BIOLOGICAL SCIENCES

BS - 01

IMMUNOSTIMULATORY EFFECTS OF *Uncaria perrottetii* (A. RICH) MERR. (RUBIACEAE) VINEBARK EXTRACT *IN VIVO*

Leonora P. Nudo[1*] and Elena S. Catap[1,2]

[1]Natural Sciences Research Institute and [2]Institute of Biology,
College of Science, University of the Philippines
Diliman, Quezon City lenpnudo@gmail.com

The immunostimulatory effects of *Uncaria perrottetii* (A. Rich) Merr. vinebark aqueous extract was evaluated by employing a number of immune response assays *in vivo*. Initially, the optimum concentration was determined *in vitro* to be 50µg/mL. This concentration was then assessed for the immunoprotective effects of the extract against immunosuppressed Balb/C mice.

Thirty-six (36) mice were divided into 3 groups: (1) Phosphate-buffered saline (PBS)-injected mice; (2) *U. perrottetii* extract and Cy-treated mice (*U+Cy*); and (3) cyclophosphamide (*Cy*)-induced immunosuppressed mice. Group 1 was injected intraperitoneally with 0.2mL of 30mg/kg body weight of cyclophosphamide (Endoxan®) at days 1, 4, 7 and 10 of the treatment period. Cyclophosphamide was also administered to group 2 one hour after each administration of the plant extract. 50mL/kg body weight of PBS and *U. perrottetti* extract (2.5 mg/kg body weight) was administered daily.

In all immune assays undertaken, group 3 showed significantly lower response when compared with group 1. Group 2 showed significant improvement in phagocytic activity (27%), proliferation of cells with (29.61%) and without (44.57%) lipopolysaccharide, superoxide production ($P<0.05$), and plasma lysozyme activity ($P<0.05$) compared with the group 3.

The study showed that the extract could potentially reverse the immunosuppressive effects of cyclophosphamide *in vivo*. There is, therefore, a great potential of the plant as a source of bioactive products and metabolites for drug development.

Keywords: *Uncaria perrottetii,* Balb/C mice, immunostimulation, Rubiaceae, murine macrophages

BS - 02

BACTERIAL PATHOGEN ISOLATED FROM WHITE SHRIMPS *Pennaeus vannamei* CULTURED IN ILOCOS NORTE

Alice Geraldine Hernando*and Prima Fe R. Franco

Mariano Marcos State University, College of Arts and Sciences
Department of Biology, Batac, Ilocos Norte

There is a need to have an alert surveillance on the microorganisms afflicting cultured shrimps to be able to curtail the spread of disease that could wipe out shrimp business ventures. This study focused on characterization and identification of a bacterial pathogen isolated from white shrimps cultured in Ilocos Norte.

White shrimps were surface sterilized for one minute prior to dissection. Swabs were obtained aseptically from the head parts after removal of the carapace and were inoculated into Nutrient broth cultured overnight. Colonies were grown in Nutrient Agar and pure cultures were further morphologically and biochemically characterized. The pure cultures were tested for hemolysin activity and the one that turned positive in the hemolysin test was identified using the API System of bacterial identification.

Results show that the isolate is a Gram negative non-fermenting aerobic rod. API identification system show that the isolate is Indole negative, nitrate reductase positive, Methyl red positive, Voges_Proskauer negative, catalase, positive, cytochrome oxidase negative, phenyl alanine deaminase positive, Tryptophan test negative, Urea positive, Casein negative starch negative, hydrogen sulfide positive and citrate agar positive. Over all characteristics reveal that the isolate is a *Proteus mirabilis*.

Results call for the need to collaborate with shrimp industry owners to become aware of the possible sources of contamination of grow-out ponds for the protection of their business as well as the consumers.

Keywords: *Proteus*,white shrimp, API, shrimp industry, biochemical characterization

BS - 03

INDUSTRIALLY IMPORTANT BACTERIAL ISOLATES FROM SWEET SORGHUM *Sorghum bicolor* L MOENCH

Prima Fe R. Franco*, Cecile A. Gaoat and Rowena Acosta
Remelyn Soliman, Samuel S. Franco and Heraldo Layaoen

Mariano Marcos State University, College of Arts and Sciences
Department of Biology, Molecular Microbiology and Biotechnology
Laboratory, City of Batac, Ilocos Norte primarfranco@gmail.com

In an effort to search for industrially important enzyme producing bacteria, specially needed for biofuels production, sweet sorghum was used as the inoculum. Three varieties of sweet sorghum namely SPV 422, NTJ2 and ICSV93046 were used in this study. Sweet sorghum flour were inoculated in Nutrient Broth and incubated overnight. Pure cultures were obtained and grown in Nutrient Agar. Colonies were morphologically and biochemically characterized. They were further screened for production of enzymes needed in the industry. Three isolates are protease producers, one is a lipase producer and one has the ability to hydrolyze starch All the isolates are Gram positive coccus except the starch hydrolysis positive which is Gram negative coccus. They can tolerate up to a maximum of 36% to 39 sugar concentration%, an indication that they are osmophiles. All the isolates are present in all of the sweet sorghum varieties. They are at present molecularly characterized to further elucidate their taxonomical positions. These are potential isolates for biotechnological production of industrially important enzymes.

Keywords: sweet sorghum, bacteria, industry, enzymes, lipase, sugar hydrolysis, protease, lipase

BS - 04

CHARACTERIZATION OF PIGMENT PRODUCING BACTERIA FOR INDUSTRIAL PURPOSES

Prima Fe R. Franco*, Cecile A. Gaoat, Rowena Acosta,
Samuel S. Franco and Heraldo Layaoen

Mariano Marcos State University, College of Arts and Sciences
Department of Biology, Molecular Microbiology and Biotechnology
Laboratory, City of Batac, Ilocos Norte primarfranco@gmail.com

This study focused on the isolation and characterization of pigment producing bacteria from various sources like larvae of insects affecting sweet sorghum, fish paste and cultured shrimps in Ilocos Norte.

Insect larvae affecting sweet sorghum include earworm, army worm and katydid. The larvae were first surface sterilized prior to dissection. Inocula were obtained aseptically from their guts and are cultured in Nutrient broth overnight. Pigmented isolates were screened and grown in pure cultures in Nutrient agar and characterized morphologically and biochemically. Isolates produce yellow pigments Fish paste was inoculated into Sea Water Complex medium and cultures were grown overnight Pigmented isolates were plated and screened in Nutrient agar supplemented with 15% NaCl The isolates from fish paste are slightly yellow. They were tested for salt tolerance and results show that they could could tolerate up to 20% salt concentration, an indication that they are pigmented halophiles.

Inocula from the head parts of shrimps *Pennaeus vanamae* were obtained and cultured in Nurient Broth. Pure cultures were grown in Nutrient agar and were screened for pigment producers. The colonies obtained are blue green.

Most of the isolates are Gram positive coccus. Pigment production ranged from the 3rd to the 6th day after inoculation and is affected by the pH of the medium but not by temperature. The blue green, yellow and slightly yellow pigments are released into the medium and therefore extracellularly secreted.

The isolates are currently characterized at the molecular level. They are potential sources of dye in the food, fabric and leather industries.

Keywords: pigment, pigment producing bacteria, industry, isolation, biochemical characterization, Biotechnology

BS - 05

PROTECTIVE EFFECTS OF FORMALIN- AND HEAT-KILLED *Aeromonas hydrophila* IN NILE TILAPIA (*Oreochromis niloticus*) INFECTION

Stephanie S. Pimentel*, Phillip Sebastian Serafin J. Golez and Eufemio Jesus R. Roldan

Biology Department, School of Science and Engineering, Ateneo de Manila
University, Loyola heights, Quezon City 1108
spimentel@ateneo.edu

Aeromonas hydrophila is an opportunistic bacterial pathogen of a number of aquatic animals that causes hemorrhagic septicemias, erythrodermatitis, fin and tail rot in fish. The pathogen affects *Oreochromis niloticus* (Nile tilapia), an economically important cultured fish in the Philippines. In this study, the protective effects of formalin- or autoclave-killed *A. hydrophila* or bacterin where injected via intraperitoneal on *O. niloticus* then challenged with live *A. hydrophila.* Data on mortality rates, serum lysozyme levels and histopathology of immune related organs, head kidney and spleen, generated responses in the challenged fish. Though fish were not entirely protected from the live form, autoclaved-killed bacteria showed stronger immune responses compared to formalin-killed bacteria.

Keywords: formalin-and autoclave-killed *A. hydrophila,* histopathology, tilapia infection

BS - 06

ESTABLISHING THE LEAF ANATOMY OF *Tamarindus indica* LINN. (TAMARIND) AND ITS IMPLICATION TO HARSH ENVIRONMENTS

Vivian S. Tolentino and Cara Paulina G. Chuidian

Department of Biology, Ateneo de Manila University, Loyola Schools,
Katipunan Road, Loyola Heights, Quezon City vtolentino2001@yahoo.com

Tamarind (*Tamarindus indica* Linn.) has not received sufficient research attention over the years though it is one of the minor fruit crops in the Philippines, with a great potential for commercialization. It is an important crop because of its varied food and medicinal uses. The young green leaves are used for tasty dishes. Crushed leaves are put on wounds and abscesses. Juice from crushed leaves is taken with porridge to stop vomiting. Several studies are reported on its taxonomy, description and distribution; properties of the species; uses and products; ecological requirements; agronomy and production areas; reproductive biology; genetic improvement; genetic resources; harvest, postharvest and processing; economics of production; marketing and trade; and current situation and needs for research and technology. None so far is reported on the anatomy of the leaves which maybe responsible for the structural responses of tamarind in environments that make it durable, robust, easy to grow and cultivate. Thus, this study aims to establish the anatomy of the leaves and other structures, and therefore contribute and hopefully complete the baseline information on tamarind. Histological techniques such as freehand , sliding, clearing and paraffin techniques on the leaves were done. The leaf anatomy is of the "non-Kranz" type. The upper and lower epidermises is uniseriate, with stomata on both epidermises (amphistomatic). The mesophyll is differentiated into three layers of long slender, compactly arranged palisade mesophyll, and a few spongy mesophyll cells. The multiseriate palisade mesophyll may be one of the reasons why tamarind is very resistant to strong winds, can tolerate violent typhoons and cyclones (von Maydell, 1986; von Carlowitz, 1986), and hurricane resistant (NAS, 1979). Its strong and pliant branches and a deep and extensive root system, which anchors it to the ground (Coronel, 1986) may also account for the overall tolerance to harsh environments.

Keywords: non-Kranz, epidermis, palisade, spongy mesophyll, stomata, amphistomatic

BS - 07

IDENTIFICATION AND CLONING OF RIBOSOME INACTIVATING PROTEIN (RIP) GENES IN *JATROPHA PANDURIFOLIA* ANDR. AND *JATROPHA PODAGRICA* HOOK.

Maricel Q. Corpuz and Vivian A. Panes

Department of Biology, School of Science and Engineering,
Ateneo De Manila University, Loyola Heights, Quezon City

Ribosome-inactivating proteins inhibit protein synthesis in eukaryotes by catalytically damaging ribosomes and been proven to have antiviral and antitumour activity because it can truncate the translation of viral coat proteins. The presence of RIP genes in *Jatrophapandorifolia* Andr., *Jatropha podagrica* Hook. was screened using 5 primer sets. *Jatropha curcas* L.was used as the control because it already has a known RIP gene as determined by Lin *et al.* (2003). PCR products obtained were analyzed through BLASTs and ClustaWofMEGA4. Cloning of the genomic DNA PCR products and RT-PCR products were performed. Sequence analysis of the cloned PCR products through BLAST showed 87%-90% homology of *J. pandurifolia*'s RIP gene sequence to *J. curcas* precursor gene, while 87%-89% homology of *J. podagrica*'s RIP gene sequence to *J.curcas*' curcin precursor gene. Nucleotide sequence and deduced amino acid sequence revealed possible ORFs in *J. podagrica* and *J. pandurifolia*'s cloned PCR products. Alignment of the deduced amino acid sequence of the cloned PCR products of *J. podagrica* and *J. pandurifolia* to *J. curcas*' curcin revealed the presence of RIP's conserved regions such as SYFF, ALD and EAA which are putative active sites. Identification of the RIP genes in *J. pandurifolia* and *J. podagrica* will shed light on the medicinal properties of the ribosomal inactivating proteins (RIPs) in these plants. It will serve as a guide for those who might be interested in the isolation and purification of these proteins which have known antitumour and antiviral properties.

Keywords: Cloning, Ribosome Inactivating Proteins, antitumour

BS - 08

CHARACTERIZATION OF *Moringa oleifera* LAM. GERMPLASM COLLECTIONS IN THE PHILIPPINES BY DNA FINGERPRINTING

Vivian A. Panes[1], **Elaine Anne L. Tandoc**[1], **Erica E. Fortuno**[1] **and Gabriel O. Romero**[2]

[1]Department of Biology, School of Science and Engineering,
Ateneo De Manila University, Loyola Heights, Quezon City
[2]Monsanto, 7th Floor, Ayala Life-FGU Center, Madrigal Business Park,
Alabang, Muntinlupa City Metro Manila

Moringa oleifera Lam. or *Malunggay* is increasing in commercial relevance because of its nutritional, therapeutic and prophylactic properties. However, in the Philippines, there are few studies of *Malunggay* on the genetic level despite the extensive genetic resources. To contribute to the knowledge about *Malunggay*, in this study, genetic variation of *M. oleifera* germplasm collections in the Philippines was analyzed using intersimple sequence repeats (ISSRs) and SRILS as genetic markers. Seventy five accessions of *Malunggay* cuttings and seeds were collected from different locations in the Philippines. From these cuttings and seeds, genomic DNA was extracted using DNAzol and Zymo plant DNA extraction kits. Twelve ISSRs and nine SRILS primers were used as markers. Genomic DNA was subjected to PCR analysis. The PCR products were run on agarose gels. Then the results were analyzed by scoring the presence (1) and absence (0) of the different bands for all the sample. Polymorphic bands were determined as the bands that were present and absent across the samples. Results showed that there are 8 polymorphic bands using the ISSR primers and 15 of polymorphic bands emerged using the SRILS primers. These results indicate that *M. oleifera* in the Philippines is genetically diverse. A preliminary analysis was done by using the PAUP software and phylogenetic tree construction is underway. More accessions are currently being optimized for PCR analysis. Hence, the extent of genetic diversity of the *Malunggay* germplasm collection in the Philippines is yet to be determined statistically.

Keywords: germplasm, *Moringa oleifera*, DNA fingerprinting

BS - 09

THE EFFECTS OF *Aglaia loheri* BLANCO, *Aleurites moluccana* LINN. AND *Ardisia pyramidalis* (CAVS.) PERS. PLANT EXTRACTS ON HEPATIC HISTOLOGY AND LIPID PEROXIDATION ACTIVITY IN CARBON TETRACHLORIDE-TREATED MICE

Elena S. Catap* and Michelle Alisa DC. Ragudo

Institute of Biology, College of Science, University of the Philippines,
Diliman, Quezon City elenacatap@yahoo.com

Three plant species, *Aglaia loheri*, *Aleurites moluccana*, and *Ardisia pyramidalis* were evaluated for their antioxidant properties by employing lipid peroxidation assays and histological analysis in carbon tetrachloride (CCl_4)-treated mice. Sixty mice were divided into six treatment groups: 1) PBS-injected mice; 2) corn oil injected mice; 3) 30% CCl_4 in corn oil intraperitoneally injected in mice every 72 hrs; 4) *A. loheri* extract + CCl_4; 5) *A. moluccana* + CCl_4; 6) *A. pyramidalis* + CCl_4. Each of the plant extract was injected intraperitoneally on a daily basis. After two weeks, liver tissues were dissected out and subjected to lipid peroxidation assays. Liver samples were also processed for histopathological examination. Lipid peroxidation in both control groups was significantly lower than in CCl_4-treated mice. Malondialdehyde (MDA) levels in *Ardisia*-treated and *Aleurites*-treated mice are higher than the CCl_4-treated mice but the *Aglaia*-treated mice had lower MDA levels than the CCl_4-treated mice. However, the difference was not statistically significant. In the histological examination, the extracts from *Ardisia* and *Aleurites* seemed to induce cellular regeneration in CCl_4-treated mice. Higher degree of vacuolization and nuclear pyknosis, ballooning degeneration and multifocal necrosis were prominent in CCl_4-treated liver tissues. Based on these results, it is recommended that further studies to confirm the antioxidant capacity of these plant species should employ varying extract dosages and longer treatment periods.

Keywords: antioxidant, histopathology, lipid peroxidation, carbon tetrachloride, malondialdehyde, ethanol plant extracts

BS - 10

AMPALAYA (*Momordica charantia* L.) LEAF EXTRACT AS TREATMENT AGAINST NON-ALCOHOLIC FATTY LIVER DISEASE

Romeo B. Rebusi, Jr.* **and Thucydides L. Salunga**

Institute of Biology, University of the Philippines,
Diliman, Quezon City rebusi.reus@yahoo.com

Non-alcoholic fatty liver disease (NAFLD) is characterized by a myriad of condition including steatosis or increase in triglyceride retention in the liver, liver inflammation, and increase in liver enzyme levels in the blood. Steatosis of the liver is due to the increase in the influx of free fatty acids (FFA) into the liver and the prevention of exportation of excess triglycerides. Increase in FFA is linked to obesity and retention of triglycerides is said to be caused by hyperinsulinemia or increase in insulin levels. Cases of NAFLD is said to be rising in an alarming rate and is said to be one of the leading cause of liver failure. A new non-genetic model of diabetes, obesity, and fatty liver was developed, called MSG-mice. This model is induced with the injection in newborn mice of monosodium glutamate (MSG). MSG-mice are regarded as the most comprehensive model described thus far based on the co-existence of multiple aspects of the human metabolic syndrome. This study was conducted to test the effectiveness of ampalaya or bitter melon (*Momordica charantia* L.) in treating various symptoms of NAFLD using the MSG-mice. Neonatal mice were subcutaneously injected for five consecutive days with MSG (2mg/g bodyweight). After eight weeks, MSG- and control-mice were fed with ampalaya leaf extract (10% and 15% gram over milliliter). At 18 weeks old, all the mice were sacrificed, along with positive and negative control groups, and their Body Mass Index (BMI), glucose levels, and liver / body weight ratio were determined. Liver samples were observed for the presence of inflammation and steatosis. It was determined that the BMI of some of the treated groups were significantly lower than that of untreated MSG-mice, that the weight change of the treated were lower than the controls, and that there is reduced steatosis in the treated groups compared to the control groups. It was concluded that both concentrations of ampalaya extract used were effective in lowering steatosis in MSG and non-MSG mice.

Keywords: fatty liver disease, steatosis, *Momordica charantia*, ampalaya, MSG-mice

BS - 11

DIVERSITY AND STATUS OF BUTTERFLIES ACROSS VEGETATION TYPES OF MT. HAMIGUITAN, DAVAO ORIENTAL, PHILIPPINES

Alma B. Mohagan

Department of Biology, Central Mindanao University,
Musuan, Bukidnon almohagan@gmail.com

An inventory was conducted to determine the diversity and status of butterflies of Mt. Hamiguitan, wildlife Sanctuary, Davao Oriental using quadrat method in five vegetation types namely; agroecosystem (10-400 masl), dipterocarp (500-900 masl), montane (900-1400 masl), mossy (1400-1500 masl) and pygmy (1500-1675 masl). Two 20m x 20m plots were established per vegetation type. These inventory techniques revealed 141 species of butterflies plus one subspecies described and illustrated. Diversity assessment using Shannon-Weiner index showed high level (4.1) in the Montane forest as compared to other vegetation types. Bray-Curtis similarity index shows low species similarity in pygmy, mossy and agro-montane forest (< 40 %). Furthermore, this study revealed 3 possible new species, 44 endemics: 2 eastern Mindanao endemic (very rare), 4 Mt. Hamiguitan endemic (very rare), 16 Mindanao endemic and 22 Philippine endemic. Seven new species were recorded in Mindanao. Mt. Hamiguitan wild life Sanctuary is the home of diverse and endemic butterfly species. The study recommends further monitoring to establish population and species richness trend. It further recommends that policies and ordinances should be formulated by PAMB and LGUs.

Keywords: butterflies species richness trend pygmy vegetation types Mt. Hamiguitan

BS - 12

INVENTORY OF EARTHWORMS IN MT. HILONG-HILONG RANGE, AGUSAN DEL NORTE, PHILIPPPINES

Mary Alona L. Murro[1], Alma B. Mohagan[1*] and Jayson Ibañez[2]

[1]Biology Department, Central Mindanao University
Musuan, Bukidnon. almohagan@gmail.com
[2]Biodiversity Coordinator, Philippine Eagle Foundation, Davao City

Earthworms play an important role in the ecosystem as keystone species on soil formation. They plough the soil, mix, till and build topsoil as they burrow through the earth. They consume large quantities of soil and fresh or partially decomposed organic matter from the soil surface, depositing it as fecal matter/casts in the lower soil horizons. They loosen and aerate the soil as they tunnel the ground affecting the hydrology cycle. Earthworms in their natural habitats improve soil structure, fertility, and the rate that organic matter gets broken down. Yet, they are less known in the Philippines. Here, inventory of earthworms in the mossy and montane forests of Mt. Hilong-hilong, Diwata Range, Agusan del Norte was conducted for 15 days to provide information on species composition and richness using digging, hand sorting and counting methods. Twenty six species were identified and 6 were undetermined. These were placed in 32 genera. Twenty species were disconcordant in the mossy and seventeen species in the montane forest. Only six species of earthworms were concordant in both vegetations. Species diversity was higher in the mossy forest H' = 2.788 compared to montane forest H' = 2.674. Similarity of species composition showed low Si=25.33% between the mossy and montane forests which implies that mossy and montane forests are two unique habitats for earthworms in Mt. Hilong-hilong. It is recommended that cast formation and biology of these earthworms be studied as future source of vermicast organic fertilizer production rather than using exotic stocks.

Keywords: inventory, earthworm, Mt. Hilong-hilong, Philippines, species diversity

BS - 13

ANTIOXIDANT PROPERTY AND HISTOLOGICAL EFFECTS OF THE ETHANOLIC PLANT EXTRACTS OF *Ficus septica* BURM, F. and *Uncaria perrottetii* (A. RICH) MERR IN MICE

Elena S. Catap* and Excelle Grace M. Canonizado

Institute of Biology, College of Science, University of the Philippines,
Diliman, Quezon City elenacatap@yahoo.com

Many antioxidants that confer protection through inhibition of cellular membrane damage have been developed from various plants. In this study, the antioxidant properties of the leaf extract of *Ficus septica*, and the vine bark extract from *Uncaria perrottetii*. were assessed by measuring the malondialdehyde (MDA) levels, as by-product of lipid peroxidation, in the liver of ICR mice. In addition, histological examination was undertaken to determine the hepatoprotective effects of the two plant extracts. Mice were grouped into five cages, as follows: 1) negative control- phosphate buffered saline (PBS); 2) negative control- corn oil only 3) positive control- intraperitoneal injection of 30% carbon tetrachloride (CCl_4) in corn oil (1 ml/kg BW) every 72 hours; 4) treatment with *U. perrottetii* + CCl_4; and 5) treatment with *F. septica* + CCl_4. Both crude extracts were injected at a dosage of 250 mg/kgBW, and were given daily through intraperitoneal injections for two weeks. Results obtained from the lipid peroxidation assay showed that the CCl_4- treated mice had the highest MDA levels. Lower MDA levels were detected in the extract-treated mice and in the negative control mice but these were not statistically significant. However, histological examinations revealed various signs of cellular repair in the hepatic tissues of the extract-treated mice relative to the degenerative and necrotic pathologies in the CCl_4-injected mice. The results show that the extracts conferred hepatoprotection but future validation studies must employ a longer period of treatment and other routes of extract application.

Keywords: antioxidant, histopathology, lipid peroxidation, *Uncaria perrottetii*, *Ficus septica*

BS - 14

EFFICACY OF FORMULAYED ANTI-MOSQUITO GEL FROM EUCALYPTUS (*Eucalyptus globules*) AND CARRAGEENAN

Charie Mayne Alcon[1], Ma.Tereza A. Blanco[1] and Shirley C. Agrupis[2]

Mariano Marcos State University, College of Arts and Sciences
Department of Biology, City of Batac, Ilocos Norte matetblanco@yahoo.com

The *in vitro* and *in situ* biocidal property of Eucalyptus gel formulation against larval mosquitoes was evaluated. The study aimed to (1) determine the effects on the morphology of mosquitoes; and (2) determine the mosquito repellant effect of the gel formulation .

Eight (8) different treatments were prepared for the *in vitro* test namely: T1 (combination of carrageenan and eucalyptus oil; T2 (combination of carrageenan and eucalyptus powder; T3 (pure carrageenan); T4 (pure eucalyptus oil);T5 (pure eucalyptus powder); T6 and T7 (positive control, commercial anti mosquito lotion for kids and adult); and T8 (negative control). Morphological effects of formulated treatments on the larval mosquitoes include the loss of body fluid, antennae and cut hair-like structures. In terms of mortality rate, eucalyptus oil formulation was not significantly different from the positive control, which both achieved 100% mortality. Based on the *in vitro* test, pure eucalyptus oil was observed as the most effective in attaining 100% mortality in just 29.33 minutes after application. The first three (3) formulations that gave the best result were used in the in situ evaluation namely T1,T2 and T4.

The *in situ* evaluation revealed that the combination of carrageenan and eucalyptus oil (T1) and commercial anti-mosquito lotion gave the best result in which no mosquito bites were observed by the human samples. Coefficient of Variation (CV) revealed that there are no significant differences between the formulated treatments and positive control on the number of mosquito bites.

The abundance of eucalyptus plant in the country can be tapped for the production of environment-friendly anti-mosquito product.

Keywords: *in vitro, in situ,* biocidal property,repellant, anti-mosquito

BS - 15

COMPARATIVE STUDY ON THE BIOACCUMULATION OF LEAD (Pb) IN THE TISSUES OF *Arachis pintoi* (PINTO PEANUT) AND *Imperata cylindrical* (COGON GRASS) GROWN IN LEAD CONTAMINATED SOIL

Flordeliz R. Estira[1*], Arlene Lu-Gonzales[2] and RenJean Esmerio[2]

[1]Biology Department, College of Arts and Sciences,
Mariano Marcos State University, City of Batac, Ilocos Norte
[2]Environmental Science Department, College of Arts and Sciences,
Mariano Marcos State University, City of Batac, Ilocos Norte
flordeliz_estira@yahoo.com

Plants have the intrinsic capability to take up heavy metals and store and accumulate these in their tissues. These capabilities are selective of plants, of the heavy metal adsorbed and on their interactions with the physico-chemical environment. This is the basis of selection of plants that could be used as potential phytoremediators. The Pb uptake potential in the root and shoot tissues of pinto peanut and cogon grass seedlings was investigated in potted greenhouse conditions and analyzed using Atomic Absorption Spectrophotometry (AAS). Percentage survival of the plants was also evaluated. Results indicated significant differences in Pb accumulation in both the root and shoot tissues of the two plants grown for 2 months in lead contaminated soil. Pinto peanut seedlings accumulated significantly greater amount of lead in their shoot (11.26 ppm) and root (8.74 ppm) than cogon grass seedlings which accumulated a noncomparably lesser amount of 1.47 ppm Pb in their shoot and 0.15 ppm Pb in their root. However both plants showed 100% survival when grown in lead-contaminated soil. The greater bioaccumulation of lead in the tissues of pinto peanut suggests that the plant may be used for the clean up and phytoremediation of Pb-contaminated soils.

Keywords: lead (Pb), bioaccumulation, pinto peanut (*Arachis pintoi*), cogon grass (*Imperata cylindrica*), phytoremediation

BS - 16

PHYTOCHEMICAL SCREENING AND EVALUATION OF AN INDIGENOUS PLANT SPECIES, PAPAIT (*Mollungo oppositifolia*) AS A POTENTIAL ANTI-DIABETIC PLANT

Leny I. Guillermo and Flordeliz R. Estira*

Biology Department, College of Arts and Sciences, Mariano Marcos State
University, City of Batac, Ilocos Norte flordeliz_estira@yahoo.com

The development and exploration of the remaining Philippine indigenous plants as a botanical resource remain a researchable area not only in medicine but also in biodiversity conservation. Papait (*Mollungo oppositifolia)* is very common weed indigenous to the Philippines. The whole plant is eaten as a vegetable by local folks and is claimed to have anti-diabetic effects and is reported lately as exceptionally rich in iron and calcium. The study aimed to determine the phytochemicals present in Papait leaf extract and evaluate its potentials to lower blood glucose level of white mice. The study was laid in Completely Randomized Design (CRD) in 5 treatments: T1 (0.0025 mlPLE/g wt of mice), T2 (0.005 mlPLE/g wt of mice), T3 (0.0075 ml PLE/g wt), T4 (0.01 ml PLE/g wt) and T5 (control-insulin). The extract was orally fed to hyperglycemic mice. Phytochemical screening of the leaf extract showed this to be very rich in phytochemicals including alkaloids, saponins, steroids, fixed oils and proteins. All the treatments exhibited comparable degrees of potency in lowering blood glucose levels in both sexes of white mice. Percentage (%) reduction of BGL as affected by Papait leaf extract in all the mice was comparable to percentage reduction caused by insulin. The results remarkably demonstrated comparable effect of the plant with the anti-diabetic drug insulin. It is concluded that Papait Leaf Extract can be used as a component material for the preparation of anti-diabetic drugs. It likewise confirms claims of folkloric medicine that the Papait plant can lower blood sugar level hence, it can be a very good complement or substitute for the expensive commercially available anti-diabetic drugs.

Keywords: phytochemical chemistry, hypoglycemic ffects, papait (*Mollungo oppositifolia),* antidiabetic

BS - 17

MOLECULAR PHYLOGENY AND A NOVEL SPECIES OF THE PHILIPPINE ENDEMIC *Antherostele* (Rubiaceae), WITH IMPLICATIONS ON ITS CONSERVATION AND BIOCHEMICAL PROPERTIES

Jasper John A. Obico*[1] **and Grecebio Jonathan D. Alejandro**[2]

[1]Department of Biology, University of the Philippines Manila, Padre Faura St., Ermita, Manila jjobico@post.upm.edu.ph; [2]College of Science and Research Center for the Natural Sciences, University of Santo Tomas, España, Manila gdalejandro@mnl.ust.edu.ph

Antherostele Bremek. is an understudied genus of Rubiaceae endemic to the Philippines comprising four species of small to medium-sized trees distributed in Luzon and Visayas. All species of the genus are threatened and categorized by the DENR (2007) under vulnerable status. Since its segregation from *Urophyllum* Wall. on the basis of morphology of limited herbarium materials, *Antherostele* has not been challenged by any molecular study to test its monophyly and determine its phylogenetic placement within Rubiaceae. To address these gaps, field collection of all *Antherostele* species was done and sequences of the Internal Transcribed Spacer (ITS) region (nrDNA) were utilized for phylogenetic analysis. Parsimony analysis of the ITS data showed that *Antherostele* is monophyletic with strong support (BS=100%) and is nested within the tribe Urophylleae. The genus is united by hairy corolla lobes and syngenesious stamens. The montane *A. luzoniensis* is found to be sister to the rest of shaded understory *Antherostele* species. The present study recognizes five *Antherostele* species including a new species (*A. samarensis* Obico & Alejandro). Three *Antherostele* species (*A. luzoniensis*, *A. callophylla* and *A. samarensis*) have restricted distributions and are considered rare based on field observation. Hence, a sustainable conservation management is herein urged for the three species of the endemic genus. By its placement under subfamily Rubioideae, *Antherostele* is a potential source of anthraquinones, an organic compound with many industrial and medicinal applications.

Keywords: *Antherostele*, conservation, nrDNA, Philippine endemic, Rubiaceae, Urophylleae

BS - 18

HISTOCOMPATIBILITY ASSESSMENT IN THE CHICKEN COLONIES CLOSED FOR SEVERAL YEARS

Marcos B. Valdez Jr.[4], Keiji Kinoshita[2], Makoto Mizutani[2], Akira Fujiwara[3], Hajime Yazawa[3], Takahiro Yamagata[1], Kiyoshi Shimada[2] and Takao Namikawa[1,2*]

[1]Laboratory of Animal Genetics and [2]Avian Bioscience Research Center, Graduate School of Bioagricultural Sciences, Nagoya University, Chikusa, Nagoya, [3]Laboratory Animal Research Station, Nippon Institute for Biological Science, Kobuchizawa, Hokuto, Yamanashi, Japan and [4]Biology Department, School of Science and Engineering, Ateneo de Manila University, Loyola Heights, Quezon City

Despite the numerous calls for the conservation of avian genetic stocks, there is still continued elimination of chicken research populations. The core objective for conservation of farm animal genetic resources is to maintain access to the adaptive genetic potential of collection of valuable research resources. The closed colonies of chicken used in this experiment are well characterized. However, assessment of isogeneity of these closed colonies has not been performed, thus, immunological methods to assess the status of the histocompatibility antigens were employed. The RIR-Y8/NU, BL-E, YL and WL-G chicken closed colonies were assessed by hemagglutination test using anti-red blood cell (RBC) antibodies (HT), skin transplantation test (STT) and formation of isohemagglutinins (FIHs) during STT. The YL individuals all showed the survival of skingrafts with no FIHs in STT and no RBC antigenic variations in HT, indicating high homogeneity at the serological loci. The BL-E as well as WL-G closed colonies were also found to be histocompatible in the STT with no FIHs, although the HT showed heterogeneities at serological locus/loci other than the *B* and *C* blood group loci which have significant effects on histocompatibility. In the RIR-Y8/NU closed colonies, STT showed early skingraft rejection with positive FIHs caused by different *B* locus alleles, suggesting relatively high heterogeneities. The YL, BL-E and WL-G are isogenic lines, however, RIR-Y8/NU needs further selection.

Keywords: chicken closed colony, chicken genetic resource, hemagglutination, isohemagglutinin, skin transplantation

BS - 19

CLONING AND SEQUENCE ANALYSIS OF PARTIAL cDNAs OF SELECTED GENES IN NORMAL AND MUTANT 'MAKAPUNO' ENDOSPERMS OF COCONUT (*Cocos nucifera* L.)

Reggie Yadao-dela Cruz[1,2*], Rita P. Laude[3], Ma. Genaleen Q. Diaz[3], Antonio C. Laurena[4], Merlyn S. Mendioro[3] and Evelyn Mae T. Mendoza[4]

[1]Graduate School, University of the Philippines, Los Baños, Laguna
[2]Department of Biology, College of Arts and Sciences, Central Mindanao University, Musuan, Bukidnon reggiecmu@yahoo.com
[3]Institute of Biological Sciences, College of Arts and Sciences and
[4]Institute of Plant Breeding, Crop Science Cluster, College of Agriculture
University of the Philippines, Los Baños, Laguna

Makapuno is a mutant coconut (*Cocos nucifera* L.) with over-proliferating endosperm, occurring in the Philippines and Dutch East Indies. Its fruit is almost full of viscous and delicious endosperm, commercially useful in making ice cream, pie and other delicacies. The science behind the makapuno phenomenon has not been completely understood. To determine the molecular basis of the makapuno phenomenon, we designed primers and cloned genes involved in cytokinin biosynthesis, polyamine synthesis, galactomannan degradation, glycolysis, alcoholic fermentation, fatty acid biosynthesis and cell cycle regulation. The total RNA from normal and makapuno endosperms of coconut were isolated and reversed transcribed. The cDNAs were used as template for PCR. The PCR products were ligated into pGEM®-T Easy vector, cloned in *Escherichia coli* JM109 cells and sequenced. A total of thirteen partial cDNA sequences were obtained. Interproscan and BLAST analyses revealed the cDNAs contain conserved domains and are highly homologous (68-98%) to equivalent sequences from other plant species. Pairwise alignment (Blossom 62) of the thirteen partial cDNAs between normal and mutant makapuno coconut revealed absence of nucleotide sequence differences implying no structural differences in the genes of the two coconut genotypes. The makapuno and normal coconut used in this study are of the same Laguna Tall variety.

Keywords: coconut endosperm, endosperm overgrowth, cDNA cloning, makapuno

BS - 20

SELECTION OF A SUITABLE INTERNAL CONTROL GENE FOR RELATIVE RT PCR-BASED EXPRESSION ANALYSIS IN NORMAL AND MUTANT 'MAKAPUNO' ENDOSPERMS OF COCONUT (*Cocos nucifera* L.)

Reggie Yadao-dela Cruz[1,2*], **Rita P. Laude**[3], **Ma. Genaleen Q. Diaz**[3], **Antonio C. Laurena**[4], **Merlyn S. Mendioro**[3], **and Evelyn Mae T. Mendoza**[4]

[1]Graduate School, University of the Philippines, Los Baños, Laguna
[2]Department of Biology, College of Arts and Sciences, Central Mindanao University, Musuan, Bukidnon reggiecmu@yahoo.com
[3]Institute of Biological Sciences, College of Arts and Sciences and
[4]Institute of Plant Breeding, Crop Science Cluster, College of Agriculture
University of the Philippines, Los Baños, Laguna

Makapuno is an abnormal coconut with over-proliferating endosperm that is softer and fluffy due to galactomannan accumulation. To perform relative RT-PCR for expression analyses of important genes possibly involved in the makapuno phenomenon, an internal control gene was initially determined for normalization purposes. Three genes were studied, namely, glyceraldehyde-3-phosphate dehydrogenase (GAPDH), enolase and actin. GAPDH and enolase were found to have considerably varied expression level in all the samples and are not recommended to be used as internal control genes. The actin gene has a uniform detectable expression in all the samples studied and is recommended for use as reference gene for normal coconut and mutant makapuno solid endosperm samples at stages 5–6, 6–7 and 7–8 months.

Keywords: actin, coconut endosperm, endosperm overgrowth, internal control, makapuno, relative RT-PCR

BS - 21

BIOPHYSIOLOGY OF *Panaeolus* spp. ASSOCIATED WITH DRIED DUNG OF DOMESTICATED RUMINANTS

Excel Rio S. Maylem, Rich Milton R. Dulay,
Sofronio P. Kalaw and Renato G. Reyes

Center for Tropical Mushroom Research and Development,
Department of Biological Sciences, College of Arts and Sciences,
Central Luzon State University, Science City of Munoz, Nueva Ecija

Panaeolus is a psychoactive mushroom usually found growing on dried dung of domesticated ruminants such as carabao and horse. In our efforts to look for alternative source of bioactive compounds for drug development, we initiated a study on domestication of this wild mushrooms hallucinogenic activity. The baseline information generated in this research is necessary for the development of production technology towards its sustainability. As a prelude to its domestication, we evaluated the nutritional and physical requirements for efficient mycelial growth of *Panaeolus* sp. and fruiting body performance on different formulated substrates under aseptic condition. Wild strain of *Panaeolus* sp. was obtained from dried carabao dung. The cell lines were aseptically rescued following the standard tissue culture protocol for mushroom.

Among the different media evaluated, results revealed that potato sucrose gulaman (PSG) was the best culture medium as indicated by vigorously thick mycelial density and shortest incubation period with a mean of 9 days. Moreover, potato sucrose gulaman with pH of 6.5 incubated under sealed and alternating light-dark condition favored the mycelial growth of *Panaeolus* sp.

Aseptic cultivation using 90% carabao dung and 10% rice bran recorded thick and fast mycelia growth and shortest incubation period with a mean of 11 days. However, 90% horse dung and 10% rice bran yielded more fruiting bodies with a mean of 0.39g, highest mean height of 33.67 mm and highest biological efficiency with a mean of 1.15%.

Keywords: Psychoactive mushroom, *Panaeolus*, domesticated ruminants, aseptic cultivation

BS - 22

MORPHO-ANATOMICAL INVESTIGATION ON STIPULES, COLLETERS AND LEAVES OF *Rhizophora* spp. IN PAGAPAS BAY, CALATAGAN, BATANGAS, PHILIPPINES

Leah E. Endonela*[1], Maribel L. Dionisio-Sese[2], Nestor C. Altoveros[3] and Teresita H. Borromeo[3]

[1]Plant Genetic Resources Conservation and Management,
e_endonela@yahoo.com; [2]Institute of Biological Science,
College of Arts and Sciences, mldsese@yahoo.com and
[3]Crop Science Cluster, College of Agriculture, University of the Philippines
Los Baños, Laguna thborromeo@yahoo.com

The morphology and anatomy of stipules, colleters and leaves of four identified *Rhizophora* species: *R. apiculata*, *R. mucronata*, *R. stylosa* and putative hybrid *R.* x *lamarckii* in Pagapas Bay, Calatagan, Batangas, Philippines were investigated. Stipule color and size vary within species. The stipule has mono-layered epidermis covered with thick cuticular wax, distinct aggregated sclereid ideoblasts, well-developed vascular bundle, and multilayered hypodermis. The variations in color, exudates consistency and series number of aggregated colleters were elucidated. Colleters have central axis composed of slender, elongated cells surrounded by palisade-like epidermis. Differences in leaf morpho-anatomy including leaf shape, leaf structure, mucus cells, vascular bundle, stomata apparatus and corkwarts were also observed in the four *Rhizophora* species. In general, *Rhizophora* leaf consists of thick cuticle, single layer epidermis, 6 to 7 layers of hypodermis, funnel-shaped mucus cells, parallel columns of four layers of palisade mesophyll, and varied layers of spongy mesophyll. The presence of druse crystals in stipule and leaves is common. Features of the stomatal apparatus incuding guard cells lignin thickness, subsidiary cells arrangement and stomatal aperture are considered diagnostic. Guard cells have a pair of inner and outer cuticular ridges. Stomata and corkwarts, which developed from stomata as the leaves mature, are concentrated only in abaxial leaf surface. These morpho-anatomical characteristics of *Rhizophora* species exemplified the structural modifications that enabled them to adapt to the physiologically dry, saline mangrove environment.

Keywords: colleter, leaf anatomy, leaf morphology, *Rhizophora* spp., stipule

BS - 23

FUNCTIONAL STUDIES OF WSSV-SHRIMP HOMOLOGS BY DS-RNA INTERFERENCE IN THE SHRIMP *Marsupenaeus japonicus*

Mary Beth B. Maningas[1,2]*, **Hidehiro Kondo**[2], **Ikuo Hirono**[2]

[1]Department of Biological Sciences, College of Science, University of Santo Tomas, Manila; [2]Laboratory of Genome Science, Tokyo University of Marine Science and Technology, Minato-Ku, Tokyo, Japan mbmaningas@mnl.ust.edu.ph

Global shrimp production tripled over the past decade from 750,000 tonnes in the 1990s to more than three billion tonnes over the past five years, severely affecting ecosystems and livelihoods. White spot syndrome virus (WSSV) is a deadly pathogen for shrimp and a major threat to shrimp farming for the last two decades. Elucidation of the underlying mechanism of shrimp-virus interaction will be of enormous help in the prevention and management of shrimp viral diseases.

Double stranded RNA interference (ds-RNAi) is a powerful tool in elucidating gene function in invertebrates. ds-RNAi allows interference with the expression of a gene in a highly sequence specific manner and thus essential genes can be targeted by design, with little or no risk of undesired off-target effects.

Eight (8) WSSV-shrimp homologs ORFs (MjORF13, MjORF17, MjORF 20, MjORF23, MjORF24, MjORF31, MjORF34, MjORF37) from Kuruma shrimp previously identified to have significant homology with WSSV were studied using dsRNA interference technology.

The eight WSSV-shrimp homologs were successfully amplified, sequenced and dsRNA have been produced. Interestingly, initial results showed that shrimp injected with MjORF31- and MjORF34-dsRNA significantly increased mortality compared to that of the PBS injected group, highlighting their importance in shrimp survival.

Challenge test results with white spot virus syndrome (WSSV) as pathogen showed that shrimp injected with MjORF17-, MjORF20-, MjORF23- and MjORF34-dsRNA have higher survival rate than those of the control. Hence, dsRNAs of these four WSSV-shrimp homologs may have protective effect on shrimp against WSSV infection. It would be interesting to pursue or assess the stability and the possible therapeutic application of dsRNAi in shrimp.

Keywords: shrimp, dsRNA interference, homologs, white spot virus syndrome, shrimp-virus interaction

BS - 24

COMPUTATIONAL RECONSTRUCTION AND CHARACTERIZATION OF ANCESTRAL PROTEINS RELEVANT TO PROKARYOTIC-EUKARYOTIC TRANSITION 2 BILLION YEARS AGO

Ma. Angelica S. Pulido[1], Evelyn M. Labastilla[1], Guia M. Guiam[1], Merilyn C. Pascua[1], Marla A. Endriga[2] Chester C. Deocaris[2] and Custer C. Deocaris[3*]

[1]College of Science, Tarlac State University, Tarlac City; [2]College of Biology, University of the Philippines-Manila and [3]Yonsei University School of Medicine, Seoul, South Korea cdeocaris@gmail.com

Anaphase Promoting Complex 11, most probable ancient Ubiquitin ligase is correlated with a former study to have emerged after the Great Oxidation Event. RPN3, most ancient non-ATPase 26S proteasome lid appeared in cyanobacteria and proteobacteria emergence. However, BCS1 in mitochondria inner membrane, found to be the most ancient protein, functions as an ATP-dependent chaperone which led to some rise of multicellular organisms. The AA sequences of these genes were retrieved with Entrez gene in FASTA format and aligned with CLUSTALW via MEGA 4. Using gamma distribution and JTT matrix rates, distance matrices were obtained and used to create a phylogenetic tree. The ancestral sequences were generated in the codeml program of PAML. ProtParam and SWISS-MODEL were used in characterizing the 1^0, 2^0, and 3^0 structures of derived ancestral proteins and reference species. Using ProtParam, physico-chemical properties of Ub ligase, mitochondrial chaperones and proteasome were known. Visualization of the 3-dimensional structures of these ancestral proteins was requested from SWISS MODEL and Rasmol. New Likelihood-based Probability Method gave 0.91, 0.86, and 0.73 for all, variable, and parsimony-informative sites, respectively, whereas the corresponding probabilities for the parsimony method were 0.84, 0.76, and 0.51, respectively. The probability that ancestral sequence was correctly reconstructed ranged from 91.3 to 98.7%.

Keywords: ubiquitin proteasomal degradation, mitochondrial chaperones, PAML, ProtParam, SWISS-MODEL

BS - 25

CRUSTACEANS AS BIOINDICATORS OF WATER IN LABAC RIVER OF CAVITE

Dickson N. Dimero* and Josefina R. Rint

Department of Biological Sciences, College of Arts and Sciences,
Cavite State University, Indang, Cavite blue_dickson@yahoo.com

The study was conducted to assess the physico-chemical status of water in Labac river in Cavite using crustaceans as bioindicators.

The occurrence of crustaceans was assessed quantitatively in eight sites of the western Labac river and nine sites of the eastern Labac river of Cavite.

The banks of the entire Labac river are highly vegetated. Some river waters are still undisturbed and clear while majority of the river stations have polluted waters.

Labac river is narrow (6.84 m) and very deep (>1.50 m) with a cold water temperature (25.9ÚC), slow water velocity (26.41 cm/sec), alkaline water (7.25), had sufficient DO (7.72 mg/L), low level of TDS (0.27Úbrix), high level of TSS (280 mg/L) and high level of salinity (2 ppt).

Shrimp population count was positively correlated with width, pH, and TDS and moderately correlated with depth and salinity of river water while crab population is moderately correlated with river water temperature. DO has a very small positive correlation with crab population.

Shrimps favor wide and deep rivers with acidic to alkaline water, and low level of TDS and salinity in water. Crabs prefer shallow, cool and shaded areas and high level of dissolved oxygen in river water.

Shrimps can be bioindicators of pH, total dissolved solids and salinity, while crabs can be bioindicators of temperature and dissolved oxygen in river water.

Keywords: Crabs, Dissolved Oxygen (DO), Labac River, pH, Salinity, Shrimps, Total Dissolved Solids (TDS), Total Suspended Solids (TSS)

BS - 26

PROCRUSTEAN ANALYSIS OF WING SHAPE DIVERGENCE AMONG SIBLING SPECIES OF *Neurothemis* DRAGONFLIES

Stephenson A. Harun, Mark Anthony J. Torres, Joey Genevieve T. Martinez, Sharon Rose M. Tabugo, Liza A. Adamat, Muhmin Michael E. Manting, and Cesar G. Demayo

Department of Biological Sciences, College of Science and Mathematics, Mindanao State University – Iligan Institute of Technology, Iligan City, Lanao del Norte

torres.markanthony@gmail.com; cgdemayo@gmail.com

Dragonflies under the genus *Neurothemis* are the common red- and brown- winged dragonflies that one sees often in drains, small ditches and ponds. There are more than four confusing *Neurothemis* species that are easily mixed up by their similar color and pattern. These species are difficult to identify in the field. The present study determined wing shape divergence in three species of *Neurothemis* dragonflies collected from Northern Mindanao, Philippines. These include *N. terminata terminata* Ris (29 females, 7 Males), *N. fluctuans* Fabricius (9 females, 4 males), and *N. ramburii ramburii* Kaup & Brauer (6 females, 7 males). A generalized least square fitting analysis was done via Procrustes superimposition of landmarks from the wings. Relative warp analysis showed significant variation among the *Neurothemis* species. The first extracted relative warp showed differences in the shape of the pterostigma and disparity in the distance between the distal end of the radial planate supplement and the distal margin of the wings bounded by the end points of the intercalary vein and the radial branch. This accounted for 26.78-42.28% of the variation in the shapes fore wing. Differences in the shape of the pterostigma were also observed and accounts for 27.98-44.18% of the variation in the shape of the hind wing. Distance matrices were constructed for the four data sets: left and right fore wing; left and right hind wing. Then, comparison was done via correlation analyses of the four matrices of distances among the species. Results showed that the shape of the fore wing contributed more in discriminating among species when compared to the hind wing. The result of the study is discussed in relation to the utility of wing morphology in the taxonomy and discrimination of sibling species of *Neurothemis* dragonflies.

Keywords: *Neurothemis*, procrustes analysis, dragonfly, wings, sibling species

BS - 27

CYTOGENETIC ANALYSIS OF SELECTED NATIVE AND ENDEMIC PHILIPPINE FRESHWATER FISH SPECIES

Roberto C. Reyes[1], Celia B. Dela Viña[1],
Nathaniel T. Carpena[1] and Pablo P. Ocampo[1,2]

[1]Institute of Biological Sciences, College of Arts and Sciences,
University of the Philippines, College, Los Baños, Laguna mbb.ibs@gmail.com
[2]UPLB Limnological Research Station, College of Arts and Sciences,
University of the Philippines, College, Los Baños, Laguna uplblrs@yahoo.com

Cytogenetic analysis may show variations in chromosome numbers and total DNA content of the nuclei which enables the identification of species origins and relationships. This project plans to establish the chromosome constitution of selected native and endemic fishes.

Ten individuals from each species were collected from Laguna, Quezon and Bicol freshwater rivers and lakes. The selected species were *Glossogobius celebius* Valenciennes (Gobiidae), *Glossogobius giuris* Hamilton (Gobiidae) and *Leiopotherapon plumbeus* Kner (Terapontidae). Metaphase chromosomes were collected from regenerating blastemas of the caudal fin following the modified procedure of Vallejo (1982). The cut caudal fin was first allowed to regenerate for 2-3 days, and then the fishes were transferred to a beaker containing a 0.003% colchicine solution for 4 hours. The regenerated fin was severed from the fish, transferred to distilled water and treated with Carnoy's solution each for 30 minutes. All the smears were stained with either 20% Giemsa or 2% aceto-orcein. The search for 20 C-metaphase cells per species is ongoing using light microscopy. Expected outputs for the first year of the project include the chromosome numbers and karyograms of the selected endemic fishes in order to provide baseline information for biodiversity and evolutionary researches.

Keywords: karyotype, *Glossogobius celebius, Glossogobius giuris, Leiopotherapon plumbeus,* karyogram

BS - 28

ISOZYME ANALYSIS OF SELECTED NATIVE AND ENDEMIC PHILIPPINE FRESHWATER FISH SPECIES

Rita P.Laude[1], Roberto C. Reyes[1], Diana Rose O. Rogelio[1] and Pablo P. Ocampo[1,2]

[1]Institute of Biological Sciences, College of Arts and Sciences,
University of the Philippines, Los Baños, Laguna mbb.ibs@gmail.com
[2]UPLB Limnological Research Station, College of Arts and Sciences,
University of the Philippines, Los Baños, Laguna uplblrs@yahoo.com

In an attempt to establish the genetic relationship and to assess the genetic variation within and in between populations of selected endemic and native Philippine freshwater fishes, isozyme analysis was done using starch gel electrophoresis for four different enzyme systems namely: esterase, alkaline phosphatase, malic enzyme and, á-glycerophosphate dehydrogenase. A total of 285 fish individuals were used that include *Leiopotherapon plumbeus* ("Ayungin"), *Glossogobius celebius* ("Biyang Bato"), *Glossogobius giuris* ("Biyang Bato") and *Ophieleotris aporos* ("Bangayngay"). The crude protein extracts were loaded onto the starch gel and the electrophoretic run followed. After the run was finished, the gel was sliced into four and each slice was soaked in a specific enzyme staining solution. The resolved bands were then scored and the relative mobility (Rf) values were computed for each band. Preliminary results include the banding patterns observed for each population and the temporarily assigned presumptive loci and alleles based on the Rf values. The loci and alleles assigned are still temporary until all the bands are confirmed for their presence and position which is now underway. These loci and alleles will be the basis for the statistical analysis using the POPGENE32 software. However, to have a more effective analysis, four more enzyme systems namely: acid phosphatase, lactate dehydrogenase, phosphogluconic acid, and isositric dehydrogenase will be added.

Keywords: isozyme, starch gel electrophoresis, esterase, alkaline phosphatase, malic enzyme, á-glycerophosphate dehydrogenase, acid phosphatase, lactate dehydrogenase, phosphogluconic acid, isositric dehydrogenase, *Leiopotherapon plumbeus, Glossogobius celebius, Glossogobius giuris,* and *Ophieleotris aporos*

BS - 29

ISOLATION AND CHARACTERIZATION OF MICROSATELLITES IN SELECTED PHILIPPPINE NATIVE AND ENDEMIC GOBIIDAE SPECIES

Aprill S. Punongbayan[1], Ma. Genaleen Q. Diaz[1], Abriel S. Bulasag[2], Ma. Cecilia S. Reamillo[1], and Pablo P. Ocampo[1,3]

[1]Institute of Biological Sciences, mbb.ibs@gmail.com, [2]UP Rural High School, and [3]UPLB Limnological Research Station, College of Arts and Sciences, University of the Philippines Los Baños, Laguna uplblrs@yahoo.com

Microsatellite markers enable species and parentage identification, assessment of genetic diversity and population genetic structure, determine relationships between populations and species and are widely used in marker-assisted breeding. These highlight the significance of the isolation, characterization and development of microsatellite markers from our own, Philippine endemic and native freshwater fishes. Five Philippine native and endemic Gobiidae species were collected. These include *Glossogobius celebius*, *G. giuris*, *Rhinogobius sp.*, *Gobiopterus lacustris*, and *Mistichthys luzonensis*. A modified protocol for the construction of microsatellite enriched genomic library was used. Genomic DNAs from each species were pooled, digested with *Rsa I*, size-selected, purified and ligated to *Mlu I* adaptors. DNA containing microsatellite fragments were captured by biotinylated oligo probes [$(TC)_{10}$, $(CA)_{10}$, and $(TG)_{10}$] and recovered using streptavidin magnetic particles. After PCR, the product was ligated to pGEM-T Easy vector, transformed into JM109 competent cells and plated on Luria-Bertani (LB) agar medium. Recombinant plasmids identified through blue-white screening were sequenced. However, there was low efficiency. To increase efficiency in selecting clones containing microsatellites, more stringent hybridization procedures are now being explored including the hybridization of colonies with non-radioactively labelled repeat oligonucleotides.

Keywords: microsatellites, library construction, *Glossogobius celebius*, *Rhinogobius sp.*, *G. giuris*, *Gobiopterus lacustris*, and *Mistichthys luzonensis*

BS - 30

CYTOTOXICITY OF FIVE ANTI DENGUE PLANTS USING *Artemia salina*

Doreen D. Domingo* **and Febee Rose D. Gamet**

Mariano Marcos State University,College of Arts & Sciences,
Biology Department, City of Batac, Ilocos Norte dr_dhee_dee@yahoo.com

The quest to prevent and cure dengue is continuous. Various researches show that *Uncaria tomentosa* or cat's claw (Yano, 2009); *Ipomoea batatas* or sweet potato (Villafuerte, 2010); *Euphorbia hirta L.* or tawa-tawa (Cabaral, 2011); *Azadirachta indica* or neem tree (Parida, 2002); and *Carica papaya* or melon tree (Ahmad et al ,2011) are all potential anti- dengue plants. These plants have been exposed to efficiently fight the disease. *Artemia salina* toxicity bioassay was used to determine and compare the mortality and LC50 (Lethal Concentration 50) of the roots, stem and leaves of the plants. Treatment concentrations of the plant parts extract were: 10,000ug/mL, 1,000 ug/mL, 100 ug/mL, 10 ug/mL, 1ug/mL and 0.1 ug/mL. Regression of Toxicity Line and Graphical Interpolation Method were used in the calculations of LC50.

Results show that 100% mortality of *Artemia salina* was obtained in the 10,000 µg/mL concentrations of each leaf extract in the five anti-dengue plants. Overall mean percentage mortality of the leaf extract concentration in *Ipomoea batatas* (47.76%), had the highest while *Uncaria tomentosa* (36.67%) being the lowest. Roots and stem extracts in all the five anti-dengue plants revealed no significant differences on the mortality of *Artemia salina*. Considering cytotoxicity of the vegetative organs among all the anti-dengue plants. *Carica papaya* (LC50 21.73 µg/mL), *Ipomoea batatas* (LC50 24.48 µg/mL) and *Azadirachta indica* (LC50 41.03 µg/mL) leaf extracts were found to be the most toxic and comparable as to its effect to *Artemia salina*. Toxicity of stem and roots were found comparable with each other among all the plants. Hence, the leaves considered as the most toxic. Leaves of the said plants can be further explored serving as benchmark on the formulation of possible drugs to prevent and treat dengue.

Keywords: cytotoxicity, LC50, Antidengue plants, *Artemia salina, mortality*

BS - 31

TAXONOMIC AND ECOLOGICAL STUDIES OF VASCULAR PLANTS AT ENTRANCES OF SELECTED CAVES IN POLILLO ISLAND, QUEZON, PHILIPPINES

Annalee S. Hadsall[1,3], Grace Angelique F. Cariño[2],
Ivy Amor F. Lambio[2], Mary Ann O. Cajano[3],
Ariel R. Larona[3] and Ireneo L. Lit, Jr. [2,3]

[1]Plant Biology Division, Institute of Biological Sciences,
[2]Environmental Biology Division, Institute of Biological Sciences, and
[3]UPLB Museum of Natural History, University of the Philippines Los Banos,
College Laguna uplbmnh@yahoo.com.ph; mnh@uplb.edu.ph

Partial results of on-going taxonomic survey and ecological studies of vascular plants found at entrances of caves on Polillo Island, Quezon, Philippines are presented. This is the first study that specifically focuses on cave entrance plants in the Philippines. The limited availability of light in cave environments consequently limits primary productivity therein. Hence, photosynthetic organisms are found mainly at the entrances and to a very limited extent, in the twilight zones of caves. Photographs were taken for documentation and common or familiar plants were identified during field surveys. Samples of uncommon as well as unidentified species were collected, brought and preserved for further study and verification at the Botanical and Forestry Herbaria of the UPLB Museum of Natural History. Species include hoyas, begonias, several aroids, ferns, palms, strangling figs, urticaceous plants, etc. Several are known lithophytes whereas a few are also known as invasive or weedy. The taxonomic list of these plants is very important as baseline information for biodiversity and conservation of Philippine caves.

Keywords: Polillo Island, taxonomic survey, lithophytes, Philippine caves

BS - 32

IN VITRO ROOTING OF *Nepenthes truncata* MACF.

Eufemio T. Rasco Jr.* and <u>Georgianna Kae R. Oguis</u>

University of the Philippines Mindanao, Mintal, Davao City
etrascojr@gmail.com; g.kae.oguis@gmail.com

In vitro grown *Nepenthes truncata* plantlets need a well-developed root system to become hardy enough to withstand changes in the environment during transfer from laboratory to nursery. The effects of rooting media, indole-3-butyric acid (IBA) concentration, method of IBA application and source of plantlets for rooting in vitro grown *N. truncata* plantlets were studied. *N. truncata plantlets* were grown in agar, vermiculite and Silvosa medium (2:1:1 coir: charcoaled rice hull:osmunda fiber). Plantlets grown in vermiculite produced the longest and the most number of roots. The number of roots in vermiculite was also significantly higher than those in agar. Apparently, the Silvosa medium can be used as a substitute for vermiculite, but vermiculite has the advantage of being easier to prepare and more aseptic. Effects of IBA concentrations, 3 mg'"L^{-1}, 6 mg'"L^{-1} and 9 mg'"L^{-1} IBA, on *N. truncata* plantlet tips and bases were investigated after a month. Root count, root length and root color ratings of shoots which were taken from the base and were placed in media supplemented with 9 mg'"L^{-1} IBA were constantly highest. *N. truncata* plantlets grown in vermiculite and supplemented with 9 mg'"L^{-1} were compared to plantlets with cut-ends dipped for 10 mins in 10, 20, 30, 40, 50, 60, 70, 80, 90, 100, 500 and 1000 mg'"L^{-1} IBA prior to growing in IBA-free vermiculite. After a month, root count in all treatments except in plantlets dipped in 50 mg'"L^{-1} were found to be significantly higher than those grown in vermiculite supplemented with 9 mg'"L^{-1} IBA. Dipping was only effective at concentration range: 10 mg'"L^{-1} to 100 mg'"L^{-1}. *N. truncata* plantlets dipped at 40 mg'"L^{-1} IBA prior to growing in vermiculite yielded the most number of roots and the highest root length rating.

Keywords: *Nepenthes truncata* Macf., rooting, indole-3-butyric acid, vermiculite, in vitro

BS - 33

COMPARATIVE ANALYSIS OF THE ELEMENTAL COMPOSITION AND FUNGALL DIVERSITY ALONG THE CALANCAN CAUSEWAY OF STA. CRUZ, MARINDUQUE

Kristina Isabel Toledo[1], Ron Edward Joson[1] and Jocelyn T. Zarate[2*]

[1]College of Arts and Sciences, University of the Philippines Manila,
Ermita, Manila and [2]National Institute of Molecular Biology and Biotechnology,
University of the Philippines Los Banos, Laguna joytzarate@yahoo.com

The Causeway, a 5-kilometer landmass formed by thirty years (1969 to 1996) of mine tailings waste from the operations of the Marcopper Mining Corporation (Marcopper), now dumped at Calancan bay, Sta. Cruz, Marinduque was subjected to soil elemental analysis and fungal diversity evaluation. The objectives of the study were to determine the level of heavy metal contamination in the site across three specific locations: 1) Along the road path; 2) beach front of Calancan bay and 3) inner landmass recently re-vegetated area. X-Ray Fluorescence (XRF) was used to determine elemental concentrations of heavy metals while fungal diversity was analyzed through dilution plating.

After three to seven days incubation in Potato Dextrose Agar (PDA), fungal colonies were counted and re-isolated on PDA plates. Fungal diversity was noted by determining the occurrence of the fungal isolate in other dilutions and sites. PDA blocks and light microscopy were used to identify the isolates. Iron, potassium, titanium, manganese, copper, vanadium and chromium were the elements which exceeded the benchmark concentration tolerable to soil organisms as recommended by the U.S. Environmental Protection Agency (EPA). *Penicillium spp., Verticillium spp., Aspergillus spp., Acremonium curvulum, Trichophyton terrestre, Chrysosporium* spp., *Chrysonilia sitophila* and *Stemphylium spp* were the fungal species isolated from the soil samples. Station 2 (beachfront) had the highest fungal population with 2330 cfu/mL. Station 1 (along road path) had 561 cfu/mL while station 3 (inner land and newly re-vegetated area) had 349 cfu/mL of fungi. Among the elements in the sites, iron and potassium showed negative correlation with the fungal population. Fungal isolates were preserved for further screening trials such as, use as biological control agents and as source of important metabolites.

Keywords: causeway, fungal diversity, heavy metals, x ray fluorescence, marinduque

BS - 34

TESTING A PRELIMINARY METAGENOMIC APPROACH TO SCREEN FOR ALKALINE PROTEASE AND XYLANASE-PRODUCING BACTERIA

Karen G. Rosal and Cynthia T. Hedreyda

National Institute of Molecular Biology and Biotechnology,
University of the Philippines Diliman, Quezon City kgrosal@ymail.com

Metagenomics is an emerging field in biotechnology that allows the genomic analysis of entire communities of microorganisms even before the need for culture of individual bacterial isolates. The objective of this study is to assess the feasibility of metagenomics as a preliminary step in screening for enzyme- producing bacteria. Luria Broth was inoculated with soil samples taken from agricultural, oil-contaminated, forest, and roadside soil as an enrichment step to increase the concentration of DNA template from viable microorganisms. The DNA extract from each sample served as templates for polymerase chain reaction using different primers that target genes for industrially- important enzymes alkaline protease and xylanase. The presence of distinct and intense bands of amplicons in the soil samples were observed, suggesting that bacteria producing such target enzymes could be present in the samples. Sequence analysis revealed that the gene from the different soil samples shows 99% homology with the endo-1,4-beta xylanase gene of two strains of *Bacillus subtillis, Bacillus cereus, Bacillus pumilus and Bacillus amyloliquefaciens*. Furthermore, the gene from the different soil samples also shows 99% homology with the *Bacillus licheniformis* bacillopeptidase gene, an alkaline serine protease. Results reveal that the preliminary metagenomic approach could be useful to provide information as to which samples will be subjected to further conventional screening to isolate enzyme-producing bacteria. Sequence analysis of amplicons could result in the isolation of novel genes exhibiting significant sequence variation from the reported genes.

Keywords: metagenomics, enzymes, alkaline protease, xylanase, PCR

BS - 35

Aedes aegypti LIFE CYCLE, TEMPERATURE EFFECTS, AND DENGUE FEVER-DENGUE HEMORRHAGIC FEVER INCIDENCE: CLIMATE CHANGE IMPACT

Franco G. Teves*, Jing B. Bautista, Edward Laurence Opena, Venessa Mae Bado, and Aubrey Valora Dulay

Molecular Genetics & Microbial Technology Laboratory,
Department of Biological Sciences, College of Science & Mathematics,
MSU-Iligan Institute of Technology, Iligan City franco_teves@yahoo.com

Climate change impact on various aspects of human existence includes worsening scenario of highly infectious vector-borne diseases such as dengue fever and dengue hemorrhagic fever. Providing an evidence-based framework for the interaction of mosquito life cycle, temperature and disease outbreaks is vital for predictive model construction and for disease control.

Hospital reports on dengue fever (DF) and dengue hemorrhagic fever (DHF) for the years 2005 to 2009 were obtained from cooperating hospitals in Iligan City, Philippines. Meteorological data (2005 to 2009) were purchased from the PAGASA Region 10 Station. Simulated environments at different temperatures (36ÚC, 38ÚC, 42ÚC, 44ÚC and 48ÚC) were constructed for larval development studies.

Results show that average minimum temperature has a direct relationship with number of DF and DHF cases. Further, that the amount of rainfall correlates positively with number of DF and DHF cases more than relative humidity. *A. aegypti* life cycle studies reveal that pupae have higher survival rates at 36ÚC and 38ÚC in a four-hour period compared to other temperatures. There was also an apparent shorter life cycle at higher temperatures seen as an adaptive strategy of the mosquito. More significantly, there was an observed deviation from the reported thermal death point of 41ÚC for 1 hour for *A. aegypti* aquatic stages. Pupae survived at 42ÚC for 72 hours and at 44ÚC and 48ÚC for 2 hours.

Therefore, warmer temperatures and increased rainfall coupled with thermal adaptive strategies significantly provide an efficient synergistic mechanism for virus transmission in the human population.

Keywords: *Aedes aegypti*, dengue fever, dengue hemorrhagic fever, climate change, adaptive strategies

BS - 36

VIABILITY OF IMMOBILIZED LACTIC ACID BACTERIA IN DIFFERENT CULTURE DILUENTS AND VARIOUS COCONUT-BASED FOOD PRODUCTS

Teresita J. Ramirez*, Jennifer D. Saguibo,
Arsenia B. Sapin, Susan A. Sedano and Mary Ann T. Tavanlar

National Institute of Molecular Biology and Biotechnology (BIOTECH),
UP. Los Baños, College, Laguna ramirez.tj@gmail.com

There is considerable interest in providing physical barrier against adverse environmental conditions to protect probiotic bacteria and improve their survival in food products. Immobilized cell technology applied to lactic acid bacteria (LAB) and probiotic cultures offers many advantages over the use of free cells.

This study investigates the use of immobilization technique using sodium alginate to determine the viability of *Bifidobacterium* strain. Cells were immobilized in different concentrations of sodium alginate with bead formation accomplished by dropwise addition of the alginate cell mixture into $CaCl_2$. Formed beads were suspended in different solutions that include coconut water, formulated coconut beverage, peptone water (0.1%) and saline solution (0.85%). They were also incorporated in reconstituted skim milk (RSM) and coconut-based food products such as ice cream and yoghurt. Viability of the probiotic strain was monitored with time.

The viability of LAB was maintained in beads, regardless of the concentration of sodium alginate used. The survival of immobilized LAB suspended in saline solution and peptone water was lower compared with the immobilized LAB suspended in coconut water and formulated coconut beverage. After two weeks storage at refrigerated temperature, there was a difference of around 2.5 log per bead in the total LAB count between beads suspended in coconut water and in the other diluents.

With incorporation into various food products, the viability of immobilized LAB was maintained at around 6 log per bead up to three weeks in coconut beverage and up to one week in yoghurt. However, this was not achieved when the beads were incorporated into RSM, a standard culture medium for LAB, and into coconut ice cream with a reduction in population to 4 log per bead after a week of storage. These preliminary results showed the potential of incorporating immobilized LABs in formulated coconut-based products.

Keywords: immobilized cells, lactic acid bacteria, coconut-based food products, *Bifidobacterium*, sodium alginate

BS - 37

EFFECTS OF MYKOVAM, BioN AND INORGANIC FERTILIZERS ON GROWTH, YIELD AND NUTRIENT STATUS OF SWEET SORGHUM [*Sorghum bicolor* (L.) MOENCH]

Nelly S. Aggangan[1]*, Miguel Luigi L. Calayag[2]
Austin Japeth R. Salvan[2], John Carlo Velandres[2]

[1]National Institute of Molecular Biology and Biotechnology, University of the Philippines Los Banos, Laguna nelly_aggangan@yahoo.com;
[2]University of the Philippines Rural High School, Paciano Rizal, Bay, Laguna

Sorghum is an important crop to dryland farmers and can be utilized for feeds and biofuel. This study was conducted to determine the effects of biofertilizers Mykovam and Bio-N on growth, yield and nutrient uptake of Sweet Sorghum. The efficiency of biofertilizers was also compared with that of chemical fertilizers. Sweet sorghum was applied with biofertilizers Mykovam (containing mycorrhizal fungi) or BioN (containing nitrogen fixing bacteria), with or without half or full rate of the recommended rate of chemical fertilizer (RRC = 60-60-60 NPK). The experiment was conducted at the Demonstration farm of BIOTECH, UP Los Baños following a RCBD with four blocks. Growth was monitored periodically and plant samples were harvested at 102 and 154 days. Fresh and dry biomass, juice from the stalk, brix level and plant N and P status were measured. Results show that Mykovam+BioN promoted the largest root collar diameter. Mykovam alone and BioN+Mykovam promoted the highest fresh and dry weights of leaves, roots and cane, cumulatively, the highest total biomass. Mykovam treated sorghum had the highest total N and P concentrations and consequently, the highest N and P uptake (1,707 g N/plant and 269 g P/plant) and the lowest was obtained from the control plants (999g N/plant and 214 g P/plant). The highest fruit yield was obtained from plants treated with RRC (16.10 g/cane) comparable with BioN+Mykovam+1/2RRC (13.24 g/cane) and the lowest was the control (3.97 g/cane). In terms of brix level at 102 days, RRC gave the highest (9.36) comparable with BioN+Mykovam+1/2RRC treated plants (8.80) and the lowest was (6.20). At 145 days, Mykovam+BioN+½RRC treated plants had the highest (14.52) brix and the lowest was the control (13.72). The high nutrient status and brix level due to Mykovam+BioN+½RRC suggests that sweet sorghum can be a feasible source of feed and biofuel.

Keywords: biofertilizers, sweet sorghum, Mykovam, BioN, mycorrhizal fungi, nitrogen fixing bacteria

BS - 38

PHOSPHORUS AND MYCORRHIZAL FUNGI AFFECT GROWTH, NUTRIENT AND HEAVY METAL ACCUMULATION IN TRANSGENIC POPLAR

Nelly S. Aggangan[1]*, Sim-Hee Han[2], Young-Im Choi[2], Eun-Woon Noh[2] and Yongsuk Lee[3]

[1]National Institute of Molecular Biology and Biotechnology, University of the Philippines Los Baños, Laguna nelly_aggangan@yahoo.com;
[2]Korea Forest Research Institute, Suwon, Korea 441-350;
[3]Pohang University of Science and Technology, Pohang, Korea

Heavy metal transgenic poplars had been developed in Korea for the immediate rehabilitation of mine tailing sites. Phosphorus fertilizers are applied to counteract the toxic effects of heavy metals on plant growth and survival in mine tailings. However, the amount of P fertilizer to be applied that conforms with the beneficial effects of mycorrhizal inoculation is not known. This experiment was conducted to determine the growth, nutrient status and translocation of heavy metals in tissue cultured transgenic *Populus alba x P. glandulosa* uninoculated or inoculated with a mixture of ectomycorrhizal (ECM) fungi *Pisolithus tinctorius, Paxillus involutus* and *Amanita pantherina* during *ex vitro*. Treated plantlets were transplanted in cups filled with 50g autoclaved peat perlite vermiculite (PPV) medium and incubated under growth room conditions. One month later, the seedlings were transferred in pots filled with150g PPV amended with 0, 8 or 16 mg P $(CaH_2PO_4)_2$ kg soil^{-1} and nil or mixture of lead (Pb), zinc Zn) and arsenic (As). Pb, Zn and As are the top heavy metal contaminants in a closed mine tailing area in Bonghwa, South Korea where rehabilitation activities has been concentrated for the last five years. After four months in a glasshouse, total plant dry weight of non-mycorrhizal plants grown at P8 and P16 was comparable with mycorrhizal plants grown at P0. Heavy metals amendment reduced plant growth and biomass but increased concentrations of all nutrients measured. Except for N, Zn and Pb, mycorrhizal plants had higher P, Ca, Mg, Na, and As than non-mycorrhizal counterpart. P16 increased plant growth and N, P, Mg and Na concentrations but reduced Zn and Pb. Highest Zn, Pb and As were in the fine roots and lowest in the stem of mycorrhizal plants. Highest (34%) mycorrhizal colonization was at P16 and reduced to 22% in the presence of heavy metals.

Keywords: mine tailing sites, *Populus*, ectomycorrhizal fungi, *Pisolithus, Paxillus, Amanita*

BS - 39

DNA BARCODING OF FRUIT FLIES *Bactrocera occipitalis* (Bezzi) AND *Bactrocera philippinensis* DREW AND HANCOCK (DIPTERA:TEPHRITIDAE) FROM CAVITE AND DAVAO DEL NORTE USING THE 5' REGION OF THE MITOCHONDRIAL CYTOCHROME C OXIDASE I (*coxI*) GENE

Michael Leonardo C. Delomen* and Merlyn S. Mendioro

Genetics and Molecular Biology Division, Institute of Biological Sciences,
College of Arts and Sciences, University of the Philippines Los Banos, Laguna

Analysis of the 5' region of the mitochondrial cytochrome oxidase I gene was done to compare the pest fruit flies *Bactrocera occipitalis* (Bezzi) and *Bactrocera philippinensis* Drew and Hancock. Adult fruit flies and larvae-infested dropped mango fruits were collected from selected orchards at Cavite and Davao del Note. Collected adults were immediately identified while larvae from infested fruits were reared to adulthood then identified. Identification was done thru the visual analysis of six diagnostic morphological characters originally described by White and Hancock (1997) and utilized by Iwahashi (1999) to differentiate between the two species.

Genomic DNA was extracted and mitochondrial *coxI* gene was amplified using custom primers. PCR products were verified thru agarose gel electrophoresis and shipped to Macrogen Inc, South Korea for sequencing. Consensus sequences were derived and analyzed using the sequence analysis softwares Clustal W2 and MEGA 4.

The generated phylogenetic trees showed that there were no significant sequence differences between *B. occipitalis* and *B. philippinensis*. Divergence values were from zero to 0.0018%; values obtained were significantly lower than the set standard of 0.5% to establish species delineation. Analysis of the 5' region of the mitochondrial *coxI* gene did not show species delineation between *B. occipitalis* and *B. philippinensis*. It is recommended that further analysis be done on more samples from other locations in the Philippines.

Keywords: DNA barcoding, cytochrome oxidase 1, *Bactrocera occipitalis* (Bezzi), *Bactrocera philippinensis* Drew and Hancock, species delineation

BS - 40

DEVELOPMENT OF PROBIOTIC VEGETABLE JUICE FORTIFIED WITH *Lactobacillus plantarum* BS

Jennifer D. Saguibo and Francisco B. Elegado*

National Institute of Molecular Biology and Biotechnology,
University of the Philippines Los Baños, Laguna fbelegado@hotmail.com

Preference for raw and minimally-processed foods and probiotic products is increasing due to obvious nutritional and non-nutritional health benefits. The country is endowed with local plant and microbial resources that can be developed into functional food products.

Various formulations of indigenous Philippine vegetables/herbs namely *alugbati* (*Bacella rubra*), *saluyot* (*Corchorus olitorius*), *okra* (*Abelmoschus esculentus*), *malunggay* (*Moringa oleifera*) and avocado (*Persea Americana*) leaf extracts, strained, pasteurized, flavored with *calamansi* (*Citrofortunella microcarpa*) or lemon grass (*Andropogon citrates*), and fortified with *Lactobacillus plantarum* BS were subjected to microbial stability and sensory testing. The selected formulation with 2% (w/v) air-dried vegetable/herb in different proportions flavored with *calamansi* and lemon grass were stored at refrigerated temperature for three months. Both flavored vegetable juice maintained a pH range of 4 - 4.5 after 14 days in contrast with the control (without flavor and *Lb. plantarum* BS only) that initially decreased to pH 4.0 at day 10 but began to elevate at day 14. *Calamansi*-flavored vegetable juice maintained a higher lactic acid bacterial count of 10^7 cfu/ml, up to 70 days of storage. In contrast, the lemon grass flavored vegetable juice maintained the lactic acid bacterial count of 10^7 cfu/ml only until day 14 but declined thereafter. However, sensory evaluation revealed that lemon grass-flavored vegetable juice was more preferred than *calamansi* flavored. Its flavor/taste, aroma and general acceptability were also significantly different at p=0.05 compared with the control after six days. This result suggests the possibility of developing vegetable/herb beverage fortified with probiotic lactic acid bacteria with enhanced health benefits.

Keywords: probiotic juice, lactic acid bacteria, *Lactobacillus plantarum*, *alugbati*, *saluyot*, *avocado*, *okra*, *malunggay*, *calamansi*, lemon grass

BS - 41

SYNERGISTIC HYPOGLYCEMIC EFFECT OF *Lactobacillus pentosus* 3G3 AND *BANABA* (*Lagerstroemia speciosa*) ON DIABETIC-INDUCED SWISS WEBSTER MICE

Jennifer D. Saguibo and Francisco B. Elegado*

National Institute of Molecular Biology and Biotechnology, University of the Philippines Los Baños, College, Laguna fbelegado@hotmail.com

Intake of herbal medicines may lead to the accumulation of toxic and anti-nutritional factors. Studies have shown that microbial action by generally regarded as safe (GRAS) and probiotic bacteria may help degrade these factors into non-insidous forms.

The hypoglycemic effect of *Lactobacillus pentosus* 3G3 pure cells on diabetic-induced male Swiss-Webster mice was tested. A saline suspension of 10^8-10^9 CFU/ml pure cells was orally administered to mice previously fed with 20 ml condensed milk and 30 ml melted butter in 50 ml sterile distilled water to induce high blood glucose levels. Results showed that *L. pentosus* 3G3 significantly decreased blood glucose level and body weight after two weeks feeding period.

In a previous study, *Lb. pentosus* 3G3 was found resistant to any antimicrobial effect of *banaba* (*Lagerstroemia speciosa*) leaf extract, reported to contain anti-diabetic compound. In this study, *Lb. pentosus* 3G3 was inoculated in 4% (w/v) *banaba* leaf and 5% (w/v) muscovado decoction, fermented for overnight, cells adjusted to 8.2×10^9 CFU/ml, and administered orally at 30 ml/kg body weight to the mice with elevated blood glucose level induced by injecting hydrochlorothiazide intraperitonially. Glibenclamide and distilled water were also administered as positive control and negative control, respectively. Single ingestion of the *banaba* decoction, fermented with *Lb. pentosus* 3G3, showed comparable potency in decreasing blood glucose level than the unfermented *banaba* decoction, but not significantly different at $p=0.05$ after 240 min monitoring of blood glucose level.

Prolonged feeding period and monitoring is being done in order to determine the long term effects of *Lb. pentosus* 3G3, mixed with *banaba* leaf decoction, in lowering blood sugar levels on mice and other positive effects of the probiotic lactic acid bacteria, such as minimizing the anti-nutritional effects of *banaba*.

Keywords: diabetes, *Lactobacillus pentosus*, probiotic lactic acid bacteria, *banaba*, *Lagerstroemia speciosa*, hypoglycemic effect

BS - 42

RANDOM AMPLIFIED POLYMORPHIC DNA – POLYMERASE CHAIN REACTION (RAPD-PCR) PRIMER DESIGN FOR THE DETECTION OF *Listeria monocytogenes*

Maria Teresa M. Perez[1], Daryl Ann Steffanie G. Torralba[2], Joel C. Mendoza[2], and Francisco B. Elegado[1*]

[1]National Institute of Molecular Biology and Biotechnology,
[2]Institute of Biological Sciences, University of the Philippines
Los Baños, Laguna fbelegado@hotmail.com

Listeria monocytogenes is a major foodborne pathogen recognized worldwide by food industries and public safety institutions because of its high fatality rate and various complications such as acute diarrhea, meningoencephalitis, septicaemia and abortion. USFDA imposes "zero tolerance" regulation for *L. monocytogenes* in raw and ready-to-eat foods. The National Meat Inspection Services of the Department of Agriculture is strictly monitoring it in processed meat.

This study reports on the design of primers through hybridization, cloning and sequencing of RAPD–PCR amplification products identified to be specific for *L. monocytogenes* reference strains (Lm2, Lm3 and Lm4) against closely related isolates: *Listeria* spp. and *Enterococcus* strains isolated from different food samples at the Food and Feed Laboratory, BIOTECH, UPLB.

Gene amplification using RAPD primer 066 resulted in a potential specific 1.2 kb marker. Molecular cloning of the isolated RAPD marker for *L. monocytogenes* was done to facilitate sequencing of the amplicon. Twenty-three presumptive positive clones were subjected to plasmid DNA extraction and restriction enzyme analysis to determine the presence of the target insert fragment. Two plasmid clones confirmed the presence of the target insert fragment through hybridization. The plasmid clone (5a1) was sequenced using the universal primers, M13. Multiple sequence alignment analysis of the 950-base marker sequence using BLAST-N revealed 100% homology with *L. monocytogenes*. The primers designed using the *Primer3 Plus* design software had good specificity with acceptable melting temperatures and GC content except that of possible formation of secondary structures.

Keywords: *Listeria monocytogenes*, RAPD-PCR, primer design, foodborne pathogen detection

BS - 43

CONSTRUCTION OF A PLANT EXPRESSION VECTOR CONTAINING THE REPLICATION INITIATION PROTEIN (REP) GENE OF THE ABACA BUNCHY TOP VIRUS (ABTV)

Patrick Louie G. Lipio, Marylette B. Roa, Kristine Ace F. Quirante, Mary Ann Robelle L. Polido and Vermando M. Aquino*

Plant Molecular Biology and Plant Virology Laboratory, National Institute of Molecular Biology and Biotechnology, College of Science, University of the Philippines Diliman, Quezon City vmaquino@mbb.upd.edu.ph*

The abaca industry in the Philippines produces an average of 68,962 metric tons per year of fiber utilized on many industries generating an income of USD 80.8M on exports alone. However viral diseases like the Abaca bunchy top virus (ABTV) cause damaging effects to the plant. A strategy to control ABTV is through pathogen derived resistance (PDR) wherein a virus component is introduced to the plant and its expression could mediate resistance to the virus. The replication initiation protein (Rep) gene, an ABTV DNA virus component is a possible candidate for PDR. For this study, the Rep gene was isolated and cloned to a plant expression vector. Primers were designed to target the Rep gene and appended with restriction endonuclease sites *Bgl*II and *Bst*EII for subsequent cloning. Amplification using the designed Rep primers and genomic DNA from ABTV infected abaca plants from Leyte yielded an approximately 900bp product. This was further subcloned to pCR2.1® TOPO® vector for sequencing. Sequencing results showed 99% similarity to the Banana bunchy top virus (BBTV) Rep gene. The pCR2.1® TOPO®-Rep construct was digested with *Bgl*II and *Bst*EII as well as the plant expression vector pCAMBIA1302. The ligated pCAMBIA1302-Rep gene construct was transformed in DH5â cells for propagation and screening. Screening using CaMV35S and NosTer pCAMBIA vector specific primers showed an approximately 1100bp product. Further sequencing is necessary to confirm if the Rep gene is inserted to pCAMBIA1302. This construct would be delivered to abaca plant cells for assessment of its expression and at most conferring ABTV resistance to the plant.

Keywords: abaca bunchy top virus (ABTV), pathogen derived resistance (PDR), replication initiation protein (Rep) gene, cloning, pCR2.1® TOPO®, pCAMBIA1302

BS - 44

SEQUENCE ANALYSIS OF THE KAPPA-CASEIN GENE IN WATER BUFFALO *(Bubalus bubalis)*

Jesus Rommel Herrera*, Amie Villanueva and Ester Flores

Molecular Genetics Laboratory, Philippine Carabao Center,
Science City of Munoz, Nueva Ecija jrvh4171@yahoo.com

The aim of the present study was to determine the sequence of the coding region of the kappa-casein gene of the water buffalo and detect potential exonic polymorphisms that are associated with milk production traits. Twenty-five genomic deoxyribonucleic acid (DNA) samples of unrelated buffalo cows coming from extremes of the population (ten high, five medium and ten low milk producers) were used in the study. Since the buffalo genome has not yet been sequenced, primers used to amplify the coding regions were based on the gene sequence of cattle. Polymerase chain reaction (PCR) amplification yielded products of similar size to that of cattle. Direct sequencing results revealed that the kappa-casein gene sequence of the water buffalo was 96% similar to that of cattle. Two potential polymorphisms were also found in nucleotides 467 and 471 located at the fourth exon. The cytosine 'C' to thymine 'T' change of nucleotide 467 resulted to an amino acid change from threonine (T)<ACC> to isoleucine (I)<ATC> at codon 156. Preliminary association studies revealed that choosing animals with 'CC' and 'CT' genotypes will be advantageous since the milk yield at 305 days (MY305D), fat yield at 305 days (FY305D) and protein yield at 305 days (PY305D) are higher ($P < 0.001$) compared with animals carrying the 'TT' genotypes. The methodology above can be used to determine the coding sequence and potential polymorphisms of other genes not only of the bubaline, but also of other ruminant species whose genome has not yet been sequenced such as the swamp buffalo, sheep, goat, anoa, cape buffalo and deer.

Keywords: water buffalo, milk production, kappa-casein, sequence analysis, polymorphism

BS - 45

MULTIGENE-LOCUS cpDNA BAYESIAN FRAMEWORKS IN RESOLVING CONFLICTS WITHIN PHILIPPINE ENDEMIC *Gardenia* ELLIS (RUBIACEAE): IMPLICATIONS TO TAXONOMY, BIODIVERSITY AND DRUG DEVELOPMENT

Jayson G. Chavez[1*] and Grecebio Jonathan D. Alejandro[1,2]

[1]The Graduate School; [2]College of Science and Research Center for the Natural Sciences, University of Santo Tomas, España, Manila jayson.chavez@gmail.com

The Philippine *Gardenia* species form a highly heterogeneous assemblage, leading to several taxonomic transfers to other Rubiaceous genera (*Ceriscoides, Discospermum, Fagerlindia, Sulitia, Vidalasia* & *Villaria*). Inspite of these taxonomic rearrangements, three remaining Philippine endemic *Gardenia* species (*G. lagunensis* Merr., *G. merrillii* Elmer & *G. negrosensis* Merr.) still exhibit disparate features against the current circumscription of the genus and remain unresolved. In order to evaluate their true generic affinities with more certitude, we conducted separate and combined Bayesian Inference (BI) of the *rps16* intron and *trnT-F* region sequences of cpDNA. The majority rule consensus tree of the combined dataset is more congruent with the *rps16* intron tree but with much higher Posterior Probabilities (PP). The four sampled Philippine *Gardenia* species including *G. elata*, appeared in two different groups of the tribe Gardenieae. *Gardenia elata* grouped within the *Gardenia* clade with high support (PP=1.00), while *G. lagunensis*, *G. merrillii* and *G. negrosensis* nested within a clade with *Rothmannia annae* and *R. capensis* with strong support (PP=0.96). These molecular findings are congruent with morphology such as the 3:3 nodal pattern of leaves, absence of domatia, bilobed stigmas, 2-locular ovaries, globular indehiscent fruits and seeds that are embedded in a pulp forming a single mass. Therefore, we recognized three novel combinations under *Rothmannia* Thunb., which established for the first time the genus to the archipelago. In relation to this taxonomic breakthrough, implications on their conservation status and possible medicinal use are presented.

Keywords: bayesian inference, cpDNA, *Gardenia*, Philippine endemic, *Rothmannia*, *rps16* intron, *trnT-F* region

BS - 46

ANTIMICROBIAL ACTIVITY OF *PARAMERIA LAEVIGATA* (A.L. JUSS.) MOLDENKE AND *VOACANGA GLOBOSA* (BLANCO) MERR. EXTRACTS

Pierangeli G. Vital[1,2] and Windell L. Rivera[1,2]*

[1]Molecular Protozoology Laboratory, Natural Science Research Institute and
[2]Institute of Biology, College of Science, University of the Philippines
Diliman, Quezon City wlrivera@science.upd.edu.ph

Ethanol extracts of leaves of *Parameria laevigata* (A.L. Juss) Moldenke and *Voacanga globosa* (Blanco) Merr. were examined for their antibacterial, antifungal, and antiprotozoal properties. *P. laevigata* and *V. globosa* extracts were tested against bacteria, namely, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhimurium*, *Bacillus cereus*, *Staphylococcus aureus*, *Micrococcus luteus*; fungus, *Candida albicans*; and protozoa, *Entamoeba histolytica*, *Trichomonas vaginalis*. Potential antimicrobial compounds were obtained through solvent extraction and rotary evaporation. To determine the antibacterial and antifungal properties of the extracts, disc diffusion assay was performed. *V. globosa* extracts revealed antibacterial and antifungal activities, inhibiting the growth of *B. cereus*, *M. luteus*, *P. aeruginosa*, *S. aureus*, *S. typhimurium*, and *C. albicans*. On the other hand, extract of *V. globosa* showed no antibacterial and antifungal activity. Lastly, to determine the antiprotozoal activity of the extracts, growth curve analysis and antiprotozoal assay were performed. The extracts were examined against *Trichomonas vaginalis* and *Entamoeba histolytica*. Results showed that both plant extracts can inhibit parasites. Thus, these plant extracts can possibly be used to produce alternative forms of antimicrobials.

Keywords: *Parameria laevigata*, *Voacanga globosa*; antibacterial, antifungal, antiprotozoal activity

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DETECTION OF WHITE SPOT VIRUS IN *Penaeus monodon* AND *Metapenaeus dalli* FROM HAGONOY, BULACAN

Rod Russel R. Alenton[2]*, Anna Dominique M. Aniag[3], Marie Veronica G. Bulaong[3], Geormae Anne K. Reblando[3], Annie Grace V. Urayenza[3], Jean Kathleen V. Yasuda[3], and Mary Beth B. Maningas[1,2,3]

[1]Research Cluster for the Natural and Applied Sciences, [2]Graduate School, [3]Department of Biological Sciences, University of Santo Tomas, España, Manila russel.alenton@gmail.com

Shrimp aquaculture is an integral source of revenue in developing tropical countries of Southeast Asia such as the Philippines. In the early 1990s, the country ranked 3rd in the shrimp international market, however the industry slid back to the 13th as it is presently plagued with outbreaks of diseases which causes massive decrease of production. Among these diseases, White Spot Virus (WSV) remains one of the most potent and infectious for most known species of cultivated penaeid shrimps. It is in this light that the presence of the White Spot Virus (WSV) in *Penaeus monodon* and *Metapenaeus dalli,* both species were collected in three commercial ponds in Hagonoy, Bulacan. In every sampling site, five shrimps of each species were collected, and from each shrimp tissue samples from gills, hemolymph, heart and hepatopancreas were obtained. DNA were then extracted from each organ and were used in the detection through Polymerase Chain Reaction (PCR) amplification. Elongation factor 1-alpha (EF1-α), primers was used to check for DNA integrity and as an internal control . Only the hemocyte samples yielded positive among all the tissues tested. Hence, hemocyte was utilized in all the 30 samples. Twenty percent (20%) of the sampled tissues of *P. Monodon* shrimps were found to be positive. However, *M. dalli* shrimps from the same sampling sites were all found negative for WSV. These results, coupled with the fact that both species were collected from the same pond, suggest that *M. dalli* is more resistant to WSV compared to *P. monodon.*

Keywords: *Metapenaeus dalli, Penaeus monodon,* white spot virus, shrimp polyculture, polymerase chain reaction, virus detection

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IMMUNOMODULATORY EFFECTS OF DRIED *Curcuma longa* POWDER ON THE EXPRESSION OF CRUSTIN AND LYSOZYME IN *Macrobrachium rosenbergii dacqueti*

Jennifer R. Alambra*[1,2], **Rod Russel R. Alcaton**[1],
Pia Clarisse R. Gulpeo[1], **Christine L. Mecenas**[1], **Abigail P. Miranda**[1],
Rey C. Thomas[1], **Maden Krista S. Velando**[1], **Lawrence D. Vitug**[1],
and Mary Beth B. Maningas [1, 2, 3]

[1]The Graduate School, [2]Research Cluster for the Natural and Applied Sciences,
[3]Department of Biological Sciences, College of Science
University of Santo Tomas, España, Manila

Turmeric, a derivative of the plant *Curcuma longa*, exhibits several pharmacological properties and its extracts were found to have a high potential to inhibit some pathogenic bacteria of shrimp and chicken. In this study, eighty (80) *Macrobrachium rosenbergii dacqueti* juvenile shrimps were used for the entire experiment. Purchased turmeric was properly identified at the UST Herbarium. The turmeric was dried and pulverized; and its powder was incorporated into commercial feeds. The shrimps were divided into three groups, D0, D1 and D7. D0 were fed with commercial feeds while D1 and D7 were fed with the enhanced shrimp feeds for one (1) and seven (7) days respectively. These were challenged with *Vibrio alginolyticus*. Relevant tissues were dissected from shrimp samples and total RNA was isolated, quantified by UV spectrophotometry and synthesized into cDNAs. RT-PCR was performed using crustin and lysozyme for target genes and EF-1α as standard control. Quality and quantity of RNA was assessed by measuring the A_{260}/A_{280} ratio and by agarose gel electrophoresis. An increasing intensity of crustin and lysozyme PCR bands relative to the duration of feeding was observed suggesting that shrimps fed with enhanced feeds after bacterial challenge had an induced expression of the mentioned antimicrobial peptides (AMPs). A remarkable increase in the expression of the two AMPs was also observed from D0 to D1 but not from D1 to D7. The results suggest that turmeric powder enhances the expression of these two peptides, but its rate of effectivity is yet to be studied.

Keywords: Shrimp biotechnology, *Curcuma longa*, turmeric, *Macrobrachium rosenbergii dacqueti*, crustin, lysozyme

BS - 49

UNIQUE NUCLEOTIDE DIFFERENCE OF TWO PUTATIVE Ty1/COPIA-LIKE LONG TERMINAL REPEAT RETROTRANSPOSONS OF ABACA (*Musa textilis* Nee)

Orlex B. Yllano*[1,2,3], **Anton Lalusin**[1], **Sheryl D. Castro**[1], **Antonio C. Laurena**[1,2], **and Evelyn Mae Tecson-Mendoza**[1,2]

[1]Institute of Plant Breeding, Crop Science Cluster, College of Agriculture, and
[2]Molecular Biology and Biotechnology Program, University of the Philippines,
Los Banos, College, Laguna, [3]Biology Department, College of Sciences and
Technology, Adventist University of the Philippines, Puting Kahoy,
Silang, Cavite orlex.b.yllano@up.edu.ph

Retrotransposons or transposons through RNA intermediates are ubiquitous mobile genetic elements that can amplify themselves in the eukaryotic genomes. It harbors regulatory signals that are responsible for chromosomal rearrangements, fragmental gene movements, alteration of gene expression and genome evolution. In this study, the putative abaca retrotransposons were cloned, verified, sequenced and characterized. The resulting PCR products were cloned using degenerate primers. Restriction digestion of the purified recombinant plasmid using EcoRI revealed a distinct band (~950 bases). Sequence analysis revealed an 868-base perfect alignment insert. Comparing and aligning the two sequences revealed a nucleotide substitution at 779 from T in Retroclone1 to C in Retroclone2. Using sequence alignment and phylogenetic analysis, the cloned putative retrotransposons of *M. textilis* grouped with Ty1-copia retrotransposon of *M. ABB*, *M. AAB* and *M. acuminata*. High homology to known retrotransposon protein of *Oryza sativa*, putative polyprotein of *Oryza australiensis*, gag-pol of *Zea mays*, integrase core domain of *O. sativa* and reverse transcriptase was noted using Blastx. This result supports the generalized structure of retrotransposon which is characterized with gag, pol and env domains in between the long terminal ends (LTRs). To our knowledge, this is the first report on the cloning and sequence analysis of putative abaca retrotransposon. These results provide insight in understanding the genome composition of the Philippine's indigenous fiber crop — abaca.

Keywords: retrotransposons, long terminal repeats, cloning, single nucleotide polymorphism, musa

BS - 50

A CANDIDATE GENE SEQUENCE FOR DOWNY MILDEW RESISTANCE IN MAIZE

Hayde F. Galvez, Eden Jane U. Tongson, Roanne R. Ripalda*, Alma O. Canama, Arma Kristal B. Malijan, and Desiree M. Hautea

Institute of Plant Breeding, Crop Science Cluster, College of Agriculture,
University of the Philippines Los Baños, Laguna rrripalda@gmail.com

We report a putative resistance gene ortholog for resistance to downy mildew caused by *Peronosclerospora philippinensis* Weston (Shaw) in Philippine maize. We designed two gene-specific markers based on sequence information of the resistance gene analog, *srga3*. BLAST analysis of the *srga3* amplicon showed 39 hits against the maize rust resistance protein *rp3-1* gene with a maximum score of 81.3 and 78% identity. The gene-specific markers were tested for co-segregation with the *srga3*-quantitative trait loci (QTL) and downy mildew resistance phenotype on BC_1F_1 mapping population consisting of 121 individuals. These will be used in map-based isolation of the downy mildew resistance gene in maize and marker-assisted selection towards the development of downy mildew resistant maize varieties.

Keywords: downy mildew, QTL, resistance gene analog, gene-specific markers, maize

BS - 51

YIELD EQUATION AND BIOMASS EXPANSION FACTOR FOR ABOVE GROUND CARBON DENSITY ESTIMATION OF SMALLHOLDER TREE PLANTATIONS IN NORTHERN MINDANAO

Richmund A. Palma*

Institute of Agriculture, Misamis Oriental State College of Agriculture
and Technology, Claveria, Misamis Oriental richmundp@yahoo.com

Regression equations for the estimation of tree volume and biomass expansion factor (BEF) in a smallholder *A. mangium* plantation were developed based on direct measurements of trees in Northern Mindanao, Philippines. *A. mangium* is an important smallholder tree plantation species and commodity for wood industry in the region and the country. Sound and effective management of smallholder tree plantation for wood production and carbon sequestration necessitates information on yield. The study was designed to develop reliable yield prediction equation and BEF according to various ages, site qualities and spacing. Fifty temporary sample plots were established in Claveria, Misamis Oriental with a total of 3,910 trees measured. The study had generated two equations: 1) yield prediction equation ($\ln YIELD = -4.250101 + 2.017218*\ln AGE + 1.079861*\ln SI + .2352149*\ln SP$); and, 2) BEF ($BEF = 1.693517*.9938733^{VB}$). The yield prediction equation can be employed as a basis for efficient management and development of smallholder tree plantations. The tree farmers could make use of yield tables in determining the volume of wood that can be harvested, economic rotation, cut schedule, plantation development schedule and business projection. The yield equation and BEF can also be used to predict above ground biomass carbon density for smallholder *A. mangium* plantation. The equations have been developed for plantations located in Claveria, Misamis Oriental particularly those with ages ranging from 3 to 18 years old, site index ranges from 12 to 25 meters and spacing of 2m x 2m, 1m x 4m, 2m x 3m, 1m x 6m, 2m x 4m, 3m x 3m, 3m x 4m, 2m x 6m and 1m x 8m. The equations were applicable outside the study area but careful validation should be done to ensure effective *A. mangium* tree plantation establishment and management.

Keywords: biomass, mangium, yield, BEF, regression, smallholder

BS - 52

TERRESTRIAL ARTHROPODS IN SELECTED CAVES OF POLILLO ISLAND, QUEZON PHILIPPINES

Ireneo L. Lit, Jr.[1,2], Sheryl A. Yap[1], Leonila A. Corpuz-Raros[1], Aimee Lynn A. Barrion-Dupo[1,2], John Mark A. Encinares[2], Veverle dA. San Juan[1], Rexele Jean F. Galvan[2], Phillip A. Alviola[1], Orlando L. Eusebio[1], Ariel R. Larona[1], Edison A. Cosico[1] and Eduardo G. Eres[1]

[1]UPLB Museum of Natural History and [2]Environmental Biology Division,
Institute of Biological Sciences, University of the Philippines
Los Banos, Laguna uplbmnh@yahoo.com.ph; mnh@uplb.edu.ph

The first set of results from the series of fieldwork focusing on the taxonomy and ecology of terrestrial arthropods found inside selected caves of Polillo Island, Quezon, Philippines are reported. The results presented and the work involved is largely pioneering as most of the existing data on cave biology have always been limited to bats, swiftlets and other vertebrates. Philippine cave arthropods from Polillo Island constitute new faunal records and probably new species that belong to various classes, orders and families. They include springtails (Collembola), cockroaches (Blattodea), ants and fairy flies (Hymenoptera), parasitic, predatory and saprophytic mites (Acari), carabid beetles and water pennies (Coleoptera), guano moths (Lepidoptera), bat flies and midges (Diptera), earwigs (Dermaptera), millipedes (Diplopoda), centipedes (Chilopoda), bat fleas (Siphonaptera), spiders (Araneae), vinegaroons (Uropygi), scorpions (Scorpionida), pseudoscorpions (Chelonethida), etc. The unique conditions and microhabitats inside cave ecosystems (most importantly the very limited availability of light, high moisture and carbon dioxide, and generally nutrient-poor underground setting) have been selected by some organisms to evolve with strikingly unique adaptations to this kind of extreme environment. The number and nature of groups and species encountered further attest to the great diversity of terrestrial arthropods on Polillo Island and in the entire Philippine archipelago. These sets of data on cave biodiversity also provide bases for protection and conservation of Philippine caves.

Keywords: terrestrial arthropods, microhabitats, cave ecosystems, cave biodiversity

BS - 53

EFFECTS OF AGROCHEMICALS ON ENDOMYCORRHIZAL DYNAMICS FOR A SUSTAINABLE SOIL IN TAMPAKAN MOUNTAIN ECOSYSTEM

Metchie Gay R. Arnaiz, Nelson M. Pampolina* and Jose Sebua*

Department of Forest Biological Sciences, College of Forestry
and Natural Resources, University of the Philippines
Los Baños, Laguna metchie_arnaiz@yahoo.com

Agrochemicals used in upland farms and nurseries in Quezon mountain range from Tampakan, South Cotabato were identified using Key Informant Interview and investigated to evaluate their effects on the dynamics of endomycorrhizal fungi. Endomycorrhizas are important link in plant nutrition and health. Population of endomycorrhizal fungi was assessed to determine soil productivity and sustainability in the area through wet-sieving decanting technique for isolation, characterization, and examination of endomycorrhizal colonization. About 95% of upland farmers extensively used fertilizers (Complete, Urea), herbicides (Power, Atrazine), fungicides (Ridomil, Daconil), and pesticides (Malathion, Lannate) for agricultural, agroforest and forest crops. Endomycorrhizas identified were *Glomus*, *Gigaspora*, and *Entrophospora*, where *Glomus sp.* mostly dominating disturbed grasslands and agricultural areas. Endomycorrhizal density isolated under Corn that received low to moderate levels of agrochemical application revealed 113-129 spores/100 g dry soil compared to only 17 spores at high level. There was also lower endomycorrhizal colonization where application of agrochemical was higher. Results suggest that heavy agrochemical use have significant effects on dynamics of endomycorrhizas, hence a good soil indicator. It is advisable to regulate the use of agrochemicals to maintain soil productivity without harming beneficial role of endomycorrhizas. Cultural practices and integrated pest management are encouraged to reduce dependency on expensive agrochemicals. Isolation, characterization, testing, and mass production of indigenous endomycorrhizas are further recommended to produce alternative organic fertilizers for upland farmers in Tampakan mountain ecosystem.

Keywords: agrochemicals, endomycorrhizas, soil indicator

BS - 54

BIOSORPTION OF LEAD, COPPER, CADMIUM, AND MERCURY BY BROWN SEAWEEDS (*Sargassum* sp. and *Turbinaria ornata*)

Riel Carlo Ingeniero, Michaela Boren, Emee Tan and Teresita Perez

Department of Environmental Science, Ateneo de Manila University,
Loyola Heights, Quezon City

Biosorption is a process that uses inexpensive biomass to sequester toxic heavy metals and is particularly useful for the removal of trace levels of contaminants from industrial effluents. Several types of biomass have been tested for heavy metal sorption capacity which includes bacteria, fungi, algae, and aquatic macrophytes. Marine macroalgae are one of the organisms proven to have high heavy metal adsorption capacity due to the functional groups such as carboxyl groups from amino acids and polysaccharides and sulfhydral groups present in the biomass of the seaweeds. In this study, dried biomass of *T. ornata* and *Sargassum* sp. were tested for its heavy metal adsorption capacity. The maximum adsorption capacity of *Sargassum* sp. and *T. ornata* (qmax range=84.09-129.87) exceeded many previously reported uptakes by other types of biomass such as that of granulated activated carbon (qmax=5.08mg/g) and *Chlorella vulgaris* (qmax=58.8mg/g). *T. ornata* adsorbed higher amounts of copper while *Sargassum* adsorbed significantly higher amounts of cadmium and mercury. Results of the experiments also showed that pH and contact time affected the adsorption of heavy metals from aqueous solutions. Based on the FTIR analysis conducted, the carboxyl and hydroxyl groups were the functional groups responsible for sorption of the heavy metals. *Sargassum* and *Turbinaria* were proven to be effective metal biosorbents.

Keywords: biosorption, brown seaweeds, heavy metals, *Sargassum*, *Turbinaria ornata*

BS - 55

THE IDENTIFICATION OF FERN SPECIES AS METALLOPHYTES AND THEIR IMPLICATIONS TO MINE REHABILITATION

Rene Juna R. Claveria*, Eryn Gayle E. de Leon and Katrina B. Teodoro

Department of Environmental Science, Ateneo de Manila University
Loyola Heights, Quezon City reneclaveria@yahoo.com

Ultramafic and hydrothermally altered terrains are geographic areas that are anomalously high in metal concentrations and these are influenced by their underlying extreme geology. Plants that grow under such conditions are called metallophytes and to study them, plant and soil samples were taken in tandem at selected sites. The plants were sent to the Botany Division of the National Museum for identification. The plants and soil samples were processed for AAS analyses.

Hydrothermally altered terrains are relative high in Cu. At Lepanto, Benguet, *Nephnolepis hirsutula, Dicranopteris linearis, Pteridium aquilinum* and *Pteris* sp were the dominant fern species. Similarly at Philex, Baguio, *Histiopteris incisa, Onychium siliculosum, Pityrogramma calomelanos, Pteris* sp and *Sphenomeris chinensis* were abundant. Cu analysis of the roots yielded high values of about 488.00 ug/g, 371.00 ug/g in the stems and about 23.90 ug/g in the leaves. These ferns accumulated higher amounts of Cu than what the soils contain, In ultramafic terrains, such as Acoje, Zambales and Brooks Point, Palawan, laterites are anomalously high in Fe. *Pteridium aquilinum* and *Sphenomeris retusa* were abundant at Acoje, while at Brookes Point, *Dicranopteris linearis* was dominant. Fe analysis of the roots yielded high values up to about 9,358.24 ug/g, about 1,935.59 in the stem and about 199.10 in the leaves. These ferns thrived well in areas with Fe reaching to about 102,803.75 ug/g in the soil.

Ferns are characteristically good metal accumulators and are able to survive in very toxic soils, thus they could be used for mine rehabilitation. Most of them are pioneering plants, and the propagation of these ferns for phytoremediation and reforestation is worth considering.

Keywords: Ferns, metallophytes, phytoremediation, mine rehabilitation

BS - 56

TOXICOLOGICAL AND HISTOPATHOLOGICAL EVALUATIONS OF THE EFFECTS OFSYNTHETIC HORMONES– 17a-ETHINYLESTRADIOL AND LEVONORGESTREL – ON TILAPIA (*Oreochromis niloticus*)

Emilyn Q. Espiritu[1*], Stephanie S. Pimentel[2],
Norman Dennis E. Marquez[3], Ma. Cathrina Margarita R. Roxas[1]
and Felix Antonio T. Domingo[1]

[1]Environmental Science Department, [2]Biology Department, and
[3]Health Sciences Program, School of Science and Engineering, Ateneo de
Manila University, Loyola Heights, Quezon City eespiritu@ateneo.edu

Due to incomplete removal in most conventional biological sewage treatment plants, human excretions containing synthetic hormones from birth control pills find their way into sewage, agricultural runoff and surface waters. In sufficiently high concentrations, these can affect the physiology and reproductive functions of exposed organisms. Unfortunately, information on their potential effects mostly comes from temperate habitats, thus, limiting their application to tropical environments.

Acute toxicity tests (i.e. 96 hrs) and histopathological analyses of gills and liver tissues were performed to determine the effects of varying concentrations of 17a-Ethinylestradiol (EE2) and Levonorgestrel on juvenile *Oreochromis niloticus* (i.e. "Tilapia"). Fish mortality increased as toxicant concentration and exposure period increased with Mean 96-hr LC_{50}s ± S.D. of 0.47 ± 0.062 mg/L for EE2 and 6.03 ± 1.03 mg/L for Levonorgestrel ($p = 0.05$). Histopathological examinations also showed massive cellular damage – e.g. disorganization of lamellae, epithelium hyperplasia and hypertrophy, and blood congestion in gill tissues as well as nuclear hypertrophy, cytoplasmic vacuolation, and cytoplasmic degeneration in the liver – relative to the controls.

The information generated in the study can be used as an aid in establishing proper waste water protocols, in risk and impact assessments of xenoestrogens and in policy formulation for public health and the environment.

Keywords: Toxicity, Histopathology, 17a-Ethinylestradiol, Levonorgestrel, Tilapia

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VARIABILITY IN FOREWING VENATION PATTERNS AND GENITAL SHAPES AMONG FOUR POPULATIONS OF RICE BLACK BUGS, *Scotinophara coarctata* (Fabricius) FROM MINDANAO, PHILIPPINES

Mark Anthony J. Torres[1], Geoffrey Reuel Q. Yañez[1], Christine Lovelle A. Mahinay[1], Gerald Manuel P. Ong[1], Jay Ar O. Esencia[1], Joey Genevieve T. Martinez, Sharon Rose M. Tabugo, Liza A. Adamat, Muhmin Michael E. Manting, Ravindra C. Joshi[2], Alberto T. Barrion[2], Leocadio S. Sebastian[2] and Cesar G. Demayo[1]

[1]Mindanao State University – Iligan Institute of Technology
Iligan City, Lanao del Norte and [2]Philippine Rice Research Institute
Maligaya, Science City of Muñoz, Nueva Ecija
torres.markanthony@gmail.com; cgdemayo@gmail.com

Rice Black Bugs are small, cryptic, and highly invasive pest species attacking all growth stages of the rice plant. This pest is extremely variable in morphology resulting to problems in its taxonomy. The present study reports on variability in wing venation patterns and genital plates among four populations of RBBs comprised of 1,1149 males and 1,164 females from Tacurong, Sultan Kudarat (44M, 45F), Lala, Lanao del Norte (132F, 102M), Sta. Josefa, Agusan del Sur (216M, 216F) and Kabacan, North Cotabato (150F, 150M).

Results showed immense inter-individual variability in venation patterns among the populations. The most important variations are in the type and number of marginal cells on the anterior distad part of the wing. The marginal cells are either closed or open and vary in number from 1-6. Geometric Morphometric analyses revealed considerable differences in the shapes of the genital plates of both sexes (@&: Wilk's Lambda: 0.0005, P: <0.000; B&: Wilk's Lambda: 0.0009, P: <0.000). Results showed differences in the concavity of the anterior and postero-lateral margins of the genital plates. The results of the current study may indicate possible genetic differentiation. Such variability may have direct bearing on the management of the RBB as a pest of rice agroecosystem.

Keywords: Scotinophara spp., rice black bug, relative warp analysis, geometric morphometrics, wing venation pattern, genital plates

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PLANT DIVERSITY, STATUS AND CONSERVATION INITIATIVES IN THREE MOUNTAIN ECOSYSTEMS IN MINDANAO, PHILIPPINES

Victor B. Amoroso* and Fulgent P. Coritico

Central Mindanao University, Musuan, Bukidnon amorosovic@yahoo.com

This research describes the vegetation types, determines the diversity, assesses the conservation status and records the distribution of vascular plants in three mountain ecosystems in Mindanao, Southern Philippines. Representative specimens were pressed, dried, poisoned and mounted as herbarium vouchers. To determine the index of general diversity for trees, 20x20 m sampling plots were established per vegetation type, and in each sampling plot, a 5x5 m sub-plot was laid to determine the species diversity for pteridophytes. Transect walk and sampling plots in 3 mountain ecosystems revealed several vegetation types with Mt. Malindang having 9 types, Mt. Hamiguitan with 5 types and Mt. Kitanglad with 3 types. Species richness was highest in Mt. Malindang (1,164 spp.), followed by Mt. Hamiguitan (878 spp.) and the lowest in Mt. Kitanglad (661 spp.). The same trend was observed when the diversity values of the 3 mountain ecosystems were computed. Regardless of the kind of mountain ecosystem, the montane vegetation had high species richness and diversity values than the dipterocarp and mossy forests. Assessment of conservation status revealed that Mt. Kitanglad had the highest number of threatened species (92 spp.) while the two other mountain ecosystems, Mt. Malindang and Mt. Hamiguitan had 34 and 35 threatened species each, respectively. As to endemism, it showed that Mt. Hamiguitan had high endemism (34 %) than Mt. Kitanglad (21%) and Mt. Malindang (16%). Moreover, the 3 mountain ecosystems showed 64 species as new record in Mindanao and 21 species in the Philippines while 2 species of *Nepenthes* are new to science. Ex-situ conservation initiatives were done to protect the remaining threatened and endemic species of plants and their habitats.

Keywords: species richness, assessment, ex-situ and in-situ conservation, protected areas

BS - 59

SPECIES DIVERSITY OF SEAGRASSES IN CAMOTES ISLANDS, CENTRAL PHILIPPINES

Serapion N. Tanduyan, Ricardo B. Gonzaga and Berenice T. Andriano

Cebu Technological University, San Francisco, Cebu Campus
standuyan@yahoo.com

Seagrasses of Camotes Islands were studied to determine its species diversity A transect-quadrat method was used where 3 transects were laid in each station taking into account its species in the four municipalities of Camotes Islands which are San Francisco, Poro, Tudela and Pilar.

Results show that there were 11 species of seagrasses found in the four municipalities of Camotes Islands which are *Halophila decipiens, Thalassia hemprichii, Cymodocea rotundata, Cymodocea serrulata, Halodule pinifolia, Halodule uninervis, Halophila minor, Halophila ovalis, Halophila ovata, Enhalus acoroides and Syringodium isoetifolium*

Halophila decipiens and *Thalassia hemprichii* are the distinct species of seagrass found in the municipalities of Poro and San Francisco, respectively while there are 9 species that are common in all the municipalities that include: *Cymodocea rotundata, Cymodocea serrulata, Halodule pinifolia, Halodule uninervis, Halophila minor, Halophila ovalis, Halophila ovata, Enhalus acoroides and Syringodium isoetifolium.*

Keywords: Seagrass, Species Diversity, Camotes Islands, Transect-quadrat method

BS - 60

MOLLUSCAN SPECIES ASSOCIATED WITH HOLOTHURIAN HABITATS IN CAMOTES ISLANDS, CENTRAL PHILIPPINES

Serapion N. Tanduyan, Berenice T. Andriano, Ricardo B. Gonzaga, Wilfredo B. Anoos and Lourdes M Garciano

Cebu Technological University, San Francisco,
Cebu Campus standuyan@yahoo.com

Mollusks inhabiting the holothurian habitats were studied. There were 3 habitats surveyed, the sandy, muddy and grassy flats.

This study used the transect quadrat method where transect was laid in the three habitats :sandy, muddy and grassy flats of Camotes Islands.

It was found out that there were nineteen species of mollusks belonging to the fifteen families namely, Family *Conidae, Phasianellidae, Strombidae, Buccinoidea, Cypraeidae, Volutidae, Cassidae, Littorinidae, Harpidae, Pectinidae, Mytilidae, Pinnidae, Arcidae, Octopodidae and Aplysiidae.*

Results show that there are six species found in sandy areas; one species in muddy and grassy areas; two in rocky areas.

Results further show that there are three species found in both sandy and grassy areas and six species in both rocky and sandy areas.

Keywords: Molluscan Species, Holothurian habitats, Camotes Islands, Transect-quadrat method

BS - 61

DETECTION OF DERMATOPHYTES FROM DIFFERENT BODY SITES OF FARMERS IN SELECTED BARANGAYS OF AMADEO, CAVITE

Sherine M. Cruzate[1], Ninibeth B. Cupino and Yolanda A. Ilagan[2]

Cavite State University, Indang, Cavite

A total of 600 skin scrapings from hands, arms, legs and feet and 300 nail clippings of toenails and fingernails were collected from 150 farmers of Halang, Banay Banay, Pangil, Tamakan and Dagatan in Amadeo, Cavite. Eighty five of the 150 farmers exhibited symptoms of dermatophytosis. All clinical samples were subjected to direct microscopic examination using potassium hydroxide (KOH) to detect the presence of fungal hyphae. Positive KOH cases were cultured on Saboraud Dextrose Agar (SDA) with streptomycin to determine the fungi that invaded the tissues.

KOH mount revealed that 79.49% of the skin scrapings and 92.31% of nail clippings from farmers with dermatophytosis were positive for the presence of fungal elements. However, opposite result was obtained on samples from farmers without dermatophytosis. Only 43% and 66.55% from skin scrapings and nail clippings, respectively, were positive in KOH mounts. Not all KOH positive samples were culture positive.

Based on the culture, 333 fungal isolates were obtained. They were commonly found in toenails, feet, legs, hands and fingernails. The least number of isolates were found in the arms. *Trichophyton* was the only dermatophyte while the rest of the fungi were non-dermatophytes belonging to the *Aspergillus*, *Penicillium* and *Fusarium*.

Statistical analysis revealed that there was a significant relationship between the age ($p<0.05$) and gender ($p< 0.01$) on the occurrence of fungi to farmers with and without dermatophytosis. However, no significant correlation was found with medical history.

Keywords: dermatophytes, dermatophytosis, KOH mounts, *Trichophyton*, hyphae

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DIVERSITY IN OUR MIDST: THE BUTTERFLY NECTAR FEEDERS OF THE DIVIDIVI TREE, CAESALPINIA CORIARIA (JACQ) WILD AT UP LOS BAÑOS, LOS BAÑOS, LAGUNA

Bonifacio F. Cayabyab* and Genaro A. Katimbang

National Crop Protection Center – Crop Protection Cluster
University of the Philippines Los Baños, Laguna bfcayabyab@yahoo.com

. The Dividivi, *Caesalpinia coriaria* (Jacq) Wild is an exotic plant from South America. This leguminous medium sized tree is usually found in windswept seashores. At a glance it looks like a tamarind tree. It is noted for its horticultural and medicinal value. The lone Dividivi tree between Biological Science and the new College of Arts and Sciences buildings at UP Los Baños exemplifies the diversity of life forms in our midst through the nectaring butterflies on its flowers.

We studied the butterfly nectar feeder of the Dividivi tree from August 2010 – December 2010 in order to contribute to the documentation of diversity inventory at Los Baños. Observations were done at 0800 hours – 0900 hours. Counts were accomplished at least once a month.

We documented the species that visited the flowers based on their flight pattern and morphological appearance. We also use sweep net to examine closely the butterflies and release them later.

A total of 36 species from 6 families were observed. The order of decreasing density of the families of butterflies was: Nymphalidae (10) > Papilionidae (9) > Pieridae (8) > Danaidae (5) > Hespiriidae (2) = Lycaenidae (2).

We plan to continue observing the butterflies of Dividivi tree on a monthly basis to enable us to have a whole year perspective of the diversity of butterfly nectar feeders of this tree.

Keywords: Diversity, Butterfly, Nectar, Feeders, Dividivi

BS - 63

UNUSUAL POPULATION INCREASE OF WOOLY BEAR CATTERPILLARS, SPILOSOMA SP. (ARCTIIDAE: LEPIDOPTERA) AT LOS BAÑOS AND VICINITY IN 2010

Bonifacio F. Cayabyab*

National Crop Protection Center – Crop Protection Cluster
University of the Philippines Los Baños, College, Laguna
bfcayabyab@yahoo.com

The wooly bear catterpillars, *Spilosoma sp.* are usually found in *Ipomea* obscura (L.) Ker-Gaw). In 2010, there was sudden and unusual increase in population of this pest. The catterpillars attacked various crops such as eggplants, corn, okra, patola, ube, paayap and a number of weeds species. The catterpillars even entered homes and pupated in ceilings, cracks and crevices. It caused anxiety to many people due to the menacing thick hairy appearance.

The prolonged dry spell in 2010 could have triggered the aggregation of adults in remaining green areas and led to subsequent population increase. We compared the 2010 agrometeorology data from that of 2005-2009 at the Central Experiment Station, UP Los Baños. The results showed low rainfall from February (3.0 mm) up to May (9.1 mm) in 2010 as compared to the previous four years. The rainfall also increased in June to July 2010 at 171.2 mm and 762.5 mm respectively. This dry spell followed by two wet months could have led to the population increase of *Spilosoma sp*. This phenomenon is similar to the locust outbreak in Central Luzon in the 90's where dry environment due to ash fall predominated in breeding areas followed by strong rainfalls led to population increase and subsequent migration/infestation in the central plains.

The population increase of wooly bear caterpillar coincided with the population outbreaks of armyworms, *Spodoptera sp*. semi-loopers and other lepidoterous species in various parts of the country.

Keywords: *Spilosoma sp.* Population, Unusual, Increase, Wooly Bear, Caterpillars

BS - 64

SOME BUTTERFLIES OF BORACAY ISLAND

Bonifacio F. Cayabyab[1]* and Edwin P. Alcantara[2]

[1]National Crop Protection Center - Crop Protection Cluster and
[2]BIOTECH - University of the Philippine Los Baños College, Laguna
bfcayabyab@yahoo.com

Boracay Island, Malay, Aklan is one of the well known and busiest tourist destination in the country at present. Owing to the increasing demand for space for commerce and domiciles, the butterflies habitat and range in this island resort are rapidly diminishing. We conducted a rapid survey of the butterflies at Boracay last April 2010 to document the butterfly fauna and contribute in the biodiversity inventory of the locality.

We noted down the species that we observed based on their flight pattern and morphological appearance. In other cases we use sweep net to examine closely the butterflies and release them later after confirming our identification.

A total of thirty two species from seven families were noted. The order of decreasing density of the seven families was: Papilionidae (8) > Nymphalidae (7) > Pieridae (6) > Danaidae (3) > Hesperiidae (4) = Lycaenidae (4).

Additional counts particularly in the watershed will be included in the future to be able to record the species that are strictly present in forested areas. We will also invite partners in the locality.

Keywords: Butterflies, Boracay, Tourist, Island, Resort

BS - 65

ISOLATION AND SCREENING FOR BACTERIA WITH AMYLOLYTIC, PROTEOLYTIC OR LIPOLYTIC ACTIVITY FROM PITCHER PLANT (*Nepenthes sp.)*

Erwin P. Elazegui

College of Science, Technological University of the Philippines-Manila

Microorganisms possess many different enzymes. The activities of these enzymes determine the nature of the organism; microbe's physiological properties by its enzymatic activities and ecological relationships. Enzymes are proteins that speed up chemical reactions. The traditional sources of many enzymes are plants and animals. These sources are no longer adequate to meet the growing demands for enzymes. Consequently, manufacturers and scientists are turning to microorganisms as sources.

Several studies have dealt with the isolation of enzyme producing bacteria from different sources. However, no similar studies have been conducted on Pitcher plant (Nepenthes sp.) or several plants which are indigenous and endangered plant to the Philippines. Enzymes produced from microbial metabolism can be useful raw materials for industrial and environmental applications.

The study focused on the isolation and screening of bacteria from the phytotelmata or digestive fluid of pitcher plant. Thirty-two bacteria were isolated in which ten were screened for enzymatic activities.

Results showed that four of the five isolates were positive for amylolitic activity as indicated by clear zones on Starch Agar. Nine isolates were lipolytic as indicated by zones of clearing and precipitation on Egg Yolk Agar while ten isolates exhibited proteolytic activity on Skim Milk Agar.

Partial characterization of bacterial isolates screened were gram positive, endospore forming, aerobic to facultatively anaerobic rods which probably belong to the Family Bacillaceae.

Keywords : bacteria, amylolitic, lipolytic, proteolytic, phytotelmata

BS - 66

AN INTERACTIVE IDENTIFICATION KEY: THE PHILIPPINE DERBIDAE PROJECT

Sheryl A. Yap[1] and Thierry Bourgoin[2]

[1]Museum of Natural History, University of the Philippines Los Baños, Laguna; sheng_yap@yahoo.com and [2]Museum National d'Histoire Naturelle, UMR 7205 Paris, France bourgoin@mnhn.fr

Identification is the process of finding unknown organism to which an organism belongs. There are several methods that are available for aiding this process. The most common is the use of a conventional paper-based identification keys - a tedious and time consuming work. The other one is with the help of a computer-aided identification keys - with more options, support back-tracking, provide pictures of excluded species as well as of those that are still under consideration, and provides drawings or pictures of characters or body parts that is/are being described (Yap and Froese, 2005).

With about 160 genera and 1500 species, Derbidae represents the third larger family of the Fulgoromorpha (FLOW, 2010). F. Muir (1917) was able to comprehensively collect and study them in the Philippines. He identified and described a total of 39 genera and 98 species based on the large collection of Prof. C. F. Baker and his own. Most hemipterist have paid less attention to the derbid fauna of the Philippines, aside from those that are found attacking economically important plants. The high endemicity of the derbids was noted by Muir (1917), and clearly showed that many more species remain to be discovered.

Using Xper2 - an open e-tool to manage descriptive data - we present here the first illustrated computer-aided identification key for the known Philippine derbid genera based on diagnostical morphological characters. This computer-aided key would facilitate much faster identification of Philippine derbids in the field and would be helpful to those studying derbids with limited access to foreign literature.

Keywords: Derbidae, computer-aided identification key, interactive identification key, identification

BS - 67

PANDAN (*Pandanus amaryllifolius* Roxb.): A PROMISING ANTIDIABETIC AGENT

Graciela R. Cuaresma[1] and Henedine A. Aguinaldo*[2]

[1]McKeough Marine Center, Xavier University,
Corrales Avenue 9000, Cagayan De Oro City
[2]College of Arts and Sciences, Mariano Marcos State University
City of Batac 2906, Ilocos Norte henedine_aguinaldo@yahoo.com.ph

The antidiabetic activity of pandan (*Pandanus amaryllifolius* Roxb) was determined in this study. Pandan is widely grown and is used for many purposes. It is known to have useful properties as antiviral, anti-allergen, antiplatelet, anti-inflammatory and anti-oxidant. Pandan contains volatile compounds, alkaloids (pandamarine, pandamerilactones) and essential oils. In this study, the antidiabetic activity of pandan leaf extract was determined in hyperglycaemic induced guinea pigs (*Cavia porcellus* Gesner K., after introduction of varying concentrations of pandan leaf extract (PALE) at 0.3 ml per 100 grams of body weight. Hyperglycaemia was induced by giving 0.3 ml glucose solution per 100g body weight after base line blood glucose determination was done.

Insulin and distilled water (DW) served as the positive and negative control, respectively. Blood glucose levels (BGL) of test animals were determined prior to hyperglycemia, at the onset of hyperglycemia and also 1, 3 and 5 hours after treatment administration of PALE.

Results showed that the PALE concentrations initiated BGL reduction in hyperglycaemic-induced guinea pigs. Significant hypoglycaemic effect was observed with different PALE concentrations: 100% PALE is as effective as insulin, followed by 30% and 50% PALE. The negative control gave a minimal decrease in BGL.

Hence, *Pandanus amaryllifolius* Roxb. Leaf extract (PALE) was found to be an effective agent in reducing blood glucose level. Economic analysis revealed 97.2% lower expense than what is incurred if insulin injection is administered. Furthermore, the use of PALE is 89.68% to 92.4% lower than the administration of Glucophage alone or Glucophage and Euglucon, respectively.

Keywords: ***Pandanus amaryllifolius*****,** pandan, antidiabetic, hypoglycaemic, BGL

BS - 68

TAXONOMIC STATUS AND LEAF EPIDERMAL FEATURES OF *AMOMUM* ROXB. (FAMILY ZINGIBERACEAE) RECORDED FROM MINDANAO, PHILIPPINES

Florfe M. Acma[1*] **and William Sm. Gruezo**[2]

[1]Department of Biology, College of Arts and Sciences, Central Mindanao
University, University Town, Musuan, Bukidnon flmacma@yahoo.com.ph
[2]Plant Biology Division, Institute of Biological Sciences, College of Arts and
Sciences and Museum of Natural History, University of the Philippines
Los Baños, Laguna asialifesciences@yahoo.com

The taxonomic evaluation of some *Amomum* species (Family Zingiberaceae) reported from Mindanao Philippines was conducted using both field and herbarium studies supplemented with leaf epidermal characters.

Botanical field studies were conducted and specimens were identified using taxonomic keys, protologues and by doing herbarium studies at the Philippine National Museum (PNH), College of Agriculture, University of the Philippines, College, Laguna (CAHUP) and Herbarium of the Singapore Botanical Gardens (SING).

Results of the study showed that a number of species originally placed under the genus *Amomum* are currently classified under the genera as *Etlingera* Giseke [*E. dalican* (Elm.) Poulsen, *E. philippinensis* (Ridl.) R. M. Smith] *Geocharis* [*G. fusiformis* (Ridl.) R. M. Sm.] and *Hornstedtia* Retzius [*H. conoidea* Ridl.] Examination of the leaf epidermal features revealed that the both *Amomum* and *Geocharis* have broad subsidiary cells while *Hornstedtia* and *Etlingera* had narrower subsidiary cells. *Amomum* have more stomata located near the veins than halfway between veins. In contrast, the stomata for *Hornstedtia* and *Etlingera* are scattered between the parallel veins of the leaf. The leaf epidermal data support the current placement of the above-listed formerly considered species of *Amomum*. Finally, this study showed that previous taxonomic treatment for the Philippine *Amomum* is quite unsatisfactory.

Keywords: Zingiberaceae, *Amomum*, epidermal features, clearing technique, stomata

BS - 69

TRANSCRIPT PROFILLING OF SELECTED GENES IN NORMAL AND MUTANT MAKAPUNO ENDOSPERMS OF COCONUT (*Cocos nucifera* L.) USING RELATIVE RT-PCR

Reggie Yadao-dela Cruz[1,2*], Rita P. Laude[3], Ma. Genaleen Q. Diaz[3], Antonio C. Laurena[4], Merlyn S. Mendioro[3], and Evelyn Mae T. Mendoza[4]

[1]Graduate School, University of the Philippines, Los Baños, Laguna
[2]Department of Biology, College of Arts and Sciences, Central Mindanao University, Musuan, Bukidnon reggiecmu@yahoo.com
[3]Institute of Biological Sciences, College of Arts and Sciences and
[4]Institute of Plant Breeding, Crop Science Cluster, College of Agriculture
University of the Philippines Los Baños, Laguna

Makapuno is an abnormal coconut with characteristic endosperm overgrowth. It has softer, fluffy endosperm compared to the hard, crisp solid endosperm of normal coconut. To gain insights on the molecular basis of the makapuno phenomenon, ten selected genes were cloned, characterized and their expression determined through Relative RT-PCR with actin as internal control gene. The ten genes showed conserved domains and have 75-98% identity to other plant sequences when analyzed with Interproscan and BLAST. Four of the ten genes were found to have lower expression in makapuno including an isopentenyl transferase gene that controls the first rate-limiting step in cytokinin biosynthesis. Three glycolytic genes, cytosolic glyceraldehyde-3-phosphate dehydrogenase, cytosolic pyruvate kinase and enolase were also downregulated in makapuno. On the one hand, three regulatory genes namely, protein kinase CK2 regulatory subunit, polyA binding protein, myb-like transcription factor were upregulated in makapuno. There was also altered expression of pyruvate decarboxylase, á-D-galactosidase, and ketoacyl-acyl carrier protein synthase 1 (KAS1). These data support previously reported differences in cytokinin levels, carbon metabolism and respiratory levels in the normal and makapuno coconut endosperms. These also provided further insights into the elucidation of the molecular mechanisms regulating gene expression in normal and makapuno endosperms of coconut.

Keywords: endosperm overgrowth, gene expression, altered carbon metabolism, relative RT-PCR, coconut

BS - 70

IN VITRO CULTURE AND ENZYME PRODUCTION OF PLASMODIAL MYXOMYCETES (SLIME MOLDS) COLLECTED FROM LUBANG ISLAND, OCCIDENTAL MINDORO

Sittie Aisha B. Macabago[1] **and Thomas Edison E. dela Cruz**[1,2*]

[1]The Graduate School, and [2]Fungal Biodiversity and Systematics Group,
Research Cluster for the Natural and Applied Sciences,
University of Santo Tomas, España, Manila sittieaisha@gmail.com;
tedelacruz@mnl.ust.edu.ph

Plasmodial myxomycetes or slime molds are often studied for the unique and interesting stages of their life cycle. Recently, these organisms were tapped for the production of anticancer metabolites. However, with their minute sizes, *in vitro* culture offers the possibility for the mass production of their natural products. In our research study, we collected plasmodial myxomycetes from ground leaf litter and twigs cultivated on moist chambers. *In vitro* culture showed spore germination of 13 species: *Craterium atrolucens, C. concinnum, Diderma effusum, Didymium ochroideum, D. squamulosum, Diachea leucopodia, Lamproderma scintillans, Oligonema schweinitzii, Perichaena microspora, Physarum bivalve, P. cinereum, P. compressum* and *P. melleum*. Ten of these species developed into amoeboflagellates while seven further grew into plasmodia. Both of these are part of the life stages of myxomycetes. The amoeboflagellates of these myxomycetes were then preserved and stored for 3 months. Revival of preserved amoeboflagellates resulted in 82% success rate. Production of extracellular enzymes was also tested from the *in vitro* grown myxomycetes. Plasmodia of *P. compressum* and one plasmodium derived from a hardened sclerotium, a resting stage of myxomycetes, showed *in vitro* the presence of amylase and protease indicating perhaps an alternative mode of nutrition for these myxomycetes in addition to the phagotrophic mode of nutrition known for these organisms. This research study is the first report of *in vitro* culture and enzyme production of plasmodial myxomycetes in the Philippines.

Keywords: slime molds, *in vitro* culture, culture preservation, extracellular enzymes, mode of nutrition

BS - 71

FUNCTIONAL ACTIVITIES OF TWO STRAINS OF *Pleurotus cystidiosus*

Daryl Juganas, Sofronio P. Kalaw and Renato G. Reyes

Center for Tropical Mushroom Research and Development, Department of Biological Sciences, College of Arts and Sciences, Central Luzon State University, Science City of Muñoz, Nueva Ecija spk31162@yahoo.com

Pleurotus cystidiosus is an edible mushroom widely distributed in subtropical and tropical countries growing on decaying logs and other agricultural wastes. In the Central Luzon, wild strains of this mushroom were collected in upland community of Llanera, Nueva Ecija and Central Luzon State University (CLSU) campus. The Center for Tropical Mushroom Research and Development has developed production technology for the cultivation of this mushroom. Because of its potential in the nutraceutical and pharmaceutical industries, our research team determined the functional activities of the basidiocarp and mycelial hot water extracts of two wild strains of *P. cystidiosus*. Carageenan induced edema and charcoal tracing method were performed to determine the anti - inflammatory and the anti - spasmodic activity, respectively, using Swiss albino mice.

Carageenan induced edema test revealed that the mycelia and basidiocarp extracts of the two strains can reduce inflammation. Basidiocarp extract of CLSU strain recorded the highest percentage of inhibition of inflammation with means of 26.47 % and 66.18 % one and three hours after the application, respectively. On the other hand, charcoal tracing assay for antispasmodic activity showed that the mycelial extract of Llanera strain registered the lowest percent traveled by charcoal in the intestine of mice with a mean of 41.83%.

Keywords: *Pleurotus cystidiosus*, functional activity, anti-spasmodic, anti-inflammatory

BS - 72

THE OCCURRENCE OF MANGROVE BRACHYURAN CRABS AND OBSERVATION ON THE FEEDING ECOLOGY OF SOME TAXA OF THE GRAPSOID FAMILIES GRAPSIDAE AND SESARMIDAE *sensu* SCHUBART *et al.* (2002) FROM SELECTED MANGALS OF LUZON, PHILIPPINES

Jimmy T. Masagca

Pacific Island Institute for Pedagogy, Technology, Arts and Sciences, Inc. (Pacifictech); De La Salle University-Dasmarinas, Cavite; and Catanduanes State Colleges, Virac, Catanduanes jmasagca@yahoo.com

Despite the obvious importance of mangrove brachyurans to ecosystem functioning in relation to their roles in retaining a large proportion of mangrove leaf-litter, the most prominent groups belonging to the grapsoid families (Grapsidae and Sesarmidae) are less studied in the Philippines. Occurrence and some observations on the feeding ecology of selected grapsoid sesarmids as to their dependence on mangrove trees as habitats, burrowing behavior and arboreal climbing skills were considered in this study. A total of 6 families, 17 genera and 21 species (9 grapsoid sesarmids and 12 non-grapsoids) of mangrove brachyurans are reported from Pagbilao (Quezon); Manila Bay area (Cavite); and Palnab-Pajo and Agojo Inlet (Catanduanes). Sesarmidae is represented by *Perisesarma, Episesarma, Selatium* and *Sarmatium;* while Grapsidae is represented by *Hemigrapsus, Pseudograpsus* and *Metopograpsus.* Non-grapsoids consist of Varunidae (*Ptychognathus*); Ocypodidae (*Dotilla, Ocypode, Uca*); Portunidae (*Charybdis, Portunus, Scylla, Thalamita*); and Eriphiidae (*Epixanthus*). The sesarmids (*Selatium elongatum* and *Episesarma versicolor*) are exclusive mangrove tree climbers or EMTC, while *Metopograpsus latifrons* and *Sarmatium germaini* are occasional mangrove tree climbers or OMTC and non-arboreal or non-mangrove tree climbers or NMTC are *Neosarmatium smithi, Perisesarma bidens.*

Keywords: Brachyurans, mangroves, Catanduanes, Quezon, Cavite, Grapsidae, Sesarmidae

BS - 73

TAXONOMY OF VEGETABLE OILS BASED ON FATTY ACID COMPOSITION

Chester C. Deocaris[1] **and Custer C. Deocaris**[2]

[1]Biology Department, College of Arts and Sciences, University of the Philippines-Manila, Padre Faura Street, Ermita, Manila chdeocaris@yahoo.com
[2]Custer's Vegginnovative Foods, Quezon City cdeocaris@gmail.com

A scheme for classifying vegetable oils based on fatty acid composition is proposed. Heatmap and dendrogram were constructed from gas-fatty methyl ester (FAME) chromatograms of vegetable oils obtained from Codex Standards 210-1999 and 33-1981; and from published and experimental data. The vegetable oils analyzed formed 5 major clusters designated as lauric (Cluster 1); linoleic (Cluster 2), high-oleic (Cluster 3); palmitic (Cluster 4) and mid-oleic (Cluster 5). Coconut oil including, the virgin coconut oil (VCO) and the refined, bleached and deodorized (RBD-CO) variants are classified under the "Lauric Cluster" along with Babassu and Palm kernel oils. This group is unique as most of its fatty acids are fall under the medium-chain (C6:0 to C12:0) category. This food oil taxonomy can be correlated with the nutritional properties attributed to specific fatty acid, fatty acid group or vegetable oil. More importantly, our simplified chemo-informatic based classification scheme can be adapted to the existing food classification systems, such as the EuroFIR and of the USDA.

Keywords: fatty acid, vegetable oil, coconut oil, chemo-informatics and cluster analysis

BS - 74

CYTOTOXICITY TEST AND PHYTOCHEMICAL SCREENING OF CRUDE ETHANOLIC AND HEXANE EXTRACT OF THE AERIAL PART OF *Hydrocotyle verticillata* (WHORLED MARSHPENNYWORTH)

Nathalie L. Daminar[1,*] **and Lydia M. Bajo**[2]

[1]Caraga State University- Cabadbaran Campus, T. Curato St.,
Cabadbaran City nathalie_daminar@yahoo.com
[2]Department of Chemistry, Mindanao State University-Iligan
Institute of Technology, Iligan City

The crude ethanolic extract of the aerial part of ***Hydrocotyle verticillata*** was subjected to solvent partitioning according to increasing polarity: n-hexane, ethyl acetate (EtOAc), dichloromethane (DCM) and water. Brine shrimp lethality test was conducted to the four partitioned solvent extracts and result showed that n-hexane and ETOAc have significant cytotoxic potentials having LC_{50} of 50.35 and 76.98 ppm respectively. Hence, n – hexane extract showed to be the most bioactive since it has the lowest LC_{50} among the four solvent extracts.

The phytochemical screening of ethanolic extract of *H. verticillata* showed the presence of bioactive components like alkaloids, saponins, steroids, flavonoids, tannins and anthraquinones. While the phytochemical test on hexane extract (the most bioactive among the partitioned solvent extract) revealed only the presence of alkaloid and saponin.

Keywords: *Hydrocotyle verticillata*; cytotoxic; phytochemicals; brine shrimp

BS - 75

SYSTEMS ANALYSIS OF X-CHROMOSOMAL miRNAs AND THEIR TARGET GENES USING INFORMATICS TOOLS

Ronie B. Mendoza[1], Evelyn M. Labastilla[1], Marla A. Endriga[2], Chester C. Deocaris[2] and Custer C. Deocaris[3*]

[1]College of Science, Tarlac State University, Tarlac City
[2] College of Biology, University of the Philippines-Manila, Manila
[3] Yonsei University School of Medicine, Seoul, South Korea
cdeocaris@gmail.com

In an attempt to investigate the potential roles of miRNAs in human intelligence, miRNAs located in the X-chromosome were identified by mining the miRBase database. Out of the 77 miRNAs, 10 were chosen for further analysis based on their differential levels in the various functional parts of the brain: cortex (executive functions, long-term/immediate memory and speech), hippocampus (learning and working memory) and the midbrain (visual and auditory systems, and body movement). Target genes of these ten brain-expressed X-chromosomal miRNAs were predicted and compared with the following randomized miRNAs: ten nonbrain-expressed X-chromosomal miRNAs, ten nonbrain-expressed miRNAs that are not found in the X-chromosome and ten brain-expressed miRNAs that are not found in the X-chromosome. For the brain-expressed X-chromosomal miRNAs, a total of 221 target genes were predicted based on their common targets from three public prediction algorithms: MiRanda, TargetScan and MiRTarget2. These target genes were then annotated and compared according to their gene ontologies (GOs). Chemo-and Bioinformatics analyses indicate that the X-chromosome has potential roles in human intelligence as indicated by the statistically significant GO terms associated with learning, memory and brain development by the target genes of the brain-expressed X-chromosomal miRNAs.

Keywords: molecular sysytems analysis, miRNAs, gene ontology, MiRanda, TargetScan