

Special Report on NPUST Research & Innovation



Subject of Research & Innovation

- NPUST CENTER FOR WATER RESOURCES EDUCATIONS AND STUDIES
Cheh-Shyh Ting 1
- EMERGING COMPOUNDS RESEARCH CENTER (ECOREC).....Kuei-Jyum Yeh, Ting-Chien Chen, How-Ran Chao 2
- PIGLET FEED INTAKE IMPROVEMENT
Liang Chou Hsia 5
- CpG DNAADJUVANT IN AVIAN VACCINES
Hso-Chi Chaung, Li-Hsiang Hung, I-Yang Lien 6
- NPUST OPENS AGRICULTURE PROGRAM FOR GAMBIAN STUDENTS
Office of International Affairs 7
- MID-AUTUMN FESTIVAL & WELCOME PARTY
Office of International Affairs 7

NPUST CENTER FOR WATER RESOURCES EDUCATIONS AND STUDIES

1.Introduction -The Development of Water Resources

The center is located in Southern Taiwan where there is abundant natural resources as well as ample hydraulic engineering resources. These superior conditions can be effectively harnessed by our center for field instruction and research. Thus, multi-developed water resources at the center have created many hydraulic engineering accomplishments for agriculture, livelihood, and industry such as reservoirs, hydropower demonstration area, and an infiltration gallery. The primary freshwater source is used for agriculture, industry, and by households. With the vision of integrating regional resources, the Center for Water Resources Education and Studies was established in 2005.

2. Teaching Place

- (1).Reservoir
- (2).Canal
- (3).Artificial Recharge Area
- (4).Construction Wetland

3. Cooperative Organization

Taiwan Power Company , Irrigation Association, Water Resources Bureau, Taiwan Sugar Corporation, and CPC Corporation, Taiwan

4. Establishment Methods

1.NPUST is determined to fully establish the Center for Water Resources Education and Studies. It combines all of the resources for strengthening research and education.

2.Educational work is detailed as follows:

- (1).Archive textbooks on water resources education
- (2).Organize conferences on water resources research
- (3).Undertake water resources education projects
- (4). Administration works

3.Technical research comprises the following:

- (1).Water resource research and development, including patent applications

(2).Project applications and implementation

5.Organizational framework

Administration Management Group

The administration secretary and the financial management officer are responsible for routine activities, salary concerns, and employee welfare.

Wetland Ecology Group

The Wetland Ecology Group mainly investigates the eco-hydrogeology and hydrological parameters toward the comprehension of wetland structure and function.



Cheh-Shyh Ting (second left) with Pingtung County Magistrate (second right) visiting Er-Fong Gallery.

This research group focuses on hydrology and ecology. Specifically, these are the areas of concern: water quality, soil, zoology and botany, and the integration of related projects.

Water Resources Group

This group focuses on the Pingtung Plain, with the following areas of study: artificial recharge of groundwater, optimization for groundwater resources pattern, investigation of groundwater and water quality, and pumping test. It focuses not only on the provision of numerical simulation capability, and also implements simulation projects, plan analysis, and pilot test analysis.

Water Resources Education Group

Ting, Cheh-Shyh, Department of Civil Engineering

This group implements water resources education, which includes groundwater and eco-hydrogeology courses. In addition, it collaborates with the Community University, colleges, and other institutes for the conduct of educational lectures. In the future, it plans to join the ecological protection and environmental consciousness advocacy. This is a crucial idea that it intends to develop in the field of water resources.

Information System Group

This group is responsible for the construction and enhancement of the central system and team work, as well as the development of the Geographic Information System (GIS) in Pingtung Plain.

6.TECHNICAL SERVICES

Investigations

1. Pumping test and analysis
2. Soil permeability pilot test
3. Hydrogeology investigation
4. Groundwater monitoring
5. River current observation
6. Meteorological observation
7. River circumstance investigation

Pilot Test and Numerical Analysis

- 1.Experiment on artificial recharge of groundwater
 - 2.Investigation on soil and chemical character
 - 3.Algae and water quality
 - 4.Hydrology groundwater monitoring
 - 5.Numerical analysis
- Equipment
- 1.Water level automatic recorder
 - 2.Manual water level recorder
 - 3.Supersonic wave overflow flow meter
 - 4.Clamp type flow meter
 - 5.Microclimate station
 - 6Automatic soil sieves

EMERGING COMPOUNDS RESEARCH CENTER (ECOREC)

1. Introduction

In the last two decades research has found that environmental chemicals can interrupt the endocrine system and generate reproductive abnormalities in wildlife, such as vitellogenin induction, feminized reproductive organs, reduced fecundity, changes in sex ratio, and developmental degeneration. These compounds are called environmental hormones, which are part of the emerging compounds.

Within the field of endocrine disruption, estrogenic effects are currently the greatest concern. A number of endocrine disrupting chemicals (EDCs) found in aquatic environments has the potential to interfere with estrogenic functioning. Of greatest concern are the natural estrogenic steroid hormones: 17 β -estradiol (E2), estrone (E1), and estriol (E3) and the synthetic estrogenic steroid hormones 17 α -ethynylestradiol (EE2) and diethylstilbestrol (DES). These chemicals can induce vitellogenin at very low concentrations, such as 1 ng/L of E2, which leads to vitellogenin induction in male trout. The formation of ova in the testis of Japanese medaka starts at a concentration of 4 ng/L for E2 and 0.1 ng/L for EE2.

Most estrogenic chemical studies in aquatic environments have focused on the influent and effluent of sewage treatment plants, the discharge of industrial wastewater and the receiving water in urban areas. For example, in a US national survey of 139 polluted streams and rivers between 1999 and 2001, estrogens were detected in the range of <5 to 112 ng/L for E1, <5 to 93 ng/L for E2, <5 to 51 ng/L for E3 and <5 to 831 ng/L for EE2. The detection frequencies were 7.1, 10, 21.4, and 15.7%, for E1, E2, E3, and EE2, respectively. The survey of three Chinese rivers in the Tianjin area found



Kuei-Jyum Yeh, Ting-Chien Chen, and How-Ran Chao
Department of Environmental Science and Engineering

estrogen concentrations ranging from 0.64 to 55.3 ng/L for E1, from not detected (ND) to 32.4 ng/L for E2, from ND to 46.4 ng/L for E3, from ND to 35.6 ng/L for EE2, and from ND to 8.51 ng/L for DES. In seven French rivers, estrogens were detected at mean concentrations in the range of 1.1 to 3.0 ng/L for E1, 1.4 to 3.0 ng/L for E2, 1.0 to 2.5 ng/L for E3, and 1.1 to 2.9 ng/L for EE2.

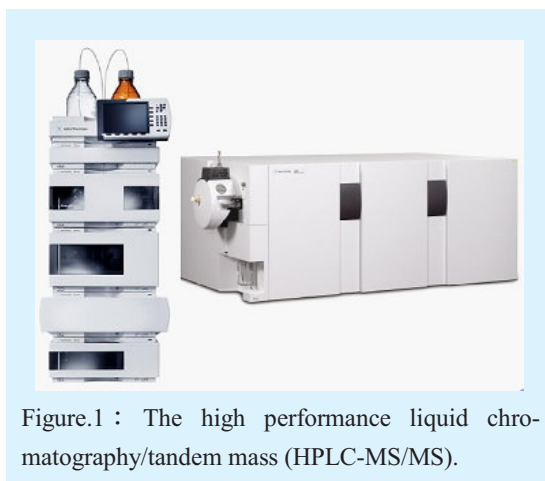


Figure.1 : The high performance liquid chromatography/tandem mass (HPLC-MS/MS).

Comparatively, the data concerned with estrogenic chemicals in receiving water affected by concentrated animal feedlot operations has been limited. It has been shown that estrogen concentrations have been much higher in water from feedlot animal operations, such as in dairy farm effluent concentrations ranging from ND to 3,123 ng/L for E1, and from ND to 331 ng/L for E2. In a swine lagoon, the maximum concentrations of E1, E2, and E3 were 24,900, 3,000, and 10,420 ng/L, respectively. In poultry waste, concentrations ranged from 14 to 533 ng/g dry weight for estrogens and in sow feces, concentrations of estrone ranged from 15 to 28 ng/g. Another major source of hormone steroids has been livestock waste, which entered waterways

through manure runoff.

These discharges need to be seriously investigated to inhibit the concentrated estrogenic chemicals generating the high estrogenic activity in receiving waters.

2. The Goals of EcoRec

The Emerging Compounds Research Center (EcoRec) was established in October 2008 and is supported by the National Pingtung University of Science and Technology. The EcoRec's main goals are to investigate the emerging compounds discharged by farm animals and determine the distribution and fate of the estrogens, androgen, and antibiotics in the aquatic environment. The EcoRec's final goal is to develop an effective technology to remove the emerging compounds from the environment. The EcoRec will assay the ecological and human risk caused by the emerging compounds in each environmental partition.

There are many kinds of emerging compounds and the standard methods for emerging compounds analyses are limited. The trace amount of chemicals and corresponding bio-responses can be detected with advanced analytical instruments and bio-technology but analytical precision is affected by clean up procedures, environmental matrix, instrumental precision, and chemical properties. In order to measure these trace chemicals in the environment, the EcoRec has purchased a high performance liquid chromatography/tandem mass spectrometry (HPLC-MS/MS), which is shown in Figure 1.

The EcoRec will also use in-vitro bio-assay because instrumental analyses Apollo Technology Co., Taiwan to vari-

can only detect compound concentrations not the toxicity to the environment, the coordination and inhabitation effects. Bio-assays can estimate the synthesized toxic effects in environments. In-vivo bioassays require long wait-times, are expensive, and use animals and the application is limited in environmental tests. Comparatively, in-vitro bioassays have the advantages of needing short wait-times, inexpensive, and are very sensitive. Although the in-vitro bio-assay is unable to determine the secretion response and metabolism in organisms, it is still a good way to screen for environmental sample bio-effects.

The ultimate goal in understanding the pollution is to eliminate the pollutants in the environment. The EcoRec, in cooperation with the University of Utah (Salt Lake City, Utah), has used a new technology, heightened ozonation treatment (HOT) as shown in Figure 2, to eliminate pollutants. The HOT is a fully new chemical oxidation process invented by the U-



Figure.2 : The heightened ozonation treatment instrument (HOT).

niversity of Utah. The HOT process includes continual enhancement and reduced pressure recycling. The process enhances the contact opportunity between pollutants and chemicals such as ozone to enable rapid pollutant destruction. The HOT working mechanism is shown in Figure 3. With future HOT applications in mind, the EcoRec has worked with the University of Utah and Apollo Technology Co., Taiwan to degrade emerging compounds in varied environmental me-

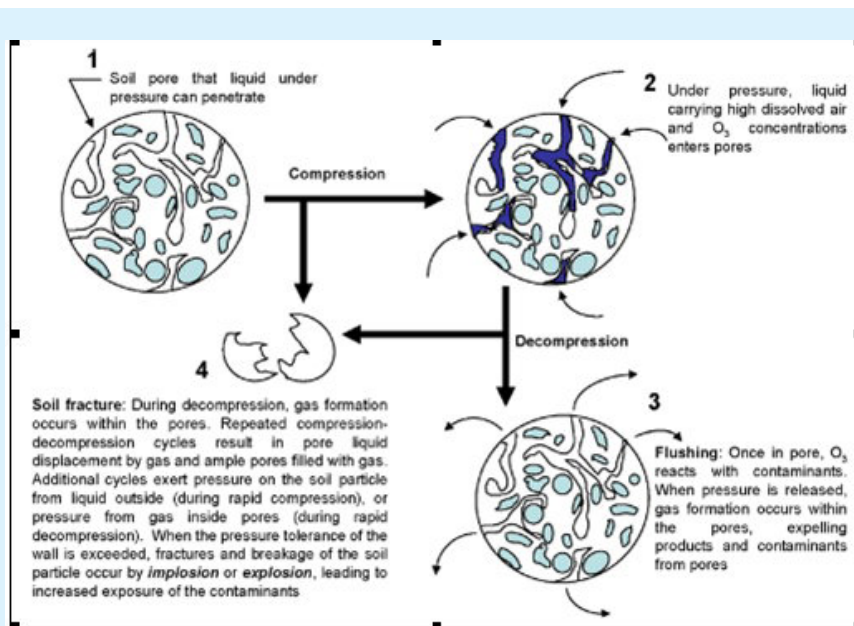


Figure.3 : Mechanisms of increased contaminant exposure to aqueous O₃ during pressure-assisted ozonation cycles.

dia.

3. The Research Results

After a year of vigorous work, the EcoRec has achieved the following results.

(1) EcoRec has established five estrogen analytical methods in aqueous phase and monitored the five estrogen concentrations in Wulo Creek (Pingtung County, Taiwan), which has a concentrated livestock feedlot upstream. The analytical methods developed in the laboratory concern the natural estrogenic steroid hormones E2, E1, and E3 and the synthetic estrogenic steroid hormones EE2, and DES. The monitored WuLo Creek flow basin has approximately 500,000 pigs and more than eight million poultry.

Samples were collected from nine sampling sites, in which eight sampling sites were along Wulo Creek. The other site was located in the Kaoping River and was used for comparison. The results showed that EE2 concentrations were less than the limit of detection (LOD) while DES concentrations were only detected twice. Concentrations ranged from 7.4 to 1267 ng/L for E1, from ND to 313.6 ng/L

for E2, and from ND to 210 ng/L for E3. E1 had the highest average mass fraction ($72.2 \pm 3.6\%$) which was much greater than E3 ($16.2 \pm 1.7\%$) and E2 ($11.5 \pm 2.6\%$). The mean E2 equivalent quotient (EEQ) in each site ranged from 17.3 to 137.9 ng-E2/L, which was much higher than the proposed non observed effect concentration of 10 ng-E2/L. E2 had a much lower concentration than E1, but E2 ($52.4 \pm 6.0\%$) had a higher contribution to the EEQ than E1 ($19.7 \pm 3.5\%$). The concentrations of three estrogens upstream were much higher than concentrations downstream. This suggested both a high attenuation effect and a fast degradation option in the study river. During the winter season concentrations were greater than in the spring season due to a low dilution effect and low microbial activity in the winter season. The results of the natural estrogens' high concentrations suggest that the wastewater discharge from the feedlot animals needs to be treated further.

(2) The application of HOT technology
The EcoRec has applied the HOT technology with the University of Utah and

ous conditions. The technology has been assessed, verified, and parameters adjusted for application in Taiwan's soil and sediment. Recently, the target chemical was Polychlorinated Biphenyl (PCB) in sediment of Erh-Jen Creek. The parameters for degrading pollutants include pressure, cycling time, O₃/air ratio, and pollutant concentrations.

(3) In-vitro bioassay system application
The CALUX bioassay is one of the in-vitro assays using dioxin toxicity in gene expression to detect dioxins and PCBs-TEQ levels. Firstly, a plasmid with specific DNA sequences called dioxin responsive elements (DREs) needs to be recombined with reporter genes from luciferase genes of fireflies. This recombinant plasmid is transfected into the human hepatoma cell. The action of dioxins/PCBs mechanism is that dioxins/PCBs bind to an intracellular receptor called the aryl hydrocarbon receptor (Ah Receptor) and activate the receptor. The dioxin/PCBs-Ah Receptor complex then travels to the cell nucleus and binds to specific DNA sequences dioxin responsive elements (DREs). The binding of the dioxins/PCBs-Ah Receptor

complex to the DREs causes the expression of the associated genes to be altered. It is the alteration in gene expression causing the toxic effects. If the DREs in the chromosome are activated by dioxins/PCBs-Ah Receptor complex, this complex also activates the DREs with luciferase genes in the recombinant plasmid to create the light.

We also established a dioxin-responsive element (DRE)-mediated Chemical Activated Luciferase eXpression (CALUX) cell line, Huh7-DRE-Luc, using stable transfection of Huh7 with a DRE-driven firefly luciferase reporter plasmid (4xDRE-TATA-Luc). Huh7-DRE-Luc and Huh7 treatments with NaAsO₂ attenuated the 2, 3, 7, 8-TCDD-induced DRE-CALUX and cytochrome P450 1A1 (CYP1A1) activations, respectively, in a dose-dependent manner. The calculated CALUX-toxic equivalent (TEQ) levels induced by cotreatment of NaAsO₂ 3.0 μM and 10 nM 2, 3, 7, 8-TCDD were significantly lower than that induced by 2, 3, 7, 8-TCDD alone (p<0.05). It was demonstrated that arsenic inhibited the TCDD-induced CYP1A1 activation and

also interfered with DRE-CALUX bioassay in human hepatoma cells. Our finding also suggests that extensive cleanup of samples for removal of any possible interfering factors is critical to guarantee the accuracy of dioxin-TEQ levels using DRE-CALUX bioassay. We used these DRE-CALUX cells to analyze the dioxin-CALUX-TEQ levels in several matrices, such as human placentas, breast milk, fly ashes, soil, and so on. Future work will be focused on establishment of standard analytical protocol for dioxins bioassay. The mechanisms and experiment equipments of DR-CALUX in vitro system is shown in Figure 4.

4. Future Work of the EcoRec

- (1) Extend the LC/MS/MS analytical method applied in various environmental media.
- (2) Develop LC/MS/MS analytical methods for other emerging compounds.
- (3) Develop in-vitro bio-assay detection methods.
- (4) Develop further HOT technology applications.

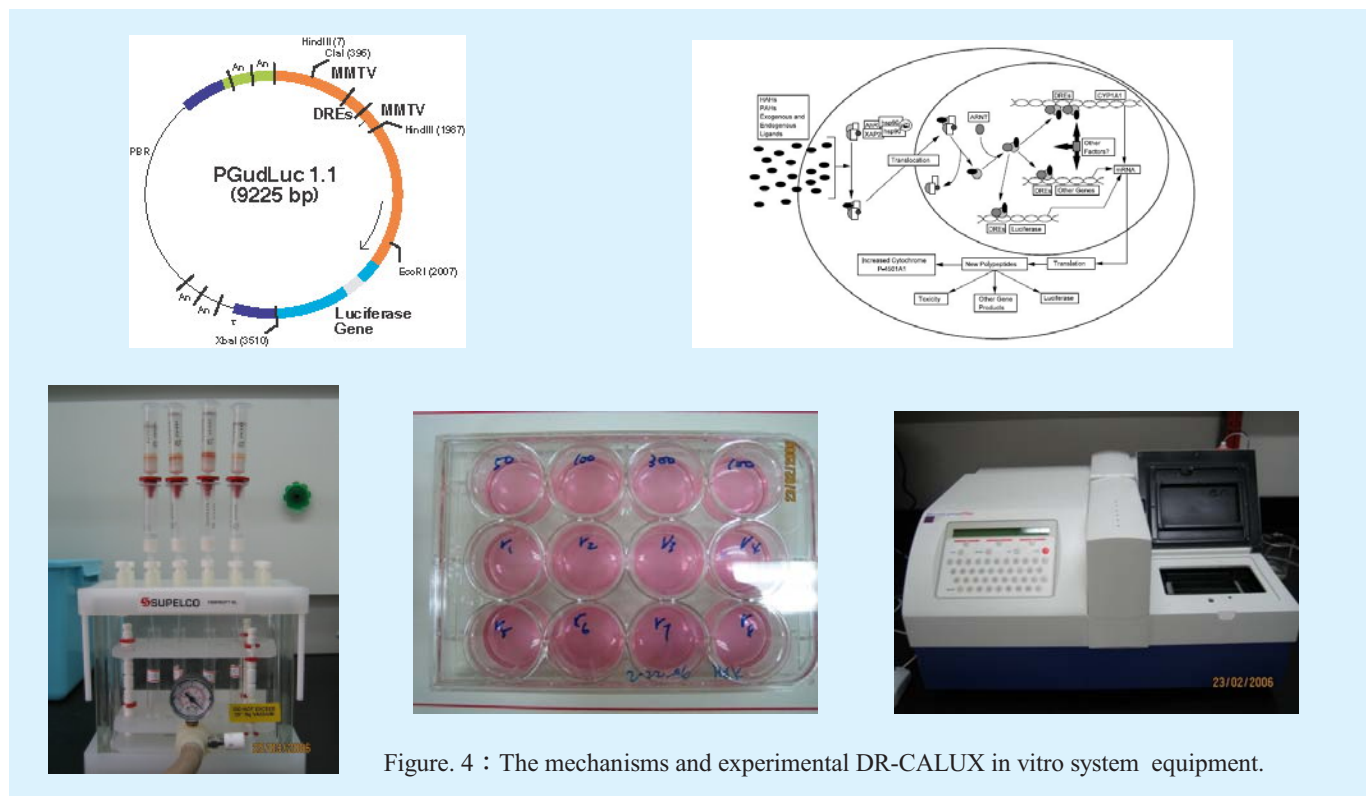


Figure. 4 : The mechanisms and experimental DR-CALUX in vitro system equipment.



1. Introduction

This paper presents a method to improve piglet feed intake. Theoretically, piglets should grow faster than today's piglet growth. The lower piglet growth causes the following three big losses: (1) economic loss due to slow growth; (2) lower piglet weight at weaning; the lower the weight gain at the growing-finishing stage, the greater the number of days to market; (3) higher mortality rate during the piglet stage. Consequently, improving piglet feed intake is a very important issue.

2. Birth weight

Heavier birth weight piglets always have higher vigorous milk suckling. The result of this higher milk intake can cause higher weight gain. The higher weight causes piglets to not get enough nutrition from milk for their maintenance and growth. Consequently, they must eat more creep feed. Once they eat more creep feed, they will become used to feed after immediately weaning. Producing piglets with heavier birth weight is the essential point. If we provide sows with more feed or a higher energy diet during the last 2-3 weeks, the sows can produce about 1.4-1.5 kg additional weight in new born piglets. There are three points we have to notice. Firstly, we should not provide too much feed for gilts. Secondly, it is important not to provide higher feed for sows for more than 3 weeks before farrowing. Thirdly, if we provide a higher energy diet, which is produced by adding a higher percent of fat or oil, we should add Vit E in the diet.

3. Spray dried plasma protein (SDPP)

Adding 5% of SDPP in the piglet diet can stimulate the piglet to eat more feed. However, the result can last about 2 weeks. This may be due to lower

isoleucine content in the SDPP.

I suggest it might be added in the creep diet and provided to weaning piglets for another 2 weeks.

4. Tryptophane

Our experimental results showed that adding enough tryptophane to the piglet diet can improve feed intake. This result may be due to tryptophane transferring to melatonin and melatonin plays an important role in feed intake.

5. Amino acid balance feed

Amino acid unbalance feed usually causes lower feed intake.

6. Sucrose

Our experimental results showed that adding sucrose in the piglet diet can improve piglet feed intake. However the minimum added percentage should be 10%. This is a very high percentage. The sugar cost is quite high. Glucose cannot replace sucrose due to its lower sweet taste.

7. Acidified feed

Slightly acid feed causes higher saliva secretion. Higher saliva secretion usually can induce piglets to eat more.

8. Vit B complex

Some members of the Vit B complex can induce higher feed intake in pigs. If we can add a proper amount of Vit B complex, it could improve pig feed intake.

9. Fresh feed

Piglets are very sensitive to bad smelling feed. They usually refuse to eat feed left in the trough for a few hours. This is the reason why we it is better to feed piglets 3-4 times during the day.

10. Conclusion

There are many methods to improve feed intake. However, the previous discussed methods are quite important.



Liang Chou Hsia,
Department of Animal Science

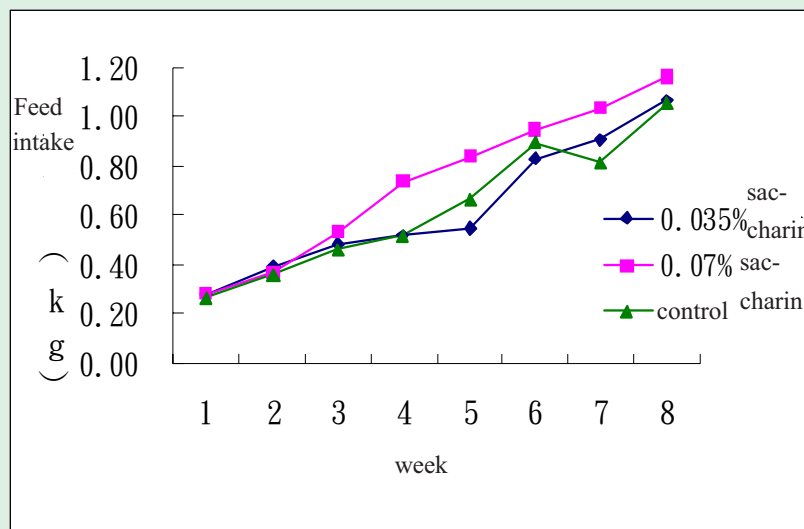


Figure.1 : Saccharin feed supplementation effect on the feed intake of weaning piglets.

Table 1 Effect of different proteins or higher tryptophane feed supplementation on piglet performance of piglets

Week	control	0.22% tryptophane	SDPP	milk pure powder	Sig.	SEM	(Unit: g)
Feed intake							
1 ~ 4	333	346	326	313	NS	15.7	
5 ~ 8	809 ^b	877 ^b	954 ^a	788 ^b	*	35.8	
1 ~ 8	571	612	640	550	NS	24.5	
Weight gain							
1 ~ 4	185	195	183	164	NS	14.6	
5 ~ 8	448 ^b	491 ^{ab}	516 ^a	440 ^b	*	17.3	
1 ~ 8	317	343	349	302	NS	13.5	
Feed efficiency							
1 ~ 4	2.02	1.93	1.86	2.07	NS	0.23	
5 ~ 8	1.81	1.78	1.84	1.79	NS	0.70	
1 ~ 8	1.92	1.86	1.85	1.93	NS	0.11	

Notes :

1. a,b significant differences within the same row while not bearing the same superscripts (P < 0.05).

2. NS: P > 0.05; *: P < 0.05 ◦

CpG DNA ADJUVANT IN AVIAN VACCINES

ABSTRACT

A CpG DNA adjuvant in avian vaccines is disclosed, which includes an immuno-stimulatory oligodeoxynucleotide (ODN) having a plurality of TCG tandem repeats at a 5' end, a poly-G structure at a 3' end, and at least one unmethylated CpG motif with avian specific flanking sequences at two ends thereof between the 5' end and the 3' end. The CpG DNA adjuvant in avian vaccines is advantageous for large-scale production; specifically enhanced avian innate and adaptive immune responses, and the CpG DNA adjuvant is difficult to be digested by DNase due to its particular structures.

The Invention

Accordingly, an aspect of the present invention provides a CpG DNA adjuvant in avian vaccines, which includes an immuno-stimulatory oligodeoxynucleotide (ODN) having a plurality of TCG tandem repeats at a 5' end, a poly-G structure at a 3' end, and at least one CpG motif with avian specific flanking sequences at two ends thereof between the 5' end and the 3'

end. The CpG DNA adjuvant in avian vaccines is advantageous for carrying out large-scale production, specifically to enhance avian innate and adaptive immune responses, and to facilitate cellular uptake of the CpG DNA adjuvant.

According to the aforementioned aspect of the present invention, a CpG DNA adjuvant in avian vaccines is provided, which includes an immuno-stimulatory ODN having a plurality of TCG tandem repeats at a 5' end, a poly-G structure essentially consisted of 4 to 6 guanines at a 3' end, and at least one unmethylated CpG motif with avian specific flanking sequences at two ends thereof between the 5' end and the 3' end thereof.

The CpG DNA adjuvant in avian vaccines is advantageous for carrying out large-scale production, specifically to enhance avian innate and adaptive immune

Hso-Chi Chung, Li-Hsiang Hung, I-Yang Lien
Department of Veterinary Medicine

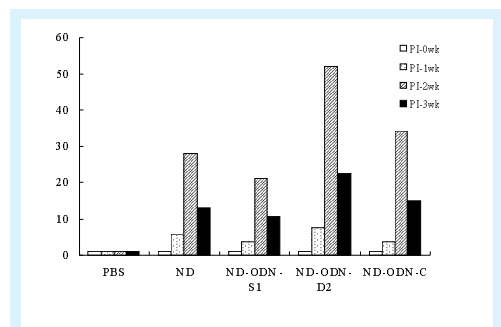


Fig. 1 : HI titer of chickens co-immunized with ND commercialized vaccine and the CpG DNA adjuvant according to a preferred embodiment of the present invention.

responses and facilitate CpG DNA adjuvant cellular uptake instead of the prior DNA adjuvant modified using complicated chemical processes.

Field of the Invention

This invention relates generally to a vaccine adjuvant and more particularly, to a CpG DNA adjuvant in avian vaccines.



✿ NPUST opens agriculture program for Gambian students ✿

National Pingtung University of Science and Technology (NPUST) set a course for students from Gambia to learn agriculture related knowledge and skills, which began on Monday Sep. 14.

The course is being attended by 25 students sent by the Gambian government. The students were divided into four groups that will study Plant Production, Animal Production, Agribusiness Management, and Agricultural Engineering and Farm Mechanization.

A ceremony was held that same day to mark the opening of classes, which was attended by the Gambian Office of the President Secretary-General Abdoulie Sallah and Gambian Ambassador to the Republic of China Mawdo C. Juwara. Also present were Chen Shih-liang, director-general in charge of African affairs at the Ministry of Foreign Affairs, along with Ministry of Education officials.



The Gambia delegation gave gifts to Dr. Mike Guu, President of NPUST.

✿ Mid-Autumn Festival & Welcome Party ✿

The moon festival celebration held on Oct. 2 at Department of Tropical Agriculture and International Cooperation. A group of senior students from Department of Modern Languages introduce the origin of Mid-Autumn Festival by short drama, Ten Suns. After foreign students understood the traditional custom on Mid-Autumn Festival in Taiwan, they had fun at the game time. The president of National Pingtung University of Science and Technology, Dr. Guu gave the opening remarks for these students who are far from their hometown and got together on Mid-Autumn Festival's eve. Finally, everybody enjoyed the moon cakes, pomelos and drinks.



International students competing in peeling pomelos.

Publisher : Yuan-Kuang Guu

Editor-in-chief : Chiwan-Wayne Hsieh

Deputy Editor-in-chief : Wu-Jang Huang, Jia-Hsin Guo

Advisory Board : Chang-Hsien Tai, Chaur-Tzuhn Chen,
Chung-Ruey Yen, Bor-Tsuen Wang, Earl-Juei Wang,
Chao-Chin Liu,

Editors : Steve Huang, Ching-Lin Hu, Yih-Wea Liu,

Huei-ling Lee

Address : No.1, Shueh Fu Road, Neipu, PingTung 912,
TAIWAN

Website : <http://www.npust.edu.tw>

Phone : 08-7703202

GPN : 2009802044