh. From the 49 API in respiratory products, 79.6% (39) were halal and 20.4% (10) were mushbooh. Similarly, from 124 excipients used, 83.1% (103) were halal and 16.9% (21) were mushbooh. Under the immunological products and vaccines, from 32 products, 96.9% (31) were mushbooh and 3.1% (1) were haram. From the 42 API under this product category, 100% were mushbooh. From the 52 excipients, 57.7% (30) were halal, 40.4% (21) mushbooh and 1.9% (1) haram. Lactose, magnesium stearate, ethanol, polysorbate 80 and amino acids are the most common musbooh ingredients. Respiratory products showed a higher percentage of halal excipients (83.1%). This study shows that most excipients are halal, but medicines became mushbooh due to the presence of certain excipients. This insight should encourage pharmaceutical manufacturers to go for halal certification to meet the demand for halal medications.

Keywords: Halal Pharmaceuticals, Haram, Musbooh, MS 2424.

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MYHALALRX: HALAL MEDICATION INFORMATION AT YOUR FINGERTIPS

N. ABD AZIZ[1]*, A. N. ABDULLAH[1], N. MOH WAHI[1], H. I. MOKTAR[1], M. B. C. M. SANI[1], N. I. MOHD SAID[1], N. S. YAHAYA[2]

[1]Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Department of Pharmacy, Hospital Sungai Buloh, Selangor, Malaysia
Email: drnoorizan2909@puncakalam.uitm.edu.my

ABSTRACT

Healthcare professionals and the public are very concerned about the halal status of medication. Presently there is no direct information on halal prescribed medication available across the world. Thus, "MyHalalRx" is the first point of reference on halal medication, a user-friendly app that provides information on the Halal status of medication. The objectives of this program are to provide accessible information at fingertips and to educate the public and health care providers on halal concepts from medication perspective. The "MyHalalRx" will have a search engine that consists of the database on the halal status of medication. With the search engine, they can just key in the brand name or the generic name of the medication to obtain the halal status of the product. Apart from that, this application will also provide brief information on the halal concept and also the guideline on how the products are evaluated. This application will also be equipped with bookmark toolbars, latest halal medication news, and a list of companies (national and international) that manufacture halal medication and also hadith and Quran verse on halal topics. Furthermore, the application will also provide other information regarding the medication such as the indication, active ingredients, excipients, and dosage. In summary, "MyHalalRx" will improve the quality of life and provide shariah compliance to the world community.

Keywords: Halal status, Medications, Active ingredient, Excipients, Apps

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PRELIMINARY SURVEY ON THE AWARENESS OF HALAL FOOD IN JAPAN

Y. SANNOMARU[1]*, J. S. YAGAMBRA[2], L. Y. MIN[2], M. FUKAYA[1], S. NUMAJIRI[1], A. TERAO[1]

[1]Faculty of Pharmaceutical Sciences, Josai University, Japan, [2]Faculty of Health and Life Science, Management & Science University, Malaysia

Email: sanchan3@josai.ac.jp

ABSTRACT

Islamic dietary guidelines require food to have a halal status. Japanese awareness of Halal is limited. Approximately 100,000 Muslims live in Japan, and, in 2020, many Muslims will visit the Olympics. However, most Japanese do not have the knowledge to prepare Halal food for Muslim visitors or residents, or even the awareness that such dietary rules are important. The current study was to explore the current level of Japanese awareness of Halal foods and their preparation requirements and examine the availability of Halal food. In 2014, a 4-part questionnaire was completed by 660 students at Josai University: 1) Characteristics of the subjects; 2) Subjects' understanding of Halal food; 3) Availability of Halal foods in Japan; 4) Awareness of Japan's policy that Halal food should be available so Muslim residents and tourists feel comfortable and respected. 88% of all participants knew that Muslims do not eat certain foods on religious grounds. Only 29% of the subjects had heard the phrase "Halal food". Though 82% of subjects knew that pork is prohibited, only about 20% of the subjects knew that alcohol is prohibited. 53% answered that there is no need for Halal food to be reasonably available. Though almost all subjects knew that the Olympic Games will be held in 2020, only about 21% knew about the Tourism Nation Promotion Basic Law (Halal food being important for promoting tourism). These results suggest that Japanese know little about Halal food and are unaware of domestic policy supportive of it. As the world is moving toward a global society, the Japanese government should take the initiative and provide more information to Japanese about the importance of understanding the customs of others.

Keywords: Halal food, Preliminary survey, Awareness, Questionnaire, Muslim tourists in Japan.

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THE IMPORTANCE OF HALAL STATUS ON VITAMINS AND SUPPLEMENTS AMONG CONSUMERS IN ULU KELANG, SELANGOR, MALAYSIA

J. ASLINDA[1], M. K., NUR ATIQAH[1], W. Z. W. SAZRINA[1]

[1]Faculty of Pharmacy, Department of Community and Pharmacy Practice, Cyberjaya University College of Medical Sciences, Malaysia
Email: aslinda@cybermed.edu.my

ABSTRACT

Nowadays, consumers in Malaysia preferred to buy products that are certified as 'Halal,' which are allowed and permitted by the Islamic Law. The Halal logo convinces consumers on the Halal ingredients used in the manufacturing of the products as well as the processes of preparing the products itself. This study aims to evaluate the differences between age, religion and level of education towards the importance of Halal status on vitamins and supplements among consumers in Ulu Kelang, Selangor. Association between knowledge and the importance of Halal status on vitamins and supplements among consumers in Ulu Kelang, Selangor were also determined. A set of questionnaire was conveniently distributed to 383 consumers in Ulu Kelang, Selangor. One-way analysis of variance (ANOVA) with post-hoc analysis was used to analyze the data to answer the research questions pertaining to the differences between age, religion and level of education towards the importance of Halal status on vitamins and supplements among consumers. There were significant differences between age ($p = 0.008$), religion ($p = 0.001$) and level of education ($p = 0.001$) towards the importance of Halal status on vitamins and supplements among consumers in Ulu Kelang, Selangor. Pearson's chi-squared test were also used to determine the relationship between consumers' knowledge on the Halal status in Malaysia and found out that there was no significant relationship between the knowledge of Halal status in Malaysia ($p>0.05$) and the importance of Halal status on vitamins and supplements. In conclusion, consumers' age, education level and religion significantly influence the importance on the Halal status on vitamins and supplements. Most of the respondents in Ulu Kelang, Selangor showed good knowledge on the status of Halal, however, knowledge was not significantly correlated to the importance of Halal status on vitamins and supplements.

Keywords: Halal pharmaceuticals, Vitamins and supplements.

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EXPLORING HALAL STATUS OF ENDOCRINE, OBSTETRICS AND GYNAECOLOGY GROUP OF MEDICATIONS AND EVALUATION OF AWARENESS AND KNOWLEDGE REGARDING HALAL PHARMACEUTICALS AMONG PUBLIC IN CYBERJAYA

A. R. SURAIYA[1], W. R. W. ROSALINA[1], M. T. A. RASHIDI[1]*, Z. Z. ANITH[1], A. MOHAMED[1], B. AMRAHI[2], B. A. HALIM[3], A. M. ADLAN[4], S. SYAZFEEZA[4], I. ZHARI[5]

[1]Faculty of Pharmacy, Department of Pharmaceutical Technology and Industry, Cyberjaya University College of Medical Sciences, Selangor, Malaysia, [2]Dept of Pharmacy, Pusat Perubatan Universiti Malaya, Malaysia, [3]Pharmacy Branch, Health Services Division, Malaysian Armed Forces HQ, Ministry of Defence, Malaysia, [4]Dept of Pharmacy, Hospital Angkatan Tentera Tuanku Mizan, Malaysia, [5]School of Pharmaceutical Sciences, Department of Pharmaceutical Chemistry, University of Science Malaysia, Penang, Malaysia
Email: rashidi@cybermed.edu.my

ABSTRACT

The development and availability of halal medication lies behind that of food products. This descriptive study aimed to determine the halal status of selected endocrine, obstetrics and gynaecology medications available in the formulary of University Malaya Medical Centre (UMMC) and Tuanku Mizan Military Hospital (TMMH). Sources of active ingredients and excipients for each product were assessed for Halal status based on available information obtained from product information leaflets or manufacturers. The products were categorized into Halal, Mushbooh, Haram and Unknown. The proportions of Halal, Mushbooh, Haram and Unknown products in UMMC was 17.2%, 20.2%, 10.1% and 52.5% respectively. Mean while, the percentage of Halal, Mushbooh, Haram and Unknown products in TMMH was 9.6%, 18.1%, 13.3% and 59.0% respectively. In this study, all the medications that categorized as Haram were due to their active pharmaceutical ingredient were from porcine or urine source. Besides that, this study also attempts to assess the awareness and knowledge of halal pharmaceuticals among the public in Cyberjaya. This was achieved using a cross-sectional study. Data was collected by distributing questionnaires to the public in housing and public areas in Cyberjaya. The study was conducted on a total of 156 respondents to assess the relationship of awareness and knowledge on halal pharmaceuticals with religion. Results revealed that public had fair level of awareness and knowledge towards halal pharmaceuticals. The association between religions and awareness level proven to be significant with p value<0.001. However, the association between religion and knowledge level showed no significant with p=0.08. Religion did give an impact on public's awareness level towards halal pharmaceuticals but not on the knowledge level.

Keywords: Halal pharmaceuticals, MS2424, Endocrine, Obstetrics, Gynaecology

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SURVEY REGARDING THE DEGREE OF SUCCESS IN ADAPTING KUESERIMUKA, AN EXAMPLE OF TRADITIONAL MALAYSIAN CUISINE, IN JAPAN

M. FUKAYA[1]*, L. Y. MIN[2], J. S. YAGAMBRA[2], Y. SANNOMARU[1]

[1]Faculty of Pharmaceutical Science, Josai University, Japan, [2]Faculty of Health and Life Science, Management & Science University, Malaysia

Email: fukaya@josai.ac.jp

ABSTRACT

Because of the value placed in Japan on hospitality, "Omotenashi", many Japanese are looking ahead to the 2020 Tokyo Olympics. Showing respect for other cultures and seeking to understand one another is extremely important. We anticipate that many tourists will come to Japan, including Muslim visitors from Malaysia. So, we created a version of Kueserimuka, a traditional Malaysian sweet, using Edamame (which are green soybeans, a traditional Japanese food). Our hope was to create the traditional cuisine of another country using traditional Japanese ingredients as a way of showing osmote nashi. We tried to make Kueserimuka with Edamame instead of Pandan leaf since Pandan is not easy to obtain in Japan. The subjects were 46 Japanese female students of Josai University. We conducted a survey of preferences regarding 2 types of Kueserimuka, traditional and one adapted to local ingredients, using a crossover study design. Using the hedonic scale, we assessed taste/flavor, texture, aroma, color, and overall appeal. The results of this survey included that the aroma and color of the traditional type were rated significantly higher than the Japanese type. These results are likely because Pandan leaf harmonizes well with coconut milk. However, the texture of the Japanese version was preferred by the Japanese students over the traditional one. Substituting Edamame improved the perceived palatability. No significant difference in the taste, flavor and overall appeal was observed. 30 subjects preferred the traditional Kueserimuka over the Japanese type though more development of the recipe may improve its appeal. These results suggest that even when some traditional ingredients from other cuisines are unavailable, we can adapt the recipes to work with local ingredients. Therefore, we can still provide foods desired and required by Muslims and guests such as Malaysians to feel comfortable and welcomed in Japan.

Keywords: Omotenashi, Kueserimuka, Muslim, Malaysian cuisine, Japanese ingredients

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EFFECTS OF AGE, HYDRATION LEVEL AND COSMETIC TREATMENT ON SKIN MECHANICAL PROPERTIES IN THAIS

A. TANSIRIKONGKOL*, D. CHARNVANICH, V. PANAPISAL, W. SUWAKUL

Department of Pharmaceutics and Industrial Pharmacy, Faculty of Pharmaceutical Science, Chulalongkorn University, Thailand
Email: nyarporn.t@chula.ac.th

ABSTRACT

Age and skin hydration influences the skin's mechanical properties. The use of cosmetic product usually improves skin hydration, but the correlation between the changes in skin hydration and mechanical properties is uncertain. This study aimed to investigate the relationship between ages, skin hydration and skin mechanical parameters. The correlation between changes in skin hydration and skin mechanical parameters after cosmetic treatment was also evaluated. A total of 123 healthy volunteers, aged 18-55, were divided into three similar age groups. Skin hydration and biomechanical properties were evaluated using Corneometer® and Cutometer®, respectively. All results were analysed with the Pearson correlation. Subsequently, 61 healthy women aged 30-55 were chosen for the second study. Skin hydration and biomechanical parameters were measured after eight weeks topical application with microemulsion or nanoemulsion. The changes in each parameter and their correlation were evaluated. The result revealed that skin capacitance correlated to some mechanical parameters only in young volunteers (<30 y old). Only gross elasticity (R2) presented strong negative correlation with age in 30-40 y old volunteers while all skin elasticity parameters (R2, R5 and R7) showed significant negative correlations with age in 41-55 y old volunteers. Application of microemulsion or nanoemulsion significantly improved skin hydration. However, increases in skin capacitance by microemulsion were correlated with the changes in skin distensibility (R0) but not with other elasticity parameters. The relationship between changes in R0 and in R2 showed a significant correlation in both treatments. The skin elasticity decreased with age and changes in gross elasticity (R2) was first observed. As the age increased, R5 and R7 showed the stronger negative relationship. Skin hydration and elasticity were not related in any age group. The increase in skin hydration by topical treatment did not always affect skin elasticity. Improvements in skin hydration and elasticity by cosmetic product are independent and are based on individual formulation.

Keywords: Age, Skin elasticity, Hydration, *In vivo*, Efficacy

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FACTORS INFLUENCING CONSUMER AWARENESS TOWARDS COSMETIC CONTAMINATION

N. MOHD HASSAN*, S. N. MA'ON, N. Z. ABD HAMID, N. I. S. ABDUL AZIZ

Faculty of Business Management, Center for Management and Administrative Studies, Universiti Teknologi MARA, Puncak Alam Campus, Malaysia
Email: naffi885@puncakalam.uitm.edu.my

ABSTRACT

A global increase in the demand for cosmetics requires awareness of safety-related issues. Consumers are generally unaware of the potential harm cosmetics may cause and their ability to induce infections. The objective of this study is to investigate the consumer's awareness about harmful substances in their cosmetics and to identify factors that affect consumer awareness based on the Health Belief Model. This cross-sectional study is carried out in Klang Valley area. The respondents were selected through purposive sampling, and 500 questionnaires were distributed among female candidates and 431 were returned and used for data analysis. The remaining 69 were deemed unsuitable for use in this study. Pearson Product Moment Correlation Coefficient and descriptive analysis were used to analyse data. The findings suggested that cosmetic consumers are aware of the types of ingredients used in cosmetics. However, there is uncertainty over the harmful substances in their cosmetics products. The study also highlighted that consumer behavior and consumer knowledge are important factors that influence awareness towards cosmetic contamination. The findings from this study are critical to helping increase the level of awareness among cosmetics consumers.

Keywords: Awareness, Cosmetic Contaminations, Consumer, Consumer knowledge.

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DERMAL EXPOSURE ASSESSMENT AMONGST FARMERS THROUGH EXPOSURE WITH IMAZAPIC IN SURFACE WATER

A. Z. MAZLAN[1]*, H. HUSSAIN[1], M. A. MOHAMED ZAWAWI[2], M. SAMENI[1]

[1]Faculty of Health Sciences, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Faculty of Engineering, Universiti Putra Malaysia, Serdang, Selangor, Malaysia
Email: aniszakiah89@yahoo.com

ABSTRACT

The pesticide is a well-known artificially synthesized substance used in agriculture practice as an effective way to increase the quantity and quality of food production by controlling pests, plague and weeds, all of which can be harmful to crops and reduces productivity. In Tanjung Karang, a new paddy strain was introduced to avoid weedy rice problem which required farmers to concurrently using its herbicides called On Duty TMWG (containing Imazapic and Imazapyr). This study was aim to estimate the risk posed by farmers through water contact exposure to compound Imazapic using dermal exposure assessment (DEA). For this purpose, water samples were taken at 6 points to gain the mean concentration of Imazapic in their water surface area to be used in the calculation for DEA. Samples were then analysed using High-Performance Liquid Chromatography-UV (HPLC-UV). As for DEA, questionnaires were distributed among 40 farmers. Among them, three of them were not using pesticide that contains Imazapic thus only 37 farmers would be taken into estimating the risk posed by them who are exposed to Imazapic compound. Through questionnaire and water sample analysis, Dermal absorbed dose (DAD) value was obtained before hazard quotient (HQ) was calculated where HQ>1 would indicate risk. DEA was done based on the exposure of farmers through certain body surface that are arms, hands, legs and feet. Based on the risk calculated, it was found that none of the farmers had a value of HQ>1 which indicate lower risk that could harm the farmers. Although the HQ for all body parts estimated does not exceed the HQ risk indicator, but it is still a concern towards the risk of Imazapic related health problem that might harm the farmers through prolonged usage of this substance.

Keywords: Dermal exposure assessment, Farmers, Imazapic, Hazard quotient, Hazard index.

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SKIN IMPROVEMENT USING LABISIA PUMILA AND FICUS DELTOIDEA COSMETIC FORMULATIONS

R. HASHAM*, N. MOHAMED NOOR, N. A. ROSLI, S. N. SYED HASSAN, N. A. AZELAN, N. JAMALUDDIN, A. ISHAK, N. ANUAR, A. A. AZIZ, R. AZIZ, M. R. SARMIDI

Institute of Bioproduct Development (IBD), Universiti Teknologi Malaysia, Johor Bahru, Johor, Malaysia
Email: rosnani@ibd.utm.my

ABSTRACT

In the tropics, continuous exposure to ultraviolet (UV) light is known to cause photoageing. UV generated reactive oxygen species can lead to collagen deficiency and eventually skin wrinkling. Previous *in vitro* studies have shown that Labisia pumila (LP) and Ficus deltoidea (FD) could protect fibroblast from UVB irradiation damage. Hence, the aim of this study was to determine the effectiveness of the cosmetic formulation based on LP and FD extracts in reducing the effect of skin ageing due to exposure to UVB irradiation. The evaluation of the active serum and placebo was carried out on 78 volunteers for two months and conducted using non-invasive bioengineering methods including melanin index, erythema index, skin moisture content, skin sebum content and skin surface pH measurements. It was found that the cutaneous hydration was increased after 8 w application of the cosmetic formulation. This is highly related to a decrease of restoration of normal skin surface pH. In addition, the formulation application also caused a significant decrease in skin melanin, erythema content and casual sebum level after 8 w of application. In conclusion, the formulations prepared from LP and FD was able to exhibit anti-ageing effects. However, further studies on LP and FD based formulations must be carried out in the tropic region such as Asia and Middle East for future natural regenerative anti-ageing cosmetic market.

Keywords: Labisia pumila, Ficus deltoidea, Cosmetic formulation

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DEVELOPMENT OF TRANSDERMAL DOSAGE FORM USING CO-PROCESSED EXCIPIENT OF XANTHAN GUM AND CROSSLINKED AMYLOSE

S. SURINI*, A. SICILIA, R. SOFIANI, S. KHOIRIAH, U. H. INDRIATIN, S. P. SARI, Y. HARAHAP

Faculty of Pharmacy, Universitas Indonesia, Depok, Indonesia
Email: silvia@farmasi.ui.ac.id

ABSTRACT

Transdermal hydrogel is a dosage form consisting of three-dimensional polymer network that is able to bind water or biological fluids in large quantities and is intended for transdermal delivery systems. In the previous study, Co-processed of Xanthan Gum and 6-Crosslinked Amylose (Co-CLA6-XG), Co-processed of Xanthan Gum and 12-Crosslinked Amylose (Co-CLA12-XG), 6-crosslinked from co-processed amylose-xanthan gum (CL6-Co-A-XG),12-crosslinked from co-processed amylose-xanthan gum (CL12-Co-A-XG) excipients were proven to have characteristics as hydrogel matrix based on sufficient gel strength, viscosity, and swelling index. The aims of this study were to formulate those excipients as a matrix and to evaluate its ability on delivering drug substances by *in vitro* and *in vivo* penetration test. *In vitro* penetration test through rat skin used Franz Diffusion Cell and *in vivo* penetration tests were performed on six Sprague-Dawley strain male rats for each formulation. *In vitro* flux of sodium diclofenac from Co-CLA6-XG, Co-CLA12-XG, CL6-Co-A-XG, and CL12-Co-A-XG transdermal hydrogels were 655.23±216.43; 655.23±216.43; 867.43±101.27 and 736.98±15.39 µg. cm-2. h-1. The value of AUC data's from *in vivo* were 32.08±5.40; 34,27±8,34; 6,20±2,90 and 14,38±2,38 µg h ml-1. Those matrices also show longer release times (shown in t1/2 and MRT value). According to the results, those excipients have potential to be developed as matrices in transdermal system.

Keywords: Crosslinked amylose, Co-processed amylose-xanthan gum, Transdermal hydrogel, *In-vitro* penetration test, *In-vivo* penetration test

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PORCINE DNA DETECTION ON COMMON TABLET FORMULATIONS AND ITS EXCIPIENTS

S. N. S. SYED SABERI[1]*, N. A. K. KHAN[1], S. MOHAMAD[1], L. C. WUEI[2]

[1]Pharmaceutical Technology, School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang, Malaysia, [2]Human Architecture Technologies, Penang, Malaysia
Email: syarifahnursyakira@gmail.com

ABSTRACT

Market demand of halal pharmaceutical products has increased due to a heightened awareness among Muslim consumers towards the availability halal products. Current halal verification methods for supplement and nutraceutical products are through on-site production and paper audits. A more definitive halal verification method would improve the quality of products and consumer confidence. With DNA verification technology, porcine content in excipients and final product can be detected by performing quantitative polymerase chain reaction (qPCR) test. In this study, tablets were chosen as they are the most commonly prescribed pharmaceutical dosage form. Three types of tablet formulation were selected; uncoated, coated and matrix type. Porcine DNA detection was assessed in each excipient used in formulating the three types of tablet and during the tableting process. Porcine gelatin was used as a spiking agent in each excipient and as a binder in positive controls for the tablets. It was found that porcine DNA was still able to retrieved and detected after the drying process for uncoated tablet production. During DNA extraction of coating excipients, increasing the pH value of the excipients to pH 9 after 1 h of incubation with Proteinase K can weaken the adsorption of DNA on kaolin in the tablet coating excipient. In a positive control matrix tablet, porcine DNA was able to be detected even with the presence of high viscosity hydroxypropyl methylcellulose (HPMC) as a matrix polymer in the formulation. Thus, when certain extraction techniques are adhered to porcine DNA detection using qPCR testing methods can be applied to detect porcine content in tablets and excipients.

Keywords: Porcine DNA detection, qPCR, Ucoated tablet, Coated tablet, Matrix tablet

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MICROEMULSION-BASED MODIFIED NANOCRYSTALLINE CELLULOSE FOR TRANSDERMAL DELIVERY OF CURCUMIN: FORMULATION AND PHYSICAL CHARACTERISATION

N. ZAINUDDIN, I. AHMAD, S. RAMLI*

School of Chemistry and Food Technology, Faculty of Science and Technology, Universiti Kebangsaan Malaysia, Bangi, Selangor, Malaysia
Email: su_ramli@ukm.edu.my

ABSTRACT

There are carriers that are widely known for sustained or controlled release and targeted delivery such as microemulsion and natural biopolymer cellulose. Curcumin, a hydrophobic polyphenol compound contain in turmeric (Curcuma Longa) was used in this study. Curcumin is known for its medicinal properties, but its usage has been limited due to poor bioavailability. The aim of this study is to formulate microemulsion-based modified nanocrystalline cellulose for controlled release and improved bioavailability of curcumin via the transdermal route of administration. The nanoscale nanocrystalline cellulose (NCC) was extracted from kenaf bast fiber using hydrolysis process with sulfuric acid. The prepared NCC was modified with a cationic surfactant of cetil trimethylammonium bromide (CTAB) to introduce hydrophobic surfaces for the binding of this hydrophobic drug. Microemulsion-based modified CTAB-NCC was formulated as a carrier of curcumin and the physical properties of the system were investigated. The system was developed using oleic acid/limonene, Cremophor RH 40 and Transcutol as oil phase, surfactant and co surfactant respectively. The pseudo-ternary phase diagrams were constructed along water dilution line and the optimum ratio of surfactant: cosurfactant was 2:1 and oleic acid: limonene was1:1. The isotropic properties of the microemulsion phases were confirmed by Polarized Light Microscopy. The effects of the addition of curcumin in both systems with-and without modified CTAB-NCC were measured at various temperatures of 4, 25 and 37 °C for 6 mo. Most of the system shows good stability and no significant changes of clarity and phase separation at 4 and 25 °C up to 6 mo. Formation of precipitation of curcumin occurred for the formulation with high water content (>60% wt. of water) after four month under high temperature due to its low water solubility properties. The average particle size of the systems was measured by Particle Size Analyzer (PSA). All systems showed small particle size which was below 200 nm. The phase transitions of the system were determined by electrical conductivity and viscosity tests. Both microemulsion-based CTAB-NCC with-and without curcumin did not show a significant difference. The phase transitions of water-in-oil to a bicontinuous phase occurred at 20% wt. of water, and a bicontinuous to oil-in-water phase at 50% wt. of water.

Keywords: Microemulsion, Nanocrystalline cellulose, Curcumin, Transdermal

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EFFECT OF PECTIN HYDROGEL ON PARTIAL THICKNESS WOUND HEALING

N. K. AZIZ[1], N. K. ANUAR[1,2]*, W. T. WUI[1,2], A. S. HAMZAH[3], W. R. WAN ENGAH[4]

[1]Particle Design Research Group, [2]Non-Destructive Biomedical and Pharmaceutical Research Centre, iPROMISE, Faculty of Pharmacy, [3]Institute of Science (IOS), Universiti Teknologi MARA, Shah Alam Campus, Selangor, Malaysia, [4]Malaysian Agricultural Research and Development Institute (MARDI), Serdang, Selangor, Malaysia

Email: al_zahra55@yahoo.com

ABSTRACT

The present work aims to investigate the effect of pectin hydrogel on the process of wound healing in rat models, following partial thickness thermal injury. The pectin hydrogel was prepared using solvent evaporation technique with glutaraldehyde as the crosslinking agent and glycerol as the plasticizer. The ability of pectin hydrogel to accelerate the wound healing process was examined by the macroscopic images, wound size reduction and histological evaluation for 14 d. The results indicated that the rats treated with pectin hydrogel had significantly smaller wound size when compared to the control group on day 7 onwards. In addition, the histological study revealed that the wounds receiving pectin hydrogel had more fibroblasts, blood vessels and collagen bundles on day 14 when compared to the control rats. The pectin hydrogel has shown promising potential to accelerate the wound healing process in partial thickness thermal burn wound.

Keywords: Hydrogel, Pectin, Wound healing

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FORMULATION AND PHYSICAL STABILITY TEST OF PHYTOSOME GEL CONTAINING ETHYL ACETATE FRACTION OF NOTHOPANAX SCUTELLARIUM LEAVES

RISSYELLY*, A. MUNIM, M. JUFRI, A. RAHMAWATI

Faculty of Pharmacy, Universitas Indonesia, Depok, Indonesia
Email: rissyelly@yahoo.com

ABSTRACT

Mangkokan leaves (Nothopanax scutellarium Merr.) contain flavonoids which have hair growth activity. Phytosome gels are carrier system which contains bioactive phyto-constituents of a plant extract surrounded and bonded by lipids. The aim of this research is to formulate and characterize the physical stability of phytosome gels containing ethyl acetate fraction of Nothopanax scutellarium with various concentrations, of 1%, 0.5% and 0.25% and to obtain the phytosome characteristics formed by the thin layer hydration method. Phytosome formulation of ethyl acetate fraction and phosphatidylcholines with a ratio of 1:2 gave an entrapment efficiency value of 74.34% with particle size distribution value of 335.4 nm, polidispersity index of 0.252 and zeta potential value of-3.50 mV. Subsequently physical stability test was performed on the phytosome gel for 8 w with accelerated stability tests. The results indicate that gels with 0.5% phytosome concentration proved to be the most stable at various storage temperatures.

Keywords: Phytosome, Flavonoid, Fraction, Gel, Nothopanax scutellarium, Physical stability

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DRUG DISTRIBUTION STUDY OF FITC-LABELLED LEVODOPA-LOADED CHITOSAN NANOPARTICLES BY INTRANASAL DELIVERY

A. F. MOHD[1], M. Z. AHMAD[1], M. H. MOHD JAAFAR[1], N. ABD LATIP[2], K. ABDUL HAMID[1]*

[1]Pharmaceutical Instrumentation Technology Laboratory, [2]Department of Pharmacology and Chemistry, Faculty of Pharmacy, Universiti Technology MARA, Puncak Alam Campus, Selangor
Email: khuriah@puncakalam.uitm.edu.my

ABSTRACT

Levodopa or L-dopa (L-3,4-dihydroxyphenylalanine) is a drug used to control symptoms of Parkinson's caused by low levels of dopamine in certain parts of the brain. L-dopa is the precursor to the catecholamine neurotransmitters that play a key role in body movement. A formulation of FITC-labeled levodopa was successfully developed and delivered via the intranasal delivery to bypass first-pass metabolism, to enhance absorption. FITC-labelled levodopa-loaded chitosan nanoparticles were prepared by ionic gelation technique. Dosage of 2.5 mg/kg of unprocessed levodopa (control) and 3.8 mg/kg of FITC-labelled levodopa-loaded chitosan nanoparticles were administered through intranasal route in rats. After 4 h of administration, five major organs including heart, lung, liver, kidney and brain were excised, weighed, homogenised and deproteinated by acetonitrile prior to UPLC analysis. The concentration of FITC-Labelled Levodopa-loaded chitosan nanoparticles was significantly higher ($P<0.05$) in the brain (149.35µg/l±10.1) as compared to lung (53.51µg/l±2.8). For the unprocessed levodopa, the concentration in brain was slightly higher (79.64µg/l±10.6) compared to lung region (61.86µg/l±4.8). The concentration of both FITC-labelled levodopa-loaded chitosan nanoparticles and unprocessed levodopa were not detected in the heart, liver and kidney regions. Concentrations of levodopa were found in the brain and lung regions but no traces in other organs. The concentration of levodopa was significantly higher in the brain compared to the lungs suggesting that more levodopa was delivered into the brain with slight permeation into the lung region. These findings propose the potential of FITC-labelled levodopa-loaded chitosan nanoparticles to be targeted and delivered to brain via intranasal delivery.

Keywords: Levodopa nanoparticles, Intranasal delivery, Parkinson's disease, Drug distribution

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FORMULATION OF LABISIA PUMILA EXTRACT AND TOXICITY STUDIES ON SPRAGUE DAWLEY RATS

M. A. A. SAEED[1], A. H. MEMON[1], M. S. R. HAMIL[1], H. K. BEH[1], Z. ISMAIL[1]*, A. SADIKUN[1], G. K. CHATAR SINGH[2]

[1]Department of Pharmaceutical Chemistry, [2]Institute for Research in Molecular Medicine, Universiti Sains Malaysia, Penang, Malaysia
Email: zhari@usm.my

ABSTRACT

Labisia pumila is a popular herb from the family Myrsinaceae, locally known as Kacip Fatimah. Traditionally it has been used to induce and facilitate childbirth and for dysentery, rheumatism and gonorrhoea. The present study aimed to prepare a nano liposome formulation of Labisia pumila extract and to study acute and repeated dose toxicity with orally administered Labisia pumila extract (LPE) and Labisia pumila-liposome (LPL). Labisia pumila extract was prepared as a nano liposome in soybean phospholipids in order to improve its solubility and permeability. Labisia pumila liposome was prepared by the conventional film method, and LPE and LPL were characterized using Fourier transform infrared spectroscopy (FTIR), transmission electron microscopy (TEM) and particle size analyser. In acute toxicity test the rats were orally treated with LPE and LPL at dose of 2000 mg/kg, and general behaviour, adverse effects, and mortality were recorded for up to 14 d. In repeated dose toxicity study, rats received LPE and LPL by oral gavage at the doses of 250, 500, and 1000 mg/kg/day for 28 d, and biochemical, hematological, and histopathological changes were determined. LPE and LPL did not produce any hazardous symptoms or death in the acute toxicity test. Repeated dose toxicity study with LPE and LPL did not show any change in body weight, food and water consumption, hematological, biochemical and histopathological profiles. In conclusion, our toxicity results showed that LPE and LPL to be safe for human use.

Keywords: Labisia pumila extract, Labisia pumila-liposome, Acute toxicity, Repeated dose toxicity

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EFFECT OF PRESSURE ON SOLID OIL PARTICLES SIZE OF GINGER RHIZOME USING RAPID EXPANSION SUPERCRITICAL SOLUTION (RESS) CARBON DIOXIDE

N. A. ZAINUDDIN[1]*, I. NORHUDA[1], I. S. ADEIB[3], A. N. MUSTAPA[1], S. H. SARIJO[2]

[1]Faculty of Chemical Engineering, [2]Faculty of Applied Science, Universiti Teknologi MARA (UiTM), Shah Alam, Malaysia, [2]Faculty of Chemical Engineering, UiTM, Johor, Malaysia
Email: nurainzainuddin@gmail.com

ABSTRACT

Recently, RESS method has been used by researchers to produce fine particles for pharmaceutical drug substances. Since RESS technology acknowledges a lot of benefits compared to conventional method for ginger extraction, it is suggested as a method of choice to explore particle formation of bioactive compounds from powdered ginger. The objectives of this research are to produce direct solid oil particles formation from grind ginger rhizome which contains valuable compounds using RESS-CO2 process and to investigate the effect of different extraction pressures to solid oil particle size that are produced at constant extraction temperature and flow rate. RESS experiments were carried out at various extraction temperatures of 40, 45, 50, 55, 60, 65 and 70 °C respectively for 40 min extraction time and at a constant flow rate of 24 ml/min. From the conducted studies, it was shown that extraction pressure is an important factor that can affect particle size. By increasing the extraction pressure, average particle size can be increased or decreased. It was found that the size of the precipitated solid oil particle obtained was smaller than the original particle size of ginger. In conclusion from this study, the smallest particle size of 2.22µm was obtained at an extraction pressure and temperature of 5000 psi and 40 °C respectively.

Keywords: Ginger, Particle size, RESS, Supercritical Carbon Dioxide, Pressure

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MICRO-ENCAPSULATED LACTOBACILLUS PLANTARUM LAB12 WITH EXCELLENT HEAT AND STORAGE STABILITY DURING AND AFTER PELLETISATION

M. F. ISMAIL[1,2], S. M. LIM[1,2], K. RAMASAMY[1,2]*

[1]Faculty of Pharmacy, [2]Collaborative Drug Discovery Research (CDDR) Group, Pharmaceutical and Life Sciences Community of Research, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: kalav922@gmail.com

ABSTRACT

Probiotics mainly lactic acid bacteria (LAB) are often incorporated in food and feed to improve their functionality. The instability of probiotic to temperature change during and after pelletisation, however, often compromises their survivability. The present study addressed this issue by encapsulating Lactobacillus plantarum LAB12 within microcapsules made up of vegetable-based protein (VP) and alginate (Alg). LAB12-loaded microcapsules were subjected to pelletisation at extremely high temperature of up to 90 °C. The storage stability of microencapsulated LAB12 post-pelletisation was also assessed over a period of four weeks. This study went on to evaluate the safety profile of microencapsulated LAB12 *in vivo*. For the acute toxicity study, Sprague-Dawley (SD) rats (n = 6/group; 8 w old; female) were being fed with a single dose of 5.0 g/kg BW microencapsulated LAB12 and observed for 14 d. For the subchronic toxicity study, SD rats (n = 6/group; 6 w old; male and female) were being fed with microencapsulated LAB12 (0.5 g/kg BW or 2.0 g/kg BW or 5.0 g/kg BW) for 90 d. The present findings indicated that microencapsulated LAB12 are effectively protected from 90 °C for an extended period of 100 sec during pelletisation, with survival of up to 89.4% (cell survivability = 9.2 log CFU g-1). Post-pelletisation storage for 4 w yielded minimal viability loss of only 24% (cell survivability = 7.8 log CFU g-1). Both acute and subchronic toxicity study revealed no treatment-related adverse effects across all vital physiological parameters. The present study has successfully produced heat and storage stable microencapsulated LAB12 which could be safely incorporated into food and animal feed.

Keywords: Microencapsulation, Lactobacilli, Pelletisation, Heat, Toxicity

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EXTRACTION AND PROPERTIES OF GELATIN FROM ASIAN SWAMP EEL (MONOPTERUS ALBUS) SKIN

Y. MD YUSOF*, B. A. NORIZAN, N. H. UYUP

Faculty of Applied Sciences, Universiti Teknologi MARA, Perlis, Malaysia
Email: yuswanie824@perlis.uitm.edu.my

ABSTRACT

The Asian swamp eel (Monopterus albus) is one of the common food fishes found in rice fields, muddy ponds and swamp areas of Malaysia. In the current work, gelatin was successfully extracted from the skin of Asian swamp eel by alkaline and acid pre-treatment method with a yield of 33.07% on the basis of wet weight. The functional group, pH value and functional properties were also studied. The gelatin obtained was snowy white and light-textured in appearance and free of fishy odor. The FTIR spectra showed major absorption band in the amide region and revealed the significant loss of molecular order of the triple helix. The pH value was 3.95. Asian swamp eel skin gelatin exhibited a slightly high emulsifying activity but low emulsifying stability, whilst the foam expansion and foam stability were considerably high. Therefore, Asian swamp eel skins could be used as a potential source of gelatin obtainment in order to replace the land animal gelatin.

Keywords: Asian swamp eel, Fish skin, Gelatin, Functional properties

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FORMULATION AND EVALUATION OF TRAMADOL HYDROCHLORIDE FLOATING TABLETS

R. NETHAJI[1]*, N, GOPAL[3], M. SENTHIL KUMAR[4], G. BABU[2]

[1]Department of Pharmaceutics, [2]Department of Pharmaceutical Chemistry, Devaki Amma Memorial College of Pharmacy, India, [3]Deparment of Medicinal Chemistry, Faculty of Pharmacy, Mahsa University, Kuala Lumpur, Malaysia, [4]Department of Pharmaceutics, Annai Veilankanni's pharmacy college, India

Email: nethajijkkncp32@gmail.com

ABSTRACT

Tramadol Hydrochloride (TH) is used for severe pain and has bioavailability about 75% and maximum absorption found in Gastro intestinal tract (GIT). Immediate releases of TH produce various side-effects. An attempt has been made to design gastric floating tablets of TH with a view to improve its oral bioavailability and reduce frequency of administration. The aim of the study is to formulate TH floating tablet using different grades of hydroxy propyl methyl cellulose (HPMC), ethyl cellulose and sodium carboxyl methyl cellulose (SCMC) in order to prolong the drug release and improve the bioavailability and therapeutic efficacy of drugs. Floating tablets of TH were prepared by direct compression method using sodium bicarbonate and HPMC K4M, HPMC K100M in different ratio as per standard procedure and evaluated for their compatibility with polymers, *In-vitro* buoyancy, *In-vitro* dissolution and accelerated stability studies. From FTIR data it has showed that there are no interactions between drug and polymers. The flow properties indicated good characteristics and have good compressibility index. In buoyancy studies, batch containing HPMC polymer showed good buoyancy lag time and total floating time. From dissolution studies, formulations containing higher HPMC viscosity grades have slower drug release rate than the formulations with lower HPMC viscosity grades. The amount of drug released from the drug polymer ratio was found to be in the order of HPMC K4M>HPMC K100M. From the drug release kinetic studies, Higuchi and peppas model exhibited best fit and predicted that release of TH from the floating formulations were of diffusion type. Stability studies showed that there were no significant changes in the parameter of tablets. It is concluded that gastro retentive tablet were controlled the delivery of TH for long period in stomach with minimum floating time and found that F12 was the best formulation.

Keywords: Gastro-retentive floating tablets, TH, *In-vitro* drug release studies, Buoyancy studies, Stability studies

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FORMULATION OF PVA AND PEG HYDROGELVIA FREEZE-THAW METHOD

A. S. AHMED, M. TAHER*, U. K. MANDAL, J. MOHD JAFFERI

Department of Pharmaceutical Technology, Kulliyyah of Pharmacy, International Islamic University, Kuantan, Malaysia
Email: mtaher@iium.edu.my

ABSTRACT

Hydrogels are three dimensional, water swollen structures composed of mainly hydrophilic homopolymers or copolymers such as polyvinyl alcohol (PVA) and poly ethylene glycol (PEG) utilized within various biomedical fields. Physically cross-linked synthetic polymers, PVA and PEG, hydrogels are excellent materials owing to their biological tissue like nature. Transparent PVA and PEG hydrogels with varying polymer concentrations were obtained by using freeze-thaw method. Gel fraction and swelling behavior were investigated. PVA (Mw 195,000, Sigma-Aldrich), PEG 400 (Merck) hydrogels prepared by dissolving PVA in deionized water with concentrations (6%, 8%, and 10%), PEG with concentrations (1%, 3%, and 5%) were added and the gel were autoclaved at 121 °C for 15 min. The clear solution was poured into a Petri dish to undergo four consecutive cycles of 16 h freezing at-80 °C and 8 h thawing at RT. Gel properties such as gel fraction, swelling index, water vapour transmission rate and mechanical strength were investigated. Four consecutive cycles of freezing and thawing formed an insoluble and entangled hydrogel. Present study demonstrates a systematic variation in physical-chemical properties of the developed using the freezing–thawing method with PVA and PEG as a function of polymer concentration. From the results with increasing PVA there is an increase in the gel fraction while increasing PEG concentration decreased gel fraction. However, increasing PVA decreased the swelling, and by adding PEG increases the swelling ratio and the water vapour transmission rate. The developed hydrogel using the freezing–thawing method with PVA and PEG was more swell-able and flexible.

Keywords: PVA, PEG, Hydrogel, Freeze-thaw.

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POLYMER SELECTION FOR NISOLDIPINE SOLID DISPERSION TO ENHANCE SOLUBILITY

M. K. OO, B. CHATTERJEE*

Department of Pharmaceutical Technology, Kulliyyah of Pharmacy, International Islamic University, Malaysia
Email: bappa@iium.edu.my

ABSTRACT

Nisoldipine is a very potent calcium channel blocker which is used as anti-hypertensive agent. Although it is widely prescribed widely as oral tablet, its use has however been limited due to poor water solubility with subsequent erratic bioavailability. Solid dispersion of poorly water-soluble drug in long chain polymer has been reported a common approach to enhance solubility and bioavailability. However a limited literature is available on the selection of polymer type and ratio which are critical parameters for solid dispersion. The present study aimed to compare the effect of different polymers on nisoldipine solid dispersion and to investigate the type and range of polymers with optimum nisoldipine solubility. Nisoldipine was solid dispersed in hydroxyl propyl methylcellulose (HPMC-K4M), polyvinyl pyrrolidone (PVP-K30), polyethylene glycol (PEG-4000) and poloxamer (Polo-188) and the solubility enhancement was evaluated by subjecting the solid dispersed nisoldipine (SDnisol) with single and combined polymers, to compatibility study and 24-h solubility study. According to Attenuated total reflectance-Fourier transform infrared spectroscopy (ATR-FTIR) results, all polymers were compatible with nisoldipine. Although solid dispersion with PVP has the highest solubility enhancement of nisoldipine, the excessive hardness of the yielded SDnisol was a major drawback. When PVP combined with poloxamer as a plasticizer in solid dispersion, it has proven optimum nisoldipine solubility with acceptable hardness of yielded SDnisol. The solubility of 1:10 SDnisol (1 g drug+4 g PVP+6 g Polo) in 0.1N HCl was 44.52±0.11µg/ml while that of pure nisoldipine was 1.27±0.1 µg/ml; a significant difference in solubility was seen between them with $p<0.05$. It was determined that the range limits for PVP was 2-4 g and for poloxamer was 5-7 g. In conclusion, it has become evident that using the three level factorial design approaches, the above mentioned ranges of the chosen polymers can be used to produce SDnisol tablets having enhanced solubility, bioavailability with subsequent increased therapeutic effect.

Keywords: Solid dispersion, Polymers, Nisoldipine, Solubility

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EFFECTS OF MIXING IBUPROFEN WITH HYDROPHOBIC AND HYDROPHILIC COLLOIDAL SILICON DIOXIDE ON POWDER FLOW PROPERTY AND SOLUBILITY: A COMPARATIVE STUDY

A. F. RUZAIDI, B. CHATTERJEE*

Department of Pharmaceutical Technology, Kuliyyah of Pharmacy, International Islamic University, Malaysia
Email: bappa@iium.edu.my

ABSTRACT

Glidants, such as colloidal silicon dioxide (CSD), are often used to improve pharmaceutical powder flow property in solid dosage form manufacturing. However, different properties and amount of glidant used affect the solubility of the powder in a characteristic way. Thus this research was aimed to compare the effects of mixing ibuprofen–a well known poorly flowing drug-with hydrophobic and hydrophilic CSD on powder flow property and solubility. Mixtures containing ibuprofen and hydrophobic CSD (0.5%, 1% and 2% w/w) were prepared using lab scale V-blender. Another three sets of mixtures were prepared with hydrophilic CSD. Powder flow properties (measured using angle of repose, Carr's Index (CI)) and solubility (medium: phosphate buffer pH 7.2 at 37 °C) were compared for all the seven samples, including unmixed ibuprofen. In general, the addition of CSD as glidant greatly improved powder flow property (32-44% and 35-39% reduction in CI and angle of repose). Although there were some differences in the level of improvement of flow property when different types and concentration of CSD were used, they were not statistically significant (at 5% significance level). Nevertheless, 2% w/w concentration for both type of CSD resulted in least improvement of flow property. It was expected that higher than 1% w/w amount of CSD produces least improvement in flow property. For both types of CSD, solubility decreased by increasing higher concentration of the CSD was added. Also, the solubility of mixtures containing ibuprofen and hydrophobic CSD was significantly higher than those containing hydrophilic CSD. With 2% w/w hydrophilic CSD, the solubility was even lower than that of unmixed ibuprofen. Therefore, hydrophobic CSD with 0.5% w/w were the best choice in improving flow property while maintaining solubility of ibuprofen. Further study will consider both CSD at 0.5% w/w for dry particle coating and hence tableting.

Keywords: Colloidal silicon dioxide, Ibuprofen, Powder flow property, Solubility

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SAFETY AND EFFICACY EVALUATION OF MICROEMULSION CONTAINING PHYLLANTHUS EMBLICA FRUIT EXTRACT FOR COSMETIC APPLICATION

D. CHARNVANICH*, S. CHANPIROM, A. TANSIRIKONGKOL, V. PANAPISAL, W. SUWAKUL

Faculty of Pharmaceutical Sciences, Department of Pharmaceutics and Industrial Pharmacy, Chulalongkorn University, Thailand
Email: dusadee.v@chula.ac.th

ABSTRACT

Phyllanthus emblica is of interest as an antioxidant and a skin lightening agent for cosmetics. The present study aimed to evaluate safety and *in vivo* efficacy of microemulsion containing the P. emblica fruit extract. Three microemulsions with the great skin penetration were chosen from literature reviews. The extract-loaded microemulsions were characterized for appearance, extract content, stability, *in vitro* release and *in vitro* skin permeation using newborn pig skin by DPPH assay. Skin irritation test of the selected microemulsions was performed in 50 healthy volunteers. Skin moisturizing, lightening and elasticity effects were evaluated in 29 healthy volunteers using Corneometer®, Colorimeter®, Mexameter® and Cutometer®. Sign test was used to analyze difference in efficacy data between after applying the product on the face at each time interval (2, 4 and 8 w) and before application. The results showed the microemulsions were stable at room temperature for at least 8 w. *In vitro* skin permeation data demonstrated that microemulsions could deliver the extract better than the extract solution. The microemulsion (ME2) was safe to use in skin care products. It showed significant ($p<0.05$) increase in skin hydration after application for 8 w. Significant skin lightening effect with increase of ITA was observed after the 2nd and 4th week whereas there was no significant difference in melanin index at every time interval. Significant decrease in R0 values after the 2nd and 4th week represented skin tightness improvement, but then R0 value significantly increased after the 8th week. Significant decrease in R2 and R7 values indicated reduction of skin elasticity which might be due to high alcohol content in the formulation. However, we observed from the volunteer photos which showed its skin lightening and wrinkle diminish effects after the 8th week. More than 95% of the volunteers were satisfied with the product for skin moisturizing, lightening and firming effects. In conclusion, the microemulsion containing Phyllanthus emblica fruit extract showed a good potential for further development to cosmetic products for anti-wrinkle and skin lightening effects.

Keywords: Phyllanthus emblica, Microemulsion, Efficacy, Skin moisturizing, Skin lightening

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PREPARATION AND CHARACTERISATION OF CHITOSAN-TRIPOLYPHOSPHATE NANOPARTICLES: OPTIMISATION OF FORMULATION AND PROCESS PARAMETERS

Z. HUSSAIN*, S. SAHUDIN

Department of Pharmaceutics, Faculty of Pharmacy, Universiti Teknologi MARA, Campus Puncak Alam, Malaysia
Email: zahidh85@yahoo.com

ABSTRACT

Chitosan (CS)–tripolyphosphate (TPP)–nanoparticles (NPs) have been extensively studied during the past few decades due to their well-recognised applicability in various fields. Thus, the present study was aimed to optimise the fabrication conditions for the preparation of CS-TPP-NPs aiming towards smallest possible size, highest zeta potential and narrow polydispersity index (PDI), simultaneously. In this study, CS-TPP-NPs were prepared via the ionic cross-linking of CS and TPP and were characterized physico-chemically (particle size, zeta potential and dispersity index) and morphologically. The influence of several formulation conditions (CS concentrations, CS: TPP mass ratio and the initial pH of CS solutions) and process parameters (stirring speed, stirring time, ultra-sonication and ultra-centrifugation) on the colloidal characteristics of the CS-TPP-NPs were also investigated. In addition, the colloidal stability of the prepared NPs was also assessed on storage. Data obtained clearly identified that the formulation and process parameters showed significant impact on the physico-chemical and morphological characteristics of the CS-TPP-NPs. The CS-TPP-NPs prepared under optimum conditions (CS concentration of 0.2 mg/ml, CS: TPP mass ratio of 7:1, initial pH of CS solution of 4.0, stirred at 700 rpm for 10 min and ultra-centrifuged at 25 000 rpm for 30 min), had shown a mean particle size of ~187±21 nm, zeta potential around+37±3.5 mV, poly dispersity index of ~0.28±0.03 as well as the smooth and round shaped morphology. The present study describes the optimal circumstances to fabricate the CS-TPP-NPs with finest physical-chemical characteristics and also explore the prospects of manipulation and optimization of the NPs for further intended applications.

Keywords: Chitosan nano particles, Ionic cross-linking, Morphology, Ultra-sonication, Ultra-centrifugation.

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STATIN ARGININE SOLUTION BINARY MIXTURE: MOLECULAR INTERACTION BY FOURIER-TRANSFORM INFRARED (FTIR) SPECTROSCOPIC ANALYSIS

M. M. AFFANDI[1,3], M. TRIPATHY[1,4]*, A. B. A. ABDUL MAJEED[2,4]

[1]Laboratory Fundamental of Pharmaceutics, [2]Pharmaceutical Life Sciences Department, Faculty of Pharmacy, [3]DDH Core, [4]Brain and Neuroscience Core, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: minaketan@puncakalam.uitm.edu.my

ABSTRACT

Studies of thermodynamic properties and molecular interactions of binary solution mixture are of considerable interest for understanding the nature of interactions between component molecules. Furthermore, these properties allow one to draw information on the molecular structure and interactions phenomenon. The present work is a part of our studies on assessing the molecular interaction of statin-arginine solution binary system by Fourier-Transform Infrared (FTIR) spectroscopic analysis. The Arginine (ARG) at concentration of 7.34 mol dm-3 systems containing saturated Simvastatin (SMV) or Atorvastatin (ATV) were subjected to spectroscopic analysis by Varian FTIR 600 series. Analysis was done at room temperature within 400-4000 cm-1 wave number with the scanning rate of 20 at a resolution of 4 cm-1. FTIR spectrum of both statins arginine binary solution system shown either broad or disappearance of the peak as compared to solution of pure compounds. Broad peak was seen at 3436 cm-1 and 3348 cm-1 for SMV and ATV respectively that corresponds to the functional group of O-H stretch. The appearance of the peak at 2075 cm-1 in the case of SMV was also observed corresponding to a functional group being attributed to ARG. This indicated the formation of weak intermolecular interactions through hydrogen bonding between component molecules in the system. Furthermore the deviation in the spectrum peak values was observed in solution form as compared to the powder form. This study successfully explains the molecular interactions in the SMV-ARG and ATV-ARG binary mixture system. This information provides clear understanding on the interactions between molecules that is important from the pharmaceutical point of view.

Keywords: Statin, Arginine, Binary solution, FTIR.

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ANTIMICROBIAL PROFILE OF TRANS-CINNAMALDEHYDE BASED MICROEMULSION: CONVENTIOL AND TEM IMAGING STUDIES

S. N. ASHAKIRIN[1], M. TRIPATHY[1,2]*, A. B. ABDUL MAJEED[2]

[1]Laboratory of Fundamentals of Pharmaceutics, Faculty of Pharmacy, Universiti Teknologi MARA, [2]Pharmaceutical and Life Sciences (PLS) Community of Research (CoRe), Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: minaketan@puncakalam.uitm.edu.my

ABSTRACT

Trans-cinnamaldehyde (TC) has been investigated extensively for its antimicrobial efficacy in food and related industries. The trans-cinnamaldehyde containing sunflower oil based oil in water type of emulsion has been prepared based on the pre-emulsion and subsequent sonication technique. The incorporation of Trans-cinnamaldehyde (4%) into the sunflower based oil in water (10% oil phase+1% surfactant+89% aqueous phase) emulsion is seen to convert the nanoemulsion into microemulsion, which can be attributed to the massive agglomeration of droplets resulting bigger size because of the changed physico-chemical characters of the oil phase, hence the subsequent alteration in the dispersion dynamics. The characterization and stability analysis has been presented elsewhere. In this communication it is intended to present the antimicrobial efficacy of the trans-cinnamaldehyde containing microemulsion (ME-TC) using conventional as well as the imaging techniques of model bacterium E. coli. The serial dilution study has been performed using 10 test tubes, the first two being used as a positive and negative control, the rest eight tubes being diluted with subsequent dilutions starting from initial concentration of 40 mg/ml trans-cinnamaldehyde. The MIC and MBC out of the above experiment are found to be 1.25 mg/ml and 2.5 mg/ml respectively. The imaging studies have been carried out using four different groups being denoted as A (5 ml LB+0.5 ml E. coli+1.5 ml dH2O), B (5 ml LB+0.5 ml E. coli+1.5 ml SF oil), C (5 ml LB+0.5 ml E. coli+1.5 ml ME-TC) and D (5 ml LB+0.5 ml E. coli+60µl TC+1440µl dH2O) respectively and incubated for 24 h after which the samples were processed for TEM imaging. TEM imaging reveals the fact that 60µL trans-cinnamaldehyde containing microemulsion (1.5 ml) is very much equivalent to its antimicrobial potential with 60µl of pure trans-cinnamaldehyde.

Keywords: Antimicrobial, Trans-cinnamaldehyde, Bacterium morphology

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COARSE GRAINED MOLECULAR DYNAMICS SIMULATION OF THE EARLIEST STAGES OF NANOPRECIPITATION AND POLYMER-NANOCRYSTALS INTERACTION

SHAHZEB KHAN[1]*, JAMSHED ANWAR[2]

[1]Department of Pharmaceutics, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor Malaysia, [2]Department of Chemistry, Faraday Building, Lancaster University, United Kingdom
Email: shahzeb_333@hotmail.com

ABSTRACT

Coarse grained molecular dynamics (MD) simulations were carried out to gain insights into the earliest stages of crystallization of the nanocrystals and their interaction with the polymers at interfaces. The starting system comprised two juxtaposed solution phases, an aqueous phase containing PVP in water, and an ethanol phase containing the drug in solution. The MD simulations were carried out using DL-POLY in the constant temperature constant pressure (NPT) ensemble. The temperature and pressure employed were 298K and 1bar respectively using the Hoover algorithm with the thermostat and barostat relaxation times set to 1.0 ps and 10.0 ps respectively. The system contained a total of 70000 molecules comprising 17500 drug molecules, 17500 ethanol molecules, 34977 water molecules, and 23 PVP molecules. The simulation covered a period of 180 ns. This simulations studies revealed that during anti solvent crystallization the solute are crystallized in organic solvent region during mixing of the solvent and anti solvent phases. This is because of the hydrophobic nature of the drug molecules which cannot accommodate the water molecules to be infused and the solvent molecules are diffused towards the water phase. As the solute molecules consolidate and crystallise we observed the slow migration of the PVP molecules to the crystal surfaces. This suggests that polymers in general are unlikely to play a significant role in promoting nucleation. This study provided physical insight into the formation of nanocrystals during anti-solvent crystallization. It explored the diffusion mechanism of solute, solvent and anti-solvent (water) molecules which lead to phase separation of the two solutions during mixing process.

Keywords: Nano precipitation, MD simulations, DLPOLY, PVP, Nanocrystals

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DRUG PERMEATION STUDY THROUGH RAT SKIN: EFFECT OF STORAGE CONDITIONS OF SKIN

S. F. ABDUL RAZAK*, N. H. ABU SAMAH

Faculty of Pharmacy, Department of Pharmaceutics, Universiti Teknologi MARA, Malaysia
Email: syahriahfadhilah194@hotmail.com

ABSTRACT

The purpose of this study was to compare the effects of different storage conditions on the permeation of two types of model compound (i.e. caffeine, ibuprofen) across the full-thickness rat skin. These model compounds were used to demonstrate the permeation of both hydrophilic and hydrophobic drugs. The full thickness rat skin was excised from dorsal part of albino out-bred Sprague-Dawley (3-months-old). The skin sample used for ex-vivo permeation study was prepared in three conditions: freshly excised, 1 w frozen stored and 6 mo frozen stored (frozen at-15 °C). The amount of caffeine and ibuprofen permeated for each skin sample was obtained from the receptor compartment of Franz diffusion cells every 2 h for over a period of 12 h and analysed by UPLC. It was found that the samples from fresh skin demonstrated less permeation than samples obtained from frozen stored skin for both drugs. These results suggest that the freezing of freshly excised rat skin for up to 6 mo does significantly affect the barrier function of the epidermal layers, resulting in significant change in the permeability of the model compound. Therefore, the use of frozen rat skin samples should be avoided in the ex-vivo permeation study.

Keywords: Caffeine, Ibuprofen, Permeation, Rat skin, Storage

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ASSESSMENT OF ACCELERATED STABILITY STUDIES OF ETHANOL EXTRACT OF ORTHOSIPHON STAMINEUS AND ITS NANO LIPOSOMES USING HIGH PERFORMANCE LIQUID CHROMATOGRAPHY AND FOURIER-TRANSFORM INFRARED SPECTROSCOPY COMBINED WITH CHEMOMETRIC APPROACH

A. SHAFAEI, M. A. AHMED SAEED, Z. ISMAIL*

Department of Pharmaceutical Chemistry, School of Pharmaceutical Sciences, Universiti Sains Malaysia, Minden, Penang 11800, Malaysia
Email: ismailzhari@gmail.com

ABSTRACT

Background: Orthosiphon stamineus (OS) Benth. (Lamiaceae) is a medicinal plant grown in Southeast Asian countries. Objectives: This study aimed to carry out accelerated stability studies on OS ethanolic extract (OS-E) and nano liposomes of OS ethanolic extract (OS-EL). Methods: The extracts were exposed to four different temperatures at 30, 40, 50 and 60 °C for 6 mo. The samples were then analyzed at 0, 1, 2, 3, 4, 5 and 6 mo by high performance liquid chromatography (HPLC) using rosmarinic acid (RA), 3'-hydroxy-5, 6, 7, 4'-tetramethoxyflavone (TMF), sinensetin (SIN) and eupatorin (EUP) as markers. Different chemical kinetic parameters of the markers were evaluated by Arrhenius equation to predict shelf life (t90) at different storage conditions at room temperature. Moreover, stability of OS-E and OS-EL were analyzes by chemical fingerprinting using Fourier Transform Infrared (FTIR) spectroscopy, Principal Component Analysis (PCA) and Hierarchical Clustering Analysis (HCA). Results: The markers in both OS-E and OS-EL followed the first order degradation. The rate constant, pre exponential factor and activation energy of the markers were found to be varying in samples stored at different conditions. The contents of the markers were found to be decreasing at high temperature with the passage of time. The predicted shelf life (t90) of the markers at room temperature was found to be 3.33, 7.57, 6.72 and 7.319 mo in OS-E and 5.48, 10.05, 10.84 and 10.08 mo in OS-EL for RA, TMF, SIN and EUP, respectively. The FTIR results showed discrimination between extracts based on the storage conditions. The results demonstrate that identification of the most similar samples based on their metabolites profile and the most different ones can be easily established by using either chemo metric analyses of PCA or HCA. Conclusion: Stability studies are necessary to understand the influence of environmental factors over the time on the quality of a plant extract substance.

Keywords: Ortho siphon stamineus, Nanoliposomes, Stability, HPLC, FTIR

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EXPLORING POWDER BLEND UNIFORMITY OF A BINARY MIXTURE CONTAINING METFORMIN HCl AND HPMC BY DIFFERENTIAL SCANNING CALORIMETRY

S. MAHMOOD, N. N. B. HILMI, N. K. B. HUSAIN, B. CHATTERJEE, U. K. MANDAL*

Department of Pharmaceutical Technology, Kulliyyah of Pharmacy, International Islamic University Malaysia (IIUM), Kuantan, Pahang, Malaysia
Email: mandalju2007@gmail.com

ABSTRACT

VBlender, also known as twin–shell mixer, is widely used in pharmaceutical industries for mixing powders, granules, and other solid materials. Its mixing mechanism involves continuous splitting, followed by intermixing of powder materials as the shell rotates on its horizontal axis. In pharmaceutical industries, mixing operation is considered a critical step in process scale updduring product development as well asroutine commercial manufacturing. In this research, HPMC and metformin HCl (95:5 w/w) at 50% and 60% occupancy of a V blender was mixed to make a binary mixture and further analyzed for their mixing uniformity by a DSC method. The results of the DSC method were further confirmed by a UV method. Powder samples were withdrawn at 5 min, 10 min, 15 min, and 20 min intervals each from the top, middle, and bottom positions of the blender. Recovery of metformin HCl after 20 min of mixing by both the UV method (2.66±1.10% and 2.60±0.32% at 50% and 60% occupancy levels, respectively) and the DSC method (2.68±0.22% and 2.29±0.14% at 50% and 60% occupancy, respectively) were found comparable. Also, maximum RSD of enthalpy (21.55 at 10 min at 50% occupancy) for sample analysis by the DSC method was comparable to that of the UV method (25.201 at 15 min at 50% occupancy). As aconclusion, DSC method can be further explored as an alternative of UV spectroscopy or HPLC method to assess the mixing uniformity of powder samples in pharmaceutical industries.

Keywords: Mixing uniformity, DSC, UV, V blender, Binary-mixture

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PALM OIL DERIVATIVES-BASED MICROEMULSION AS HAIR SERUM

S. RAMLI*, C. L. HONG

Faculty of Science and Technology, School of Chemical Sciences and Food Technology, Universiti Kebangsaan Malaysia, Bangi, Selangor, Malaysia
Email: su_ramli@ukm.edu.my

ABSTRACT

This research was performed to study the formation of microemulsion based on palm oil derivatives for the production of hair serum. Microemulsion was produced by using polyoxyethylene (20) sorbitan monooleate (Tween 80) as a surfactant (S), propylene glycol as a co-surfactant (CoS) and isopropyl myristate (IPM) as an oil phase. Ternary phase diagram was constructed to determine the area of microemulsion formed using the water titration method. Formulation with ratio 1:9 (IPM: S/CoS) of Km 3:1 was chosen for further characterization because it formed the biggest microemulsion area on ternary phase diagram. Characterization was performed to determine the stability, particle size, viscosity and electrical conductivity for microemulsion system with-and without additives. The results of viscosity and electrical conductivity showed that the changes of the type of microemulsion from water-in-oil (w/o) to bicontinuous and then to oil-in-water (o/w). Particles size for the microemulsion formulations were in a range of 10-200 nm. Microemulsion systems with additives showed good stability properties at temperature 4, 25 and 40 °C. Observation on the hair cuticle's surf was carried out using Scanning Electron Microscope (SEM) for the formulation that contains 30% and 50% water only. It was performed in order to determine the efficiency of the formulations. The observation showed that the formulations can improve moisturizes and smooth the hair effectively.

Keywords: Microemulsion, Palm oil derivatives, Hair serum, Ternary phase diagram

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TRANSDERMAL PERMEATION MECHANISM OF SODIUM DEOXYCHOLATE AIDED NANO-TRANSFERSOMES BY DIFFERENTIAL SCANNING CALORIMETRY (DSC)

S. MAHMOOD, M. TAHER, U. K. MANDAL*

Department of Pharmaceutical Technology, Kulliyyah of Pharmacy, International Islamic University Malaysia (IIUM), Kuantan, Pahang, Malaysia
Email: mandalju2007@gmail.com

ABSTRACT

Transfersomes are lipid based nano-vesicles made of phospholipid and surfactant. Their drug permeation mechanism through the skin has been attributed to the moisture seeking tendency (xerophobia) of the lipid vesicles followed by destabilization of lipid bi-layer in the stratum corneum (SC) by surfactant. However, structural changes of SC due to surfactant need further elucidation to know the specific role of the surfactant in doing so. The objective of this study was to evaluate drug permeation mechanism of Raloxifene loaded nano-transfersomes containing sodium deoxycholate as a surfactant. Phospholipon® 90G was used as a lipid composition in the nano-formulation. Three different types of skin i.e. mice, guineapig and rabbit were used in this study. The SC was microtomed from the rest of the skin layers with chemical treatment, thoroughly washed, and kept for drying in a vacuum desiccator. The SC samples were then subjected to an ex-vivo permeation study of the transfersomal formulation for a period of 8 h. A control sample was prepared in a similar way, without any formulation treatment. A sample of the SC section was cut, sealed in aluminum hermetic pans and scanned using DSC at a scanning rate of 5 °C per min over the range of 25 °C–125 °C. The characteristic bi-layer lipid and keratin transition peaks found in the control SC samples were: 75 °C, 78 °C, and 95 °C for mice; 820C, 900C, and 990C for guineapig; and 840C, 920C, 990C for rabbit. However, upon treatment with sodium deoxycholate aided Raloxifene nano-transfersomes, most of the peaks were shifted towards lower melting points and some of them were disappeared. This finding confirmed disruption of lipid bi-layer and denaturation of keratin in the SC layer of the investigated skin samples by nano-transfersomes with sodium deoxycholate. The results of the present study clearly highlighted the role of sodium deoxycholate in transdermal permeation of drug loaded nano-transfersomes formulation.

Keywords: Transfersomes, Differential scanning Calorimetery, Raloxifene, Sodium deoxycholate

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PHARMACOKINETIC PROFILES OF ENTERIC-COATED INSULIN NANOPARTICLES IN RAT PLASMA AFTER ORAL ADMINISTRATION

M. H. MOHD JAAFAR[1], K. ABDUL HAMID[1,2]*

[1]Pharmaceutics Department, Faculty of Pharmacy, [2]Brain and Neurosciences Communities of Research, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: khuriah@puncakalam.uitm.edu.my

ABSTRACT

Double-functional nanoparticles with pH-sensitivity and mucoadhesivity were developed to prevent insulin from contacting the highly acidic medium in the stomach, prolong the intestinal residence time and increase the permeability of insulin to the systemic circulation. Four different formulations of enteric-coated insulin nanoparticles were prepared using polyelectrolyte complexation and ionotropic gelation methods with varying concentrations of alginate and calcium chloride. Their pharmacokinetic profiles were characterized after oral administration. Four groups of rats were administered orally with 100 mg/kg of enteric-coated insulin nanoparticles while a control group was given 10 mg/kg of pure insulin solution. Blood samples (200 µl) were collected using heparinized syringes at predetermined time intervals over 240 min. 100 µl of plasma samples were introduced into HPLC and insulin concentration was measured at 210 nm. Pharmacokinetic profiles of insulin in each group were calculated using trapezoidal method. The formulation that contained 1.0% alginate and 1.16% calcium chloride showed the highest absorption profile with AUC value of 2135.71±68.64 µg/ml. min. The control group exhibited the lowest AUC value of 192.49±10.92 µg/ml. min. In conclusion, delivery systems which exhibit pH-sensitivity and mucoadhesive properties could have an excellent synergistic effect to enhance insulin absorption via oral delivery.

Keywords: Insulin nanoparticle, Pharmacokinetic profiles, Oral administration, Alginate, Chitosan

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SOLUBILITY ENHANCEMENT OF SIMVASTATIN IN THE SOLUTION OF α-, β-, AND γ-CYCLODEXTRINS: A COMPARATIVE PHASE SOLUBILITY STUDY

M. A. ISMAIL[1], M. TRIPATHY[1,2]*, A. B. ABDUL MAJEED[2], S. AOKI[3]

[1]Laboratory of Fundamentals of Pharmaceutics, Faculty of Pharmacy, [2]Pharmaceutical and Life Sciences (PLS) Community of Research, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [3]Department of Medicinal and Life Sciences, Faculty of Pharmaceutical Sciences, Tokyo University of Science, Japan
Email: minaketan@puncakalam.uitm.edu.my

ABSTRACT

Solubility enhancement of poorly aqueous soluble drugs by cyclodextrins has been the main focus of many researchers. The cyclodextrins such as α, β and γ possess anomalous pattern as a result of which; it may affect the enhancement phenomenon of a poorly soluble drug. Simvastatin (SMV) is classified under BCS class II drug, extensively used for its cholesterol lowering activity. Several methods have been proposed to enhance its solubility. In this presentation, we report the differential solubility behaviour of model drug SMV, in presence of α-, β-, and γ-cyclodextrins. Phase solubility study was carried out using the method of Higuchi and Connor. Briefly, different molar concentrations such as 1 mM to 7 mM concentrations of α-, β-, and γ-cyclodextrins were prepared separately and correspondingly denoted as S1 till S7 respectively. S0 has been denoted as blank. Excess amount of SMV was added to different molar solutions of α-, β-, and γ-cyclodextrins to prepare saturated solutions using a shaking water bath maintained at 37 °C for 72 h and so also for blank. At the end of experiment, solutions were filtered and SMV content was determined spectrophotometrically at 238 nm. The intrinsic aqueous solubility (S0) of SMV is observed to be 8.89 µM. The solubility of SMV is observed to decrease for S1 till S5 followed by an increase (S6 and S7) in case of α-cyclodextrin solutions as compared to that of S0. A linear increase (R2=0.991, AL pattern means 1:1 inclusion complexation) of SMV solubility is seen for all the studied concentrations of β-cyclodextrins concentration dependently. However a similar pattern (R2=0.976) is found in case of γ-cyclodextrins solutions (with higher solubilisation compared to that of β-cyclodextrin solutions) but with a deviation in of S7, where the solubility is observed to decrease.

Keywords: Cyclodextrin complexation, Simvastatin, Solubility enhancement

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POLYVINYL PYRROLIDONE WRAPPED SINGLE WALL CARBON NANOTUBES (PVP-SWNTs): SOLID STATE CHARACTERISATION

M. WAN CHIK[1], M. TRIPATHY[1,2]*, A. B. ABDUL MAJEED[2,3]

[1]Laboratory Fundamental of Pharmaceutics, [2]Department of Pharmaceutical Life Sciences, Faculty of Pharmacy, [3]Brain Degeneration and Therapeutics Group, Pharmaceutical and Life Sciences Community of Research, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: minaketan@puncakalam.uitm.edu.my

ABSTRACT

Polymer Polyvinyl Pyrrolidone (PVP) wrapped single walled carbon nanotubes (SWNTs) (PVP-SWNTs) have been synthesised as presented elsewhere. In this presentation, we report the solid state characterizations such as X-Ray Diffraction (XRD), Fourier-Transform Infrared Spectroscopy (FTIR) and Differential Scanning Calorimeter (DSC). The aim of the study is to characterise the solid state of synthesised PVP-SWNTs. XRD pattern for PVP, SWNTs and PVP-SWNTs have been recorded using Rigaku Ultima IV with Cu-Ka radiation generated at 40 kV and 40 mA. For similar samples FTIR and DSC have been carried out using Varian Pro Excalibur 3100 FTIR spectrophotometer at room temperature within 400-4000 cm-1 wave number with scanning rate of 40 at a resolution of 4 cm-1 and Perkin-Elmer power-compensated Hyper DSC 8500 in purified nitrogen atmosphere with heating rate of 20 °C/min respectively. As presented, result of PVP-SWNTs showed disappearance of peak 2θ at 25.44 which apparently appeared in SWNTs, while having peaks at 2θ angles of 4.49 and 6.77. The FTIR spectrum of PVP-SWNTs revealed the characteristic peaks of PVP is dominant as it wrapped around the SWNTs. The results showed its characteristic peaks at (cm-1) 1096, 1292, 1375, 1443 and 1465, and 1654 representing the functional groups of C-C stretching of the ring, C-N stretching, shifting of C-H bending, moiety of pyrrolidone, and C=O stretching respectively. The DSC thermogram of PVP showed a broad endothermic peak at 85.40 °C with enthalpy of fusion 793.24 J/g which was suggested as a dehydration profile of a hygroscopic amorphous material and may be attributed to the endothermic relaxation. In case of samples PVP-SWNTs the melting transition in DSC thermogram appeared at 72.29 °C and can be considered non-significant. Hence, this study explains the solid state of the synthesised PVP-SWNTs. This information provides better understanding for future references in biological research focusing in the targeted drug delivery system.

Keywords: PVP-SWNTs, SWNTs, PVP, Solid state characterization

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FOOD SECURITY STATUS: ASSOCIATION WITH ENDOTHELIAL DYSFUNCTION AND BODY MASS INDEX AMONG YOUNG ADULT

A. NUR ATIQAH[1], N. MD. NOR[2]*, M. F. MASTUKI[3], K. A. MD. ISA[1]

[1]Department of Basic Science, [2]Department of Nutrition and Dietetic, [3]Department of Medical Lab Technology, Faculty of Health Sciences, Universiti Teknologi, MARA, Puncak Alam Campus, Selangor, Malaysia
Email: azmir2790@puncakalam.uitm.edu.my

ABSTRACT

Food insecure, inadequate quality and quantity of nutritious food are associated with adverse health outcome. Food security status is divided into two groups, food secure and food insecure. These studies investigate endothelial dysfunction marker (E-selectin) and body mass index (BMI) for both groups among young adult. A cross-sectional comparative study was carried out among participants aged between 18 to 25 y old (n=124) were selected through Adult Food Security Survey Module (AFSSM) and participated in blood draw procedure. Sandwich-ELISA principle used in Human SELE (E-Selectin) enzyme-linked immunosorbent assay (ELISA) kit with a detection range of 78.13 to 5000 pg/ml. Of the 124 participants, 40 were selected for data analysis after checking outliers and normality distribution. An independent-samples t-test was conducted to compare the food security status for E-selectin and BMI. There was no significance difference on E-selectin for food insecure (3867.28±793.34 pg/ml) and food secure (2809.81±568.38 pg/ml), t(38) =-4.917, p<0.001 (two-tailed). The magnitude of the differences in the means (mean difference =-1057.47, 95% CI: -1492.8 to-622.11) and BMI for food insecure (20.08±2.59 pg/ml) and food secure (19.82±2.14 pg/ml), t (38) =-0.344, p= 0.733 (two-tailed). The magnitude of the differences in the means (mean difference =-0.259, 95% CI: -1.785 to 1.267). Mean for food insecure (p>0.05) was slightly higher in E-selectin and BMI compared to food secure which indicated that young adult also has potential risk for chronic illnesses. This study described knowledge of food insecurity experienced to allow better understanding of healthy food choices. This study will continue further in depth in gene expression of peroxisome proliferator-activated receptor gamma (PPAR-y).

Keywords: Food security, Food insecure, E-selectin, BMI, Young adult

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A RETROSPECTIVE COHORT STUDY ON THE EFFICACY OF HOMEOPATHY PLUS CONVENTIONAL MEDICINE COMPARED TO CONVENTIONAL MEDICINE IN THE TREATMENT OF HYPERLIPIDAEMIA

I. U. MHAISKER*, F. M. H. GIRACH

Faculty of Traditional and Complementary Medicine, Cyberjaya University College of Medical Sciences, Cyberjaya, Selangor, Malaysia
Email: ibrahim@cybermed.edu.my

ABSTRACT

Some of the medicines used in homeopathy are well proved whereas some have not been proved in details and have been verified only clinically by some authors. Cholesterinum is one such medicine where research in its use as lipid lowering medicine is lacking in the literature. Also, today's sedentary life style has brought about increase cases of hyperlipidaemia and concurrent cardiovascular and cerebrovascular diseases. Standard treatments available in modern science have adverse effects. While the demand for traditional and complementary medicine (T&CM) is on the rise, more evidence is needed to evaluate whether the clinical use of homeopathy will bring any beneficial effects to the community at large. The aims of this study were 1) to evaluate the effects of hypolipidaemic activity of homeopathy medicine Cholesterinum and to compare integrated treatment using homeopathy plus conventional pharmacotherapy, homeopathy medicine alone and conventional treatment alone in order to determine its efficacy in the treatment of hyperlipidaemia and 2) to evaluate the efficacy of homeopathy in the treatment of hyperlipidemia. This was a retrospective cohort study. The study was done at the Yerala Homeopathic Medical College located in Mumbai, India. Cases diagnosed and treated for hyperlipidaemia under the outpatient department affiliated to Yerala Homeopathic Hospital and Research Centre were assessed for eligibility, classified and analyzed. Patient records were retrieved from the archives of Yerala Homeopathic Hospital & Research Centre, Mumbai, India. Cases treated for hyperlipidaemiain between the years 2010 to 2012 were reviewed before and after twelve weeks of treatment were included. The subjects were categorized into two groups according to the treatment they received i.e. integrated group (homeopathy plus conventional medicine) and conventional group. The change in reducing of hyperlipidaemia was observed twelve weeks after exposure to treatment. The data was then analyzed descriptively and statistically. Statistical results suggest that there is a significant difference between the integrated and conventional group in the outcome of hyperlipidaemia treatment at week twelve. It is concluded that integrated approach is more efficacious than conventional pharmacotherapy alone in the treatment of hyperlipidaemia.

Keywords: Homeopathy, Hyperlipidaemia

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A REVIEW OF TRADITIONAL MEDICINE PRACTICES AMONG THE INDIGENOUS ETHNIC GROUPS IN SABAH

D. MATANJUN*, Z. A. MUSTAPHA, U. J. A. D'SOUZA, M. S. RAHMAN

Faculty of Medicine and Health Sciences, Universiti Malaysia Sabah, Sabah, Malaysia
Email: matanjun@gmail.com

ABSTRACT

As in most other parts of the world, there are many types of traditional medicines that have been used for hundreds of years among the various indigenous ethnic groups in Sabah. However, due to a scarcity of large clinical trials of acceptable quality, a meta-analysis in traditional medicine would be difficult. Traditional medicine in Sabah consists mainly of herbalism, manipulative practices and spiritualism. Herbalism is practiced by all the indigenous races in Sabah. Usually such practice would be based mainly on raw plants and plant extracts; many plants can also be found in herbal products. Herbal medicine is a popular component of the traditional Sabah health care system because of their perceived therapeutic efficacy. Traditional massage and other manipulative-practices, based on manipulation and movement of body parts, are also popular because of their simplicity and apparent immediate relief that can be experienced by the patient or client. In traditional massage therapy, the practitioner would usually explain that the mechanism of action leading to the effectiveness of the treatment would be mainly dependent on getting "wind" out the body, a concept not immediately comprehensible to westerners. Spiritualism, which maybe considered as a peculiar dimension of the mind-body medicine subset of traditional medicine, continues to have its adherents among the natives of Sabah; ceremonies, chants and other attendant healing modalities are important components in the traditional health system in some cultures. A mixture of traditional healing methods may come into play with some healers. The degree of success of any traditional therapy usually depends on the skill and experience of any traditional healer.

Keywords: Traditional Medicine, Indigenous Sabah herb

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STUDY OF ADVERSE DRUG REACTIONS RELATED TO UNREGISTERED TRADITIONAL MEDICINES IN MALAYSIA

N. H. ABD. RAHIM[1]*, N. AHMAD SA'AD[2], M. T. ABDULLAH[3], N. ABD. AZIZ[4]

[1]Jabatan Farmasi, Hospital Angkatan Tentera Tuanku Mizan, Kuala Lumpur, Malaysia [2]Unit Farmasi, Klinik Kesihatan Bandar Botanic, [3]Unit Farmasi, Klinik Kesihatan Bukit Goh, Pejabat Kesihatan Daerah Kuantan, Pahang, Malaysia, [4]Faculty of Pharmacy Universiti Teknologi MARA (UiTM), Puncak Alam Campus, Selangor, Malaysia
Email: idaujala@gmail.com

ABSTRACT

Adverse drug reaction is the fourth to sixth leading cause of death in US. Studies suggested that 6-7% of admission was due to ADRs and the overall fatality was 0.15%. In Malaysia, admission due to ADRs was even higher (8.4%) than other countries. Rising numbers of ADR cases resulted from the use of traditional product triggers a signal that there are some issues need to be investigated. To study the adverse drug reactions related to unregistered traditional medicines in Malaysia. This is a descriptive retrospective study where all ADR data related to unregistered traditional medicines, from 2010 to 2014, were extracted from national pharmaco-vigilance Quest 2 database. The extent of ADR for most cases were reported as severe (n=205; 41.75%). The top five ADR reported according to system organ class was lead by liver and biliary system disorders (n=156), followed by urinary system disorders (n=117), body as a whole–general disorders (n=111), skin and appendages disorders (n=100) and finally gastro-intestinal system disorders (n=96). Out of 417 products reported with ADR cases, 114 products were tested and confirmed adulterated with illegal and controlled substances such as dexamethasone, chlorpheniramine, sildenafil and sibutramine. Increasing trends in the usage of unregistered traditional medicines is a sign lack of awareness regarding their harmful effect amongst the community. Holistic approach in strategies of future pharmaco-vigilance should involve the healthcare professionals, pharmaceutical company, authority agencies and public to prevent ADRs due to the use of unregistered traditional medicines.

Keywords: Unregistered traditional medicine, Adverse drug reactions, ADR

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VIRGIN COCONUT OIL PROTECTS RETINAL GANGLION CELLS FROM APOPTOSIS INDUCED BY SERUM DEPRIVATION

H. IBRAHIM[1,3]*, S. S. DAHHAM[2], S. S. ABDULHAMEED[2,3], A. S. ABDUL MAJID[1], A. M. S., ABDUL MAJID[2]

[1]Integrative Medicine Advanced Medical and Dental Institute (AMDI), Universiti Sains Malaysia, Kepala Batas, Penang, Malaysia, [2]Pharmacology Department, School of Pharmaceutical Sciences, Universiti Sains, Malaysia, Penang, Malaysia, [3]Department of Biology, Faculty of Science, University of Zakho, Duhok, Iraq
Email: abraihimi@yahoo.com

ABSTRACT

Virgin Coconut oil is an edible coconut oil, which is being utilized by the people in Malaysia. It has various applications in food, medicine and industry. Nufera is a trade mark for a commercially available edible coconut oil. It is meticulously extracted through bacteria fermentation process, yielding up to 45% extraordinarily pure lauric acid. Lauric acid is a fatty acid having 12 carbon chains (C12) which is a fat soluble organic compound. The aim of our study was to demonstrate whether FVCO is able to reduce cell death of retinal ganglion cells (RGC-5) caused by serum deprivation. Cells were subjected to serum deprivation and then were treated by using FVCO at different concentrations. Various procedures were used to demonstrate the effect of FVCO such as cell viability using MTT, resazurin fluorescent dye assay, cells morphology, and apoptosis assay. The results of this study show that FVCO decreases apoptotic cell death induced by serum deprivation and significantly increases neuronal viability. Moreover, FVCO decreased the expression of cleaved caspase-3/7 and caspase 8/9 at 25µg/ml. This study demonstrates that FVCO is having anti-apoptotic effects in serum deprived RGC5 cell line, suggesting that FVCO can be considered as a good candidate for neuroprotective therapy.

Keywords: Fermented virgin coconut Oil (FVCO), Anti-apoptotic, Aeuroprotective therapy

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POTENTIAL USED OF ACACIA HONEY IN AMELIORATING STRESS DISORDER: PRELIMINARY RESEARCH RESULTS

Q. H. ZAINOL ABIDIN[1], W. I. WAN ISMAIL[2], N. E. ISMAIL[3], Z. ESHAK[1]*

[1]Department of Pharmacology and Chemistry, [2]Clinical Bio Pharmaceutical Research Group (CBRG), Pharmaceutical and Life Sciences Community of Research, [3]Department of Pharmacy Practice, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: zolkaplieshak@gmail.com

ABSTRACT

Honey has been reported to reduce pre-natal stress. It has been shown that Malaysian Tualang honey and African Idanre honey had antidepressant activity that causes the reduction of distress of expectant mothers. Thus, this study was conducted to observe the anti-stress properties of another Malaysian honey, Acacia honey on stress-induced rats. Acacia honey moisture content, free acidity and pH were obtained for physicochemical parameters and Total Phenolic and Flavonoid Content (TPC and TFC) were conducted to measure the honeys' antioxidant properties. The haemolysis inhibition activity was also carried out by using oxidative haemolysis inhibition assay (OxHLIA). Chronic Mild Stress (CMS) animal model was used to induce stress disorder on male Sprague Dawley rats for 28 consecutive days with 6 different stressors applied randomly each day. Four groups of test rats were applied with CMS: normal control group, stress model group, stress treated with Acacia honey (1.2 mg/kg, orally) and stress treated with amitriptyline (tricyclic antidepressant) group. At the end of experiment, rats were subjected to Sucrose Preference Test and Open Field Test for behavioral study and blood was taken to measure blood biochemistry and stress-related hormone parameters. Our results showed that the quality of Acacia honey was within the acceptable parameters set by International Honey Commission and CODEX Alimentarius. TPC and TFC values for Acacia honey were 57.85 mg GAE/100g and 14.15 mg CE/100g, respectively. As for OxHLIA results, Acacia honey had low haemolysis activity which suggest greater anti-oxidant protective effect. There were no significant differences in behavioral and blood biochemistry parameters compared to normal control group. However, there were significant differences compared to stress-induced rats. These results suggested that Acacia honey may have anti-stress properties and further studies need to be conducted to elucidate the mechanism of action.

Keywords: Acacia honey, Stress, Chronic mild stress, OxHLIA

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CYTOTOXIC AND ANTIANGIOGENIC EFFECT OF BARRINGTONIA RACEMOSA FRUIT AND SEED EXTRACTS

N. AMRAN[1]*, A. N. ABDUL RANI[1], R. MAHMUD[1], K. B. YIN[2], A. M. S. ABDUL MAJID[1]

[1]Department of Pharmaceutical Chemistry, School of Pharmaceutical Sciences, [2]Institute for Research in Molecular Medicine, Universiti Sains Malaysia, 11800 Minden, Penang, Malaysia
Email: liyana_amran@yahoo.com

ABSTRACT

Barringtonia racemosa Roxb (Lecythidaceae) or locally known as putat sungai has been used as a traditional in the treatment of cough, asthma, ulcer and diarrhea. This study investigates the anticancer and anti angiogenic activities of Barringtonia racemosa fruit and seed methanol crude extract, respectively. The anticancer activities of fruit and seed extracts were investigated by *in vitro* cytotoxicity on human breast cancer cell lines (MCF-7), human colon cancer (HCT-116) and human umbilical vein cell line (EA. hy926) by MTT assay. The ex vivo anti angiogenic effect of fruit and seed extracts were determined using rat aorta ring assay. The extraction of fruits and seed were carried out by maceration at room temperature. The results demonstrated that B. racemosa seed extract exhibited a potent cytotoxic effect on MCF-7 cell line (IC50 15.74±1.52 µg/ml) and it is found to be safe on normal cell line (IC50 180.91±1.09 µg/ml). The antiangiogenic activity of B. racemosa fruit extract showed no microvessels outgrowth in rat aortic rings when tested at 100 µg/ml. The present study indicated that the B. racemosa seed extract may be potent anticancer agent while the fruit extract of the same plant possessed antiangiogenic effect. Following this promising finding, further fractionation and purification of the active extract are underway to uncover associated potent plant components as a viable source of anticancer therapeutic and/or preventive agents.

Keywords: Barringtonia racemosa, Cytotoxic, MTT assay, Antiangiogenic, Rat aorta ring assay.

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RATIONALE FOR THE USE OF CAMOMILE LEAVES IN THE TREATMENT OF MIGRAINE

S. AGATONOVIC-KUSTRIN[1]*, D. W. MORTON[2]

[1]Faculty of Pharmacy, Universiti Teknologi MARA (UiTM), Puncak Alam Campus, Selangor, Malaysia, [2]School of Pharmacy and Applied Science, La Trobe Institute of Molecular Sciences, La Trobe University, Bendigo, Australia
Email: snezana@ salam.uitm.edu.my

ABSTRACT

Feverfew leaves have been traditionally used in the treatment of migraine with the parthenolide as the main bioactive component responsible for its effectiveness. However, due to similar flowers, feverfew is sometimes mistaken for German chamomile. Although German chamomile belongs to the same Asteraceae family, its use in migraine prophylaxis is not evidenced. The pharmacological effect of German chamomile is mainly associated with its essential oils extracted from the flower heads, with bisabolol and chamazulene as the main components. The aim of this study was to compare and analyse active components in feverfew, chamomile and calendula using High Performance Thin Layer Chromatography as the analytical method. Calendula was included in the study for comparison since it belongs to the same family. HPTLC chromatographic fingerprints are used to quantify apigenin, chamazulene, bisabolol and parthenolide in extracts from leaves and flower heads of feverfew, German chamomile and calendula. Antioxidant activity in sample extracts was compared by superimposing the chromatograms obtained after post chromatographic derivatization with DPPH• free radical and post chromatographic derivatization with anisaldehyde. Antioxidant activity was expressed in Gallic acid equivalents. Bisabolol and chamazulene, were found to be present in higher concentrations in chamomile and feverfew flowers, and were detected in leaves from flowering German chamomile, while parthenolide was present in higher concentration in leaves and was not detected in flower head extracts. Apigenin was detected and quantified only in chamomile extracts (highest concentration in flower head extracts). Our study suggests that the parthenolide pathway is favoured in leaves, while formation of matricin and bisabolol is favoured in flowers. HPTLC-DPPH• method has enabled quantification and comparison of free radical savaging activity of the major phenolic compounds in extracts. Chamomile leaves and flower heads have the most prominent antioxidant activity, due to the high concentrations of chamazulene, bisabolol and apigenin. The method reported in this work provides the advantage of both identification and quantification of antioxidant constituents in a multi-component sample.

Keywords: Feverfew, German chamomile, Calendula, HPTLC fingerprinting, Reversed phase HPTLC-DPPH bioassay

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THE ANTIDEPRESSANT-LIKE EFFECT OF GINSENG EXTRACT G115 IN MICE BY AN ALTERATION OF SERUM CORTICOSTERONE

R. RODSIRI[1]*, W. BOONLERT[1], J. UMKA-WELBAT[2]

[1]Faculty of Pharmaceutical Sciences, Department of Pharmacology and Physiology, Chulalongkorn University, Thailand, [2]Faculty of Medicine, Department of Anatomy, Khon Kaen University, Thailand
Email: ratchanee.r@pharm.chula.ac.th

ABSTRACT

Ginseng extract G115 has been used as a nutraceuticals worldwide. The benefits of ginseng extract and ginsenosides in the central nervous system have been widely investigated. The present study aimed to determine the antidepressant-like effect of ginseng extract G115 in mice using forced swimming test. Alteration of serum corticosterone levels in mice was also investigated. Mice were divided into 6 groups receiving either water (10 ml/kg), amitriptyline (10 mg/kg) or ginseng extract G115 (100, 200, 400 or 800 mg/kg) orally for 8 d. Forced swimming test and locomotor activity were performed one day after the last treatment. Blood was collected immediately after forced swimming test to determine serum corticosterone levels. It was shown that repeated administration of amitriptyline and ginseng extract G115 (100, 200 and 800 mg/kg) significantly decreased immobility time in forced swimming test ($p<0.001$, 0.01, 0.05 and 0.001, respectively) while there was no effect of amitriptyline and ginseng extract G115 on mice locomotor activity. The results indicated that ginseng extract G115 has antidepressant-like effect in mice. In addition, ginseng extract G115 (800 mg/kg) significantly increased serum corticosterone ($p<0.05$). The result suggested that alteration of serum corticosterone is one of the mechanisms of antidepressive action of ginseng extract G115 as an adaptogen. The alteration of brain amines and brain-derived neutrophic factor (BDNF) levels should be further investigated.

Keywords: Ginseng extracts G115, Depression, Corticosterone, Forced swimming test, Mice

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IN VITRO ANTIOXIDANT, ANTIMICROBIAL AND CYTOTOXIC POTENTIAL OF THE ESSENTIAL OIL OF TEUCRIUM STOCKSIANUM BIOSS

S. M. M. SHAH[1,2]*, S. KHAN[1]

[1]Department of Pharmacy, University of Malakand, Chakdara Dir, Khyber Pukhtoon Khawa (KPK), Pakistan, [2]Department of Pharmacy, Sarhad University of Science and Information Technology, Khyber Pukhtoon Khawa (KPK), Pakistan
Email: shahpharmacist@yahoo.com

ABSTRACT

Essential oils are most frequently used in a variety of cosmetics, food and medicinal preparations. Essential oil of Teucrium stocksianum exhibited profound antinociceptive potential. The current study aimed to explore the antioxidant, antimicrobial and cytotoxic potentials of the Essential oil of Teucrium stocksianum. Hydro-distillation process yielded pale yellowish aromatic essential oil. The essential oil was dried with anhydrous Sodium sulfide (Na2SO4) and was stored in refrigerator at 4 °C before analysis using GC-MS. The chromatogram confirmed the presence of 26 chemical constituents. The main constituents, observed were δ-cadinene (12-pinene.92%), (10.3%), myrcene-caryophyllene (8.64%), (8.23%), germacrene β D (5.18%), limonene (2.36%), and elemol (2.13%). Radical scavenging ability of the essential oil of T. stocksianum was ascertained by using stable free radical of 2, 2-Diphenyl-1-picrylhydrazyl (DPPH). Essential oil exhibited profound antioxidant activity in a concentration dependent manner. Maximum activity (84.33 µg/ml, IC50; 28.5 µg/ml) was observed at100 µg/ml. The essential oil demonstrated moderate to excellent antibacterial potential against both Gram positive and Gram negative bacterial strains in well diffusion method. Pseudomonas aeruginosa has shown marked (MIC, 250 µg/ml) susceptibility to the constituents of the essential oil followed by Bacillus cereus and Escherichia coli (MIC, 450 µg/ml each). In cytotoxic assay the test sample indicated moderate lethality (LC50, 1200 µg/ml) against brine shrimp. Maximum activity (62%) was observed at 2000 µg/ml. Our observations suggest that the essential oil of Teucrium stocksianum possesses marked antioxidant, broad spectrum antimicrobial and moderate cytotoxic potential.

Keywords: Teucrium stocksianum, Essential oil, Antioxidant, Cytotoxic, Antibacterial

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DAILY INTAKE OF PINEAPPLE HONEY REDUCED OBESOGENIC PARAMETERS AND ADIPOCYTOKINES LEVEL IN OBESE MALE SPRAGUE DAWLEY RATS INDUCED WITH HIGH FAT DIET

S. SAMAT[1,2], Z. ESHAK[2], F. N. HUSSEIN[3], Z. ADAM[4], W. I. WAN ISMAIL[1,2]*

[1]Clinical BioPharmaceutical Research Group (CBRG), Pharmaceutical and Life Sciences Community of Research, [2]Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [3]Faculty of Veterinary Medicine, Universiti Putra Malaysia, Serdang, Selangor, Malaysia, [4]Medical Technology Division, Malaysian Nuclear Agency, Kajang, Selangor, Malaysia
Email: waniryani@gmail.com

ABSTRACT

Obesity is one of chronic public health problems affecting people globally. It alters adipocyte-derived hormones or cytokines expression, which promotes obesity-linked disorders such as type 2 diabetes, dyslipidemia, and cardiovascular diseases such as hypertension and atherosclerosis. Apart from drug treatment, healthy diet such as daily intake of honey may solve the problem. Pineapple honey, a local honey in Malaysia, may provide beneficial effects in controlling obesity. Obesogenic parameters and adipocytokine secretion in high fat diet-induced obese male Sprague Dawley rats fed with pineapple honey for eight weeks were investigated. The results on obesogenic parameter showed that weight gains of obese rats were significantly reduced and their food intakes were not affected. The adiposity and Lee's index's, total cholesterol level, triglycerides and glucose in the honey treated group decreased significantly compared to high fat diet (HFD) group alone. Furthermore, the results obtained from adipocytokines level showed a significant decrease in leptin and resistin concentrations in pineapple honey treated group when compared to HFD group. In conclusion, administration of pineapple honey could decrease obesity related parameters and regulate adipocytokine secretion in order to suppress high fat diet-induced obesity. The findings from this study could suggest the potential of honey in controlling obesity and associated complications when consumed daily.

Keywords: Adipocytokines, Obesity, High fat diet, Honey, Rats

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EFFECT OF PERSEA AMERICANA FRUIT PUREE AS FAT REPLACER ON THE MICROSTRUCTURE CHARACTERISTICS OF CUPCAKE

A. M. MARINA*, H. N. HADAINA

School of Health Sciences, Health Campus, Universiti Sains Malaysia, Kubang Kerian, Kelantan, Malaysia
Email: mareena@usm.my

ABSTRACT

The consumption of cupcake is increasing over the years. Thus, there is an opportunity to further improve the nutrient contents of cupcakes. Avocado or Persea Americana is an edible fruit that contains a variety of essential nutrients such as monounsaturated fatty acids, phytochemicals, phenolics and phytosterols. The purpose of this study was to investigate the microstructure characteristics of chocolate cupcakes using avocado fruit puree as partial replacement for fat in the cake formulation. The cakes were prepared by replacing 0%, 25% and 75% of butter with local avocado fruit puree. The analysis of the crumb structure was performed by scanning the cake crumbs using a scanner with 200 dpi of resolution. The scanned images were analysed using Image J software programme. Scanning electron microscope (SEM) was used to study the micrograph structure of the cake crumbs. There was no significant difference ($P>0.05$) between the number of cells, average cell sizes, total area of cells and cell density of chocolate cupcakes' crumb. The scanning electron microscopy shows that as fat replacement increased, the pores were larger with less consistency in their arrangement. The protein matrix forms no continuous sheet but rather discontinuous and ruptured structure. With fat replacement, more gelatinized starches were scattered on the crumb. The higher water content contributed by the avocado puree aided in the gelatinization process in the fat-replaced samples. In conclusion, partial fat replacement in chocolate cupcakes with avocado fruit puree was possible without compromising the morphological characteristics of the cakes.

Keywords: Avocado, Cake, Microstructure, Scanning electron microscopy

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RAW HONEY IN MALAYSIA: PROPERTIES, QUALITY CONTROL AND SENSORY ACCEPTANCE

M. BASIR[1,2], W. I. WAN ISMAIL[1,2]*

[1]Clinical Bio Pharmaceutical Research Group (CBRG), Pharmaceutical and Life Sciences Community of Research, [2]Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: waniryani@gmail.com

ABSTRACT

Malaysia is rich with diversity of flora which provides incredible nectar sources for bee to produce various types of honey including honey from wild harvesting and beekeeping industry. However, most of the honeys available in the market are already processed with heat treatment to extend the sensory quality. In this study, five types of raw honey were selected where wild harvesting honey (Gelam and Tualang honeys) were collected from the forest at Marang, Terengganu and Tasik Pedu, Kedah respectively while beekeeping honey (Acacia, Pineapple and Kelulut honeys) were all obtained from the bee farms in Johor. Manuka honey was purchased at the local store for comparison. Selected physicochemical parameters, antioxidant and nutritional properties were investigated. All honey samples had low pH ranging from 3.64 to 4.21. Moisture content ranged from 19.4–34.0 %. In terms of colour, Pineapple honey was the lightest (34 mm Pfund) and Kelulut was the darkest (150 mm Pfund). Electrical conductivity was as low as 0.299–2.003 mS/cm. Hydroxymethyl furfural (HMF) content were all below 80 mg/kg. All honeys showed good antioxidant capacity using DPPH assay (32.43-69.208 %) and total phenolic content (TPC) (132.98–446.27 mg GAE/kg). Nutritional content of these honeys were quite low including ash (0.09–1.07 g/100g), protein (0.05–0.14 g/100g) and vitamin C (0.16–1.51 mg/100g). The keeping quality study showed that there were an increase in moisture and TPC but a decrease in pH of honey samples stored in the refrigerator for six months. Acacia honey was the most preferred in terms of sensory. Physicochemical, antioxidant and nutritional properties of the honeys were quite comparable. Quality was well preserved under low temperature and it could be envisaged high demand of Acacia honey by the consumers.

Keywords: Antioxidant, Nutritional, Physicochemical, Raw honey, Sensory acceptance

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CONSUMPTION OF FRUITS AND VEGETABLES, AND THE LEVELS OF ANTIOXIDANT ENZYMES IN DOWN SYNDROME INDIVIDUALS

M. MAHAMOOD[1,2]*, A. ADAM[1,2], M. H. HASAN[1,2], M. ALIMUKHTI[1], C. SUNGGIP[3], A. MARZUKI[4], W. Z. WAN NGAH[5]

[1]Faculty of Pharmacy, [2]Research on Affinity, Safety and Efficacy Studies (OASES), Brain and Neuroscience Communities of Research, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [3]School of Medicine, Universiti Malaysia Sabah, Sabah, Malaysia, [4]Faculty of Pharmacy, Cyberjaya University College of Medical Sciences, Cyberjaya, Kuala Lumpur, Malaysia, [5]Department of Biochemistry, Faculty of Medicine, Universiti Kebangsaan Malaysia, Kuala Lumpur, Malaysia
Email: maziana_mahamood@yahoo.com

ABSTRACT

Down syndrome (DS) is a common chromosomal abnormality occurring in about 1 in 700 live births. Neuromotor deficiency in DS was reported and influencing the chewing frequency and therefore, the food choices for DS persons was limited. Previous studies in epidemiological, *in vitro* and *in vivo* showed that oxidative stress is elevated in DS individuals. The aim of this study was to determine the correlation between fruits and vegetables consumption with the activities of antioxidant enzymes in DS. The frequency of fruits and vegetables consumption by individuals with DS and the influence of variations in the level of this consumption on biomarkers of oxidative stress were reported by the parents or caregivers through questionnaires during the interview sessions. The questionnaire included a checklist of the different types of fruits and vegetables, portion sizes and frequency intake. Blood sample was collected with informed consent from the participants and their parents or guardians. Activities of antioxidant enzymes in erythrocytes such as superoxide dismutase, glutathione peroxidase and catalase were also measured using established techniques. There was a significant correlation between the frequency of fruit consumption with the levels of catalase ($P<0.01$), showing that the action of catalase is important in H2O2 breakdown in DS. While there were no significant relationships were found between consumption of fruits and vegetables with the level of other antioxidant enzymes. Consumption of fruits and vegetables as an essential part of healthy diet was low in individuals with DS. This seemed can be related to the difficulties patients with DS report chewing. Parents of children with DS should be encouraged to give more fruit and vegetables to their children and choose soft fruits and preparation methods that make them easier to consume.

Keywords: Down syndrome, Fruits, Vegetables, Antioxidant enzymes

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EFFECTS OF SELENIUM-RICH GREEN AND BLACK TEAS ON BONE HEALTH IN YOUNG GROWING MALE RATS

N. H. NORDIN[1,2]*, W. H. CHUA[2], A. L. MOLAN[3], M. C. KRUGER[2]

[1]Faculty of Pharmacy, Universiti Teknologi MARA Pulau Pinang, Bertam Campus, Kepala Batas, Penang, Malaysia, [2]School of Food and Nutrition, Massey Institute of Food Science and Technology, College of Health, Massey University, New Zealand, [3]Department of Biology, College of Sciences, Diyala University, Diyala, Iraq
Email: noorhazarina046@ppinang.uitm.edu.my

ABSTRACT

Bone health and integrity may be greatly affected by nutrition during growth and aging. Tea consumption has been shown to be effective in minimizing age-related bone loss. However, it is unclear whether tea enhances bone mass and strength during the rapid growth phase. Currently, no information is available about the beneficial effect of selenium-rich teas on bone health. This study investigated the effect of selenium-rich green tea extract (Se-GTE) and selenium-rich black tea extract (Se-BTE) on bone mass using a young growing rat model. The effect of four-week's tea consumption on bone mass and strength was examined in young growing male Sprague-Dawley rats. Four-week-old rats were randomised into three groups: control (deionised water), Se-GTE (1%), and Se-BTE (1%). Rats were fed standard rat chow pellets (0.5% calcium), and provided with deionised water or tea water extracts, ad libitum throughout the study period. At the end of the study, animals were euthanased and blood serum, femurs and the lumbar spine were collected. The serum bone resorption biomarker C-telopeptide of type I collagen (CTx-I), was analysed. Ex vivo bone mineral density (BMD) of femurs and lumbar spine were measured using dual energy X-ray absorptiometry. Right femurs were tested biomechanically using the three-point bending test. There were significant higher fluid and feed intake in Se-BTE rats than the control rats ($p = 0.0002$ and $p = 0.0024$, respectively). Even though serum CTx-I concentrations ($p = 0.0459$) and some parameters of bone mass and strength differed significantly among all groups, rats given Se-GTE and Se-BTE showed no significant differences relative to the control rats. In conclusion, Se-GTE and Se-BTE did not significantly improve the bone parameters evaluated in the present study. A longer tea intervention could possibly allow these tea extracts to exert significant effect on bone parameters.

Keywords: Selenium-rich green tea, Selenium-rich black tea, Peak bone mass, Bone, Young growing rats

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ANTIDEPRESSANT EFFECT OF COMMERCIAL OLIVE OIL ON DEPRESSION INDUCED MICE MODEL

Z. YUSUF*, A. S. RUSLAN, S. ABDUL RAHMAN

Faculty of Pharmacy, Department of Pharmaceutical Sciences, Cyberjaya University, College of Medical Sciences, Cyberjaya, Selangor, Malaysia
Email: zauyah@cybermed.edu.my

ABSTRACT

Olive oil is a popular Mediterranean diet and it is widely used in cooking throughout the world. It is believed to contain many health benefits and many reported research done have actually proved the health benefits of consuming olive oil regularly on daily basis. This study is designed to determine any possibility of antidepressant effect of commercial olive oil on depression induced mice model. The olive oil used are of 3 different brands that can easily be found in Malaysian market. A total of 80 male mice were divided into 5 groups (n=8) which received different treatment. A total of 40 mice were tested using tail suspension test (TST) and another 40 mice using for forced swimming test (FST). All treatments were given by force feeding method one h prior to testing. Group 1, the negative control group, were stress induced but without any treatment given. Group 2, the positive control group, was treated with the dose of 25 mg/kg fluoxetine hydrochloride. Groups 3, 4 and 5 were treated each with Palestine olive Oil, Minsyam Olive Oil, and Colavita Olive Oil, respectively, with the dose of 10 ml/kg. The same groupings and treatments were conducted for TST and FST. The time duration of mice being immobile was recorded and analysed. Reduced immobility time indicated antidepressant effect. The result of the experiment revealed that the immobility time was reduced significantly in FST for all olive oil groups. However, there was no significant reduction of immobility time in TST. In conclusion, olive oil showed some potential to become an alternative to antidepressant medications. Further research needs to be done to confirm the antidepressant effect of olive oil.

Keywords: Olive oil, Antidepressant, tail suspension test, Forced swimming test

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TUALANG HONEY MITIGATES OVALBUMIN-INDUCED AIRWAY INFLAMMATION IN A MICE ALLERGIC ASTHMA MODEL

N. S. S. SHAMSHUDDIN[1], R. MOHD ZOHDI[1,2]*

[1]Wound Healing Research Group (WHRG), [2]Atta-ur-rahman Institute, Universiti Teknologi MARA, Puncak Alam, Selangor, Malaysia
Email: rozainizohdi@puncakalam.uitm.edu.my

ABSTRACT

Allergic asthma is a chronic inflammatory disorder of the pulmonary airways. Administration of an anti-inflammatory agent is an important way to reverse the effect of inflammation and one of the well-known natural products that possess this property is honey. Thus, this study aimed to investigate the effect of Tualang honey on immunomodulatory and histological changes in the lungs of a mice model of allergic asthma. Forty female ICR mice were divided into four groups. All mice except the control group (Group 1) were sensitized and challenged with ovalbumin (OVA) via intraperitoneal and intranasal routes. Group 2 and 3 were administered with 80% Tualang honey and 3 mg/kg dexamethasone (DXN) respectively, via oral gavage once a day for 5 d of the challenged period. The negative control group (Group 4) received no treatment. Mice were sacrificed 24 h after the last OVA challenged and histological parameters such as tissue inflammation and mucin production were assessed. Our data showed that Tualang honey significantly inhibited inflammatory cell infiltration and mucus overproduction compared with the OVA-induced group. Also, Tualang honey significantly reduced the production of ovalbumin-specific IgE in serum and beta hexosaminidase in bronchoalveolar lavaged fluid (BALF) comparable to dexamethasone. In conclusion, our results suggested that Tualang honey administration attenuates the pathological changes of mice model of allergic asthma.

Keywords: Honey, Ovalbumin, Allergic asthma, Dexamethasone

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MUSHROOM ENDOPHYTIC FUNGI AS A SOURCE OF BIOACTIVE METABOLITES

F. LEMBUK*, N. MANSHOOR, S. SULTAN, J-F. F. WEBER

Atta-ur-Rahman Institute for Natural Product Discovery (AuRiND), Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: fatma_potter@yahoo.co.uk

ABSTRACT

A mushroom is the fruiting body of a fungus. It harbours endophytic bacteria and fungi. Host-endophyte communication utilises secondary metabolites, thus intrinsically bioactive. Fungi are known as a great source of new bioactive metabolites. They serve a wide range of application as antimicrobial agents, anticancer agents, antioxidants, immunosuppressants, etc. In this study, we are focusing on secondary metabolites produced by endophytic fungi of mushrooms collected from Sungai Chilling, Kuala Kubu Bharu, and Selangor. Endophytic fungi were isolated and growth rate estimations of each fungus were obtained by measuring the colonies size on agar in standardised culture condition. The fungi were cultured in 12 different media. Half amount of the culture extracts were pooled and analyzed by high performance liquid chromatography (HPLC). The culture extracts were subjected to preliminary screening for antimicrobial activity against Escherichia coli ATCC 25922, Pseudomonas aeruginosa ATCC 27853, Staphylococcus aureus ATCC 6538, Bacillus subtilis ATCC 6633, Candida albicans ATCC 10031 and Aspergillus niger ATCC 16404. Another half amount of the extracts were extracted by solid phase extraction for HPLC profiling. The best media were chosen for large scale cultures. The large scale cultures were extracted by standard operating procedure. Bioactive crude extracts were separated by preparative HPLC and purified by recycling PLC. The isolated compounds were elucidated by Nuclear Magnetic Resonance (NMR), mass spectrometry (MS) and infra red (IR) analyses. The species of mushrooms and endophytic fungi obtained were identified. 103 endophytic fungi were isolated from 22 species of mushrooms. Growth rates classification; 39 fast, 38 medium and 26 slow. Bioactive crude extracts towards selected microorganisms; Staphylococcus aureus–9, Escherichia coli–3, Candida albicans–26, Aspergillus niger–20, Bacillus subtilis–19, Pseudomonas aeruginosa–31. Endophytic fungi grown in different media gave different HPLC profiles. Mushroom endophytic fungi grown in different media may give different types of compounds.

Keywords: Antimicrobial activity, Endophytic fungi, Secondary metabolites

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THE ANTIDIABETIC ACTIVITIES OF PERESKIA BLEO LEAVES EXTRACTS

A. N. ABDUL RANI[1]*, N. AMRAN[1], R. MAHMUD1, M. Z. ASMAWI[2]

[1]Department of Pharmaceutical Chemistry, [2]Department of Pharmacology, School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang, Malaysia
Email: anisnajwa_abdulrani@yahoo.com.my

ABSTRACT

Pereskia bleo which belongs to the family Cactaceae is a well-known traditional remedy in Malaysia for the treatment of diabetes, cancer, high blood pressure and diseases associated with rheumatism and inflammation. This study investigated the hypoglycaemic effects in normal rats, the intraperitoneal glucose tolerance test (IPGTT) in normal rats and the antihyperglycemic effects in streptozotocin (STZ)-induced diabetic-rats of the crude leaf plant extracts. Four different extracts of differing degrees of polarity of P. bleo leaf are selected namely petroleum-ether extract, chloroform extract, methanol extract and aqueous extract respectively. The hypoglycaemic effect was measured by the ability of the extracts to lower blood glucose level to below the normal level of normal rats treated with 1000 mg/kg extracts following 7-h study. All extracts were found to be insignificant compared to the control group while the positive control glibenclamide, a clinical diabetic drug was found to be significant ($p<0.05$). For the IPGTT, the effect was measured by the ability of the extracts to lower the blood glucose level in normal rats upon intraperitoneally glucose loading. None of the extracts showed significant effect compared to control while the positive control metformin, a clinical diabetic drug has shown significant ($p<0.05$) compared to negative control. In antihyperglycemic test, the diabetic type-2 rat was induced by injecting 50 mg/kg of STZ. The effect was measured by the blood glucose level in diabetic rats treated with 1000 mg/kg extracts two times daily for 12 d. The antihyperglycemic effect was found to be significant ($p<0.05$) in the positive controls, as early as day 3 while aqueous extract showed significant effect on day 6 and continued to be consistent throughout the study. The petroleum-ether and chloroform extracts only showed significant effects on the last day of the treatment while methanol extract showed insignificant effect throughout the study. These results clearly suggest that the aqueous extract of P. bleo is effective in lowering the blood glucose level in STZ-induced diabetic rats.

Keywords: Pereskia bleo, Hypogycaemic, Glucose tolerance test, Antihyperglycemic

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PHENOLIC COMPOUNDS FROM DIPTEROCARPUS SEMIVESTITUS

R. RAMLI, N. MANSHOOR*, N. ISMAIL

Atta-ur-Rahman Research Institute for Natural Product Discovery, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: nurhuda15@salam.uitm.edu.my

ABSTRACT

Identification of phenolic compounds from Dipterocarpus semivestitus acetone stem extract was conducted using de replication strategy; a strategy to differentiate unknown compound from the known. Known compound was identified prior to the fractionation process using liquid chromatography tandem mass spectrometry. Stem extract was injected into LC-MS system and MS data were collected for each compound. Two compounds, hopeaphenol and hemsleyanol D, were identified by comparing their mass fragmentation patterns with in-house library. α-viniferin and a coumarin, scopoletin were isolated and since their MS data were not available in the library, its structure was confirmed by NMR analyses and comparison with published data.

Keywords: Dipterocarpaceae, Dipterocarpus semivestitus, Resveratrol oligomers, Tandem MS, Dereplication

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THE EFFECT OF ZINGIBER ZERUMBONE IN ATTENUATING SEROTONIN INDUCED ITCH IN MICE

K. RAJENDRAN, E. P. PERIMAL, M. R. SULAIMAN, A. AKIRA*

Faculty of Medicine and Health Sciences, Department of Biomedical Sciences, Universiti Putra Malaysia, Serdang, Selangor, Malaysia
Email: ahmadakira@upm.edu.my

ABSTRACT

Itch, also known as pruritus, is a sensation that causes the desire to scratch. It serves an essential physiologic function. However, when it becomes chronic, it affects one's quality of life. Zingiber zerumbet Smith is type of ginger found and used in Southeast Asia and is a natural remedy for minor aches, rashes and itchiness. The objective of this experiment was to investigate the antipruritic effects of EOZZ in a murine model of serotonin induced itch. Studies were conducted upon 6-8 w old male ICR mice. The mice were divided into 10 groups (n=6). Mice were treated via intraperitoneal administration of EOZZ at dosages of 30, 100, 300 (mg/kg). Pruritus was then induced by subcutaneous injections (10 uL) of pruritogenic agent serotonin, into the rostral back. Observation of scratching of the injected site by mouse hind-limb for a 30 min period was taken as indices of pruritus. Oxatomide acted as a positive control group whereas vehicle was used as a negative control. This study found that EOZZ could significantly suppress the pruritus induced by serotonin (percentage of inhibition by EOZZ: 30 mg/kg-36%, 100 mg/kg-82%, 300 mg/kg-92%) in the dose dependant manner. The results suggest that EOZZ might be effective in treating pruritus especially itch that is not histamine related. Further research is required to study the mechanism of action of EOZZ in pruritus.

Keywords: Pruritus, Essential oil of Zingiber zerumbet (EOZZ), Antipruritic

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MAHANIMBINE IMPROVES COGNITIVE FUNCTION VIA ELEVATING CHOLINERGIC ACTIVITY IN AGED MICE

N. S. MOHD AZAHAN[1,2], V. MANI[1,2]*, K. RAMASAMY[1,3], A. PRAKASH[1,2], A. B. ABDUL MAJEED[1,2]

[1]Faculty of Pharmacy, [2]Brain Degeneration and Therapeutics Group, [3]Collaborative Drug Discovery Research Group, Pharmaceutical & Life Sciences CoRe, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: vasudevan@puncakalam.uitm.edu.my

ABSTRACT

Aging is a complicated multifactorial progression that refers to a continuing slow decline in the physiological function of an organism. Aged rodents have shown age-related impairment of dynamic aspects in cholinergic neurotransmission. In fact, the cholinergic neurotransmission plays an important role in monitoring the transitions toward more vigilant brain that is involved in learning, memory and neuropathology of brain. Concentration of acetylcholine (ACh) in the central nervous system attempts to improve memory. Meanwhile, the acetylcholinesterase (AChE) enzyme relates to loss of cortical cholinergic neurotransmission. The inhibition of AChE activity enhanced cholinergic neurotransmission by increasing the availability of ACh in the synaptic cleft. Mahanimbine is a major bioactive carbazole alkaloid from Murraya koenigii leaves. Numerous reports are supporting the precise role of various alkaloids in enhancing the learning and memory process. The present study was designed to investigate the potential of mahanimbine from Murraya koenigii leaves, in aging mouse models for improving the memory. Thirty male ICR mice were divided randomly into five groups (n=6): control young, control aging, piracetam and mahanimbine (1 and 2 mg/kg, p. o.). The entire groups were administered orally for 30 d consecutively. Morris Water Maze (MWM) test was conducted on the 27th day of treatment to evaluate cognitive functions. Based on MWM results, treatment with mahanimbine significantly enhanced the memory in aging mice evidenced by a decrease in escape latency and escape distance to reach the hidden platform. Furthermore, mahanimbine was able to boost the memory consolidation based on the probe test during MWM task. Moreover, pre-treatment with mahanimbine improved the central cholinergic transmission by increasing the ACh level through the inhibition of AChE as compared to aging control. As a conclusion, mahanimbine can enhance the memory in aging mice by improving the cholinergic neurotransmission in the brain.

Keywords: Dementia, Mahanimbine, Morris water maze, Acetylcholine, Acetylcholinesterase

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CYTOTOXICITY EFFECT OF MARINE ENDOPHYTIC FUNGI FROM MALAYSIAN SEAWEED AGAINST HepG2 AND HeLa

H. WAHID*, S. A. ARIFFIN

Marine Pharmaceutical Research Group (MaReG), Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: hmayumi@sirim.my

ABSTRACT

Liver cancer is major concern which accounts for millions of deaths annually. Liver cancer seems to be resistance against current available synthetic drugs. Previous study reported that chemotherapeutic effects of endophytic fungi-based resulted acute efficiency against many type of cancer cell lines. The objective of this present study was to investigate the effect of methanol extracts isolated from Malaysian seaweeds against cancer cell, human hepatocellular carcinoma (HepG2) and non cancerous cell, human epithelial normal liver (HeLa). In the present investigation, five species of marine endophytic fungi from Gracilaria coronopitolia coded as CN, Gracilaria arcuata zanardin coded as MV, Acanthophora picifera (M. Vahl) Borgesen coded as ED1, Caulerpa sertularioides coded as CS and Chaemotorpha minima coded as UF, were grown on potatoes dextrose agar (PDA) at two different salinity (1% and 3%) of artificial sea salt (ASS). HepG2 and HeLa cells were exposed to methanol extracts with different concentrations at 24, 48 and 72 h, and viability of cells were determined by (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT) agent. Amongst the 10 extracts, CN1-1%, CN-3% and MV-3% induced cytotoxicity in HepG2 cells after 24 h exposure, with IC50 values of 36.7±2.9 µg/ml, 38.3±5.8 µg/ml, and 45.0±5.0 µg/ml respectively, however non-toxic to the HeLa. MV species grown in higher salinity of 3% AAS showed potent cytotoxicity effect compared to 1% AAS. This preliminary data suggest that CN and MV species of marine endophytic fungi from Malaysian seaweeds have a potential in cytotoxicity against HepG2 without toxic to the HeLa. However, further investigation is needed to explore this potential bioactive as chemotherapeutic agent against liver cancer cell.

Keywords: Cytotoxicity, Marine endophytic fungi, Liver cancer, HepG2, HeLa

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MICROBIAL TRANSFORMATION OF ETHYNODIOL DIACETATE AND MEDROXYPROGESTERONE BY A SERIES OF FUNGI

S. N. WAN YUSOP[1,2]*, J-F. F. WEBER[1,2], L. K. TEH[1,3], S. A. A. SHAH[1,2], S. SULTAN[1,2]

[1]Faculty of Pharmacy, [2]Atta-ur-Rahman Institute for Natural Product Discovery (RiND), [3]Integrative Pharmacogenomics Centre (iPROMISE), Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: sharifahnurfazilah@hotmail.com

ABSTRACT

Natural products continue to be the top sources of drugs and drug leads, and this remains true today. Some of these are sourced from fermentation of microorganisms. Microbial transformation is one of the developments in microbial natural product discovery. It is a method of chemical alteration of organic compounds that are too complex to be achieved through conservative chemical techniques. Enzyme configurations in the biological systems of microorganisms enable region-selective and stereospecific chemical transformations. Microbial transformation also promotes the growth of sustainable technologies for the production of chemicals and drugs, which means green chemistry. In the present work, biotransformations of ethynodiol diacetate and medroxyprogesterone have been investigated with a series of fungi from different biotopes. Screening experiments were performed in 100 ml conical flasks containing 40 ml media and were autoclaved at 121 °C for 15 min. sterile culture media were inoculated with a collection of fungi. After 3-5 d of inoculation, substrates were introduced aseptically into fermented liquid media (0.5 mg/40 ml media) and further fermentations were allowed for 4-12 d. Preparative scale started upon detection of bio-transformed products. Metabolic changes were observed by comparing the HPLC chromatograms of starting compounds, control cultures and fermented extracts. Botrytis cinerea was selected for large scale fermentation of ethynodiol diacetate. Mucor plumbeus was selected for large scale fermentation of medroxyprogesterone. Incubation of ethynodiol diacetate with B. cinerea for twelve days afforded one new and two known metabolites. The new metabolite was characterized as 17α-ethynyl-17β-acetoxyestr-4-en-3-one-15β-ol. Incubation of medroxyprogesterone with M. plumbeus (large scale) for eight days afforded two major peaks and several additional peaks of suspected bio-transformed products. Complete spectrometric analysis will be performed to verify the structures of transformed products. Ethynodiol diacetate and medroxyprogesterone are successfully transformed by Botrytis cinerea and M. plumbeus, respectively.

Keywords: Microbial transformation, Ethynodiol diacetate, Medroxyprogesterone, HPLC, Spectrometric analysis

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

F. B. M. HUSSAIN[1,2], S. SULTAN[1,2], J-F. F. WEBER[1,2], K. RAMASAMY[3], N. MANSHOOR[1,2]

[1]Faculty of Pharmacy, [2]Atta-ur-Rahman Institute for Natural Product Discovery (AuRIns), [3]Collaborative Drug Discovery Research (CDDR), Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: fati_mah2000@yahoo.com

ABSTRACT

Plant endophytes microorganisms are an important and novel resource for natural bioactive compounds. In the past two decades, many valuable bioactive compounds with antimicrobial, cytotoxic and anticancer activities have been successfully discovered from endophytic microorganisms. Studying microorganisms from a variety of biotopes increase the chances to isolate novel bioactive metabolites. For this reason, extremophiles have always been deemed as endowed with a greater potential for drug discovery than microorganisms from "normal" ecological niches. In this respect, psychrophiles from the Arctic fulfill the criteria of an interesting source of potential new drugs. In this research, a total of 23 endophytic fungi (13 psychrophilic and 10 psychrotrophic fungi) were isolated from leaves, stems, and flowers of 8 plants collected from Longyearbyen, Svalbard Island, Norway. The objective of this study was to isolate secondary metabolites from cultures of the above fungi. Each culture was fermented and extracted using ethyl acetate based on our standard operation procedure. The crude extracts were screened for antibacterial activity against Staphylococcus aureus ATCC 25923, Enterococcus faecium ATCC 51585, Pseudomonas aeruginosa ATCC 27853 and Escherichia coli ATCC 25922, using the MTT method. The crude extracts showing significant results were analysed and fractionated by high performance liquid chromatography (HPLC). Pure compounds were subjected to standard spectrometry analyzes and their structures established.

Keywords: Arctic, Psychrophilic fungi, Psychrotrophic fungi, Antibacterial, HPLC, LC/MS, NMR

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HISTOLOGICAL STUDY OF WOUNDED SKIN OF DIABETIC RATS TREATED WITH TOPICAL AQUEOUS METHANOL EXTRACT OF HIBISCUS ROSA SINENSIS LEAVES

N. Z. TAJUDDIN[1], I. T. ISMAIL[1], M. S. ABD WAHAB[1], R. MOHD ZOHDI[1], A. A. ALI [1, 2]*

[1]Faculty of Pharmacy, [2]Reseach on Affinity, Safety and Efficacy Studies, Brain and Neuroscience Communities of Research, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: aidaaz2790@puncakalam.uitm.edu.my

ABSTRACT

Wound healing involves restoration of the injured tissues to its normal function and structure. However, wound healing in diabetic patients are impaired and delayed. The present study aimed to investigate the macro-and microscopic changes of diabetic wounded skin after treated with aqueous methanol extract (MeOH) of Hibiscus rosa sinensis leaves. Fifty rats were used in the study that were randomly divided into four groups with 5 rats in the normal control group and 15 rats each in the negative control, positive control and treatment groups. Except for the rats in the normal control group, all other rats were streptozocin induced-diabetic and received a wound (2 cm x 2 cm) on the thoracolumbar spine. Topical silver sulphadiazine served as the treatment in the positive control group, while wounded rats in the negative control group were left untreated. The treatment group received 0.3 ml of 10 mg/kg MeOH extract. The treatment was provided daily. On the 3rd, 7th and 14th day of the study, a group consisting of five rats from the positive and negative control and the treatment groups were sacrificed. Our findings showed that rats that were treated with Hibiscus rosa sinensis MeOH extract showed significant improvement in wound closure in addition to absence of exudates or signs of infection. Histologically, improved structural changes by an elevation of well-arranged collagen fiber and increment of fibroblast accumulation were noted in the treatment group. As a conclusion, Hibiscus rosa sinensis MeOH extract may improve wound healing in diabetic rats, and may have potential as an alternative treatment for wound healing.

Keywords: Hibiscus rosa-sinensis, Wound healing, Diabetic, Excision wound, Aqueous methanol extract

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TENSILE STRENGTH AND COLLAGEN CONTENT EVALUATION OF WOUNDED SKIN OF DIABETIC RATS FOLLOWING TOPICAL TREATMENT WITH HIBISCUS ROSA-SINENSIS LEAVES AQUEOUS-METHANOLIC EXTRACT

I. T. ISMAIL[1], N. Z. TAJUDDIN[1], M. S. ABD WAHAB[1], R. MOHD ZOHDI[1], A. A. ALI[1,2]*

[1]Faculty of Pharmacy, [2]Reseach on Affinity, Safety and Efficacy Studies, Brain and Neuroscience Communities of Research, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: aidaaz2790@puncakalam.uitm.edu.my

ABSTRACT

The effects of the aqueous methanol extract of Hibiscus rosa-sinensis leaves on tensile strength and collagen content of wounds on diabetic male Wistar rats were investigated. Fifty rats were randomly divided into four groups: (1) normal control; (2) negative control group with no treatment; (3) positive control group with topical silver sulfadiazine treatment; and (4) treatment group that received topical treatment of 0.3 ml of 10 mg/kg aqueous methanol (MeOH) extract. Diabetes in the rats was streptozotocin-induced. A 4-cm2 wound was created on each rat except for those in the normal control group. The treatment was given daily. A group consisting of five rats from the positive and negative control and the treatment groups were sacrificed on the 3rd, 7th and 14th day of the study. The skin around the wound was taken for collagen content and tensile strength measurement. Our findings showed a significant increase in tensile strength and collagen content in rats treated with topical aqueous methanol extract compared to those treated with silver sulfadiazine and those that received no treatment. As a conclusion, a topical application of aqueous methanol extract of Hibiscus rosa-sinensis leaves may improve wound healing in diabetic rats.

Keywords: Hibiscus rosa-sinensis, Wound healing, tensile strength, Collagen content, Excision wound

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ANTI-ANGIOGENIC EFFECT OF ARDISIA CRISPA ROOT IN HUMAN UMBILICAL VEIN ENDOTHELIAL CELLS (HUVECS)

W. J. LIM, R. ABD HAMID*, P. F. CHAN

Department of Biomedical Science, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia, Serdang, Selangor, Malaysia
Email: roslida@upm.edu.my

ABSTRACT

Angiogenesis, the growth of new blood vessels from the existing one, is part of the crucial events required for tumor progression, where the growth is dependent on vascular induction. Ardisia crispa is an evergreen shrub widely found across Asia and traditionally used by the locals for medicinal purposes. Previous studies revealed that the hexane fraction of the plant's root (ACRH) and the isolated benzoquinone compound (AC2) can suppress murine skin tumorigenesis, inflammation-induced angiogenesis and possess anti-inflammatory, anti-hyperalgesic and anti-ulcer properties *in vivo*. This study aimed to investigate the anti angiogenic effect of ACRH and AC2 on human umbilical vein endothelial cells (HUVEC) in various *in vitro* assays. After 24 h exposure to ACRH and AC2 (1.53-100µg/ml), HUVEC growth reduced at all tested concentrations with the IC50 of 1.21µg/ml and 0.95µg/ml for ACRH and AC2, respectively. Cell cycle distribution analysis revealed that most cells were arrested at G0/G1 phase (64.14±1.30%), followed by S phase (18.84±0.79%) and G2/M phase (6.12±0.51%) without significant difference across all groups. ACRH and AC2 (0.1-100µg/ml) also showed significant inhibition of cell migration in wound healing assay in independent concentration manner. Cell invasion assay was performed using Matrigel coated invasion chamber. The number of invaded cell were reduced ranging from 18.2 up to 97.0% across all tested concentrations of ACRH and AC2 in comparison with the negative control group ($p<0.001$). In tube formation assay, the total tube length was significantly reduced about 30% when treated with the lowest concentration (0.1 µg/ml) of both ACRH and AC2. Whilst, at higher concentrations (1, 10 and 100 µg/ml), both treatments achieved total inhibition when compared with negative control group ($p<0.001$). Further studies are still underway to elucidate their potential pathway, but meanwhile, both ACRH and AC2 have shown their anti-angiogenic potential via these preliminary observations.

Keywords: Angiogenesis, Ardisia crispa, Benzoquinone

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β-CARYOPHYLLENE, A NATURAL COMPONENT OF AGARWOOD: INHIBITS GROWTH AND METASTASIS OF HUMAN COLORECTAL CANCER *IN VITRO* AND *IN VIVO*

S. S. DAHHAM[1], A. H. IBRAHIM[1,2], S. S. AL-RAWI[1,2], A. M. S. ABDUL MAJID[1]

[1]School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang, Malaysia, [2]Biology Department, University of Zakho, Kurdistan Regin, Iraq
Email: hawk_dijla@yahoo.com

ABSTRACT

Colorectal cancer (CRC) is one of the most lethal malignant diseases which remain a rampant killer across the world. Toxic drugs have failed to reduce the mortality and morbidity rate of CRC patients, thus new agents that are safe, available and effective are urgently needed. In our previous communications, we reported that Aquilaria crassna and its active principle β-caryophyllene (BC) possessed a broad spectrum of antiproliferative, apoptogenic and antioxidant activities. The present study focuses upon the molecular mechanism by which BC mediates its effects on cell growth and metastasis of CRC. The structure of BC was elucidated using FT-IR, 1H and 13C NMR and MS spectral. Anti-colon cancer effect was investigated on HCT116 human colorectal carcinoma cells including cytotoxicity, anti tumorigenicity and antimetastatic. In addition, xenograft model was conducted in orthotopically implanted colon cancer cells in nude mice. The mechanism of the cell death has been assessed using Human Apoptosis Antibody Array. BC significantly inhibited the proliferation of HCT116 cells with an IC50 of 6.3±1.2 µg/ml. The main three steps in tumour metastasis including cell invasion, cell migration and clonogenicity were significantly obstructed. Transmission electron microscope revealed further supportive information on the typical singe of apoptosis morphological changes including nuclear shrinkage, chromatin condensation and nuclear fragmentation. Furthermore, BC was mediated through the down regulation of apoptosis inhibitor such as XIAP, survivin and P53 in HCT 116 cells which indicated the apoptogenic properties of BC. Administration of BC (200, 100 and 50 mg/kg/day) dose-dependently inhibited the growth of colon cancer in an orthotopic model. The intensity of prosense and angiosense probes were identified by fluorescence molecular tomography (FMT). Collectively, our results revealed a new mechanism of action of β-caryophyllene and suggest that it may be a potential chemotherapeutic agent selectively against CRC.

Keywords: Agarwood, β-caryophyllene, Metastasis, Apoptosis, Colorectal cancer

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OPTIMISATION OF FUCOXANTHIN EXTRACTION FROM MALAYSIAN BROWN SEAWEEDS AND EFFECTS ON 3T3-L1 CELLS

H. AHMAD[1], I. JASWIR[2]*, D. SUSANTI[1], M. TAHER[3]

[1]Kulliyyah of Science, [2]International Institute of Halal Research and Training (INHART), [3]Kulliyyah of Pharmacy, International Islamic University Malaysia (IIUM), Kuantan, Malaysia
Email: irwandi@iium.edu.my

ABSTRACT

Over 11.6 billion of the population worldwide are overweight (BMI>25 kg/m2), and 400 million are obese (BMI>30 kg/m2). Malaysia has also seen an increase in obesity prevalence over the past few decades, with 7.7% men and 33.4% women being obese, according to the Malaysian Third National Health and Morbidity Survey. This trend is worrying as obesity has been reported as a risk factor for high blood pressure, diabetes, and cardiovascular disease, several cancers disabilities and death. Despite advances in the development of more effective weight loss drugs, safety and dependence remains an issue. Natural products have long been an alternative to drugs in the treatment of diseases. Fucoxanthin, a natural compound found in brown seaweeds, has been documented to exhibit several activities, including anti-obesity potential. This study aimed to establish the anti-obesity potential of Malaysian brown seaweeds Sargassum oligocystum, Padina australis and Dictyota dichotoma, and to optimise extraction parameters to retain maximum fucoxanthin content in the extract. Briefly, the study consisted of two major parts: firstly, optimizing the extraction process of the brown seaweeds by varying temperature, particle size and solvent to ensure high fucoxanthin yields and secondly, applying the extracts to 3T3-L1 cells to establish the adipogenic and adipolytic activity of the locally sourced brown seaweeds. Optimal parameters for extraction varied between seaweeds, while Dictyota dichotoma showed both the highest anti-adipogenic and pro-adipolytic potential when applied to 3T3-L1 cells. The results showed that under optimal conditions, fucoxanthin content in the seaweed extracts can be maximised and that local brown seaweeds Sargassum oligocystum, Padina australis and Dictyota dichotoma can be further developed as potential natural agents in the search for safer and more effective treatment for obesity.

Keywords: Brown seaweeds, Fucoxanthin, Extraction, Adipogenesis, Adipolysis

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ANTIBACTERIAL ACTIVITIES OF PYROLIGNEOUS ACID EXTRACTS FROM RHIZOPHORA APICULATA AND RHIZOPHORA MUCRONATA

N. H. UYUP*, W. M. A. WAN MOHAMAD, Y. MD YUSOF, M. L. KAMAL, S. HASAN

Fakulti Sains Gunaan, Universiti Teknologi MARA, Malaysia
Email: hafizah802@perlis.uitm.edu.my

ABSTRACT

The objective of this study was to investigate antibacterial activity of pyroligneous acid extracts of Rhizophora apiculata and Rhizophora mucronata plants against Escherichia coli, Salmonella typhimurium, Staphylococcus aureus and Bacillus subtilis. By using the paper disc diffusion method, the antibacterial activity of the pyroligneous acid extracts was evaluated by determination of the diameter of zone of inhibition against two Gram-negative (i.e. Escherichia coli, Salmonella typhimurium) and two Gram-positive (i.e. Staphylococcus aureus, Bacillus subtilis) human pathogenic bacteria. The results showed that the notable inhibition of the bacterial growth was shown against the four tested microorganisms, with the largest diameter of inhibition zone was 34 mm when evaluated against Staphylococcus aureus and Salmonella typhimurium. The significance of this investigation suggested that the pyroligneous acid extracts of these plants have the potential source as a nontoxic and natural antibacterial agent and also can be used to discover for evolving new pharmaceuticals research and invention in Malaysia.

Keywords: Pyroligneous acid, Antibacterial activity, Rhizophora apiculata, Rhizophora mucronata

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DISCREPANCY EFFECTS OF LABISIA PUMILA VAR. ALATA AQUEOUS EXTRACT VIA ESTROGEN RECEPTOR BINDING ON HUMAN BREAST CANCER AND NORMAL CELL LINES

M. F. ZULKIFLI[1,2], C. Y. CHOO[2], W. I. WAN ISMAIL[1,2]*

[1]Clinical Bio Pharmaceutical Research Group (CBRG), Pharmaceutical and Life Sciences Community of Research, [2]Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: waniryani@gmail.com

ABSTRACT

Labisia pumila var. alata has intensively consumed and investigated to reduce menopausal symptoms. It has potential to solve problems related to hormone replacement therapy (HRT) in menopause women. In contrast to the HRT, which is believed to develop cancer, the plant extract also exhibited anticancer effects by inhibiting proliferation of cancer cell lines. Mechanism of the extract in displaying both antiprolirative effects and reducing menopausal syndrome is hypothesized through the binding activity on estrogen receptor and yet to be observed in the study. Two estrogen receptor (ER)-positive cell lines, human breast adenocarcinoma cell lines (MCF-7) and human normal epithelial breast cell lines (MCF-10A) were treated with several concentrations of Labisia pumila var. alata aqueous extract using methylthiazolyldiphenyl-tetrazolium bromide (MTT) assay. The cells were then incubated for 72 h, either with or without ER-antagonist (ICI 182,780). 17 beta-estradiol (E2) and phosphate buffer saline (PBS) were used as a positive and negative control respectively. The plant extract significantly increased the proliferation of MCF-10A cell at concentration of 25%, 50%, and 100% of 1µg/ml ($p<0.001$). In contrast, it significantly exhibited antiproliferative effect to MCF-7 cell by inhibiting the cell growth at 50% and 100% of 1µg/ml ($p<0.001$) with the IC50 value of 94.42. E2 displayed significant proliferation effects on both cell lines. Meanwhile, ER-antagonist blocked both cell proliferation induced by E2 and Labisia pumila var. alata respectively indicating estrogen receptor involved in the mechanism. Interestingly, estrogen receptor binding activity by the plant extract in different cell lines demonstrated divergent effects i.e. inhibited cancer cell growth (MCF7) but increased proliferation of normal cell (MCF10A). Details of the mechanism however required further observation. Labisia pumila var. alata aqueous extract has potential to replace current treatment for menopausal symptoms. Binding activity of estrogen receptor by the plant extract show selectivity effects, which is crucial in establishing an effective and safe treatment to the menopause women.

Keywords: estrogen receptor, Labisia pumila aqueous extract, MCF-7, MCF-10A, selectivity effects

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CLINACANTHUS NUTANS EXTRACT PROTECTS AGAINST DOXORUBICIN-INDUCED CARDIOTOXICITY IN RATS

K. ESMAILLI[1], A. SHAFAEI[1], E. FARSI[2], Z. ISMAIL[1]*, M. Z. ABDUL SATTAR[3]

[1]Department of Pharmaceutical Chemistry, [2]Department of Pharmacology, [3]Department of Physiology, School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang, Malaysia
Email: ismailzhari@gmail.com

ABSTRACT

Clinacanthus nutans (CN), or Sabah snake grass, is small shrub belong to Acanthaceae family. It has been used in traditional medicine to cure various kinds of ailments such as snake bites, kidney failure and cancer. Clinical uses of doxorubicin (DOX), a highly active anticancer agent, are limited by its severe cardiotoxic side effects associated with increased oxidative stress and apoptosis. In this study we aimed to investigate whether standardised water extract of CN (CN-W) has protective effects against doxorubicin-induced free radical production and cardiotoxicity in male rats. Thirty Sprague Dawley male rats were divided into five equal groups consisting of six animals. Two groups (I and II), were used as control and received distilled water orally. Groups (III, IV and V) were received CN-W at doses of 125, 250, 500 mg/kg orally for 27 d. Groups (II, III, IV and V) were received a single i. p. dose of DOX (25 mg/kg) on day 27th, after successive administration of distilled water or CN-W. At the end of the experiment period (29 d), 48 h, after DOX injection, rats were anesthetized and blood samples were collected from ophthalmic artery in the orbital rim prior to sacrifice. Serum was separated and heart specimens were fixed in 10% formalin for histopathological examination. A single dose of doxorubicin (25 mg/kg) caused increscent in both serum cardiac enzymes LDH and CPK activities and a significant increase malonyl dialdehyde (MDA) in plasma (group II). However, pretreatment of rats with CN-W for 27 d before doxorubicin therapy, reduced the activity of both enzymes, and significantly decreased of MDA production in plasma (groups III, IV and V). Total antioxidant activity was increased in groups III, IV and V after oral administration of CN-W compare to untreated group II. Histopathological examination of heart tissue showed that DOX treatment resulted in alteration of cardiac tissue structure in the form of peri arterial fibrosis and apoptotic changes in cardiomyocytes. Pre-treatment with CN-W for 27 d ameliorated the effect of DOX administration on cardiac tissue; cardiomyocytes looked more or less similar to those of control (group I). Our results suggest that CN-W is potentially protective against doxorubicin-induced cardiotoxicity.

Keywords: Clinac anthus nutans, Doxorubicin, Cardiotoxicity

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CHROMATOGRAPHIC PROFILING OF SECONDARY METABOLITES FROM PANDANUS EXTRACTS

A. R. ZULKIFLI, M. F. SADIRAN, R. S. JALAL*, H. F. MOHSIN, I. ABDUL WAHAB

Department of Pharmacology and Chemistry, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: ibtisam@puncakalam.uitm.edu.my

ABSTRACT

The secondary metabolites in plants have numerous medicinal properties which are health beneficial. These secondary metabolites can be found in a variety of plants around the world including the Pandanus species (family: Pandanaceae). Pandanus amaryllifolius (P. amaryllifolius or the fragrant screw pine) is well known in the South East Asia countries. This plant is also referred as "Vanilla of the East" and is synonymous to Pandanus odorus. The leaves are locally used as flavouring and colouring agent. However, few information is available about the usage of the secondary metabolites containing in the Pandanus amaryllifolius. Therefore, this study was conducted to obtain the chromatographic profile of secondary metabolites in Pandanus amaryllifolius. The leaf samples were collected from various locations, including Bandar Utama (Kuala Lumpur), Cameron Highland (Pahang) and Sungai Besar (Selangor). In addition, the root part of the plant was taken from Seri Kembangan (Selangor). These research materials were extracted with methanol and subjected to a reverse phase high performance liquid chromatography (RP-HPLC), with water and acetonitrile as the mobile phase. The separation was detected by using diode array detector at λ = 220-260 nm. The chromatograms were analysed via Chem Station software. From the result, a sharp, single and symmetrical chromatographic peak was observed, which could indicate the presence of polar secondary metabolites, such as polyphenols. The separation of the significant, baseline-resolved peaks was achieved after optimising the RP-HPLC conditions. The experimental parameters were improved, which include the mobile phase composition, flow rate, the time of analysis and the gradient elution, to obtain better chromatographic resolutions. Meanwhile, the column temperature was maintained throughout the sample injections. Nevertheless, the natural Pandanus' compositions could not be briefly identified, as there are absences of standards. Upon comparison, the best chromatographic profile was shown by the sample obtained from Bandar Utama, Kuala Lumpur.

Keywords: Chromatography, Pandanus

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EURYCOMA LONGIFOLIA EXTRACT EXHIBITS ANTI-ATHEROSCLEROTIC EFFECTS IN AN *IN VIVO* MODEL

A-J. FAKHRIA[1]*, A. K. SAXENA[1], I. M. AL-ANI[1], T. A. NORLELAWATI1, M. R. HANIM[2], K-Z. NORSIDAH[1]

[1]Department of Pharmacology, Faculty of Medicine, International Islamic University, Malaysia, Kuantan, Pahang, Malaysia, [2]Faculty of Medicine, Universiti Sains Islam Malaysia
Email: mrsfofo.2010@gmail.com

ABSTRACT

Hyperlipidemia remains the most important risk factor for cardiovascular events mainly the atherosclerosis, a progressive process occurs within the walls of large and medium arteries. Eurycoma longifolia (EL), Simaroubaceae is a herb that has been used since ancient times to combat many health problems and for other medicinal purposes. Its effects are generally attributed to its bioactive constituents. To examine the effects of EL on lipid profile and intima media thickness (IMT) in aorta of Sprague Dawley (SD) rats by high-fat induction. Healthy male SD rats were divided into 4 groups of 6 animals and treated for 12 w as follow: Group I was given only normal diet, Group II was given normal diet and EL, Group III was given only high fat diet (HFD), Group IV was given HFD and EL extracts. The thicknesses of the intima media of aortas were photographed and measured with Dino-Capture® 2.0. Statistical significance was determined by one-way (ANOVA). P values less than 0.05 were considered significant. In this study, the aortic IMT was significantly increased in the HFD group than that of ND ($p<0.05$), on the other hand the aortic IMT was significantly reduced in HFDEL group than that of HFD group ($p<0.05$) Results represent mean (SD) of 5 rats per group. Results of this study showed that the aqueous extract of EL attenuated the formation of atherosclerotic plaques in aorta of rats by HFD induction.

Keywords: Atherosclerosis, Intima media thickness, Aorta, High fat diet

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ALPHA-MANGOSTIN ATTENUATES LPS-INDUCED NEUROINFLAMMATION AND MEMORY IMPAIRMENT IN MICE

N. E. YAACOB[1,2], V. MANI[1,2]*, K. RAMASAMY[1,3], A. PRAKASH[1,2], A. B. ABDUL MAJEED[1,2]

[1]Faculty of Pharmacy, [2]Brain Degeneration and Therapeutics Group, [3]Collaborative Drug Discovery Research (CDDR) Group, Pharmaceutical and Life Sciences Community of Research, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: vasudevan@puncakalam.uitm.edu.my

ABSTRACT

α-Mangostin is a polyphenolic xanthone, that is found in the Garcinia mangostana (Mangosteen) pericarp. Several *in vitro* studies indicated that this bioactive compound is capable to inhibit inflammation. The present study aimed to investigate the effects of α-Mangostin on LPS-induced neuroinflammation in mice. In this study, thirty male ICR mice were used and they were divided into 5 groups randomly (n=6); among them 3 groups were administered orally with α-Mangostin at doses of 1, 3, and 5 mg/kg, whilst control group and the LPS group received 0.5% w/v CMC orally. The treatment continued for 30 d and the last 4 d, lipopolysaccharide (LPS) was injected intraperitoneally except for the control group. The spatial memory was assessed using a Morris water maze test. From this test, the animal received LPS injection depicted memory deficits as compared with the control group, where escape latency remain increased in 3 consecutive days of the test. However, administration of α-Mangostin indicated reversal of the memory deficit with a decrease in escape latency time. Apart from that, the pro-inflammatory (TNF-α, IL-6, and IL-1β) and anti-inflammatory (TGF-β1 and IL-10) cytokines also were examined. Most of the pro-inflammatory cytokines exhibited significant reductions; however, treatment with 5 mg/kg of α-Mangostin indicated a failure in suppression of IL-6 production. Consider anti-inflammatory cytokines, treatment with α-Mangostin elevated these levels with significant as compared to LPS group. In conclusion, α-Mangostin was promised a good candidate in reducing the neuroinflammation and further study should be conducted on the pathway levels in which it inhibits inflammation.

Keywords: α-Mangostin, Memory impairment, Neuroinflammation, Inflammatory cytokines

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EPIGENETIC MODIFIERS AS TOOLS TO DIVERSIFY SECONDARY METABOLITE PRODUCTION IN ARCTIC FUNGI

S. H. SADIRAN[1], S. A. ALIAS[2], S. SULTAN[1], J-F. F. WEBER[1]

[1]Atta-ur-Rahman Institute for Natural Product Discovery, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Institute of Ocean and Earth Sciences, University of Malaya, Kuala Lumpur, Malaysia
Email: are_jar2009@yahoo.com

ABSTRACT

Secondary metabolite production of fungi can be exploited by different approaches such as modification of the fermentation medium, genetic manipulation and epigenetic modifiers. In this study, epigenetic modifiers were applied to polar fungi isolated from soil samples. Soil samples were collected from Svalbard Island, Arctic, and direct inoculation method was used to isolate the fungi. Thirty-eight pure cultures were isolated and then grown in 96-deep well microtitre plate using the laboratory standard operating procedure. They were grown in different media containing combinations of epigenetic modifiers at different concentrations. The epigenetic modifiers used were DNA methyltransferase inhibitors (S-adenosylhomocysteine and 5-azacytidine) and histone deacetylase inhibitors (HDAC) (i.e. valproic acid, suberoylanilide hydroxamic acid and sodium butyrate). Psychrotrophic fungi were incubated at 28 °C for 2 w and the psychrophilic fungi were incubated at 10 °C for 5 w. The cultures were extracted with ethyl acetate and crude extracts were subjected to HPLC analysis. The chromatographic profiles were examined to detect epigenetic elicitor's effects on the secondary fungi. Cultures A1C3, metabolite B2C2, B1-3, D2CD2 and D1D1 showed significant results where the production of some metabolites was increased in the presence of epigenetic inhibitors. The selected fungi were grown at large scale to isolate the compounds. The above results demonstrate that the epigenetic manipulation is a promising method to stimulate the silent or little expressed biosynthetic pathways in fungi and thus spur the production of secondary metabolites from Arctic fungi.

Keywords: Arctic fungi, Psychrotrophic, Psychrophilic, Epigenetic modifiers

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CHITINOPHILIC AND KERATINOPHILIC FUNGI AS A SOURCE OF SECONDARY METABOLITES

S. N. S. ZUBIR[1,2]*, J-F. F. WEBER[1,2], S. SULTAN[1,2], N. MANSHOOR[1,2]

[1]Faculty of Pharmacy, [2]Atta-ur-Rahman Institute for Natural Product Discovery, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: sitinursarah@hotmail.com

ABSTRACT

Fungi, recognised as frequent producers of secondary metabolites, occupy virtually all possible ecological niches. The present study focuses on studying chitinophilic and keratinophilic fungi as a potential source of secondary metabolites. The objective of this study is to isolate secondary metabolites from the above mentioned fungi using a modified protocol named MECSUS (Microtiter plate, Elicitors, Combination, Solid phase extraction, UHPLC, Statistical analysis) recently developed in the Microbial Metabolite Laboratory of Atta-ur-Rahman Institute for Natural Products Discovery (AuRIns). Chitiniphilic fungi were isolated by collecting insects that were sick or dead due to fungal infection. Keratinophilic fungi were isolated using 'Tokava' hair-baiting method. Soil samples were collected from the Biological Research of AuRIns at Puncak Alam, Sungai Chilling Fish Sanctuary at Selangor, Endau-Rompin National Park at Johor and Tanah Aina Bentong at Pahang. Eleven fungi were isolated namely, TOWB-F2, SA-F1, SC14a-1, SC14a-2, SC14b-1, ERS, ERI, ER2a-1, ER2a-2, ER2b-1, TOH and BENTONG. A growth study was conducted over month on five strains which showed that two weeks were suitable fermentation duration. The isolated fungi were grown simultaneously on 96-well microtiter plates in 8 media made of a common standard composition and supplemented with various elicitors. Liquid-liquid extraction was used to extract the secondary metabolites. All the fungi were analysed by HPLC. After repeating chromatogram analysis, two compounds were isolated namely, penicillic acid and pseurotin A.

Keywords: Chitinophilic, Keratinophilic, Tokava method, MECSUS

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CYTOTOXIC AND ANTIMICROBIAL EFFECTS INDUCED BY ETHYL ACETATE EXTRACTS OF MALAYSIAN MARINE ENDOPHYTIC FUNGI

S. M. LIM[1,2], N. S. ABDUL RASHID[1], N. MOHD TAHIR[1], S. S. HASNI[1], S. F. S. BAHRIN[1], M. S. IBRAHIM[1,2], K. RAMASAMY[1,2]*

[1]Faculty of Pharmacy, [2]Collaborative Drug Discovery Research (CDDR) Group, Pharmaceutical and Life Sciences Community of Research, Universiti Teknologi MARA, Selangor, Malaysia
Email: kalav922@gmail.com

ABSTRACT

Endophytic bioactive compounds are progressively becoming significant in drug discovery given the diversity of their biological activities which include anticancer and antimicrobial effects. Capitalising on the abundance of unexplored Malaysian endophytes that reside within marine plants grown along the Morib Beach, the present study was undertaken to assess the cytotoxic and antimicrobial profiles of ethyl acetate extracts of endophytic fungi, namely MBS3.2 (stem; Sonneratia sp), MKS3 and MKS3.1 (stem; Terminalia sp) as well as MBL (leaf; Avicennia sp). For cytotoxic assay, HCT116 (human colorectal carcinoma cells) were treated with the ethyl acetate endophytic fungal extracts (0.01-100 µg/ml) for 72 h. SRB assay was then performed to generate data from which the IC50 (concentration required to achieve half maximal inhibition) was determined. The antimicrobial assay, on the other hand, was carried out using the Minimum Inhibitory Concentration (MIC) Broth Microdilution Method. Both Gram positive (Staphylococcus aureus) and negative (Escherichia coli, Salmonella typhimurium and Pseudomonas aeruginosa) bacteria were exposed to ethyl acetate endophytic fungal extracts (0.01-100 µg/ml) for 24 h after which MIC was determined. The present findings found the ethyl acetate extracts of endophytic fungi to be active cytotoxic agents (IC50<20 µg/ml). The relative potency of the extracts can be ranked (in decreased potency) as MBS3.2>MKS3.1>MKS3>MBL. MBS3.2 (IC50 = 0.16 µg/ml) is approximately 3-fold more potent than that of 5-FU, the positive control. MBS3.2 also demonstrated potential antimicrobial activity against S. aureus, P. aeruginosa and S. typhimurium (MIC = 0.1 mg/ml, 0.7 mg/ml and 1 mg/ml, respectively). Its MIC against S. typhimurium, in particular, was comparable to that of gentamicin, the positive control. The rest of the extracts elicited weak antimicrobial activity. MBS3.2 has emerged as the lead ethyl acetate endophytic fungal extract and the results warrant in-depth cytotoxic and antimicrobial studies using its pure compounds.

Keywords: Cytotoxic, Antimicrobial, Marine, Endophytes, Fungi

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ANTIOXIDANT, ANTIDIABETIC AND ANTIGLYCATION PROPERTIES OF METHANOLIC EXTRACTS OF LOCAL AND IMPORTED PUNICA GRANATUM

S. ABDUL RAHMAN[1]*, N. S. MOHD SUHAIMI[1], W. ABDUL HAKEEM[1], Z. ADAM[2]

[1]Faculty of Pharmacy, Department of Pharmaceutical Sciences, Cyberjaya University College of Medical Sciences, Cyberjaya, Malaysia, [2]Agensi Nuklear Malaysia, Medical Technology Division, Kementerian Sains, Teknologi dan Inovasi, Bangi, Kajang, Selangor, Malaysia
Email: shamima@cybermed.edu.my

ABSTRACT

Punica granatum, commonly known as pomegranate, has been proven to exhibit many pharmacological activities, and among them are antioxidant and antidiabetic activities. P. granatum that is available in Malaysia is mostly imported, but it is grown locally too. Many studies had been done on imported P. granatum, but limited research performed on local P. granatum. In this study, *in vitro* antioxidant and antidiabetic activities of methanolic extracts of pulp and peel of both local and imported P. granatum were evaluated. The antioxidant study was done by evaluating the total phenolic content (TPC), total flavonoid content (TFC), and reducing power ability. The TPC and TFC were done using the Folin–Ciocalteu assay and aluminum chloride colorimetric assay, respectively. The antidiabetic study was done using alpha glucosidase inhibition assay and anti glycation assay. The results showed that the peel extracts have higher phenolic and flavonoid content than the pulp extracts. In the antidiabetic study, the peel extracts also showed higher alpha glucosidase inhibition and antiglycation activity. In general, local P. granatum showed to have better antioxidant, and antidiabetic activities compared to the imported P. granatum.

Keywords: Punica granatum, Antioxidant, Antidiabetic, Antiglycation

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ANTIPROLIFERATION EFFECT OF CLINACANTHUS NUTANS ETHANOLIC EXTRACT ON MCF-7 BREAST CANCER CELLS

N. H. R. AHMAD[1], I. ABDUL WAHAB[2] AND A. H. JAHIDIN[1]*

[1]Pharmacology and Toxicology Research Laboratory, [2]Atta-Ur-Rahman Institute For Natural Product Discovery, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: aisya735@salam.uitm.edu.my

ABSTRACT

Clinacanthus nutans Lindau is a small shrub of the family Acantaceae. This native of Southeast Asia has been used traditionally to treat numerous ailments including dysentery, fever, skin rashes, insect and snake bite, herpes simplex virus infection and varicella-zoster virus lesions, inflammation, haematoma and gout. Many studies have demonstrated its efficacy as immunomodulatory, antioxidant, antiviral and antiproliferative agents. Its potency to relieve minor skin inflammation has also been proven clinically. Recently, the efficacy of C. nutans in treating various types of cancer has been reported. However, the mechanism by which C. nutans exerts its anticancer effect on breast cancer remains to be elucidated. This study seeks to elucidate the effect of C. nutans ethanolic extract on the proliferation of the MCF-7 human breast cancer cell line, which represents the estrogen receptor-positive subtype of breast cancer. The extract of C. nutans was prepared by soaking dried fresh leaves in 50% ethanol for 72 h. Studies using crude extracts are critical due to possible additive and synergistic effects of phytochemicals in the extracts. The effect of C. nutans on cell proliferation was elucidated using MTS assays. The ethanolic extract of C. nutans significantly ($p<0.05$) inhibits the proliferation of MCF-7 cells after 72h of treatment. Further studies need to be perfomed to understand the molecular mechanisms underlying its antiproliferative activity against MCF-7 cells. Apart from expanding and enriching knowledge on medicinal plants and traditional medicine, the results help to define the molecular mechanism of C. nutans anticancer activity and provide scientific-based evidence on the effectiveness of C. nutans as the basis for potential application in adjuvant therapy for breast cancer.

Keywords: Clinacanthus nutans, Breast cancer, Belalai gajah, Sabah snake grass

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PHYTOSTEROLS FROM PANDANUS CONOIDEUS AND PANDANUS LERAM

U. A. ARMAYNI, M. S. AHMAD, N. YUSOF, H. F. MOHSIN*, I. ABDUL WAHAB

Department of Pharmacology and Chemistry, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: hannisfadzillah@puncakalam.uitm.edu.my

ABSTRACT

Pandanus conoideus and Pandanus leram (genus: Pandanus, family: Pandanaceae) are known for their fruits, which have been traditionally used among the indigenous Asia Pacific populations. However, in this study the phytochemicals of Pandanus conoideus root and Pandanus leram leaves were investigated. The roots of P. conoideus (PCR) and the leaves of P. leram (PLL) were both introduced to the non-acidic extraction. Subsequently, the fractionation was performed by using the conventional, non-automated chromatographic techniques. The structures of five pure compounds (PCR1, PCR2, PCR3, PLL1 and PLL2) were elucidated via 1D and 2D Nuclear Magnetic Resonance (NMR) spectroscopic methods and by comparison with the literatures. PCR1 was tentatively identified as steroidal alkaloid. Meanwhile, PCR2 and PLL1 was tentatively identified as stigmastenone which was observed as major constituent of PCR and a minor constituent in PLL extract. The PCR3 and PLL2 were tentatively identified as beta-sitostenone and were observed as the minor component of PCR and the major component from PLL extract. Results from this study recorded of the occurrences of phytosterols, from P. conoideus and P. leram.

Keywords: Pandanus, Phytosterols

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CALLUS INDUCTION OF TACCA INTEGRIFOLIA

A. A. ZAKARIA*, N. ANILIZAWATIMA

Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: anieskuzzer@gmail.com

ABSTRACT

Medicinal value of Tacca integrifolia plant has been widely used in the tropical region of Asia to treat gastric ulcer, enteritis, and hepatitis. The aim of this study was to establish an effective protocol for callus induction from the explants of Tacca integrifolia and to identify the best medium using different types of plant growth hormones for callus induction. A callus is an undifferentiated mass of tissue which appears on explants after a few weeks of transfer onto a medium with suitable plant growth hormones. Callus can be an alternative way in producing desired bioactive compounds without sacrificing the whole mother plant. In this study, 8 different types of full strength Murashige and Skoog (MS) media with different concentration of 6-benzylaminopurine (BAP) were used to induce callus. Each of the media was supplemented with 30 g/l of sucrose and 0.5 mg/l 1-naphthaleneacetic acid (NAA). The source of the explants was taken from the matured *in vitro* culture of Tacca integrifolia which was 6 mo old. The leaf and stem explants were cultured onto the media and put in the dark to examine callus formation. After 3 mo, the formation of callus can be observed from 5 different types of media supplemented with 0.5, 1.0, 2.0, 3.0 and 4.0 mg/l BAP. No formation of callus was obtained from other media supplemented with 0, 6.0 and 8.0 mg/l BAP. Callus formation can be obtained from the stem explants while the leaf explants showed no formation of callus. Calli were further subcultured onto new media with the same concentration to allow the development of callus. Only media with 2.0 mg/l BAP managed to develop a clump of callus. The results showed that full strength Murashige and Skoog media supplemented with 30 g/l sucrose, 0.5 mg/l NAA and 2.0 mg/l BAP using the stem explants were the best medium to induce callus.

Keywords: Tacca integrifolia, Callus induction

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CHEMOPREVENTIVE EFFECT OF CLINCANTHUS NUTANS EXTRACT ON CERVICAL CANCER CELLS AND ITS MORPHOLOGICAL CHANGES

N. A. MAT JUSOH[2], N. A. BAKHARI[2], A. SEENI[3], C. N. OTHMAN[1]*

[1]Faculty of Pharmacy, Universiti Teknologi MARA (UiTM), Bertam Campus, Kepala Batas, Penang, Malaysia, [2]Faculty of Applied Sciences, UiTM Permatang Pauh, Penang, Malaysia, [3]Advanced Medical and Dental Institute, Universiti Sains Malaysia, Kepala Batas, Penang, Malaysia

Email: chenoriah.othman@ppinang.uitm.edu.my

ABSTRACT

Cervical cancer remains the second most common cancer in women worldwide despite the various treatments available to date. Currently, chemoprevention approach using natural products is of great interest among researchers. Clinacanthus nutans (C. nutans) has been used traditionally as a herbal medicine to treat various diseases including cervical cancer. In this study, antiproliferative activity of C. nutans extract on cervical cancer using human cervical adenocarcinoma (HeLa) cell line and the morphology changes was explored. 500 gram of dried leaves and stem of C. nutans powder were immersed in ethanol for 3 d. After filtration, the extracts were label as ethanol extract of C. nutans (EECN). The extracts were concentrated by using rotary evaporator. EECN were prepared to various concentration and treat on HeLa cells to get the IC50 value. Morphology changes were observed. EECN inhibit the growth of cancer cells at IC50 value of 500 µg/ml. Microscopic observations showed that EECN treatment induced apoptotic features. These preliminary results suggest that EECN exhibits a new potential candidate as chemopreventive agent in cervical cancer cells due to its antiproliferative effect on Hela cells.

Keywords: Clinacanthus nutans, HeLa, Antiproliferative, Morphology, Apoptotic

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NEUROPROTECTIVE EFFECT OF MYRMECODIA PLATYTYREA TUBER EXTRACT ON LIPOPOLYSACCHARIDE-INDUCED NEUROINFLAMMATION

N. A. NORDIN*, M. H. HASAN, N. S. A. SURATMAN, A. ADAM

Pharmaco-Toxicology Research Laboratory, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Malaysia
Email: mizatonhazizul74@gmail.com

ABSTRACT

Neuroinflammation is a state of brain responding to diseases, infections and neuronal injuries that may be a contributing factor for many neurodegenerative diseases such as Alzheimer's disease (AD). Myrmecodia platytyrea (M. platytyrea), locally known as Sarang Semut used by old folks in treating inflammatory-related diseases such as ulcer, tumour and cancer, haemorrhoid and coronary heart problem. Therefore, this study was aimed to investigate the potential of M. platytyrea aqueous tuber extract (MPAE) on lipopolysaccharide (LPS)-induced neuroinflammation, *in vivo*. There are six groups assigned in this experiment including two groups of control (untreated with LPS and treated with LPS (3 mg/kg), piracetam (a nootropic drug, 400 mg/kg) and three groups of MPAE (100, 200 and 400 mg/kg). Morris water maze (MWM) test was carried out for 3 d followed by probe test (PB) to test for memory and learning. Our results showed that there were no significant difference between control (treated with LPS) and the treated groups (MPAE and piracetam) in terms of average speed and total travelled distance but MPAE showed slight memory improvement in term of escape latency. Meanwhile, PB test of MPAE showed significant difference between control (treated with LPS) and control (untreated with LPS) group in the number of entries in the targeted zone (Zone D) but no significant difference between all group for time spending in the targeted zone. In conclusion, MPAE may have neuroprotective effect on LPS-induced neuroinflammation but further study need to be conducted to clarify the molecular mechanism.

Keywords: Morris water maze, Myrmecodia platytyrea, Neuroinflammation

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ANTIPROLIFERATIVE ACTIVITY OF MYRMECODIA PLATYTYREA

N. IBRAHIM, M. H. HASAN*, M. ABDUL SAMAD, A. H. JAHIDIN, A. ADAM

Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: mizaton_hazizul@puncakalam.uitm.edu.my

ABSTRACT

Ant nest plant is a member of Rubiaceae family with five genera, however only two of them including Myrmecodia (42 species) and Hydnophytum (94 species) are related to ants. Myrmecodia spp. are scattered from Peninsular Malaysia to the Asia Pacific region and they are well known for their tuber that are inhabited by ants. Tuber of the Myrmecodia spp. is used as decoction, herbal preparation for mild diseases such as backaches, ulcer, nosebleed, allergy, and haemorrhoid, uric acid disorder to a much severe diseases such as tuberculosis, coronary heart disease, stroke, tumour and cancer by the locals in the West Papua. Nevertheless, there is only limited research on Myrmecodia platytyrea which is locally known as sarang's. Antiproliferative activities of M. platytyrea methanolic tuber extract against several tumour and normal cell lines have been carried out using MTS assay. To further affirm the mode of cell death of M. platytyrea methanolic tuber extract, apoptosis analysis was conducted by fluorescent microscopy (Acridine Orange/Propidium Iodide staining). The extract was found to be most potent in inhibiting the growth of liver cancer cell line compared to other cancerous cells. M. platytyrea methanolic tuber extract is significantly cytotoxic against HepG2 cell compared to Chang cells. A HepG2 cell treated with M. platytyrea methanolic tuber extract was found to produce typical apoptotic characteristic when observed under fluorescent microscope at 400x magnification at a dose-dependent manner. M. platytyrea methanolic tuber extract inhibited the proliferation of HepG2 cells without affecting normal cells, showing high selectivity. In conclusion, M. platytyrea methanolic tuber extract possess potential antiproliferative activity that may be developed as a potential anticancer therapy.

Keywords: Myrmecodia platytrea, Rubiaceae, Antiproliferative activity, Fluorescent microscopy

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ANTIDIABETIC POTENTIAL OF METHANOLIC EXTRACT OF MYRMECODIA PLATYTYREA ON STREPTOZOTOCIN-INDUCED DIABETIC RATS

H. ZAKARIA, M. H. HASAN*, I. ABDUL WAHAB, A. ADAM

Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam, Selangor, Malaysia
Email: mizaton_hazizul@puncakalam.uitm.edu.my

ABSTRACT

Tubers of Sarang-Semut, Myrmecodia platytrea (Rubiaceae) have been used traditionally as an alternative therapy for the management of cancer. This plant is also believed to have the ability to lower blood glucose level. Nevertheless, no scientific proof is available on its antidiabetic effect. Type 2 diabetes mellitus (T2DM) is one of the main non-communicable chronic diseases. Individuals suffering in T2DM have insulin resistance and usually develop relative insulin deficiency. The purpose of this study was to investigate the potency of M. platytyrea methanolic tuber extract (MPMTE) as anti-hyperglycemic agent by evaluating their effects on STZ-induced diabetic rats. Hyperglycemia was induced in fasted rats with STZ (45 mg/kg) and then the male Sprague Dawley rats were treated orally with of MPMTE (100, 200 and 400 mg/kg) and metformin (positive control, 100 mg/kg) daily for 14 d. Blood were taken from day 0 to 15 to measure fasting blood glucose level and also to analyze the lipid profile. The results showed that the STZ-induced diabetic rats (control group) had significant increased ($p<0.05$) of fasting blood glucose, total cholesterol, triglycerides and low-density lipoprotein (LDL), and low in levels of high-density lipoprotein (HDL) compared to the normal rats (non-STZ-induced rats). The STZ-induced diabetic rats given MPMTE 200 and 400 mg/kg showed significant decreased ($p<0.05$) in fasting blood glucose, total cholesterol, triglycerides and LDL (0.7±0.1 and 0.5±0.1 mmol/l, respectively) levels compared to control rats. However, the HDL levels were not different from the control rats. Therefore, our results suggested that MPMTE has the potential as an anti-hyperglycemic agent by reducing blood glucose levels and lowering the cholesterol and triglyceride levels of STZ-induced diabetic rats.

Keywords: Myrmecodia platytrea, Steptozotocin, T2DM, Diabetic rats, Hyperglycemic

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IN VITRO ANTI-INFLAMMATORY ACTIVITY OF ERYTHROXYLUM CUNEATUM AQUEOUS EXTRACT

S. R. SALEH[1], M. H. HASAN[1]*, M. I. ADENAN[2], A. ADAM[1]

[1]Department of Pharmacology and Toxicology, [2]Atta-ur-Rahman Institute for Natural Product Discovery, Faculty of Pharmacy, Universiti Teknologi MARA (UiTM), Puncak Alam Campus, Selangor, Malaysia
Email: mizatonhazizul74@gmail.com

ABSTRACT

Erythroxylum cuneatum was used in traditional medicine to treat diabetes mellitus, bodily discomfort and used as tonics for miscarriages. This study was performed to evaluate the ability of E. cuneatum aqueous extract (ECAE) as anti-inflammatory agent using RAW 264.7 macrophage cell line. Macrophages were first screened for cytotoxicity effect using MTT assay. Then, cells were induced with lipopolysaccharides prior treatment with ECAE to induce inflammation. Evaluations on the cytokines, cyclo oxygenases and antioxidants activities were later performed. MTT assay revealed that ECAE had low cytotoxic effects on the RAW 264.7 (IC50 =141.25 µg/ml). Compared to the non-treated group, treatment of macrophages with ECAE (35, 70 and 140 µg/ml) significantly reduced interleukinIL-1β and IL1β-6 levels and at the same time increased the concentration of tumour necrosis factor-α TNF(-α). Besides that, significant reduction in cyclooxygenase activity such as COX-1 and COX-2 also were observed in cells treated with ECAE (17.5, 35 and 140 µg/ml) and (35 and 140 µg/ml), respectively. In addition, superoxide dismutase (SOD) activity has increased in activated cell treated with ECAE (17.5, 35, 70 and 140 µg/ml) in a dose-dependent manner. No significant changes were observed in the catalase activity. Lastly, decreased in glutathione peroxidase (GPx) activity was observed in cell treated with ECAE (35, 70 and 140 µg/ml) in a dose-dependent manner. In conclusion, ECAE showed anti-inflammatory activity by reducing cytokines (IL-1βand IL-6) and COXs levels; and also increased in SOD activity.

Keywords: Erythroxylum cuneatum, RAW 264.7, Anti-inflammatory

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ANTI-INFLAMMATORY ACTIVITY OF MYRMECODIA PLATYTREA ON CARRAGEENAN-INDUCED RAT PAW EDEMA

M. MOHD ZIN, M. H. HASAN*, T. PONTO, A. ADAM

Department of Pharmacology and Toxicology, Faculty of Pharmacy, University Technology MARA (UiTM), Puncak Alam Campus, Selangor, Malaysia
Email: mizatonhazizul74@gmail.com

ABSTRACT

Several studies on pharmacological importance of Myrmecodia sp. revealed that this plant have potent antioxidant activities, anti-proliferative activities and were able to upregulate the immune response system. Myrmecodia sp. has been traditionally used as a remedy to treat diseases such as hyperuricemia, tuberculosis and cancer. In this study, the anti-inflammatory effect of Myrmecodia platytyrea tuber aqueous extract (MPAE) was investigated. Carrageenan-induced rat paw edema model was used to measure the effect of this extract on the edema volume and immunological parameters. Adult male Sprague Dawley rats were classified into six groups, group I received the vehicle (normal saline), group II received indomethacin (5 mg/kg), group III received acetylsalicylic acid (ASA) (200 mg/kg), and group IV, V and VI each received 100 mg/kg, 200 mg/kg and 400 mg/kg of MPAE, respectively. The edema was quantified by measuring the hind paw thickness before sub plantar injection, and after the injection at 1, 2, 3 and 4 h. Blood samples were withdrawn after the 4th h of carrageenan induction and the serum were used for analysis of pro-inflammatory cytokines, which includes tumour necrosis factor-α TNF(-α), interleukin-1βIL-(1β)and IL-6. Administration of MPAE (100, 200 and 400 mg/kg, p. o.), significantly reduced the edema thickness in a time-and dose-dependent manner. MPAE showed reduction in the edema thickness with percentage inhibition of inflammation in a range between 11 to 12%. However, MPAE was not able to reduce secretion of inflammatory mediators in all determined parameters compared to the control group. Based on histological examination on liver, kidney and stomach, there are no abnormalities in the cell morphology. In conclusion, MPAE may be a potent anti-inflammatory agent in diminishing edema caused by carrageenan. This may be due to the presence of flavonoid and phenolic in the extract and it is vital for further investigation to be carried out to elucidate the active compounds responsible for the pharmacological activities.

Keywords: Anti-inflammatory, Cytokines, Myrmecodia, Carrageenan

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ANTIPROLIFERATIVE AND CYTOPROTECTIVE EFFECTS OF 3,4,10-TRIMETHOXYSTILBENE

M. ZULKEFLEE, M. H. HASAN*, I. ABDUL WAHAB, A. ADAM

Department of Pharmacology and Toxicology, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: mizatonhazizul74@gmail.com

ABSTRACT

Stilbenes are well known polyphenols that are normally found in plants. They are recognized as one of the polyphenols that have been identified as having potential cancer chemo preventive properties. One of the most studied stilbene is resveratrol (3,5,4'-trihydroxy-trans-stilbene). However, resveratrol was found to have low bioavailability. Therefore, new derivatives of stilbene have been developed to enhance its potential. 3, 4, 10-Trimethoxystilbene is a stilbene analogue that has been synthesized through Heck reaction in our laboratory. We were looking into the potential of this compound to reduce the progression of several cancer cell lines, *in vitro* and protect normal cells against free radical produced by the current chemotherapy. Thus, this study was carried out to evaluate the antiproliferative activity against human hepatocellular carcinoma (HepG2) and human colorectal carcinoma (HT29). The cytoprotective effect of this compound against normal cells treated with hydrogen peroxide (100µM) and DPPH radical scavenging assay were also carried out. The antiproliferative and cytoprotective effects were determined via MTT assay. Our results showed that 3,4,10-trimethoxystilbene was able to inhibit the proliferation of HepG2 and HT29 with IC50 values less than 20µg/ml. 3,4,10-Trimethoxystilbene showed a significant decreased in cell death compared to control on hydrogen peroxide-induced Chang cells treated with 3,4,10-trimethoxystilbene. However, this compound was unable to scavenge DPPH radical. As a conclusion, 3,4,10-trimethoxystilbene demonstrated antiproliferative activity on HepG2 and HT29 cell lines. It also showed cytoprotective effect against hydrogen peroxide-induced oxidative stress but, not via free radical scavenging mechanism. Thus, further investigation is needed to identify mechanism of action of 3,4,10-trimethoxystilbene as an antiproliferative agent against hepatocellular carcinoma and colorectal carcinoma and its cytoprotective effect against hydrogen peroxide-induced oxidative stress.

Keywords: 3,4,10-trimethoxystilbene, Antiproliferative activity, Protective effect, Hydrogen peroxide HepG2

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ANTIOXIDANT AND LIPID LOWERING EFFECT OF KAPPAPHYCUS ALVAREZII ON HYPERCHOLESTEROLEMIC INDUCED SPRAGUE-DAWLEY RATS

M. MAHADI[1,2], Z. ESHAK[1], N. A. N. AHMAD NAZRI[2], A. H. IBRAHIM[3], F. OTHMAN[4]

[1]Department of Pharmacology and Biochemistry, Faculty of Pharmacy, Universiti Teknologi Mara, Malaysia, [2]Department of Pharmaceutical Sciences, Faculty of Pharmacy, Cyberjaya Universiti College of Medical Sciences, Malaysia, [3]Department of Food Sciences and Technology, MARDI, Malaysia, [4]Department of Human Anatomy, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia, Malaysia

Email: aniyaus@gmail.com

ABSTRACT

Research focusing on seaweed has been increased in recent years due to its valuable nutritional contents. Seaweed contains a significant amount of soluble dietary fibers, proteins, minerals and fatty acids which are essential for human nutrition. Kappaphycus alvarezii is a red seaweed claimed to have anti-inflammatory, anti-oxidant, anti-microbial, anti-obesity as well as lipid lowering properties. The present study evaluated the antioxidant level and lipid lowering effect of K. alvarezii on hypercholesterolemia-induced rats. 30 male Sprague-Dawley rats were used in this study. Diet-induced hypercholesterolemia rats were fed with 5% and 10% K. alvarezii in comparison with normal control, high fat diet (HFD) and atorvastatin treated rat. Body weight and lipid profile were measured every two weeks and upon sacrificed, liver were collected for the antioxidant measurement. Antioxidant activity was analyzed using DPPH, FRAP and catalase test. Atorvastatin treated rats and rats fed with 5% and 10% K. alvareazii had significantly reduced weight gain, TC, LDL cholesterol and increase HDL level compared to HFD group ($p<0.05$). TG level was slightly reduced when compared to normal group. Group fed with 10% K. alvareazii shows highest antioxidant activity ($p<0.05$) compared with other groups. K. alverazii is suggested to possess anti-hypercholesterolemia and antioxidant properties which probably due to the antioxidant content as well as the soluble dietary fibers.

Keywords: Nutritional value, Seaweed, Antioxidant.

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A PRELIMINARY STUDY ON ANTIOXIDANT ACTIVITY OF ENTADA SPIRALIS RIDL LEAVES

S. N. A. SYED MOHAMAD[1], S. Z. MAT[1]*, N. S. MOHD HASSAN[1], N. M. I. NIK YUSOFF[1], A. HARUN[2]

[1]Kulliyyah of Pharmacy, Pharmaceutical Chemistry Department, International Islamic University, Malaysia, [2]Faculty of Applied Sciences, Chemistry Department, Universiti Teknologi MARA, Pahang, Malaysia
Email: dszaiton@iium.edu.my

ABSTRACT

Entada spiralis Ridl. (Leguminoceae), locally known as Sintok' 'Beluru' grows wildly in Malaysia. This plant has been traditionally used as body soap, washing agent, and natural shampoo to treat dandruff. It also has been used to treat syphilis and insect bites. The use of the leaves of E. spiralis is quite rare compared to its stem and roots. However, leaves have been used traditionally to treat abdominal complaints, whereas chewed or pounded leaves are externally applied as a remedy for filariasis or elephantiasis. This study aims to discover the potential of antioxidant activity and responsible active components in the leaves so that its traditional use can be supported scientifically. This research involved preliminary phytochemical screening test, *In-vitro* antioxidant activity assay, and fractionation of leaves extract of E. spiralis by chromatographic techniques. The consecutive extraction with petroleum ether, chloroform, and methanol was employed. The phytochemicals screening test revealed the presence of saponin, tannin, triterpenoid, steroid, flavonoid and antioxidant. Each crude extract was screened rapidly by dot-blot assay and DPPH staining method. All types of extracts exhibited the antioxidant activity. Methanol extract was found to be the most active antioxidant extract which produced intense yellow spot due to the presence of high radical scavengers. While, chloroform extract possessed moderate antioxidant activity followed by petroleum ether extract. All fractions of methanol extract shows positive results due to the appearance of yellow spots detected via Thin Layer Chromatography sheet. The result supports the radical scavenging activity possessed by E. spiralis Ridl. Leaves and in-depth research is needed to further investigate the compounds responsible for its antioxidant activity by bioassay guided isolation and compound characterization for development of cosmetic product.

Keywords: Leguminoceae, Entada spiralis Ridl, Antioxidant, Radical scavenger, DPPH.

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EFFECTS OF GYNURA PROCUMBENS LEAVES AQUEOUS EXTRACT ON SERUM LIPID PROFILES AND HISTOLOGICAL ANALYSIS OF NEW ZEALAND WHITE RABBITS INDUCED WITH HIGH CHOLESTEROL DIETS

M. A. H. ISMAIL, E. A. BAHARI, F. S. IBRAHIM, R. DASMAN, Z. AMOM*

Faculty of Health Sciences, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: zulkha2992@puncakalam.uitm.edu.my

ABSTRACT

Gynura procumbens is also known as "Sambung Nyawa" in Malay contains active chemical constituents such as flavonoids, saponins, tannins, terpenoids and sterol glycosides and have been used traditionally in treating eruptive fever, rash, kidney disease, migraines, constipation, hypertension, diabetes mellitus, and cancer. Thirty six (36) males New Zealand white rabbits were divided into 6 groups as normal diet (ND), high cholesterol diet (HCD), simvastation group (SG) and treatment groups (TGA, TGB and TGC). TGA, TGB and TGC were given Gynura procumbens leaves aqueous extract at 100 mg/kg, 200 mg/kg and 400 mg/kg per day. All groups except for ND were given 0.5% cholesterol diet to induce hypercholesterol. The effects of Gynura procumbens extract towards hypercholesterolemic induced rabbits were determined by measuring the body weight, lipid profiles (Total Cholesterol, Triglycerides, HDL, and LDL) where the serum samples were analysed by CPDRL, UiTM Sungai Buloh. Total cholesterol at week 10 was 1.163±0.186 mmol/l, 40.153±3.958 mmol/l, 9.830±mmol/l, 4.303±0.856 mmol/l, 16.750±4.304 mmol/l and 22.668±2.833 mmol/l respectively. Triglycerides at week 10 were 0.790±0.092 mmol/l, 1.945±0.355 mmol/l, 0.563±0.071 mmol/l, 0.453±0.061, 0.527±0.090 and 0.610±0.125 mmol/l respectively. HDL level for ND, HCD, SG, TGA, TGB and TGC for week 10 were 0.505±0.089 mmol/l, 2.840±0.433 mmol/l, 1.777±0.276 mmol/l, 1.544±0.235 mmol/l, 1.899±0.315 mmol/l and 2.510±0.391 mmol/l respectively. LDL for ND, HCD, SG, TGA, TGB and TGC on week 10 were 0.233±0.020, 32.917±5.956 mmol/l, 9.249±0.702, 8.162±0.065 mmol/l, 16.240±4.366 mmol/l and 32.063±7.184 mmol/l respectively. Supplement of Gynura procumbens extracts (TGA, TGB and TGC) shows positive effects as compare to HCD. Rabbits that induced with high cholesterol diets with supplementation of the extract reduce the deposition of cholesterol in aorta. Consumption of Gynura procumbens leaves aqueous extract for a long term give beneficial effects in lipid profiles and reduce the formation of atherosclerosis as it can work against high cholesterol.

Keywords: Gynura procumbens, Antioxidants, Lipid profiles, Liver function test.

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EVALUATION OF THE TERATOGENICITY OF AQUOEUS EXTRACT OF LIGNOSUS RHINOCERUS IN RATS

H. S. Z. BAYANI[1]*, A. NURUL ASMA[1], S. H. GAN[2], M. F. WAN EZUMI[1]

[1]School of Health Sciences, [2]School of Medical Sciences, Universiti Sains Malaysia, Health Campus, Kubang Kerian, Kelantan, Malaysia
Email: wanezumi@usm.my

ABSTRACT

Lignosus rhinocerus (LR) or locally known as "tiger milk mushroom" is a valuable herb with abundant medicinal properties but unfortunately, its safety profiles is still lacking. This study was conducted to provide evidence on the potential teratogenic effects of LR on rats. The aqueous extract at doses of 0 (control), 250, 500, 1000 mg/kg/day were orally administered to four groups of female Sprague Dawley rats during the two weeks of premating, mating and throughout gestation periods and sacrificed on day 21 of pregnancy. Results obtained indicated that the females displayed regular oestrous cycles. The maternal body weight (MBW) and corrected maternal body weight (CMBW) of control animals were slightly higher than treated groups but no statistical differences were found. The gravid uterus of females receiving 1000 mg/kg exhibited the highest weight compared to others due to the highest foetal weight but statistically, there were no significant differences. Number of corpora lutea, number of implantation sites, percentage of implantation losses, and number of foetal resorptions, number of life or dead foetuses and foetal sex ratio showed no significant differences among all groups of animals. Overall, the live foetuses at autopsy have no signs of any external congenital malformations. These findings may suggest that the aqueous extracts of LR of up to 1000 mg/kg/day did not cause any teratogenic effects towards rat foetuses.

Keywords: Lignosus rhinocerus, Teratogenicity, Aqueous extract

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PROTECTIVE EFFECTS OF PHYLLANTHUS GOMPHOCARPUS ON MALE FERTILITY AGAINST GENITOTOXICITY OF BISPHENOL-A

E. A. BAHARI[1], A. H. ISMAIL[1], R. DASIMAN[1,2], N. HARON[3], Z. AMOM[1,2]*

[1]Natural Products and Phytomedicine Biotechnology Research Laboratory (NPPBRL), [2]Department of Basic Sciences, [3]Department of Medical Laboratory Technology, Faculty of Health Sciences, Universiti Teknologi MARA, Puncak Alam, Selangor
Email: zulkha2992@puncakalam.uitm.edu.my

ABSTRACT

Bisphenol A (BPA) is a chemical used in epoxy resin and polycarbonate plastic production and constant exposure to BPA may induce infertility in male. Phyllanthus gomphocarpus or Cermela hutan in Malay is an herb that was traditionally used to treat male infertility problem particularly among Orang Asli community. This study was designed to gain scientific evidence of the efficacy of Phyllanthus gomphocarpus root extract (PGR) in experimental male infertility. Ten percent aqueous extraction of PGR was prepared by incubating 100 g of dried ground root of Phyllanthus gomphocarpus in 1L of distilled water at 40ºC for 12 h and the decoction were freeze dried to obtain crude extract. Total numbers of 24 male rats were randomly divided into 4 groups and labeled as NEC, POC, PGR50 and PGR800. Except for the NEC group, other groups were induced genitotoxicity by oral supplementation of 200 mg/kg of BPA for 21 consecutive days. For PGR50 and PGR800 groups, the animals were concomitantly supplemented with 50 mg/kg and 800 mg/kg of PGR extracts, respectively. Sperm and blood samples were collected for sperm and hormonal analysis. The results demonstrated that the quality of sperm in both PGR50 and PGR800 groups were significantly increased in terms of its count, motility and morphology ($p<0.005$) compared to the rats in POC group. Circulating testosterone level also was found to significantly increased in both PGR groups ($p<0.05$), PGR50 (46.48±0.86 ng/ml) and PGR800 (130.24±6.2 ng/ml) compared to POC group (17.42±1.47 ng/ml). In conclusion, PGR was suggested to possess a protective effect against genitotoxicity of BPA towards male fertility through the improvement of sperm quality and testosterone hormone production.

Keywords: Phyllanthus gomphocarpus, Testosterone, Sperm quality, Genitotoxicity, Bisphenol-A

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ANTI-ANGIOGENESIS STUDIES OF SCOPOLETIN ISOLATED FROM NICOTIANA GLAUCA AGAINST COLON CANCER USING XENOGRAFT MODEL

Y. M. TABANA, L. E. A. HASSAN, S. S. DAHHAM, A. M. S. ABDUL MAJID*

EMAN Research and Testing Laboratory, School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang, Malaysia
Email: aminmalikshah@gmail.com

ABSTRACT

To investigate the *in vivo* properties of Scopoletin isolated from tree tobacco (Nicotiana glauca) using ex-vivo ring assay model and *in vivo* tumour angiogenesis in athymic nude mouse xenograft model. Isolation of Scopoletin using bio-guided anti-angiogenesic rat aorta ring assay. Elucidation of Scopoletin was done using UV, IR, NMR and GC-MS. Ex-vivo ring assay was performed applying different concentrations of the compound. Tumour xenograft model of angiogenesis was conducted (Human colorectal tumors were grown in athymic (Ncr-Nu/Nu) nude mice by injecting HCT 116 cells (107 cells/150 µl DMEM) subcutaneously on dorsal side of the animals. To further demonstrate the activity, Matrigel sponge model was carried out by injecting subcutaneously 0.3 ml of Matrigel supplemented with 100 µl of (106 27ƨ 1 cells /ml) HCT 116 to NU/Nu immunocompromised nude mice. The mice were treated orally with three different concentrations 50, 100 and 200 mg/kg. Vehicle (0.1% tween 80) and Imatinib (100 mg/kg) were used as negative and positive controls. Scopoletin inhibited significantly the microvessel in rat aortic with IC50 0.06 µM. Treating the nude mice for seven days showed strong suppression of the vascularization in matrigel plugs implanted in nude mice whereby the dose 50 and 100 mg/kg showed 59.72% and 89.4% inhibition, respectively. After chronic treatment (21 d) of the Scopoletin to nude mice using xenograft model, Scopoletin displayed a remarkable suppression of HCT 116 tumour growth when compared to that of negative. Scopoletin displayed 33.01%, 34.4% and 94.7% 573 inhibitions at 50, 100 and 200 mg/kg doses, respectively. In addition, morphological investigation of the harvested tumours demonstrated drastic reduction effect of Scopoletin on tumour vasculature. The results were compared with the imatinib as standard reference. In the present study, Scopoletin has been purified from Nicotiana glauca and it demonstrates potential anti angiogenesic activity using ex-vivo and *in vivo* models. Scopoletin could be useful as treatment for colon cancer.

Keywords: Scopoletin, Nicotiana glauca, Xenograft model, Angiogenesis

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THE POTENTIAL OF LYGODIUM MICROPHYLLUM AS AN ANTIBACTERIAL AGENT

R. AHMAD[1]*, Z. ZAKARIA[1], R. MAHMUD[2]

[1]School of Distance Education, [2]Department of Pharmaceutical Chemistry, School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang, Malaysia
Email: radiahahmad88@yahoo.com

ABSTRACT

Pteridophytes (fern and fern allies) are found scattered all over the world including Malaysia. This plant group has abundant source of natural products with therapeutic potentials, but current knowledge of pharmacological properties of ferns is limited. Lygodium microphyllum (Lygodiaceae) is a medicinal fern used for the treatment of skin diseases, swellings, curing of hiccups and control of dysentery. The aim of the study was to quantitatively assess the antibacterial activities of methanol extract of the L. microphyllum fronds. The Minimum Inhibitory Concentration (MIC) values obtained from tetrazolium microplate assay at initial concentration of 1 mg/ml was used to determine the antimicrobial activity against gram-positive bacteria Staphylococcus aureus and Bacillus subtilis; and a group of gram-negative bacteria frequently reported to cause enteric infection in humans which are Escherichia coli, Klebsiella pneumonia, Shigella dysenteriae, Salmonella typhi and Proteus mirabilis. The MIC values ranged from 0.125 to 1 mg/ml were almost comparable to the standard antibiotics cefepime, amoxicillin and Gentamicin. The growth of all the gram-negative bacteria was inhibited at varying degrees, thus justifying the use of this fern in treating enteric infections. The results demonstrated that L. microphyllum is a potential candidate to be used for the treatment of infections caused by susceptible bacteria.

Keywords: Medicinal fern, Lygodium microphyllum, Antibacterial activity, Enteric bacteria

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EFFECTIVENESS OF PEPEROMIA PELLUCIDA AS ACE INHIBITOR IN *IN VITRO* AND *IN VIVO*

A. MUN'IM*, I. HUTAHAEN, S. F. CHANY

Faculty of Pharmacy, Universitas Indonesia, Depok, West Java, Indonesia
Email: munimabdoel@gmail.com

ABSTRACT

Antihypertensive properties of plants can be evaluated *in vitro* method on inhibition of angiotensin converting enzyme (ACE) activity. In this study, we investigated the inhibitory activity of some Indonesian medicinal plants traditionally used as antihypertensive agents. The plant which showed the strongest ACE inhibitory activity, then was evaluated its antihypertensive activity on rats according to Two-kidney one-clip (2K1C) Goldblatt model. Methanolic extract of Peperomia pellucida demonstrated the strongest ACE inhibitory activity with IC50 of 7.17 mg/ml. After fractionation of the extract, ethyl acetate fraction showed the best ACE inhibitory activity, so the fraction was evaluated their antihypertensive activities *in vivo* using 2K1C method with some modification. 2K1C method can increase blood pressure rats until>150/100 mmHg after 6 w. After treatment for 2 w, the ethyl acetate fraction at dosage 50 mg/kg BB decreases blood pressure from 155/123 mmHg to 124/100 mmHg and the concentration of angiotensin II in plasma decreases from 39.83 pg/ml to 34.88 pg/ml. As a positive control, captopril decreases blood pressure from 155/122 mmHg to 112/95 mmHg and the concentration of angiotensin II in plasma decrease from 39.88 pg/ml to 31.8 pg/ml. Plasma renin concentration was not influenced by treatment of the fraction and captopril.

Keywords: Antihypertension, ACE inhibitor, Peperomia pellucida, Renin

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ANTI-DIABETIC PROPERTIES AND NUTRIGENOMIC BASIS OF CURCULIGO LATIFOLIA

I. N. AKMAL[1]*, I. MAZNAH[1,2]

[1]Nutricosmeceutical and Nutrigenomic Programme, Laboratory of Molecular Biomedicine, Institute of Bioscience, [2]Department of Nutrition and Dietetics, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia, Serdang, Selangor, Malaysia
Email: nur_akmal@upm.edu.my

ABSTRACT

Curculigo latifolia (C. latifolia) plant grows wildly in tropical Asia especially in Malaysia. C. latifolia fruit has 9000 times the sweetness of sucrose and it is due to a protein known as curculin. This indicates that C. latifolia plant has the potential to be used as an alternative low-calorie sweetener for diabetic patients. In the present study, anti-diabetic properties of C. latifolia fruit: root and the pertinent mechanism were investigated on diabetic-induced rats. Diabetes rats were developed by feeding high fat diet (HFD) which contained 56.9% calorie contributed by fat and low dose STZ (40 mg/kg bw) injection. Rats were administered with C. latifolia fruit: root at graded oral doses 50, 100 and 200 mg/kg b. w for 30 d. Before and after treatments, blood glucose, plasma lipid and adiponectin were measured to evaluate its anti-diabetic effects. The regulatory effects of C. latifolia fruit: root extracts on genes involved in glucose and lipid metabolisms were further studied on glucose transporter (GLUT 4), peroxisome proliferator-activated receptor (PPARγ) and adiponectin receptor (AdipoR1) in skeletal muscle tissue. The C. latifolia fruit: root possessed anti-diabetic activities as shown by the decreased of blood glucose, total cholesterol (TC), triglyceride (TG) and low density lipoprotein-cholesterol (LDL), as well as increased plasma level of adiponectin. Treatment with 200 mg/kg b. w of C. latifolia fruit: root extracts significantly improved glucose metabolism in diabetic-induced rats compared to other concentrations by increased gene expression of GLUT 4 (3 fold) and PPARγ (4 fold) tissue. Furthermore it improved lipid metabolism by increased the expression of AdipoR2 (3 fold). Based on the current findings, C. latifolia fruit: root extracts exhibit anti-diabetic properties on hyperglycemia and hyperlipidemia in diabetic rats possibly through regulating the levels of GLUT4, PPARγ and AdipoR2 tissue. Findings also provide an opportunity to food industries in designing product for diabetic complications.

Keywords: Curculigo latifolia, Diabetes, Gene expression, High fat fed diet, Streptozotocin

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EFFECT OF PROLONGED PHOSPHATE SUPPLEMENT CONTENT ADMINISTRATION OVER VERTEBRAL BONE VOLUME

U. F. HASHIM*, S. DOM

Faculty of Health Sciences, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: ummifarhanahashim@yahoo.com

ABSTRACT

Phosphate administration mediates the parathyroid hormone synthesis by promoting the genesis of the parathyroid gland hyperplasia. Excessive phosphate retention will increase the serum calcium levels. Ten (10) healthy New Zealand White rabbits aged 4 mo with body mass between 2.0 to 2.5 kg were used. They were divided into 2 groups which are the control group and experimental group. In experimental group, each rabbit was injected intravenously with 25 milligram of inorganic phosphorus, three times a day for 5 mo resulting in hyperparathyroidism which can detected by marked high serum calcium level. Following treatment, blood were collected for serum calcium examination and rabbit's 3rd lumbar spine were scan using Sky Scan 1176 Micro CT scanner for bone analysis Micro CT for trabecular bone volume (TBV), trabecular thickness (Tb. Th), trabecular number (Tb. N), and trabecular separation (Tb. Sp). In experimental group, the serum calcium was 4.61±0.601 mmol/l, TBV 109.74±21.327 mm3 Tb. Th 0.2157±0.022 mm Tb. N 0.273±0.629 1/mm and Tb. Sp 3.824±0.344 mm. While the normal group the serum calcium was 3.49±0.147 mmol/l, TBV 140.98±7.368 mm3, Tb. Th 0.238±0.029 mm Tb. N 0.380±0.040 1/mm and Tb. Sp 2.527±0.121 mm. Prolonged administration of phosphate supplement content show an adverse effect on rabbit 3rd lumbar, altering the trabecular bone architecture by increasing the calcium level resulting in the increasing of bone Tb. S and reducing the TVB, Tb. N. Altering the trabecular bone architecture will reduce the bone strength and quality which can increase the risk of fracture.

Keywords: Bone volume, Calcium, Phosphorus, Micro-CT

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ANIMAL ALTERNATIVES: PRACTICES AND INITIATIVES IN SINGAPORE

M. V. F. VALLARTA*

National University of Singapore, Institutional Animal Care and Use Committee (IACUC), Centre for Life Sciences, Singapore, Singapore
Association for Laboratory Animal Science (SALAS), Singapore
Email: vallarta@nus.edu.sg

ABSTRACT

Singapore is renowned for its thrust towards nurturing world-class scientific research and talent to drive economic growth transform the country into a vibrant knowledge-based and innovation-driven economy. With numerous multinational companies shifting focus to faster-growing emerging markets in Asia, Singapore has had its fair share of businesses either making the country their regional hub or completely transferring their manufacturing operations to the city-state. Together with this move also came the corporate laboratories which, in addition to Singapore's existing research in the academy, have brought about a sharp increase in the number of animals used in research and development (R&D). In recent years, as an ever-increasing number of countries make moves to end the testing of pharmaceuticals, nutraceuticals and cosmetics on animals, the Singapore R&D community has made innovations in the application and development of alternative methods. An overview of the progress in Singapore on animal replacement in research, teaching and testing is presented, with particular reference to technologies that have been adopted and developed locally.

Keywords: Alternatives, Animal Replacement, Refinement

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INTERNATIONAL TREND OF 3RS AND ASIAN PROSPECTIVE

T. M. KUROSAWA*

Animal Welfare Research, Kagoshima University, Japan
Email: japanawr@gmail.com

ABSTRACT

The tenet of 3Rs in animal research is well recognized internationally. Many advanced countries amend their animal welfare regulations and they emphasize 3Rs. This trend reflects to International Standard Guidelines of laboratory animal welfare. The OIE; World Animal Health Organization sets use of animals in research and education in its international standard; Terrestrial Animal Health Code in 2010. CIOMS with ICLAS revised its guiding principles in 2012. ISO10993-2 Animal Welfare Requirement was revised in 2014 and may be revised again soon. EU Directive for laboratory animal protection was revised in 2010 and ILAR guide of USA was revised and published as the 8th edition in 2011. OECD keeps rabbit eye test as TG405 in 2012 but the laboratory animal care for the testing animals became more stringent. The academic associations for 3Rs are very few apart from JSAAE which was founded in 1984. Now EUSAAT was founded in 2010 and ASCCT was founded in 2012. These academic associations could lead this field with the World Congress of Alternatives. In Asia, apart from Japanese activities, Koreans are keen to advance this field and founded KSAAE in 2006. P. R. China sent a plenary speaker for WC9 in 2014 and there are various activities in 3Rs reported in Asian countries. The main activities of 3Rs in various Asian countries are lead by the laboratory animal science associations acting as AFLAS members. AFLAS is keen in laboratory animal welfare and the next AFLAS congress will be held in Singapore. AAALAC International is keen to propagate 3Rs tenet to PAC RIM countries including Asia and Oceania and the number of accredited research institutions in this region is increasing remarkably. In near future the number of accredited institutions will reach to 200 hundred. With these Asian activities, JSAAE decided to organise Asian Congress of Alternatives in 2017. Asian prospective of 3Rs and laboratory animal welfare could have bright future with Asian specialists in this field.

Keywords: 3Rs, International, Asia

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OECD *IN VITRO* SKIN SENSITIZATION DRAFT GUIDELINE: HUMAN CELL LINE ACTIVATION TEST (h-CLAT)

T. ASHIKAGA*

Shiseido Co. Ltd. Japan
Email: Takao.ashikaga@to.shiseido.co.jp

ABSTRACT

The activation of dendritic cells (DC), typically assessed by expression of specific cell surface markers, is the key event of Adverse Outcome Pathway (AOP) of skin sensitization. Evaluation of changes in cell surface marker expression induced in dendritic cells (DC) or DC-surrogate cell lines following exposure to sensitizers represents one approach for the development of *in vitro* skin sensitization test. The h-CLAT is one of these assays. The h-CLAT examines by flow cytometry the level of expression of CD86 and CD54 on the surface of THP-1 cells (human monocytic leukemia cell line), following 24 h of chemical exposure. About 100 chemicals were evaluated in the h-CLAT and compared to LLNA. The accuracy of the h-CLAT vs. LLNA was 84%. Most chemicals were evaluated correctly, but a few chemicals failed. Most of the false negative chemicals were low solubility chemicals, pro/pre haptens and so on. Next, we calculated the estimated concentration of the positive threshold for both induction of CD86 and CD54 as RFI=150 for CD86 (EC150) or 200 for CD54 (EC200). The Minimum Induction Threshold (MIT) was determined as a smallest value of either EC150 or EC200. The MIT showed a good correlation with EC3. From these data, the h-CLAT has a possibility of classifying the allergic potency of chemicals. The h-CLAT has been evaluated by five independent laboratories in COLIPA and seven laboratories in Japan using same chemicals. These inter-laboratory studies have found the h-CLAT to be transferable and reproducible. These above data confirms the reliability and relevance of the h-CLAT. Currently OECD opens the draft guideline. Current situation of standardization of the test will be presented.

Keywords: Skin sensitization, *In vitro*, Sensitizer

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A SURVEY ON THE INTEREST AND THE KNOWLEDGE OF PHARMACY STUDENTS AT UNIVERSITI TEKNOLOGI MARA, PUNCAK ALAM ON VETERINARY PHARMACY

F. KOLENUS, V. SUBRAMANIAM*

Pharmacology and Toxicology Research Laboratory, Faculty of Pharmacy, Universiti Teknologi MARA (UiTM), 42300 Bandar Puncak Alam, Selangor Darul Ehsan, Malaysia
Email: vellayan@salam.uitm.edu.my

ABSTRACT

The roles of the veterinarians and the pharmacists were mutually supportive whereby the role of diagnosing and prescribing drugs is undertaken by the veterinarians while compounding and dispensing drugs to the animals is undertaken by the pharmacist. The lack of veterinary and animal health education in the pharmacy curriculum may influence the graduate's choices of their career path. So this study is aimed to survey the interest and knowledge of the pharmacy students at UiTM Puncak Alam toward Veterinary Pharmacy. Survey through the use of a questionnaire consisting 4 different sections was formulated to identify pharmacy students at UiTM Puncak Alam, who have the interest and knowledge in Veterinary Pharmacy. Surveys at Faculty of Pharmacy UiTM suggest an interest in Veterinary Pharmacy is high in second year students and low in fourth year students. Results also indicate that animal lovers, students and students having pets at home expressed interest in Veterinary Pharmacy. Knowledge of pharmacy students was shown to be deficient in these areas, since only five knowledge statements hit more than 50% and the other four knowledge statements hit less than 50%. The interest toward veterinary pharmacy was shown to be high from second year students. This group of students was also found to be interested to work as a veterinary pharmacist upon their graduation. This survey finding indicates that there is a need for education in this field. The school of pharmacy in Malaysia must identify the needs of this subject "Veterinary Pharmacy" as an optional or necessary subject to pharmacy students.

Keywords: Veterinary pharmacy, Pharmacy students, Malaysia

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CIPROFLOXACIN STIMULATES STEMNESS-RELATED SIGNAL IN HUMAN DERMAL PAPILLA CELLS

C. KIRATIPAIBOON[1], P. TENGAMNUAY[2], P. CHANVORACHOTE[3,4]*

[1]Pharmaceutical Technology (International) Program, [2]Department of Pharmaceutics and Industrial Pharmacy, [3]Department of Pharmacology and Physiology, [4]Cell-Based Drug and Health Product Development Research Unit, Faculty of Pharmaceutical Sciences, Chulalongkorn University, Bangkok, Thailand
Email: pithi.c@chula.ac.th

ABSTRACT

An improved expansion method for adult stem cell may enhance its use in regenerative therapy. Ciprofloxacin has been used for the prevention of bacterial infection in patients receiving stem cell transplant and in stem cell research; however, its effects on the stem cells, especially dermal papilla cells have not been fully elucidated. This study aimed to investigate the effect and underlying mechanism of ciprofloxacin in maintaining the stemness of dermal papilla cells. Human dermal papilla cell line and primary human dermal papilla cells were selected as representatives of multipotent stem cells. Various stem cell molecular markers, epithelial to mesenchymal markers and Wnt/b-catenin-associated proteins were evaluated by western blot analysis and immunocytochemistry to prove the beneficial effect of ciprofloxacin on the cells. The present study demonstrated that treatment of dermal papilla cells with nontoxic concentrations of ciprofloxacin could prevent the loss of stemness during culture. The ciprofloxacin-treated cells were found to maintain stem cell morphology, clonogenicity and stem cell markers in the culture. The results revealed that ciprofloxacin possessed its effect through ATP-dependent tyrosine kinase/glycogen synthase kinase3b dependent mechanism which in turn up-regulated b-catenin. Furthermore, the down-stream stemness-related transcription factors of Wnt/b-catenin pathway, namely, Oct-4 and Nanog were significantly up-regulated in the ciprofloxacin-treated cells. Also, ciprofloxacin was shown to induce epithelial to mesenchymal transition in dermal papilla cells as the transcription factors ZEB1 and Snai1 were significantly up-regulated. The effects of ciprofloxacin in preserving stemness features were also exhibited in the primary dermal papilla cells directly obtained from human hair follicles. These results unveiled a novel application of ciprofloxacin for the cell therapeutic approaches and provided its molecular mechanisms for maintenance of the stemness features in human dermal papilla cells.

Keywords: Ciprofloxacin, Dermal papilla cell, Stem cell, Epithelial to mesenchymal transition, β-catenin

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ANTI-PROLIFERATIVE AND ANTI-INVASION EFFECT OF ASIATIC ACID AGAINST ORAL SQUAMOUS CELL CARCINOMA*IN VITRO* AND IN ORTHOTOPIC XENOGRAFT MODEL STUDIES

N. CHE AZIZ[1], F. KHAN[2], J. MAHMOOD[2], A. ADAM[1]*

[1]Pharmacology Toxicology Research Lab, Faculty of Pharmacy, OASES, Pharmaceutical & Life Sciences CORE, Universiti Teknologi MARA (UiTM), Puncak Alam, Selangor, Malaysia, [2]Department of Radiation Oncology, University of Maryland School of Medicine, Baltimore, Maryland

Email: aishah_adam@puncakalam.uitm.edu.my

ABSTRACT

Oral cancer is among the most common cancers in South East Asia with a survival rate of 50% for the past thirty years due to poor prognosis. Oral squamous cell carcinoma (OSCC) represents 80-90% of all malignant neoplasms of oral cavity. However, effective therapy against OSCC is a great challange. This study was aimed at evaluating the anti-proliferative effect of Asiatic acid (AA), a pentacyclic triterpene against OSCC *in vitro* and its protective effect in orthotopic xenograft model of oral cancer. Antiproliferative activity was determined by MTS assay. Expression of genes that account for cell death namely Bax and Bcl-2 were determined by quantitative RT-PCR with AA (30 and 40 µM). Protective effect of AA (1-10 mg/kg, i. p.) *in vivo* was studied by its daily administration for 20 d prior to tumour inoculation on day 21 to tongue of severely compromised immunodeficiency (SCID) mice. Treatment with AA was then continued for a further 24 d. IC50 for AA in OSCC at 24, 48 and 72 h were 36±4, 38±1.7 and 12±3.5 µM, respectively. AA significantly ($p<0.05$) initiated apoptosis by down regulation of Bcl-2 gene by 68% and up regulation of Bax gene by two folds. Eventhough AA did not affect tumour formation; the tumours that were formed were smaller in their area of invasion. In conclusion, AA showed promise as an antiproliferative agent against oral squamous cell carcinoma *in vitro* and prevented invasion of tumour *in vivo*. This study provided a theoretical reference for the exploration of new anticancer agents and may be useful for the design of potent adjuvant chemotherapeutic drugs.

Keywords: Asiatic acid, Oral squamous cell carcinoma (OSCC), Apoptosis, Orthotopic xenograft model, Tumour invasion

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INHIBITORY EFFECT OF 2-BENZOYL-6-(3,4 DIHYDROXYBENZYLIDENE)CYCLOHEXEN-1-OL ON CHEMICAL-INDUCED NOCICEPTION MODEL IN MICE

A. F. A. AZMI, E. K. PERIMAL, A. AKIRA, D. A. ISRAF, T. A. S. TENGKU MOHAMAD, M. R. SULAIMAN*

Department of Biomedical Science, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia, Serdang, Selangor, Malaysia
Email: mrs@upm.edu.my

ABSTRACT

Some analgesic drugs like morphine still remains as the most effective narcotic analgesic over the years to treat severe and chronic pain. The continuous and heavy applications of drugs have often led to severe health problems like gastropathy, kidney failure and liver damage. These adverse side effects of the contemporary painkillers have accelerated the studies of other analgesic compounds with comparable effects yet limited undesirable side effects. In the present study, we have evaluated the antinociceptive property of synthetic Diaryl-penta-noidanalogue, 2-benzoyl-6-(3,4-dihydroxybenzylidene)cyclohexen-1-ol (BDC) using acetic acid-induced abdominal writhing test in mice. BDC (1, 3, 10 and 30 mg/kg), vehicle (10 ml/kg) or aspirin (ASA, 10 mg/kg) were pre-administered intraperitoneal, 30 min before intraperitoneal injection of 0.8% acetic acid. The number of abdominal writhes was recorded for 30 min, starting from 5 min following acetic acid injection. It was demonstrated that BDC significantly inhibited ($P<0.05$) the writhing response induced by acetic acid in dose dependent manner. The percentage of inhibition produced by BDC at 1, 3, 10 and 30 mg/kg was 64.42, 80.52, 97.19and 99.78% as compared with control, respectively. Such effects were also observed in mice pre-treated by ASA (63.42%, $P<0.05$). Findings suggested that 2-benzoyl-6-(3,4-dihydroxybenzylidene)cyclohexen-1-ol exerted marked antinociceptive activity when assessed in chemical model of induced nociception in mice.

Keywords: 2-benzoyl-6-(3,4-dihydroxybenzylidene)cyclohexen-1-ol, Antinociceptive

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SPATIAL MEMORY ENHANCEMENT OF CIPROXIFAN AGAINST SCOPOLAMINE-INDUCED AMNESIA IN ICR MICE VIA BDNF SIGNALING PATHWAY

S. MOHD NOH[1,2], V. MANI[1,2]*, K. RAMASAMY[1,3], A. PRAKASH[1,2], A. B. ABDUL MAJEED[1,2]

[1]Faculty of Pharmacy, [2]Brain Degeneration and Therapeutics Group, [3]Collaborative Drug Discovery Research Group, Pharmaceutical and Life Sciences CoRe, Universiti Teknologi MARA (UiTM), Puncak Alam Campus, Selangor, Malaysia
Email: vasudevan@puncakalam.uitm.edu.my

ABSTRACT

The well-known pathological hallmark of Alzheimer's disease (AD) is neuronal loss as well as amyloid-beta (Aβ) deposition and neurofibrillary tangles (NFT). There are few drugs from acetylcholinesterase (AChE) antagonist and NMDA antagonist that have been approved by FDA to treat AD. However, no exact treatments were able to stop the disease progression. H3-receptor is an auto receptor that controls the brain histamine release as well as other neurotransmitters such as dopamine and acetylcholine (hetero receptor). We used a drug called as ciproxifan (H3-receptor antagonist) that functions by blocking the H3-receptor and allows more histamine and other neurotransmitters including acetylcholine to be released. Recently, ciproxifan has been shown to promote vigilance and attention as well as enhances cognition and memory in behavioral studies. In the present study, we used scopolamine to induce memory impairment in ICR mice. Groups of mice were injected with ciproxifan (1 & 3 mg/kg, i. p.) for 30 consecutive days and scopolamine (3 mg/kg, i. p) on the last 4 d of treatment, 30 min before the behavioral task. We have identified that ciproxifan (3 mg/kg, i. p.) increased the memory ability and learning assessment by Morris Water Maze task, increased the level of acetylcholine (ACh), and decreased the activity of AChE. Besides that, a significant elevation of brain-derived neurotrophic factor (BDNF) level was noted in ciproxifan-treated groups as compared to scopolamine-induced group, which showed ciproxifan may increase learning and memory through the BDNF signaling pathway. In conclusion, the present results of ciproxifan may promise, a potential agent for the treatment of AD and other neurodegenerative diseases.

Keywords: Ciproxifan, Memory impairment, Acetylcholine, Brain-derived neutrophic factor, Alzheimer's disease.

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THE EFFECTS OF EQUAL DOSES OF SULFORAPHANE, CURCUMIN, QUERCETIN, BUTYLATED HYDROXYANISOLE AND INDOLE-3-CARBINOL ON GLUTAMATE-CYSTEINE LIGASE CATALYTIC SUBUNIT (GCLC) GENE AND PROTEIN EXPRESSION IN MICE LIVER

A. ABDULLAH*, N. ALRAWAIQ, A. ATIA

Faculty of Medicine, Department of Pharmacology, Universiti Kebangsaan Malaysia, Medical Centre (UKMMC), Kuala Lumpur, Malaysia
Email: manlah1969@yahoo.com

ABSTRACT

Glutamate-cysteine ligase catalytic subunit (Gclc) is an enzyme that in mouse is encoded by the Gclc gene. Glutamate-cysteine ligase is the first rate limiting enzyme of glutathione synthesis. The objective of this study is to determine the effects of administration of equal dose (50 mg/kg) of several dietary chemicals on the gene and protein expression levels of Gclc in mice livers. Adult male ICR white mice were divided into 7 groups (N=6 per group), i.e. sulforaphane, quercetin, curcumin, butylated hydroxyanisole (BHA), indole-3-carbinol (I3C), vehicle 1 (control 1) and vehicle 2 (control 2) groups. The chemicals were administered intraperitoneally for 14 d at a dose of 50 mg/kg body weight. Vehicle 1 (DMSO, tween-20 and normal saline in the ratio of 0.05:0.1:0.85) was the solvent for sulforaphane, curcumin and quercetin. Vehicle 2 (corn oil) was the solvent for BHA and I3C. At day 15, mice were sacrificed and their livers harvested. Total RNA was extracted from the liver, reverse transcribed and subjected to quantitative real-time PCR to detect Gclc gene expression. Agarose gel electrophoresis was performed to verify the specificity of amplification. Western blots were performed to detect Gclc protein expression. Sulforaphane treatment resulted in 3.5-fold and 2.6-fold induction of Gclc gene and protein respectively, compared to controls ($P<0.05$). Curcumin treatment resulted in 2.6-fold and 1.7-fold induction of Gclc gene and protein respectively ($P<0.05$). Quercetin treatment resulted in 2.5-fold and 1.4-fold induction of Gclc gene and protein respectively ($P<0.05$). BHA treatment resulted in a 2.8-fold and 1.9-fold induction of Gclc gene and protein respectively ($P<0.05$). I3C treatment resulted in a 2.8-fold and 1.9-fold induction of Gclc gene and protein respectively ($P<0.05$). At 50 mg/kg, sulforaphane is the most potent in inducing Gclc expression, followed by I3C and BHA (equipotently), curcumin and quercetin.

Keywords: Gclc, Gene/protein expression, Dietary chemicals

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DIFFERENTIAL EXPRESSION OF SERUM METABOLITE PROFILES IN OBESE AND HEALTHY LEAN OF KUALA SELANGOR VOLUNTEERS

S. N. HUSSIN[1]*, J. K. S. SHIA[1], A. AHMAD[2], M. OMAR[2]

[1]Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Faculty of Medicine, Universiti Teknologi MARA, Selangor, Malaysia
Email: nooraishah0352@puncakalam.uitm.edu.my

ABSTRACT

Metabolomic studies on obesity are believed to bring insights into discovery of novel biomarkers with comprehensive understanding at metabolic levels. In conjunction to this, global or untargeted LCMS-QTOF based comprehensive metabolomics profiling was employed in this study. A total of 80 aged-matched respondents, that include 40 obese and 40 healthy lean as control, were selected and the serum metabolites were profiled. The differentially expressed metabolites were determined and analysed using Principal Component Analysis (PCA) and Receiver Operative Characteristic Curve Explorer and Tester (ROCCET). A total of 225 metabolites were identified, in which 48 metabolites passed the univariate significant analysis at corrected p-value<0.01 with fold change cut-off of = 2. A total of 27 metabolites demonstrated an up-regulation in the serum of obese volunteers. Classes of metabolites include steroids, fatty acyls, benzene and substituted derivatives, linoleic acids and derivatives, prenol lipids, carboxylic acid and derivatives, pronol lipids, glycerophospholipid, and lineolic acids and derivatives. In contrast, a total of 21 metabolites demonstrated a down-regulation in the serum of obese volunteers. Classes of metabolites include fatty acids and conjugates, quarternary ammonium salts, fatty acyls, azolines, indoles and derivatives, cinnamaldehydes, carboxylic acids and derivatives, sphingolipids, prenol lipids, tetrapyroles and derivatives, and carbohydrates. In relation with the regulation of these metabolites in the analysis, the highest AUC value for up-regulated metabolites in obese is 7,7-dimethyl-5Z,8Z,11Z-eicosatrienoic acid which is 0.887 with range of confidence intervals of 0.811-0.951 and 90 percent of sensitivity and 80 percent of specificity. In contrast, the down-regulated metabolite in obese that contribute to the best performance classifier is the phosphorylcholine. Several pathways are suggested to be closely involved with these potential biomarkers such as the glycerophospholipid metabolism and steroid hormone biosynthesis in the up and down regulatory process.

Keywords: Metabolomic, Obesity, Global profiling, PCA, ROCCET

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COMPARATIVE STUDY OF CONVENTIONAL AND SUPERCRITICAL FLUID EXTRACTION METHODS FOR THE ISOLATION OF SECONDARY METABOLITES FROM SYZYGIUM CAMPANULATUM KORTH

A. H. MEMON[1], F. RITHWAN[2], S. ZHARI[2], M. S. R. HAMIL[1], M. A. AHMED SAEED[1], Z. ISMAIL[1]*, A. M. S. ABDUL MAJID[3]

[1]Department of Pharmaceutical Chemistry, School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang, Malaysia, [2]Centre of Lipids Engineering and Applied Research (CLEAR), Ibnu Sina Institute for Scientific & Industrial Research (Ibnu Sina ISIR), Universiti Teknologi Malaysia, Johor, Malaysia, [3]EMAN Research and Testing Laboratory, School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang, Malaysia
Email: zhari@usm.my

ABSTRACT

Two flavanones; (2S)-7-hydroxy-5-methoxy-6,8-dimethyl flavanone (HMDF), (S)-5,7-dihydroxy-6,8-dimethyl-flavanone (DDF), chalcone; (E)-2',4'-dihydroxy-6'-methoxy-3',5'-dimethylchalcone (DMC) and two triterpenoid; betulinic and ursolic acids (BA and UA) are present in Syzygium campanulatum Korth (Myrtaceae). In this work, the recovery of five compounds from leaves of S. campanulatum was studied using different solvents based and conventional methods of extraction, namely maceration (MAC), soxhletion (SOX) and reflux (REF) using ethanol, ethyl acetate, methanol, n-hexane, mixture of n-hexane: methanol (1:1) and water as solvents. Furthermore, preliminary and pilot scale studies of Supercritical Fluid Extraction (SFE) of S. campanulatum leaves were also performed. The solvent based conventional extraction techniques (MAC, SOX and REF) recovered five compounds in the range 0.1-15.0%, respectively. An enriched extract with five compounds was carried out using n-hexane: methanol (1:1) solvents using SOX technique. SFE method produced higher recoveries (0.1-60.0%), of all five compounds as compared to conventional extraction techniques. The addition of food grade ethanol as modifier/co-solvent produced prominent improvement of both chalcone and betulinic acid recovery.

Keywords: Secondary metabolites, Supercritical fluid extraction, Syzygium campanulatum, Conventional extraction

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DESIGN AND SYNTHESIS OF ORTHO-, META-AND PARA-XYLYLGUANIDINIUM Zn^{2+}-CYCLEN COMPLEXES AND ITS INTERACTION TOWARDS DEOXYRIBONUCLEIC ACID (CYCLEN-1, 4, 7, 10-TETRAAZACYCLODODECANE)

N. A. HASSAN[1,2]*, M. Z. MAT JUSOH[1,2]

[1]Atta-ur-Rahman Institute for Natural Product Discovery, [2]Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: aminmerican@gmail.com

ABSTRACT

The three of new Zn2+complexes C1, C2 and C3 were designed and synthesised by coordination of Zn2+into the integrated 1,4,7,10-tetraazacyclododecane (Cyclen) with ortho-, meta-and para-bromoxylylguanidinium pendants group at one of the nitrogen atoms. All the Zn2+complexes structures have been characterized by using 1H and 13C NMR spectroscopy, infrared spectroscopy and elemental analysis. The aims of synthesising these Zn2+complexes are to investigate the position effect of guanidinium group of Zn 2+-cyclen complexes towards interaction with deoxyribonucleic acid (DNA) and to explore its potential application in the field of medicinal chemistry. Hence, ethidium bromide (EB) fluorescence assay and circular dichroism (CD) spectroscopy have been used to ascertain the interaction between Zn2+complexes towards natural calf thymus DNA (ctDNA) and its mechanism. By implying the emission of EB at 590 nm and excitation at 510 nm, the addition of 12-15 μM of C1, C2, and C3 have decreased the emission intensity of EB-ctDNA solution and its complexation concentration were reduced to 50%. The addition of these Zn2+complexes did replace the EB until the complete intercalations in ctDNA occurred. CD spectroscopy results also revealed that C1 has disturbed both base stacking as well as right handed helicity properties of ctDNA at 277 nm positive and 245 nm negative bands respectively, but retained the B-form of ctDNA structures. Meanwhile, C2 and C3 have transformed the conformation of ctDNA from B-form into Z-form. Further study on the thermal melting of ctDNA, cleavage of the P—O bonds or phosphate hydrolysis within the model phosphodiester bis (p-nitrophenyl) phosphate (BNPP) and supercoiled pBR 322 plasmid DNA will be determined by thermal melting UV analysis and Gel Electrophoresis.

Keywords: Cyclen, Guanidinium, Zinc complex, DNA, Medicinal chemistry

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SERUM METABOLIC PROFILING OF MALAYSIAN ALZHEIMER'S DISEASE PATIENTS UNRAVEL PERTURBED SPHINGOLIPID AND AMINO ACID PATHWAYS

C. N. A. ENCHE ADY[1,2], S. M. LIM[1,2], A-V. CHIN[5], S. B. KAMARUZZAMAN[5], M. P. TAN[5], P. J. H. POI[5], L. K. TEH[4], M. Z. SALLEH[4], I. K. S. YAP[6], A. B. ABDUL MAJEED[1,3], K. RAMASAMY[1,2]*

[1]Faculty of Pharmacy, [2]Collaborative Drug Discovery Research (CDDR) Group, Pharmaceutical and Life Sciences Community of Research, [3]Brain Degeneration and Therapeutics Group, [4]Integrative Pharmacogenomics Institute (iPROMISE), Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [5]Division of Geriatric Medicine, Department of Medicine, [6]Department of Life Sciences, International Medical University, Kuala Lumpur, Malaysia

Email: kalav922@gmail.com

ABSTRACT

Alzheimer's disease (AD) is a neurodegenerative disorder that is characterised by loss of memory and cognitive function. As such, there is a dire need for early diagnosis of AD so as to effectively delay disease progression. The present study profiled differential metabolomics of Malaysian AD patients (n=55) vs healthy subjects (n=55) utilising combined platforms of Liquid Chromatography/Mass Spectrometry (LCMS)/Nuclear Magnetic Resonance (NMR). Analyses were performed by using Mass Profiler Professional (MPP), ROCCET, MetPA and SIMCA softwares. The results of the combined platforms were found to be complementary in nature. LC/MS analysis revealed a total of 4 lipid metabolites [fold change>2; $p<0.05$; Area under Receiver Operating Characteristic] that were found (AUROC) to be differentially =0.8] that were found to be differentially up-regulated amongst AD patients. These lipid metabolites included N-(2-hydroxyethyl) palmitamide, N-(2-hydroxyethyl) icosanamide, C2-dihydroceramide and phytosphingosine. NMR analysis, on the other hand, identified 4 differential metabolites ($p<0.05$; AUROC=0.6). Isoleucine was significantly up-regulated whereas creatinine, very low density lipoprotein (VLDL) and an unidentified lipid were significantly down-regulated in AD patients. Metabolic pathway analyses of results generated from the combined platforms indicated involvement of sphingolipid (impact value: 0.298) and amino acid metabolism (impact value: 0.013). These pathways are linked to apoptosis, oxidative stress and brain oxidative metabolism impairment, all of which could give rise to AD. The present investigation, has uncovered potential AD biomarkers which could serve as an important insight into understanding of the pathogenesis of AD.

Keywords: Metabolomics, Serum, Alzheimer's, Pathway

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PROLIFERATIVE EFFECTS OF CENTELLA ASIATICA ON *IN VITRO* HUMAN MESENCHYMAL STEM CELLS (hMSCs)

N. OMAR[1], Y. LOKANATHAN[1], Z. R. MOHD RAZI[2], A. MOHD ILHAM[3], R. IDRUS[1,4]*

[1]Tissue Engineering Center, Universiti Kebangsaan Malaysia Medical Center, [2]Department of Obstetrics and Gynecology, Faculty of Medicine, Universiti Kebangsaan Malaysia, Kuala Lumpur, Malaysia, [3]Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, [4]Department of Physiology, Faculty of Medicine, Universiti Kebangsaan Malaysia, Kuala Lumpur, Malaysia
Email: ruszyidrus@gmail.com

ABSTRACT

Centella asiatica (L.) is a traditional medicinal herb that has been used since ancient times. It had been documented to have numerous pharmacological effects on different cell types. Nevertheless, its proliferative effects against human mesenchymal stem cells (hMSCs) are largely unknown. Keeping emphasis on its properties, this present study was conducted to evaluate the *in vitro* effects of raw extract of Centella asiatica (L.), (RECA) on the proliferation of human mesenchymal stem cells (hMSCs), for the subsequent neural induction. RECA was prepared in the ethanolic form and hMSCs were isolated from human Wharton's jelly umbilical cord. MTT assay was used to measure cell viability upon treatment with different concentrations of RECA (400, 800, 1200, 1600, 2000 and 2400 µg/ml) and RECA LD50 value was determined directly from dose-response curve. Cell proliferation rate was measured on the 1st, 3rd, 5th to 7th day of post-RECA treatment below the LD50 value. The result showed that RECA has cytotoxic effects against hMSCs above 1600 µg/ml with LD50 value of 2200 µg/ml. The proliferative effects of RECA on hMSCs are concentration-dependent too. It was found that RECA concentration up to 800 µg/ml reduced the hMSCs proliferation rate, while concentration beyond 800 µg/ml inhibited cell proliferation totally. This pilot study suggests that even the lowest tested concentration (400 µg/ml) RECA is anti-proliferative to hMSCs. However, RECA concentration 800 µg/ml and below are not cytotoxic and will be used as a preliminary test dose for neural induction that will be performed. Further investigations are ongoing to verify the findings before proceeding to downstream analysis.

Keywords: Pegaga, Cytotoxic, Wharton's jelly mesenchy.

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PLANT-PRODUCED RABIES VIRUS NEUTRALISING SINGLE-CHAIN ANTIBODY FOR THERAPEUTIC DELIVERY

W. PHOOLCHAROEN*

Pharmacognosy and Pharmaceutical Botany, Faculty of Pharmaceutical Sciences, Chulalongkorn University, Bangkok, Thailand
Email: waranyoo.p@chula.ac.th

ABSTRACT

Rabies virus (RABV) is a neurotropic virus that can cause fatal disease in humans and animals. Once RABV enters the central nervous system, antibodies cannot pass through the blood-brain barrier to neutralise the virus. Therefore there is no effective treatment for the patient at this stage. The goal of this study is to develop plant-produced rabies single-chain antibody (ScFv) to be able to cross the blood-brain barrier (BBB) and neutralise RABV. The ScFv was fused with a peptide from rabies glycoprotein (RVG). RVG was previously reported to be a specific target peptide for the brain because it interacts specifically with nicotinic acetylcholine receptor (NAchR) occured in high density at the neuromuscular junction and presented in the central nervous system. The rabies ScFv-RVG fusion protein (ScFv-RVG) is produced from Nicotiana benthamiana leaves by transient expression and showed the neutralisation activity to RABV. Comparing to ScFv alone, ScFv-RVG increases the binding to NAchR and the entry in to neuronal cells. In addition, hCMEC/D3 cell line, human brain endothelial cell, was used as an *in vitro* BBB model. The result demonstrated that the plant-produced ScFv-RVG is able to transport across the hCMEC/D3 cells on the trans well filters. In conclusion, the plant-produced ScFv-RVG fusion protein is able to neutralise RABV, bind to NAchR and transport across the BBB *in vitro*. The transport of the plant-produced ScFv-RVG across the BBB *in vivo* will be performed in the future.

Keywords: Rabies virus, Plant-produced rabies single-chain antibody, Nicotinic acetylcholine receptor, Neuronal cells

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SYNTHESIS AND ANTI-BACTERIAL EVALUATION OF 2-(4'-SUBSTITUTEDPHENYL)-3-[4-(N-(1-DISUBSTITUTED AMINO) METHYL-2-OXO INDOILIN-3-YLIDENE] BENZENESULFONYL QUINAZOLIN-3-(4H)-ONE DERIVATIVES

G. NATESAN*, Z. X. LAI, J. KAUR, S. MENON, C. H. LIM, A. MAHMUD

Department of Medicinal Chemistry, Faculty of Pharmacy, MAHSA University, Kuala Lumpur, Malaysia
Email: gopal@mahsa.edu.my

ABSTRACT

The multidrug resistant is one of great concerns for the healthcare and it could be a life threatening situation. It is essential to synthesise novel antimicrobial agents to deal with increased number of multidrug resistance organisms (MDRO) and limited antimicrobial agents. Literature survey showed that quinazolinone possesses varied biological activities; 2nd and 3rd positions are the target for substitution with other moieties. On the other hand, sulphanilamide derivatives have been reported to possess significant antibacterial activities through competitive inhibition of dihydropteroate synthetase enzyme and isatin moiety also displayed valuable biological activities. Hence, it was thought worthwhile to study the effects of three pharmacophoric moieties like quinazolinone, sulphanilamide and isatin in a single molecule of the antibacterial activity. Series of novel 2,3-disubstituted quinazolin-4(3H)-one derivatives have been synthesized from the intermediate Schiff base of 2-(4'-substitutedphenyl)-3-[(N-2-oxoindolin-3-ylidene)-4"-sulphonamidophenyl]-quinazolin-(3H)-one derivatives, which was prepared from reacting 2-(substituted phenyl)-4H-benzo[d][1,3]-oxazin-4-one with sulphanilamide. The required benzoxazinone derivates was prepared by reacting anthranilic acid with benzoyl chloride. All the compounds structures were characterised by using H1 NMR and melting point was recorded. The intermediate Schiff base and final Mannich base compounds were evaluated for their antibacterial activity against Staphylococcus aureus, Bacillus cereus, Escherichia coli, and Pseudomonas aeruginosa at a concentration of 50µg/ml and 100µg/ml by agar well diffusion method using Norfloxacin (50µg/ml) as standard drug. From the results of the study, it has been observed that final Mannich base showed a better activity when compared to the parent Schiff bases except compound with diethyl substitution, which is devoid of antibacterial activity. The substitution of N-methyl piperazine at the 1st position of the indole nucleus of the final quinazolinone derivatives increases the antibacterial activity against the entire microorganism. In a conclusion, all the test compounds exhibited lower degree of antibacterial activity when compared to standard drug.

Keywords: Quinazolin-4(3H)-one, Sulphanilamide, Isatin, Mannich, Antibacterial.

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SYNTHESES, CHARACTERISATION AND ANTIPROLIFERATIVE ACTIVITIES OF MORPHOLINE AND COUMARIN ANALOGUES

M. A. MOHD FARIDZ[1,2]*, S. A. A. SHAH[1,2], M. T. HUSSIN[1,3], S. SULTAN[1,2], M. Z. MAT JUSOH[1,2]

[1]Atta-ur-Rahman Institute for Natural Products Discovery (AuRIns), [2]Faculty of Pharmacy, Universiti Teknologi MARA (UiTM), Puncak Alam Campus, Selangor, Malaysia, [3]Faculty of Applied Sciences, UiTM Shah Alam, Selangor, Malaysia
Email: fifa200208@gmail.com

ABSTRACT

Drug discovery is the process through which potential new medicines are identified. It involves a wide range of scientific disciplines, including biology, chemistry and pharmacology. Organic synthesis has overcome the limitations of the compounds from natural products, which provides and develops new pharmaceutical sciences field and lead compounds drug design program. In order to develop biologically active synthetic analogues, coumarin, a plant-based phenolic compound, and morpholinothiophene were chosen as the starting material due to their wide variety of biological activities such as urease inhibition, anti-inflammatory, antioxidant, CYP3A and antifungal activities. Research was focused on molecules which were synthesized from Schiff bases chemistry. It has been expanded enormously biological activities such as antimicrobial, antifungal, antibacterial, anticancer, antimalarial, antitubercular, antiviral, anti-inflammatory etc. Schiff bases are condensation products of primary amines with carbonyl compounds used as the basic synthetic method forming the hydrazone analogues of morpholine and coumarin. All synthetic analogues were screened for antiproliferative activity through MTT assay in hepatocellular carcinoma cell line (HepG2) and breast cancer cell lines (MCF-7). Several synthetic candidates of morpholine and coumarin showed potent inhibition of MCF-7 and several showed potent inhibition to HepG2 cell lines, while several of the candidates were showed potency to inhibit both of cell lines. The standard drugs used for MCF-7 was tamoxifen and for HepG2, doxorubicin in order to calculate IC50 values and cisplatin as the universal drug. The potential candidates underwent cytotoxicity MTT assay test towards the normal cell lines of liver and breast. Analogues of morpholine and coumarin showed potent inhibition of HepG2 and MCF-7 cell lines. This study will offer the possibility of expedient additional modifications with enhanced selective inhibition towards HepG2 and MCF-7 cell lines.

Keywords: Morpholine, Coumarin, Analogues, Anticancer, Antiproliferative, Cytotoxicity, Synthesis

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STRUCTURE ELUCIDATION OF F70HAB16, A NOVEL β-SECRETASE INHIBITOR FROM A MALAYSIAN ENDOPHYTE

S. F. ROSLI[1]*, R. M. J. JAMES[1,2], N. MANSHOOR[1], J-F. F. WEBER ABDULLAH[1]

[1]Faculty of Pharmacy, [2]Brain Degeneration and Therapeutics Group, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia

Email: bellezza_hana@yahoo.com

ABSTRACT

Alzheimer's disease is a brain disease which affects memory, thinking, language, skills and eventually, the ability to carry out the simple tasks. The disease usually begins after age 65 and the risk goes up with age. Currently, it has no cure. Natural products played and are still playing a major role as a source of novel chemical entity in the drug discovery pipeline. B-secretase (also known as BACE-1) is a promising molecular target for the development of anti-Alzheimer drugs according to the amyloid plaques hypothesis. Moreover, endophytes were recently shown to be excellent source BACE-1 inhibitors. A novel compound, F70HAB16 (MW=382 m/z) was isolated from Cytospora rhizophare, a local endophytic strain and was found to inhibit β-secretase. Our studies have shown that this neuroactive compound improved spatial memory and learning in scopolamine-induced and transgenic mice models of AD. Cytospora rhizophorae will be cultured prior to a semi-polar extraction in ethyl acetate. F70HAB16 will be isolated or fractionated by using a preparative C-18 column on a ultra high performance liquid chromatography (UHPLC), performance liquid chromatography (PLC) or recycling HPLC until the targeted compound obtained in a pure form. This compound will then be subjected to LC-MS to confirm its unknown compound that focusing on M+H ion at m/z 383. The unknown compound will be subjected to complete UV, IR, MS and NMR analysis in order to contribute generating a viable structure. Further tests, crystallisation will be achieved by derivatisation of the above halogenated derivatives and/or co-crystallisation followed by X-ray diffractometry. In case the X-ray analysis would not be able to provide an absolute stereochemistry, circular dichroism (CD) combined to quantum mechanics calculations would provide the required information.

Keywords: Endophyte fungi, β-secretase, UHPLC, IR, LC/MS, NMR

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DEVELOPMENT AND VALIDATION OF BIOANALYTICAL METHOD FOR AMLODIPINE IN HUMAN PLASMA WITH OPTIMISED EXTRACTION METHOD USING DESIGN OF EXPERIMENT (DOE)

A. B. M. HELAL UDDIN*, M. ALAAMA, H. J. MOHAMAD, N. S. AMIRUDDIN

Analytical and Bio-Analytical Research Laboratory, Department of Pharmaceutical Chemistry, Faculty of Pharmacy, International Islamic University Malaysia (IIUM), Kuantan, Pahang, Malaysia
Email: abmhelal@iium.edu.my

ABSTRACT

Amlodipine is antihypertensive drug belonging to calcium antagonist group. This study aimed to apply experimental design for the optimization of extraction method of amlodipine from human plasma for higher and constant recovery. A chromatographic method involving High Performance Liquid Chromatography (HPLC) with Diode Array Detector (DAD) was developed and validated for the determination of amlodipine in human plasma with cetirizine as internal standard. Sample preparation for the extraction of amlodipine and cetirizine from plasma was optimised using DOE (Design of Experiment). Three different parameters namely solvent type, solvent volume and pH were monitored at different level for the optimisation of sample preparation that would produce best recovery of the analytes. For this purpose, three-level full factorial design, which involves 27 experiments, was conducted. The recovery of amlodipine was determined using the developed and pre-validated HPLC-DAD method with an Agilent 1100 HPLC system. Chromatographic separation of the analytes was obtained using phenomenix (150 x 4.5 mm, 5 µm) C18 column. The mobile phase consists of 35:65 v/v of ACN: 0.3% triethylamine with pH adjusted at 3 using phosphoric acid. Detection of analyte was conducted using a DAD at 237 nm wavelength. Design Expert 8.0 software was used to interpr*et al.* l data obtained from the optimisation study. For the parameters solvent and solvent volume "acetonitrile" and "1 ml" was found to be the optimum values required to extract amlodipine and internal standard while pH had no effect on the optimum recovery. The recovery of amlodipine and internal standard was 80-88% and 85-99% respectively.

Keywords: Amlodipine, Bioanalytical method, HPLC, Validation, Experimental design

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INSULIN RESISTANCE CROSS LINKS LIPID METABOLISM AND CELL MEDIATED IMMUNE SYSTEM IN GESTATIONAL DIABETES MELLITUS

S. H. ABDUL AZIZ*, C. M. JOHN, F. MOHD FAUZI

Department of Pharmacology-Toxicology, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: anaasrar92@yahoo.com

ABSTRACT

Insulin resistance (IR) coupled with an inadequate insulin response are the pathophysiological mechanisms underlying the development of Gestational Diabetes Mellitus (GDM). The enhanced IR in most GDM cases reported due to the disturbance in glucose metabolism and immune system. The underlying reasons were not established till now. This current study tried to evaluate the role of lipid metabolism along with cell mediated immune system in developing IR in GDM. Experimental GDM animal model was developed in SD rats fed with fatty-sucrose diet and by inducing Streptozotocin (STZ) 35 mg/kg bw, nicotinamide (NA)120 mg/kg bw 15 min prior to STZ on day one of gestation (G1). GDM state was spotted with hyperglycemia along with decrease in serum insulin level and C-peptide level. Animals were sacrificed on Gestational day 21(G21). Insulin resistance was evaluated by measuring resistin, leptin, insulin, and C-peptide from the blood plasma collected on G21, along with evaluating lipid profile and cytokines (TNF-α and IL-6). T cells and B cells and NK cells were measured from the spleen cells. GDM groups showed an increase in glucose, resistin and leptin, where as insulin and C-peptide showed a reduction. Increased levels of triglycerides, total cholesterol, LDL-cholesterol were observed. In addition, levels and tumour necrosis factor-alpha (TNF-α) and IL-6 showed an increase ($p<0.5$) along with T and NK cells. This study demonstrates the insight relationship between lipid metabolism and cell mediated immune system in developing insulin resistance and suggests that these two parameters to be monitored in pregnancy as a marker for GDM.

Keywords: Insulin resistance, Lipid metabolism, Immune system, Gestational diabetes mellitus

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RHOA ATTENUATION BY PEDIOCOCCUS PENTOSACEUS LAB6 AND LACTOBACILLUS PLANTARUM LAB12 IN NEUROBLASTOMA CELLS IS ACCOMPANIED BY AMYLOID-β INHIBITION

R. M. ZAKI[1,2], K. RAMASAMY[1,2], S. M. LIM[1,2]*

[1]Faculty of Pharmacy, [2]Collaborative Drug Discovery Research (CDDR) Group, Pharmaceutical and Life Sciences Community of Research, Universiti Teknologi MARA, Selangor, Malaysia
Email: stvlsm@gmail.com

ABSTRACT

Alzheimer's disease (AD) is the most common form of dementia characterized by extracellular aggregation of amyloid beta (Aβ) plaque. The mechanisms underpinning these hallmarks, however, remain unclear, thus making the design of AD specific therapy very challenging. There is increasing evidence, which supports the role of RhoA activation in pathogenesis of AD. RhoA may serve as a potential therapeutic target for AD as it is involved in the development of AD. Our preliminary screening of MRS broth fermented with Malaysian lactic acid bacteria (LAB; five Lactobacillus spp and eight Pediococcus spp) yielded significant inhibition of RhoA activation in SK-N-SH neuroblastoma cells. The recent emergence of gut-brain axis has linked LAB to neuroprotection against neurodegenerative diseases. The present study was undertaken to assess the correlation between LAB-induced inhibition of RhoA activity and Aβ. For this purpose, four LAB (two Lactobacillus spp and two Pediococcus spp) that exhibited the highest RhoA inhibition during preliminary study were selected. The RhoA inhibitory properties of this LAB in SK-N-SH were first validated using Rhodamine-phalloidine dye and Fluorescence Microscopy. For Aβ study, SK-N-SH was transfected with plasmid carrying amyloid precursor protein (APP) gene. The transfected cells were then treated with LAB at their respective highest subtoxic concentration for 24 h and subjected to evaluation using the Aβ1-42 ELISA kit. The current study found SK-N-SH, which was induced with Rho activator to be presented with abundant stress fibers (indication of RhoA activation). Cells treated with LAB in the presence of Rho activator showed reduced number of stress fibers. Aβ study, on the other hand, indicated Pediococcus pentosaceus (LAB6) as the most potent Aβ inhibitor (30.7%). The present findings indicated that LAB-induced neuroprotection is mediated via attenuation of RhoA activation and Aβ accumulation. This warrants in depth validation using *in vivo* models.

Keywords: Amyloid beta, Lactobacillus, Pediococcus, RhoGTPases, RhoA

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CXCL10 AND CXCR3 MEDIATE NEUROINFLAMMATION AND NEURONAL CELL DEATHS IN ALZHEIMER'S DISEASE

S. M. H. DAYANA[1,2], S. M. LIM[1,2], A-V. CHIN[4], M. P. TAN[4], P. J. H. POI[4], S. B. KAMARUZZAMAN[4], A. B. ABDUL MAJEED[1,3], K. RAMASAMY[1,2]*

[1]Faculty of Pharmacy, [2]Collaborative Drug Discovery Research (CDDR) Group, Pharmaceutical and Life Sciences Community of Research, [3]Brain Degeneration and Therapeutics Group, Pharmaceutical and Life Sciences Community of Research, UiTM, Puncak Alam Campus, Selangor, Malaysia, [4]Division of Geriatric Medicine, Department of Medicine, Faculty of Medicine,University of Malaya, Kuala Lumpur, Malaysia

Email: kalav922@gmail.com

ABSTRACT

There is emerging studies suggesting that cytokines released from chronically-activated microglia could result in neuroinflammation, a condition strongly associated with AD. In our recent clinical study, cytokine profiling of serum samples obtained from 39 AD patients and 39 healthy controls (recruitment at UMMC) had successfully identified CXCL10, a non-classical pro-inflammatory cytokine as potential AD biomarker. CXCL10, also known as IP10, was significantly up-regulated ($p<0.001$) in AD patients with strong MMSE correlation ($r = -0.7844$), high specificity and sensitivity (AUC = 1). The role of CXCL10 in neuroinflammation and its relationship with microglia and neurons, however, remains poorly understood. More recently, our preliminary Gene Globe Analysis of CXCL10 and CXCR3 knockdown in the presence of lipopolysaccharide (LPS) revealed down-regulation of transcription factors (e. g. NFkB, STAT, SP1 and Notch1) which are reported to be involved in neuroinflammation and apoptosis. The present study was thus undertaken to elucidate the roles of CXCL10 and its receptor, CXCR3, in apoptosis under LPS-induced inflammation using *in vitro* cell-based models. In order to mimic neuroinflammation, EOC20 (microglial cells) as well as SKNSH and SHSY5Y (neuroblastoma cells) were induced to over express CXCL10 and CXCR3, respectively. Knockdown of CXCL10 and CXCR3 led to significant reduction ($p<0.001$) of calcium dysregulation in micoglial (-6 fold when compared to LPS control) and neuroblastoma cells (-14 fold when compared to LPS control), respectively. Knockdown of CXCL10 and CXCR3 also resulted in significant reduction ($p<0.05$) of DNA fragmentation (indication of early apoptosis) in EOC20 as well as SKNSH and SHSY5Y (-2 fold when compared to LPS control), respectively. The present findings confirmed that CXCL10, a potential AD biomarker, mediate neuroinflammation as well as neuronal cell deaths via calcium dysregulation and apoptosis.

Keywords: Alzheimer's disease, CXCL10, CXCR3, Calcium dysregulation, Apoptosis.

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DIFFERENTIAL CYTOTOXICITY OF F8268 A3 AGAINST MCF7 AND MDA468 HUMAN BREAST CANCER CELLS

Z. M. ZAKI[1,2], K. RAMASAMY[1,2], V. MANI[1,3], S. M. LIM[1,2]*

[1]Faculty of Pharmacy, [2]Collaborative Drug Discovery Research (CDDR) Group, [3]Brain Degeneration and Therapeutics Group, Pharmaceutical and Life Sciences Community of Research, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: stvlsm@gmail.com

ABSTRACT

Breast cancer has emerged as one of the most prevalent cancer types in the world. Chemotherapy is one of the treatment modalities commonly used for treatment of breast cancer. Its efficacy, however, is often compromised by undesirable side effects and cancer resistance. There is a dire need for discovery of new anti-cancer agents with optimal therapeutic effect and minimal toxicity. Our research group has isolated a novel peptide (patent number = US2011/0201642A1), F8268-A3, from the endophytic fungus, Aspergillus sclerotiorum strain HAB10R12. Given that HAB10R12 extract was previously found to exhibit potent cytotoxicity against MCF7 (IC50 = 0.04 µg/ml), the present study was undertaken to assess the differential cytotoxicity of F8268-A3 against MCF7 (oestrogen receptor-positive) and MDA468 (oestrogen receptor-negative) human breast cancer cells. For this purpose, F8268-A3 (0.01-50 µM) was tested against MCF7 and MDA468 for 24 h. SRB assay was then performed and data generated from the spectrophotometer (520 nm) was used to plot the dose-response curve from which IC50 (concentration that inhibit 50% of the cells) was determined. Subsequently, this study examined the acute toxicity profile of F8268-A3. Briefly, balb/c mice (n=3/group; 8 w old; female) were being administered with F8268-A3 at fixed doses (5, 50, 100 mg/kg). The treated mice were then being observed for signs of mortality over 14 d. Current results indicated that F8268-A3 was relatively more selective against MCF7 (IC50 = 0.58 µM) when compared to MDA468 (IC50 = 2.7 µM). Interestingly, the IC50 of F8268-A3 against MCF7 was found to be 50 fold more potent when compared to conventional cytotoxic agent, paclitaxel (IC50 = 29 µM). On the other hand, acute toxicity study F8268-A3 showed no sign of mortality even at the highest tested concentration. As such, the present findings warrant in-depth mechanistic study of the anti-cancer effect (i.e. anti-angiogenic) of F8268-A3 against breast cancer *in vivo*.

Keywords: Cytotoxicity, Breast cancer cells, Endophytic fungus, Acute toxicity

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PRIMARY BLOOD BRAIN BARRIER CELL CULTURE MODEL: COMPARISON OF HUMAN BRAIN MICROVASCULAR ENDOTHELIAL CELL AND PORCINE BRAIN ENDOTHELIAL CELL

J. SU, A. F. DALI*, M. N. MOHD NAJIB, S. SULAIMAN

Faculty of Pharmacy, Universiti Teknologi MARA, Bertam Campus, Penang, Malaysia
Email: fauzi.dali@ppinang.uitm.edu.my

ABSTRACT

The blood-brain barrier (BBB) remains to be the most challenging obstacle for central nervous system (CNS) drug delivery as this structure restricts more than 98% of small molecules into the brain. This led to challenge the development and the discovery of drugs for CNS treatment disorders. Early prediction of CNS drug delivery at the BBB can be determined by *in vivo*, *in silico* and *in vitro* approaches. Thus, good BBB models are crucial for assessing and screening the permeability of CNS active drugs and new developed drugs to overcome the BBB. The present reviews the *in vitro* approach which is one of the simplest designed methods to be applied and it can provide data at lower cost with high-throughput screening compared to *in silico* and *in vivo*. Two different origins of primary BBB cell culture models such as Human Brain Microvascular Endothelial Cell (HBMEC) and Porcine Brain Endothelial Cell (PBEC) were compared. Both cells performed very well and built the characteristics of the BBB as reported in previous studies. This review focuses on the comparison in terms of the feasibility of these cell lines as BBB models, the tight junction formation measured by Trans endothelial Electrical Resistance (TEER) value, paracellular permeability, distribution of transporter proteins, drug permeability assay, and the need of co-culture with astrocytes and pericytes. In conclusion, both cells were reliable to be a promising cell line for drug permeability studies of the BBB.

Keywords: Blood brain barrier, Cell cuture model, HBMEC, PBEC

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PREFERENCES OF COLOUR, SIZE, SHAPE AND TASTE OF ORAL SOLID DOSAGE FORM IN MALAYSIA

E. USIR *, N. Y. ROCKIE

Faculty of Pharmacy, Department of Pharmacy Practice, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: ezlin365@puncakalam.uitm.edu.my

ABSTRACT

Oral solid dosage forms consist of tablets, capsule, soft gel and caplet. The objectives of this study were to determine the 1) most preferred oral solid dosage form, 2) perceptions towards therapeutic benefit of medicine in a relation of colour, size, shape and taste of oral solid dosage form and 3) factors associated with preferences of oral solid dosage form. The instrument used was a self-administered 36-items questionnaire and distributed to 130 individuals at four retail pharmacies in Kuala Terengganu. Data was analyzed using SPSS version 20.0. The results showed that 52.3% respondents preferred tablet over any other solid dosage forms followed by capsule (26.2%), soft gel (13.8%) and caplet (7.7%). About a third of the respondents (36.9%) reported that tablet is a very "easy to swallow" dosage form, followed by soft gel (28.5%), capsule (27.7%) and caplet (4.6%). Statistically significant differences in respondents' perception found towards gender, age, monthly income, marital status and occupation of the respondents. In conclusion, tablet is the most preferred oral solid dosage and a form very easy form to swallow. Majority of respondents preferred oral solid dosage form to be white in color, round in shape, small in size and without any taste. The most important physical characteristics of oral solid dosage form were size, followed by taste, color and shape.

Keywords: Preference, Colour, Size, Shape, Taste, Oral solid dosage form

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THE UNREGISTERED DRUGS IN MALAYSIA MARKET: PHARMACIST PERCEPTIVES

N. W. ZULKIFLI[1]*, N. ABD. AZIZ[1], Y. HASSAN[1], M. A. HASSALI[2], N. L. Z. BAHRIN[1]

[1]Faculty of Pharmacy, Department of Pharmacy Practice, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Faculty of Pharmacy, School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang, Malaysia
Email: nurwah dazulkifli@gmail.com

ABSTRACT

Unregistered drug is one of the worldwide phenomena and Malaysia is not spared. The problem becomes more sophisticated and complicated when the technology becomes more advance. There are limited studies to get overview of the current situation. The objective is to explore pharmacist perceptive towards unregistered drugs existence in Malaysian markets. Sixteen pharmacists were interviewed in depth using semi-guided interviewed. The interviews were tape-recorded and analyzed qualitatively using constant comparative method. Most of the pharmacists believed that there is an abundance of unregistered drugs in the Malaysia market. They believed that the abundance of unregistered drugs is influenced by the influx of immigrants that come to Malaysia, such as from Indonesia. Moreover, the obstacles faced in eliminating unregistered drugs by enforcement are the lack of awareness and knowledge among public citizens on drug registration and self-medication. These two themes are related because if a person is not aware and not well-educated about registered drugs, he/she would buy unregistered drugs and practice self-medication. Majority of the pharmacist believed that, the abundance of unregistered drugs in the market does not only give adverse health impacts to the consumers itself, but also to the country because the unregistered drugs might affect local pharmaceutical companies' businesses. From the findings, it can be concluded that in order to eliminate unregistered drug, we need to manage consumers, manufacturers and technology. These three factors have to be managed from every aspect. Additionally, to make this plan successful, every organization related to this matter has to cooperate with one another in one joint force.

Keywords: Unregistered drugs, Market, Pharmacist, Perceptive

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PERCEPTION OF HEALTH-CARE PROFESSIONALS REGARDING THE ROLE OF PHARMACISTS IN CANCER CARE

A. SUNDUS*, N. E. ISMAIL, S. GNANASAN

Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: ahlam_sundus@hotmail.com

ABSTRACT

Anti-cancer medications are ranked 2nd in causing most fatal medication errors and have the highest incidence of drug related problems. Oncology pharmacists could potentially assist by detecting errors in prescribing, reducing poly-pharmacy and individualizing the dose. The objective was to explore the perception of healthcare professionals on the role of clinical pharmacist for ensuring safe and effective use of cancer medications. Qualitative methods were employed in this study. Semi-structured interviews were conducted with 9 doctors, 7 nurses and 4 pharmacists from Hospital Kuala Lumpur and Hospital Selayang. Interviews were audio-recorded, transcribed verbatim and were analysed using thematic analysis and constant comparison method. Ethical approval was sought from Malaysian Medical Research & Ethics Committee. All of the healthcare professionals interviewed had a strong consensus that pharmacists possess optimum knowledge and skills for the provision of patient-oriented cancer care. Data obtained from doctors suggested that pharmacists can assist them by screening chemotherapy regimens for correct doses and managing the sequence of treatment protocol. They can also help in reducing drug toxicities by predicting drug-drug interactions, by modifying doses according to renal and hepatic function and by informing rare but possible side effects. According to doctors pharmacists act as their back-up in designing safe and effective treatment plan. Nursing staff expressed a high degree of convenience in working with pharmacists and they designated pharmacists as the source of accurate drug information for them. Pharmacists verified that they do posses optimum skills to screen chemotherapy prescriptions for inappropriate regimens and that they often intervene to ensure patient safety and treatment efficacy. Integration of pharmacists into cancer care team is vital to facilitate pharmaceutical care and improve treatment outcomes.

Keywords: Health-care professionals, Role of pharmacists, Cancer care

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UiTM PHARMACY STUDENTS–WHAT THEY KNOW ABOUT ORAL CARE

N. A. ABDUL RAHIM, A. MOHD SUHAIMI*

Faculty of Pharmacy, Pharmacy Practice Department, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: drazyyati@puncakalam.uitm.edu.my

ABSTRACT

The significance of oral health has long been recognised to be an important and essential part of overall body health. Good self-care of oral health among students would improve their daily performance and personal hygiene, hence potentially alleviating the likelihood of getting oral diseases which is strongly associated with the increment in oral health knowledge. The main objective of this study is to identify the consumer behaviour of students on purchasing of oral care products and its association with their knowledge in oral health. For this study, quantitative research analysis was adopted using simple random sampling technique. A survey was conducted and 186 participants responded to the questionnaire which consisted of general background information, oral health behaviour and oral health knowledge. It was found that majority of pharmacy students in UiTM Puncak Alam were generally concerned about the significance of oral health care and selecting appropriate oral care products of high quality to maintain their general well-being. It could be foreseen that this study might assist pharmacists in the community pharmacy to provide better services in relation to oral care products for the young consumers.

Keywords: Pharmacy, Oral health, Dental care

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PERCEPTION OF DIABETES AMONG PATIENTS AND THEIR UNDERSTANDING ON INSULIN USAGE

A. R. AIMAN ROSLI, Y. GOPALAN*

Department of Pharmacy Practice, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: yoghes@gmail.com

ABSTRACT

Diabetes is a metabolic disorder which affects population all over the world with its prevalence increasing yearly. Insulin was introduced as one of the treatment for diabetes. However, the effectiveness of the insulin has been compromised due to lack of knowledge and proper education on insulin usage. Hence, this study was conducted in order to gauge the perception on diabetes and knowledge on insulin among diabetic patients. A questionnaire-based survey was done on randomly selected 100 diabetic patients on insulin therapy. Subjects were recruited from the Malaysian Diabetic Society, Petaling Jaya and community pharmacies in Selangor, Kelantan and Terengganu. Insulin Treatment Satisfaction Questionnaire (ITSQ) and Patients Diabetes Knowledge Questionnaire were used to identify perception and knowledge on diabetes and insulin. Majority of the subjects were 40 y and above (90%) with normal BMI (53%) and 77% of them had family history of diabetes. From 100 respondents, it was only 27% of them who had the proper education on insulin usage. 59% of the insulin users claimed that they had low satisfaction on blood glucose control by using the insulin. Meanwhile from the perception aspect, about 58% admit that the insulin treatment causes slight problem or inconvenience to them. About 97% respondents claimed that they had experienced hypoglycemic (low blood glucose level) signs and symptoms during the treatment. Significant association were found with proper diabetic education counselling with knowledge on insulin type and insulin storage ($p<0.05$). With the low level of exposure related to diabetes management, it is recommended that concerted effort focused on awareness and educating the patients reinforced in order to enhance the knowledge and understanding to achieve better diabetes management and improve their quality of life.

Keywords: Diabetes mellitus, Insulin treatment satisfaction questionnaire, Patients diabetes knowledge questionnaire

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STUDY OF AMLODIPINE INDUCED BLURRED VISION IN MALAYSIA

H. MAT NOJI[1]*, W. Z. W. A. MANAN[1], R. M. ZIN[1], N. ABD. AZIZ[2]

[1]Department of Pharmacy, Ministry of Health, Malaysia, [2]Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: scarletpills84@gmail.com

ABSTRACT

In 2014, NPCB was alerted by WHO collaboration center on the possibilities of occurrence of blurred vision induced by the consumption of Amlodipine. This study was to determine possible associations between amlodipine and blurred vision. This is a descriptive retrospective study whereby all ADR data related to amlodipine, from 1st January 2002–31st December 2014, were extracted from national pharmacovigilance Quest 2 database. A total of 24 cases have been reported over the last 12 y. More than half of the reports involved female patients (64.5%) and 32.3% for males, however one report did not specify the patient gender. Highest report involved malay patients (45.2%), followed by others (29.0%) and Chinese (25.8%). Patient age ranged from 30–83 y old with mean age 50.3 y old. Majority (67.7%) of patients fell in the age range of 45-64 y. The extent of reaction for majority of the reported cases were classified as moderate (n=15; 48.4%), followed by mild with 7 cases (22.6%) and severe with 6 cases (19.4%). There were 3 reports (9.7%) that did not mention the extent of reaction. Significant number of blurred vision cases reported with the use of amlodipine which warrant further study and investigations.

Keywords: Amlodipine, Blurred vision, Adverse drug reactions, ADR

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SELF-MEDICATION PRACTICE AMONG CONSUMERS IN SANA'A CITY

S. M. ALBAWANI*, Y. HASSAN, N. ABD AZIZ, S. GNANASAN

Faculty of Pharmacy, Department of Pharmacy Practice, Universiti Teknologi MARA, Malaysia
Email: samibwani@gmail.com

ABSTRACT

Self-medication is a common practice worldwide. However, drug abuse can lead to serious side effects, toxicity, drug interaction, antibiotics resistance or could delay accurate diagnosis and treatment. In Yemen, many drugs are dispensed without medical supervision. Regardless of the intensive research spent on self-medication, little information has been available about its major determent. In Yemen, very few studies about self-medication were found in literature. The purpose of this study was to determine the prevalence of self-medication practice and its associated risk factors in Sana'a City. A cross-sectional study was carried out using a validated self-administered questionnaire distributed to consumers attending community pharmacies in Sana'a City. Chi-square test and multiple logistic regressions were used in this study. A total of 400 consumers (57.8% males and 42.2% females) were involved in this study. The majorities were married (62.8%), non-smokers (77.3%), Qat chewers (72.5%), with low income (74.2%), without medical insurance (85.8%), and have access to medical centres (83.2%). The prevalence of self-medication in the past six months was 90.7 % (94.1% of males and 88.3% of females). Drug commonly used were analgesics (90.1%), antimicrobials (87.1%), antipyretics (64.2%), antacids (51.2%), vitamins and minerals (50.7%) and common cold (72.2%) medications. Common reasons for self-medication were the high cost of doctor consultation (92.3%), case was not serious (65.6%) and being easily managed (43.5%). Common source of information were found to be the community drug dispensers (56.2%), family members (28.7%) and media (16.3%). In multivariate analysis, perception about death due to self-medication and chewing Qat were significant predictors of self-medication practice. Those who did not chew Qat were more likely to use self-medication compared to those who did chew Qat (OR= 2.4, CI 95% 1.4-4.1, p= 0.02). Those who did not believe that self-medication leads to death are more likely to use self-medication compared to those who believe that self-medication may lead to death (OR=13.9, CI 95% 8.2-23.5, p= 0.001). Self-medication practice was high among consumers in Sana'a City and chewing Qat and perception about death due to such practice was significantly associated with self-medication.

Keywords: Self-medication, Community pharmacies, Sana'a City

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ASSESSING THE IMPACT OF MEDICATION ADHERENCE THERAPEUTIC CLINIC ON KNOWLEDGE OF ASTHMA IN ADULT ASTHMA PATIENTS

S. AHMAD[1], A. I. ISMAIL[2], M. A. MOHD ZIM[2], W. AKRAM[1], N. S. A. BANGASH[1], S. A. ABBOUD[1], N. E. ISMAIL[1]*

[1]Faculty of Pharmacy, Clinical BioPharmaceutics Research Group (CBRG), Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Faculty of Medicine, Respiratory Unit, Universiti Teknologi MARA, Selayang Campus, Selangor, Malaysia
Email: elkudssiah77@yahoo.com

ABSTRACT

The knowledge of asthma patients is very imperative to the optimal management of asthma. Medication Therapeutic Adherence Clinics (MTAC) are introduced in Malaysian healthcare settings in order to improve the self-management of asthma patients. The objective of this study was to determine the impact of MTAC on knowledge of adult asthma patients. In this cross-sectional study, 55 adult asthma patients were recruited from Hospital Selayang, Selangor, Malaysia. The level of knowledge of the adult asthma patients was determined by the self-administered questionnaire. The questionnaire was developed from different publications and permission to adopt and translate the questionnaire was obtained from the respective authors. The self-designed questionnaire was translated into Malay language using international translation guidelines. The questionnaire was administered first before the patient's visit to the MTAC, and secondly on their next scheduled visit to the patients' respiratory clinic. Patients' responses were recorded on dichotomous scale of true and false. One mark was given to the correct answer to the statement. Statistical Package for Social Science (SPSS®) version 21 was used for paired t-test. The enrolled asthma patients showed moderate level of knowledge at pre and post visit to MTAC. The findings of paired t-test showed that there is a significant improvement in the mean score of asthma patients before their visit to MTAC in the study hospitals i.e. 18.65,±2.34 to the next scheduled visit to the study settings i.e. 19.65,±2.09; t =-4.949 (54), p<0.001). The asthma patients showed significant improvement in their knowledge of asthma on second administration of the questionnaire. The findings of the study suggested that the MTACs are not only helpful for improving the medication adherence but the level of knowledge of asthma also in the adult asthma patients. Such improvements consequently lead to the better asthma control and management in asthma patients.

Keywords: Asthma, Medication Therapeutic Adherence Clinic

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REDUCING DOSING ERROR IN PAEDIATRIC PATIENTS: AN INTERVENTION THROUGH THIS (PAEDIATRIC POP UP DOSE)

L. Y. CHONG*, P. M. WONG, R. N. ZAINOL ABIDIN

Pharmacy Department, Serdang Hospital, Malaysia
Email: leann6198@gmail.com

ABSTRACT

Children are vulnerable to medications error. There is an increased need for calculations, dilutions and manipulations of paediatric medicines. In Serdang hospital itself, 38% of errors found in year 2013 were contributed by paediatrics ward. Out of it, 50% were dosing errors. Serdang Hospital is a fully Information Technology (IT) based system where the prescribing and supplying of medications are done through Total Hospital Information System (THIS). Thus this study aims to reduce dosing error in paediatric patients by implementing an intervention through THIS. This is a prospective interventional study where all prescriptions received from general paediatric wards during office h in June 2014 were screened for dosing errors and classified according to pharmacological group. This is followed by intervention phase in July to October 2014 where incorporation of Paediatric Pop up Dose (dose-assisted prescribing) into THIS was done. The post intervention data was then collected in November 2014. Rate of dosing errors before and after dosing-assisted system implementation were calculated and significance level was calculated using Chi-Square. The number of dosing error decreased from 59/4345 (1.36%) to 23/3721 (0.6%) following intervention. (p value=0.01). In general, antibiotic prescription has the most errors followed by antipyretic (paracetamol). Paediatric Pop up Dose reduces paediatric dosing error from 1.36% to 0.6%. The process of medication delivery involves many stages and personnel. To ensure zero medication error, interventions should be comprehensive in view that error causes are multifactorial.

Keywords: Dosing error, Paediatric, Intervention, Electronic

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MANAGEMENT OF DIABETES MELLITUS IN TUBERCULOSIS PATIENTS IN MALAYSIA: A RETROSPECTIVE STUDY

A. A. IBRAHIM, S. GNANASAN*

Department of Clinical Pharmacy, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Malaysia
Email: shubashini.gnanasan@gmail.com

ABSTRACT

The prevalence of diabetes mellitus (DM) and tuberculosis (TB) as well as DM in TB cases keeps increasing every year in most of the countries around the world. Many studies have been reported to explore the association between DM and TB. Due to this convergence, the World Health Organization (WHO) and the Union have initiated a guideline called Collaborative Framework for Care and Control of Tuberculosis and Diabetes to guide and educate the healthcare practitioners in the management of TB-DM patients. The aims of this study were to determine the prevalence of DM in TB patients, compare the characteristics and differences between TB and TB-DM patients, and investigate the current management of DM in TB patients. The study was conducted at Institut Perubatan Respiratori (IPR) in Kuala Lumpur. 100 medical records from January 2014 to June 2014 were reviewed. The data was recorded in data collection form and the prevalence of DM in TB was quantified. The prevalence of DM in TB patients was 18%. The number of TB-DM patients was found to be almost equal for both male and female at 53% and 47% respectively. The mean age of TB-DM patients was 55.5 y which is significantly higher compared to TB only patients with mean age of 39.5. Almost half of the TB patients were Malay. Most of them were married and employed. The percentage of TB-DM patients having co-morbid condition was higher (61.1%) as compared to TB only patients with only 23.3%. In addition, there was lack of regular blood glucose monitoring at the TB clinic whereby 43% were not screened for random or fasting blood glucose and insulin was underutilized in the management of DM in TB patients.

Keywords: Tuberculosis, Diabetes Mellitus, Malaysia

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GRADUATES' PERCEPTION ON THE POSTGRADUATE COURSEWORK PROGRAMS IN UNIVERSITI TEKNOLOGI MARA

F. I. ABDUL JALIL*, M. KARUPPANNAN

Department of Pharmacy Practice, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: farahizzah3993@gmail.com

ABSTRACT

Pharmacy Practice and Clinical Pharmacy (by coursework) are new programs in UiTM. These two programs were introduced in 2009 and were offered for pharmacists. Being new, the programs were never reviewed and evaluated for improvement. This study aimed to identify the perceptions of graduates on these two programs. This was a quantitative survey study involving 70 postgraduate students who have graduated from both programs. The questionnaire was distributed using Google form. The data was analysed using SPSS version 22. 70 completed questionnaires were received. Majority of the graduates (n= 34, 49%) were within the age range of 31-35 y. 79% of the respondents (n=55) were graduates of pharmacy practice program. The main reason for taking up a masters degree program was for personal development (n=62) and the reasons for choosing UiTM was because of the location (57%) and reasonable and affordable fees (43%). The respondents agreed that the program had increased their confidence level (n=70,100%), had great impact on their personal development (n=69, 99%) and improved their skills (n=67, 96%). Respondents also agreed that the program helped to prepare them to further their studies (n=63, 93%) in which 74% (n=52) of respondents reported planning to further studies to PhD. Many agreed that the duration of the program (3 semester) was appropriate (n=67, 96%) however a small number claimed that the sequencing of the courses in the program was inappropriate (n=8, 11%) and the classroom facilities were inadequate (n=3, 4%). In addition, 63% of the respondents (n=44) claimed that they will definitely recommend these programs to others. The graduates from both programs have good perception on these programs however some improvisations are still needed. Nevertheless, these programs are achieving their main goal that is to produce graduates with improved self-skills.

Keywords: Postgraduate, Pharmacy graduates, Perception

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KNOWLEDGE CONCERNING AEDES MOSQUITO, PREVENTIVE MEASURES OF DENGUE, AMONG GENERAL POPULATION IN THE STATE OF PENANG, MALAYSIA

K. ALAM[1]*, S. A. SYED SULAIMAN[2], A. F. DALI[1]

[1]Faculty of Pharmacy, Universiti Teknologi MARA, Bertam Campus, Penang, [2]Department of Clinical Pharmacy, School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang
Email: muhdkhurshid79@gmail.com

ABSTRACT

This study aimed to evaluate the basic knowledge of dengue, and its prevention among the general population. A cross-sectional study was carried out to evaluate the knowledge about dengue fever among the general population of Penang. Knowledge was assessed through self-administered questionnaire regarding Aedes mosquito, breeding sites, spreading source and preventive measures. One Way ANOVA was used to assess the association and differences of knowledge concern to dengue among different age groups and on employment level. Over all 862 of the respondents participated. Most of the respondents (78.8%) had knowledge that Aedes mosquito breed in water. Majority of the people (68.8%) knew that Aedes mosquito bites during day time. In this study 53.4 % of the respondents incorrectly responded that dengue is a contagious disease. Knowledge about the preventive measures was found to be highly significant in the age group 25-34 y old with mean score 7.01 (95% CI: 6.77–7.25), while lower knowledge found in the teenagers 6.44 (95% CI: 6.12–6.67) and older age groups 6.33 (95% CI: 5.68-6.98). Government employees revealed to have sufficient knowledge with mean score of 5.13 (95% CI: 4.84-5.42), 5.08 (95% CI: 4.93-5.23), 7.44 (95% CI: 7.14-7.73) on breeding sites, spreading source and preventive measures of dengue respectively. In this study it was found that teenagers, unemployed and older age group need more attention to encourage them to participate in dengue awareness campaigns.

Keywords: Dengue, Knowledge, Breeding sites, Preventive measures, Malaysia

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PERCEIVED PRACTICE AND BARRIERS TOWARDS ASTHMA MANAGEMENT AMONG URBAN COMMUNITY PHARMACISTS OF LAHORE, PAKISTAN

M. A. NAWAB KHAN[1]*, W. AKRAM[1], S. AHMAD[1], N. S. ALI BANGASH[1], M. JAMSHAID[2], N. E. ISMAIL[1]

[1]Clinical BioPharmaceutics Research Group (CBRG), Inhalational Delivery Research Unit and Biomedical Analysis Lab (BAL), Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Faculty of Pharmacy, University of Central Punjab, Khyaban-e-Jinnah, Lahore, Pakistan
Email: anwarrkhan2003@yahoo.com

ABSTRACT

The aim of asthma management is to gain and maintain control of disease. Pharmacists could assist asthma patients to achieve and maintain asthma control by providing suitable information and training about asthma medication and facilitating adherence to controller medication. Previous studies have explored the pharmacists' perception of their role in asthma management in different parts of the world. This study aimed to assess the perceived practice and barriers toward the provision of asthma management services among urban community pharmacists of Lahore, Pakistan. One hundred fifty (170) urban community pharmacists in Lahore, Pakistan, were randomly selected and recruited for this baseline study. Self-designed questionnaires (47 items) were directly distributed and collected by the investigator himself. The extracted data from the completed questionnaires were analysed descriptively and inferentially using Statistical Package for Social Science (SPSS) version 19. Pharmacists perceived their roles in asthma management along three major dimensions: -patient self-management, medication use and asthma control. The mean average score of the perception towards asthma management was 79.29±3.21 over a maximum possible score of 110 which was thought to be good perceived practice. There was no significant difference in the perceived practice among age, gender, pharmacy education level, practicing year and type of pharmacy. Significant difference in the perceived practice was observed among different working hs. Pharmacists identified some of the most common barriers towards asthma counselling including lack of time, lack of asthma knowledge, lack of counselling space and cost of asthma drugs. Overall urban community pharmacists of Lahore, Pakistan had good perceived practice of better management of asthma. The main contribution of this research is in understanding the perceptions of pharmacists' role in asthma management. Participants also highlighted both pharmacists and patient related barriers towards the asthma counselling.

Keywords: Perceived practice, Barriers, Community pharmacists, Asthma

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PERCEPTION ON QUALITY OF LIFE (QoL) AMONG GOVERNMENT SERVANTS AND FACTORS AFFECTING IT

W. Z. W SAZRINA*, Z. B. N ZUHAIDA, A. R. SURAIYA, Z. FAUZIAH

Faculty of Pharmacy, Department of Pharmaceutical Technology and Industry, Cyberjaya University College of Medical Sciences, Selangor, Malaysia
Email: wansazrina@cybermed.edu.my

ABSTRACT

Quality of life is one of the important aspects that may affect someone's daily life. There are many factors that may affect an individual's quality of life and may change their perception regarding this matter. Therefore, this study was done to know the perception on quality of life and factors affecting it on government servants. A quantitative cross sectional study was conducted in Selangor and Putrajaya from 20 April 2014 until 25 August 2014. Questionnaires were distributed to government servants around Selangor and Putrajaya. A total of 311 respondents were recruited through convenient sampling. The results revealed that respondents perceived good quality of life on most of the components except for component of life enjoyment. The mean value for component of overall quality of life show significant difference between male and female ($p= 0.03$). There was a negative correlation between age and quality of life components of life enjoyment ($p= 0.004$, $r=-0.163$). In addition, the mean values for physical state, life enjoyment and overall quality of life showed significant difference between the groups of marital status ($p= 0.023$, $p<0.05$, $p=0.011$). Mental/emotional state showed significant difference on the mean values for demographic factors of education qualification ($p= 0.017$) and job designation ($p= 0.019$). This study has found out that there were certain factors that will influence employee's quality of life. As a conclusion, the results of this study have led to the need of the future research focusing on the other factors influencing one's quality of life.

Keywords: Quality of life, Government servant

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A MANAGERIAL QUALITY STUDY TOWARDS EMPOWERING MALAYSIA PHARMACEUTICAL FISCAL SUSTAINABILITY

N. L. ZAINAL BAHRIN*, Y. HASSAN, A. B. ABD MAJEED, N. W. ZULKIFLI, A. AHMAD

Department of Pharmacy Practice, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Malaysia
Email: liyanazb@gmail.com

ABSTRACT

Emphasise on the proper management of monetary is important as the operational of the healthcare system depends on the available fund. In Malaysia, operation of the public healthcare system is through a fully subsidized mode, thus proper management on the procurement and pharmaceutical inventory are crucial. This is because as the Malaysian population grows, the pharmaceutical expenditures increases, hence the total financial allocation for MOH will also expand yearly. This study assessed and reviewed the current procedures in the procurement of pharmaceuticals in Malaysia and its correlation towards the rotation stock control. This is a cross-sectional study involving 13 major public hospitals in Malaysia. A quantification and statistical analysis of the current pharmaceutical procurement and the stock rotation were performed for the outlined results. SPSS and Microsoft Excel were used to analyze the data statistically. Spearman correlation test were used for the correlation analysis. The outcome of the study managed to identify the correlating factors that determine the stock rotation rates towards the expenditures spend. The present findings in the component of medicine stock rotation rate indicated that it did not have a strong correlation with the hospitals medicine financial allocation. The p value observed for the rotation rates towards the expenditure spend was 0.745 for year 2012 and 0.506 for year 2013 respectively. As this particular criterion has been classified as fundamental characteristics towards a powerful force of pharmaceutical industry, the guidelines of controlling medicines stock cycle should be adhered. The majority of the hospitals' medicines stock cycle lies in the range of 7-10. The outcomes obtained have fulfilled the research gap in the managerial quality towards the financial sustainability of the pharmaceutical services in Malaysian public healthcare institutions. Further evaluation should be conducted on the individual healthcare institution.

Keywords: Pharmaceutical, Stock rotation, Malaysia, Managerial quality, Fiscal sustainability

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DETERMINATION OF APPOINTMENT ADHERENCE RATE OF PHARMACY DISPENSING VALUE-ADDED SERVICES

A. AHMAD*, Y. HASSAN, N. ABDUL AZIZ, N. L. ZAINAL BAHRIN, N. W. ZULKIFLI

Department of Pharmacy Practice, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: 25azlan@gmail.com

ABSTRACT

Malaysian public healthcare is fully funded by government and medicines are supplied in a month duration basis. For those patients who require a long life treatment management with repeated prescription like cardiovascular patients, Pharmaceutical Service Division (PSD), Malaysia implemented a new service called Value-Added Services consist of Phone & Take, SMS & Take, Drive-Through Pharmacy and Pharmacy Home Delivery System. This innovation has been taken up as a new Key Performance Indicator from year 2014. This study reviewed the enrollment rate of patient registered with Pharmacy Appointment Medication Dispensing of Value-added Services. The retrospective data from PSD in year 2013 was analysed. A quantitative study approach was adopted by focusing on facilities that implemented all services of Phone & Take, SMS & Take, Drive-Through Pharmacy and Pharmacy Home Delivery System. As being quantified, the percentage of patients enrolled with these services was 24.06 in year 2013. This value was indicated as there were 338,459 patients with repeated prescriptions who used these services in the 1,407,018 total of patients who received treatment nationwide. From National Health and Morbidity Survey (NHMS) 2011, 2.6 millions of diabetic patient, 5.8 millions of hypertension patient and 6.2 millions of hypercholesterolemia patient were estimated having this kind of illnesses. From this statistic, 80% of these patients will come to the public hospitals or health clinics for the treatment. The finding of the study has highlighted that there is only 24.06 % of patients enrolled for the Value Added of Pharmacy Services in the year 2013. Thus, the finding of this research will affirm more indicators and factors to be studied in the future study as to justify the competencies of the Value Added of Pharmacy Services offered.

Keywords: Medication dispensing, Adherence rate, Value-added, Pharmacy

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MORISKY MEDICATION ADHERENCE SCALE AMONG ADULT DIABETES PATIENTS IN HOSPITAL KUALA LUMPUR (HKL)

S. A. ABBOUD[1], N. E. ISMAIL[1]*, N. S. A. BANGASH[2], S. AHMAD[1], M. B. L. BIDIN[2]

[1]Clinical BioPharmaceutics Research Group (CBRG), Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Hospital Kuala Lumpur, Endocrine Unit, Medical Department, Kuala Lumpur, Malaysia
Email: elkudssiah77@yahoo.com

ABSTRACT

Diabetes mellitus (DM) causes dangerous complications if not managed properly. Adherence is a common daily and complex challenge in patients living with DM. Medication non-adherence is a growing concern to both healthcare professionals. The aim of this study was to assess the medication adherence in DM patients using Morisky Medication Adherence Scale (MMAS-8). Permission was obtained from respective authors to use the Malay MMAS-8. This cross-sectional study recruited 62 diabetes patients aged =18 y old, either having type 1 DM or type 2 DM, who attended Endocrine Clinic at Hospital Kuala Lumpur. Majority of the patient have type 2 DM (67.7%). The mean age was 35.89 (SD =±11.81, range: 19–57) and majority of them were females (60%). The mean value of HbA1c was 8.89 (SD =±2.34, range: 6–13). Only 30% of the enrolled patients had good glycaemic control. The majority of the patients (65%) had poor adherence to their medication regimen, 20% had medium adherence and only 15% had high adherence. In type 2 DM, the mean value of HbA1c was 9.48 (SD =±2.09, range: 6–16). There were only 21.4% have good glycaemic control whereas 50% of patients had poor adherence to their medication regimens, 38.1% had medium and (11.9%) had high adherence. In both type 1 and type 2 DM, majority of the enrolled patients had poor medication adherence.

Keywords: Adherence, MMAS-8

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SURVIVAL IN ADVANCED NON SMALL CELL LUNG CANCER AT HOSPITAL KUALA LUMPUR, MALAYSIA

N. S. A. BANGASH[1], S. A. ABBOUD[1], S. AHMAD[1], N. E. ISMAIL[1]*, N. M. HASHIM[2]

[1]Clinical Bio Pharmaceutics Research Group (CBRG), Department of Pharmacy Practice, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Department of Oncology and Radiotherapy, Hospital Kuala Lumpur, Malaysia
Email: nahlah@puncakalam.uitm.edu.my

ABSTRACT

The impact of chemotherapy on survival in patients with advanced non-small cell lung cancer (NSCLC) was investigated. A review of medical records of the patient with NSCLC hospitalized in year 2009 to 2012 at Hospital Kuala Lumpur was carried out. The socio-demographic and medical data of the patients was abstracted including the type of chemotherapy received, date of diagnosis, date of first and last follow-up and date of death. Statistical analysis was conducted using Chi-square for categorical data. For survival estimation, Kaplan Meier was used in univariate analysis. For comparison between groups, log rank was used. In multivariate analysis Cox regression was used. Of a total 163 lung cancer patients, 65% were at stage IV, 24% were at stage IIIB and 11% were at stage IIIA. The median survival time was 377 d for platinum chemotherapy and 409 d for non-platinum chemotherapy. With the non-platinum based chemotherapy, the risk of death increased as the cancer stage progressed 0.20 times at stage IIIA, 0.61 times at stage IIIB and 0.74 times at stage IV. There is no chemotherapy regimen to be considered as the treatment of choice at advanced lung cancer, probably due to comparable prognosis at locally advanced and metastatic disease.

Keywords: Advanced lung cancer, Chemotherapy treatment, Survival

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UTILIZATION OF CHEMOTHERAPY REGIMEN FOR ACUTE MYELOID LEUKAEMIA (AML) PATIENTS IN AMPANG HOSPITAL

J. JAMARI[1]*, M. M. MANAN[1], W. S. PING[2]

[1]Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, [2]Pharmacy Department, Ampang Hospital, Selangor, Malaysia

Email: jamiecrabie85@yahoo.com

ABSTRACT

Acute Myeloid Leukaemia (AML) is one of the most common and acute diseases among different types of leukaemia. Due to increased number of AML cases and high cost of treatments, drug utilization evaluation of AML is important for healthcare allocation. This is a retrospective drug utilization study of chemotherapy regimen for AML patients in Ampang Hospital. The objective of the study is to describe the utilization of chemotherapy regimen drugs for AML patients. The specific objectives are to describe patients characteristic, types of chemotherapy regimen drugs used, side effect of chemotherapy used, examine the complete remission (CR) rate and overall survival for AML patients. A total of 133 records were retrieved with the median age of 41 y old ranged from 13 to 74 y old. The most commonly prescribed induction regimen was DA regimen. The most commonly used consolidation chemotherapy was HIDAC regimen and for salvage chemotherapy was FLAG-Ida regimen. Of the 108 patients achieved CR, 39 patients relapsed at a median of 9 mo, 22 patients died in CR at a median of 5.5 mo and 47 patients remain alive in CR1 with a median of 34 mo. Only transplant had a significant effect to the CR ($P<0.05$) and increased odds of CR by 2.8 times. Total of 83 AML patients had died during the study period (62.4%). AML patients who achieved CR have decreased odds of mortality by 66% than AML patients who are not achieved CR. The median OS was 20 mo (95% CI: 15.4 to 24.6 mo). From this study, all chemotherapy regimens in either induction phase, consolidation phase or salvage phase have been used in accordance with their appropriate indications and follow the international guideline. These results should increase understanding of chemotherapy utilization pattern in making evidence based decision among physician and pharmacist.

Keywords: Drug utilization, Acute Myeloid Leukaemia

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UTILIZATION OF NON-STEROIDAL ANTI-INFLAMMATORY DRUGS IN ORTHOPAEDIC WARD AT AMPANG HOSPITAL

N. A. SALAMAT*, M. M. MANAN

Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor
Email: aini241086@gmail.com

ABSTRACT

Non-steroidal Anti-Inflammatory Drugs (NSAIDs) are among the group of drugs which contribute to most-reported cases of adverse drug reactions and their safety profiles always become a concern. Despite its therapeutic effects, many studies have shown that chronic NSAIDs use heightens the risk of gastrointestinal complications, acute renal failure, and cardiovascular event. This is a retrospective drug utilization study of NSAIDs available at Ampang Hospital. The objective of the study is to describe the utilization pattern of NSAIDs in orthopaedic ward at Ampang Hospital. The specific objectives for this study are to assess the pattern of NSAIDs prescribed, to estimate Defined Daily Dose (DDD) for each NSAID prescribed and to identify potential NSAIDs prescription related problem. A total of 210 patient records were retrieved from eHIS system in Ampang Hospital with the median age of 31 y old. Based on the DDD's calculated, the most prescribed NSAIDs were diclofenac tablet (80.46), diclofenac injection (19.25), meloxicam (1.25) and ibuprofen (0.35). Majority of patients received NSAIDs for 4 to 7 d. Types of injury with p-value of 0.007 showed that it was statistically associated with types of analgesic prescribed during study period. Among the NSAIDs user, 8.4% had chronic conditions such as hypertension, diabetes mellitus and cardiovascular disease. All potential drug-drug interactions with NSAIDs were involving diuretics and ACE Inhibitors. Within one year after the study period, none of the patient has been admitted due to gastrointestinal complications but there were 3.2% patients being started on gastro protective agent. Nevertheless, the gastrointestinal complications were not dependent on age group, gender and ethnicity factor (p=0.606, p=0.269, p=0.163) respectively. These results should raise awareness to prescribers and pharmacist of such risks and work collaboratively to encourage safe prescribing and use of NSAIDs. Pharmacist also plays a key role in the overall operation of the drug utilization program to provide a more comprehensive understanding of NSAIDs utilization pattern.

Keywords: Drug utilization, Non-steroidal anti-inflammatory drugs, Orthopaedic, Define daily dose, Usage pattern

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QUALITY OF LIFE OF CONTRACEPTIVE USERS AT URBAN COMMUNITY PHARMACY

W. M. H. WAN SULAIMAN[1], M. M. MANAN[1], S. MOHD ALI[2]

[1]Faculty of Pharmacy, Department of Pharmacy Practice, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Faculty of Pharmacy, MAHSA University, Malaysia
Email: wmh859@gmail.com

ABSTRACT

Contraception provides many benefits to married couples or couples on a consensual relationship. This study expects to evaluate quality of life among contraceptive users at community pharmacy settings and in addition to that will provide evidence to community pharmacists to improve their pharmaceutical care in the context of sexual and reproductive health among its customers. A cross-sectional study was conducted by administering the WHOQOL-BREF questionnaires to customers who obtain condom or contraceptive pills at urban community pharmacies in Kuala Lumpur and Selangor. Socio-demographic questionnaires were also administered to the respondents. Respondents'-demographic characteristics were further analyzed and interpreted with appropriate statistical tests. All of 375 respondents reported high quality of life scores in all domains. The mean score for each item were in the range from 3.33 to 3.96. The top three most positive QOL items were self-esteem (mean = 3.96, SD = 0.68), spirituality/religion/personal beliefs (mean = 3.94, SD = 0.71), and personal relationships (mean = 3.87, SD = 0.82). The QOL score among domain were highest in the social domain (mean = 15.37, SD = 2.71) and the lowest score were in the physical domain (mean = 13.56, SD = 1.81). Overall quality of life scores were high in both contraceptive groups and the highest score were reported in the social domain and the lowest in the physical domain.

Keywords: Contraceptive, Pharmaceutical care, Community pharmacy

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LINGUISTIC AND PSYCHOMETRIC VALIDATION OF THE MALAYSIAN VERSION OF DIABETES QUALITY OF LIFE BRIEF CLINICAL INVENTORY (DQoL-BCI)

S. SAMAH[1,2], C. F. NEOH[1,2]*, Y. Y. WONG[3], M. A. HASSALI[4], A. A. SHAFIE[4], Y. W. HAN[5], N. MAT NASIR[5], S. M. LIM[1,2], K. RAMASAMY[1,2]

[1]Faculty of Pharmacy, [2]Collaborative Drug Discovery Research (CDDR) Group, Pharmaceutical and Life Sciences Community of Research, Universiti Teknologi MARA (UiTM), Puncak Alam Campus, Selangor, Malaysia, [3]Faculty of Pharmacy, UiTM, Bertam Campus, Kepala Batas, Penang, Malaysia, [4]School of Pharmaceutical Sciences, Universiti Sains Malaysia, Penang, Malaysia, [5]Department of Primary Care Medicine, Faculty of Medicine, UiTM, Selayang Campus, Selangor, Malaysia
Email: neohchinfen@puncakalam.uitm.edu.my

ABSTRACT

Quality of life (QoL) assessment provides valuable outcome to support clinical decision-making, particularly for patients with chronic diseases that are incurable. A brief, 15-item diabetes-specific tool [i.e. Diabetes Quality of Life Brief Clinical Inventory (DQoL-BCI)] is an instrument designed with actionable items to improve treatment adherence and satisfaction, and patient-provider communication. It is known to be developed in English and validated for use in clinical practice. This simplified tool, however, is not readily available for use in the Malaysian setting. The present study was undertaken to translate and validate the Malaysian version of DQoL-BCI. A forward-backward translation, involving professional translators and experts was conducted according to the principles of good practice delineated by the International Society for Pharmacoeconomics and Outcome Research task force. A total of 202 Type 2 diabetes mellitus (T2DM) patients were then recruited from Diabetic Clinics in Selangor State from June to August 2015. T2DM patients who fulfilled the inclusion criteria were invited to complete the translated DQoL-BCI and data were analyzed using SPSS and AMOS software. Findings from exploratory factor analysis indicated that a 4-factor structure of the Malaysian version of DQoL-BCI was optimal, explained 50.9% of the variance; confirmatory factor analysis further confirmed the 4-factor model fit. There was a positive, moderate correlation between the scores of the Malaysian version of DQoL-BCI and ED-5Q descriptive system score ($r = 0.527$, $p<0.001$). Patients with diabetes macrovascular complications ($p = 0.017$) and microvascular complications ($p = 0.013$) reported poorer QoL Cronbach's alpha 0.703, indicating good reliability of the translated DQoL-BCI. This study has validated the linguistic and psychometric properties of the Malaysian version of DQoL-BCI, thus providing a reliable brief tool for assessing the QoL of Malaysian T2DM patients.

Keywords: Validity, Reliability, Diabetes mellitus, Malaysian

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LEARNING STYLE PREFERENCES OF UNDERGRADUATE PHARMACY STUDENTS IN UNIVERSITI TEKNOLOGI MARA

E. M. JOHN[1], C. F. NEOH[1,2]*, C. M. LONG[1,3], Y. HASSAN[1]

[1]Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, [2]Collaborative Drug Discovery Research (CDDR) Group, Pharmaceutical and Life, Sciences Community of Research, [3]Brain Degeneration and Therapeutics Group, Pharmaceutical and Life Sciences Community of Research, UiTM, Shah Alam Campus, Selangor, Malaysia
Email: neohchinfen@puncakalam.uitm.edu.my

ABSTRACT

Learning style is defined as a manner in which individuals choose to or are inclined to approach a learning situation". The lack of knowledge and understanding on the learning style preferences of pharmacy students can lead to mismatch of learning style and teaching method by instructors which can bring frustration to both parties. The identification of differences in learning styles among pharmacy students is important as this would promote optimal teaching-learning process. The learning style preferences of pharmacy students of Universiti Teknologi MARA (UiTM), however, remain unknown. The present study was undertaken to assess the learning style preferences of pharmacy students, to compare the learning style preferences among year 1, 2, 3 and 4 pharmacy students and to determine demographic variables that significantly influenced the learning style preferences of pharmacy students. A 24-item questionnaire which consisted of two sections was administered to pharmacy students from March to June 2015, at Faculty of Pharmacy, UiTM. Section 1 consisted of socio-demographic items while section 2 was the validated Pharmacists' Learning Styles Inventory (PILS) tool. Data collected were entered and analysed using SPSS version 20. A total of 469 students completed the questionnaire; the majority were female (385/469, 82.1%) and year 4 pharmacy students (156/469, 33.3%). The dominant learning style preferred by the pharmacy students was assimilator (n = 189/469, 40.3%), followed by converger (n = 115/469, 24.5%). The secondary learning style preferred by students was mainly converger (n = 119/469, 25.4%). There was no significant association between dominant learning style preferences and all the demographic variables ($p>0.05$). Similar trend was observed in secondary learning style ($p>0.05$). A variety of teaching methods should be implemented in pharmacy education to compliment all learning style preferences, ensuring an optimal learning environment for all type of learners.

Keywords: Learning style, Pharmacy students

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JOB CHARACTERISTICS OF PHARMACY GRADUATES FROM UNIVERSITI TEKNOLOGI MARA

N. A. OMAR*, M. KARUPPANNAN

Faculty of Pharmacy, Department of Pharmacy Practice, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: aienomar93@yahoo.com

ABSTRACT

Faculty of Pharmacy was established in 2001 and the first batch for Bachelor of Pharmacy was recruited for the May-November 2002 academic session consisting of only 29 students. In 2003, the faculty started to offer postgraduate courses. To date, there are 13 batches of undergraduate students with 9 batches that have already graduated. The aim of this study was to explore the job characteristics and education plans among the pharmacy graduates from Universiti Teknologi Mara (UiTM). Participants were pharmacy graduates from UiTM Puncak Alam. The questionnaires were distributed via online survey using Google form and a reminder was sent after 2 w. The collected data was analysed using SPSS version 22. A total of 95 completed questionnaires were received. The respondents were those who graduated in 2006 to 2014. More than half of them were female (n = 59, 61%) and in the age range of 25-30 y old (n = 74, 78%). About 62% (n = 59) of the respondents went through the 1+1 training (1 y of provisionally registered pharmacist+1 y government service) and the rest (n = 36, 38%) went through the 1+3 training where they served the government for 3 y. All the respondents are currently employed. Majority are working in the government sector (n = 91, 96%) where 70% (n = 66) are in the government hospitals. Only 4% (n = 4) are in the community pharmacy. About 64% (n = 61) of the respondents have no interest or have not decided to further their studies while 36% (n = 33) are currently looking forward to further their studies. Government sectors have attracted many graduates to stay on. With the increasing number of pharmacists in the government sector, it is time to encourage more pharmacists to further their studies to specialize in specific area.

Keywords: Pharmacy graduates, Job characteristics

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EVALUATION OF SAFETY PROFILE OF A FIXED DOSE COMBINATION OF PIOGLITAZONE AND TELMISARTAN IN TERMS OF ORAL SUBCHRONIC TOXICITY STUDY IN RAT

P. SENGUPTA*, A. DAS, F. IBRAHIM

Faculty of Pharmacy, Lincoln University College, Malaysia
Email: pinaki@lincoln.edu.my

ABSTRACT

Hypertension occurs in nearly 50% of type-2 diabetes mellitus patients and is the major contributor to cardiovascular mortality in diabetic subjects. The cardiovascular complications of diabetes are the leading cause of death in people with diabetes. Hence, it is necessary to include antihypertensive medication in the antidiabetic therapy regimen for effective control of complications in diabetes. The safety profile of the established drugs may alter when they combined and can change the toxicity profile of each ingredient. In the present investigation, safety profile of the fixed dose combination of Pioglitazone, an antidiabetic drug and Telmisartan, an antihypertensive drug was evaluated. Toxicological profiling of the combination was assessed to identify any additional toxicity raised when they combined. Pioglitazone and Telmisartan were administered orally at daily doses (low, medium and high dose levels) alone and in combination to rats for evaluation of 28-day oral toxicity of individual drugs and their combination. Two drugs in combination showed no evidence of potentiation of toxicity in 28-day repeated dose toxicity study. There were no significant changes in hematology (Hemoglobin %, Platelets %, Reticulocyte %, total WBC), serum biochemistry (SGOT, SGPT) and organ weight in the oral toxicity study of Pioglitazone-Telmisartan combination in rats. No detectable abnormalities were found in the histopathology of the selected organs (liver and kidney). Pioglitazone and Telmisartan combination can primarily be stated safe in terms of present toxicity findings as no additional toxicity was generated when they combined. This preliminary toxicological result suggests that the short-term treatment of the fixed dose combination of these drugs is devoid of any harmful effects and the combination can be evolve as promising regimen for treatment of diabetic antihypertensive patient.

Keywords: Safety profiling, Toxicity study, Pioglitazone, Telmisartan, Fixed dose combination

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ANTI-INFECTIVES IN CHILDREN BELOW 2 Y OLD: ANALYSIS OF SPONTANEOUSLY REPORTED ADVERSE DRUG REACTIONS CASES

R. ROSLI*, M. M. MANAN

Department of Pharmacy Practice, Faculty of Pharmacy, Universiti Teknologi MARA (UiTM), Puncak Alam Campus, Selangor, Malaysia
Email: rosliana_rosli@yahoo.com

ABSTRACT

Anti-infectives were among the most widely prescribed therapeutic agents in children below 2 y old. Despite its extensive use, the information on safety profiles of anti-infectives used in this population is not well documented. The aims of this study were to describe the adverse drug reactions (ADRs) related to anti-infectives and to identify the common drugs associated with ADRs reported for children below 2 y old in Malaysia. Reports on ADRs related to all anti-infectives for children aged from birth to 2 y old from 2000 to 2013 were retrieved from the national ADRs database system. Data were analysed with respect to gender, type of reporters, suspected agents (Anatomical Therapeutic Chemical (ATC) Classification) and System Organ Class (SOC). 802 ADRs were observed from 561 Malaysian ADR reports. Of these reports, 90.2% were related to infants whilst only 9.8% reports on neonates. A large proportion (95.4%) of the reports was received from healthcare providers, mainly pharmacists (51.7%) in the government health facilities. Reports encompassed medicine from various anti-infectives groups: antibacterials (95%), antivirals (3.7%), antimycotics (0.9%) and antimycobacterial (0.4%). Penicillins and other beta-lactam antibacterials accounted for more than 70% of all drugs implicated in ADRs. For antibacterials, only 1.2% of reports belonged to newer antibacterial generations. The most common SOCs associated with ADRs was skin and appendages disorders (70.8%) and the frequently reported symptoms were rash maculo-papular (n=165) followed by rash (n=153) and urticaria (n=115). A small proportion of cases were classified as severe (7.5%) but there were no fatalities seen in this study. ADRs related to anti-infectives in children below 2 y old were predominantly classified to have possible causality with large proportion of mild and moderate ADRs which recovered without sequalea. However, more research is needed as studies have shown higher incidence of ADRs in real hospital setting than in national databases.

Keywords: Adverse drug reactions, Children, Pharmacovigilance, Anti-infectives

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ATHEROSCLEROSIS AND THE CHOLESTEROL THEORY: A REAPPRAISAL

A. P. WONG*

Cyberjaya University College of Medical Sciences (CUCMS), Cyberjaya, Selangor, Malaysia
Email: wongangpeng@hotmail.com

ABSTRACT

Atherosclerosis is the precedent to ischemic heart disease, which may lead to angina, myocardial infarct, or heart failure; or to ischemic cerebrovascular disease, which may lead to stroke. The prevailing belief underlying conventional approaches to treatment of atherosclerosis and its sequel is that a diet high in cholesterol and saturated fat is the main contributory factor, triggering cholesterol build up in the intima of the blood vessels. Over the last 60 y, the blame has shifted from fats, to saturated fats, to low-density lipoprotein (LDL), and finally to oxidized LDL (Ox-LDL). Therapy has been predominantly aimed at lowering cholesterol and control of risk factors. However, there is an alternative hypothesis about the cause of heart disease linking it to the weakening of the vascular collagen matrix at the sites of high hemodynamic stress (coronary arteries) which triggers the infiltration of lipoprotein(apo) [Lp(a)] and plaque development. Accordingly, the vascular deposition of large molecules such as Lp(a) and atherosclerosis is the result of the body's endogenous protective mechanism to reinforce the weakened artery walls. Understanding this mechanism may guide the natural prevention of this disease and form the basis for developing effective therapeutic strategies aiming at natural reversal of atherosclerosis through the reinforcement of the vascular wall structure as its primary goal. This reappraisal of atherosclerosis and the cholesterol theory looked at the historical development of the theory, and the Rath and Pauling unified theory of cardiovascular disease.

Keywords: Atherosclerosis, Low-density lipoprotein, Oxidized low-density lipoprotein, Lipoprotein

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ADVERSE DRUG REACTIONS OF IBUPROFEN IN MALAYSIA

R. ABD. MANAN*, N. M. SHARIF, M. H. OTHMAN, M. MUKHTAR, N. ABD. AZIZ

Faculty of Pharmacy, Universiti Teknologi MARA (UiTM), Puncak Alam Campus, Selangor, Malaysia
Email: ayu_ppf@yahoo.com

ABSTRACT

Ibuprofen is widely available and extensively used in the world, including Malaysia. Unfortunately health care providers do not have a real picture of adverse reactions of this drug in Malaysia. The objective was to study the adverse drug reactions of Ibuprofen in Malaysia. This is a descriptive retrospective study where all ADR data related to unregistered traditional medicines, from 2000 to 2014, were extracted from national pharmacovigilance Quest 2 database. A total of 675 Ibuprofen induced adverse drug reaction were reported from 2000 to 2014 but only 583 reports was analyzed after fulfilled the inclusion criteria. The trend keeps increasing from 2007 until 2014. Most of ADR induced by Ibuprofen reported by young adult with 44.60% (260) followed by children (n=105, 18.01%), middle age group (n=89, 15.27%) and adolescent (n=76, 13.04%). There were 1246 drug adverse reactions from a total of 583 reports. Based on adverse drug reaction (ADR) classification of Pharmacovigilance Unit, Malaysia 412(33.07%) affected body as whole-general disorders followed by skin (n=407, 32.66%), respiratory (n=172, 13.80%) and urinary system (n=129, 10.35%). Only 0.16% of ADR affected cardiovascular (CVS), vascular and heart rate respectively. Out of 172 reaction affected respiratory disorders, based on preferred term the highest frequency was dyspnea reaction with 124 (72.09%) which is included dyspnea (n=9), breath shortness (n=96) and breathing difficult (n=19). More local data is needed to approve the safety signal. Health care professionals should apply information on ADRs of local data when choosing the most appropriate therapy for patient.

Keywords: Ibuprofen, Adverse drug reactions, Malaysia

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EXPLORING THE CURRENT MANAGEMENT OF TUBERCULOSIS AND DIABETES MELLITUS AMONGST HEALTHCARE PRACTITIONERS AND PATIENTS: A QUALITATIVE STUDY

S. MOHD NASIR, S. GNANASAN*

Department of Clinical Pharmacy, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia
Email: shubashini.gnanasan@gmail.com

ABSTRACT

Diabetes mellitus (DM) is the most common comorbidity in tuberculosis (TB) patients with prevalence as high as 15-30% in Malaysia. The convergence of these two diseases is linked to poorer TB treatment outcomes, increased chances of relapse, reactivation of latent TB infection and higher risk of death during TB treatment. Realising this joint burden, World Health Organisation (WHO) and the International Union Against Tuberculosis and Lung Disease (The Union) have developed a provisional collaborative framework to guide national programmes, researchers, and those directly involved in the care, prevention, and control of TB and DM in order to establish a coordinated response to both diseases. In order to improve the provision of pharmaceutical care that is suited to every TB-DM patient, a thorough understanding of obstacles and limitations in the existing healthcare system must be attained. This study aims to explore the current management of TB and DM amongst healthcare practitioners and patients and seeks to unravel new ideas of creating awareness and imparting knowledge about the link between TB and DM amongst healthcare practitioners and patients. Data were collected through face-to-face interviews with healthcare practitioners and patients at Institute Perubatan Respiratori which were audio-recorded before being transcribed verbatim and analysed thematically. The themes included TB-DM management, directly observed treatment short-course (DOTS), knowledge about TB-DM link and barriers in healthcare system. The study revealed that the integration of DM management into the care of TB patients with DM comorbidity was still found to be lacking especially concerning DM monitoring in TB patients. DOTS visits were not utilised for DM monitoring due to time constraint and patient reluctance. Educational tools are imperative to educate both healthcare practitioners and patients effectively.

Keywords: Tuberculosis, Diabetes mellitus, Pharmacist, DOTS, Malaysia

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A QUALITATIVE STUDY ON PHARMACIST-PHYSICIAN COLLABORATIVE PRACTICE AT MUAR DISTRICT HEALTH CLINIC

F. A. SANI[1], Y. HASSAN[1], N. UJANG[2]

[1]Faculty of Pharmacy, Department of Pharmacy Practice, Universiti Teknologi MARA, [2]Pejabat Kesihatan Daerah Muar, Jabatan Kesihatan, Johor

Email: anursha2001@yahoo.com

ABSTRACT

Collaboration among healthcare providers is a relatively new concept for pharmacy especially at the primary care health clinic. Effective collaborative practice highly depends on the good relationship and teamwork between interprofessional team members especially between pharmacist-physician. The objective of the study is to describe the contributing factors and challenges for the successful implementation of the Medication Therapy Adherence Clinic (MTAC) in the current situation of diabetes management at the primary care health clinic setting. A qualitative study of four focus groups consisting of pharmacists and physicians were conducted at four different health clinics in Muar district. A semi-structured interview questionnaire had been used to facilitate discussions. Conversations were tape-recorded, transcribed verbatim and coded by themes. Twelve pharmacists and ten clinicians with an average 6 y of service in health clinic setting participated in the study. The respondents highlighted some factors that contribute to the effective collaboration between health care professionals. The most common factors are effective feedback and attitude of the healthcare professionals. In addition, working culture of certain clinics, and enough manpower may also affect the collaboration. Existing protocols and guidelines should be revised and improved in certain aspects to make it more practical to be used locally. This study also gives an overall view to the pharmacist of the physicians' expectation for collaboration in providing DMTAC service. Pharmacists should be more open and willing to share their findings in order to improve beneficial care to the patients. Pharmacists especially in the government health clinic may have some limitations in servicing the public, but they have the capability to cultivate and implement drug therapy management by MTAC in managing type-II diabetes patients. Effective communication and collaboration between pharmacist and physician may help to improve the glycemic control of diabetic patients at the primary care health clinic.

Keywords: Collaboration, Pharmacist-physician collaborative practice, Primary care health clinic, Medication Therapy Adherence Clinic, Qualitative

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MEDICATION USE BEHAVIOURS AND HEALTH OUTCOMES OF TYPE 2 DIABETES MELLITUS HEALTH CARE IN THE OCCUPIED PALESTINIAN TERRITORIES

R. S. MOSLEH[1], S. ZYOUD[2], W. SWEILEH[2], S. MOHD ALI[3], M. M. MANAN[1]*

[1]Department of Pharmacy Practice, Faculty of Pharmacy, Universiti Teknologi MARA, Puncak Alam Campus, Selangor, Malaysia, [2]Department of Pharmacy, An-Najah National University, Nablus, Palestine, [3]Department of Pharmacy Practice, Faculty of Pharmacy, MAHSA University, Malaysia
Email: mmmanan2002@yahoo.com

ABSTRACT

The prevalence of Type 2 diabetes mellitus (DM2) patients has increased rapidly worldwide, especially in Middle East Countries. In Occupied Palestine Territories (OPT), there were problems in health care of DM2 in OPT, such as lifestyle, inappropriate perspectives and medication use and lack of research related to health care. The present study aimed to identify the medication behaviours and outcomes among DM2 patients in OPT. The study was an observational retrospective study. Data from 330 DM2 patients were collected from National Centre of Chronic Diseases and Dermatology in Ramallah from November 2012 through November 2013. More than half of the participants were male (51.2%, N = 169; female: 48.8%, N = 161). Sixty nine of the participants were low income patients. Two hundred seven (62.73%) participants had been diagnosed with DM2 for more than ten years. One participant was under diet control without any antidiabetic medications. Seventy four (22.4%) participants received only Insulin injections, and seventy four (22.4%) participants were receiving only oral hypoglycemic medications. More than half of the participants (N=181, 54.8%) received combined oral Insulin injection medications The mean and median numbers of total medications per day were 6.3848 (SD = 2.9) and 6, respectively. HbA1c testing was not measured for fifty nine participants (17.87%). Based on WHO medical standards of diabetes care, only 4.43% of the participants who have been pursuing their HbA1c levels was well controlled (i.e. optimal control: HbA1c<6%). The control of almost forty one percent of them (40.96%, N = 111) were acceptable but need improvement (HbA1c 6%–8%), and 54.61% (n=148) were in poor control and needed intensive treatment (HbA1c>8%) and intervention. Although there were some positive signs in some of the factors, DM2 care management in OPT was relatively poor. Various interventions from health care professionals should be developed to improve the quality of healthcare services for DM2 patients in OPT.

Keywords: Type 2 diabetes mellitus, Health care, Occupied Palestinian Territories (OPT), Medication use behaviors

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INVESTIGATION ON THE IMMEDIATE EFFECTS OF MALAY CUPPING THERAPY ON SOME HORMONAL AND BIOCHEMICAL PARAMETERS

S. A. ABDULLAH[1], M. N. MOHD NAJIB[1]*, S. SULAIMAN[1], S. MOHAMED[2]

[1]Faculty of Pharmacy, Bertam Campus, Universiti Teknologi MARA, Kepala Batas, Penang, [2]Institut Pergigian dan Perubatan Termaju, Universiti Sains Malaysia, Kepala Batas, Penang
Email: mohdna2857@ppinang.uitm.edu.my

ABSTRACT

Cupping therapy is one of the oldest methods used mainly for health maintenance as well as treatment all over the world. It could be interpreted as the act of suction and removal of blood after some superficial incisions made on the skin at various points on the body. Due to positive perception and belief, the therapy is still widely practiced among the Malay community until today. However, the exact mechanism of how cupping work is still unclear, with only little researches tackling the area. The objective of this study is to investigate the immediate effect of Malay cupping therapy on some hormonal and biochemical properties. Twenty consented subjects between the ages of 26-72 y old were randomly selected for this study. Five milliliters blood from the medial cubital was collected one h before cupping and another 5 milliliters one h post cupping for each session. The analyzed hormones (endorphine and enkephaline) and biochemical parameters showed some changes which were found to be statistically significant. In conclusion, there are some immediate hormonal and biochemical changes due cupping therapy. This study hopes to provide further information on the effects of cupping therapy on physiological properties. Further studies with larger sample size and of longer duration would reveal more conclusive results.

Keywords: Cupping, Hormonal, Biochemical

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EFFECT OF WET CUPPING THERAPY ON BIOCHEMICAL PARAMETERS OF VENOUS BLOOD COLLECTED FROM OBESE MALE INDIVIDUALS

M. A. MD YASSIN[1], M. MOHAMAD LOCMAN[2], Y. MOHD YUNUS[3], N. S. A. SURATMAN[1], M. M. MANAN[1]*, A. ADAM[1]

[1]Faculty of Pharmacy, [2]Health Centre, Universiti Teknologi MARA, Puncak Alam, Selangor, Malaysia, [3]Al-Jawahir Therapy Centre, Shah Alam, Selangor, Malaysia
Email: mmmanan2002@yahoo.com

ABSTRACT

Cardiovascular Disease (CVD) is a leading health problem caused by several risk factors which include obesity. Obesity is defined as abnormal or excessive fat accumulation that may impair health and is the fifth leading risk for global deaths. Wet cupping therapy (WCT) is a recognized Malay traditional medicine that was shown pre-clinically to be effective against CVD parameters. There was no study to determine effect of WCT on venous blood parameters. This study was conducted to determine effect of WCT on blood of obese male individual's pre-and post-wet cupping sessions. Obese male participants (n=31) aged 22 to 39 y old were recruited with informed consent to receive two WCT sessions. All participants were subjected to venipuncture at baseline (BASE, day=0), before cupping (BWCT, day=21) and after cupping session (AWCT, day=42). Wet cupping sessions were conducted on days 28 and 35. For each WCT session, 3 cupping points were applied on participant's skin. Venous blood and cupping blood were collected and analysed. Paired sample t-test was utilized to determine mean differences at baseline, before and after WCT. Independent sample t-test was used to measure differences between venous and cupping blood samples. Level of significance was set to 5% and data analysis was performed by using Statistical Package for Social Science (SPSS) v.20 Windows version. There were significant changes in alanine transaminase ($p<0.05$), aspartate transaminase ($p<0.05$), glucose ($p<0.05$), total protein ($p<0.001$) and uric acid ($p<0.05$) between BWCT and AWCT sessions. No significant changes following WCT were observed in levels of calcium, cholesterol, creatinine, high density lipoprotein, low density lipoprotein, total bilirubin and triglyceride. Two sessions of WCT significantly reduced glucose, total protein and uric acid thus may have a role in management of hyperglycaemia and hyperuricaemia.

Keywords: Obesity, Cupping therapy, cardiovascular disease