



Faculty of Science, Kasetsart University, Bangkok Thailand

I-KUSTARS 2017

1 - 3 June, 2017

The International Kasetsart University
Science and Technology
Annual Research Symposium



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Dear Colleagues

On behalf of the organizing committee of the International Kasetsart University Science and Technology Annual Research Symposium 2017 (I-KUSTARS 2017), it is my great pleasure to extend a warm welcome to you to participate in this Symposium which is one of the significant occasions to highlight our new vision, “Excellent in Natural Science in ASEAN” by 2022.



I-KUSTARS 2017@Kasetsart is an exciting conference in science to provide opportunities for senior students to present their research work in a dedicated forum at the symposium. Leading by Plenary and invited speakers who pioneered their research field and made significant contributions in the area. This symposium is also a platform to strengthen current networks and to establish new collaborative links within Asian community and with the rest of the world. The scientific program comprises sessions that illustrate the relevance and value of modern science and technology and innovation.

We are looking forward to giving a warm welcome to you, and colleagues at I-KUSTARS 2017. We hope that you will find the Symposium both enjoyable and valuable. We thank you in advance for participating and contributing to the success of the event to mirror “*Knowledge of the Land*” for sustainable world.

With best wishes

Supa Hannongbua
Dean of Faculty of Science

“SciKU”

S: Sustainability

C: Creativity

I: Integrity

K: Knowledge

U: Unity

VISION: Excellence in Natural Science in ASEAN by 2022

VALUE: Create Science for All





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PROGRAM SCHEDULE

June 1, 2017	
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09.30 A.M. - 10.00 A.M.	Opening Ceremony
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1:00 P.M. - 4:00 P.M.	Poster Session (Ground Fl., Davi Yannasugondha Bldg.)
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PLENARY SESSION





Xing-Ming Zhao

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Xing-Ming Zhao obtained his PhD from University of Science and Technology of China and has since conducted research as a member of Hongkong Baptist University, The University of Tokyo, Institute of Systems Biology at Shanghai University and The European Molecular Biology Laboratory. Today, he is a Professor in the department of Computer Science, School of Electronics and Information Engineering, Tongji University, China. His research has focused on the construction and analysis of molecular networks, including protein - protein interaction network and gene regulatory networks, the recognition of disease associated molecular pathways and gene modules and drug-protein interaction, drug - drug interactions and the prediction of drug combination.





Network medicine: Understanding diseases and designing drugs based on molecular networks

Xing-Ming Zhao

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The biological systems can be described by molecular networks. Accordingly, the aberrant functions of the system that lead to diseases can be detected with molecular networks. With the accumulation of huge amount of omics data, it is becoming possible to construct molecular networks in an accurate way, which in turn can help detect the disease genes and aberrant pathways. In this talk, I'll present our recent works on algorithms and models for identifying disease genes from molecular networks. In addition, I'll showcase our works on drug developments based on molecular networks.

Keywords: bioinformatics research, systems biology, disease association, combination drug design

References:

- XJ Zhang, KQ Liu, ZP Liu, B Duval, JM Richer, XM Zhao, JK Hao, and LN Chen. *NARROMI: a noise and redundancy reduction technique improves accuracy of gene regulatory network inference*. *Bioinformatics* (2013) 29 (1): 106-113.
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Dehydrating Paddy by Solar Thermal Energy using a Flat Plate Solar Collector

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Thailand always receives solar energy throughout the year because it is located near the equator and has the solar zenith angle all the year about 17.58 degree. In this experiment, we designed and made a flat plate solar collector in order to convert solar energy into thermal energy for reducing moisture in the paddy. The latitude of Bangkok, 13 degrees 45 minutes north, is used. The experiment ran between 10:00 a.m. - 2:00 p.m. daily with the average declination angle is 9.54 degree for 4 days consecutively. The result shows that the flat plate solar collector can heat the temperature of the evacuated air up to 66.80 degree Celsius and can reduce the wet basis moisture percentage of paddy up to 9.21%

Keywords: flat plate solar collector, reduce moisture paddy, wet basis moisture percentage, solar energy





Portable Paddy Rice Moisture Content Meter

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A portable paddy rice moisture content meter (PRMM) is useful for the indication of the amount of water content of a given paddy sample. This information is especially useful to the farmers who need to measure the moisture content of paddy rice. This project, we developed a digital PRMM based on the 555 integrated circuit timer as a major component. The 555 timer was configured with the probes connected to the paddy rice container indicate the capacitance of the paddy rice. The digital PRMM was calibrated and the reading was displayed on a liquid crystal 20 x 4 LCD display panel. Paddy rice was packed in a 450 cm³ cylindrical plastic container with the diameter of 10 cm and 10 cm height. Weight of each sample of paddy rice was approximately 270 g. The moisture content range of the paddy rice was 6 % to 26 % wet basis. The PRMM was developed for Thai farmers.

Keywords: paddy rice, moisture, 555 timer





Bird Community in Different Landscape Structures: Lesson from Urban Areas in Bogor, Indonesia

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The presence of birds in an urban habitat, mainly diversity and abundance, has been known to be affected by habitat conditions and the landscape structures of the urban area. Birds have different responses to landscape modification, depends on its adaptation to the environment. The purpose of this study was to identify bird communities in several landscape types (including patch sizes, edge characteristics) and to analyze the landscape structures that affects bird communities.

The study was conducted from April to June 2016 at 29 landscapes in the city of Bogor and its surrounding. The landscapes was categorized into four types based on their fragmentation stages: intact, variegated, fragmented and relictual ($n = 7$ or 8). The bird survey was conducted using a standard point count method. Species richness, diversity (Shannon-Wiener, H'), and evenness (Brillouin evenness index, E) were calculated, as well as community similarity (using Bray-Curtis Index of Similarity, IS). The landscape structures were quantified by using 16 variables, and measured by using ArcView patch analyst. The relationship between landscape structures and bird communities was analyzed by using Pearson correlation test with 95% confidence level.

Total 8,967 individuals belonging into 77 species and 36 families were observed, with H' 3.123 and E 0.298. Similarity analysis of the bird community suggested that intact landscapes were clustered separately (IS 0.63), while other three types were clustered together. The correlation analysis revealed that the species composition of bird communities in each landscape types was influenced by various landscape structures. Four variables (i.e. mean shape index, number of patches, median patch size, mean patch edge) significantly affected the bird diversity. The number of species were positively affected by total edge and edge density.

Keywords: avian diversity, intact landscape, landscape ecology, patch analysis, urban fragmentation





Acoustic Communication Variation of *Leptophryne cruentata*

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For the last decades, Java has been highly degraded by human activities that led to the fragmentation of the remaining forests which mostly retained as protected areas. The geographical barriers between each remaining forest patch can lead to the isolation of local populations and pushed species to adapt to its environment. Many amphibian species are negatively affected by loss of habitat. Acoustic communication in frog is an important element in the intra-sexual selection and can be the best model for studying the evolution of acoustic signals due to geographic constraints. The variation of frog sounds is influenced by local environmental factors and may be critical for geographic variation in sexual traits. The main goal of our study was to provide a detailed description of the spectral and temporal parameters of the calls of *Leptophryne cruentata*.

Research is conducted at three mountains known as the last remaining habitat of *Leptophryne cruentata*: the Mount Gede Pangrango, Ciremai and Slamet from February to May 2017. Calls were recorded using a digital audio recorder H4N ZOOM series. During each recording session, we measured air temperature, SVL, and weight. Acoustic analyses were performed in Raven Pro 64 1.4 for Windows (Cornell Lab of Ornithology). We analyzed frequency range, peak of dominant frequency, call duration, number of pulses per note, and pulse rate. We calculated the variation in quantitative acoustic variables through the coefficient of variation (CV; SD/mean) for both the among-males and within-males level. By analyzing temporal and spectral parameters of the calls, we report that *Leptophryne cruentata* has two type of tone: pure tone and pulse tone. Pooling the available data, we can conclude that call traits (call structure, dominant frequency and call duration) of *Leptophryne cruentata* recorded from different geographic locations were highly variable. This may be caused by some proximate factors such as different body sizes and air temperatures. The duration of the advertisement call of *Leptophryne cruentata* showed a positive correlation with air temperature. However, SVL data from each population was not yet examined and it was thus unable to examine the relationship between call traits. Preliminary data from Mount Gede pangrango and Ciremai showed that each individual from different population has a distinct significant differences (e. g., call structure, dominant frequency, call duration, pulse/call rate). Further work (for example, molecular survey) should help us better understand why the variations occurred in the populations with large distribution areas.

Keywords: frog call, geographic variation, *Leptophryne cruentata*





Effects of flower nutritional resources on the longevity and parasitism of *Cotesia vestalis* (Hymenoptera: Braconidae) on *Plutella xylostella*

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Parasitoid is known to play an important role to control insect pest and to reduce negative-impact of chemical pesticides in crops. In Viet Nam, *Cotesia vestalis* is a native species and has been demonstrated its applicability as natural enemy. Additionally, many adult parasitoids require food resources such as nectar, pollen to optimize their life histories in the field. We investigated the potential effects of three flowering plants, yellow cosmos (*Cosmos sulphureus*), shrub verbena (*Lantana camara*), and creeping-oxeye (*Sphagneticola trilobata*), and different honey solutions concentration 100%, 30%, 5%, and water (control) on the longevity of *Cotesia vestalis*, which is an important solitary parasitoid of *Plutella xylostella*. Four females parasitoid were used for each treatment under laboratory conditions. The results showed that 100% and 30% concentration of honey yielded the highest longevity of *Cotesia vestalis* at $5,00 \pm 0,25$ days for male and $5,75 \pm 0,50$ days for female; $4,88 \pm 0,38$ days for male and $5,63 \pm 0,38$ for female compared to water as a control, respectively. Moreover, nutrition resources from yellow cosmos (*Cosmos sulphureus*) significantly increased the longevity of *C. vestalis* in comparison with that in the water and other flowers ($P < 0.05$), with $4,13 \pm 0,19$ d for male, $4,25 \pm 0,25$ d for female. Then, we tested the parasitism efficiency of *C. vestalis* on 10 larvae of diamondback moth *Plutella xylostella* at three different food resources: 5% honey, yellow cosmos flower (*Cosmos sulphureus*) and water as control. The larvae of *P. xylostella* were replaced at every 24h for parasitism until the parasitoid died. The number of parasitized larvae was highest at the first day of trial and there is no significant difference among the food resource treatments, then it was significantly reduced after the second days. These results imply that food resources from flowers can benefit to parasitoid in the field.

Keywords: *Cotesia vestalis*, longevity, yellow cosmos (*Cosmos sulphureus*), nectar, honey solution





Endophytic bacteria isolated from elephant grass (*Pennisetum purpureum*) promote biological control ability of brown plant hopper *Nilaparvata lugens*

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Currently, biological control of brown plant hopper, *Nilaparvata lugens* considered as a threat to rice production in Asia, plays an important role in reducing yield losses and enhancing rice quality. In this study, 25 endophytic bacteria were isolated from rhizome, stem and leaf of the elephant grass (*Pennisetum purpureum*). The screening of bacterial virulence in *Nilaparvata lugens* was conducted with 25 bacterial isolates at 10^8 CFU/ml concentration and distilled water was used as a control. Five adults of *Nilaparvata lugens* were released into a three 20 days-old rice seedlings plastic cup that was covered by plastic tube and muslin cloth. Each endophytic bacterial isolate was tested with totally 20 adults of *Nilaparvata lugens*. The number of living and dead brown plant hoppers was recorded and the mortality rate was analyzed by using Abbott's formula. The results indicated that three endophytic bacterial isolates including VBL1, VBT1, VBT5 showed the highest biological control of *Nilaparvata lugens* at the mortality rates of 46.95%, 55.02% and 55.02% respectively after 8 days of screening and gave significant difference compared to others isolates ($P < 0.05$). Additionally, the screening of insecticidal activity of three bacterial isolates was conducted at different concentrations (10^6 , 10^7 , 10^8 CFU/ mL) and we found that the highest mortality rate in this experiment was significantly observed at 10^8 CFU/ mL (63.83%) after 10 days trial ($P < 0.05$). The results obtained here indicated that the identification of three endophytic bacterial isolates (VBL1, VBT1, VBT5) showing significant insecticidal activities towards *Nilaparvata lugens*, promises a significant biological approach in controlling the threat of brown plant hoppers in rice fields.

Keywords: Biological control, insecticidal activity, endophytic bacteria, *Nilaparvata lugens*, *Pennisetum purpureum*





**Biological characterization of *Stethorus pauperculus* Weise
(Coleoptera:Coccinellidae) feeding on *Tetranychus urticae* Koch
(Acari: Tetranychidae) and survival on non-mite foods under laboratory
conditions**

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Two-spotted spider mites *Tetranychus urticae*, the most serious pest on various crops and weeds such as, gourd crops, beans, sweet pepper, cassava, and water hyacinth in Vietnam. The present studies were focused to predators because of the insecticide resistance potential of this pest. This study focused on the *Stethorus pauperculus* Weise, a promise predator, two-spotted spider mites in cassava field. The life cycle, fecundity, longevity, survivorship and sex ratio of *S. pauperculus*, were performed under laboratory conditions ($27\pm 2^{\circ}\text{C}$, $60\pm 5\% \text{RH}$). Two-spotted spider mites were collected from natural then feeding to the predator. Three types of host plants, including cassava leaves, bean leaves, water-hyacinth, were used to evaluate the most effect of different host plants on the life cycle of *S. pauperculus*. The life cycle of predators was completed successfully in 15.73 ± 1.53 , 18.4 ± 1.59 and 15.07 ± 1.49 days on cassava leaves, bean leaves, with water-hyacinth, respectively. The daily and total fecundity were recorded 3.5 ± 0.62 and 50.9 ± 12.26 eggs, respectively. The longevity of *S. pauperculus* was more than 30 days with an average ovipositional period of 14.6 ± 2.27 days. The survival rate from egg to adult was 51.7%. The sex ratio ($\text{♀}:\text{♂}$) of *S. pauperculus* is 1.1:1. In addition, in the experiment without mites (non-mite foods) showed that the larva of *Stethorus pauperculus* could feed by larva of *Thrips palmi*. The results suggested that *Stethorus pauperculus* developed their life cycle better when using water-hyacinth. In addition, *Stethorus pauperculus* larva could eat other pests to survive.

Keywords: Biological characterization, *Stethorus pauperculus*, life cycle, non-mite foods, host plants





**Potential Biological Control Agents of the Lanzones Mussel Scale,
Unaspis mabilis Lit & Barbecho (Hemiptera: Diaspididae) in CALABARZON,
Philippines**

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Mussel scales insect (*Unaspis mabilis* Lit & Barbecho) caused high yield losses to lanzones (*Lansium domesticum* L.) producing provinces in the Philippines specifically in the Cavite, Laguna, Batangas, Rizal and Quezon (CALABARZON). The objective of this study is to identify the ladybird beetles associated with the mussel scale insects. Specimens of mussel scales and their associated predators were collected in the CALABARZON area. Both the scale insects and the natural enemies were placed in 10X12 inches zip-locked bags and brought to the laboratory for rearing and identification. Representative samples of mussel scales were mounted and preserved in Hoyer's mounting media. Associated predatory beetles were reared to adult stage if larvae were collected. Representative samples of the predators were preserved as dried specimens which were mounted in paper points and stored in airtight Cornell boxes. The predators were identified to the nearest possible taxa using pertinent taxonomic keys. Documentation and illustrations of diagnostic characters are provided. The beetles were identified to be *Acarinus philippinensis* Kapur, *Chilocorus circumdatus* (Gyllenhal), *C. nigrita* (Fabr.), *Scymnus (Neopullus) hoffmani* Weise, *Nephus phosphorus* Lewis, *Microweiseinae* sp., *Pseudoscymnus* sp., *Scymnus (Pullus)* sp., and *Telsimia nitida* Chapin. Further studies and rearing researches are recommended to evaluate these potential biological control agents against mussel scale insects.

Keywords: mussel scale insect, *Lanzium domesticum* L., Coccinellidae, predatory ladybird beetles





Synthesis of a novel fluorescent sensor based on dansyl derivative for anion detection

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A novel fluorescent sensor 2'-hydroxy-5,5'-dinitro-[1,1'-biphenyl]-2-yl 5-(dimethylamino)naphthalene-1-sulfonate (T_1) was synthesized in two steps and characterized by ^1H NMR, ^{13}C NMR, ESI-MS, FT-IR spectroscopy and elemental analysis. The complexation of T_1 and various anions (F^- , Cl^- , Br^- , CH_3COO^- , $\text{C}_6\text{H}_5\text{COO}^-$ and H_2PO_4^-) was evaluated by ^1H NMR and fluorescence spectroscopy in DMSO solution. Addition of F^- , the color of solution T_1 changed from pale yellow to red. For ^1H NMR titration experiment, the disappearance of OH proton at 10.5 ppm of T_1 occurred due to the deprotonation process. The aromatic protons were induced to upfield, resulting the increase of electron density. Moreover, the fluorescence spectrum of T_1 obviously found the enhancement of a new emission wavelength at 450 nm. The 1:1 ratio of complexation between T_1 and fluoride ion was confirmed by Job's method.

Keywords: fluoride, anion sensor, fluorescent, hydrogen bonding





Total Synthesis of Dart Frog Indolizidine **209B** and **209D**

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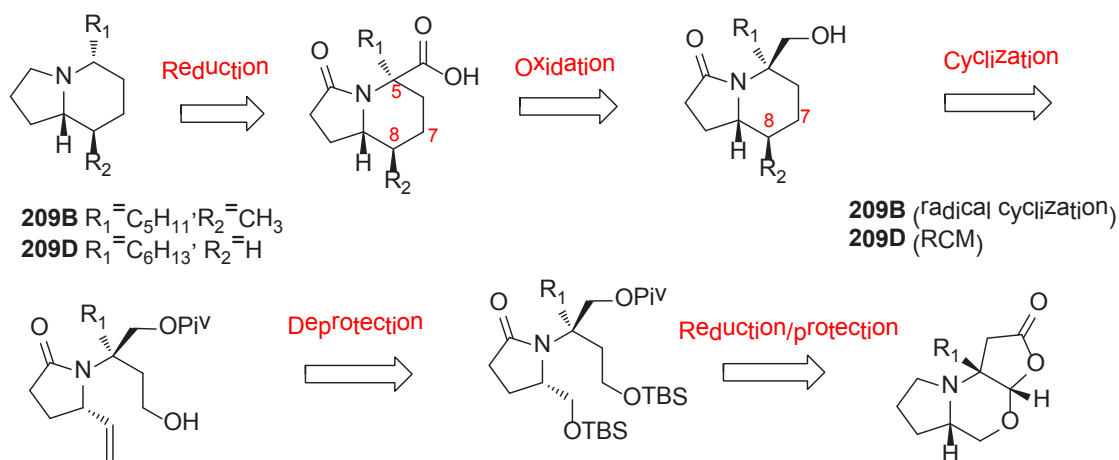
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The systematic investigation of extracts from the skin of neotropical amphibians, most notably from frogs belonging to the Dendrobatidae family, has resulted in the isolation of a number of alkaloids. Highly stereo-controlled total synthesis of dart frog indolizidine alkaloids (—)-**209D** and (—)-**209B** was accomplished, with a common tricyclic lactone intermediate as the starting compound. The key reaction of the synthesis of (—)-**209D** was ring closure metathesis followed by hydrogenation to construct indolizinone. The key reaction of the synthesis of (—)-**209B** was radical cyclization to construct indolizinone. The excess carbons on C5 position of both two lactams were removed through Barton decarboxylation. LAH reduction of lactams completed the total synthesis of (—)-alkaloid **209B** and (—)-alkaloid **209D**. Total synthesis of (—)-alkaloid **209B** and (—)-alkaloid **209D**, both in 19 steps, in overall yields of 8% and 6%, respectively.



Keywords: natural products, total synthesis, alkaloids, Barton decarboxylation, RCM





Assessment the metabolic capacity for biodiesel from insects biomass

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Biodiesel is an alternative potential energy source for petroleum diesel in the context of the energy crisis global, however, biodiesel price is expensive. To reduce production costs, the source of cheap materials has been more considered. Insect is one of the most promised source because of its huge and abundant biomass in the world. The parameters of the reaction trans-esterification were optimized at a reaction temperature of 85°C, with 6:1 methanol to oil ratio (v/v), 0.4% (w/w) NaOH catalyst, and reaction time of 60 minutes. The maximum efficiency of the *Zophobas morio* larvae oils (ZMLO) reaction was 84.33% and the *Hermetia illucens* larvae oils (HILO) reaction was 81.67%. The ratio of saturated fatty acids in the oil was 38.31% for ZMLO and 52.33% for HILO. Biodiesel from insects has relative oxidation stability. The ratio of saturated fatty acids C16 and C18 reached 37.40% (ZMLO), 15.52% (HILO) has great potential in the production of biodiesel. Methyl ester content various in products, can be mixed to get the desired fuel. The biomass residual after extraction can be used as protein feedstuff for animal feed industry. Through assessment the metabolic capabilities biodiesel found, the production of biodiesel from insects can be applied in practice.

Keywords: biodiesel, esterification, transesterification, *Zophobas morio*, *Hermetia illucens*, insects





Synthesis of Amphiphilic Nanoparticles Using a Cell-Penetrating Pentapeptide, KTTKS, Linked with Stearic Acid and Rhodamine to Enhance the Toxicity in *E. Coli* and Zebrafish Embryo

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Nanoparticles have been used widely for drug delivery both for diagnostic and therapeutic applications. Liposomes are one of the nanoscale materials commonly used for drug delivery because they are biocompatible, possess the permeability needed for drug uptake, and have a particle size suitable for the encapsulation of drugs. To increasing the effective concentration inside the cell, liposomes consisted of cell-penetrating peptides was prepared. A pentapeptide of Lys-Thr-Thr-Lys-Ser (KTTKS) was used as hydrophilic moiety which is known to be capable of stimulate the regeneration of collagen in skin. For the lipophilic moiety, stearic acid and rhodamine were used to link with a pentapeptide through the formation of an amide bond. The preparation of peptide and its derivatives were achieved by solid-phase synthesis. After the identification with NMR and Mass spectroscopy, these modified amphiphiles were capable of self-assembling to be functional liposomes in water under the sonication condition. The morphology of nanoparticles was confirmed by Transmission Electron Microscope (TEM) images. The toxicity of peptide-derivatives liposomes was evaluated in zebrafish embryos that were staged according to their age (dpf) with a standard procedure. The formation of nanoparticles by the rhodamine which linked with a short peptide (KTTKS) was capable of enhancing the cell penetration to induce the specificity in zebrafish neuromasts for maintaining a longer time. As a result, the formation of liposome is crucial for increasing the toxicity of peptide-derivatives in the transportation of target compounds into cells to exert its biological effect.

Keywords: cell-penetrating pentapeptide, KTTKS, amphiphilic nanoparticles, rhodamine, zebrafish embryo





A Novel Synthesis of Dual-responsive Polymer Nanoparticles as Antitumor Drug Carrier

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First research developed a dual temperature- and pH-responsive drug carrier which was synthesized through radical copolymerization of methacrylic acid (MAA), N-isopropylacrylamide (NIPAm), and N- (methacryloyl)glycylglycine 4-nitrophenyl ester (MA-GG-ONp). The monomer of NIPAm has the property of lower critical solution temperature and the monomer of MAA can response to phase transition under acidic condition. When the anticancer agent gemcitabine or antibiotic sulfamethoxazole was conjugated with the polymer, we can adjust the composition ratio to form the dual temperature- and pH-responsive polymer-drug. The structure and properties of polymer-drugs were investigated using nuclear magnetic resonance, acid base titration, ultraviolet–visible absorption, dynamic light scattering and turbidity measurement. When heated beyond its lower critical solution temperature (40°C), a dual temperature- and pH-induced phase transition was observed. This temperature was considered ideal for activating drug aggregation under hyperthermic and acidic conditions for cancer treatment.

Second research was based on previous study and was under different method. This study reports the rapid synthesis of two biodegradable and amphiphilic block copolymers through reversible addition–fragmentation chain transfer (RAFT) polymerization under microwave irradiation. Through use of 2-dodecylsulfanylthiocarbonylsulfanyl-2-methyl propionic acid (DMPA) as a RAFT agent, the microwave-assisted polymerization rate of N-isopropylacrylamide was faster than that observed under conventional heating conditions, and the resulting homopolymer can be reactivated as a macroinitiator to produce pNIPAm(DMPA)-*b*-pMAA and pNIPAm(DMPA)-*b*-pAA block copolymers through a similar method. Cisplatin was loaded to the polymeric carrier through a ligand exchange to form a macromolecular prodrug. The observed critical micelle concentration was 0.1 mg/mL and 0.15 mg/mL. Overall, these polymers offer considerable potential for developing a new multifunctional drug delivery system.

Keywords: cisplatin, polymer-drug, drug delivery, microwave-assisted synthesis, RAFT polymerization





Nanoparticle or Thin Membrane of Carboxymethyl Chitosan for Fibroblast Growth Factor 2 Release

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Fibroblast growth factor 2 (FGF-2) is a multi-functional protein regulating many biological processes, especially those involved in wound healing. However, FGF-2 has short half-life and is easily degradable *in vivo*, which limited its use for wound treatment. In this study, we investigated a drug-delivery model for FGF-2 via incorporation with biodegradable carboxymethyl chitosan (CMCS), a substance classified as safe for use in human by the Food and Drug Administration. We showed that CMCS nanoparticles (NPs) could be synthesized by ionic gelation method using CaCl_2 as a crosslinking reagent with the CMCS: CaCl_2 ratio of 1:0.8. Hydrogel film from CMCS and hydroxyethyl cellulose (HEC) was prepared by blending and casting method with CMCS:HEC weight ratios 4:1. Synthesized CMCS NPs were spherical with an average diameter of 31.96 ± 5.31 nm, which was determined by Field Emission Scanning Electron Microscope (FE-SEM), and were non-toxic at concentrations up to 2.5 mg/ml, while CMCS/HEC hydrogel was non-toxic at concentrations up to 5 mg/ml, had a porous structure and its swelling behavior sensitive with pH. Incorporated CMCS:FGF-2 NPs had an average diameter of 33.55 ± 5.39 nm and FGF-2 incorporation efficiency was 95%. CMCS:FGF-2 NPs and hydrogel film had ability to release FGF-2 with releasing rate over 50% after 48 hours and the release process could be affected by pH of solution. Moreover, the incorporation and release processes did not have a significant effect on FGF-2 activity, which was measured via its induced proliferation of NIH-3T3 cell line using MTT assay. Simultaneously, CMCS:FGF-2 NPs and hydrogel film could protect FGF-2 from the degradation of trypsin *in vitro*. Taken together, our results laid the groundwork for the manufacturing of delicate protein incorporated CMCS NPs and hydrogel film for bio-applications.

Keywords: carboxymethyl chitosan, fibroblast growth factor 2, hydroxyethyl cellulose, nanoparticles, thin film





The Weather during a Low-Level Wind Shear

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The purpose of this research is to study the weather during a low-level wind shear by comparing cloud types and rainfall when the low-level wind shear occurs and then analyze what is the weather like that possibly to create a low-level wind shear.

This research use the aviation routine weather reports data from 2012-2015 from Suwannaphum airport. This finding indicate that the cumulonimbus cloud will appears every time when the low-level wind shear occurs. When the low-level wind shear occurs sometime there is no rain, sometime there is drizzle and sometime there is heavy rain. Low-level wind shear usually occurs on July to August, and usually occurs between 6.00 pm.-0.00 am.





The Study of Hydrogeology and Groundwater Potential in KMUTT (Ratchaburi learning park) Using Detail Resistivity Imaging

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KMUTT (Ratchaburi learning park), a particular drought zone with mainly metamorphic rock aquifer zone where located at Rangbua subdistrict, Chombueng district, Ratchaburi province, is found a limit of surface water. As a result, the most alternative water supply must be from groundwater resources. Then the study of hydrogeology and groundwater potential, detail 2D resistivity measurement was applied to locate high potential zone using Geomative resistivity instrument, GD-10 model with 84 multi-electrodes. Six survey lines were designed in E-W covering the campus area of about 2 km in E-W and 800 m in N-S with 720-840 m length. Data collecting in the field, Schlumberger configuration was taken with resolution of 10 m for continuing detail along survey line with depth of >120 m with electrodes spacing of 10 m. In data processing stages, both 2D and 1D inversion models have been done for details vertical resistivity change in every 10 m. From hydrogeological study groundwater can be found as groundwater flow through the campus are S-N direction. At least 3 location of high yield groundwater can be indicated of northern part of the campus. Aquifer may found in the fracture of local bedrock with depth of 50-80 m. These high yield groundwater positions are recommended for groundwater drilling in the future water supply in the campus.

Keywords: 2D Resistivity, Groundwater, Potential Groundwater Zone, Detail Resistivity Imaging.





Depositional environment of Permian limestone, Khao Erawan, Lopburi

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Every sedimentary rock represents its depositional environment, which has factors specific to sediment transport, deposition, and diagenesis evident in rock texture. Permian limestone that was deposited in Khao Erawan area, Lopburi, contains abundant and diverse fossils and shows different texture throughout the study area. These texture and fossils can reflect ancient environments during limestone deposition. However, limestone in this area has not been studied in detail. Here we show a depositional model and stratigraphy of the Permian limestone, which was deposited during Artinkian to Kungurian age enhances (about 290 - 272 million years ago) in coastal environments. It can be divided into 4 units chronologically: lime mud conglomerate unit, diversely fossiliferous limestone unit, fossiliferous limestone unit, and slightly fossiliferous limestone unit. The lime mud conglomerate unit was deposited in supratidal environments, whereas the other three limestone units were deposited in intertidal environments. The latter consists of fossil skeletons as a major allochems. However, their abundance and their diversity vary among the three units. This study not only has academic significance, but also enhance basic knowledge among local people in order to better develop their hometown as a valuable and interesting nature, especially geology, learning center.

Keywords: Depositional environment, Permian limestone, fossil, Lopburi





MY AEC: Mobile Application for Traveling in AEC Country

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AEC is developed from Association of Southeast Asian Nations (ASEAN). There are five countries to be start 1. Indonesia 2. Malaysia 3. Philippines 4. Singapore 5. Thailand, followed by Brunei, Vietnam, Laos, Myanmar and Cambodia join the members. As a result to be the ASEAN Economic Community (AEC) that was established for political cooperation, economic security society and culture. In current, Thailand is one of ten AEC member countries. There are many Thai people who travel or work in countries that are members of the AEC community. It is important to know the language and culture of that country in order to make communication faster and easier.

From the above, we get the idea to create the application for travelling called “MY AEC” in android operating system for traveler for tourists who are interested in traveling to countries that are members of the AEC Community. The application can support from all countries because this application is mainly used in English, and use the national language in AEC. If user choose Thailand flag, in the function will display the function statement both English and Thai. The design of this application is designed to be easy to use, so that users can easily understand and in this application has various functions but there is also a clear classification of this application. In this application will gathers the information needed for tourists, including: 1. Basic information about that country. In this function will show simple information for example: the capital name of that country. 2. Common vocabulary used in everyday life. In this function will have be karaoke language to read as the main language selected, and there are sounds of vocabulary from native speakers to make it easier for users to communicate with native speakers. 3. The attractions of the country. In this function will show simple information that the nature of the place, such as entrance fees and location so that users can choose and decide to travel more easily. 4. National food of that country 5. Culture that should do and should not do in order to make users understand the culture of that country and users will behave correctly. 6. Airport. In this function will show name and location from Google map of the airport in the capital or nearby for users to choose the nearest airport. 7. Currency conversion system will make users can calculate the cost roughly that the monetary value will be the current value.

In this application will help users to know vocabulary and language of that country in order to communicate with native speakers have more understanding and users can travel to places and users can choose food by yourself that make travel in AEC to be easy.

Keywords: AEC, Travel, Vocabulary, Attractions, National food, Culture, Airport, Currency





Loop mediated isothermal amplification (LAMP) technical application in detection of Hepatitis B virus in human

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Hepatitis B virus (HBV) is considered as one of the major causes of acute hepatitis, chronic hepatitis and hepatocellular carcinoma (HCC). According to the World Health Organization, there are 2 billions of worldwide people who have been infecting by this virus and 686,000 people died as consequences a result of hepatitis B infection in each year. Currently, there are several diagnostic tools based on serological tests and molecular techniques, which have been applied domestically and internationally for the diagnosis of hepatitis B virus. However, these tests require expensive equipment, high-skilled personnel, and time. Recently, many studies have been indicated significant advantages of loop - mediated isothermal amplification (LAMP) based method that could be over serological tests and PCR for rapid detection of microbial pathogens. The objective of this study was to develop a rapid LAMP assay for the detection of HBV from DNA and particularly blood samples from infected patients by using a commercially available master mix and a mobile real-time fluorometer. The final optimized fluorescence-based LAMP assay provided significant amplification time of less than 15 minutes in comparison with over 1 hour for PCR and even an opened tube LAMP system as described previously (*Bst* polymerase based LAMP assay, gel electrophoresis and colorimetric assay). Additionally, the productivity and the diagnostic sensitivity of LAMP assay were conducted with donor blood samples (n = 30). The result of this experiment revealed that LAMP assays detected 19 of 30 donor blood samples (63%) as HBV positive. This result was consistent with with clinical validation of those samples using realtime PCR and better than PCR method tested in this study. Therefore, fluorescence-based LAMP assay is more sensitive than PCR as a comparator and can be used for rapid detection of HBV directly with heat-treated blood samples resulting a rapid, sensitive, efficient and high reliable approach that could reduce time, labor and costs in the implementation of rapid detection of HBV in human.

Keywords: Hepatitis B virus, diagnostic, LAMP, Rapid detection





The methylation status of *Deleted in lung and esophageal cancer 1 (DLEC1)* gene promoter: a potential biomarker for Nasopharyngeal carcinoma in Vietnamese patients

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Background: The silence of *Deleted in lung and esophageal cancer 1 (DLEC1)*, which is belonged to the tumor suppressor gene, located 3p.22.3, has been observed in several types of human cancer, including Nasopharyngeal carcinoma (NPC), the most common and highly incident cancer of head and neck cancer in Asian countries, especially in Vietnam. Promoter hypermethylation, one of the epigenetic mechanism, has been shown to be responsible for the silencing of *DLEC1*.

Purposes: To investigate the frequency of *DLEC1* gene promoter hypermethylation status as well as whether or not an association between patterns of *DLEC1* hypermethylation and NPC.

Methods: A total of 20 NPC biopsy samples and 20 non-cancerous nasopharyngeal brushing samples were enrolled into current study. The methylation of the *DLEC1* gene promoter was determined using methylation nested-specific PCR (nested-MSP) with the outer primer yielded the product within 260-bps in length, from nucleotide 19572 to 19831 of *DLEC1* promoter (Accession number: AP006309).

Results: The frequency of aberrant promoter methylation and unmethylation of *DLEC1* gene in NPC samples were 75.0% (15 of 20 samples) and 25.0% (5 of 10 samples), respectively. In the case of non-NPC brushing sample was 15.0% (3 of 20 samples) and 85.0% (17 of 20 samples) for methylation and unmethylation, respectively. Moreover, a trend toward positive association was found between hypermethylation of *DLEC1* gene and NPC was observed ($p = 0.0005$). Moreover, the odds ratio (OR) and relative risk (RR) were found in statistical significant value: OR = 17 (95%CI = 3.464 – 83.438, $p = 0.0005$) and RR = 5.0 (95%CI = 1.709 – 14.626, $p = 0.0033$), respectively.

Conclusion: Our study indicated that *DLEC1* hypermethylation was significant associated with NPC in Vietnamese patients. However, it is necessary to a larger number of samples in order to confirm the characteristic of *DLEC1* hypermethylation could to be considered as the promising biomarker for early diagnosis and screening for NPC in Vietnamese population.

Keywords: Nasopharyngeal carcinoma, *DLEC1*, hypermethylation, nested-MSP.





Variations of *Epstein-Barr virus nuclear antigen (EBNA-1)*: V-val type preferentially exists in biopsies of nasopharyngeal carcinoma from Vietnamese patients

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Background: *Epstein-barr virus nuclear antigen (EBNA1)*, encoded by *EBNA-1* gene, has been shown as one of the most frequently detected protein in Nasopharyngeal carcinoma (NPC), which the most common and highly incident cancer of head and neck cancer in Asian countries. The geographically-associated polymorphisms of EBNA-1 have been observed in East-Southern Asia, including Vietnam. The subtype V-val EBNA-1 has been demonstrated that it may contribute to the oncogenesis of NPC. This current study is performed to characterize the variations of EBNA-1 in NPC biopsy samples from Vietnamese patients.

Methods: A systematic literature analysis was conducted based on the comprehensive search of observational studies. Experimentally, for the variations characterization, Nested PCR-sequencing was applied to amplify and characterize the C-terminal region of *EBNA-1* gene by the designed oligonucleotide primers.

Results: Total of 6 previous published studies were identified and accessed for eligibility from the literature research and enrolled into systematic revision. The variants of EBNA-1 have been described based on the amino acid signature at codon 487, which is classified into five subtypes: P-ala, V-ala, P-thr, V-val and V-pro. The nested PCR-sequencing method, counting for 100% (6 of 6 studies), was the majority technique for EBNA-1 variants characterization. Experimentally, 20 NPC biopsy samples were enrolled, as the results, only two patterns of EBNA-1 variations: P-ala and V-val were observed. In addition, of these two subtypes, the frequency of V-val and P-ala were determined counting for 80% (16 of 20 cases) and 20% (4 of 20 cases), respectively.

It indicated the V-val subtype preferentially exists in biopsy NPC samples, which collected from Vietnamese patients.

Conclusion: We successfully designed the nested PCR-sequencing primer for detection of EBNA-1 variations based on the data collected from previous study. In our initial study, the sub-strain of EBV with V-val subtype of EBNA-1 infects NPC preferentially to those from biopsies of NPC patients. In further study, it is necessary to a larger number of samples and non-cancerous samples in order to confirm the characteristic of EBNA-1 variations, as well as determination of the association V-val subtype with NPC in Vietnamese patients.

Keywords: V-val, P-ala, variations, EBNA-1, nasopharyngeal carcinoma.





The association between glutathione *S*-transferase *P1* gene hypermethylation and breast cancer: A meta-analysis

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Worldwide, the correlation between aberrant methylation of *GSTP1* gene and breast cancer have been highlighted in many scientific researches, notably, the results are arguable as it should be a present tense here. Therefore, this study was carried out to evaluate the association between the *GSTP1* hypermethylation with breast cancer by meta-analysis.

Previous studies, which were collected from many databases, including PubMed, PudMed Central, Google Scholar were enrolled in present study. As the results, total of 11 studies were systematically collected, by the date of October, 2016. In this data collected samples, it composed 446 cases of breast cancer and 361 controls. The correlation was evaluated by the odds ratios (OR), which were appropriately calculated from fixed-effects (F) or random-effects (R) models and 95% confidence interval (95% CI).

At the overall analysis, the *GSTP1* hypermethylation was proven to be significantly associated to breast cancer under a random effect model (OR = 9.846 (95%CI = 2.619-37.014; $p = 0.001$)). Additionally, the hypermethylation of *GSTP1* promoter was significant associated to late stage of breast cancer (OR = 1.685 (95%CI = 1.007-2.818; $p = 0.047$)), high histological grade (OR = 2.409 (95%CI = 1.286-4.51; $p = 0.006$)), lymph node positive at diagnosis (OR = 1.851 (95%CI = 1.095-3.126; $p = 0.021$)) and the down-expression of GSTP1 protein (OR = 86.549 (95%CI = 3.167-2365.073; $p = 0.008$)).

These results suggested that, the hypermethylation of *GSTP1* promoter could be served as the potential biomarker in diagnosis and monitoring breast cancer.

Keywords: *GSTP1*, hypermethylation, breast cancer, meta-analysis





Identifying the genomic elements associated with drug resistance based on text mining

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The emergence of drug resistance in clinic leads to the failure of standard treatments, thereby making the treatments of diseases more complex and costly. Therefore, understanding the mechanism of drug resistance may help to improve the drug therapy. Here, we performed a text mining method to extract the potential associations between human drug resistance and genetic factors including genes, single-nucleotide polymorphisms (SNPs) and microRNAs (miRNAs) from a comprehensive perspective. For each drug, all possible synonyms from compound were adopted as its drug names. For genes/miRNAs/SNPs, symbols as well as all their possible synonyms were used to query the MEDLINE. For each paper, only the title and abstract were considered in this work and the associations between drugs and genes/miRNAs/SNPs were derived based on their co-occurrence. If a drug and a gene/miRNA/SNP co-occur in the same sentence in the abstract or title of one published paper, they are possibly related to each other. Furthermore, if the key word resistance/resistant/chemoresistance occurs around the drug name in the same sentence in which the gene/miRNA/SNP also occurs, we supposed this gene/miRNA/SNP is possibly responsible for the resistance to the drug. Due to the high false positive rate of text mining, all associations between drug resistance and genes/miRNAs/SNPs were manually curated to improve the precision. As a result, 1631 associations between 201 drugs and 758 genes, 106 associations between 29 drugs and 66 miRNAs, and 44 associations between 17 drugs and 22 SNPs were kept and deposited into GEAR. In addition, to facilitate choosing associations between drugs and genes/miRNAs/SNPs, all associations are ranked based on the probability of their co-occurrence in MEDLINE database with mutual information. Moreover, we choose the resistant genes as an example to investigate the molecular mechanisms underlying drug resistance. The results on both pathway and Gene Ontology enrichment analysis imply that the resistant genes tend to be enriched in biological processes related to drug effects. By investigating the genetic factors and drug targets, it is found that the relationships between them can help to understand the molecular mechanisms underlying drug resistance. Moreover, it is confirmed that drugs with same resistant genes tend to have similar therapies which further indicated that similar drugs may have similar mechanism when develop resistance. In addition, considering the drug pairs sharing same genetic factor, it is also found that drug resistome can help predict new indications for old drugs and can be useful for narrowing down candidate drug combinations.

Keywords: drug resistance, text mining, genomic elements





Control Strategies on a TB-HIV/AIDS Co-infection Model

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Bhunu *et al* (2009) developed a Tuberculosis and HIV/AIDS (TB-HIV/AIDS) co-infection model considering antiretroviral therapy for the AIDS cases and treatments for latent and active forms of TB. These treatment rates are determined beforehand, and the model is analyzed using stability analysis. In this research, we analyze the model with treatments using optimal control, so the treatment rates are modified from static to dynamic parameters. We aim to see the effects of these treatment controls over a period of 10 years of implementation to the population. Pontryagin maximum principle was applied to derive the differential equations system as a condition that must be satisfied by the optimal control variables. Furthermore, the fourth-order Runge-Kutta method was exploited to determine the numerical solution of the optimal control problem.

The numerical results show that treatments on a TB-HIV/AIDS co-infection model provide good effects to the population after 10 years of control application. In the presence of treatment, TB-exposed and TB-infective populations are decreasing, while the TB-recovered population is increasing significantly. HIV-positive population increasing due to some AIDS patients improving their health following antiretroviral treatment, and recovering from TB for those HIV-positive individuals dually infected with TB. Individuals exposed to TB and infected with HIV decreasing due to treatment of latent TB. Individuals exposed to TB and sick from AIDS also decreasing due to treatment of latent TB and antiretroviral treatment. HIV-positive individuals dually infected with TB are decreasing due to treatment of TB, and lastly the number of individuals dually sick from TB and AIDS also decreasing due to the combination of TB and AIDS treatment. From these results we can conclude that TB treatment is effective not only on TB-only patients but also on TB patients co-infected with HIV/AIDS.

Keywords: optimal control, antiretroviral therapy, tuberculosis, HIV/AIDS, Pontryagin maximum principle, Runge-Kutta method





Optimal Control of Malaria Transmission in Two Regions Using Treatment and Spraying

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Malaria is caused by one of the four species of protozoa in the genus *Plasmodium*, named *P. falcifarum*, *P. vivax*, *P. malariae*, and *P. ovale*. The environmental condition in the tropics is the prime factor for it being endemic. In 1890, Sir Ronald Ross had demonstrated the life-cycle of the malaria parasite in mosquito, and he developed a simple model that is the Ross Model. The model has been developed by many researchers, some of them are Johansson dan Leander (2010).

In this study, a model of the transmission of malaria in two regions, which is developed by Johansson dan Leander, was equipped with two control variables, namely the treatment effort to reduce the number of susceptible humans who become infected by infectious mosquitoes bites and the effort of reducing mosquito population through spraying. We consider two scenarios related to the application of control strategies under different rate of movement between regions. Pontryagin maximum principle was applied to derive the differential equation system as optimality conditions which must be satisfied by optimal control variables. The 4th-order Runge Kutta method was then performed to solve the differential equations system.

It is shown by numerical simulation that controls application applied to both regions provides better performance than that applied to one region. When controls applied to both regions, integrated control is most optimal to minimize the control performance in the two regions. Generally, giving spraying in the first 7 days is enough, when treatment can be given to 30-40 days, and after that the controls can be decreased until the last periode.

Keywords: malaria two regions, optimum control model, Pontryagin maximum principle, Runge-Kutta method





Impact of monochromatic lights on the growth, biosynthesis of bioactive substances and expression of *Cmwc-1* in *Cordyceps militaris*

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Cordyceps militaris is a precious medicinal fungus, typically parasites on larvae or pupae in some insects. Light is an essential environmental factor for the stroma's production, accumulation of bioactive substances and pigmentation in *Cordyceps militaris*. Light also affects the expression of the *Cmwc-1* gene in *Cordyceps militaris*, a blue-light receptor for *wc-1* in *Neurospora crassa*. In this study, difference of LED monochromatic lights (blue, green, red), with the intensity of lights including 400 lux, 600 lux and 800 lux, were used during *Cordyceps militaris* cultivation. Effects of various monochromatic lights on bio-efficiency of stroma, biomass, and contents of cordycepin and adenosine were evaluated. Also, RT-PCR technique was used to confirm the presence of gene *Cmwc-1* in different light conditions.

The result showed that blue-light of 400 lux had highly affected stroma formation (16,97%) compared with blue-light of 600 lux (13,67%) and 800 lux (6,82%), green-light of 400 lux (8,06%), green-light of 600 lux (10,71%) and 800 lux (11,92 %), red-light of 400 lux (10,42%), red-light of 600 lux (10,37%) and 800 lux (10,91%). The white-light of 800 lux highly affected stroma formation (17,00%) compared with white-light of 600 lux (14,42%) and 400 lux (9,65%). In addition, these stromata weren't formed in dark condition.

Red-light of 800 lux was optimal for biomass increasing (32,05 g) compared with monochromatic lights, green-light of 800 lux (29,95 g), blue-light of 400 lux (25,26 g), darkness (13,89 g), and normal growth condition, which is white-light (29,33g). Blue-light of 400 lux obtained the highest content of cordycepin (4329,766 µg/g) compared with green-light of 800 lux (3363,998 µg/g), red-light of 800 lux (2797,771 µg/g), white-light (2293,196 µg/g) and darkness (1413,910 µg/g), respectively.

RT-PCR results confirmed the expression of *Cmwc-1*, which was only expressed in the light conditions (white-light or monochromatic light) but not under dark condition.

Taken together, the results demonstrated the function of monochromatic lights on the growth and biosynthesis of bioactive substances in *Cordyceps militaris*. These results also suggested a possibility of substitution of the white-light by monochromatic lights for improving the quality and bioactive substance enrichment in *Cordyceps militaris*.

Keywords: adenosine, cordycepin, *Cordyceps militaris*, monochromatic light, stroma





Expressional and functional diversity of *Bombyx mori* Opsins

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Opsins, the components of animal visual pigments, are members of the rhodopsin family of the super family of G-protein-coupled receptors (GPCRs). Opsins were suggested as photoreceptors that interpret photic input that plays an important role in circadian rhythm as well as photoperiodism. In insects, photoreceptor opsins have been characterized in term of cellular expression and sequence. Several types of opsins were confirmed by sequence analysis that may respond to different wave length of light. In *Bombyx mori*, Long-wave opsin (LWO) was cloned and investigated its cellular localization. We have identified other *B. mori* opsin genes including wave-length specific opsins (Short-wave opsin (SWO), Ultra-violet opsin (UVO)), Opsin3, and Pteropsin. Their expression pattern was investigated in both mRNA and protein levels by using northern blot, *in situ* hybridization, and immunohistochemistry. Northern blot results showed that *B. mori* opsin mRNAs were expressed only in *B. mori* brain tissue but not in muscle, fat body, ovary or testis tissues. Spacial localization of all *B. mori* opsin genes were found on deuto-lateral cells in silkworm brain and on sub-oesophageal ganglion cells. Cellular co-localization of opsins and diapauses hormone and the key circadian clock protein (PER) revealed a functional diversity of these opsins not only in photoperiodic-related phenomena but also in circadian rhythm in *B. mori*.

Keywords: circadian rhythm, immunohistochemistry, *in situ* hybridization, Opsin, photoperiodism, photoreceptor





Bioconversion of AHX to AOH by *Buttiauxella gaviniae* A111

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A compound that stimulates the growth of bentgrass, 2-azahypoxanthine (AHX), was isolated from a mushroom, *Lepista soldida*. AHX is metabolized to 2-oxohypoxanthine (AOH) in rice. Both compounds show plant growth-promoting activity and are present in rice at levels similar to those of plant hormones. AOH also imparts drought stress tolerance and cold stress tolerance to *Arabidopsis thaliana*. AOH has been synthesized from AHX by enzymatic method because it is difficult to synthesize AOH by a chemical method. At first, AOH was synthesized by xanthine oxidase (XOD) from buttermilk. The commercially available XOD is not suitable for large-scale preparation of AOH because the enzyme is very expensive and the concentration of substrate, AOH is very low (70 mg/L). In the previous study, we found that *Burkholderia contaminans* CH-1 converted from AHX to AOH at 2 g/L but this strain was not enough to prepare AOH in a large scale. In this study, to develop effective bioconversion method of AHX to AOH in a large scale, we screened for microorganisms that show higher conversion activity and showed properties of the conversion enzyme.

The conversion of AHX to AOH was carried out in PBS (pH 7.4) at 30 °C in various conditions. AHX and AOH were analyzed by HPLC (Develosil C30-UG-5 column, 0.02% TFA, 30°C, 254 nm.) For the enzyme purification, the cells were disrupted by sonication. The AOH conversion enzyme was purified through ammonium sulfate precipitation follow by a multistep purification process as hydrophobic interaction chromatography and anion-exchange chromatography.

Four strains (A13, A82, A111 and A112) isolated from soil showed high AOH conversion activity at 2 g/L of AHX in resting cell reaction for 4 hours. Among these strains, strain A111 was selected as a strain showing the highest AOH conversion activity. Although strain CH-1 converts 2 g/L of AHX to AOH for 24 hours, strain A111 completely converts AHX to AOH at 2 g/L within 4 hours. Strain A111 can convert 5 g/L of AHX to AOH by the resting cell reaction within 24 hours in 10 liter jar fermenter. The yield of this reaction is 92.8%. Conversion activity of strain A111 is 8-10 times higher than *B. contaminans* CH-1. Strain A111 is presumed as *Buttiauxella gaviniae* by 16S rDNA analysis. Most of the xanthine-oxidizing enzymes from microorganisms are xanthine dehydrogenases. AOH conversion enzyme from *B. gaviniae* A111 shows a xanthine oxidase activity.

Keywords: *Buttiauxella gaviniae*, 2-Oxohypoxanthine (AOH), 2-Azahypoxanthine (AHX), Xanthine oxidase





A comparative genomic analysis of cell wall degrading enzymes encoded proteins in *Magnaporthe oryzae*, *Neurospora crassa* and *Aspergillus niger* and *Aspergillus oryzae*

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Filamentous fungi have a diverse array of cell wall degrading enzymes (CWDEs) to depolymerize the main structural polysaccharide of plant cell wall including cutin, cellulose, hemicellulose and pectin. The role of CWDEs and systemic analysis of these proteins have been previously reported in the rice blast fungus, *Magnaporthe oryzae* but a few studies have been demonstrated in filamentous fungi. In this study, a comparative genomic analysis of CWDEs in *Magnaporthe oryzae*, *Neurospora crassa* and *Aspergillus s.p* including *Aspergillus niger* and *Aspergillus oryzae* have been done. For *M. oryzae*, *N.crassa*, *A.niger*, *A. oryzae* respectively, we have identified 179, 79, 166 and 180 genes encoding CWDEs by crossing BLAST between CAZy database and Ensembl fungi database. Particularly, the function of 303 putative genes encoding CWDEs were revealed based on their conserved domain analysis and phylogenetic topology. The results of this study have been provided a comprehensive database resource on all CWDEs in *M. oryzae*, *N.crassa*, *A.niger* and *A.oryzae* to obtain further insight into the composition and evolutionary of CWDEs in filamentous fungi.

Keywords: CWDEs, comparative genomic, *Magnaporthe oryzae*, *Neurospora crassa* and *Aspergillus sp*





Optimization of semi-solid medium for simultaneous culture of lactic acid bacteria isolated from corn silage

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Ensiling is one of the most effective methods for preservation of livestock feed, in which lactic acid bacteria (LAB) plays an important role in the fermentation. This study aimed to isolate and identify of LAB from corn silage as well as optimize growth conditions of selected LAB strains. Based on colony morphology, Gram staining, biochemical test and 16S rRNA gene sequence analysis, four out of six wild strains which have highest acid-producing ability were identified as *Lactobacillus plantarum*, *Lactobacillus casei*, *Lactobacillus buchneri* and *Enterococcus faecium*. All of these LAB strains were able to assimilate starch, protein, cellulose and glycerol and could be grown simultaneously on a plate without any competition. After 48 h incubation on MRS medium, the bacterial biomass (CFU/ml) of single culture of *L. plantarum*, *L. casei*, *L. buchneri*, and *E. faecium* was 3.2×10^{10} , 1.2×10^{10} , 30×10^9 , and 2.4×10^9 , respectively. Optimizing semi-solid medium for simultaneous cultures of these strains was performed using Response Surface Methods (RSM) and Central Composite Designs (CCD) methods. Our results showed that concentrations of the medium components (g/L) were determined as molasses 20.0; soybean curd residue 86.7; 50-60% moisture content on a wet weight for achieving biomass at 4.28×10^9 CFU/g after 48 h of incubation at room temperature under aerobic conditions. In this study, semi-solid medium have been successfully found for future scale up of LAB production to improve the silage fermentation.

Keywords: Lactic acid Bacillus, MRS, Ensiling, RSM, CCD, semi-solid medium.





Antibacterial Activity of Green Bottle Fly (*Lucilia sericata*) Extracts on Antibiotic-Resistant Bacteria

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Recently, modern medicine is facing many difficulties and challenges from antibiotic-resistant bacteria. Many doctors and researchers have returned to ancient therapies, such as the use of larvae of flies to treat wounds, also known as Maggot Debridement Therapy (MDT). The testing of inhibitory activity against antibiotic-resistant bacteria was conducted with the live green fly larvae of *Lucilia sericata* at 3rd stages on the Nutrient Agar media and observed the inhibition after 24h, 48h and 72h. After 24h, the result indicated that the live green fly larvae of *Lucilia sericata* at 3rd stages were resistant to tested bacteria, including the common pathogenic bacteria and antibiotic-resistant bacteria which produce carbapenemase and Extended Spectrum Beta-Lactamase (ESBL). Based on this observation, we extracted compounds from 3rd stage larvae of green fly with n-Hexane, Chloroform, Ethanol and Methanol, then tested the extracts against a collection of bacterial pathogens by agar diffusion, plate count, Turbidometric (TB) and minimum inhibitory concentration (MIC) assay. The results showed that only n-Hexane extract has antibacterial activity against all groups of test bacteria including *S. aureus* and MRSA (Methicillin Resistant *S. aureus*) and the extract also has the strongest activity against the multidrug-resistant bacteria *Acinetobacter spp.* with the diameter of inhibitory zone is 18.00 ± 0.32 mm by disk diffusion method. Additionally, the testing of antibacterial activity against antibiotic-resistant bacteria was conducted in the plate count and Turbidometric assay, we found that the highest rate of bacterial inhibition is 98.26 ± 0.71 % – 100.19 ± 0.56 % in the TB assay and 93.58% in the plate count assay was analyzed by using TB formula and plate count formula. In the concentration test, we found that the minimum inhibitory concentration (MIC) is in the range of $5 \mu\text{g/mL}$ – $8.33 \mu\text{g/mL}$.

Keywords: maggot debridement therapy, *Lucilia sericata*, antibacterial, antibiotic-resistant bacteria





**In Vitro Efficacy of Fungal Endophytes and Medicinal Plant Extracts Against
Fusarium proliferatum (Matsush.) Nirenberg,
Causal Organism of Leaf Blight of Coconut (*Cocos nucifera* L.)**

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Alternative methods of managing plant diseases, such as the use of antagonistic biological agents and phytochemicals are being studied extensively due to emerging concerns on the negative impacts of synthetic fungicides. In this study, efficacies of endophytic fungi (EF) and medicinal plant extracts (MPE) as potential biological and botanical control, respectively, against *Fusarium proliferatum* causing leaf blight of coconut (*Cocos nucifera* L.) were evaluated *in vitro*. Five endophytic fungal isolates from asymptomatic coconut leaves collected from Los Banos, Laguna showed promising antagonistic activity against the ten isolates of *Fusarium proliferatum* (Fm) in dual culture test. Among the isolates *Penicillium* sp. SCB-EF13 showed moderate antagonism covering more than two-thirds of the mycelial growth of all Fm isolates suggesting a potential biological control. The ability of this promising EF to grow rapidly, overlapped the growth of the pathogen and compete with space and nutrients were the common modes of antagonism observed. On the other hand, from the twenty medicinal plant extracts (MPE) assayed against the same pathogen, only three crude extracts of MPE at 10,000 ppm exhibited zones of inhibition using agar well diffusion method. They were *Chrysanthemum indicum*, *Piper betle* and *Pseuderanthemum reticulatum* that showed antagonistic potentials against all isolates of *F. proliferatum* with high average inhibition zones of 24.67mm, 24.33mm and 31.67mm, respectively where the latter was comparable to the average inhibition zone expressed by the positive check, Difenoconazole (fungicide; recommended rate of 3 g or ml/liter of water). Other MPE with potential promising inhibitory effects on Fm were *Blumea balsamifera*, *Annona squamosa*, *Morinda citrifolia*, *Cymbopogon citratus* and *Artemisia vulgaris*. Antifungal property of these promising MPE was perhaps attributed to active secondary metabolites. Analysis of Variance (ANOVA) of independently conducted experiments showed highly significant results at $P < 0.05$. Comparing statistically the performance of utilization of endophytic fungi with that of the plant extracts, however, was not done.

Keywords: endophytic fungi, medicinal plant extract, *Fusarium proliferatum*





Isolation and characterization of plant growth-promoting diazotrophs from soil, plant rhizosphere, and plant roots collected from salt-affected areas in the Philippines

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Soil salinity is the major cause limiting plant productivity worldwide. This study focused on the isolation and characterization of diazotrophs present in saline soils, plant rhizosphere and plant roots which can be used to enhance the growth of crops on saline soil. A total of 247 nitrogen-fixing bacteria were isolated from soil, plant rhizosphere and plant root samples obtained in four different sites in the Philippines namely Pampanga, Batangas, Iloilo, and Palawan. Of the 247 isolates, 147 were selected on the basis of their colony characteristics, cell shape, and gram reaction. The plant growth-promoting activity (ACC deaminase, IAA production, and siderophore production) of the isolates were tested. Ten isolates, D7.3RS, DS8.1RS, SDSO11.2, and AS6.3ES isolated from loose soil, AS10.2GPR, BAPR5.1, BBVPR9.2, and BPRP18.1 isolated from rhizospheric soil, and DGE17.2, and AGE18.1 isolated from plant roots, tested positive in all or most of the plant growth promoting traits. The corn (*Zea mays*) seedling, specifically the Institute of Plant Breeding variety 11 (IPB var 11), a saline-tolerant variety of corn, treated with AS10.2GPR showed the highest percent increase with respect to the uninoculated, no-nitrogen control (71.78%), followed by BPRP18.1 (69.59%). Pairwise comparison of the means of the biomass of seedlings treated with these isolates with respect to the uninoculated control showed a significant difference ($p < 0.05$) using Fisher's Least Significant Difference (LSD) test. BLAST analysis of the *16S rDNA* showed that the microbial isolates were highly similar to members of *Enterobacter* sp., *Serratia* sp., *Pseudomonas* sp., *Haererehalobacter* sp., *Salinicola* sp., and *Mangrovibacter* sp. Isolates AS10.2GPR and BPRP18.1 were identified to be *Serratia* sp. *NH10* and *Enterobacter sacchari* strain *HX148S*, respectively. The *nifH* gene was detected in eight out of the ten selected isolates. Further studies on the plant growth promotion by these isolates on other crops can be done.

Keywords: soil salinity, diazotroph, 1-aminocyclopropane-1-carboxylic acid (ACC) deaminase activity, indole acetic acid (IAA), siderophore production, phosphate solubilization, BLAST, NifH





Supporting for Identification of Sample DL0038A & DL0038B by the Prediction of Secondary Structure of Nuclear Ribosomal Large Subunit (*nrLSU*) Region

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Background: Many fungi belonged to the ascomycetes genus *Cordyceps*, entomopathogenic fungi have long been used as traditional herbal medicine in various Asian countries, including Vietnam with the height of known species diversity. In our previous study, we have tentatively concluded that our fungal specimens DL0038A and DL0038B, which were collected at the Langbian mountain, Vietnam, were *Cordyceps takaomontana*. In order to further support the morphological identification, we continued to predict the secondary structure of nrLSU (nuclear ribosomal large subunit) DNA sequences.

Methods: Pure mycelia were obtained from PGA cultured DL0038A and DL0038B. DNA was extracted by phenol/chloroform method. Then, PCR assay was carried out to amplify *nrLSU* region by LR0R (Forward primer) and LR5 (Reverse primer). The PCR product was to be sequenced and confirmed proofreading before the molecular phylogenetic analysis. For secondary structure prediction, Mfold 3.1 software was used to predict the secondary structures of *nrLSU* sequences. The maximum delta G (ΔG) was included to support the secondary structure.

Results: Our result have shown that the secondary structure of sense and antisense of DL0038A and DL0038B were similar to the structure of reference sequence, *Cordyceps takaomontana* (AB044637). In detail, the structure of sense sequence formed a special structure with three large stem loops interleaved with many small stem loops with the same ΔG : (DL0038A ($\Delta G = -108.71$ Kcal/mol), DL0038B ($\Delta G = -105.74$ Kcal/mol), *Cordyceps takaomontana* (AB044637) ($\Delta G = -109.41$ Kcal/mol). The structure of anti-sense sequence also formed the similar structure within two large stem loops with the same ΔG : (DL0038A ($\Delta G = -105.74$ Kcal/mol), DL0038B ($\Delta G = -105.35$ Kcal/mol), *Cordyceps takaomontana* (AB044637) ($\Delta G = -105.85$ Kcal/mol).

Conclusion: We concluded that the prediction of the *nrLSU* secondary structures reconfirmed the DL0038A and DL0038B, identified as *Cordyceps takaomontana*. Therefore, this research will be applied with larger number of samples in near future.

Keywords: *Cordyceps takaomontana*, *nrLSU*, molecular phylogenetics, secondary structure prediction





Identification of Vip3A proteins in *Bacillus thuringiensis* isolates in Vietnam

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Bacillus thuringiensis (*Bt*) is a Gram-positive, spore-forming bacterium that can synthesize parasporal crystalline inclusions containing Cry and Cyt proteins, some of which are toxic against a wide range of insect orders, nematodes, and cancer cells. These toxins have been successfully used as bioinsecticides against caterpillars, beetles, and flies, including mosquitoes and blackflies. *Bt* also synthesizes insecticidal proteins that are subsequently secreted into the growth during the vegetative growth phase. These proteins are commonly known as vegetative insecticidal proteins (Vips) and hold insecticidal activity against *lepidopteran*, *coleopteran* and some homopteran pests. Therefore, it is important and urgently to isolate and identify toxic genes, especially Vip genes. In this study, we performed the presence of vip3A genes in ten *Bacillus thuringiensis* (*Bt*) isolates using PCR method with specific vip3A primers. These *Bt* isolates were collected from different regions in Vietnam and checked their biochemical tests as well as the presence of spores and crystal proteins. Five of ten *Bacillus thuringiensis* (*Bt*) isolates showed 3.4 kb band as the control (*Bacillus thuringiensis* var *kurstaki*) indicating the potential presence of Vip3A proteins in such *Bt* isolates. Additionally, the expression of Vip3A proteins was also conducted by using the SDS-PAGE method with five *Bt* isolates including VP9.1, VP10.1, TG5.1, TG6.2 and BT 10.2 that showed the presence of vip3A gene as shown above. The results indicated that all of them expression the target protein with 66 kDa as shown in positive control resulting potential application of these *Bt* isolates containing Vip3A protein for controlling pests in crops.

Keywords: *Bacillus thuringiensis*; Vip3A gene; crystal proteins, spores, SDS-PAGE





Effect of Agitation Speed on Chitin-degrading Enzyme Activity by *Pseudomonas stutzeri* PT5 on Fermentor

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This study aimed to know the effect of agitation speed in fermentation of *Pseudomonas stutzeri* PT5 on chitin-degrading enzyme activity and to determine the optimum agitation speed on the production of chitin-degrading enzyme using fermentor. Chitin-degrading enzyme was produced by a liquid fermentation method using 2 L fermentor with liquid chitin medium added with maltose (0.1%) and ammonium phosphate (0.1%). The condition of pH 6, temperature 37°C, aeration 1 vvm and variation of agitation speed (200, 350, 500 rpm) were maintained during 48 hours of fermentation. The observation was conducted every 8-hour. The design used was a completely randomized design with three replication. Data were analyzed by one way ANOVA test followed by DMRT test. The result showed that agitation speed has an effect on chitin-degrading enzyme activity, N-acetylglucosamine concentration on the medium and cells aggregate. Based on the experiment result, the most optimum agitation speed for producing chitin-degrading enzyme using fermentor was 350 rpm with chitinase activity reached 0.0051 U/ml and N-acetylglucosamine concentration was 35.161 µg/ml on 24-hour fermentation. Chitin-degrading enzyme activity produced by a fermentor from this experiment was a 4 times higher than the enzyme produced by a shaken-flask method.

Keywords: agitation speed, chitinase activity, fermentation, fermentor,
Pseudomonas stutzeri PT5





Molecular cloning, expression and characterization of a cDNA encoding MKS2-like protein from soybean (*Glycine max*)

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2-Methylketones found in plants have multiple potential functions including as defense compounds to protect plants from pests and as feedstocks for biofuel production. Methylketones have not only favourable cetane numbers but also lower hydrophilicity and melting points than fatty acids which have been used in biodiesel production. Two key enzymes of methylketone biosynthetic pathway, which were first identified in wild tomato *Solanum habrochaites*, include methylketone synthase 1 (ShMKS1) and methylketone synthase 2 (ShMKS2). ShMKS2 has a thioesterase activity, hydrolyzing the thioester bond in 3-ketoacyl-ACPs (plastid-localized intermediates of fatty acid biosynthesis) to produce the corresponding 3-keto acids, and ShMKS1 has a decarboxylase activity towards 3-keto acids, resulting in the production of 2-methylketones. As methylketones were shown to have various applications depending on their carbon chain length, there has been great interest in looking for new MKS2 natural variants each of which is capable of hydrolyzing a specific 3-ketoacyl-ACP with respect to carbon chain length from various plant species.

Taking advantage of the recently released soybean genome sequence, we conducted a genome-wide search for the homologues of *ShMKS2* in *Glycine max* and identified a gene encoding for a protein of 23.5 kDa with more than 50% identity to previously reported ShMKS2 sequence. The gene is designated as GmMKS2. A full-length *GmMKS2* cDNA was isolated from the immature seeds of soybean and sequenced. The deduced amino acid sequence contains a conserved aspartate residue essential to the catalytic core of the hotdog-fold thioesterases like ShMKS2 as shown in protein sequence alignment.

To characterize the activity of GmMKS2 protein in *E. coli*, we constructed an ORF of GmMKS2 that began at Met-65 to remove the entire transit peptide of GmMKS2. The ORF was cloned into pETDuet-1 vector and the resulting vector was transformed into *E. coli* C41(DE3) and induced for expression in this host strain by 0.25mM IPTG at 18°C for 16 hours. Gas chromatography - flame ionization detector (GC-FID) analysis of culture medium of *E. coli* C41 (DE3) expressing GmMKS2 following heat - activated decarboxylation revealed the production of odd-chain methylketones ranging from C7 to C13 with 2-heptanone (C7) being the most abundant. This suggests that their corresponding 3-keto acid ranging from C8 to C14 were present in the culture medium. These results are consistent with *in silico* analysis predicting that GmMKS2 is a 3-ketoacyl-ACP thioesterase: it could hydrolyze endogenous 3-ketoacyl-ACPs in *E. coli* to generate 3-keto acids in the culture medium, and thus leads to the formation of the 2-methylketones after decarboxylation.

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Keywords: 3-ketoacyl-ACP thioesterase, 2-methylketones, methylketone synthase 2, *Glycine max*





Use of PCR protocol to detect animal-derived ingredients in vegetarian foods

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The consumption and demanding of vegetarian foods have seen a dramatic increasing, thus, it is required to have a higher standard in foods production, which is remained to be without any animal-derived ingredients in vegetarian food processing. Thus, to verify food ingredients is of great importance in order to ensure for whether or not contains animal-derived ingredients, it is necessary to establish a rapid method for detecting meat ingredients in processed vegetarian foods. The PCR assay, based on the amplification of mitochondria cytochrome b by primer Cyto-F and Cyto-R, that are specific to detection of the animal-derived ingredients presence was designed. Notably, Cyto-F and Cyto-R were designed to be specific to most of animals. A total of 50 vegetarian samples, which were collected from local market and supermarket, were enrolled into current study. As the result, 17/50 (counting for 34%) vegetarian samples were positive, that meant having animal-derived ingredients in their foods processing. In addition, representative PCR products were conducted sequencing, that showed the animal-derived ingredients originated from *Sus scrofa* and *Gallus gallus*. According to these sequencing results, the specificity of amplification of Cytochrome b was confirmed. In summary, we affirmed that cytochrome b – primer-specific amplification in current study could be to apply on the large number of samples in the reality.

Keywords: *Cytochrome b*, vegetarian food, PCR, animal-derived ingredients





Rapid and sensitive detection of *Burkholderia pseudomallei*, *Burkholderia thailandensis* and *Burkholderia vietnamiensis* by loop mediated isothermal amplification (LAMP) assay

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Gram-negative *Burkholderia* bacteria of which *B. pseudomallei* has been known as an opportunistic human pathogen causing melioidosis so called “Whitmore disease “. Although *B. thailandensis* and *B. vietnamiensis* rarely cause human disease, cases of disease have been documented when they are exposed at high dose. Due to the high mortality and virulence in human, the need for rapid and accurate detection method of those *Burkholderia* bacteria is necessary for potential uses in diagnostic and environmental testing. Additionally, the genomes of *B. thailandensis* and *B. vietnamiensis* are closely related to that of *B. pseudomallei* resulting the difficulty in distinguishing among these *Burkholderia* species. Many previous efforts have been conducted for specific detection of *B.pseudomallei* by using realtime PCR. However, this approach requires well-equipped lab with high-skilled personnel, labour and time consuming. Therefore, we report an alternative method so-called loop-mediated isothermal amplification (LAMP) that can allow less than 30 minutes in the detection of three *Burkholderia* species. For this purpose, specific LAMP primers for the detection of each *Burkholderia* species were designed by LAMP designer software of Optigene (<http://www.optigene.co.uk/lamp-designer/>), then followed by validating those primers using Vector NTI software (Thermo Scientific, US) and *in silico* PCR amplification. The LAMP assays were conducted with Master mix (Optigene, UK) at the single temperature of 63⁰C for 30 minutes. Samples tested in this study, were genomic DNA of 5 *Burkholderia* species including three *B.pseudomallei* strains (MSH487, B11, K9), one *B.mallei* strain (ATCC), one *B. thailandensis*, one *B. vietnamiensis*, and two *B.cepacia*. All experiments were carried out by using PCR and GenieIII detection units. The results indicated that LAMP based detection method can distinguish three species of *Burkholderia* (*B.pseudomallei*, *B.thailandensis* and *B.vietnamese*) but not *B.mallei* due to cross amplification caused by high similarity of genomic sequences between *B.pseudomallei* and *B.mallei*. This study provide potential development of new and promising diagnostic system for on-site detection of bacterial pathogens in human and environment.

Keywords: *B. pseudomallei*, *B. mallei*, *B. thailandensis*, *B. vietnamiensis*, melioidosis, opportunistic pathogens, loop-mediated isothermal amplification (LAMP) assay





Development of Sequence Characterized Amplified Region (SCAR) Marker for Identifying Virulence of The Rice Blast Fungus, *Magnaporthe oryzae*

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Magnaporthe oryzae is a causal agent of blast disease, one of the most devastating disease in rice resulting significant crop losses worldwide. Understanding of fungal virulence and its markers have been known to play important role of the improvement of strategies in management of the rice blast disease. In previous study, genetic characterization using sequence-related amplified polymorphism (SRAP) and fungal virulence were performed with 30 *M.oryzae* isolates collected from rice fields in South, Central and North Vietnam. The results of this experiment indicated blast isolates collected in Binh Thuan province, Vietnam (BT samples) can break-down the blast resistance in Thai and Vietnamese rice varieties, JHN and IR50404 respectively. Herein, approximately 400 bp of specific band was observed in at least four BT samples but not in others that do not involve in the break-down of blast resistance in rice varieties when using the pair of primers, me3 and em4. The sequencing of specific band indicated the presence of partial sequence of AVR-Pita2 gene and was used to design primers of SCAR markers. Re-amplification of SCAR primers was conducted to generate trait specific fingerprint linked to the blast resistance break-down of *M.oryzae* isolates in rice by PCR method. The results showed that 400 bp specific band was observed only in BT samples but not in other *M.oryzae* isolates indicating a potential application of SRAP-SCAR marker for screening of DNA fingerprinting and for controlling blast disease in the field.

Keywords: Rice blast, *M.oryzae*, SCAR marker, SRAP





Evaluating DNA integrity for human sperm cryopreservation by Sperm chromatin structure assay (SCSA)

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Background: Human sperm cryopreservation can have detrimental effects on sperm DNA integrity, as well as semen quality. It can decrease mobility and viability of human sperm which have been reported in previous studies.

Objective: To evaluate the impacts of our in-house cryopreservation method on human sperm DNA integrity, mobility and viability.

Methods: Thirty fresh semen samples were evaluated sperm quality according to World Health Organization (2010). After semen analysis, each sample was mixed 1:1 with freezing medium (SpermFreeze, Fertipro, Belgium). The mixture was aspirated into sterile cryotube. After incubated for 10 minutes at room temperature for equilibration, the cryotube was placed in horizontal position at 15cm above and parallel to the surface of liquid nitrogen for 15 minutes to cool down to -80°C and then plunged into liquid nitrogen (-196°C) for storage. Thawing of spermatozoa was carried out slowly at 37°C or room temperature and then mobility, viability and DNA integrity were determined. Determination of sperm DNA integrity using SCSA procedure described by Evenson et al (2000), each sample was diluted to a final concentration of 2×10^6 sperm/ml with TNE buffer. This cell suspension was treated with an acid detergent solution (pH 1.2) for 30 seconds, and then stained with 6 mg/l of purified Acridine Orange. Approximately 20,000 cells of each sample were measured by a flow cytometer.

Results: No significant difference in DNA fragmentation was found between before and after cryopreservation ($12.40 \pm 7.32\%$ vs $10.77 \pm 6.73\%$; $p = 0.25 > 0.05$). However, the mobility and viability of post-thawed sperm after cryopreservation were significantly reduced ($p < 0.05$) (from $49.02 \pm 9.02\%$ to 26.20 ± 8.46 and from $78.10 \pm 8.93\%$ to 31.30 ± 8.31 , respectively). Besides, the percentage of cryosurvival rate of sperm was decreased to $53.57 \pm 15.10\%$.

Conclusion: Our in-house cryopreservation method had not significantly detrimental effect on sperm DNA integrity. However, sperm mobility and viability were reduced by half between before and after freezing.

Keywords: human sperm cryopreservation, SCSA, DNA integrity, mobility, viability





Potential Bacteria as Biological Control and Biological Fertilizer Used in Agricultural Applications

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The overuse of chemical pesticides or fertilizer in agriculture has also created environmental problems, such as air pollution, reduced biodiversity, and suppressed ecosystem function. Current effort to control plant disease mostly is emphasized on the use of resistant varieties, however, the pathogens strains that differ among seasons and among locations could cause resistant plant varieties cannot survive longer. Therefore the need for development of non-chemical alternative methods to control plant diseases is necessary. Among biological control agents, bacteria such as *Pseudomonas*, *Serratia*, and *Bacillus* had shown activities in suppressing fungal infection. We have developed formulation of bacterial consortium (*Bacillus firmus*, *B. cereus*, *Pseudomonas aeruginosa*, and *Serratia marcescens*) to control multiple rice diseases, such as sheath blight, neck blast, and bacterial leaf blight, as an alternative biological control strategy. The formulation of bacterial consortium using talcum as carrier agent showed to reduce sheath blight lower than control without application at field experiments in West Java. The formula also showed better effect to reduce the length of lesion of bacterial blight diseases which had inhibition percentage 45.76% compared to other treatments and control without application. *In vivo* tested of the formula showed reduction of neck-blast attack (39.5%) compared to control treatment without application (79.47%). Application of the formula on rice fields at South Sulawesi influenced to reduce intensity to leaf blast disease, neck blast, bacterial leaf blight, and sheath blight. The formula also produced higher of grain in the range 615.16 g / 25 clumps or conversion of grain yields in the range 3.89 tons/Ha. Viability test of the formula showed stability as much as 10^8 - 10^9 cfu/ml up to three months. In conclusion, the bacterial consortium formula could control single or multiple rice diseases and tend to improve rice yield may have been due to indirect effect of antagonism as well as competitions with pathogens for essential nutrients.

Keywords: bacterial formulation, field experiments, microbial pathogens





Promoter Motif Differences Cause Different Expression Level of *OsGERLP* Gene in Aluminum Tolerant and Sensitive Rice

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A rice gene encoding a protein containing ribosomal L32-like motif (*OsGERLP*) is suggested as one of the aluminum (Al) tolerance gene in rice. The expression of the gene is induced by Al and the gene expression level under Al stress in Al-tolerant rice (cv. Hawara Bunar) is higher than that of Al-sensitive rice (cv. IR64). The expression differences might be due to the differences in promoter sequences. This study aimed to analyze the promoter sequence of the *OsGERLP* gene in rice cv. Hawara Bunar and IR64. We characterized multiple *cis*-elements in the *OsGERLP* promoter using bioinformatic tools, cloned the promoter using Gateway system, and constructed *OsGERLP::GFP* to analyze the expression differences. In this study, we found that *OsGERLP* gene promoter sequences of rice cv. Hawara Bunar was different with that of cv. IR64 in 15 SNPs. The promoter isolated from cv. Hawara Bunar had more motifs related to abiotic stresses and pathogens than that of cv. IR64. Green Fluorescent Protein expression level in Hawara Bunar driven by *OsGERLP* promoter is also higher compared to IR64. It was suggested that the promoter sequence differences cause the differences in the *OsGERLP* gene expression in both cultivars.

Keywords: aluminum, gene, promoter, tolerance





Variation of Iron Tolerance Level and Mechanism of Several Rice Genotypes

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Plant tolerance mechanism to iron (Fe) toxicity involve complex physiological processes and depend on the genotypes. The objectives of the research were (1) to analyze growth response of several rice genotypes to Fe toxicity, and (2) to analyze the relationship between rice growth characters and tolerance mechanism to Fe toxicity. The research was conducted in 2 experiments. The 1st experiment was conducted to compare the effectiveness of three nutrient culture media (YHS, YHSA, and YFSA) to distinguish between Fe-tolerant (Mahsuri) and -sensitive (Inpara 5) genotypes in response to Fe toxicity. The 2nd experiment evaluated ten rice genotypes (IR64, IRH108, Danau Gaung, Hawara Bunar, Indragiri, Pokkali, Mahsuri, Inpara 2, Inpara 5 and Inpara 6) grown in nutrient culture media that were treated with and without 400 ppm Fe. To distinguish the tolerance mechanism of those rice genotypes to Fe toxicity, plant growth characters, leaf bronzing score (LBS), Fe content, and physiological responses were observed. The result showed that Yoshida half strength with agar 0.2% (YHSA) medium and 400 ppm FeSO₄·7H₂O for 10 days stress was effective and efficient nutrient culture medium to analyze growth response and tolerance mechanisms of rice to iron toxicity. The LBS-based rice responses to Fe stress was divided into 2 response groups, i.e.: early response (LBS1-LBS6) and late response (LBS7-LBS10). Fe toxicity decreased shoot and root growth characters. The Fe content in root plaque and shoot varied among genotypes. Analysis of relationship between genotypes and growth characters divided the genotypes into 3 groups of Fe-tolerance mechanism, i.e.: sensitive-includer (IR64 and Inpara 5), tolerant-includer (Danau Gaung, Inpara 6, Inpara 2, Mahsuri, and IRH108), and tolerant-excluder (Hawara Bunar, Indragiri, and Pokkali).

Keywords: genotype, iron tolerance, rice





Morpho-physiological Characteristics of Indonesian Rice Cultivar under Drought Stress Condition

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Drought stress causes the changes in anatomical, morphological and physiological characters of rice, which affects rice growth and production. The objectives of the research was: (1) to do a rapid test for selecting genotypes that were tolerant to drought stress, (2) to validate the rapid drought stress selection test to several rice genotypes, and (3) to study the morphological and physiological characters of rice genotypes to drought stress. This research used 15 rice genotypes consisting of 8 upland rice cultivars, 6 Indonesian local varieties, and c.v. IR64 as a drought-sensitive control cultivar. Polyethylene Glycol (PEG) 6000 was used to adjust water potential of 0, -0.19, and -0.67 MPa for drought stress simulation. Morphological and physiological analyzes were conducted on 14-day-old plants after being grown on PEG-6000 nutrient solution. Observations were carried out at 3, 5, and 8 days after treatment. The results showed that the characters that was positively correlated between early stage screening and IRRISSES is the seedling root-shoot length ratio. Based on the early stage screening technique we chose three rice genotypes, i.e.: INPAGO10, Hawara Bunar, and Situ bagendit for further study. In addition, IR64 also included as a drought-sensitive control. Morphological analysis showed that Hawara Bunar was the most tolerant to drought. It has the longest root and lowest leaf rolling under drought stress. Physiological analysis showed that leaf relative water content was highest in Hawara Bunar, followed by INPAGO10, Situ Bagendit, and IR64, respectively. Proline concentration of the leaves showed similar level between Hawara Bunar and INPAGO10, and was higher than that of the other two genotypes. Hawara Bunar also had lower levels of lipid peroxidation and chlorophyll breakdown than that of the other genotypes. Interestingly, Hawara Bunar, INPAGO10, and Situ Bagendit showed no significant different of the leaf ABA concentration under drought stress. The results are very interesting to reveal the tolerance mechanism of rice under drought conditions at the level of molecular physiology.

Keywords: drought stress, rice, early screening, abscisic acid (ABA)





Haploid *in vitro* from Anther Culture in traditional tomato *Solanum lycopersicum* L.

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Tomato (*Solanum lycopersicum* L.), belongs to *Solanaceae*, is one of the most important vegetables in the world. Considered to the breed of tomato, it is based on the self-pollinated homozygous generation, takes a long time in production.

Traditional tomatoes were collected from the botanic garden at Ho Chi Minh City Open University. In current research, many conditions, including the effect of size of buds (3-4 mm, 5-6 mm and 7-8 mm), 7 °C-treatment duration (0, 24h, 48h, 72h), phytohormones concentration changes (BA 0-1 mg/L and NAA 0-1 mg/L) in MS medium, were used in finding the optimal condition for the induction of callus from anther culture. Besides, the BA levels (0 – 2 mg/L) was experimentally studied for the regeneration shoots of callus from anther culture in MS medium. Finally, the growth of plantlets from shoots *in vitro* in MS with NAA (0-0.9 mg/L), active carbon (0-4 g/L), casein (0-400 mg/L) or reduce sucrose (30-0 g/L) were studied.

The results showed that, after one month cultivation, callus induction was obtained 30% from 5-6 mm-long buds, after pre-treatment at 7 °C, 24 hours, in MS medium contain 0.2 mg/L BA and 0.2 mg/L NAA. Rapid proliferation shoots *in vitro* from callus with 13 shoots/callus have effect in MS with BA 1.5 mg/L. Photoautotrophic culture *in vitro*, sucrose concentration down from 30 g/L to 15 g/L to 0 g/L, have 100% live plantlets when move to *ex vitro*. One haploid tomato line seeds are obtained from androgenesis method after 3 months.

Keywords: haploid, anther culture, homozygous, phytohormones, callus, plantlet, photoautotrophic





***In vitro* propagation of Ginger (*Zingiber officinale* Rosc.)**

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Background: Ginger (*Zingiber officinale* Rosc.) is the perennial herb, which has been commercially cultivated and medical used in most tropical regions of the world, including Vietnam. Notably, the underground rhizomes have been used as planting material, resulting in high-cost of seeding and without the disease clearance. Notably, in Vietnam, there are still limit researches, that associated to the *in vitro* propagation of the ginger. Therefore, the aim of current study was to establish the protocol and guidance for *in vitro* culture of Ginger.

Methods: Regeneration from apical meristem, regeneration by thin layer of cells, regeneration by protocorm creating indeterminate shoots were enrolled in current study to study the protocol for *in vitro* culture of Ginger.

Results: A simple protocol was

described for high frequency plant regeneration from protocorm, which was isolated from shoot meristem tip embryogenic callus of Ginger. The protocorm was successfully divided to form bud differentiation by culturing in MS Agar in which protocorm were embedded in 8 g/l gellan gum MS medium, which contains 1.0 mg/l NAA, 3.0 mg/l BA, 20g/l sucrose. For the continuous growth well of the bud differentiation, it was essential to add 1% (v/v) fresh coconut water in the liquid reservoir medium from the later stage of the culture. Moreover, in this culture condition, the buds started to growth into *in vitro* seedlings of culture. The regenerated plants were successfully grow in to the greenhouse and showed normal morphology. Moreover, the survival rate of seedlings in the nursery stage in the first months reached to 95%.

Conclusion: The results of this study have improved ginger *in vitro* propagation. Successfully researched *in vitro* ginger seedlings and can produce on orders.

Keywords: *Zingiber officinale* Rosc, propagation, protocorm, *In vitro*





Karyotype Variability in Species of the Genus *Zephyranthes* Cultivated in Yogyakarta, Indonesia

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The objective of this study were to identify chromosomal variability in three most cultivated species of the genus *Zephyranthes*, in Yogyakarta. In this work, the cytotaxonomic implications of the chromosomal characterization of cultivated *Zephyranthes* species described in Yogyakarta, Indonesia were studied. The genus of *Zephyranthes*, represented by about 60 species is one of the evolutionary dynamic genera of the family *Amaryllidaceae*. In Indonesia, especially Yogyakarta, the genus of *Zephyranthes* were cultivated for its ornamental and medicinal properties. *Zephyranthes* species used in this work were characterized by the color of the flower. Somatic metaphase of chromosomes cells were prepared according to the Sharma and Gupta method with minor modification and stained using 1% *acetocarmine*. Karyotype data were analyzed in the number, short and long scales, absolute scales, scales ratio of chromosomes. Homologous chromosomes were arranged by scatter diagram (Scatter Plot). For karyotypical symmetry were determined by chromosomal asymmetry indexes.

All individual karyotype formed by a set of metacentric chromosomes. In addition to submetacentric and acrocentric chromosomes. In *Zephyranthes candida* with white flower, $2n = 38$ were observed, by karyotype formula of $2n = 14M + 24SM$ in somatic cells, and showed a ploidy level of diploid. *Zephyranthes rosea* with pink flower showed chromosome with $2n = 24$ in somatic cell with karyotype formula of $2n = 4M + 16SM + 4A$, and it showed a ploidy level of tetraploid. *Zephyranthes ajax* with yellow flower showed $2n = 42$ in somatic cell chromosome with karyotype formula of $30M + 12SM$, and formed a ploidy level of hexaploid. The most asymmetrical karyotype showed in *Z. rosea* and *Z. candida*. The most advanced evolution were showed by *Z. rosea* by the most asymmetrical karyotype, while the advanced evolution also showed by *Z. candida* considered by variation in chromosomes size. The chromosome count and karyotype from previous research were confirmed. Taken together, data from this study partially confirm previous counts for epithets and further enhance the cytological variability data previously reported for the genus.

Keywords: *Zephyranthes*, chromosome, karyotype, variability, evolution





Extensions of the Fibonacci-Lucas relations

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Let F_n and L_n be the Fibonacci and Lucas numbers, respectively. Sury [3] proved the Fibonacci-Lucas relations

$$2^{m+1}F_{m+1} = \sum_{i=0}^m 2^i L_i,$$

and Marques [1] found an analogue identity that replaces 2 by 3.

Elhameed and Zeyada [4] extended the above identities on the Fibonacci and Lucas numbers to the generalized Fibonacci and Lucas numbers.

Recently, Martinjak [2] gave an identity with alternative signs:

$$\sum_{i=0}^m \frac{(-1)^i}{2^i} L_{i+1} = \frac{(-1)^m}{2^m} F_{m+1} \quad (1)$$

In this project, we will generalize the identity (1). to the generalized Fibonacci numbers.

Keywords: Fibonacci numbers, Lucas numbers

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Preparation and Characterization of Clay-Rhodamine B Nanopigments

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In this research, montmorillonite-Rhodamine B (MMT-RhB) nanopigments were prepared via the ion-exchange reaction with the variation of Rhodamine B concentrations in aqueous solution. Nanopigments were investigated by Fourier Transform-Infrared Spectroscopy (FT-IR), X-ray Diffraction (XRD), Thermogravimetric Analysis (TGA) and Elemental Analysis (EA, CHN-mode). 60% CEC of Rhodamine B in aqueous solution is the optimal concentration that maximized exchangeable Rhodamine B into the montmorillonite interlayer with the d-spacing of 18.8 Å. Based on the elemental analysis, the amount of Rhodamine B was equal to 29.1% CEC of montmorillonite. The orientation of Rhodamine B in the interlayer is related to dimeric aggregation. Thermogravimetric analysis suggests the improvement in the thermal stability of the intercalated dye. In addition, nanopigments were used for the dyeing process on cotton fabrics. Scanning Electron Microscopy (SEM) shows the sheet-like structure of nanopigments adsorbed on cotton fabrics.

Keywords: Nanopigments, Rhodamine B, Montmorillonite, ion-exchange, thermal stability





Revealing the Structural and Dynamic Properties of Betaine Aldehyde Dehydrogenase 2 from Rice (*Oryza sativa*): Simulation Studies

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Betaine aldehyde dehydrogenase 2 (BADH2) is an enzyme that inhibits the accumulation of 2-acetyl-1-pyrroline (2AP), a potent flavor compound in rice fragrance. The malfunction of BADH2 results in the increase in rice fragrance. BADH2 contains three domains: NAD-binding domains (225 amino acids), substrate-binding domains (201 amino acids), and oligomerization domains (28 amino acids). The previous studies revealed that Tyrosine 420 (Y420) exhibits a less catalyzed efficiency toward γ -aminobutyraldehyde but not for betaine aldehyde. Cysteine 294 (C294), Glutamic acid 260 (E260), and Asparagine 162 (N162) are found to be important for BADH2 function. To date, the understanding on BADH2 structure and function are unclear. An insight into the nature of BADH2 can serve as one of key starting points for the production of high quality fragrant rice. In this study, we therefore constructed the homology model of BADH2 and employed 500-ns Molecular Dynamics simulations (MD) to primarily understand the structural and dynamic properties of BADH2. Initially, the Ramachandran plot confirms the good quality of modeled protein structure. Principle Component Analysis (PCA) was also calculated to capture the protein dynamics. Among 3 domains, the results show that NAD binding and substrate binding sites are the most flexible. Moreover, the roles of key amino acids (N162, E260, C294, and Y419), on protein structure and function are investigated.

Keywords: betaine aldehyde dehydrogenase 2, fragrant rice, homology modeling, molecular dynamics simulations





Synthesis and Characterization of Oligopeptides for Bone-Targeted Liposomal Delivery

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Liposomes have been widely used as nanocarriers for therapeutic and cosmetic applications. Modification of liposome surface using targeting moiety could increase specificity of the nanoparticles, leading to enhanced efficacy of encapsulated agents. The aim of this study is to design a targeting moiety that is specific to bone cells. Peptides composed of aspartic acids or glutamic acids are the targeting moiety of interest as they showed high affinity to hydroxyapatite, the mineral predominantly found in bones. In this study, synthesis of the oligopeptides was carried out on resin using standard Fmoc procedure. HBTU and HOBt were used as coupling agents. The resulting compounds were recrystallized, freeze-dried, and further purified by semi-preparative HPLC. The purified compounds were characterized by mass spectrometry and their purities were verified by reversed-phase HPLC. Binding of the oligopeptides to hydroxyapatite was performed in buffer (pH 7.4) at 37 °C. The unbound oligopeptide was analyzed by UV-Vis spectroscopy.

Keywords: Oligopeptide, Solid-phase peptide synthesis, Hydroxyapatite, Binding, Targeting moiety





Quantitative Analysis of Cellulose from Mulberry Plants

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Cellulose is one of the most widely used natural substances and has become one of the most important commercial raw materials. It is used to make paper, film, and plastics. This study is extracting cellulose from Mulberry Plants with different species. Cellulose is isolated by acid hydrolysis method. Furthermore, quantities of lignin, ash and extractive have been analysed. The results show that cellulose in the part of branches of all species is not much different. For the part of leaves, Yai Boroh species has much more cellulose than others and more fresh leaves have less cellulose too.

Keywords: Cellulose, Mulberry tree, Morus, Acid hydrolysis





Effect of Copper-promoted Silica Supported Cobalt Catalysts on Product Selectivity from Fischer-Tropsch Synthesis

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Un-promoted and copper-promoted cobalt catalysts supported on Aeroperl 300/30 were studied on their catalytic activity in Fischer-Tropsch synthesis (FTS). All catalysts were prepared by the impregnation method with the mass ratio (Cu : Co : Aeroperl 300/30) of 0, 10, 20 : 20 : 100 and characterized by X-ray diffraction (XRD) and X-ray absorption spectroscopy (XAS). Their catalytic activity testing was performed in a fixed bed reactor with the feeding H₂ and CO at the volume ratio of 2 : 1 at 190°C and 10 bar. The FTS products were analyzed by gas chromatograph (GC) in terms of the C₁, C₂-C₄, C₅-C₁₂, C₁₃+ and methanol selectivity.

Keywords: catalyst for Fischer-Tropsch synthesis



Synthesis of Benzoxepino[3,2-*b*]pyridine via Nucleophilic Aromatic Substitution

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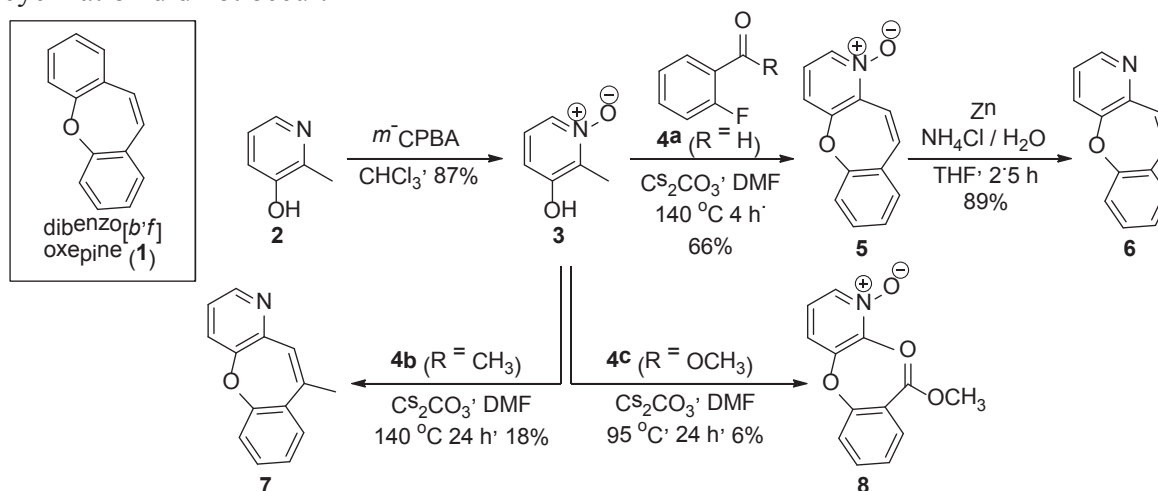
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Dibenzo[*b,f*]oxepine (**1**) and its derivatives can be found as a core structure of many biologically active natural products. Although benzoxepino[3,2-*b*]pyridine (**6**) is structurally similar to dibenzo[*b,f*]oxepine (**1**), the synthesis and biological evaluation of this compound have never been reported. We are interested in synthesizing both benzoxepino[3,2-*b*]pyridine (**6**) and its *N*-oxide derivative (**5**) since many natural products are in the *N*-oxide form. The benzoxepino[3,2-*b*]pyridine-*N*-oxide (**5**) could be synthesized in moderate yield from 2-methyl-3-pyridine-*N*-oxide (**3**), which was prepared from the oxidation of 2-methyl-3-pyridinol (**2**), and 2-fluorobenzaldehyde (**4a**) by nucleophilic aromatic substitution and Knoevenagel condensation. Reduction of this *N*-oxide derivative yielded the desired benzoxepino[3,2-*b*]pyridine (**6**) in good yield. In addition, we tried to synthesize benzoxepino[3,2-*b*]pyridine's derivatives by changing aldehyde **4a** to 2-fluoroacetophenone (**4b**) and methyl-2-fluorobenzoate (**4c**). However, only **4a** gave desired product **7** whereas **4b** gave undesired product **8**, which cyclization did not occur.



Keywords: Benzoxepino[3,2-*b*]pyridine, nucleophilic aromatic substitution, Knoevenagel condensation.





Determination of Toxicity in Vehicles Used Lubricant oil by Microwave Digestion and Atomic Absorption Spectroscopy

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A determination of heavy metal in used lubricant oil was performed by microwave digestion of the sample and analysis by Atomic Absorption Spectroscopy (AAS). The detected heavy metals included iron (Fe), copper (Cu), chromium (Cr) and lead (Pb) were those contained in vehicle compartment. The method was started from mixing a lube oil sample with concentrated nitric acid and hydrogen peroxide and put the mixture into a microwave digestion system with a four-stage program. A yellowish-clear liquid sample was obtained. Either of organic solvents, namely ultrapure water, 1,4-dioxane, methanol, acetone and acetonitrile, was added to the liquid extract as an additive. It was found that among 4 selected metal ions, Cu ion gave the highest sensitivity of AAS. However, ultrapure water could be solely used during sample preparation without causing inhomogeneity of the solution and giving similar absorption to those with organic solvent additive. From the method optimization, it was confirmed that the determination of heavy metals in used lubricant oil by the method was feasible, leading to the next stage of method development and resulting quantitative analysis of the heavy metals.

Keywords: Atomic absorption spectroscopy; heavy metal; hydrogen peroxide; method optimization; microwave digestion; nitric acid; used lubricant oil.





Application of porous Metal Organic Framework for preparation anion sensor materials

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Composite materials $ZnQ_2@BIO-MOF1$ has been synthesized for anions sensor. It has been prepared by ion exchange between dimethylamine cation(DMA^+) in porous of BIO-MOF1 with Zn^{2+} provided $Zn^{2+}@BIO-MOF1$. This compound was soak in methanol solution of quinoline to obtain $ZnQ_2@BIO-MOF1$. To confirm the structure of compound, it was characterized by IR, UV-visible, elemental analysis and Powder X-ray diffraction(XRD). This material was applied for anion sensor by using fluorescence property of $Zn(quinoline)_2$ at $\lambda_{em} \sim 530$ nm (When $\lambda_{ex} \sim 420$ nm). The fluorescence intensity of $ZnQ_2@BIO-MOF1$ was gradually increase by anions F^- , Cl^- , Br^- and I^- especially Cl^- when increasing its concentration.

Keywords: metal organic framework, fluorescence properties, ion exchange, anion sensor





Trace Metals Analysis in Herbal Medicine and Cosmetic Samples by AAS

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Recently, the herbal medicines and cosmetics are widely sold and used. The herbals have many properties such as skin whitening, deodorant, inhibit degradation of knee bones and muscle, and treat mouth ulcer. The consumers buy freely. The herbal medicines and cosmetics are cheap and not strictly control. Many people pay attention to use the herbal medicines and cosmetics because they trust that herbals are safe and good for health and skin. Actually, the herbal medicines and cosmetics may be contaminated with heavy metals. If the concentrations of heavy metals were above the suggested safe limit, it cause make human health risks. The objective of this research provided information on the risk associated with human exposure to heavy metals in the herbal products and studied pros and cons of digestions which were digested by hot plate and microwave. This research aimed to quantify 7 trace metals in 6 herbal medicine and cosmetic samples, sold in Thai markets, and analyzed by Atomic Absorption Spectroscopy. The concentration of Iron (Fe), Manganese (Mn), Copper (Cu), and Cadmium (Cd) were digested by hot plate and microwave analyzed using Flame Atomic Absorption Spectroscopy (FAAS). The concentration of Mercury (Hg) and Arsenic (As) were digested by microwave and were analyzed by Hydride Generation with Flow Injection Atomic Absorption Spectroscopy (FIAS). The result study revealed that the amount of concentration of essential metals (Fe, Mn, Cu, and Zn) seem to be high in samples and the concentration of heavy metals (Cd, As, and Hg) were below the suggested safe limit. The advantages of microwave using time, chemicals, and samples saving. In conclusion, the levels of heavy metals in samples were low and within acceptable and safe concentrations that were proposed by the WHO, U.S.FDA, EU, and FDATHAI

Keywords: The herbal medicines and cosmetics, Atomic Absorption Spectroscopy, Heavy metals, Hot plate, Microwave





Application of Nanoporous Silicon as Drug Delivery Systems

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In this research, Porous Silicon (PSi) was applied in drug delivery system. Curcumin, a nutraceutical used for suppressing or treating certain chronic diseases, was selected in this study due to its instability. Monocrystalline silicon wafer is the starting material for PSi fabrication via electrochemical etching technique. A current density of 157 mA/cm^2 was used to produce the PSi layer of 90% porosity. The obtained PSi layer was crushed to fine powder and calcined at $500 \text{ }^\circ\text{C}$ for 1 hour to obtain hydrophilic SiO_x surface. Then, the curcumin was dissolved in ethanol and loaded into PSi powder for 3 hours. The release behavior was studied by stirring loaded powder in 5 different media: ethanol, H_2O and phosphate buffer saline (PBS) of pH 5.8, 7.8 and 8.0 for 24 hours. UV-Visible Spectroscopy was employed to monitor the concentration of curcumin in solutions. The results show that 53.73 % of curcumin can be loaded into the PSi powder, but can be released very few.

Keywords: Curcumin, Electrochemical Etching, Porous Silicon





Curcumin encapsulation by poly(lactic-co-glycolic acid)-cyclodextrin polymer nanoparticles

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Curcumin, a natural yellow compound, showed many excellent pharmacological activities. It has very poor water solubility (11ng/mL) and photostability resulting its pharmacological potentials are limited. In this research, curcumin encapsulation by nanoparticles have been studied. Poly(lactic-co-glycolic acid) (PLGA) is a core and poly-beta-cyclodextrin polymer (pbCD) is a shell have been prepared. The PLGA nanoparticles were prepared by nanoprecipitation method. The pbCD shell has obtained by adsorption from pbCD aqueous solution. The ratio of PLGA-pbCD-curcumin was varied. The particles sizes and dispersion in water were investigated by dynamic light scattering (DLS). The structures were studied by ¹H-NMR, UV-visible spectroscopy, and scanning electron microscope (SEM). The results showed that by using low molecular weight PLGA (50,000-75,000 Da) the sizes of particles increased when the amount of pbCD were increasing. The average sizes were around 224-263 nm. The nanoparticles which contained curcumin, the size of particles were around 356 nm that is a properly size for drug delivery system.

Keywords: curcumin; encapsulation; poly(lactic-co-glycolic acid); poly-beta-cyclodextrin





Preparation of Magnetic photocatalyst BaFe₁₂O₁₉/SiO₂/TiO₂

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In this research, the hybrid materials containing BaFe₁₂O₁₉, SiO₂ and TiO₂ were developed via sol gel technique for using as magnetic photocatalyst. Initially, the magnetic BaFe₁₂O₁₉ was incorporated into polyethyl methacrylate (PMMA) template. Surface modification of BaFe₁₂O₁₉ was carried out by coating with silica or silica-polyacrylic acid (PAA). Then, titanium butoxide was added. To obtain BaFe₁₂O₁₉/SiO₂/TiO₂ and BaFe₁₂O₁₉/SiO₂-PAA/TiO₂ products, the samples were calcined at 500 °C for 2 hours. The results of XRD, FTIR, SEM and VSM characterization of the obtained samples confirmed that there was anatase TiO₂ phase on magnetic BaFe₁₂O₁₉ surface. This indicated that magnetic photocatalyst was successfully prepared. Preliminary studied on degradation of Edical tartazine solution under UV light, the prepared BaFe₁₂O₁₉/SiO₂/TiO₂ and BaFe₁₂O₁₉/SiO₂-PAA/TiO₂ showed photocatalyst activity and could be removed by external magnet.

Keywords: Magnetic, Photocatalysts, Titanium dioxide, Barium ferrite





Synthesis of $Zn_2(\text{fumarate})_2(\text{bpy})$ and its surface modification by hydrophobic coating

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Metal-organic framework (MOF) materials have been widely studied as solid adsorbents due to their highly porous nature. Despite the excellent sorption properties of many MOFs, the utilization of MOFs have been limited in real application because of their moisture sensitivity. In this research, modification of MOF surface by hydrophobic coating was employed as a strategy to tackle this problem. $Zn_2(\text{fumarate})_2(\text{bpy})$ MOF was synthesized. We are study hydrophobic coating on MOF surface. $Zn_2(\text{fumarate})_2(\text{bpy})$ is MOF via liquid-assisted grinding (LAG) in the presence of small amount of dimethylformamide (DMF) and using zinc acetate dihydrate, fumaric acid and 4,4'-dipyridyl as precursors. $Zn_2(\text{fumarate})_2(\text{bpy})$ was characterized by X-ray powder diffraction (XRD), Fourier transform infrared spectroscopy (FTIR), and transmission electron microscopy (TEM). The surface of $Zn_2(\text{fumarate})_2(\text{bpy})$ was modified by polydimethylsiloxane coating using a facile vapor deposition technique (VDT). The presence of polydimethylsiloxane on the surface of the MOF was investigated by FTIR and SEM techniques and the hydrophobicity of the coated MOF was visualized by dropping water onto the coated sample.

Keywords: metal-organic framework, hydrophobic, coating





Preparation of Surface-Modified Zinc Oxide Nanoparticles by Silane Coupling Agent to Enhance Light Stability of Dyes Composition

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In this research, the preparation of modified surface zinc oxide nanoparticles by silane coupling agent in various conditions in order to retard the photodegradation were studied. The properties of coated particles under different pH condition and type of silane coupling agent between 3-aminopropyltriethoxysilane (APTES) and Isobuthyltrimethoxysilane were studied. The modified surface of ZnO were characterized by elemental analysis (EA), X-ray diffraction (XRD), Fourier transform infrared (FT-IR) and Thermogravimetric analyser (TGA). The photodegradation rate of Basic Blue 41 by sunlight was studied and was found that the optimised pH for the modification method of ZnO with APTES is 12. The amine group in APTES structure can reduce free radical such as peroxide from photodegradation reaction of dye. The surface modified photocatalyst by silane coupling agent with electron with drawing group can increase light stability of dyes.

Keywords: ZnO, ZnO dope ATPES, Light stability of dyes





Binding Study of Calix[4]pyrroles to Steroids

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Two calix[4]pyrroles were successfully prepared from Electrophilic Aromatic Substitution reaction between pyrroles and two kinds of ketones including acetone and cyclohexanone. ^1H NMR titration was used to study their binding properties to steroids including cholesterol, β -estradiol, testosterone, lithocholic acid, prednisolone, and cholic acid.

Keywords: Calix[4]pyrroles, Steroids, Binding Study, H^1NMR titration

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Binding Investigation of Glycan to norovirus using Molecular Modeling

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Norovirus (NoV) is the cause of gastroenteritis and highly contagious among human. *The most* common NoV infecting in humans is genogroup II (GII) which is also the major cause of outbreak gastroenteritis. In this work, we study NoV GII.4 strain which *was discovered* in Japan around 2012. Because of the lack of *crystal structure*, *homology modelling approach* is applied to build 3D structure of NoV GII.4 2012 strain based on the template structure (PDB code: 3SEJ) taken from Protein Data Bank. Early studies showed that blood group-related glycans determining ABO and Lewis blood groups are specific to the infection of norovirus. In order to investigate the structural effect of this binding, the aim of this work is to preliminary study the binding between NoV GII.4 2012 strain and Le^b tetrasaccharide using molecular modeling. Molecular dynamics (MD) simulations for 100 ns are performed by using GROMACS program with AMBER99SB forcefield.

From the results, MD simulations reveals the stabilized complex structure after 30 ns. Root-mean-square deviation (RMSD) of NoV GII.4 2012 strain and Le^b tetrasaccharide in MD trajectories are investigated and no significant change in the structure is found after the 30 ns MD trajectories. Five conformational clusters posed the high contribution covers 99.67% of total conformations between 30-100 ns MD trajectories are selected for analyzing the binding interaction in NoV GII.4 2012 strain/Le^b tetrasaccharide. The key amino acids which are GLN182, GLN181, TRP183, ALA36, PHE37, VAL184 are found to form H-bond interaction to Le^b tetrasaccharide. Information from the study can be helpful to explain specific binding in NoV to blood group-related glycan(Le^b tetrasaccharide) and can be used as a guideline for studying the other blood group-related glycans.

Keywords: MD simulations, homology *modelling*, blood group-related glycan





Isolation and structure elucidation of bioactive compounds from the fungus *Menisporopsis theobromae* BCC 4162

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Menisporopsis theobromae BCC4162 is a seed fungus producing two bioactive compounds, menisporopsins A and B. These compounds show antimalarial, antibacterial and cytotoxic activities. As fungus are one of the major natural resources for bioactive compounds, this makes us believe that a large number of bioactive compounds produced by this fungus are still unexplored. In this work, the production medium was changed from Fructose Meat Salt Medium (FMSM) to medium containing D-Mannitol, KNO₃, KH₂PO₄ and trace elements. After the fungus was grown at 28 °C for 14 days. The mycelium and medium were extracted with ethyl acetate and both crude extracts were further purified using sephadex LH-20 column chromatography and silica gel column chromatography. To date, two compounds could be isolated from both mycelium and medium and their structures will be elucidated by NMR spectroscopy and mass spectrometry techniques. Moreover, these compounds will be tested for their biological activities.

Keywords: *Menisporopsis theobromae* BCC4162, bioactive compounds





Preparation of Ce-substituted in anatase for reactive red dye photocatalytic degradation

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Water pollution from the dyeing process in the textile industries consists of surfactants, chelating reagents, pH regulators, residual dyes and etc. The azo dyes are commonly used and biologically non-degradation dyes. The photocatalytic degradation of azo dyes is considered to eliminate the residual dyes in waste water. In this research, the Ce/TiO₂-powder and Ce/TiO₂-3DOM (Three-dimensionally ordered macroporous, 3DOM) were prepared using the sol-gel method and studied the photocatalytic degradation of reactive red dye under UV irradiation. The Poly methyl methacrylate, PMMA, was the template of TiO₂-3DOM formation. XRD and SEM-EDS, techniques were employed to characterize the structure, morphology and the photocatalytic activity was evaluated by the UV-Vis spectroscopy. The XRD patterns of all samples showed only TiO₂ anatase without CeO₂. The morphology of catalysts presented the small particles of TiO₂-powder and the 3DOM structure for the TiO₂-3DOM. The photocatalytic performance of TiO₂-powder showed the highest photocatalytic activity comparing to others due to their morphology and particle size.

Keywords: Ce-substituted, Photocatalytic degradation, TiO₂ anatase, 3DOM





Electrochemical detection of capsaicin by using Sn reduced graphene oxide modified glassy carbon electrode

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Food is one of factors for living. Variety of food flavors depends on raw materials and culture of each country. Chilli is a raw material that has been used for cooking around the world. Capsaicin is an organic substance that causes spiciness in chilli. Determination concentration of capsaicin is useful for food industries in order to show the degree of spiciness. Electrochemical method has been used for determination the concentration of capsaicin. In this work, tin electrodeposited on reduced graphene oxide was employed to modify the surface of glassy carbon working electrode. All potentials in this work respect on Ag/AgCl electrode. The experiment was carried out in sodium acetate buffer with certain pH. Cyclic voltammetry was used to determine concentration series of capsaicin to obtain a standard calibration curve. The effects of potential scan rates, pH of electrolyte and accumulation time on the activity of the electrode were also studied. Scanning electron microscopy (SEM) was employed to study the electrode surface. Energy dispersive x-ray spectroscopy (EDS) was used to confirm the electrode surface composition.

Keywords: Capsaicin, Electrochemical, Cyclic voltammetry, Tin graphene oxide





Removing of Calcium ion in Biodiesel from using of solid catalyst

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The aim of this study is to remove the calcium ion Ca^{2+} contained in biodiesel. The leaching of calcium methoxide ($\text{Ca}(\text{OCH}_3)_2$) catalyst in biodiesel production is the case of Ca^{2+} contaminated in biodiesel. So, our method use sodium carbonate (Na_2CO_3) to precipitate Ca^{2+} into the solid form of calcium carbonate that can be easily remove from biodiesel. In addition, we analyze the Ca^{2+} concentration in biodiesel by complexometric titration with ethylenediaminetetraacetic acid (EDTA)

Keywords: Biodiesel, Heterogeneous catalyst, Calcium soap, Sodium carbonate, Calcium methoxide





Electrochemically detection of capsaicin by surface modified halloysite electrode

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Thai food is worldwide known to be “spicy”. In Chemistry, the term “spicy” is defined by the amount of one chemical found in various type of chilli, called capsaicin. Numerous methods have been presented to quantify the amount of capsaicin in samples. However, they mostly require complicated procedures or expensive instruments. This project aims for developing an electrochemical sensor for capsaicin detection. We incorporated a mesoporous compound of halloysite nanotube into the carbon electrode to increase the internal surface area, hoping to enhance the detection limit. In addition, the surface of halloysite nanotube was modified with 3-aminopropyl triethoxysilane (APTES) to chemically attach amino functional group inside the mesopores. This modification, in turns, significantly increases the sensitivity of capsaicin detection. We also compared the results from electrochemical method with a traditional measurement and found an acceptable agreement. We will later test this modified electrode on the real food samples to quantify the “spiciness” in each food. We believe that this developed technique could be beneficial for the food industry in the foreseeable future.

Keywords: Capsaicin, Electrode, Modified electrode, Electrochemical, Halloysite nanotube





Preparation of Silica-Graphene Oxide Hybrid Particles in Natural Rubber Composites

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Silica-graphene oxide (Si-GO) hybrid particles were prepared by the hydrolysis of tetraethyl orthosilicate (TEOS) in the presence of graphene-oxide (GO) obtained from a modified Hummers method. Scanning electron microscopy (SEM) images provided visible evidence of the silica nanoparticles grafted on the surface of GO, resulting in Si-GO hybrid particles. Energy dispersive X-ray spectroscopy (EDX), Fourier Transform Infrared spectroscopy (FT-IR) and X-ray diffraction (XRD) spectra indicated the coexistence of silica and GO in the particles. The Si-GO hybrid particles showed better thermal stability than that of GO according to thermogravimetric analysis (TGA). The Si-GO hybrid particles were then incorporated in natural rubber by two roll mill. It was found that the Si-GO hybrid exhibited much better dispersion in NR compared with neat SiO₂ and neat GO nanofillers. The Si-GO hybrid exhibited good interaction with NR matrix, which resulted in the improvement in the tensile properties (tensile strength, 300%Modulus and elongation at break). With the addition of only 1 phr of Si-GO hybrid, the NR composite exhibited apparently enhanced tensile properties and hardness compared with the neat one.

Keywords: Graphene oxide, Silica-Graphene oxide hybrid





Effect of Modified Eggshell Content on Mechanical Properties of Natural Rubber Composites

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For industrial egg producers, shells have to be disposed of in landfill because the egg waste attached to them rots quickly, causing a smelly by-product. Eggshell is made of calcium carbonate. The aim of this work was to investigate the potential of using the eggshell waste as a filler in rubber composite instead of commercial calcium carbonate. In order to improve the interaction between and natural rubber, eggshell was modified by coumarone indene resins at 1 %wt. The amount of filler was varied from 0-40 phr. Natural rubber and filler were mixed by using internal mixer and two-roll mill. The cure characteristic was studied by moving die rheometer. The mechanical properties were performed by tensile testing. The results showed that the hydrophobic properties of eggshell were increased by modified with coumarone indene resins. The cure characteristics of natural rubber did not change with adding calcium carbonate or eggshell. Hardness and modulus of vulcanizate were increased with increasing the amount of filler. Tensile strength of vulcanizate was decreased with increasing the amount of filler. However, the filler content did not affect much on the elongation at break and tear strength of vulcanizate. Then, the amount of filler can be added up to 40 phr in rubber composite. Moreover, the mechanical properties of eggshell filled rubber were almost the same as commercial calcium carbonate filled rubber. Therefore, eggshell had the potential to use as a filler in natural rubber instead of calcium carbonate.

Keywords: rubber composite, eggshell, calcium carbonate, coumarone indene resins





Removal of industrial dyes from aqueous solution using geopolymer as a potential adsorbent

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Geopolymer is the new material which can be made from several aluminosilicates material. In this work, geopolymer was applied to remove brilliant green (BG), a cationic dye, from aqueous solution. Optimum parameters for adsorption including dosage and adsorption time were investigated. Adsorption isotherm and thermodynamic properties were studied. The obtained results indicated that at initial dye concentration at 300 mg/L, optimum dosage of adsorbent is 10 g/L and optimum adsorption time is 30 minute with 99 % of dye adsorption. Moreover, to understand thermodynamic properties of the adsorption, free energy of adsorption (ΔG°), enthalpy (ΔH°) and entropy (ΔS°) changes were performed. Based on the obtained results, geopolymer is recommendable to use as highly effective adsorbents in removing industrial dye contaminated in wastewater because of its low cost and environmentally friendly chemical process.

Keywords: geopolymer, brilliant green, adsorption, isotherm, thermodynamic properties





Elucidating the Key Structural Features of 1)-Benzimidazol-1-yl-(3)-2,3-Dihydro-1*H*-inden-5-yloxy (Propan-2-ol of Enoyl-ACP Reductase Inhibitor as a Novel Anti-tuberculosis Agents using Molecular Docking Calculations and Molecular Dynamic Simulations

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1) -benzimidazol-1-yl-(3) -2,3-dihydro-1*H*-inden-5-yloxy (propan-2-ol was discovered as novel anti-tuberculosis agent by structure based virtual screening onto enoyl-ACP reductase)InhA (binding site and mycobacterial whole cell assay .Here, this compound was used as the template for developing of highly potent anti-tuberculosis agents .In this study, bioisosterism was applied to modify the structure of 2,3-dihydro-1*H*-inden-5-yloxy ring and molecular docking calculations was applied as screening tools to obtain new structures of 1) -benzimidazol-1-yl-(3) -2,3-dihydro-1*H*-inden-5-yloxy (propan-2-ol derivatives . The obtained results show that 72 novel compounds were highly predicted binding energy in InhA binding site than template compound . Moreover, molecular dynamic simulations were applied to elucidate the binding mode and binding interactions into the InhA binding site . The crucial interaction of highly active compound formed hydrogen bond interactions between hydroxyl group with hydroxyl group of nicotinamide ribose cofactor and Tyr158 . The binding affinity of highly active compound was improved by hydrophobic interactions of modified substituents onto 2,3-dihydro-1*H*-inden-5-yloxy ring with amino acid surrounding binding site .Accordingly, the obtained results aid to understand the crucial interactions of 1)-benzimidazol-1-yl-(3)-2,3-dihydro-1*H*-inden-5-yloxy (propan-2-ol derivatives of InhA inhibitors and provide information structure concept for designing of novel compounds with better potency against *M. tuberculosis*.

Keywords: anti-tuberculosis agents, enoyl-ACP reductase, molecular docking calculations, molecular dynamic simulations





Preparation of Nanocellulose Particles from Water Hyacinth for use as a Reinforcing Fiber in Natural Rubber

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Water hyacinth is one of the world's worst aquatic weeds. Water hyacinth mats clog waterways. Water hyacinth mats also degrade water quality by reducing oxygen level in water. The objective of this work is to prepare nanocellulose from water hyacinth for use as a reinforcing agent in natural rubber. Fresh water hyacinth was extracted by juice blender. The percent yield of fiber was 6.91. Then, lignin in cellulose fiber was removed by 1M NaOH for 1 hour at 60°C with the percent yield of 71.72. After that, the nanocellulose particle was prepared by acid hydrolysis (hydrochloric acid) and high shear homogenization. The percent yield of nanocellulose was 39.58. We also studied the acid hydrolysis by using sulfuric acid with different concentrations: 10, 20, 30, 37 and 47 %. The results showed that the percent yield of nanocellulose was 13.08, 14.56, 20.37, 62.86, and 70.64, respectively. The color of nanocellulose particle by using sulfuric acid 47% was black while the other conditions were brown. In the final step, the film from rubber latex and nanocellulose particle was prepared and mechanical properties will be investigated.

Keywords: nanocellulose, natural rubber, water hyacinth, acid hydrolysis





Modification of Eggshell by Coumarone indene Resin for use as Filler

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Eggshell is one of the solid wastes. The disposal of this waste can lead to a very important problem such as public health, water source contamination and polluting the environment. Eggshell contains mainly calcium carbonate. This research aimed to study the effect of modification of the calcium carbonate from eggshell on the mechanical properties of natural rubber composite. The eggshell was modified by coumarone indene resin with 0, 0.5, and 1.0% by weight. The mechanical and physical properties of natural rubber with eggshell compared to calcium carbonate were investigated. The results showed that the modification of eggshell with coumarone indene resin increased the hydrophobic properties of eggshell. Modification of eggshell did not affect the scorch time and cure time of rubber compound. Hardness of eggshell with and without modification filled natural rubber was increased when compared gum (unfilled natural rubber). The mechanical properties of filled natural rubber were slightly improved by modification in terms of modulus and tensile strength. The suitable amount of coumarone indene resin was 0.5% by weight. Moreover, the rubber composite with eggshell and commercial calcium carbonate have similar mechanical properties. Hence, eggshell has the potential to use as filler in natural rubber.

Keywords: eggshell, natural rubber, calcium carbonate





Study of Antioxidant Properties of Thai Herbs in North Eastern of Thailand

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The antioxidant activities of fourteen Thai herbs in North Eastern of Thailand were investigated. There were extracted using hexane, ethyl acetate and methanol as solvents. The antioxidant activity evaluated by DPPH and ABTS assay show that methanol crude extracts showed the highest antioxidant activity. Thai herbs namely Phaya Mueling, Bai Yanang Daeng, Kua Roi Ru, Fang Daeng, Kampaeng 7 Chan, Thaowan En-on, Ta Kwang, Pra Dong Dam, Toom Ka Daeng and Kamlung Wua Thaleng shows the more than 90 % antioxidant activity for DPPH and ABTS assay. The antioxidant activities for Kampaeng 7 Chan from methanol crude extracts showed the highest antioxidant activity is 95.20 % inhibition for DPPH assay and Fang Daeng from methanol crude extracts showed the highest antioxidant activity is 99.92 % inhibition for ABTS assay. For the reducing antioxidant powder (FRAP), the crude extracts in methanol of Phaya Mueling, Bai Yanang Daeng, Fang Daeng, Thaowan En-on, Ta Kwang, Pra Dong Dam and Kamlung Wua Thaleng shows the more than 1800 mg of VCE/g. The methanol crude extract of Pra Dong Dam showed the highest antioxidant activity is 2118.84 mg of VCE/g extract.

Keywords: Thai herb, antioxidant activity, DPPH assay, ABTS assay, FRAP assay





The Equilibrium Study of Adsorption Brilliant Green Dye Solution using Water Sludge and Synthetic Zeolite as Highly Potential Adsorbent

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This research, water sludge was used as raw material to synthesize zeolite using alkali fusion method. These synthetic zeolites were applied to remove brilliant green dye of aqueous solution. The obtained results implied that synthetic zeolites were higher adsorption efficiency (> 90 % adsorption efficiency) than water sludge. The optimal parameters of synthetic zeolite No. 1, the most suitable adsorbent on dye adsorption process were further investigated. The obtained results showed that the optimal parameters of adsorption were 4 g/L of adsorbent dosage with 60 minutes of adsorption times. The adsorption isotherm was corresponded well to Langmuir adsorption isotherm with r^2 of 0.9786. The pseudo second order kinetic models were well fitted with experimental data with r^2 of 0.9994, exhibiting a maximum adsorption capacity of 79.40 mg/g. Therefore, we presented the potential adsorbent using synthetic zeolite as a suitable adsorbent for the removal of brilliant green dye from aqueous solution.

Keywords: zeolite, brilliant green dye, water sludge





A novel natural clay-polymer composite for Application of Slow-releasing Plant Hormone

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In this study, a novel clay-polymer composite was prepared via polymerization of poly (vinyl alcohol) (PVA), glutaraldehyde (GA) and zeolite in the presence of zinc nitrate for slow-released plant hormone application. Evidence of clay intercalation and component interactions, structure, and morphology was characterized by FTIR, XRD and SEM techniques. Moreover, the maximum absorption of each reactant was investigated by UV-Visible spectroscopy. The water absorption and retention behaviors were investigated. After the characterization, the potential applications have been verified through the ability of the composited film on slow release naphthalene acetic acid (NAA), which affects adventitious root development of moong bean (*Vigna radiata*). The obtained results could be beneficially for agricultural application because of low cost, good water and chemical absorbency properties and show environmentally friendly.

Keywords: poly vinyl alcohol (PVA), glutaraldehyde (GA), naphthalene acetic acid (NAA), clay-polymer composite





**Binding Mode of Bioactive Compounds of Marine-Derived Fungus
Aspergillus unguis in *M. tuberculosis* PknG Binding Site :
Structure Based Drug Design**

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Mycobacterium tuberculosis Serine/Threonine Protein Kinase G, PknG has been focused as attractive target for anti-tuberculosis drug development. This enzyme involves in signal transduction partway that is essential to promote mycobacterial survival within macrophages. Herein, we aim to develop new anti-tuberculosis agents based on PknG inhibitors with short treatment durations from Thai natural products. Bioactive compounds which isolated from marine-derived fungus *Aspergillus unguis* were docked into PknG binding pocket to elucidate the binding mode, binding interactions, and binding affinity. The obtained result based on docking score suggested that aspergillusidone A was strongest bound to PknG active site with docking score of -9.08 kcal/mol. Based on MD simulations, hydrogen bond interactions of hydroxyl group with carbonyl backbone of Glu233 and Val235 were found as the crucial rule for binding in PknG binding site. In addition, molecular behavior aspergillusidone A/PknG complex was provided the key information to develop highly specific PknG inhibitors from bioactive compounds. Therefore, the obtained results from this study guide to design the new potential bioactive compounds and biological assay evaluation against PknG.

Keywords: PknG inhibitors, natural products, molecular docking





Removal of industrial dyes from aqueous solution using melanin nanoparticles as a potential adsorbent

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In this study, the adsorption efficiency of brilliant green (BG), congo red (CR) and methyl orange (MO) dyes onto bentonite and modified bentonite as melanin nanoparticles from aqueous solution were investigated by batch adsorption technique. The obtained results show that the adsorption efficiency of modified bentonite adsorbent for adsorb BG dry show higher 90 % adsorption efficiency than CR and MO at 500 ppm of initial dye concentration 30 min. of adsorption time and 20 g/L of adsorbent dosage. Moreover, the optimal adsorption condition based on the optimal adsorbent dosage was investigated. The optimal dosage of modified bentonite adsorbent to remove CR dye at 500 mg/L of initial dye concentration is 20 g/L. Bentonite and modified bentonite as melanin nanoparticles were characterized by X-ray diffraction method. The results indicate that the modified bentonite as nanomelanin adsorbent is a potential low-cost effective adsorbent for dyes removal from contaminated wastewater.

Keywords: bentonite, melanin nanoparticles, adsorption, brilliant green (BG), congo red (CR), methyl orange (MO)





Determination of Total Content of Phenolic Compounds and its Antioxidant Properties in Star Grass

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In this study, determination of antioxidant compounds from macerated star grass powder with various solvents; acetate, ethanol, acetone, methanol and distilled water was investigated by using DPPH Radical Scavenging assay. Moreover, determination of content of total phenolic compounds was done by using Folin-Ciocalteu colorimetric method. The results indicated that the ethanol crude extract obtained from DPPH assay showed highest antioxidant activity with 44.47%. In addition, the highest content of total phenolic compound is 95.74 mgTAE/g. Therefore, these results could be beneficially used in cosmeceutical, pharmaceutical and nutritive information for consumers.

Keywords: star grass, DPPH assay, Folin-Ciocalteu colorimetric method, total phenolic compound, antioxidant activity





Determination of antioxidant properties and total phenolic compound of various pigmented Thai rice varieties grown in Northeastern region of Thailand

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In this study, the antioxidant activity of eight glutinous Thai rice sample including three glutinous rice; Dang, Dam and Ko-Kho 6. Moreover, five normal rice; Dang, Lueng, Homnin, Insri and Mali 105 were investigated. The uncooked seed of rice samples were grinded to powder and extracted by hexane, ethyl acetate and methanol. Antioxidant activity using DPPH Radical Scavenging Activity, ABTS Cation Radical Scavenging Activity and Ferric Reducing Antioxidant Power (FRAP) were investigated. Moreover, total phenolic compounds were studied. The results indicated that the antioxidant activity obtained from DPPH and ABTS assays show methanol crude extracts of Dang glutinous rice with highest antioxidant. The activity antioxidant activity of Dang glutinous rice with 68.61% and 99.52% for DPPH and ABTS assays when compare with Trolox[®] reagent, respectively. Moreover, the total phenolic compound of Dang glutinous rice of methanol crude extract showed highest total phenolic compound is 63.07 mgTAE/g when compare with Tannic acid. Based on the FRAP assay results, the methanol crude extract of Dang glutinous rice is highest value with 1419.69 mg. Therefore, these results could be beneficially used as nutrition information for consumers.

Keywords: glutinous Thai rice, DPPH Radical Scavenging Activity, ABTS Cation Radical Scavenging Activity, Ferric Reducing Antioxidant Power (FRAP), Total phenolic compound and Antioxidant activity





Synthesis of co-polymeric thermoresponsive chitosan-graf- poly(N-isopropylacrylamide) encapsulated magnetic-silica as a nanocarrier for drug delivery with improve therapeutic efficiency

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The exploration of multifunctional particles based theranostics provides powerful tools for efficiency of cancer therapies. Here, we report a magnetic-silica core nanoparticle modified with thermo and pH sensitive polymers shell comprising of temperature sensitive polymer poly(N-isopropylacrylamide) (PNIPAAm) with chitosan (Mag-MSN@Chi-g-PNIPAAm). The Mag-MSN@Chi-g-PNIPAAm nanocomposites exhibited structured spherical particles with diameter about 20 nm as seen by TEM. To future confirm about the magnetic properties, the synthesise particles showed superparamagnetic behavior at room temperature with saturation magnetization of 20.20 emu/g. The DOX was released from Mag-MSN@Chi-g-PNIPAAm highly sustainably (over 25h) in the physiological environment and room temperature (pH 7.4 and 37°C). While the synthesise particles highly sensitive to reduced pH (4.0) and at hyperthermia (T=45°C) environment effectively releasing DOX drug (>90%) was obtained after 5h. The in vitro cell culture experiments shoed DOX-loading Mag-MSN@Chi-g-PNIPAAm particles more biocompatible and less cytotoxic than DOX only, but had a higher concentration of DOX-Mag-MSN@Chi-g-PNIPAAm particles of 250µg/ml inhibitory effect on HeLa cell growth was obtained. All these features indicated the Mag-MSN@Chi-g-PNIPAAm particle has potential as therapeutic agents for future in vivo drug delivery systems.

Keywords: magnetic drug delivery, chitosan, poly(N-isopropylacrylamide), dual pH and thermo responsive.

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Fabrication of salinity monitoring sensor

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Most Thai people love to eat meals with strong flavors. Based on Department of Health under the Ministry of Public Health report, Thai people consume sodium/salt more than twice as much as they need. In general, a healthy adult shouldn't exceed 2,300 mg which translates to about 5.5 grams of salt daily. Too high salt consumption causes high blood pressure that is a risk factor for problems with the kidneys, the heart and the brain. In this project, we present the low-cost fabrication of salinity monitoring sensor. Effects from pH and temperature have been investigated for correction of salinity monitoring. Based on the results, the salinity in liquid samples can be estimated via detection of electrical conductivity in liquid samples. The electrical conductivity exponentially increases with increasing amount of salts. Temperature and pH strongly affects on the electrical conductivity in liquid samples. The electrical conductivity increases when temperature increases and pH is in acids. A mathematical model will be proposed for correction of electrical conductivity that leads to estimation of salinity with high accuracy in liquid samples.

Keywords: NaCl, Conductivity of NaCl, Salinity sensor





Surface Angle Measurement using Adapted Autocollimator Combined with Fizeau Interferometer

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We have designed an adapted autocollimator which can be combined with Fizeau interferometer. Our objective is to take advantage of both devices to measure small tilt angle of a surface without touching a workpiece. Autocollimator is a standard device used to measure small tilt angle of a flat surface. However, when the surface is rough, image from the device can be distorted. The result is that we cannot measure the midpoint of the image. To overcome this problem, we found that the configuration of autocollimator is very similar to Fizeau interferometer. The main difference is that we have to insert a reference flat plate to get an interference pattern. So we have designed the device that can work in both modes. For a flat surface we will use autocollimator mode since it gives higher precision angle measurement. When the surface is rough or curved we can use the interference pattern to resolve the distortion of autocollimator image to get, with less precision, tilt angle of the surface.

Keywords: Surface Angle Measurement, Autocollimator, Fizeau Interferometer





A Small-Scale Homemade Vertical Axis Wind turbine for Generating Electricity

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This research studies on the aerodynamics of the vertical axis wind turbine's blades and the movement of air flow attacking the blades. The point is to design the blades for performing at low wind speed, and to find the power efficiency of the wind turbine at the varying wind speeds. In the experiment, the electricity power is calculated, and the turning speed of the wind turbine is measured, and forces attacking the blades are found out. The result shows that the designed blade has a turning curve for the air flow to attack. The air will turn inside and turn out of the blades. This gives a drag force to spin the turbine at lower wind speed if it is compared to other types of the blade design. Moreover, a lift force is perpendicular with a drag force. The turbine is measured at a constant wind flow at the average speed of 3.5 meters per seconds that is called gentle breeze in the wind rate. It gives an average electrical power of 23 mill watts per hours or 566 watts per day and the average turning speed of 44 rounds per minute.

Keywords: Aerodynamics, Vertical Axis Wind Turbine, Power Efficiency, Lift Force, Drag Force





Measurement of Radioactive Iodine-131 Airborne Dispersion at Radioactive Center of Thailand Institute of Nuclear Technology

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Radioiodine-131 is low stability radioactivity that can be sublimate or evaporate for all states. The physical half life of Iodine-131 is 8.04 days, after which it disintegrates into Xenon-131 through beta(maximum beta energy: 606 keV (90%),334 keV (7%),248 keV (2%)) and gamma(gamma energy: 723 keV (2%),637 keV (7%),365 keV (82%),284 keV (6%),80 keV (3%))emissions. That beta ray will destroy the tissue when the radionuclide is adsorbed into human organ. Radiological specialist must be mind themselves to adsorbed radionuclide. The research main objective is to measure the radioiodine-131 airborne dispersion by 13 samples collected at Radioactive Center.They were measured and evaluated by using a high-purity Germanium (HPGe) detector and gamma spectrometry analysis system at the Thailand Institute of Nuclear Technology. The volume Iodine –131 standard sources were used to compare the efficiency for calculating specific activities. The measurement time of each sample is 10,000 seconds. Radiological specialist intake of radioiodine should be less than 20 m Sv/yr.

Keywords: radioactivity, Iodine-131, gamma ray, beta ray, specific activity





Synthesis of graphene by thermal chemical vapor deposition method

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Graphene is an allotrope of carbon in the form of a two-dimensional, atomic-scale, and hexagonal lattice. Graphene owns many unique properties such as 200 times stronger than the strongest steel, good transparent, high heat conductivity, and great electrical conductivity. Graphene can be applied to several applications including solar cells, light-emitting diodes (LED), touch panels, sensors and smart windows or phones. In this project, we report the synthesis of graphene by using a thermal chemical vapor deposition method. Growth parameters such as temperature and flow rates of gases have been investigated in order to find an optimum parameter for graphene growth. C_2H_2 and H_2 were used to synthesize the graphene. H_2 was used for removing oxides while C_2H_2 was used for carbon gas source. From the results, the best temperature for graphene growth is $\sim 700^\circ C$. At too low and high temperature, it leads to form a carbon clusters. Mixed air flow of 1,400 sccm H_2 and 20 sccm C_2H_2 can be resulted in formation of graphene sheets. The quality of graphene has been confirmed by Raman spectroscopy.

Keywords: Graphene, Chemical vapor deposition method, Raman spectroscopy





Fitting Rotation curve of galaxies by pressure from dark matter annihilation

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There are many evidences for the existence of dark matter. We found that a quarter of the Universe is composed of dark matter. The properties of dark matter are still being huge problems of cosmology and particle physics. Therefore, physicists try to identify the characteristics of dark matter. Dark matter can be interacted by gravitational force and can be weakly interacted by weak force without any electromagnetic force or strong force. Although, we can precisely measure the amount of dark matter by gravitational force, we still cannot identify type of dark matter.

However, we can measure the properties of dark matter by indirect detection which based on the assumption that dark matter and anti-dark matter can annihilate into standard model particles which can be detected such as electrons, positrons, protons, anti-proton, neutrinos and photons. The signature of dark matter particles can be investigated from the productions of dark matter annihilation.

In this research, we have studied the effect of dark matter pressure to the rotation curves of low surface brightness galaxies. We assume that dark matter particles annihilate into electrons and positrons. After the annihilation, electrons and positrons can lose their energy by inverse Compton scattering, Coulomb collision, Bremsstrahlung, Ionization and Synchrotron radiation. However the electrons and positrons can create a gas pressure, called dark matter pressure, against the gravitational contraction which effect to the rotation curves of galaxies. We have studied the effect of dark matter pressure by using the Navarro Frenk and White (NFW) profile and varying the ionization fraction of the gas in the galaxies.

Keywords: Astrophysical signatures of dark matter, Dark matter, Dark matter indirect detection





First-principles study of the optical property of Ti-doped α -Al₂O₃

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Corundum (α -Al₂O₃) is well known as the gemstone which is attractive and valueable. It is known that colors in the gemstone are mostly related to the impurities existing in the crystal. There are several attempts to improve the color quality of the gemstone without adding other elements to increase its value. However, the understanding of the cause of color in the gemstone is still limited. In this research, we investigated the effect of Ti doped in corundum on the optical property of the crystal. By calculating the defect formation energy under two extreme growth conditions, i.e., Al-rich and O-rich growth conditions, to study the possibility of Ti defect formation, we found that under Al-rich growth conditions, Ti could substitute for Al (Ti_{Al}) and O (Ti_O) sites or insert into the crystal at interstitial site (Ti_i) depending on the Fermi-energy position. Regarding O-rich growth condition, we found that Ti is likely to substitute for only Al site. Based on the optical absorption and emission calculations, we found that Ti_{Al} defect can absorb the energy at the wavelength of \sim 688 nm, which is in the visible light region.

Keywords: first-principles calculations, corundum, DFT, defect





First-principles study of the optical property of Fe-doped in α -Al₂O₃

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In this research, we used first-principles calculations based on density functional theory to investigate the effect of Fe doped in α -Al₂O₃ or corundum on the physical properties, especially optical property. It is well known that colors in corundum are directly related to the defects created in the crystal. Consequently, the study of Fe doped in α -Al₂O₃ could help us to gain more understanding about the optical property of the crystal doped. Here, we investigated the likelihood of Fe defect formation by considering the defect formation energy under two extreme growth conditions, i.e., Al-rich and O-rich growth conditions. Our calculation results revealed that under Al-rich growth condition Fe could exist in many forms, such as substitution for Al site (Fe_{Al}), substitution for O site (Fe_O), and Fe interstitial (Fe_i) depending on the Fermi-level position. However, under O-rich growth condition, we found that Fe prefers to substitute for only Al site. Based on the optical absorption and emission calculations associated with Fe defects in several configurations, we found that Fe_{Al} defect can absorb the energy at the wavelength of \sim 597 nm, which is in the visible light region.

Keywords: first-principles calculations, corundum, DFT, defect





Engineering scaffold of bioactive glass incorporated biphasic calcium phosphate /PCL/Chitosan-collagen nanocomposite: A platform for bone tissue engineering

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In the present study, scaffolds for bone tissue engineering applications were made by immersing the inorganic phases of biphasic calcium phosphate (BCP) mixing bioactive glass (15Ca:80Si:5P) (BG) with different ratios of polycaprolactone (PCL) in an organic phase of chitosan/collagen (ChiCol) matrix. Porous scaffolds were obtained by freeze-drying the combinations. The mechanical properties and *in vitro* growth of rat osteoblast-like UMR-106 cells were investigated. The investigation indicated that the compressive strength was controlled by the ratios of organic phases. The highest compressive modulus of the composites was 644.92 MPa (20.31 MPa for compressive strength) which is for the BCPBG@PCLChiCol10 composite. Due to its better stability and mechanical strength BCPBG@PCLChiCol11 composite scaffold was chosen for bioactivity tests. *In vitro* cell availability and proliferation tests confirmed the osteoblast attachment and growth on the BCPBG@PCLChiCol11 surface and non-significant differences when compared to that on the glass substrate after 7 days of culturing. To test its bioactivity, BCPBG@PCLChiCol11 was chosen for MTT and ALP assays on UMR-106 cells. The results indicated that the UMR-106 cells were viable and had higher ALP activity as the culturing times were increased. Therefore, BCPBG@PCLChiCol scaffold shows promise for future in bone regeneration.

Keywords: calcium phosphate, bioactive glass, biopolymers, bone tissue engineering





Uncertainty Analysis in Basic Physics Laboratory 1

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We investigate sources of error and source of uncertainty in basic physics laboratory. So far scoring a laboratory report is quite subjective. This makes grading system unreliable. To get an quantitative grading system we can apply the metrology principle to analyze the procedures in each experiments. Source of errors are first investigated. These informations should be informed to the students to avoid them. We can also use these informations to develop the laboratories procedures and equipments. Source of uncertainties are then analyzed. Propagation of uncertainty is used to analyze the theoretical uncertainties of the experiments. Scoring criteria can be defined base on statistic. The information obtained can be used as a quantitative and reliable grading system.

Keywords: Basic Physic Laboratory, Uncertainty, Metrology





Surface Angle Measurement from Fringe Analysis using Combined Fizeau Interferometer and Autocollimator

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We have designed a surface analysis instrument based on the principle of Fizeau interferometer to measure small tilt angle of a surface without touching a workpiece. The tilt angle can be obtained using fringe analysis technique. We use an optical flat plate with 0.2 μm accuracy as a reference for Fizeau interferometer. A beam splitter is used as a tested surface. The interference fringes are produced using a calibrated 633 nm laser light source. The fringe images are captured by digital camera. They are then analyzed via a python program with OpenCV routines. The images of red fringes are transformed to grey scale. The separation of the fringes together with the wavelength of the light source can be used to find surface tilt angle. Our design is meant to combine with an autocollimator, developed by Metrology and Physics of Instrument Research Unit, to overcome the disadvantage of autocollimator in measuring tile angle of a convex or concave small surface. We expect that the combined results of these two devices can be used in the small angle analysis in high precision industrial and research measurements.

Keywords: Fizeau interferometer, fringe analysis, surface tilt angle





Schwinger pair creation in graphene wormhole

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We investigate the behavior of Dirac Particle (electron and hole) confined on the surface of graphene wormhole, specifically, curvature induced by the geometry of graphene wormhole. For calculation, we use numerical shooting method of finding eigenvalue of the Dirac equation in a curved space of graphene wormhole. Our numerical results demonstrate that energy of band structures of the particle depend on the geometrical parameters of graphene wormhole and appear to the periodic spin-orbit potential, corresponding to Zeeman effect. The geometrical parameters and quantization of momentum in the circumference direction specify a quantum state of particle as a bound state particle or a unbound state particle. Finally effects of the geometrical parameter of graphene wormhole on the Schwinger pair creation of particle are presented.

Keywords: Condensed Matter, Graphene, Wormhole, Curved space





Synthesis and Properties of Ni doped ZnO nanostructures

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This work was studied the possibility of growing Ni doped ZnO nanostructure by electrochemical method. Ni doped zinc oxide ($Zn_{1-x}Ni_xO$, $x=0, 0.02, 0.04, 0.06, 0.08, 0.10$) nanostructures have been synthesized by hydrothermal method. Upon successful synthesizing the samples, they were characterized using various techniques. The structure of Ni doped into ZnO nanostructure was studied by X-ray diffraction (XRD). Scanning electron microscopy (SEM) and dispersive spectrum (EDS) were used to investigate the morphology and composition of Ni doped into ZnO nanostructures. The optical property of those samples was analyzed by UV-visible spectroscopy.

The XRD result showed that undoped zinc oxide has wurtzite structure and the Ni substituted ZnO nanostructure maintained the hexagonal wurtzite structure for all levels of Ni substitution. Doping of ZnO with Ni^{2+} was intended to enhance the surface defects of ZnO. The incorporation of Ni^{2+} in the place of Zn^{2+} provoked an increase in the size of nanocrystals as compared to undoped ZnO. The morphology of Ni doped ZnO nanostructure was investigated by SEM. The result showed that pure ZnO has short rods with small particle. When Ni 1% and 2% doped into ZnO; the structure was hexagonal with a small rod and the nanorods were agglomerate together. For Ni 4% and 5%, the results revealed that the nanorods were still short but they are bigger that the other concentrations. Moreover, the surface of nanorods was covered by thin film. EDS measurements of the Ni doped ZnO nanostructure indicated that the Ni substitution created additional Zn vacancies in the wurtzite structure which is reflected in the enhanced photoluminescence emission in the visible light spectra between 450 to 550 nm. Furthermore, the UV-visible spectra of the Ni doped ZnO nanostructure was also investigated. The band gap of the Ni doped ZnO nanostructures can be tuned in the range of 3.227–3.1468 eV by the use of the dopants. The observed red shift in the band gap from UV-visible analysis and near band edge UV emission with Ni doping may be considered to be related to the incorporation of Ni ions into the Zn site of the ZnO lattice.

Keywords: ZnO, Ni doped ZnO, hydrothermal





Epidemic modeling of Dengue fever using one-dimensional lattice model

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Dengue fever is one of the main tropical diseases that threatens world population in more than 110 countries. Currently there is still no effective way to help prevent and control the disease from spreading. The spreading of Dengue fever can be considered as a non-equilibrium phase transition process that depends on the interactions between the population of two species, i.e. humans and mosquitoes, which contract the disease from one another. The disease can exist in one of two phases, i.e. active phase (spreading phase) and absorbing phase (not spreading phase), that depends on many parameters, such as the number of people contracting the disease, the diffusion rate of mosquitoes and the number of mosquitoes.

In this study, we used one-dimensional lattice model to investigate the spreading patterns and the phase transition of Dengue fever in two following models, i.e. 1) a static human model with each human fixed on each lattice site, and 2) a dynamic human model with humans being able to move to other lattice sites and being initially distributed in specific patterns. For each time step, there are five events happening, i.e. human infection and recovery, and mosquito infection, diffusion, and replacement. By starting our model with a single infected site, we can observe the time evolution of the epidemic and study the phase transition by specifically varying the number of mosquitoes in the system. In our static model with 5,000 humans (lattice sites), we found that the phase transition occurred when the number of mosquitoes reached 10,600. Moreover, we also found that the Dengue fever spreaded like a wave with its wave-front moving at a constant speed depending on the diffusion rate of mosquitoes. With the understanding of the principal features of Dengue fever epidemic based on our simple lattice model, this may allow us to be help prevent the disease from spreading in the near future.

Keywords: Epidemic, spreading, Dengue fever, lattice model





Apply IoT for protecting to forget to turn off the car lights

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This research applies the Internet of Things (IoT) technology for car security. In this technical paper, we are focus in how to protect car from unexpected activity of car owner, is forget to turn off headlights, interior light and or tail light that will cause lost power of battery. These unintentional events are call daily bad habit car's owner. To help car owner to stop these habits, we will applies Internet of Things (IoTs) detect that habit then with mobile application, we can monitor and alarm the owner before they lost that cars.

Internet of Things (IoTs) is technology which it's combine 3 technology systems together include Embedded Technology (ET), Network Technology (NT) and Information Technology (IT). Embedded Technology is many devices or embedded system that communication in machine to machine (M2M). We use various devices to solve the problems, the forget to close headlight, interior light or tail light therefore use the light sensor install under headlights and tail light and near interior light for we know status turn on/off of various light. The problem will happen depend on engine working then use the noise sensor to help to check from sound of the working engine. All devices in this major communicate through mainboard, the Arduino Mega board. Network Technology is technology can access internet or cloud server which is 3G-Shield. It role is backbone of the access to cloud server when each devices need send data into cloud. The cloud to use is MQTT Protocol, which stores information divided into 2 threads are "topic" and "message". The topic is domain name of the device and the message is status of the device. The 3G-Shield send information to the cloud every 1 second by PUBLISH command, but send information to the mobile application when the mobile application choose topic interested to the cloud by SUBSCRIBE command. The last one is Information Technology is technology receive various data which depending on each devices. It shows data on mobile application.

We create simulator car for test function of devices that describe detail are as follow: 1.) the light sensor detects light that value is analog and scale 0 – 1,023. While headlights turn on; the values are during 900 – 1,000, interior light turn on; values are during 800 – 900 and tail light turn on; the values are during 800 – 900. 2.) the noise sensor detects sound of engine and can change sound sensitive on device. When sound engine loud, values are "1" or it quite, values are "0". The mobile application will notify to him when the engine is off, but the car lights is turn on.

Keywords: Car Security; IoT; Mobile Application; Smart Car





Smart Dog Collar (Tag) Tracking

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In recent year, the most of people raise the dog popularly but the losing rate of their dog increase also. Because of this problem, So we research and develop Smart Dog Collar(Tag) Tracking. Collar(Tag) we design by assembling Internet of Things(IoT) device 3 devices: Microcontroller board(Arduino), 3G module, GPS module. So we develop the application for preventing losing rate. The collar would receive location data from GPS module consist of latitude and longitude. And the 3G module would send these data to the database server. Finally, the application gets the data from the server and show on Google map API. Our application has 3 functions. The first function is tracking pet. The second function is inspecting that pet is in safe zone. The last is showing the route of pet.

In the experimentation, we test and collect history data and then plot the route of pet in different duration time such as 1 minutes, 5 minutes and 10 minutes. It is found that the error of the collected data is less error. Deviation of data is not significant. Working of GPS Module within the open areas is considered precise. We can receive data from livestock continuously through using 3G. From the experiment, we have tested preciseness of the Smart Tag and found the following weakness of its working:

1. GPS Module has high accuracy in the open areas but quality of detection and accuracy will decrease immediately when it is in the dense closed areas, such as, in the dense forest of in the building.
2. Sending data depends on the mobile phone network providers. If it is the no-signal area, it will cause a problem to sending the data. In this case, it also depends on SIM Card service provider.
3. Our Alpha test Smart Tag is still too big to be used for working really. We have to improve the Smart Tag and design a new model of the Smart Tag.

Conclusion, Our Smart Collar(Tag) can track a position and plot set of position really, but it still several limitations, such as quality of GPS module which there are still better ones than. Those better ones can solve a signal-receiving problem in the dense closed areas. For designing Tag, making a suitable specific form of Tag will have to spend many expenses. We cannot make it and in the aspect of application, now, there is still only Android Application. In the future, we will study, research, fix the problems and develop new functions for the best of Smart Tag. This research can study with other research, such as, Studying weight gain of pet or Studying behavior of pet.

Keywords: Smart Tag, Dog tracking, GPS Smart Tag, Arduino, Mobile Application





Parking Slot Searching System using Internet of Things

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At present, in Bangkok Metropolitan area number of cars has increased especially for the past two years due to the past government's promotion policy. Hence, parking spaces become an issue all over especially in the city. At any organization; for example, Kasetsart University, Bangkaen campus, parking slots are scarce resource. There are not enough parking places for teachers, staff and students and it is difficult to manage to find a parking slot at rush hours. It is time-consuming and a waste of energy in making frequent trips through the parking buildings and parking lots.

Parking slot search application is an application that allows drivers to find an available parking slot within an organization. This application will allow drivers to navigate to the selected parking lot or building. It also displays information on available and taken slots on parking lot map. The driver does not have to drive around to find the parking slot himself. What makes our application different from other applications is that a driver can check for an available parking slot using mobile application on his/her smart phone and then reserves one without having to drive to that particular location. The driver must park his/her car within 10 minutes after reserving otherwise the reserved slot will be released and available for others again. The driver can only reserve one slot at a time. This makes the commuting routine more comfortable and efficient and lowers power consumption. The drivers can estimate commuting time without wasting time in parking his/her car. The application includes an ability to remember the parking location in the building where the user may forget and takes time to look for his/her car. This application is a good choice for any organizations with a few parking lots and buildings. The development of the parking slot searching system is done using the Android Studio program. Android Studio is the latest IDE from Google for developing Android applications specifically. The system uses Arduino IDE to control the sensors, such as a barrier detector or an ultrasonic sensor in order to determine if a car is parked at that particular time. It uses Message Queuing Telemetry Transport (MQTT) Protocol, designed for machine-to-machine (M2M) connections for devices supporting IoTs (Internet of Things) technology. The Arduino Uno Board, an open-source AVR micro-controller board is used together with Arduino Wifi Shield. It allows our Arduino Uno Board to connect to the Internet via wireless network 802.11 (WiFi). Measure the distance at the parking slot is done using the Ultrasonic Sensor module which will start working by first signaling to activate the sound source. Then it sends out the sound waves and waits for an echo in order to send out the sound waves.

The Parking slot searching system application and hardwares will help car drivers reserve parking spaces and navigate them to the reserved parking spaces in at various places conveniently. It can also show the availability of parking spaces at that request time.

Keywords: Parking, Internet of Things, Car, Application, Reserve, Search





Creating and Tracking Memorandum of Scholarship Website

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Nowadays, technology is very necessary in many ways. In addition to the technology makes the work is convenient, it also makes fast and accurate working. Especially the government in Thailand have to use the documents as evidences. Our faculty is the same, there are many students, professors, and staffs who have problems with the document processing. In this project, we call document that memorandum. For example, the memorandum format is not correct, the memorandum is not complete, the memorandum is delayed because it does not track. From the problems mentioned above, we bring knowledge and technology to solve the problem to benefit the Faculty of Science.

Generally, the process of approving activities of the Faculty of Science will use the form provided by the Faculty of Science. The student or contact person must prepare the documents with a memorandum to give details about the activity and will send the memorandum to the Project Advisor, Head of Department, Dean of the Faculty of Science and Director of International Affairs Division of Kasetsart University. The considerations will be considered in that order.

To send a memorandum, the faculty do not store and save data to the database, including no formal memorandum form. So the contacts have to edit the information in their document several times, which effect to data collection. On our website, users can create a memorandum and fill information in the correct form, and the site has an example of filling the correct information. And users can track the status of their memorandums. Moreover, the Faculty of Science can also provide information about the scholarship, view the statistics of sending memorandum in each semester or year, and update the status of the memorandum. All of us hope that the site will solve the problems that occur. It is helpful and a starting point to develop processes in the faculty and university.

The purposes of this project were to provide convenience to staffs and students in the faculty in the part of documentation and document tracking, and to create the correct memorandum for easy archiving into the database. Therefore, we will use the knowledge from the Department of Computer Science to help complete the project and develop it in the future. Other than, our project will benefit to the faculty and also help us to solve problems outside the classroom, which is the inspiration for further study and the starting point to improve ourselves.

Keywords: Memorandum, Document Tracking, Documentation





KU Alert (Android Mobile Application)

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Because of the problem. When problems arise, students or staff at the university. I do not know who to report it. Or with any agency and it is not known whether the problems they have reported have been resolved yet. So I have developed this application to collect problems and deliver them to those who are responsible for correcting the problem in a systematic and appropriate manner. And fast you can also verify that the issue has been fixed or has not been resolved. For Kasetsart University to be even better.

KU Alert is an application for help in reporting problems within Kasetsart University, the principle is to let users report problems through the application. And then the application will bring these problems to the various agencies efficiently.

Why choose this project? Because of the many unexpected events today. Cause a loss of both life and property. From the mistakes of human beings themselves such as path with less fire Cause robbery Cause harm or water pipe in the bathroom leak Cause accident or Low power lines may cause harm to the passers-by.

The study aimed at a project. From the above mentioned problems, the problem has not been solved and solved. Resulting from when a problem occurs. The person experiencing the problem I do not know the agency that will report the problem after encountering the problem. Or have difficulty in reporting problems Cause the patch to ignore the problem. Make no edits or not as good as it should be. Make this project come up for Edit the above

This project will cause problems. Found on campus the tricky thing to do is to simply report the problem through the application. The application will forward the problem to relevant agencies quickly and efficiently.

Scope of the project

This project will bring the information that the person who needs it, The problem was solved using, Enter a name that you can call. Date and time of event. Press the SAVE GPS coordinates at the scene. Press the photo or video of the scene and the surrounding location. Fill out event details. Fill in what you want to edit





Smart Insurance for Android Application

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Nowadays, paying for insurance is apart of an investment as a guarantee for an unprecedented emergency case of a payer. As known by many, how important insurance is to our lives. Many people realize it is important and they pay for different types of insurance for a more secure assets and future. The importance of an insurance is based mainly because of our realization called life insurance or of that other people realize its importance and make us do it for some reasons called forced insurance such as insurance for an accident by car insurance for goods paid by money such as car insurance and an insurance for life in households.

This research is subjected to the development of mobile application called “Smart Insurance” on Android to manage an insurance system. It is developed through Android Studio for a usage on Android mobile devices. JAVA is used as a language on AndroidStudio and SQLiteis used as a database specifically targeting on storing data regarding information of the insurance such as life insurance, car insurance, both compulsory third party insurance and voluntary motor insurance. It is set to alert the user so that the payment goes exactly as planned to secure the maximum benefit of the insurance. Compulsory third party insurance and voluntary motor insurance are both important to insured person such as cost distribution for danger on life, body and asset of the insured person. This, for a convenience of managing the insurance user has applied to. The application itself has various functions to record and alert the user to conveniently manage the insurance payment. User can also store information of the insurance they have recorded such as picture and retrieve the insurance details of the company via each company website.

The result show that the usage of the application is easy to use and short response time. The application can also be used swiftly on its various function and stores, records, alerts and retrieves all the data completely as targeted. The application is suitable for all mobile phones with Android5.0 and above it is downloadable for free. Shortly, the application is useful for data management on wide range of operating system.

Keywords: Mobile Application, Android OS, Insurance Management.





Medicine Machine and Medical Reminder Application

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The objective of this project is to be a part of the way to solve a problem about skipped or too much medication is taken in elderly people. As you see in common, most family in the world has elderly people. Most elderly people have to take the medicine for their health, but If they didn't have a person who takes care of them, they might be skipped or take overdose medicine and its very danger for their health because the result can be deadly at worst. Our project will help elderly people who have to stay at home alone during the day and must take the medicine can medication taken correctly.

In our project, we will build the products that including one hardware, one application and connected it together by using IoT devices, ionic framework, firebase mobile and web application platform to help elderly people to take the medicine and to let their children knows if they skipped taking the medicine. This project will go on by following steps. First, we created hardware named "Medicine Machine" in a machine we build it with Arduino mega board, the sensor of IoT such as a buzzer, LCD, force sensor, servo, Node MCU. Second, we build an android application named "medication reminder" using the ionic framework. Third, we create the firebase platform for manage about authentication and real-time database. Fourth, the most important of my project is make three part in each step connected. we decided to connect a machine and an application via firebase. The flow process of our project starts with the user who has a medicine machine download a medication reminder application for connecting application and medicine machine. After the device connected, a user will able to set the time of taking a medicine for let the machine know the time to drop a medicine for elderly people. Moreover, if elderly people don't pick up the medicine in the tray, the machine will send the notification to their children who downloaded the application .Their children can double-check their parent by making a call to prevent elderly people skipped taken the medicine. We will test all of the processes in our project to ensure that our project can work correctly.

Keywords: Medical, Medicine, IoT, Ionic, Firebase, Reminder





Mobile Application for Korea language Translation and Tourism

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This objective of this abstract is to explain mobile application is called Mobile Application for Korea language Translation and Tourism. This application was developed by software Android Studio version 2.2.3 for windows, used database in SQLite 2.2-a to store data and supported the smartphones operating system Android. Otherwise, this application was invented for Thailand tourists who want to visit Korea forasmuch as Korea is popular tourist attraction of Thai people nowadays and they are further interested about Korean, culture, mini-series and food. However, the tourist often confronted with problems that cannot communicate with Korean and do not information about tourist attraction. So, this application gather information necessary for tourism. E.g. Korean vocabulary, conversation in daily life, meaning of vocabulary, word reading in Thai language and can be read aloud in Korean. So, these data allow a user to learn vocabulary in Korean. This application there is information of popular tourist attractions in Seoul, South Korea (history of tourist attractions, admission fees, opening time, route to the location), rules and etiquette, currency exchange rates Which allows tourists to be able to use the application to communicate with Korean. User will abide by the rule and etiquette of the tourist attractions in Korea. The application can connect to application Google map to allow users to travel to tourist attractions. And can calculate currency exchange rates to help tourists decide which items to purchase.

Keywords: Mobile, Application, Android, Translation, Tourist, Tourism, Korea, Tourist Attraction, Communicate





Smart Expense

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In the present, many expense applications have been developed to help users to record any expenses on the smartphone easily., allows the user to record new payments or to observe previous payments. The expense recording on the smartphone can be recorded anywhere but the main cause why many users stopped using expense applications is the user behavior.

The objective of this study was to help users not to forget to record their daily payments. The problems mentioned above is the main cause that why some users stopped using expense applications. However, other causes may be due to the complicated recording procedure. People have many activities each day, it is not easy to record the payment and numerous immediately. That makes the user forget to record the previous payment. There are a lot of people have ever used expense applications, but most users always forget to record some payments. When they forget more often, they would lazy to record some previous payments. That is the reason why most people stopped using expense applications. In addition, setting budgets do not respond to the needs of users as well.

This study plans to develop an expense application for Android operating system effective and easier to use. This expense application called Smart Expense. This study was very helpful to people who wants to know or to control their behavior about their payments. It is a convenient expense application because users can add their payments easily. There are many useful functional that will facilitate the user. For example, Smart Expense has a function to create many accounts for many budgets. In contrast, other expense applications may have only one budget in the application. They may use money in cash, credit cards or makes some financial transactions such as transfer. This function allows users to classify which accounts that they spend money. Moreover, this expense application can predict the user's behavior that might cause potentially spending from the location that the user was. For example, if the user was at the shopping mall, the application will detect a type of user's location and prompt users for any payment there. This function works as a reminder to remind users not forget to record. Besides, the device will get messages from the bank when the user makes some financial transactions. The application will check the number that sent the message if the number is match with numbers were set. It will process the message and get only the cost to add in the expense. The process will automatically classify type of expense which is income or expense. This expense application simplifies the recording process and makes it easier for users to manage or to observe their expenses. The result of this expense application helps to plan user's spending and to change their spending behaviors.

Keywords: Android, Budget, Expense





Class Scheduling Management System

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At present, university provides education under many curriculums and offers many courses for students each semester. The common problem each university faces is class scheduling and mapping them to class rooms. Class scheduling management system is proposed in order to help each department's administrators manage their classes and allocate classroom accordingly. If student want to register for a class but it has a time conflict with another, they shall not be able to register for both classes. The incident must be identified to the administrators immediately.

The proposed class scheduling system is developed as a Web-based. The application allows the professors to determine which courses they will offer at coming semester and select the time slot. The administrators manage the course information and assign a classroom to each offered course. The courses will be arranged such that there would be the least number of overlapping courses for students in the same year. It also make sure the availability of the room and the exam room and date/time. The information about classes, rooms, buildings and professors is stored in the database. The department administrator is responsible for managing the buildings, a classroom for each class, a classroom for exam as well as can manage new courses and departments. Professors select which classes to offer and specify the exam dates for each class. Then the administrators assign building and room to the subject. The highest priority is for senior year subjects which will be scheduled first then junior-year, then sophomore year and first year classes. Professors can submit his/her available time that they can teach. During the semester, there are a number of main subjects that students must register for if students do not take those classes, they will not graduate within four years of study plan. The application will report course schedule for students, analyze them if there exists any conflicts. And then the class schedule as well as exam date and time for each semester can be print out in the form of PDF file. The application allows the administrator and professors to reserve a classroom for doing other extracurricular activities. Teacher can reserve a room for teach extra slot if there is not enough time to cover all class materials. The reserve room will be shown after finishing the schedule the class. Teacher can also check the availability of classrooms.

Our Class scheduling management web application is expected to help solve the existing class scheduling problem. It reduces the class time conflicts and helps manage classroom for efficiently and effectively.

Keywords: class scheduling, class management





Smart Microgreen

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This project included the process of applying Internet of Thing (IoT) into the smart farming, which specify in hydroponic farming in order to plant organic vegetables at home. Our project will support none-professional farmer to do vertical planting in very small area in the city such as the building top, the balcony of small room in high-rise building and in the small office spaces. In hydroponic farming, it is difficult to plant and manage many factors during the farming processes. For example, water and air temperature, PH value, concentration and Electrical Conductivity (EC) value of nutrient solutions. The farmer have to measure all factor then take action when that metric is not proper, for example good temperature range for planting is between 20 to 30 °C, farmer have to reduce temperature by open water springer in a specific time, no more and less, but people in the city cannot take action as well as professional farmer.

In this project; we, project owner, will focus on learning how to build Hydroponic Farming System (HFS) by using IoT devices to support city people with limited knowledge in farming to do their own organic vegetables farm in their home.

This project will cover 3 steps, designing, building and testing HES model. First step, designing model HES. We have designed our HES by using the IoT to link throughout our hydroponic farm by making channels between the sensors and the server. An example of some functions: the server can receive then analyze data from the temperature sensors to control the sprinkler watering in order to reduce the air temperature. In addition, the server can receive data from the PH and EC sensors to control values concentration of nutrient solutions for modify values to stay in a constant balance or stay in range of threshold. Furthermore, the data will be collected in a database server or send it to the mobile application. A mobile application makes IoT concept to be completed because it can be alert to mobile's user if the farm is in the bad situation such as higher temperature or the nutrient solutions at the plantation is not appropriate and also the users can control all sensors which are installed at the plantation. At the part of things in IoT which is in the second step of this project. The part of building or the making of all the parts in our product. We will be using the Arduino board and a microcontroller board (to connect all the sensors, and to connect to the network). The data that we will use in this project include temporary and humidity values of the air temperature, data about the nutrient solution and water flow at planting vegetables rails. We make sensors for controlling the nutrient solution and reducing the temporary in farming area. Final step we install all sensors in HFS. This system was tested to make sure that our system can run properly through a prototype that we made, and we plant green oak with HFS and follow the results of the testing.

Keywords: Smart Farm; Internet of Things; Arduino; Automatic Planting; Mobile Application





Apply RFID for Trolley Management System (TMS) for small retail business

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Statistic of using trolleys significantly increase due to the fact that trolleys are the tools for assisting people in many businesses or services. Supermarket and airport are an examples of those businesses. Many of problems are caused by the huge number of customers. Missing trolleys is the main problem that each businesses have to concern about. Therefore, Trolley Management System (TMS) has been created. In this systems, combined the idea of Internet of Things(IoT) with Radio Frequency Identification (RFID). RFID technology can identify data by using radio frequencies. There are many advantages of RFID for example, it can read wirelessly and record a huge number of data. Equipment is also easy to maintain. This system provides many functions to help the staff, particularly through the handling of trolleys. The Queueing Theory will also be mentioned in this paper, the formula shows the optimal solution to collect trolleys. With the help of our TMS, we can allocate a practical system to manage the resources of trolleys. Due to the fact that trolleys are dispersed all over the place when customers are finished with them, we have a very good reason to make a Trolley Management System (TMS). Staff have to spend a lot of time to figure out where they have been put and slowly collect all the trolleys. Devices that we use are RFID Reader, RFID Tag, Arduino Data Logger Shield, Arduino Board and Module Wifi. In this project, we test the system by creating a small scenario. A scenario is divided into 4 area. In each area will install reader in an appropriate area for reading data through RFID tag. These data will be real-time updating to database which collect all necessary information for Trolley Management System(TMS). The way we send data to database is using module wifi. Information in database will be analyzed in propose to show those data on TMS website. Staffs will use these information in order to manage trolley, searching trolley's data also provide in this website. TMS website restrict for staff in this working area.

In conclusion, Trolley Management System(TMS) is the management system for trolley by using RFID which is a part of IoT. This system will reduce people to work in area and increase staff efficiency in working area. This project still has a limitation. Now TMS can use in a small scenario area , in nearly future TMS will able to apply in another place for instance airport. There are many airport that start using RFID apply on their area for example, Suvarnabhumi Airport and Donmuang Airport in Thailand. More feature that we expect to add more in this project is weight detector. This feature will help to indicate that in those trolley are used or not and also can use in the security. In the work field of security, weight detection may indicate object that is packed in the suspicious bags.





Dietary for Kidney care Application

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Kidney disease or failure is a condition where patients lose their kidney functions. This results in the body being unable to drive water and waste out of the blood resulting in congestion to the point of poisoning the body. It also affects the electrolyte balance and blood pH including causes some hormone created by the kidneys being depleted. All these changes lead to abnormalities of almost every part of the body. Renal disease is caused by complications of diabetes and high blood pressure that has seriously gone untreated on most patients. Diabetes and high blood pressure are caused by the lack of planning and control of the amount of food eaten at each meal.

Currently, there are roughly 8 million Thai people suffering from chronic kidney disease or 17.6% of the population. There are two hundred thousand people in the last phase. The number of patients increases more than 7,800 per year. Dr. Supachai (Nongkhai Hospital Director) said that kidney disease is currently a major global problem. The number of Thais being sick with kidney disease is likely to increase. Thai people with kidney disease is ranked 3rd in ASEAN behind Malaysia and Singapore. 70% of kidney disease is caused by diabetes and high blood pressure with the total number of patients is nearly 15 million. The result is faster kidney failure if fail to correctly behave. There are only 500 kidney transplant surgeries per year so the focus must be on slowing down the kidney degeneration to slow down the dialysis stage.

The problem of chronic kidney disease patients is reduced Glomerular Filtration Rate (GFR). Normal GFR for healthy people is over 90. If the GFR is roughly 90, it indicates that kidney is starting to degenerate. This results in decreased kidney function such as balancing the body fluid by filtering excess fluid from the blood, controlling blood water and minerals such as sodium, potassium, phosphorus, and calcium, removing waste from the blood, removing drugs and toxic from the body, and secreting hormones into the bloodstream. This includes hormones that help control blood pressure, produce red blood cell production, and retain bone strength. Therefore, controlling food consumption by taking the kidney health into account is of primary importance for kidney disease patients or those at risk of kidney disease.

From the mentioned problem, the researcher came up with the idea of developing an application that helps process the nutritional value of food that patients consume as to the criteria that would be dangerous for kidney disease patients, recommend food and drink consumption, and prohibition for kidney disease patients to guide kidney disease patients practice.





Food Ordering Online Application

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This project was created for customers and restaurants to reduce a time to ordering foods. Otherwise, this project will help the restaurants to have more convenient managing their restaurant. In the past, ordering foods take a long time, sometime the customers hadn't got foods that they ordered yet, or they didn't get their foods correctly. But on this day, many restaurants was established, and they confront with the ordering problem that made the customer didn't appreciate so far. Developers know the way of this problem is "The food online ordering". The customers can order foods and reserve the table though this application before they coming. And the employees that work in the restaurant can have the customer orders or can track foods status and make out a receipt for the customers either. Otherwise, the owner can manage restaurant, control employees and summarize circulation through this application. The food online ordering supported with website and Android smartphone by developed website with Visual Studio Code, and programing with PHP language and developed the application with Android Studio version 2.2.3 for windows. Database was created in phpMyadmin. So, "The food online ordering" will help many customers to have more convenient and will help restaurants to have more profits. In the future this application also applies for other restaurants.

Keywords: Restaurants, Customer, Order foods, Convenient, Reserve the table, Circulation, Receipt, Website, Application, Android





The Study of Hydrogeology and Groundwater Potential in KMUTT (Ratchaburi learning park) Using Detail Resistivity Imaging

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KMUTT (Ratchaburi learning park), a particular drought zone with mainly metamorphic rock aquifer zone where located at Rangbua subdistrict, Chombueng district, Ratchaburi province, is found a limit of surface water. As a result, the most alternative water supply must be from groundwater resources. Then the study of hydrogeology and groundwater potential, detail 2D resistivity measurement was applied to locate high potential zone using Geomative resistivity instrument, GD-10 model with 84 multi-electrodes. Six survey lines were designed in E-W covering the campus area of about 2 km in E-W and 800 m in N-S with 720-840 m length. Data collecting in the field, Schlumberger configuration was taken with resolution of 10 m for continuing detail along survey line with depth of >120 m with electrodes spacing of 10 m. In data processing stages, both 2D and 1D inversion models have been done for details vertical resistivity change in every 10 m. From hydrogeological study groundwater can be found as groundwater flow through the campus are S-N direction. At least 3 location of high yield groundwater can be indicated of northern part of the campus. Aquifer may found in the fracture of local bedrock with depth of 50-80 m. These high yield groundwater positions are recommended for groundwater drilling in the future water supply in the campus.

Keywords: 2D Resistivity, Groundwater, Potential Groundwater Zone, Detail Resistivity Imaging.





Contamination of Heavy Metals in Leachate, Groundwater and Soil at Khon Kaen Municipal Solid Waste Landfill, Khon Kaen Province

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Khon Kaen municipal solid waste landfill is a major landfill that it supports large amounts of waste from many areas in Khon Kaen. In addition, the Khon Kaen municipal solid waste landfill is also close to the community and agricultural areas. Therefore, contamination of pollutants, as heavy metals, should be occurred. This study aimed to study the contamination of heavy metals in leachate, groundwater and soil at Khon Kaen municipal solid waste landfill. All samples were collected during rainy season in September 2016 and dry season in December 2016. Leachate samples were collected in leachate treatment plant, as influent and effluent, groundwater samples from 3 monitoring wells and soil samples from 5 stations. For the leachate and groundwater samples were divided into 2 parts, as filtered through filter paper no. 1 and no.42 and digested with HNO_3 and HCl as following with USEPA 3005A method. For the soil samples were performed according to the USEPA guideline (3050B method). Heavy Metals (As, Cd, Cr, Mn, Ni, Pb and Zn) were analyzed by Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES). The results showed concentration of most heavy metals in leachate during rainy season were higher than during dry season. Moreover, concentration of Cr (7.44 mg/l) was higher than the leachate quality standard (5.0 mg/l) in influent leachate during rainy season. During dry season, the highest efficiency of the leachate treatment system could reduce about 100% in Cd, Cr, Mn and Zn in filtrate leachate samples. In groundwater, Pb were higher than the groundwater quality standard (0.01 mg/l) in all groundwater samples, during both rainy and dry season. Particularly high concentration during rainy season were 1.84, 2.13 and 1.23 mg/l in monitoring wells no.1, 2 and 3, respectively. In soil samples, the result of Enrichment Factor (EF) showed values of EF are indicated to no to extremely severe enrichment, the highest values of EF are found in soil no.5 by Cd, both during rainy and dry season, at 117.11 and 64.74, respectively. Geo-accumulation Index (I_{geo}) values are indicated to uncontaminated and uncontaminated to moderately contaminated, except for Cd during dry season in soil no.1 and 4 are indicated to extremely (5.67) and strongly contaminated (3.44), respectively. Contamination Factor (CF) values are indicated to low and moderate contaminated, except for Cd during rainy season in soil no.5 (3.39) and during dry season in soil no.2 (3.58) are indicated to considerable contaminated. Moreover, CF of Cd during dry season in soil no.1 (16.25) and 4 (10.23) are indicated to very high contaminated. However, Pollution Load Index (PLI) values in each sampling site are indicated to no pollution. In conclusion, the concentration of heavy metals are generally quite low. However, some toxic heavy metals, such as Cr, Pb and Cd, are higher than standard values. Therefore, the contamination of these heavy metals in the environment at Khon Kaen municipal solid waste landfill should be strictly controlled.

Keywords: Heavy Metals, Contamination, Leachate, Groundwater, Soil, Enrichment Factor, Geo-accumulation Index, Contamination Factor, Pollution Load Index





Internal Structure of Hydrophane Opal from Ethiopia

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Opal is one of the famous gemstones in the world with specific phenomena called play-of-color. It could be found in many gem deposits such as Australia, Brazil, and Ethiopia. Ethiopian opal shows not only play-of-color but also a hydrophane character an ability to absorb water and to change the transparency from opaque or semi-translucent to translucent or transparent. The objectives of this research are to study physical properties, spectroscopic properties, and the internal structure of hydrophane opal after soaking in water, acetone, and olive oil. Thirty-nine samples were determined using scanning electron microscope (SEM), fourier transform infrared (FTIR) spectrometer, UV-Vis-NIR spectrophotometer, and colorimeter. Most of the samples soaked in water and acetone are higher specific gravity than unsoaked samples. The samples soaked in olive oil are lower specific gravity than unsoaked samples. The infrared spectra of the samples before and after soaking showed a board band of water molecule approximately $5,300\text{ cm}^{-1}$ and silanol (SiOH) about $4,600\text{ cm}^{-1}$. According to UV-Vis-NIR spectra, all the samples showed absorption peaks about 1,420 nm. In fact, after immersing in water and acetone, the opal samples became transparent and the play-of-color effect was disappeared. Some of the white opals changed to brownish orange body color after immersing in olive oil. Opal samples consisted of hydrated silica spheres with 100 nm to 470 nm diameter. After the experiment, it could be concluded that hydrophane opal is a porous material, whereas, the immersing method did not effect to its internal structure.

Keywords: Ethiopia, Hydrophane opal, internal structure, spectroscopy, SEM





Crystal Structure of Zircon Samples before and after Heat Treatment

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The aim of this research is to study the zircon crystal structure from different origins before and after heat treatment. The samples were collected from six localities including Ratanakiri, Pailin, Preahvihear in Cambodia; Ratnapura in Sri Lanka; Chanthaburi and Kanchanaburi in Thailand. The samples could be divided into two groups by specific gravity. The first group is high zircon (crystalline zircon) from Ratanakiri, Pailin, Preahvihear in Cambodia and Chanthaburi, Kanchanaburi in Thailand with a specific gravity as 4.47 – 4.48 showing brown to reddish brown color. The second group is metamict zircon samples from Ratnapura in Sri Lanka with a specific gravity as 3.93 – 4.03 showing green color caused by the trace radioactive elements to destroy in crystal structure relating to geology. The Raman spectra of unheated showed peak position around 1007.7 cm^{-1} – 1009.0 cm^{-1} . After heat treatment at 1000°C in reducing atmosphere for 60 mins, the high zircon group change to blue color to near colorless and the FWHM of the Raman spectra are decreased and shifted to higher peak position. For the metamict zircon group, the samples change from green to yellowish green and the Raman spectra were changed from broad band to narrow peak base on higher peak position. Consistent with XRD results, it could be concluded that heat is predominantly zircon due to the recrystallization.

Keywords: Zircon, Heat Treatment, Raman Spectroscopy





A Study Meteorological Drought in Thailand using the Standardized Precipitation Index

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Thailand has been frequently confronted with drought as main causes of an erratic distribution of rainfall and dry spells in the rainy season. The Standardized Precipitation Index (SPI) is a widely used index to characterize meteorological drought on a range of timescales. The objectives of this study were to study spatial pattern of meteorological drought in Thailand using SPI. Monthly rainfall data for the period 1980 to 2016 from 114 meteorological stations distributed in Thailand were collected. Meteorological drought maps were produced by using Geographic Information System (GIS) by the Kriging interpolation method. The results revealed that in the period 7 years (2010 to 2016), the meteorological drought occurrence distributed in the whole region of a country. The area that no drought during the period of study was 0.01% of the country, the area that drought occurred 1 to 2 times was 45.3% of the country in scattered the whole region, the area that drought occurred 3 to 5 times was 53.9% of the country in Upper Northern, Northeastern and Southern and the area that drought occurred 6 to 7 times was 0.7% of the country in Northeastern and Southern. The SPI index relates to the drought analysis in Upper Thailand but, it had an error in the Southern. Therefore, the SPI values should be adjusted for relating to the area of the Southern region.

Keywords: Meteorological Drought, Standardized Precipitation Index (SPI), Thailand





Attractive Geotourism in Phrae Province

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This research aimed to describe the geological characteristics of the tourist attraction in Phrae province. There are many natural attractions in this province. However, the geological information is not enough for educating and informing the tourists. This investigation studied the geological features, field survey and rock sample collections for petrography analysis. The result from this study shows that tourist attractions can be divided into three types by landforms and geological process. First, the weathering and erosion landform, there are 13 places. They are consist of karst landform (cave and dissolve limestone), waterfall and fluvial process landform. Second, the volcanic landform, there are 5 places. They are consist of hot springs at the Phrae fault zone, the south western of Phrae province and the columnar basalt at Den Chai and Wang Chin districts. Finally, the fossils attraction, there is a good exposure at Bo Fossil Hoi Nguang Chang. In addition, some places in this study can be both karst landform and fossil attraction, such as Pha Klong cave and Suan Hin Maharaj. Understanding of the geological process can also be applied to make plans for conservation, protection, rehabilitation and development of a sustainable tourism.

Keywords: Geotourism, Tourist attraction, Phrae





Seismic Reflection Survey for Fault Detection in Chiang Rai Province, Northern Thailand

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Chiang Rai province in northern Thailand contain several know active faults that have caused many earthquakes in the region. This study aims to conduct a shallow seismic reflection survey to explore the hidden fault in the study area in Mae Suai district, Chiang Rai province. This study consists of 2 seismic survey lines along southeast-northwest direction with total length of 5,470 meters. The result of this study suggests that there are 20-50 meters thick of sediment layer or weathered rock overlying on the bedrock which may be related to the deeper fault that has caused earthquake in this region.

Keywords: Seismic Reflection, Fault, Chiang Rai Province





Seismicity of Chom Thong and Mae Wang Districts, Chiang Mai Province, Northern Thailand

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This study aims for collect and analyze earthquake from 20 seismic stations occurring in Mae Wang and Chom Thong districts, Chiang Mai province during 21 January 2016 to 27 February 2017 using SEISAN to understand the nature of these earthquake swarm. The result indicate that there were 127 earthquakes with magnitude (MI) from 1.0-4.2. (34 earthquakes of MI 1.0-1.4, 44 earthquakes of MI 1.5-1.9, 30 earthquakes of MI 2.0-2.4, 13 earthquakes of MI 2.5-2.9 and 7 earthquakes of MI greater than 3.0. These earthquakes were located in Ban Luang sub-district, Chom Thong district and Mae Win and Mae Gad sub-districts, Mae Wang district of Chiang Mai Province with the majority of them located in Mae Win sub-district, Mae Wang district, Chiang Mai. The biggest earthquake of MI 4.2 were located at Ban Luang sub-district, Chom Thing district, Chiang Mai at latitude 18.592 °N and longitude 98.546 °E at 2.7 km depth with oblique slip motion.

Keywords: Earthquake, Magnitude, Oblique slip motion





Contamination of Heavy Metals in Soil and Farm Plants near Ban Kam Bon Landfill Khon Kaen Province

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Contamination of heavy metal in soils and crops around the landfill tends to continuously increase due to many factors. Khon Kaen has a huge landfill for supporting garbage and waste from the province, called Khon Kaen municipality landfill. Around the landfill site, there are many crops, especially cassavas and canes which are industrial drop. Thus, crops may be contaminated by heavy metals from the field. This research aimed to study the potential of contamination and distribution of heavy metals in soils and crops in order to compare the concentrations of heavy metals with the standard and control the heavy metals contamination. Soils and plants were collected during rainy season (September 2016) and dry season (December 2016) from agricultural area around landfill site. Cassava samples were collected from 3 stations, 3 replications, divided into 3 part; roots, stems and leaves. Cane samples were collected from 1 station, 3 replications, divided into 2 part; stems and molasses. The soil and plant samples were air dried and digested by HNO₃, HCl and H₂O₂, as following by USEPA 3050B method. 8-heavy metals in the samples (Arsenic, Cadmium, Chromium, Iron, Manganese, Nickel, Lead and Zinc) were analyzed by Inductivity Coupled Plasma-Optical Emission Spectroscopy (ICP-OES). The results show that manganese concentration is high in soils and in cassava leaves, with 105.75 mg/kg in soil and 132.13 mg/kg in cassava leaf. The high level of nickel concentration was detected in cassava leaves, with 66.875 mg/kg Zinc accumulates in stems and cassava leaves, with 70.33 mg/kg in stem and 106.25 mg/kg in cassava leaves. However heavy metals in all samples are not exceed to the standard.

Keywords: Heavy metal, Contamination, Soil, Cassava, Crop





Mapping Shallow Groundwater in Dan Chang-Nong Ya Sai area, Suphan Buri Province by Using 2D Resistivity Imaging Technique

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The old sand channel, presented as significant shallow groundwater for support in agriculture during dry season, hardly found at a margin of central Chao Phraya basin. The study area, Dan Chang - Nong Ya Sai area is selected by GIS analysis of sandy soil map. The trend of sand deposit (or shallow groundwater zone) in East - West direction which following the lineament of a surface stream. Subsurface feature of shallow groundwater can be investigated with 2D resistivity measurement. Three survey lines covering in Northern to Southern of Huai Krasiao stream (located from GIS mapping) with the length of 2 km and with line spacing of 200 m. 2D resistivity imaging technique with multi-electrode system with Schlumberger and Dipole-Dipole array was applied with electrode spacing of 5 m and target depth of 20 m. The result can display the channel of shallow sand deposit zone which present as slightly high resistivity ($>40 \Omega\text{m}$) with depth about 10 m and with dimension of 10 m deep and about 40m - 120m wide with lineament in West to East direction. The sand deposit zone is found as a high yield of shallow groundwater. However, geoelectrical section obtained from data processing is generally found as low resistivity ($<10 \Omega\text{m}$) which present as clay deposit. Interpretation of the data revealed that the layers of buried sand channel which is deposit from the old stream with the alluvial fan from West to East and became a potential shallow groundwater for agricultural activity.

Keywords: Sand channel, Shallow groundwater, 2D resistivity imaging





Geology of gem deposits in Bo Rai District, Trat Province

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In the past, Bo Rai District, Trat Province, was the famous in Gem deposits such as Siam Ruby, which closely associated with Cenozoic basalt. At present, the quantity of gem in Trat province has been decreased considerably. Therefore, additional exploration and geological mapping are required to study the distribution and rock units. This study focuses on the details of aerial photograph interpretation, field surveys and collecting samples to create the update geological map. Other techniques are needed to use with the study for obtain the accurate information and increase the resolution. For example, Petrographic analysis and X-ray diffraction technique (XRD)

From aerial photography interpretations, it appears the landform of Bo Rai district consists of six units. Highland is located at the eastern of Bo Rai district near Cambodia border. Deposition of colluvium and talus are distributed at the eastern foothills of Bo Rai district. Alluvial fan, Undulating terrace and Floodplain spread throughout the Bo Rai district. Moreover, the rock units in this area were divided into three rock types: graywacke, siltstone and basalt. The basalt in Bo Rai consists of three basalt areas: Ban Nong Bon - Ban Suea Dao, Ban Khlong Yo and Ban Nonsi. Particularly, gemstones, such as ruby, spinel, and garnet were found at Ban Khlong Yo. In addition, basalt were analyzed by petrography analysis. Mineral compositions are olivine, pyroxene and feldspar. X-ray diffraction was employed for the mineral compositions in this work and confirm the accuracy of the study. The results showed that mineral compositions are augite, nepheline and analcime.

Keywords: Trat, Basalt, Bo Rai, Gem deposits





Carbon capture and storage (CCS) by mineral carbonation using carnallite from Mahasarakham Formation, Khorat Group

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Carbon capture and storage (CCS) is an alternative method to reduce carbon dioxide (CO₂) emission to atmosphere. There are many methods have been developed over a decade to store carbon dioxide (CO₂). However, in this study, storage of carbon dioxide (CO₂) in the form of mineral carbonates by mineral carbonation is selected using the local geological material in Thailand, which is carnallite mineral. Carnallite mineral is designated as a waste in potash mining process at the Mahasarakham Formation, Khorat Group. Totally 15 samples from the northeast of Thailand were preliminary examined by X-Ray diffraction analysis (XRD) and thin section through the polarizing petrographic microscope. The results showed that samples consist mainly of halite. More samples are collecting and further processing.

Keywords: Carbon capture and storage (CCS), Mineral carbonation, Carnallite





Developing 3D S-Shape Resistivity Approach to Determine Shallow Cavities at Limestone Quarry of SCCC Limestone Mine, Kaeng-Khoi District, Saraburi Province

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Shallow cavity found at the quarry in limestone mining of Siam City Cement Public Company Limited, at Kaeng-Khoi district, Saraburi province, may cause sinkhole collapse due to the heavy vehicle working. The known of shallow cavity in limestone mining can be help to manage safety in mining activity. 2D resistivity can successfully be applied to detect shallow cavity. 3D S-Shape configuration approach is a new challenge to locate cavities in more details than 2D resistivity imaging. In data acquisition stage, location of taking measurement located at the previous cavity zone, survey lines were arranged by continue 3 parallel lines into S-Shape. The length of each survey line is 180 m with line spacing of 20 m, and electrode spacing of 5 m. 2D and 3D resistivity measurements were operated with Schlumberger and Dipole-Dipole configuration. Format of data collection of 3D S-Shape is gathering data between survey lines by electrode in other line which different to 2D resistivity imaging. In data processing, the 3D inversion model was produced in 3 types i.e. pseudo-3D model (from combined 2D resistivity data), Schlumberger 3D model, and Dipole-Dipole 3D model. The results of 3D inversion model show shallow empty (air) cavity zone with very high resistivity ($>5000 \Omega \cdot m$), surrounded limestone layer with lower resistivity zone ($100-2000 \Omega \cdot m$). The depth of cavities found by 3D inversion model is 8 m beneath the ground surface with dimensions of 25 m long and 20 m wide. 2D inversion model was shown shallow cavity zone at 6 m beneath the ground surface with a length of 10-25 m. As the results of 3D S-Shape inversion model provide deeper and display in 3D view much better than 2D resistivity imaging.

Keywords: 3D S-Shape Resistivity, Resistivity, Shallow Cavity, Limestone





Characteristic of Au-mineralization of Huai Kham On Gold deposit, Phrae Province, Northern Thailand

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Huai Kham On gold deposit is located in Ban Mae Kra Thom, Soi sub-district, Wang Chin district, Phrae Province about 545 km north of Bangkok in Thailand. Huai Kham On gold deposit is situated in Sukhothai fold belt. Orebody of Huai Kham On consists of veins, veinlets, and stockwork breccias ranging from 5 to 250 cm. The gold-bearing quartz vein is hosted in andesitic tuff, volcanic sedimentary rock, and polymictic rhyolitic breccias of Permian-Triassic age.

The gold-bearing quartz vein in Huai Kham On gold deposit is divided into 6 stages based on the cross-cutting relationships and mineral assemblages. Stage I: quartz-adularia-calcite-pyrite-galena-chalcopyrite-sphalerite-electrum vein, Stage II: quartz-calcite-adularia-pyrite-chalcopyrite-galena-sphalerite-electrum vein, Stage III: quartz-adularia-dolomite-calcite-chlorite-pyrite-chalcopyrite-galena vein, Stage IV: quartz-dolomite-calcite-adularia-laumontite-chlorite-pyrite-chalcopyrite-sphalerite-galena, Stage V: quartz-dolomite-calcite-pyrite, and Stage VI: dolomite-quartz.

Intense of gold mineralization occurred in Stages I and II. The mineral assemblages of Stages I and II are characterized by a large amount of pyrite, galena, chalcopyrite, quartz, and a small amount of calcite. Electrum coexists with pyrite, galena, and chalcopyrite.

Keywords: Huai Kham On gold deposit, Gold-bearing quartz vein, Electrum





Surface Morphology of Irradiated Sapphire Samples from Phrae, Thailand and Sri Lanka

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The irradiation is one of the methods to improve the quality of gemstones, for example, diamond, zircon, topaz and sapphire. This technique is applied for increasing the gem price. Nowadays, it is hardly to identify the irradiated gemstone by the naked eyes. While the potential methods such as fade testing used for detecting the irradiated gemstone are the sample-destructive method. The purpose of this study is to analyze the surface morphology before and after irradiated sapphire by atomic force microscopy (AFM). In this study, the sapphire samples from Phrae, Thailand and Sri Lanka were divided into 3 groups for high energy electron beam irradiation at 40,000 kGy, 60,000 kGy and 80,000 kGy. The sapphire samples were irradiated at Thailand Institute of Nuclear Technology (Public Organization). As the results, the surface morphology of irradiated sapphire has been changed especially the roughness. It could be summarized that irradiation also destroys some of the micro-structure showing as a defect on the sample surface. Therefore, this study will be useful in the gemstone enhancement identification.

Keywords: Sapphire, Irradiation, Atomic Force Microscopy (AFM)





Critical Impact of Contamination Level in Mangrove Sediment Intervention Project; in Case of Chu Lai Economic Zone, Quang Nam Province, Central of Vietnam

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Chu Lai economic zone is located at coastal environment in An Hoa lagoon, Quang Nam province, Vietnam. An Hoa lagoon has river which connects between Quang Nam province and Hoi An city. Around Chu Lai economic zone has many industrial activities and agricultural activities. Thus, the anthropogenic activities can cause the impacts on the environmental condition, as increasing of level of heavy metals in mangrove sediment. Accordingly, sediments in this area should be monitored the level of the pollution that caused by heavy metals. Mangrove sediment samples from 8 sites in and around Chu Lai economic zone were collected in October 2015 and analyzed heavy metals (As, Cd, Cr, Pb, Mn, Ni, and Zn). The aim of this study was to determine the level of heavy metals contamination by using geo-accumulation index (I_{geo}), enrichment factor (EF), contamination factor (CF), and pollution load index (PLI). In present study, sediment samples from this area were air-dried and digested with nitric acid, hydrochloric acid, and hydrogen peroxide, as following by USEPA 3050B method. Sample solutions were analyzed by Inductive Coupled Plasma Optical Emission Spectrometer (ICP-OES). The measured concentration in mangrove sediment was compared to the Canadian sediment quality guidelines for the protection of aquatic life (CCGQ). The concentration of heavy metal in sediment are lower than the CCGQ, except Pb exceed to the CCGQ for the threshold effect level (TEL) and Zn is also higher than the probable effect level (PEL). The Geo-accumulation index (I_{geo}) show that the sediments quality is unpolluted to moderately polluted (class 0 - class 1) in sediment samples. The high values of EF show in all of the studied metals, especially in Zn, Ni, Cr, Pb, and As. The CF values for Zn are very high contamination (>6) and other studied metals are ranged from 1-6 (low to considerable contamination). The PLI indicated that all samples have no metal pollution, except the sample 5, which taken from Chu Lai economic zone seaport. Heavy metal contamination in mangrove sediments is derived from human and industrial activities, especially the metals released from industrial seaport and the point source of wastewater. Therefore, the heavy metal contamination should be controlled, such as improvement of wastewater treatment system and using phytoremediation in order to remove heavy metals in coastal sediment.

Keywords: Heavy metals, Contamination, Sediment, Geo-accumulation index, Enrichment factor, Contamination factor, Pollution load index, Vietnam





Geology and geochemistry of volcanic rocks in Huai Kham On gold deposit, Phrae Province, Northern Thailand

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Huai Kham On area is located in Sukhothai fold belt about 545 km north of Bangkok in Thailand. Geology of the area consists of three units such as polymitic andesitic breccias, volcanic sedimentary breccias and andesitic tuff in ascending order. There is granite intrusion in the South of the area. The gold bearing quartz veins cut all the units.

The lower part of polymitic andesitic breccia have thickness is about 60 meters thick, it overly by subhorizontally of volcanoclastic rocks (1-5 m). The upper part is andesitic tuff. Polymitic andesitic breccias is characterize by reddish brown to brown, consist of andesite clasts (2-5 cm) and fine-grained matrix (0.5-1 mm). Clasts are composed of andesite, fragment of quartz. The groundmass consists of quartz, feldspar, plagioclase, and hornblend is replaced by chlorite.

Volcanic sedimentary rock is characterized by reddish brown, pebble to silt grain size and rounded shape. Matrix consists of fine-grained (0.2-0.4 mm) of plagioclase, quartz and feldspar. There are hematitic alteration in matrix.

Andesitic tuff is characterized by grey to green, showing porphyritic texture. It is composed of hornblende, feldspar and plagioclase phenocrysts (1-3 mm), microlith of plagioclase (0.3-0.5 mm). Quartz filled in groundmass. Sericite replaced K-feldspar and plagioclase. Chlorite replaced hornblende. There are present of pyrite and ilmenite.

Keywords: Sukhothai fold belt, polymitic andesitic breccias, volcanic sedimentary rock





Orientation of Silica Spheres in Precious Opal Samples

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In gem market, opal is commercial gemstones because of its beautiful and rare. It is deposited at a relative low temperature and may occur in the fissures of any kind of rock. Australian opal has often been cited as accounting for 95-97% of the world's supply of precious opal and it shows an unique and beautiful play of color. The aims are to study physical properties, spectroscopic properties and the orientation of silica spheres in precious opal. Eight opal samples were studied using scanning electron microscope (SEM), fourier transform infrared (FTIR) spectrometer and UV-Vis-NIR spectrophotometer. The infrared spectra of the samples showed the board band of water molecule around 5,200-5,400 cm^{-1} and SiOH peaks tentatively 4,300-4,500 cm^{-1} . According to UV-VIS-NIR spectra, all samples show strong absorption peaks of H₂O or OH on about 1405 and 1912 nm. Scanning electron microscope showed morphology of hydrated silica spheres.

Keywords: Opal, Precious opal, Silica sphere, Morphology





Imaging Plant Root Form by Resistivity and Induced Polarization Monitoring with 3D S-Shape Configuration

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Resistivity and induced polarization (IP) measurements with 3D S-shape configuration were successfully techniques that have been proved as a non-destructive method to picture root form for understanding ecological processes and behavior of plant root that will apply to use in bioengineering for protect landslide occurrence. Sunflower were observed with repeated 2D and 3D S-shape configuration resistivity and IP measurement in lab experiment planting with wood frame covering the area of 1 m wide \times 1 m long \times 0.1 m high. Field resistivity and IP instrument of Geomative (GD10 model) was modified for lab scale by using new designed accessories e.g. wiring box and copper electrode. In lab scale experiment, 5 survey lines with length of 1.6 m were designed to covering wood frame with spacing of 0.3 m. Each survey lines contain 85 electrodes with electrode spacing of 0.1 m in resistivity measurement and 0.2 m in IP measurement. 2D and 3D resistivity and IP measurement were measured with schlumberger and dipole-dipole configuration every week in lab scale experiment that planting sunflower in the half plot cover area of 0.5 m wide \times 1 m long. The results of 3D measurement show that resistivity and IP data illustrate a slightly change from beginning planting to after 5 weeks. Resistivity inversion show the resistivity change from 10-30 $\Omega\cdot\text{m}$ to high *value* about 20-50 $\Omega\cdot\text{m}$. While IP result can not show clearly variation value in the area of sunflower but the area not have planting resistivity and IP value is quite constant.

Keywords: 3D S shape configuration, Resistivity, Induced Polarization, Landslide





Mineral Inclusion in Sapphire Samples from Sri Lanka after Thermal Enhancement

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An inclusion is any minerals or features trapped in the host mineral such as liquid, minute particles and crystals of minerals. Inclusion affects to the clarity of the gemstone. Nowadays, heat treatment is a famous method to improve gemstone quality. The aims of this study are to identify the mineral inclusions in sapphire samples and to analyze the effects of heat treatment on inclusions by using Raman spectroscopy combined with Gemolite (Gemstone microscope). Twenty sapphire samples from Sri Lanka have been characterized before and after heat treatment in reducing condition. Besides, the spectroscopic properties are applied to study the samples by the advanced instruments including Fourier transform infrared spectroscopy (FTIR), Ultraviolet-Visible-Near Infrared spectrophotometer (UV-VIS-NIR spectrophotometer) and Colorimeter. As the results, the FTIR absorption spectra of sapphire samples showed the pattern of CO₂ at 2339 cm⁻¹, the C-H stretching at 2921 cm⁻¹, H₂O bonding shown as broad band at 3500-4000 cm⁻¹. Milky to colorless samples showed tiny absorption in a visible range. Fluid inclusion, two-phase inclusions, rutile (TiO₂) and abundant colorless negative crystals commonly found in Sri Lanka sapphire samples. According to some of the negative crystals, the Raman spectra showed peaks of spinel (MgAl₂O₄) which is indicated to be associated mineral of sapphire from Sri Lanka. After heat treatment, some inclusions were dissolved or changed their shape. Since, the gemstone clarity could be improved by heat treatment.

Keywords: Sapphire, Sri Lanka, Heat treatment, Inclusion, Raman spectroscopy





Hydrological Characteristic of Upper Khwae Noi Watershed at Thong Pha Phum District, Kanchanaburi Province

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The aim of this study to analyze hydrological characteristics of Upper Khwae Noi watershed. The result revealed that the study area has an average annual rainfall of 1,789 millimeters that affect to 7 sub-watersheds at upper Khwae Noi watershed that include Ban Sai Yok sub-watershed (K.22B), Ban Nam Chon sub-watershed (K.31), Huai Lin Thin sub-watershed (K.38A), Ban Ung Thi sub-watershed (K.39), Ban Hin Laem sub-watershed (K.50), Ban Lin Thin sub-watershed (K.54) and Ban Kui Mang sub-watershed (K.60) that analyzed into 3 groups. Group I contain K.38A, K.39, K.50 and K.60. The ratio between runoff and rainfall is 16.76% to 23.61%. The runoff yield is 9.75-13.76 liters/second/square kilometer. Group II contain K.22B and K.31. The ratio between runoff and rainfall is 42.66% to 61.72%. The runoff yield is 24.81-35.86 liters/second/square kilometer. And group III is K.54. The ratio between runoff and rainfall is 73.54%. The runoff yield is 41.23 liters/second/square kilometer. The difference of 3 groups is land uses that watershed area of group I is the forest with community and agriculture. As to watershed area of group II is a National park and dense forests. The group III (K.54) had difference of flow and data because this group is influenced from Vajiralongkorn Dam.

Keywords: Hydrological characteristic, Watershed, Khwae Noi watershed





Carbon Capture and Storage (CCS) by Mineral carbonation using Epsom salt from Samut Songkhram Province

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The purpose of this experimental research is to study Carbon Capture and Storage (CCS) and to remediate the severe carbon dioxide (CO_2) release into the atmosphere. In this research, Carbon Capture and Storage (CCS) by Mineral carbonation is selected. Carbon dioxide (CO_2) in the form of bicarbonate (HCO_3^-) will be reacted with local geological material that contains metal ion +2 in this case is Mg^{2+} in epsom salt. This method is expect to change carbon dioxide (CO_2) which causes global warming to be carbonate mineral phases for instance magnesite and dolomite that are stable in earth atmosphere and do not affect environment. This method can reduce carbon dioxide (CO_2) that release from CO_2 emission sources i.e. fuel power station, steelworks, cement factory etc. The advantage is an invention of technology for Thailand in order to be ready for law of carbon tax in the future. The material used in this study is imitate geological processes from Thai wisdom is Epsom salt from salt evaporation pond. The substrate is not only inexpensive but also find easily in Thailand. Epsom salt were analysed by X-ray diffractometer (XRD) and the result showses that the sample is composed of Halite (NaCl), Hexahydrate ($\text{Mg}(\text{SO}_4) (\text{H}_2\text{O})_6$). However, the result of mineral carbonation reaction is process.

Keywords: Carbon Capture and Storage (CCS), Mineral carbonation, Epsom salt





Thermoluminescence dating of burial jars from Ban Muang Bua Archaeological site, Roi Province.

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Thermoluminescence (TL) dating is an important technique for determining absolute age in archeology. The 10 samples, burial jars, were excavated in 2016 from Ban Muang Bua archaeological site, Roi Province. This research methods included to petrography and TL dating. THE 10th REGIONAL OFFICE OF FINE ARTS DEPARTMENT, ROEI reported the soil profile of Pit 1 was divided into 7 layers and the soil profile of Pit 2 was divided into 6 layers. The study collected 3 samples in Pit 1 from layer 2 (sandy clay loam) and layer 4 (loam) and 7 samples in Pit 2 from layer 4 (sandy clay loam), layer 5 (clay loam) and layer 6 (clay loam). The petrography interpretation of burial jars showed the different mineral of quartz, opaque and clay minerals contains in each sample. TL age results of Pit 1 were 2100-2960 year-old and the TL age results of Pit 2 were 1720-1770 year-old and 2400-2650 year-old. The TL age results were separate to 2 periods of Ban Muang Bua Archaeological site as 2650-2100 years ago (pre-historical period) and 1770-1720 years ago (historical period).

Keywords: Thermoluminescence dating, Ban Muang Bua archaeological site





Contamination of Heavy Metals in Supply Water near Khon Kaen Municipal Landfill, Khon Kaen Province, Thailand.

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Heavy metals contamination in supply water is a municipal problem in local communication. Khon Kaen municipal landfill is large landfill in Khon Kaen. Hence, heavy metals from landfill may be leaked to supply water of the community near landfill. This present study aimed to study the contamination and the distribution of heavy metals in surface and groundwater in community near Khon Kaen municipal landfill, Khon Kaen Province and to compare the concentrations of heavy metals with the water quality standard from Notification of the National Environmental. Field study and sampling were conducted in September (wet season) and December (dry season) 2016. Groundwater samples were collected from 3 groundwater supply, and 4 surface water were collected in is areas as well. The experiment groundwater and surface water was divided into 2 methods: acid digestion, taked 100 ml of the water samples added HNO_3 , HCl and digested by hot plate $90-95^\circ\text{C}$ from method 3005a, and filtrated through filter paper No.1 and No.42 respectively. 7 heavy metals (As, Cd, Cr, Mn, Ni, Pb and Zn) was analyzed by Inductively Coupled Plasma-Optical Emission Spectroscopy (ICP-OES). The results were illustrated as followed: concentration of Pb and Cr were exceeded than surface water quality standard at 0.05 mg/l by Pb, 0.05 mg/l by Cr. Pb in digestive surface sample No.1 (9.04 mg/l), surface sample No.2 (8.52 mg/l), surface sample No.3 (1.03 mg/l), surface sample No.4 (3.03 mg/l) in September 2016, and Cr in sampl eNo.2 (0.16 mg/l), at 0.05 mg/l and surface sample No.3 (0.08 mg/l) in December 2016. For groundwater samples, In all samples, Cd concentration in September was higher than the groundwater quality standard at 0.003 mg/l, and Pb in all digestive Groundwater samples are higher than groundwater quality standard at 0.01 mg/l in September 2016. Moreover, Mn exceed the groundwater quality standard at 0.5 mg/l in filtrate water sample and digestive water sample in groundwater sample No.2 (2.38 mg/l, 2.14 mg/l respectively) in September 2016. To control heavy metal contamination of Ban municipal landfill should be improved, and heavy metal contamination in environment should be monitored.

Keywords: contamination, heavy metals, supply water





Application of Deep 2D Resistivity Imaging to Study Pu-Kham Hot Spring Occurrence, Wichainburi Phetchabun

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The hot spring is found as an important resources, which is generally for tourist attraction and community water supply. Pu-Kham hot spring zone (observed temperature of 40°C) located in Saraburi-Lomsak road, Wichainburi district, Phetchabun province has found an interesting newly current tourist attraction with convenient accessibility. Pu-Kham hot spring occurrence can be performed subsurface geology feature by application of deep 2D resistivity imaging. Three survey lines with length of 840 m, 1320 m and 960 m and with survey lines spacing of approximately 500 m lay in the East-West direction crossing geology lineament in the North-South direction. Multi-electrodes resistivity measurement with a resolution of 10 m was carried out with the Geomative instrument (GD-10) with Schlumberger array for target deep more than 120 m. Interpretation of 2D resistivity inversion models clearly indicate contact zone between very thick (> 100 m) sediment layers presented as very low resistivity ($< 10 \Omega \cdot \text{m}$) found in the Eastern part of section and slightly high resistivity ($> 60 \Omega \cdot \text{m}$) found in the Western part of section which presented as shallow sedimentary bedrock. The location high potential of Pu-Kham hot spring is positioned at this contact zone. It is noted that the heat source of Pu-Kham hot spring must be from the deep volcanic rock underneath sedimentary bedrock. It is suggestion that the development of Pu-Kham hot spring is possible to setup at the contact zone.

Keywords: Deep 2D Resistivity, Hot Spring Occurrence, Pu-Kham, Subsurface Geology





Forecasting of the Exporting Quantity of Ribbed Smoked Sheets

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The purpose of the research is to study and forecast the exporting quantity of the smoked sheet during January 2551 to December 2557 .The data consist of 84 monthly of the exporting quantity of the smoked sheet. The three forecasting methods are studied: Holt-Winter, Decomposition and Box-Jenkins method which all of three methods are for the data that have trend and seasonal, In comparison, Mean Absolute Percent Error (MAPE) are considered which the lower value is the better method.

In the research, it is found that Holt-Winter is the appropriate forecasting method which MAPE is 12.1950 %. The forecasting equation: when t = quantity of year, n = quantity of year, L = level, s = quarter and = seasonal.

Keywords: Holt-Winter method, Decomposition method, Box and Jenkins method, Ribbed smoked sheet the 3th





Comparison of Data Transformation Methods for Normal distribution

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The objective of this research was to compare three data transformation methods: the dual power transformation, the exponential transformation, and the Modified Box-Cox transformation. The criterion used for the study was the percentage of acceptances the null hypothesis to the data having a normal distribution, after the three data transformation methods were applied with Chi-square and t distributed data. For the studied data, Chi-square and t distributed dataset was simulated using R version 3.3.1 with three levels of sample size (n): small (10, 20), medium (30, 40), large (50, 100). Each situation was repeated 1000 times and the significance level was set at 0.05.

The result of this study was summarized into two parts. The first part was generated chi-square distributed data: it is found that when the data has small sizes, Dual power transformation method and Modified Box-cox method are the best method. For medium sizes and large sizes, Modified Box-cox method were the best method. The second part was generated t-distributed data: for the data with small sizes, Modified Box-cox method was the best method. The Modified Box-cox method and Exponential method were the best method for data with medium and large sample sizes.

Keywords: Transformation, Dual power transformation, Exponential transformation, Modified Box-Cox transformation.





Comparison of Efficiency of Screening Factor between Fractional Factorial Design and Plackett Burman Design

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This research aims to compare the efficiency of screening factor between fractional factorial design and plackett Burman design. These designs were applied with the trials data which has 11 factors and one response from experiment of the new chemical product. We used regression analysis to find the appropriate model. The situation was divided into with two cases. In the first case, the distance of each factor was set up from low level to middle level of their range. In the second case, the distance of each factor was set up from middle level of their range to their high level of the first case. In each case, the errors for the response are three types. The error was normal distribution with zero mean and various variances that are 1, 0.1 and 0.01, respectively. Fractional factorial design and Plackett Burman design were screening for important factor. The important factor consisted only 5 factors (D, E, F, L, H). Normal plot of the effect and Pareto chart of the effect are considered to indicate important factors. This research shows percent of accuracy by fractional factorial design and plackett Burman design. Obviously, in each situation, fractional factorial design has high performance with the largest percent accuracy.

Keywords: Fractional factorial, Plackett Burman, Screening factor





Robustness Comparison of Correlation Coefficient for Data Containing Outliers

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The objective of this research is to compare the robustness of four correlation coefficients—Pearson, Spearman, Kendall Tau and Blest correlation coefficients—for data containing outliers. The criteria for this comparison are the absolute bias and mean square error. The simulations data in the form of standard bivariate normal distribution are generated with six levels of the correlation between two variables are set at 0, 0.2, 0.4, 0.6, 0.8 and 1.0. In addition, the sample sizes (n) in this study are determined at 10, 20, 50, 100 and 200, and the percentages of mild outliers are set at 0%, 10%, 20% and 30% of the sample size. The totals of 120 situations are studied. The experiment is repeated 1,000 times for each situation. The conclusions of this research are as follows: In the case of no-outliers in Y variable, the robust correlation coefficient is Pearson correlation coefficient. When Y variable is contaminated with outliers and the two variables are not correlated ($\rho = 0$) or high correlated ($\rho = 1$), the robust correlation coefficient for all sample sizes is Kendall tau correlation coefficient. However, when correlation of two variables are between 0.2 and 0.8 and outliers in the Y variable for all the sample sizes, Spearman or Blest correlation coefficients tend to be the most robust correlation coefficients, they tend to give the efficiencies which are not different from each other.

Keywords: Correlation coefficient, outliers, robustness, absolute bias,
mean square error





A Comparison of Confidence Interval Estimation for Binomial Proportions

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The confidence interval estimation for the proportion of binomial distribution is investigated based on three methods in this study, i.e., Wilson's, exact and Bayesian methods. In this research, we consider some circumstances, such as, sample sizes (n) in the study are 5, 10, 50, and 100, the population proportions (p) are 0.08, 0.1, 0.2, 0.3, 0.4 and 0.5 based on the level of confidences 95% and 99%. The Monte Carlo simulation technique is employed for efficiency comparison of the estimation methods. Each situation is run with 1,000 repetitions. The finding results based on three estimation methods in this study are summarized.

Keywords: Binomial proportions, Confidence interval estimation, Wilson's method, Exact method, Bayesian method





Comparison of Forecasting Techniques for the quantity of Sugar Export in Thailand

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The objective of this research is to find the suitable model for forecasting the quantity of sugar export in Thailand. Three methods are considered which are the Decomposition, Holt-Winter and Box-Jenkins methods. Then, the efficiency is compared by the Mean Square Error and the Mean Absolute Percentage Error. The data is separated into two parts. The first part consists of fifty-eight data from January 2011 to October 2015 for determining the models. There are twelve data of the second part from November 2015 to October 2016 for forecasting the quantity of sugar export in Thailand.

The results show that the Box-Jenkins is the appropriate method because of the lowest in Mean Square Error and Mean Absolute Percentage Error. The suitable model is $ARIMA(0, 0, 1)(1, 1, 0)_{12}$ without constants.

Keywords: Sugar export, Decomposition Method, Holt-Winter Method, Box-Jenkins Method, Mean Absolute Percentage Error, Mean Square Error





A Comparison between Logistic and Probit Regressions for Factors Affecting the Further Study in Master's Degree of Students in Faculty of Science, Kasetsart University

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The aim of the study was to investigate the performance of logistic and probit regressions to determine factors affecting the further study in master's degree of students in faculty of Science, Kasetsart University. The suitable of the regression model and percent of correct were considered. Logit model was more appropriated than probit model. The percent of correct in logit model was 65.0%. The factors affecting the further study in master's degree of students were someone in family graduated in master's degree, need to get more knowledge and experience and obtain the scholarships.

Keywords: logistic, probit, regression analysis





A Comparison of Classification Methods of Learning Achievement on the Principles of Statistics Course of Kasetsart University Students

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The purpose of this research is to predict the learning achievement of the principles of statistics course of Kasetsart University students. It found that important independent variables in determination of student success in prediction of learning achievement are sex, faculty, year and GPA. Student are determined by grouping into two group : 1) success group, student who got grade A, B+, B, C+, C, D+ and D 2) failure group, student who got grade W and F. Modeling is applied Discriminant Analysis and Logistic Regression Analysis.

The result of this research shows that analysis of logistic regression method has the most percent accuracy of 76.2 and the discriminant analysis method has the percent accuracy of 69.0

Keywords: Discriminant Analysis, Logistic Regression





Sensitivity Comparison of Quality Control Charts for Process Mean Shift Detection when the Data are Normally Distributed

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The objective of this research is to compare the sensitivity for process mean shift detection of three control charts, namely exponentially weighted moving average control chart (EWMA), exponentially weighted moving average control chart with fast initial response (FIR- EWMA) and \bar{x} control chart when the data are normally distributed. The studied factors consist of a sample size (n) for each subgroup is equal to 3,5,10,15,20 and 50, the shift from process mean ($\delta\sigma$) is set at $0.2\sigma, 0.4\sigma, 0.6\sigma, 0.8\sigma, 1\sigma, 2\sigma, 5\sigma$. A data simulation is conducted 10,000 times for each situation by Monte Carlo technique to compare the efficiency in term of the out-of-control average run length (ARL_1). The results of this research show that the efficiencies for process mean shift detection of FIR - EWMA and EWMA control charts tend to be better than that of \bar{x} control chart for the following situations: ($\delta \leq 1$ and $n = 3, 5, 10$) or ($\delta \leq 0.6$ and $n = 15, 20$) or ($\delta \leq 0.4$ and $n = 50$). For such other situations, the efficiencies of all three control charts tend to have no difference. That is, all three control charts tend to give the efficiencies that are not different from each other when the shift from process mean increases.

Keywords: Exponentially weighted moving average control chart, average control chart, process mean shift, average run length





Willingness to pay for studying in the special program of Statistics at Kasetsart University under the hypothetical situation of increasing tuition fee and no student loan supports

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The aim of this study was to estimate the highest value of willingness to pay (WTP) and find the factors affecting the WTP for studying in the special program of statistics at Kasetsart University (KU), under the hypothetical situation of increasing tuition fee and no student loan support. The factor of interest consisted of gender, year of study, parent's income and expenditure, and number of dependency children. A random sample of 82 students from freshmen and sophomores were requested to evaluate the highest value of WTP from the double closed - end questions and also stated the real value of WTP from the opened - end questions. Data analysis were performed using descriptive statistics and applying Cameron's regression models under log - normal and weibull distributions.

The results showed that the average real value of WTP is about 33,808.54 baht (95% CI: 31,935.18 - 35,681.90). The log - normal model fit well with this data in estimating the average highest values of WTP as 41,349.09 baht (95% CI: 40,623.32 - 42,074.86) at a significant level of 0.05. Moreover, the factor that was statistically significant final in the model was the year of study (p-value ≤ 0.0001).

Keywords: Log - normal and Weibull distributions





Performance Comparison of Tukey's control charts under Gamma and t distributions

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The propose of this study is to evaluate the performance of Tukey's control chart under Gamma and t distributions for detecting the process mean shift with the out-of-control Average Run Length (ARL_1) when the process mean changes to small, medium and large shift. The ARL_1 should be very small to get the out-of-control signal. The results of this study show that Tukey's control chart is the best performance to monitor the large process mean shift for Gamma and t distributions. For Gamma distribution, a sample size of 10 shows better performance than a sample size of 30. On the other hand, a sample size of 30 shows better performance than a sample size of 10 for t distribution.

Keywords: Tukey's Control Chart, Average Run Length, Mean Shift, Gamma Distribution, t Distribution





A Comparison of Efficiency of Ratio Estimators for Population Mean in Simple Random Sampling

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The purpose of this statistical project is to compare the efficiency among three ratio estimators for population mean in simple random sampling (SRS). The three ratio estimators are proposed by Cochran (1942), Muneer Et. Al. (2015), and Brar and Kaur (2016), respectively, which these estimators were not referenced and compared to each other before. Population of size 1,000 ($N=1000$) consisting of one studying variable (Y) and two assistant variables (X and Z) are simulated. Variables Y , X and Z are imposed to have normal distributions with means 10, 20 and 30, respectively, with equal variances of one. The correlation coefficient (ρ) between two variables of Y , X and Z are 0.95, 0.75, 0.50, 0.25, -0.95, -0.75, -0.50 and -0.25. Samples of size $0.10N$ are randomly drawn from the population by SRS 1000 times, repeatedly.

Based on considering the mean square error (MSE) the ratio estimator proposed by Cochran (1942) has the best efficiency in all cases considered. The second estimator comes from Muneer Et. Al. (2015). The poorest estimator is the ratio estimator given by Brar and Kaur (2016). Two ratio estimators of Cochran (1942) and Muneer Et. Al. (2015) have very similar efficiency. In addition, these three estimators have a similar performance in the case of the correlation coefficient between studying variable Y and assistant variable X has a medium size. (in this paper $\rho_{yx} = 0.50$)

Keywords: simple random sampling, ratio estimator, mean square error





Comparison of Power of Test for Normal, Lognormal and Exponential Distribution

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This research's objective is to compare an ability of controlling type I error and power test of the test statistic that use in testing Normal Distribution, Lognormal Distribution and Exponential Distribution. Which test statistic that uses in this research are Kolmogorov -Smirnov (KS), Anderson – Darling (AD) and Kuiper (V). In this research the preferences of population distribution use 2 parameters Normal Distribution, 2 parameters Lognormal Distribution and 1 parameter Exponential Distribution. With an amount of sample of 10 and 50 test for 1000 times repeatedly test in each situation. By considering the ability of controlled probability type I error, then comparing the efficiency of the tests with power of test from 3 different statistic tests under significant level (α) 0.05 and 0.10 . Which the test results summarized as follow.

The ability consideration of controlled probability type I error founded that Kolmogorov – Smirnov (KS) test has the best ability of controlled probability for type I error. The result of comparing power test shown that the test power of Anderson – Darling (AD) test has the highest distribution value. But for the Normal Distribution and the Lognormal Distribution, Kuiper (V) statistic and Anderson – Darling (AD) statistic test value are approximately similar. For Exponential distribution, Kolmogorov -Smirnov (KS) statistic test is approximately similar to Anderson – Darling (AD) statistic test on test power. And Kolmogorov -Smirnov (KS), Anderson – Darling (AD) and Kuiper (V) statistic test 's power of test will rise when sample increase at every significant level and every distributions.

Keywords: Type I error, Power of test, Normal Distribution, Lognormal Distribution, Exponential Distribution, Kolmogorov -Smirnov (KS), Anderson – Darling (AD) and Kuiper (V)





Study of Claims and Number of Claims Distributions for Motor Insurance in Thailand

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The purpose of this project studied probability distribution of claims and the number of claims for type 1 of motor insurance in Thailand. The data used in the project are between 2011 to 2015. The interested probability distributions for the claims are Weibull, Log-normal, Pareto and Gamma distribution. In addition, the interested probability distribution for the number of claims are Poisson distribution and Negative Binomial distribution. The probability distribution tests are Kolmogorov-Smirnov test, Anderson-Darling test and Chi-Square test at 0.05 significant levels. Then, we compare the efficiency of the probability distribution by using mean square error (MSE) and the results shown that Gamma distribution is appropriate for probability distribution for claims which gives estimated shape parameter ($\hat{\alpha}$) is 485.08046 and estimated scale parameter ($\hat{\beta}$) is 0.03782. Moreover, Poisson distribution is appropriate to probability distribution for the number claims which gives estimated parameter ($\hat{\lambda}$) is 1.2.

Keywords: Probability distribution, Kolmogorov-Smirnov test, Anderson-Darling test, Chi-Square test, Mean square error





Forecasting the Quantity of Durian and Longan Exports

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The purpose of this study is to compare three forecasting methods which are Holt-Winter method, Decomposition method and Box-Jenkins method for optimal and forecasting the quantity of durian and longan. The appropriate forecasting method is considered by Mean Square Error (MSE). The method which has the smaller value of MSE indicates the best method to use forecast. The results show that the Decomposition method is the most appropriate method for forecasting the quantity of durian. On the other hand, to forecast the quantity of longan is to use Box-Jenkins method which give the lowest MSE.

Keywords: Quantity, Mean Square Error, Holt-winter method, Decomposition method, Box-jenkins





Comparison of Time Series Forecasting Method of the Stock Price: A Case Study of Stocks in the Sector of Construction Services

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The purpose of this study is to compare three forecasting methods on the stock price of construction services which are Double Moving Average, Double Exponential Smoothing, and Box-Jenkins. Firstly, the fundamental analysis is used to select two interesting stocks, which are BJCHI and STPI. Then, use technical analysis to guide an investment in the stock market. To find the suitable forecasting model, the MSE and MAPE are the criteria for the comparison. The data of BJCHI and STPI are divided into two parts. The first set is used for creating the model and the second set is used for forecasting. The first set of BJCHI is from November 29, 2013 (started entering the stock market) - October 14, 2016 and the second set is from October 17 - 31, 2016. The first set of STPI is from November 4, 2013 - October 14, 2016 and the second set is from October 17 - 31, 2016.

The results of the study are as follow: the Box-Jenkins is the suitable method used in forecasting. The models are $ARIMA(1,1,1)$ without constant term for BJCHI and $ARIMA(2,1,2)$ without constant term for STPI because they provide the lowest MSE and MAPE among the Double Moving Average method and Double Exponential Smoothing method.

Keywords: Stock price, Double Moving Average, Double Exponential Smoothing, Box-Jenkins





Performance Comparison of Forecasting Methods for the Appropriate Parameters with Exponential Smoothing Technique for Various Time Series Data.

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In this research aim to study time series forecasting methods for finding the appropriate model. The time series data applied in this research were divided into 3 patterns each pattern consist of 10 series such as stationary, trend and trend and seasonal. Forecasting methods are used in this research consist of exponential smoothing fixed parameters and used optimization for finding appropriate parameters include of single exponential smoothing, double exponential smoothing and Holt-Winter. The other methods are moving average, double moving average, decomposition and Box-Jenkins. Two criteria are used to compare the efficiency of each method are mean squared error (MSE) and mean absolute percentage error (MAPE).

In this study, we found that Box-Jenkins method is higher performance than other methods for stationary pattern. For the pattern series with trend, trend and seasonal we found that exponential smoothing by used optimization for find appropriate parameters is higher performance than other methods.

Keywords: exponential smoothing, moving average, decomposition, Box-Jenkins and optimization





A Study Fundamental and Technical Analysis for Stocks Price in The Stock Exchange of Thailand: A Case Study of Property Development Sector

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This research is to study the property development sector of SETHD: five stocks of the property development of the Stock Exchange of Thailand during October 2014 – 2016 by the fundamental analysis of quantities are the total assets, the total liabilities, the profit to asset ratio, the return on equity, the net profit margin, the price-earning ratio, profit to asset ratio, the book value per share and the dividend yield ratio by comparing the annual database for 5 years. The result is to analyze the companies which have the good factors and being in trend of high profits found that Sansiri Public Co.,Ltd is the most proper stock. In addition by the technical analysis comparison: the weighted moving average and the relative strength index, shown that in the past this stock was the closing price and downstream on the database tracing back since 13 July 2016- 27 December 2016. The price was 1.95 bath on the way down until to 1.64 bath which accounted for 17.27 percentage then the price was upstream from 1.64 bath up to 2.04 bath which accounted for 27.74 percentage on 7 March 2017. In the future, this stock should be on the way up in such short a period.

Keywords: Fundamental analysis, Technical analysis, Risk, The property development stock





Comparisons of Type I Error Rates and Power of the Tests in Post Hoc Comparison Procedures for Completely Randomized Design

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The purpose of this study was to compare the type I error rates and power of the test in completely randomized design for four Post Hoc comparison procedures. The four Post Hoc comparison procedures include Least Significant Difference (LSD), Duncan, Waller-Duncan and Games-Howell with $\alpha = 0.05$. The population had equal variances and normally distributed. Data were simulated by Monte Carlo technique with 500 times. The number of treatments (k) were 3 and 5 with small, medium and large sample sizes which they were equal in each treatment.

The result showed that LSD, Duncan and Waller-Duncan methods can control the Type I error rate however Games-Howell method cannot control the Type I error rate for all sample sizes. When considering the power of the test, when $k = 3$, the LSD method provided the highest power of the test when $k = 5$, Duncan methods gave the highest power of the test for all sample sizes. Furthermore, the result show that the power of the test increase according to the number of treatments and sample sizes.

Keywords: Type I error rate, Power of the test, Post Hoc comparison, completely randomized design





Inverse Sampling and Its Efficiency

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This research studies inverse sampling: sampling scheme and estimation. To investigate its efficiency, inverse sampling is compared to simple random sampling without replacement by simulation study, which consisting of two parts. For the first and second part, both sampling designs are applied to population data generated by using R program and real data, respectively. The estimates of the population proportion and estimated variance are calculated under two sampling methods. Relative efficiency is considered in comparison.

The simulation result shows that simple random sampling using an appropriate sample size is more efficient than inverse sampling when the number of interested unit in the sample is set to be small value under inverse sampling. However, if the researchers set the number of interested unit in the sample close to the total number of interested in the population, inverse sampling is more efficient than simple random sampling.

Keywords: simple random sampling, inverse sampling, relative efficiency





An Appropriate Sampling Design for a Survey of Smart Phone Usage amongst Students in the Department of Statistics, KU

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The purpose of this research is to study an appropriate sampling design for survey of smartphone usage amongst students in the Department of Statistics, KU. The efficiency of simple random sampling, systematic sampling, stratified sampling and cluster sampling are compared by considering the estimated variance of the estimators of parameters.

The result of this research shows that systematic sampling is more efficient than simple random sampling, stratified sampling, and cluster sampling. The survey result is following. The proportion of sampled male students is 0.32 and of sampled female students is 0.67. Considering smart phone brand, the proportion of students using iPhone, Samsung, Sony, Oppo, and other brands is 0.63, 0.22, 0.02, 0.02, and 0.08, respectively. About telephone networks, the proportion of students using AIS, TRUE, DTAC, and other telephone networks is 0.29, 0.35, 0.34, and 0.01, respectively. The average number of minutes per day that a student uses smart phone is 378.53 minutes. The average number of minutes per day that a student plays games on smart phone is 76.37 minutes. The average number of minutes per day that a student plays internet is 270.72 minutes. The average number of minutes per day that a student makes phone calls is 31.44 minutes. The average service charge per month is 539.94 baht. Total service charge for all students per month is 247,296 baht. The proportion of students using the smart phone while studying is 0.72. The proportion of students who think that using smart phone for a long time has a negative effect on studying is 0.49.

Keywords: Sample random sampling Systematic sampling Stratified sampling
Cluster sampling





Production of mono-specific polyclonal antibodies against *Brucella abortus* and *Brucella melitensis*

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Brucellosis, a bacterial disease caused by members of *Brucella* spp., is an important zoonosis and a significant cause of reproductive losses in animals. Ten species of *Brucella* spp. such as *Brucella abortus* (*B. abortus*), *B. melitensis* and *B. suis* are identified according to the types of infected hosts. Moreover, within these species, sub-species classifications as biovars have been indicated following of bacterial cell surface antigenicity and biochemical characteristics. For example, *B. abortus* and *B. melitensis* have been divided into 8 biovars and 3 biovars, respectively. Different biovars of each *Brucella* spp. can cause of dissimilar infection capability, virulence, pathogenicity and host ranges. Therefore, biovars classification of *Brucella* spp. species is essential for developing of effective disease control and prevention programs. Mono-specific polyclonal antibodies (MPAs) against to M and A antigens of *Brucella* spp. were useful tool for bacterial serotyping that it is one of important parts for bacterial species biovars classification. The aim of this study is to produce MPAs against *B. abortus* and *B. melitensis* containing confluent A and M antigens respectively. The developed MPAs will be used for *Brucella* spp. sub-species typing in National Institute Animals Health (NIAH) of Thailand that they can compensate the imported commercial MPAs costly. To obtain the MPAs against *B. abortus* and *B. melitensis*, two rabbits were immunized with *B. abortus* B99 (biovar 1) and *B. melitensis* 16M (biovar 1) for 4 and 2 times, respectively. The antibody titers were measured by ELISA that it revealed the immune response potency. Both injected bacteria showed high immunogenicity. Undesired antibody remained in serum was removed by heterogeneous absorption. Compared between pre- and post-heterogeneous absorption, measured antibody titers obtained after immunization of *B. abortus* and *B. melitensis* showed decreasing from 1:40 to 1:10 and 1:25,600 to 1:200, respectively. The results indicated that heterogeneous absorption could remove most of non-specific antibody from anti-serum. Correlated to commercial MPAs, each developed MPA showed high specificity with homogeneous *Brucella* spp. for slide-agglutination serotyping. Bacterial number and diluted serum reacting properly for agglutination test were 3×10^4 cells and 1:4 respectively. In conclusion, developed in-house MPAs against *B. abortus* and *B. melitensis* can use for *Brucella* spp. species serotyping for supporting of *Brucella* spp. species biotyping.

Keywords: mono-specific polyclonal antibody, *Brucella abortus*, *Brucella melitensis*





Bacterial isolation from mushrooms and their ability to increase mushrooms yield

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Pleurotus mushrooms are popular as a commercial mushrooms. In this study, the methods to increase mushroom yield by bacteria were studied. Bacteria which lives on all part of mushrooms such as fruiting body and stalk were isolated. The bacteria were isolated by spread plate method and the ability of bacteria to increase mushrooms yield was measured. The isolated bacteria were screened by co-cultivated with three strains of Pleurotus mushrooms; *Pleurotus ostreatus*, *Pleurotus pulmonarias* and *Pleurotus flabellate* using dual culture technique. Five bacterial strains were chosen for the next step. The mushrooms were cultivated on para rubber substrate and 2 treatments were performed. First, the suspension of five bacterial strains was added to the substrate simultaneously with mushroom cultures and second, after the mushroom mycelia was fully grown on the substrate. The mushrooms yield was measured and compared with control. As the result, 61 bacterial strains were isolated and from the dual culture technique, five strains were chosen, isolate 4, 32, 42, 69 and 72, because these five isolates were the best bacteria that could activated mycelium growth. The result showed that the first treatment provided higher mushrooms yield than the second treatment. *Pleurotus ostreatus* when co-cultivated with the bacteria isolate 42 produced the highest mushroom yield (32.85%). *Pleurotus pulmonarius* when co-cultivated with the bacteria isolate 72 produced the highest mushroom yield (23.76%). *Pleurotus flabellatus* when co-cultivated with the bacteria isolate 4 produced the highest mushroom yield (50.95%). This result indicated that the bacteria isolate 4, 42 and 72 might be able to use for increase the yield of mushrooms.

Keywords: Pleurotus mushrooms, Bacteria, Dual culture technique, Mushrooms yield





Detection of *Salmonella* Typhi by loop-mediated isothermal amplification (LAMP) technique

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Typhoid is infectious disease in gastrointestinal tract of human, easily transmitted through food and drinking water or fecal contaminated with *Salmonella* Typhi. Rapid method for detection of *Salmonella* Typhi is important for typhoid treatment and prevention. The objective of this study is development of loop-mediated isothermal amplification (LAMP) technique for detection *Salmonella* Typhi in Thailand. Target gene for LAMP test is STY2879 gene which is putative reverse transcriptase. LAMP test uses 4 primer specific 6 region of STY2879. The condition of LAMP technique was at 64 °C for 70 minutes. Reading of result was done by naked eye observation of hydroxynaphthol blue (HNB) color change from purple to blue in the positive sample and no color change in negative sample. Confirmation of the result was done by 3% agarose gel electrophoresis and followed by staining with ethidium bromide which shows ladder band of DNA. The LAMP technique for STY2879 gene can specifically detect *Salmonella* Typhi and have no cross reaction with 7 other bacteria (*Escherichia coli*, *Enterobacter aerogenes*, *Serratia marcescens*, *Pseudomonas aeruginosa*, *Proteus mirabilis*, *Bacillus cereus*, and *Klebsiella pneumonia*). Sensitivity of LAMP technique was very high could detect 0.0001 ng/μl of DNA. Therefore loop-mediated isothermal amplification (LAMP) technique is rapid, specific and sensitivity of detect *Salmonella* Typhi. The LAMP technique developed in this study may be a useful alternative for routine detection of *Salmonella* Typhi in the food industry and hospital.

Keywords: loop-mediated isothermal amplification (LAMP) assay, *Salmonella* Typhi, Typhoid fever.





The studies of relationship between aeration and biotin levels on glutamic acid production and phosphoenol pyruvate carboxylase, pyruvate carboxylase and lactate dehydrogenase gene cloning of *Corynebacterium glutamicum* strain DS50, CS176 and mutant UV-BB9

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Corynebacterium glutamicum is a microorganism in actinobacteria that is highly accumulated intracellular glutamic acid. It has been recognized that the aeration has an effect on glutamic acid production, which is a primary metabolite. In the past, it is understood that for increasing glutamic acid production a highly aeration must be applied which in turn promoting cell growth. However, recently Zhang *et.al*, 2014, found that the glutamic acid production in micro-aerobic growth gave higher glutamic acid than that of high oxygen level. In addition, biotin level will be another important factor that effect to glutamic acid production, which relates to pyruvate carboxylase and phosphoenol pyruvate carboxylase activities. Thus, this research aims to study the relationship between aeration and biotin levels on glutamic acid production of three strains of *Corynebacterium glutamicum* isolated in Thailand. The two wild type strains, namely, DS50 (fast growth in glucose medium), CS176 (an arabinose utilizing strain) and mutant derived from DS50, namely, UV-BB9 (more sensitive to lysozyme) were characterized in term of growth and glutamic acid production using basal salt medium pH 7.2 with different biotin levels and culturing at room temperature (33-35°C) and 37°C with shaking at 150 rpm (low aeration) and 300 rpm (high aeration). The growth and level of glutamic acid production were determined by measuring the optical density at 600 nm (OD₆₀₀) and paper chromatography, respectively. The results showed that *C. glutamicum* DS50 and *C. glutamicum* UV-BB9 gave better growth when cultured at room temperature with high aeration as compared to low aeration. On the other hand, *C. glutamicum* CS176 gave the same growth when cultured at high or low aeration. In addition, both *C. glutamicum* CS176 and *C. glutamicum* UV-BB9 showed better growth at 37 °C under low aeration whereas *C. glutamicum* DS50 gave the same growth. In terms of glutamate production, culturing at room temperature or 37°C revealed that all three strains produced higher level of glutamic acid at low aeration condition and 5 micrograms per liter of biotin gave the highest yield. Moreover, genes encoded phosphoenolpyruvate carboxylase, pyruvate carboxylase and lactate dehydrogenase of these three strains will be further studied.

Keywords: *Corynebacterium glutamicum*, aeration, biotin levels, glutamic acid production, gene cloning





Isolation and Physiological Characterization of Xylose-Utilizing Yeasts Isolated from Soil Around Khlong Lan Waterfall

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Bioethanol is a renewable energy source, it is expected that a source for low-cost ethanol production will be lignocellulosic biomass such as agricultural and forestry residues. The aim of this study is to isolate yeast that capable of xylose fermentation. Therefore, this study concerned in finding yeasts from soil around Khlonglan waterfall. The enrichment technique in yeast extract-peptone containing 2% D-xylose (YPX) medium supplemented with 0.4 mM sodium azide (NaN_3), a respiratory inhibitor, was employed to isolate yeasts from 11 soil samples. A total of eight yeast isolates were obtained from soil samples. The shapes of isolated yeast cells were mostly ellipsoidal. To confirm whether all yeast isolates obtained really resist to NaN_3 , we determined their growth in the YPX agar plate supplemented with various concentrations of sodium azide (0, 0.2, 0.4, 0.6 and 0.8 mM). The highest concentration of sodium azide that the isolated yeasts can grow was 0.6 mM and cell growth of all isolates was completely inhibited in the presence of 0.8 mM NaN_3 . Notably, the growth of all 8 strains was better than the control yeast strains *Scheffersomyces stipitis* and *Kluyveromyces marxianus* in the presence of NaN_3 0.2-0.6 mM. Next we tested for their ability to ferment glucose and xylose at room temperature in flasks under a shaking speed of 160 rpm. In YPD medium, cell growth of all 8 strains was found to be higher than that of the *Saccharomyces cerevisiae* SC90. In YP medium containing 4% xylose, all strains showed a lower cell growth compared to that of *S. stipitis*. In contrast, the growth of all 8 strains was found to be higher in xylose medium supplemented with 0.4 mM NaN_3 . This indicated that all 8 strains were possibly able to ferment xylose to ethanol. The investigation of ethanol production from xylose and identification of strains are underway.

Keywords: ethanol fermentation, xylose-utilizing yeast, sodium azide, respiratory inhibitor





Screening and identification of phytase-producing fungi

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Forty-one soil samples collected from agriculture area, community area, forestry area, husbandry area and other places in Thailand were screened for phytase-producing filamentous fungi. A phytase-screening medium supplemented with chloramphenicol (antibiotic) to inhibit bacteria was used and the phytase-producing filamentous fungi were selected by observing clear zone formation around the fungus colony. Ninety mold isolates producing phytase and forming clear zone around the colony were selected. These mold isolates will be compared for their ability to degrade phytate and the fungi with high phytate-degradation ability will be identified by morphological study under microscope using slide culture technique.

Keywords: phytase, phytase-producing fungi, phytate-degradation





Detection and isolation of antimicrobial-resistant *Staphylococcus aureus* on surfaces of personal smartphone

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Recently, smartphones have become one of the most popular devices used in daily life. Poor personal hygiene and irregularly cleaning of the touch screen of the mobile phones may lead to bacterial accumulation on smartphone surfaces, especially *Staphylococcus aureus* which is a common opportunistic pathogen found on skin, nose and respiratory tract. *S. aureus* can cause disease in humans and cause skin and internal infections. In addition, it has been reported that *S. aureus* resists to several kinds of antibiotics, which is a problem with the current medical treatment. This study investigated the presence of *S. aureus* on surface screen of smartphone (n=15) belonged to students of Kasetsart University, Bangkok. A total number of 68 isolates were isolated from Trypticase soy broth (TSB) containing 10% Sodium chloride and 1% Sodium pyruvate, and Baird-Parker agar. The isolates were identified to be *S. aureus* by morphological characteristics, catalase test and agglutination test. The ability of antibiotic resistance was performed by disc agar diffusion method with six antibiotics included ciprofloxacin, gentamicin, kanamycin, oxacillin and vancomycin.

Keywords: *Staphylococcus aureus*, multidrug resistance, personal smartphone





Hand hygiene evaluation of cook by 3M Clean-Trace Luminometer

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Nowadays, many human diseases are caused by eating contaminated foods or by preparation procedure that's not clean. One of the most important causes of bacterial contamination is the hygiene of the cook. Especially at the hands of the cook because it is a direct contact with the food. If the cook's hand is dirty, have a wound or have a bad cleaning method, it will cause the food to be contaminated by the microorganisms from the hands and diseases in human, so it brings to this research. The objective of this research is analyzed hand hygiene evaluation of cook in the canteen at Faculty of Science, Kasetsart University. This research analyzed amount of microorganisms and the value of RLU by Colony Count Technique and 3M Clean-Trace Luminometer respectively from palm and prong of finger of cook's hands. Collected from 21 canteens during a period of 2 months (February-March). The study analyzed by comparing the amount of microorganisms and the value of RLU from the surface at palm and prong of finger of cook's hands between cleaned by using their's normal method and cleaned by using products that have ability to clean and reduce the accumulation of bacteria or products that have contain of antiseptic as ingredients. Overall results presented in this research shown that the amount of microorganisms and the value of RLU after cleaned cook's hand by using their's normal method is more higher than the amount of microorganisms and the value of RLU after cleaned by using products that have contain of antiseptic as ingredients. When compared the value between cleaned cook's hand by using their's normal method and cleaned by using products that have contain of antiseptic as ingredients. Found that, the product that have contain of antiseptic as ingredients can clean and reduce the accumulation of bacteria more than cleaned cook's hand by using their's normal method. From this research can't find a relationship between the amount of microorganisms and the value of RLU because 3M Clean-Trace Luminometer was analyzed ATP luminescence intensity of the samples from bacteria and rest of food after cleaning. It's different from Colony Count Technique that can find the amount of microorganisms of the total bacteria. However, 3M Clean-Trace Luminometer is a tool that useful and can be used to support for decided about risk-based hygiene interventions indirectly because it's convenient, quick, easy to use and portable.

Keywords: Hygiene, Contaminate, Colony Count Technique, 3M Clean-Trace Luminometer





Screening for Plant Growth Promoting Bacteria from Nam Dok Mai Mango's Bark and Leaf in Bangkachao Plantation

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Mango is considered as one of the most important agronomical fruit of Thailand since Thailand economic still depends on agricultural products. One of the outstanding varieties of mango is called “*Nam Dok Mai Mango*” grow in Bangkachao, the plantation area located in Samutprakarn province nearby Bangkok. Nam Dok Mai mango possesses remarkable sweet flavour and fragrant. One hundred and ninety-nine epiphytic bacteria obtained from Mango's tree (barks and leaves) were tested for plant growth promoting traits such as production of indole-3-acetic (IAA), siderophore and phosphate solubilisation. IAA production was conducted by cultivating bacteria in PYDM broth supplemented with 0.1% L-Tryptophan and IAA was quantitatively measured using Salkowski's reagent (Colorimetric method). Moreover, the bacteria were spot-inoculated onto Pikovskaya's agar and CAS (Chrome Azurol S) agar to observe the clear zone and halo zone appearance as a result of solubilisation of phosphate and siderophore production, respectively. Results indicated that 163 isolates (85.4%) produced IAA in the range of 0.24 to 275.97 $\mu\text{g/ml}$. The highest IAA producing bacteria was *Micrococcus luteus*. Sixty-eight isolates (34.2%) and 55 isolates (27.6%) were able to solubilize phosphate and produce siderophore, respectively. The highest phosphate solubilizing and siderophore producing bacteria were *Pantoea sp.* and *Pseudomonas stutzeri*, respectively. The bacteria with ability to produce siderophore were further investigated on their capabilities to inhibit the growth of Mango's pathogenic fungi, *Colletotrichum gloeosporioides* which cause the mango anthracnose. This finding will lead to an application of these potential bacteria by promoting the growth of mango tree and inhibiting pathogenic fungi in mango plantation and then increase the yield of Nam Dok Mai mango.

Keywords: Plant Growth Promoting Bacteria, Mango, *Colletotrichum gloeosporioides*, Antifungal





Effect of carbon sources on Degradation of PAHs by bacterial-fungal co-cultivation

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Polycyclic aromatic hydrocarbons (PAHs) are organic compounds consisting of two or more fused benzene rings, such as phenanthrene and pyrene, which are concerned pollutants contaminated in environment. Main sources of PAHs are incomplete combustion of organic compounds and contamination of petroleum. Moreover, they are highly toxic, carcinogenic to human and other organisms. Nowadays, microbial bioremediation is considered as an environment-friendly method to remove PAHs. Therefore, the aim of this research was to improve phenanthrene and pyrene biodegradation by bacterial-fungal co-cultivation and additional carbon sources in medium. Previously, bacteria and white-rot fungi with capability of phenanthrene degradation were isolated in our laboratory. We selected phenanthrene-degrading bacterial and fungal strains to co-cultivate in minimal media with various additional carbon sources. These bacterial and fungal strains showed the highest growth in phenanthrene containing minimal medium with glycerol as additional carbon source. The bacterial-fungal co-culture showed an improved PAH biodegradation than those of bacterial or fungal single cultures.

Keywords: PAHs biodegradation, Carbon sources, Co-cultivation





Detection of *Listeria monocytogenes* by loop-mediated isothermal amplification (LAMP) technique

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L. monocytogenes is an important foodborne pathogen commonly found in environments of seafood processing that causes the infection listeriosis. The manifestations of listeriosis include septicemia, meningitis, encephalitis, corneal ulcer, pneumonia and intrauterine or cervical infections in pregnant women. In addition *L. monocytogenes* can be found in the vast majority of food such as meat, eggs, poultry, seafood and dairy products. In this study detection of *L. monocytogenes* was developed by using loop-mediated isothermal amplification (LAMP) assay which is highly sensitive and specific method for amplification of the target genes. Target gene for LAMP assay is lmo0460 gene sequence encoding membrane-associated lipoprotein and hlyA gene that hly is one of the prfA virulence gene cluster and encodes a hemolysin called listeriolysin O. LAMP uses 4-6 primers specific to 6 regions of the target genes. In addition the result can be observed by hydroxynaphthol blue (HNB) color change from purple to sky blue color and no color change in negative sample. Confirmation of result was done by agarose gel electrophoresis followed by staining with ethidium bromide which showed ladder band of DNA. LAMP technique is rapid, specific, sensitive method for detection of *L. monocytogenes*, easy to perform, inexpensive and it will be a potential useful and powerful tool for detecting foodborne pathogens.

Keywords: Loop-mediated isothermal amplification, *L. monocytogenes*





Study on the Properties of Xylose-Utilizing Yeasts Isolated from Peat Swamp Forest for Agricultural Applications

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The peat swamp forest is a unique ecosystem, where waterlogged conditions prevent full decomposition of plant debris. A total of 44 xylose-utilizing yeast strains were isolated from organic soil at Sirindhorn Peat Swamp Forest Nature Research and Study Center (Pa Pru To Daeng). They were previously identified to be *Candida sanyaensis* (2 isolates), *Cyberlindnera subsufficiens* (32 isolates), *Candida pseudolambica* (1 isolate), *Blastobotrys illinoisensis* (1 isolate), *Candida tropicalis* (3 isolates) and *Pichia manshurica* (5 isolates). In this study, their ability to produce indole-3-acetic acid (IAA), the major member of the plant growth promoters in the auxin class, and extracellular enzymes including phytase, pectinase and xylanase, were investigated. A production of extracellular enzymes was tested on an agar medium. All strains were found to produce IAA in a range of 9.82 to 16.08 mg/L, with the highest IAA concentration produced by *Candida sanyaensis* TD (1)01-1. In addition, 25 and 1 yeast strains were found to produce phytase and pectinase, respectively. The highest phytase production was by *C. subsufficiens* TD (1)05-1 (enzyme activity index, EAI = 1.93). *C. subsufficiens* TD (2)09-1 is the only one yeast strain which can produce pectinase. None of the yeast strains tested in this study had the ability to produce xylanase. Our investigation on the production of extracellular enzymes revealed that all 44 yeast strains produced at least two extracellular enzymes, phytase and pectinase. Taken together, all of 44 xylose-utilizing yeasts had potentials for agricultural applications.

Keywords: extracellular enzymes, indole-3-acetic acid, xylose-utilizing yeasts





Isolation and characterization of multidrug resistant *Staphylococcus aureus* from dishwashing scrubber

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Staphylococcus aureus is one of normal flora on human skin and nasal which has chance to be an opportunistic pathogen. It has been found to resist to several kinds of antibiotics, causes nosocomial infection in hospital worldwide. This study aimed to isolate and characterize the multidrug resistant properties of *S. aureus* on dishwashing scrubbers (n=15) from four cafeterias in Kasetsart university, Bangkok. *S. aureus* strains were screened and isolated on Trypticase soy broth (TSB) supplemented with 10% Sodium chloride and 1% Sodium pyruvate and Baird-Parker agar, respectively. A total number of 48 isolates were obtained and confirmed to be *S. aureus* by morphological characteristics, catalase test and agglutination test. Antibiotic susceptibility test was examined on the following antibiotics: ciprofloxacin, gentamicin, kanamycin, oxacillin and vancomycin.

Keywords: *Staphylococcus aureus*, multidrug resistance, dishwashing scrubber





Study of heavy metal resistance of petroleum hydrocarbon degrading bacteria

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Many harmful pollutants come from heavy metals and petroleum. They widely spread in environments and affect human health both directly and indirectly. Some microorganisms are able to withstand heavy metals and can degrade petroleum. They are important to remove and reduce pollution in the environments. In this study, we tested ability of selected bacteria to degrade the hydrocarbons in diesel fuel and resistant to heavy metals. The bacterial strain was isolated from zinc mining soil from Mae Sot, Tak by selective enrichment medium supplemented with 10% v/v of diesel oil. It showed resistance to zinc (Zn), cadmium (Cd) and copper (Cu) on nutrient agar (NA). Moreover, bacterial growth in nutrient broth (NB) supplemented with different concentrations of each and mixed heavy metals (10-50 mM Zn, 2 mM Cd and 2 mM Cu) was determined. It was found that, this strain was able to grow in 20 and 10 mM Zn, and 2 mM Cd. Then, the study was performed to test the abilities of both petroleum degradation and heavy metal resistance in Bushnell-Haas (BH) broth supplemented with diesel oil. The results revealed that, this bacterial strain can grow by utilizing hydrocarbon source in the diesel oil contaminated with 10 mM Zn and 2 mM Cd. The strain was then identified by nucleotide sequencing of 16S rRNA gene and it was related to *Serratia marcescens*. The bacterial strain from this study was potential material for bioremediation of heavy metals contaminated petroleum in environments.

Keywords: heavy metals, petroleum hydrocarbon degrading bacteria, diesel fuel, zinc mine, *Serratia marcescens*





Optimization of biotreatment of pulp mill effluent by a white rot fungus *Trametes polyzona*

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Pulp and paper industries produce large quantities of toxic and intensely colored waste effluents, causing severe water pollution. Thus, it is important to treat the industrial effluents before their final discharge. The present study deals with the bioremediation of pulp mill effluent by a white rot fungus, *Trametes polyzona* isolated from Thailand. To improve the efficacy in biotreatment of pulp mill effluent by the fungus, factors affecting the biotreatment were studied. A five-level Box–Behnken factorial design was employed to find the optimal condition in biotreatment. A mathematical model was developed to show the effect of each factor and their interactions on the biotreatment of effluent. Reduction of color by the fungus was found to be influenced by various factors such as initial chemical oxygen demand (COD) in the effluent, aeration rate, interaction effect of COD and aeration rate, and interactivity between concentration of fungal inoculum and aeration rate. Interactivity between concentration of fungal inoculum and aeration rate had high significant effect on reduction of COD in the effluent. The mathematical model estimated the optimal conditions for biotreatment. These predicted conditions were verified by validation experiments. Under the optimized condition, efficacy in biotreatment of pulp mill effluent by the fungus was higher than that obtained in non-optimized conditions.

Keywords: Bioremediation, Box–Behnken Design, Pulp mill effluent, *Trametes*, White rot fungus





Gene Expression of Bid in Hepatoma cell line with Hepatitis B Virus Infection

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Hepatitis B virus (HBV) is a major cause of chronic hepatitis that can lead to cirrhosis and hepatocellular carcinoma. Bid (BH3-interacting domain death agonist) is a member of the BH3-domain-only subgroup of BCL-2 family members, which is a protein family involved in the regulation of apoptotic cell death. There are four isoforms of Bid (Bid_{EL}, Bid_L, Bid_S and Bid_{ES}) that are regulated by alternative splicing. These isoforms might involve in HBV pathogenesis. Thus, we applied reverse transcriptase polymerase chain reaction (RT-PCR) technique to investigate the mRNA expression of these Bid isoforms in two hepatoma cell lines including HepG2 and HepG2 transfected with HBV (HepG2.2.15). GAPDH gene was used as internal control and ImageJ Program was used to determine DNA band intensity of gel electrophoresis. Our study showed that mRNA expression of Bid_L and Bid_{ES(2)} were increased in HepG2.2.15 cell line as compared to HepG2 cell line with the fold change of 1.10 and 1.27, respectively, but the expression of Bid_{ES(3)} was decreased 0.92 times. Furthermore, Bid isoforms were decreased in HepG2.2.15 starved cell line when compared to HepG2 starved cell line with 0.61 (Bid_L), 0.50 (Bid_{ES(3)}) and 0.40 (Bid_{ES(2)}) times. In addition, we found that the most abundant isoform was Bid_L following with Bid_{ES(3)} and Bid_{ES(2)}, respectively. Distinct expression of these Bid isoforms may influence the function of Bid to interact with other pro-apoptotic Bcl-2 family proteins and lead to the fate of cell. Nevertheless, the function of Bid isoforms involving in HBV infection is needed to further clarify in the future.

Keywords: Bid, Hepatitis B virus (HBV)





Production of recombinant raw starch degrading enzyme by *Escherichia coli* BL21(DE3) and its application for syrup production from broken mill rice

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This work was investigated the optimal conditions for the maximum expression of intracellular recombinant raw starch degrading enzyme (LsA175) by *Escherichia coli* BL21(DE3), which harboring *lsa175* gene from *Laceyella sacchari* LP175. The optimization was investigated from the mid-logarithmic phase of recombinant *E. coli* cells at OD_{600 nm} of 2.0, grown at 37 °C in basal medium supplemented with glucose 5 g/L and 100 µg/mL ampicillin, using response surface methodology with a central composite design. The investigated optimal factors, temperature, IPTG concentration and induction time, on the expression of recombinant protein were at 22 °C, 1 mM IPTG and for 31 h, respectively, with P value < 0.05. The optimal condition gave the highest enzyme production at 4,472.17 U/g cell dry weight in 250 mL shaking flask and 4,545.93 U/g cell dry weight in a 2.0 L aerated stirrer fermenter. The obtained crude enzyme from the of reducing sugar after incubated at 50 °C for 9 h. Scanning electron microscope images showed the loss of starch granule structure after digested with the recombinant enzyme as compared with recombinant strain hydrolyzed the broken-milled rice (300 g/L) with 150 U/mL of enzyme resulting 60.96 g/L the native granule of broken-milled rice powder. These results showed the possibility for application of recombinant enzyme produced from *E. coli* BL21 (DE3) to produce the sugars syrup from a low cost of substrate at low temperature (50 °C) which can reduce the energy consuming and reduce the cost of operation in starch processing industries.

Keywords: *E. coli*; Recombinant raw starch degrading enzyme; Broken mill rice





Genetic diversity of culturable bacteria isolated from termite guts

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Termites are detritivores or detritus feeders. Termite gut contains organisms from all three domains of life: Bacteria, Eukarya and Archaea. There is a great diversity of microbes in the termite gut. Termites rely upon the microbes in their gut to digest the complex sugars, cellulose and organic fiber found in wood and plant into simpler molecules that they can use for food. Base on morphological study, 24 isolated bacteria were isolated from the guts of 2 higher-termite species including wood-feeding termite, *Pericapritermes semarangi* (16 bacterial isolates) and lichen-feeding termite, *Hospitalitermes* sp. (8 bacterial isolates). We studied the ability of the isolated bacteria to digest cellulose, xylan, pectin, amylose, protein, gelatin and lipid. This research was conducted to classify and identify the culturable bacteria from the termite guts using restriction fragment length polymorphism (RFLP) and sequence analyses of 16S rRNA gene. The bacterial isolates were extracted their genomic DNA using enzymatic method. Then, polymerase chain reaction (PCR) was used for amplification of 16S rRNA gene using *Bacteria*-universal primers (616V and 1492r). The PCR products (approximately 1.5 kb) were digested by each of restriction endonucleases (*MspI* and

AluI) at 37°C for 1 hour. The results showed that, the bacterial isolates were assigned into diverse groups on the basis of their restriction fragment (RF) patterns. Then, the representative isolates of each group were selected for identification by nucleotide sequencing of 16S rRNA gene. This research presented new knowledge of diverse population of culturable bacteria from the guts of termites, *Pericapritermes semarangi* and *Hospitalitermes* sp.

Keywords: Termite gut, 16S rRNA gene, Polymerase chain reaction (PCR), Restriction fragment length polymorphism (RFLP), Sequence analysis





Survey of *Listeria* in Sc KU Food Court

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Listeria monocytogenes(LM) is the species of pathogenic bacteria that can also be transmitted transplacentally from animals to humans by food and found widely tested for in environments and food such as soil and water. There is species characteristic which contaminated and grew on acid foods, low Aw foods in vacuum package or MAP package and has the ability to grew at temperature refrigerator. Addition when LM infection in humans lead to severe symptoms of diseases such as meningitis approach bloodstream and pregnancy thus disallow found in ready to eat foods. This study was to investigate the prevalence and contaminate LM in SC KU food court by inspect om floor that crowded place, meat packeaging and vegetable packaging of store. There are in spect by swab technique and conventional. This result were isolated and identified base on colony morphology and color colony on ALOA medium and plate count agar. There have 237 bulish colonies ,147 colonies from floors (61.60 %) and 91 colonies from packaging(38.40%) respectively.However I found LM in a few colonies but be careful and preventive measures by GMP for preventive post-contaminatio , cross-contamination and preventive dispersed system in SC KU food court

Keywords: *Listeria monocytogenes*, ALOA





Isolation and optimization of microalgae *Schizochytrium* sp. for Astaxanthin production

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Astaxanthin is a carotenoid pigment. It provides the red color of crustaceans and salmon. Therefore, commercial cultivation of aquatic animals require astaxanthin in feed for color development, growth and survival. Furthermore, astaxanthin also benefits poultry production and added in feed to increase mating rate and red color in egg yolks. For medical applications, astaxanthin has strong antioxidants properties that can inhibit development of cancer, prevention of heart disease and protect skin and eyes from sunlight. Nowadays, several studies on astaxanthin production from microalgae *Schizochytrium* sp. has been reported. *Schizochytrium* sp. is a heterotrophic unicellular alga, cells are globose with thin wall, produced high astaxanthin, easy to cultivation and growing fast. The objectives of this research were to isolate *Schizochytrium* sp. from mangrove forests which has ability to produced astaxanthin and optimization of the culture conditions for growth and astaxanthin production. Isolation was carried out on GYP agar supplement with chloramphenicol. Pure cultures were picked up and maintained on GYP agar with 3% glucose. One hundred and thirty-eight isolates were obtained for screening. The isolate SA19-37 showed ability to accumulate high astaxanthin content. Four experiments for optimization of the culture conditions were investigated. The different in salinity of the medium at 0, 10, 15, 20, 30, 40, 50 and 60 ppt showed that *Schizochytrium* sp. SA19-37 has high astaxanthin 1.34 mg/L with dry biomass 4.90 g/L at salinity 60 ppt. Study for optimal temperature at 15, 17, 20, 23, 26, 28, 30, 33, 36, 37, 40, 43 and 46°C in L-tube revealed that 15°C yielded the highest dry biomass 3.39 g/L and highest astaxanthin concentration at 0.97 mg/L. Among light intensity at 0, 1000, 2000, 3000, 4000 and 5000 lux, 5000 lux yielded the highest astaxanthin concentration at 1.51 mg/L and dry biomass at 11.56 g/L. Study on peptone as nitrogen source at concentration 0, 0.25, 0.5, 0.75 and 1.0% found that peptone 1.0% yielded the highest astaxanthin concentration at 1.41 mg/L and dry biomass 11.09 g/L.

Keywords: Astaxanthin, *Schizochytrium* sp., mangrove forests, optimization





Isolation and Optimization of Squalene Production from Microalgae *Aurantiochytrium* sp.

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Squalene is a triterpene hydrocarbon that can be synthesized in living organisms. It is the precursor for cholesterol, steroid hormones, and vitamin D synthesis. Human body contains about 13% of squalene in the form of lipid. Squalene is used in cosmetics and immunological adjuvant in vaccines for medical benefits. It was reduced to cholesterol in our blood and able to inhibit development of cancer. It was found in high content in the liver of deep sea sharks about 400-800 g/kg. Currently, the number of wild shark is decreasing at alarming rate in the nature. Growing concerns over shark hunting have motivated to search for other alternative sources such as extraction from vegetables or biosynthetic processes. Recently there were reports showing that the microalgae, *Aurantiochytrium* sp. was able to produce high concentration of squalene. The objectives of this research were to isolation and optimization for squalene production from microalgae *Aurantiochytrium* sp. Isolation was carried out on GYP agar and the colonies of *Aurantiochytrium* sp. were picked up and maintained on modified GYP agar plates containing 3% glucose. One hundred and sixty-five isolates of *Aurantiochytrium* sp. were obtained for screening and isolate SA7-17 show relatively high squalene contents. Optimization revealed that, *Aurantiochytrium* sp. SA7-17 produced high at squalene 9.60 mg/L and biomass 1.30 g/L at temperature 40°C. The optimal medium composition was 3% glucose, 2% peptone, 0.5% yeast extract and 2% monosodium glutamate yielded 22.50 mg/L of squalene with 2.80 g/L biomass. The concentration of sea water 0 ppt improved squalene content to 25.22 mg/L and biomass 4.19 g/L. Cultivation of *Aurantiochytrium* sp. SA7-17 in 2 liter Jar fermentor produced highest squalene at 25.76 mg/L and biomass 3.76 g/L after cultivation for 48 hours.

Keywords: *Aurantiochytrium* sp., Squalene, Optimization





Antifungal activity of actinomycetes to control *Alternaria* sp. and *Colletotrichum gloeosporioides*

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Nowaday, chemical fungicides are widely used to control fungal pathogens. Although, the chemical fungicides have benefit to inhibit plant pathogenic fungi but they are also dangerous to the environment and humans. Therefore, the study of using biological controls are interesting. Actinomycetes are microorganisms that can produce many kinds of metabolites such as antibiotics and enzymes. They have been reported for their ability to inhibit fungi that caused plant pathogens. Thus the purpose of this research was to investigate the capability of actinomycetes, isolated from soil samples in Bang-Kachao, Samutprakarn province, to control *Alternaria* sp. and *Colletotrichum gloeosporioides*. *Alternaria* sp. caused many diseases of plants especially in family Brassicaceae. *Colletotrichum gloeosporioides* caused anthracnose disease in mango. In this research, a total of 57 actinomycetes which were isolated from previous research by using isolation media supplemented with 1.5 % NaCl. All the isolates were separated into 5 spore-color groups and non-spore forming group according to their spore color on oatmeal (ISP3) agar. The majority of spore color was white by 42.1 % and the minority were black and non-spore forming by 1.75 %. All the isolates growing on ISP2 supplemented with 1.5% NaCl were tested for their antifungal activity to inhibit *Alternaria* sp. and *Colletotrichum gloeosporioides* by using dual culture method. The results revealed that *Alternaria* sp. was inhibited by actinomycete isolates 4BHN1 and 3BSN17 by 100 % and 85 % respectively. Actinomycetes isolates 4BHN1 and 3BSN17 also had the widest antifungal spectra of activity by 90 % inhibition of *Colletotrichum gloeosporioides*. Identification of potential actinomycete isolates 4BHN1 and 3BSN17 based on 16S rRNA gene sequences revealed that both strains were closely related to *Streptomyces violaceusniger* (100 % similarity).

Keywords: actinomycetes, antifungal, *Alternaria* sp., *Colletotrichum gloeosporioides*





Characterization of Auxin hormone produced from Halotolerant bacteria and their potential as Plant growth promotion

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Salt-tolerant bacteria play an important role in improving the quality of arid saline soils for suitable use as plant cultivation land. Especially, for those saline-tolerant bacteria possessing the property of promoting plant growth. This research aims to select salt-tolerant bacteria previously isolated from soils that can produce indole-3-acetic acid (IAA) and test their potential as plant growth promoting bacteria. From previous studies, the isolation of salinity-tolerant bacteria from soil samples in Bang-Kachao, Samut Prakan province and Kanchanaburi Research Station was carried out on Haloalkaliphile agar (HA) pH 7.6 using a spread plate technique 49 of IAA producing strains were determined for IAA in the culture HA supplemented with 5 mM tryptophan. All 49 isolates produced IAA concentrations of 10-200 micrograms per milliliter. The P2-23 isolate produced the highest IAA (200 micrograms per milliliter). 10 out of 49 showing high IAA production were selected for plant growth promoting. They belonged to 3 groups based on cell morphology and Gram-staining. 7 isolates, namely, 4-8, SP46, 3-13/3, BK78, P12-12, SP45 and SP158 were Gram positive, rod shape and spore forming bacteria. One isolate, namely, SP52 was Gram-positive, rod shape bacteria and 2 isolates, namely, P2-23 and BK 100 were Gram-positive cocci bacteria. For testing the germination ability of corn kernels, the seeds were immersed in cell suspensions (10^8 CFU/ml) or in the bacterial cultures of each isolate. Seeds were germinated at 25 °C for 7 days. The maize hardness index was compared with the control. For plant growth promoting by auxin-like hormone, the bacterial IAA were substituted with commercial IAA used in plant tissue culture. Carrots were cultured in Murashige & Skoog' medium containing 2 formulas. The first formulas were supplemented with IBA 0.1 mg/l and the second formulas were supplemented with 50% cultured, and then measured the number of roots and root lengths of carrots compared to the control plants. The results of this study demonstrate the potential of IAA-producing bacteria from soils as applied to plant growth promoting bacteria. In addition, if the bacteria could be evaluated to promote the growth of cultivated plants in saline soils or in drought-depleted soils, it will clearly demonstrated that these salinity-tolerant bacteria could be applicable in plant cultivation in areas with saline soils.

Keywords: Halotolerant bacteria, Indole-3-acetic acid (IAA), Plant growth promoting bacteria (PGPB), *Zae may*, *Daucus carota var sativar*





Study of spontaneous acidification and humidity of cooked soy bean for property Tempe microbiological

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Tempe is traditional Indonesian food product obtained from legumes by means of solid-state fermentation with *Rhizopus* sp. In Tempe production consist of soaked and cooked soybean soaked soy bean for 24 hour dehulled and cooked soybean mixed single culture inoculums and incubated in perforated plastic bag at 35 °c for 36 hour. The objective of research is to study the parameter is effected to spontaneous acidification, after soaked soy bean for 24 hour. The physical of soaked water have change such as form bubble or film on surface soaked water form mucus, it made soaked water like slime and sweet and/or sour smell. This physical made by many microorganisms in soaked water. This research is to study the correlation of spontaneous acidification and factor consist of sugar volume in cooked soy bean water (% ° brix), measure by refractometer high water level above high soy bean level and amount of microorganisms in soaked water, after fermentation for 24 hour. We founded the sugar volume in cooked soy bean water (%°brix), cooked soy bean water combined with eatable water for diluted for 1-3 %°brix are not affected to acidity. High water level above high soy bean is higher than 2 inch physical soaked water are low mucus, *Enterobacteriaceae* are decrease and increase acidity in soaked water (pH less than 5) in 35 °c. The second objective research is to study property Tempe microbiological with ph in soaked water which control moist of dehulled soy bean before mixed single culture inoculums less than 50% by Amount of total microorganism and *Enterobacteriaceae* and ammonia (NH₃) have detected in Tempe, there are indicator. We founded if Tempe is high ammonia level, Amount of *Enterobacteriaceae* have dominant in Tempe but if Tempe is low ammonia level, Amount of Total Microorganism is higher than *Enterobacteriaceae* 1 log cycle which Amount of Total Microorganism is 10⁶-10⁸ cfu/g.

Keywords: Tempe, spontaneous acidification





Yeast isolation from mushrooms and their ability to increase mushrooms yield

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Yeast is a microorganism that classified in fungi has various shape and can be found in natural, soil, water, parts of plants or even in mushrooms. The growth of yeast that lives with mushroom may affect the growth of mushroom in both positive and negative. The objective of this study is to isolate yeast from mushroom and study their ability to increase mushrooms yield. Mushroom samples were added in YM medium volume 9 ml. then dilute and culture on YM agar with 0.025% Sodium propionate and 0.02% Chloramphenicol. Yeasts that are different in morphology are purified. Yeast 4 strains from screening EY2, EY3, EY4, and EY5 were co-cultivated with three strains of commercial mushrooms; *Pleurotus ostreatus*, *Pleurotus pulmonarius* and *Pleurotus flabellatus* by Dual culture techniques. The result showed that yeast strain EY5 could promote the growth of all three types of mushrooms, but EY2, EY3, and EY4 yeast inhibited the growth of *Pleurotus flabellatus*. Yeasts were cultivated on para rubber substrate and 2 treatments were performed. First, yeast suspensions were added to the substrate simultaneously with mushroom cultures and second, yeast suspensions were added after the mushroom's mycelium was fully grown on the substrate. The mushrooms yield was measured and compared with control. The result showed that the first treatment provided higher mushrooms yield than the second treatment. *Pleurotus ostreatus* when co-cultivated with yeast strain EY2 produced the highest mushroom yield (146.182 g) when compared with control. *Pleurotus pulmonarius* when co-cultivated with yeast strain EY5 produced the highest mushroom yield (96.128 g) when compared with control. *Pleurotus flabellatus* when co-cultivated with yeast strain EY2 produced the highest mushroom yield (132.802 g) when compared with control. Therefrom the weight of mushroom yield can be used to calculate the percentage of biological efficiency. The result showed that the first treatment provided higher mushrooms biological efficiency percentage than the second treatment. *Pleurotus ostreatus* when co-cultivated with yeast strain EY3 provided the highest mushroom yield (47.083%) when compared with control. *Pleurotus pulmonarius* when co-cultivated with yeast strain EY5 provided the highest mushroom yield (27.773%) when compared with control. *Pleurotus flabellatus* when co-cultivated with yeast strain EY2 provided the highest mushroom yield (29.848%) when compared with control. This result indicated that yeast strains EY2, EY3, EY4, and EY5 might be able to use for promoting or increase the yield of mushrooms.

Keywords: Yeast, *Pleurotus ostreatus*, *Pleurotus pulmonarius* and *Pleurotus flabellatus*





Cultivation of Lactic acid bacteria using Coconut waste water and Rice extract

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Synthetic MRS Medium is commonly used for cultivation of Lactic acid bacteria (LAB) but it is quite expensive and need to be imported from other countries. In this study, Coconut waste water (CW) and Jasmine rice extract (RE) were modified as a cheap medium for LAB growth. *Pediococcus lolii* KU-E1 is chosen as a tested LAB. This strain is noted as bacteriocin producer which is sensitive to inhibit *Listeria monocytogenes* DMST 17303. The highest antilisterial activity of cell free supernatant (CFS) based on the spot-on-lawn assay was 51,200 AU/ml when grown in MRS broth incubated at 37°C for 16 h under microaerophilic condition. On the other hand, *P. lolii* is capable of growth in CW as well as RE with maximum cell number of 8 Log CFU/ml in the same condition as MRS broth (9 Log CFU/ml) but bacteriocin formation was absent. Supplementation of 1% yeast extract (YE) to CW increased bacteriocin formation to as high as 25,600 AU/ml while no activity was observed in RE +1% YE. This may be due to insufficient vitamin or growth factors remain in polished rice. Therefore, unpolished brown rice (URE) was tested for growth and bacteriocin production and 100 AU/ml antilisterial activity was observed. Mixing of CW and URE with the ratio 1: 1 increased antilisterial activity to 200 AU/ml whereas in CW only antilisterial activity was zero. Modified medium of mix CW+URE (1: 1) with 0.5% YE gave increased antilisterial activity to 25,600 AU/ml at 22h of incubation at 37°C. The results shown that modified medium of CW+URE+0.5%YE could be a good medium for LAB cultivation to replace the synthetic MRS medium.

Keywords: Coconut waste water, Lactic acid bacteria, *Listeria monocytogenes*, *Pediococcus lolii*, Rice extract





Effect of weak acids on cell growth and relationship between heat generation and glucose concentration in the thermotolerant yeast *Kluyveromyces marxianus* DMKU 3-1042

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At present, the production of ethanol from lignocellulosic biomass, which is by-products from agriculture and industry, is interesting. Lignocellulose feedstock was first pretreated and hydrolyzed into the liquid hydrolysate containing the fermentable sugars such as glucose and xylose. The pretreatment and hydrolysis processes possibly produce some toxic molecules such as formic acid and acetic acid that inhibit the growth of yeast cells during the fermentation process. In this study, the effect of weak acid on cell growth of a thermotolerant yeast *Kluyveromyces marxianus* DMKU 3-1042 was investigated. Yeast cells were cultivated in yeast extract-peptone-dextrose (YPD) agar supplemented with 0.10%, 0.30% and 0.50% (v/v) formic acid or 0.10%, 0.30% and 0.50% (v/v) acetic acid, and the initial pH was adjusted to 3.5, 4.5 and 7.0 for each acid. The results revealed that different types and concentrations of weak acids affected the growth of yeast differently. Formic acid showed more negative effect on cell growth than that of acetic acid. The concentration of formic acid greater than 0.10% (v/v) completely inhibited the yeast growth. In the other work, a relationship of glucose concentration and external heat generation was also studied. Experiments were performed in flasks containing YP broth with 2%, 8% and 16% w/v glucose under a shaking speed of 160 rpm and uncontrolled room temperature. Cell growth, ethanol concentration and temperature of culture broth were measured every 6 h until 48 h of incubation. Lag phase was observed for the first 6 h under the three different glucose concentrations. Cell growth and ethanol production increased proportionally to the increase of glucose concentration. The temperature of culture medium was 1-2°C increment, especially during the exponential phase (6-12 h), and subsequently decreased until 24 h. Thus the increase in temperature of culture broth was simply related to the growth phase and cell activities. This information is possibly useful for scale-up process

Keywords: *Kluyveromyces marxianus*, weak acid, thermotolerant yeast, heat generation





Screening for Spore-forming Bacteria From Surimi and Surimi processing of raw fish

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It has been reported that endospore-forming bacteria such as *Bacillus simplex*, *Bacillus subtilis* and *Sporosarcina aquimarina* which isolated from surimi affected to shelf life of surimi product. The purpose of this study was to determine whether these endospore-forming bacteria are originated from marine fish that used for surimi production or not. Samples were collected from 2 parts. The first part was sampling from both water and fishes of each surimi processing step, heat shocked at 80 degree Celsius 10 minutes and enumerated on Plate Count Agar (PCA) by pour plate technique without serial dilution. The second part was sampling from various raw surimi products by the following method. One hundred grams of surimi was mixed with 100 ml of sterile water, homogenized with Stomacher, centrifuged at 10,000 rpm and then pellet was suspended in 5 ml sterile water, heat shocked and enumerated on PCA as above. Different colonies from each part were reisolated and classified by morphological study. Fifty-four isolates were obtained from the production process, they are belonged to group IA (1 isolate), IB (32 isolates), II (11 isolates), and III (10 isolates). Twenty-six isolates, from surimi products, belonged to IA (17 isolates), IB (7 isolates), and II (2 isolates). Large number of endospore-forming bacteria, belonged to group IA, were collected from surimi samples are expected to be *Bacillus cereus* related-group. These isolated bacteria were tested for their biochemical characters that were emphasized on Proteolysis property. Samples from both parts found bacteria which suspected to be *Bacillus* sp. and/or *Sporosarcina* sp.

Keywords: Surimi, Endospore-forming bacteria





Screening of Phytase Producing Yeasts Isolated from Peats in a Peat Swamp Forest

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Phytase is an important enzyme in the feed industry. It has efficacy for hydrolysis of phytate to inorganic orthophosphate, free *myo*-inositol or *myo*-inositol mono-, di-, tri-, tetra- and pentaphosphate. A major of phosphorus source in crops is phytate, which are used as feed. All animals can not produce phytase enzyme. However, in ruminants such as cow, microorganisms in the rumen produce phytase, so phytate can be utilized as phosphorus source for them. But it does not occur in monogastric animals. Therefore, to make phosphorus source available in feed form phytate, phytase is needs. Phytase is produced by various microorganisms including yeasts. We aimed to screen phytase producing yeasts from 111 yeast stains in 41 species, which were isolated form peat in a swamp forest in Thailand. In the first step, growth on agar medium containing phytic acid as a sole source of phosphorus at 25 °C for 5 days was investigated. The result showed that 38 stains grew well. The stains showing good growth was tested for their ability of phytase production on agar medium containing sodium phytate as a sole source of phosphate by cultivation at 25 °C for 5 days. Yeast colonies with clear zone indicated phytase production. Seven yeast stains, namely stain DMKU PS 4-1, DMKU Y28-C, DMKU YNB28-2, DMKU YNB32-2, DMKU YMB 35-1, DMKU YNB38-3 and DMKU38-B showed high phytase production. Phytase production in liquid medium of the seven stains was determined by cultivation in a 100 ml of medium containing sodium phytate, in a 500 ml Erlenmeyer flask, on a rotary shaker at 150 rpm and 25 °C for 3 days. The enzyme activity was determined by estimation of inorganic phosphate released from sodium phytate. Three strains namely *Cyberlindnera saturnus* DMKU PS 4-1, *Saitozyma podzolica* DMKU Y28-C and *Cyberlindnera subsufficiens* DMKU-YNB 32-2 showed high enzyme activities of 39, 35 and 26 mU/ml, respectively. The present study suggested that *C. saturnus*, *S. podzolica* and *C. subsufficiens* can producing the enzyme in large quantities. It can be used for further study on phytase enzyme production.

Keywords: Phytase; Phytic Acid; yeast; Phytase screening





Antimicrobial Test of Thai Medicinal Plant in the genus *Piper*

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The member of genus *Piper* (Piperaceae) or Sa Kan (Thai name) has long been regarded as vegetables and it is also considered as a medicinal plants. Many common species were reported as antimicrobial agents. The aim of this work is to investigate the antimicrobial activity of six species of *Piper* from Thailand as *P. boehmeriifolium*, *P. longum*, *P. retrofractum*, *P. ribesioides*, *P. umbellatum* and *P. wallichii*. Antimicrobial properties of these piperaceous plants have not been studied. Antimicrobial activity of different solvent crude extracts from leaf and stem of the *Piper* were comparatively studied by disc diffusion method. Minimum inhibitory concentration of the crude extract was also determined. The crude extracts of *P. boehmeriifolium*, *P. longum* and *P. retrofractum* exhibited greater antimicrobial activity against the selected bacterial and fungal strains. The extract from leaf has antimicrobial activity against broad spectrum of microorganisms. Ethylacetate leaf extract of *P. retrofractum* have the highest antimicrobial power against *Staphylococcus aureus*, *Bacillus cereus*, *Escherichia coli*, *Salmonella typhimuriam*, *Shigella* sp., *Fusarium* sp. and *Aspergillus flavus*. This report suggests that the test plants could be used as natural antimicrobial agents in both food preservation and human health.

Keywords: antibacterial activity, antifungal activity, antimicrobial activity, medicinal plant Piperaceous plant





Effect of *Moringa oleifera* leaves extract for growth inhibition of enteropathogenic bacteria

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Moringa oleifera is one of the most useful trees that known as “the tree of life” which belonging to the genus *Moringa* in the family Moringaceae. It is the plant that commonly found in South and Southeast Asia, which has highly nutritious and powerful anti-oxidant, anti-inflammatory, and tissue-protective properties. Objective of this study is to test ability of *Moringa Oleifera* leaves extract for inhibition of 8 enteropathogenic bacteria (*Escherichia coli* O157:H7, *Serratia marcescens*, *Enterobacter aerogenes*, *Staphylococcus aureus*, *Vibrio parahaemolyticus*, *Shigella flexneri*, *Salmonella Typhi* and *Listeria monocytogenes*). *Moringa Oleifera* leaves extract in different condition include water extract of powder leaf, methanol extract of powder leaf and water extract of fresh leaf, were determined by disc agar diffusion method and minimum inhibitory concentration (MIC) method. Water extract of powder leaf and methanol extract of powder leaf displayed inhibition zone only in *S. aureus* with inhibition zone diameter of 0.75 and 0.85 millimeters, but do not inhibit other 7 bacterial strains. Water extract from fresh leaf can inhibit *Serratia marcescens*, *Staphylococcus aureus* and *Vibrio parahaemolyticus* with inhibition zone diameter of 0.75, 1.70 and 1.20 millimeters, respectively.

Keywords: *Moringa oleifera*, Enteropathogenic bacteria, Disc agar diffusion, Minimum inhibition concentration (MIC)





Study on antimicrobial activity of metal organic framework MIL-101

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The metal organic framework of chromium (III) terephthalate (MIL-101) is comprised of trimeric chromium (III) octahedral clusters interconnected with 1,4-benzenedicarboxylates, resulting in a highly porous 3-dimensional structure. The aim of this study was to evaluate the antimicrobial activity of MIL-101 against eight microorganisms, including *Bacillus cereus*, *Staphylococcus aureus*, *Escherichia coli*, *Salmonella Typhimurium*, *Saccharomyces cerevisiae*, *Aspergillus niger*, *Aspergillus oryzae* and *Fusarium oxysporum*. The antimicrobial activities tested against bacteria and yeast were estimated using agar well diffusion, minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) methods. From the MIC and MBC tests, the concentrations that inhibited the growth of bacterial and yeast cells were 200 ppm and 400 ppm. In order to investigate fungal inhibition caused by MIL-101, poisoned food method was employed in the experiment. The result showed that at 500 ppm of MIL-101 decreased the mycelium growth rate of *A. oryzae* and *F. oxysporum* greater than at 100 ppm. In contrast, low concentration (100 ppm) of MIL-101 suppressed the growth of *A. niger* better than high concentration (500 ppm). The finding of this study indicates that the antimicrobial activity of MIL-101 varied depending on the microbial groups and species. However, factors that affect improvement or declination of MIL-101 efficacy for controlling microbial growth need to be further studied.

Keywords: antimicrobial activity, metal organic framework, MIL-101





Inhibition activity of actinomycetes from Bangkrachao to inhibit *Alternaria* sp. and *Colletotrichum gloeosporioides*

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Bangkrachao, which located nearby Bangkok in Samut Prakarn Province, is called Bangkok's Green Lung. It's a large green area with varieties of agricultural plant and vegetable species. In this study, total of 70 actinomycetes that isolated from Bangkrachao in previous research were studied for their ability to inhibit phytopathogen. These isolates were divided into groups on the basis of their aerial spore mass color on ISP3 agar. The result showed that all the isolates were separated into 9 color groups and a non-spore forming group. In antifungal bioassay, all isolates were evaluated for antifungal activities against two species of pathogenic fungi, 1.) *Alternaria* sp. that caused leaf spot in many types of crucifers, including kale, cabbage and broccoli and 2.) *Colletotrichum gloeosporioides* which caused anthracnose that is the most wide-spread disease in mangoes. The result of antifungal activity by dual culture method showed that 10 actinomycete isolates (14.28%) and 14 isolates (20%) had antifungal efficiency more than 50 % to inhibited *Alternaria* sp. and *Colletotrichum gloeosporioides*, respectively. Isolates 2BS15 and 2BS5 had the highest antifungal efficiency to inhibit *Alternaria* sp. (100% inhibition) and *Colletotrichum gloeosporioides* (73% inhibition), respectively. Molecular identification of the potent isolates, 2BS15 and 2BS5, were performed based on 16S rRNA gene nucleotide sequences. The result showed that isolates 2BS15 and 2BS5 were closely related to *Streptomyces misionensis* 100% and *Streptomyces albidoflavus* 99.88%, respectively. This study was a primary step to screen for potential actinomycetes to use as biocontrol agents in the future.

Keywords: actinomycetes, *Colletotrichum gloeosporioides*, *Alternaria* sp., antifungal activity, Bangkrachao





The efficiency of bacterial chitinase in control of pathogenic fungi causing disease in chili

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Chili is an important economic vegetable, and spices. It has many benefits and high nutrition. The problems of chili cultivation are insect pests and diseases. Therefore, farmers desire to use excessive chemicals to eliminate losses caused by those pests. A biological control agent is an alternative to synthetic pesticides, which have remaining toxic residue on produces, are able to enter the food chain, and seriously cause environmental problems. Biological control, in particular, is the application of microbial antagonists for controlling pathogenic microorganisms. The objective of this study was to evaluate the efficiency of bacterial chitinase in controlling of pathogenic fungi causing diseases on chili. *Paenibacillus phyllosphaerae* PALXIL04, the antagonistic bacterium, is a gram-positive, aerobic rod-shaped and endospore-forming bacterium. Inhibitory activity of *P. phyllosphaerae* PALXIL04 culture and its crude chitinase enzyme against *Fusarium oxysporum*, *Rhizoctonia solani*, *Sclerotium rolfsii*, *Colletotrichum capsici*, and *C. gloeosporioides* was examined on Trypticase soy agar (TSA) amended with 0.3% colloidal chitin using dual culture technique. The results showed that, the bacterial culture of *P. phyllosphaerae* PALXIL04 was not able to suppress the growth of all tested fungi. The microscopic study revealed physiological abnormalities of *R. solani* mycelium when *R. solani* was grown with *P. phyllosphaerae* PALXIL04 on the dual culture plate. Preliminary characterization of crude chitinase showed that the enzyme could digest colloidal chitin in the TSA medium. However, the efficacy of crude chitinase is not yet evaluated for its efficiency to inhibit the growth and infection of the fungal pathogens on chili fruits.

Keywords: chili, chitinase, *Paenibacillus phyllosphaerae*, pathogenic fungi





Detection of *Shigella flexneri* by Loop – Mediated Isothermal Amplification (LAMP) technique

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Shigella are members of the family Enterobacteriaceae, This is a microorganism that causes major intestinal diseases. *Shigella* caused Shigellosis, which are health problem all over the world. *Shigella* are transmitted through a fecal-oral route, person-to-person contact, or contaminated from water and food. *Shigella* only 200-1,000 cells can cause infection. The most common symptoms of shigellosis after exposure 1-2 days such as abdominal pain and stomach cramps, diarrhea, tenesmus, fever (over 38.5 degree Celsius), loss of appetite, nausea, vomiting, fatigue, malnutrition. In this study, Loop-mediated isothermal amplification (LAMP) assay was developed to detect *Shigella flexneri*. Four sets of LAMP primers was designed to specifically target invasion plasmid antigen H (*ipaH*) gene, Which can recognize 6 distinct regions of the target gene. LAMP assay is performed under isothermal conditions, could be completed within 60 minute and reaction carried out in a 25 μ l. In this assay, parameters tested were the temperature and time taken to amplify DNA. Sensitivity test was performed using different concentrations of DNA template (10 fold serial dilutions), to determine the minimum concentration of DNA template of *Shigella flexneri* that can be detected. Specificity test using bacteria 10 strains (*Shigella flexneri*, *Shigella sonnei*, *Escherichia coli*, *Enterobacter aerogenes*, *Serratia marcescens*, *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Bacillus cereus*, *Klebsiella pneumonia*, *Salmonella Typhi*) to evaluate specificity for detection of the *ipaH* gene. Sensitivity test of LAMP amplification with spiked food. Result of the LAMP reaction can be seen with naked eye by observing hydroxynaphthol blue dye change (positive reaction is indicated by dye change purple to blue). The LAMP products were confirmed by electrophoresis on 3% agarose gel and visualized under ultraviolet light after staining with ethidium bromide. LAMP technique is high efficiency, simple, rapid, low cost effective equipment and sensitive method.

Keywords: Loop-mediated isothermal amplification (LAMP), *Shigella flexneri*, *ipaH* gene





Optimization of succinic acid production by *Corynebacterium glutamicum* isolated in Thailand

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Succinic acid is an important chemical because it can be used as a substrate for the production of various chemicals such as 1,4-butanediol, Tetrahydrofuran, Succinamide. To date, it is currently produced by biological methods using succinic acid producing bacteria. Recently, there is a few papers review the production of succinic acid in *Corynebacterium glutamicum* via the reductive TCA cycle, which occurs under limited oxygen condition involving two related enzymes, phosphoenolpyruvate carboxylase and pyruvate carboxylase, with carbon dioxide as co-substrate. Thus, this research aims to characterize the succinic acid production of *Corynebacterium glutamicum* strains isolated in Thailand. Three strains have been investigated namely, DS50 (fast growing strain in glucose), BB9 (mutant with UV radiation of DS50) and CS176 (arabinose utilizing strain). Two-state fermentation using basal salt MM or BT medium consisting of the first state to increase biomass and the second state to produce the succinic acid under either the oxygen-limited condition or adding bicarbonate were carried out. The experiments were done in tested medium containing glucose as a carbon source and in a screw cap tube. The optimized condition was screened by the detection of succinic acid using Thin Layer Chromatography with Ethanol: NH₃: water ratio of 75.5: 12.5: 12 as a mobile phase. The results showed that CS176 could produce succinic acid better than the other two strains. The optimum condition under oxygen limit is in a 16 x 125 mm screw cap containing 15 ml of 11 ml of culture in BT medium (turbidity at 600 nm was 13.81) and 4 ml of BT without nitrogen source. Moreover, the production of succinic acid of CS176 in the BT adding bicarbonate (0-32 mM) with varying the time of adding (0-4 hours) and harvesting (1-6 hours) were further investigated. The results indicated the succinic acid is increased according to the bicarbonate concentration and harvesting time. The efficiency of succinic acid production of CS176 in the optimize condition and quantitative detection of succinic acid using High Performance Liquid Chromatography (HPLC) will be elucidated. The results of this study are preliminary data to further study the process of bio-based succinic acid production by *Corynebacterium glutamicum* especially by CS176 strain.

Keywords: 2-state fermentation, bio-based succinic acid production, *Corynebacterium glutamicum* CS176, succinic acid producing bacteria





Solid state culture of *Streptomyces* sp. 8ER183 for the production of bioactive compound to control soft rot disease in orchids caused by *Erwinia carotovora*

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Orchids is the most important exported flower in Thailand, however, the orchid disease such as soft rot disease caused by *Erwinia carotovora* subsp. *carotovora* resulting in poor orchid quality and low yield and economic losses. Currently, biological control by actinomycete group is proposed to be an alternative effective, safe and ecologically friendly approach for disease management. The objective of this research was to investigate the optimization of solid state culture on the production of bioactive compound by *Streptomyces* sp. 8ER183 to control soft rot disease. The strain cultivated on 10 g raw materials in the 250 ml Erlenmeyer flask consisting of rice bran and coconut husk at the ratio 7.5: 2.5, at initial moisture content of 55 % with the supplementation of 4 ml of inorganic salts solution (2g/l KH_2PO_4 , 1g/l K_2HPO_4 , 5g/l $(\text{NH}_4)_2\text{SO}_4$, 0.1 g/l $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, and 1 g/l NaCl) at 37 °C gave the highest bioactive compound, diameter of inhibition zone 2.4 cm, and growth with 5.45×10^7 CFU/g solid at 9th day culture. Ammonium sulfate in mineral salt solution required for the production both bioactive compound and growth. In addition, the bioactive compound produced by *Streptomyces* sp. 8ER183 had the ability to inhibit both Gram-positive and Gram-negative bacteria.

Keywords: Orchids, *Streptomyces* sp. 8ER183, Solid-state fermentation, Bioactive compound, *Erwinia carotovora*





Antagonistic mechanisms of vetiver rhizospheric and phylloplane yeasts to control plant pathogenic fungi

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Rice, corn and sugarcane are known as economic plants in Thailand. During growing these crops diseases caused by various organisms including bacteria and fungi are highly possible to damage these crops. That's why control of plant disease is very importance. Fungi are the casual agents of various plant diseases. Nowadays to control plant diseases there are two types including chemical and biological controls. Antagonistic yeasts are possible to control these diseases by various mechanisms. The aims of this research were to study antagonistic mechanisms of the antagonistic yeasts against plant pathogenic fungi and to determine their plant growth promoting activity. A total of 45 antagonistic yeasts strains isolated from rhizospheric and phylloplane of vetiver grass were examined for their antagonistic mechanisms against fungi causal rice disease (*Fusarium moniliforme*, *Helminthosporium oryzae*, *Curvularia lunata*, *Pyricularia grisea* and *Rhizoctonia solani*), corn disease (*Bipolaris zeicola*, *Macrophomina phaseolina* and *Fusarium moniliforme*) and sugarcane disease (*Fusarium moniliforme*). The results showed that *Candida tropicalis* DMKU-LV 45 effectively inhibited fungal pathogens namely *F. moniliforme* (rice), *C. lunata*, *R. solani* and *M. phaseolina* by the mechanism of competition of nutrient and space. In addition *C. tropicalis* DMKU-LV 45 also effectively inhibited *C. lunata* by mechanism of volatile compounds production with the efficiency of 60% and *C. tropicalis* DMKU-LV 11 effectively inhibited *P. grisea* by the competition of nutrients and space and volatile compounds. The production of fungal cell wall degrading enzyme i.e. protease, chitinase and glucanase by all antagonistic yeasts were investigated. Five strains produced protease on skimmed milk agar and *C. tropicalis* DMKU-LV 06 had the highest enzyme activity index. Four strains produced glucanase on carboxymethyl cellulose agar (CMC) and *Candida metapsilosis* DMKU-LV 07 had the highest enzyme activity index. No chitinase was produced by any yeast strains. Investigation of plant growth promoting activity revealed that five strains showed the ability to solubilize phosphate and *C. tropicalis* DMKU-LV 45 showed the highest ability. Two strains solubilized zinc oxide and *C. tropicalis* DMKU-LV 45 showed the highest ability. Seven strains produced siderophore and *C. tropicalis* DMKU-LV 11 showed the highest ability. The biological control mechanisms of plant pathogenic fungi by the antagonistic yeast could be summarized as, seven yeasts strains showed the ability to control *F. moniliforme* (rice), eight strains showed the ability to control *R. solani*, 29 strains showed the ability to control *C. lunata*, 10 strains showed the ability to control *P. grisea* and four strains showed the ability to control *M. phaseolina*. Twelve yeast strains showed the ability to be plant growth promoting yeasts. Moreover, the competition of nutrients and space was the main mechanism to control *H. oryzae*, *B. zeicola*, *F. moniliforme* (corn) and *F. moniliforme* (sugarcane). The production of volatile compounds was the main mechanism to control *F. moniliforme* (rice), *H. oryzae*, *R. solani*, *B. zeicola*, *F. moniliforme* (corn), *M. phaseolina* and *F. moniliforme* (sugarcane).

Keyword: Biocontrol, antagonistic yeast and plant growth promotion





Optimization of lipase production by yeast isolated from grease trap

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Lipase catalyzes hydrolysis of triglycerides yields glycerol and free fatty-acids. In the present work, we optimized medium components through one-factor-at-a-time (OFAT) approach in submerged cultivation of extracellular lipase producing yeast strain namely L1-3. This yeast was isolated from grease trap at the canteen of the Faculty of Science, Kasetsart University. To optimize, medium composition, different carbon, nitrogen and oil type were varied. Of all the substrates evaluated, we found that lactose, peptone and soybean oil are the best carbon source, nitrogen source and oil type for extracellular lipase production by the isolate L1-3, respectively. Using the optimal medium containing 0.7% (w/v) lactose, 0.3% (w/v) peptone and 1.5% (v/v) soybean oil, 0.3% (w/v) yeast extract and 0.3% (w/v) malt extract at 30 °C for 3 days, the lipase activity of 3010.51 U/ml was obtained. In addition, the culture supernatant (with extracellular lipase) was shown to be stable at 4 °C for 2 day by remaining 100% relative activity to the original sample.

Keywords: Lipase, Yeast, Lipase production, Optimization, Effect of surfactant





Study of ability to digest sugarcane bagasse of *Paenibacillus* isolated from termite gut

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There is a large amount of agricultural waste in Thailand, but it is used to bring benefits relatively low. In this study, an ability of the isolated gut bacteria to digest sugarcane bagasse was tested. The bacterial strain was isolated from the gut of soil-feeding termite, *Pericapritermes semarangi*. Molecular identification based on 16S rRNA gene showed that, the isolate was related to *Paenibacillus taichungensis* strain BCRC 17757. It has the ability to digest various substrates: carboxymethyl cellulose (CMC), xylan, pectin and starch. To test its ability to digest the selected agricultural waste, Berg's medium was added with pretreated bagasse (1% w/v) for study of production of various enzymes including cellulase, xylanase, pectinase and amylase, in neutral pH (pH 7) at room temperature. The activity of each enzyme was tested by DNS method and detected at every 24 hours for 7 days. The experiment found that, the bacterial growth and enzyme production of this bacteria were related to the primary metabolite. Unit of the amount of particular enzyme was difference and the xylanase activity was dominant. In addition, the bacterial growth was tested in different conditions such as temperature, pH and salinity. The results of this study suggested that, the *Paenibacillus* related strain has high ability to produce the enzymes from the agricultural waste, sugarcane bagasse. The bacteria and enzymes can be applied for industry and biotechnology.

Keywords: Sugarcane bagasse, *Paenibacillus*, Agricultural waste, Cellulase, Xylanase, Pectinase, Amylase, Termite gut





Expression profile of Bid gene in liver tissues of patients with hepatitis B virus-related hepatocellular carcinoma

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Hepatitis B virus (HBV) infection is a major public health problem and causes acute and chronic hepatitis. Chronic HBV infection can lead to cirrhosis and hepatocellular carcinoma (HCC). Apoptosis is a program cell death which occurs in multicellular organism. Bid (BH3 interacting-domain death agonist) is a pro-apoptotic member of the Bcl-2 protein family that induces apoptosis by interacting with other pro-apoptotic Bcl-2 family proteins at mitochondrial surface. The objective of this research is to compare the expression profile of Bid gene in liver tumor tissues and adjacent non-tumor tissues. In our study, we extracted RNA from liver tissues and then convert into cDNA. The expression profile of Bid gene was detected in liver tissues by using quantitative real-time reverse transcriptase-polymerase chain reaction (RT-PCR). In the present study, we found that the mRNA expression of Bid gene was decreased in HCC tissues when compared to adjacent non-tumor tissues. This finding suggests that the decreasing of Bid might be an important mechanism leading to uncontrolled growth of tumor cell. However, the functional study of Bid molecule in liver cancer is required in the future.

Keywords: *Bid, Hepatitis B virus, Hepatocellular carcinoma*





Screening of Ammonia Producing Bacteria from Tempe

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Over fermented tempe and low quality tempe usually have unpleasant ammoniacal odor that almost people are not familiar. In tempe production, *Rhizopus oligosporus* was inoculated, however other microorganism such as various kinds of bacteria and yeast were also contributed resulted in hydrolysis of soybean protein to peptide and amino acid lead to ammonia. This experiment was aimed to find out the ammonia causal microorganism from tempe products. Our tested samples were included of good quality, poor quality and also overripe tempe products. Total bacteria, enterobacteria and yeast were counted on Plate count agar, MacConkey agar and YM agar, respectively. Heat shock of 100 times diluted tempe samples were performed on Plate count agar for determination of endospore forming bacteria. Different bacteria colonies of each medium were collected and tested for proteolysis activity. The results showed that 61 isolates from 66 isolates could produce ammonia from peptone broth, soytone broth, tempe broth (detection by Nessler's solution) and/or urea broth. Those were 36 isolates of enterobacteria and 17 isolates of endospore forming bacteria. Eighteen of 36 isolates of enterobacteria were strong positive in peptone broth, soytone broth, tempe broth and urea broth. They were identified by conventional method as *Klebsiella pneumoniae* subsp. pneumonia, *Klebsiella planticola* and *Enterobacter cloacae*. Whereas other 18 isolates were positive in peptone broth, soytone broth, tempe broth were identified as *Klebsiella pneumoniae* subsp. ozaenae and *Enterobacter aerogenes* and unidentified sp. Fifteen of 17 isolates of endospore forming bacteria which were identified as *Bacillus thuringiensis* also showed proteolysis activity on those media only exception of urease activity.

Keywords: Tempe, Ammonia producing bacteria





Characterization and optimization of microalgae *Schizochytrium* sp. for DHA production

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DHA is an omega-3 polyunsaturated fatty acid. Its structure is a carboxylic acid with 22 carbon atoms and six *cis* double bonds. DHA plays a significant role in human cognitive ability and brain physiology. It is a primary structural component of the human brain, cerebral cortex, skin, sperm, testicles and retina. In aquatic animals, DHA is important in the development of juvenile animals such as molting. There are reports showed that the marine microalgae within the genus *Schizochytrium* sp., is a primary producer of DHA in large amounts. The objectives of this research were isolation and screening of *Schizochytrium* sp. from the mangrove forests along the Gulf of Thailand, and optimization of culture conditions for DHA production. Isolation was carried out on Glucose Yeast extract Peptone agar (GYP) supplemented with chloramphenical. One hundred and sixty-eight isolates of *Schizochytrium* sp. were obtained for screening by using GYP broth containing 3% glucose and incubated at 25°C for 42 hours in a rotary shaker at 115 rpm. The results showed that 20 isolates contained relatively high DHA contents. Tertiary screening revealed that one strain namely *Schizochytrium* sp. SA17-28 had the highest DHA content at 26.50% of total fatty acids. Optimization of culture condition were conducted as monosodium glutamate (MSG) concentration at 0, 0.25, 0.5, 0.75 and 1.0 %, temperature at 21, 23, 25, 27, 28, 30, 32, 35 and 37 °C, and salinity at 0, 1.0, 1.5, 2.0, 3.0 %. The results showed that medium containing 0.25% MSG, temperature 30°C salinity 2.0% enhanced highest DHA production by *Schizochytrium* sp. SA17-28 at 4.70, 4.02 and 5.43 g/L, highest DHA content 34.21, 35.32 and 26.39% of total fatty acids, respectively. Cultivation in 2 liter fermenter was carried out. The highest DHA production 14.75 g/L and DHA content 42.87% of total fatty acids were obtained at 36 hours of cultivation. However, the highest biomass 15.12 g/L was obtained at 24 hours of cultivation.

Keywords: DHA, *Schizochytrium* sp., Mangrove forest, Thailand





Study of Antitumor Activity of Hydrophobin Extracts from Mushroom Culture

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Hydrophobins are small amphipathic proteins about 7-15 kDa sized only produced by mushroom and filamentous fungi. The properties of these proteins are able to self-assemble into amphipathic monolayer at hydrophobic/hydrophilic interfaces. Previous research has found that the Hydrophobins SC3 from *Schizophyllum commune* have an antitumor activity against sarcoma and melanoma. The objective of this research was to investigate antitumor activities of hydrophobin crude extracts from cultures of *Agrocybe cylindracea* and *Ganoderma lucidum* with TFA extraction by examining their anti-proliferation on hepatocellular carcinoma (HepG2). HepG2 cell lines were cultivated in 96-well plate at the concentration of 3×10^3 cells per well. The MTS assay was used to determine the inhibitory effects of hydrophobin crude extracts. Putative hydrophobins from *Agrocybe cylindracea* and *Ganoderma lucidum* were approximately 8 kDa sized. They were subjected to examine their antitumor activity.

Keywords: Hydrophobins, HepG2, Mushroom





Study on phytase production of molds isolated from soil

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The objectives of this study are to study the optimal conditions and the stability of the phytase produced by molds that were isolated from soil samples of Thailand regions. Forty-one mold isolates which are able to degrade phytin were cultivated in the modified phytase screening medium and measured the phytase activity in order to select the fungi with the highest phytase activity. Among forty-one mold isolates, the isolate labeled “PY22-4” showed the highest phytase activity (0.622 units/ml) after cultivation for 7 days. The phytase of this mold was studied for the optimal conditions and the stability of phytase at various temperatures and pH. It was studied with the pHs and temperatures from 2.0 to 10.0 and 30 to 70°C, respectively. The optimal pH for the phytase activity was 2.0 and the enzyme shows the highest stability at pH 2.5 and 3.0 after incubated at 4°C for 20h. The optimal temperature for the enzyme activity and heat stability of enzymes are under investigation.

Keywords: Phytase · Phytate · Fungi





Optimization of indole-3-acetic acid production by *Enterobacter* sp.DMKU-RP206

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Indole-3-acetic acid (IAA) is plant hormone of the auxin group which produced by plants and microorganisms including bacteria, actinomycetes, filamentous fungi and yeast. IAA is the main auxin in plant, controlling many important physiological processes including responses to light and gravity, cell division and enlargement and tissue differentiation. The purpose of this research is to study an optimum condition of IAA production by *Enterobacter* sp. DMKU-RP206 isolated from rice phyllosphere. The most appropriate carbon and nitrogen sources were investigated by one factor at a time (OFAT) method which each factors were varied one at a time while keeping other factors at constant levels. The result showed that *Enterobacter* sp. DMKU-RP206 produced the highest amount of IAA (1,014.4 $\mu\text{g/ml}$) when lactose was used as a carbon source compared with the others (starch, dextrose, mannitol and galactose). This bacteria produced the highest worth of IAA (597.695 $\mu\text{g/ml}$) when yeast extract (technical grade) was used as a nitrogen source compared with other nitrogen sources (yeast autolysates and yeast extract laboratory grade). For both experiments, IAA was confirmed by colorimetric method (Salkowski's Test). More results indicated that sweet whey which is the byproduct of cheeses making can be used to replace lactose as a carbon source by *Enterobacter* sp. DMKU-RP206. A Box-behnken design was used in optimization of IAA production experiments. The optimum conditions of IAA production was found when *Enterobacter* sp. DMKU-RP206 was cultured in test medium (1.48% sweet whey, 1.42% yeast extract technical grade, 0.88% L-tryptophan and pH was adjusted to 6.0) incubated on a rotary shaker at 200 rpm and 30 °C for 3 days The bacteria produced 4,084.8 μg IAA/ml confirmed by High performance liquid chromatography (HPLC). In addition, the effect of IAA on seeds germination were studied. Rice seeds cultivar HomMali 105 were planted in a sterilized boxes with IAA supernatant varying concentration (0.1, 1, 10, 100 and 1000 μg IAA/ml). The result showed that the supernatant at 10 μg IAA/ml is the best sample to increase germination rate of the rice seeds when compared with other concentration and control. The results of this study indicate the potential of *Enterobacter* sp. DMKU-RP206 as a significant producer of plant growth promoting substance and a good inoculant to promote the growth of rice seedlings.

Keywords: IAA, l-tryptophan, optimization, *Enterobacter*, auxin.





Detection and isolation of coliform bacteria and *Pseudomonas* spp. from dishwashing scrubber: An examination of multidrug resistant *Pseudomonas* spp.

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Survival and cross contamination of food borne pathogens are crucial factors of food poisoning outbreaks in kitchen environment. Dishwashing scrubber frequently contacts with dishes, cooking utensils and other food-contacted surfaces. Accumulation of organic matters and/or fat on surface of the scrubbers, and moist environment around kitchen sink, may lead to growth and colonization of microorganisms. The aims of this work were to survey and study the accumulation of coliform bacteria and *Pseudomonas* spp. in dishwashing scrubbers from four cafeterias of Kasetsart University, and to examine antimicrobial resistant ability of isolated *Pseudomonas* spp. As a detection of coliform bacteria by most probable number (MPN) method with 15 dishwashing scrubber samples, 13 samples were detected with coliform bacteria and none fecal coliform in all samples. Isolation of *Pseudomonas* spp. was performed on *Pseudomonas* CN agar and confirmed by morphological characteristics and catalase test. A total number of 54 *Pseudomonas* strains were isolated from 10 dishwashing scrubber samples. Antimicrobial resistant test was examined by disc agar diffusion method. The following antimicrobial were tested: 30 µg Azetreonam, 30 µg Cefotaxime, 5 µg Ciprofloxacin, 10 µg Meropenem, and 100 µg Piperacillin.

Keywords: coliform bacteria, *Pseudomonas* spp., multidrug resistance, dishwashing scrubber





Molecule Characterization and Functional Analysis of Lectin in Black Tiger Shrimp

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Black tiger shrimp is one of the economic creatures of great value to Thailand. The spread of bacteria in the shrimp farming industry is a major cause of shrimp loss. Therefore, to understanding the innate immune system of the shrimp is important to prevent loss of shrimp production. Lectin is a protein found in shrimp and is associated with the immune system. Shrimp relies on the innate immune system like other invertebrates lacking the adaptive immune system. The Innate immune system is the first defense against infection, which consists of humoral and cellular immune respond system to against invasion of invading pathogens. The cellular immune responses include phagocytosis, nodulation and encapsulation while humoral immune responses involve the synthesis and secretion of proteins in the immune system. Invertebrates will recognize invading pathogens using pattern-recognition proteins (PRPs), which recognize and bind to pathogen-associated molecule pattern (PAMPs). The primary response to invasive pathogens in shrimp is associated with multiple PRPs including lectins. C-type lectin (calcium-dependent lectin), containing a carbohydrate recognition domain (CRD), can bind to carbohydrate moieties on viruses and bacteria. Therefore, this research focuses on the study of the mannose binding protein in the black tiger shrimp. Expression of recombinant MBP was carried out and the protein function will be elucidated. The knowledge from this research may lead to further development of disease control in shrimp.

Keywords: Lectin, Black tiger shrimp, Immune system





Characterization and improvement of thermostable DNA ligase for applications in biotechnology

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At present, molecular cloning has become routine in laboratory. The ligase that is the most frequently used in molecular biology is DNA ligase from T4. T4 DNA ligase is a 68 kDa monomer that requires Mg²⁺ and ATP as cofactors. T4 DNA ligase, which is essential in both DNA replication and DNA repair processes, plays a key role in formation of phosphodiester bonds between the 5'-phosphoryl and 3'-hydroxyl end-groups. T4 DNA ligase is normally used for ligating recombinant DNA molecules contained inserted DNA fragments to the appropriate vector molecule. This study aimed to overexpress T4 DNA ligase in *E. coli* using pTTQ18 expression vector containing the *tac* promoter, which is a weaker promoter compared that of the T7 bacteriophage. Previous study had shown the expression of T4 DNA ligase gene in *E. coli* with strong T7 promoter resulted in formation of inclusion bodies. T4 DNA ligase gene was amplified by PCR reaction, and PCR fragment was purified by Geneaid® and inserted to pGEM cloning vector using *E. coli* strain DH5 α as a cloning host. The recombinant plasmid obtained, designated as pGEM-T4 DNA ligase, was propagated in DH5 α , and the plasmid was purified by alkali lysis method. To confirm that pGEM-T4 DNA ligase contains the correct inserted DNA, The plasmid was digested by restriction endonucleases (*Sac*II and *Nde*I) that pGEM and T4 DNA ligase was separated. The result showed that the obtained plasmid contained inserted T4 DNA ligase. Furthermore, the T4 DNA ligase gene was digested by *Xma*I and *Hind*III and the gene was purified by using Geneaid®. This fragment was ligated into pTTQ18 expression vector which have been digested previously with *Xma*I and *Hind*III. The ligation mixture was transformed in *E. coli* strain BL21 (DE3), and the cell was grown at 37°C. The expression of T4 DNA ligase was induced by addition 1 mM lactose.





Production of *Plasmodium falciparum* Histidine-rich protein II for Plasmepsin V activity assay

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Malaria is still the biggest killer of all the parasitic disease caused by female *Anopheles* mosquitoes infected with *Plasmodium* parasites. *P. falciparum* and *P. vivax* have been identified as the most virulent species causing human malaria. During the erythrocytic stage of infection, erythrocyte remodeling via expression of many hundreds of exported protein begins immediately. Histidine-rich protein II (HRPII), composed of 40% alanine, 36% histidine and 11% aspartate, is a specific protein expressed only by *P. falciparum*, so detection of HRPII provides evidence for active or recent infection and is utilized for both diagnostic and surveillance purposes. HRPII contains a PEXEL motif (R-x-L-x-E/D/Q) required for export, which acts as a recognition site for an ER-resident aspartic protease, called plasmepsin V (PMV). This study therefore aimed to produce recombinant HRPII for develop the enzymatic assay of *Pf*PMV by using HRPII as a natural substrate. The *hrpII* gene was recodonized and synthesized by GenScript® and inserted into pET32a, generating pET32a-HRPII-GS. Recombinant HRPII (Trx-HRPII) was overexpressed in *E.coli* BL21 (DE3), induced with 0.2 mM IPTG at OD₆₀₀ ≈ 0.5, 16°C for 16 h. The protein was purified by a nickel affinity column chromatography followed by Q sepharose column and a size exclusion chromatography. The purified protein was analyzed by Western blot and LC/MS-MS. Western blot analysis of purified Trx-HRPII probed with either anti-His and anti-Trx antibodies revealed two bands of proteins at the molecular weight of 55 and 72 kDa. LC/MS-MS of these two proteins showed that both proteins contained Trx protein but no amino acid sequences of HRPII were detected. Interestingly, three PEXEL motifs (RGLLQ, RDLLE and RNLQQ) were identified. Four protein fragments were observed when Trx-HRPII was incubated with *Pf*PMV. In order to test if this problem has stemmed from the synthetic plasmid, the *hrpII* gene from pET32a-HRPII-GS was amplified by PCR and subcloned into pET32a, generating pET32a-HRPII-His. The overexpression and purification of HRPII from pET32a-HRPII-His are being carried out and the results will also be presented. In conclusion, the recombinant HRPII produced in this work will be utilized as a substrate in PMV activity assay, and as a standard protein for developing HPRII kits to detect HRPII from malaria-infected blood samples.

Keywords: Malaria, *Plasmodium falciparum*, Histidine-rich protein II, PEXEL, Plasmepsin V





Expression and Purification of Structural Proteins in New Emerging Virus in Tilapia Nile tilapia (*Oreochromis niloticus*) in *Escherichia coli* and Neutralization Assay

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Nile tilapia (*Oreochromis niloticus*) is an important global food source because their omnivorous diet, tolerance for high-density aquaculture and relative disease resistant. Since 2009, found substantial mortality of wild tilapia has been observed in the Sea of Galilee and in commercial ponds in Israel and Ecuador. Symptoms of tilapia after getting infected are black discoloration, skin abrasions, ocular degeneration and dead. It was caused by a novel RNA virus infection. The virus, denominated tilapia lake virus (TiLV) was isolated from diseased fish, propagated in cell culture and it induced a cytopathic effect. Recently, in Thailand, the emerging of similar virus infection was mentioned. Therefore, in this study, we prepared cDNA from diseased fish and used as template for viral structural genes amplification. Ten viral open reading frames (ORFs) were amplified by using polymerase chain reaction (PCR). The amplified DNAs were ligated to TA cloning vector and sequenced. Corrected viral ORFs were transferred from TA cloning vector to *Escherichia coli* (*E. coli*) expression vector (pET-22b) for recombinant protein production. The recombinant expression vector harboring target ORFs were transformed to *E. coli* BL21 and induced recombinant protein expression by using IPTG. Recombinant proteins were purified by the affinity column. Recombinant protein (ORF7) was used to determine the inhibition of viral infection on fish cell line by using neutralization assay. This study will provide important molecular information of novel emerged virus in Tilapia which prevention or protection methods would be developed.

Keywords: virus, TiLV, Tilapia, neutralization assay, PCR





Design and over-expression of MMP-9-sensitive peptide hydrogels

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Breast cancer is a one of the most common cancer, which is the leading cause of death among women worldwide. Invasive breast cancer can invade to other tissues in the body which make it difficult to treat. One of the factor involve metastasis in breast cancer is the over-expression of matrix metalloproteinase-9 (MMP-9) to degrades extracellular matrix around the cells. In this study, we designed MMP-9-sensitive hydrogels for studying metastasis of breast cancer. The hydrogel consisted of three-stranded peptides incorporated with MMP-cleavage site, which are PDGQV and PRRLT. The sequences of the peptides were converted to polynucleotides, which were codon-optimized. Then, the genes were transformed into *Escherichia coli* BL21(DE3) strain. Isopropyl β -D-1-thiogalactopyranoside (IPTG) was used to induce expression. SDS-polyacrylamide gel electrophoresis (SDS-PAGE) was employed to investigate the expression level. Peptide's purity was analyzed by SDS-PAGE and mass spectrometry. The peptide form this study will be utilizes as artificial matrix to study the metastasis of breast cancer.

Keywords: MMP-9, peptide, hydrogel, breast cancer





Roles of Polypyrimidine tract binding protein (PTB) in the functions of the Hepatitis B virus post-transcriptional regulatory element (PRE)

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Hepatocellular carcinoma (HCC) is one of the most prevalent cancers in the world. Its incidence is increasing in many parts of the world. The major risk factor of HCC is hepatitis B virus (HBV) infection. At present, current vaccines effectively protect new infection, but anti-HBV drugs have associated with severe side effects and viral resistance. Therefore, a better understanding of how HBV replicate in the host cells is essential especially for a development of novel anti-HBV drugs. Recently, a proposed core element of HBV post-transcriptional regulatory element (HBV PRE 1151-1410) has been reported to export intronless HBV transcripts and also facilitated inhibition of HBV mRNA splicing. However, another fraction of HBV PRE has been reported to bind with endogenous polypyrimidine tract binding protein (PTB) *in vitro* and enhanced the nuclear export of HBV. Therefore, the role of PTB on function of the HBV PRE core element is unknown. This study therefore aimed to study the effects of overexpression of PTB on the function of HBV PRE 1151-1410. In order to achieve the aim, different amounts (0-400 ng) of the plasmid expressed full-length of PTB protein was co-transfected with the luciferase reporter plasmid that either fused with the full-length HBV PRE or HBV PRE 1151-1410 in liver cancer cell (HepG2). The results from the luciferase assay confirmed the previous report that both full-length HBV PRE and HBV PRE 1151-1410 could enhance the nuclear export of the intronless luciferase transcript. Interestingly, the overexpression of PTB in the presence of full-length HBV PRE did not lead to a dose-dependent induction of HBV PRE, but it resulted in the repression of luciferase activity. The inhibitory effect was observed with 100 ng of PTB overexpression. Likewise, 100 ng of PTB also decreased luciferase activity in the presence of HBV PRE 1151-1410. The results from this study implied that PTB was involved in the regulation of function of HBV PRE 1151-1410.

Keywords: HCC, HBV, PRE, PTB, Overexpression, luciferase assay





Analysis and biological activities of resveratrol in mulberry fruit extracts

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Mulberry (*Morus* spp.) is one of economic plants grown widely in Northeastern region of Thailand. It has been exploited commercially for the silk industry to rear silkworms (*Bombyx mori*). Mulberry fruits are rich in vitamins, minerals, fibers and high levels of antioxidants. Among phytochemicals that have antioxidant properties, resveratrol (3,5,4'-trihydroxystilbene) has recently gained much scientific interest. Resveratrol is one of the major stilbene phytoalexin produced by plants in response to biotic and abiotic stresses. It has lots of human-health benefits such as cardio-protection, anti-aging, cancer-chemoprevention and is thus widely used in dietary supplement industry. Therefore, the aims of this study were to determine the resveratrol contents in fruits of mulberry, Kamphaengsaen 42 (KPS) variety and investigate the expression of genes involved in resveratrol biosynthesis. Firstly, the resveratrol was extracted from mulberry fruits using chloroform: ethyl acetate (1:1 v/v). High Performance Liquid Chromatography (HPLC) was used to quantify resveratrol content in the fruit extracts. Then, total RNA were isolated from leaves and fruits of mulberry plants, which grown under well-watered (control) and drought conditions, then converted to cDNA. The expression of *phenylalanine ammonia lyase (PAL)*, *stilbene synthase (STS)* and *4-coumarate:CoA ligase (ACL)* were determined by qRT-PCR using specific primers. Lastly, the antibacterial activity was also checked against *Escherichia coli* and the antioxidant activity was examined by DPPH assay. The study of resveratrol and its biosynthesis will bring benefits for quality improvement and industrial exploration of mulberry fruits.

Keywords: Gene expression, Mulberry, Resveratrol, Stress response





Molecular Characterization of Nile tilapia (*Oreochromis niloticus*) Aromatase and Inhibitor Identification

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Nile tilapia (*Oreochromis niloticus*) is an important farmed fish in Thailand. Faster growth and uniform fish size is the most important requirement for successful tilapia aquaculture. Therefore, farmers prefer to culture “all male tilapia or monosex culture” because female tilapia always stunt growing from the reproductive cycle. Currently technique used to produce monosex tilapia culture is the steroid hormone application to reverse fish sex from female to male during fry stage. However, the utilization of hormone is restricted in human food consumption, then, a novel method of safely fish sex reversal is being required. In this study, we are interested to control sex reversal mechanism by targeting to a key enzyme. Aromatase is an important enzyme in fish sex differentiation by catalyzing the conversion of androgen to estrogen resulting in changing male tilapia to female tilapia. The purpose of this study was to identify the inhibitors for tilapia aromatase to prevent sex differentiation to female which will increase in the male tilapia population. In this study, the total RNA of female tilapia were extracted from brain and ovary and used to prepare the full-length of P450arom cDNA by molecular cloning. Then, the sequence of full-length P450arom cDNA was used to find the molecular properties of aromatase enzyme by bioinformatics analysis such as Compute pI/Mw, MEGA (version 6.0) and so on. The structure of aromatase and its inhibitors were constructed by molecular docking. In addition, the purified aromatase will use to determine the inhibitory efficiency of those inhibitors by measured the decreasing of enzymatic activity. The identification and characterization of brain aromatase (On450AromB) demonstrated that the open reading frame of the gene composed of 495 amino acid residues with predicted molecular mass of 56.33 kDa and pI of 7.22. Phylogenetic tree analysis and multiple sequence alignment of On450AromB showed sequence similarity to other animal aromatase family and grouped in the same clade with other fish aromatase that control sex reversal. The homology structure of On450AromB was created and used for screening the potential inhibitors. The results showed that the structures of inhibitors are similar to hormone, which is the substrate of enzyme, and mainly bind to the active site with hydrophobic interactions. Those inhibitors were ZINC01669828, ZINC01163259, ZINC01749571, ZINC23118752, ZINC23118770, ZINC01668253, ZINC03954311, ZINC19923695, and ZINC19794765. They will be used *in vitro* analysis of aromatase enzymatic activity to determine the efficiency of inhibition by these inhibitors. Finally, those inhibitors will be used *in vivo* analysis to reverse female to male tilapia during fry state.

Keywords: Nile tilapia; monosex tilapia culture; aromatase; bioinformatics analysis; molecular docking





Study of catalase promoters from *Chlamydomonas reinhardtii*

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Salinity is one of the main factors that affects the growth of plants. Free radicals, which are generated and accumulated by plants under salt stress, will damage the cells. Therefore plants must have mechanisms to protect cells by producing more antioxidant molecules as well as antioxidant enzymes. From our previous proteomic study revealed alterations of proteins in the salt adapted (SA) line compared to the wild type *Chlamydomonas reinhardtii*. One of the protein exclusively found in the SA cells was Catalase. Catalase converts hydrogen peroxide to oxygen and water thus helping the cells to cope oxidative environment. There are 2 catalase genes in *Chlamydomonas*, *CAT1* and *CAT2*. At transcription level, *CAT1* and *CAT2* are found to be up-regulated in the SA cells when compared to the wild type culture pointing out the control of gene expression occurred at this level. Thus, this research aimed to study the function of full length and deleted promoters of *CAT1* and *CAT2*. The full-length of *CAT1* with 638 bp was cloned into expression vector. For the *CAT2* promoter, 4 different constructs were generated with the following length of promoters; 473, 380, 283, 181 bp respectively. All of the promoters were used to drive expression of luciferase reporter gene. Transgenic *Chlamydomonas* lines carrying each construct were generated. The cultures of each lines were induced the salt-adaptation by culturing the cells in 300mM NaCl for few weeks. Expression of *LUC*, *CAT1* and *CAT2* was determined in these transgenic lines both under normal growth (salt-sensitive) and salt adapted conditions. The promoter of *CAT1*(638) show no significant alteration in expression of *LUC* and *CAT1* between normal growth condition compare with salt-adapted condition. But the *CAT2* expression was clearly induced in the SA cells. Similar to *CAT1* promoter, *CAT2* promoter and its deletions displayed no change in expression of *LUC*, *CAT1* and *CAT2* respectively.

Keywords: *CAT1*, *CAT2*, Catalase, *Chlamydomonas reinhardtii*, Deleted promoters, *LUC*, Salt stress.





Expression analysis of iron transporter genes under iron deficient conditions in *Chlamydomonas reinhardtii*

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Iron is an essential element of many proteins in plants. Plants have two major iron-utilizing pathways, photosynthesis and respiration which are very important pathways in plants. Iron transports and storage therefore are tightly controlled. Iron transporters are found in membrane and transports the ferric ion into cytosol including *IRT1*, *IRT2* and *FOX1*. The *FER1* and *FER2* encoding ferritin subunits are the major iron storage protein complexes in *Chlamydomonas*. In our previous study, the salt adapted (SA) *Chlamydomonas* strain CC-503 displayed increased in expression of many iron transporter genes. However, the effect of iron deficiency to the growth and gene expression in the SA have not yet been investigated, therefore this research aimed to study the growth of *Chlamydomonas* strain CC-503 both wild type and salt adaptation under iron deficient conditions. The cells were inoculated in modified TAP medium containing 0.1 μM , 0.25 μM , 1 μM , 17.5 μM FeSO_4 and normal TAP medium with 17.5 μM FeSO_4 . The growth of both strains did not effected by the varied concentration of iron though the slower growth rate has been found in the SA cells. Expression analysis of iron transporter genes and iron storage protein genes in SA and WT have been carried out. All of the genes displayed increased in the expression level under iron deficient conditions. Interestingly iron deficiency conditions strongly influences the expression of *IRT1*, *FER2* and *FOX1* genes in SA cells compared to wild type in medium containing same as iron concentrations. But the *IRT2* and *FER1* genes in SA was found to be similar to the wild type in medium containing the same iron concentrations. Thus, iron deficient conditions indeed can induce expression of iron transporter genes and effects of iron concentrations to their expression were differ in SA and WT. The higher in expression of iron transport proteins as well as iron storage proteins in the SA pointed out the importance of iron in this SA strain. Requirement of iron and the contribution of iron to the salt adaptation process will further be investigated.

Keywords: iron deficient, iron transporter genes, *IRT1*, *IRT2*, *FOX1*, *FER1*, *FER2*, *Chlamydomonas reinhardtii*





Expression and characterization of recombinant β -glucanase from *Bacillus* sp. C4 SS-2013

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β -1, 3-1, 4-Glucans are components of the cell walls of plants, fungus, yeast and bacteria. This enzyme belongs to the endogenous glycosyl hydrolases family 16 and plays an important role in the cleavage of β -1,3-1,4-glycosidic bond in 3-O-substituted glucopyranose units. In the previous report, the β -1,3-1,4-glucanase, isolated from *Bacillus* sp. C4 SS-2013, was successfully purified by several steps of FPLC columns.

In this study, the recombinant β -1,3-1,4-glucanase was cloned into pET32a(+) vector and expressed in *Escherichia coli* BL21 (DE3). It was successfully expressed in a soluble form at 16 °C with 0.1 M IPTG induction. The calculated molecular weight of recombinant glucanase as 31 kDa which was similar to the apparent molecular weight on SDS-PAGE. The recombinant glucanase was purified by HisTrap FF column and exhibited high activity with azobarley substrate. Characterization of the recombinant enzyme is in progress. The enzyme will be useful as biological control of some fungal in the field of agriculture.

Keywords: β -1,3-1,4-glucanase, *Bacillus subtilis*, protein expression, properties





The study of synergistic effect of antioxidative peptides extracted from rice bran

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Generation of excess reactive oxygen species (ROS) such as peroxy, hydroxyl, and nitric oxide radicals results in oxidized environments causing damages to cells. Several diseases such as coronary artery disease, cancers, and Alzheimer's disease have been reported as a consequent effect of excess ROS. Several natural antioxidants are such as vitamin A, C, and E. Recently, peptides from rice bran, a byproduct obtained from rice production, have also shown antioxidant activities. Consequently, peptide extracts from rice bran would add value to rice bran byproduct. Here, the aim of this study is to investigate synergistically antioxidative effects of antioxidative peptides extracted from rice bran on a variety of ROS. Three different peptides including FK13, KR10, and CK9, previously computationally predicted to have antioxidant activities, were chemically synthesized using a solid phase synthesis. Next, all three peptides were tested individually and in combination on four different antioxidant assays including DPPH, ORAC, DNA damage, and hydroxyl radical with salicylic acid assays. For individual peptide tests, each peptide possesses its own capacities in removing different ROS. In DPPH, CK9 was the best peptide in scavenging DPPH with IC_{50} value of $25\mu M$. In ORAC assay, The ORAC values of FK13, KR10, CK9 were 27, 37, 40 μM ascorbic acid equivalent/L respectively. So, the best peptide was also CK9. For DNA damage assay generated by hydroxyl radicals, FK13 rendered the best protection on the DNA plasmid. The results from hydroxy radical test using salicylic acid, is still being investigated. To investigate the synergistic effects from these 3 peptides, a variety of peptide combination was prepared and tested on the same antioxidant assays. The combination between FK13 and CK9 was the best in DPPH and ORAC assay. However, we did not see the synergistic effect of these peptide combinations at the concentration tested.

Keywords: reactive oxygen species, radical, peptide, rice bran, DNA damage, DPPH, ORAC, salicylic acid





Study of cytokine expression in breast cancer cell line treated with protein extracts from silkworm pupae

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A silkworm pupae is a by-product from silk drawing. Previous studies showed that silkworm pupae are a source of high-quality proteins, vitamins and fats. Proteins from silkworm pupae have been discovered to be involved in medical use such as reducing risk of Alzheimer's disease and decreasing blood lipids. Breast cancer is the most common cancer among Thai women and is the second leading cause of cancer death in patients. Inflammation is a potential leading cause of cancer. It also can induce secretion of cytokines that affect cancer cell formation and progression. This study therefore aimed to study effect of protein extracts from silkworm pupae on cytokine expression including interleukin-1 β (IL-1 β), interleukin-6 (IL-6) and tumor necrosis factor- α (TNF- α) in breast cancer cell line. Two species of silkworm pupae were used in this study, Thai native silkworm: Nanglai (mulberry silkworm, *Bombyx mori*) and Eri silkworm (non-mulberry silkworm; Eri silkworm, *Samia ricini*).

By performing ELISA, breast cancer cell line from human (MCF-7) and normal kidney cell line from monkey (Vero) were treated with crude protein extracts. The results showed that secreted IL-1 β and IL-6 were significantly decreased when compared with control. Likewise, cellular protein expression of IL-1 β and IL-6 were significantly decreased when treated with protein extracts. The results from real-time polymerase chain reaction (RT-PCR) showed that messenger RNA levels of TNF- α , IL-1 β , and IL-6 were significantly decreased when compared to the control. Interestingly, protein extracts from Nanglai exhibited more effective changes of cytokine expression than Eri. Moreover, protein extracts from silkworm pupae showed no significant effect on cytokine expression at any protein levels and mRNA levels in normal kidney cell line. In conclusion, protein extracts from Nanglai and Eri could decreased IL-1 β , IL-6 and TNF- α expression in breast cancer cells that could offer a possibility of treatment of breast cancer in the future.

Keywords: breast cancer, silkworm pupae, cytokine, protein extracts





Characterization of immune response genes in nuclear polyhedrosis virus (NPV) infected silkworm, *Bombyx mori*

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The sericulture induction is important agroindustry of Thailand, but many problems of agriculturist that can waste the silk product is silkworm diseases. The most severe disease is grassery disease. Grassery can destroy the silk product 30-50% or probably kill all silkworms. The grassery was caused by nuclear polyhedrosis virus (NPV) which was a genus of the family Baculoviridae. The symptoms of grassery will represent when the worm near death, therefore it is difficult to control disease. The understanding of silkworm immune system is necessary to develop techniques that can lower the lost from NPV. In this study, genes involve in immune response to NPV of silkworm were identified. RNA from normal and NPV infected silkworms will be extracted and converted to be cDNA. The gene expression will be carried out using reverse transcription PCR. The results of the study will help to understand the silkworm's response to viral infection leading to further protection.

Keywords: grassery, nuclear polyhedrosis virus, *Bombyx mori*





Initial Characterization of Cisplatin-resistant Nasopharyngeal Carcinoma Cell lines

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Nasopharyngeal carcinoma (NPC) is a cancer of a nasopharynx. A major problem of the treatment is a multidrug resistance. As a result, mechanisms involved in the multidrug resistance need to be investigated for the efficient treatment. Here, the aim of the study is to initially characterize cisplatin-resistant NPC cell lines for further studies on multidrug resistant mechanisms. In the study, NPC cell line (6-10B) was continuously treated with increasing concentrations of cisplatin, a well-known chemotherapeutic drug to develop cisplatin-resistant 6-10B (6-10B_{cisR}) cells. The half maximal inhibitory concentration (IC₅₀) values determined by MTT assay were used to confirmed the resistance of cisplatin on 6-10B_{cisR}. After increasing the concentration of cisplatin to 4 μ M, the IC₅₀ of 6-10B_{cisR} increased from 10 to 200 μ M compared with 6-10B. The 6-10B_{cisR} also demonstrated differences in cellular morphologies. Cell proliferation curves and doubling times of both cell lines were also compared. The results demonstrated that 6-10B_{cisR} exhibited a faster growth rate than 6-10B with doubling times of 20.19 and 21.53 hours, respectively. Next, differential expression of specific genes involved in multidrug resistance and oxidative stress were investigated by qPCR. The expression of multidrug resistance protein 1 (ABCB1), multidrug resistance-associated protein 1 (MRP1/ABCC1), ATP-binding cassette subfamily G member 1 (ABCG1) and ATP Binding Cassette Subfamily G Member 2 (ABCG2) of 6-10B_{cisR} were 30.13-, 2.28-, 2.67- and 2.85-fold higher than 6-10B, respectively. The oxidative stress-related genes including catalase (CAT) and manganese superoxide dismutase (MnSOD) also showed 1.34- and 2.10-fold higher differential expression in 6-10B_{cisR} compared to 6-10B. On the contrary, glutathione peroxidase (GPx) in 6-10B_{cisR} were relatively down-regulated with 0.68-fold expression. In addition, to further explore on cross-resistance, we found that the 6-10B_{cisR} cells also demonstrated greater resistance to another chemotherapeutic drug, lapatinib than 6-10B. As a result, we succeeded in initially establish and characterize 6-10B_{cisR} cell line by investigating its multidrug resistance, morphology, growth, and specific gene expressions.

Keywords: Nasopharyngeal carcinoma, cisplatin-resistant, multidrug resistance, ABC transporter, oxidative stress, cross-resistance





Effects of the D400N and D400N/S454F mutations in beta-glucosidase from Blackwood

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The soybean isoflavones, such as genistin and daidzin, can act as phytoestrogens, as they resemble human estrogen. Therefore, soybean isoflavones are beneficial to human health, such as prevention of osteoporosis, cancers, postmenopausal symptoms and coronary heart disease. Soybean isoflavones are mostly present as isoflavone glucosides, which are very poorly absorbed in the small intestine as compared with the free isoflavones forms. So, these isoflavone glucosides need to be cleaved by β -glucosidase to release free isoflavones before consumption. Three leguminous isoflavone β -glucosidases, which are Dnbglu2 from *Dalbergia nigrescens* Kurz (blackwood), dalcochinase from *Dalbergia cochinchinensis* Pierre (Thai rosewood) and GmICHG from *Glycine max* (soybean), share between 60-80% amino acid sequence identity, but show different kinetic properties. Dnbglu2 hydrolyzes daidzin and genistin with the highest efficiency, while GmICHG was most efficient in hydrolysis of soybean isoflavone glucosides with a modified group at the C-6 of position, such as malonylgenistin and malonyldaidzin. The objective of this study is to identify the amino acid residues that are responsible for their kinetic differences. So, in this study, we changed the amino acids located in the substrate binding pocket of Dnbglu2 to the same residues of GmICHG, generating single and double mutants, namely the D400N and D400N/S454F mutants. These mutant constructs were transformed into *Escherchia coli* (DH5 α) by electroporation and selected on media containing zeocin. The mutated sequences were confirmed by DNA sequencing before transformation into *Pichia pastoris* by electroporation. The mutants were expressed into culture media after induction with 0.5 % (w/v) methanol, and purified by using a phenyl sepharose column followed by a Ni²⁺ sepharose column. The purified proteins appeared as 66 kDa protein bands on a 10% SDS-PAGE. The kinetic results showed that hydrolysis efficiencies of both Dnbglu2 mutants decreased as compared with that of the wild-type Dnbglu2.

Keywords: isoflavone glucosides, mutantion, Dnbglu2, β -glucosidases





Expression and Purification of Recombinant SAL1 from Thai Aromatic Rice KDML 105

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Rice (*Oryza sativa* L.) is the staple food of the Thai people that is rich in carbohydrates. *Thailand* has long been one of the world's major producers and exporters of rice. The most popular aromatic rice for local in Thailand is Khao Dawk Mali 105 (KDML 105). *Rice growing, planting* and harvesting are popular in northeastern region of *Thailand*. However, the rice fields in this area have been severely damaged by drought. One of the enzymes, which responses to drought stress is SAL (Inositol polyphosphate 1-phosphatase) 1. Previous study in *Arabidopsis thaliana* demonstrated that SAL1 is a negative regulator of drought response. Moreover, SAL1 has been identified in other crops, including wheat and rice. This research therefore aimed to express and purify recombinant SAL1 from Thai aromatic rice, KDML 105. Firstly, recombinant plasmid of SAL1 was constructed in pET28b vector and introduced into *E.coli* strain BL21 (DE3) cells. Then, protein production was induced by 0.5 mM IPTG at 16 °C overnight. The recombinant protein was purified by HisTrap column with nickel bead. The protein band on SDS-PAGE appeared to be ~42 kDa. In addition, enzymatic assay was performed using PAP as a substrate. Taken together, the results from this study will contribute to precise understanding of the role of SAL1 protein in drought tolerance mechanism in rice plants.

Keywords: Rice (*Oryza sativa* L.), SAL1(Inositol polyphosphate 1-phosphatase) 1, PAP, Drought stress





Effects of the Y286A and Y492A mutations in beta-glucosidase from *Aspergillus niger*

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Bioethanol is produced by degradation of biomass materials into sugars followed by fermentation of sugars into ethanol. In particular, cellulose, a long-chain glucose polymer, is a useful starting material for bioethanol production, because it is not needed for consumption. Degradation of cellulose to glucose requires activities of many carbohydrate-active enzymes, including β -glucosidases, which catalyze the hydrolysis of β -1,4-glycosidic bonds in cellobiose. Our previous research have identified a β -glucosidase from *Aspergillus niger* (AnBG) with a high efficiency in degradation of cellobiose. Structural data of a β -glucosidase from *A. aculeatus* (AaBGL1), sharing 81% amino acid sequence identity with AnBG, showed that aromatic amino acids in the active site were important for substrate binding. So, in the present study, we chose to study the role of tyrosine at positions 286 and 492 in the active site of AnBG by substituting each of them with alanine, to produce the Y286A and Y492A mutants, respectively. These mutant constructs were transformed into *Escherichia coli* (DH5 α) by electroporation and selected on media containing zeocin. The mutated sequences were confirmed by DNA sequencing before transformation into *Pichia pastoris* by electroporation. Expression of the two mutants in *P. pastoris* cultures was induced by 0.5 % (w/v) methanol. Purification of the Y286A and Y492A mutants from the culture media was achieved by hydrophobic interaction chromatography. The purified proteins appeared as 130 kDa protein bands on a 10% SDS-PAGE. Kinetic studies of the two mutants revealed that the catalytic efficiency toward pNP-xyloside of the Y286A mutant was similar to that of the wild-type AnBG, while that of the Y492A mutant was lowered. Moreover, both mutants exhibited decreased catalytic efficiencies toward pNP-glucoside compared with the wild-type AnBG.

Keywords: *Aspergillus niger*, Bioethanol, β -glucosidases, Mutation





Construction and characterization of the deletion mutant of *SNF1* ortholog gene in *Yarrowia lipolytica*

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Yarrowia lipolytica is oleaginous yeast with the huge accumulation of lipid in cells. This yeast was widely used to study the production of biodiesel for an alternative energy. On the other hand, the control of lipid metabolism in *Y. lipolytica* has not been completely understood. Previous studies have reported that fatty acid metabolism of *Y. lipolytica* is controlled by the major transcription factors Por1p and Cfu1p. These major transcription factors regulate gene expression in beta-oxidation. However, the expression of genes in β -oxidation was remained in double deletion mutant of POR1 and CFU1 genes. This revealed that other proteins might control in this process together with Por1p and Cfu1p. To identify another factor, this research generated a deletion mutant of *YALI0D02101g* genes in *Y. lipolytica*. *YALI0D02101g* is an ortholog of Snf1p which plays roles in controlling various cellular processes, including catabolic repression in the yeast *S. cerevisiae*. The deletion cassette of the *YALI0D02101g* gene (101A cassette) was transformed into *Y. lipolytica* strain CXAU1. After that, the transformants were selected on selective medium and first-screened by the Colony PCR. The result showed that target product from transformant is 5.4 kb- DNA fragment, thereby indicating that this strain was the deletion mutant of *YALI0D02101g* gene. Next, Southern blot technique will be used to validate this mutant again. Finally, the growth of yeast mutants will be analyzed on media containing various carbon sources, such as glycerol, decane, hexadecane and oleic acid, to characterize the gene functions.





***In silico* nanobody engineering to improve protein-protein interaction for alternative cancer immunotherapy**

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Epidermal growth factor receptor (EGFR) is one of important protein causes cancer. The activation of EGFR owing to the biological consequences in human malignancies including increased cell proliferation and reduced apoptosis. Several drugs against EGFR were used for many types of cancer treatment but resistant is developed after long period of treatment. The immunotherapy is one of the efficiency remedy that can be an alternative treatment. Nanobodies are the choice because it is consisting of a single monomeric variable antibody domain with a full antigen binding capacity. This research aim to rectify and learn binding mode of nanobody against kinase domain of EGFR, V_HH18, which was identified in our lab by Bioinformatics techniques. The homology model of V_HH18 was built. The protein-protein docking was performed between V_HH18 and tyrosine kinase of EGFR. The complex structure was used as a starting conformation for further engineer of V_HH18 to make more efficiency. We discover V_HH 18 use CDR3 as a binding site and V_HH 18's mutation some amino acid can increase the efficiency of binding property between EGFR and V_HH 18. From the prediction V_HH18 flavors to binding in front of the EGFR's pocket site. It matches to the previously mimotope results from our laboratory.

Keywords: EGFR; nanobody; kinase domain; protein Bioinformatics





Inhibition of Melanogenesis and Antibacterial Property of Sericin Hydrolysate and Application

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Sericin is a globular protein comprising eighteen types of amino acid of which the majority is serine. The previous studies revealed that sericin is capable of inhibiting the activity of tyrosinase which is an enzyme responsible for melanogenesis in the skin. Many studies have also reported antioxidant and antibacteria properties of sericin. In this study, four sericin samples derived from different isolation methods were biochemically investigated. The protein and amino acid contents in the samples were quantified by biuret and ninhydrin assays. The results indicated that sericin samples: savinase enzyme, alkaline, protease from *Bacillus subtilis* C4/pH adjustment (pH6), and protease from *Bacillus subtilis* C4/pH adjustment (pH7) had protein content 12%, 21%, 6% and 6%, and the amino acid contents were 21%, 27%, 9% and 18%, respectively. Non peptide larger than 10 kDa was found in any samples when analyzed by SDS-PAGE, suggesting that the samples contain only small peptides and free amino acids. The result from HPLC analysis revealed a large variation of peptides in the samples. The capability of inhibiting melanogenesis was tested using mushroom tyrosinase and its substrate, L-DOPA. The result indicated that all four sericin samples can efficiently inhibit the tyrosinase enzyme. DPPH radical scavenging assay showed that all sericin sample also possessed antioxidant activity, in which savinase-derived sericin was the most potent antioxidant compared to the other samples. When testing the antibacterial property of sericin samples against *E. coli* and some species of bacterial organism isolated from human skin, the result indicated that savinase-derived sericin exhibit antibacterial activity to some extent, while the other sericin samples had no activity at all, and even promoted the bacterial growth. Therefore, savinase-derived sericin was the most suitable sample for developing skin cares in the future work.

Keywords: Sericin, Melanogenesis, Antibacteria, Antioxidant





Cloning and production of restriction endonuclease *Bgl*III for applications in biotechnology

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By exploitation of type II restriction endonucleases (*Eco*RI, *Bam*HI, *Nde*I, etc.) that recognize and cleave the specific DNA sequences, it is possible to create recombinant DNA molecules. Genes of interest can be inserted into a multiple cloning site of cloning vectors to increase the copy number of genes or expression vectors to express the gene into protein. *Bgl*III endonuclease (*Bgl*III) is the one of commonly-used restriction endonucleases. In order to produce *Bgl*III in large quantity, expression vectors (usually pET expression system) are used to overexpression of *Bgl*III. However, previous research that used pET expression vector to overexpress *Bgl*III was found to produce insoluble protein (inclusion body). This research will use pTTQ18 expression vector, instead of pET expression vector, to overexpression of *Bgl*III and develop the fluorogenic substrate to assay the *Bgl*III activity.

pTTQ18, which will be used as expression vector, was transformed to competent *E. coli* (DH5 α) cells by heat-shock and the plasmid was purified by alkali-lysis and precipitation with polyethylene glycol to increase the quantity of pTTQ18 for further experiments. *Bgl*III gene was amplified from *Bacillus globigii* chromosomal DNA by PCR reaction, using forward and reverse primers an added on with *Eco*RI and *Hind*III restriction sequences respectively. *Bgl*III gene then ligated to pGEM-T-easy vector and transformed to competent *E. coli* (DH5 α) cells by heat-shock and screen by blue-white selection. The presence of *Bgl*III gene in recombinant plasmid of the positive clone was confirmed by colony PCR. To ligate the *Bgl*III gene into pTTQ18, pGEM-*Bgl*III and pTTQ18 will digested with *Eco*RI and *Hind*III, then ligated with ligation reaction and transformed to competent *E. coli* (DH5 α) cells and confirmed with colony PCR. However, *Hind*III was found to loss of enzyme activity after prolonged storage. Therefore, the ligation of *Bgl*III gene to pTTQ18 couldn't be done and so the subsequent experiments.

Keywords: restriction endonuclease, cloning, expression, *Bgl*III, *E. coli*





Analysis of effective characteristic features of miRNAs involved in hepatitis B virus research

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MicroRNA is a small non-coding RNA molecule that containing about 22 nucleotides. It has been found to play an important role in the development of organisms and the regulation of cellular function and gene expression. Moreover, microRNA has a potential to develop a cancer therapies including infectious diseases such as Hepatitis B. At present, there are many programs that can predict microRNA structure. It uses similar prediction principles such as complementary pairing and thermodynamics. In this study was focused on microRNA of Hepatitis B virus by considering the complementary of MicroRNAs with their targeted molecules, thermodynamic, conservation and predict novel microRNA that binds to the Hepatitis B viral genome. By compiling the microRNA data that has been reported in Hepatitis B virus in 2010-2016 that be divided 2 type of target are Cellular and HBV and predict the structure found that most of the structure of microRNA are 5' dominant canonical. And compare energy and relation of MiRNA. Afterwards By doing sequence alignment using clustal omega and T-coffee programs, part 1 research do for want to know property of MiRNA that can resistance in Heptitis B viruses part 2 is create MiRNA that can resistance in Heptitis B viruses start from designed microRNA from compare microRNA Accession No. AM282986 Position 1186-1684. Then, perform sequence alignment by Clustal Omega and T-coffee the result found that Hepatitis B virus has Genotype A-J. Afterwards select target of MiRNA and predict structure. The result showed that this microRNA is 5' dominant canonical. This result is based on the properties of the microRNA structure that can withstand the hepatitis B multiple varieties. Overall, the result indicated that designated microRNA maybe able to be used in resistance in Hepatitis B viruses in the future.

Keywords: Hepatitis B virus, miRNA,





Screening of cyclic peptide inhibitor for reverse transcriptase of HIV-1

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The acquired immune deficiency syndrome is one of the deadly infectious disease spread around the world caused by infection with human immunodeficiency virus (HIV). This infection destroy white blood cell in the body result in the low body's immunity lead to another infection is easier than normal. As a result, AIDS patients receive an abundant pain and suffering. Presently, there are no medicine to cure, so all AIDS patients die with this disease. The proliferation of HIV virus requires a reverse transcriptase which is a enzyme to convert a RNA genomes into a DNA then incorporate into the host cells. For this reason, the invention of drugs that inhibit the reverse transcriptase occur, but the proliferation of virus is still there because HIV virus is resistant to drug. Therefore, the research and development of new anti-HIV drugs have been continuing, including looking for new techniques that will help in research and development of drugs to better efficiency. The one of the interesting technique is a phage display. The phage display is a selection technique in which peptides or proteins that display on the surface of viral particle can bind to target and inhibit target's activity. Thus, this research aimed to use the phage display technique for screening cyclic peptide that inhibit the reverse transcriptase of HIV-1. The result found that 30 samples could bind with reverse transcriptase. Then all 30 samples were tested for inhibiting activity of reverse transcriptase by using Reverse Transcriptase Assay Kit. There are only 9 samples which inhibit reverse transcriptase of HIV-1. In addition, PCR products of DNAs of cyclic peptides from 9 samples were sequenced and translated to amino acid sequences. There are 7 unique cyclic peptides. Consequently, these cyclic peptides may be used as a guideline for new anti-HIV drug development together with invention of vaccines that can protect HIV virus.

Keywords: human immunodeficiency virus (HIV), reverse transcriptase, phage display, cyclic peptide





Cloning, Expression, and Characterization of pH-responsive Protein Materials

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Hydrogel is a polymeric material that can store large amount of water and have been used in several clinical applications such as tissue engineering and drug delivery. Properties of hydrogel created from protein-based polymers can be designed to response to external stimuli such as pH of solution. The aim of this study was to produce pH-responsive polypeptide-based hydrogel by over-expression in *Escherichia coli*. Here, we designed two polypeptides with repetitive sequences of glutamic acid and valine residues flanked by four-residue turn. To construct the gene encoding these repetitive polypeptides, concatemerization was performed by the ligation of a synthetic monomeric DNA based on seamless cloning method. These concatamers were ligated into modified pET19b vector and then were transformed into *Escherichia coli* strain BL21(DE3) by heat shock method. The colonies were initially selected by ampicillin treatment prior to performing colony PCR. The constructs were analysed further by agarose gel electrophoresis and DNA sequencing. Protein expression was induced by isopropyl β -D-1-thiogalactopyranoside (IPTG). Polypeptides were identified by SDS-polyacrylamide gel electrophoresis (SDS-PAGE) and were purified using fast protein liquid chromatography (FPLC). The hydrogel properties were studied at different pH by table-top gelation. The results in this study will provide some information and utility for investigation of protein-based polymer in future applications.

Keywords: Concatemers, Hydrogel, Polymers, Protein





Optimum condition for sericin degrading with recombinant protease from *Bacillus subtilis* C4

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Sericin is a glue protein derived from the silkworm's protein filaments. Sericin is composed of amino acids that are close to the type of amino acids that constitute the human body, so sericin can be used in a variety of industries such as cosmetics, medicine, drugs apart from being used in research. The application of sericin depends on size. The sericin that is smaller than 20 kDa is used as a cosmetic ingredient. This research was aimed at finding optimal conditions for degrading sericin with recombinant protease from *Bacillus subtilis* C4.

The protease activity was determined by using sericin as a substrate by o-phthalaldehyde (OPA) method. The percentage of sericin hydrolysis was determined by OPA method. The optimum condition for degrading sericin protein was 2% substrate, the recombinant enzyme 20 U (substrate to enzyme ratio of 1:50), at 50 °C, and pH 8.0 for 4 h. The sericin peptides will be further purified by HPLC technique and the biological properties of peptides will be examined.

Keywords: sericin peptide, protease, o-phthalaldehyde, properties





Cloning and expression of *CFU1* gene in *Yarrowia lipolytica*

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Currently, the production of renewable energy, especially biodiesel from renewable resources such as vegetable oils, animal fats may not be sufficient to future demands. Then, biodiesel production from oleaginous microorganisms has been an interesting alternative and widely study both domestically and internationally. *Yarrowia lipolytica* is one of the most extensively studied “non-conventional” yeasts that able to accumulate large amounts lipids in form of lipid body to levels exceeding 50% of cell dry weight. Therefore, it has been studied of this yeast species as a model for generate of renewable energy sources in the future. However, the control of lipid metabolism in *Y. lipolytica* has not been completely understood. From previous studies, the transcription factors, Por1p and Cfu1p, are involved in the regulation of the fatty acid metabolism in *Y. lipolytica*. Hence, this research will construct *Y. lipolytica* with *CFU1* protein that can track location with green fluorescent protein (GFP). First, the two DNA fragments of *CFU1* gene, the first is 4.5 kb- fragment containing the promoter, 5' UTR and core gene of *CFU1* gene. Another is 1 kb-fragment containing 3'-UTR and terminator of this gene, were amplified by polymerase chain reaction (PCR) with specific primers. After that cloned into pGEM-T Easy vector by TA cloning technique and subcloned the 4.5 kb and 1 kb gene fragments of *CFU1*, obtained by cleavage with restriction enzyme from the pGEM-T easy vector, into pSUT5 by welded into position EcoRI and XbaI of the plasmid pSUT5 with the enzyme T4 ligase. Next, insert Entrance Green fluorescent protein (EGFP) between 4.5kb and 1kb *CFU1* gene in the position NsiI and StuI. When cloning of *CFU1* gene fragments and insertion of EGFP gene was successfully. In the next step, plasmids were transferred to *Yarrowia lipolytica* yeast by electroporation. Finally, the localization analysis of *CFU1* gene was investigated within *Y. lipolytica* yeast fed with different carbon sources by fluorescence microscope.

Keywords: *Yarrowia lipolytica*, *CFU1*, oleaginous microorganisms, biodiese





Effect of UV-C Irradiation on Growth and Lipid Accumulation of *Chlorella* sp.

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Microalgae are an alternative energy source for biodiesel production because they have an advantage of using less area to culture. Microalgae can be cultivated in shorter time than other oil crops and their oil can be produced throughout the year. This research aimed to study the growth and lipid accumulation of green microalgae, *Chlorella* sp., after exposed to UV-C for 60, 180, 300 and 420 seconds. The treated *Chlorella* sp. was cultured in liquid TAP medium for 13 days. The study was divided into 3 parts. 1) The effect of UV-C on growth of *Chlorella* sp. showed that UV-C significantly reduced the growth when compared to the control unit. 2) The effect of UV-C on the lipid accumulation showed that in the late stationary phase of growth, the lipid content of *Chlorella* sp. tend to be increased after exposed to UV-C for 60 and 420 seconds. 3) Triacylglycerols (TAGs) were found in all treatments of *Chlorella* sp. when determined with thin-layer chromatography (TLC) by using soybean oil as the standard.

Keywords: Microalgae, *Chlorella*, Lipid, UV-C radiation





Fractionation, Isolation and Purification of Active Compounds of *Flacourtia indica* (Burm.f.) Merr.

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Flacourtia indica (Burm.f.) Merr. is a tree in the family Salicaceae, found in the deciduous dipterocarp forest. This plant has been used as a medicinal plant for a long time. In this study, we isolated fractions from the *Flacourtia indica* stem crude extract and tested for its anti-HIV activities. Crude chloroform extract was fractionated with chromatography. The fractions were investigated with Thin Layer Chromatography (TLC). The fractions with similar TLC profiles were combined. Total of seven combined fractions were obtained and tested for their anti-HIV activities. Two fractions showed about 80% and 50% inhibition against HIV-1 RT. For further isolation we chose one combined fraction to fractionate with chromatography. Four potentially pure compounds were obtained and structure elucidated will be further analyzed.

Keywords: Anti-HIV Activities, *Flacourtia indica* (Burm.f.) Merr., Isolation





Evaluation of Antioxidant Activities of *Homolium tomentosum* & *Hymenodictyon orixense* Fractions

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The present study was aimed at investigating the antioxidant activities of the various fractions of the wood of *Homolium tomentosum* and the bark of *Hymenodictyon orixense*. Fractions were evaluated for total phenolic and total flavonoids content. Antioxidant activities of fractions were determined by using DPPH test and Ferric reducing antioxidant power assay. *Homolium tomentosum* extract was fractionated into 15 fractions. For *Homolium tomentosum*, total phenolic content of 15 fractions, was found to be 23.60 – 50.45 mg of GAE/g. Fraction no. 15 had the highest amount of phenolic content. Total flavonoids content of 15 fractions, was found to be 392.25 – 417.94 mg of QE/g. Fraction no. 15 had the highest amount of flavonoids content. DPPH, expressed in term of IC₅₀, was found to be 67.05 – 236.65 ug/ml. Fraction no. 14 showed the highest activity with IC₅₀ value 67.05 ug/ml. Ferric reducing antioxidant power, was found to be 83.75 – 89.08 mg TE/g. Fraction no. 4 showed the highest activity with the value 89.08 mg of TE/g. *Hymenodictyon orixense* extract was fractionated into 5 fractions. For *Hymenodictyon orixense*, total phenolic content of 5 fractions, was found to be 22.00 – 32.89 mg of GAE/g. Fraction no. 4 had the highest amount of phenolic content. Total flavonoids content of 5 fractions, was found to be 316.76 – 323.63 mg of QE/g. Fraction no. 2 had the highest amount of flavonoids content. DPPH, expressed in term of IC₅₀, was found to be 192.76 – 306.24 ug/ml. Fraction no. 4 showed the highest activity with IC₅₀ value 192.76 ug/ml. Ferric reducing antioxidant power, was found to be 83.25 – 88.08 mg of TE/g. Fraction no. 5 showed the highest activity with the value 88.08 mg TE/g. In conclusion, fraction no. 14 and 15 of *Homolium tomentosum* and fraction no. 2 and 4 of *Hymenodictyon orixense* have high value of antioxidant activities. These fractions may contain interesting bioactive compounds. So, these fractions should be focused to isolate and identify for further drug developing.

Keywords: *Homolium tomentosum*, *Hymenodictyon orixense*, Total phenolic content, Total flavonoids content, Antioxidant activities





Anatomy of some woods in the family Dipterocarpaceae in Thailand

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Dipterocarpaceae woods are major material for wood industry in Thailand. Wood timbers and wood products are utilized in the country and export. Ten wood species belong to the family Dipterocarpaceae were collected and used in this study. The wood samples were prepared into permanent slides using a sliding microtome technique and a cell maceration method. It was found that all species have growth ring boundaries indistinct with diffuse-porous wood. Vessels arrange in a diagonal pattern. There are 3 patterns of intervessel pit arrangement including scalariform, alternate and opposite. There are 4 patterns of axial parenchyma distribution including vasicentric, winged-aliform, confluent and diffuse apotracheal. Multiseriate rays were observed in all species. Fibers are libriform with very thick – walled except *Shorea leprosula* Mig. which has thin to thick – walled fibers.

Keywords: Anatomy, Anatomy of some woods, Dipterocarpaceae





Comparative Phytochemistry of *Salix tetrasperma* Roxb. and It's bioactivity

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Comparative Phytochemical analyses of *Salix tetrasperma* Roxb. (Salicaceae) was conducted during September 2015 to March 2016. The lipophilic extracts of 3 plant parts i.e. leaves, stem bark and root bark were analyzed by means of Thin Layer Chromatography (TLC) and High Performance Liquid Chromatography (HPLC). The chemical profiles of stem bark and root bark are similar, but different from the chemical profile of leaves. From Phytochemical screening test, coumarins and alkaloids could be detected in stem bark and root bark. However, terpenoids were found in all 3 plant parts. Antibacterial activity of lipophilic extracts from 3 plant parts was screened with 8 digestive track bacteria i.e. *Escherichia coli* (*E. coli* O157:H7), *Serratia* sp., *Enterobacter* sp., *Staphylococcus aureus*, *Listeria monocytogenes*, *Salmonella typhi*, *Shigella flexneri*, *Vibrio parahaemolyticus*. It was found that the lipophilic extracts from stem bark and root bark could inhibit *Serratia* sp. at the concentration 80.20 mg/ml with 0.82 cm clear zone, and the lipophilic leaves extract could inhibit *Staphylococcus aureus* at the concentration 264.80 mg/ml with 0.85 cm clear zone. The anti-Alzheimer disease was screened by anticholinesterase activity test, but the results showed less than 10% inhibition. Thus, these *Salix* extracts are not appropriate for drug development against Alzheimer.

Keywords : *Salix tetrasperma*, Comparative Phytochemistry, bioactivity





Effect of Polyethylene Glycol on Carotenoid Accumulation of Some Microalgae

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Study on the effect of polyethylene glycol (PEG) at concentrations 5, 10, 15 and 20% (w/v) on growth and carotenoid content of green microalgae which was cultured in liquid TAP medium. The growth of the algae was significantly inhibited by 15 and 20% PEG treated cells after 20 days of treatments. The carotenoid contents were significantly less than the control but likely to increase. Beta-carotene was found in the algae when determined with thin-layer chromatography (TLC) technique by using beta-carotene as standard.

Keywords: Microalgae, Polyethylene glycol, Carotenoid, TLC





Effect of Chemical Fertilizer 18-6-12 on Growth of Holy Basil

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The study of chemical fertilizer 18-6-12 on growth of Holy Basil was done. The experiment design was completely randomized design (CRD) with 5 treatments as followed 1) no chemical fertilizer (Control) 2) chemical fertilizer 10 g per water 20 L spray every week 3) chemical fertilizer 20 g per water 20 L spray every week 4) chemical fertilizer 10 g per water 20 L spray two week intervals and 5) chemical fertilizer 20 g per water 20 L spray two week intervals. The duration of experiment was 8 weeks. The results showed that chemical fertilizer 20 g per water 20 L spray every week and chemical fertilizer 10 g per water 10 L spray two week intervals tended to have the highest relative growth rate from week 4th to week 8th. Consideration to SPAD-value all chemical fertilizer application tended to maintain higher leaf greenness than control, especially in week 4th and week 6th. Chemical fertilizer had significantly leaf greenness than control. Consideration to leaf branching chemical fertilizer 20 g per water 20 L spray two week intervals tended to have the highest number of new shoot. The results indicate that application of chemical fertilizer could increase the growth of Holy Basil by increase relative growth rate, increase shoot number and maintain high leaf greenness.

Keywords: chemical fertilizer, growth, Holy Basil





Lichens Richness Varies with Host Tree Size and Species

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Lichens are the one of the most unique organisms. However, there are relatively a few studies about them. In the present, most of the studies about lichens are from the temperate zone, and most studies in Thailand are about diversity, morphology, and physiology. Only a few studies are on ecology, especially the effect of the host trees on lichens. To find the relationship between host tree characters and lichens richness, we surveyed epiphytic lichens on 218 trees of 5 host species in the area of Peninsular Botanical Garden Khao Chong, in the permanent plot of Center for Tropical Forest Science. We found a total of 29 species from 15 genera of lichens. With ANOVA and Regression, we found that lichens species richness was lower on trees with rough barks and larger diameter. All these results could be references for the future studies about lichens ecology.

Keywords: Lichen, Tree size, Tree species





Qualitative Analysis of Jerusalem Artichoke (*Helianthus tuberosus* L.) and It's Bioactivity

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Qualitative analysis and bioactivity test of Jerusalem Artichoke (*Helianthus tuberosus* L. family Asteraceae) were conducted by chromatographic techniques during September 2016 to March 2017. One of Jerusalem Artichoke product from Petchaboon research station, Faculty of Agriculture, Kasetsart University was in freshly rhizome and another product was purchased from the market as Jerusalem Artichoke tea. By mean of chromatographic techniques, the chemical profiles of lipophilic extracts from both samples were analyzed by High Performance Liquid Chromatography (HPLC) and they were resemble. From Phytochemical screening test using Thin Layer Chromatography (TLC) technique, terpenoids and coumarins could be detected in both samples, but their chemical profiles were rather dissimilar. Alkaloids were found on the starting lines of both samples, so it could not be compared with each other. The results from TLC reveals that the process of tea preparation affected to chemical compounds in Jerusalem Artichoke. Antibacterial activity of lipophilic extracts from both samples was screened with 8 digestive track bacteria i.e. *Escherichia coli* (*E. coli* O157:H7), *Serratia* sp., *Enterobacter* sp., *Stephylococcus aureus*, *Listeria monocytogenes*, *Salmonella typhi*, *Shigella flexneri* and *Vibrio parahaemolyticus*. It was shown that the lipophilic extracts from Jerusalem Artichoke rhizome and Jerusalem Artichoke tea at the concentrations 244.4 mg/ml and 391.9 mg/ml respectively could inhibit *Serratia* sp. and *Stephylococcus aureus*. The anti-Alzheimer disease was screened by anticholinesterase activity test, but the results showed less than 10% inhibition. Thus, both extracts are not appropriate for drug development against Alzheimer.

Keywords: Jerusalem Artichoke, Qualitative Analysis, Bioactivity





Comparative Anatomy of *Lumnitzera littorea* (Jack) Voigt Grown in Saltwater and Freshwater

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Lumnitzera littorea (Jack) Voigt, a mangrove species belongs to family Combretaceae. It's medium to large tree with fissured bark. It has knee root. *L. littorea* distributes in South Asia, South East Asia and Malay Archipelago. It is used as ornamental plant. Interestingly that *L. littorea* can grow in both salt and freshwater. So this study, we aim to investigate and compare anatomical characters of *L. littorea* which grows in different habitats. In this study, root, pneumatophore, shoot tip, stem, leaf, petiole and flower of *L. littorea* were observed. It was found that the anatomical characters of *L. littorea* which grows in different habitats are not different. We found druse crystals and tannin in all of the structure. We conclude that saltwater and freshwater are not affect anatomical characters. Therefore, *L. littorea* can grow in fresh water normally. However, this study has shown anatomical characters of *L. littorea* thoroughly, and can be used for identification of *L. littorea*, or for baseline data in botany.

Keywords: Anatomy, Comparative anatomy, *Lumnitzera littorea* (Jack) Voigt





Pollination and Fruit Set in Durian (*Durio zibethinus* Murr.) cv. “Monthong” by Cross- Pollination

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Thailand is a major producer and exporter of durian in the world. However, there are still have some production problems such as low fruit set and fruit quality is not as good as it should be. This experiment was aimed to investigate pollen tube growth in style and fruit set in durian ‘Monthong’ cultivar by open pollination, self-pollination, cross-pollination by using pollen of durian ‘Kradum Thong’ and ‘Phaung Manee’. The results showed that of pollen tube growth reached stigma within 1 hour and reached the base of the style within 12 hours. The amount of pollen tube in self-pollination, cross-pollination by using pollen of durian ‘Kradum Thong’ and ‘Phaung Manee’ were higher than open pollination. After 56 days of pollination, the fruit set in self-pollination and cross-pollination by using pollen of durian ‘Kradum Thong’ and ‘Phaung Manee’ tended to better than open pollination. This experiment indicated that self-pollination, cross-pollination by using pollen of ‘Kradum Thong’ and ‘Phaung Manee’ tended to increase fruit set in durian cv. ‘Monthong’ than open pollination.

Keywords: Durian ‘Monthong’, Fruit set, Pollination, Pollen tube growth reached stigma





Morphology and Pollen Morphology of the Genus *Dolichandrone* (Fenzl) Seem. (Bignoniaceae) in Thailand

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Morphological study of the genus *Dolichandrone* (Bignoniaceae) in Thailand was conducted from March 2016 to March 2017. Three species, *Dolichandrone columnaris* Santisuk, *D. serrulata* (Wall. ex DC.) Seem, and *D. spathacea* (L. f.) Seem. are enumerated. Morphological descriptions, distribution and ecological information are provided. The differences based on morphological characters are leaflet margin, comparison between length of lower cylindrical tube and upper campanulate tube, width of upper campanulate tube, winged seed character, fruit shape and character, width of septum, character and width of pseudoseptum. The distribution of three species are different, *D. columnaris* occurs only in the peninsular region. *D. serrulata* occurs in the eastern, central and peninsular regions. *D. spathacea* occurs in the central, south-eastern and peninsular regions. Pollen morphology of these three species were studied. The result showed that the pollen morphological data do not provide characters for identification within the genus.

Keywords: *Dolichandrone*, Bignoniaceae, morphology, pollen





Effects of Organic Additives on *In Vitro* Shoot Regeneration of *Dendrobium spectabile* (Blume) Miq.

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Dendrobium spectabile (Blume) Miq. is a beautiful orchid with distinctive characteristics, red spot sepals, petals and lip and have high economic value. The aim of this study was to investigate the effect of organic additives (banana and potato) on shoot regeneration of *D. spectabile*. The treatments comprise of modified Vacin and Went (VW) media supplemented with banana pulp and/or potato extract at different concentrations and cultured for 12 weeks. The result showed that the best formulation for shoot and root multiplication was modified VW media supplemented with 50 g/l potato extract, which produce 4.00 shoots and 12.40 roots, respectively. The longest root was 5.52 mm when cultured on media supplemented with 150 g/l potato extract. The results from this studied indicate that the suitable media for shoot multiplication was modified VW media supplemented with 50 g/l potato extracts.

Keywords: *Dendrobium spectabile*, Modified VW media, Banana, Potato





Evidence of pig origin of the Ban Dung archaeological site at the ancient city of Udon Thani by the analysis of molecular techniques

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Pig is a popular domestic animal for food. Pig domestication occurred several thousand years ago. Thailand was one of the centers of pig husbandry, but the beginning of pig husbandry in Thailand is not clear. Therefore, we aimed to study of the origin of domestic pigs from Ban Dung archaeological site, Ban Dung District, Udon Thani. After we got pig's bones, the specimens were cleansed using 10% Clorox and rinsed with sterilized water twice. Then they were exposed to UV for 30 min and ground into fine powder. The bone powder was DNA extracted using a silica-based method. Then, a partial control region in mitochondrial DNA was amplified by polymerase chain reaction (PCR) technique. The PCR product was checked by agarose gel electrophoresis. In the gel, there was a 179-bp DNA band indicating that we can extract DNA from the ancient bone. In the future, the PCR produce will be purified and sequenced. Subsequently, the sequence will be analyzed using blast program and phylogenetic analysis.

Keywords: domestication, Ban Dung archaeological site, mitochondrial DNA, polymerase chain reaction (PCR)





Peptidomes and Proteomes of gastrointestinal tracts, testes, ovaries and stem cells of abalone *Haliotis diversicolor*

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Variously colored abalone (*Haliotis diversicolor*) is one of the highly-valued marine snails. Fully integrated abalone farming structures have been operated in the southern Thailand. Thousands of abalones are farmed and processed to various healthcare, cosmetics and medical products. The leftover of breeding and meat processing (i.e. gastrointestinal (GI) tracts, testes, ovaries, and embryonic stem cells) have not been effectively used. Therefore, this study aimed to optimize protein extraction methods for abalone embryonic stem cells and visceral mass (GI tracts, testes, and ovaries) by comparing TCA/acetone and RIPA buffer extractions. Protein yields were quantified by Lowry assay. Peptidomes and proteomes of these samples were separated in triplicate and compared using 1D-SDS-PAGE. Results showed that the RIPA buffer extraction was a more effective method than the TCA/acetone extraction. Gel analysis showed complex and distinct band patterns. The bands will be further identified by LC-MS/MS and analysed by bioinformatics techniques. The results provide basic knowledge and will potentially be useful for the future commercial application.

Keywords: Abalone, *Haliotis diversicolor*, Peptidome, Proteome





Characterization of HL-tolerant mutants of *Chlamydomonas reinhardtii*

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Chlamydomonas reinhardtii is a unicellular green alga that is used as a model organism for biological research especially in the areas of photosynthesis and oxidative stress. One advantage of using *C. reinhardtii* is its fast generation time. Lately, it has also been used as a model organism to study microalgal biofuel production. Acclimation to oxidative stress may be involved in biofuel production, because biofuel tends to accumulate under environmental stress conditions. Here, this study used high-light tolerant mutants from the *Chlamydomonas* CAL collection. These mutants were characterized under oxidative stress conditions. Growth of these mutants was compared to wild-type strain. CAL028_01_28 and CAL034_01_48 were found to grow better than the wild type. These strains were also resistant to H₂O₂ at 1 mM concentration whereas the wild type was not. The tolerance of these mutants might be caused by an increase in antioxidant levels. Oxidative stress tolerance of these high-light tolerant mutants may be used to maximize biofuels and bio-product yields. Therefore, we could utilize environmental stresses such as wastewater to improve biofuel productivity.

Keywords: *Chlamydomonas reinhardtii*, oxidative stress, biofuel





Isolation of leaf variegated genes from *Dendrobium lindleyi* Steud (Orchidaceae)

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Orchid is one of the most popular Thailand's export product with an export value of up to 3,000 million baht per year. *Dendrobium lindleyi* is one of the famous orchid species due to their beautiful flowers and sweet honey-like aroma. Not only they have been used as ornamental plant, their flowers have also been used to make perfume and tea. In this research, we aim to identify genes involved in the control of variegated leaf phenotype in *D. lindleyi*. Identification of the leaf variegated genes in orchids will facilitate the improvement of desire traits and thus increase the value of the plants. To identify the genes involved in variegated phenotype, the NCBI database was searched for leaf variegated gene orthogues from other species. Five nuclear genes, *VAR1* (*Yellow variegated1*), *VAR2* (*Yellow variegated2*), *IM* (*Immutans*), *CHM* (*Chloroplast mutator*), and *SWL 1* were identified. RT-PCR was performed to identify these genes in *D. lindleyi* using specific primers designed from sequences obtained from the database. Only *VAR1* and *VAR2* partial sequence were successfully amplified using the RT-PCR. However, the *VAR1* sequence matched to the mitochondrial *FTSH* gene with 91% identity. *VAR2* matched to the mitochondrial *FTSH2* gene with 91% identity. We aimed to isolate the genes from the nucleus of *D. lindleyi*, but only the genes from mitochondria were amplified. This could probably be because the primers designed for these two genes also have a conserved region in the mitochondrial *FTSH* genes and mitochondrial DNA is more abundant than a nuclear DNA in the extracted DNA solution. In the future, new set of primers will be designed to increase amplification specificity.

Keywords: *Dendrobium lindleyi*, leaf variegation





Transformation of a dual reporter plasmid (pSRG) into pig isolates of *Pasteurella multocida* in Thailand

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Pasteurella multocida is a Gram-negative bacterium and a causative agent of infectious diseases in various birds and mammals. Roles of this bacterium in complex respiratory diseases, particularly pneumonic pasteurellosis in pigs, remains poorly understood. This study aimed to transform a previously-published dual reporter plasmid (pSRG) into porcine isolates of *P. multocida* in Thailand. This plasmid contains genes coded for a red fluorescent protein (RFP) controlled by a prokaryotic promoter, a green fluorescent protein (GFP) controlled by a eukaryotic promoter, and kanamycin resistant protein. The pSRG plasmid was successfully transformed into the competent porcine isolates using electroporation. The transformed bacteria could grow on brain and heart agar (BHIA) supplemented with kanamycin, while the wild type was not. Extracted DNAs of the transformed and normal isolates were confirmed by agarose gel electrophoresis and showed two bands of genomic DNA and plasmid only in the transformed isolate. These transformed isolates of *P. multocida* will be useful for further genetic characterization and *in vivo* experiments to understand cellular mechanisms of this bacteria.

Keywords: *Pasteurella multocida*; Reporter plasmid; Plasmid transformation





Investigation of nucleocapsid expression in *E.coli*

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Porcine epidemic diarrhea virus (PEDV) is an enveloped virus that causes severe diarrhea, vomiting and death in swine of all age. PEDV encodes four structural proteins such as, spike (S), nucleocapsid (N), envelope (E) and membrane (M). Nucleocapsid protein is an important protein of the PEDV. The aim of this study was to investigate the conditions that could change the recombinant N protein from insoluble to soluble form. The expression of N protein was analyzed. The recombinant N protein was expressed in *E.coli* under IPTG induction. The cell pellet was collected and suspended in 1XPBS buffer. The cell suspension was treated by lysozyme and left on ice at different time points before sonication to lyse the cells. The supernatant and the inclusion body fractions of recombinant N protein were identified using SDS-PAGE. The result showed the longer time that the sample was left on ice, the higher amount of soluble protein was obtained.

Keywords: PEDV; Nucleocapsid; Protein expression





Protein Stability of Vegetative Insecticidal Protein and PEDV Nucleocapsid Protein

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Protein stability is the net balance of forces in a protein, which determine whether it will be in native folded conformation or a denatured state. The net stability of a protein is defined as the difference in free energy between the native and denatured state. In this study, we investigated the stability of two recombinant proteins, Vegetative insecticidal protein (Vip3A) and PEDV nucleocapsid protein (N). These two recombinant proteins were expressed in *E.coli* under IPTG induction. Vip3Aa and N were then purified by affinity chromatography. The purified Vip3Aa was mixed with the various concentrations of denaturant, guanidine hydrochloride (GuHCl). The emission fluorescence spectra of each mixture were determined using the excitation wavelength at 280 nm. The native and denatured states of Vip3Aa gave the emission maximum wavelengths at 336 nm and 346 nm, respectively. This result will be calculated to obtain the difference in free energy between the native and denatured state which will reveal the stability of Vip3Aa. The N protein will be investigated by the similar methods to acquire its stability.

Keywords: Vegetative insecticidal protein, *PEDV*, Protein stability





Identification of microflora on facial skin of monozygotic twins and dizygotic twins

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Normal flora are local microbes living on human's skin. The microbes do not cause any disease and they protect our skin from infectious organisms. Species and amount of microbes rely on either genetic background or environmental factors. In this study, we examined bacterial species colonized on facial skin of monozygotic twins and dizygotic twins. Firstly, facial bacteria were swabbed from both twins and cultured in TSA medium. Secondly, numbers of bacterial colonies were counted and colonies with different characteristics such as color and size, were selected for identifying bacterial species. After that, the bacterial colonies were used as templates in polymerase chain reaction (PCR) and a partial 16s rRNA gene was amplified by universal primers. PCR products were purified and sequenced. Then, bacterial species were identified using blast program. The results showed that both monozygotic twins and dizygotic twins have the same and different bacterial species. The same bacteria on facial skin of monozygotic twins and dizygotic twins were *Staphylococcus capitis* and *Staphylococcus epidermidis*, respectively. The different bacteria on facial skin of the monozygotic twins were detected. *Bacillus firmus*, and *Staphylococcus epidermidis* were isolated from one of the monozygotic twins. In the case of the dizygotic twins, *Enterobacter aerogenes*, *Staphylococcus capitis* were found in older twin and *Staphylococcus hominis* was found in younger twin. In conclusion, the environment seemed to have more effect on facial bacterial diversity than genetic background.

Keywords: microflora, facial skin, monozygotic twins, dizygotic twins





Detection and genotyping of Dengue virus in *Aedes aegypti* mosquitoes by partial NS5 gene

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Dengue disease is a mosquito-borne infectious disease which is caused by dengue virus (DENV) and *Aedes* mosquito is the vector. Dengue disease is widespread in the tropical and subtropical regions, including Thailand. DENV is a member of genus *Flavivirus* in the family *Flaviviridae*. There are 4 serotypes of DENV: DENV-1, DENV-2, DENV-3 and DENV-4. DENV genome is approximately 11 kilobases in length of a single stranded RNA which encodes three structural proteins (capsid [C], premembrane [prM], and envelope [E]) and seven nonstructural proteins (NS1, NS2A, NS2B, NS3, NS4A, NS4B, and NS5). NS5 is the largest nonstructural protein which functions in four activities necessary for cap synthesis, RNA dependent RNA polymerase (RdRp), interleukin-8 induction (IL-8) and nuclear localization. In this study, we aimed to identify serotype and genotype of DENV infected in field caught *Aedes aegypti* mosquitoes. The mosquitoes were collected from endemic areas of dengue disease in Thailand. Fourteen samples of *Aedes* mosquitoes were collected from Bangkok, Chanthaburi, Chasengsao, Lop Buri, Pathum Thani, Nakhon Ratchasima, Nonthaburi, Songkhla, Suphanburi and Trat Provinces. RNA was extracted using xxxxxx and cDNA was synthesized by xxxxxx. Then, the cDNA was used as a template in polymerase chain reaction (PCR). The PCR product was visualized by gel electrophoresis and stained with ethidium bromide. The results showed that the partial NS5 gene of two samples from Chanthaburi and Lop Buri were amplified. Subsequently, they were sequenced and the blast results showed that DENV in the mosquitoes collected from Chanthaburi and Lop Buri were the NS5 gene of DENV-4 and DENV-3, respectively. Next, DENV genotype will be examined using phylogenetic analysis.

Keywords: Dengue, *Aedes aegypti*, serotypes, NS5 gene





Ligation Independent Cloning of *Carboxypeptidase B* gene

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Malaria is one of the epidemic diseases in Thailand, especially on the border areas. The malaria transmission can be inhibited by transmission blocking compounds to disrupt the function of *Anopheles* proteins. CarboxypeptidaseB (CPB) is an important protein for *Plasmodium* development in an *Anopheles* midgut. Therefore, CPB is a potential protein target for blocking. The aim of this study is to clone the *cpb* gene into 2 expression vectors, pET28a vector and pH6HTC His6HaloTag T7 vector, by using ligation independent cloning (LIC). The synthesized *cpb* gene was amplified using forward and reverse primer that contained partial sequences of the vector and insert. The digested vector and insert are mixed with T4 DNA polymerase and incubated at room temperature. For annealing, the reaction mixture was placed on ice and then transformed into *Escherichia coli*. However, the cloning of CPB is still unsuccessful. When the *cpb* gene is successfully cloned into the expression vectors, the next step will be expression and purification. The small compounds will be screened against the CPB to discover the potential drugs that could block the malaria.

Keywords: *Carboxypeptidase B*, Ligation independent cloning (LIC), Malaria





Comparison of Methods of Preparing Fossil for DNA Extraction

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The study about DNA from ancient bone often has plenty of problems, especially DNA degradation. Therefore, the DNA extraction should be done carefully with considering all the factors that cause the DNA degradation. The objective of this study is to compare the efficiency of two methods of preparing sample from ancient bone and an evaluation of the usefulness of these methods for the study of ancient DNA. The bone samples were ground into fine powder by two different tools: a pestle and mortar and a mini-drill. Then, the bone powder was DNA extracted using a silica-based method and amplified by polymerase chain reaction (PCR) technique. After that, the PCR product was checked by gel electrophoresis and spectrophotometry. The efficiency of each sample preparation method was evaluated with PCR product concentration obtained from the same extraction and amplification method. And the difference efficiency between two methods will help us to choose better method for ancient DNA study.

Keywords: DNA degradation, DNA extraction, ancient DNA, Polymerase chain reaction (PCR), Gel electrophoresis, Spectrophotometry





Phenotypic study on *Jatropha curcas* *AGAMOUS* overexpressed *Arabidopsis*

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Oil consumption has skyrocketed while new discovery of oil or petroleum sources have decreased. Biodiesel has gained extensive research as an alternative source of energy. Biodiesel can be produced from *Jatropha curcas* which is a monoecious plant whose fruits can be directly squeezed to obtain oil. In *J. curcas*, male and female flowers are in the same inflorescence, and normally the female flowers are outnumbered by male flowers by many folds. This lack of female flowers lead to low fruitset and thus low yield. To increase number of female flower, a construct containing *J. curcas* *Agamous* gene under the control of 35S promoter was transformed into *Arabidopsis thaliana*. Transgenic plants were analyzed for flower structure and expression of the *JcAG* was analyzed by Real-Time PCR. The research is still ongoing.

Keywords: *Jatropha curcas*, *Arabidopsis thaliana*, Biodiesel, Real-Time PCR, 35S promoter





***SCALLOPED* and *SEPIA* gene Knockouts in *Drosophila melanogaster* using CRISPR/Cas9**

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CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) and CRISPR-associated (Cas) is the new effective technique to modify genetic material in organisms and to study function of genes that are related to the trait of interest. CRISPR-Cas9 consists of two parts, a guide RNA (gRNA) and a non-specific CRISPR-associated endonuclease (Cas9). The gRNA is a sequence designed specifically for the gene of interest. The Cas9 protein is the endonuclease that is responsible for cutting the DNA in that specific position and resulting in a double strand break (DSB). In this study, *Drosophila melanogaster* was used because of its phenotypic variations and a relatively large database. Two genes, *SEPIA* and *SCALLOPE* controlling the body color and the wing shape were selected to test the CRISPR/Cas9 technique. The eggs and larvae of *D. melanogaster* were either microinjected or fed with *in vitro* transcribed gRNA and the Cas9 protein. The survival rate and the phenotype of these injected fruit flies were observed comparing to the control. We found that the survival rates of the control groups injected with loading dye were 35% when injected into the eggs and 42.86% when injected into the larvae. The survival rates of the control groups (fed with distilled water) were 5% (egg), 94% (larvae). The experiment groups (fed with Cas9 and gRNA) have the survival rate of 3% for eggs and 94% for larvae. However, we couldn't observe the phenotypic changes in the experimental group. In further experiments, both eggs and larvae will be injected with Cas9 and gRNA at different concentrations. This technique can also be adapted to be used to test gene functions in other insects in the future.

Keywords: Gene editing, Insect, CRISPR-Cas9, gRNA





Identification bacteria on facial, oily and dry skin in family

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Many microorganisms that live on human skin and do not cause the disease to host are called normal flora. The different areas of our body's skin also have a different diversity of microbe that depends on host skin characteristics, endogenous and exogenous environmental factors. In this study, we aimed to identify bacteria on facial skin of five individuals from the same family. There are two groups of the skin characteristics, oily and dry skin. We used swabbing technique for collecting samples from facial skin of each individual and used a TSA medium for growing bacteria. Then, 16s rRNA gene was amplified and sequenced to identify bacteria. Sequences were blast against GenBank and EzTaxon database. In the result, the most of bacteria found in this study was *Staphylococcus epidermidis*. This bacterial species is one of normal flora which found in both types of skin. The result showed that bacteria found on oily facial skin were genus *Staphylococcus*, *Bacillus*, *Pseudomonas*, *Enterobacter* and *Micrococcus* while those on dry facial skin were *Brevibacillus*, *Staphylococcus* and *Micrococcus*. From the result, we couldn't see the difference of bacterial species between oily and dry skin because only a few of bacteria could be isolated. One thing we found in this result was women's facial skin have only *Staphylococcus sp*. It might be caused by using cosmetics and facial cleansers resulting in lower diversity of bacteria on women's facial skin. Therefore, men have higher diversity of facial bacteria than women. We summarized that exogenous environmental factors can affect to diversity of bacteria on facial skin.

Keywords: normal flora, facial skin, oily skin, dry skin





Comparative transcriptome analysis of mantle and foot tissues of giant African snail (*Achatina fulica*)

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Giant African snail (*Achatina fulica*) is one of the largest land snails and an agricultural animal pest worldwide. Even though there are several reports on medical and pharmaceutical applications of this snail secretion, the mucus biosynthesis of *A. fulica* foot and mantle tissues remains unknown. Therefore, this study aimed to optimize the RNA extraction techniques for preparing total RNAs from *A. fulica* foot and mantle tissues. Trizol reagent and commercial RNA extraction kit (E.Z.N.A.® Mollusc RNA Kit) were compared in terms of quantity and quality of the total RNAs. Results showed that RNAs extracted by the Trizol reagent of the mantle tissue (concentration of 1.65 $\mu\text{g}/\mu\text{l}$ and A260/A280 ratio of 2.03) had higher quantity and quality compared to those extracted from the foot tissue (concentration of 1.40 $\mu\text{g}/\mu\text{l}$ and A260/A280 ratio of 1.98). The extraction by Trizol reagent (concentration of 1.65 $\mu\text{g}/\mu\text{l}$ and A260/A280 of 2.03) was also better than the commercial RNA extraction kit (concentration of 9.3 $\text{ng}/\mu\text{l}$ and A260/A280 of 2.02). The best quality of the total RNAs has been submitted for RNA sequencing. Furthermore, the comparative transcriptomes of these two tissues will be performed to reveal the mucus biosynthetic pathways of *A. fulica* which will be useful for medical and cosmetic applications.

Keywords: RNA extraction, Trizol, *Achatina fulica*, mantle tissue, foot tissue





Development of Gold-Iron Nanoparticles as a Contrast Agent for MRI

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Contrast agents play an important role in biomedical applications such as targeted drug delivery, biochemical sensing, and sensitive diagnosis. Metal nano-sized magnetic particles can be used as a contrast agent for medical imaging. The metallic nanoparticles leads to increase of its reactive and their magnetic properties are controlled by superparamagnetic and surface effects. Gold-coated magnetic iron oxide nanoparticles (Au-FeNPs) result in increased stability and robustness of particles. In this study, the nanoparticles of iron coated by gold were synthesized by chemical method and their structural as well as magnetic characteristics were analyzed. Gold nanoparticles (AuNPs) were prepared by reduction of Au^{3+} to Au^0 using tri-sodium citrate ($\text{Na}_3\text{C}_6\text{H}_5\text{O}_7 \cdot 2\text{H}_2\text{O}$). Then, Au-FeNPs were synthesized by surface ionic interaction. The morphology, size, shape and contrast properties were investigated using UV-visible spectroscopy scanning electron microscopy (SEM), transmission electron microscopy (TEM) and magnetic resonance imaging (MRI). The results showed that the size of AuNPs was 10 nm and absorbed UV at 520 nm. The images from SEM demonstrated that FeNPs were coated with AuNPs. Size distribution of the bi-metallic nanoparticles was $\sim 30\text{nm}$. The molar ratio of Au:Fe nanoparticles was 9.9 : 0.05. The results of MRI are in the process of investigation.

Keywords: Gold-Iron nanoparticles, Contrast agent, Magnetic Resonance Imaging (MRI), UV-visible spectroscopy, Scanning Electron Microscopy (SEM) and Transmission Electron Microscopy (TEM).





Natural Rubber / Iron (II,III) Oxide Composites for Use as Gamma Radiation Shields

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This work investigated the gamma shielding, cure characteristics, and mechanical properties of natural rubber (NR) with additions of iron (II,III) oxide (Fe_3O_4) for potential use as flexible, lead-free, gamma-shielding materials. The results showed that increases in the contents of Fe_3O_4 from 0 to 100, 300, and 500 parts per hundred parts of rubber by weight (phr) and increases in thickness of the composites increased the values of the gamma attenuation coefficients, tensile modulus at 100% elongation, and hardness (Shore A), but reduced the values of the tensile strength and elongation at break. All composites also underwent thermal aging tests at 70 °C for 96 hr to investigate the deterioration of mechanical properties with time caused by heat. The results indicated only slight decreases in the overall tensile properties. Specifically for the composites with 500 phr of Fe_3O_4 , the linear attenuation coefficients (μ_l) were $8.0 \pm 1.2 \text{ m}^{-1}$ for 662-keV γ and $4.4 \pm 0.6 \text{ m}^{-1}$ for 1.25-MeV γ , while the mass attenuation coefficients (μ_s) were $2.6 \times 10^{-3} \pm 0.4 \times 10^{-3} \text{ m}^2/\text{kg}$ for 662-keV γ and $1.7 \times 10^{-3} \pm 0.2 \times 10^{-3} \text{ m}^2/\text{kg}$ for 1.25-MeV γ

Keywords: Gamma rays shielded rubber, radiation shielding materials, Iron (II,III) Oxide





Radioprotective Effect of Apigenin against Gamma Radiation-Induced Micronucleus in Mice

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The objective of this study aimed to evaluate the radioprotective effect of apigenin, a potent anti-oxidant in flavonoids, against radiation-induced micronucleus in peripheral blood *in vivo*. Micronucleus assay (MN) stained with fluorescent dye, acridine orange (AO), was used in the present study. Blood samples were collected from the mice that were pre-treated with various concentrations (0, 10, 20 and 40 mg/kg) of apigenin for 3 days and 10 days, respectively. After treatment, the mice were irradiated with 3 Gy for one time. They were sacrificed at 3rd day after irradiation. Their blood were collected and then smeared on the slides. All slides were stained with AO and observed micronucleus under light microscope. The results showed the reverse correlation between the concentration of apigenin and frequency of micronucleus. Our present results indicate that apigenin may be an effective radioprotective against gamma radiation.

Keywords: Radioprotective, Micronucleus assay, Apigenin, Acridine orange





Radio-protective Effect of *Momordica cochinchinensis* (Lour.) Spreng: Against Gamma Irradiation – Induced Dicentric Chromosome in Human Peripheral Blood Lymphocytes *in vitro*

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Gac fruit (*Momordica cochinchinensis* (Lour.) Spreng) has been known to contain high levels of beta-carotene and lycopene which are potent anti-oxidants and can prevent the chromosomal damage from radiation exposure. The purpose of this study aimed to evaluate the radio-protective effect of gac fruit extracts against 3 Gy gamma ray on human lymphocytes.

In this study, the various parts of gac fruit (aril oil, aril solid and skin) were extracted and used at concentrations of 0, 25, 50, 75 µg/ml. The results showed that pre-treatment of lymphocytes with aril oil, aril solid and skin extracts of gac fruit altered the frequency of dicentric chromosome mediated by gamma ray when compared to the lymphocytes treated with radiation alone. These findings revealed the reverse correction between the concentration of gac fruit extracts and frequency of dicentric chromosome. Our study indicated that gac fruit extracts might be an effective radio-protector against gamma radiation.

Keywords: Dicentric assay, Lymphocyte cell, Gac fruit, Chromosome, Radio-protective





Development of Gold Nanoparticles as a Contrast Agent for MRI

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Imaging techniques play a critical role in medical applications such as diagnosis, treatment planning and evaluation. CT, MRI and ultrasound are standard clinical imaging modalities. X-ray CT is one of the most convenient and efficient tools used in hospital. However, CT has limitation in clarifying between soft tissues that have similar densities. Thus, it is important to use a contrast agent to improve quality of CT imaging. Metal nano-sized particles are attractive in CT contrast agent field because their unique physical and chemical properties. In this study, we focus on synthesize gold nanoparticle for X-ray CT imaging. The gold nanoparticles (AuNPs) were prepared by Turkevich chemical method. Au (III) ions were reduced by trisodium citrate to form Au⁰. The metallic nanoparticles were stabilized by 5000 da polyethylene glycol (PEG). UV-visible spectroscopy scanning electron microscopy (SEM), transmission electron microscopy (TEM), magnetic resonance imaging (MRI) and X-ray computed tomography (X-ray CT) were used to investigate morphology, size, shape, and contrast properties of AuNPs. The results showed that the size of AuNPs was 10 nm and absorbed UV at 520 nm. The images from SEM indicated that there were Na and Au in dried phase. The results of MRI and X-ray CT are in the process of investigation.

Keywords: Gold nanoparticle, Scanning electron microscopy (SEM), Transmission electron microscopy (TEM), Magnetic resonance imaging (MRI), X-ray computed tomography (X-ray CT)





The quantitative measurement of organophosphate insecticides using gold nanoparticles

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Pesticide contamination in environment is a serious problem in agricultural based countries. Normally, the organothiophosphate pesticide is always found in the general surface water. Highly sensitive and accurate evaluation of the contamination in environment samples are importance for environmental monitoring. In this study, we synthesized gold nanoparticles (GNPs) to detect Malathion insecticide using gamma radiation synthesis. The sulfhydryl group (-SH) of malathion can react with GNPs, leading to change the color of the solution and increasing Plasmon resonance absorption. The absorption depends on concentration of the insecticide and can be visually detected by UV-visible spectroscopy. The GNPs were synthesized in an aqueous solution, containing 0.5 mM $\text{HAuCl}_4 \cdot 6\text{H}_2\text{O}$ in presence of 4 % polyvinylpyrrolidone (PVP) as a stabilizer and 0.2 M 2-isopropanol as a scavenger of oxidizing agents. The sample solutions were irradiated 4 kGy at room temperature in a ^{137}Cs gamma irradiator with dose rate $4.15 \pm \text{Gy/min}$. The dose rate was measured by Fricke dosimeter. The total volume of the mixed solutions was 1 ml. The detection limit of this technique to measure the insecticide was 2 mM. The color of GNPs solutions in presence of malathion changed from pink to purple. The color of the solution depended on the concentration of malathion.

Keywords: Gold nanoparticles, Gamma ray, Pesticides, organothiophosphates, Malathion, polyvicyl alcohol, polyvinylpyrrolidone, acrylamide





Natural Radionuclides in *Zingiber cassumunar* measured by Gamma Spectrometry

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Activity concentration of naturally occurring radioactive materials (NORMs) in medicinal plants for *Zingiber cassumunar* were investigated in this work by means of gamma spectrometry analysis using a HPGe detector. The results showed that the activity concentration of ^{238}U , ^{226}Ra and ^{40}K in *Zingiber cassumunar* were found to be 146.29 ± 0.0101 Bq/kg, 6.4888 ± 0.0076 Bq/kg and 339.49 ± 0.0062 Bq/kg, respectively. The average annual committed effective doses (AADED) for each radioactive isotope due to the ingestion of *Zingiber cassumunar* were 0.0118 mSv/y, 0.00327 mSv/y and 0.00378 mSv/y for ^{238}U , ^{226}Ra and ^{40}K , respectively. The results indicated that the AACED values from the interested plant were far below the worldwide average annual committed effective dose of 0.3 mSv/yr for an individual provided in UNSCEAR 2000 report, implying that the associated radiological health risk resulting from the intake of radionuclides in the medicinal plants was insignificant.

Keywords: medicinal plants, gamma spectrometry, HPGe, radionuclides





Effects of Radiation Dose Rate on Gold Nanoparticles by Gamma Irradiation

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Synthesis of Gold nanoparticles (AuNPs) of desired sizes and shapes has enormous importance because small changes in the sizes or shapes of nanoparticles can have great effects on a variety of properties and different applications. This study aimed to assess effects of radiation dose rates on gold nanoparticle synthesis. AuNPs were synthesized by gamma irradiation in an aqueous solution, containing 1mM HAuCl₄.6H₂O in presence of 4 % polyvinyl alcohol (PVA) as a stabilizer and 0.2 M 2-isopropanol as a scavenger of oxidizing agents. The sample solutions were irradiated 4, 6 and 8 kGy at room temperature in a ¹³⁷Cs gamma irradiator with dose rate 4.15 Gy/min and a ⁶⁰Co gamma irradiator with dose rate 59.9 Gy/min. The dose rate was measured by Fricke dosimeter. The results showed that at low dose rate the values of λ_{max} were 539, 536 and 534 nm for irradiation doses 4, 6 and 8 kGy, respectively. At high dose rate the absorbance wavelengths were 520, 518 and 522 nm, respectively. In addition, the size of AuNPs was indicated using transmission electron microscopy (TEM).

Keywords: Gold nanoparticles synthesis, Gold nanoparticles (AuNPs), Gamma irradiation, Radiation dose rate





Gold nanoparticles synthesis by chemical methods

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Metal nanoparticles have been attractive in many fields because their special physical and chemical properties can be used in many applications such as biochemical biomedical and industrial applications. In this project, we prepared gold nanoparticles (GNPs) using chemical method. 0.25 mM choloauric acid (HAuCl_4) was reduced by 2.5 mM tri-sodium citrate. The gold chloride solution was heated to the boiling point then tri-sodium citrate was added to the solution with rigorous stirring to maintain a homogenous solution. The color of the solution changed to red wine color. It indicated that the Au^{3+} ions were reduced to Au^0 atom to form GNPs. The morphology, size, shape and contrast properties were investigated using UV-visible spectroscopy scanning electron microscopy (SEM), transmission electron microscopy (TEM). The results showed that the size of GNPs was 12.5 ± 2.5 nm and absorbed UV at 520 nm. The images from SEM indicated that there were Na and Au in dried phase.

Keywords: Gold nanoparticles synthesis, Trisodium citrate, PEG





Natural Radionuclides in *Andrographis paniculata* (Burm.f.) Wall ex Nees. measured by Gamma Spectrometry

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Andrographis paniculata (Burm.f.) Wall ex Nees. is an important medicinal plant in Thailand. This study aimed to investigate natural radionuclides in *Andrographis paniculata* (Burm.f.) Wall ex Nees. and soil from Sunee garden in Banglen District of Nakhonpathom. All samples were analyzed for activity concentrations of natural and artificial radionuclides using HPGe gamma spectrometry. The average annual committed effective dose (AACED) due to the ingestion of radionuclides from *Andrographis paniculata* (Burm.f.) Wall ex Nees. were also estimated. The activity concentrations of ^{238}U , ^{226}Ra , and ^{40}K in soil samples were found to be 180.57 Bqkg^{-1} , 144.33 Bqkg^{-1} and 339.98 Bqkg^{-1} , respectively, while the activity concentrations of ^{238}U , ^{226}Ra , and ^{40}K in *Andrographis paniculata* (Burm.f.) Wall ex Nees. were 133.00 Bqkg^{-1} , 100.55 Bqkg^{-1} and 950.77 Bqkg^{-1} , respectively. The soil to plant transfer factors (TF) varied from 0.77, 0.70 and 2.80 respectively for ^{238}U , ^{226}Ra , and ^{40}K , respectively. The AACED due to the ingestion varied from $0.0106 - 0.0507 \text{ mSvy}^{-1}$. The AACED values reported in this study are much below the world average value of 0.30 mSvy^{-1} for an individual. This indicates that there is no radiological health risk in using these plants for medicinal purposes. This study may also contribute data on local medicinal plants to formulate regulations related to radiological healthcare.

Keywords: medicinal plants, radionuclides, gamma spectrometry, HPGe





Principles and calibration of a radon detector (RAD 7)

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Radon is radioactive inert gas that found in natural samples such as rock, soil, water and air. Radon is produced by the decay of uranium-238 series. It is due to the fact that radon emits alpha particles and half-life is 3 days. Thus, the radioisotope gas is very dangerous, leading to deposit all energy to internal organs, especially lung. RAD 7 is one of the most efficient alpha spectrometry instrument. The hand-held instrument can be used to measure radon content in natural samples. The device is a small size to carry to work in the field. However, the instrument is very sensitive and need to set up calibration. In this study, the instrument were calibrated by radon chamber and compared with a calibrated device. It can be performed in the field and reliable. The radon instrument will be available to people who are interested in studying radon measurement in environmental samples.





Effects of Acute Gamma Irradiation on *In vitro* Culture of *Globba sp.*

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Globba (Globba sp.), native to Thailand, are recently gained much popularity in the world cut flower market. Tuberos rhizomes are produced commercially for export and gradually increase in demand every year. The present investigation was undertaken to broaden the genetic variability of *Globba* using gamma ray as well as to employ micropropagation to bypass the slow rate of multiplication of traditional methods. Acute gamma irradiation on *in vitro* culture was carried out; the mutation effect, the medium lethal dose (LD_{50}) and the 50% growth reduction (GR_{50}) were examined. *In vitro* culture of *Globba* grown on MS proliferation medium were exposed to acute gamma radiation from Cs-137 at Nuclear Research Center, Faculty of Science, Kasetsart University at the doses of 0, 10, 20, 30, 40 and 50 Gy. After irradiation, the irradiated plantlets were subculture on MS medium supplemented with 1.0 mg/L BA. The results in M_1V_1 generation showed that percent of survival plantlets and growth of *Globba* seedling were decreased as radiation dose increased. At 45 days after irradiated, the $LD_{(50/45)}$ and $GR_{(50/45)}$ were estimated to 33 and 38 Gy, respectively. Some abnormalities observed in the M_1V_1 generation of the irradiated plantlets were variegated leaf, changed in leaf color and white stripe leaf. The most mutation frequency was found at dose of 20 Gy. These characteristics that detected in M_1V_1 generation will be further studied on the heritability in M_1V_2 generation.

Keywords: Acute Gamma Irradiation, *In vitro* Culture, *Globba sp.*





Frequency of Micronuclei in Human Lymphocytes after Exposure to Low-dose Gamma Rays *in vitro*

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The radiation biodosimetry is used to assess the radiation absorbed dose that people might expose from several activities such as routine working with radiation, nuclear accidents, or living in high-level background radiation areas. Micronuclei were used as a biomarker to indicate the chromosomal damage caused by ionizing radiation exposure. In this study, the cytokinesis-blocked micronucleus (CBMN) assay was employed to analyze the correlation between the radiation absorbed dose and the frequency of micronuclei in 1,000 binucleated cells in human lymphocyte cells after *in vitro* exposure to low-dose gamma rays. The whole blood was exposed to a Cs-137 gamma source at the dose rate of 1.75 Gy/min for the dose of 0, 0.2, 0.4, 0.6, 0.8 and 1.0 Gy. The micronuclei were analyzed with respect to the guidelines recommended by the IAEA. The results show that the frequencies of micronuclei were increased related to radiation absorbed dose from 0-1 Gy. This study indicated the potential of using micronuclei to assess the absorbed dose in case of low-dose exposure.

Keywords: Low-dose gamma rays, Lymphocyte cells, Micronuclei, Radiation Biodosimetry





Synthesis of Graphene Oxide/Chitosan Composite for Radionuclide Adsorption from Aqueous Solution

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Iodine-131 is the most common radioactive iodine found in wastewater from medical facilities including nuclear medicine. The removal of radioiodine is needed before discharging into the environment. This research aimed to explore the capacity of graphene oxide/chitosan (GO/CS) composites to remove iodine-131 from aqueous solution. GO was synthesized by modified Hummer's method, then mixed with CS in a ratio of GO to CS 2:1. Iodine-131 was added into the graphene composites with varying adsorption time. The radioactivity of iodine-131 in supernatant was analyzed by gamma spectrometry with HpGe detector. The adsorption efficiency was calculated by comparing the activities of iodine-131 before and after mixing with graphene composites. The results show that GO/CS composites can remove up to 99 percent of iodine-131 from aqueous solution. This provides information on the potential of graphene composites for removal of radioiodine from waste water.

Keywords: Iodine-131, radioactive iodine, Hummer's method, Graphene oxide, Chitosan, Graphene oxide/chitosan, potential of graphene composites





Development of flexible gamma shielding materials from natural rubber/tungsten oxide composites

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This research aimed to develop a flexible gamma-ray shielding material from composites of tungsten oxide (W_2O_3) and STR20 natural rubber (NR). The contents of W_2O_3 that were added to NR varied from 0, 100, 300, and 500 parts per hundred parts of rubber by weight (phr) and the composites were tested for mechanical properties (tensile modulus at 100% elongation, elongation at break, tensile strength, hardness, specific gravity, morphology, and gamma shielding properties). The results showed that tensile modulus, hardness, and gamma shielding properties increased as contents of W_2O_3 increased. In particular for the composites with 500-phr W_2O_3 , the linear attenuation coefficients (μ_l) were $13.8 \pm 0.8 \text{ m}^{-1}$ for 662-keV γ and $7.1 \pm 0.8 \text{ m}^{-1}$ for 1.25-MeV γ , while the mass attenuation coefficients (μ_s) were $5.0 \times 10^{-3} \pm 0.3 \times 10^{-3} \text{ m}^2/\text{kg}$ for 662-keV γ and $2.7 \times 10^{-3} \pm 0.2 \times 10^{-3} \text{ m}^2/\text{kg}$ for 1.25-MeV γ , respectively.

Keywords: gamma-ray, tungsten oxide, natural rubber STR20,





Measurement of the radionuclides in milk and dairy products by Gamma Spectrometry

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The objective of this study was to investigate the radionuclides in milk and dairy products from Japan and Thailand. Two samples of each product such as Japanese milk (Hokkaido Gyunyu and Milkland Hokkaido), Thai milk (Foremost and Thai-Denmark), and Japanese dairy products (UHA milk candy and Jersey milk candy) have been measured and analyzed by a high-purity germanium (HPGe) detector for gamma spectrometry analysis. The results of this study showed that the specific activities of K-40, Ra-226, and U-238 in Japanese milk exhibited in a range of 1.65-42.25 Bq/L, 1.93-2.26 Bq/L and 34.79-40.31 Bq/L, respectively. The specific activities of K-40, Ra-226, and U-238 in Thai milk exhibited in a range of 39.93-43.33Bq/L, 1.90-2.12Bq/Land 44.94-49.57Bq/L, respectively. The specific activities of K-40, Ra-226, and U-238 in Japanese dairy products exhibited in a range of 57.85-67.84 Bq/kg, 2.29-2.35 Bq/kg and 41.89-42.99 Bq/kg, respectively. These finding indicated that the specific activities of radionuclides in these samples was lower than the standard of International Atomic Energy Agency (IAEA). Therefore, these samples could be safe for consumption.

Keywords: Radionuclides, Milk, Dairy product, Gamma Spectrometry





Gamma Shielding materials by natural rubber/bismuth oxide composites.

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This research developed a flexible and lead-free gamma-ray (γ) shielding material using natural rubber and bismuth (III) oxide powders (Bi_2O_3) with the contents of Bi_2O_3 varied from 0 to 100, 300, and 500 parts per hundred parts of rubber by weight (phr). The results showed that the increases in contents of Bi_2O_3 and thickness of the composites increased the values of the gamma attenuation coefficients, tensile modulus at 100% elongation, and hardness (Shore A), but reduced the values of the tensile strength and elongation at break. All composites also underwent thermal aging tests at 70 °C for 96 hr to investigate the deterioration of mechanical properties with time caused by heat. The test showed that only small changes in overall mechanical properties were observed. For gamma shielding tests, a Sodium Iodide (NaI) detector with Co-60 and Cs-137 sources were used to characterize gamma shielding properties. Specifically for the composites with 500 phr of Bi_2O_3 , the linear attenuation coefficients (μ) were $20.4 \pm 1.2 \text{ m}^{-1}$ for 662-keV γ and $10.5 \pm 1.0 \text{ m}^{-1}$ for 1.25-MeV γ , while the mass attenuation coefficients (μ_s) were $6.1 \times 10^{-3} \pm 0.4 \times 10^{-3} \text{ m}^2/\text{kg}$ for 662-keV γ and $2.9 \times 10^{-3} \pm 0.3 \times 10^{-3} \text{ m}^2/\text{kg}$ for 1.25-MeV γ , respectively.

Keywords: radiation shielding; gamma shielding; natural rubber; radiation protective agent





Assessment of Absorbed Radiation Doses in Human Peripheral Lymphocytes using *In Vitro* Micronucleus Assay

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The ionizing radiation is used in a wide variety of fields such as medicine, industry and energy. The potential effects of radiation on human health are of great concern. Micronucleus assay is extensively used to evaluate the extent of chromosomal damage in humans exposed to genotoxic agents including ionizing radiation. This study aimed to assess the absorbed doses in human peripheral lymphocyte after exposure to gamma radiation using cytokinesis-block micronucleus (CBMN) assay. The blood sample received from a healthy male donor was collected and irradiated with gamma rays at doses of 0, 1, 2, 3, 4 and 5 Gy using a Cs-137 source. The CBMN assay was performed to determine the relationship between the micronuclei frequencies and the absorbed doses. The dose-response curve was established to be applied to assess the absorbed radiation doses in the exposed individuals.

Keywords: Ionizing radiation, Gamma rays, Cytokinesis-block micronucleus assay, Human peripheral lymphocytes, Dose-response Curve, Biological dosimetry





Radio-Protective Effect of *Momordica cochinchinensis* (Lour.) Spreng Against γ -Radiation-Induced Cytotoxicity in Human Colorectal Adenocarcinoma Cell Line (HT-29)

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Momordica cochinchinensis (Lour.) Spreng is commonly known as gac fruit, highly carotenoid pigment containing fruit especially lycopene and beta-carotene. Both lycopene and beta-carotene are the antioxidant agents that can resist the negative effects of free radical from gamma ray exposure. However, the different part of gac fruit e.g.aril, skin and pulp has a different containing of carotenoid and other phytochemicals. This study conducted to examine the radio-protective property of the different part of gac fruit on HT-29 viability. Seeding HT-29 cell at densities about 5×10^4 cell/ml in 96 well plate and incubated for 24 hours. Part of gac fruit extracts, aril oil, aril solid and skin was tested in various concentrations (0, 20, 40, 80,100 $\mu\text{g/ml}$) of vitamin E and incubated for 1 hour before gamma irradiation (3Gy). Viability of cells was determined by MTT-assay. The result shows that cell viability of gamma irradiated treatments were increased with an increase in concentration of gac extracts.

Keywords: Radio-protective, Ionizing radiation, Gamma rays, Gac fruit, Antioxidant, MTT-assay,





Radioactivity Level in Marine Sediment after Fukushima Nuclear Power Plant Accident

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All people are generally exposed to natural and man-made radioactivities depending on their living environment. The radioactivity levels in marine and coastal environment are of great concerns. The research main objective is to measure and analyze the specific activities of natural and man-made isotopes in marine sediment collected in Chonburi Province after the Fukushima Daiichi disaster in 2011. The sediment samples were collected by a grab sampler from 10 sites along the seashore in three periods during 2016 - 2017. The radionuclides were analyzed using a high-purity Germanium (HPGe) detector and gamma spectrometry analysis system. The measuring time of each sample was 15,000 seconds. It was found that no artificial radionuclide was detected in this study. The mean specific activities of natural radionuclides, i.e. ^{40}K , ^{226}Ra and ^{232}Th in sediment samples were not significantly different with those found before Fukushima accident. The experimental results were compared to the previous data of the Office of Atoms for Peace (OAP) in Thailand and the global radioactivity measurements.

Keywords: U-238 Th-232 K-40 HPGe







Effect of stabilizers on the formation of gold nanoparticles using gamma irradiation.

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Currently, gold nanoparticles (GNPs) have been extensively interesting in many scientific fields such as biochemical, biomedical and industrial applications. The size and shape are important factors for chemical and physical properties to apply these nanoparticles. It is well known that the radiation-induced generation of metal cluster has been showed a powerful techniques to synthesize at room temperature nano-colloids and to control their size as well as shape. The three main components of the metal solutions are gold ions, a stabilizer and an oxidizing scavenger. In this study, we focused on radiation synthesis of GNPs in aqueous biopolymer dispersion, using 100000 da polyvinyl alcohol (PVA), 10000 da polyvinyl pyrrolidone (PVP) and acrylamide monomer (ACM) as a stabilizer. The Au-nanoparticles were synthesized by a ^{137}Cs gamma chamber irradiator with dose rate $4.15 \pm \text{Gy}/\text{min}$. The dose rate was measure by Fricke dosimeter. The sample solutions were composed of 0.5 mM HAuCl_4 , 0.2 M iso-propanol used as an oxidizing scavenger and 4 % a stabilizer. The solutions were purged nitrogen gas 15 minutes to remove oxygen before irradiation. The optimum absorbed dose was 4 kGy. The GNPs solutions were investigated by UV-visible absorption spectroscopy. The maximum absorbance wavelengths (λ_{max}) of the sample solutions were 520-530 nm. The λ_{max} of GNPs in acrylamine-polymer was 520 nm, while the maximum wavelength of gold nanoparticles in PVA was 530 nm. However, the GNPs in 4 % PVP were not observed in these wavelengths. In order to study effect of PVP on formation of GNPs, we varied concentration of PVP 2, 4 and 6 %. The results showed that at 2 % PVP the maximum absorbance wavelength was 532 nm, while at 6 % PVP the λ_{max} was 530 nm. These results indicated that the size of gold nanoparticles depended on stabilizers and concentration of stabilizers. In addition, the morphology of gold nanoparticles in dried phase were analyzed by Transmission Electron Microscopy (TEM).

Keywords: gold nanoparticles, gamma radiation, polyvinyl alcohol, polyvinyl pyrrolidone, acrylamide monomer





Improvement of Vetiver for Salt Tolerance through Gamma Ray and Tissue Culture Technique

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Vetiver grass is generally used as protection against soil erosion and slope stabilization due to its extraordinary morphological and physiological characteristics. In Thailand, approximate 3.61 million ha of land are affected by salt, with about 2.8 million ha covering the northeastern part of the country and about 0.58 million ha covering coastal areas. This problem is predicted to be worsen as a result of climate change. Although Vetiver is relatively highly tolerant to salinity as compared with some common crop and pasture species, its effectiveness in soil erosion control and slope stabilization is sometimes affected under highly saline conditions. The aim of the present field study was to improve the salt tolerance of vetiver grass.

The plantlet of native Thai vetiver ecotype Mai Huay Wai was carried out aseptically with tissue culture. After 4 weeks, the plantlet were exposed to acute gamma radiation from Cs-137 at Nuclear Research Center, Faculty of Science, Kasetsart University at the doses of 0 (control) and 15 Gy. Hundred and twenty M₁V₂ plantlets were screened for their salt tolerance level using 0, 0.5, 1 and 1.5% NaCl under *in vitro* condition. The results showed that the concentration level of NaCl increase causing growth rate and number of new tillers decrease. Moreover, the M₁V₂ plantlet under saline condition with 1.5% NaCl of irradiated treatment demonstrated higher survival rate compared to non-irradiated treatment. Further screening on M₁V₃ generation employing 0, 1.5, 2 and 2.5% NaCl to study the effect of salt on plant growth and development would be evaluated.

Keywords: Vetiver, Gamma Irradiation, Tissue Culture, Salt tolerance





Studying the Characteristic of DNA Damage after Gamma Irradiation using Gel Electrophoresis

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Biological organisms are under continuous attack from chemical and physical agents of both natural and manmade origin, which can result in cell death, mutations or cancer induction. DNA is the repository for the information that a cell requires to function and reproduce, and is almost always the critical target. The damage inflicted upon DNA can take many forms that depend on the nature of the mutagen. Gamma ray is one of the low linear energy transfer (LET) of ionizing radiation. The mechanism of damage is mainly given indirect action to the cell, however, unclearly understood. Thus, the purpose of this study was to study the characteristic of DNA molecule after exposed by gamma radiation.

DNA extract from fresh tomato leaves using the CTAB extraction buffer were directly exposed to gamma radiation from Cs-137 at Nuclear Research Center, Faculty of Science, Kasetsart University at the doses of 0 (control), 200, 400, 600 and 800 Gy. The concentration of plant DNA was dilute to 100, 200, 300 and 400 ng / μ l, prior to gamma irradiation. The characteristic of DNA damage and DNA migration were observed by 1.0% agarose gel electrophoresis, which was then stained with SYBr safe for visualization. The results showed that increasing of radiation dose enhanced DNA stand break regarding to migration fragments. The information from this research can be further study the effects of gamma rays on biological organism.

Keywords: DNA damage, Gamma irradiation, Gel electrophoresis





Radon measurement of water samples from Kamphaeng Phet province using RAD7

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The study aimed to measure ground water re and analyze ^{222}Rn Radon concentration in ground water samples. The samples were collected ground water from eight positions with 50 m depth and one surface water sample from upper Chao phraya river at Kamphaeng Phet province. ^{222}Rn in these samples was measure by Alpha spectrometer (RAD7) and measure immediately after sampling. This technique is easy, convenient, precisely and suitable for the field work. The results showed that the concentration of radon in the surface water sample was 1.94 pCi/L, while levels of the radioactive gas in ground water samples were 14.46 – 203.39 pCi/L. the highest concentration of Rn-221 in ground water was found at Khlong Mae Lai station (SGW36)

Keywords: RAD7, ^{222}Rn Radon, Ground water, Surface water, Alpha spectrometry





Antimutagenic Potential of Mangosteen Peel Extract on Chromosomal Aberrations in *Allium cepa*

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The growing interest in the replacement of synthetic food antioxidants by natural ones has fostered research on the screening of plant-derived raw materials for identifying new antioxidants. Mangosteen peel is an important source of natural phenolic antioxidants. The mangosteen peel contains various bioactive substances, i.e., phenolic acids and flavonoids, which possess biological and medicinal properties, especially antioxidant properties. In the present study, the antimutagenic potential of mangosteen peel was evaluated in *Allium cepa* root meristem cells. So far there is no report on the biological properties of mangosteen extract in plant test systems. The roots were soaked in tap water for 3 days. Then the root tip cells were exposed to acute irradiation with gamma ray from Cs-137 at dose of 3 Gy. Mangosteen peel extract was given at 10, 20 and 30 $\mu\text{g/ml}$ for 24 hours, prior to gamma irradiation. The tips were squashed after fuelsen treatment and the cells were analyzed for chromosomal aberrations.

Gamma radiation induced chromosomal aberrations including fragment, bridge and micronucleus significantly with non-irradiated control. The total number of aberrations was significantly reduced in root tip cells pretreated with mangosteen peel extract. The study reveals that mangosteen peel has antimutagenic potential against gamma radiation induced chromosomal aberrations in *Allium cepa* root meristem cells. The antimutagenic potential of mangosteen peel extract is effective at 30 $\mu\text{g/ml}$ in *Allium cepa* root meristem cells.

Keywords: Chromosome aberrations, gamma ray, *Allium cepa*





Synthesis of Graphene Oxide/Chitosan Composites for Adsorption of Soluble Iodine-131 in Aqueous Solution

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The immobilization of radioiodine from wastewater using different materials is of interest in radioactive waste management. This research aimed to investigate the capacity of graphene oxide/chitosan (GO/CS) composites for the adsorption of iodine-131 from aqueous solution. GO was synthesized by the modified Hummer's method, then mixed with chitosan in a ratio of GO to CS 2:1. The composite with different volume varying in 200-5,000 μl was mixed with iodine-131 for 24 hours. The radioactivity of supernatant was determined by HPGe gamma spectrometry. The result revealed that the more volume effected the higher adsorption of iodine-131 solution. However, the adsorption abilities of iodine-131 at volume of 800 μl were saturated with 99% adsorption efficiency.

Keywords: Ionizing radiation, Gamma rays, Hummer's method, Graphene, Graphene oxide, Chitosan, Graphene oxide/chitosan, Iodine-131, Management of waste





Measurement of radionuclides in Japanese snacks by Gamma Spectrometry

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This study aimed to determine the amount of radionuclides in Japanese snacks which have been imported and distributed to the supermarket in Thailand. Six Japanese snack (Kitano Aye, Pocky strawberry, Ramenmini chicken flavour, Pocky Almond crush, Nabisco and Panpoko yakisoba sauce) were randomly chosen for measurement of their radionuclides by HPGe for Gamma spectrometry. The result demonstrated that the specific activities of radionuclides, e.g. K-40, Ra-226 and U-238, of Japanese Snacks were in the range of 41.65-42.25 Bq/L, 1.93-2.26 Bq/L and 34.79-40.31 Bq/L, respectively. Our results showed that the specific activities of radionuclides in those Japanese snacks were lower than the standard value of International Atomic Energy Agency (IAEA) . In conclusion, these Japanese snacks might be safe for human consumption due to undetectable radioactive residue in these product.

Keywords: Gamma spectrometry, Japanese snack, Radionuclide





Development of gamma shielding materials from W₂O₃/carbon black/natural rubber composites

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This research aimed to investigate the gamma shielding, cure characteristics, and mechanical properties of natural rubber (NR) with additions of Tungsten (III) oxide (W₂O₃) and carbon black powders for potential use as flexible, lead-free gamma-shielding materials. The investigated properties included radiation protection properties, cure times, torque differences, and mechanical properties. The results showed that the increases in contents of W₂O₃ from 0 to 100, 300, and 500 parts per hundred of rubber by weight (phr) and the increases in thickness of the composites increased the values of the gamma attenuation coefficients, torque differences, tensile modulus at 100% elongation, and hardness (Shore A), but decreased the values of the tensile strength and elongation at break. However, the increases in W₂O₃ contents had no effect in cure times. All composites also underwent thermal aging tests at 70 °C for 96 hr to investigate the deterioration of mechanical properties with time caused by heat, the results indicated only slight decreases in the overall tensile and elongation properties. Specifically, for the NR composites with 500 phr of W₂O₃, the linear attenuation coefficients (μ_l) were $13.8 \pm 2.3 \text{ m}^{-1}$ for 662-keV gamma rays from Cs-137 and $7.1 \pm 2.5 \text{ m}^{-1}$ for 1.25-MeV gamma rays from Co-60, while the mass attenuation coefficients (μ_m) were $5.6 \times 10^{-3} \pm 0.3 \times 10^{-3} \text{ m}^2/\text{kg}$ for 662-keV gamma rays from Cs-137 and $2.9 \times 10^{-3} \pm 0.3 \times 10^{-3} \text{ m}^2/\text{kg}$ for 1.25-MeV gamma rays from Co-60. When compare the values of μ_m of 500-phr W₂O₃/NR composites and lead sheets, they were comparable, implying possible and effective alternative lead-free gamma shielding materials of W₂O₃/NR composites to lead sheets.

Keywords: Gamma ray/Radiation shielding/Tungsten oxide/Natural rubber





Integrated Analysis Between Protein - Protein Interaction Network and Gene Expression Profile of *Cordyceps militaris*

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Cordyceps militaris is one of entomopathogenic fungi being used as herbal drugs because it produces several bio-metabolites with different pharmaceutically and biologically active functions. Despite the medicinal importance of *C. militaris* as mentioned, there is a relatively little known information about its fundamental process associated in protein-protein interactions (PPI) network which is essential for understanding cell functions and biological processes. Moreover, molecular studies on the developmental stages for both mycelial and fruiting body in *C. militaris* are less focused at systems level. In this study, we aimed to perform integrative data analysis of PPI network and gene expression data for identification of important co-regulated genes associated in mycelial and fruiting body stages. Initially, we constructed PPI network of *C. militaris* based on identification of putative components through comparative genomics and interactomics between *C. militaris* and *Saccharomyces cerevisiae*. To the end, the constructed PPI network was then used for identification of significant co-regulated genes in a transcriptional response to alteration of these two development stages. As a result, the constructed PPI network of *C. militaris* contains 2,880 individual proteins with 85,071 putative interactions of protein pairs. Once integrating with significant gene expression data achieved from the comparative analysis of two development stages (FDR<0.05) and further used Maximum Neighborhood Component (MNC) method for identification of essential *proteins/hubs*, consequently the 50 essential proteins/hubs were significantly found. These proteins/hubs were majorly involved in metabolic and regulation processes, especially regulation of nucleobase, nucleotide, nucleoside and nucleic acid metabolisms. The integrated PPI network with gene expression data of *C. militaris* provides as an important resource for growth and developmental research of this promising fungus.

Keywords: *Cordyceps militaris*, Protein-protein interaction network, Entomopathogenic, Mycelial, Fruiting body





The Inductive Effect of Giant African Snail's Mucus on Mesenchymal Stem Cell Osteogenic Differentiation on Polycaprolactone Biomaterial

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Giant African snail (*Achatina fulica*) is a major pest in Thailand. The mucus of this snail has several medical applications. Previous research found that *A. fulica* mucus stimulated development and mineralization of bone cells, potentially useful in bone repairs. Other studies have revealed effective use of biodegradable and tissue-compatible polycaprolactone (PCL) as a transplant material and components in medical devices. Taken together, this study aimed to examine inductive effect of *A. fulica* mucus on osteogenic differentiation of mesenchymal stem cells (MSCs) grown on the PCL biomaterials. The MSCs were cultured in 6-well plate at a density of 8×10^4 cells per well for 2 weeks in triplicate. The experimental group was treated with frozen-dried mucous powder dissolved in the culture medium at a concentration of 50 $\mu\text{g}/\text{ml}$. The control was grown in the medium alone. The MSCs treated with the snail mucus had higher number of mineralized nodules stained with Alizarin Red S spreading across the PCL and showed cell expanding configuration with better PCL attachment after staining with Coomassie Brilliant Blue. Moreover, expression of osteogenic differentiation-related genes in the MSCs after the mucus treatment for five days, *RUNX2* was upregulated while *OSTERIX* and *SOX9* were down regulated compared to the control. Notably, the expressions of *COL1A1* and *ALP* were undetectable after 2 weeks of the treatment. Thus, our results demonstrated that the snail mucus could induce calcium accumulation, cell expanding and attachment on the PCL of MSCs, molecularly confirmed by osteogenic-related gene expression. The information will be useful for bone engineering and encourage medical use of *A. fulica* mucus.

Keywords: *Achatina fulica*, snail mucus, mesenchymal stem cells, polycaprolactone





A comparative study of different carbon sources on growth and cordycepin production in *Cordyceps militaris*

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Cordyceps militaris is one of the most popular *Cordyceps* species which is widely used for cordycepin (3'-deoxyadenosine) production. However, it is hard to produce large amounts of this bioactive compound for industrial production. To explore cellular responses underlying the cordycepin biosynthesis in *C. militaris* strain CMRU01, a comparative study of the biomass and cordycepin production on different carbon sources i.e. glucose, sucrose and xylose was performed using surface liquid cultivation mode. Among three carbon sources tested, sucrose is the best carbon source for both biomass and cordycepin production in *C. militaris* strain CMRU01 in terms of the highest maximum specific growth rate, biomass and cordycepin productivities. This study provides a useful information for further optimizing the yields and productivities of the cordycepin and the other targeted compounds with industrial interest.

Keywords: *Cordyceps militaris*, Cordycepin production, Biomass, Carbon sources, Surface liquid cultivation





Microscopic structures of the anterior digestive system and accessory organs of the butterfly lizard (*Leiolepis ocellata*)

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The microanatomy of the anterior digestive tract and accessory digestive organs of the butterfly lizard, *Leiolepis ocellata*, was studied. The digestive tract is elongated and consists of 4 concentric tissue layers: the mucosa, submucosa, muscularis and adventitia/serosa, as in other reptiles. The esophagus is lined by a pseudostratified ciliated columnar epithelium with goblet cells. The mucosa extends into the lumen, thus forming longitudinal folds. There are no esophageal glands. The stomach is divided into two histological regions: the anterior and posterior regions. The gastric lining structure is of a simple columnar epithelium. The anterior region is composed of gastric glands, which contain oxynticopeptic cells and mucous cells, while the posterior region contains prominent gastric rugae and mucous gastric glands without oxynticopeptic cells. The tongue is covered by a keratinized stratified squamous epithelium and consists of multidirectional musculatures. In addition, the tongue contains serous glands, mucous glands and cartilaginous axis at the posterior lingual region. The liver comprises 3 incomplete hepatic lobes. The hepatic parenchyma is constituted by two-cell-thick plates flanked by sinusoids. Numerous lipid vacuoles are present in the hepatocyte cytoplasm. The pancreas is divided into two secretory regions: exocrine and endocrine pancreas. The exocrine pancreas houses acini. Acinar cells have the basal nuclei and eosinophilic supranuclear zymogen granules. The endocrine pancreas (islets of Langerhans) is interspersed with the exocrine pancreas. Improved basic knowledge on the digestive structures of the lizard will make a valuable contribution to better understanding the comparative digestive system of reptiles.

Keywords: *Leiolepis ocellata*, microanatomy, digestive system





Effect of p-coumaric acid on brain oxidative status in chronic cerebral hypoperfusion mice

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The present study, aimed to investigate the effect of p-coumaric acid on brain oxidative status in chronic cerebral hypoperfusion mice. Twelve male ICR mice were randomly divided into 3 groups (RCO, RCO25, RCO50). Chronic cerebral hypoperfusion was induced by permanent right common carotid artery (RCO). P-coumaric acid 25 and 50 mg/kg were orally administered for 3 weeks. Brain oxidative status was evaluated by using colorimetric method. The result showed that p-coumaric acid 50 mg/kg significantly decrease calcium level and significantly increase catalase activity. The present study suggest that p-coumaric acid 50 mg/kg ameliorated oxidative status in brain tissue of chronic cerebral hypoperfusion mice.

Keywords: Oxidative status, Brain oxidative status, P-coumaric acid, Chronic cerebral hypoperfusion, ICR mice





Inhibitory effect of Golden apple snail (*Pomacea canaliculata*) extract on *Escherichia coli* and *Staphylococcus aureus* biofilm formation

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Escherichia coli and *Staphylococcus aureus* are recognized as normal flora bacteria in human. However, they can also be opportunistic pathogens and contaminated in medical devices that cause nosocomial infection. These bacteria have ability to form biofilms, surface adhesive community of bacteria, enabling them to survive from immune, antibiotics, and other environmental stresses. Golden apple snail (*Pomacea canaliculata*) is an invasive species that rapidly increase population and cause damages to agricultural products. Recently, there were several reports on antibacterial activities of snail mucus and extract. The study of these extracts against bacterial biofilm formation remains limited. This study aimed to evaluate the inhibition effect of the snail extracts from the golden apple snail on *E. coli* and *S. aureus* biofilm formation. The snail extracts were separated and collected in five fractions by C18-reverses phase HPLC column. Antibiofilm formation activity was tested at 100 µg/ml of the crude extract and their fractions in 96-well microtiter plates for 72 hour at 37°C. Crystal violet (CV) assay was performed to evaluate the bacterial biofilm formation followed by visualization under the inverted microscope and measuring the absorbance at 630 nm. Results indicated that the fractions F2 and F3 had the highest inhibitory effect on biofilm formation. In *E. coli*, the antibiofilm activity of the F2 were dose independent while the IC₅₀ value of the F3 was 241.30 µg/ml. In *S. aureus*, the IC₅₀ value of the F2 and F3 were 30.67 and 324.53 µg/ml. Our research showed potential application of the pest snails as valuable antibiofilm agents.

Keywords: biofilm, snail extract, *Escherichia coli*, *Staphylococcus aureus*, golden apple snail





Effects of p-coumaric acid on hippocampal neuron in chronic cerebral hypoperfusion mice.

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Chronic cerebral hyperpofusion associated with cognitive decline. This study investigated effect of chornic cerebral hypoperfusion and effect of p-coumaric acid on dorsal hippocampal neurons. Thirteen-male ICR mice were randomly divided into 4 groups of Sham, UCO, UCO-PQ25 and UCO-PQ50. The chornic cerebral hypoperfusion caused by permanent right common carotid artery occlusion. UCO-PQ25 and UCO-PQ50 were orally treated with p-coumaric acid after the arterial occlusion for 2 weeks. Histological analysis of the hippocampus was studied by using Cresyl violet staining. The results indicated that the viable cell of dorsal hippocampus in Sham significantly higher than UCO. Viable cell in UCO-PQ25 and UCO-PQ50 were significantly higher than UCO. The persent study concluded that permanent right common carotid artery occlusion induce the reduction of the dorsal hippocampal neurons and p-coumaric acid prevent the reduction of dorsal hippocampal neurons in chornic cerebral hyperpofusion mice.

Keywords: p-coumaric acid, hippocampal neuron, chronic cerebral hypoperfusion





***In silico* Analysis of Thiostrepton Efficacy in FOXM1 Inhibition in Breast Cancer**

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Breast cancer was found to be the highest incidence in female cancer patients in Thailand. This has been reported to be the second cause of the death among Thai women. Breast cancer can be caused by the oxidation by free radicals that can lead to the DNA damage and mutations. These lead to the altering of the cells division and increase cell division rates. Various studies showed that Fork head box M1 (FOXM1) protein has a higher expression in many cancer cells, which is one of the causes of cancer cells progression. FOXM1 was also found to contribute to the chemotherapy resistance in breast cancer. This is because the FOXM1 protein promotes DNA damage repair, especially one from the chemotherapeutic drugs. The purpose of this research is to analyze the inhibition activity of FOXM1 protein by thiostrepton, a natural antibiotic from *Streptomyces*. There was a study that showed thiostrepton plays an important role in the inhibition of FOXM1 expression. Here, Gromacs program was used to analyze, *insilico* (Gromos force field; 53A5/53A6 version). This program is a common tool for analyzing protein drug interaction by molecular dynamics simulation. In this study the molecular interaction between DNA binding domain (DBD) of FOXM1 protein and target DNA was observed in both conditions; with and without thiostrepton. The result showed that there were hydrogen bonds between DBD of FOXM1 protein and the DNA as following; 241-Tyr, 260-Lys, 280-Gly, 283-Asn, 286-Arg, 290-Ser, 292-His, 304-Lys, 306-Ser, 308-Trp, 316-Arg. This study also established the lower number of the hydrogen bonds between FOXM1 protein and DNA after applying thiostrepton. Therefore, this study can be assumed that thiostrepton might have the effect in inhibiting the molecular interaction between DBD of FOXM1 protein and DNA in human breast cancer.

Keywords: FOXM1, Breast cancer, Thiostrepton, Gromacs, DNA binding domain





A study of Thiostrepton antibiotic on FOXM1 inhibition in chemoresistant breast cancer cells

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Breast cancer is one of all dangerous cancer in humans from the studies in many cancer. FOXM1 protein is a major protein involved in cell proliferation. It promotes the cell cycle and incorporates in repairing the DNA damage that caused by chemotherapeutic drug. In addition, FOXM1 induce breast cancer cells to grow rapidly and it has an ability to induce chemotherapy resistance. A thiasole ring containing antibiotic, Thiostrepton, has an ability to inhibit the activity of FOXM1. It has been reported to decrease the binding of FOXM1 to the targeted DNA. The aims of this study were to investigate the cytotoxicity and activity of Thiostrepton in inhibiting the activity of FOXM1 in breast cancer cells. This study also examined the capability in down-regulation of *FOXM1* expression and its activity by Thiostrepton that lead to cell apoptosis and cellular senescence. The cytotoxicity of Thiostrepton was determined using MTT assay in breast cancer cell line (MCF-7). The cytotoxicity effect (IC_{50}) 25 and 50 μ g/ml, at 48 and 72 h, respectively. In addition, results from the study of conventional reverse transcription PCR showed the decrease in the activity of *FOXM1* and the targeted genes.

Keywords: FOXM1, Thiostrepton, Breast cancer, cellular senescence, apoptosis





Study of effect of *Tiliacora triandra* leaf extract and oxidative status in chronic high glucose diet mice

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The present study aimed to investigate the effect of *Tiliacora triandra* leaf extract and oxidative status in chronic high glucose diet mice. Nine male ICR mice were randomly divided into 3 groups of HGD, HGDT300, and HGDT600. High glucose diet (HGD) was induced by orally given of 30% glucose and/or *T.triandra* leaf extract 300, 600 mg/kg for 4 weeks. Oxidative status was evaluated by using colorimetric method. The result showed that *T.triandra* leaf extract has different effect on different organ tissue oxidative status. *T.triandra* 600 mg/kg significantly increased calcium level in brain, while significantly decreased in kidney. Moreover, *T.triandra* 600 mg/kg significantly decreased superoxide dismutase (SOD) in brain and significantly increased catalase in stomach. The present study suggested that *T.triandra* 600 mg/kg increased brain oxidation, but reveals ameliorate effect in other organ tissue such as kidney and stomach in chronic high glucose diet mice.

Keywords: hyperglycemia; oxidation stress; *Tiliacora triandra*; antioxidant

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Effect of p-coumaric acid on anxiety in chronic cerebral hypoperfusion mice.

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Anxiety is a nervous disorder characterized by a state of excessive uneasiness and apprehension, typically about an imminent event or something with an uncertain outcome. The exact cause of anxiety disorders is unknown, but in some study suggested that a chronic cerebral hypoperfusion may lead to an anxiety disorder. An excessive or persistent state of anxiety can have a devastating effect on the physical and mental health of living being. The present study investigated the effect of a chronic cerebral hypoperfusion and p-coumaric acid on anxiety in mice. Male ICR mice were divided into 2 groups of normal (SHAM) and chronic cerebral hypoperfusion mice cause by permanent unilateral common carotid artery occlusion (UCO). These 2 groups were further subdivided into 3 groups of Control (2% DMSO), 25 mg/kg p-coumaric acid, and 50 mg/kg p-coumaric acid. Treatment was orally administered 1 week after chronic cerebral hypoperfusion induction in both groups for 3 days. The anxiety evaluation was performed by elevated plus maze (EPM) for 7 days. The result revealed no significant difference on anxiety between normal and chronic cerebral hypoperfusion mice groups but found a significant difference in chronic cerebral hypoperfusion mice treated with 25 mg/kg p-coumaric acid. The result showed that a chronic cerebral hypoperfusion caused by permanent unilateral common carotid artery occlusion has no effect on anxiety in male ICR mice, but 25 mg/kg p-coumaric acid revealed anxiolytic effect in chronic cerebral hypoperfusion mice.

Keywords: Mice behavior, Anxiety, p-coumaric, Plus maze, Chronic cerebral hypoperfusion





Comparative image analysis of mucous tissues in *Achatina fulica* and *Pomacea canaliculata*

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Giant African snails (*Achatina fulica*) and golden apple snails (*Pomacea canaliculata*) are important agricultural pests in Thailand. Both snails have been found to produce mucus which may be useful for medical or cosmeticeutical applications. The aim of the study was to develop a procedure for mucus productivity evaluation in different age of both species. The digital pictures of mucosal tissue stained with alcian blue (AB) and Periodic acid Schiff (PAS) dyes were taken from both species during the age of 1 month, 2 months and 3 months at 10x 20x and 40x objective lens magnification. The stained mucosal areas were measured by manual and automate methods of ImageJ program. The results showed that 20x magnification objective lens was the most proper for data acquisition and analysis. Notably, the mucus productivity from foot and mantle in giant African snails was much higher compared to golden apple snails and significantly positive correlated with age. The automate method was able to estimate the percentage of mucous per mucosal area significantly higher than the manual method. Our results recommend that the mucus from giant African snails at higher age should be considered as the proper source for the commercial snail mucus production. This research can be developed further to enhance the application of snail pests in the most advantage of sustainable ways.

Keywords: *Achatina fulica*; *Pomacea canaliculata*; image analysis; mucous tissue; snails





Study on the Genetic Diversity of five wedge bivalve species from Thailand.

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Wedge shell or *Donax* is under family Donicidae, marine bivalve mollusks worldwide distributed intertidal sandy beaches. Members of *Donax* are similar in shell morphology, triangular hard shell with the variability in pattern and color. Five *Donax* species were collected from Thailand, including *Donax tinctus* Gould, *Donax faba* Gmelin, *Donax incarnatus* Gmelin, *Donax (Deltachion) semigranosus* Dunker and *Donax scrotum* Linnaeus that was focused on the study of the Genetic Diversity by molecular biological analysis. DNA was extracted by soft tissue and DNA extraction was performed with GF-1tissue (DNA extraction, vivantis). Then DNA extraction was carried out prior to PCR amplification using universal primer for Cytochrome c oxidase I (COI) gene. The nucleotide sequences of COI gene were aligned against *Donax* five species using Clustal Omega program. The result showed significant mitochondrial genetic different within genus level, sequences that are proximity to genus level and quey cover are in range of 43-92% similarity.

Keywords: *Donax*, Cytochrome c oxidase I (COI), Class bivalvia, Thailand, Genetic variation



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