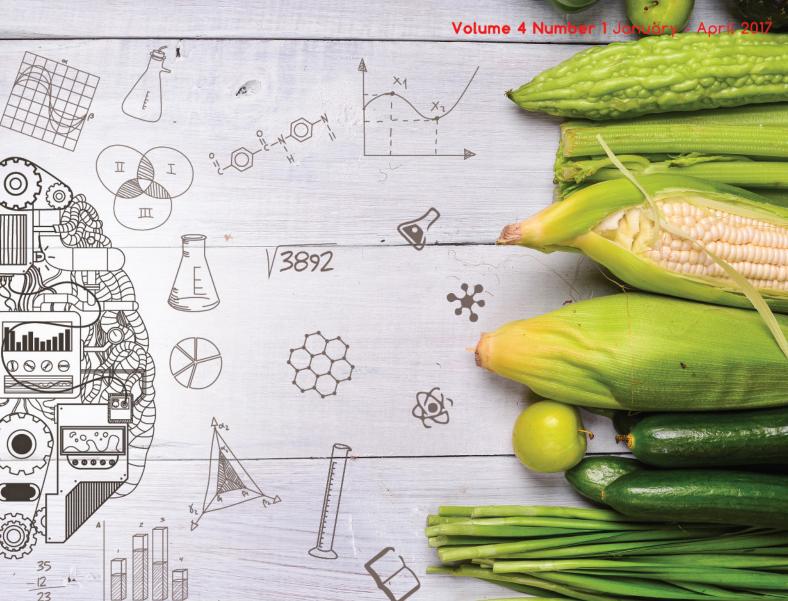


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The Enhancing Quality Control of Raw Milk at Milk Collection Centers

Diversity of Parasitic Wasps in Thailand: DNA Barcode and Taxonomy

Use of Cement By-Product (Silicon Dioxide) for Swine Production

Bacterial Blight of Rice in Thailand:

Current Status of Disease Epidemics

Roles of Microorganisms in the Nitrogen Cycle and the Hinetic Model for Aquaculture Systems

Soil Microbiome in the Upper Northeast of Thailand





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ด้วยเกล้าด้วยกระหม่อม ข้าพระพุทธเจ้า ผู้บริหารและเจ้าหน้าที่ศูนย์บริการวิชาการแห่งจุฬาลงกรณ์มหาวิทยาลัย

actions.

# Editor's note



# JNISEARCH

# JOURNA

Our world today faces growing existential challenges from mega-trends such as climate change, global population growth and urban expansion, an increasingly critical water shortage, together with ever-dwindling natural resources. In particular, soil and water resources are fundamental to global food security, and their inadequacy disproportionately impacts on developing countries whose populations are predominantly poor and dependent on agriculture.

The challenges highlight the importance and urgency of research in these critical areas in order to improve efficiency of use of scarce resources such as land and water, improve resilience to climate change impacts, manage pests and diseases and improve productivity while conserving the environment and natural biodiversity.

Moreover, utilization of existing research, innovation and technology should be enhanced in order to establish a robust and sustainable foundation for Thailand's agricultural sector.

Accordingly, relevant agencies in all sectors should prioritize and promote research to resolve some of the country's most pressing problems. How to revitalize our rural economies, conserve our natural resources and enhance quality of life for the country's farmers? How also to add value to agricultural production, boost agro-innovation and drive differentiation in Thailand's exports to the world? How, in summary, to transform Thailand's agricultural sector to preserve Thailand's unique natural and social capital?

Part of our role as research and educational institutions, in expanding our understanding at the convergence of scientific and social research, is to support an evidence-based development trajectory for the country that offers a fair deal, justice and quality of life for all segments of our population. Thailand's widening chasm of inequity between rich and poor offers a stark reminder of how far we are from this ideal, and how much remains to be done. Boosting our agricultural sector will be an essential first step in this direction.

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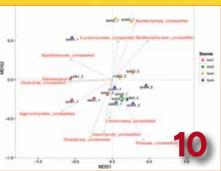
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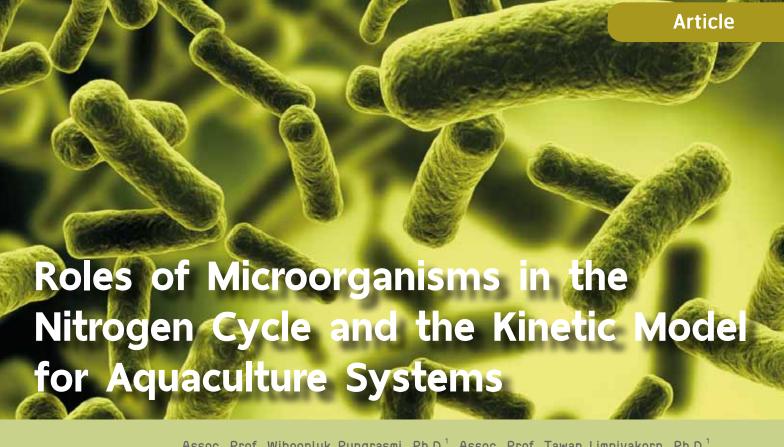
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# Introduction

This research is a further study of the role of microorganisms in biological treatment within aquaculture systems, under cooperation between staffs from the Department of Environmental Engineering, Faculty of Engineering, Chulalongkorn University and a senior researcher from the Center of Excellence for Marine Biotechnology, National Center for Genetic Engineering and Biotechnology (BIOTEC). This study comprises 2 sub-projects. The first concerns a follow-up to an earlier study on "Dissimilatory nitrate Reduction to Ammonium (DNRA) in the denitrification process which has a role in converting nitrate to ammonia". DNRA leads to elevated levels of an unwanted microbial activity in wastewater treatment and aquaculture systems. Yet, formation of ammonia in anaerobic systems is found only under certain conditions. So, further study is needed to understand the factors that contribute to conditions that allow effective system control and efficient ammonia treatment in aquaculture systems. The second sub-project is a kinetic study of nitrification, an important process in aquaculture systems because of its impact on rate-limiting steps. The specific bacteria responsible for nitrification have a slow growth rate and are highly sensitive to environmental change. Kinetic knowledge and data have a major role in designing and controlling nitrogen levels and optimize system management. Furthermore, an understanding of system kinetics provides an additional tool to help explain changes and remediate problems occurring in the system.

### **Aquaculture Systems in Thailand**

Thailand's aquaculture industry has grown rapidly to meet burgeoning domestic and international demand. An emerging technology: the Intensive Aquatic System (IAS) is gaining popularity and provides an effective system for management of aquaculture systems. Today IAS is reported to account for 85 percent of all aquaculture systems in Thailand. In this system, the excess ration of pelleted feeding dosages and good water quality controls were continuously managed throughout the culture period. In addition, the aquaculture system is converted into a semi-closed or closed recirculating aquaculture system (CRAS) in order to save water, prevent contraction and reduce environmental impact from releasing wastewater from the tanks. However, such closed systems frequently suffer from accumulation of inorganic nitrogen compounds in the system caused by unconsumed feed, fecal matter and degradation of the protein from dead aquatic animals. Accumulation of ammonia and nitrate at high levels can be toxic to aquatic animals even at low concentrations. Thus, the current study aimed to identify methods to control nutrient levels in wastewater and organic sediments in aquaculture tanks, as an essential means for water quality improvement in recirculating systems.

## **DNRA Roles in Aquaculture System**

DNRA is biological mechanism of microorganism which converts inorganic nitrogen from nitrate to ammonia, with nitrite ( $NO_2$ ) as mediator in the reaction. DNRA microorganisms grow in similar conditions to denitrifying bacteria; i.e. anaerobic conditions with nitrate as the electron acceptor. However, the products of both processes are considerably different since denitrification converts nitrate to gaseous nitrogen ( $N_2$ ) while DNRA converts nitrate to ammonia- a waste product that requires further treatment before discharge. Typically, ammonia

which is treated will be transformed to nitrate by nitrifying microorganisms under aerobic conditions. Then, nitrate is converted to gaseous nitrogen by denitrifying microorganisms under anaerobic conditions. DNRA microorganisms, found in denitrification processes can regenerate ammonia in the system and are therefore unwanted in wastewater treatment and aquaculture systems. Nevertheless, DNRA microorganisms are sometimes detected under anaerobic conditions.

This research aimed to gain an understanding of the activity of DNRA microorganisms under conditions prevailing in recirculating aquaculture systems, which recirculate treated wastewater to the aquaculture tank in order to minimize water use and system waste. Moreover, ammonia is highly toxic to aquatic animals even at low concentrations, so that the DNRA process is extremely detrimental to overall water quality. So, an understanding of the factors influencing presence of these microorganisms and the conditions that trigger the DNRA process is important to efficient system operation and ammonia treatment, especially in CRAS aquaculture systems.

Laboratory experiments were conducted at room temperature at the aquaculture laboratory at the Center of Excellence for Marine Biotechnology, Faculty of Science, Chulalongkorn University. The experiment collected samples of denitrification sediments from a tilapia culture tank from 2-system housing with different COD/NO<sub>2</sub>-N ratio, in order to test the DNRA reaction or reduction of nitrate into ammonia. That is, sediments with low COD/NO<sub>3</sub> were collected from the freshwater tilapia culture tank with a closed circulating system. The high COD/NO sediment was collected from a Biofloc technology tilapia culture tank in which organic carbon is added to activate growth and increase populations of microorganisms in the system. The microorganisms form suspended biological sediments in water called "Biofloc". The details of the 2-system aquaculture tanks are shown in Table 1.

Table 1 Characteristics of tilapia culture system used in the experiment to determine the DNRA reaction

	NH <sub>4</sub> + (mg-N/L)	NO <sub>2</sub> (mg-N/L)	NO <sub>3</sub> (mg-N/L)	DO (mg/L)	рН	Temp (°C)	SS (mg/L)	Tank volume (L)	Carbon added
Low COD/NO <sub>3</sub>	0.04±0.04	0.09±0.03	10.0±0.7	4.2±0.1	6.8±0.0	29.7±0.1	126.0±17.4	4,000	-
High COD/NO <sub>3</sub>	0.13±0.02	0.35±0.01	32.8±1.0	7.1±0.2	7.0±0.0	29.6±0.1	498.3±30.1	700	Starch

Source: Chutivisut et al. (2014)

In the experiment, 10 grams (dry weight) of sediment samples from the 2-system tilapia culture tank was separately placed into 1-litre reactors. The systems were operated under batch condition by adding different amount of sodium nitrate at the concentration of 25 mg. and 100 mg. nitrate-nitrogen/litre, respectively. This is to test the nitrate reduction activities when different initial nitrate concentrations are used (Figure 1).

The composition of Wastewater used in the experiment (per litre) are as follows: 3.8 g. of disodium hydrogen phosphate (Na $_2$ HPO $_4$ ), 1.5 g. of dipotassium phosphate (K $_2$ HPO $_4$ ), 0.1 g. of magnesium sulphate (MgSO $_4$ ·7H $_2$ O) and 2 ml. of supplementary nutrients. The component per litre of nutrients, comprising the following ingredients:

50.0 g. of ethylene diamine tetra-acetic acid: EDTA 7.34 g. of calcium chloride dehydrate:  $CaCl_2 \cdot 2H_2O$  4.99 g. of Iron (II) sulfate heptahydrate:  $FeSO_4 \cdot 7H_2O$  1.57 g. of Copper (II) sulfate pentahydrate:  $CuSO_4 \cdot 5H_2O$  22.0 g. of zinc sulphate heptahydrate:  $ZnSO_4 \cdot 7H_2O$  5.06 g. of manganese (II) chloride tetrahydrate:  $ZnSO_4 \cdot 2H_2O$  0.195 g. of sodium molybdate dihydrate:  $ZnSO_4 \cdot 2H_2O$  1.61 g. of cobalt (II) chloride hexahydrate:  $ZnSO_4 \cdot 2H_2O$  1.61 g. of cobalt (II) chloride hexahydrate:  $ZnSO_4 \cdot 2H_2O$ 

The sediments and wastewater in the reactors were stirred throughout the experiment using a magnetic stirrer, in order to ensure even nutrition for the microorganisms in the sediments. After beginning the experiment by adding nitrate, water samples were collected every 30 or 60 minutes to analyze inorganic nitrogen levels. Analysis of nitrate, nitrite



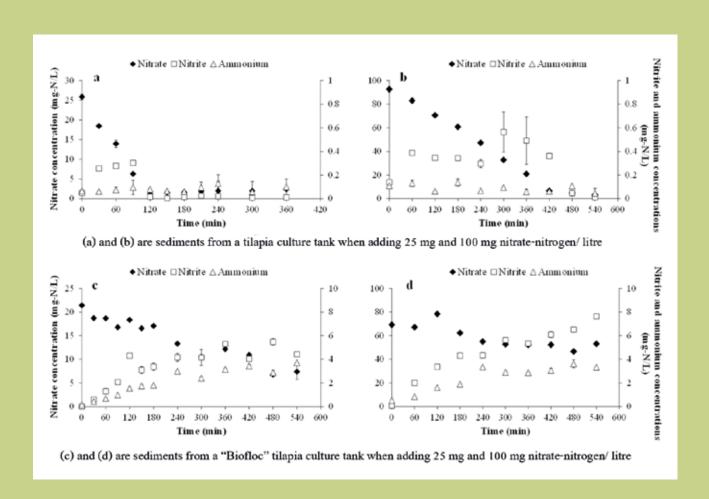
Figure 1 Reactors for testing microorganism roles in nitrogen cycle Source: Pokchat Chutivisut, taken in March 2013

and ammonia in water samples was done via the standard method for water and wastewater (APHA, 2005), nitrate analysis by UV-spectrophotometry method (APHA, 2005) and nitrite and ammonia analyses by the sulfanilamide (APHA, 2005) and salicylate-hypochlorite (Bower and Holm-Hansen, 1980) methods.

According to the DNRA activity test in sediment samples from aquaculture tanks with low and high COD/NO<sub>3</sub>, the changing result of nitrate, nitrite and ammonia values in water from reactors which adding sodium nitrate at different initial concentrations with 25 and 100 ml. nitrate-nitrogen/litre was shown in Figure 2. Nitrates in sediments from low COD/NO<sub>3</sub> system can be reduced rapidly with a little nitrite

accumulation and without ammonia formation. The result indicates that denitrification occurred, with nitrate transformed to gaseous nitrogen via the following sequence:  $NO_3^- -> NO_2^- -> NO_2^- -> N_2^- O_3^- -> NO_2^- -> N_2^- O_3^- -> NO_2^- -> N_2^- O_3^- -> NO_3^- ->$ 

Concerning sediments from high COD/NO<sub>3</sub> system, nitrate reduction occurs slowly compared



**Figure 2** Changes in nitrate, nitrite and ammonia in water from reactors into which sodium nitrate at different initial concentrations is added

Source: Chutivisut et al. (2014)

with low COD/NO $_3$  systems and the accumulation of nitrite and ammonia were found at both initial nitrate concentrations (25 and 100 ml. nitrate-nitrogen/litre). Besides, nitrite accumulation indicates incomplete denitrification and ammonia accumulation also indicates DNRA reaction and nitrate reduction to ammonia as the steps followed;  $NO_3^- -> NO_2^- -> NH_4^+$ . The occurrence of DNRA reaction when nitrate is added into reactors shows the presence of DNRA microorganisms in high COD/NO $_3^-$  aquaculture system. According to the toxicity of ammonia to aquatic culture, the existence of DNRA microorganisms indicated high impact to aquaculture system. Details and the development of the microorganism classification need to be studied further.

It can be said that COD/NO<sub>3</sub> ratio is an important factor affecting nitrogen transformation occurring in the system. Under normal conditions, denitrification usually precedes for the energy from the transformation of nitrate to nitrogen is higher than DNRA process. Besides, yield value of Denitrifier microorganism growth is higher than those using DNRA process. Yet, in the condition that has a lot of organic substances (electron-donor substances) but limited nitrate (electron-acceptor substances), DNRA process can occur because, when compared the calculated energy values that microorganisms receive per mole of nitrate, energy from DNRA has higher value than those from denitrification. However, apart from COD/NO<sub>2</sub> ratio that triggers DNRA process, there are other factors affecting the competition among the processes occurred like bacterial populations, environment etc.

# The Study of Kinetic Model of Nitrogen Treatment in Aquaculture System

Nitrogen control in aquaculture systems generally relies on biological conversion of nitrogen

species as the principal mechanism. Under aerobic condition, nitrification precedes; ammonia-oxidizing bacteria and archaea (AOB and AOA) oxidize nitrogen waste in form of ammonia to nitrite and nitrite-oxidizing bacteria (NOB) oxidize nitrite to nitrate. Then, bacteria in denitrification process in anaerobic condition reduce nitrite to nitrogen gas which is released into atmosphere. Nitrification is considered to be rate-limiting step in nitrogen elimination, since the microorganisms in this process have very slow growth rate and are highly sensitive to environmental changes. Therefore, kinetic study of microorganisms in nitrification process is very important to nitrogen control in aquaculture system, because it has a major role in designing nitrogen controlling system and enhances more efficient operation. Moreover, kinetic knowledge can explain abnormalities in the system including solving problems when the system cannot run properly.

Kinetic of ammonia-oxidizing microorganisms who oxidize ammonia to nitrite is important to nitrification. According to the past researches, ammonia concentration directly impacts the population of ammonia-oxidizing microorganisms which apparently affects the kinetic of ammonia oxidation. To solve the ammonia accumulation in aquaculture tanks, microorganism cultures which have ability to treat ammonia and nitrite are added into the system to reduce the accumulation of inorganic nitrogen in aquaculture tanks. Normally, the seeds used is usually derived from the enrichment under high ammonia concentration (ammonia is higher than 20 mg. nitrogen/litre). However, since normal aquaculture tanks usually have not so high ammonia contamination, the seeding that is derived from the enrichment under high ammonia condition might not be so effective because kinetic factors don't support the seeds to grow and work with full efficiency in actual condition. Regarding the issue, currently, there are no researches clearly asserting the hypothesis mentioned above about using ammonia—oxidizing seeds enriched with high ammonia condition in low ammonia system or, conversely, using ammonia-oxidizing seed enriched from low ammonia concentration in high concentrated environment.

This research project aims to study the change of ammonia-oxidizing and nitrite-oxidizing population after changing ammonia concentration. Initially, both groups of microorganisms are enriched by feeding 50 mg. nitrogen/litre ammonia (reactor A) and 1 mg. nitrogen/litre ammonia (reactor B). Later, when the systems are stable, the ammonia concentration of both reactors is switched. Here, kinetic parameters are analyzed for, example maximum substrate utilization rate,  $q_{max}$  and substrate half saturation constant ( $K_{\epsilon}$ )

At the early stage of the experiment, seeds are produced by culturing in different ammonia concentrations. It turns out that ammonia-oxidizing microorganisms from both reactors have clearly different kinetic characteristics of ammonia oxidation. In reactor A, low affinity to ammonia microorganisms are mainly found while high affinity to ammonia

microorganisms are found in reactor B. Nitrite-oxidizing microorganisms in both reactors are not so different with high affinity to nitrite as main population.

After switching the concentration of both reactors for a short time (8 days); reactor A from 50 ml. nitrogen/litre ammonia to 1 ml. nitrogen/litre ammonia and reactor B from 1 ml. nitrogen/litre ammonia to 50 ml. nitrogen/litre ammonia, kinetic parameters for ammonia oxidation of reactor A apparently changes; that is,  $K_s$  reduces substantially while reactor B still has low  $K_s$  (Table 2).

Concerning engineering benefits, the experiment result demonstrates that we can culture microorganisms in high ammonia environment for treating ammonia and nitrite in low ammonia and nitrite aquaculture tanks. Culturing microorganisms in high ammonia condition can be done more easily as it form the seed more quickly.

### **Acknowledgement**

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Table 2 Kinetic parameter of ammonia oxidation for reactors A and B before and after switching ammonia concentration

	Reactor A swit concentration f nitroge		Reactor B switches ammonia concentration from 1 to 50 ml. nitrogen/litre		
Parameter	Before	After	Before	After	
K <sub>s</sub> (mg. nitrogen/litre)	8.78	1.50	1.19	0.25	
q <sub>max</sub> (mg. nitrogen/litre/ ml. MLVSS.hour)	0.128	0.091	0.0089	0.20	
$R^2$	0.90	0.68	0.91	0.69	

**Note:** MLVSS is Mixed Liquor Volatile Suspended Solids **Source:** Tarnpetch Charoanwoodtipong (2015)

Project of fiscal year 2014, Ratchadapiseksomphot Endowment Fund, Chulalongkorn University (CU-57-023-FW) and the project is supported with devices, equipment, chemicals and work place from the Center of Excellence for Marine Biotechnology, Faculty of Science, Chulalongkorn University.

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# Soil Microbiome

# in the Upper Northeast of Thailand

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# Introduction

Thailand is agriculture-based country, primarily in the Northeast of Thailand (NE). Major cultivar plants in Thailand are rice (>50%), followed by assorted vegetables and fruits (e.g. tea, corn and banana) (NESDB, 2005).

NE encompasses one-third of the country's land (168,854 km²), and contains a majority of population. The NE population nominates in agriculture for living. In contrast to other regions of Thailand, the NE soil is rather inferior for cultivation (Fukai, Sittisuang and Chanphengsay, 1998). The soil is of fine-grained sand texture. The NE geography has limited to water access and the annual rainfall is unpredictable. Examples include a long drought season and then a flood. Irrigation relieves the problem but leaving the soil salinity problem. Besides, many cultivar lands come from deforestation. Together, the crop yields per area are poor, and promote farmers to use industrial agriculture (IA), which further accelerates the soil quality problems (Fedra, Winkelbauer and Pantulu, 1991; NESDB, 2005).

A modern era of industrialization arises a conventional farming, also known as industrial agriculture (IA). IA refers to farming system that utilizes chemical fertilizer, pesticide, herbicide and other synthetically inorganic inputs, as well as genetic modified organism, concentrated animal feeding, and extensive irrigation or tillage. This allows a large sum of food production on less land and with less human labor (Seufert, Ramankutty, and Foley, 2012). However, many activities of this non-naturally high resource and energy intense IA affect environmental health, such as chemical leaching, land degradation and loss in biodiversity resources. Researches comparing between IA and organic agriculture (OA), a contrast method to IA, also reported generally more species diversity and abundance in OA than IA (Bengtsson, Ahnström, and Weibull, 2005). In addition, the chemical inputs, e.g. pesticide, pose harm to human health. Nonetheless, with the instant great yield, IA remains commonly used by Thai farmers, and poor soil quality problems occur increasingly (Fukai, Sittisuang and Chanphengsay, 1998).

# Organic agriculture and importance of soil microbiome

OA, also known as sustainable farming, prohibits those chemical and artificial inputs, and genetic modified organisms, albeit the use of agricultural machines is still allowed for energy and human labor efficient (Hole et al., 2005; Paull, 2011). For instances, OA utilizes animal and plant product fertilizers, hand weeding and biological pest control, in replace of chemical fertilizer, herbicide and pesticide. OA also uses polyculture (or companion planting), and crop rotation to increase soil diversity. The goal of OA is to maintain the healthiness of nature, including, but not limited to, soil, water, animals and humans. The OA processes rely on ecological biodiversity and metabolic food networks that suit local geography (Rigby and Cáceres, 2001). Office of Agricultural Economics of Thailand reported the OA farms in Thailand comprise of rice (approximately 68%), vegetables (12%), fruits (8%), tea (8%), and herbs and others, and the OA produces can generally have the accepted higher market price than the non-OA produces due to their merit in green agriculture.

Microorganisms serve the major beginning and end (recycling) steps in almost every food network routes. Recently, researches attempt to increase arable yield through the role of prokaryotic and eukaryotic (mainly fungi, nematodes and annelids) microbiome. When farmers grow and harvest crops, some nutrients were removed from the soil. Continuingly, the land suffers nutrient depletion and become non-arable soil. To replenish nutrients in soil, maintenance of suitable microbial diversity balance, including prokaryotic bacteria and archaea, and eukaryotic fungi, protists and small animals i.e. nematodes, serve primary roles in nutrient cycles. Fertile (arable) soil microbiome is an ultimate goal of OA, hopefully to provide sustainably cultivar land with fertile microbial abundance and metabolic food networks. Studies demonstrated agrotechniques affected soil microbial diversity, and suggested long-term

IA change soil microbial structures likely in a way to decrease species abundance and activities compared with OA (Bossio et al., 1998; Letourneau and Goldstein, 2001; Berkelmans et al., 2003). OA soils also showed decreasing plant pathogens through the arm of beneficial soil flora (Griffiths et al., 1994). Whilst individual microbe application showed inconsistent results, the databases of diverse soil microbiome suggestive to core microbiome applications have resolved various agriculture plant and soil dilemma, i.e. salt-tolerant plant and improved soil health (Vandenkoornhuyse et al., 2015; Qin et al., 2016; Schmidt, Bowles and Gaudin, 2016). However, soil microbiome vary across geographic regions and crop types (Bulgarelli et al., 2012; Lakshmanan, 2015; Agler et al., 2016).

Therefore, the database and knowledge of soil microbiome in cultivar farms in Thailand is priceless. In Thailand, the agriculture soil microbiome database is still a tip of an iceberg since the next generation sequencing is a recent advance technique. Several essential studies thereby rely on bacterial culture, individual clone sequencing and non-high throughput sequencing (i.e. community profiles separation by automated ribosomal intergenic spacer analysis and terminal restriction fragment analysis) (Chawanakul et al., 2009; Doi and Ranamukhaarachchi, 2009; Sooksa-Nguan et al., 2010; Chaiyasen et al., 2014; Chunjaturas et al., 2014; Siripattanakul-Ratpukdi et al., 2015); hence, the true microbiota remains to fulfill the missing puzzles.

# **Study objectives**

To support the understanding and sustainable management of the NE soil, we obtained a complete database of soil prokaryotic (bacteria and archaea) and eukaryotic (fungi, protists, plants and animals) microbiome in an upper NE named Sakon Nakhon province using metagenomics combined with 16S and 18S rRNA genes next generation sequencing. The correlations between microbial metabolic networks

and microbiota were also analyzed to retrieve putative links of key metabolic pathways and microbes that may be lost or overgrowth by IA. As the agrotechnical soil microbiome database in Thailand remains limited, the present study represents an initial database of the NE (Sakon Nakhon province), and this article includes a part of our results on prokaryotic and eukaryotic diversity (excluded soil characteristics correlation, and metabolic potential correlation analysis). Sakon Naknon is one of the 19 provinces in NE, with 0.3 m. ha rice cultivar area in 2005. This considers moderately high compared to other provinces in the NE.

### **Methods**

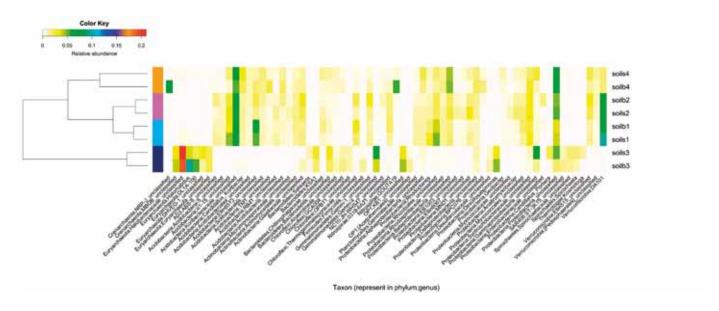
Four different agriculture sites in Sakon Nakhon province were selected to cover the soil agricultural diversity in the NE: (1) OA rice farm with polyculture and crop rotation in Kusuman District (site 1); (2) OA plus IA rice farm (near salt pan) with crop rotation in Wanorn Niwat District (site 2); (3) salt pan in Wanorn Niwat District (site 3); and (4) IA rice farm with crop rotation in Phanna Nikhom District (site 4). Samples were collected on 21-22 June 2014. Of each site, 10 independently soil sampling collections of 2 depths (~1-5 cm represent 'surface' microbiota and ~20-30 cm belowground represent 'root' associated microbiota) of 15 g each 1 m apart were collected in sterile containers. The surface and root microbiota are abbreviated 's' and 'b', respectively. With four total sites, soils1 represents surface at site 1, soilb1 represents root at site 1, soils2 represents surface at site 2, and so on.

Metagenomic extraction for prokaryotes and eukaryotes were performed on freshly collected soil samples using Power Soil DNA Isolation Kit (MoBio, Carlsbad, CA, USA), and determined quality and concentration via agarose gel electrophoresis and nanodrop spectrophotometry. The library constructions were performed using universal 16S rRNA gene (for prokaryotes) and 18S rRNA gene

(eukaryotes) primers with appended 5' Illumina adapter and 3' Golay barcode, following Caparaso et al. (2012). Each amplicon library was size-purified using GenepHlow<sup>™</sup> Gel Extraction Kit (Geneaid Biotech Ltd., New Taipei City, Taiwan) and quantified with Picogreen (Invitrogen, Eugene, Oregon, USA), and a minimum of independent triplicates were performed and equally pooled to minimize stochastic PCR bias. Sequencing was performed by MiSeq300 platform (Illumina, San Diego, CA, USA) at Chulalongkorn Medical Research Center, along with appropriate sequencing primers and index sequence (Caporaso et al., 2012). For bioinformatic analyses, Mothur's standard operating procedures (Schloss et al., 2009) were followed unless noted. Pearson product-moment correlation coefficient was used to evaluate an association between community structures and soil characteristics. For representative species of the community structures, Spearman's correlation was performed.

# Diversity of bacteria and archaea populations

Among groups, the bacteria of Proteobacteria phylum was dominated (avg. 31.43%), followed by Acidobacteria, Verrucomicrobia, Actinobacteria and Chloroflexi. For archaea, phyla Crenauchaeota, Euryarchaeota and Parvarchaeota were found. According to statistic computation, independent replicate sequencing results were merged (data not shown). Surface- and root-associated microbiota showed relatively close community structures (Yue & Clayton theta similarity coefficient, p-value 0.780; Jaccard similarity coeffecient, p-value 0.824), while of different agriculture sites showed disparate structures. While the soil microbiota between (1) and (4) should be of relative due to the land characteristics (data not shown: geography, temperature, pH and texture identity of soil) and crop plantation, the soil microbiota of (1) and (2) were most related. Examples of dominated species in (1) are several members in



**Figure 1** Percent composition of prokaryotic genera, along with the dendrogram clustering on the left (branch in different color denotes different soil groups). Of each group, relative percent abundance was shown bases on a color chart on the top left. To fit a limited space, genus with less than 1% relative abundance was omitted.

Source: Somboonna (2015)

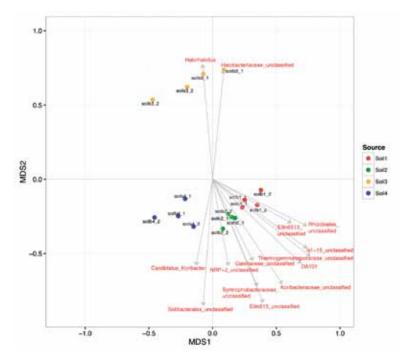


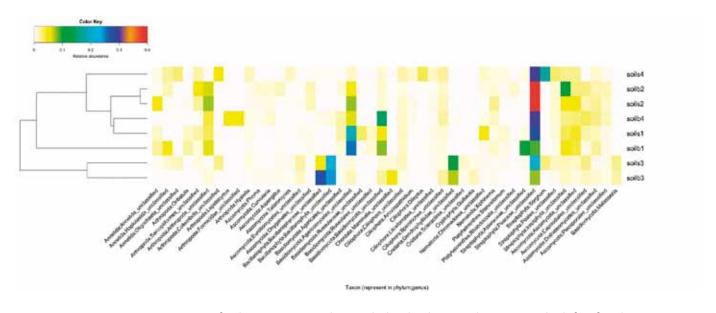
Figure 2 NMDS showing prokaryotic communities relatedness, and representative genera calculated by Spearman's correlation Source: Somboonna (2015)

phylum Acidiobacteria (e.g. iii1-15 and Ellin6513), unclassified in family Thermogemmatisporaceae (phylum Chloroflexi), Gemm-1 in phylum Gemmatimonadetes, 0319-6A21 in phylum Nitrospirae, Rhodoplanes and unclassified genus in order Rhizobiales (phylum Proteobacteria), unclassified in order Myxococcales (phylum

Proteobacteria), and DA101 in class Spartobacteria (phylum Verrucomicrobia) (Figure 1). This despites the fact that site 2 is a neighbor to site 3, a salt pan, and hence share soil characteristics to site 3. The separate cluster of site 4 potentially highlights OA apart from IA factors, give more extensive chemical inputs in site 4 (synthetic fertilizer, pesticide and

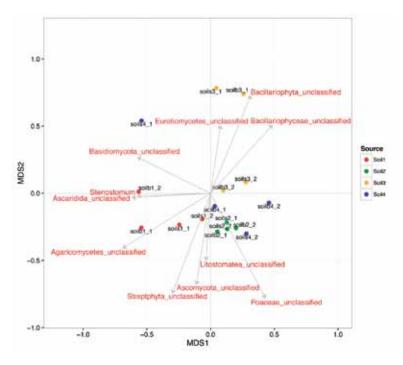
herbicide) than in site 2, which only experienced synthetic fertilizer. On the other hand, the uniqueness of site 3 highlighted the specific structure of the salt farm characteristics and hence soil microbial structure, as demonstrated by the more common archaea (e.g. family Halobacteriaceae and Halorhabdus in

phylum Euryarchaeota), bacterial members of phyla Bacteroidetes, Firmicutes and Spirochaetes. Soil of site 3 also denoted none to minute presence of Acidobacteria, Actinobacteria, Nitrospirae, Proteobacteria (the common phylum for soils of site 1 and 2, and fewer in 4), and Verrucomicrobia (Figure 1).



**Figure 3** Percent composition of eukaryotic genera, along with the dendrogram clustering on the left. Of each group, relative percent abundance was shown based on a color chart on the top left. To fit a limited space, genus with less than 1% relative abundance was omitted.

Source: Somboonna (2015)



**Figure 4** NMDS showing eukaryotic communities relatedness, and representative genera calculated by Spearman's correlation

Source: Somboonna (2015)

Prokaryotic community structures were plotted onto a non-metric multidimensional scale (NMDS) that showed IA may affect the soil characteristic in a way to separate microbiota site 4 from the others (data not shown). Figure 2 demonstrated many bacterial flora (e.g. Ellin6513, Rhizobiales, iii1-15 and DA101) that were statistically suggested to drive the OA soil of site 1 characteristics, diverged from site 44, given the vector length indicates the strength of the association and the vector direction indicates the direction of the effect. For the salt pan soil microbiota, family Halobacteriaceae (including genus Halorhabdus) in phylum Euryarchaeota were representative. These microbiota results allow prediction of microbial metabolic networks, and analysis for key metabolic pathways and associated microbes to OA (data not shown). This opens a venue for IA minimal core microbiome remediation, attempt to convert soil to once an OA soil microbial resources.

# Diversity of microbial eukaryote populations

Figure 3 demonstrated detail genera composition of microbial eukaryotes among sites, with independent replicate sequencing results (not shown) were merged. The community structures of soils of site 2 were clustered, embraced by soils1 and soilb4, and soils4, and then soilb1, in orderly. This pinpoints a partial community relationship of site 4 to 1 and 2, alternatively the closer between site 4 and 1 for the soil eukaryotic compared with prokaryotic microbiome.

The dissimilarity indices by the Jaccard (jclass) and Yue & Clayton theta (thetaYC) showed a minor closer of site 1 to 2 than 4 (jclass: soilb1 and soilb2 = 0.510, soils1 and soils2 = 0.503, soilb1 and soilb4 = 0.552, soils1 and soils4 = 0.548; thetaYC: soilb1 and soilb2 = 0.735, soils1 and soils2 = 0.218, soilb1 and soilb4 = 0.639, soils1 and soils4 = 0.448), consistent with the community structure plots in NMDS in Figure 4. Note the dissimilarity index ranges

from 0.000 to 1 and the closer to 0.000 infers the closer community similarity. Possibly, the eukaryotic profiles include organisms of the greater size than bacteria, hence more resistant to chemical inputs in site 4. This reflects the complex of nature microbiota. Signature microbes to site 1 was animal Ascaridida (phylum Nematoda) and fungi Agaricomycetes (phylum Basidiomycota) (Figure 4), and moderate abundance of plant Poaceae (phylum Streptophyta) (Figure 3). Similar to prokaryotes, the eukaryotic community structures of site 3 remain distinct, with relatively low frequency of annelids (earthworms), arthropods and fungi Basidiomycota, but abundance in Bacillariophyta (phylum) plants (Figure 3). Annelids, nematodes and fungi (e.g. Basidiomycota and Ascomycota) that were more common in crop farms (soil of site 1, 2 and 4) (Figure 3) function significant regulators for soil organic nutrient dynamics, including tillage, fertilization, and soil texture (Fonte, Winsome, and Six, 2009). Soil organic matters is crucial to metabolic food networks in soil ecosystem (Lal, 2004; Fonte, Winsome, and Six, 2009).

### Conclusion

Today, people and government are aware of sustainable environments as well as microbial resources, the fundamentals to the diverse metabolic food networks. The market and agrotechnique for organic crops arise. This study represents the first to uncover the agrotechnique-associated microbiota, both prokaryotes and eukaryotes, in the NE of Thailand. The data are useful to academic and public interests, support elucidation of the core microbiome to convert soil health to crop yield in an environmental safe manner.

# Acknowledgement

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# **Diversity of Parasitic Wasps in Thailand:**

# **DNA Barcode and Taxonomy**

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# Introduction

Thailand is still a largely agricultural-based country in terms of employment, where agriculture employs some 40–45% of the population (from ca. 70% in 1980), although much of this is subsistence agriculture by the poorest sections of society, and so although agricultural products are still an important income of the country, it now accounts for ~8% of the GDP. However, the advent of large monocultures has led to a dramatic expansion of pests, including predominantly insect pests, of these crops. Such pests cause serious problems to all agricultural countries, where if they are left unabated they will destroy the agricultural products. The larvae of Lepidoptera, known as caterpillars, are important pests of many crops, where they cause a high loss of crop yield and quality, and hence economic loss. Therefore, farmers have increasingly relied on the use of pesticides to control pest populations on the crop, but this leads to many problems, such as higher production costs, selection for insect resistance to the pesticides, and environmental and ecological problems, including negative impacts on both the farmers' and consumers' health.

Parasitic wasps have been used as natural enemies to control the population levels of insect pests in biological control programs. This method has many benefits, such as a low production cost and no adverse affects to the environment, including the farmers' and consumers' health. However, before using parasitic wasps for biological control, both the insect pest must be accurately identified to the species level and suitable parasitoids be identified that will control the pest population. Such host-parasitoid databases are not available for many current and potentially emerging future pests, while classical rearing and morphological-based taxonomics are expensive, time consuming and problematic in terms of rearing mortalities, leading to no fast alternative solution to pesticide treatment for emerging pests.



However, nowadays molecular techniques can be used to rapidly identify insect pests to the species (or molecular operational taxonomic units, MOTU) level, as well as their natural (indigenous) parasitoids.

In this research DNA barcoding was used to identify both caterpillars and their natural associated parasitoids, and so their relationships, at the species (MOTU) level. Caterpillars were collected from many areas, but principally from Chulalongkorn University, Kaeng Koi District, Saraburi Province and the Center of Learning Network for the Region (CLNR), Wiangsa







Figure 1 Collecting larvae within Chulalongkorn Area, Kaeng Khoi, Saraburi: Storing preserved larvae in 95% ethanol (top); Beating sheet (middle); larvae (bottom) Photographed by: Butcher, July 2014

district, Nan Province. Wild-caught caterpillars were examined for any ectoparasitoids or emerged endoparasitoid larvae/cocoons, and then preserved in 95% (v/v) ethanol. Subsequently each caterpillar specimen was dissected to see whether it had been parasitized by endoparasitoids or not. In case of parasitism, tissues of both the caterpillar and parasitoid were then collected into 96 well-plates for commercial DNA barcoding analysis by BOLD (Barcode of Life Data System), Canada. Thus, a preliminary database of the diversity and relationships of caterpillars and parasitoids in Thailand is being established.

Relationships between caterpillars and their associated parasitoids have co-evolved for a very long time. Normally, parasitoids are smaller compared to their insect hosts and have a high ability (efficiency) to search for their hosts, even at low densities. Some species of parasitoids can alter the behavior and development of their insect hosts (Hassell and Waage, 1984). Parasitoid larvae develop, eat and live either on or inside their hosts, and once they reach maturity, the hosts are killed (and hence the biocontrol aspect, since the host is killed prereproduction). Insects that have parasitoid life histories are found in the orders Hymenoptera, Diptera and Strepsiptera, but most parasitoids are classified in the order Hymenoptera. Due to their life histories as parasitoids and their prevalence (~25% of all insects are parasitoids), these group of insects are important in natural ecosystems, and this is also true for agricultural habitats (Godfray, 1994), because they can be used as natural enemies to control the population level of their hosts down to a low threshold limit (LaSalle and Gauld, 1993; Hassell, 2000). Parasitic wasps are all classified in the order Hymenoptera, the highest evolved group of insects and second most diverse to beetles (order Coleoptera), or may even be more diverse than beetles (Quicke, 1997, 2015). From their life history, parasitic wasps are divided into the two groups of endoparasitoids, where the larvae develop inside the host's body, and ectoparasitoids, where the larvae develop outside the host's body, (Godfray, 1994; Quicke, 1997, 2015). Shaw and Huddleston (1991) studied the biology and diversity of parasitoids and revealed that overall parasitic wasps could attack insect hosts in at least 11 insect orders, including other arthropods, such as spiders, ticks and mites (Gauld and Bolton, 1988) and centipedes (Newman, 1867). However, any given parasitoid species is typically restrained to one to a few related species of hosts. Many species of parasitic wasps are natural enemies of agricultural insect pests, and so can be used to control the population level of the pest in biological control programs with a high success rate compared to other natural enemies. This is because parasitic wasps have a well-developed nervous system and semiochemical/vibration detecting sensors that impart a high searching ability even at low host densities. Therefore, using parasitic wasps as natural enemies to control pest population levels, typically as part of an integrated pest management program, would lead to a reduction in the use of pesticides. The study of the relationship between insect hosts and parasitoids would lead to a database of suitable (natural indigenous) parasitoids for existing or emerging agricultural pests to be applied in biological control programs, as well as have conservation related benefits.

Overall, insect hosts in every stage of development can be utilized by parasitoids, although any given parasitoid species will be specialized for and restricted to using just one or two stages, such as egg, larvae, pupae, larvae-pupae and adult attacking parasitoids. Parasitoids may be either solitary, where only one parasitoid will develop per host, or gregarious, with up to 1,000 parasitoids in a single host, (Quicke, 1997, 2005). In this research, the study focused on caterpillars (Lepidoptera larvae), because caterpillars are the major pests of most agricultural crops, and, due to climate changes, are currently

subject to rapid population increases. This overpopulation of some lepidopteran species that are agricultural pests will impact on the quality and quantity of agricultural products in the future (Dokchan, 2013). At present, 4,087 lepidopteran species have been recorded in Thailand, of which 1,291 species are butterflies (classified into 10 families) and 2,796 species are moths (classified into 64 families) (Hutacharern et al., 2007; Ek-Amnuay, 2006).

Clearly, parasitoids are important to terrestrial ecosystems and economics. However, knowledge of their diversity, distribution and biology, especially the relationship between insect hosts and parasitoids, is limited not only in Thailand but worldwide (Quicke, 1997, 2015; Jones et al., 2009; Baselga et al., 2010; Santos et al., 2010). The study of insect host-parasitoid relationships by conventional means is difficult due to the limitation of the requirement to rear all the collected insect hosts to adults and then identify these to species level. This is not only time consuming, expensive and space restricted, but culture mortality, which can be high, imposes a strong restriction on data completeness and validity. In contrast, using molecular techniques of DNA barcoding of both the collected caterpillar hosts and associated parasitoids in any developmental stage (Traugott et al., 2013) removes these problems. DNA barcoding can accurately identify the relationship of caterpillars and parasitoids to the species (MOTU) level without the time and consumable expense and mortality-induced loss of data of culturing, and so can reliably answer ecological questions. Because DNA barcoding technique is very popular in the study of insect host-parasitoid relationships and molecular identification (Hajibabaei et al., 2006; Smith et al., 2008; Wirta et al., 2014) a large database of known species' sequences is already available and rapidly expanding, making identification to species or closely related MOTU, and phylogenetic placement unambiguous and rapid, in contrast to specialist morphology-based taxonomy.





Figure 2 Dissection of larvae, revealing gregarious parasitism

Photographed by: Butcher, January 2015

### Research methodology

### 1. Insect sampling

Two methods were used to collect caterpillars. The first was hand collection, which allows detailed examination of the caterpillar and area without disturbance for attached or nearby ectoparasitoids or emerged endoparasitoid larvae/pupae, but is restricted to sampling within the visual range and limits and is slow. The second method was the use of a beating sheet to increase the chance to collect more caterpillars and from areas not within the visual range (Deutscher et al., 2003). These are shown in Figure 1. Caterpillars were collected during 06.00–09.00 h and 16.00–18.00 h for 2 d every 2 weeks for 13 months. GPS data was recorded using a GARMIN eTrex 30, and other physical factors were recorded by Digital Hygro-Thermometer HTC-2.

# 2. Specimens photographing and dissecting

Wild-caught caterpillars (preserved in 90% (v/v) ethanol) were brought back to Integrative Ecology Laboratory for further studies. The caterpillar was then briefly rehydrated in water to soften the tissues and measured for length and width using Vernier calipers. Voucher numbers were given to all specimens, along with the matching record of

collecting location, method and date of collection, etc. The caterpillars were then photographed using an Olympus digital camera TG-4 for identification using the Manual of Butterflies and Caterpillars (Lewvanich, 2001) and Plant Diseases and Insect Pests of Economic Crops (Ek-Amnuay, 2010).

Each caterpillar was then dissected, the presence of parasitoid larva(e)/egg(s) inside the parasitized caterpillar was examined for carefully and recorded, and discriminated between gregarious (more than one parasitoid larva inside a caterpillar) (Figure 2) or solitary (single parasitoid larva found inside a caterpillar) (Figure 3). Sometimes a parasitized caterpillar can be detected by the oviposition wound in the exoskeleton of the caterpillar, which is caused by female parasitoid's ovipositor puncturing inside the caterpillar body to transfer eggs (Figure 3), or by the encapsulated remains of eggs or larvae within the caterpillar, and so these were also searched for and recorded.

# 3. Using DNA barcoding to identify caterpillars and parasitoids

If the caterpillar was parasitized, the tissues from eggs/larvae of the parasitoid(s) and the caterpillar were collected separately in 100% (v/v) ethanol in 96 well-microtiter plate and then sent for commercial

sequencing to the Canadian Centre for DNA Barcoding (CCDB), Biodiversity Institute of Ontario, University of Guelph, Canada. At that commercial institute the DNA was extracted by Chelex extraction by Proteinase K digestion and then the mitochondrial cytochrome oxidase 1 gene fragment ( $\sim$ 668 bp) was PCR amplified using Platinum® Taq DNA Polymerase (Invitrogen $^{\text{TM}}$ ) and sequenced. The sequence was sent back to the Integrative Ecology Laboratory, Department of Biology, Faculty of Science, Chulalongkorn University.

# 4. Data analysis and preliminary DNA barcoding database of caterpillars and parasitoids

DNA sequences were compared from the GenBank and BOLD databases using the BLAST version 2.2.31 (NCBI; www.ncbi.nlm.nih.gov) and BOLD Systems version 3 (Barcode of Life Data System; http://www.boldsystems.org), respectively. The data were then analyzed for establishing the DNA barcoding database of the caterpillars, parasitoids and their relationships for the samples collected in this research.

### Results

Currently, more than 10,000 lepidopteran larvae have been sampled from within Chulalongkorn

University, Kaeng Khoi, Saraburi. Larval dissections have revealed an average parasitism rate of 10% from parasitic wasps and parasitic flies (Diptera: Tachinidae). For the parasitic wasp larvae, most (70%) were from the subfamilies Microgastrinae, Agathidinae, Eulophinae, Aphelininae and Pteromalidae. Microgastrinae (Genus Cotesia, Diolcogaster and Apanteles) and Agathidinae (Genus Therophilus) are classified in the superfamily Ichneumonoidea, while the others (Eulophinae, Aphelininae and Pteromalidae) are classified in the superfamily Chalcidoidea. Most of the parasitized wild-caught caterpillars were from the families Noctuidae and Geometridae.

Preliminary results show that parasitic wasps in the subfamily Microgastrinae are the most common parasitoid that parasitized caterpillars when compared with other groups of parasitoids. Therefore, they could be considered as potential natural enemies to control populations of insect pests in biological control programs. However, this is only a preliminary result. Sample collections from agricultural areas and natural habitats are needed to further study the host-parasitoid relationships, while the affect of seasons on the parasitism rate and the population dynamics of caterpillars and parasitoids also requires more attention.



Figure 3 Trace of parasitism from external wound punctured by female parasitic wasp's genitals (Left) with parasitic wasp larva inside (Right)

Photographed by: Butcher, January 2015

### Conclusion

From a collection of over 10,000 wild-caught caterpillars, 10% were found to be parasitized by parasitic wasps or tachinid flies. Most parasitic wasps found were classified in the subfamily Microgastrinae and the most parasitized caterpillars were Noctuidae and Geometridae. Still, this is only preliminary data, more samples from Thai's agricultural areas are needed to assess the overall levels of parasitism and the relationships between caterpillars and parasitoids on the economic crops, in order to select suitable parasitic wasps as natural enemies to manage insect pest populations in biological control program.

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# Bacterial Blight of Rice in Thailand: Current Status of Disease Epidemics

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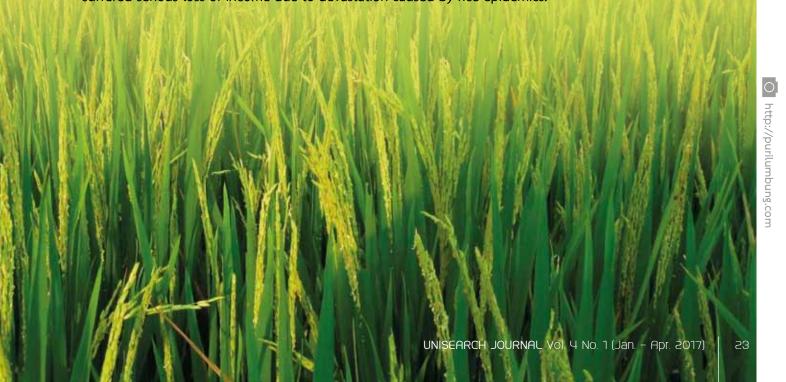
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# Introduction

Rice (*Oryza sativa* L.) is significant for the food security of humanity because rice is one of the most important staple foods for more than half of the world's population, especially in Asia (Chakravarthi and Naravaneni, 2006). Thailand is the world's sixth largest rice producing countries in the world, following China, India, Indonesia, Bangladesh and Vietnam. Thailand used to be the world's top rice exporter of high quality rice (Ministry of Agriculture and Cooperatives, 2004). However, in 2012-2013, Thailand's world ranking fell to third place, and was overtaken by India and Vietnam (USDA, 2013). However in 2014, Thailand regained its pole position as the world's top rice exporter and most recently, Thailand is now ranked second place among rice exporting countries (USDA, 2016).

In 2012-2013, Thailand had approximately 70 million rai of rice growing areas including both irrigated and rainfed lowland rice fields, together producing about 36 million tons of paddy rice nationwide. Productivity for irrigated and rainfed lowland rice averages 414 kg/rai and 668 kg/rai respectively (Rice Department, 2013); these yields are below those achieved by other Asian countries, resulting in higher production cost per tonne, and a resulting lower competitiveness compared to neighboring countries.

In addition, climate change has brought higher average temperatures and a rise in sea level; among the impacts of climate change are a rise in the incidence of important rice diseases. Farmers have recently suffered serious loss of income due to devastation caused by rice epidemics.



### **Bacterial blight disease**

Bacterial blight disease caused by *Xanthomonas* oryzae pv. oryzae (ex Ishiyama) (Swings et al., 1990) (Syn. *X. campestris* pv. oryzae (Ishiyama Dye)) (Swings et al, 1990) is a major disease of rice, with epidemics affecting all rice growing areas in Thailand, especially irrigated rice. In rainfed lowland rice, where there is favorable environment for disease such as high water level in the field, poor drainage, storms, floods, continuous monocropping with the same cultivar, or high nitrogen application and close spacing, severe and rapidly-spreading epidemics can devastate crops (Distaporn, 1989; Rice Research Institute, 1996; Srichumpa et al., 1998).

Epidemics of bacterial blight disease in rice occur in tropical and subtropical rice-growing areas worldwide. The first bacterial blight in rice epidemic was found in Japan in 1884 (Tagami and Mizukami, 1962). In 1960, bacterial blight epidemics were reported in several rice-growing areas in Asia including the Philippines, Indonesia, India, Sri Lanka, China, Taiwan, Thailand and Vietnam. Epidemics also

spread to Australia, USA, the African continent, Latin America and the Caribbean (Robert, 1992).

Bacterial blight disease was first found in Thailand in 1963, in Pathum Thani province in 1963 (Eamchit, 1982) with widespread leaf blight symptoms (Figure 1) at maximum tillering stage. Another bacterial blight symptom presented at seedling or transplanting stages is kresek or wilt, first reported in Indonesia (Reitsma and Schure, 1950) and then in Korea (Yu and Cho, 1978).

In August 2003, an epidemic of bacterial blight with kresek or wilt symptoms (Figure 2) was found in Khao Dawk Mali 105 (KDML 105) at 1–1.5 month post-transplanting grown in rainfed lowland conditions in Ban Khakom, Pa Ow subdistrict, Muang district, Ubon Ratchathani province in northeast Thailand. Recently, bacterial blight with kresek or wilt symptoms (Figure 3) was again found in KDML 105 rice in September 2016, this time in Ban Srangmeg, Moo 3, Nonghang subdistrict, Benjaluk district, Sisaket province.

# leaf blight

# Founfed maximum tillering stages



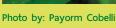




Figure 1 Bacterial blight symptoms at maximum tillering stage Photographed by: Cobelli, September 2016 (left) and October 2004 (right)

# Kresek or wilt

# Founfed in seedling tillering stages





Photo by: Payorm Cobelli

Figure 2 Kresek or wilt symptoms of bacterial blight, 1-1.5 months post-transplanting.

The epidemic was found in 2003 in Ubon Ratchathani province.

Photographed by: Cobelli, August 18, 2003

Currently (2015-2016), severe epidemics of bacterial blight are widespread, especially in areas growing susceptible rice varieties. In northeast Thailand, susceptible rice varieties such as KDML 105, RD15, RD6, RD12 and Sakon Nakhon were damaged by bacterial blight which reduced rice production by 10-30%. In the lower North and Central regions, severe epidemics presented in RD31, RD41, RD47, RD49, RD53, RD61 and Phitsanulok 2 with 100% epidemic and disease severity score level 9 was found in RD49. In the Southern region of the country, a severe epidemic was found in September 2016 at the flag leaf to harvesting stage in RD41 and RD49 rice. This epidemic and stages of infection were similar to bacterial blight disease presented in Sunpatong 1 and RD49 in the North. The damage of bacterial blight on flag leaves will affect rice production by decreasing yield, resulting in unfilled grains, low grain weight and poor quality grain. The flag leaf is a primary contributor to grain filling because it supplies photosynthetic products mainly to the panicle (Figure 4). In conditions favouring the disease (high water level in field, poor drainage, storms and floods), bacterial ooze, small yellow exudates of bacterial cells, will be presented at the edges of the disease lesion (Figure 5). These bacterial oozes will dry out and turn brown, before spreading through water and rain.

Currently, bacterial blight disease epidemics in Thailand are intensifying, and urgent strategies for disease management are needed in the short, medium and long terms. Short-term or urgent measures generally depend on chemical sprays, which are only partially effective and environmentally harmful. Medium term management strategies rely on integrated pest management or biological control- which are difficult, time-consuming and location-specific. In the long term, selection of

# Kresek or wilt

# Founfed in seedling tillering stages





Photo by: Payorm Cobelli

Figure 3 Kresek or wilt symptoms of bacterial blight, 1-1.5 months post-transplanting.

The epidemic was found in 2016 in Sisaket province.

Photographed by: Cobelli, September 7, 2016



Photo by: Payorm Cobelli

Figure 4 Bacterial blight symptom on flag leaf will affect rice production by decreasing yield, resulting in unfilled grains, low grain weight and poor quality grains.

Photographed by: Cobelli, September 21, 2016

## Bacterial ooze on bacterial blight disease lesion edge





Photo by: Payorm Cobelli

**Figure 5** Bacterial ooze on bacterial blight disease lesion edge. **Photographed by:** Cobelli, October 6, 2004 (left) and September 28, 2016 (right)

resistant varieties will be the most effective and the most environmentally friendly and sustainable strategy, which is also easy for farmers to adopt.

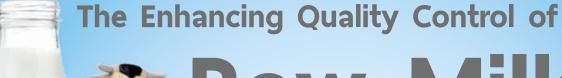
### **Acknowledgements**

This article is part of the research project entitled "Diversity Analysis of Xanthomonas oryzae pv. oryzae Causing Bacterial Leaf Blight of Rice from North, Northeast, Central and Southern Thailand" by conducting both phenotypic

and genotypic analyses to study the effect of plant diseases on near-isogenic-lines (NILs) of rice. The lead researcher for this project is Dr. Payorm Cobelli who also serves as a research committee member under the project of "Diversification and Data Base Management for Major Rice Pathogens". The project was approved by the National Research Council of Thailand and funded by the Rice Department, Ministry of Agriculture and Cooperatives (2016–2018).

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# Raw Milk at Milk Collection Centers

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# Introduction

Dairy farming in Thailand has been promoted both by the government and private sector. Since an introduction, the industry has constantly expanded. In 2015, it was reported that there were 509,524 dairy cows in Thailand, of which 235,829 were being milked, producing 1,084,162 tons of raw milk per year (Information Technology and Statistics Division, Information and Communication Technology Centre, Department of Livestock Development, Ministry of Agriculture and Cooperatives). Most of Thailand's raw milk comes from 16,248 small farm households. The produce is collected and transported to 158 milk collection centers nationwide before processing or transfer to dairy processing factories. The quality of raw milk is paramount for producing and processing high quality dairy products, and consumer trust is crucial. Therefore, milk collection centers are the first step to inspect and control the quality of raw milk, which consist of 1) milk composition including fat, protein and lactose content; 2) safety such as microbial and chemical contamination; and 3) somatic cell count (indicating health of the cow's udders e.g. evidence of mastitis).



Figure 1 Father of Thai Dairy Farming

King Bhumibol Adulyadej and King Frederick IX of Denmark officially chaired the opening ceremony of the Thai-Danish Dairy Farm and Training Centre on 16 January 1962, what is now a state enterprise under Ministry of Agriculture and Cooperatives. It is now called "Dairy Farming Promotion Organization of Thailand" (DPO).

Source: Dairy Farming Promotion Organization of Thailand (DPO)

# Factors affecting quality of raw milk and how to set the control

The key factors affecting the quality of raw milk include optimal animal nutrition management, good farm hygiene practices, farming, and milking. These factors affect the health of cows and can lead to abnormal conditions such as mastitis. Therefore, quality checks and control at the milk collection centers are essential quality processes that ensure the product complies with international standards and thus enhance its value. Effective quality checks could turn a page for dairy farming and milk production for the entire country. In order to maintain the quality of raw milk to meet international standards, the National Bureau of Agricultural Commodity and Food Standards, Ministry of

Agriculture and Cooperatives, established Good Manufacturing Practices for Milk Collection Centers.

The Good Manufacturing Practices for Milk Collection Centers consist of requirements on the following issues: (The National Bureau of Agricultural Commodity and Food Standards, 2015)

- 1. Facilities
- 2. Operation controls
- 3. Maintenance and sanitation
- 4. Personal hygiene
- 5. Transportation
- 6. Audit
- 7. Training
- 8. Member promotion system
- 9. Documentation and recording system.

The project to enhance quality control of raw milk at milk collection centers originated from the cooperation with Khao Yai Dairy Co., Ltd. which operates medium sized milk collecting center situated in Pak Chong Subdistrict, Pak Chong District, Nakhon Ratchasima Province. The project team sent specialists and experts to train staff at the milk collection center, establish understanding and participation in creating awareness relating to milk quality among dairy farmers. These personnel also introduced the quality check system and a feedback and response system. All these efforts aim to ensure

best practices are followed at milk collection center, which will lead to fewer losses in raw milk prices.

Since the milk collection centers are the first checkpoint for quality inspection of farms to sort the grades and determine the buying price, appropriate methods are required to ensure effective quality checks. Staff performing this task need to be qualified with proper training and ability to undertake the checks accurately. Staff should also possess relevant knowledge in the scientific basis of diagnostics of mastitis, laboratory quality of raw milk, and information from relevant research, especially factors



Figure 2 Selection of dairy cow breeds; most Thai farmers prefer crossbred Holstein Friesian Photo taken at World Dairy Expo 2016, Madison Wisconsin, USA Source: Ajariyakhajorn (2016)



Figure 3 Age and milking period of dairy cows
Photo taken at World Dairy Expo 2016, Madison Wisconsin, USA
Source: Ajariyakhajorn (2016)



Figure 4 Feed management; the proportion of roughage and concentrate affects milk quality and productivity

Photo taken at Dairy Farm, Wisconsin, USA

Source: Ajariyakhajorn (2016)



Figure 5 Good barn management with sufficient ventilation for cow comfort

Photo taken at Dairy Farm, Wisconsin, USA

Source: Ajariyakhajorn (2016)



Figure 6 Free stall is floored with fine sand for cow comfort and cleanliness Photo taken at Dairy Farm, Wisconsin, USA Source: Ajariyakhajorn (2016)



Figure 7 Types of milking system; milking tools must be installed properly and well-maintained for good performance; good milking hygiene is also important for milk quality

Source: http://www.interpuls.com/products/milking-clusters.html

(October 1, 2016)

affecting raw milk quality (Figures 2-12). For example, they should have a good practical understanding of dairy cattle breeds, milking season, size of farm, farm management, bull selection, milking, quality grading, and buying prices for raw milk. In addition, staff should be trained to educate farmers in order to

PALATTER WILL FIFE LINE

STALL FIFE LINE

STALL COCK

Figure 8 Bucket type milking system

Source: http://www.old-engine.com/empire.htm (October 1, 2016)

fulfill the process of raw milk quality check, feedback system, and giving advice to farmers.

During the one-year research period, the researcher and the entrepreneur (Khao Yai Dairy Co., Ltd.) have an excellent good opportunity for developing human resources in quality control, process development, and quality checks which are more accurate and reliable. A quality check manual will be developed as well as a feedback and response process and basic instructions to elevate the quality of raw milk at the farm level and at milk collection center. Successful project implementation is expected to result in significant reduction of losses suffered by dairy farmers and milk collection center. Moreover, the project's approach to milk quality control could also be extended and applied to upgrade quality of other milk collecting centers.



Figure 9 Pipeline milking system of a large dairy farm in Vietnam Source: Ajariyakhajorn (2016)



Figure 10 Rotary milking system of a large dairy farm (2,000 cows) in Wisconsin, USA

Source: Ajariyakhajorn (2016)



Figure 11 Robotic milking system

Source: http://robotsinsider.com/wp-content/
uploads/2013/07/Mark-Redeker3.jpg (October 1, 2016)



Figure 12 Milk collection centre with milk quality grading and pricing of Khao Yai Dairy Co., Ltd.

Source: Ajariyakhajorn (2016)

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# Use of Cement By-Product (Silicon Dioxide) for Swine Production

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# Introduction

Silicon dioxide or silica (SiO<sub>2</sub>) comprises of silicon and oxygen and is found naturally in many forms, particularly sand and quartz, and in the cell membrane of diatoms. In nature, it may contain a range of heavy metals at various levels depending on its origins. SiO<sub>2</sub> is virtually insoluble in water or acids, with the exception of hydrofluoric acid. However, it dissolves well in base. SiO<sub>2</sub> is highly stable at normal temperatures and is non-combustible. Amorphous silica is more chemically reactive than crystalline silica due to its larger surface area. It is used in many industries such as mirror, glass, crystal, ceramic, stoneware, cement etc.

In animal industry, SiO<sub>2</sub> is widely used as a carrier, diluent, or feed additive. For pig production, losses occur during the early stages of growth due to a range of causes such as small piglets (dwarfism) and low growth rate before weaning due to weakening of the animals. The piglets are also highly sensitive to any infection or environmental stress, such as unsuitable temperature and moisture.



### Use of silicon dioxide to reduce neonatal piglet mortality

The use of piglet powder to preserve heat production and to reduce incidence of diarrhea has proven successful in reducing piglet mortality. Silicon dioxide, as a by-product of the cement industry, absorbs moisture. Its appearance is quite similar to commercial piglet powder already available in the global market, including chemical composition, physical characteristics, binding capacity, and moisture absorption capacity. Piglet powder is used mainly to maintain body temperature and absorb moisture in order to reduce the number of pathogen such as *Escherichia coli (E. coli)*. It has been hypothesized that silicon dioxide powder is retained on the piglet skin, reducing heat loss, reducing skin moisture and infections by pathogens. Silicon dioxide therefore has potential for use as piglet powder at birth.

A preliminary study was conducted to evaluate the possibility of using this cement by-product as piglet powder. The  ${\rm SiO}_2$  used in this study was a very finely ground powder resulting from grinding clay shale with particle size smaller than 0.75 millimeter (Figure 1).



Figure 1 Silicon dioxide appears as a very fine yellow-brownish powder.

Photographed by: Jamikorn, January 2016

When applied over the skin of newborn piglets (Figure 2), as compared to commercial available piglet powder, after 24 hours post-application, no significant difference was observed between the two powders in adhesion (Figure 3), body temperature (Figure 4), drying of umbilical cord, and appearance of piglets. All piglets reached the sow's udders and nipples normally.



Figure 2 Newborn piglet was applied with silicon dioxide powder Photographed by: Jamikorn, January 2016



Figure 4 Measurement of piglet skin temperature that fell in normal range Photographed by: Jamikorn, January 2016



Figure 3 Adhesion of powder at 24 hours after birth Photographed by: Jamikorn, January 2016

However, general use of piglet powder would be more effective during low ambient temperature and/or high relative humidity. Unfortunately this current study was conducted during hot and dry weather therefore the efficiency of powder was not well recognized. Nevertheless, there appears to be a possibility to use silicon dioxide powder as a byproduct of the cement industry, as piglet powder for value added in commercial market.

### **Acknowledgement**

This article is part of the research project entitled "Pretest of SiO<sub>2</sub> Powder with Piglet" funded by The Siam Cement Public Company Limited.

# Prof. Mongkol Techakumphu, DVM.



# From Research Knowledge to Agriculture and Smart Farming

Prof. Mongkol Techakumphu, DVM. is an expert in biotechnology for animal reproduction with more than 30 years of experience. He broadens his research knowledge to apply to livestock management towards smart agriculture and transfer his research to livestock industry for implementation.

### **Smart Agriculture**

"...Bringing the concept of smart agriculture to improve the way farmers work will help them reduce costs, improve quality, and mitigate agricultural risks..."

Global changes in geography and climate are forcing farmers around the world, including Thailand, to reform the way agriculture is done. Scientific knowledge, technology, and research data are increasingly being used to improve crop and livestock farming under the concept of smart agriculture.

Urban expansion has resulted in increasing labour shortages in the agricultural sector as well as a declining trend in agriculture, despite an increasing demand for food due to

the world's growing population. By 2050 the world population will have increased from 6 billion to 9 billion people. We will face increasing competition for resources, especially food and water, to sustain the planet's population.

The concept of smart farming concerns the interpretation of information technology, analyses of available data, and the convergence of new technologies in order to optimize production precisely. For example, satellite technology and aerial photography using drones are today used extensively to survey and analyze farm areas and assess production and crop damage from pests or floods. The survey and analysis data are then used to determine suitable zoning for specific types of crops/breed, when



to grow them, how many to grow, how the soil should be improved, and which area is suitable to breed what kind of livestock animals. The purpose of this is to prevent oversupply or undersupply, which inevitably affects the price of the next season of produce. Therefore, the application of technology for livestock and crops, can substantially help farmers in terms of cost reduction, yield improvement, reducing the risk of farming, maximizing productivity with limited and finite natural resources.

### **Smart Farming in Thailand**

"...Smart farming is the application of information technology to interpret and analyze the data for the most accurate and optimized agriculture..."

In Thailand, only a few big companies actively apply the concept of smart farming for example in industrial pig and chicken farming. These 'smart farms' take into account the needs for each age of livestock and apply the technology with genetic innovative

improvement in order to have good breeds suitable for farming conditions. In addition, the organic concept is also employed to enhance value-added. However, 90% of farmers have yet to apply the concept. Therefore, in order to apply smart farming concept in Thailand, it is important to use academic knowledge to support small farmers in four topics: production, housing, management, and nutrition.

Formerly, the critical problems in livestock farming in Thailand were nutrition and animal diseases. For example, foot and mouth disease in dairy cattle and pigs having a high impact on Thailand's livestock industry. Porcine epidemic diarrhea virus (PED) and shrimp early mortality syndrome (EMS) have also caused extensive impacts. To address these problems, private companies still have to rely on the latest research. Therefore, it is important to conduct talent mobility by promoting connection and collaboration among universities and industries or private businesses. To do so, they have to learn from each other in order to share data and knowledge. It is another way to improve the performance and development of Thai agriculture and enhance its global competitiveness. In particular, universities need to encourage researchers and lecturers to spend time in the field with businesses and industries. This is because class lessons should be based on practical as well as theoretical experience. Also, lecturers should also function as researchers in order to drive the country toward the concept of Thailand 4.0.

### Transfer of knowledge from universities to farms

"...It is important to emphasize to small farmers and develop a model by working with government agencies and determining clear responsibilities from upstream, midstream, to downstream..."

The role that universities must play in order to drive smart farming forward is to work closely with small farmers and develop a model by working with government agencies and determining clear responsibilities. To clarify, universities should play a role in the upstream part. That is to say, it is the job of universities to research and provide information to government agencies, such as Ministry of Agriculture and Cooperatives, which are the midstream. To apply the knowledge with the farmers is the downstream role. To do so, the government has to support universities by giving the research funding. Objective problems should be brought up to analyze and prioritize, considering the problem expected to consume low cost with high impact. Such problems shall be proposed as a long term project expected to help farmers in a long run.

The part that Chulalongkorn University should play is to engage in research to create new and relevant knowledge to address society's demand and real needs. Therefore, it is important to establish a clear research question and brainstorm experts from many different disciplines in order to develop a model. The model shall be then applied to the actual area to form a clearer model for a specific place, such as for Saraburi or Nan provinces. Some areas should receive special emphasis, and it is important to highlight

that when the knowledge from Chulalongkorn University is used for the area, it makes the area better. For example, the knowledge from the university was used to develop

a model of organic pig production in Nan Province. Also, the innovation of artificial insemination is introduced to farmers in the province. After that, it has been proven that farmers can repay some of their debts. Some farmers have a better life. It is regarded as a great success and the same process has been improved ever since.

It is true that Chulalongkorn University alone cannot distribute the help to farmers all over the country. But what it can do is to help farmers around the university. The fact that Thailand has many universities in every region will certainly help farmers in each area by sharing knowledge and cooperating among the universities so that research is most beneficial to the society.



### Problems and Obstacles of Smart Farming in Thailand

"...Failure to look at the problem in overview, together with a lack of cooperation among researchers, slow down the development and practical application of research.

Also, the resultant operation is not continued or sustainable..."

One obstacle to uptake of smart farming is the failure to look at the problem in overview. The lack of cooperation among researchers in universities makes them work in different directions. Instead of aligning and working together, researchers fight for budget to conduct research. The research findings could not be then applied. The resultant operation is not continued and sustainable while the country faces the same problems over and again. If researchers can overcome their ego and open up to the opinions of others in the interest of the public or the nation and work together on beneficial research, this will enable research institute to survive.

While the government needs to allocate budget to meet the country's critical needs, the questions should be clear and the criteria should be revised. In the past, criteria for granting research funding focused on the publications in an international journal. Of course, publication in international journals is necessary for all researchers. But the real essence should be the knowledge learned from that research. Formerly, there were many unpublished research works, whose findings could be applied in practice to solve real-world local problems. Some could be made into policy for area management.

When combined with the knowledge beneficial to the country, such research can be as relevant and influential as internationally published research. Supporting agencies should be able to distinguish between research that is beneficial to the country and one can be applied with others.

### Trends in smart agriculture and smart farming in Thailand

"...The trend of agriculture in Thailand should be a combination between smart farming and sustainable farming..."



Thailand has limited areas for agriculture while the demand for product is higher to satisfy the growing population. The trend of agriculture in Thailand should be the combination between smart farming and sustainable farming. The problems should be solved from the root cause. Productivity should be improved. The development to support the policy of the world's kitchen is inevitable. Smart farming does not only improve productivity, but also reduces the risk for farmers. In this, Thailand is already ready at some level. However, with more cooperation among researchers both within Thailand and with other countries, the pace of national development and improvement in the well-being of our people could both be greatly accelerated.

### Prof. Mongkol Techakumphu, DVM.

Prof. Mongkol Techakumphu, DVM. is a former vice president responsible for research and innovation at Chulalongkorn University and Dean of Faculty of Veterinary Science, Chulalongkorn University. He graduated with a Ph.D. in Physiology of Reproduction (Doctorat de 3e cycle, Paris VI (Pierre et Marie Curie)), France. Many of his works have been great contribution to the society, especially in term of livestock such as pigs, cows, goats, and sheep. He has published more than 200 publications. Prof. Mongkol Techakumphu, DVM. originated the cooperation among universities in Thailand to help each other solve problems. The collaboration is called Research University Network (RUN) which has now attracted cooperation from other countries.

Prof. Mongkol Techakumphu, DVM. is currently an advisor to Chula Unisearch, lecturer at the Department of Obstetrics, Gynnaecology and Reproduction, Faculty of Veterinary Science, Chulalongkorn University, and director of RUN office.





# Farmer's Quality of Life: Progress towards A Sustainable Future

Thailand's unique cuisine and the quality and diversity of its agricultural products and exports to all corners of the world have given Thailand its well-earned reputation as the 'Kitchen of the World'. Agriculture has always been and remains a mainstay of the national economy; however, the sector must face mounting challenges to its global competitiveness, particularly in boosting productivity and creating new value added through downstream processing to create innovative agri-food products. Farmers are highly vulnerable both to fluctuations in domestic and global market prices for raw agricultural commodities, and also to exploitation by merchant middlemen. In real terms, farm incomes and quality of life are on the decline, causing an

exodus of farm labour to urban centres and a steady rise in the average age of Thailand's farmers.

Thailand also needs to consider growing risks of food insecurity. The increasing frequency and intensity of extreme weather events such as the recent floods and droughts experience in Thailand serve to highlight the issue. Nevertheless, there appears to be a complacency both at government policy level and among the general public to this critical issue, and the country has no concrete long-term policy to address the root causes.

Thailand has more than 100 million rais of farmland, of which only 30 million rais are irrigated. The government's efforts to increase the irrigated area have met with limited success due to

unfavorable local topography and the high costs of rehabilitation of existing irrigation infrastructure often a burdensome legacy of inappropriate, poorly designed systems and corruption. The supply of water is often insufficient and not continuous. Irrigation systems are often damaged and land management frequently inappropriate; although the government has attempted to organize land management systems, many farmers remain untrained in, or even indifferent to the requirements of irrigated farming to conserve long-term soil structure and fertility. Most farmers take a short-term perspective and will do what it takes to meet market demand, regardless of the longer-term impacts on the sustainability of soil resources and the environment.

Exacerbating the situation is the increasing and inappropriate use of chemical fertilizers to boost yields and manage pests and diseases. Thailand has become one of the world's most intensive users of chemicals in agriculture, on a per rais basis. Despite their high costs, use of fertilizers and pesticides is rising, leading to a range of critical impacts aside

from the increased direct costs of production. Impacts include adverse effects on soil quality, farmers' health, water quality and biodiversity, as well as economic damage through rejection of exits at destination ports due to excessive levels of pesticide residues or even detection of banned products.

The concept of 'Sufficiency Agriculture' advocated by His Majesty the Late King Bhumibol Adulyadej embraces the concept of a more sustainable approach to farming; however, its meaning is not well understood by farmers, who still live according to a short-term perspective. Village loans intended to improve the farmer's production system are often diverted to purchase consumer goods such as mobile phones, motorcycles, etc. Farmers who are then unable to boost their productivity and incomes, risk defaulting on loans. A downward spiral inevitably follows, with recourse to the informal sector to raise funds, e.g. though loan sharks to repay the original debt. These usurious loans are subject to enormous daily interest rates, which subject the farmer to stress-social problems,





violent crime, suicides and landlessness are the inevitable result of this spiral.

Not all is gloom and doom, however. Major efforts are under way to enhance the country's agricultural competitiveness The government, private sector, research institutions, development agencies and grass-roots community organizations and farm communities are working together to develop innovative, practical solutions. The common goal of these efforts is to improve quality of life of farmers, including environment-friendly farming methods that conserve our natural resources and improve farmer health.

We can also learn from our neighbours. In only a few years, Viet Nam initiated a systematic reform of its agriculture sector, including farm size and the use of biochar to improve agricultural soils. Biochar has already been used for this purpose in many countries, especially in China, where its use is part of a major programme to reduce use of chemical fertilizers and pesticides in rice production. China's



research and support programme includes development of new biochar fertilizers, production tools and machinery. Despite impressive results indicating crop yield increases from biocharaugmented soils, Thailand has yet to prioritize research into the potential of biochar to enhance Thailand's agricultural productivity.

There is also a need for more sophisticated approaches to marketing of Thailand's agricultural products, including real-time price and market information, simulation models to forecast market scenarios, and differentiation of Thai product in the face of competitive challenges from its neighbours and other food producers and exporters around the world.

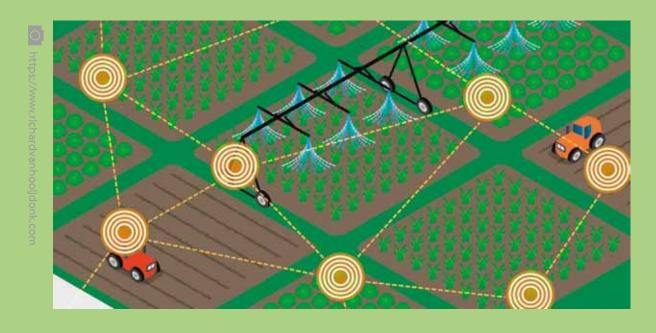
Policies also are in need of major reform. Recent history has highlighted the short-sightedness of supply-driven, and often politically-motivated programmes to boost production of specific crops. Market gluts and collapsing prices are a frequent consequence of such government-driven programmes.

Thai farmers are however innovative. Although naturally risk-averse, they are open to adopting new technologies to improve productivity and boost incomes, e.g. through restructuring their operations. However, government-promoted approaches such as 'Smart Farming'- a new style of agriculture aimed

at improving farming methods, can only be realistically adopted by farmers with an adequate budget, or large corporate farmers. Smallholders are marginalized by this approach, and cannot compete.

In order to ensure Thailand's farms and the agricultural sector can be sustainable in the long term, the government and relevant agencies most think anew to do a better job of helping farmers. We need to identify and implement bold measures to diversity production, ensure resilience to climate change impacts such as droughts or floors, protect soil fertility and the environment. Only through such measures can we preserve the quality of life for farmers and stem the exodus of rural people to the country's already over-crowded cities.

Transformation of the agricultural sector will be essential if Thailand's rural economy, farmers and deep-rooted cultural legacy are to survive. Under the philosophy of the "Sufficiency Economy" of His Majesty the Late King Bhumibhol Adulyadej, and including the widespread promotion of sustainable, climate-smart best practices such as biochar, together with effective conservation and management of water for agriculture, Thailand can indeed expect to remain recognized as the 'Kitchen of the World' and our treasured farmers can continue to live with pride and dignity.



# Application of Non-Steroidal Anti-Inflammatory Drugs to Enhance Artificial Insemination Efficiency in Pigs

Prof. Padet Tummaruk, DVM., MSc., Ph.D.



The development of Thailand's livestock industry, and especially the swine industry has focused on breeds, feed, and sanitation managements to meet international standards. However, high risks in swine breeding include pain for sows and piglets, with the possibility of injury, trauma and inflammation during and after breeding. Dystocia in particular is considered as a major cause of adverse health effects, animal welfare and productivity. Anti-inflammatory drugs or non-steroidal anti-inflammatory drugs (NSAIDs) are effective in reducing pain, inflammation and toxic endotoxins and as an antipyretic in postpartum sows. NSAIDs have the potential to significantly improve health of both sows and piglets, thereby enhancing productivity outcomes. To our knowledge, the use of NSAIDs to reduce pain and/or inflammatory response post-breeding has never been done.

Previous research indicates that NSAIDs reduced pre-weaning mortality of piglets and help increase efficiency of postpartum sows. The mechanism of action was studied by giving a dose of meloxicam to pregnant sows and determine for levels of Immunoglobulin G (IgG) subsequently passed from sow to piglet. The researchers studied 30 sows divided

into 2 groups; a control group and a treatment group. The sows were given a dose of meloxicam immediately after the onset of parturition and the 325 piglets subsequently delivered were tracked by weighing birth weight and weaning weight at 21 days after birth. Mortality rates were also recorded for both sows and piglets. In addition, the 4 piglets in each litter were randomly selected to measure the quantity of blood immunoglobulin G in the bloodstream at 1, 2 and 20 days after birth.

The study found that an oral dose of meloxicam given to sows resulted in higher live weight of weaning piglets, significant at P < 0.01 (6.56 and 6.15 kg in treatment group and control group, respectively). Piglet growth rate was also higher (236 and 217 g/day in the treatment and control group, respectively, significant at P < 0.01). However, piglet mortality rate in the treatment and control groups showed no significant difference (6.7 and 6.8 percent in the treatment group and control group, respectively P = 0.89) (Mainau et al., 2016). Immunoglobulin level was higher in treatment than control group on Day 1 (31.9 and 27.9 mg/ml, P < 0.01) and Day 2 (7.6 and 7.1 mg/ml, P = 0.59). Thus, sows that was provided anti-inflammatory



(NSAIDs) soon after parturition increased the volume of immunoglobulin in their piglets and helped boost growth rate in weaning piglets.

In the current study, researchers studied the use of non-steroidal anti-inflammatory drugs (NSAIDs) to increase the effectiveness of artificial insemination in pigs and study conception rate and litter size of pigs after injection of NSAIDs compared with normally inseminated sows. Cooperation is emerging among universities and the industry to develop guidelines to improve performance of artificial insemination in swine, as well as guidelines for research in using NSAIDs in the field. The study also examined patterns, problems, and defects of swine artificial insemination in Thailand and standards for artificial insemination of swine in commercial farms.

The medication selected for this study ('Tolfedine') is a non-steroidal anti-inflammatory drug (NSAIDs) that can reduce inflammation, pain, fever and combat the effects of endotoxins. NSAIDs have a very high level of safety compared to traditional steroid-based treatments such as 'Dexamethasone'. Tolfedine is effective in reducing inflammation because it inhibits prostaglandin synthesis and also competes to bind to prostaglandin receptors. Recent information available from 'The National Committee for Pig Production Annual Report of Denmark in 2005' claims that the concentration of prostaglandins in the bloodstream is elevated during 30-40 min after artificial insemination compared to regular insemination; this may prevent or interfere with sperm transport, thus reducing efficiency of insemination. Thus,

if the effects of prostaglandin can be suppressed for a brief period immediately after insemination, conception rates and litter size using artificial insemination may be improved.

In the study, the researchers investigated the effect of Tolfedine on breeding performance including litter size and farrowing rate, using 2 groups of sows: Group 1 (n = 300) were treated with injections of 'Tolfedine' at 2 mg per kg of body weight (or 1 cc/ 20 kg) about 30 minutes before artificial insemination. All sows in Group 2 (n = 300) were inseminated without injection of 'Tolfedine'. The study was repeated twice, the first during the cool season between November and January, and the second during hot the season from March to June.

The study results contribute to a deeper understanding of the potential utility of NSAIDs such as Tolfedine for swine breeding. This product has not been previously researched in Thailand or optimized for use in artificial insemination; however, pilot experiments have yielded encouraging results, pointing to increased return on investment in development of effective swine artificial insemination process for primary breeding. However, it is too early to recommend broader commercial use and further in-depth research is needed.

#### Reference

Mainau, E., Temple, D., and Manteca, X. 2016. Experimental study on the effect of oral meloxicam administration in sows on pre-weaning mortality and growth and immunoglobulin G transfer to piglets. **Prev. Vet. Med.** 126: 48-53.

# 1<sup>st</sup> Academic conference of Cognitive Behavior Therapy Alliance of Thailand (CAT) - Keep Calm and Learn CBT





excellent networking forum for therapists and service providers to meet and identify opportunities to Speakers presented a wide range of

or CBT.

the latest developments in

psychotherapy techniques in the area of Cognitive Behavior Therapy

The event also served as an

Chula Unisearch together with the Cognitive Behavior Therapy Alliance of Thailand (CAT) held the "1<sup>st</sup> Conference of Cognitive Behavior Therapy Alliance of Thailand (CAT) - Keep Calm and Learn CBT" during 22-24 August, 2016 at the Narai Hotel, Bangkok.

The conference brought together experts and practitioners in the field to exchange knowledge on

collaborate. Speakers presented a wide range of relevant topics including roles and CBT learning points for Thailand, prevention and treatment of depression in children and adolescents, a case study of treatment with CBT and workshops for delegates to assist them to apply the technique in their work and daily lives.

# Stakeholder Consultation for Preparation of (Draft) National Geo-Informatics Master Plan B.E. 2559



The National Geo-Informatics Board, Ministry of Science and Technology, Geo-Informatics (GIS) and Space Technology Development Agency (GISTDA), together with **Chula Unisearch** held a stakeholder consultation to seek opinions, ideas and perspectives as part of preparation of the (Draft) National Geo-Informatics Master Plan B.E. 2559 at Centra by Centara Government Complex Hotel & Convention Centre Chaeng Watthana, Bangkok on Friday 19 August, 2016.

A total of 120 delegates attended the workshop, including representatives and experts from government, together with manufacturers, users, GIS

solution providers and policymakers, IT experts, researchers and representatives from educational institutions with GIS-related research and teaching courses, as well as civil society organizations.

The delegates provided detailed comment and approaches to the application of the National Geo-Informatics Master Plan, which comprises 4 key strategic components as follows: 1) A set of GIS Basic (Fundamental Geographic Data Set, FGDS); 2) A spatial data Clearinghouse; 3) GIS standards; and 4) GIS policy for Thailand.

The 1<sup>st</sup> National Geo-Informatics Master Plan 2016 defines a comprehensive strategy appropriate to developing countries for the period 2017-2021, including GIS infrastructure information which is an essential prerequisite for Thailand to be able to access and use spatial information sharing via the Internet. The Plan also aims to provide a network system for all sectors to trace, browse and download data and generate complete basic geo-information, with accurate and updated details in a versatile, business-friendly format.

# Signing of a Memorandum of Understanding (MOU) for R&D cooperation between Chulalongkorn University and The Siam Cement Public Company Limited (SCG)







On Wednesday 7 September, 2016, Chulalongkorn University President Prof. Bundhit Eua-arporn, Ph.D. and Mr. Roongrote Rangsiyopash, Chairman and Director of the Siam Cement Public Company Limited (SCG) signed an agreement to amend their existing MOU on R&D cooperation. The signing ceremony was witnessed by Prof. Kiat Ruxrungtham, M.D. (Vice President for Research and Innovation), Assoc. Prof. Thavivongse Sriburi, Ph.D. (Managing Director of Chula Unisearch), Supichai Tangjaitrong, Ph.D. (Deputy Managing Director, Chula Unisearch), Mr. Kiti Madilokkovit, HR Director of SCG and Board of SCG Directors.

The MOU was signed as a result of a continuing

project: the "CU Industrial Linkage Partners Program", managed by Chula Unisearch. The project focuses on cooperation in R&D, particularly with innovative industry sectors, to extend networks and stimulate technical cooperation between research and industry. The expanding to the MOU paves the way for further technical cooperation in the social, environmental and cultural arena across ASEAN Member States. This is an essential development that will broadly complement the scope of the original collaboration in research and development that has so far focused on scientific and engineering projects that contribute to innovation and productivity.

# NRCT-IOC/WESTPAC International Meeting on Management and Rehabilitation of Corals after the Bleaching Events





The National Research Council of Thailand (NRCT),
Department of Marine Science, Faculty of Science
(Chulalongkorn University), Department of
Marine and Coastal Resources, UNESCO-IOC/WESTPAC
and Chula Unisearch recently held an International
Conference "NRCT-IOC/WESTPAC International
Meeting on Management and Rehabilitation of
Corals after the Bleaching Events" from
15-17 September, 2016 at S31 Sukhumvit Hotel.

The meeting aimed to provide opportunities for participants to understand new concepts, techniques,



and methods to manage and rehabilitate coral reefs after bleaching, learning from the countries in the Western Pacific. The meeting also triggered the creation of a network to rehabilitate coral reefs, among key institutional actors including government agencies, representatives of the marine resources and researchers working in the countries of the Western Pacific.

## Chulalongkorn University and Inspur Group Co., Ltd., (People's Republic of China, PRC) signed MOU in R&D





On 27 September, 2016, Assist. Prof. Naebboon Hoonchareon, Ph.D., Assistant to the President for Academic Affairs, Chulalongkorn University, and Mr. Peter Huang, General Manager of Inspur Oversea signed an MOU to expand their cooperation with China's Inspur Group Co., Ltd. in R&D in network technology for cloud computing and Big Data. Inspur is a world leading provider of cloud computing solutions. The signing took place at the Guest House of Inspur Group's offices.

The board of directors of **Chula Unisearch**, Assoc. Prof. Thavivongse Sriburi, Ph.D. (Managing Director), Supichai Tangjaitrong, Ph.D. (Deputy Managing Director), Sornnate Areesophonpichet, Ph.D. (Manager of Coordinate

Academic and Industry Collaboration Office) as well as the Administrative board of Chulalongkorn University and Board of Directors of the Inspur Group witnessed the signing ceremony.

On this occasion, administrators and faculty members from Chulalongkorn University traveled to study the educational system at Jinan city, Shandong Province, and Hefei City, Anhui Province. The delegation visited the Smart Education and Education Cloud Service Platform in Hefei No.8 High School and at Shandong University they observed the advances made in the development of speech technology and software development in the Smart Education Platform of the PRC education system at the High Performance Computing Center (HPC) and Data Center of the Inspur Group and iFLYTEK Co., Ltd. The delegation returned to Thailand with many ideas that could be applied to reform Thailand's educational learning process for Thailand's digital future (Thai Education 4.0) as part of Thailand 4.0 strategy.

# Opening ceremony of workshop "Regional Trade Policy Course for Asia and Pacific 2016 (RTPC 2016)": Cooperation between WTO, ITD and Chulalongkorn University



The World Trade Organization (WTO), International Institute for Trade and Development (ITD) and Chulalongkorn University, represented by the Office of International Affairs and Chula Unisearch recently convened a joint international workshop: "Regional Trade Policy Course for Asia and Pacific 2016 (RTPC 2016)."

The event was honored by the presence of Assist. Prof. Pomthong Malakul Na Ayudhaya, Ph.D., Vice President for Academic Affairs, Assist. Dr. Kamalinne

Pinitpuvadol, Director and Secretary of the International Institute for Trade and Development (ITD) and Dr. Luanga Mukela Faustin, Head, Asia and Pacific Regional Desk, Institute for Training and Technical Cooperation, WTO, who presided over the opening ceremony, held at the Chaloem Rajakumari 60 Building, Chulalongkorn University. On this occasion, Assoc. Prof. Suchana Chavanich, Ph.D., Deputy Managing Director, Chula Unisearch also attended the opening ceremony.

The workshop took place from 3 October to 25 November, 2016, and was intended to allow a total of 25 primary level officials in the field of trade and commerce from 17 countries in the Asia-Pacific region to exchange knowledge and experiences relating to trade policies in order to enhance trade policies in each country.

### Special Ceremony to Commemorate King Rama VI Memorial Day







On Friday 25 November, 2016, representatives from the Executive Board of **Chula Unisearch**, Assist. Prof. Saowanee Wijitkosum, Ph.D., and Assoc. Prof. Suchana Chavanich, Ph.D., Deputy Managing Director of Chula Unisearch, joined a special ceremony to pay homage on King Rama VI Memorial Day by laying garlands at the King Rama V and King Rama VI statues in front of the Chulalongkorn University Auditorium and the Statue of King Rama VI in Lumpini Park, Bangkok.

#### Chula Unisearch staff volunteer to make merit for the Great Father













Assoc. Prof. Thavivongse Sriburi, Ph.D., Managing Director of Chula Unisearch, together with staff members of **Chula Unisearch**, made merit by volunteering for His Majesty the Late King Bhumibol Adulyadej, together with the Bhumipalung Phandin club (BPP club) that is dedicated to promoting the virtue of His Majesty King Rama IX.

The volunteers gave food, drinking water and snacks to people who come to pay homage to His Majesty the Late King Bhumibol Adulyadej at Sanam Luang, Bangkok, from 1 November, 2016 onwards.

