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ABSTRACTS

Oral Presentation

Poster Presentation

O-01

Genetic Polymorphism of *hOGG1* (C1245G) and the Risk of Cervical Cancer

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Abstract

Human 8-oxoguanine glycosidase 1 (hOGG1) is a DNA repairing enzyme which is encoded from *hOGG1*. Polymorphism at the position of C1245G of this gene has 3 genotypes; CC, GG, and CG genotypes. Moreover, this polymorphism may leads to cervical cancer development. So, the aim of this study was to investigate association between *hOGG1* polymorphism and cervical cancer risk among northeast Thais. From this study, we conducted an age-matched case-control study with 182 cervical cancer cases and 182 healthy controls. The peripheral white blood cell samples were collected for DNA extraction and analyzed for the *hOGG1* polymorphism by using real-time polymerase chain reaction (RT-PCR) method. Result of this study shows the genotype distribution of *hOGG1* polymorphism in controls and cervical cancer group did not show a significant difference for the risk of cervical cancer (P>0.05). The frequency of the CG genotype can reduce risk of cervical cancer development in subjects who have partner with smoking behavior (OR=0.38, 95%CI: 0.17-0.85, P=0.01 and adjusted OR=0.38, 95%CI: 0.18-0.81, P=0.01) and subjects who never use oral contraceptive pills (OR=0.44, 95%CI: 0.20-0.96, P=0.02 and adjusted OR=0.45, 95%CI: 0.22-0.94, P=0.03). Then, after analysis with two risk factors, the data shows, CG genotype reduces risk of cervical cancer in the subjects with oral contraceptive use and smoking habit of partner (OR=0.30, 95%CI: 0.10-0.86, P=0.01 and adjusted OR=0.30, 95%CI: 0.11-0.79, P=0.02). So, we can conclude that the polymorphism of *hOGG1* is the associated factor of cervical cancer in women who have partner with smoking behavior. They should avoid receiving smoke from other people and they should examine their genetic polymorphism for preventing and monitoring treatment of cervical cancer.

Keywords: *hOGG1*, polymorphism, oral contraceptive use, smoking habit of partner, cervical cancer.

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O-02

Detailed Analyses of Sexual & Smoking Behaviors and Risk of Cervical Cancer in Northeastern Thailand

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Abstract

Cervical cancer is a serious public health problem in Thailand. We investigated possible risk factors for cervical cancer including HPV infection, p53 polymorphism, smoking and reproductive history among women in Northeast Thailand using a case control study with 177 cases and age-matched controls. Among the HPV carriers, a significantly increased risk for cervical cancer with an OR of 36.97 (P<0.001) and an adjusted OR of 38.07 (P<0.001) were observed. Early age at first sexual exposure, and multiple sexual partners increased the risk of cervical cancer with ORs ranging 1.73-2.78 (P<0.05). The interval between menarche and first sexual intercourse <6 years resulted in a significant increase in the risk for cervical cancer with ORs ranging 3.32-4.09 and the respective adjusted OR range for the 4-5 and 2-3 year-old groups were 4.09 and 2.92, respectively. A higher risk was observed among subjects whose partner had smoking habits, whether currently or formerly; with respective ORs of 3.36 (P<0.001) and 2.17 (P<0.05); and respective adjusted ORs of 2.90 (P<0.05) and 3.55 (P<0.05). Other smoking characteristics of the partners including smoking duration≥20 years, number of cigarettes smokes ≥20 pack - years and exposure time of the subject to passive smoking≥5 hrs per day were found to be statistically significant risks for cervical cancer with adjusted ORs of 3.75, 4.04 and 11.8, respectively. Our data suggest that the risk of cervical cancer in Thai women is substantially associated with smoking characteristics of the partner(s), the interval between menarche and first sexual intercourse as well as some other aspects of sexual behavior.

Keywords: Menarche, sexual exposure, HPV infection, cervical cancer, Northeast Thailand.

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O-03

Improvements of Vascular Dysfunction and Baroreflex Sensitivity by Tetrahydrocurcumin and Deferiprone in Iron Overloaded Mice

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Abstract

Iron is an essential element; however, excess iron is a major cause of iron-induced oxidative stress, vascular dysfunction and disruption of baroreflex sensitivity (BRS). Tetrahydrocurcumin (THU), a strong antioxidant hydrogenated from curcumin of *Curcuma longa* and deferiprone (L1), an oral iron chelator, have been used in this study for evaluating the effects on vascular function and BRS in iron overloaded mice. An iron overload condition was induced in adult male ICR mice by daily iron sucrose injection at a dose of 10 mg/kg for eight consecutive weeks. Propylene glycol (a vehicle), THU (50 mg/kg), L1 (50 mg/kg) or THU plus L1 (50 and 50 mg/kg, respectively) were intragastrically administered once daily throughout the period of iron sucrose administration. After eight weeks, arterial blood pressure, vascular reactivity to vasoactive agents and BRS were measured. Aortic superoxide anion production, plasma malondialdehyde, urinary nitric oxide metabolites, blood glutathione (GSH) and serum iron indices were also evaluated. The results indicated that iron overloaded mice showed a marked increase of serum iron indices, especially non-transferrin bound iron (NTBI) (P<0.001 *vs* untreated controls) and revealed deleterious effects on inducing high blood pressure which were related with impairment of vascular function and lowering bradycardic response of BRS (P<0.05 *vs* untreated controls). Moreover, iron toxicity also increased oxidative stress whereas blood GSH was depleted. THU or L1 partly alleviates these effects. A combined therapy of THU and L1 exerted a greater effect than THU or L1 monotherapy by improving vascular function, increasing BRS, decreasing oxidative stress, enhancing antioxidant status and reducing NTBI levels. Overall findings suggests the synergistic effect of THU and L1 on improvement of vascular dysfunction, BRS and reducing oxidative stress in iron overloaded mice. THU may be used as an iron chelating agent in excessive iron condition.

Keywords: Iron overload, tetrahydrocurcumin, deferiprone, vascular dysfunction, baroreflex sensitivity.

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O-04

Protective Effect of Tetrahydrocurcumin on Phenylhydrazine-Induced Hemolytic Anemia and Oxidative Stress in Rats

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Abstract

Phenylhydrazine (PHZ) is a known hematotoxic and strong oxidant agent. PHZ oxidation causes free iron release resulting in free radicals generation, oxidative stress and tissue damage. Tetrahydrocurcumin (THU) is an antioxidant derived from curcumin by hydrogenation. Recently, attention has been focused on THU because this compound appears to exert greater antioxidant activity in various conditions. The aim of present study was to investigate whether THU could protect against PHZ-induced hemolytic anemia and oxidative stress in rats. Male Sprague-Dawley rats were exposed to PHZ (15 mg/kg/day; i.p.) and orally administered with THU (30 and 100 mg/kg/day) for 8 days. After 8 days of treatment, arterial blood pressure, hematocrit, superoxide ($O_2^{\bullet-}$) production and non-transferrin bound iron (NTBI) were assessed. The oxidative stress markers, malondialdehyde (MDA) and protein carbonyl in plasma and tissues such as heart, liver and kidney were determined. Our results showed that the systolic pressure, diastolic pressure, mean arterial blood pressure and hematocrit of PHZ treated rats were markedly decreased whereas serum NTBI level was dramatically increased as compared to the normal controls. Interestingly, the improvement of blood pressure, and hematocrit during PHZ exposure was associated with alleviation of free iron release and oxidative stress in THU-treated rats by reducing NTBI, $O_2^{\bullet-}$ production in thoracic aorta, MDA and protein carbonyl in plasma and tissues. Results in this study reveal the beneficial effect of THU on prevention of cardiovascular damage and oxidative stress in PHZ treated rats.

Keywords: Tetrahydrocurcumin, phenylhydrazine, hemolytic anemia, oxidative stress.

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O-05

Modification of Electrical Brain Wave by *Citrus sp.* Essential Oil Inhalation

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Abstract

The essential oil of orange (*Citrus sp*.) has been widely used in aromatherapy according to its anxiolytic effect. Electroencephalography (EEG) is one of the reliable neurological techniques used to study the brain and behavioral functions. This method has been used to record brain waves for frequency analysis. The present study examined the effect of citrus essential oil (EO) on EEG pattern in adult male Wistar rats. Animals were anesthetized with Zoletil® (60 mg/kg i.m.). Stereotaxic apparatus was used to fix rat skull for the implantation of 4 stainless steel screw electrodes over the frontal and parietal cortices using bregma as the landmark. Ampicillin (145 mg/kg i.m.) was daily injected for 4 days after the surgery and the animals were allowed to recover for 10 days. On the day of the experiment, individual rat EEG was recorded using PowerLab®/4s system. The EEG signals were displayed on LabChart® software and processed on a personal computer. The inhalation of EO (20 and 200 μl) and distilled water as control were performed in the EEG recording chamber. Frontal EEG analysis showed that EO (20 μl) increased the percent baseline powers in low frequency bands ranging from theta (4.7-6.6 Hz), alpha1 (7-9.4 Hz), alpha2 (9.8-12.5 Hz) to beta1 (12.9-18.4 Hz) waves. However, in the parietal cortex, only alpha2 and beta1 powers were significantly increased. In addition, EO 200 μl increased the percentage of the baseline powers only in the theta and the alpha2 bands in the frontal cortex. In conclusion, this study demonstrates EEG pattern in response to inhalation of citrus EO. The data may represent EEG biomarker of the EO in the central nervous system (CNS). However, it remains unknown in terms of its CNS mechanism.

Keywords: Essential oil, citrus oil, electroencephalogramphy, rat.

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O-06

Curcumin Attenuates Oxidative Stress and Improves Hemodynamic Status in 2K-1C Hypertensive Rats

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Abstract

Curcumin (CUR), a dietary spice and strong antioxidant, is a natural polyphenol compound isolated from *Curcuma longa*. The aim of this study was to investigate the potential antihypertensive and antioxidative effects of CUR in a rat model of two-kidney, one-clip (2K-1C) renovascular hypertension. Male Sprague-Dawley rats were induced 2K-1C hypertension by placing a silver clip (0.2 mm, i.d.) around the left renal artery. Sham-operated rats served as normotensive controls. 2K-1C and sham-operated rats were treated with CUR at doses of 50 and 100 mg/kg/day or vehicle continuously for six weeks. Systolic blood pressure (SP) was measured weekly by using tail-cuff plethysmography. At the end of experiments, the effects of CUR on hemodynamics and oxidative stress status were evaluated. SP was significantly increased in rats with renal artery clipping as compared to the sham-operated rats ($P<0.05$). The 2K-1C hypertensive rats also showed a significant elevation of mean arterial pressure (MAP), hindlimb vascular resistance (HVR) and oxidative stress ($P<0.05$). Administration with CUR prevented hemodynamic changes by decreasing MAP and HVR as compared to the 2K-1C untreated controls ($P<0.01$). The higher levels of oxidative stress, such as superoxide production, malondialdehyde, protein carbonyl were also attenuated in 2K-1C rats treated with CUR. In addition, treatment with CUR also improved the level of antioxidant glutathione ($P<0.05$). Overall findings suggest that CUR protects against the development of hypertension and exerts antioxidative effect in 2K-1C renovascular hypertensive rats.

Keywords: Curcumin, 2K-1C, renovascular hypertension, antioxidant, glutathione.

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O-07

Immunohistochemical Analysis of Chondroregulatory Protein Expression in the Epiphyseal Plate of Bromocriptine-Treated Lactating Rats

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Abstract

Endochondral bone growth is enhanced in lactating rats, leading to bone elongation. This lactation-induced bone elongation could be inhibited by administration of dopamine D2 receptor agonist, bromocriptine. However, the effect of bromocriptine on the alteration of regulatory proteins in the growth plate, the principal region controlling longitudinal bone growth, remained unknown. In the present study, we quantified immunohistochemical signals of chondroregulatory proteins in the growth plate of bromocriptine-treated lactating rats. The studied proteins were parathyroid hormone-related peptide (PTHrP), PTHrP receptor, Indian hedgehog (Ihh), sex determining region Y-box 9 (Sox9), and *runt*-related transcription factor 2 (Runx2). The results showed that bromocriptine upregulated Ihh expression in the hypertrophic chondrocytes during early and mid-lactation, whereas the expression of PTHrP receptor, but not PTHrP, was increased in the proliferative and hypertrophic chondrocytes during mid- and late lactations. The expression of Runx2 was markedly suppressed in the hypertrophic zone, while Sox9, an important transcription factor for promoting chondrocyte proliferation, was upregulated in the hypertrophic zone of bromocriptine-treated lactating rats. Taken together, it could be concluded that bromocriptine increased Ihh, PTHrP receptor and Sox9 expressions but suppressed Runx2 expression, which might, in turn, accelerate chondrocyte proliferation loop but delay chondrocyte hypertrophy, thereby decelerating bone elongation.

Keywords: Bromocriptine, chondrocyte, growth plate, Indian hedgehog (Ihh), PTHrP.

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O-08

Effects of Curcumin on Diabetes-Induced Leukocyte-Endothelium Interaction: Roles of NOX2 and Thioredoxin-Interacting Protein

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Abstract

Hyperglycemia increases thioredoxin-interacting protein (Txnip) expression that causes cellular redox imbalance and also enhances intercellular adhesion molecule-1 (ICAM-1) which leads to increase leukocyte-endothelium interaction to promote vascular inflammation. Moreover, the leukocyte accumulation in microcirculation causes endothelial damage by NADPH oxidase enzyme (NOX2). Curcumin exhibits antioxidant, and anti-inflammatory properties. The study aimed to evaluate the mechanisms of curcumin on preventing diabetes-induced leukocyte-endothelium interaction associated with its actions on NOX2 and Txnip expressions. The rats were divided into four groups: control group (CON), control-treated with curcumin group (CONCUR, 300 mg/kg BW), diabetic-treated with corn oil group (DM; streptozotocin (STZ), i.v. 55 mg/kg BW), and diabetes-treated with curcumin (DMCUR). The curcumin supplementation was started at 10 days after STZ-injection. 12th week after STZ injection, iris blood perfusion and leukocyte adhesion were assessed by using laser Doppler and intravital fluorescent microscopy, respectively. Plasma glucose and HbA1c were also determined by using enzymatic and turbidimetric immunoinhibition methods, respectively. P47phox expression (marker of NOX2 activation), Txnip expression, and malondialdehyde (MDA) level at eye fundus were determined by Western Blot analysis and TBAR assay, respectively. The results showed that body weight and iris blood perfusion of DM and DMCUR were decreased significantly as compared to CON (P<0.001), but plasma glucose and HbA1c of DM and DMCUR were increased significantly as compared to CON (P<0.001). Interestingly, the leukocyte adhesion, p47phox expression, and MDA level in DMCUR were decreased significantly when compared to DM (P<0.05). It is concluded that 300 mg/kg BW curcumin could ameliorate diabetic vascular inflammation by decreasing ROS overproduction, reducing leukocyte-endothelium interaction associated with NOX2 inhibition. These data suggest that NOX2 activity should be a primary factor in diabetes-induced vascular inflammatory reactions. Curcumin might be a novel therapeutic strategy for treatment of the diabetic retinopathies associated with vascular inflammation in the future.

Keywords: Diabetes, curcumin, NOX2, Txnip, endothelial dysfunction, leukocyte adhesion.

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O-09

Effects of Avian Influenza Virus (H5N1) on Cytokine and Chemokine Gene Expression in Human Respiratory Epithelium

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Abstract

One important feature of influenza A/H5N1 immunopathogenesis is the appearance of hypercytokinemia (cytokine storm) which is characterized by the extreme production and secretion of large numbers and excessive levels of pro-inflammatory cytokines. H5N1 targets specific host cells, i.e., bronchiolar cells, macrophages and type 2 alveolar pneumocytes. These cells, but not those in the tracheal or the upper path of the respiratory tract, express the α2-3 sialic acid receptors, the binding site for H5-hemagglutinin. It has been reported that H5N1 infection induced the production of TNF-α, IP-10, IL-6 and IFN-β in human alveolar and bronchial cells. Recent data, however, suggest that H5N1 can also induce cytokine release via an infection-independent mechanism. This study aimed to investigate the mechanism by which H5N1 triggers expression of cytokines/chemokines in the respiratory epithelium. Expression of the mRNAs of interest in 16HBE14o cells treated with inactivated-H5N1 was analyzed by quantitative RT-PCR. Our results showed that the inactivated-H5N1 increased mRNA expression of IL-6 and IL-8 but not of TNF-α, RANTES and IP-10. Since cytochalasin D treatment, which disrupts actin cytoskeleton and, hence, inhibits endocytotic uptake of the virus, had no effect on the effect of the inactivated-H5N1 on cytokines mRNA expression, this effect of H5N1 did not require the presence of the virus particles in the cytosol. Cytokines production can, therefore, be initiated by an interaction between H5N1 capsule and surface membrane of the respiratory epithelial cells. The underlying mechanism by which H5N1 generates this infection-independent effect on cytokine production is currently under investigation.

Keywords: H5N1, respiratory epithelium, cytokine, chemokine.

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O-10

The Effects of Natural and Semi-Synthetic Ecdysteroids on C2C12 Proliferation and Differentiation

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Abstract

Phytoecdysteroids, which are structurally similar or identical to insect molting hormones, produce a range of effects in mammals, including enhancements of growth and physical performance. 20-Hydroxyecdysone (20E), which is the main phytoecdysteroid, enhanced proliferation of myoblast cells *in vitro* suggesting that analogs of ecdysteroids may enhance proliferation of muscle cells. In this study, we investigated the effect of 20E and its semi-synthetic analogs on mouse myoblast (C2C12) cell proliferation and differentiation using MTT assay and myosin heavy chain immunostaining and Western blotting, respectively. The effects on cell proliferation of 20E and its 7 analogs, using testosterone as a positive control, at doses of 100 nM and 1 μM were studied at 24 and 48 hrs. The results showed that testosterone, 20E and its 5 analogs increased the percentage of C2C12 cell proliferation compared with control (vehicle). Then, the dose-response of 20E and its 5 analogs were studied at 0.1, 1, 10 and 100 nM, 1 and 10 μM, respectively. Results showed that most compounds except compound AS-CP012, AS-CP014, AS-CP015 exhibited optimal effect at the concentration 10-100 nM. However, differentiation study revealed only 20E and AS-CP015 had positive effects on C2C12 cells. It is concluded that only 20E enhances both proliferation and differentiation whereas other analogs increase either proliferation or differentiation of C2C12 cells. This study suggests that 20E and some of its analogs have potential for promoting muscle growth and regeneration, which may be of clinical values in muscle repairs after injury or in sarcopenia.

Keywords: Phytoecdysteroids, cell proliferation, cell differentiation.

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O-11

The Combined Endothelial Progenitor Cells (EPCs) and Mesenchymal Stem Cells (MSCs) Accelerate Wound Healing in Diabetic Mice by Enhancing Angiogenesis and Reducing Neutrophil Infiltration

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Abstract

Non-healing diabetic ulcer was the most common cause of lower extremity amputation in diabetes. Several studies showed that EPCs can increase angiogenesis, release of proangiogenic factors and promote wound healing. However, there is no report for the combination of EPCs and MSCs for diabetic wound healing. Therefore, the objective of this study was to access the effects of combined EPCs and MSCs on angiogenesis and wound healing in diabetic mice. Balb/c nude mice were divided into five groups; control group (Control), diabetic group (DM, injection of streptozotocin 45 mg/kg i.p. daily for 5 days), diabetic group with MSCs (DM+MSCs; $1x10^6$ cells), diabetic group with EPCs (DM+EPCs; $1x10^6$ cells), and diabetic group with combined MSCs and EPCs (DM+MSCs+EPCs; $0.5x10^6$ cells each). After 6-7 weeks, mice were anesthetized and created bilateral full-thickness excisional skin wounds on the dorso-rostral back (0.6x0.6 cm^2). Each mouse received fibrin gel or stem cells injection into wound bed. On day 7 and 14 post-wound, the percentage of wound closure (%WC), the percentage of capillary vascularity (%CV), neutrophil infiltrations and VEGF level were determined by using Image Pro-Plus, intravital fluorescence microscopy, H&E staining and ELISA, respectively. The %WC of all groups increased significantly compared with DM on day 7 and 14. On day 7, the %CV of all groups increased significantly compared with DM. The %CV of DM+EPCs+MSCs group was significantly higher than DM+MSCs and DM+EPCs groups. On day 14, the %CV of DM+EPCs+MSCs group was increased significantly compared with DM. Number of neutrophil infiltration in all groups were decreased significantly than DM at day 14. The VEGF levels of all groups increased significantly compared with DM on day 7. In conclusion, the present study has demonstrated that the combined EPCs and MSCs can increase VEGF level, angiogenesis and reduce neutrophil infiltration, which lead to enhance wound healing in diabetic mice.

Keywords: Endothelial progenitor cells, mesenchymal stem cells, angiogenesis, wound healing, diabetes mellitus.

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O-12

Transepithelial Chloride Secretion via CFTR Is a Major Determinant of Intestinal Fluid Loss During *Vibrio cholerae* Infection

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Abstract

Cholera is a diarrheal disease causing significant morbidity and mortality worldwide. This study aimed to establish an adult mouse model of *Vibrio cholerae*-induced diarrhea and to investigate pathophysiology of cholera diarrhea using this model. Ligated ileal loops of adult mice were inoculated for 6, 9, 12 and 18 h with a classical O1 hypertoxigenic 569B strain of *Vibrio cholerae* (10^7 CFU/loop). Time-course studies demonstrated that optimal period for inducing diarrhea was 12 h post-inoculation, when peak intestinal fluid accumulation (loop/weight ratio of ~ 0.2 g/cm) occurred with the highest diarrhea success rate (90 %). In addition, pathogenic numbers of *V. cholerae* (~ 10^9 CFU/g tissue) were recovered from ileal loops at all time points between 6-18 h post- inoculation with the diarrheagenic amount of cholera toxin being detected in the secreted intestinal fluid at 12 h post-inoculation. Interestingly, repeated intraperitoneal administration of $CFTR_{inh}$-172 (20 µg every 6 h), an inhibitor of cystic fibrosis transmembrane conductance regulator (CFTR), completely abolished the *V. cholerae*-induced intestinal fluid secretion. As analyzed by *ex vivo* measurement of intestinal electrical resistance and *in vivo* assay of fluorescein thiocyanate (FITC)-dextran trans-intestinal flux, *V. cholerae* infection had no effect on intestinal paracellular permeability. Furthermore, histological examination of *V. cholerae*-infected intestinal tissues illustrated edematous submucosa, congestion of small vessels and enhanced mucus secretion from goblet cells. This study established a new adult mouse model of *V. cholerae*-induced diarrhea, which could be useful for studying the pathogenesis of cholera diarrhea and for evaluating future therapeutics/cholera vaccines. In addition, our study confirmed the major role of CFTR in *V. cholerae*-induced intestinal fluid secretion.

Keywords: Animal model, cholera, *Vibrio cholerae*, CFTR, paracellular transport.

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O-13

PRL Stimulates Duodenal Calcium Transport in Male Rats

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Abstract

In female rats, prolactin (PRL) has been known to be a calcium-regulating hormone by enhancing maternal calcium absorption during pregnancy and lactation to restore bone mineralization. However, the role of PRL in intestinal calcium transport in male rats has been unknown. In the present study, the effect of PRL on the transepithelial calcium transport, and the signaling pathways of PRL in regulating calcium transport were demonstrated in the duodenums of male rats by Ussing chamber technique. The duodenums were exposed with various doses of PRL (50, 100, 200, 400, 600, 800, and 1,000 ng/ml) added directly in the chamber. PRL was found to stimulated intestinal calcium absorption in a biphasic dose-dependent fashion with the maximum effective dose of 200 ng/ml. The stimulatory effect of PRL on male duodenal calcium absorption was abolished by several inhibitors, including AG490 (JAK2 inhibitor), U0126 (MAPK inhibitor), LY294002 (PI3K inhibitor) and SU6656 (Src kinase inhibitor). The PRL-stimulated transcellular active calcium transport in male rats was totally abolished by transcellular calcium inhibitors, namely nifedipine (voltage-dependent L-type calcium channel inhibitor), vanadate and trifluoperazine ($PMCA_{1b}$ inhibitors), and KB-R7943 (NCX1 inhibitor). PRL also enhanced the paracellular passive calcium transport in the duodenums of male rats, and such the PRL effect was completely abolished by PI3K inhibitor. The present results thus indicated that PRL could stimulate duodenal calcium absorption in male rats via both active and passive mechanisms in a PI3K-dependent manner.

Keywords: Epithelial calcium transport, prolactin, Ussing chamber.

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P-01

Effects of *Kaempferia parviflora* Liquor on Male Fertility and Histopathology of Testes and Epididymis in Rats

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Abstract

Kaempferia parviflora (KP) has traditionally been used as KP liquor for enhancement of male sexual function. However, scientific information supporting this folklore claim is limited. Therefore, this study determined the effects of KP liquor on fertility and histopathology of testes and epididymis of male rats. Thirty-two Sprague-Dawley rats, aged 6 weeks old (200-230 g), were divided into 4 groups (8 rats/group). Group1: intact control. Group2: vehicle-treated control received 25% ethanol at 4 ml/kg/day per os (equivalent to 800 mg/kg/day). Groups 3 and 4: KP liquor-treated groups received ethanol extract of KP in 25% ethanol at 4 ml/kg/day per os at the doses of 200 and 1,000 mg/kg, respectively. At week-4 after treatment, male fertility was assessed by cohabiting each male with two pro-oestrus females. Successful mating was confirmed by the presence of spermatozoa in vaginal smear. A week later, male rats were sacrificed, and testes and epididymis were excised, weighed and prepared for histopathology studies. The percentage fertility was calculated from the ratio of number of fetuses to the number of corpora lutea x 100. Ethanol suppressed fertility from 84.4±4.0% in the intact control group to 71.7±4.3% (15.5% reduction). Treatments of ethanol extract of KP significantly reversed the fertility (P<0.05) back to the intact control level (80.0±3.1% and 85.0±3.1% in the 200 and 1,000 mg/kg KP-treated groups). Histopathology studies revealed that spermatogenesis was arrested in ethanol-treated group. On the other hand, different stages of spermatogenesis in the seminiferous tubules and spermatozoa-filled lumen were observed in the testes of intact control and KP treated groups suggesting normal spermatogenesis. Ethanol had small adverse histological effect to the epididymis. The present study provides experimental evidence for the beneficial effects of KP liquor on male sexual function claimed by folklore medicine.

Keywords: *Kaempferia parviflora*, male fertility, spermatogenesis, histopathology, testes, epididymis, rats.

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Acknowledgement: Supported by the Thailand Research Fund

P-02

Association of *MDR1* Polymorphism and Cervical Cancer in Contraceptive Pill Users

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Abstract

Cervical cancer is a serious public health problem in Thailand. The use of genetic risk factors to control cervical cancer among high-risk groups promises to be a key strategy for decreasing both incidence and mortality. The multidrug resistance1 gene (*MDR1*) plays an important role in regulating the expression and function of P-glycoprotein (P-gp), an efflux-pump on the cell membrane, to excrete toxins and drugs from cells. In humans, a nucleotide at position 3435 on exon 26 of *MDR1* is reportedly polymorphic, since it can substitute cytosine (C) with thymine base (T), yielding 3 genotypes: CC genotype (wild type), CT genotype (heterozygous type) and TT genotype (mutant type). Therefore, the purpose of this study was to investigate the association between *MDR1* (C3435T) polymorphism and the risk of cervical cancer among contraceptive pill users in northeastern Thailand. The 348 age-matched study subjects were divided into two groups, normal controls (174 persons) and cervical cancer patients (174 persons). Genomic DNA, which had been extracted from the peripheral blood leukocytes of each subject, was analyzed for *MDR1* (C3435T) polymorphism by polymerase chain reaction restriction fragment length polymorphism (PCR-RFLP) technique. It was found that individuals with the TT genotype were at 1.98-fold higher risk of cervical cancer than those with the CC genotype (OR=1.98, 95%Cl=1.08-3.65). Moreover, individuals with the TT genotype and taking oral contraceptive pills were at 2.74-fold higher risk of cervical cancer than those with the CC genotype (adjusted OR=2.70, 95%Cl=1.08-6.76). In conclusions, our results indicated that *MDR1* polymorphism is associated with cervical cancer risk among women who live in the northeastern Thailand and the TT genotype of *MDR1* (C3435T) polymorphism may be a useful genetic risk factor for screening for the women at high risk of cervical cancer. This can help physicians detect early-stage cases that are susceptible to successful treatment in the future.

Keywords: *MDR1*, contraceptive pill, cervical cancer.

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P-03

NSAID As an Inhibitor of CFTR-Mediated Chloride Secretion in Human Intestinal Epithelia and Potential Therapy of Cholera

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Abstract

Cholera is a life-threatening secretory diarrhea caused by cystic fibrosis transmembrane conductance regulator (CFTR)-mediated chloride secretion across intestinal epithelia. The present study aimed to investigate the effect of a non-steroidal anti-inflammatory drug (NSAID) on CFTR-mediated transepithelial chloride secretion in human intestinal epithelial (T84) cells and to evaluate its therapeutic potential in the treatment of cholera. Using short-circuit current analysis, it was found that NSAID inhibited CFTR-mediated transepithelial chloride secretion across monolayers of T84 cells with an IC_{50} of ~ 20 μM. Apical chloride current analysis in basolaterally permeabilized T84 cell monolayers demonstrated that NSAID inhibited CFTR-mediated chloride transport activated by forskolin, a cell-permeable cAMP analog CPT-cAMP and a CFTR activator genistein. In addition, potency of CFTR inhibition by NSAID was not affected by pretreatment with compound C, an AMP-activated protein kinase (AMPK) inhibitor, indicating that the mechanism of CFTR inhibition by NSAID does not involve AMPK. Surprisingly, NSAID failed to inhibit CFTR-mediated apical chloride efflux in Fisher rat thyroid cells stably transfected with wild-type human CFTR. Interestingly, intraperitoneal administration of NSAID significantly reduced the cholera toxin-induced intestinal fluid secretion in mouse closed-loop models. Our results have revealed an unprecedented action of NSAID as an inhibitor of CFTR-mediated chloride secretion in human intestinal epithelial cells and indicated that NSAID represents an inexpensive and effective therapeutic option to reduce intestinal fluid loss in cholera.

Keywords: Cholera, cystic fibrosis transmembrane conductance regulator (CFTR), NSAID.

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P-04

Physical Fitness Testing in Southern Thai Population During PSU Science Week 2008-2012

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Abstract

Department of Physiology, Faculty of Science, Prince of Songkla University (PSU) has performed academic services for nearby community including physical fitness testing since 2000. Our physical fitness testing has been served for southern Thai population during PSU Science Week or PSU Academic Week in August annually. This work aimed to report the collecting data of the physical fitness testing during 2008-2012 systemically. Physical fitness testing protocol was divided into to 6 steps; 1) registration with personal data and fill up information about personal health, illness, operation or accident history, 2) recording body weight and height for calculated body mass index (BMI), measuring waist and hip circumference for calculated waist to hip ratio, the waist to height ratio was determined to evaluate the risk of obesity, 3) measuring resting arterial blood pressure and resting heart rate, 4) measuring vital capacity for primary pulmonary function test, 5) determination of hand grip and leg muscle strength and 6) measuring submaximal oxygen uptake (VO_2 max) during 5-6 min cycling according to P.O. Astrand method to determine the subject aerobic endurance. Fitness scores were analysed and catagorized according to sex and age. Five groups of the age interval were designed including 17-19, 20-29, 30-39, 40-49 and over 50 years old group. Each subject fitness score was informed with the health improvement instructions. All data were collected during the year 2008-2012 and kept as preliminary database of southern Thai population health fitness scoring. These data would also reflect their quality of life, nutritional status, community medical services and exercise technology providing. It is also be the useful information for Thai government to direct the public health policy in the southern part of the country.

Keywords: BMI, waist-to-hip ratio, waist to height ratio, grip strength, leg strength, VO_2 max.

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P-05

Rice Bran Peptide Attenuates Hypertension and Oxidative Stress in High-Carbohydrate, High-Fat Diet-Induced Metabolic Syndrome in Rats

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Abstract

Rice bran (RB) is a by-product of the rice milling, which derives from the outer layer of the rice grain. Previous studies have demonstrated that peptide-derived from RB (RBP) contains large amounts of fiber and bioactive molecules with special interest as antioxidants. The aim of this study was to investigate the antihypertensive and antioxidative effects of RBP in a rat model of metabolic syndrome (MetS). Male Sprague Dawley rats (200-220 g) were randomly divided into three groups (n=6-8 each). One group was fed with standard chow diet. Two groups of rats were fed with a high-carbohydrate, high-fat diet (HCHF) with 15% fructose in the drinking water for 16 weeks. One group of HCHF was supplemented with 1% RBP in the food for the final 6 weeks of the treatment protocol. The HCHF-fed rats showed the symptoms of MetS, including hypertension, dyslipidemia, hyperglycemia and impaired glucose tolerance. Supplementation with RBP significantly reduced fasting blood glucose, heart rate and blood pressure ($P<0.05$). Plasma malondialdehyde and vascular superoxide production were also decreased in MetS rats fed with RBP. These results suggest the antihypertensive, antihyperglycemic and antioxidative effects of RBP in a rat model of MetS.

Keywords: Rice bran peptide, high-carbohydrate, high-fat diet, metabolic syndrome, oxidative stress.

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P-06

Mechanisms of Vasorelaxation Induced by Thai Rice Bran Protein Hydrolysate in Conduit and Resistance Vessels Isolated from Metabolic Syndrome Rats

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Abstract

Our previous study was first demonstrated the vasorelaxant effect of Thai rice bran protein hydrolysate (TRBPH) in normal rats. This study characterized the effect of TRBPH on vascular tone and its mechanism in thoracic aortae and mesenteric vascular beds isolated from metabolic syndrome rats induced by high-fat, high-carbohydrate (HFHC) diet. Male Spraque-Dawley rats were fed with HFHC diet and 15% fructose in drinking water for 16 weeks to induce insulin resistance conditions. Fasting blood glucose (FBG), oral glucose tolerance test (OGTT), lipid profiles, and systolic blood pressure were evaluated. The vasorelaxation of TRBPH and its mechanism were assessed in pre-constricted aortic rings and mesenteric vascular beds. Results showed that rats received HFHC diet had mild hypertension (SP = 142.42±0.91 mmHg), dyslipidemia, an increase in FBG, and impairment of OGTT, indicating metabolic syndrome in these rats. TRBPH at dose 0.001-1 mg/ml produced vasodilation by dose dependent in endothelium-intact aortic rings and mesenteric vascular beds. Removal of endothelium abolished the vasorelaxant effect of TRBPH in aortic rings and partially inhibited the vasorelaxation in mesenteric vascular beds. In addition, there was no TRBPH relaxation in endothelium-intact aortic rings in the presence of L-NAME. While pretreatment with capsaicin, depleting calcitonin gene-related peptide (CGRP)ergic-nerve, in the presence of L-NAME abolished the vasorelaxation response to TRBPH in endothelium-intact mesenteric vascular beds. In conclusion, TRBPH vasorelaxation in the aortic rings and mesenteric vascular beds of metabolic syndrome rats was endothelium-dependent through NO relaxation pathway. In addition to endothelium involvement, vasorelaxation induced by TRBPH in resistance vessels was also partially attributed by TRPV1 receptor located at perivascular nerves.

Keywords: Thai rice bran protein hydrolysate, vasorelaxation, metabolic syndrome rats.

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P-07

Association Between Arterial Stiffness and Metabolic Risk Factors

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Abstract

Metabolic syndrome (MetS) is an increasing epidemiologic challenge and a cluster of cardiovascular risk. MetS is a major health problem with high prevalence in Thailand. Increased arterial stiffness is a cardiovascular outcome of MetS. Pulse wave velocity (PWV) is a good marker for evaluating arterial stiffness. The purpose of this study was to assess the arterial stiffness by measuring PWV in Thai subjects with MetS. A total of 208 subjects, aged 35-75 years, 135 with and 73 without MetS according to IDF and NCEP-ATPIII criteria were included in this study. Brachial-ankle pulse wave velocity (baPWV) and aortic ankle pulse wave velocity (aaPWV) were measured. Routine anthropometric and serologic data were collected. The baPWV and aaPWV were significantly increased in MetS subjects when compared with non MetS (P<0.01). Both of PWVs were associated with age, systolic blood pressure and waist circumference (P<0.05). PWV, especially aaPWV increased progressively with increasing number of MetS risk factors (r=0.202, P<0.01). Results in this study suggest that MetS risk factors are strongly associated with increased arterial stiffness in a Thai population with MetS.

Keywords: Metabolic syndrome, pulse wave velocity, arterial stiffness.

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P-08

Asiatic Acid Alleviates Hemodynamic Alterations and Vascular Dysfunction in L-NAME-Induced Hypertensive Rats

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Abstract

It is well established that asiatic acid, a natural triterpenoid compound derived from *Centella asiatica*, exhibited biological effects including, antiinflammation, wound healing, antioxidant and antihyperglycemia. In the present study, we assessed the effect of asiatic acid on hemodynamic changes and vascular responses in N^{ω}-nitro-L-arginine-methyl ester (L-NAME)-induced hypertensive rats. Male Sprague-Dawley rats were fed with L-NAME (40 mg/kg/day) in drinking water daily for 5 weeks while normal control rats received distilled water. Hypertensive rats were intragastrically administered with asiatic acid (20 mg/kg/day), L-arginine (100 mg/kg/day) or vehicle in the last 2 weeks of the experimental period. Systolic blood pressure was monitored using a tail cuff method once a week. At the end of the experimental day, mean arterial blood pressure (MAP), heart rate (HR), hindlimb blood flow (HBF), and vascular responsiveness by acetylcholine (ACh), sodium nitroprusside (SNP), and phenylephrine (Phe) were measured under anaesthetic conditions. Hindlimb vascular resistance (HVR) was calculated. Results showed that rats treated with L-NAME for 5 weeks had high blood pressure (MAP=173.29±6.3 mmHg *vs* 91.63±1.9 mmHg), heart rate (413.01±9.9 beat/min *vs* 346.88±7.1 beat/min), HVR (40.8±2.3 mmHg/min/100 g/ml *vs* 12.8±0.5 mmHg/min/100 g/ml) and low HBF (4.3±2.3 ml/100g tissue/min *vs* 7.3±0.3 ml/100 g tissue/min) compared to those of control ($P<0.05$). In addition, vascular responses to ACh (49.7±4.5 % *vs* 65.3±1.1 %) and Phe (34.2±5.1 % *vs* 64.9±3.2 %) were significantly blunted in hypertensive rats ($P<0.05$). However, supplementation of asiatic acid significantly reduced MAP about 19.2% which was associated with reductions in HR and HVR ($P<0.05$). The impairment of vascular responses to ACh and Phe was restored in hypertensive rats treated with asiatic acid ($P<0.05$). Furthermore, treatment with L-arginine also exhibited antihypertensive effect which related to improvement of hemodynamic parameters and vascular responses in hypertensive rats. These results suggest that asiatic acid had anti-hypertensive effects in rats treated with L-NAME, which was associated with improving of hemodynamic alterations and vascular function.

Keywords: Asiatic acid, vascular dysfuction, hypertension.

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P-09

Anti-Oxidative Stress Activity of Dried Banana Syrup in Mice

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Abstract

Dried banana syrup is syrup from pressing sun soaked or dried bananas. This processing can increase a value added of dried banana. Bananas are a great source of good nutrition such as vitamins, minerals, sugars (like fructose, glucose and oligosaccharide) as well as tryptophan and tyrosine. There were evidences that the consumption of banana reduced the plasma oxidative stress and enhanced the resistance to oxidative modification of LDL in healthy individuals, and banana flavonoids showed antioxidant activity. The present study aimed to determine the effects of dried banana syrup on weight gain, food intake, plasma glucose level, lipid profiles and lipid peroxidation in mice. ICR mice were divided into 3 groups and treated for 3 months with normal saline, honey and dried banana syrup. These results showed that dried banana syrup and honey had no effect on weight gain, food intake, plasma glucose, lipid profiles whereas it was significantly decreased lipid peroxidation as indicated by lower malondialdehyde than that control group. These results suggest that there do appear to be health benefits associated with consuming dried banana syrup for a prolonged period of time.

Keywords: Dried banana syrup, anti-oxidative stress activity, lipid profile, plasma glucose.

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P-10

Development of Freshly Dissociated Human Umbilical Vein Endothelial Cell Preparation for Electrophysiological Studies

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Abstract

Alterations of ion channel expression in cultured cells are well known and hamper the generalization of electrophysiological data obtained from primary culture cells. The present study aimed to develop a freshly dissociated human umbilical vein endothelial cells (HUVECs) preparation, in order to obtain cells that have been least modified from in situ conditions (compared to primary culture cells) for electrophysiological studies. Human umbilical cords were randomly collected after normal delivery from normal pregnant women who were admitted at the Department of Obstetrics and Gynecology, Siriraj Hospital. These subjects must not have had history of any disease prior to and during pregnancy, as ascertained by an obstetrician. The umbilical cords were cleaned with normal saline to remove all traces of blood. Then endothelial cells were isolated under sterile conditions by using 0.2% collagenase A solution. Isolated cells were grown on gelatin-coated glass coverslips placed in petri dishes containing M199 medium, 10% FBS, 50 i.u./ml penicillin, 50 μg/ml streptomycin and, with 5% CO_2 at 37°C. After 4 hours of isolation, cells were single, round-shaped and well attached to the coverslips. Immunofluorescent staining was used to demonstrate the presence of von Willebrand factor and vascular endothelial growth factor receptor-2 (VEGFR-2, KDR, flk1), the specific markers of endothelial cells, to assure the purification of HUVECs. On the other hand, α-actin of vascular smooth muscle cells (VSMCs) was not detected. Therefore, we have formulated the optimal conditions for freshly isolating HUVECs. The cells obtained consisted of endothelial cells with virtually no contamination from VSMCs in this preparation.

Keywords: HUVECs, fresh dissociation, immunofluorescent staining.

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P-11

Reduced Neovascularization in Aged Rats: A Study Using Lipopolysaccharide-Induced Inflammation

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Abstract

Aged tissues are associated with reduced capillary density and delayed neovascularization. The aging environment is characterized by increased oxidative stress, which damages macromolecules, and thereby causes apoptosis of endothelial cells. Lipopolysaccharide (LPS) has been shown to have angiogenic property. Therefore, the objective of this study was to examine whether neovascularization after LPS-induced inflammation in aged rats is decreased when compared to adult rats. Male Wistar rats were divided into four groups: adult (aged 6-8 months), aged (aged 22-24 months), adult + LPS, and aged + LPS. Blood perfusion, capillary vascularity, plasma and tissue MDA, and tissue VEGF levels were measured. Plasma MDA in aged group was higher than in adult group ($P < 0.05$). Blood perfusion and capillary vascularity (%CV) in aged group were lower than in adult group ($P < 0.05$). Blood perfusion and %CV in aged + LPS group were significantly lower than those in adult + LPS group. Aged rats have reduced capillary vascularity when compared to adult rats. LPS could induce neovascularization in both adult and aged rats. However, aged rats have reduced neovascularization after LPS pre-treatment when compared to adult + LPS rats.

Keywords: Aging, neovascularization, lipopolysaccharide, oxidative stress.

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P-12

Lipidemic, Glycemic and Organ Protective Actions of Tea Seed Oil in Rats Fed With High Fat and High Carbohydrate Diet

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Abstract

The present study was conducted to investigate the lipidemic, glycemic and organ protective effects of tea seed oil (TSO) in rats fed with high fat and high carbohydrate (HFHC) diet. Three groups of male Wistar rats were used including normal control group, group fed with HFHC diet (cholesterol+lard+fructose) for three months, and HFHC group treated with TSO. At the end of the experiment, serum lipid profile, blood glucose, oral glucose tolerance test (OGTT), serum AST, ALT, LDH, CK-MB, creatinine and BUN were determined. Individual fatty acids containing in TSO was assayed by Gas chromatography. The results showed that oleic acid was the primary fatty acid in TSO (83.36%). HFHC diet increased serum lipid profile and atherogenic index (AI). The high serum levels of lipid profile and AI were decreased in HFHC rats treated with TSO. The elevation of area under the curve of OGTT was alleviated by TSO. TSO also normalized the high serum levels of AST, ALT, LDH, CK-MB, creatinine and BUN. It can be concluded that TSO was able to decrease the high serum levels of lipid profile, and high blood glucose level of OGTT in rats fed with HFHC, indicating its therapeutic potency to prevent atherosclerosis and hyperglycemia. It also protects liver, heart and kidney in HFHC group. Oleic acid containing in TSO might be responsible for these activities.

Keywords: Tea seed oil, high fat diet, liver function, cardiac function, fructose.

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P-13

Preferences and Opinions of Second Year Medical Students on Learning Activities in the Organ System Block

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Abstract

Many medical schools have adopted a systems based preclinical curriculum rather than discipline orientated. This is believed to encourage integrated teaching of preclinical subjects and its linkage to the clinical atmosphere early in medical training. A questionnaire following the respiratory system-I block sought the views of second year medical students on improving their learning activities; the return rate was 26%. Apart from normal lectures and laboratories, the majority of respondents approved of *the clinical correlation* lecture (100%); *the concept map writing* (91%); *peer-assessment* for group activities (85%); *problem-based learning* (PBL)(83%); *pre-learning exercises* (83%); and *the lab conference* (80%). All students saw the significance and linkage of preclinical knowledge to the clinic and desired to learn more from lectures. Concept mapping after PBL sessions enabled them to integrate their thoughts, reason causes-effect relations and draw conclusions from knowledge extracted from the discussion and PBL self-learning on one page. Peer-assessments provided each student a good feedback on their group performance and reflected their values such as responsibility, communicative skill, tactfulness, punctuality, etc. PBL improved their quest for knowledge and team work. The pre-learning exercises introduced technical terms and the content framework which stimulated review or pre-reading the learning content which augmented their understanding during lectures. The lab conference bridged the lecture and laboratory experiences. Students also suggested a wrap-up session for each of PBL, lab conference and concept mapping session. These findings suggest various learning processes on which the student reflected on their opinions. The teaching approach that relates content to clinical scenarios stimulates learning and may ultimately be a cost-effective way underpinning evidence-based medicine.

Keywords: Clinical correlation, concept map, PBL, pre-learning exercise, evidence-based medicine.

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P-14

Autonomic Dysfunction Following Acute Stroke: A Preliminary Study

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Abstract

Autonomic nervous system (ANS) disturbances or autonomic dysfunction often occurs after cerebrovascular events. Many studies proposed that stroke in different populations may vary in characteristics and associate with autonomic dysfunction. Therefore, we aimed to determine the prevalence and severity of autonomic dysfunction in Thai acute stroke patients. Twelve consecutive acute stroke patients who were admitted into the Stroke Unit, Siriraj Hospital, were subjected to cardiovascular autonomic function tests, as well as standard physical examination, investigation and treatment. Cardiovascular autonomic function tests consisted of 1) heart rate variability (HRV) analysis of 5-min resting electrocardiogram recorded by PowerLab acquisition system, 2) baroreflex sensitivity measurement by using Finapres equipment, the continuous non-invasive beat-to-beat blood pressure recorder, and 3) Ewing test, the cardiovascular autonomic function tests. Ten male and two female acute stroke patients, aging 71±15 years, were recruited, with NIHSS score of 9±8. Nine patients had hypertension, while dyslipidemia and diabetes mellitus were found in eight and three patients, respectively. The mean BP was 144±23/85±11 mmHg. Heart rate variability parameters were: average SDNN 28.7±19.9 ms, average RMSSD 18.7±13.6 ms, average pNN50 4.6±8.2%, average low frequency component (LF) 232.1±401.2 ms^2 (LFnu 55.2±21.0), average high frequency component (HF) 141.2±188.9 ms^2 (HFnu 33.5±18.1) and average LF/HF ratio 2.3±1.4. The mean baroreflex sensitivity was 7.8±6.3 ms/mmHg. However, the Ewing test could be performed in only one patient; the other eleven patients had poor cooperation due to their conditions. These preliminary data suggested possible autonomic dysfunction in acute stroke patients and showed that it may not be appropriate to perform Ewing test in these patients.

Keywords: Stroke, cardiovascular autonomic dysfunction.

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Effects of Lutein and Sericin-Derived Oligopeptides Extracted from Yellow Silk Cocoons (*Bombyx mori*) on Vascular Function in Aged Rats

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Abstract

This study was aimed to investigate the effects of silk lutein (SL) and sericin-derived oligopeptides (SDO) extracted from yellow silk cocoons (*Bombyx mori*) on isolated aorta of aged rats. Male Sprague-Dawley rats were randomly separated into 4 groups. Each group was orally administered with a single dose of propylene glycol (negative control), 10 mg/kg lutein (SL), 100 mg/kg SDO, or 1 mg/kg Donepezil (positive control) once a day for 8 weeks. At the end of experiments, rat's thoracic aorta was removed and suspended in an organ bath filled with oxygenated physiological balance-salt solution (PBSS). The vascular tone was monitored using an isometric force transducer coupled to a bridge amplifier and a PowerLab unit, and displayed on a computer using ChartPro (v. 5.4). Prior to all tests, the aortic ring was induced to reach a maximal contraction with 10 µM phenylephrine. Then, either 0.0001-10 µM acetylcholine (ACh) or 0.00001-10 µM sodium nitroprusside (SNP) were added into the PBSS in order to produce graded magnitudes of vascular relaxation. We found that the isolated aortic rings of SL or SDO treated group showed greater magnitudes of relaxation mediated by endothelial and smooth muscle cell when compared to the control groups. These preliminary results suggest that the actions of SL and SDO on vascular function are mediated by both endothelial cell and smooth muscle pathways and that these silk extracts may help to improve vascular dysfunction in aging rats. Currently, we are conducting an additional experiment in order to elucidate the underlying mechanism(s) of these two compounds.

Keywords: *Bombyx mori*, lutein, sericin-derived oligopeptide, vascular function.

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Neuroprotective Effects of the *Asparagus racemosus* Root Extract on Ovariectomized Rats

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Abstract

Estrogen decline in menopausal women may impair the cognitive function. This change can be ameliorated by estrogen replacement therapy (ERT). However, ERT is often overshadowed by the serious side effects of estrogen use in menopause women. *Asparagus racemosus* (AR) is well known for its phytoestrogenic properties while neuroprotective effects of AR in ovariectomized model are unknown. This study aimed to investigate effects of AR root extract on serum estradiol level, learning and memory, and neuronal viability in hippocampus and medial prefrontal cortex (mPFC) of ovariectomized rats by using electrochemiluminescence immunoassay, the novel object recognition test and histological analysis, respectively. Adult female Wistar rats were divided into five groups and gavaged for 90 days with vehicle (propylene glycol) for sham and OVX groups and another 3 groups of OVX rat were gavaged with 100 or 1000 mg/kg B.W./day of AR root extract or 0.1 mg/kg B.W./day of 17α-ethynylestradiol (EE), respectively. There was a significant decrease in recognition index of the OVX rats and AR root extract could reverse this effect. The serum estradiol level was significantly decreased in OVX group whereas AR root extract did not statistically change from that demonstrated by OVX group. The OVX rats showed a marked decrease in the number of neurons in hippcampus and mPFC. AR root extract and EE could reduce the neuronal loss in hippocampus and mPFC. The results obtained suggest that AR could improve cognitive ability in ovariectomized model which associated with increase neuronal viability of hippocampus and mPFC. AR may be a beneficial agent for prevention cognitive decline induced by ovariectomy.

Keywords: *Asparagus racemosus*, estrogen, learning, memory, ovariectomy.

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Effects of Sericin-Derived Oligopeptides Extracted from *Bombyx mori* Cocoons on Locomotor Activity and Cognitive Function in Mice

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Abstract
Sericin-derived oligopeptides (SDO) are short-chain and small molecular weight peptides extracted from coating materials of yellow silk cocoon (*Bombyx mori*). The present study aimed to study the effects of subchronic treatment of SDO on brain functions. For the locomotor activity test, adult male mice were randomly divided into 5 groups: negative control group received distill water, SDO-1 group received 1 mg/kg SDO, SDO-10 group received 10 mg/kg SDO, SDO-100 group received 100 mg/kg SDO, and positive control group received 30 mg/kg i.p. of pseudoephridine. Animal's locomotive behavior was continuously monitored at a 10-min epoch using an infrared sensor behavioral box. About 30 min after SDO administration, SDO-1 and SDO-10 groups showed a small, yet significant, increase in their locomotor activities when compared to negative control group. For the cognitive function tests, the mice were divided into 5 groups: 1) sham+vehicle group which was fed with 0.5 ml distill water (vehicle) once a day for 7 weeks and received an intracerebroventricular (i.c.v.) injection with normal saline at 4th week, and groups 2)-4) are 3 AB+SDO groups (fed with 1, 10, or 100 mg/kg/day SDO for 7 weeks), and 5) AB+Don group (fed with 1 mg/kg/day donepezil for 7 weeks) and then received an i.c.v. injection with amyloid beta peptides at 4th week. We found that the mice treated with SDO showed the better cognitive indices for both spatial memory and object recognition tests when compared to AB+vehicle group. These preliminary results suggest that SDO has a mild stimulant action and may help to improve the memory deficits caused by beta amyloid peptide plagues in the brain.

Keywords: *Bombyx mori*, sericin-derived oligopeptide, locomotor, memory.

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Assessment of Arterial Stiffness in Elderly Thai

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Abstract

Most studies suggested that augmentation index (AI) and pulse wave velocity (PWV) increased linearly with age, and used these two parameters as the indicators of arterial stiffness. The objective of this study was to evaluate the arterial stiffness by measuring the AI and PWV in elderly Thai. A total number of 216 Thai (96 men, 120 women) aged from 50 to 70 were recruited in this study. The augmentation pressure (AP), AI, brachial ankle PWV (baPWV) and major cardiovascular risk factors of the participants were measured. The AI values have been normalized at a heart rate of 75 beat/min and expressed as AI@HR75. AI@HR75 and baPWV trend to increase with age. The female subjects had significantly greater mean values of AP and AI@HR75 than those male subjects (P<0.001). In contrary, the baPWV was significantly higher in male than female subjects (P<0.001). These data suggest that AI and PWV do not always show a linear correlation, but rather they are different affected by ageing. Changes in AI were more prominent in younger individuals, whereas the changes in PWV were more marked in those older than 50 years. This study is reported for the first time that increased AI@HR75 and baPWV are found in elderly Thai. Further investigation is needed in large population at different age and gender. Information about arterial stiffness in Thai population will be beneficial to the public health interventions as arterial stiffness is a powerful predictor of cardiovascular disease.

Keywords: Arterial stiffness, Augmentation index, brachial ankle pulse wave velocity, ageing.

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Hippocampal EEG Biomarkers of Novel Exploration

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Abstract

The hippocampus plays critical roles in episodic memory, spatial/contextual processing, and novelty detection. Hippocampal novelty detection requires the encoding of new information into memory process. Hippocampal electrical activity produces several oscillatory patterns. Previously, theta-related activity (8-12 Hz) in the hippocampal formation supports spatial memory and novelty detection. In addition, beta oscillations (15-30 Hz) during the learning in novel environments was observed. However, the relationship between hippocampal oscillations and novel processing remains unclear. This study aims to identify hippocampal electrical biomarkers that represent neural activity involving novel processing. Male Swiss albino mice (35-40 g) were implanted with Teflon coted silver wire electrode into dorsal hippocampus (CA1 region). Electroencephalography (EEG) was used to record hippocampal field potential while animals were placed in ~6 cm wide circular track chamber with difference in floor and wall textures. Fast Fourier Transform (FFT) algorithm was used to analyze the EEG signal. The results revealed the sudden increase in power of 2 frequency ranges which were 7–10 Hz and 15–20 Hz simultaneously with explorative behaviors as soon as the animals entered the novel space. These findings were found to be progressively decreased by time especially during re-exposures. This study demonstrated the emerging of hippocampal EEG components during novelty exploration. However, further studies are needed to investigate whether these oscillations specifically reflect novelty recognition process and not secondary to exploratory behaviors.

Keywords: Electroencephalography, hippocampus, novel environment.

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Effects of Morelloflavone from *Garcinia dulcis* on Vasorelaxation of Isolated Rat Thoracic Aorta

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Abstract

Morelloflavone is a flavonoid isolated from *Garcinia dulcis*, Kurz., an Asian medicinal plant used to treat a sore throat, scurvy and cough. Previous *in vitro* experiments have shown that morelloflavone possesses anti-inflammatory, anti-oxidant and anti-bacterial properties while other pharmacological effects have never been researched. This work aimed to investigate the action and mechanism(s) of morelloflavone from *G. dulcis* on relaxation of isolated rat thoracic aorta precontracted with norepinephrine (1 g resting tension). Cumulative addition of 10^{-9}-10^{-5} M morelloflavone significantly relaxed the precontracted aortic rings in a dose-dependent manner with a maximal relaxation of 77.5±5.4% and an EC50 of approximately 10^{-7} M. In denuded rings, this effect was abolished. Pre-incubation of endothelium-intact aortic rings with 10^{-6} M L-NOARG, a nitric oxide synthase inhibitor, significantly abolished morelloflavone-induced vasorelaxation, while the cyclooxygenase inhibitor indomethacin (10^{-6} M) had no effect. Morevover, either 10^{-5} M glibenclamide (an ATP-sensitive K^+ channel inhibitor) or 10^{-3} M TEA (a non-selective Ca^{2+}-activated K^+ channel blocker) could partially inhibited morelloflavone-induced vasorelaxation. Therefore, the vasorelaxation mechanisms of morelloflavone was endothelium-dependent which involved nitric oxide signaling pathway and, partly, ATP-sensitive and Ca^{2+}-activated K^+ channels.

Keywords: Morelloflavone, flavonoid, thoracic aortic ring.

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