# ABSTRACTS AND PROGRAM


## Organized by

**School of Science, KMITL**

**School of Agricultural Technology, KMITL**

**Chinese Society for Microbiology**

**Heilongjiang Province Society of Microbiology**

**Heilongjiang Province Society of Bionic**

**Harbin Institute of Technology**

# THE 18th INTERNATIONAL SYMPOSIUM ON BIOCONTROL AND BIOTECHNOLOGY

## ABSTRACTS AND PROGRAM

**June 16-17, 2022**

**via Zoom**

***Organized by***

**School of Science, KMITL**

**School of Agricultural Technology, KMITL**

**Chinese Society for Microbiology**

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**Heilongjiang Province Society of Bionic**

**Harbin Institute of Technology**

# Message from Acting President of King Mongkut's Institute of Technology Ladkrabang

Dear Distinguished Invited Speakers, Participants, Ladies and Gentlemen,

As the Acting President of King Mongkut's Institute of Technology Ladkrabang or KMITL, it is my great honor to welcome the invited speakers and all participants who attend the 18th International Symposium on Biocontrol and Biotechnology or ISBB 2022. The first symposium was organized in 2002 by the collaboration between Harbin Institute of Technology and KMITL. Since then, there have been a steady growth in the number of institutions, universities, and partners for the organization of the symposium from China, Egypt, India, Poland, the Philippines, and Thailand. Along the way, several remarkable collaborations were created between researchers who attended the symposia. All of these have contributed to the progress of knowledge in the field of biocontrol and biotechnology which play crucial parts in the advance of technology and the improvement of human's life and the environments as a whole.

The 17th symposium on Biocontrol and Biotechnology was jointly organized with the 3rd International Symposium on Agricultural Technology by the School of Agricultural Technology at KMITL on July 2nd – 5th, 2019. This symposium was originally planned to be held in 2020. Unfortunately, because of the COVID-19 pandemic, the symposium has to be postponed for two years. From late 2021 to early 2022, the School of Science and the School of Agricultural Technology at KMITL, the Chinese Society for Microbiology, Heilongjiang Province Society of Microbiology, Heilongjiang Province Society of Bionic and Harbin Institute of Technology from the People Republic of China have come together to make ISBB 2022 possible.

ISBB 2022 comprises of two full days of academic activities. We are honored to have our special invited speakers to share with us on several topics in the field of biocontrol and biotechnology. There are also have presenters from many institutions and universities to share with us their wonderful experiments and discoveries. Additionally, the symposium provides a special opportunity to meet and greet with other scientists. I believe that this will be the first step for finding new friends and building new collaborations. I wish all participants the fruitful outcomes during their attendance at ISBB 2022 and hope that we will meet again in the 19th Symposium on Biocontrol and Biotechnology in the very near future.

Sincerely Yours.

Associate Professor Anuwat Jangwanitlert, Ph.D.,
Acting President of
King Mongkut's Institute of Technology Ladkrabang

# Message from Dean of School of Science, King Mongkut's Institute of Technology Ladkrabang

Dear Distinguished Invited Speakers, Participants, Ladies and Gentlemen,

It gives me a great pleasure to have the opportunity to welcome all participants to the 18th International Symposium on Biocontrol and Biotechnology or ISBB 2022. Despite the COVID-19 pandemic, scientists and researchers around the world are still working on their remarkable projects to advance our knowledge in the field of biocontrol and biotechnology. A significant factor for such achievements is the domestic and international collaborations between laboratories, research units, institutions and universities, and all ISBB symposia, since the first one in 2002, were organized for exactly this reason.

For this year ISBB 2022, we have high-school, undergraduate and graduate students, young and senior researchers as well as prestigious invited speakers to share with us their discoveries and experiences in various fields of biocontrol and biotechnology. The topics range from the discovery of important genes and useful molecules in microbes, the application of plant bioactive compounds for pest management, the synthesis of new materials for environmental solutions to the innovation of biotechnological products. I believe that this symposium is a great opportunity for all of us to learn and exchange our knowledge. Additionally, it provides us a platform to form new friendships and collaborations for future exciting scientific endeavors. All of these will ultimately lead to the development of new technologies to better our life and solve our facing environmental issues.

On behalf of the School of Science at KMITL, I would like to express my sincere gratitude to KMITL's School of Agricultural Technology, the Chinese Society for Microbiology, the Heilongjiang Province Society of Microbiology, the Heilongjiang Province Society of Bionic and Harbin Institute of Technology for their continuous support and help that have made ISBB 2022 possible. I hope that the symposium will be beneficial to all participants and contribute to the success of their career in the future.

Sincerely Yours,

Associate Professor Sutee Chutipaijit, Ph.D.,
Dean of Science School

# About the symposium

The International Symposium on Biocontrol and Biotechnology was first held in 2002 at Harbin Institute of Technology (HIT), P.R. China to provide an opportunity for students, scientists, and researchers to exchange and share their knowledge in the field of biocontrol and biotechnology. Since then, the symposia were held regularly by many universities and institutions in various countries. King Mongkut's Institute of Technology Ladkrabang (KMITL), Bangkok, Thailand and Huazhong Agricultural University, Wuhan, P.R. China, also co-hosted the 2nd and 3rd symposia in 2003 and 2005, respectively.

In 2006, Lady Doak College, Madurai, India was the host of the 4th symposium. This was followed by the 5th symposium that was co-organized by KMITL and Khon Kaen University, Nongkhai Campus, Thailand, in 2007. Luzon State University, Philippines, HIT and KMITL were the co-host of the 6th symposium in 2008. Subsequently, the 7th symposium was held in 2009 by HIT, Weihai Campus, P.R. China. KMITL hosted the 8th Symposium in 2010. This was followed by the 9th symposium, in 2011, that was co-organized by the Institute of Tropical Biology Technology of the Chinese Academy Tropical Sciences, Hainan University, State Key Laboratory of Agricultural Microbiology and National Engineering Research Center of Microbial Pesticides, P.R. China.

The 10th symposium was organized by HIT in 2012. The 11th symposium was co-hosted by Alexandria University, Biblotheca Alexandrina and the Egyptian Development & Technology Transfer Association, Egypt, HIT, P.R. China and KMITL in the following year. The 12th symposium was held in Thailand with the Faculty of Science, the Faculty of Agricultural Technology and KMITL's Prince of Chumphon Campus as the host in 2014. HIT Shen Zhen Campus, P.R. China, was the host of the 13th symposium on November 6th – 8th, 2015. In the following year, the 14th symposium was held on November 6th – 9th, 2016 in Saint-Petersburg, Russian Federation by All-Russian Institute of Plant Protection, All-Russian Institute of Agricultural Microbiology in collaboration with HIT and KMITL.

The 15th symposium was hosted by Alexandria University at Hurghada, Egypt on October 17th – 20th, 2017 with KMITL as a co-host. The 16th symposium was organized by HIT in Beijing under the International Congress of Biological Control in 2018. The School of Agricultural Technology, KMITL, hosted the 17th symposium jointly with the 3rd International Symposium on Agricultural Technology (ISAT 2019) in Thailand, on July 2nd – 5th, 2019.

The symposia provide continuous opportunities for undergraduate students, graduate students, researchers and scientists in the field of biocontrol and biotechnology to collaborate and interact with each other to promote and advance our knowledge in the field. Many successful partnerships and collaborations were also established from the meetings of the participants of the symposia.

The 18th International Symposium was previously planned to be hosted in Poland by Warsaw University of Life Science. However, the symposium has to be postponed for two consecutive years because of the COVID-19 pandemic. Therefore, the School of Science and the School of Agricultural Technology at KMITL, in collaboration with the Chinese Society for Microbiology, the Heilongjiang Province Society of Microbiology, the Heilongjiang Province Society of Bionic and Harbin Institute of Technology, have decided to jointly organize the 18th International Symposium on Biocontrol and Biotechnology online via Zoom during June 16th – 17th, 2022. The 19th International Symposium will be hosted in Poland when the COVID-19 situation is resolved.

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# Organizing Committee

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# Program schedule

## The 18th International Symposium on Biocontrol and Biotechnology (ISBB2022)

## June 16-17, 2022 (Online conference via ZOOM)

(Time indicated here is GMT+7)

| | |
|---|---|
| ***Day 1; June 16, 2022*** | |
| 8:30 – 9:00 | Participant joining via zoom |
| **Opening session 9:00 – 9:30**<br>**Master of Ceremony:** Dr. Bunyarit Meksiriporn | |
| 9:00 – 9:10 | Report and welcome address<br>by Assoc. Prof. Dr. Sutee Chutipaijit, Dean of School of Science, King Mongkut's Institute of Technology Ladkrabang |
| 9:10 – 9:20 | Welcome address<br>by Prof. Liu Hong, Vice President of Harbin Institute of Technology |
| 9:20 – 9:30 | Opening address<br>by Assoc. Prof. Dr. Anuwat Jangwanitlert, Acting President of King Mongkut's Institute of Technology Ladkrabang |
| **Plenary session I 9:30 – 12:00**<br>**Chairperson:** Assoc. Prof. Dr. Chokchai Kittiwongwattana | |
| 9:30 – 10:30 | **Topic I**: "Control of plant and food pathogens by vapor-phase vinegar"<br>by Prof. Dr. Warawut Krusong, School of Food Industry, King Mongkut's Institute of Technology Ladkrabang, Thailand |
| 10:30 – 11:30 | **Topic II**: "An engineered genetic selection strategy to uncover productive binders against difficult-to-drug targets"<br>by Dr. Bunyarit Meksiriporn, School of Science, King Mongkut's Institute of Technology Ladkrabang, Thailand |
| 11:30 – 12:30 | **Topic III**: "COVID-19 mRNA vaccine development: ChulaCov19"<br>by Prof. Dr. Kiat Ruxrungtham, Chula Vaccine Research Center (ChulaVRC), School of Global Health, Faculty of Medicine, Chulalongkorn University, Thailand |
| 12:30 – 13:00 | Lunch break |
| **Plenary session II 13:00 – 15:00**<br>**Chairperson:** Dr. Bunyarit Meksiriporn | |

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| 13:00 – 14:00 | **Topic IV**: "Extension of the biotechnique for recycling the waste from potato starch industry"<br>by Prof. Dr. Yang Qian, School of Life Science and Technology, Harbin Institute of Technology, P.R. China |
| 14:00 – 15:00 | **Topic V:** "Biological control of erophyoid mites and their use as biological agents for weed control"<br>by Assoc. Prof. Dr. Mariusz Lewandowski, Department of Plant Protection, Institute of Horticultural Sciences, Poland |
| 15:00 – 15:15 | Session break |
| **Oral session I** 15:15 – 16:45<br>**Chairperson:** Asst. Prof. Dr. Wipawee Dejtisakdi | |
| 15:15 – 15:45 | **Topic I:** "Production of microbial biocontrol agents from *Bacillus thuringiensis* for targeting plant-parasitic nematodes"<br>By Mr. Pasin Jammor |
| 15:45 – 16.15 | **Topic II:** "Effects of fermentation on black soldier fly as animal feed additive"<br>by Dr. He Liu |
| 16.15 – 16.45 | **Topic III** "Study and utilization of chlamydospores of *Trichoderma*"<br>by Prof. Li Mei |
| ***Day 2; June 17, 2022*** | |
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| 8:30 – 9:00 | **Topic I:** "Chemical analysis and bioactivity of essential oil from *Salacca wallichiana* Mart.: herbicidal, antibacterial and antioxidant activity"<br>by Mr. Nutcha Manichart |
| 9:00 – 9:30 | **Topic II:** "From an in-depth interview on pesticide use behavior to the development of a pesticide residue test kit"<br>by Mr. Kotchakorn Krikaew |
| 9:30 – 10:00 | **Topic III:** "Improvement of *Cananga odorata* essential oil-based pre-emergence bioherbicide using an ultrasonic emulsification method: bioherbicidal activity against *Amaranthus tricolor*"<br>by Ms. Naphat Somala |

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| 10:00 – 10:30 | **Topic IV:** "Impact of degraded agrochemical and heavy metal bio-products on soil microbiota"<br>by Ms. Paweena Tamsamear |
| 10:30 – 11:00 | **Topic V:** "Synbiotic beverage production from mangosteen juice mixed with yam bean juice using *Lactobacillus casei* 431"<br>by Ms. Yuwadee Sinlapajan |
| 11:00 – 11:30 | **Topic VI:** "Characteristics and antioxidant activities of kombucha from black tea and roselle by a mixed starter culture"<br>by Ms. Tanyarat Sutthiphatkul |
| 11:30 – 12:00 | **Topic VII:** "Effect of bioproduct for degraded chemical residues on inhibited *Sclerotium rolfsii* cause root and stem rot of green oak"<br>by Ms. Warisara Surattaseranee |
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| 8:30 – 9:00 | **Topic I:** "Nanoporous carbon derived from oil palm leaves via hydrothermal carbonization for adsorbent"<br>by Mr. Sirayu Chanpee |
| 9:00 – 9:30 | **Topic II:** "Synthesis of nanoporous carbon derived from vinasses wastes via carbonization for adsorbent"<br>by Ms. Phetcharat Nenyoo |
| 9:30 – 10:00 | **Topic III:** "Nanoporous carbon derived from lignin extracted from black liquor via pyrolysis carbonization for catalytic support"<br>by Mr. Tassanai Tempiam |
| 10:00 – 10:30 | **Topic IV:** "Development of carbamate and organophosphate residue detection kits in durian"<br>by Ms. Ponchanok Datmanee |
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| 13:15 – 13:30 | **Title:** "Actinomycetes from organic rice field soil, their antagonistic activity and antimicrobial activity"<br>by Dr. Khanungkan Klanbut |
| 13:30 – 13:45 | **Title:** "Production of yeast extract powder from spent yeast of bioethanol plant and its potential use as a nitrogen source in fermentation"<br>by Ms. Sikarinthan Lhakot |
| 13:45 – 14:00 | **Title:** "Quality control of six ingredients in Prasamawaeng traditional anti-cough formula using thin-layer chromatography technique"<br>by Ms. Phatchada Chunhavacharatorn |
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| 14:15 – 14:30 | **Title:** "Medium effects on antifungal activity and biosynthetic gene expression of *Bacillus velezensis* 2211, a plant-growth-promoting bacterium"<br>by Assoc. Prof. Dr. Chokchai Kittiwongwattana |
| 14:30 – 14:45 | **Title** "Solid phase extraction with the UHPLC–MS/MS for detection of tetracyclines antibiotics residues in composting system of black soldier fly larva"<br>by Ms. Xia Yang |
| 14:45 – 15:00 | Session Break |
| **Closing Session 15:00 – 15:30**<br>**Master of Ceremony:** Dr. Bunyarit Meksiriporn | |
| 15:00 – 15:15 | Awards ceremony for best oral and poster presentation and<br>Announcement of the host for the 19th International Symposium<br>by Assoc. Prof. Dr. Mariusz Lewandowski, Department of Plant Protection, Institute of Horticultural Sciences, Poland |
| 15:00 – 15:30 | Closing speech<br>by Assoc. Prof. Dr. Sutee Chutipaijit, Dean of School of Science, King Mongkut's Institute of Technology Ladkrabang |

# Medium effects on antifungal activity and biosynthetic gene expression of *Bacillus velezensis* 2211, a plant-growth-promoting bacterium

Nantawan Niemhom[1], Chokchai Kittiwongwattana[2,*]

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[2] Department of Biology, School of Science, King Mongkut's Institute of Technology Ladkrabang, Bangkok, Thailand

## Abstract

Members of the genus *Bacillus* produced a diverse group of antimicrobial compounds. Here, we present the antifungal activity and genome sequence analysis of *Bacillus* sp. 2211, a potential plant-growth-promoting bacterium. Cell-free supernatants from strain 2211 cultures in nutrient broth (NB) and potato dextrose broth (PDB) suppressed the mycelial growth and spore germination of *Pyricularia oryzae*, *Colletotrichum aenigma* and *Colletotrichum fructicola.* However, the supernatant from PDB displayed a significantly higher inhibition activity than NB. Additionally, the supernatant from PDB significantly reduced the disease severity caused by *P. oryzae* on rice seedlings. The genome of strain 2211 was sequenced. The highest digital DNA-DNA hybridization (80.1%) and average nucleotide identity (97.57%) levels indicated that strain 2211 was a member of the species *Bacillus velezensis*. The phylogenomic analysis showed that it clustered with *B. velezensis* NRRL B-41580$^T$, *B. velezensis* KACC 13105 and *B. velezensis* subsp. *plantarum* FZB42. The gene expression analysis showed that *fenA* and *bmyA*, fengycin and bacillomycin D biosynthesis genes, respectively, were significantly up-regulated in PDB, compared to NB. This work demonstrated that the culture media may affect the expression of biosynthetic genes and consequently the antagonistic activity of strain 2211.

**Keywords**: *Bacillus,* antagonistic activity, *Pyricularia oryzae, Colletotrichum aenigma, Colletotrichum fructicola.*

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# Cytotoxicity effect of acetone extracts from *Physalis angulata* L. fruits on acid-fast *Mycobacterium tuberculosis* $H_{37}Ra$ relation with lung cancer cell (NCI-H 187)

Thanavadee Boonchaidee[1,*], Kamonaphach Nantakarn[1], Sirapat Akkhosuwan[1], Siriporn Bumpenphol[1]

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## Abstract

This study interested to evaluate DPPH antioxidation and cytotoxicity effect of acetone extracts from *Physalis angulata* fruit using ultrasonic extraction at 40°C for 30 mins. The cytotoxicity activities were tested with *Mycobacterium tuberculosis* (pathogen of tuberculosis) and *Klebsiella pneumonia* (pathogen of pneumonia disease), *Magnaporthe grisea* (pathogen of rice blast disease), *Alternaria brassicicola* (pathogen of black spot disease), lung cancer cell (NCI-H187), hepatoma cancer cell (HepG2), oral cancer cell (KB) and colon cancer cell (CaCo2) *in vitro*. It was found that, DPPH scavenging of *n*-hexane extracts of *P. angulata* were not significantly different from ascorbic activity ($p < 0.05$). $IC_{50}$ of *n*-hexane extracts of roots, stems, fruits, leaves and ascorbic acid were 0.09 ± 0.03, 0.12 ± 0.05, 0.10 ± 0.02, 0.11 ± 0.01 and 0.03 ± 0.00 µg $mL^{-1}$. DPPH activity of acetone extracts of stems and methanol extract of stems and leaves were significant difference with ascorbic activity ($p < 0.05$). Acetone extracts exhibited the highest antimicrobial effect on *M. tuberculosis* than antimicrobial effects on *M. grisea, K. pneumonia* and *A. brassicicola.* $IC_{50}$ of antimicrobial activities were 5.19 ± 0.19, 10.00 ± 0.00, 35.71 ± 0.00 ug/ml and 50.00 ± 0.00 ug $ml^{-1}$, respectively. The cytotoxicity effect against NCI-H187 cell exhibited significant difference with HepG2, KB and CaCo2 cell cytotoxicity ($p < 0.05$) and $IC_{50}$ were 7.36 ± 0.14, 22.73 ± 0.00, 28.08 ± 0.44 and 28.13 ± 0.63 ug $mL^{-1}$, respectively. This study suggests that phytochemicals of *P. angulata* fruits have an amphiphile property for lipid solubility. High lipid levels are susceptible to pathogen infections and cancer diseases.

**Keywords**: *Physalis angulata* fruit, acid fast bacteria, *Mycobacterium tuberculosis*, high lipid levels, tuberculosis, lung cancer

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# Nanoporous carbon from oil palm leaves via hydrothermal carbonization for adsorbent

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## Abstract

In the present study, nanoporous carbon (NPC) was completely obtained from oil palm leaves via hydrothermal carbonization (HTC). This research studied the effect of hydrothermal temperature from 160-200 °C and reaction time for 6-24 hours. Afterwards, carbonization was obtained at the temperature of 600-900 °C for 1 hour under nitrogen atmosphere for developing the porosity and even removing the contaminants of hydrothermal char to obtain the porous carbon. The physico-chemical properties of nanoporous carbon were comprehensively characterized through scanning electron microscope (SEM), Fourier transforms infrared (FT-IR), Raman spectroscopy, X-ray diffraction (XRD), CHN elemental analyser, and BET analysis. The sponge-liked structure, functional group, amorphous matrials, carbon content, adsorption capacity, and surface area of nanoporous carbon from oil palm leaves were increased with increasing hydrothermal carbonization temperature and time. HTC at 200 °C for 12 hours and carbonization at 900 °C for 1 hour under nitrogen flow are the optimum condition for the synthesis of starting materials for good adsorbent.

**Keywords**: Oil palm leaves; Hydrothermal carbonization; Nanoporous carbon; Adsorbent

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# Synthesis of nanoporous carbon derived from vinasses wastes via pyrolysis carbonization for adsorbent

Phetcharat Nenyoo[1], Atthapon Srifa[2], Worapon Kiatkittipong[3], Apiluck Eiad-Ua[4,*], Suttichai Assabumrungrat[1]

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## Abstract

The vinasse waste is a large byproduct of the sugar or ethanol industry. This waste is acidic, thus affecting soil and water quality. Therefore, it is essential to dispose of this waste to prevent any environmental impact. In this study, nano porous carbon was synthesized from vinasses waste via pyrolysis carbonization and chemical activation process for adsorbent. This research studied the effect of potassium hydroxide (KOH) concentration by a mass ratio from 1 to 3 at activation temperature from 600 °C to 900 °C for 1 hour under the nitrogen atmosphere to improve the nano porous structure. The physical and chemical properties of nano porous carbon was characterized by Energy Dispersive X-Ray Spectroscopy (EDS), thermogravimetric analysis (TGA), X-ray fluorescence (XRF), infrared spectroscopy, Raman spectroscopy and Nitrogen sorption isotherm, respectively. Activation at temperature 900 °C and the 1:1 activator (KOH)- precursor ratio provided the maximum surface area 1,018 m2/g which was suitable to be applied in making an adsorbent. The adsorbent is used for Tetracycline adsorption application.

**Keywords**: Vinasses; Nano Porous Carbon; Chemical Activation; Pyrolysis Carbonization; Adsorbent

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# Nanoporous carbon derived from lignin extracted from black liquor via pyrolysis carbonization for catalytic support

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## Abstract

In the chemical pulping industry, black liquor is the byproduct from kraft process. This waste can reduce oxygen availability and negatively affect water resources. Therefore, it is significant to seek a viable, economical and environmentally friendly method to utilize and dispose of lignin from black liquor. In this work, lignin was extracted from black liquor with potassium alum solution by precipitation. Nano porous carbon was synthesized from the lignin via carbonization assisted chemical activation process for catalytic support. This research studied the effect of potassium hydroxide (KOH) concentration by a mass ratio from 0.5 to 1.5 at activation temperature from 600 °C to 900 °C for 1 hour under the nitrogen atmosphere to improve the nano porous structure. The physical and chemical properties of nanoporous carbon was investigated by field-emission scanning electron microscope (FESEM), X-ray diffractometer (XRD), thermogravimetric analysis (TGA), X-ray fluorescence spectroscopy (XRF), Raman spectroscopy, and Nitrogen sorption isotherm, respectively. Activation temperature at 700 °C and the 1:1 activating agent (KOH)- precursor ratio provided the maximum surface area of 1,676.5 m2/g and a total pore volume of 1.091 cm3/g, which was a suitable condition for the excellent catalytic support. The catalyst is used in the transesterification reaction for green diesel applications.

**Keywords**: Nano Porous Carbon; Black liquor; Chemical Activation; Carbonization; Catalytic Support

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# Synbiotic beverage production from mangosteen juice mixed with yam bean juice using *Lactobacillus casei* 431

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## Abstract

Mangosteen (*Garcinia mangostana* L.) is a fruit plant that grows in Southeast Asia. The fruit pulp is slightly sour and sweet. In this study, mangosteen juice was fermented with probiotic bacterium *Lactobacillus casei* 431, and the physicochemical, antioxidant activity and sensory and survival characteristics of samples during the fermentation process were investigated. The yam bean gave the higher growth of this bacterium at 37 °C for 15 hours (6.98 log CFU $mL^{-1}$) than mangosteen juice and pineapple juice and was used prepare the inoculum. The synbiotic beverage was prepared by blending mangosteen juice with yam bean juice and pineapple juice for substrate in various ratios, using 10%, 20%, and 30% of inoculum. The mixture of mangosteen juice, yam bean juice, and pineapple juice was 80:10:10, and using a 30% inoculum for 10 hours resulted in the highest viable cell counts of probiotic (7.89 log CFU $mL^{-1}$). The total acidity and pH slowly changed to 0.58±0.00% and 3.27±0.01, respectively. Both total phenolic content and antioxidant activity increased slightly after 10-20 hours of fermentation time. The highest antioxidant activity with the DPPH value was 93.68±0.01%. The mixed mangosteen beverage was kept at 4-10 °C for 30 days. The pH, total sugar, antioxidant activity, and phenolic compound of this beverage slightly decreased from the initial product. Cell viability was reduced and remained 6.02±0.04 Log CFU $mL^{-1}$ on day 5 of the storage period. The fermented juice was shown to be an acceptable and stable product during storage.

**Keywords**: *Lactobacillus casei*, Mangosteen juice , Synbiotic

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# Improvement of *Cananga odorata* essential oil-based pre-emergence bioherbicide using an ultrasonic emulsification method: bioherbicidal activity against *Amaranthus tricolor*

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## Abstract

Essential oil (EO)-based bioherbicide is widely used for sustainable management of weeds. EO from *Cananga odorata* flowers was prepared into nanoemulsion and evaluated for its pre-emergence herbicidal activity on Amaranthus tricolor seeds. In this study, *C. odorata* EO was developed into an ultrasonic nanoemulsion formulation with non-ionic surfactant Tween 80 (polysorbate 80) and DI water. The effects of sonication amplitude (20 – 60 %) and sonication time (2 – 10 min) were evaluated and optimized on droplet size and polydispersity index (PI) of oil-in-water (O/W) nanoemulsion. The smallest droplet size of the nanoemulsion with 44 nm (PI 0.219) in diameter was formulated by the ultrasonic emulsification method at a sonication amplitude of 40% to for 6 min. 21 days after preparation. Droplet size of the nanoemulsion showed 130.6 nm when stored at room temperature. The herbicidal activity of the *C. odorata* nanoemulsion was evaluated against A. tricolor compared to the coarse emulsion. The coarse emulsion and nanoemulsion at different concentrations (62.5, 125, 250 and 500 µL $L^{-1}$ of EO) reduced seed germination and seedling growth in dose-response. This study presented that the nanoemulsion has high effectiveness than the coarse emulsion at the same concentration. The percentage of seed germination inhibition was 100% and 63.75% when treated with 250 µL $L^{-1}$ of EO of the nanoemulsion and coarse emulsion, respectively. A decrease in seed imbibition and α-amylase activity represented a disruption of the germination process. Hence, the ultrasonic nanoemulsion of *C. odorata* EO could be used as a bioherbicide candidate in green agricultural systems.

**Keywords**: *Cananga odorata*, weed control, allelopathic plant, natural herbicide, ultrasonic nanoemulsion, Tween 80

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# Chemical analysis and bioactivity of essential oil from *Salacca wallichiana* Mart.: herbicidal, antibacterial and antioxidant activity

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## Abstract

Salak (*Salacca wallichiana* Mart.) essential oil (EO) was identified by GC–MS for its chemical constituents. Its biological activities (herbicidal, antibacterial and antioxidant) were studied and reported for the first time. The main compound of EO was methyl salicylate. In the herbicidal experiment, the oil was evaluated for its effect on *Echinochloa crus–galli* and *Amaranthus tricolor* by a Petri dish method. The 100% essential oil at 6 µL was added to each Petri dish without direct contact with the weed seeds. The results showed that the EO significantly reduced germination and seedling growth of both weeds. However, inhibitory effects on *A. tricolor* were higher than on *E. crus–galli*, 10 – folds higher. Foliar spray, bioassays were performed by preparing EO solution with different concentrations. At 8% of EO, both weed seedlings were highly susceptible to foliar spray within three days after treatment, suggesting a potent allelopathic potential for the evaluated oil. Antioxidant activity was assessed using a DPPH assay; EO showed weak scavenging activity with >20,000 ppm $IC_{50}$ compared with standard BHT ($IC_{50}$ of 25.23 ppm). The antibacterial activities of pure EO were tested using the disc diffusion method. The zone of 100% essential oil inhibition against bacterial test strains at 6 µL per disc showed *Escherichia coli* TISTR 780 (0.79 ± 0.08 mm) and *Staphylococcus aureus* TISTR 1466 (2.39 ± 0.09 mm). At the same time, the zone of inhibition of standard antibiotic amoxicillin was 1.92 ± 0.11 mm and 3.38 ± 0.17 mm in *E. coli* and *S. aureus*, respectively. The obtained results provided valuable information about the possible oil from *S. wallichiana* contributing to the excellent bioactivity for sustainable agricultural applications.

**Keywords**: essential oil, *Salacca wallichiana*, herbicidal activity, antibacterial activity, antioxidant activity

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# Production of microbial biocontrol agents from *Bacillus thuringiensis* for targeting plant-parasitic nematodes

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## Abstract

Recently, many plant-parasitic nematodes (PPNs), one of the major agricultural pests in Thailand, have been reported to cause significant damages to different parts of agricultural crops. Moreover, the use of chemical nematicides is under certain concerns since they can cause several adverse effects to environment and non-target organisms. Therefore, the development of alternative treatments of chemical nematicides is highly crucial for PPNs control. Nematicidal crystal proteins produced by *Bacillus thuringiensis* (Bt), such as, Cry5B, Cry6A and Cry21A, have been used as a bionematicide due to their capability to intoxicate a wide range of free-living nematodes. Unfortunately, these proteins are susceptible to several environmental conditions including UV radiation, high temperature, and proteolytic degradation as opposed to chemical nematicides. Accordingly, nanotechnology has been used to overcome formulation instability and to deliver the nematicidal proteins to target sites. Therefore, this study aims to produce Cry5B protein as a bionematicide to control PPNs population together with enhanced protein stability and nematicidal efficiency through the development of Cry5B-loaded nanoparticles either with silica nanoparticles (SiNPs) or sulfur nanoparticles (SNPs). The nanoparticles are synthesized via green synthesis method by using fungal filtrates of nematode-trapping fungi, *Arthrobotrys oligospora*, as a capping agent. The biophysical properties of the nanoparticles were characterized using Field Emission Scanning Electron Microscopy (FESEM), Energy Dispersive X-ray Spectroscopy (EDX) and Dynamic Light Scattering (DLS). In addition, nematode pathogenic analysis using *Caenorhabditis elegans* as a model organism will be investigated to understand the intracellular mechanism underlying the nematicidal activity of the free Cry5B protein and Cry5B- loaded nanoparticles.

**Keywords**: Plant-parasitic nematodes, *Bacillus thuringiensis*, Cry5B, Nanoparticles, *Caenorhabditis elegans*

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# Peanut shell nanoporous carbon from hydrothermal-carbonization assisted KOH activation for glyphosate adsorption

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## Abstract

The objective of this study was to optimize the adsorption performance of nanoporous carbon derived from peanut shells by hydrothermal-carbonization assisted KOH activation. The process of deriving nanoporous carbon from raw peanut shells was performed at 200°C for 4-24 hours (Hydrothermal). The nanoporous carbon was then activation with KOH at ratio of 1:1 for 24 hours. Finally, hydrochar was pyrolyzed at 600-900 °C for 1 hour (Carbonization) under a nitrogen gas atmosphere. The nanoporous carbon products were comprehensively characterized by their relevant physical and chemical properties that are important for their novel utilization, using a Scanning Electron Microscope (SEM), an X-ray Diffractometer (XRD), thermogravimetric analysis (TGA), and Fourier-Transform Infrared Spectroscopy (FTIR). The optimum conditions of hydrothermal carbonization were 200°C for 12 hours. The carbonization at 800°C for 1 hour was suggested for the synthesis of nanoporous carbon effective for glyphosate removal from wastewater. The physical-chemical properties and the performance of the nanoporous carbon are presented and discussed.

**Keywords**: Peanut shell, Hydrothermal carbonization, Nanoporous carbon, Glyphosate adsorption

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# Characteristics and antioxidant activities of kombucha from black tea and roselle by a mixed starter culture

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## Abstract

Kombucha is a functional beverage fermented by the symbiosis of bacteria and yeasts and comprised mainly of bioactive compounds and acids that benficial effects on health benefits. Our research aims to improve the consistency of kombucha quality so that it can be used on a mass production. In this study, the isolated bacterium *Acetobacter pasteurianus* AJ605, and the yeast *Zygosaccharomyces bailii* YN403 were used as the co-culture starter. The characteristics of microbial, chemical characteristics and antioxidant activities in kombucha fermentation were analyzed. Results showed that the co-culture ratio of 8:2 (v/v) between *A. pasteurianus* AJ605 and *Z. bailii* YN403 had the highest antioxidant activity after 10 days of fermentation, with a DPPH $IC_{50}$ value of 25.76 µL $mL^{-1}$ and an ABTS $IC_{50}$ value of 8.84 µl/ml. Additionally, during fermentation, the pH of kombucha was decreased to 3.16, with an increase in titratable acidity and alcohol content of 7.00 g $L^{-1}$ and 7.96 g $L^{-1}$, respectively. To enhance the antioxidant activity and taste quality of kombucha. The black tea and roselle were mixed in various ratios used as fermentation substrate and inoculated with 10% (v/v) optimal co-culture. The results showed that the highest antioxidant activity was obtained using an 8:2 (w/w) ratio of black tea and roselle, at day 10 of fermentation, with a DPPH $IC_{50}$ value of 23.88 µl/ml and ABTS $IC_{50}$ value of 6.11 µL $mL^{-1}$. The sensory evaluation revealed that the mixture of black tea and roselle kombucha was perceived favorably.

**Keywords**: Antioxidant; Black Tea; Co-culture; Kombucha; Roselle

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# Production of yeast extract powder from spent yeast of bioethanol plant and its potential use as a nitrogen source in fermentation

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## Abstract

Yeast extract is one of organic nitrogen sources widely used in microbial cultivation. Yeast extract is often produced from spent brewery yeast. There are several processes that are used to produce yeast extract. This study aimed to produce yeast extract from alternative source, which was spent yeast from a molasses-based ethanol plant. Autolysis was chosen as the main process to obtain the cell's extract. A fixed condition of 50 ℃, initial pH of 5.0, and 60 h incubation time was used in autolysis. A single parameter of cell concentration was varied between 20-60% w/v (wet basis). Total soluble solids (by hand refractometer) and total protein of clear autolysate increased with increasing cell concentrations. At 60% w/v cell concentration, the autolysate contained the maximum total soluble solids of 5.1 ˚Brix and 24.5 g $L^{-1}$ of total protein. When the whole autolysate was subjected to a bead mill, total soluble solids increased 34% and total protein increased 29%. Yeast extract powder obtained from spray drying had a moisture content of 0.1 g $g_{powder}^{-1}$ and protein content of 0.64 $g_{protein}$ $g_{powder}^{-1}$. A 40 g $L^{-1}$ solution of the yeast extract powder corresponded to 4.8 ˚Brix. The prepared yeast extract (YE) powder was tested in a cultivation of *Rhodotorula glutinis* TISTR5159 with AR-grade yeast extract used as a control. By using YE as a sole nitrogen source at 3 g $L^{-1}$, cell dry weight of *R. glutinis* was 5.66 ± 0.36 g $L^{-1}$ using the prepared YE powder, compared with 9.98 ± 0.16 g $L^{-1}$ obtained using the AR-grade YE. This study successfully showed a value-added use of spent yeast from molasses-based ethanol plant as an effective organic nitrogen source for microbial cultivation. The application also demonstrated the concept of circular- and bio- economy by utilizing waste from bioindustry to be reused in the process.

**Keywords**: Spent yeast, Yeast extract powder, Autolysis, Bead milling, Fermentation, Nitrogen source

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# Quality control of six ingredients in Prasamawaeng traditional anti-cough formula using thin-layer chromatography technique

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## Abstract

The use of herbal medicines and nutraceuticals continues to expand rapidly. For approximately 80% of the world's population used plant materials as a primary source for treatment of various diseases in primary healthcare. However, there are some factors that limit the rational use of herbal medicine and limit the sustainable development of such biological products. The limiting factors are uncertain efficacy, uncertain safety, and uncontrolled products quality. Incorrect identification of the plant materials have led to serious side effects or deaths. Prasamawaeng is a Thai tradition anti-cough formula containing six plant meterials. Each plant material was purchased from local herbal drug stores as crude drugs. Identification of crude drug is the most important step in compounding or making herbal medicines. The major problems affecting the quality of crude drug are adulteration or substitution, degradation due to faulty collections, drying or storage. For identification of the plant materials, macroscopic identification was done and Thin layer chromatography (TLC) was used to verify the identity of plant materials by determining the chemical fingerprints of the extracts comparing with the authentic plants and reconfirmed by the plant specialists.The results found that TLC technique can be useful as a complementary technique that can distinguish one plant from another under visible or UV light and specific spray reagents that allowed visualization of the colorless components on a TLC plate. By comparing the Rf values of chemical compounds found in the plant meterials with authentic plant species we can simply confirm the quality of Prasamawaeng formula.

**Keywords**: Thin-Layer Chromatography, Prasamawaeng, *Caesalpinia bonduc*, *Curcuma* sp., *Ocimum tenuiflorum*, *Solanum trilobatum*, *Solanum violaceum*, *Tarlmounia elliptica*

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# Effects of cyanobacterium *Nostoc* sp. extract on growth of Brahmi (*Bacopa monniera*)

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## Abstract

Brahmi (*Bacopa monniera*) is a medicinal herb that can boost memory retention and protect against depression and Alzheimer's disease. This study aimed to compare the growth of Brahmi in hydroponic systems supplemented with various concentrations of Nostoc sp. extract. The experimental design was RCBD (Randomized Complete Block Design) with five treatments and three replications including a control solution (Bristol medium, pH 6.0) 0.001%(w/v) Nostoc sp. extract (NT), 0.01%(w/v) NT, 0.1%(w/v) NT, and 1.0%(w/v) NT, respectively. The survival rate, shoot height, root length, bud numbers, and chlorophyll contents of Brahmi were investigated. The results showed that the optimal concentration of NT extract for Brahmi growth is at 0.01%(w/v) that causing the highest survival rate, shoot height, root length, bud numbers, and chlorophyll content. Nostoc sp. extracts were subjected to determine Indole acetic acid (IAA) contents. The results showed that Nostoc sp. extracts contain IAA. This finding suggests that cyanobacteria extract contains IAA that could be used as biostimulants for plant growth and development and hence its utilization in agriculture.

**Keywords:** *Nostoc*, extract, Brahmi, Growth, IAA

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# Stability of the key substances in *Isaria tenuipes* extracts in cosmetic products

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## Abstract

*Isaria tenuipes* extracts were analyzed for key substances yields on a dry mass basis were cordycepin (0.32 mg $g^{-1}$), adenosine (5.62 mg $g^{-1}$) and polysaccharides (65.8 mg/g). Antioxidant activity measured as IC 50 was 0.27 mg/g by DPPH and 0.04 mg $g^{-1}$ by ABTS methods. Polysaccharides and adenosine, extracted from *I. tenuipes*, stored at 4 °C and 30 °C, was stable for 12 weeks, *i.e.*, they changed by less than 10%, except cordycepin. The optimum condition, for added *I. tenuipes* extract in the serum, was 2.0% on a weight basis. For *I. tenuipes* serum, stored at 30 °C for 12 weeks, the key substances had changed less than 10%. and at the lower tested temperature (4 °C), the pH, viscosity, and color were not significantly different, *i.e.*, now worse than 10% different. Microbiological analysis showed acceptable levels of bacteria following industrial standards. Responses to a questionnaire from 100 volunteers revealed that the volunteers preferred our new serum over a commercial product.

**Keywords**: Adenosine, Cordycepin, *Isaria tenuipes*, Polysaccharide, Stability

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# Production of geranylgeraniol by metabolic engineered *Escherichia coli*

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## Abstract

Geranylgeraniol (GGOH) is an acyclic diterpenoid that induces apoptosis in cancer cells. It originates from the dephosphorylation of geranylgeranyl diphosphate (GGPP) by geranylgeranyl diphosphate phosphatase (PDPase). GGPP is the diterpenoid precursor, which is synthesized from farnesyl diphosphate by geranygeranyl diphosphate synthase (GGPPS). This study aimed to produce GGOH by the metabolic engineered *E. coli* as an environmentally friendly method of reducing chemical consumption. Since *E. coli* strain is one of the factors affecting the gene overexpression level, two strains of *E. coli* BL21(DE3)pLysS and *E. coli* Origami harboring PDPase and GGPPS (pETDueT-PDPase-GGPPs) were used as the expression system to produce GGOH in this study. The expression of both genes was induced by 1 mM IPTG at 18 $^{O}$C for 24 h. The pellet and culture media were partitioned with EtOAc and detected for GGOH by TLC densitometry using mobile phase benzene: EtOAc (9:1). The results showed the presence of GGOH in the culture media of the induced *E.coli* Origami, which indicated that it was extracellular secreted after formation, and this strain was more suitable for producing GGOH. The expression of PDPase and GGPPS in *E.coli* Origami was needed for further optimized to achieve more amount of GGOH.

**Keywords**: Geranylgeraniol, Geranylgeranyl diphosphate phosphatase, Geranylgeranyl diphosphate synthase, *Escherichia coli*, TLC densitometry

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# Development of carbamate and organophosphate residue detection kits in durian

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## Abstract

Environmental concerns are becoming more and more severe, driven by advances and growth in economic, industrial, scientific, and technological sectors to increase productivity to meet the demands of an ever-growing population. Based on such information, it directly affects the use of chemicals to increase agricultural production. As well, it was found that the production of durian used higher amounts of pesticides. Those pesticides result in residues in the produce. To ensure the quality and safety of durian produce before selling to manufacturers, distributors and consumers from consuming durian produce that may be contaminated with pesticide residues. The objective of this research was to develop a pesticide residue test kits in durian produce consisting of a carbamate and organophosphate derivative chemical test kit to determine pesticide residues, before selling or exporting durian products according to GAP standards.The developed test kits consist of TU one-shot organophosphate test kit and TU one-shot carbamate test kit. These test kits are development based on the working principle of Cholinesterase inhibition technique. The efficacy test found that when tested with 30 durian samples, the TU one-shot organophosphate test kit had a sensitivity, specificity, and an accuracy with 83.33, 95.00, and 93.33%, respectively. In the direction of similar results in the carbamate detection of the TU one-shot carbamate test kit, it had a sensitivity, specificity, and accuracy of 83.33, 95.00, and 93.33%. The 2 test kits take approximately 5-10 minutes to check, which can provide accurate and quick results compared to standard inspections.

**Keywords**: durian, pesticide test kit, carbamate, organophosphate

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# Improving the quality of lunar regolith simulant soil for future food security

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## Abstract

At present, the world is facing many chaos related to war, new epidemics, food shortages and climate change. When that day comes, humanity must prepare for the factors of exploration and life such as food security (fresh vegetables). Food security is an important issue in the present because of rapid changes in the world. This research aims to explore the possibilities of using the lunar soil resources in agriculture. The effectiveness of humic micro-encapsulated was investigated for physical and chemical quality improvement of lunar culture applications by using TLS-01 (Thailand artificial lunar regolith Simulant) as a test substance. Sunflower (*Helianthus annuus* L.) was used as a test plant. TLS-01 particles were evenly distributed, uncluttered and consisted of 54.55% polygons and 45.45% rods, with soil density levels before and after crushed at 1.72 and 2.30 g/cm3, respectively, without the highest water holding capacity. The plants were be grown in TLS-01, but their physical characteristics were not acceptable. Sunflower seedlings grown in TLS-01 soil showed a lower percentage of germination, root, and stem length compared commercial planting material. After an improvement of physical properties and nutrients, the results showed that 1:1 mixture of TLS-01: Coconut-Coir with addition of 2 times the recommended dose of humic acid was able to promote sunflower seedling quality comparable to commercial planting material. Germination percentage and growth index were 85.00% and 6.62 respectively. However, in an actual usage for interplanetary application, coconut fiber-like materials and the humic substances were applied as in microcapsules.After the formation of the microcapsules using Whey Protein Isolate (WPI) encapsulation, fine powder particle were observed under the microscope. Additionally, releasing ability test showed that, after 6 hours, the release of humic substance by fickian diffusion was at the rate of 75% and subsequently slowed down. The encapsulation efficiency was 90.37%. In summary, with some additives, with some additives, TLS-01 could be utilized as planting material.

**Keywords**: TLS-01, sunflower, nanocapsule, coir, humic

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# Impact of degraded agrochemical and heavy metal bio-products on soil microbiota

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## Abstract

The objective of this research was to study the effect of bioproduct on soil population structure. Bacteria can be screened from soil samples before and after using bioproduct, average amount of bacterial before-after bioproduct application was $1.47x10^{15}$ and $1.28x10^{15}$ cfu $g^{-1}$ of soil, respectively. The amount of bacteria after using the bioproduct, there was a difference in the number of bacterial populations. It was found that before using the bioproduct, group 3 bacteria were classified as *Bacillus mycoides* with the highest amount of 31.77%, followed by groups 4, 2, and 1 *Saccharomyces* sp., *Xanthomonas campestris* (cause of black rot disease) and *Erwinia carotovora* (soft rot disease) were 24.76, 13.13, and 14.54%, respectively. In addition, found that group 8 is *B. subtilis* was the lowest at 0.60%. More over after using bioproduct the population structure has changed with group 8 bacteria increasing from 0.60 up to 45.86%. The experiments demonstrated that bioproduct contained *B. subtilis* TU089 can effectively multiply and occupy agricultural soil and also reduce the number of pathogenic pathogens present in the soil. It effects the structure of the soil bacterial community, causing a positive change by increasing beneficial bacteria, and reducing the number of plant pathogenic bacteria.

**Keywords**: plant pathogenic bacteria, beneficial bacteria, plant disease control

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# From an in-depth interview on pesticide use behavior to the development of a pesticide residue test kit

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## Abstract

Pests, weeds, and diseases are important factors that affect the yield and quality of durian. As a result, farmers must turn to chemical pesticides. This has led to an over-utilization of the pesticides without the consideration of the risk of contamination of the pesticides. This research was conducted by an in-depth interview on the agricultural practice of durian farmers in Chanthaburi Province. The sample size of this research was 30 people. The questionnaire consisted of social, economic, knowledge, and related to the pesticides. We found that the use of pesticides in the durian production system of the farmers in the study area was divided into two groups: 1) growth stage and 2) flowering and fruiting stage. Pesticides of the pyrethroid group were the largest group in this study. Inspection of pesticide residues in produce is very important. Currently, pesticide residue monitoring kits are difficult, inconvenient, complicated, and time-consuming. Thus, we developed "TU one-shot pyrethroid test kit" for the detection of pesticide residues in the pyrethroid group. The development relies on the principles of compatibility of chemical reactions and physics. It's a simple test set. It only takes 5-10 minutes to check. The efficacy test results showed that the detection of pyrethroid residues in 30 durian samples with the TU one-shot pyrethroid test kit had an 88.88% sensitity,95.00% specificity and 81.00% accuracy. Developed with an application called "Agriculture Pro", the color reaction values could be read from the residual pesticide content expressed in numerical units in the ppm or mg/kg test object. The experiments indicated that the developed test kits were accurate. In addition to being able to inspect multiple samples at a time, the developed pesticide test kit could also detect pesticide residues in other agricultural products as well as soil and water.

**Keywords**: Test kit, Agriculture Pro, application, pesticides

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# Effect of bioproduct for degraded chemical residues and the inhibition of *Sclerotium rolfsii*, the cause of root and stem rot of green oak

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## Abstract

A bioproduct for degrading chemical residues was developed by encapsulating three beneficial bacteria (SP-TU-2-12Y, SP-TU-C, and SP-TU 15-2), using the spherification method. The product was stored at 4 and 25°C for six months. The viability of all 3 strains of beneficial bacteria was tested. The degradation efficiency of carbosulfan, chlorpyrifos and cypermethrin and the growth inhibition of *Sclerotium rolfsii*, the cause of root and root rot of green oak lettuce, were also examined. The result showed that the viability of beneficial bacteria was not statistically different at the two storage temperatures. Bacterial populations of strains SP-TU-C, SP-TU 2-12Y and SP-TU-15-2 were 8.48 x $10^{19}$, 2.93 x $10^{19}$ and 2.51 x $10^{19}$ cfu $mL^{-1}$, respectively. Bacterial populations of strains SP-TU-C, SP-TU 2-12Y and SP-TU-15-2 were 8.48 x $10^{19}$, 2.93 x $10^{19}$ and 2.51 x $10^{19}$ cfu $mL^{-1}$, respectively. It is still effective in degrading the chemicals carbosulfan, chlorpyrifos and cypermethrin. We also found that the biochemical biodegradation was effective in inhibiting mycelial growth of *S. rolfsii* on average between 27.00±0.25 − 28.00±0.27%, which has better performance compared to the chemical control (10.78±0.14%). The study showed that the bioproduct is not only effective in degrading the chemicals but also in root system disease control.

**Keywords**: Beneficial microorganism, antagonistic bacteria, plant disease control

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# The antibacterial and antifungal activities of phenolic compounds extracted from various parts of *Mangifera indica* L. fruits against pathogenic bacteria and *Aspergillus niger*

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## Abstract

The present work aimed to investigate the effect of phenolic compounds extracted from various parts of *Mangifera indica* L. fruits on the antibacterial and antifungal activity evaluation against various bacteria including *Bacillus subtilis* TISTR 1984, *Bacillus cereus* TISTR 1474, *Escherichia coli* TISTR 073, *Pseudomonas aeruginosa* TISTR 1287, *Staphylococcus aureus* TISTR 746, *Enterococcus aerogenes* TISTR 1540, and the pathogenic fungus *Aspergillus niger* which normally contaminated agricultural products. The crude extract from the seeds showed the inhibition zones of *B. cereus* TISTR1474 (15 mm), *B. subtilis* TISTR 1984 (14 mm), *E. coli* TISTR 073 (13 mm), *P. aeruginosa* TISTR 1287 (10 mm), and *S. aureus* TISTR 746 (10 mm). After seven days of incubation, the crude derived from the seed and peel also suppressed *A. niger* mycelial growth. Gallic acid, Ferulic acid, Apigenin, and Mangiferin levels were higher in the crude extract from the seed than that from the peel and fruit pulp. Finally, the antibacterial and fungal activity of phenolic compounds isolated from *Mangifera indica* L. fruits are attractive bioresources with prospective applications in agricultural, food, and other industries.

**Keywords**: *Mangifera indica* L., antibacterial, antifungal, phenolic compounds

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# Actinomycetes from endophytic and epiphytic root of five medicinal plants and their antimicrobial activity

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## Abstract

The objective of this study was to isolate endophytic and epiphytic actinomycetes from roots of five medicinal plants in Thailand, including gombo (*Abelmoschus esculentus* (L.) Moench), bergamot (*Citrus hystrix* DC.), carunda (*Carissa carandas* L), papaya (*Carica papaya* L.) and marigold (*Tagetes erecta* L.). A total of fifty-five strains of actinomycetes were isolated and their morphological and biochemical characteristics were examined. Preliminary screening of antimicrobial activities against six microorganisms; *Staphylococcus aureus* ATCC 25923, *Bacillus subtilis* ATCC 6633, *Kocuria rhizophila* ATCC 9341, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853 and *Candida albicans* ATCC 10231 found that six strains could inhibit test microorganisms including AM231, CA131, CA331, RO232, RO321 and RO331. Antimicrobial activities of crude ethyl-acetate extracts were examined against test microorganisms using disc diffusion method. The result showed that only strains AM231 and CA131 were able to inhibit *Kocuria rhizophila* ATCC 9341. The inhibition zones were 34.5 and 4 mm., respectively with the concentration of crude extract at 50 mg/ml.

**Keywords**: Actinomycetes, Antibacterial activity, Ethyl acetate extraction, Thai medicinal plants

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# Actinomycetes from organic rice field soil, their antagonistic activity and antimicrobial activity

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## Abstract

Fifty-three actinomycete strains were isolated from organic rice field soil samples collected from Im Sook organic Farm, Pathum Thani province, Thailand. All isolates were tested for antagonistic activity against rice blast disease fungus *Pyricularia* spp. on potato dextrose agar by the dual culture technique. Isolate PTN21021 showed the highest percentage radial inhibition growth of the fungus (%RGI) at 60.63% (moderate antagonistic activity). Isolate PTN21021 and five potent isolates, PTN10712, PTN20322, PTN21123, PTN10112 and PTN21024 were selected for their phenotypic characterization and gene sequence analysis. Molecular identification by sequencing of 16S rRNA gene revealed that the isolate PTN10712 shared 99.66% similarity with *Amycolatopsis rhizosphaerae* DH51B-4-3$^T$. PTN21021 shared 99.83% similarity with *Kitasatospora cineracea* DSM 44780$^T$. PTN20322 and PTN21123 shared 99.84% similarity with *Streptomyces shenzhenensis* 172115$^T$. PTN10112 shared 99.84% similarity with *Streptomyces malaysiense* MUSC 136$^T$. PTN21024 shared 99.75% similarity with *Streptomyces bungoensis* DSM 41781$^T$. On primary screening for antimicrobial activities, nine strains showed the activities against six microorganisms, including *Kocuria rhizophila* ATCC 9341, *Staphylococcus aureus* ATCC 25923, *Bacillus subtilis* ATCC 6633, *Pseudomonas aeroginosa* ATCC 27853, *Escherichia coli* ATCC 25922 and *Candida albicans* ATCC 10231. The agar disc diffusion was performed with the concentration of 50 mg/ml. The highest inhibition zones were found on isolate PTN10711, PTN21126 and PTN21129 at 50 mg/ml against *Bacillus subtilis* ATCC 6633 at 25, 8 and 14 mm. respectively, from crude ethyl acetate extraction.

**Keywords**: Actinomycete, Antagonistic activity, Rice blast disease, *Pyricularia* spp., Base sequence and Antimicrobial activity

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# Identification of *bla*$_{\text{OXA-72}}$ gene associated with high-level carbapenem resistance in a multidrug-resistant *Acinetobacter baumannii* isolate in Thailand

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## Abstract

Dissemination of antimicrobial resistance genes (ARGs) between bacteria from different origins may promote the evolution of drug-resistant pathogens, indicating the urgency to carefully monitor and control the spread of ARGs. The emergence of carbapenem-resistant *Acinetobacter baumannii* through the acquisition of resistance determinants has become an ongoing public health threat. This study investigated the presence of carbapenemase genes in a multidrug-resistant (MDR) *A. baumannii* isolate in Thailand. The MDR *A. baumannii* strain MTC0706 collected from a tertiary hospital in Northern region of Thailand was subjected to antimicrobial susceptibility testing and the PCR technique was used to identify resistance genes encoding four major groups of OXA-type carbapenemase. The isolate exhibited relatively high MIC to meropenem (128 μg $mL^{-1}$), imipenem (128 μg $mL^{-1}$), and doripenem (64 μg.$mL^{-1}$). Amplification of resistance genes showed positive results for the presence of OXA-51-like and OXA-24/40-like carbapenemase, while the OXA-23-like and OXA-58-like carbapenemase were absent. Moreover, sequencing of the *bla*$_{\text{OXA-24/40-like}}$ fragment confirmed the occurrence of *bla*$_{\text{OXA-72}}$ gene which may have an important role to confer carbapenem resistance in the tested isolate. The *bla*$_{\text{OXA-72}}$ was notably detected in the plasmid DNA as well, suggesting its localization and the potential spread. Collectively, the results of this study demonstrate the occurrence of *bla*$_{\text{OXA-72}}$ gene in an MDR *A. baumannii* isolate with high level of non-susceptibility to carbapenems. Possible plasmid localization of the gene also underlines the necessity to track the horizontal transfer of this ARG to other isolates in hospital and beyond.

**Keywords**: *antimicrobial resistance, Acinetobacter baumannii, carbapanemase, bla*$_{\text{OXA-72}}$

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# Characterization of a gene cassette structure of *bla*$_{\text{imp-65}}$ gene, from an MDR *Pseudomonas aeruginosa* in Thailand

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## Abstract

In recent years, the development of multidrug resistance in bacteria has contributed significantly to severe cases of infection and health complications. A great portion of multidrug resitant-related (MDR) diseases around the world are caused by MDR *P. Aeruginosa*. In this study, the total of a strain of *P. Aeruginosa* produced metallo beta-lactamase was extracted and sequenced by whole-genome sequencing. The sequence data around metallo beta-lactamase gene was assembled by the Geneious Prime software and blasted with the database of NCBI. The annotation was performed using Benchling. We have discovered that the *bla*$_{\text{IMP-65}}$ gene was surrounded by various transposable elements, such as Tn3 and IS genes, which led to the conclusion that this resitant gene is part of a gene cassette. Additionally, we have observed the presence of resistant genes that encoded for enzymes known for neutralizaing aminoglycosides and sufonamides. These resistant genes are located within a few hundred base pairs away from the *bla*$_{\text{imp-65}}$ gene. In this study, the results also demonstrated the significance of transposons in dissembinating resistant genes. Sutdies have shown the potential of beta-lactamase genes in transferring via mobile elements such as Tn3 genes. It can also be observed that other resistant genes are typically also present with beta-lactamase genes which have added to a bacterium's resistance against beta-lactams and other classes of agents. In conclusion, the genetic composition of the IMP-65 gene cassette shown in this study is likely to affect the resistance profile of this *P. aeruginosa* strain. Further study on the transferability of *bla*$_{\text{imp-65}}$ gene cassette will be required to understand and quantify the full impact of this gene.

**Keywords:** IMP-65 gene, MDR *P. aeruginosa*, metallo-beta-lactamase, transposons.

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# Biosynthesis of silver nanoparticles using of *Chlorella* sp. extract and antibacterial activity investigation

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## Abstract

Silver nanoparticles (AgNPs) is important for antibacterial activity used in various application, such as food and medicine. Nanoparticles have been synthesized in many methods including physical and chemical method. Biosynthesis was chemical method employed to synthesize silver nanoparticles using bioactive compounds of extracts. In this study, synthesis of AgNPs from reducing the silver ions present in the silver nitrate ($AgNO_3$) solution were synthesized using *Chlorella* sp. ethanolic extract. The obtained AgNPs were studied for antibacterial properties. Two common grams bacteria: gram-positive (*Staphylococcus aureus*) and gram-negative (*Escherichia coli*) were used for the antibacterial effect of synthesized AgNPs production by agar diffusion technique. The result showed, the growth of *S. aureus* and *E. coli* were inhibited by AgNPs. The greater inhibition zones for those bacteria were 13.80 and 8.10 mm in diameter respectively. *Chlorella* sp. ethanolic extraction was used reduce silver nitrate to AgNPs showed great antibacterial property compared to the *Chlorella* sp. ethanolic extract; however, less than the $AgNO_3$ solution. The result show that AgNPs from *Chlorella* sp. ethanolic extract can potentially kill resistant bacteria. In addition, *Chlorella* sp. was microalgae have been bioactive metabolites, nutrients, and chemical compounds to importance in aquaculture, the use of algae extracts to synthesis AgNPs which contributes to its application in the aquaculture as antipathogens, feed, and transport packaging in agriculture.

**Keywords**: Biosynthesis; Silver nanoparticles; *Chlorella*; Antibacterial activity

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# Effects of fermentation on black soldier fly as animal feed additive

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## Abstract

In recent years, *Hermetia illucens* larva has gained popularity as an animal feed supplement or protein source. It contains a high level of crude protein, fatty acid, and balanced amino acid composition, making it a good substitute for costly fish meal and bean pulp in animal diets. However, excess of chitin in larva skin limits its application as an animal feed additive, consequently, employing fermentation with zymocytes to remove chitin is necessary. In this study, *Bacillus subtilis* and *Aspergillus niger* were applied to co-ferment *Hermetia illucens* larva paste. Label-free quantitative proteomic analysis revealed that the expression of multiple enzymes from *B. subtilis* and *A. niger* involved in polysaccharide hydrolysis, amino acid biosynthesis, and fatty acid metabolism have either increased or decreased significantly as compared to the control group. the findings demonstrated that both zymocytes concentrated on disintegrating macromolecular substances, producing active small molecule substances, and avoiding biosynthesis of surplus amino acids under starvation conditions. Production of free amino acids, acid-soluble proteins, and short-chain fatty acids was increased after fermentation. 13 out of 17 amino acids were increased, with MET increased 84-folds, LYS increased more than 1000 mg/kg, ASP, GLU, and other acids increased from 1-fold to 22-folds, and total free amino acids were increased from 0.08 g/100 g to 0.3 g/100 g. Organic acids such as ethanoic, propionic, isobutyric, ethyl acetic, isovaleric, and pentanoic acid were increased up to 4.81, 8.2, 59.49, 61.45, 47.21, and 17-folds through fermentation, respectively. While actual protein content declined from 3.03 g/100 g to 1.81 g/100 g, peptide content increased from 1.3 g/100 g to 2.46 g/100 g, total sugar decreased from 1.399 mg/mL to 0.527 mg/mL, chitin degradation rate was 62.3%.

**Key words:** *Hermetia illucens*, *Bacillus subtilis*, *Aspergillus niger*, chitin, fermentation

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# Study and utilization of chlamydospores of *Trichoderma*

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## Abstract

*Trichoderma* spp. can control almost all plant fungal diseases. Existing *Trichoderma* biopesticides are all made from conidiospores which are poor stability, short shelf life (six months), and high production and application costs. Compared with conidiospore, chlamydospore has larger volume, thicker cell wall and stronger stress resistance. However, there is no chlamydospore fermentation process and products because the induction conditions and chlamydospore -producing mechanism are still unknow. In this study, a mixture named Ha01 was found to induce the formation of chlamydospore of *Trichoderma*, and low nitrogen environment promoted the transformation of chitin and the formation of chlamydospore, which was regulated by aminoglycose metabolic pathway. N-acetyl-d-glucosamine was the key intermediate and inhibited sporulation. The mass fermentation process of chlamydospore was developed. On the basis of analyzing the chlamydospore production mechanism, the mass fermentation process of chlamydospore was developed. The first chlamydospore product of *Trichoderma* chlamydospore named Shibeijian was developed. The shelf life of Shibijian is 2 years, and showed better stability than RootShield, product of Bioworks inc., the largest manufacturer of *Trichoderma* biopesticides in the world. We developed 11 series of *Trichoderma* chlamydospore products for controlling different diseases and established 6 sets of soil-borne disease control technologies. In the past 6 years, the application area of chlamydospore products reached 300,000 hectares. The average control efficiency against over 20 soil-borne diseases was 50%-70% and the average yield increased by 10%-20%. The products have won 5 industry honors such as China's microbial industry leader brand, Baijia fertilizer star product title.

**Keywords**: *Trichoderma* spp., biopesticides, chlamydospores

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# Solid Phase Extraction with the UHPLC–MS/MS for detection of tetracyclines antibiotics residues in composting system of black soldier fly larva

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## Abstract

Tetracycline antibiotics are the most extensive residual antibiotics in feces. Black soldier fly (BSF), *Hermetia illucens*, which is capable of efficiently converting manure into insect biomass,is at risk of accumulating antibiotics, causing widespread concerns of food safety as a protein feed. Thus, it is necessary to monitor the dynamic changes of antibiotics in the BSF composting system in order to determine the safety of larvae as protein feeds and the safety of frass as organic fertilizer. The present paper described the development and optimization method for the analysis of tetracycline antibiotics (TC) in matrix of BSF based on UHPLC–MS/MS for the first time and monitored the degradation dynamics and the bioaccumulation law of tetracyclines in the composting system. The results showed that there was a good linear relationship between the concentration of TC and the corresponding chromatographic peak area from 0.1 μg $L^{-1}$ to 500 μg $L^{-1}$, R2 = 0.9999 and 0.9975, respectively. At a 50 μg $kg^{-1}$ spiked level, the recovery rates of TC in BSF were between 80.66% and 101.16%, and method repeatability featured by a general relative standard deviation below 6%, the LOD and LOQ of TC was 0.23 μg $kg^{-1}$ and 0.76 μg $kg^{-1}$ respectively. The resulting methodology was applied to to monitor the degradation of tetracycline by BSF larvae under stress of TC, we found that after 14 days of exposure, tetracycline was undetectable in the feces of 0.01 mg $kg^{-1}$ initial concentration, but the concentration of TC in larvae was still as high as 2.93 ng/g; The TC decreased to 5.93 mg/kg as the rate of degradation reached 96% in group with 100 mg $kg^{-1}$ TC initial concentration and the bioaccumulation concentration of TC in vivo increased to 10 mg $kg^{-1}$, which definitely hint the risk of bioaccumulation antibiotics and feed security issues. This method plays a importent role to monitor TC concentration in BSF composting system as its highly sensitive and this paper first warned that BSF transformed from animal manure may be at risk of accumulating antibiotics and requires further treatment before it can be used as protein feed.

**Keywords**: Black soldier fly, Antibiotic residual, Antibiotic accumulation, Liquid chromatography tandem mass spectrometry, Solid-phase extraction.

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