ISSN 0115-8848

TRANSACTIONS
of the
NATIONAL ACADEMY OF SCIENCE AND TECHNOLOGY
Philippines

ABSTRACTS of PAPERS
Presented during the
28th NAST Annual Scientific Meeting

The Century of Biology

12-13 July 2006

2006
Volume 28
Issue No. 1

Republic of the Philippines
National Academy of Science and Technology
Philippines

ISSN 0115-8848

TRANSACTIONS
of the
NATIONAL ACADEMY OF SCIENCE AND TECHNOLOGY
Philippines

ABSTRACTS of PAPERS
Presented during the
28th NAST Annual Scientific Meeting

The Century of Biology

12-13 July 2006

2006
Volume 28
Issue No. 1

Republic of the Philippines
National Academy of Science and Technology
Philippines

28th ANNUAL SCIENTIFIC MEETING

The Century of Biology

28th ASM: 12-13 July 2006; The Manila Hotel

28th ASM Steering Committee

Academician Emil Q. Javier (Chair)
Academician Evelyn Mae Tecson-Mendoza
National Scientist Clare R. Baltazar
National Scientists Jose R. Velasco
Academicians Angel C. Alcala
Academicians Filomen F. Campos
Academicians Magdalena C. Cantoria
Academicians Veronica F. Chan
Academicians Salcedo L. Eduardo
Academicians Edgardo D. Gomez
Academicians Asuncion K. Raymundo
Academicians Prescillano M. Zamora

Transactions of the
National Academy of Science and Technology
Philippines

ABSTRACTS of PAPERS
Presented during the
28th NAST Annual Scientific Meeting

The Century of Biology

28th ASM: 12-13 July 2006; The Manila Hotel

2006
Volume 28 (No. 1)

Republic of the Philippines
National Academy of Science and Technology
Philippines

ISSN 0115-8848

Trans. Nat. Aca. Sci. Tech. Philippines
Vol. 28, No. 1
Published by the National Academy of Science and Technology Philippines
2/F Philippine Science Heritage Center
DOST Complex
Bicutan, Taguig City 1631 Philippines
secretariat@nast.ph; nast@dost.gov.ph
Tel. No. (632) 837-2071 loc 2170 to 73; Telefax: (632) 837-3170

Issue Editor: Academician Evelyn Mae Tecson-Mendoza

TABLE OF CONTENTS

PLENARY SESSIONS

POSTER SESSIONS

AGRICULTURAL SCIENCES

BIOLOGICAL SCIENCES

CHEMICAL, MATHEMATICAL, AND PHYSICAL SCIENCES

ENGINEERING SCIENCE AND TECHNOLOGY

HEALTH SCIENCES DIVISION

SOCIAL SCIENCES

PLENARY SESSIONS

BIO-RESOURCE MANAGEMENT AND OUR COMMON FUTURE

Agnes C. Rola, Ph.D.

Professor and Director
Institute of Strategic Planning and Policy Studies
College of Public Affairs, University of the Philippines Los Baños
Email: arola@laguna.net

Societies survive or collapse depending upon how their inhabitants are able to manage the bio resources. Bio-resources management involves responsible use of our living resources- plants, animals and the natural environment that support these, for both traditional and new applications. Bio-resources' utility vary temporally and spatially. In the early times, when customary rules prevailed and societies were able to check population growth, bio resources were managed in a sustainable manner. These approaches were rooted on religious beliefs and myths, legends, and cultural norms. Collective action evolved in the villages to share responsibilities to safeguard the land, water and the biological resources for sustainable use.

As population grew, the state became the more powerful steward of the bio resources. Protected areas in forests and marine sanctuaries were set-up. But the weak property rights, the lure of commercialism and the seemingly lack of collective action to protect these resources contributed to bio resources' destruction. Recent data (2005) show that the Philippines ranked 125 out of 146 countries in terms of environmental sustainability index. This reveals the sorry state of bio resources management in the country.

In the paper, we hypothesize that policies and institutions influence bio resources conditions and reforms of these will contribute to sustainability. The paper is divided into three parts. Part 1 describes the situation of bio resources in the country and the circumstance that led to their current state. Part 2 discusses the current management strategies and notes the impact of the recent laws including the devolution of some of the environmental services to local governments. Part 3 answers the question -where do we go from here? What are some of the optimal strategies for bio-resources management that will ensure our society's survival? Discussions will revolve on institutional reforms and mainstreaming bio resources management into the development agenda; changing mindsets, attitudes and practices; and pursuing innovative partnerships, including more active participation in international treaties governing bio resources.

Keywords: bio-resource management, environmental sustainability, international treatise

PROTEOMICS: WEIGHING THE EVIDENCE

Leopold L. Ilag, Ph.D

Director, Proteomics Facility
Department of Analytical Chemistry
Stockholm University, Sweden
Email: leopold.ilag@anchem.su.se

There are an estimated 1,000,000 proteins coded by the genes of the human genome. This staggering number coupled to the inherent variety of activities afforded by post-translational modifications and several potential interactions make the whole world of genomics look simple in comparison to the goals of proteomics. Not only is the proteome more diverse, but it is also dynamic in contrast to the static genetic constitution of a given organism. This is expressed not only in the temporal regulation of protein function but also in the modulation of spatial distribution. In other words, a single gene could give rise to a variety of functional proteins each of which may be expressed at different levels at different times and in different sub-cellular sites. To make matters more challenging, proteins often act as part of large complexes. Among these dynamic assemblies are multi-subunit molecular machines which are products of billions of years of evolution. These machines often prove to be elusive targets for most biochemical and biophysical scrutiny. In part this is because these multi-subunit assemblies are not practical to over-express hence most studies depend on isolation of materials from natural sources. Furthermore, many of these macromolecular complexes tend to asymmetric making them less straightforward to study by techniques that profit from data averaging such as NMR and crystallography.

A major boost in the advance of proteomics is the application of mass spectrometry that was made possible by the development of soft ionization methods namely Matrix-assisted Laser desorption (MALDI) and electrospray ionization (ESI). Nano-electrospray conditions provide optimal sensitivity for individual proteins and their peptidic fragments. Electrospray used in this way coupled to tandem mass spectrometry approaches to analyse peptide fragments formed the backbone of most efforts in proteomics worldwide. In parallel to developments in proteomics, mass spectrometry also evolved as an important tool for looking at intact biopolymers. Non-covalent complexes were possible to maintain inside the spectrometer by manipulating solution conditions. Taken together, these advances have allowed mass spectrometry to identify important biological interactors and analyze the largest of complexes.

Keywords: proteomics, MALDI, ESI, human genome, protein, Electrospray

BIOREMEDIATION: A PROVEN AND COST EFFECTIVE TOOL FOR REPAIRING THE ENVIRONMENT

Stephen S. Konigsberg, Ph.D[1].and Asuncion K. Raymundo, Ph.D[2]

[1]Partner, Environmental Strategies Consulting LLC
Irvine, California, U.S.A.
Adjunct Professor, The University of California and
The California State University
Email: skoenigsberg@escirv.com
[2]Academician, National Academy of Science and Technology
Professor, University of the Philippines Los Banos
College, Laguna 4031
Email: akr@laguna.net

About twenty years ago, bioremediation, the art and science of harnessing the natural metabolic processes of everything from simple microorganisms to plants, to destroy or sequester contaminants, became a real option. The potential to engineer these processes created a new alternative to capital and management intensive mechanically-driven protocols and a thriving industry arose to serve this new strategic approach. We are now experiencing a maturation of this "era of bioremediation", which is further being accessorized with in-situ thermal and chemical oxidation processes for more rapid initial site impacts. Taken all together we are moving into an "era of in-situ treatment", but regardless of how one intervenes with thermal or chemical energy in a contaminated area, biological processes ultimately are needed to finish the operation. Further, there have been many exciting new developments in molecular biology that are now enhancing the science of bioremediation, such that with a more refined understanding of key biological players and processes we can better design and manage bioremediation engineering.

The Republic of the Philippines is like any other with a modern economy- it has severe environmental problems that have reached crisis proportions. However, as an island nation with limited territory, these problems have a special insidious feature; there is no place to run. Fortunately, with the help of powerful natural processes like bioremediation that work around the clock without supervision we have the tools to heal the environment cost-effectively. Consequently, the National Academy of Science and Technology formed a Bioremediation Research Team (BRT), which, after a series of meetings, decided to focus on unattended sources of hazardous wastes, with a special emphasis on the problems of the mining industry.

The issuance of Executive Order No. 270 – the National Policy Agenda on Revitalizing Mining in the Philippines, (January 16, 2004), and the favorable Supreme Court decision on the Philippine Mining Act 7942 of 1995, have led to the

revitalization of the local minerals industry, which had become nearly dormant prior to the decision. Such decisions elicited hope in both public and private sectors that a revitalized mining industry could boost the economy; however, there are two main issues attached to the mining industry: abandoned mines and the management of mine wastes. How can bioremediation help? Well, from the top down, we have phytoremediation which harnesses the healing aspects of plants for the more surficial aspects of these abandoned mines while subsurface engineered biological barriers, which rely on microorganism activity, address contaminated aquifers.

The BRT proposes to conduct a project on Field Test Applications of Phytoremediation and Microbial Technologies for the Rehabilitation of Contaminated Mine Sites. This project will utilize established heavy metal resistant plants. Jatropha curcas, commonly known as "tubang bakod", is a prime candidate as it thrives in marginalized land and the nuts can be used as source of biodiesel.

Other projects on "The Use of Local Bioremediation-based Technologies for the Management of Wastes from the Mining Industry" are also being proposed. The BRT, composed of microbiologists, chemists, botanists, foresters, and plant biologists, believes that bioremediation projects are worth pursuing should funds become available.

Keywords: bioremediation, waste management, mining, BRT, Philippine Mining Act 7942

PHILIPPINE BIODIVERSITY: ECOLOGICAL ROLES, USES AND CONSERVATION STATUS

Angel C. Alcala, I.E. Buot Jr., A. Diesmos, M. L. Dolar, E.S. Fernando J.C. Gonzalez, and B. Tabaranza[1]

Angelo King for Research Center and Enviornmental Management
Siliman, University, Dumaguete City
Email: suakcrem@yahoo.com

The Philippines has been recognized as having one of the world's mega-biodiversity centers for terrestrial and near-shore marine fish fauna. This is due to a number of factors: insular condition providing barriers to faunal and floral dispersal, isolated high mountain areas promoting high levels of endemism as a result of geographic isolation, tropical rain forests providing equable climatic conditions year round, and unique geological origin of islands. The paper deals with species of seed-bearing and non-seed-bearing flowering plants, freshwater and top carnivorous fish, amphibians and reptiles, birds, and terrestrial and marine

mammals. The main topics discussed are conservation status at the species level, values and uses of biodiversity, threatened and endangered species, and recommendations on their conservation.
**Names of authors arranged alphabetically

Keywords: Biodiversity, endemism, conservation, endangered species

BRIDGES OVER TROUBLED WATERS: CONNECTING REEF SYSTEMS WITH LARVAL DISPERSAL

Cesar A. Villanoy

Marine Science Institute, University of the Philippines
Diliman, Quezon City 1101, Philippines
Email: c.villanoy@gmail.com

Marine resources in the Philippines are in a state of rapid decline. Efforts to stem this decline are being made at different levels. A common management measure is the setting up of protected areas to conserve biodiversity and to ensure a supply of both larvae and adults from spillover effects to traditional fishing grounds. A network of such protected areas can potentially contribute more due to synergistic effects but designing such networks requires a variety of considerations such as larval dispersal trajectories and local hydrodynamics. Surface circulation patterns simulated from hydrodynamic models forced by mesoscale circulation, wind and tides are used to drive Lagrangian models of larval dispersal in selected areas around the Philippines. The larvae are represented as passive, neutrally-buoyant particles and attributes such as release location, age since release and settling location for each particle are stored. These attributes are used to build exchange matrices which show the degree of larval exchange and connectivity between reef areas and can be used in bio-economic models (e.g. FISH-BE) to parameterize larval spillover effects.

Keywords: Marine resources, reef system, larval dispersal, hydrodynamics

PRIORITIES IN THE BIOTECHNOLOGY INDUSTRY CLUSTER

Fortunato T. Dela Peña

Undersecretary for S&T
Department of Science and Technology
DOST Complex, Bicutan, Taguig City
Email: ftdp@dost.gov.ph

The Philippine National Science and Technology Plan (2002-2020) states that the development priorities in biotechnology are its applications in agriculture, forestry and natural resources, health/medical sciences, environment, energy and manufacturing and process engineering. Applications have been identified in these sectors as follows:

For agriculture and fisheries – varietal improvement and sustainable production for food security; for forestry – maintenance and maximization of forestry resources; for health/medical sciences – prevention, diagnosis and treatment of diseases; for marine sciences – development of marine bioindustries; for energy – development of renewable energy resources; for manufacturing and process engineering – development of novel processes of manufacture using life forms as miniature factories to produce the desired products and for environment – bioremediation.

The plan also indicates that the priority research and development areas for biotechnology processes and technologies are in the following: plant tissue culture; immuno-based diagnostics, protein and DNA markers; microbial transformations; plant transformations; genome mapping; cloning single genes; and mammalian tissue culture.

The sectoral R&D councils have formulated their medium term plans consistent with the NSTP priorities. In agriculture R&D in biotechnology is geared to generate breakthroughs such as fertilizer substitutes, genetically engineered plants and animals, biological control of pests and diseases, and plant and animal diagnostics. In industry R&D will be focused in the production on specialty chemicals which includes food additives and intermediate chemicals used in food and chemical processing more particularly enzymes, organic acids, polymers for films, coatings and flavoring agents. In health diagnostics which aid in the treatment of diseases as well as development of vaccines especially on emerging diseases and other infectious diseases have been identified.

Consistent with the above priorities and pursuing the strategy of niching and clustering the Department of Agriculture, Department of Trade and Industry and the Department of Science and Technology pushed the creation of a Biotechnology Cluster under the Export Development Council. This is seen as a

boost to the development and growth of the biotechnology industry in the country. The two subsectors identified with their corresponding lines of products and services considered to offer good opportunities are the agricultural and health bioindustry subsectors.

Keywords: biotechnology, agriculture, bioindustry, health, clusters

GLOBAL SITUATION OF AVIAN INFLUENZA (AI) IN HUMANS AND IN ANIMALS

Takeshi Kasai

Regional Adviser on Communicable Diseases Surveillance and Response
World Health Organization - Western Pacific Regional Office
Email: MINAJ@wpro.who.int

In 1997, earliest cases of AI in humans were reported in China, coinciding with outbreaks of highly pathogenic H5N1 in animals. From then, no human cases were reported until 2003 in Hong Kong (February) and Viet Nam (December). In 2005, cases of H5N1 in animals and humans spread from Asia to Europe, Africa and the Middle East.

From December 2003 to 20 June 2006, 228 human cases of AI were reported to the World Health Organization. Cases were distributed in four regions of the WHO: Western Pacific (52%), South-East Asia (32%), Europe (9%) and Mediterranean (7%). Ten countries were affected, Viet Nam has the most cases (41%) followed by Indonesia (22%).

Ages of cases ranged from 3 months to 81 years, (median 20 years, n=216). Twenty-six percents' were below 20 years old. Cases were almost equally distributed by gender (F=51%, M=49%, n=216). Thirty-two percents' (34/104 cases) were farmers. Two cases were pregnant. There were 130 fatalities (CFR 57%). Half were below 20 years old. Duration from date of onset to death ranged from 2 to 28 days (median 9, n=111). Indonesia has the highest CFR (76.5%) among countries with 10 cases and more. CFR trend has declined in early part of 2005 and has slightly increasing since then.

In 2003, Korea reported the first outbreak of H5N1 in poultry. In 2004, many countries in South-east Asia were affected. H5N1 spread dramatically in 2005 to East Asia, Central Asia, East Europe, and in 2006, to Africa. The OIE or FAO has recorded more than 40 countries that officially reported H5N1 outbreaks in poultry or wild birds as of 20 June 2006. Wild birds were considered responsible for the introduction of H5N1 virus at considerable geographical distance from known outbreaks in poultry. Trades were also believed as a factor.

In mid-2005, WHO and APEC assessed the status of country's pandemic preparedness. Since then, WHO spearheaded the acceleration of pandemic planning process, invited multi-sector participation, and advocating international collaboration. Some countries have shown progress through animal surveillance, culling/compensation and community awareness, but there remains much work to be done.

Avian influenza virus has already entrenched and is expanding to neighboring regions. The reported number of human cases is still small considering the size of the spread in birds. Increased exposure of humans to H5N1 resulting from global spread multiplies the opportunity to adapt or mutate. Containment of a pandemic influenza is possible only if earliest signals are promptly detected by surveillance and vigorous measures are implemented rapidly.

Keywords: avian influenza virus, H5N1,

AGRICULTURAL BIOTECHNOLOGY TRENDS AND NICHES FOR THE PHILIPPINES

Eufemio T. Rasco, Jr.

Professor, College of Science and Mathematics
University of the Philippines Mindanao
Email: eufemio_rasco@yahoo.com

The main objective of this paper is to assess the leading edges of today's knowledge in agricultural biotechnology at the global scale, and offer some recommendations on the possible niches of the Philippines. Until recently, biotechnology is neatly classified as agricultural (including forestry and aquaculture), health, industrial and environmental. Presently, however, a great revolution is going on. Agricultural biotechnology is invading the other fields of biotechnology! We can call this the third agricultural revolution. The first revolution started the process we now call civilization 10000 years ago; the second (the Green Revolution) saved civilization from hunger about 40 years ago. The third hopes to save us from the problems created by the first and second revolutions and provide the material needs of future generations in a sustainable manner.

The scope of agriculture is now being extended from provision of basic needs, namely, food, fiber and clothing to include needs of modern civilization such as energy, materials, drugs, and industrial products such as enzymes. The definition of agricultural crops is being extended to include not only higher plants, but all photosynthesizing organisms. Techniques traditionally used for industrial scale culture of bacteria and fungi are being applied for single cell, tissue and

organ cultures of higher plants and other photosynthesizing organisms. Thus, we are looking forward to a new generation of biofactories and production systems using photosynthesis as the main engine. These biofactories will produce traditional and non-traditional products cheaper, faster, safer and better. It is an exciting future with a lot of promises but many challenges and unknown perils, too.

The niche for the Philippines is dictated by the reality that its land area, the traditional basis of agriculture is limited. In addition, its climate is generally less favorable for traditional agriculture than many other environments. On the other hand, the Philippines has a huge surplus of unemployed manpower, sunshine and water. Review of recent literature suggests the following leading edges suitable for the Philippines for scientific and technological development in the field of conventional and modern agricultural biotechnology: 1) new agricultural crops that are less susceptible to the vagaries of local climate and limitations of arable land, 2) new approaches for recombinant DNA technology, specifically plastid engineering; and 3) bioreactors and less sophisticated production systems using higher plant cells and organ cultures, and other photosynthesizing organisms such as mosses and algae.

Scientific literacy is a prerequisite for the third agricultural revolution. A scientifically literate nation will formulate policies that will encourage innovation, deploy its best minds to the service of science and technology, and create a public that is receptive to new ideas. Even as we look to the future, the struggle for public acceptance of the third agricultural revolution is taking place today. There are existing biotechnologies waiting to be used, such as transgenic crops, livestock, forest trees and fishes. These will not prosper if public reaction and corresponding government regulation is guided by imagined risks rather than demonstrated benefits. The paper argues for a system of regulation that will achieve an appropriate balance between the need to assure the public of the safety of agricultural biotechnology and the imperative to explore new technology for solving the problems of modern living.

Keywords: agricultural biotechnology, biotechnology, green revolution, transgenic crops, recombinant DNA

POSTER SESSION

AGRICULTURAL SCIENCES

ASD No. 1

DEVELOPMENT OF BACTERIAL BLIGHT RESISTANT HYBRID RICE PARENTAL LINES THROUGH BI-DIRECTIONAL MARKER-AIDED SELECTION

Joan Marie S. Agarcio[1], Lucia M. Borines[2], Samuel A. Ordoñez[1], Marina P. Natural[3], Casiana M. Vera Cruz[4], Hei Leung[4], and Edilberto D. Redoña[1]

[1]Philippine Rice Research Institute, Muñoz, Nueva Ecija
Email: jmsagarcio@philrice.gov.ph
[2]Department of Pest Management, Leyte State University
Visca, 6521-A, Baybay Leyte;
[3]Department of Plant Pathology, University of the Philippines
Los Baños, Laguna, 4031,
[4]Entomology and Plant Pathology Division
International Rice Research Institute, Los Baños, Laguna

Although hybrid rice (*Oryza sativa* L.) technology has been widely adopted in China, it is relatively new in tropical Asia. Bacterial blight (BB), caused by *Xanthomonas oryzae* pv. *oryzae* (Ishiyama 1922) Swings et al. 1990 *(Xoo)*, has posed serious threats in the success of hybrid rice seed production and cultivation in the Philippines. To improve the level of resistance, BB resistance from donor lines carrying *Xa4*, *Xa7*, and *Xa21* genes were successfully introgressed into IR58025B, IR62829B, LianB, 913B and BoB maintainer lines. To further refine the selection process, bi-directional marker-aided selection (MAS) was utilized in selecting for the advanced generation (BC_4F_1) of IR58025 progenies. Morpho-agronomic evaluation facilitated by molecular markers enabled the selection to be more efficient. To determine the level of parental genotype recovery among the improved progeny lines, DNA fingerprints were generated. Results indicate a high level of similarity with the original parental genotype. While the rest of the improved maintainer lines will serve as potentially useful hybrid rice germplasm with enhanced level of bacterial blight resistance for future breeding purposes, the selected IR58025B and IR58025A with improved BB resistance can now be used to produce a BB-resistant *Mestizo*, the most widely used hybrid in the Philippine government's hybrid rice commercialization program. Moreover, IR58025A is the female parent of most hybrids released in Asia. With the AxB and AxR seed production technology

well worked out and found to be economically feasible, these BB-resistance enriched lines thereby promise to create both national and international impact.

Keywords: hybrid rice, bacterial blight, marker-aided selection, *Oryza sativa* L., *Xanthomonas oryzae* pv. *oryzae*

ASD No. 2

INULIN AVAILABILITY IN TRANSGENIC RICE

Rhodora R. Aldemita[1]* Salim Al-babili[2], Peter Beyer[2]

[1]Philippine Rice Research Institute, Science City of Muñoz, Nueva Ecija, Email: rraldemita@hotmail.com
[2]Albert-Ludwigs Universität, Freiburg, Germany
Email: peter.beyer@biologie.uni-freiburg.de

Inulin is a linear fructan abundant in 15% of flowering plants. Inulin lowers blood lipids, reduce serum triglycerides and most importantly enhance bifidobacteria and lactobacilli for increased Fe, Mg, Ca, and Cu absorption. The staple rice is a good target to contain inulin since it lacks many micronutrients needed in the human diet, most specially, vitamins, the essential amino acids and iron. The two inulin synthesizing enzymes saccharose-saccharose-1 fructosyltransferase (SST) and fructan-fructan-1-fructosyltransferase (FFT) were isolated from artichoke and obtained from Max-Planck Institute. The two genes were cloned in a binary vector containing the phosphomannose isomerase selectable marker gene and placed under two different endosperm-specific promoters. This binary vector pInulin 2 was introduced to *Agrobacterium tumefaciens* LBA4404 and used to transform immature embryos of Taipei 309. A total of 128 putatively transformed plants were obtained and 7 transformation events were confirmed through Southern blot analysis. Reverse transcriptase PCR of the T1 seedlings proved the presence of the 3 transgenes. Fructose analysis of the T2 seeds that is being continuously conducted in the University of Stuttgart showed that line 4-1 has 0.5 mmol/L and line 5-1 has 1.5 mmol/L inulin. Highly expressing lines that will be obtained will be seed-increased for possible feeding test in animals.

Keywords: inulin, rice, saccharose-saccharose-1 fructosyltransferase, fructan-fructan-1- fructosyltransferase, *Agrobacterium tumefaciens*-mediated transformation

ASD No 3

DEVELOPMENT OF LOCALLY-ADAPTED RICE VARIETIES WITH HIGH BETACAROTENE CONTENT IN THE GRAINS (GOLDEN RICE)

Antonio A. Alfonso*, Nelson S. Garcia, Christine Jade A. Dilla, Eleanor S. Avellanoza, Marlou C. Abalos, Robenzon E. Lorenzana, Rhodora R. Aldemita and Leocadio S. Sebastian

Plant Breeding and Biotechnology Division
Philippine Rice Research Institute
Maligaya, Science City of Muñoz, Nueva Ecija
Email: aaalfonso@philrice.gov.ph

A two-pronged strategy is being implemented to develop locally-adapted Golden Rice, a genetically modified rice capable of producing high amounts of pro-Vitamin A (beta-carotene) within the endosperm. In the first strategy, Golden Rice versions of popular varieties PSB Rc82 and Mabango1 are being developed through molecular marker-aided backcrossing with emphasis on foreground and background selection. In the second strategy, new elite Golden Rice lines that are resistant to tungro and bacterial blight diseases are being bred through multiple crosses, anther culture and marker-aided selection. A total of 26 cross combinations were performed using three Syngenta Golden Rice 1 events (with up to 8 mg/g beta-carotene content) as donor of the beta-carotene biosynthetic genes. All three events are in the background of US rice variety Cocodrie and have a single copy of the transgene cassette. Morphological evaluation and PCR assay using primers specific to the transgenes *phytoene synthase* and *phytoene desaturase* confirmed the hybrid nature of the F1 plants. BC1F1 progenies were produced by crossing the F1 plants from selected cross combinations to their respective recurrent parents. Variations in grain color (yellow) intensity, an indicator of beta-carotene content, were observed both in the BC1F1 and F2 grains suggesting gene dosage effect. Individual BC1F1 plants that will be selected based on genetic similarity to their recurrent parent and/or resistance to tungro and bacterial blight will be used for further backcrossing and selection. Locally-adapted Golden Rice varieties are envisaged as an additional strategy to combat the persistent problem of vitamin A deficiency (VAD) in the country.

Keywords: Golden Rice, vitamin A deficiency, betacarotene, marker-aided backcrossing, biofortification

ASD No. 4

EFFICIENT *IN VITRO* SEED CULTURE, GERMINATION, SHOOT REGENERATION AND MUTANT INDUCTION TECHNIQUES IN LANSONES (*LANSIUM DOMESTICUM* CORREA)

Renato A. Avenido*

Plant Cell and Tissue Culture Laboratory, Institute of Plant Breeding
College of Agriculture, University of the Philippines Los Baños, Laguna 4031
Email: raavenido@yahoo.com

Langsat or lansones (*Lansium domesticum* Correa) is a very popular fruit in South-East Asia. In this species parthenocarpy is the rule and apomixis as evidenced by multiple seedlings due to polyembryony is quite common. No breeding has been attempted, and propagation is limited to seeds and through grafting to achieve fruiting in half-time (5-7 years). Mutation breeding by ionizing radiation is a potent strategy to induce and select for improved traits particularly early fruiting, dwarfing, improved fruit quality and disease resistance in lansones. This study involved the establishment of *in vitro* culture requirements for aseptic seed culture, germination and shoot regeneration in lansones 'Paete' followed by gamma-irradiation of cultured seeds.

The first two replicated experiments involved the culture of 81 seeds in MS basal medium with 30 g L^{-1} refined sugar and 2 mg L^{-1} BAP (L1 medium). On the average, 86.6% germination was achieved in 30 d of culture with 1.5 and 2.1 shoots produced per seed at 30 and 60 d, respectively. The third experiment involved the comparison of germination and shoot regeneration as influenced by four culture media namely L0 (MS basal medium with no growth regulator), L2 (B5 basal medium with 1 mg L^{-1} each of BAP and GA_3, and L3 (WPM medium with 1 mg L^{-1} each of BAP and GA_3) with L1 medium as check. Higher % germination was achieved using L0 (92.9%) and L2 (92.6%) as compared with L3 (81.3%) and L1 (80.4%). However, earlier shoot germination was recorded in L2 (24.4 d) and L3 (25.2 d) as compared with L1 (34.6 d) and L0 (33.0 d). Highest number of shoots produced after 60 d was obtained in L1 (2.5 shoots) followed by L2, L3 (1.9 shoots) and lowest in L0 (1.5 shoots). Addition of BAP alone or BAP and GA_3 had significant effects on days to germination, number of shoots and shoot lengths. The fourth experiment involved exposure of cultured lansones seeds in L1 medium to gamma rays (0, 10, 20 and 40 Gy) followed by shoot regeneration. The number of shoots regenerated was significantly reduced from 2.6 (control) to 2.0 (10 Gy), 2.1 (20 Gy) and 1.6 (40 Gy). Shoot tip and nodal explants from irradiated seedlings were successfully advanced through micropropagation.

Keywords: langsat, *Lansium domesticum* Correa, micropropagation, mutation breeding, tissue culture, tropical fruit tree

ASD No. 5 ✓

MICROGRAFTING TECHNIQUE FOR SOMATIC EMBRYO RESCUE AND MUTANT RECOVERY IN AVOCADO (*PERSEA AMERICANA* MILLER)

Renato A. Avenido*, Julita G. Dimaculangan and Julieta N. Welgas

Plant Cell and Tissue Culture Laboratory,
Institute of Plant Breeding College of Agriculture
University of the Philippines Los Baños College, Laguna 4031
Email: raavenido@yahoo.com

The successful plant regeneration via somatic embryogenesis of the two Philippine avocado varieties 'RCF Purple' and 'Semil' at the Institute of Plant Breeding (IPB-CA, UPLB) has promoted the use of plant biotechnology through somaclonal variation and *in vitro* mutation as potential strategies for avocado crop improvement. In the last three years, we have produced over 250 shoot regenerants as putative variants/mutants following tissue culture and gamma-irradiation treatments. These materials are being micropropagated, and screening for genetic variations using molecular markers has started with very promising results. Among the challenges met were the limited recovery of rooted (bipolar) plantlets from somatic embryos, slow growth of shoot regenerants and the losses due to contamination and callusing problems among long-term avocado tissue cultures. Micrografting and *ex vitro* grafting are viable tools available to promote shoot growth of putative mutant lines and fast-track the screening process for mutant selection. For the purpose of developing avocado micrografting technique, we have used two zygotic embryo-derived shoot cultures namely 'Semil' as scions and the *Phytophthora* root rot-resistant and faster growing line 'Mainit' as rootstocks. Scion shoots with matching V-shaped base were inserted into slit made on the rootstock shoots, with grafts secured using sterile filter paper strip and thread. The micrografts were re-cultured in agar-solidified B5 basal medium with 1 mg L^{-1} each of BAP and GA_3. Depending on scion type, micrografting was 72-100% successful. A modified procedure of micrografting either zygotic and somatic embryo-derived shoots of 'Semil' onto rooted *in vitro* germinated seedlings as rootstocks resulted in 90 and 61% success, respectively. Growth of successful shoot micrografts in shoot cultures and in the rooted rootstocks are being evaluated for subsequent *ex vitro* grafting and potting out, respectively.

Keywords: Avocado, *ex vitro* grafting, mutation breeding, micropropagation, *Persea Americana*, shoot cultures, somatic embryogenesis, tissue culture

ASD No. 6

SEARCH FOR TUBER-ASSOCIATED PROTEIN GENES IN CASSAVA

Vivian A. Azucena[1] and Antonio C. Laurena[2]

[1]Division of Physical Sciences and Mathematics
University of the Philippines Visayas, Miag-ao, Iloilo;
[2]Institute of Plant Breeding, College of Agriculture
University of the Philippines Los Baños, College, Laguna 4031 Philippines
Email: aclaurena@yahoo.com

Cassava ranks second among the five major tuber crops in terms of worldwide production as an important source of starch or carbohydrates for human food, animal feed and industry. But the protein content of the cassava tuber is relatively low (1 to 2%) compared to other cereal crops such as rice and corn (8 to 12%). Conventional breeding efforts to increase protein content in cassava from 2 to 7% were successful but with major drawbacks such as poor agronomic traits and increased cyanide content.

A molecular approach to increase the protein content without such drawbacks is through genetic engineering. The primary requirement is to isolate a gene encoding a tuber-associated protein from cassava and regulate the expression of this gene using a strong constitutive plant promoter. Primers were designed and constructed based on the N-terminal sequence of a non-glycosylated 40 kDa globulin protein (pI of 6.5) isolated and purified from the cassava tubers. PCR and RT-PCR of genomic DNA and total RNA from cassava leaves/tubers generated a 600 bp PCR fragment. Based on DNA sequence analyses, two genes (CSV1 and CSV2) encoding putative tuber-associated proteins were identified. CSV1 is approximately 187 amino acids of 20.6–21.5 kDa molecular mass while CSV2 consisted of 183 amino acids with a molecular mass of 19.5 kDa. Phylogenetic analyses suggest that CSVI and CSV2 are more related to patatin than dioscorin, sporamin and tarin. Unexpectedly, the deduced amino acid composition showed that both are highly charged basic (cationic) proteins (pIs of 9.8 and 11.6). Two highly conserved domains (LSGRQ and WISAEFAL) were identified in both CSV1 and CSV2.

Keywords: cassava, *Manihot esculenta*, tuber proteins

ASD No. 7

TOWARDS MAP-BASED ISOLATION OF DOWNY MILDEW RESISTANCE GENE IN MAIZE

Hayde F. Galvez*, Alma O. Canama and Desiree M. Hautea

Institute of Plant Breeding,
College of Agriculture
University of the Philippines
Los Baños 4031 College, Laguna, Philippines
Email: hfgalvez@ipb-uplb.org.ph
hgalvez_4031@yahoo.com

Downy mildew caused by *Peronosclerospora philippinensis* Weston (Shaw) is one of the most important diseases limiting maize production in the country. The use of resistant varieties remains the most effective control measure against downy mildew.

Bulk segregant analysis (BSA) coupled with simple sequence repeat (SSR) and resistance gene analog (RGA) marker analyses was employed to saturate the quantitative trait loci (QTL) for downy mildew resistance (DMR) and to identify tightly linked SSR/RGA markers. Nine RGA markers were mapped in DMR QTL regions especially in chromosomes 2 and 3, and in the major QTL region in chromosome 8. RGA marker *srga3* mapped exactly within the interval of the major QTL, which is flanked by RFLP markers *umc150* and *asg52*. Four EST- derived SSRs were also mapped in this region and confirmed 100% linked to DMR by BSA.

The *srga3* fragment in maize is currently being cloned for sequencing. This is to characterize this putative plant resistance gene ortholog, develop DMR-specific DNA markers for marker-assisted selection, and to isolate the underlying R genes via a QTL map-based approach. RGA *srga3* marker is derived from a plant disease resistance gene sequence of soybean.

Keywords: maize, downy mildew resistance, QTL, disease resistance gene sequence

ASD No. 8

NEW TECHNIQUES IN HYBRID RICE CULTIVATION: PARACHUTE OR SCATTERED TRANSPLANTING METHOD

Rodolfo L. Castro[1], Romeo B. Gavino[1], Cheng Liangji[2], Carlos C. Abon[2,3], Emmanuel V. Sicat[2,3], Yuan Guangjie[2], Wang Chuntao[2], Tang Lixin[2], Huang Baizhang[2], Trojane J. Velasquez[2], Francis E. Mina[2], Maribel Managuit[2], Erickson Frediles,[2] Aldo Valdez[2] and Michael Dizon[2]

[1]Department of Agriculture,
[2]Philippine Sino Center for Agricultural Technology and
[3]Central Luzon State University,
Science City of Muñoz, Nueva Ecija
Email: rodytah@yahoo.com

Parachute or scattered transplanting is a new method used widely in China specifically in hybrid rice production. It is a technique where rice seedlings are grown in plastic trays, pulled or uprooted from the tray, with a soil ball and tossed like a parachute toy on a puddle field. In 2003, because of the interest shown by PhilSCAT researchers on the method, 30,000 trays were sent to the Philippines through the center. Thus formal research for the adoption of parachute transplanting has now started in the country.

This research aimed to compare three transplanting methods commonly used for hybrid rice production. It specifically aimed to seek the advantages of the parachute transplanting method over mechanical transplanting and conventional (manual) method in terms of yield and economics of production.

Parachute transplanting showed significantly better results than manual or conventional transplanting and mechanical transplanting in terms of number of productive tillers, number of tillers per hill, yield and net income. With the parachute transplanting method, the seedlings were able to reach maximum tillering faster and thus they had more productive tillers.

In the two-season trials, parachute transplanting gave the highest net income of P 23,657.34 for wet season (WS) and P 32,465.55 for the dry season (DS). Mechanical transplanting gave an income of P 22,827.74 (WS) and P 28,872.13 (DS) while conventional transplanting was the lowest as it registered P 19,710.32 (WS) and P 23,766.21 (DS).

Based on the two season trials, parachute transplanting is worth considering due to the following reasons: (a) seedlings can be established easier and (b) grow faster, (c) it promotes tillering (more number of tillers per hill and productive tillers), (d) it provides higher yield, and (e) higher net income.

Keywords: parachute, hybrid, tillers, panicles, rice cultivation

ASD No. 9

DESICCATION AND FREEZING TOLERANCE OF MANDARIN (*Citrus reticulata* Blanco) SEEDS

Olivia P. Damasco and Luzminda C. Refuerzo

Institute of Plant Breeding, College of Agriculture
University of the Philippines Los Banos, College, Laguna 4031
Email: oliedamasco@yahoo.com

Citrus seeds are classified as intermediate, thus, seed conservation under conventional storage conditions defined for orthodox seeds (3 to 7% moisture content (MC) at -20°C storage) cannot be applied. Cryopreservation is the only viable option for long-term storage of *Citrus* germplasm. One of the critical factors ensuring success in cryopreservation is the amount of water present in the cell prior to freezing. This study aimed to determine the moisture content that would allow seeds of mandarin species to survive cryopreservation without deleterious effects on germination and seedling recovery. The tolerance of seeds of mandarin (*Citrus reticulata* Blanco) varieties 'Calamandarin', 'Ladu', 'Szinkom', and 'Tai Cat' to desiccation and liquid nitrogen freezing was determined.

Surface sterilized seeds excised from mature fruits were dried over silica gel (100 g) in an airtight container for varying time intervals. Desiccated seeds were subjected to rapid freezing in liquid nitrogen (-196°), rapid thawing at 50 °C, and cultured onto MS basal medium for seedling recovery and germination.

Mandarin seeds desiccated to about 9% MC showed germination rates similar to fresh seeds (80 to 100% germination). Lowering the MC to <5% resulted to significant reduction in germination rates (<50% germination). The highest survival after cryopreservation, ranging from 21.5 to 27.8%, was observed only for seeds with 9.1% MC for 'Calamandarin', 6.1% MC for 'Szinkom', and 3.8% MC for Tai Cat', respectively. Among the different mandarin varieties tested, 'Ladu' showed the least survival (13%) after cryopreservation. Results show that mandarin seeds were moderately tolerant to desiccation (could be desiccated to ~9% MC without significant reduction in germination), but sensitive to liquid nitrogen freezing. Hence, cryopreservation using seed desiccation technique is not a viable option for long-term conservation of mandarin seeds.

Keywords: mandarins, cryopreservation, seed desiccation, citrus

ASD No. 10

FIELD PERFORMANCE OF SELECTED BBTV RESISTANT MUTANT LINES OF BANANA CV LAKATAN

Olivia P. Damasco*, Teodora O. Dizon, Judith B. Estrella, Leila S. Caymo, Efrelito Jay M. Guittap, Felipe C. Dela Cruz Jr. and Evelyn Mae Tecson-Mendoza

Institute of Plant Breeding, College of Agriculture
University of the Philippines Los Banos, Laguna 4031
Email: oliedamasco@yahoo.com

Banana Bunchy Top Virus (BBTV) is a major problem of the Philippine banana industry. It has gravely reduced yields for the small-scale farmers and even wiped out the Lakatan industries in some regions of the country.

Bananas are not amenable to sexual breeding because they are male and female sterile. Thus, bunchy top virus resistance cannot be introgressed into banana by conventional breeding methods. Gamma irradiation and *in vitro* technologies were used to develop BBTV resistance in banana cv Lakatan. Thirty two (32) selected BBTV resistant mutant lines obtained from the earlier study were micropropagated and planted in BBTV infected field under high disease pressure for confirmation of stability of BBTV resistance and evaluation of agronomic characters of Generation 1 and 2 (G1 and G2) mutant lines.

Of the 32 mutant lines evaluated, ten (10) promising lines consistently showed lower BBTV disease incidence compared with the tissue culture (TC) control plants 3 to 19 months after planting. The %BBTV-free G1 plants at harvest was significantly higher for the ten selected mutant lines (58.3% to 95.8%) compared with the TC control plants (32.4%). Yield parameters such as bunch weight, number of hands per bunch, weight per hand and number of fingers per hand were comparable with the TC control plants. Some selected mutant lines have shorter number of days from planting to harvest; harvest period of 15 to 32 days earlier than the TC control plants.

Keywords: gamma irradiation, *in vitro* techniques, BBTV resistance, mutant lines

ASD No. 11

IN VITRO TECHNIQUES FOR MICROPROPAGATION AND LONG-TERM CONSERVATION OF INDIGENOUS CITRUS SPECIES

Olivia P. Damasco* and Luzminda C. Refuerzo

Institute of Plant Breeding, College of Agriculture
University of the Philippines Los Banos, College, Laguna 4031
Email: oliedamasco@yahoo.com

Indigenous citrus species are important part of the culture and livelihood of the native and rural communities in the Philippines. These species are becoming endangered, and at present found only in the wilds and remote areas of the country. Collection, propagation and conservation are urgently needed for further utilization and possible commercialization of these species. This study aimed at developing *in vitro* techniques for conservation and use of indigenous citrus species.

Indigenous citrus species such as 'Gapas-gapas' (*Citrus hystrix* var *boholensis*), 'Kubot'/'Kulubot'/'Kalpi' (*Citrus* spp), 'Dalayap'/'Dayap' (native lime, *C. aurantifolia*) and 'Suwa' (*C. limon*) were collected in different areas in Luzon (Quezon, Isabela, Nueva Viscaya, Quirino) and Mindanao (Cagayan de Oro). *In vitro* techniques for micropropagation and long-term conservation through cryopreservation are being developed for the different species.

Somatic/nucellar embryogenesis in 'Gapas-gapas' and 'Kalpi' was obtained using immature and mature seeds cultured onto MS basal medium or MS basal medium supplemented with 2,4-D and BAP. Techniques for long-term conservation through cryopreservation of desiccated seeds were developed. The % germination after cryopreservation ranged from 68 to 75% for native lime 'Dalayap'/'Dayap', 74 to 93% for 'Suwa' and 34 to 80% for a range of 'Kubot'/'Kulubot' genotypes. Results of the study show that seeds of indigenous citrus species, though heterogeneous, could be cryopreserved and this technique would allow long-term conservation of these species.

Keywords: indigenous citrus species, cryopreservation, micropropagation, *in vitro* techniques

ASD No. 12

A CANDIDATE GENE SEQUENCE FOR BACTERIAL WILT RESISTANCE IN TOMATO

Hayde F. Galvez*, Josefina O. Narciso, Nenita L. Opina, Alma O. Canama, Eden Jane U. Tongson, Marivi G. Colle and Desiree M. Hautea

Institute of Plant Breeding, College of Agriculture
University of the Philippines Los Baños, 4031 Laguna, Philippines
Email: hfgalvez@ipb-uplb.org.ph; hgalvez_4031@yahoo.com

Tomato is one of the most important vegetable crops grown in the Philippines. Bacterial wilt is a serious production constraint in tomato production especially during the off-season planting. Genetic sources for bacterial wilt resistance (BWR) have been identified and quantitative trait loci (QTL) have been mapped. However, the BWR-QTL identified need to be fine-mapped and validated for use in marker-assisted breeding and to isolate the BWR genes.

Integrating mapped SSR as anchor markers, six (6) of the 12 linkage groups previously constructed for BWR were assigned to their respective chromosome numbers in the tomato genome. The major QTL for BWR (AFLP *af02*) was assigned in tomato chromosome 6. The QTL is being fine-mapped to possibly characterize and/or clone the underlying resistance gene. Through bulk segregant analysis (BSA), 20 RGA and one (1) SSR markers were identified linked to BWR.

Gene-specific markers are being developed based on *af02* marker. Based on sequence homology, *af02* was found to be highly identical with a gene sequence of the signal recognition particle (SRP54) specific protein of tomato, EST and other tomato exon sequences, and with a putative plant disease resistance protein of *Solanum demissum*. For further applications, SCAR markers were designed based on the sequence information of *af02*. These markers are being used to screen and validate the susceptible mutants induced by physical and chemical mutagenesis. The markers are also used as starting regions in the map-based isolation of BWR genes.

Keywords: tomato, bacterial wilt resistance, QTL, fine mapping, bulk segregant analysis, map-based gene isolation

ASD No. 13

GERMPLASM COLLECTION, MULTIPLICATION, MAINTENANCE AND EVALUATION OF NATIVE AND EXOTIC VARIETIES OF UBI AND TUGUI UNDER ILOCOS CONDITION

Noralyn B. Legaspi* and Beatriz S. Malab

Research and Development Directorate
Mariano Marcos State University
Batac 2906, Ilocos Norte
Email: rddirectorate@yahoo.com

The collection of different accessions started in 2001. To date, a total of 19 tugui and 26 ubi accessions have been collected and maintained at MMSU for conservation. Characterization using the IPGRI Descriptor for Yam was undertaken to establish authenticity. For tugui, the most prominent differentiating characteristics were vine color and tuber characteristics such as hairiness, shape, size and presence/denseness of thorns. On the other hand, variability of the ubi collections were apparent both on the foliage and on the tubers. These include leaf shape, color, density and the presence of aerial tubers. For the tubers, flesh color were either purple, white, yellowish, off-white, a combination of white and purple with either color as the primary flesh color, purple with a shade of white or white with a shade of purple. Among the collections, four tugui accessions coded as MMSU Tugui # 3, 6, 7 and 9 and two ubi accessions coded as MMSU Ubi # 2 and 4 were identified promising. These could yield an average of 4.59-5.54t/ha for tugui and more than 11 t/ha for ubi. In addition, these accessions are very acceptable to consumers. Moreover, the farmers' cultural management practices which only include planting and harvesting were documented and fine-tuned to develop a technology for increased productivity. With such, farmers could engage to a more profitable yam production, making the marginal and idle lands productive and thereby be used as a means to support the governments program on food security.

Keywords: yam, ubi, tugui, accession

ASD No. 14

INDIGENOUS SEMI-TEMPERATE VEGETABLES OF THE HIGHLAND CORDILLERAS

Lorenza G. Lirio*, Lito O. Ayyokad, and Joyce N. Paing

Semi-Temperate Vegetable Research and Development Center,
Benguet State University, 2601 La Trinidad, Benguet, Philippines
Email: lglirio@yahoo.com

Indigenous vegetables are hardy, nutritious and a storehouse of desirable traits. A thorough survey and inventory of the indigenous vegetables in the various municipalities in Benguet and Mountain Province and including Baguio City was undertaken through a participatory approach with the local folks. Each plant was closely studied and aptly described botanically.

This step hopes to come up with strategies for the genetic conservation, propagation and eventual commercialization of the most promising (in terms of nutrient content, hardiness, acceptability, etc) indigenous semi-temperate vegetables in the Cordillera. Results of the chemical analysis revealed that carbohydrates, fat, fiber, crude protein; vitamins, minerals, etc of the indigenous semi-temperate vegetables were comparable with those of the cultivated crops.

Forty-nine (49) plants belonging to 23 families were eaten as vegetables utilizing the tops or young shoots. Few make use of the fruits and flowers for food. Of the 23 families, a greater number (18.4%) belong to Asteraceae, followed by Solanaceae (10.2%), then Amaranthaceae (8.2%).

A very important output of this project is a handbook with the title "Indigenous Semi-Temperate Vegetables of Cordillera" which will surely boost the awareness and on the utilization of indigenous vegetables.

Keywords: indigenous, vegetables, semi-temperate, highland, cordillera, sustainable, conservation, propagation.

ASD No. 15

GERMINATION AND GROWTH OF COCONUT EMBRYOS

Pablito M. Magdalita[1], Olivia P. Damasco[1], Lolita D. Valencia[1], Abigail May R. Oropesa[1] and Stephen W. Adkins[2]

[1]Institute of Plant Breeding, College of Agriculture
University of the Philippines Los Baños, College, Laguna 4031
[2]School of Land and Food Sciences
The University of Queensland, Brisbane, Australia 4072
Email: pablitomagdalita@yahoo.com

Gibberellic acid (GA_3) concentrations ranging from 30–40 µM significantly promoted germination and fresh weight of 'Laguna Tall' embryos in comparison with the control. Shoot emergence of germinated embryos was significantly enhanced with the addition of 10–50 µM GA_3 but did not significantly promote root and shoot growth *in-vitro*. In addition, various in vitro soil support systems including coconut coir dust, vermiculite and garden soil were tested for suitability during in vitro acclimatization to improve growth of germinated seedlings in comparison with the hybrid protocol. These support systems had the same effect as the control (hybrid protocol) on the promotion of leaf production, leaf width expansion, leaf elongation, girth expansion and enhancement of leaf quality (greening of leaves). Seedlings acclimatized in vitro in coconut coir dust and vermiculite, and in the control had similar leaf quality four months after culture, but those in garden soil had pale green or yellowish leaves and had lower leaf quality rating. This quality of seedlings was carried up to the ex vitro acclimatization phase where seedlings previously acclimatized in vitro in coconut coir dust and vermiculite had greener leaves than those in garden soil. Percent survival of the ex vitro acclimatized seedlings previously grown in coir dust (in vitro acclimatization) and the control is higher (83.3%) than those acclimatized in vitro in vermiculite and garden soil (66.7%). Furthermore, the collective acclimatization method applied to newly ex vitro acclimatized seedlings, using a wooden box covered with a transparent plastic lid, produced better quality seedlings with greener and shinier leaves than was possible using the plastic bag method used in the hybrid protocol.

Keywords: coconut, gibberellic acid, in vitro soil

ASD No. 16

ORGANIC, VIRUS-FREE AND TRUE-TO-TYPE TISSUE-CULTURED GARLIC (*ALLIUM SATIVUM* L.)

Lilian F. Patena, Gloria S. Rodulfo, Lolita M. Dolores, Roberta N. Garcia, Jonathan L. Juanillas, and Ramon C. Barba

Institute of Plant Breeding, College of Agriculture
University of the Philippines Los Banos, Laguna
Email: lilian@patena.net
velet@laguna.net

Garlic is one of the most economically important crops in the Philippines. Average yield is low compared to other countries due to low quality planting materials which can be traced to its conventional method of propagation, i.e. by cloves. Traditionally, farmers use previous crop's harvest as planting materials on a continuous basis. This poses a high risk of transmitting diseases from one generation to the next and of losing varieties. To solve this problem, we developed the technology of producing virus-free and true-to-type garlic planting materials and adapted it for mass production of bulbs for distribution to farmers. In addition, we incorporated organic farming in increasing bulb production to add value to the crop.

The technology consisted of (1) improved tissue culture protocol (with cold pre-treatment and thermotherapy) to produce small-tissue-cultured bulbs (in vitro bulblets we referred to as G_0 bulblets), (2) Enzyme-Linked ImmunoSorbent Assay or ELISA to index the materials for presence/absence of virus, (3) isozyme (protein) markers to check if the tissue-cultured materials were true-to-type, i.e. without genetic variations from the initial materials and (4) field planting (using organic farming) of tissue cultured bulblets to produce more bulbs ($G_0 > G_1 > G_2 > G_3$) and increase bulb size. Small bulbs were obtained from in vitro bulblets in the first generation (G_1) while normal size of bulbs was obtained in the 2nd generation (G_2). Increase of planting materials from G_2 to G_3 was 6x for Ilocos White, 5x for Mindoro and 7x for Tan Bolter. While low cost, imported garlic is available in the market, what we have produced are bulbs which are disease-free, true-to-type and with added value of having been produced organically.

Keywords: *Allium sativum* L., garlic, thermotherapy, tissue culture, virus-free

ASD No. 17

SOMATIC EMBRYOGENESIS IN COCONUT (*COCOS NUCIFERA* L.) USING PLUMULE EXPLANTS OF 'MAKAPUNO', 'LAGUNA TALL', AND 'BAYBAY TALL'

Lilian F. Patena, Claudette N. Dacumos, and Ramon C. Barba

Institute of Plant Breeding, College of Agriculture
University of the Philippines Los Banos, College, Laguna
Email: lilian@patena.net
velet@laguna.net

Coconut is among the woody species which is very difficult to tissue culture. It is propagated only by seed. Through biotechnology, we are developing other systems for its propagation.

Sixteen batches of plumule explants from fresh and pre-germinated embryos of coconut var. 'Makapuno', 'Laguna Tall' and 'Baybay Tall' were inoculated onto four media for calloid formation and somatic embryogenesis. The media consisted of Y3 (Euwen's, 1976), BP (Barba and Patena, 2002), modified BP and MS (Murashige and Skoog, 1962) basal salts supplemented with different levels of 2,4-D, BAP and 2-iP. Fruit weight, embryo size, plumule and calloid weight, calloid color and degree of calloid formation were noted. 'Laguna Tall' had the highest increase in embryo length, followed by 'Baybay Tall' while 'Makapuno' remained very slow growing except in the first 14 days of pre-germination. Calloid formation was observed one month after inoculation. Calloid weight increase was greatest with 42-day pre-germinated embryos of 'Laguna Tall' and 'Baybay Tall' while the effect of media was variable on these varieties. Calloid weight increase in 'Makapuno' embryos was greatest using Y3 medium, then BP medium and last, MS medium. White calloids were observed on the three varieties using BP medium. Few to profused (rating of 1 to 3) calloid formation was observed using BP medium while cultures turned brown using Y3 and modified BP media.

Keywords: calloid formation, coconut, *Cocos nucifera* L., somatic embryogenesis,

ASD No. 18

TISSUE CULTURE OF DIFFERENT STRAINS OF 'CARABAO' MANGO (*MANGIFERA INDICA* L.) AND THEIR CHARACTERIZATION USING SSR MARKERS

Lilian F. Patena[1], Darshan Brar[2], Dinah D. Tambalo[2], and Ramon C. Barba[1]

[1]Institute of Plant Breeding, College of Agriculture
University of the Philippines Los Banos, Laguna
[2]International Rice Research Institute
Los Banos, Laguna
Email: lilian@patena.net, velet@laguna.net

'Carabao' mango is the variety of commerce in the Philippines, both for local consumption and for export. There are several strains of the 'Carabao' mango and SSR markers were used to characterize them.

Somatic embryos (1°, 2°, 3°, ... n°) were produced on a continuous basis using the protocol of Pateña, et al. (2002). Depending on the strain, primary (1°) somatic embryo induction ranged from 33 to 100% while succeeding somatic embryo proliferation after 23 months (subculture cycle 8, S8) ranged from 25-100%. Cotyledonary leaf and root formation was best in modified Gamborg's B5 and Barba and Patena's BP basal media supplemented with either Ki or BAP and maintained in lighted condition. An increase of 40-50% in frequency of shoot formation was observed in BAP-containing media. True leaf formation was obtained 4–8 weeks after subculture onto the regeneration medium compared to 1–2 years the previous years. 2,4-D inhibited root formation. Initial transplant of plantlets to soil was done and more plantlets are being regenerated for subsequent transplant. Approximately 6-gram leaf samples were used for DNA extraction. DNA extraction was better using yellowish green, young leaves and adding PVP and mercaptoethanol to the extraction medium. The modified CetylTrimethylAmmonium-Bromide (CTAB) Method used in genomic DNA extraction from rice leaves was successfully used for genomic DNA extraction of mango. Different primers were tested for DNA amplification using PCR and characterization of the different 'Carabao' strains using SSR markers is in progress.

Keywords: mango, *Mangifera indica* L., tissue culture, SSR markers

ASD No. 19

MICROSATELLITE POLYMORPHISM AND DISEASE RESPONSE TO BACTERIAL BLIGHT (*XANTHOMONAS ORYZAE* PV. *ORYZAE*) OF RICE AMONG RESTORERS, MAINTAINERS AND THERMOSENSITIVE GENIC MALE STERILE LINES IN PHILIPPINES HYBRID RICE BREEDING

Loida M. Perez[1]*, Edilberto D. Redoña[1], Merlyn S. Mendioro[3] Casiana M. Vera Cruz[4] and Hei Leung[4]

[1]Philippine Rice Research Institute
Maligaya, Science City of Muñoz, Nueva Ecija 3119
Email: lmperez@philrice.gov.ph
[2]Genetics and Molecular Biology Division, Institute of Biological Sciences
University of the Philippines Los Baños, Laguna 4031
[4]Entomology and Plant Pathology Division, International Rice Research Institute, DAPO 7777, Metro Manila

Many of the problems associated with different breeding programs based on phenotypic characterization of an agronomic trait can be eliminated by the use of DNA-based diagnostics. The analysis of genetic relationships among lines is necessary in determining widely diverse genotypes for developing heterotic hybrid combinations and ensuring wide genetic base of future hybrids that will be developed. In this study, 61 hybrid rice materials including 28 restorer (R) lines, 15 maintainer (B) lines, 6 thermo-sensitive genetic male sterile (TGMS) lines and 12 advanced breeding lines were analyzed for extent of genetic relationships in 64 microsatellite loci spanning the 12 rice chromosomes. UPGMA cluster analysis showed that both IRRI-bred and PhilRice-bred and developed B and TGMS were distinct in terms of genetic background. B lines of IRRI clustered at 59.2% genetic similarity and are separated from the B lines of PhilRice hybrid genepool except for IR73328B and IR78367B. The TGMS materials including IR68301s, IR73827-23s and IR73834s, grouped at 76.7% genetic similarity. IR73827-23s and IR73834s showed 96.4% genetic similarity in 64 loci and 129 alleles. TGMS1 exhibited 63.5% similarity with the TGMS lines from IRRI while TGMS4 and TGMS6 clustered at 90.1% similarity. All 61 parental cultivars were susceptible to *X. oryzae* pv. *oryzae* race 6, PXO99. IR62161-184-3-1-3-2R, pollen parent of NSIC114H demonstrated high level of resistance to *Xoo* race 10, PXO341 including IR68897B, IR79123B, IR78378B, PR3B, IR68301s (TGMS) and three advanced breeding lines.

Keywords: restorer, maintainer, TGMS, hybrid rice, *Oryza sativa*, bacterial blight, microsatellites, genetic diversity

ASD No. 20

STORAGE ROOT DEVELOPMENT OF FIVE VARIETIES OF SWEETPOTATO *(Ipomoea batatas)* UNDER LA TRINIDAD CONDITION

Leoncia L. Tandang* and Leah D. Pacio

Benguet State University La Trinidad, 2601 Benguet
Email: lltandang@yahoo.com

The storage root development of five varieties of sweetpotato was evaluated at the BSU-Institute of Plant Breeding (UPLB-CA) Highland Crops Research Station in Benguet State University, La Trinidad, Benguet from September 2004 to March 2005 to identify early and high yielding variety.

Sweetpotato storage root development increased with time of harvest from two to five months after planting exhibiting a sigmoid curve which varied among the five varieties of sweetpotatoes studied such as: SG 98-01-03, PSBSp 17, NSICSp-27 (Bengueta), Kalbo-oy and Haponita. Sweetpotato varieties showed significant differences on the number and weight of storage root per plot, diameter and length of storage root, plant weight and harvest index. SG 98-01-03 and PSBSp 17 significantly produced the highest number and weight of storage root , storage root diameter and plant weight. These varieties were identified early maturing and high yielding under La Trinidad, Benguet condition during September to March production season. Haponita was medium maturing variety while NSICSp-27 and Kalbo-oy were late maturing varieties based on the storage root diameter, weight of storage root per plant and computed total yield per hectare.

(SG-sweetpotato genotype; PSBSp-Philippine Seed Board Sweetpotato; NSICSp-National Seed Industry Council Sweetpotato)

Keywords: sweetpotato, storage root, development, early, medium and late maturing varieties

ASD No. 21

BIOEFFICACY OF TOBACCO (*Nicotiana tabacum* L.) SEED POWDER AGAINST CORN WEEVIL (*Sitophilus zeamayz*)

Shirley C. Agrupis and Shirley B. Gudoy

Mariano Marcos State University, Batac, Ilocos Norte
Email: shirleyagrupis@yahoo.com

The use of botanical pesticides for agricultural stored food stuffs is the most environment-friendly way to safeguard the consuming public from the risk of possible pesticidal residue poisoning.

The laboratory and *in situ* bioefficacy tests of tobacco seed powder (TSP) against corn weevil were evaluated. Dry tobacco seeds were powderized and different TSP quantities at 1, 3, 5, 7, 9, 11, and 15 g were prepared and mixed separately with 7 g of uninfected corn kernels previously placed in a petri dish. Percent (%) mortality of the test insects was evaluated 6 h after application and every 6 h thereafter for 72 h.

The 15 g TSP outperformed the other treatments including the positive control. The commercial insecticide and the 15 g TSP showed 100% mortality of the test insects after 66 and 72 h, respectively.

The *in situ* test used the best treatment from the laboratory experiment. The 15 g TSP was then tested and was evaluated in two preparations: the TSP in powder form and TSP in tea bag. Each of the TSP preparations was used to treat 100g uninfected corn kernels placed in a rectangular box of 6.5 x 4.5 inches dimension and which were previously infested with 30 healthy corn weevils. After one month observation period, TSP in pure form showed the highest and fastest insecticidal action at par with the TSP in tea bag including the commercial pesticide.

Furthermore, percent (%) damage of the corn kernels was least pronounced in pure TSP. Corn kernels treated with pure TSP gave 100% germination while those seeds treated with the commercial insecticide, negative control, and TSP in tea bag gave 98%, 45% and 82%, respectively.

Keywords: Tobacco seed powder (TSP), pesticidal, mortality, bioefficacy

ASD No. 22

ECOTYPIC VARIATION OF THE ASIAN CORN BORER, *OSTRINIA FURNACALIS* (GUENEE) POPULATIONS IN THE PHILIPPINES

Merdelyn T. Caasi-Lit[1], Myrna G. Ballentes[2], Reny G. Gerona[3], Reynaldo H. Legaste[4] Ma. Dulce A. Jovillano-Mostoles[5], Mercedes D. Masipequeña[6], Ma. Luz J. Sison[1], and Franco G. Asoro[1]

[1]Institute of Plant Breeding, College of Agriculture,
University of the Philippines Los Baños, Laguna 4031, Philippines
[2]Central Mindanao University, Musuan Bukidnon;
[3]Leyte State University, Baybay, Leyte
[4]Provincial Agriculture Office, Koronadal, South Cotabato;
[5]Camarines Sur Agricultural State College, Pili, Camarines Sur;
[6]Isabela State University, Echague, Isabela
Email: mtcaasi_lit@yahoo.com

Barrion *et al.* (1981) and Mendoza *et al.* (1994) reported differences in terms of morphometric, DNA and isozyme analysis in the Asian corn borer populations from Laguna, Leyte, Bukidnon and South Cotabato. There is the possibility also that other differing populations may be occurring in other corn growing areas. What is the significance or what are the implications of these possible different populations particularly on the management of the corn borer during production. Likewise, what is its implication in corn breeding program for ACB resistance and release of commercial varieties?

Each geographical region(s) has its own set of environmental conditions including climatic and other factors. Hence, a crop variety's responses may vary for each site. In corn, the expression of resistance may differ not only because of the abiotic factors in the agroecosystem but more importantly it is because of the varying levels of diversity among local populations of its primary pest, the Asian corn borer, Therefore, the differential varietal responses against the ACB can be a reliable and direct indicator in confirming the presence of significantly different populations of ACB in the major corn growing areas of the country. This study is aimed to establish the occurrence of these populations and assess their infective ability through the field responses of selected corn varieties in six major corn-growing areas of the Philippines.

This study was conducted in six (6) selected sites representing different corn growing areas as follows: Laguna, Isabela, Bicol (Camarines Sur), Leyte, Bukidnon, South Cotabato using the following corn varieties: Yellow corn hybrid (Opaque) – IPB 911, Yellow corn hybrid (Flint) – NK 8840, Sweet Corn – IPB Philippine Supersweet, White corn hybrid – C 818, Tiniguib, Lagkitan.

Wet season trial results showed that Philippine Supersweet was the most susceptible variety against ACB across locations followed by Lagkitan and Tiniguib. The most ACB resistant varieties were the two hybrids NK 8840 and C818. Intermediate response (tolerance to ACB feeding) was exhibited by IPB 911. NK 8840 and C818 had the highest yield followed by IPB 911. Philippine Supersweet followed by Lagkitan had the lowest yield.

Keywords: ACB resistance, IPB 911, Yellow corn hybrid (Flint) – NK 8840, Sweet Corn – IPB Philippine Supersweet, White corn hybrid - C 818

ASD No. 23

QUADRASTICHUS ERYTHRINAE KIM (HYMENOPTERA: EULOPHIDAE), A NEW INVASIVE PEST INFESTING, GALLING AND KILLING DAPDAP TREES (*ERYTHRINA* SPP.) IN THE PHILIPPINES

Ireneo L. Lit, Jr.,*[1,3] Juancho B. Balatibat[1] and Armando M. Palijon[2]

[1]Department of Forest Biological Sciences and [2]Institute of Renewable Natural Resources College of Forestry and Natural Resources, and
[3]Entomology Section, UPLB Museum of Natural History
University of the Philippines Los Baños, College, Laguna 4031, Philippines
*e-mail: illit_jr@yahoo.com

Dapdap trees (*Erythrina* spp.) are commonly planted as ornamental trees and their usually bright red flowers and attractive form make them important landscape material. In the last two years at least, however, many dapdap trees have died and only a few trees are left and struggling to recover. The culprit is an insect that causes masses of galls on leaves, stems and shoots, eventually defoliating trees and drying up new and old growth. Damage assessments place the percentage of mortality among trees from around 20 to as high as 60%. Affected plants range from seedlings to pole stage young trees to very mature ones. Recovery, if at all, is very slow and usually death of trees follows, often signaled by the final attack of scavenging maggots. This paper reports the identity of the species as *Quadrastichus erythrinae* Kim, a wasp belonging to the order Hymenoptera, family Eulophidae and recently described in 2004 from Singapore, Reunion Island and Mauritius. The species is believed to be Afrotropical in origin but its manner of introduction into the Philippines and the penultimate source is still unknown. The adult insect, its larvae and the nature, development and extent of damage are described and photodocumented. There are large- and small-type galls which seem to correspond with the sizes of the haploid males and diploid females, the species apparently exhibiting haplodiploidy like most Hymenoptera. This is the

third invasive insect species that entered the country in a span of less than 5 years, following the buff coconut mealybug, *Nipaecoccus nipae* (Maskell) and long palm leaf beetle, *Brontispa longissima* Gestro. Poor implementation and/or widespread non-compliance to quarantine rules and regulations are always suspect underlying causes of these new pest problems.

Keywords: dapdap, dapdap gall wasp, Eulophidae, forest insect pests, Hymenoptera, invasive species, *Quadrastichus erythrinae* Kim

ASD No. 24

IDENTIFICATION OF MUTATION SITES IN THE ACETOLACTATE SYNTHASE (ALS) GENE OF THE RESISTANT *Rotala indica*

Maria Estela B. Detalla[1]*, Kenji Usui[2], Kazuyuki Itoh[3] and Hiroyuki Shibaike[4]

[1]College of Agriculture, Central Mindanao University, Musuan, 8710 Bukidnon
[2]Institute of Applied Biochemistry, University of Tsukuba
Tsukuba City, Ibaraki, Japan
[3]Japan International Research Center for Agrobiological Sciences
Tsukuba City, Ibaraki, Japan
[4]National Institute of Agro-Evironmental Sciences, Kannondai
Tsukuba City, Ibaraki, Japan
Email: meabdetalla@yahoo.com

Rotala indica, a lowland weed, developed resistance to an ALS-ihibiting herbicide, bensulfuron methyl(BSM). Most studies on the resistance mechanism to ALS inhibitors report that resistance is due to mutations in the ALS gene. This study aimed to identify amino acid substitutions in conserved regions of the resistant *R. indica* ALS gene that could explain its resistance to ALS inhibitors.

First strand cDNA was synthesized from the total RNA extracted from leaves of the resistant and susceptible *R. indica*. These served as templates for the polymerase chain reaction (PCR) amplifications. The target was a 230-bp sequence that contains two conserved regions of the ALS Domain A. Amplification was verified by agarose gel electrophoresis, and the excised product was cloned uing the TA-cloning technique. Double-stranded plasmid DNA was purified and DNA sequencing was performed using an ALF autosequencer.

Comparing the partial DNA and amino acid sequences of the resistant and susceptible ALS, seven nucleotide substitutions were detected but these did not result in mutations within the conserved regions. There was, however, an obvious difference in a single amino acid right after the AFQETP region (Isoleucine$_{211}$ in the

R-biotype; $Threonine_{211}$ in the S-biotype). There was no amino acid substitution at $Proline_{197}$ of the AITGQVPTKVIGT and at $Alanine_{205}$ of the AFQETP sequences. Instead, a three-amino acid sequence after $Proline_{197}$ of the AITGQVP***TKV***IGT was found. This sequence is common for both biotypes but not with other plants. This is the first report of a unique sequence within this conserved region. It is not conclusive yet to state that the resistance mechanism of *R. indica* is attributed to that single amino acid substitution after the AFQETP region, but it can be hypothesized that it may have an influence on the responses of the weed to ALS inhibitors.

Keywords : *Rotala indica*, herbicide resistance, acetolactate synthase inhibitor, mutation, bensulfuron methyl

ASD No. 25

INSECT RESISTANCE MANAGEMENT (IRM) OF BT CORN IN THE PHILIPPINES

Bonifacio F. Cayabyab*[1], Wilma R. Cuaterno[2], Emiliana N. Bernardo[3], Lilian Teaño, Godfrey Ramon[4], Melvin D. Ebuenga[1], and Pablito G. Gonzales[1]

[1]Plant Quarantine Support Laboratory, National Crop Protection Center University of the Philippines Los Baños (UPLB), College, Laguna 4031; [2]Bureau of Plant Industry, San Andres, Manila; Corn R, D & E Network, UPLB; [4]Biotech Coalition of the Philippines, Diliman, Quezon City
Email: bfcayabyab@yahoo.com

Bt corn was commercially sold in the Philippines market in 2002. The Bt corn has the ability to withstand the Asian corn borer infestation as compared to traditional varieties and hybrids that are susceptible to it. In order to prolong the use of this new technology, insect resistance management (IRM) is needed as a proactive method on risk mitigation.

In 2005, the Bureau of Plant Industry, the technology providers namely: Monsanto, Pioneer and Syngenta, non-government organization represented by the Biotech Coalition of the Philippines, the academe through the U.P. Los Baños, selected local governments and corn farmers planned and joined efforts to address IRM issues. This group recently launched concerted actions related to insect resistance management on Bt corn (IRM) in the Philippines thru an awareness briefings to farmers in five provinces of Mindanao and four provinces of Luzon from July 18-27. A total of 926 Bt corn adoptors participated. They were informed on modern biotechnology and Bt corn. The 80:20 bag in a bag method to prolong

the technology of Bt corn usage was discussed. This insect resistance management strategy involves the planting of 80% Bt corn and 20% non Bt corn. Different planting designs were presented to the farmers. Later they were asked if they were willing to practice this IRM strategy or not. About 56.40% were not willing to implement such scheme while 43.60%. The figures are quite close to each other as far as the acceptance and non acceptance of 80:20 IRM strategy is concerned. The results of this briefing cum survey were used in the review of DA Memorandum Circular # 17 (S 2003) which provides for the requirements for the IRM of Bt corn in the

Keywords: Bt corn, insect resistance management, risk mitigation, alliance

ASD No. 26
PRODUCTION AND UTILIZATION OF *TRICHOGRAMMA EVANESCENS TRICHOGRAMMA* PARASITOIDS IN TARLAC

Bonifacio F. Cayabyab*[1], Pablito G. Gonzales[1] Manuelo V. Agsaoay[2], Marietta C. Soriano[2] and Ana Maglalang[2]

[1]National Crop Protection Center
University of the Philippines Los Baños, Laguna;
[2]Tarlac College of Agriculture, Camiling, Tarlac
Email: bfcayabyab@yahoo.com

This study is a joint undertaking among corn farmers, the Provincial and Municipal governments of Tarlac, the Tarlac College of Agriculture and the National Crop Protection Center - UP Los Baños on the production and use of Trichogramma in Tarlac. It is a continuation of the project on *Community Based Mass Production and Utilization of Trichogramma Parasitoids Against Lepidopterous Pest of Corn.*

The National Crop Protection Center which is now a part of the Crop Protection Cluster, College of Agriculture, UPLB, extended a technology transfer scheme to the Tarlac College of Agriculture in 2004 through a memorandum of agreement. Four staff and five students of the college were trained on the production and utilization of Trichogramma. The Tarlac College of Agriculture on the otherhand forged a partnership with the governor's office of Tarlac to provide farmers Trichogramma parasitoids as a part of an IPM package for corn borer.

Trichogramma evanescens Westwood releases were done three times during the 2004-2005 growing season. These were at 30, 45, and 60 days after corn emergence. Twenty five farmers tilling sixty four hectares of Dekalb hybrid corn used this technology . A total of 4,445 trichogramma cards were released.

As of April 2006 the information drive for farmers was continued at the Tarlac College of Agriculture. A total of 50 farmer leaders from ten towns of Tarlac participated. More farmers are targeted for this year. Meanwhile the production of Trichogramma using *Sitotroga* as host commenced last December 2005 in addition to the initial *Cocyra* host that was utilized. Likewise *Trichogramma chilonis* will be reared for use against corn earworm, tomato fruit worm and eggplant fruit and shoot borer.

Keywords: Central Luzon, corn borer, partnership, *Trichogramma*, Tarlac, corn

ASD No. 27 X

IDENTIFICATION OF MUTATION SITES IN THE ACETOLACTATE SYNTHASE(ALS) GENE OF THE RESISTANT *Rotala indica*

Maria Estela B. Detalla[1]*, Kenji Usui[2], Kazuyuki Itoh[3] and Hiroyuki Shibaike[4]

[1]College of Agriculture, Central Mindanao University
Musuan, 8710 Bukidnon
[2]Institute of Applied Biochemistry, University of Tsukuba
Tsukuba City, Ibaraki, Japan
[3]Japan International Research Center for Agrobiological Sciences,
Tsukuba City, Ibaraki, Japan
[4]National Institute of Agro-Evironmental Sciences, Kannondai
Tsukuba City, Ibaraki, Japan
Email: meabdetalla@yahoo.com

Rotala indica, a lowland weed, developed resistance to an ALS-ihibiting herbicide, bensulfuron methyl(BSM). Most studies on the resistance mechanism to ALS inhibitors report that resistance is due to mutations in the ALS gene. This study aimed to identify amino acid substitutions in conserved regions of the resistant *R. indica* ALS gene that could explain its resistance to ALS inhibitors.

First strand cDNA was synthesized from the total RNA extracted from leaves of the resistant and susceptible *R. indica*. These served as templates for the polymerase chain reaction (PCR) amplifications. The target was a 230-bp sequence that contains two conserved regions of the ALS Domain A. Amplification was verified by agarose gel electrophoresis, and the excised product was cloned uing the TA-cloning technique. Double-stranded plasmid DNA was purified and DNA sequencing was performed using an ALF autosequencer.

Comparing the partial DNA and amino acid sequences of the resistant and susceptible ALS, seven nucleotide substitutions were detected but these did not

result in mutations within the conserved regions. There was, however, an obvious difference in a single amino acid right after the AFQETP region (Isoleucine$_{211}$ in the R-biotype; Threonine$_{211}$ in the S-biotype). There was no amino acid substitution at Proline$_{197}$ of the AITGQVPTKVIGT and at Alanine$_{205}$ of the AFQETP sequences. Instead, a three-amino acid sequence after Proline$_{197}$ of the AITGQVP***TKV***IGT was found. This sequence is common for both biotypes but not with other plants. This is the first report of a unique sequence within this conserved region. It is not conclusive yet to state that the resistance mechanism of *R. indica* is attributed to that single amino acid substitution after the AFQETP region, but it can be hypothesized that it may have an influence on the responses of the weed to ALS inhibitors.

Keywords : *Rotala indica*, herbicide resistance, acetolactate synthase inhibitor, mutation, bensulfuron methyl

ASD No. 28

EFFICIENCY OF HIGH FREQUENCY RESONANT PEST-KILLING LAMP IN CONTROLLING COTTON BOLLWORM, *Helicoverpa armigera* (Hubn.)

A. D. Solsoloy, F. Calamaan, I. C. Paraoan,

Cotton Research Center, Cotton Development Administration
Batac 2907, Ilocos Norte
Email: aidasolsoloy@yahoo.com

The efficiency of the imported light trap from china, known as the high frequency resonant pest killing lamp for pest control was evaluated on cotton. Ten insect orders with various number of families were noted, namely, coleoptera (23 families), hemiptera (11 families), homoptera (2 families), hymenoptera (4 families), lepidoptera (6 families), odonata (1 family), orthoptera (4 families), dermaptera (1 family), diptera (4 families) and neuroptera (1 family), with all described for their trophic roles and some insects identified to the species level.
An increasing trend in *helicoverpa armigera* population was noted from 45 dap – 78 dap while *amrasca biguttula* population was minimal.

About 20% increase in seedcotton yield was obtained from cotton with the light trap over the comparative farm. Further, there was only one chemical spraying for cotton with the trap while the comparative farm received 3x sprayings, thus, savings of two sprayings and six labor man-days for the former. Partial budget analysis showed an advantage of ca. Php 2000 for the new technology vis-à-vis chemical insecticides.

It is highly recommended that the technology be marketed soon in the country through government intervention so that its merits as pest control tactic are harnessed for other economic crops.

Keywords: high frequency resonance lamp, cotton bollworm, seedcotton yield, chemical insecticides

ASD No. 29

PLANT RESOURCES USED AS BUILDING MATERIALS IN TINOC, IFUGAO CORDILLERA ADMINISTRATIVE REGION, LUZON, PHILIPPINES

Teodora D. Balangcod

Department of Biology, College of Science
University of the Philippines Baguio, Baguio City 2600
Email: balangcod@yahoo.com

The towering mountains and dense forests surrounding Tinoc, Ifugao, one of the provinces of the Cordillera Administrative Region, has made it one of the remotest areas in the country. As such, the Kalanguya inhabiting the area has preserved their culture. The tribe members have managed to survive the harsh life in the mountains. This study highlights their ingenuity on plant use especially those used as building materials. Information regarding traditional or indigenous use of plant resources among the Kalanguya was gathered using interview schedules, focused group discussions and direct observation. The information gathered from interviews was enhanced by field observations. Results showed that there are 38 plant species distributed in 36 genera and 29 families identified for building purposes. These plants are further classified into their specific uses, i.e. house construction, fencing, kitchen paraphernalia,baskets, chicken cages and coffin.

Key words: traditional or indigenous uses, Kalanguya, plant resources, Tinoc, Ifugao

ASD No. 30

PRELIMINARY ASSESSMENT OF LAND USE AND WATER QUALITY OF SOME LAKES IN SAN PABLO, LAGUNA, PHILIPPINES

Rene Juna R Claveria*, Teresita R Perez, Jewel Racquel S. Unson, Jillian T. Roque, Gillian Mari S. Bayugo, and Joselito B. Aquino

Department of Environmental Science
Ateneo de Manila University, Loyola Heights, Quezon City
Email: rclaveria@ateneo.edu

Land conversions brought about by increasing population and the apparent increase of agricultural, residential and commercial activities in San Pablo have affected the quality of the water resources in the area. Agriculture and built-up areas comprised the majority of land use. Their assessment generated new interpretative maps after integrating remotely sensed data such as Landsat image and aerial photographs to various land use studies and surveys.

The degradation of surface water quality was manifested by the physico-chemical analyses of sediments deposited in selected rivers and lakes. Sediments are important repositories of information regarding water quality and chemical composition of water bodies. Sediments from Palacpaquen Lake and adjoining rivers were assessed. The sulfate values of samples taken from different sites range from 47.79 to 292.29 ppm, phosphates from 0.79 to 5.53 ppm, nitrates from 146.98 to 370.50 ppm, and organic carbon from 0.98 to 4.56 ppm. The observed variability of nutrient concentrations were attributed mainly to anthropogenic activities around the lake and rivers which include agriculture, domestic and aquaculture.

Land use has also affected groundwater resources being one of the principal sources of drinking water. Untreated spring and well water are vulnerable to industrial, domestic and agricultural pollution. Spring and well water from selected lakes such as Sampaloc, Mohicap and Palacpaquen were monitored for total and fecal coliform contamination levels. There were varied levels of contamination. In most cases the microbiological water quality in the sampled sites did not pass the drinking water standards of the Department of Health and the Department of Environment and Natural Resources.

Proper land and water resource planning and management should be implemented to address the problem of water quality degradation in San Pablo.

Keywords: land use, water quality, San Pablo City, remote sensing, nutrient analysis, coliform analysis

ASD No. 31

EFFECTS OF FILAMENTOUS GREEN ALGAE (*ENTEROMORPHA INTESTINALES* LINN.) AND COMMERCIAL FEEDS TO MILKFISH (*CHANOS-CHANOS* FORSSKAL) FINGERLINGS REARED IN CAGES AT THE MARINE WATERS OF TIGUIS, PORO, CEBU

Lacbayo Michelle C.[1], Bensig , Quincyann P.[1]Oteda, Mirasol G.[1]Gonzales, Jonar D.[1] and Tanduyan, Serapion N.[2]

[1] Bachelor of Science in Fisheries student
Cebu State College of Science and Technology
Fishery and Industrial College
6050 San Francisco, Cebu Campus
[2] Professor I, Cebu State College of Science and Technology
Fishery and Industrial College
6050 San Francisco, Cebu Campus
Telefax: 032-4970318 e-mail: standuyan@yahoo.com

Bangus or milkfish being our national fish is the focus of some aquaculturists to be reared in any system of culture in order to meet the demands of the consuming public. Camotes Islands as one of the fishing grounds in the Philippines have areas where fishpen and cage culture can be done That is why this study was conducted in order to find out the effects of the filamentous green algae and commercial feed to bangus fingerlings in cages in terms of body weight, total length and weight to supply acclimated fingerlings to the prospective fishpen and fishcage owners.. This study was also conducted in order to find out the survival and mortality rate of the fish.

Bangus fingerlings were reared in cages and there were four treatments in the study with four replicates using Randomized Complete Block Design (RCBD). Treatment 0 (control) no feed given; Treatment 1 (fed with filamentous green algae) ; Treatment 2 (Commercial feeds) and Treatment 3 (Combination of filamentous green algae and commercial feeds). Feeding was done twice a day based on 25% of its biomass Sampling was done every 15 days for 1 1/2 months.

Results show that in terms of body weight, Treatment 2 has gained 7.1 grams followed by Treatment 3 which has a gain of 5.5 grams. Third is Treatment 1 which has 2. 7 grams and the last is Treatment 0 which is 2 grams. For the total length, Treatment 2 got first which has an increase of 4.5 cm, Treatment 3 has 3.4 cm; Treatments 1 has 2.7 cm and Treatment 0 has 2 cm respectively. For the body depth, Treatment 2 got first with and increase of 0.8 cm followed by Treatment 3 , Treatment 1, and Treatment 0 which is 0.4.cm For the survival and mortality rate , it shows that Treatment 2 and Treatment 3 has 100% survival rate compared to Treatment 0 and Treatment 1 which is 97.73%.

Results further show that there was no significant mean difference on the rate of growth of milkfish fingerlings in terms of body weight, body depth, total length and mortality rate.

Keywords: *Chanos chanos*, Poro, Cebu and *Enteromorpha intestinales*

ASD No. 32 31

TOLERANCE OF GIANT FRESHWATER PRAWN (*MACROBRACHIUM ROESNBERGII* DE MAN) POSTLARVAE TO HIGHER SALINITIES REARED IN AQUARIAAT THE LABORATORY OF CEBU STATE COLLEGE OF SCIENCE AND TECHNOLOGY ,SAN FRANCISCO, CEBU CAMPUS

Abellanosa, Melvin[1], Tumanda Harold[1], Mabulay Rey[1] Tampus Roy[1] and Tanduyan, Serapion[2]

[1] Bachelor of Science in Fisheries student Cebu State College of Science and Technology- Fishery and Industrial College
6050 San Francisco, Cebu Campus
[2] Professor 1
Cebu State College of Science and Technology- Fishery and Industrial College
6050 San Francisco, Cebu Campus
Telefax: 032-4970318 e-mail: standuyan@yahoo.com

The tolerance of the postlarva of giant freshwater prawn to higher salinity was studied using behaviour, movements, and feeding characteristics as indicators. There were five treatments in the study where Treatment 0 (control 0- 0.5 ppt); Treatment 1 (1-5ppt); Treatment 2 (6-10ppt);Treatment 3 (11-15 ppt) and Treatment 4 (16-20ppt). The postlarvae having a weight of 0.9 to 1.2 grams were stocked in the aquaria using 8 grams of prawn per liter of water as the ratio. Increase in salinity in each treatment was done gradually every two days by adding saltwater in each treatment measured by using refractometer until the desired salinity level in each treatment is reached. Movements, behaviour, feeding characteristics and mortality were observed throughout the experimental process.

Results show that the scattered swimming behaviour of the freshwater prawn has increased from 2. 5 cm per minute in the Treatment 0 (0-5 ppt) to 2. 9 cm per minute in the Treatment 4(16-20 ppt).Rapid up and down movements has reduced from17 times per second in T0 to 9 times per second in T4.Motionless behaviour has increased form 25 times per second in T0 to 27 per second in T4.As to the bodily movements, it showed that there was an increase of the movements of the walking legs from 21 times per minute in the T0 (control) to 29 times per minute in T4. Antennae movements have also increased fro 29 times per minute in T0 to 41

times per minute in T4. And on the swimmerets movements it decreased from 501 times per minute in T0 to 204 times per minute in T4.On the effects of salinity to the feeding characteristics of the shrimp it was found out that there was an increase on the time the fish reacted to the feed. That is from 15 seconds in T0; 48 seconds in T1; 120 seconds in T2; 58 seconds in T3 and 65 seconds in T4.For the survival rate it showed that the highest survival mean was in T0 which is 94.12 % followed by T4 which is 84. 98 %; T3 is 80.45 %; T1 is 80.44 % and the last is T2 61.26 %.

Results further show that increase in salinity has affected the physiological characteristics of the freshwater prawn that the higher the salinity, the faster are the movements of its antennae and walking legs. On the contrary slower movements of the swimmerets were observed at increasing salinities. Food consumption appears to be lower at higher salinity.

Keywords: Tolerance, Giant Freshwater prawn and Salinity

ASD No. 33

EFFECTS OF FIRST FOOD ON THE HEART OF 30-DAY OLD NILE TILAPIA *Oreochromis niloticus L.*

Annabelle A. Herrera*, Evangeline Jaravata and Marc Bueser

Institute of Biology, College of Science, UP Diliman, Quezon City
Email: annabelle.a.herrera@gmail.com

First food for fish culture is the initial feed material introduced to the fry stage of the fish upon yolk resorption. This study was conducted to determine if the quality of first food, based on five different diets has any effect on the histology of the heart of month-old tilapia. The diets used were: I – zooplankton Moina; II – 40% fish meal and 60% rice bran; III – commercial brand fry mash; IV – combination of I and II; and V – combination of I and III. After the treatment period of one month, the heart of the fish specimens were dissected out and processed for histological analysis. Thickness of heart layers and spaces between muscle fibers were compared to those of the control, the fish given diet I. Body weight, length and muscle fiber diameter were subjected to statistical analysis. Diet I produced the greatest length and weight while diets IV and V induced the formation of the largest muscle fiber diameters. Differences were statistically significant using SPSS ver 10.0.1 program.

Keywords: heart, tilapia, first food quality, fish meal, rice bran, commercial brand fry mash

ASD No. 34

LEAF-LITTER PATTERN OF MAJOR COMPONENT SPECIES IN SECONDARY TROPICAL RAINFOREST IN PHILIPPINES

Amelita C. Luna[1]* and Katsuhiro Osumi[2]

[1]College of Forestry and Natural Resources,
University of the Philippines
Los Baños 4031 Laguna, Philippines;
[2] Senior Researcher, Kansai Research Center,
FFPRI, Kyoto 612-0855, Japan;
Email addresses: trr472@yahoo.com; osumi@fsm.affrc.go.jp;

Continuous litterfall measurements were made from 1993-1997 at the secondary tropical rainforest of Mt. Makiling Forest Reserve, Philippines. Mean annual litterfall was 15.3 t ha^{-1} yr^{-1}, composed of 64.79% leaves, 18.68% woody materials, 3.46% reproductive organs and 12.99% trash. Total litterfall and total leaf fall exhibited a bimodal pattern while wood and trash fall have a unimodal litterfall pattern. The bulk of the litterfall consisted of the leaves and maximum leaf shedding was during dry season. The dominant tree species had a unimodal leaf pattern characterized by a semi-deciduous trees while evergreen trees showed a bimodal pattern of leaf fall. The major leaf producing species were *Celtis luzonica*, *Diplodiscus paniculatus* and *Parashorea malaanonan*. Based on dissimilarity grouping, phenological observations on the species and climatic characteristics, the study indicated that the tropical rainforest of Mt. Makiling Forest Reserve comprised primarily of evergreen trees and to a lesser degree, semi deciduous tree species. Compared with similar studies on other old growth and secondary forest in SouthEast Asia, the litterfall value of this study is unusually high. The pattern of leaf shedding was species-specific and varied from markedly seasonal to nearly continuous shedding with small seasonal fluctuations.

Keywords: bimodal, leaf-shedding pattern, litterfall, unimodal

ASD No. 35

THE COMMUNITY STRUCTURE OF LOGGED-OVER TROPICAL RAIN FOREST IN MT. MAKILING FOREST RESERVE, PHILIPPINES

Amelita C. Luna[1]* and Katsuhiro Osumi[2]

[1]College of Forestry and Natural Resources,
University of the Philippines Los Baños 4031 Laguna
[2] Senior Researcher, Kansai Research Center, FFPRI, Kyoto 612-0855, Japan
E-mail addresses: trr472@yahoo.com; osumi@fsm.affrc.go.jp

Community structure and species diversity were studied at a 4-ha tropical rain forest in Luzon Island, Philippines, 50 years after selective logging. The once selectively logged forest had high species diversity and complex guild structure. The 4-ha plot recorded 3,648 trees and palms of 179 species with larger than 5cm in diameter breast height (dbh). The dbh size of all trees and palms showed an inverse-J distribution with a maximum of about 150 cm. Species diversity indices for 4-ha plot fell within the range of the indices previously reported for old-growth tropical rainforest in Southeast Asia . The dbh size distribution in the population of each component indicated that the community was composed primarily of shade-tolerant species. Most species with more than 1% density or basal area of the plot total had an aggregate distribution, and nearly half of them showed a significant topographic preference. The smaller presence of dipterocarp species, which were the dominant in the original flora, indicated that the species have suffered heavy utilisation in the past, with the result that numerous non-dipterocarp tree species now formed a species-rich secondary tropical rain forest.

Keywords: community structure, guild structure, shade-tolerant species, species diversity

ASD No. 36

OPTIMIZING CONDITIONS FOR KABIR POULTRY RAISING

Reneecilia B. Paz de Leon

College of Science, University of Rizal System- Morong, Rizal
Email: renee_pdl@yahoo.com

Today, poultry raisers are facing crisis due to rising cost of antibiotics, feeds, labor, and infrastructure requirements. Birds if not fully supplied with medicines become vulnerable to disease and stress, thus, requiring expensive raising. An

attractive alternative for today's mass-produced industrial broiler is "Kabir", strong, antibiotic free chicken breed with firm and savory meat.

The study sought to verify Kabir's optimum responses when subjected to varying temperature and space conditions. Kabir's feeding, water, sanitation requirements and reproduction behaviors were also taken into account. Experimental research design and descriptive methods were used to establish the optimum temperature and space requirements for chicks ranging from 0-4 weeks.

Kabir is a dynamic breed of chicken whose ancestral basis can be traced from Israel. The breed is superior for meat conversion because of its rapid growth, good body conformation and efficient feed conversion. Study revealed that the optimum temperature depends on the age of the chicks. Temperature ranging from 33–35 °C gave best survival rate for week old chicks, 31–33 °C for two weeks, 29–31 °C for three weeks and 27–29 °C for four weeks. In terms of space, 1 m x 1 m is the most ideal space measurement for twenty chicks whose age ranges from 0–3 weeks.

The type of feeds depends on the age of the chicks. Beyond four weeks, Kabir can be exposed to a free range environment. One male Kabir is capable of breeding with 15 to 20 females. The breed begins lying after 26 weeks and can lay 200 eggs/cycle.

Kabir raising is a potential livelihood opportunity to families that desire to have an additional income, and a source of "native style" meat with low cholesterol table eggs.

Keywords: poultry, Kabir, feeding

ASD No. 37

DETECTION OF FOREIGN GENES INTRODUCED INTO *Zea mays* L. BY DUPLEX PCR ANALYSIS

April B. Cabang* and Cynthia T. Hedreyda

National Institute of Molecular Biology and Biotechnology
Albert Hall, Lakandula corner Jacinto Sts.
University of the Philippines Diliman Quezon City, Philippines 1101
Email: april.cabang@up.edu.ph

Polymerase Chain Reaction (PCR) - based detection of genetically modified organisms (GMO) requires the use of oligonucleotide primers targeting transgenes introduced into GM varieties. In this study, new primers were designed and evaluated by PCR that can be used as basis for developing a GM corn seed detection kit. Duplex PCR procedures were developed for detecting structural gene and terminator DNA sequences in three transgenic maize lines: insect-resistant Yieldgard®

MON810, glyphosate-tolerant Roundup Ready® GA21 and Roundup Ready® NK603.

Three sets of primers CrF/CrR, AbcF/AbcR and NosMf/Nosr targeting the *cry1Ab* delta-endotoxin gene from *Bacillus thuringiensis* subs. *kurstaki*, optimized transit peptide and modified maize enolpyruvylshikimate-3-phosphate synthase (*OTP-mepsps*) gene and Nos termination sequence from *Agrobacterium tumefaciens* respectively were used for the specific detection of recombinant DNA sequences in GM corn samples. One set of primers Zmf/Zmr was used to amplify the endogenous corn gene *zein* which served as an internal control target in non-GM IPBVar1 and GM corn. Duplex PCR combining: Zmf/Zmr and CrF/CrR primers resulted in the amplification of *zein* (589bp) and *cry1Ab* (400bp) fragments in Yieldgard® MON810, Zmf/Zmr and AbcF/AbcR produced the expected *zein* and *OTP-mepsps* (213bp) PCR products in Roundup Ready® GA21 and Roundup Ready® NK603, Zmf/Zmr and NosMF/Nosr primers generated the expected *zein* and *nos* (162bp) amplicons in all transgenic corn samples. The combination of primers targeting native and recombinant gene sequences amplified the target DNA segments distinguishing non-GM from GM corn varieties.

Keywords: genetically modified corn, transgenic maize, GM corn detection

ASD No. 38

MULTIPLEX PCR DETECTION OF FOREIGN GENES INTRODUCED INTO SOYBEAN (*GLYCINE MAX* L.)

Diana Rose E. Rafioa*, Trisha Hannah R. San Diego and Cynthia T. Hedreyda

National Institute of Molecular Biology and Biotechnology
College of Science, University of the Philippines Diliman, Quezon City, 1101
*Email: deranoa@up.edu.ph

Polymerase chain reaction (PCR) primers targeting foreign genes introduced into genetically modified (GM) soybean (*Glycine max*) were designed and used in this study for multiplex PCR detection of transgenic soybean Roundup Ready® GTS 40-3-2. Three primer sets that resulted in distinguishable profile for transgenic soybean Roundup Ready® soybean line GTS 40-3-2 were simultaneously used to amplify a 300-bp fragment traversing the cauliflower mosaic virus (CamV) 35S promoter and a modified form of the plant enzyme 5-enolpyruvylshikimate-3-phosphate synthase (*m-epsps*), a 173-bp fragment of the nopaline synthase (*nos*) 3' terminator, and a 430-bp native lectin fragment that is present in both GM and non-GM seed samples. As expected, only the lectin gene fragment was amplified in the non-GM control soybean IPB PSBSY2. The three primers were also tested on

non-GM soybean samples containing 5%, 2%, 1%, 0.5%, 0.1%, and less than 0.0.3% transgenic Roundup Ready® soybean. Detection limits for the 300-bp CamV 35S/*m-epsps* and the 173-bp *nos* terminator gene fragments by the multiplex PCR procedure used in this study were 0.5% and 1.0%, respectively. The three new primers designed and tested in this study can now be used for multiplex PCR detection of GM soybean containing the transgenes *CamV/EPSPS* and *nos* terminator and can be utilized in developing a commercial detection kit for such transgenes in GM soybean.

Keywords: genetically modified (GM) soybean, *Glycine* max, multiplex PCR, PCR detection limit

BIOLOGICAL SCIENCES

BSD No. 1

CHALLENGES IN THE STUDY AND CONSERVATION OF PHILIPPINE *RAFFLESIAS*

***Julie F. Barcelona[1,4], MaryAnn O. Cajano[2], Leonardo L. Co[3] and Danny S. Balete[4]**

[1]Philippine National Herbarium (PNH), Botany Division
National Museum, P. Burgos St., Manila, P.O. Box 2659;
[2]College of Agriculture Herbarium, Museum of Natural History
University of the Philippines Los Baños, College, Laguna 4031
[3]Conservation International-Philippines,
6 Maalalahanin St., Teacher's Village; Diliman, Quezon City 1101;
[4]Laksambuhay Conservation, Inc.
10241 Mt Bulusan St. Umali Subd., Los Baños, Laguna;
Email: BarcelJF@hotmail.com

Rafflesia (Rafflesiaceae) is a genus of plants obligately parasitic to *Tetrastigma* (Vitaceae), lianas of the lowland rainforests in tropical and subtropical Asia. To date, 21 species of *Rafflesia* have been described: one species in Thailand, eight in Indonesia (five in Sumatra and three in Java), eight in Malaysia, and four in the Philippines including the most recently described, *R. mira*, from Compostela Valley in Mindanao. Our recent explorations in Luzon have added two more new species. Three potentially new species, yet to be described, are currently known from photographic evidence only. Once formally described, they would make the Philippines the global center of *Rafflesia* with a total of at least nine species, the

highest species concentration relative to land area. The lowland rainforest, home to rafflesias, is the most threatened due to deforestation and conversion to other landuses. Because of their rarity, diversity, unique reproductive biology, and ecology, studies on Philippine *Rafflesia* have not yet gone beyond discovery, nomenclature, and mapping. Research on *Rafflesia* is currently being hampered primarily by the generally tedious and slow permitting process of the Department of Environment and Natural Resources or by the absence of competent botanists at the time of discovery. In order to make sound conservation action plans for this enigmatic genus and its host, collection and scientific studies of Philippine *Rafflesia* must be supported by all stakeholders. Furthermore, further explorations of the lowland rainforests in the Philippines should be facilitated and collaboration between Filipino and foreign parasitic plant experts should be encouraged. If the scientific studies remain to be hampered by the lack of collecting permits and the destruction and disappearance of the lowland rainforests in the Philippines is unabated, perhaps the diversity of this plant group may disappear even before all the species are discovered.

Keywords: conservation, lowland rainforests, parasite, Philippines, *Rafflesia*, *Tetrastigma*

BSD No. 2
BUTTERFLY SPECIES IN LOWLAND AND MOUNTAINOUS LOCALITIES

Bonifacio F. Cayabyab

National Crop Protection Center, Crop Protection Cluster
College of Agriculture, University of the Philippines
Los Baños, College, Laguna 4031, Philippines
Email: bfcayabyab@laguna.net

Butterflies have diminishing range due to habitat degradation brought about by land developments and deforestations. Observations regarding butterflies in lowland and mountainous locality will be elucidated in this paper. This study will show the disparity of species in the said localities and the reasons for such contions.

A typical developed lowland like in Los Baños, Tarlac and Pampanga neighborhood with few surviving flora will yield the following family and species: Family Pieredae: *Eurema hecabe, Catopsilia pomona, Catopsilia pyranthe, Catopsilia scylla, Leptosia nina, Appias olferna,* Family Lycaenidae: *--Zizina otis oriens, Eucrysops cnejus,* Family Papilionidae: *Papilio demoleus,* Family Nymphalidae: *Junonia almana, Junonia hedonia ida, Hypolimnas bolina, Melanitis leda leda,* Family Satyridae: *Bibasis gomata, Amathusia phidippus* and

Erionota thrax. In contrast mountainous surroundings such as Mt. Arayat and Mt. Makiling with remaining forest covers can yield more than one hundred species. These species seldom venture outside the said mountains. The host plants land use patterns and natural enemies affect the scarcity or abundance of a given species.

Keywords: Butterflies, family, species, lowland, mountainous, localities

BSD No. 3

THE AMPHIBIAN FAUNA OF MT. MALINDANG MINDANAO, PHILIPPINES

[1]Olga M. Nuñeza* and [2]Fritzie B.Ates

[1]Department of Biological Sciences, College of Science and Mathematics
MSU-Iligan Institute of Technology, Tibanga, Iligan City
Email: olgamnuneza@yahoo.com
[2]Department of Biology, University of the Philippines Mindanao, Davao City

The Philippines is one of the world's top 17 megadiverse countries, however, it is not en exception to the general trend of forest destruction. Mt. Malindang is one of the upland ranges where biodiversity has been severely threatened due to forest loss. To assess amphibian faunal species richness for better understanding and management of critical resources, fieldwork was conducted in Mt. Malindang, Mindanao from October 2003 until December 2004 in different vegetation types (mossy, montane, almaciga, submontane dipterocarp, lowland dipterocarp, mixed dipterocarp and plantation forest as well as agroecosystem sites) from elevation of 120 meters to over 1700 meters above sea level. Primary data were collected through opportunistic technique with the participation of Subanens (indigenous people in the area) as local researchers. Assessment revealed 26 species of amphibians, 11 of which are endemic species with seven found only in Mindanao. Nine species are in the threatened category where eight are vulnerable and one endangered. *Philautus surrufus*, a Mindanao endemic listed in the endangered category was found to be abundant in the Mt. Malindang. All endemic species captured were found to be forest dwellers. It is apparent from the results of this present study that despite habitat degradation in Mt. Malindang, species richness of amphibians is high where 26% of amphibians recorded in the Philippines and 74% recorded in Mindanao are found in Mt. Malindang, indicating the conservation importance of this mountain range.

Keywords: amphibians, endemic, species, richness, Mt. Malindang

BSD No. 4

SURVEY OF ENDEMIC, INDIGENOUS AND INTRODUCED (FOREIGN) SPECIES IN THE FRESHWATER ECOSYSTEMS OF NUEVA ECIJA

Annie Melinda Paz-Alberto*, Erlene S. Castro and Vincent Lainard E. Flores

Central Luzon State University, Science City of Muñoz, Nueva Ecija
Email Address: ralbrtco@mozcom.com

The study was conducted to describe, identify, classify and to determine the occurrence and diversity of the endemic, indigenous and introduced (foreign) species in the freshwater ecosystems of Nueva Ecija. It also aimed to determine the sources and level of impacts of environmental degradation in the freshwater ecosystems of Nueva Ecija.

This survey was conducted in five freshwater ecosystems of Nueva Ecija, namely, Talavera River in Talavera, Pampanga River in Palayan City, Tabuating River in San Leonardo, Paitan Lake in Cuyapo and Pantabangan Dam in Pantabangan. Three sampling stations were selected in each of the five study areas and in each station ten quadrats measuring 10 m x 10 m were laid out randomly. Freshwater fishes, mollusks and macroflora were collected in the five study sites. An interview with the residents and a survey questionnaire was distributed to the 30% of the total population of fishermen in each study area. An assessment for the environmental disturbance affecting the freshwater ecosystems of Nueva Ecija was done by means of a checklist to survey the sources and level of impact of environment degradation of the freshwater ecosystems.

Twelve (12) species of indigenous and endemic species were identified and classified in the different freshwater ecosystems of Nueva Ecija which include four (4) species of freshwater fishes, two (2) species of freshwater crustaceans, one (1) species of freshwater mollusk and five (5) species of freshwater macroflora. *Leiopotherapon plumbeus* (Ayungin or Lukaok) and *Glossogobius giuris* (Biya) registered the highest percentage occurrence among the identified freshwater animal species. *Ipomoea aquatica* got the highest percentage occurrence with regards to macroflora. Pantabangan Dam registered the highest species diversity value among the freshwater ecosystems surveyed.

Sixteen (16) introduced species are described, identified and classified in the freshwater ecosystems of Nueva Ecija which include seven (7) species of freshwater fishes, five (5) freshwater mollusks and four (4) freshwater macroflora. Nile tilapia "*Tilapia nilotica*" is the most common introduced freshwater fish while *Pomacea canaliculata* also called golden snail is the most common introduced freshwater mollusk in the selected freshwater ecosystems of Nueva Ecija. Moreover, *Eichornia crassipes* also known as water lily is the most common introduced freshwater

macroflora. Paitan Lake got the highest species diversity of introduced species which include fishes, mollusks and macroflora because the people near the lake are dependent on those species as their source of food and source of income.

Based on the study, pollution such as animal wastes and solid wastes, biological pollution of introduced or alien species, recreational development, establishment of the hydroelectric power plant, toxic chemical hazards and recreational development have significantly contributed to environmental degradation of the freshwater ecosystems in Nueva Ecija.

At present, only a few number of indigenous and endemic species are present and the introduced species dominate the native species in the freshwater ecosystems of Nueva Ecija. This condition may result into the extinction of the endemic and indigenous species and to the destruction of ecological balance in the freshwater ecosystems in the upcoming years as well as loss of biodiversity.

Keywords: Endemic species, indigenous species, introduced/foreign species, lake, river, dam, biodiversity, environmental degradation, biological pollution

BSD No. 5

ASSESSMENT OF MYCORRHIZAL DIVERSITY IN ABANDONED MINE SITES IN TOLEDO, CEBU FOR BIOREMEDIATION

Nelly S. Aggangan[1], Nelson M. Pampolina[2], Nina M. Cadiz[3] and Asuncion K. Raymundo[3]

[1]National Institute of Molecular Biology and Biotechnology
[2]Department of Forest Biological Sciences
College of Forestry and Natural Resources
[3]Institute of Biological Sciences College of Arts and Sciences
University Philippines Los Baños, College, Laguna 4031
Email: nea@laguna.net

A survey of mycorrhizal diversity associated with selected plant species within the 35,000-hectare Atlas Mines in Toledo, Cebu, was conducted with the goal of rehabilitating abandoned copper mines through bioremediation by utilizing indigenous plants and mycorrhizal fungi. The Atlas Mine site is generally covered by vegetation classified as a disturbed grass-shrub-agroforest plant community. Plants growing in marginal and stressed environments similar to that in mine sites usually are associated with mycorrhizal fungi. Plants were identified and rhizosphere soil (including fine roots) were collected in five sites delineated by the mining company where ten samples were randomly collected from each site. Mycorrhizal colonization was assessed after the roots were cleared with potassium hydroxide

and stained with tryphan blue. Mycorrhizal spores were separated from the soil using the wet sieving and decanting procedures and counted following the grid line intersect method.

In the survey conducted, out of the 50 plants, only five (i.e. *Lycopodium*, *Saccharum spontaneum*, *Nephrolepis*, *Acacia mangium* and *Stachytarpheta jamaicensis*) collected from two sites showed colonization by vesicular-arbuscular mycorrhizal (VAM) fungi, with infection ranging from 10 – 100%. All roots of kandi-kandilaan (*Stachytarpheta jamaicensis*) were colonized solely by *Glomus* sp. Kamachile (*Pithecellobium dulce)* harboured the highest spore density (2,575 spores/plant/30g soil), consisting of *Glomus*, *Acaulospora* and *Entrophospora.* Datiles (*Muntingia calabura)*, on the other hand, was the only plant associated with *Glomus, Gigaspora, Scutellospora, Acaulospora* and *Entrophospora,* but with low spore population. *Glomus* was the most prevalent among the VAM fungi. Fruit bodies of ectomycorrhizal (ECM) fungi: *Pisolithus, Scleroderma, Thelephora* and *Bolettellus* were found under *Acacia auriculiformis, A. mangium, Eucalyptus urophylla* and *E. camaldulensis. Pisolithus* was the most prevalent. These VAM and ECM fungi can be isolated and mass-produced as inoculants for the successful rehabilitation of mine sites. Nursery and field trials have shown that mycorrhizal fungi can enhance plant growth, promote survival in infertile and acidic soils and tolerance to heavy metals.

Keywords: mycorrhizal diversity, VAM fungi, ECM fungi, rehabilitation, mine sites

BSD No. 6

GROWTH AND HEAVY METAL ABSORPTION OF MYCORRHIZAL AND NON-MYCORRHIZAL *JATROPHA CURCAS L.* IN MARGINAL GRASSLAND AND MINE WASTE SOILS

Eusebeia Joy B. Mendoza[1], Clarice Meryl M. Ocampo[1] and Nelly S. Aggangan[2]

[1]University of the Philippines Rural High School and
[2]National Institute of Molecular Biology and Biotechnology (BIOTECH),
University of the Philippines Los Baños College, Laguna 4031
Email: nea@laguna.net

This study determined the effect of mycorrhizal inoculation on the growth of *Jatropha curcas L.* in 9 mine waste soils and compared it with that in marginal grassland and garden soil. Soil pH ranged from 2.4 to 6.8 and Cu concentration of 3.5 to 200 mg/kg soil. One-week old seedlings were inoculated with mycorrhizal fungus *Gigaspora margarita*. The initial height of the plants was measured a week

after transplanting and the succeeding height measurements were done once a week for ten weeks. At harvest, root collar diameter, leaf area, plant dry weight, mycorrhizal infection and Cu and P concentration and uptake of different plant parts were determined. Results showed that out of the 11 soils studied, only seven sustained growth of *Jatropha* throughout the ten-week period. Plants grown in garden soil exhibited the best growth and highest P uptake, followed by those grown in a grassland soil. The poorest growth was observed in plants grown in mine soils collected from Antamok, Benguet and Paracale, Camarines Norte. Mycorrhizal inoculation promoted better growth of *Jatropha* in all soils except in garden and grassland soils. Phosphorus and Cu concentrations and uptake were highest in the leaves and lowest in the roots which were comparable with that in the stem. This implies that mycorrhizal *Jatropha* grew better in heavy metal sites and may contribute more in the extraction of Cu and other heavy metals in grassland and mine waste soils than the non-mycorrhizal ones. Moreover, better growth of mycorrhizal plants will definitely mean more fruits where biodiesel can be extracted.

Keywords: heavy metals, mycorrhiza, *Jatropha curcas*, mine waste soil, marginal grassland, copper

BSD No. 7

MYCORRHIZAL FUNGI AND HEAVY METAL TOLERANCE OF FAST GROWING REFORESTATION TREE SPECIES

Nelly S. Aggangan[1] and Ma. Concepcion P. Segismundo[2]

[1]National Institute of Molecular Biology and Biotechnology (BIOTECH)
University of the Philippines Los Banos, College, Laguna 4031
[2]Lipa City National Science High School, Lipa City, Batangas
Email: nea@laguna.net

Rehabilitation and reforestation of heavy metal contaminated sites can significantly contribute to our country's economical, agricultural and ecological stability.

Three fast growing tree species were used in the experiment namely: *Acacia mangium, Acacia aulacocarpa* and *Swietenia macrophylla* and inoculated with either *Gigaspora margarita, Glomus etunicatum,* a commercial mycorrhizal inoculant "Mykovam" and mycorrhizal fungi from mine site coded as Paracale isolates. Mykovam is a soil inoculant comprising of *G. margarita, G. etunicatum* and *G. macrocarpum* whereas Paracale isolates were a mixture of *Glomus, Entrophospora, Scutellospora* and *Acaulospora.*

The study was conducted to determine heavy metal tolerant fast growing reforestation tree species, and to determine the growth promoting and ameliorating effects of mycorrhiza in fast growing trees grown in mine waste soil. The experiment was one in a Randomized Complete Block Design with 10 replicates.

Inoculated *A. mangium* seedlings showed higher height and biomass than the uninoculated ones. Mycorrhizal *A. aulacocarpa*, showed higher height, biomass and phosphorus uptake than non-inoculated ones. *Swietenia macrophylla* seedlings were significantly affected in their height and biomass but the phosphorus content was higher than the non-mycorrhizal counterpart. Inoculation with mycorrhizal fungi enhanced the heavy metal tolerance of *A. mangium, A. aulacocarpa* and *S. macrophylla* and could be potential species for the rehabilitation and reforestation of heavy metal contaminated areas.

Keywords: *Acacia mangium, Acacia aulacocarpa*, mycorrhiza, copper, heavy metals, mine sites, phosphorus, *Swietenia macrophylla.*

BSD No. 8

ANALYSIS OF HEAVY METAL UPTAKE OF SOME POTENTIAL PLANTS FOR PHYTOREMEDIATION IN AN ABANDONED MINE AREA

Nina M. Cadiz[1], Nelly S. Aggangan[2] Nelson M. Pampolina[3] and Asuncion K. Raymundo[1]

[1]Institute of Biological Sciences, College of Arts and Sciences,
[2]National Institute of Molecular Biology and
[3]Department of Biological Sciences
College of Forestry and Natural Resources
University of the Philippines Los Baños, College, Laguna 4031
Email: nm_cadiz@yahoo.com

Phytoremediation, is offered as a "green" solution to environmental problems and may be applied *in situ* or *ex situ*, to soils, sludges, sediments, other solids, or groundwater. The study primarily aimed to investigate the tolerance mechanism of selected dominant plants as potential phytoremediatiors thriving in an abandoned mine area in Toledo, Cebu.

Tissue analysis of the various plant organs of silver fern, malatungaw and Japanese acacia revealed their various capacities to take up high concentrations of copper (Cu), lead (Pb), cadmium (Cd) and zinc (Zn), the usual HM contaminants in a copper and gold mining sites. Japanese acacia and silver fern showed high concentrations of Cu in the roots. Japanese acacia, as a nitrogen fixer, has rhizobial symbionts while the silver fern has mycorrhizal root association. In spite of high

concentration (435.17 mg kg^{-1}) of Cu in the roots of Japanese acacia, it was notable that the metal was not translocated in the fruits. Moreover, the levels of Cd, Pb and Zn in the fruits were very much below the allowable limits. Malatungaw, on the other hand, showed the lowest uptake of the HMs among the dominant species in the area, and thus, may be considered as a true excluder of Cu, Cd, Pb and Zn.

Key words: heavy metals, tolerance, uptake, phytoremediation, abandoned mine area

BSD No. 9

PHYTOMONITORING OF Rhoeo discolor AS DETERMINANT OF AIR QUALITY IN TWO DIFFERENT SITES

Erwin P. Elazegui , Vanessa R. Garcia , Timothy Z. Riva

College of Science , Technological University of the Philippines, Manila
Email: ngsepe@yahoo.com

Phytomonitoring is the use of plants as determinants of environmental pollution. Plants respond to pollutants in the form of changes in certain parameters such as chlorophyll level , leaf-extract pH , relative water content and stomatal density. These changes can be measured and used as a reliable indicator of exposure to airborne pollutants . Phytomonitoring as its is called can thus be used as a possible alternative and substitute to the usual physico-chemical methods and costly instruments used in air quality monitoring.

The study focuses on the potentials of Rhoeo discolor as biological indicator of air quality by phytomonitoring methods . Sample leaves were collected in two sites , Laguna and Manila . Four phytomonitoring methods were used : total chlorophyll content , leaf extract pH , relative water content and stomatal density.

Results showed that sample leaves from manila have lower values of total chlorophyll content , 5.11 mg/g ; leaf extract pH 5.55 ; relative water content , 93.25% ; and stomatal density , 19.82 stomata per cm2 compared to those from Laguna total chlorophyll content 10.0 mg/g , leaf extract pH 6.68; relative water content 99.08% and stomatal density 29,519 stomata per cm2 . Based on the results , Rhoeo discolor is a good biological indicator of air quality.

Keywords: Phytomonitoring , Rhoeo discolor , biological indicator , stomatal density

BSD No. 10

DEVELOPMENT OF A STANDARD TOXICITY TEST PROTOCOL FOR THE MARINE ENVIRONMENT USING THE BLACK TIGER SHRIMP, *PENAEUS MONODON*

Emilyn Q. Espiritu*, Katrina Lynn B. de Guzman and Carlos Celso V. Quevedo

Department of Environmental Science
Ateneo de Manila University, Loyola Heights, Quezon City 1108
Email: eespiritu@ateneo.edu, katjadeguzman@yahoo.com.ph; morisato54@yahoo.com

Biological assays are an essential tool for the determination of potential adverse effects of xenobiotics in the environment. However, many of the protocols for toxicity testing use temperate organisms and could not be directly applied to Philippine studies. The study explored the potential of the marine Black Tiger shrimp, *Penaeus monodon*, as a test organism for toxicity testing. The investigations focused on the following aspects: (a) standardization of laboratory conditions; (b) determination of the appropriate larval stage for use in toxicity testing; (c) selection of the appropriate reference toxicant; (d) comparison of test sensitivity between the *Penaeus monodon* test and the Standard *Artemia franciscana* toxicity test; and (e) application of the developed test to environmental monitoring.

The results showed that the post-larval stage of the shrimp is the most suitable stage to be used for toxicity testing in comparison to the protozoeal and mysis stages. Moreover, among the toxicants tested (i.e. potassium dichromate, copper sulfate, and mercuric chloride), the organism exhibited the most stable response to potassium dichromate, $K_2Cr_2O_7$, which was consequently chosen as the reference toxicant. Inter-calibration tests done with the standard *Artemia* toxicity test showed that the *Penaeus* test is comparable in sensitivity with the ArTox test. With these promising results, the *Penaeus monodon* test can be a useful addition in the list of toxicity tests for the marine environment which are applicable to the Philippine conditions.

Keywords: bioassays; xenobiotics; toxicity testing; *Penaeus monodon*; *Artemia franciscana*; potassium dichromate, post larva.

BSD No. 11

GROUNDWATER QUALITY ASSESSMENT IN PAYATAS DUMPSITE, PHILIPPINES

Glenn L. Sia Su

Biology Department, College of Science, De La Salle University- Manila
Email: glss76@lycos.com

Groundwater is an important resource providing man with numerous uses. However, it has often been neglected and overly exploited. The concern over today's contamination of groundwater and surface water sources resulting from open dumpsites had been identified as a potential threat to Metro Manila's water supply. The study assessed and compared the groundwater quality of fourteen (14) selected wells continuously used in the "with" (Payatas) and "without dumpsite" (Holy Spirit) areas of Payatas estate, Philippines. Water quality monitoring and analyses of the bio-physico-chemical parameters: pH, total suspended solids (TSS), total dissolved solids (TDS), total coliform, turbidity, conductivity, salinity, nitrate-nitrogen, sulfate, color, total chromium, total lead and total cadmium were carried out for six consecutive months from April to September 2003, covering dry and wet seasons. Results showed that most of the groundwater quality parameters in both study areas were within normal water quality standards of the DENR-EMB and the DOH except for the observed high TDS (> 1000 mg/L), TSS (>50 mg/L), total coliform (MPN > 0) and low pH levels (< 6.5). No significant differences in the following parameters: nitrate-nitrogen, total cadmium, total lead, total chromium and total coliform were observed between the "with" and "without dumpsite" areas. The parameters: TDS, turbidity, conductivity, salinity and sulfate concentrations in the "with dumpsite" groundwater sources were significantly higher. The significant differences at the levels of the pollutants strongly indicate the existence of pollution at the Payatas open dumpsite. Continuous water quality monitoring is encouraged. Increasing the frequency of sampling and analyses on the study areas are needed to effectively monitor impact of dumpsites on the environment and human health.

Keywords: water quality, groundwater, dumpsite, Payatas, Philippines

BSD No. 12

ECONOMIC AND BIOTECHNOLOGICAL POTENTIAL OF MUNICIPAL SOLID WASTE: LESSONS FROM DUMPSITE SELECTION PRESSURES AND EMERGENCE OF UNIQUE MICROBIAL STRAINS

Franco G. Teves*, Suzanne E. Go and Aniline D. Sacdalan

Molecular Genetics and Microbial Technology Laboratory,
Department of Biological Sciences, College of Science and Mathematics
MSU-Iligan Institute of Technology, 9200 Iligan City
*Email: franco_teves@yahoo.com

We focused our study on a ten-hectare, thirty four-year old dumpsite in Iligan City, where approximately 120 tons of non-segregated municipal solid waste are dumped daily. A consequence of the diverse compounds present is the selection of a unique microbial biodiversity.

Four composite sampling sites were identified representing 1.0% of the whole dumpsite area. Soil samples were obtained and processed for the isolation of facultative and aerobic bacteria and fungi. Isolates were pre-screened for their production of bioactive compounds by testing their metabolites for anti-bacterial and anti-fungal properties. Selected microorganisms were tested further for the production of DNA-binding metabolites using a two-dimensional thin layer chromatography (2D-TLC) set up.

Of the 26 bacterial isolates, four exhibit potential for an industrial application, with nine of the 73 fungal isolates also showing the same. While common microbial strains can be purchased from culture collections at a very low price (PhP 250.00 to PhP 300.00), special and industrial strains can run from PhP 10,000.00 to PhP 20,000.00 per strain, based on average international prices. Granting that the ratio we obtained from our two-step screening is the actual picture when applied to the whole dumpsite area, we calculated a total of roughly 1,300 microbial isolates with diverse capabilities waiting to be discovered in the ten-hectare site. If we place the number of industrially useful strains to be only 10% of the total, we would get 130 special strains. Translating these figures to commercial value of the microbial strains would give an estimate of PhP 1,300,000.00 to PhP 2,600,000.00 from sales of the cultures alone. This new paradigm should be considered in order to give added value to what we refer to as municipal solid waste.

Keywords: municipal solid waste, microbial biodiversity, DNA-binding, 2D-TLC

BSD No. 13

UTILIZATION OF AN INDIGENOUS DYESTUFF FROM *Basella rubra* (ALUGBATI) AS MICROBIOLOGICAL STAIN

Lorna T. Enerva, Winrich A. Bati, Arby B. de Jesus, Angelito T. Uldo[1] and Julius L. Leaño Jr.[2]

College of Science and SciTech R and D Center[1]
Polytechnic University of the Philippines
Philippine Textile Research Institute[2]
Email: lornaenerva@yahoo.com

This study was undertaken to extract anthocyanin from Basella rubra berries and utilize the extract as microbiological stain. It is an inexpensive, indigeneous and abundant raw material.

The alugbati berries were macerated in a blender and extracted with 1% HCl in 95% methanol. The extract obtained was filtered and then concentrated. Thin layer and column chromatography methods were used to isolate and purify the anthocyamin. The samples were analyzed using infrared spectra and ultraviolet spectra. FT-IR revealed the presence of a hydroxyl group which is prominent in the structure of anthocyanin pigment at 3385cm^{-1}. The C=O stretching for aromatic ring were indicated by a peak at 1635.20 cm^{-1}, 1513.38 cm^{-1} for C=O bending, 1439.91 cm^{-1} for O-H bending, 1207.42 cm^{-1} for C-O stretching, 1151.92cm^{-1} for alkane and 1100.77 cm^{-1} for C-C stretching. The structure of anthocycanin was further established by the ε maximum of the ultraviolet spectrum at 510.0 nm.

For the application, the crude extract was used as a stain for *Staphyloccus aureus*, a gram positive bacteria and *Escherichia coli*, a gram negative bacteria. The staining process for the microorganism used mordants like potassium alum, calcium oxide and copper sulfate for fixing the color. Only copper sulfate and lime responded positively as a mordant that gave favorable outcome in fixing the color of alugbati. The samples were screened based on the criteria of color retention and evenness. The structure of the microorganisms with respect to shape and size and certain cellular components were identified using a microscope and photomicrographs. The alugbati extract produced stain that was comparable with synthetic stains like crystal violet and safranin and can, therefore, be used as an alternative stain.

Keywords: anthocyanin, microbiological stain, mordant, dye.

BSD No. 14

PLANT DIVERSITY AND ABOVE-GROUND CARBON BUDGET OF BROADLEAF SPECIES ALONG NORTHEASTERN SLOPE OF MT. MAKILING FOREST, LUZON PHILIPPINES

[1]DIXON T. GEVAÑA and [2]NELSON M. PAMPOLINA

[1]Environmental Forestry Program. College of Forestry and Natural Resources
Email: enfor@laguna.net,
[2]Department of Forest Biological Sciences,
College of Forestry and Natural Resources, University of the Philippines
Los Baños, College, Laguna 4031;
Email: nelmpampolina@yahoo.com

This study aims to assess the plant diversity resources and carbon budget along elevation gradient in Mt. Makiling forest. Three elevation ranges were identified along the northeastern slope of the mountain: low elevation (50-300 masl), middle elevation (301-700 masl), and high elevation (701-1114 masl). Within these ranges, vegetation samplings for broadleaf were done on tree, intermediate, and wildling layers using the Quadrat Sampling Technique. A total of 110 species, 75 genera and 35 families were identified. Family Moraceae and genus *Ficus* have the highest number of occurrence in all elevation ranges, while *Swietenia macrophylla* was the most abundant species that had occurred from low to middle elevation. Vegetation and biometrics were generally denser in low and middle elevation while partially open and low on high elevation. The ecological values also showed that density, frequency and dominance were higher on these elevations where *Parashorea malaanonan* and *Pterocymbium tinctorium* had the highest Importance Value (IV) recorded. The diversity values in all ranges were generally low to moderate. The highest diversity was observed in high elevation that exhibited a trend of increasing from low to middle elevation then decreasing from middle to high elevation. Similarly, aboveground biomass and carbon density exhibited a same trend and found highest at the middle elevations where plantations of *S. macrophylla* and Dipterocarps are located. The highest estimate for carbon density was 451.62 ± 50.07 at 400 masl while lowest at the 900 masl with 94.58 ± 24.12. Overall, plant diversity resources and the corresponding potential carbon sequestration are important ecological values that should be considered as input in sustainable management of Mt Makiling forest.

Keywords: Plant diversity, broadleaf species, carbon budget, Mt Makiling Forest

BSD No. 15

DEVELOPMENT AND QUALITY CONTROL OF A 10% TINCTURE OF *IPOMOEA MURICATA* L. (JACQ.) (CONVOLVULACEAE)

***Sandra Cases Sy[1,2,3] and Marina O. Osi[2]**

[1]Faculty of Pharmacy, [2]Graduate School,
[3]Research Center for the Natural Sciences,
University of Santo Tomas, España, Manila
Email: scsy@mnl.ust.edu.ph

Phytochemical and clinical studies have shown that the seed of *Ipomoea muricata* L. possessed promising antibacterial activities and analgesic effects. This investigation dealt with the quality control of a tincture developed from the crude ethanolic extractof the dried seeds of *Ipomoea muricata* L. for purposes of standardization in the interest of monograph preparation and drug formulation. The 10% tincture is a thin, yellow liquid turning brown to dark brown with age; with a tamarind-like and slight alcoholic odor. It has a specific gravity of about 0.9185 and a pH of 5.70. It had a mean potency of 9.89% (specification: 9-11% w/v) when assayed by a spectrophotometric method developed in the course of the research. Such method was validated for precision, accuracy and linearity.

The assay procedure may not be stability-indicating since regression analysis of accelerated stability data revealed inconsistencies of correlation data that made them unsuitable for predicting shelf life using Arrhenius equation. In view of this, a long-term stability study remains the best approach to determine stability. Based on real-time data of 12 weeks, the tincture has a predictive shelf life of 18.5 weeks.

As final output of this study, a monograph on the finished product "Tinctura Ipomoea Muricatae Semen" (Tonkin Seed Tincture) has been generated.

Keywords: *Ipomoea muricata* L., tincture, TLC profile, ultraviolet spectra, validation, stability, monograph

BSD No. 16

SUPERCRITICAL (SC)-CARBON DIOXIDE (CO_2) EXTRACTION AND FRACTIONATION OF BIOACTIVE METABOLITES FROM *Ganoderma applanatum*

James G. Mercado[1], Aldrin Wendell H. Suarez[1], Roberto M. Malaluan[2] and Franco G. Teves[1]*

[1] Molecular Genetics and Microbial Technology Laboratory,
Department of Biological Sciences College of Science and Mathematics
[2] Department of Chemical Engineering Technology,
School of Engineering Technology
MSU-Iligan Institute of Technology, 9200 Iligan City;
* Email: franco_teves@yahoo.com

Traditional solvent extraction of bioactive compounds from biological materials such as the shelf fungus *Ganoderma applanatum*, usually requires an array of extraction solvents. The use of supercritical-carbon dioxide (SC-CO_2) demonstrates a time-and-cost efficient procedure for extraction of polar and non-polar components of *G. applanatum*.

Fruiting bodies of the shelf fungus were collected from a semi-forest area in Tandag, Surigao del Sur, cleaned of extraneous materials, and ground using a pre-washed laboratory mill. Extraction of the polar fraction was done by setting the temperature at 40 °C, a pressure of 300 atmospheres (atm) and a flow rate of 0.5 m^3/h at normal conditions (T = 25°C, P = 1 atm). The non-polar fraction was obtained using a temperature of 40 °C, a pressure of 120 atm, and a flow rate of 0.5 m^3/h. Total extraction time was four hours each, for the polar and non-polar fractions.

More than 50 ml each of the polar and non-polar fractions were obtained from separate 100 grams ground samples of *G. applanatum*, sufficient volume for use in the various bioassays. Overall, the extraction period using SC-CO_2 was much shorter (total of eight hours) compared to the traditional solvent extraction (36 to 48 h). Moreover, usually only amounts in milligrams are obtained after concentration by rotary evaporation. With SC-CO_2, concentrated amounts in solution are readily obtained. Additionally, since toxic and obnoxious chemical solvents are no longer employed, SC-CO_2 proves to be a cheaper and safer way to fractionate metabolites and extracts for bioactivity testing. In so far as literature is concerned, this is the first report on the use of SC-CO_2 extraction of metabolites from *G. applanatum*.

Keywords: Supercritical-CO_2, *Ganoderma applanatum*, bioactive, bioassay, metabolites

BSD No. 17

EVALUATION OF LEAF EPIDERMAL CHARACTERISTICS OF SOME *PLUMERIA SPECIES*

Vivian S. Tolentino, Haziel Adrielle P. dela Rosa*, and Vivian A. Panes*

Department of Biology, School of Science and Engineering
Ateneo de Manila University, Loyola Schools, Quezon City
Email: vtolentino@ateneo.edu

Plumeria species commonly called Kalachuchi are generally planted for ornamental purposes, and are now widely cultivated throughout the country because of their resilience, robustness and versatility in different types of environments. In some countries, it is cultivated for its fragrance for perfumes, but is rarely cultivated for its medicinal properties. Plumerias are commonly found in sacred places, such as churches, monasteries, temples and even in schools. Nowadays, they are usually found in roadsides and in parking areas. It is for this reason that the main objective of this study is to look into the leaf epidermal characteristics of the plant which may be responsible for the wide adaptability of this plant in different environments. The main objective of the study is to characterize the leaf anatomy of the epidermises of the different varieties of Plumeria variants. The different cells of the epidermises, such as the hairs or trichomes, stomatal apparatus, its location, stomatal index and type were studied. Histological techniques such as leaf clearing, epidermal imprints and cross-sections of leaves using free hand, sliding and rotary microtome were used. Illustrative slides were used for anatomical characterization of different epidermal structures, observed and photomicrographed using the BH-epiflourscence and the CK-2 inverted Olympus microscopes. Results showed that both the upper and lower epidermises are cutinized. Stomata are found on both epidermises, which is amphistomatic, and stomatal type is anomocytic. Trichomes are present on both epidermises and are multicellular. Trichome length ranges from 10-20 mm in length and distributed in clusters and rarely in isolation. These structural features of the leaf epidermises may be responsible for the adaptability of Plumerias to different environments especially along dusty, open, arid roadsides.

Keywords: *stomata, trichomes, amphistomatic, anomocytic, multicellular, epidermis*

BSD No. 18

INDUCTION OF CALLUS IN LEAF EXPLANTS OF *Lagerstroemia speciosa* (L.) Pers. AND DETECTION OF COROSOLIC ACID

Maria Reichenber C. Arcilla[1], Lilian B. Ungson*[2,3], and Almario D.J. Baltazar[4]

[1] Faculty of Medicine and Surgery, University of Sto. Tomas Espana St., Manila
[2]Institute of Biology, College of Science and
[3] Natural Sciences Research Institute, University of the Philippines Diliman
[4]USE TECHNO Corp. 11-30 Mekong St., Riverside Subd. Angeles City Pampanga
*Email: lilian.ungson@up.edu.ph

Lagerstroemia speciosa (L.) Pers. (Family Lythraceae) is a deciduous tree that grows in the Philippines, India, and other subtropical and tropical countries. This species, commonly known as crape-myrtle and locally called "banaba" in the Philippines, is known to have hypoglycemic effects. Corosolic acid is among its active components. This study aimed to induce callus development in leaf explants *in vitro* and to detect presence and concentration of corosolic acid.

Leaf explants from one-year old seedlings were treated with ascorbic acid to prevent browning prior to surface sterilization with 70% ethanol (EtOH) and 20% sodium hypochlorite (NaOCl). Leaf explants were grown in Murashige and Skoog (MS) medium supplemented with naphthalene acetic acid (NAA) and kinetin (KIN) at combinations of 5 iM and 20 iM of NAA and 0 iM, 1 iM, 5 iM, and 20 iM of KIN. Ten explants were used per treatment. Data were collected on abundance of callus pinheads (calli < 1mm dia.) and clumps (calli e" 1mm dia.).

Calli developed on the explants, especially along the margins, seven days after inoculation except at hormonal combination of 5 iM NAA: 0 iM KIN. Treatment with 20 iM NAA: 5 iM KIN ratio produced the highest mean number of pinheads (1.89) while that with equal concentrations of NAA and KIN (20 iM:20 iM treatment) produced the highest mean number of clumps (1.60). Fourteen days after inoculation, largest mean size of the clumps (2.46 mm) was also observed in the 20 iM NAA: 20 iM KIN treatment. After 53 days in culture, 20 iM NAA: 1 iM KIN treatment produced the largest amount of calli in terms of fresh weight (ca 300 mg).

Approximately 50 mg of oven-dried calli per treatment were subjected to corosolic acid analysis by high performance liquid chromatography (HPLC). Percent yield of corosolic acid found in calli of all treatments ranged from 0.20 to 0.39%; the highest value was in 5 iM NAA: 0 iM KIN treatment.

Keywords: *Lagerstroemia speciosa*, callus, corosolic acid, naphthalene acetic acid (NAA), kinetin (KIN)

BSD No. 19

PREPARATION AND CRYOPRESERVATION OF CYTOPLASTS FOR THE PRODUCTION OF BOVINE AND BUBALINE CLONAL EMBRYOS

Edwin C. Atabay*, Eufrocina P. Atabay, Felomino V. Mamuad and Libertado C. Cruz

Philippine Carabao Center, Central Luzon State University,
Science City of Muñoz, Nueva Ecija
Email: atabay22@yahoo.com

One major factor influencing success in cloning by nuclear transfer (NT) is the recipient oocyte. The development of efficient procedure for preparation and utilization of in-vitro matured oocytes as recipients for NT is therefore essential. This study was conducted to: prepare cytoplasts with high enucleation rate and developmental competence, determine the effects of activation treatments, and utilization of vitrified bovine oocytes as recipients for intraspecies and intergeneric somatic cell NT. In study 1, vortexing cumulus-oocyte complexes in hyperosmotic media for a long period moved the first polar body and reduced enucleation rate. Removal of 20% cytoplasmic volume increased enucleation rate and showed no deleterious effect on subsequent development of reconstructed bovine embryos. In study 2, NT embryos were constructed with bovine oocytes of 3 different preparations: enucleated oocytes treated with calcium ionophore for 5 min and cycloheximide for 5 h; treated with ethanol for 7 min and cycloheximide for 2 h, and those without treatment. NT embryos constructed with ethanol-treated oocytes showed similar rates of fusion, cleavage and blastocyst formation to those of the non-treated oocytes. NT embryos constructed with ionophore-treated oocytes did not show any pronucleus-like structure and showed lower cleavage rate and no development to blastocysts. In study 3, *in-vitro* matured bovine oocytes were vitrified in microdrops. After warming, around 90% of the vitrified oocytes were morphologically normal. The enucleation rate and development to blastocysts were similar to those constructed with fresh oocytes. Intergeneric NT embryos had lower cell numbers than the intraspecies NT blasocysts. In conclusion, aspiration of the first polar body and adjacent 20% cytoplasmic volume after removing cumulus cells by vortexing for a short period using hypo-osmotic media increased enucleation rate of bovine oocytes. This study also demonstrated the efficacy of microdrop vitrification procedure and the successful production of buffalo clonal embryos using vitrified bovine oocytes.

Keywords: Activation, bovine oocytes, enucleation, intergeneric, vitrification

BSD No. 20

VITRIFICATION AND IN VITRO CULTURE OF MOUSE PREANTRAL FOLLICLES: A MODEL FOR MAXIMUM UTILIZATION OF OVARIAN FOLLICLES IN WATER BUFFALOES

Eufrocina P. Atabay*,[1] Edwin C. Atabay[1] and Libertado C. Cruz[1]

[1]Philipppine Carabao Center, National Headquarters and Genepool
Science City of Munoz, Nueva Ecija
Email: bingay2003@yahoo.com

In-vitro culture and cryopreservation of ovarian pre-antral follicles is a new tool for rescuing genetic materials in mammalian ovary for offspring production. The present study demonstrated successful production of pups from vitrified pre-antral follicles in mice which would then be applied in buffalo species. Preantral follicles mechanically isolated from ovaries of 12 day old mice were exposed to 2 mol ethylene glycol for 2 or 5 min and then to vitrification solution (VS) containing 6 mol ethylene glycol and 0.3 mol raffinose for 0.5, 1.0, or 2.0 min before vitrification. The vitrified and fresh preantral follicles were treated with collagenase, and the oocyte-granulosa cell complexes (OGCs) obtained were cultured in-vitro for 10 days in membrane inserts. Preantral follicles exposed to 2 mol ethylene glycol for 5 min then to VS for 0.5 or 1.0 min showed highest survival rates after warming. The follicular loss after warming was approximately 20%. After in-vitro culture, the proportion of viable OGCs from vitrified follicles was 10% lower than that of fresh preantral follicles. There were no differences in the rates of maturation, fertilization and development to blastocysts between the oocytes derived from vitrified follicles and those from fresh preantral follicles; however, the developmental competence of oocytes derived from both vitrified and fresh preantral follicles grown in-vitro was lower than that of oocytes grown in-vivo. One of the recipient mice that received 20 blastocysts derived from vitrified preantral follicles gave birth to six live pups. The ability to rescue preantral follicles and produce offspring is of great benefit in buffaloes because of the inherently low number of preantral follicles in their ovary compared to cattle. The vitrification and culture of preantral follicles is therefore a potentially valuable reproductive technology as this will allow maximum utilization of the limited number of ovarian follicles in water buffaloes.

Keywords: Buffaloes, preantral follicles, vitrification, mouse, ethylene glycol, in vitro culture

BSD No. 21

AN INITIAL INVESTIGATION ON THE MORPHOLOGY, ULTRASTRUCTURE AND DEVELOPMENT OF THE ELASMOID SCALES OF MULLET FISHES (PERCIFORMES: MUGILIDAE) WITH EMPHASIS ON LUDONG (*CESTRAEUS PLICATILIS* VAL. 1836)

Pacifico Eric E. Calderon, Marivic A. Leon, Anna Cristina V. Suarez, Ma. Bida Olivia S. Tamayo, Julie Ann M. Valencia and Emerita C. Simeon

Department of Biological Sciences, College of Science
University of Santo Tomas España Blvd., Manila
Email: creeknipacey@yahoo.com

Comparative morphologic and molecular characters of adult mullet fishes in the Philippines were investigated, with emphasis given on *ludong* (*Cestraeus plicatilis* Valenciennes 1836), a rare, catadromous species suspected to be endemic to the Cagayan River and its tributaries. *Ludong* commands a price of about Php 3000 per kilogram for its unique palatability, aroma and seasonality. Although *ludong* has been pronounced endangered by the Bureau of Fisheries and Aquatic Resources, its biology and taxonomy are, however, yet to be fully established. This study was taken as an initial investigation on *C. plicatilis* and effort was made to present its unique morphology for its proper identification, with emphasis on the histology and ultrastructure of the scale ornamentation pattern. First, five (5) mullet species were identified by morphological characterization. Secondly, the histology and ultrastructure of scale ornamentation pattern were investigated by combining ocular inspection, eosin-Y staining, light and scanning electron microscopy (SEM). Finally, the possible role of *fibroblast growth factor 8 (fgf-8)* in scale development was investigated by genomic DNA isolation, gene amplification by polymerase chain reaction, direct DNA sequencing, sequence alignment and phylogenetic tree construction. In conclusion, mullet fishes were shown to exhibit significant variations in the sub-oral regions and in the squamation patterns, among other morphometric and meristic data. Intra-species regularity and inter-species variation of squamation pattern, scale morphology and ultrastructure were observed. Epithelial microridges were observed on the outer surface of the scale epidermis, forming a species-specific ornamentation pattern. The discreteness between morphologic and molecular data suggests the existence of both genetic cascades and epigenetic factors in scale organogenesis and scale ornamentation development. The observed scale ornamentation pattern may be employed to further analyze phylogenies and infer modes of aquatic adaptation among mullets. Descriptive data obtained in this baseline study are deemed essential for further studies on *ludong* conservation.

Keywords: *Cestraeus plicatilis*, catadromous, conservation, elasmoid scale, endemic, scale ornamentation, meristic, morphometric, SEM, *fgf-8*, ultrastructure

BSD No. 22

THE FRESHWATER AND SEMI-TERRESTRIAL BRACHYURANS OF SELECTED AREAS IN LUZON ISLAND, PHILIPPINES

Jimmy Tevar Masagca

Graduate School of Education, Arts and Sciences
De La Salle University Dasmariñas, Cavite 4115
Email: jtmasagca@mail.dasma.dlsu.edu.ph

The freshwater and semi-terrestrial brachyurans of selected areas in Luzon were studied on the basis of field collections carried out in 2004 to 2005 in Cavite, Nueva Ecija, Tarlac, Camarines Sur, Albay and Catanduanes. A total of 116 crab samples were obtained, preserved and diagnosed in the laboratory for taxonomic characters. Eight (8) taxa are reported and described here. The crabs identified belong to four families, namely: Potamidae Ortmann, 1896; Parathelphusidae Alcock, 1910 (sensu Ng, 1988); Grapsidae Mcleay, 1838; and Gegarcinidae Mcleay, 1838. Two unidentified endemic potamid crabs are tentatively described as *Ovitamon* sp. 1 from Ternate, Cavite and *Ovitamon* sp. 2 from Sulong Falls in San Miguel, Catanduanes. Although the specimens examined from Cavite showed striking similarities with *Ovitamon artifrons* Burger, 1894, the structure of male gonopodium (G_1) does not resemmble that of the specimens described previously from Tagaytay by Ng & Takeda in 1992. A new endemic species *Ovitamon cavitensis* (sp. ov.?) is proposed here. Two riverine crabs belonging to the family Parathelphusidae are reported here as *Sundathelphusa* sp. 1 and 2 from Guimba, Nueva Ecija and Moncada, Tarlac, respectively. Out of the three species of the widely distributed grapsid crabs (of the genus *Varuna* H. Milne Edwards, 1830), two species are reported here as *Varuna altimana* and *V. literata*. Among the semi-terrestrial crabs, *Sesarmops (Sesarma) impressum* H. Milne Edwards, 1887 (from Lictin River, San Andres, Catanduanes Island) and *Cardisoma carnifex* Herbst, 1794 (from Balayan, Batangas) are also described in this continuing project on the "Biodiversity of Freshwater Crabs of Luzon, Philippines" at De La Salle University-Dasmariñas.

Keywords: Brachyurans, crapbs, taxonomic characters

BSD No. 23

MERISTIC AND MORPHOMETRIC CHARACTERS OF AN ENDEMIC FRESHWATER CRAB, *OVITAMON CAVITENSIS* (SP. NOV. ?) (BRACHYURA: MALACOSTRACA) FROM CAVITE LUZON ISLAND, PHILIPPINES

Jimmy T. Masagca[1] and Cherry Z. Cuevas[2]

[1]College of Education, Professional Education Department-Graduate Studies
[2]Biological Sciences Department, College of Science
De La Salle University-Dasmariñas
Cavite 4115, Philippines
E-mail: jtmasagca@dasma.dlsu.edu.ph

Given the paramount importance of the biodiversity in freshwater systems of the country to generate more information on the crab faunal diversity and the need to conserve the remaining natural populations of these groups of crustaceans, a research project under the research program on the *Patterns of Biodiversity in Aquatic Systems* is being carried out at De La Salle University in Dasmarinas (DLSU-D). Several crab specimens which are wholly freshwater species in three families, Potamidae Ortmann, 1896, Parathelphusidae, Alcock, 1910 (both *sensu* Ng, 1988), and Grapsidae McLeay, 1838 (*partim*) were collected by handpicking and with the use of crab traps ("pukot" and "bintol") from different riverine and lacustrine environments of southern Luzon (Cavite, Quezon, Camarines Sur, Albay, Sorsogon). These specimens are currently being diagnosed taxonomically for new records and possibly new species. The freshwater crab fauna of the Philippines is poorly known, thus more systematic studies are needed since many more species undoubtedly still await for discovery. During field collections in Cavite, specimens of an endemic wholly freshwater potamid species (tentatively identified as *Ovitamon cavitensis* (sp. nov.?) was obtained from a forest stream in Mts. Palaypalay/Mataas Na Gulod National Park (200 to 360 meters above sea level or asl). A total of sixty (30 males and 30 females) were described and subjected for meristic and morphometric analyses. Diagnostic analysis shows that carapace ovoid, branchial, cardiac, urogastric and cervical grooves very shallow; postero-lateral regions appear smooth; external orbital angle broadly triangular, epibranchial tooth low, but clearly separated from external orbital angle; antero-lateral margin convex, postero-lateral margin appears converging; male abdominal distinctly triangular, segment 7 slightly longer than segments 6 and 5, lateral margin of segment 7 straight, lateral margin of segment 6 slightly convex; third maxilliped exopod with well-developed flagellum, longer than the width of the merus; ischium with sulcus, not medial; and dorsal margins of chela not serrated, and finger slightly longer than palm. The first gonopodium (G1) stout, cylinder-shaped, curved towards the inside, and surfaces

covered with hairs. Meristically, *O. cavitensis* (sp. nov.?) in both sexes consist of 7 abdominal segments, 2 anterolateral teeth protected by the third maxilliped, numerous well-developed inner distal spines in the cheliped and maxilliped; and spinnules in the cheliped and maxilliped. Morphometric analysis includes 7 characters (carapace length, *CaL*; carapace width, *CaW*; maxilliped length, *MaL*; cheliped length, *ChL*); propodus length, *PrL*; frontal margin, *FrM*; and eye diameter, *EyD*). Results indicate an observed difference between male and female samples (as to *CaL*, *MaL* and *FrM*) of the endemic crab under study (*O. cavitensis* sp. nov. ?), which belongs to the genus *Ovitamon* Ng et Takeda 1992 (Potamidae Ortnmann, 1896). Further investigations are needed, particularly on the morphology (ultrastructures) of the sperm transfer tubes in the male gonopodium (G1).

Keywords: *Ovitamon cavitensis*, Cavite, lacustrine,

BSD No. 24

MONSOONAL WINDS INFLUENCE THE ASSEMBLAGE STRUCTURE, ABUNDANCE AND DISTRIBUTION OF NET PHYTOPLANKTON IN ILIGAN BAY, NORTHERN MINDANAO

Rosabeth Morandarte and Ephrime B. Metillo*

Department of Biological Sciences, Mindanao State University-Iligan Institute of Technology, A. Bonifacio Ave., Iligan City 9200
Telephone: (063) 221 4050 up to 55 local 137
E-mail: ovcre-ebm@sulat.msuiit.edu.ph

The coastal and oceanic waters of the Philippines are mainly exposed to two dominant wind patterns annually, the north easterlies during the northeast (NE) monsoon and the south westerlies during the southwest (SW) monsoon. We studied the structure, abundance and distribution of surface layer net phytoplankton assemblages in Iligan Bay during the southwest and northeast monsoons. Vertical samples were collected at 24-station sampling grid on July-August 2000 (SW monsoon) and November-December 2000 (NE monsoon). The abundance of each phytoplankton Genus was estimated following the Utermöhl method. Assemblage structure and distribution were analyzed using multivariate and univariate routines available in the PRIMER software (Warwick and Clarke 2001). Abundance was significantly higher in the NE monsoon. Although assemblages in the two monsoons have similar generic richness, cyanobacteria (*Trichodesmium*) dominated the NE monsoon assemblages while diatoms (*Chaetoceros*) in the SW monsoon. Bray-Curtis similarity dendrograms and multidimensional scaling plots revealed a bay-wide contour of assemblages that

have high, moderate, and low levels in generic diversity and relative abundance. All three levels were observed in the SW monsoon assemblages, but only two (high and low) in the NE monsoon samples. High level stations are found in the southwestern sector of the bay, the low levels near mouths of major river systems, and the moderate level in other locations. Factors including salinity, temperature, and total dissolved phosphate weakly explain the spatial variations observed, thus contrasting wind patterns and other hydrodynamic processes during the two monsoons are also important in structuring phytoplankton assemblages in Iligan Bay.

Keywords: phytoplankton ecology, *Trichodesmium*, *Chaetoceros*, northeast monsoon, southwest monsoon, multivariate analysis, Iligan Bay

BSD No. 25

MODULATION OF ANGIOGENESIS IN THE CHICK EMBRYO CHORIO-ALLANTOIC MEMBRANE (CAM) BY THE POLAR AND FRACTIONAL POLYSACCHARIDE EXTRACTS OF *Ganoderma applanatum*

Mikhael Cereno M. Bajo[1], Lydia M. Bajo[2], Roberto M. Malaluan[3] and Franco G. Teves[1]*

[1] Molecular Genetics and Microbial Technology Laboratory,
Department of Biological Sciences,
[2] Department of Chemistry, College of Science and Mathematics
[3] Department of Chemical Engineering Technology,
School of Engineering Technology
MSU-Iligan Institute of Technology, 9200 Iligan City
*Email: franco_teves@yahoo.com

Ability to regulate angiogenesis, or *de novo* formation of blood capillaries, finds its significant role both in physiological and pathological conditions. The treatment of certain neoplastic cases with extracts of *Ganoderma* sp. has been documented in China and other Asian countries, although the specific mechanism is still not very clear.

A polar fraction and four fractional polysaccharide extracts from *Ganoderma applanatum* were tested for angiogenesis modulating effects using the chick embryo chorioallantoic membrane (CAM) assay. One-day old fertilized chicken eggs were treated with three concentrations of the *G. applanatum* extracts in three replicates. One square centimeter "windows" were aseptically created, by removing shell materials within specified areas. Two to three milliliters of albumen was removed using sterile pipettes to expose the CAM, which was then treated with 0.1 ml of the

extracts. The "windows" were sealed with 16-ply sterile gauze taped onto the shell and the eggs incubated at 37°C until the seventh day. Semi-quantitative scoring was done by removing shell materials from the periphery of the "windows" and comparing the degree of vascularization with the control.

The polar fraction exhibited a significant angiogenic stimulation whereas three out of four fractional polysaccharide extracts showed significant inhibition of angiogenesis as analyzed using Scheffe's post hoc test and t-test. These differential effects seem to demonstrate the presence of more than one type of bioactive components in *G. applanatum*. It is therefore possible that one mechanism for the anti-neoplastic effect of this shelf fungus' extract is through angiogenesis modulation. Tumors cannot thrive without abundant blood supply.

Keywords: Angiogenesis, chorioallantoic membrane, polysaccharide, *Ganoderma applanatum*

BSD No. 26

GEOMETRIC MORPHOMETRIC APPROACHES TO THE DISCRIMINATION OF STOCK POPULATIONS OF FRESHWATER COMMERCIAL FISHES FROM BALOI LAKE IN LANAO DEL SUR

Nasheba Arumpac, Mark Anthony J. Torres and Cesar G. Demayo

Department of Biological Sciences
College of Science and Mathematics
MSU-Iligan Institute of Technology, Iligan City
Email: c_gdemayo@yahoo.com

The truss network in fishes has been used as a fisheries management tool effective in the description of morphological variation and identification of stocks, thus improving the biological basis of management. Traditionally, this is analyzed and compared by collecting a series of measurements between landmarks that form a regular pattern of connected quadrilaterals or cells across the body form using a vernier caliper. In this paper, however, the truss networks in five freshwater commercial fishes are analyzed using the methods of geometric morphometrics, which includes the generation and analysis of thin-plate spline transformation grids, partial warp scores and interlandmark distances. This study aimed to present an alternative computerized approach in the collection and multivariate analysis of fish truss network morphometric data.

A total of two hundred forty-four (244) individuals were sampled. Sample sizes for each species are as follows: *Glossogobius* sp. (n = 40) , *Hypseolotris agilis* (70), *Anabas testudineus* (42), *Puntius tumba* (69), and *Cyprinus carpio*

carpio (23). These fishes were scanned at 600 dpi. Then, a total of fifteen landmarks that are distinctive and homologous from specimen to specimen were selected around the outline of the fish form. The X-Y coordinate value for the positions of landmarks is digitized for each fish with the use of the *ScionImage* image digitizing and processing software. The X-Y coordinate data is then transformed into linear distances by computer using the Euclidean Distance Matrix Algorithm (EDMA). This transformed data was then used as morphometric variable for multivariate methods of statistical analysis. Such as principal component analysis, discriminant function analysis and cluster analysis.

The original coordinate data were also used to compare shape differences among the different species. Because, all samples for all populations were taken from different ages, there is a need to eliminate the size effect in the data set. This was done by subjecting the coordinate data to procrustes fitting. Then, the information contained in the procrustes-fitted coordinates were used to reconstruct the truss networks of individual fish samples using thin-plate spline transformation grids. Morphological variations within and among the different species were summarized using PCA and other multivariate methods of statistical analysis. Global and localized variations were also determined through the analysis of partial warps.

This study showed variations in the sizes and shapes of the individuals within and among the different species as evident from the thin-plate spline grids and supported by the results of the PCA and dendrogram generated through Cluster Analysis. Discriminant analysis showed that the identification accuracy was between 93% and 100%, and global identification accuracy was 98.6%. Morphological differentiation was shown to vary between the sexes in all fish species as a result of sexual dimorphism. This suggests that females and males should be treated separately in truss network analyses to remove the effect of sex from the result. The results of this study proved the usefulness of geometric morphometric approaches to the study stock populations of freshwater fishes, which are powerful in reconstructing and comparing the truss networks of fishes.

Keywords: truss network, geometric morphometrics, cluster analysis, discriminant analysis

BSD No. 27

BIOLOGY OF *Eocanthecona furcellata* (Wolff) AND ITS PREDATORY CAPACITY AGAINST CORN BORER (*Ostrinia furnacalis*)

Somkid Nuchphun and Teotimo M. Aganon*

Plant Protection Service Division, Department of Agricultural Extension,
Bangkok, Thailand and Central Luzon State University
Science City of Muñoz, Nueva Ecija, Philippines
Email: tmaganon@lycos.com

The biology of *Eocanthecon. furcellata* was studied under laboratory conditions. The insect oviposition period ranged from 20 – 29 days with an average of 23.4 + 3.657. The female laid eggs ranging from 7 to 104 (X = 72.88 + 25.11).

The egg incubation period was 5.20 + 0.837 days. The nymph molted five times with a total nymphal stage days of 18.84 + 2.160 broken down as: 1st instar, 2.92 + 0.846; 2nd instar, 3.56 + 0.501; 3rd instar, 3.46 + 0.503; 4th instar, 3.50 + 0.496 and 5th instar, 5.40 + 0.496. The total life cycle was 24.52 + 1.787 days. The adult male lived within a range of 36.40 days after emergence whereas the female lived 42.49 days after emergence.

The total predatory period from the second nymphal instar to death of adult was 45.23 days.

Functional response of adults as predator on second and third larval instars of *O. furnacalis* increased with an increased in prey density. The same trend was observed on the predatory capacity of the nymphal stages. Adults predatory capacity is higher than the nymphal stages.

The results showed that *E. furcellata* was not a good predator if the corn borer density is high, whereas if the corn borer density is low, *E. furcellata* could be a good predator.

Keywords: *Eocanthecona furcellata*, corn borer, *Ostrinia furnacalis*, biological control

BSD No. 28

FIGHTING BEHAVIOR OF THE PHILIPPINE DERBY SPIDER *Neoscona punctigera* (Doleschall)

Aimee Lynn A. Barrion

UPLB Museum of Natural History, Upper (Forestry) Campus
University of the Philippines Los Baños, College, Laguna 4031
Email: aranea95@yahoo.com

Filipinos are game-loving people. They have managed to utilize an assortment of animals in their games and leisures like cockfighting, horse racing, dog racing, carabao fighting and spider derby. In most spider games, females of the species *Neoscona punctigera* (Doleschall) appear to be commonly used. However, the predatory nature of this spider is yet to be explored. Thus, this study provides preliminary documentation and explanations for spider fighting behavior. A wooden T-shaped rod with a catch at the bottom was used as a fighting arena. Spiders whose body sizes ranged from 13 – 15 mm were grouped as the large mature females while those whose body sizes ranged from 10 – 12.5 mm were designated as small mature females. Reproductive status was determined by observing the epigynum. Oviposited spiders were allowed to fight 2 days after oviposition. Complete sequential steps of the fights were observed, described and recorded in video. Details of the spider matches were presented in kinematic graphs. Matches involving spiders of different sizes showed that larger spiders win in all encounters that resulted in actual combat (55 of 75 matches). Only in 21 of the 75 total matches between two large female spiders actually resulted in an encounter. On the other hand oviposition status did not affect the winning chances of the spider. In view of these observations, two hypotheses on the origins of fighting behavior of derby spiders were proposed. One is that the aggressive behavior of derby spiders is due to maternal care, while the other is linked to territoriality.

Keywords: behavior, derby, epigynum, kinematic graphs, *Neoscona punctigera* (Doleschall), matches, oviposition, spider

BSD No. 29

ALTERNATE HOSTS OF CORN EARWORM, *Helicoverpa armigera* (HB.) AND CUTWORM, *Spodoptera litura* (F.) IN ONION REFUGIAS

B.F. Cayabyab[1,2], C.L. Padilla[1], J.R. Deanon, Jr.[1], D. Vargas, Jr.[1] and R.G. Bayot[1]

[1]National Crop Protection Center, Crop Protection Cluster
[2]Crop Science Cluster, College of Agriculture
University of the Philippines Los Baños
College, Laguna 4031, Philippines
Email: bfcayabyab@laguna.net

This study was conducted from 2002-2005 in four towns of Nueva Ecija which are the main onion growing areas in the country. It aimed to identify alternate host plants corn earworm, *Helicoverpa armigera* (Hb.) and cutworm, *Spodoptera litura* (F) are major pests of vegetables and grains in the Philippines in onion refugias of Laur, Gabaldon, Pantabangan, and Bongabon towns.

The presence of these two pests was done through ocular observation on the boundaries of onion fields where weeds abound. The results showed that the following species harbor these two pest: *Amaranthus spinosus* L., *Stachytarpheta jamaicensis* (L) Vahl., *Polygonum barbatum* L., *Cassia tora* L., *Melochia corchorifolia* L., and *Chromolaena odorata* L.

Onion farmers apply sixteen times spraying on their fields to control these pests. Knowledge of alternate host plants or where to find these pest can lessen pest management cost by directing control measures on refugias where these alternate host and these two larval pest are present. Hence, population build up of these pest that infest onions is reduced.

Keywords: refugias, corn earworm, cutworm, indicator plants, onion, larvae

BSD No. 30

ONION MAGGOT/FLY, *Delia antigua* MEIGEN (DIPTERA: ANATHOMIXIDAE): A NEW PEST OF ONION IN THE PHILIPPINES

Bonifacio F. Cayabyab[1] MarioV. Navasero[1], Daniel G. Vargas[1], Paul Rommel Elvira[1], Rolando G. Bayot[1], Melvin D. Ebuenga[1], Mar Antineo[2], and Michael Caballes[3]

[1] Crop Protection Cluster, University of the Philippines
Los Baños, College, Laguna
[2] Office of the Provincial Agriculturist, Lingayen Pangasinan
[3] Allied Botanical Corporation, Quezon City
Email: bfcayabyab@laguna.net

Since 2001 to date onion farmers in Pangasinan complained of damages in their onions described as root rot of seedlings and bulb rot. The onion leaves become brown and eventually dry up.

Field collected damaged bulbs from Amancosiling, Bayambang, Pangasinan were brought to the Plant Quarantine Support Laboratory, UP Los Baños in 2004-2005 cropping season. Each bulb with visible larva was separated and these were reared in the laboratory. Emerging adults were put in cyanide bottles. These adults were pinned and stored in standard insect boxes. Collection of adults through sweep net was also done in infested fields. The adults onion fly were separated and stored in insect boxes.

The male is large (6 – 7 mm in length), with densely grey to yellow-grey dust on thorax and abdomen. The eggs are about 1.25 mm long white and elongate-ovoid, resembling grains of rice. The larvae are typical elongate creamy-white maggots, tapering towards the head end and truncate at the posterior end where two breathing spiracles are situated. The pupae are ovoid or seed-shaped, reddish-brown. The posterior spiracles are situated on small bosses. Adults are slender yellowish-grey flies, 6 – 7 mm in length. The signs of attack are yellowing and wilting of the outer leaves. Green and apparently healthy leaves will become flaccid, and the whole plant may collapse. Later generations of larvae tunnel into the onion bulbs as well as attack the roots. This new pest of onion is prevalent in onion growing areas of Pangasinan such as Alcala, Bayambang Basista and nearby towns. It has also been found in Nueva Ecia and Nueva Viscaya onion areas.

Keywords: Onion maggot/fly, New Pest, Onion, Delia antiqua

BSD No. 31

PHILIPPINE PREDATORY MITES OF THE GENUS *CHEIROSEIUS* (ACARI: ASCIDAE)

Ireneo L. Lit, Jr.*[1,2] and Leonila A. Corpuz-Raros[2,3]

[1]Department of Forest Biological Sciences
College of Forestry and Natural Resources,
[2]Entomology Section, Museum of Natural History, and
[3]Crop Protection Cluster,
College of Agriculture, University of the Philippines Los Baños
College, Laguna 4031, Philippines; *
Email: illit_jr@yahoo.com

A taxonomic study of Philippine mites belonging to the genus *Cheiroseius* (Acari: Ascidae) was conducted. This is a group consisting of predators of soil arthropods and other small animals like nematodes and are, therefore, potential biological control agents against soil-inhabiting pests. Habitat-wise, they can be found in soil, decaying plant litter and mosses. From only three previously reported species, namely: *Cheiroseius curtipes* (Halbert), *C. nepalensis* (Evans & Hyatt) and *C. serratus* (Halbert), there are now 11 species, including four described as new to science and four new faunal records. Two of the new species are from Mount Banahaw de Lucban, coded *Cheiroseius* spp.nov. BDL1 & BDL2, and the two others are from Polillo island, coded *Cheiroseius* spp.nov. POL1 & POL2. BDL1 is unique in having a prominent dorsal shield, so produced as to form a "humpback" effect, whereas BDL2 has very unusual ornate genital shield. POL1 has intricate reticulate patterns on the dorsal shield and POL2 has scale-like patterns on the sternal and genital shields. The four new records are *C. browningi* (Evans & Hyatt), *C. cassiteridum* (Evans & Hyatt), *C. phalangioides* (Evans & Hyatt) and *C. politulus* Tseng. A key to Philippine species, descriptions and illustrations are provided.

Keywords: Ascidae, Acari, *Cheiroseius*, mites, predatory mites, soil-litter fauna

BSD No. 32

THE PHILIPPINE ENDEMIC STICK-INSECT GENUS *OBRIMUS* STÅL, 1875 (PHASMATODEA: HETEROPTERYGIDAE: OBRIMINI)

Ireneo L. Lit, Jr.*[1,2] and Orlando L. Eusebio[2]

[1]Department of Forest Biological Sciences
College of Forestry and Natural Resources
[2]Entomology Section, UPLB Museum of Natural History
University of the Philippines Los Baños
College, Laguna 4031, Philippines;
* E-mail: illit_jr@yahoo.com

Studies on Philippine stick insects have recently gained considerable interest from many parts of the world, especially with the recent tide of discoveries of new species and genera. Among Philippine stick insects, the tribe Obrimini stands out as being endemic at a suprageneric level. As a contribution to the systematics of this group and to the documentation of Philippine terrestrial arthropod biodiversity, a taxonomic review of the genus *Obrimus* Stål, 1875 was conducted. Six species, including two possibly new ones, are described and illustrated. They are *Obrimus bicolanus* Rehn & Rehn (Isarog), *Obrimus bufo* (Westwood), *Obrimus* **sp.1** *ex* Northern Sierra Madre, *Obrimus* **sp. 2** *ex* Southern Luzon, *Obrimus mesoplatus* (Westwood) and *Obrimus uichancoi* Rehn & Rehn (Greater Sipit Watershed, Mt. Makiling). The differences of these species lie in the pattern of their armature which consists mainly of spines and tubercles. All species are of narrow endemic distribution and may be possible candidates for listing as threatened or vulnerable species, considering the rapid destruction of forest habitats and their current status as among favorites of hobby collectors in Europe and other advanced countries.

Keywords: Heteropterygidae, Obrinini, *Obrimus*, Phasmatodea, Philippine endemic arthropods, stick insects, terrestrial arthropod biodiversity

BSD No. 33

BACTERIAL INHIBITION OF *Aspergillus flavus* GROWTH AND AFLATOXIN BIOSYNTHESIS: *Bacillus amyloliquifaciens* ET2004, A POTENTIAL BIOCONTROL AGENT

Sharon May D. Erasquin, Franco G. Teves*, Ermelinda G. Tobias and Faith B. Amorado

Molecular Genetics and Microbial Technology Laboratory
Department of Biological Sciences, CSM
MSU-Iligan Institute of Technology, 9200 Iligan City
*Email: franco_teves@yahoo.com

Damage to crops by fungal agents like *Aspergillus flavus* and *Aspergillus parasiticus* not only lower their yield and quality but also continue to pose health hazards to consumers as a consequence of aflatoxin production. While genetic modification bestows resistance against pathogenic fungi, the use of appropriate biological control agents may also provide an equally effective alternative.

Five pre-selected bacterial isolates (*Staphylococcus epidermidis*, *Agrobacterium radiobacter*, *Bacillus megaterium*, *Enterobacter aerogenes*, and *Bacillus amyloliquifaciens* ET2004) were screened for inhibition of growth in *Aspergillus flavus* and/or inhibition of aflatoxin biosynthesis. Replicated co-cultures on potato dextrose agar (PDA) plates, which supported the growth of the fungus and bacteria, were prepared by parallel streaking. The plates were incubated at 30 °C and examined for fungal overgrowth on the bacterial streak after 48 h.

Inhibition of aflatoxin biosynthesis was determined by co-cultures in potato dextrose broth (PDB). One milliliter of 10^6 to 10^7 bacterial cells/ml and 56 ml of sterile water-suspended *A. flavus* conidia (10^7 to 10^8/ml), were mixed in four milliliters of PDB and incubated with shaking initially at 37 °C for day one and at 30 °C for the next six days. The cultures were centrifuged at 3,000 rpm for five min. The supernatants were collected aseptically and analyzed for aflatoxin content using Competitive-Direct Enzyme Linked Immunosorbent Assay (CD-ELISA) with the VeratoxÔ System (Glenwood Technologies).

Results show significant fungal growth inhibition exhibited by *Bacillus amyloliquifaciens* ET2004, but not by the other bacterial isolates. Interestingly, no aflatoxin was detected in the latter tubes although substantive growth of *A. flavus* was evident. Growth inhibition may be attributed to the production of subtilisin by *B. amyloliquifaciens* which is known to degrade proteins extracellularly. Presence of fungal growth without the concomitant production of aflatoxin may indicate the synthesis of cell products that specifically inhibit the aflatoxin biosynthetic pathway, but not growth.

B. amyloliquifaciens is not known to exhibit pathogenicity or toxigenicity, and is therefore a very promising candidate for the biocontrol of fungal invasion of crops. Pending verification, the other pre-selected bacterial isolates may be utilized for targeting only aflatoxigenic fungi, while leaving unaffected other fungi that may play important ecological roles.

Keywords: *Aspergillus flavus*, *Bacillus amyloliquifaciens* ET2004, aflatoxin, subtilisin, biocontrol

BSD No. 34

MITOCHONDRIAL DNA ANALYSIS OF GENETIC INTROGRESSION IN SELECTED PHILIPPINE CATTLE POPULATIONS

Genevieve Mae B. Aquino[1]*, Rita P. Laude[2], Jianlin Han[3], and Olivier Hanotte[3]

[1] Animal & Dairy Sciences Cluster, College of Agriculture, and
[2] Institute of Biological Sciences, University of the Philippines
Los Baños, College, Laguna, Philippines
[3] International Livestock Research Institute, Nairobi, Kenya
Email: gm.aquino@gmail.com

For the future improvement of beef cattle breeding programs in smallholder farming systems, mitochondrial DNA (mtDNA) displacement loop (D-loop) sequence variation was examined to supplement existing basic information on the genetic composition of Philippine Native cattle. To evaluate the degree of genetic introgression in native cattle populations representing the Ilocos, Batangas, Ilo-ilo and Philippine Native types, a fragment of the hypervariable region in the mitochondrial D-loop was amplified and sequenced from a total of 100 individuals used in a previous study on autosomal microsatellite variation. Multiple sequence alignment with published D-loop sequences of foreign pure breds and subsequent phylogenetic analysis revealed three major clades of zebu (*Bos indicus*), taurine (*B. taurus*) and banteng (*B. banteng*) maternal ancestry. Individual sequences did not cluster significantly into geographic groups, providing evidence of migration between populations though trade relationships between these regions. However, haplotypes of Native cattle were distinct from those of the foreign breeds, implying the evolution of the Philippine populations from their foreign ancestral populations. Patterns of mitochondrial DNA variation indicate that the Ilo-ilo type was of taurine ancestry with zebu genetic introgression, and the Philippine-Bali type was of zebu ancestry with banteng introgression. The hybrid zebu-taurine composition of mtDNA from the Ilocos and Batangas populations confirmed the results of previous

autosomal microsatellite analysis. Molecular characterization based on combined mitochondrial and microsatellite analysis provides a clearer perspective on the genetic composition of Philippine cattle populations and showed the potential of existing populations in the geographically-isolated islands of the Visayas for future genetic conservation and breed development programs.

Keywords: cattle, mitochondrial DNA, D-loop, genetic diversity, Philippine Native breed

BSD No. 35

SEQUENCE ANALYSIS AND CHARACTERIZATION OF THE MOVEMENT PROTEIN-ENCODING COMPONENT OF ABACA BUNCHY TOP NANOVIRUS (ABTV)

Vermando M. Aquino* and Maria Armila D. Ruiz

National Institute of Molecular Biology and Biotechnology
University of the Philippines Diliman, Quezon City
Email: vermando.aquino@up.edu.ph

Abaca bunchy top nanovirus (ABTV) is the most important viral pathogen of abaca. The virus causes stunting and significant reduction in yield. In this study, the DNA component of abaca bunchy top nanovirus (ABTV) coding for the movement protein was isolated and characterized. Oligonucleotide primers based on published sequence of a related virus, banana bunchy top virus (BBTV), were used for PCR amplifications. Total DNA from infected leaves was extracted from different ABTV isolates using a modified CTAB extraction method. A 380 bp fragment was amplified, cloned and sequenced. Sequence analysis showed that the movement protein gene includes regions that codes for hydrophobic protein. The nucleotide sequences were compared with published BBTV sequences and showed 94 to 96% nucleotide identities with isolates from Australia and Asian countries.

Keywords: abaca, movement protein gene, abaca bunchy top nanovirus

BSD No. 36

THE RELATIONSHIP BETWEEN ABACA BUNCHY TOP NANOVIRUS (ABTV) AND BANANA BUNCHY TOP NANOVIRUS (BBTV) BASED ON SEQUENCE ANALYSIS OF DNA-3 AND DNA-4 COMPONENTS

Vermando M. Aquino* and Maria Armila D. Ruiz

National Institute of Molecular Biology and Biotechnology
University of the Philippines
Diliman, Quezon City
Email: vermando.aquino@up.edu.ph

Bunchy top is the most devastating disease of both abaca and banana caused by abaca bunchy top nanovirus (ABTV) and banana bunchy top nanovirus (BBTV) respectively. ABTV is assumed to be synonymous to the bunchy top virus that infects banana but this has never been confirmed. BBTV is considered to be a variable virus with strain-complexity. Previous reports on transmission experiments showed dissimilarity between these two viruses, although they exhibited strong serological relationship. In this study, a comparative analysis was made on the nucleotide sequences of DNA-3 and DNA-4 components of both BBTV and ABTV. These two DNA components contain the major ORFs encoding for the coat protein and movement protein. Primers targeting a 586 bp gene fragment of the DNA-3 component and a 380 bp gene fragment of the DNA-4 component were used for PCR amplifications. The resulting PCR products were cloned, sequenced and subjected to pairwise alignments. The mean sequence difference between BBTV and ABTV was 4 to 7% for the DNA-3 fragment and 2 to 4% for the DNA-4 fragment. Comparisons were also made for specific regions of the ORF and the encoded amino acids. The low sequence heterogeneity obtained for the two major ORFs suggests that these two viruses are strains or biotypes and not distinct Nanovirus species.

Keywords: abaca, banana, abaca bunchy top nanovirus, banana bunchy top nanovirus

BSD No. 37

SHELL SHAPE CHANGES IN NINE SPECIES OF MARINE, FRESHWATER AND LAND SNAILS USING SUPERIMPOSITION AND THIN-PLATE SPLINE ANALYSIS OF LANDMARKS IN DIGITIZED IMAGES OF SHELLS

Rizalyn T. Borra and Cesar G. Demayo
Department of Biological Sciences, College of Science and Mathematics
MSU-Iligan Institute of Technology, Iligan City
Email: c_gdemayo@yahoo.com

Morphometrics involves the quantitative study of form. Form is intuitively understood that is consisting of size and shape which are never biologically independent but are instead inextricably interrelated. In this study continuous data was obtained to assess variations in the form of the shell of the snail. This was determined by employing geometric Morphometrics, a method used to obtain detailed shape information. In general, geometric morphometric methods provide greater power than the traditional methods because the position of the landmarks can be retained and can be graphically reconstructed. Meaning, it preserves geometry of object studied and it allows visualization of shape differences between specimens and between group means in specimen shape thus was used in the study. Shell shapes of a marine snail *Marmorostoma chrysostomum,* freshwater snails namely *Pila polita, Belamaya angularis, P pomacea, Pseudosuccinae peregrina* and an unknown species (designated as "unknown sp. e") and three species of land snails namely *Achatina fulica* and two unknown species (designated unknowns b and c) were used since the shell of this group is either spherical or heliciform or elongate ovate having three to five sutures with wide oval or circular aperture. It has no siphonal canal and the outer lip of the aperture is not reflected. These nine species have twenty-one (21) homologous landmarks identified thus were the bases for their use in this study. Intraspecies and between species shape variations were examined using procrustes superimposition and thin-plate spline (TPS) analysis to examine local and global sources of variations in shapes. Results of procrustes superimposition showed the landmark points in all the snails fall on the same location and the general shape of the shell structure was determined. Distinct differences can now be determined and variations in the deformation between the species can be visually detected. Procrustes analysis was therefore an important means of shape comparison because it removes or eliminates the differences in rotation, translation and scaling of forms. After the landmark configurations were superimposed, residuals were modeled with the Thin-Plate Spline (TPS) transforming one coordinate system into another. The parameters of the TPS transformation were used to explore patterns of shape changes between objects. Shape changes as deformation within species of snail's shell included in this group was made possible by fitting an interpolation function to the aligned

landmark coordinates of each specimen against the reference configuration, so that all homologous landmarks coincide. Variations between species were observed as the shells vary in landmark points where local and global deformations have occurred. These can easily be observed from the graphical presentation of the changes in individual shapes.

Keywords: *Superimposition, Thin-plate Spline Analysis, Geometric Morphometrics*

BSD No. 38

SIZE AND SHAPE VARIATION IN POPULATIONS OF *Achatina fulica* AND *Amphidromus maculiferus*: A MULTIVARIATE MORPHOMETRIC APPROACH

Ken P. Dadula, Mark Anthony J. Torres and Cesar G. Demayo

Department of Biological Sciences, College of Science and Mathematics
MSU-Iligan Institute of Technology, Iligan City
Email: c_gdemayo@yahoo.com

Size and shape variation in selected populations of the land snails *Achatina fulica* and *Amphidromus maculiferus* was determined using landmark- and distance-based morphometric analysis. Three hundred thirteen *A. fulica* and ten *A. maculiferus* were sampled from Mt. Agad-agad, Iligan City and from Balangao, Zamboanga-Sibugay. All samples were scanned at 600 dpi with the snails carefully oriented with the aperture resting on the bed of the scanner. The two land snail species have obvious spiral ornamentation which allows determination of biologically meaningful landmarks, of which twenty-two points were digitized using the ScionImage software. This study used distance variables based on the twenty two landmarks with an objective of comparing the sizes of the different samples. Distances between points were calculated using the Euclidean Distance Matrix Algorithm. The measurements include the shell's total length, which measures the maximum distance parallel to the axis of coiling from the shell apex to the basal apertural edge; total width of the snail shown as the maximum distance perpendicular to the axis of coiling; height of the spire which is also the distance from the apex to the aperture–suture junction; distances between the included major spiral chords; and the angulations on the corner of the aperture. These measurements give a balanced overview of the shape of the snail's shell, with detailed representation of the whorls characteristic of each species. Principal Component Analysis (PCA) was done to determine components with defined biological meanings that account for the maximum variation of the snail samples. This multivariate method allows numerous variables to be collected, and the significant fine-scaled variation to be

extracted. Loadings for each variable in each principal component represent that variable's contribution to the variance explained by that principle component. Cluster analysis was also employed to group snails with similar sizes together. Then, test for significant differences among the snails was done using the Discriminant Function Analysis (DFA). Shape differences among the different snail samples was determined by subjecting the raw x and y coordinates of the twenty two digitized landmarks to Procrustes fitting. This procedure eliminates both rotational and size translation allowing for the analysis of snail shape variation only. The Procrusted-fitted coordinate values were also subjected to PCA, Cluster analysis and DFA.

Results showed no significant size differences among the snail samples from the two locations based on the computed interlandmark distances. A variation in shell shapes between the two species is evident from the reconstructed images of the snails. PCA of the Procrustes-fitted values returned a total of thirty-six significant components with the first component explaining ninety-eight percent (98%) of the observed variation. This variation is summarized in a scatter plot with the Zamboanga population shown to have broader apices, more expansive aperture and wider spire base. Cluster analysis of the Procrustes-fitted values separates the three populations into three well-defined groups. DFA of the data reveals that the variations observed are statistically significant. These results are further discussed in the light of ontogenetic allometry and parasite-induced changes in the morphology of the snail.

Keywords: procrustes analysis, superimposition, multivariate morphometrics

BSD No. 39

HOMOSEXUAL ORIENTATION IN FILIPINO GAY MEN IN RELATION TO RELATIVE LENGTH OF THE SECOND AND FOURTH FINGERS (2D:4D RATIO), DEVELOPMENTAL INSTABILITY AND FRATERNAL BIRTH ORDER

Cesar G. Demayo*, Mark Anthony J. Torres, Christian Mae T. Tuanzon and Ruffy Allan V. Genilo

Department of Biological Sciences, College of Science and Mathematics
MSU-Iligan Institute of Technology, Iligan City
Email: c_gdemayo@yahoo.com

Variations between homosexual male, heterosexual male and heterosexual female populations were assessed based on relative lengths of the second and fourth fingers (2D:4D ratio), fraternal birth order and developmental instability.

Our study on homosexual and heterosexual 2D:4D ratios revealed high heterosexual male 2D:4D ratio than homosexual men. Comparison of the frequency distribution of the 2D:4D ratios also showed that for most homosexuals, the ratio is lower than of the heterosexual men. It was shown that homosexual men's ring finger lengths were shorter than heterosexual men's. The index fingers however show that homosexual men's index finger lengths did not differ from heterosexual men. It is suggested from this study that there is a correlation between homosexual orientation and the ratio of relative length of the second and fourth fingers of the hand thus confirming previous reports.

Many other studies on sexual orientation in humans also argued to be possibly influenced by levels of prenatal sex steroids which canalize neurodevelopment along sex-typical (heterosexual) or sex-atypical (homosexual) lines. We measured developmental instability by subtracting the left from the right value of the nineteen (19) bilaterally symmetrical traits (FA) among one-hundred (100) homosexual males, fifty (50) heterosexual males and fifty (5) females. These include digits on the right and left hands and feet, ears and face, lengths and width of right and left hands and feet. It is argued by many researchers that higher FA values indicate higher incidence of developmental instability. Seven (7) fluctuating asymmetry (FA) indices were used. The normality of the frequency distributions was evaluated through frequency histograms that test various aspects of distributions such as skewness, leptokurtosis and platykurtosis. Results showed variations between males, females and homosexuals based on FA values. However, homosexual males exhibited the highest FA values in all the indices used indicating high developmental instability in homosexuals than heterosexual males and females which showed low FA values.

Many earlier studies have repeatedly shown that older brothers increase the probability of homosexuality in later-born males. The collective findings suggest that birth order is perhaps the single most reliable demographic difference between homosexual and heterosexual men.

We conducted a study in 2004 and 2005 involving one-hundred seventy five (175) homosexuals and 100 heterosexual males assessing their fraternal birth order. The odds and probability of homosexuality as a function of number of older brothers and attributable fraction with zero (0) to *n* number of older brothers were computed. The attributable fraction, an index referring to the difference between the prevalence of homosexuality among men with one older brother and the prevalence among men with no older brothers calculates how much of the probability is attributable to fraternal birth order. The results of the study showed there is an increase in the probability of being a homosexual as the number of older brother increases. The results of the current study confirmed earlier reports that the older the brothers a boy has, the more likely he is to develop a homosexual orientation.

Keywords: fraternal birth order, 2D:4D ratio, developmental instability, fluctuating asymmetry

BSD No. 40

SCREENING FOR AMPICILLIN RESISTANCE GENES FROM *VIBRIO HARVEYI* NBRC 15634 AND PHILIPPINE *VIBRIO* ISOLATES

Prima Fe R. Franco[1,2] and Cynthia T. Hedreyda[1]

[1]National Institute of Molecular Biology and Biotechnology
College of Science, University of the Philippines
Diliman, Quezon City1101
[2]College of Arts and Sciences, Department of Biology
Mariano Marcos State University
Batac, 2906, Ilocos Norte;
Email: pfrfranco@digitelone.com

Vibrio harveyi, the bacterium implicated in luminous vibriosis, causes mass mortalities in hatchery-reared larvae of the black tiger shrimp *Penaeus monodon*. Antibiotics are commonly used in hatcheries and grow-out ponds as prophylactic agents for the prevention of larval bacterial infection. *V. harveyi* isolates from moribund shrimps are resistant to antibiotics. The ability of such isolates to develop resistance to antibiotics like ampicillin was explored in this study.

Preliminary results of our studies showed that a strain of *V. harveyi* becomes resistant to ampicillin when transformed with pUC18 with ampicillin resistance. Confirmation of the transformation process, however, showed that the genomic DNA of the untransformed strain contains an ampicillin resistance gene. This was done via Polymerase Chain Reaction (PCR) using primers designed based on the ampicillin resistance genes in pUC18. The presence of the gene in the reference and some Philippine *Vibrio* isolates was observed when expected amplicons approximately 700 bp were obtained. Optimized PCR conditions for 30 cycles include initial denaturation at 94 °C for 5 minutes, denaturation at 94 °C for 1 min, annealing at 57 °C for 1 min, elongation at 72 °C for 1.5 min and final elongation at 72 °C for 5 min. The PCR products were purified using NucleoSpin purification kit and submitted for sequencing. BLAST search results show that the obtained genes were 98% similar to the ampicillin resistance gene in pUC18.

Keywords: *Vibrio harveyi*, Phillipine *Vibrio* isolates, ampicillin resistance genes

BSD No. 41

Relative Warp Analysis to Explore Scapular Shape, Mandible and Pelvic Girdle in Fruit and Insectivorous Bats

Cesar G. Demayo, Mark Anthony J. Torres, Angela Marie Cariaga, Gianhope Silao, Putri Hadjariah Baser and Leonard Patrick Gabato

Department of Biological Sciences, College of Science and Mathematics
MSU-Iligan Institute of Technology
Email: c_gdemayo@yahoo.com

One method that can be powerful in establishing structure-function relationships is the use of relative warp analysis of digitized images. Relative warp analysis is essentially a principal component analysis of the covariance matrix of partial warp scores produced by thin-plate splines. The technique produces the power of thin-plate splines to produce a visualization of morphospace via the transformation grids. We elucidated the functionality of this method to explore scapular, mandibular and pelvic girdle shapes in fruit- and insectivorous bats. Relative warp analysis can provide both a descriptive and quantitative description of shape. Adult specimens of bats that were accidentally caught and injured in sampling by researchers and by fruit farmers who consider the bats as pests in their farms and from the museum were processed and the images generated by digitization at 600 dpi using high resolution computer scanners. Ten (10) landmark coordinates of the mandible, eleven (11) from the scapula and twelve (12) from the pelvic girdle were identified and transported in a processing software providing translation, rotation, and resealing of landmarks with reference to a specified baseline. Mean shapes for each species were determined by averaging shape coordinates for each landmark and these means were imported to a paleontological statistics software developed by Hammer in 2002 for relative warp analysis. The mean shapes for each species were compared to the reference form to generate partial warps on the same partial warps. Shape differences for each species were then revealed by examination of associated partial warps in the transformation grids. A model for the deformation of one shape to another was then made possible by superimposition method calculating the "best fit" between the shapes. A UPGMA (unweighted pair group method using averages) cluster analysis was performed on the resulting matrix to explore patterns of shape in the scapula, mandible and pelvic girdle.

Results of relative warp analysis of the mandibles of the bats show that the evolutionary shifts in diet are reflected on the changes in mandibular shape especially in the coronoid process and angle of the jaw. Significant differences in shapes of the mandible between species, families and between the frugivorous and insectivorous group were observable in these regions. Higher coronoid process

can be found in jaws of hard object diet specialist such as frugivorous bats and are reduced in the insectivorous bats. The scapular changes within and between groups of bats were similar and were characterized by the enlargement of the teres major process, a landmark that serves as attachment of the teres major muscle which is very important in flight. This result is expected since all the bats are strong flyers. For the pelvic shape, results show major changes in the posterior interior spine, dorsal margin of the crest of the ilium, anterior margin of the ilium, ventral margin of the crest of the ilium iliopectineal eminence, both anterior and posterior margins of the obturator foramen. This result on the pelvic girdle variations reflects group status in bats. The shape differences in these landmarks also indicate the launching differences between the insectivorous and frugivorous bats. The results of this study indicate that new methods like the relative warping method in morphospace studies can be of prime importance in establishing structure function relationships in living organisms.

Keywords: *relative warps, frugivores, insectivores*

BSD No. 42

GENETIC ANALYSIS OF *Allium* ALLIINASES BY SEQUENCE ALIGNMENT AND PHYLOGRAM CONSTRUCTION

Ralph Arnold S. Lasala*, Vivian A. Panes, Vivian S. Tolentino and Merab A. Chan

Department of Biology, Ateneo de Manila University
Loyola Heights, 1108 Quezon City
Email: rlasala@ateneo.edu

Bioactive compounds in garlic (*Allium sativum*) and other *Allium* species originate from thiosulfinates such as allicin, which are produced by the action of the enzyme alliinase (EC 4.4.1.4) on cysteine derivatives, through cleavage of carbon-sulfur bonds. In this study, sequences of alliinases from member species of the genus *Allium* were used in order to describe phylogenetic relationships among *Allium* plants. Using BLAST (Basic Local Alignment Search Tool) searches on NCBI (the USA National Center for Biotechnology Information) databases along with searches on MapViewer and ExPASy (Expert Protein Analysis System) servers, alliinase and alliinase-related sequences within the genus *Allium* (*A. ascalonicum, A. cepa, A. chinense, A.fistulosum, A. giganteum, A. sativum, A. schoenoprasum, A. tuberosum, and A. wakegi*) were obtained and analyzed. By multiple sequence alignments using ClustalW and various distance matrix computations, cladograms and phylograms were constructed to characterize the

genetic distances among *Allium* species. Ninety nine (99) residues were found to be identical in all sequences used in the alignment. Sixty five (65) conserved substitutions and 43 semi-conserved substitutions were observed. The phylogram, a branching diagram (tree) assumed to be an estimate of a phylogeny wherein branch lengths are proportional to the amount of inferred evolutionary change, clustered the alliinase sequences from *A. fistulosum, A. giganteum, A. sativum, A. tuberosum* under one clade, while the *A. cepa* and *A. wakegi* alliinases were clustered separately. Molecular structures of the various alliinase sequences were also generated using Swiss-PDB Viewer or Deep View, which showed similar patterns of the arrangement of the component modules (a homodimeric structure with each monomer composed of the N-terminal domain including the EGF-like domain, followed by the central domain residues, and then the C-terminal domain). This study significantly contributes to our understanding of the genetic structure of alliinase expression among members of the genus *Allium*. Studies on the detailed structure of the conserved domains are required in order to elucidate further the evolution of the alliinase gene family.

Keywords: alliinase, Allium, alignment, phylogeny, genetic distance, garlic, onion, leek, chive

BSD No. 43

ISOLATION OF PLASMIDS FROM PHILIPPINE *NAEGLERIA* SP.

Fides Marie DS. Francisco[1] and Windell L. Rivera[1,2]

[1]Institute of Biology, College of Science
University of the Philippines Diliman
[2]Molecular Protozoology Laboratory
Natural Sciences Research Institute,
University of the Philippines Diliman
Email: windell.rivera@up.edu.ph

Naegleria is a genus of free-living soil amoebae. One species, *Naegleria fowleri*, is opportunistically pathogenic in mammals and is the etiological agent of primary amoebic meningoencephalitis (PAM). The species is the third known eukaryotic organism with ribosomal RNA gene (rDNA) plasmid. The rDNA structure has been described in the strain of *Naegleria gruberi*; the genes carried on a 14-kilobasepair, circular, extrachromosomal DNA plasmid. Soil and water samples were obtained and tested for the presence of *Naegleria* through the enflagellation test and polymerase chain reaction (PCR). Samples were then subjected to plasmid extraction, and visualized using agarose gel electrophoresis (AGE). Bands with a

size of 14-kb were observed in all samples after AGE of the extracted plasmid. This study substantiates the presence of extrachromosomal plasmid in Philippine *Naegleria* species.

Keywords: *Naegleria*, ribosomal DNA plasmid, enflagellation test, polymerase chain reaction

BSD No. 44

RESTRICTION ENZYME ANALYSIS OF THE 18S SSU rDNA OF PHILIPPINE *ACANTHAMOEBA* ISOLATES

Jessica Anne A. Gandionco[1] and Windell L. Rivera[1,2]

[1]Institute of Biology, College of Science
University of the Philippines Diliman
[2]Molecular Protozoology Laboratory
Natural Sciences Research Institute
University of the Philippines Diliman
Email: windell.rivera@up.edu.ph

Acanthamoeba, the most common protist found in soil, are a genus of small, free-living, and ubiquitous amoebae that exhibit a biphasic life cycle composed of vegetative trophozoite stage and a physiologically static cyst stage. They are the causative agents of *Acanthamoeba* keratitis (AK) and granulomatous amoebic encephalitis (GAE). However, despite its medical importance, the subgenus classification of *Acanthamoeba* is still problematic. Riboprinting is a promising method for the rapid ideotification of unknown *Acanthamoeba* isolates. It involves the use of polymerase chain reaction (PCR) coupled with restriction fragment length polymorphism (RFLP) of the nuclear small subunit ribosomal RNA gene (SSU rDNA). Philippine *Acanthamoeba* isolates were collected from soil samples obtained from different geographic locations in the country. These were analyzed based on morphological characteristics as well as through the restriction enzyme analysis of 18S SSU rDNA. Majority of the isolates exhibited the group II cyst morphology. Data from riboprinting were analyzed using cluster analysis. Using this method, this study was able to prove that genetic diversity exists in Philippine *Acanthamoeba* isolates, but this diversity is independent of the location from where the isolates were obtained.

Keywords: *Acanthamoeba*, riboprinting, genetic diversity

BSD No. 45

UTILIZATION OF PCR FINGERPRINTING FOR THE DISCRIMINATION OF A PHILIPPINE HYPERVIRULENT *RALSTONIA SOLANACEARUM* TOMATO STRAIN

Jocelyn T. Zarate,[1] Asuncin K. Raymundo*[2] and Caitilyn C. Allen[3]

[1]National Institute of Molecular Biology and Biotechnology (BIOTECH) and
[2] Institute of Biological Science, College of Arts and Sciences,
University of the Philippines Los Baños, College, Laguna 4031,
[3]Department of Pathology, University of Wisconsin-Madison, USA
Email: joytzarate@yahoo.com ; cza@plantpathwisc.edu; akr@laguna.net

Ralstonia solanacearum is the causal agent of the serious disease known as bacterial wilt affecting greater than 200 plant species. We are working on the characterization of a hypervirulent *R. solanacearum* substrain (T731) that wilted previously resistant tomato varieties compared to its wild type strain (T523). The presence of additional virulence factor might impact disease management. Different PCR- based DNA fingerprinting techniques were evaluated for differentiation of the hypervirulent mutant strain from the wild type strain. BOX and ERIC PCR were unable to differentiate the two strains while REP PCR and Pulse Field Gel Electrophoresis (PFGE) generated differential bands unique to the hypervirulent strain. Three rare cutting restriction enzymes used in generating the macrorestriction patterns in PFGE showed that strain T523 and T731 were very similar to each other except for one to three differential bands. Cloning and sequencing of a 414 bp REP PCR generated DNA fragment which was present only in the hypervirulent strain showed that the sequence is present in the megaplasmid of the sequenced *R. solanacearum* strain GMI1000, a tomato isolate from French, Guyana in three different portions. The highest homology was present at the 5' side of the transposase protein of the insertion element 13 (ISRso13). G+C content of the DNA fragment was 57%, as differentiated from the average G+C content of 67% in the rest of the megaplasmid and chromosome. *In silico* protein translation of the DNA fragment showed it was 61% identical to a hypothetical prjohnny depp otein in *Burkholderia cepacia.* The clone REP–PCR fragment might have a possible role in the hypervirulence of strain T731.

Keywords: *Ralstonia solanacearum*, DNA fingerprinting

BSD No. 46

POTENTIAL OF *DESMODIUM CINEREA* AND MYCORRHIZAL FUNGI FOR BIOREMEDIATION OF COPPER RICH SOILS

Joden M. Adiova[1], Nelson M. Pampolina[1] and Nelly S. Aggangan[2]

[1]Bachelor of Science in Forestry Student
Associate Professor
College of Forestry and Natural Resources,
University of the Philippines Los Baños, College, Laguna
[2]University Researcher
National Institute of Molecular Biology and Biotechnology
UPLB, College, Laguna 4031
Tel/Fax: 049 536 0563, Email: nea@laguna.net

This study was conducted to determine the effect of arbuscular mycorrhizal fungi (AMF) for the phytoremediation of copper (Cu) rich soils using *Desmodium cinerea* and to assess if this forage legume is safe for livestock grazing. Seeds of *D. cinerea* were sown in pots containing soil amended with increasing levels of Cu from nil to 400 ppm or from nil to 1600 ppm Cu in two experiments. Half of the seedlings were inoculated or uninoculated with AMF during sowing. In the third experiment, inoculated and uninoculated seedlings were plantd in mine tailing soil collected in Marinduque with 82 ppm Cu. The experiments were established in a screenhouse following Randomized Complete Block Design with ten replicates. Height was monitored forth nightly while diameter, leaf area and biomass were measured at harvest (two months).

Generally, AMF inoculated plants were taller, heavier, greater leaf area and diameter than the uninoculated plants. In the first experiment, Cu concentrations of 100 to 200 ppm improved the growth of *D. cinerea* implying that this forage crop requires 100 to 200 ppm Cu for maximum normal growth. In the second experiment, highest height, diameter, leaf area and biomass were obtained from plants with no added Cu. Addition of Cu gave corresponding decreases in height, diameter, biomass and leaf area of inoculated and uninoculated plants. Cu level of 800 ppm seemed to be critical for both inoculated and uninoculated plants because beyond this level, leaf chlorosis, stunted growth and early leaf fall were observed although these were more evident in uninoculated plants. Furthermore, the roots were confined in the original potting soil. Seedling survival rate at 1600 ppm Cu was 30% in the uninoculated while 70% with inoculation. In mine tailing soil, mycorrhizal *D. cinerea* survived throughout the two months period whereas all the uninoculated ones died within one month. The results show that AMF alleviated Cu toxicity of *D. cinerea*, although it is unclear whether AMF does this by improving nutrient absorption or by binding mechanisms. Plant tissue analyses indicate that transport

of Cu within the plant is prevented by the presence of mycorrhizal fungi, thus, *D. cinerea* planted in Cu mine sites is safe for livestock.

Keywords: Arbuscular mycorrhizal fungi, *Desmodium cinerea*, copper, bioremediation

BSD No. 47

ANTIBODY-BASED DETECTION OF MONODON BACULOVIRUS (MBV) IN *Penaeus monodon*

Ma. Michelle D. Peñaranda[1], Elena S. Catap*[2], James Torres[3], and Ikunari Kiryu[4]

[1]Department of Pathobiology, University of Washington
Seattle, WA, USA,
[2]*Institute of Biology, College of Science, University of the Philippines
Diliman, 1101 Quezon City, Philippines
[3]Department of BiologyUniversity of the Philippines in the Visayas
Miag-ao, 5023, Iloilo, Philippines
[4]Japan International Research Center for Agricultural Sciences (JIRCAS) /
Fisheries Division,1-1, Ohwashi, Tsukuba, Ibaraki, 305-8686 Japan
Tel. No. (02) 981-8500 loc. 3727; Telefax No. (02) 920-5471
E-mail: escatap@up.edu.ph

Spherical baculovirosis due to monodon baculovirus (MBV) infection is one of the crustacean diseases listed by the Office International des Epizooties (OIE) that is of social and economic importance. In the Philippines, monodon baculovirus (MBV) is a prevalent virus in the aquacultured tiger shrimp, *Penaeus monodon* Although DNA-based techniques, such as the polymerase chain reaction (PCR), had been very useful in screening broodstock and postlarvae these techniques require sophisticated equipments and very skilled staff in very clean laboratories, which cannot be established easily in rural shrimp farm areas. On the other hand, antibody-based methods could be developed in simple but field-friendly formats with comparable sensitivity and specificity. Therefore, these experiments were undertaken to produce polyclonal antibodies that could be used as a tool in antibody-based diagnosis of MBV infection in *P. monodon*

Polyclonal antibodies (PAbs) against MBV were developed by immunization of rabbits with purified virus. Using indirect fluorescent antibody test (IFAT), intense reaction was observed in hepatopancreatic impression smears and paraffin-embedded sections of MBV-infected postlarvae (PL-15 to 20) but not in smears and/or sections infected with hepatopancreatic parvovirus (HPV) and white spot

syndrome virus (WSSV). Similarly, immunohistochemical tests on paraffin-embedded sections showed positive coloration (brownish red to rose red precipitate) in MBV-infected cells not found in sections infected with either HPV or WSSV. Indirect ELISA revealed that the antisera could detect 4 to 100 ng of purified MBV. Overall, the PAbs obtained in this study have potential applications in the rapid, sensitive and simple detection of MBV provided that further purification of the antisera is undertaken to further minimize some background reactions.

Keywords: Monodon baculovirus, MBV, polyclonal antibodies, IFAT, ELISA, immunohistochemistry, Penaeus monodon, immunodetection assays

CHEMICAL, MATHEMATICAL, PHYSICAL SCIENCES

CHEMISTRY

CMPSD No. 1

BIMOLECULAR REACTION RATE CONSTANT OF THREE-BODY SYSTEMS: AN APPLICATION TO TRANSITION STATE THEORY

Christopher E. Ambe[1], Edgar W.Ignacio[2], Dante D. Dinawanao[3], and Jingle B. Magallanes[4]

[1]Department of Chemistry, CSM
MSU-Iligan Institute of Technology, 9200 Iligan City
[2] Northern Mindanao State Institute of Science and Technology,
Ampayon, 8600 Butuan City
[3] Department of Computer Science, SCS,
MSU-Iligan Institute of Technology, 9200 Iligan City
[4] Department of Physics, CSM
MSU-Iligan Institute of Technology, 9200 Iligan City
Email: csm-cea@sulat.msuiit.edu.ph

Transition state theory (TST), also known as Activated Complex Theory (ACT), pictures elementary reaction and presumably stated that there exists an activated complex having a higher Gibbs free energy in between the reactants and product. The sole basis of the transition state theory in determining the rate constant of bimolecular reaction is on the application of statistical mechanics to reactants and activated complexes. In this study, the potential energy surface (PES) for H + HF à H_2 + F (1), H + HCl à H_2 + Cl (2), and H + CH_4 à H_2 + CH_3 (3) were theoretically

scanned using two potential energy functions, the London-Eyring-Polanyi-Sato (LEPS) and Lennard-Jones 12-6 potential.

Preliminary mathematical calculations, as well as the visualization and plotting of the PES for these reactions, were carried out using Mathematica 3.0 running on PC workstation. The results showed that calculation of the rate constant of these bimolecular reactions using TST is inappropriate when Lennard-Jones potential is used. This observation is due to the absence of a real transition structure or a first order saddle point (manifested by the single imaginary frequency) along the minimum energy path (MEP) of the reaction.

In this paper, the rate coefficients of these reactions were determined using the LEPS potential. The optimization of the equilibrium structures for the first order saddle point was performed using Newton-Raphson method. Furthermore, vibrational analysis on the saddle point of the reactions (1), (2) and (3) revealed a single imaginary vibrational frequency of 336.443*i*, 1009.99*i*, and 1198.81*i*, respectively. The respective thermal rate coefficient obtained using TST for these reactions at 298.15 K were 7.4912×10^{-11} $cm^3mol^{-1}s^{-1}$, 4.0300×10^9 $cm^3mol^{-1}s^{-1}$, and 9.3708×10^6 $cm^3mol^{-1}s^{-1}$. These results were supported by the calculated activation energy (E_a) of 132.988 kJ/mol, 26.1519 kJ/mol and 35.7400 kJ/mol, respectively.

Keywords: Lennard-Jones potential; rate coefficient; minimum energy path; activation energy.

CMPSD No. 2

SYNTHESIS OF GOLD NANOCRYSTALS IN 1-BUTYL-3-METHYL IMIDAZOLIUM LAURYL SULFATE VIA SEEDING GROWTH APPROACH

[1]Susan D. Arco, [1,2]Judy M. Obliosca, and [2]Michael H. Huang*

[1]Institute of Chemistry, University of the Philippines
Diliman, Quezon City, Philippines
Email: sarco@chem.upd.edu.ph
[2]Department of Chemistry, National Tsing Hua University,
Hsinchu, Taiwan
*Email: hyhuang@mx.nthu.edu.tw

We present the first synthesis of gold nanocrystals via seeding growth approach using the greener ionic liquid, 1-butyl-3-methylimidazolium lauryl sulfate, [BMIM][$C_{12}H_{25}OSO_3$], as a surfactant. The ionic liquid was synthesized by reacting 1-methylimidazole with 1-bromobutane and exchanging the bromide with lauryl sulfate ion to afford the product, [BMIM] [$C_{12}H_{25}OSO_3$]. Seeding growth approach was used to prepare larger Au nanoparticles from 2.5-nm Au seeds with the

successive addition of a growth solution containing $HAuCl_4$ and ascorbic acid. SEM and TEM results showed the presence of Au nanocrystals with particle sizes ranging from 30 to 50 nm. Selected-area electron diffraction and PXRD pattern confirmed that the particles produced are gold nanocrystals. Direct addition of ionic liquid to Au seeds in 3:1 volume ratio was also investigated to see if larger Au nanoparticles can be synthesized in one single step. TEM image and UV-vis data revealed the formation of 20–50 nm Au nanocrystals. These results show that the greener ionic liquid, [BMIM] [$C_{12}H_{25}OSO_3$], is a suitable reaction medium for the direct synthesis of larger Au nanoparticles from Au seeds.

Keywords: greener ionic liquid, 1-butyl-3-methylimidazolium lauryl sulfate, gold nanocrystals, seeding growth approach

CMPSD NO. 3
A BENZODIOXATETRAAZA CYCLIC ALCOHOL FROM THE MARINE SPONGE *Halisarca sp.*

Lydia M. Bajo[1]*, Jean Theresa O. Go[2], Mylene M. Uy[1], Franco G. Teves[3], and Myrna H. Salarza[1]

[1]Department of Chemistry, Mindanao State University-Iligan Institute of Technology, 9200 Iligan City,
[2]Department of Chemistry, Silliman University
6200 Dumaguete City, Philippines
[3]Department of Biological Sciences, Mindanao State University-Iligan Institute of Technology, 9200 Iligan City,Philippines
Email: csm- lmb@msuiit.sulat.edu.ph; lmb8557@yahoo.com

Bioassay-guided fractionation of the butanol-soluble portion of the marine sponge *Halisarca sp.* collected from Bais City, Negros Oriental afforded a cytotoxic yellow and red extracts. These extracts were isolated by subjecting the sponge biomass to methanolic extraction followed by sequential solvent partitioning using hexane, ethyl acetate and 1-butanol. The more cytotoxic BuOH-soluble portion was purified through a series of flash column chromatography using the gradient elution technique (MeOH/DCM & MeOH/H_2O). A benzodioxadiazo cyclic alcohol with an IUPAC name of (3Z,5Z) 7-methyl-2-propyl-7,8-dihydro-2H-benzo[1,9,3,4,5,6]dioxatetraazacycloundecin-8-ol was isolated from the yellow extract of the marine sponge. Its structure was elucidated on the basis of FTIR, EIMS, various one-dimensional, i.e. ^{13}C, ^1H, and two-dimensional NMR experiments, i.e. COSY, HMQC and HMBC. The compound exhibited high cytotoxicity towards the nauplii of brine shrimp *Artemia salina* with an estimated chronic LD_{50} of 323.59 ±

1.23 ppm. It was inactive against gram- positive cocci *Streptococcus pyogenes* and gram negative bacilli *Escherichia coli* and *Pseudomonas aeruginosa.* It revealed marginal antibacterial activity against gram-positive bacteria *Staphylococcus. aureus.* Its high clastogenic potential was determined using the micronucleus test and results were evaluated using statistical tools, i.e. one-way ANOVA and DMRT. The red fraction of the butanol-soluble portion exhibited a lower toxicity towards brine shrimp with an acute LD50 = 582.10±1.49 ppm but displayed significant antibacterial activity against gram-positive cocci *Str. Pyogenes* and *S. aureus.* Its DNA-breaking capacity was only demonstrated at a concentration of ¾ LD50 (LD50 was based on BSLT).

Keywords: bioassay-guided, micronucleus, clastogenic, benzodioxatetraaza, FTIR, EIMS, NMR, COSY, HMBC, HMC.

CMPSD NO. 4
SYNTHESIS OF CARBAZOLE, PHENANTHRIDINE AND PHENANTHRENE FROM *N*-ALKYLATED 2-AMINOBIPHENYLS

Evelyn C. Creencia*[a] and Takaaki Horaguchi*[b]

[a]Dept. of Chemistry, College of Science and Mathematics,
MSU-Iligan Institute of Technology, 9200 Iligan City, Philippines
[b]Dept. of Chemistry, Faculty of Science, Niigata University,
Ikarashi, Niigata 950-2181, Japan
Email: csm-ecc@sulat.msuiit.edu.ph

Due to their many practical uses, carbazoles and other heterocycles have received considerable attention particularly in terms of their synthetic methodology [1, 2]. Freeman, *et al.* reported a novel modification of the Cadogan reaction for the synthesis of a series of substituted carbazoles, with reaction times ranging from 21 to 39 h [1]. This paper reports the synthesis of carbazole, phenanthridine and phenanthrene by thermal intramolecular cyclization reactions. Thermal cyclization reactions were examined by passing vapors of *N*-alkylated 2-aminobiphenyls over calcium oxide at 450–600°C under nitrogen carrier gas, for 40 min. The results showed that the major product of the reactions of 2-methylaminobiphenyl, 2-dimethylaminobiphenyl and 2,2'-bis(dimethylamino)biphenyl was phenanthridine (38% at 500°C, 60% at 500°C, 92% at 450°C, respectively) while that of 2-benzylaminobiphenyl was carbazole (69% at 500°C). Phenanthrene was a minor product of the thermal cyclization of 2-benzylaminobiphenyl. Reaction pathways of product formation were studied by subjecting 9-methylcarbazole and 9-benzylcarbazole to similar reaction conditions. When 9-methylcarbazole was heated

at 500°C, carbazole (36%) and phenanthridine (46%) were obtained. These results show that carbazole and phenanthridine are produced via 9-methylcarbazole, which is one of the reaction pathways. When 9-benzylcarbazole was heated at 500°C, carbazole (64%), phenanthridine (20%), and phenanthrene (13%) were obtained. In this case, elimination of the benzyl group was a major reaction and yielded phenanthrene. Therefore, thermal cyclization reaction is an effective method for the preparation of nitrogen-containing heterocylces, giving good yields at shorter reaction time.

Keywords: heterocycle, synthesis, thermal reaction, carbazole, phenanthridine, phenanthrene

[1] A.W. Freeman, M. Urvoy and M.E. Criswell, *J. Org. Chem.*, **70**, 5014 (2005).
[2] A. Kuwahara, K. Nakano and K. Nozaki, *J. Org. Chem.*, **70**, 413 (2005).

CMPSD NO. 5

FLOW INJECTION ANALYSIS OF PROTEINS USING A POLYPYRROLE-METHYLORANGE COATED PLATINUM ELECTRODE

Florian R. del Mundo

Institute of Chemistry, College of Science
University of the Philippines
Diliman 1101, Quezon City
E-mail: fdelmund@yahoo.com

In this study, the potential application of the polypyrrole-methyl orange (PMO) coated Pt electrode for routine electroanalysis of three proteins was demonstrated using flow injection analysis (FIA) with pulsed amperometric detection. This technique couples process of anodic detection and cathodic reactivation where the potential is pulsed from E_1 to E_2 during detection. This was found to be more suitable for our system than controlling the potential of the PMO electrode at a fixed value. The effect of carrier stream pH on the FIA response of the different proteins was studied using buffers. It was found that the response of myoglobin is essentially independent of the carrier pH while with bovine serum albumin (BSA) and ovalbumin the optimum responses were observed around their isoelectric points. A carrier stream of I M Na_2SO_4 (~ pH 6.5) was used for myoglobin. However small, insensitive responses were observed. This observation could be due to the low signal to noise ratio because of high concentration of background electrolyte. We then decreased the carrier stream concentration to 10^{-3} M and sensitive responses were then obtained. With BSA and ovalbumin, we used a 0.1

M potassium phosphate buffer carrier stream of pH 5.0. Tripticate to quadruplet injections of 250 mL each of the protein standards were made sequentially into a 0.001 M sodium sulfate (pH 6.5) or 0.1 M phosphate buffer (pH 5.0) carrier solution at an optimum flow rate of 2 mL/min. Results showed that BSA gave the most sensitive response (49,666 mA/mM) and the lowest limit of detection (5.9×10^{-5} mM) while ovalbumin gave the least sensitive response (0.0023 mA/mM) and highest limit of detection (89 mM) among the proteins considered in this FIA study. The FIA method gave reproducible response for all proteins with precisions ranging from 0.2–14 % relative standard deviations.

Keywords: electroanalysis, polypyrrole sensor, proteins, flow injection analysis

CMPSD NO. 6

PURIFICATION OF LIPASE FROM HAUSTORIUM OF GERMINATING COCONUT *(COCOS NUCIFERA)* BY HEPARIN-BINDING AFFINITY CHROMATOGRAPHY

Lydia M. Bajo[1]*, Gina V. Batoy[2], Roberto Malaluan[3]

[1]Department of Chemistry, MSU- Iligan Institute of Technology,
Tibanga Iligan City 9200 Philippines,
[2]Deparment of Chemistry, Central Mindanao University
Musuan Bukidnon, Philippines,
[3]Chemical Engineering Technology Department
Mindanao State University-Iligan Institute of Technology
Tibanga, Iligan City, Philippines
Email: csm- lmb@msuiit.sulat.edu.ph
lmb8557@yahoo.com

Crude lipase was extracted from the haustorium of germinating coconut using phosphate buffer as extracting medium and was purified to apparent homogeneity by 90% $(NH_4)_2SO_4$ precipitation and affinity chromatography. The purified lipase (PL) bound to the heparin ligand of the column was eluted by increasing the ionic strength of the binding buffer from 0.01 M NaCl in phosphate buffer (pH 7) to 0.5 M NaCl in phosphate buffer (pH 7). This indicates that PL contains the heparin-binding site and the lipase-heparin ligand interaction is electrostatic in nature.

The obtained PL had a specific activity of 5567 U/mg with 411-fold purification and yield of 11.67%. SDS-PAGE analysis of PL showed single protein band with estimated molecular weight of 57 kDa.

The crude lipase extract (CLE) and PL had maximal activities at pH 7 and 30 min incubation time. Purification shifted the maximal activity of CLE at 40°C to 60°C.

At 0.005 mmol, their activities were enhanced greatly by Zn^{++} and to a lesser extent by Ca^{++} and Mn^{++}. Hg^{++}, Fe^{++}, and Mg^{++} inhibited lipase activity. After 27 days, CLE and PL retained 55% and 50% respectively of their initial activities. K_M and V_M values of PL were 0 .6667 M and 3333.5 U/mg, respectively. Gas chromatographic analysis of the hydrolysis products of coconut oil by PL showed 10 fatty acids with lauric acid (49.27%) as the most abundant.

Keywords: coconut, lipase, haustorium, affinity chromatography, heparin-binding, SDS-PAGE, K_M, V_M

CMPSD NO. 7
SPECTROMETRIC ANALYSIS OF THE ETHANOLIC EXTRACT FROM MAHOGANY (*Swietenia macrophylla*) SEEDS

Abigail P. Cid[1] and Lorna T. Enerva[1,2]

[1]Department of Natural Sciences, College of Science
[2]SciTech Research and Development Center
Polytechnic University of the Philippines
Sta. Mesa, Manila
Email: niabru1@yahoo.com

Swietenia macaphylla, locally known as mahogany, seeds is eaten to treat various illnesses. This study aimed to investigate the ethanolic extract of the seeds of this tree.

The mahogany seeds were extracted twice with ethanol using soxhlet method. The ethanolic extracts were fractionated by silica gel column chromatography with thin layer chromatography (TLC) confirmation. A single spot on the TLC of the chloroform: ethyl acetate: ethanol fraction was subjected to spectral analysis. The isolated extract absorbed at 247 nm, 286 nm, 296 nm, 327 nm, and 337 nm. Infrared spectra shows peaks indicating the presence of C-H bonded alkanes and alkenes, C-H aromatic rings, and C=O aldehydes, ketones, carboxylic acid, and esters. Gas chromatography/mass spectra revealed the presence of 14 known compounds.

Keywords: *Swietenia macaphylla*, mahogany, seeds

CMPSD NO. 8

BENZOYL ISOTHIOCYANATE (BITC) LEVELS OF GM AND NON-GM PAPAYA AT DIFFERENT STAGES OF FRUIT DEVELOPMENT

CS Cabanos, Evelyn Mae T. Mendoza, Roberta N. Garcia and Antonio C. Laurena

Institute of Plant Breeding (IPB), College of Agriculture
University of the Philippines Los Banos
Email: aclaurena@yahoo.com

Several transgenic lines of papaya containing the capaccs2 gene in the antisense orientation were generated from the IPB BL2 greenhouse. The antisense orientation of the ripening-related gene (capaccs2) is supposed to inhibit the biosynthesis of the ripening hormone ethylene through the down regulation of the ACC synthase gene. The presence of the capaccs2 gene were confirmed by PCR and Southern gel blot analyses using genomic DNA isolated from the transgenic lines. Preliminary studies suggest that several of these lines also exhibit the delayed ripening (DR) trait through changes in both color and tissue softening.

Unintended effects may result from the random and stable integration of the transgene (capaccs2) and therefore compositional analyses of the macro- and micronutrients, anti-nutritional and toxic factors should be provided to confirm substantial equivalence between the GM papaya and its conventional counterpart. One of the anti-nutrient or toxic principles in papaya is the presence of benzoyl isothiocyanate (BITC) especially in the fruit latex at the green immature stage. This study aims to determine the levels of BITC at different stages of maturity between GM and non-GM papaya to confirm no unintended effects during transgenesis.

Keywords: Benzoyl Isothiocyanate (BITC), papaya, delayed ripening (DR), DNA

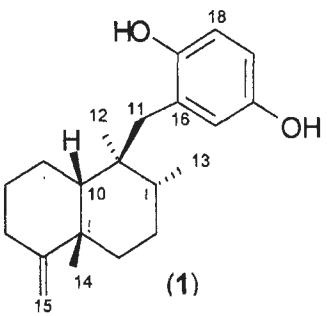
CMPSD NO. 9

A SESQUITERPENE FROM *DYSIDEA* sp. COLLECTED IN MANTIGUE ISLET, CAMIGUIN

Alexander O. Mosqueda[1*], Lydia M. Bajo[1], Franco G. Teves[2], Roberto M. Malaluan[3], Ian van Altena[4], and Mihiko Yanai[5]

[1]Department of Chemistry,
[2]Department of Biological Sciences,
[3]Department of Chemical Engineering Technology
MSU-Iligan Institute of Technology
[4]Chemistry Department, School of Environmental and Life Sciences,
University of Newcastle, Australia
[5]Natural Science for Basic Research and Development,
Hiroshima University
Email: alexmosqueda1980@yahoo.com

A sesquiterpene compound (**1**) known as arenarol was isolated from the marine sponge, *Dysidea* sp. collected from the waters of Mantigue Islet (9° 10' N, 124° 49'E), Camiguin Province. Extraction was done by Supercritical Fluid Extraction (SFE) with conditions set at 300 atm and 40°C, using CO_2 as the supercritical fluid. The metabolite was purified through reversed-phase High Performance Liquid Chromatography (HPLC) with UV detector at l_{max}, 229 nm to give a single sharp



(1)

peak with a retention time of 8.948 min. The molecular structure was elucidated on the basis of spectral evidences including Ultraviolet-Visible (UV), Infrared (IR), Electron Impact Mass Spectroscopy (EIMS), and extensive 1D and 2D Nuclear Magnetic Resonance (NMR) experiments: ^{1}H-NMR, ^{13}C-NMR, DEPT (Distortionless Enhancement by Polarization Transfer), Proton Homonuclear Correlated

Spectroscopy (COSY), Heteronuclear Multi-Quantum Correlation (HMQC) and Heteronuclear Multi-band Correlation (HMBC). The isolated secondary metabolite exhibited a very weak antimicrobial activity at 15 µg against *Staphylococcus aureus*, *Bacillus subtilis, Escherichia coli* and *Pseudomonas aeruginosa*, and showed a DNA-binding trait with a retention factor ratio of 0.76 by 1D Thin-Layer Chromatography (TLC).

Key words: *Dysidea*; NMR; sesquiterpene; arenarol; antimicrobial; DNA-binding

CMPSD NO. 10

CHEMICAL COMPOSITION OF INDUSTRIAL TREE PLANTATION SPECIES (ITPS) BARKS

Jennifer P. Tamayo[1] and Maxima E. Flavier[2]

[1]Forest Products Research and Development Institute (FPRDI-DOST)
College, Laguna 4031
[2]Institute of Chemistry, University of the Philippines
Los Baños, Laguna 4031
Email: jenny_tamayo7@yahoo.com

The bark consists of an outermost corky layer (epidermis), a layer of food-conducting tissues (phloem), and a zone between these two layers (cortex). In several species, a layer of fibrous strips (bast fiber) form an inner bark. Oils, resins, tannins, waxes, and phenolic substances may also be present in the bark and when extracted may be useful in the manufacture of certain chemicals and medicinal products.

The chemical composition of ITPS barks was analyzed using the standard methods of the Technical Association of the Pulp and Paper Industry (TAPPI). ITPS barks studied were *Acacia mangium, Eucalyptus deglupta, Eucalyptus camaldulensis, Paraserianthes falcataria, Endospermum peltatum, Anthocephalus chinensis, Samanea saman* and *Gmelina arborea.*

ITPS barks were found to contain the following components: ash (3.37 to 11.87%), hot water extractives (4.22 to 20.44%), alcohol-cyclohexane extractives (0.65 to 7.58%), caustic soda solubles (14.93 to 49.76%), lignin (14.43 to 41.04%), holocellulose (37.65 to 62.05%), total sugars (1.19 to 21.94%) and tannin (2.41 to 14.93%).

P. falcataria bark had the highest hot water extractives, total sugars and tannin content. This can a source of dyes and pigments. *A. chinensis* bark had the highest holocellulose and lowest lignin content and is a potential raw material for the pulp and paper industry. *E. peltatum* bark has the lowest total sugars and

tannin content while *E. camaldulensis* bark had the lowest hot water and alcohol cyclohexane extractives. These two species did not impart color when placed in solution and could be good adsorbent for removing heavy metals in wastewater.

Keywords: chemical composition, ITPS barks, ash, hot water extractives, alcohol cyclohexane extractives, caustic soda solubles, total sugars, holocellulose, lignin, tannin

CMPSD NO. 11

BIOMOLECULAR-CHEMICAL PROFILING OF METABOLITES FROM SEDIMENT MICROORGANISMS

Haide May G. Aloot[1], Franco G. Teves[1]*, Lydia M. Bajo[2] and Carmelita G. Hansel[3]

[1]Molecular Genetics and Microbial Technology Laboratory,
Department of Biological Sciences
[2]Department of Chemistry
MSU-Iligan Institute of Technology9200 Iligan City
[3]Department of Biology, College of Natural Sciences and Mathematics
Mindanao State University-Main Campus, Marawi City
Email: franco_teves@yahoo.com

Sediment samples from an estuarine environment were obtained near the mouth of the Layawan River, Oroquieta City, Misamis Occidental, Philippines. Three groups of marine sediment microorganisms, namely, actinomycetes, non-filamentous heterotrophic bacteria, and filamentous fungi, were targeted for isolation using standard microbiological procedures. Isolates showing antibiotic properties in two-level screening were massively grown on plates from which were obtained extracts using ethyl acetate and concentrated by rotary evaporation. Two-dimensional thin layer chromatography (2D-TLC) formats were then employed to screen for DNA-binding properties using salmon sperm DNA, by comparing the mobility indices of isolated spots (Rf_2) with control (Rf_1) visualized under ultraviolet light.

Metabolites from antibiotic-producing heterotrophic marine bacteria all showed positive but weak DNA-binding properties (average Rf_2/RF_1=0.9). Among the estuarine antibiotic-producing actinomycetes, *Streptomyces* sp. showed the highest DNA-binding activity (Rf_2/Rf_1 = 0.8). Overall, the best result for DNA affinity was obtained with the estuarine strain of *Penicillium* sp. (Rf_2/Rf_1 = 0.6). We have shortened significantly the screening process for new potential drugs based on their antibiotic and DNA-binding properties. DNA-binding is a powerful

indicator of gene regulatory functions as exemplified by specific peptide pharmaceuticals.

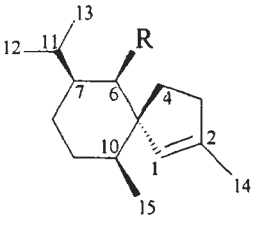
Keywords: sediment, metabolite, DNA-binding, thin layer chromatography, actinomycetes, fungi

CMPSD NO. 12
NEW BIOACTIVE SPIROCYCLIC SESQUITERPENES FROM THE MARINE SPONGE *GEODIA EXIGUA*

Mylene M. Uy[a*], Shinji Ohta[b*], Mihoko Yanai[c], Emi Ohta[b]
Toshifumi Hirata[d] and Susumu Ikegami[b*]

[a]Department of Chemistry, MSU- Iligan Institute of Technology
9200 Iligan City, Philippines
[b]Nagahama Institute of Bio- science and Technology
1266 Tamura-cho,Nagahama, Shiga 526-0829, Japan.
[c]Natural Science Center for Basic Research and Development
Hiroshima University, 1-3-1 Kagamiyama,Higashi-Hiroshima 739-8526, Japan.
[d]Department of Mathematical and Life Sciences, Graduate School of Science,
Hiroshima University, 1-3-1 Kagamiyama, Higashi-Hiroshima 739-8526, Japan.
*Email: csm-mmu@sulat.msuiit.edu.ph

The sea urchin embryo provides an excellent model system to study the molecular mechanisms of cell fate specification. The MeOH extracts of the marine sponge *Geodia exigua* Thiele collected off Oshima, Kagoshima Prefecture, Japan, inhibited the formation of normal plutei during the development of the sea urchin *Hemicentrotus pulcherrimus*. Bioassay-guided fractionation of the MeOH extracts resulted in the isolation of three new spirocyclic sesquiterpenes designated exiguamide (**1**), exicarbamate (**2**) and exigurin (**3**) together with (-)-10-*epi*-axisonitrile-3 (**4**). All four compounds possess the spiro[4.5]decene skeleton and



1 R : -NHCHO
2 R : -NHCOOCH$_3$
3 R : -NHCOCH$_2$N(CH$_3$)CH$_2$COOCH$_3$
4 R : -NC
5 R : -NH$_2$

their structures were determined on the basis of spectroscopic data. The structure of **1** was confirmed by X-ray crystallographic analysis and the absolute configuration was determined by applying the modified Mosher's method on its amine derivative (**5**).

This research work illustrates the first occurrence of an isonitrile co-existing with formylamino, a methoxycarbonylamino, and a 2-(methoxycarbonylmethyl-methyl-amino)-acetylamino groups.

When fertilized eggs of the sea urchin (*H. pulcherrimus*) were cultured in the presence of 0.4 – 12.0 mM **1**, they divided equally to form 16-cell embryos that were comprised of sixteen cells of the same size. After passing through the blastula and then gastrula stages, the **1**-treated embryos developed to form spicule-deficient plutei. There are very few substances having the biological activity. The analogous sequiterpene having the same spiro[4.5]decene skeleton with a methoxycarbonylamino (**2**), a 2-(methoxycarbonylmethyl-methyl-amino)-acetylamino (**3**) or an isonitrile(**4**) group in place of the formylamino group of **1** did not exhibit such activity. Exiguamide (**1**) is considered a useful tool for elucidating the mechanism of cell fate specification during sea urchin embryogenesis.

Keywords: biologically-active compounds; natural products; sesquiterpenes; marine sponges; sea urchin

CMPSD NO. 13
INSTRUMENTAL ANALYSIS OF THE SEEDS OF *Artocarpus heterophyllus Lam. Moracea* (JACKFRUIT)

Lorna T. Enerva and Abigail P. Cid

College of Science and SciTech R and D Center
Polytechnic University of the Philippines, Sta. Mesa, Manila
Email: lornaenerva@yahoo.com

Jackfruit is a medium-sized tropical fruit tree reaching 15-20 meters in height. Inside, the fruit is made up of large, yellow bulbs enclosing an oval light-brown seeds. All parts of the tree produce sticky, white latex, but gum-free genotypes have been identified in India. Different parts of the jackfruit tree have medicinal properties. This study was undertaken to elucidate the structure of the seeds of langka using instrumental methods of analysis which is used as a tonic agent.

Ten grams of the air dried seeds was extracted with ethanol for 48 hours by soxhlet extraction. Two extractions were made and the combined extracts were concentrated to fifteen milliliters using a rotary evaporator. Column chromatography

was used to separate the constituents of the extract using $CHCl_3$-ethylacetate-ethanol as solvent.

The isolates were colored yellow, insoluble in water but soluble in organic solvents. The infrared spectra of isolates 4 and 9 showed the presence of OH, CH stretch, C=O, aromatic ring, CH_3 and C-O bonds. The maximum wavelength for isolate 4 was 245 nm and isolate 9 was 239 nm respectively. Gas chromatography-mass spectra were used to further elucidate the structure of the compound. Using GC-mass spectra for isolate 4 the following compounds were identified: 9,19-cycloergost-24(28)-en-3-ol. 4,14-dimethyl-acetate(3b,4a,5a) $C_{32}H_{52}O_2$ MW 468, silicic acid; dietheyl bis(trimethylsilyl)ester, MW 468, $C_{10=}H_{28}O_4Si_3$,MW 296; 9,19 cycloergos-2(28)-ene-3-ol, 4, 14-dimethyl-acetate $C_{32}H_{52}O_2$ MW 468; squalene $C_{30}H_{50}$ Mw 410 and cyclotrisiloxane, hexamethyl $C_6H_{18}O_3Si_3$ MW 222.

For isolate No 9, the following compounds were identified 1,16-cycloreynan-17-oic acid, 19,20-didehydro, methyl ester $C_{20}H_{34}O_2$ MW 294; hexanedioic acid,bis(2-rthylhexyl)ester $C_{22}H_{42}O_4$ MW 370; 1,2 benzenedicarboxylic acid, diisooctyl ester $C_{24}H_{38}O_4$ MW 390; 2,4,6-cycloheptatriene-1-one,35-bis-trimethylsilyl $C_{13}H_{22}OSi_2$ MW 250; 9,19-cyclolanost-24-ene-3-ol, acetate $C_{32}H_{52}O_2$ MW 468 and 9,19-cycloergost-24(28)-ene-3-ol,4,14 dimethyl-acetate (3b,4a, 5a) $C_{32}H_{52}O_2$ MW 468. The structure of the isolates can be further elucidated using nuclear magnetic resonance.

Keywords: tonic, infrared spectra, ultraviolet spectra, mass spectra, gas chromatography.

CMPSD NO. 14
ENGINEERING OF ARGININE AND GLUTAMIC ACID ON ANTIBODY CH3 DOMAIN INTERFACES PROMOTES HEAVY CHAIN HETERODIMERIZATION

Maria Aiko Angela A. Diaz[1], Maria Katrina C. Canlas[1]
Eduardo A. Padlan[2], and Ameurfina D. Santos*[1]

[1]National Institute of Molecular Biology and Biotechnology;
[2]Marine Science Institute (MSI), University of the Philippines
Diliman Quezon City, Philippines 1101
Email: ameurfinas@gmail.com

Redesigning domain interfaces of antibodies to promote heterodimer formation and prevent assembly of homodimers is crucial for bispecific antibody (bsAb) production. Moreover, extensive protein-protein interactions leading to dimer formation of antibody molecule occurs at the C-terminal domains of the

heavy chains (CH3). Here, complementary charges were engineered on the CH3 domain interfaces of two IgG1 antitumor antibodies, CC49 fused with interleukin-2 (IL2) and HuCOL-1. The CC49IL2 antibody was engineered to contain a positively charged arginine residue at amino acid position 407 originally containing a tyrosine. On the other hand, HuCOL-1 antibody was engineered to have a negatively charged amino acid, either glutamic acid or aspartic acid, at position 366 which originally is a threonine. The CC49IL2 and HuCOL-1 gene constructs with the introduced charges were cloned downstream of the p10 and polH promoters, respectively, of a single pAcUW51 baculovirus expression vector to produce the gene constructs pAc CC49IL2 Y407R / HuCOL-1 T366E and pAc CC49IL2 Y407R / HuCOL-1 T366D. These were transfected and expressed in *Spodoptera frugiperda* (Sf9) insect cells. Western blot of the amplified transfection supernatants showed that more of the CC49IL2/HuCOL-1 heterodimer was formed for the construct with the introduced arginine-glutamic acid pair but not with the arginine-aspartic acid mutation. The result indicates that the arginine and glutamic acid mutation may have been in an orientation which favors the maximum interaction of the complementary charges which leads to more of the heterodimer. Moreover, introduction of arginine and glutamic acid at positions 407 and 366, respectively, can be useful in promoting association of antibody chains for the generation of a bispecific antibody.

Keywords: bispecific antibody, heterodimerization, complementary charges

CMPSD NO. 15

GENOMIC DNA IN WINGED BEAN ENCODES PUTATIVE REGULATORY PROTEINS WITH POLYASPARTIC ACID-REPEATS

I. B. Domingo [1], Maria Jamela R. Revilleza[1] and Antonio C. Laurena[2]

[1]Institute of Chemistry, College of Arts and Sciences and
[2]Institute of Plant Breeding, College of Agriculture,
University of the Philippines Los Baños, College, Laguna 4031
Email: aclaurena@yahoo.com

The molecular mechanisms underlying the cell division/cell expansion phase of early seed development in angiosperms are poorly understood but previous studies have shown that the temporal expression of a diverse group of soluble proteins known as 2S albumins coincides with the initiation of cell expansion in developing seed embryos. A 2S albumin (Gm2S-1) gene was isolated and purified in midmaturation soybean seeds and encode a mature 8 kDa which consisted of a linker peptide, a signal peptide and a unique 43-amino acid peptide which contains a polyaspartic acid carboxyl terminus, an RGD cell adhesion motif and a predicted

conserved helical region with structural homology to chromatin-binding proteins Homologues of this small polypeptide were PCR-screened in other plants such as winged beans, peanut, velvet bean, yardlong bean, cowpea and coconut. Six oligonucleotides (SP2, NP2+d, IBDF1,ANP3, LS12a and IBDRev1) were designed to amplify various regions of the DNA that encode for the signal peptide, the amino and carboxyl terminal (polyaspartic motif) of the 43-amino acid peptide. Various PCR products were generated only from winged bean (180, 270 and 440 bp) and the yardlong bean (80 and 240 bp) using the primer pairs NP2+d/ANP3. DNA sequence analyses of some of the PCR products from winged bean showed that Ptet-1 consisted of 414 bases and Ptet-2 has 239 with a 30 bp consensus sequence to Gm2S-1 in soybean. This consensus sequence represents the poly-aspartic acid rich repeat while the RGD cell adhesion motif and the helical motif were absent in winged bean. This study suggests that there is no common regulatory protein during seed development (cell division/expansion interface) but rather the presence of other regulatory proteins with the highly conserved polyaspartic acid rich repeats.

Keywords: polyaspartic acid repeats, winged bean, PCR, seed development and 2S albumin.

CMPSD NO. 16
A STUDY ON THE CHARACTERISTICS AND ION-EXCHANGE POTENTIAL OF CLINOPTILOLITE IN IMMOBILIZING HEAVY METALS

Anna Marie M. Hufemia*, Rene Juna Claveria and Zharina Ahne M. Corpus

Department of Environmental Science,
Ateneo de Manila University
Loyola Heights, Quezon City
Email: ahufemia@ateneo.edu

When industrial effluents containing heavy metals are discharged untreated into the environment, it will cause adverse impacts both on ecological life and human health. Heavy metals are known to bioaccumulate in the food chain and remain persistent in the environment. Although various physico-chemical methods for heavy metal ion removal have already been developed, there is a need to find a practical and cost-effective means of doing it commercially. This study explored a possible alternative treatment of wastewater containing high concentrations of heavy metal ions through ion-exchange. Clinoptilolite, a type of zeolite mineral, was used as the ion-exchange medium for the removal of particular heavy metal ions namely, copper, lead, and zinc. The locally-available clinoptilolite was

characterized according to its porosity, density and cation-exchange capacity. Modified clinoptilolite was also prepared by immersing it in NaCl solution for 7 days. The ion-exchange potentials of natural and modified clinoptilolite were tested by subjecting the minerals in aqueous single-ion solutions of copper, lead and zinc. One hundred milliliter solutions at an initial concentration of 20 ppm were allowed to come in contact with 10 g of 250 micrometer of clinoptilolite for 24 h while being shaken at 20 rpm. Using atomic absorption spectrophotometry (AAS), the metal ion concentrations were determined at a retention time of 6 h and 24 h. Initial results showed that natural clinoptilolite can achieve a removal efficiency of 26% and 46% of copper ions at 6 h and 24 h retention time, respectively. Similar trends are expected with the removal of zinc and lead using clinoptilolite, making this method a promising means of protecting the environment from heavy metal contamination.

Keywords: heavy metal removal, cation-exchange capacity, zeolite, clinoptilolite

CMPSD NO. 17
CONDUCTIVE AND CATALYTICALLY ACTIVE POLYPYRROLE/PLATINUM COMPOSITE GAS DIFFUSION ELECTRODE FOR A POLYMER ELECTROLYTE MEMBRANE FUEL CELL

***Leon M. Payawan, Jr. and Ian Harvey J. Arellano**

Institute of Chemistry
University of the Philippines
Diliman, Quezon City 1101
Email: lpayawan@chem.upd.edu.ph

Synthesis and characterization of a possible substitute for commercial carbon cloth as gas diffusion electrode (GDE) for a polymer electrolyte membrane fuel cell (PEMFC) is reported. Polypyrrole (PPy) is an intrinsically conducting polymer that shows stability over a wide range of operating conditions. Dense PPy was successfully deposited via *in situ* oxidation of the monomer in the presence of a textile substrate with poly(sodium-4-styrene sulfonate) as dopant. Analysis of the Fourier Transform Infrared (FTIR) spectrum confirmed the formation of the polymer with concomitant incorporation of the dopant. Nano-sized platinum (Pt) was deposited on the PPy cloth via electrochemical and formaldehyde oxidation-assisted platinum reduction (FOAPR). The conductivity values of the resulting PPy/Pt composite range from 1.0–2.0 $\Omega^{-1}cm^{-2}$. Scanning Electron Microscopy (SEM) studies show polydispersed platinum nanoparticles, with 80–500 nm size distribution, on a discontinuous overlapping PPy morphology. Cross sectional SEM images showed

that FOAPR produces smaller Pt particles but it is destructive to the PPy layer which results to decrease in conductivity. The proton exchange membrane, Nafion 112, was activated by soaking it to a series of solutions, H_2O-H_2O_2-H_2SO_4-H_2O, at 80°C for 1 hour in every solution. The membrane electrode assembly (MEA) was constructed by sintering two PPy/Pt electrodes with the activated Nafion 112 membrane in between. A single unit H_2/O_2 PEMFC was fabricated using Plexiglass with single-serpentine flowfield geometry. The PPy/Pt/Nafion MEA was incorporated as GDE of the fabricated PEMFC. The operation outputs were 0.18 V and 0.29 A, which correspond to 25% and 42% of the outputs generated using a platinum-coated carbon cloth based-PEMFC used as reference.

Keywords: polypyrrole, PEMFC, MEA, platinum nanoparticles

CMPSD NO. 18
POTENTIAL PURIFICATION PRODUCTS (CHROMATOGRAPHIC SILICA GEL AND ZEOLITE) FROM RICE HULL

Leni L. Quirit, Elma C. Llaguno, Fernando C. Pagdanganan and Karen N. Hernandez

Institute of Chemistry
University of the Philippines
Diliman, Quezon City
Email:lquirit@chem.upd.edu.ph

Rice hull is an abundant agricultural waste material which could be a renewable energy source when combusted. The combustion residue (called rice hull ash or RHA) contains a significant amount (20% of the hull) of potentially high grade silica for many industrial uses. This paper presents the preparation and characterization of two types of silica products from rice hull: a silica gel for possible chromatographic use and zeolites X and Y.

A silica gel sample was prepared (at acid pH gelling conditions) from rice hull ash and compared to three commercial chromatographic silica gels using quantitative elemental x-ray fluorescence analysis. Elemental levels in the rice hull gel were within the range of levels or close to the detection limits of corresponding elements in the chromatographic gels. Water vapor adsorption, x-ray diffraction, infrared spectroscopy and scanning electron microscopy showed that the rice hull gel was similar to the commercial chromatographic silica gel Davison 12.

Zeolites are crystalline aluminosilicates used as molecular sieves for purification and catalytic purposes. Zeolites X and Y were synthesized from rice hull silica gel and aluminum hydroxide. For comparison, controls were synthesized

from commercial silica gel. The samples and controls exhibited characteristic infrared peaks corresponding to the vibrations of the TO_4 (T= Si, Al) of the zeolite framework. The x-ray diffraction patterns of the zeolite samples are similar to the controls with respect to the 2 theta peak values.

Keywords: chromatographic silica gel, FTIR, rice hull silica gel, zeolites, infrared spectroscopy, x-ray fluorescence, x-ray diffraction

CMPSD NO. 19
FABRICATION OF TEXTURED (BI,PB)2SR2CA2CU3O10+X/AG THICK FILMS BY ELECTROPHORETIC DEPOSITION METHOD

Meredith Anne C. Sotto[1], Raymond.V.Rivera-Virtudazo[2] and R.V. Sarmago[3]

[1]Physics Division, Institute of Mathematical Sciences and Physics,
University of the Philippines Los Baños
4031 Laguna, Philippines
[2]Department of Ceramic Engineering (Materials Science and Engineering)
College of Engineering
Mariano Marcos State University
Batac Ilocos Norte 2906, Philippines
[3]Materials Science and Engineering Program
National Institute of Physics
College of Science, University of the Philippines Diliman,
Quezon City 1101, Philippines
Email: rv200476@yahoo.com

This study explores the preparation of textured $Bi_2Sr_2Ca_2Cu_3O_{10+x}$ (Bi-2223) thick films by electrophoretic deposition (EPD). The EPD Method involves the dispersion and stabilization of precursor powder in a suitable liquid medium so that the former achieves a surface electric charge.

Pure and Pb-doped (Pb=0.3) $Bi_2Sr_2Ca_2Cu_3O_{10+x}$ (Bi-2223) thick films (~5–30mm) were electrophoretically deposited on high purity silver foil using ethanol as the suspending liquid. The deposition was done at applied voltage of 400 V/cm and current of ~0.02 mA/cm^2 for a period of 10 min. Short sintering time of 1 h at 820°C was performed to reduce film porosity and allow vacancy diffusion of the deposited grains.

The present study showed that pure and Pb-doped (Pb=0.3) $Bi_2Sr_2Ca_2Cu_3O_{10+x}$ (Bi-2223) thick films (~5–30 ìm) were successfully fabricated unto high purity silver substrates using electrophoretic deposition method. X-ray diffraction and surface

image analysis shows that the deposited films had a high grain alignment with a preferred orientation along the *c*-axis. These results indicate texturing of the deposited films.

Keywords: superconductors, electrophoretic deposition , Bi-2223, doped, thick films

MATHEMATICS

CMPSD NO. 20

CONSTRUCTION OF A QUASI-REGULAR SELF-COMPLEMENTARY GRAPH

Esperanza Blancaflor Arugay

Department of Mathematics
MSU-Iligan Institute of Technology Tibanga, Iligan City 9200
Email: csm-eba@sulat.msuiit.edu.ph

A self-complementary graph is a graph whose complement is isomorphic to itself. A quasi-regular self-complementary graph is of order 4k, k a positive integer, where 2k vertices have degree 2k and the remaining vertices have degree 2k-1. This paper presents a procedure for constructing a quasi-regular self-complementary graph. The following results are proven:

1. Let ó = (1,2,3,….,4k) be a complementing permutation of a self-complementary graph G with vertices V(G) = {1,2,3,…,4k}. Let K_{4k} be the corresponding complete graph where K_{4k} = GUG'. If [1,2], [1,3],…,[1,2k+1] are colored black or red and each time apply ó thereafter, the resulting graph is K_{4k} whose edges have colors red and black where G and G' graphs are each induced by a monochromatic color class.
2. Let G be a totally disconnected graph of order 4k and let the vertices of G be labeled 1,2,3,…,4k and arranged in a circular manner. If [1, j], j even, j<2k+1 is colored black or red and applying ó = (1,2, 3,….,4k) thereafter, all vertices of G gain by one degree. If [1, j], j odd, j<2k+1 is colored black and applying ó = (1,2,3,….,4k) thereafter, odd-labeled vertices of G gain by two degrees while even-labeled vertices gain 0.
3. Let G be a totally disconnected graph of order 4k and let the vertices of G be labeled 1,2,3,…,4k and arranged in a circular manner. If [1, 2k+1] is colored black and applying ó thereafter, odd-labeled vertices of G gain one degree while even-labeled vertices gain 0.

Keywords: self-complementary graph, quasi-regular self-complementary graph, complementing permutation.

CMPSD NO. 21

FOLDING and SOME GRAPH INVARIANTS

Romulo C. Guerrero

Department of Mathematics
MSU-Iligan Institute of Technology, Tibanga, Iligan City 9200
Email: romy1325@yahoo.com

If two non-adjacent vertices of a connected non-complete graph G that have a common neighbor are identified and the resulting multiple edges are reduced to simple edges, then we obtain another connected graph of order one less than that of the original graph. If a similar process is applied to the new graph, and we repeat the operation until there are no more nonadjacent vertices, we finally get a complete graph. We say that the graph is folded into a complete graph. This process of folding a graph into a complete graph induces in a very natural way a partitioning of the vertex-set of G into pairwise independent sets. We denote by $F(G)$ the set of all complete graphs, up to isomorphism, into which a graph G can be folded.

Let G be a graph. The largest of the vertex degrees of G is called the maximum degree of G and is denoted by D(G) or simply D.

A *coloring* of G is an assignment of colors to its vertices (exactly one color for each vertex) so that no two adjacent vertices in G have the same color. A coloring of G which assigns c colors to its vertices is called a c-coloring.

The *chromatic number* of G is the minimum number, denoted by c(G), for which G has a c- coloring. Equivalently, c (G) is the minimum number n for which G is an n-partite graph.

A set S of vertices in G is *independent* if no two vertices of S are adjacent in G. The maximum cardinality of an independent set of vertices in G is called the *independence number* of G and is denoted by a(G).

A *clique* in G is a maximal complete subgraph of G. The maximum order of a clique is called the *clique number* of G, and is denoted by v(G).

A set S in G is a *dominating set* if every vertex not in S is adjacent to a vertex in S. The *domination number* g(G) is the minimum cardinality of a dominating set in G.

G is called a *unit graph* in the Euclidean space $\boldsymbol{R}^d$ if there is a one-to-one mapping f: $V(G)$®$\boldsymbol{R}^d$ such that $|f(x) - f(y)| = 1$ whenever $[x, y] \hat{I} E(G)$. The mapping f is called a *unit representation* of G in $\boldsymbol{R}^d$.

The *dimension* of G, denoted by $dim(G)$, is the smallest integer d such that G is a unit graph in $\mathbf{R}^d$.

Let G be a unit graph in $\mathbf{R}^d$. The *span* of G in $\mathbf{R}^d$, denoted by $span_d(G)$, is the real number s such that for every $e > 0$, the following conditions are satisfied:

1. there exists a unit representation of G in $\mathbf{R}^d$ which is contained in some open ball of diameter $s + e$;
2. no unit representation of G in $\mathbf{R}^d$ is contained in any open ball of diameter s.

This study seeks to enumerate some relationship of folding graphs to several graph invariants such as chromatic number, clique number, span, independence number, domination number, maximum degree, and dimension.

Among the main results generated are the following:

1. If $K_p \hat{I} F(G)$, then $p £ a(G)c(G)$

2. $min\{r \mid K_r \hat{I} F(G)\} ³ c(G)$.

3. $min\{r \mid K_r \hat{I} F(G)\} ³ \left\lceil \frac{2}{2-[span_{2\chi(G)}(G)]^2} \right\rceil$, where $2 ¹ [span_2 c(G)]2$.

4. $min\{r \mid K_r \hat{I} F(G)\} ³ v(G)$.

5. $min\{r \mid K_r \hat{I} F(G)\} > \left\lceil \frac{\gamma(G)}{\alpha(G)} \right\rceil$.

6. $min\{r \mid K_r \hat{I} F(G)\} ³ \left\lceil \frac{n}{\alpha(G)} \right\rceil$.

7. $max\{r \mid K_r \hat{I} F(G)\} £ \left\lfloor \frac{1+\sqrt{1+4n\Delta}}{2} \right\rfloor$.

8. $min\{r \mid K_r \hat{I} F(G)\} ³ \left\lceil \frac{\dim(G)}{2} \right\rceil$.

Keywords: fold of a graph, chromatic number, clique number, span, independence number, domination number, maximum degree, dimension

CMPSD NO. 22

FOLDING REGULAR GRAPHS

Severino V. Gervacio* and Romulo C. Guerrero**

*Department of Mathematics, De La Salle University
2401 Taft Avenue, 1004 Manila
Email: gervacios@dlsu.edu.ph; gervacio@pacific.net.ph
**Department of Mathematics
MSU-Iligan Institute of Technology
Tibanga, Iligan City 9200
Email: romy1325@yahoo.com

Graph folding is a unary operation introduced by Dr. Severino V. Gervacio in 1992 which he later modified in 1999. It is defined as follows. If two non-adjacent vertices of a connected non-complete graph G that have a common neighbor are identified and the resulting multiple edges are reduced to simple edges, then we obtain another connected graph of order one less than that of the original graph. If a similar process is applied to the new graph, and we repeat the operation until there are no more non-adjacent vertices, we finally get a complete graph. We say that the graph is folded into a complete graph. This operation induces in a very natural way a partitioning of the vertex set of a graph into pairwise linked and independent sets. We denote by $F(G)$ the set of all complete graphs, up to isomorphism, into which a graph G can be folded. Here, we shall give results involving the folding of regular graphs. Specifically, we will show that the famous Petersen graph folds only into the complete graphs K_3, K_4, and K_5. In addition among others, we will show that $max\{t \mid K_t \hat{I} F(G)\} \pounds \left\lfloor \frac{1+\sqrt{1+4rn}}{2} \right\rfloor$, where G is a connected r-regular graph of order n.

A graph G is called *regular* if all the vertices of G are of equal degree. If " $x \hat{I} G$, $deg(x) = r$, we say that G is an r-regular graph.

Among the major results obtained in this study are the following:

1. The Petersen graph G^* shown in Fig.1 folds only into K_3, K_4, and K_5, i.e., $F(G^*) = \{K_3, K_4, K_5\}$.

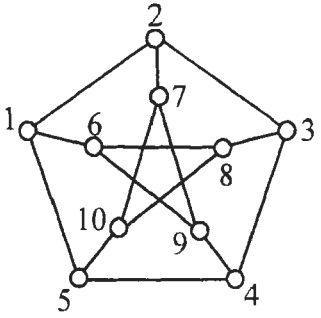


Figure 1: The Petersen graph G^*.

2. Let G be a k-regular graph of diameter 2, where k ³ 3. If G folds into K_r, r ³ $k+2$, then G necessarily folds into K_{r-1}. In particular, any 3- regular graph of diameter 2 that folds into K_r, where r ³ 5, necessarily folds into K_{r-1}.
3. Let G be a connected r-regular graph of order n. Then

$$max\{t \mid K_t \hat{I} F(G)\} \pounds \left\lfloor \frac{1+\sqrt{1+4rn}}{2} \right\rfloor.$$

4. Let G be a connected r-regular graph of order n and such that its complement $\widetilde{G}$ is also connected. Then

$$max\{t \mid K_t \hat{I} F(\widetilde{G})\} \pounds \left\lfloor \frac{1+\sqrt{1+4n(n-r-1)}}{2} \right\rfloor.$$

Keywords: fold of a graph, regular graph, r-regular graph, Petersen graph, complement of a graph

CMPSD NO. 23
ON THE EXISTENCE OF HOMOGENEOUS FACTORIZATION OF THE HAMMING GRAPH $H_1(n,q)$ USING THE IMPRIMITIVE ACTION OF Nd"Sq ON THE ARC SET OF THE COMPLETE GRAPH Kq.

Aurea Z. Rosal

SciTech R&D Center & Mathematics and Statistics Department
Polytechnic University of the Philippines
Sta. Mesa, Manila
Email: azrosal@yahoo.com

If Ã=(V(Ã), A(Ã)) is a digraph with vertex set V(Ã) and arc set A(Ã), then a homogeneous factorization of Ã of index n is the 4-tuple (M,G, V(Ã), $\tilde{N}$) such that

1. P= {P_1, ..., P_n} is a partition of A(Ã).
2. G d" Aut(Ã) acting transitively on P.
3. M is the kernel of the action of G and is transitive on V(Ã).

In a previous paper [Poster Paper, 27th ASM - NAST], it has been shown that there exists

1. a subgroup N of Sq that acts imprimitively on A(Kq). In this paper, it will be shown that for each d, there exists a unique homogeneous factorization (M,G, V(Ã), $\tilde{N}$) of type [$Im(N)$, T, n, d] of the Hamming graph $H_1(n,q)$ = $(K_q)^n$ = $K_q \times \ldots \times K_q$ (n factors of K_q) of index n using the imprimitive action of N on $A(K_q)$. This factorization satisfies the following:
2. $M = N^n = N \times N \times \ldots \times N$ (n factors of N)
3. $G = M \times T$, where Td" S_q is abelian acting regularly on {1,2, ..., n}
4. $d = q(q-1)$ " [N : H], where $N_{(u,v)} < H < N$, (u,v) is any element in $A(K_q)$.

Keywords: Hamming graph, imprimitive action, homogeneous factorization, action, complete graph.

CMPSD NO. 24

EMBEDDING THE JOHNSON NETWORKS INTO THE HAMMING NETWORK

Geoffrey A. Solano[1] and Jaime D.L. Caro[2]

[1] Department of Physical Sciences and Mathematics,
University of the Philippines, Manila
[2] Department of Computer Science, University of the Philippines, Diliman
Emails: solange1611@yahoo.com
jdlcaro@gmail.com

Embedding of graphs is an important and interesting approach to parallel computing. Generally it can be used to model simulation of networks and algorithm structures on different networks. This paper shows that there is an embedding of the Johnson Networks into the Hamming Network.

The vertex set of the Johnson Scheme $G(n,k)$ is the set of all k-subsets of a fixed n-set. Two vertices A and B in $G(n,k)$ are adjacent if $|A) "B| = k-1$. The *ith* graph of the Johnson Scheme $G_i(n,k)$ is an extension of $G(n,k)$ such that vertices are adjacent if they are they are i–related, that is, if $|A) "B| = k-i$. i is referred to as the Johnson Distance. The combined *ith* graphs of the Johnson Scheme $SG_i(n)$ is the graph formed by the union of all the graphs $G_i(n,k)$ where $0<k<n$. A Johnson Network is a network modeled after Johnson graph $G_i(n,k)$.

The Hamming Scheme $H(n,q,r)$ is an association scheme whose vertex set Q^n is the set of all words of length n over the alphabet Q of q symbols. Two vertices are adjacent if and only if they are r-related, that is, if they differ in exactly r coordinate positions where r is referred to as the Hamming Distance. A Hamming Network is a network modeled after the Hamming graph.

This paper shows that there is an embedding of Johnson Networks, or the combined ith graphs of the Johnson Scheme $SG_i(n)$, into the Hamming Network, when $q=2$ and $r=2i$. This is done by showing that $SG_i(n)$ is a spanning subgraph of the Hamming Graph $H(n,2,2i)$. It is also shown that every Johnson Network can also be embedded into the said Hamming Network.

Keywords: embeddings, hamming scheme, johnson scheme, association schemes

CMPSD NO. 25
PRIME CIRCUMFERENCE OF GRAPHS RESULTING FROM SOME UNARY OPERATIONS

Joselito A. Uy

Department of Mathematics, College of Science and Mathematics
MSU-Iligan Institute of Technology, Iligan City
Email: csm-jau@sulat.msuiit.edu.ph

The *prime circumference* ë(*G*) of a simple graph *G* is the length of a longest induced cycle of *G* if *G* has a cycle; otherwise, ë(*G*) = 0. Graph *G* is a cycle if and only if it has a cycle and ë(*G* " *v*) = 0 for each $v \in V_G$. It is a cycle or consists of a cycle and isolated vertices if and only if it has a cycle and ë(*G* " *e*) = 0 for each $e \in E_G$. If it is an *n*-cycle, $n \geq 6$, then $\lambda(G^2) \in \{\frac{2}{3}n, \frac{2}{3}(n-1), \frac{2}{3}(n-2)\}$. If it is a tree, then $\lambda(G^2) = 0$ or 3. If it is the complement of a line graph, then $\lambda(G) \leq 6$. Finally, if G_1, G_2, Λ, G_m are its components and *H* is the line graph of $K_1 + G$, then $\lambda(H) \in \{0, 3, \max\{\lambda(H_i)\}_{i=1}^{m}\}$, where H_i is the line graph of $K_1 + G_i$.

Keywords: induced cycle, prime circumference, complement, line graph, edge and vertex deletion, square

CMPSD NO. 26
AUTOMATED PRESCHOOL LANGUAGE AGE EVALUATION

Ma. Sheila A. Magboo*, Ronald G. Magcalas

Dept. of Physical Sciences and Mathematics, College of Arts and Sciences
University of the Philippines, Manila, Padre Faura, Ermita, Manila
Email: sheilaabad@yahoo.com

Language age is the age of a person with regards to speech and communication skills. In the Philippines, we have a number of communicatively handicapped patients who are mostly very young children. Preschool Language

Scale-3 (PLS-3) test is the standard test that is used to assess receptive and expressive language skills of children ages 2 weeks through 6 years, 11 months. The two subscales, Auditory Comprehension and Expressive Communication, enable a speech therapist to evaluate a child's relative ability in receptive and expressive language. When comparing the scores, deficiencies are determined whether primarily receptive or expressive in nature, or if they reflect an overall delay or disorder in communication. PLS-3 also assesses behaviors considered to be language precursors. A report about the child's language ability is then prepared by the therapist including frequency and intensity of services and the interdisciplinary team who will work with the parents. However, with only a few speech therapists available in the country and so many tests to process, the release of speech language evaluation test takes several weeks, especially at the Philippine General Hospital, hence, there is a need to automate the generation of the evaluation report. To address this problem, the "Automated Preschool Language Age Evaluation" was created. This prototype is a web-based database application capable of a) determining the language age of children ages 2 weeks through 6 years and 11 months using the PLS-3 test, b) monitor the results of the PLS-3 tests of each patient, c) update PLS-3 test questions, d) display and update patient records, and e) perform user account management. A line graph of the patient's language age (determined from PLS-3) vs. time (in months) to visualize the patient's progress with regards to language age is likewise generated. The questions can be updated in order to tie in with the available testing materials (ex. change "teddy bear" to "rag doll" if a teddy bear is currently unavailable). The system should be administered only on children within the age range specified in PLS-3. Also, the system should not be used to determine whether or not a child is gifted.

Keywords: language age, Preschool Language Scale-3

CMPSD NO. 27

DOCUMENT EXCHANGE SYSTEM FOR CRITICALLY APPRAISED TOPICS

Ma. Sheila A. Magboo* and Leo Tenmatay

Dept. of Physical Sciences and Mathematics, College of Arts and Sciences
University of the Philippines Manila, Padre Faura, Ermita, Manila
Email: sheilaabad@yahoo.com

A new medical paradigm called Evidence-based Medicine (EBM) that uses current best evidence is now being used in making decisions about the care of

individual patients. EBM integrates individual clinical experience with the best available external clinical evidence. To make it easier for clinicians to learn EBM and put it into practice, Critically Appraised Topics (CATs) which summarize the procedures done in an evidence-based approach to a literature were developed. A CAT defines the clinical question, presents the methods and results of the study, describes the details of the appraisals, and allows a clinician to give a conclusion based on the strength of the evidence. Although there are a lot of free CATs posted on the web for any clinician to use, finding one that is truly relevant to his needs is not easy. Those without websites use email as a means of sending and receiving CATs. In the Philippines, there is currently no working system yet for managing CATs. As a response, a Java application was made that employed exchanging and searching CATs without a need for email nor websites. In particular this prototype system is capable of a) creating, editing, storing and printing CATs, b) searching for CATs stored in other computers' database, c) sending and retrieving complete CAT documents from one computer to another, and d) initiating a chat session with appraisers. The application integrates the web browser, instant messenger and a critically appraised topics document maker in one package. All of these are done via a hybrid peer-to-peer (P2P) network where there is central server that keeps information on peers and responds to requests for that information. Peers on the other hand are responsible for hosting the information (as the central server does not store files), and for letting the central server know what CATs they want to share. The CATs are formatted using Extensible Markup Language (XML) to facilitate the sharing of data across different systems. The ability to send a document back to its owner is a plus since it makes CATs more useful and notifies the owner that his article has already been reviewed.

Keywords: evidence-based medicine, critically appraised topics, hybrid peer-to-peer (P2P) networks, Extensible Markup Language (XML)

CMPSD NO. 28

SIZE AND SHAPE VARIATION IN POPULATIONS OF *ACHATIVA FULICA* AND *AMPHIDROMOUS FURCILLA*: A MULTIVARIATE MORPHOMETRIC APPROACH

Ken P. Dadula, Mark Anthony J. Torres and Cesar G. Demayo

Department of Biological Sciences, College of Science and Mathematics
MSU-Iligan Institute of Technology, Iligan City
Email: c_demayo@yahoo.com

Size and shape variation in selected populations of the land snails *Achatina fulica* and *Amphidromous furcilla* was determined using landmark- and distance-based morphometric analysis. Three hundred thirteen *A. fulica* and ten *A. furcilla*

were sampled from Mt. Agad-agad, Iligan City and from Balangao, Zamboanga-Sibugay. All samples were scanned at 600 dpi with the snails carefully oriented with the aperture resting on the bed of the scanner. The two land snail species have obvious spiral ornamentation which allows determination of biologically meaningful landmarks, of which twenty-two points were digitized using the ScionImage software. This study used distance variables based on the twenty two landmarks with an objective of comparing the sizes of the different samples. Distances between points were calculated using the Euclidean Distance Matrix Algorithm. The measurements include the shell's total length, which measures the maximum distance parallel to the axis of coiling from the shell apex to the basal apertural edge; total width of the snail shown as the maximum distance perpendicular to the axis of coiling; height of the spire which is also the distance from the apex to the aperture–suture junction; distances between the included major spiral chords; and the angulations on the corner of the aperture. These measurements give a balanced overview of the shape of the snail's shell, with detailed representation of the whorls characteristic of each species. Principal Component Analysis (PCA) was done to determine components with defined biological meanings that account for the maximum variation of the snail samples. This multivariate method allows numerous variables to be collected, and the significant fine-scaled variation to be extracted. Loadings for each variable in each principal component represent that variable's contribution to the variance explained by that principle component. Cluster analysis was also employed to group snails with similar sizes together. Then, test for significant differences among the snails was done using the Discriminant Function Analysis (DFA). Shape differences among the different snail samples was determined by subjecting the raw x and y coordinates of the twenty two digitized landmarks to Procrustes fitting. This procedure eliminates both rotational and size translation allowing for the analysis of snail shape variation only. The Procrusted-fitted coordinate values were also subjected to PCA, Cluster analysis and DFA.

Results showed no significant size differences among the snail samples from the two locations based on the computed interlandmark distances. A variation in shell shapes between the two species is evident from the reconstructed images of the snails. PCA of the Procrustes-fitted values returned a total of thirty-six significant components with the first component explaining ninety-eight percent (98%) of the observed variation. This variation is summarized in a scatter plot with the Zamboanga population shown to have broader apices, more expansive aperture and wider spire base. Cluster analysis of the Procrustes-fitted values separates the three populations into three well-defined groups. DFA of the data reveals that the variations observed are statistically significant. These results are further discussed in the light of ontogenetic allometry and parasite-induced changes in the morphology of the snail.

Keywords: multivariate analysis, cluster analysis, *Achatina fulica*, *Amphidromous furcilla*, snails

CMPSD NO. 29

ON THREE-PARAMETER SIGMOID-SHAPED NEGATIVE EXPONENTIAL VOLUMETRIC EQUATIONS OF STATE USING MARQUARDT METHOD

Derick Erl P. Sumalapao

Department of Physical Sciences and Mathematics,
College of Arts and Sciences
University of the Philippines Manila
Telefax: 526-5858; Email: derickearl@yahoo.com

Ideal gas law predicts well the properties of gases only under the wide range of ideal conditions that are commonly experienced and it works with some degree of accuracy necessary for safety engineering, the pressure and temperature in car tires and commercial gas cylinders. This law is reasonably accurate for gases at low pressures and high temperatures. However, it becomes increasingly inaccurate and fails miserably at higher pressures and lower temperatures. Despite its egregious and glaring shortcomings, the ideal gas law is still used extensively in multifarious fields of science and engineering because of its traditional entrenchment. Due to its uncomplicated form, forthright solutions to a number of problems involving the equation of state can be realized if the system of interest is postulated to exhibit the features of an ideal gas. But the overly simplified model cannot satisfactorily capture the more realistic, intricate, and complicated phenomenon. It is therefore necessary that the volumetric properties of gases and fluids be predicted as precise as possible so as to obtain an accurate computation of the different thermal and mechanical properties of the system such as work, thermal efficiency, energy, and heat rate.

This study is one such effort as it had investigated and compared the different three-parameter sigmoid-shaped negative exponential models, namely the Gompertz and the Logistic models, aimed at determining which model is more viable in capturing the intrinsically complex volumetric behavior of saturated fluids and gases. The thermodynamic property tables of saturated liquid water, ammonia, acetone, carbon dioxide and bromine were used for testing purposes.

Nonlinear models, in general, are more difficult to specify, identify and estimate than linear models, and they involve nonlinear iterative processes in determining the values of these parameters. In estimating the parameters of the Gompertz and the Logistic models, the classic NLIN (nonlinear) procedure of SAS (Statistical Analysis System) and the Marquardt Method were employed. Selection of the best model was made based on the prescribed set of criteria, such as p-values, coefficient of determination (R^2) and sum of squares residuals (SSR).

The class of three-parameter sigmoid-shaped negative exponential models unquestionably has a better model fit to the randomly chosen volumetric properties

tables of saturated fluids and gases. The properties of Gompertz and the Logistic models were imperative to note such as having points of inflection and being sigmoid in shape, which suggest that these models are indeed suitable for modeling the series of pressure that demonstrate a sigmoid pattern over the entire pressure values, implying further that the ratio of temperature to volume reflects a relatively fast initial and terminal increase rates. Analysis of the differential forms and second order derivatives of the Gompertz and the Logistic models pointed out that it is negative over the entire range of the pressure values where the deviations from ideality is positive and the second order derivatives of the models are positive over the range of pressure values where the deviations from ideality are negative. In the class of three-parameter sigmoid-shaped negative exponential models, the study found out that the Gompertz model is the more viable model in explaining the thermodynamic property values of saturated fluids and gases.

Keywords: Negative Exponential Models, Sigmoid-Shaped Models, Gompertz Model, Logistic Model, Saturated Liquids and Gases, NLIN Procedure, Marquardt Method

CMPSD NO. 30

VERTEX INDEPENDENCE IN GRAPHS UNDER SOME BINARY OPERATIONS

Rolito G. Eballe*, Emmeline Victoria M. Llido, and Melona T. Nocete

Department of Mathematics, Central Mindanao University
University Town, Musuan, Bukidnon, 8710 Philippines
Email: rgeballe@cmu.edu.ph; eballe2004@yahoo.com

A set of vertices in a graph G is said to be independent if no two of them are adjacent. The largest number of vertices in such a set is called the independence number of G and is denoted by $b(G)$. In this paper, the authors attempted to characterize the independent sets of vertices in the sum $G+H$, composition $G[H]$, and Cartesian product $G'H$ of graphs G and H (in the context of finite, undirected graphs with no loops nor multiple edges).

Experimentation was first done by drawing samples of graphs under the binary operations sum, composition, and Cartesian product, in order to gain insights as to the characteristics of these graphs in relation to the notion of independence.

Two of the major results of this study are listed below.

1. The independent sets of vertices in $G+H$ are the independent sets of vertices in G and the independent sets of vertices in H. Consequently,

the independence number of $G+H$ is given by $b(G+H) = maximum\ (b(G), b(H))$.

2. The independent sets of vertices in $G[H]$ are those of the form $S = \Upsilon_{i=1}^{k}(\{u_i\} \times H_i)$, where the set $\{u_1, u_2, \ldots, u_k\}$ is independent in G and each H_i is independent in H. Consequently, the independence number of $G[H]$ is given by $b(G[H]) = b(G)\ ' b(H)$.

The results show that as long as the independent sets of vertices in both G and H are known, the independent sets of vertices in $G+H$ and $G[H]$ can surely be identified. Though this study also produced partial results in the Cartesian product of graphs, completely describing the independent sets of vertices here seemed to be very difficult. In complexity theory, we say that in the Cartesian product the problem is probably NP-hard. (The problem, in general, has already been reported to be NP-hard.)

Keywords: independence, independence number, sum, composition, Cartesian product

CMPSD NO. 31
THE ESSENTIAL CUTSET NUMBER AND VERTEX CONNECTIVITY OF THE SUM AND COMPOSITION OF GRAPHS

Rolito G. Eballe'[1] and Sergio R. Canoy, Jr.[2]

[1]Department of Mathematics, Central Mindanao University
University Town, Musuan, Bukidnon, 8710 Philippines
Email: rgeballe@cmu.edu.ph / eballe2004@yahoo.com
[2]Department of Mathematics, College of Science and Mathematics
MSU-IIT, Iligan City, 9200 Philippines
Email: csm-src@sulat.msuiit.edu.ph

In a connected graph G a subset $Y \hat{I} V(G)$ is a cutset if the removal of Y from G yields a disconnected subgraph. A cutset Y of G is essential if for each $y \hat{I} Y$ the subgraph induced by $(V(G) \backslash Y) \dot{E} \{y\}$ is connected. The essential cutset number $ecs(G)$ of G is the maximum cardinality of an essential cutset of G; thus, if G is a non-complete connected graph of order $n^3 3$, then $1 £ k(G) £ ecs(G) £ n-2$, where as usual $k(G)$ is the vertex connectivity of G.

In this paper, the essential cutsets of the sum $G+H$ and composition $G[H]$ of two connected graphs G and H are characterized, and the formulas for their essential cutset numbers, as well as their vertex connectivity, determined. These are done by first drawing samples of the sum of graphs and composition of graphs, and

then investigating what the notion of an essential cutset amounts to in these graphs.

Four of the main results in this study are listed below. For simplicity of enumeration, we let G and H be non-complete connected graphs and K_n the complete graph of order n.

1. The essential cutsets of the sum $G+H$ are of the form $V(G)\dot{E}S_1$ and $V(H)\dot{E}S_2$, where S_1 and S_2 are essential cutsets of H and G, respectively. Consequently,
$$ecs(G+H) = maximum\{|V(G)|+ecs(H), |V(H)|+ecs(G)\},$$
$$k(G+H) = minimum\{|V(G)|+k(H), |V(H)|+k(G)\}.$$
2. The essential cutsets of $G+K_n$ are of the form $V(K_n)\dot{E}S_2$, where S_2 is an essential cutset of G. Consequently, $ecs(G+K_n) = ecs(G)+n$, and $k(G+K_n) = k(G)+n$.
3. The essential cutsets of the composition $G[K_n]$ are of the form $S'V(K_n)$, where S is an essential cutset of G. Consequently, $ecs(G[K_n]) = ecs(G)'n$, and $k(G[K_n]) = k(G)'n$.
4. The essential cutsets of $G[K_n]$ are of the form $S'V(K_n)$, where S is an essential cutset of G. Consequently, $ecs(G[K_n]) = ecs(G)'n$, and $k(G[K_n]) = k(G)'n$.

Keywords: essential cutset, essential cutset number, vertex connectivity, sum, composition

CMPSD NO. 32

SHARPNESS OF CRITICAL CONSTANT OF HARDY-SOBOLEV INEQUALITY

Alnar L. Detalla

Department of Mathematics, Central Mindanao University
Musuan, 8710 Bukidnon
Email: al_detalla@cmu.edu.ph, al_detalla@yahoo.com

Let Ω is a bounded domain in "!^{n}, $n \geq 2$, with $0 \in \Omega$. For any $1 < p < n$, the well-known Hardy-Sobolev inequality

$$\int_{\Omega} |\nabla u(x)|^p \, dx \geq \left(\frac{n-p}{p}\right)^p \int_{\Omega} \frac{|u(x)|^p}{|x|^p} dx \qquad (1)$$

holds for any $u \in W_0^{1,p}(\Omega)$, where $W_0^{1,p}(\Omega)$ is the completion of $C_0^{\infty}(\Omega)$ in the norm

$$\|u(x)\|_{1,p,\Omega} := \left(\int_{\Omega} |u(x)|^p \, dx + \int_{\Omega} |\nabla u(x)|^p \, dx \right)^{\frac{1}{p}}.$$

In the recent work of Adimurthi, Chaudhuri and Ramaswamy, they proved that for the case $n = p$ (critical case) an inequality can be derived from Hardy-Sobolev inequality (1) such that

$$\int_{\Omega} |\nabla u(x)|^n \, dx \geq \left(\frac{n-1}{n} \right)^n \int_{\Omega} \frac{|u(x)|^n}{|x|^n} \left(Log \frac{R}{|x|} \right)^{-n} dx. \quad (2)$$

It remains to show the sharpness of the critical constant $\left(\frac{n-1}{n} \right)^n$ in inequality (2). In this paper, we shall prove the sharpness of the critical constant .

Assume the domain Ω to be unit ball B_1 and the function u radial in $W_0^{1,n}(B_1)$. Then by the symmetrization argument, we can extend B_1 to the general domain Ω. Using the test function

$$u_{\varepsilon} = \left(Log \frac{R}{|x| + \varepsilon} \right)^{\frac{n-1}{n}} - \left(Log \frac{R}{1+\varepsilon} \right)^{\frac{n-1}{n}}$$

and by Taylor expansion, we can show the sharpness of the critical constant

$$\left(\frac{n-1}{n} \right)^n.$$

Keywords: Hardy-Sobolev inequality, symmetrization argument, critical constant, sharp constant

CMPSD NO. 33

SHARP REMAINDER TERMS OF HARDY INEQUALITIES

Alnar L. Detalla[1*], Toshio Horiuchi[2], and Hiroshi Ando[2]

[1] Department of Mathematics, Central Mindanao University
Musuan, 8710 Bukidnon
[2] Department of Mathematical Sciences
Ibaraki University, Mito City, Ibaraki, Japan
Email: al_detalla@cmu.edu.ph, al_detalla@yahoo.com

In this paper we shall prove the existence of finitely many sharp remainder terms of Hardy inequality of the following type:

$$\int_{\Omega}\left|\nabla u(x)\right|^2 dx \geq \left(\frac{n-2}{2}\right)^2 \int_{\Omega}\frac{\left|u(x)\right|^2}{\left|x\right|^2}dx \qquad (1)$$

for any $u \in W_0^{1,2}(\Omega)$, Ω is a bounded domain in $\mathbb{R}^n$, $n \geq 2$, with $0 \in \Omega$. By $W_0^{1,2}(\Omega)$ we denote the completion of $C_0^{\infty}(\Omega)$ in the norm

$$\left\|u(x)\right\|_{1,2,\Omega} := \left(\int_{\Omega}\left|u(x)\right|^2 dx + \int_{\Omega}\left|\nabla u(x)\right|^2 dx\right)^{\frac{1}{2}}.$$

The best constant $\left(\frac{n-2}{2}\right)^2$ in inequality (1) is given by the $Inf_{W_0^{1,2}(\Omega)} I(u)$ where

$$I(u) = \frac{\int_{\Omega}\left|\nabla u(x)\right|^2 dx}{\int_{\Omega}\frac{\left|u(x)\right|^2}{\left|x\right|^2}dx}.$$

Moreover there exists no extremal function in $W_0^{1,2}(\Omega)$ which attains the infimum of this ratio. Roughly speaking the candidates of the extremals are singular at the origin which are not in the class. Hence it is natural to consider that there

exists "missing terms" in the right hand side of (1). In view of this, we shall investigate the Hardy inequality (1) and find out the remaining terms involving singular weight $\left(Log\frac{R}{|x|}\right)^{-2}$. The number of remaining terms depends on the choice of R.

Before stating the main result, let us introduce the following notations: For $t > 0$ and $k \geq 2$, $A_1(t) := Log\frac{R}{t}$, $A_k(t) := LogA_{k-1}(t)$, $e_1 := e$, $e_k := e^{e_{k-1}}$.

The study has generated the following results:

1. Let $n \geq 2, K \geq 1$ and $R \geq e_k Sup_\Omega |x|$. For any $u \in W_0^{1,2}(\Omega)$, there exist sharp remainder terms such that

$$\int_\Omega |\nabla u(x)|^2 dx \geq \left(\frac{n-2}{2}\right)^2 \int_\Omega \frac{|u(x)|^2}{|x|^2} dx \qquad (2)$$

2. In inequality (2), $\frac{1}{4}$ is best constant for all k-missing terms.

3. In inequality (2), the exponent 2 of the weight function is optimal.

Keywords: Hardy Inequality, extremal functions, sharp remainder term, missing term

CMPSD NO. 34

ON QUASI-C-REGULAR GROUPS

Jocelyn P. Vilela and Agnes T. Paras

Math Department, College of Science and Mathematics
MSU-Iligan Institute of Technology, Iligan City
Math Department, University of the Philippines Diliman, Quezon City
Email: csm_jpv@sulat.msuiit.edu.ph

An abelian group **G** is a module over the center **C** of its endomorphism ring **E** since **C** is a subring of **E**. This paper characterizes torsion-free abelian groups

satisfying the condition that every cyclic **C**-submodule is a quasi-summand called the **quasi-C-regular groups (qCr)**. It is shown that strongly irreducible groups are qCr and that these groups describe the structure of qCr groups. More specifically, a qCr group **G** is quasi-equal to a finite direct sum of strongly irreducible groups $\mathbf{H}_i$ such that there are no nonzero homomorphisms from $\mathbf{H}_i$ to $\mathbf{H}_j$ for i "" j .

Keywords: regular groups, quasi-regular groups, endomorphism ring, torsion-free groups, quasi-equality, quasi-summand, strongly irreducible groups, homomorphism.

PHYSICS

CMPSD NO. 35
PATH INTEGRATION FOR QUANTUM RELATIVISTIC PARTICLES IN A UNIFORM MAGNETIC FIELD: A WHITE NOISE FUNCTIONAL APPROACH

Jinky B. Bornales*[1], Christopher C. Bernido[2], Maria Victoria C. Bernido[2]

[1]Department of Physics, MSU-Iligan Institute of Technology, Iligan City
[2]Research Center for Theoretical Physics, Central Visayan Institute Foundation, Jagna, Bohol
Email: jbornales@yahoo.com

The Green function for a Dirac equation and Klein-Gordon equation in the presence of a constant magnetic field are obtained using the white noise functional approach. The Green functions for both systems are first expressed as a path integral. The path integral is then explicitly evaluated in the framework of white noise analysis. Unlike many calculations in the literature, the white noise functional approach allows us to evaluate the path integral in real time and the time - slicing of Feynman is not anymore necessary.

In the framework of white noise analysis, the evaluation of the path integral is made by first parametrizing the paths of the relativistic particle in terms of the Brownian motion $B(\lambda) = \int \omega(\lambda) d\lambda$, where $\omega(\lambda)$ is a random white noise variable. With the parametrization, the integration over all paths is converted into a well defined Guassian white noise measure $d\mu(\omega)$ and the interaction potential in the Lagrangian yields an expression similar to Levy's stochastic area encountered in stochastic calculus. Evaluation of the path integral is then carried out by using the T-transform in white noise analysis. The following results are obtained:

$$G(t''x''y''z'';t'x'y'z') = -\frac{\gamma}{(2\pi)^3}\exp\left[\frac{i\gamma}{2}(x'y''-y'x'')\right]\exp\left\{-\frac{\gamma}{2}\left[(x''-x')^2+(y''-y')^2\right]\right\}$$

$$\times\sum_{n=0}^{\infty}\sum_{m=0}^{n}\frac{(-1)^{-n}2^{-2n}}{m!(n-m)!}H_{2m}\left(\sqrt{\frac{\gamma}{2}}(x''-x')\right)H_{2(n-m)}\left(\sqrt{\frac{\gamma}{2}}(y''-y')\right)$$

$$\times\lim_{\varepsilon\to 0}\int_{-\infty}^{+\infty}dE\int_{-\infty}^{+\infty}dp_z\,\frac{\exp\left[iE(t'-t'')+ip_z(z'-z'')\right]}{\varpi+i\varepsilon}$$

(1)

for a Klein- Gordon particle in a uniform magnetic field, where $\gamma = eB/2$, $\varpi = 1/2\left[E^2 - M^2 - p_z^2 - (n+1/2)2eB\right]$, B is the magnetic field and M is the mass of the particle. The Green function for the iterated Dirac equation takes the form

$$g(\mathbf{r}',\mathbf{r}'') = \frac{i\gamma}{(2\pi)^2}\sum_s \eta_s\eta_s^+\exp\left[iM\gamma(x'y''-y'x'')\right]\exp\left\{-\frac{M\gamma}{2}\left[(x''-x')^2+(y''-y')^2\right]\right\}$$

(2)

The energy spectrum for the Klein-Gordon and Dirac particle in a uniform magnetic field can be derived from the poles of Eq.(1) and Eq. (2) respectively. The energy spectrum obtained agrees with the known result.

Keywords: Brownian motion, white noise analysis, T-transform, Feynman integral

CMPSD NO. 36

FABRICATION AND CHARACTERIZATION OF PANI/ZnO DIODE VIA CBD

Edmond B. Salomsom[1], Editha P. Jacosalem[2], Reynaldo M. Vequizo[2,4], Rosario L. Reserva[2], Jingle B. Magallanes[3]

[1]Physics Department, Xavier University (XU)
Ateneo de Cagayan 9000 Cagayan de Oro City
[2]Material Science Laboratory (MSL), Department of Physics
Mindanao State University Iligan Institute of Technology
(MSU-IIT) 9200 Iligan City
[3]Computational High Energy Physics (IITCompHEP), Department of Physics,
Mindanao State University
Iligan Institute of Technology (MSU-IIT) 9200 Iligan City
[4]Faculty of Engineering, Niigata University, Japan
Email: rose@physics.msuiit.edu.ph

PANI/ZnO heterojunction diodes are fabricated by preparing a p-type HCl doped Polyaniline (PANI) on an n-type Zinc oxide (ZnO) films via chemical bath deposition method (CBD) at room temperature. Annealing of ZnO films at 500°C is done before coating a portion of it with the green conducting emeraldine salt (ES) form of PANI film by CBD. Electrical properties and characteristics of individual PANI films and ZnO films, and PANI/ZnO diodes are then determined at MSL, MSU-IIT while X-Ray Diffraction (XRD), operating at 20kV, 20mA, measurements are performed in Niigata Univerity, Japan. Measurements gave conductivities of the PANI films (10-15 ìm thickness) under dark condition in the order of 10^{-2} S/cm while that of ZnO (20-35 ìm thickness) in the order of 10^{-5} S/cm. The conductivity of the samples increased when illuminated with a 365nm wavelength UV light lamp illumination in the voltage ranging from 0.025 to 6 volts. The PANI/ZnO contact demonstrated a rectifying behavior by current-voltage (I-V) curves measured at room temperature. The forward bias I-V characteristics of the diode samples have exhibited ideality factors ranging from 4.25 to 5.40, a cut-in voltage from 0.27 to 0.68 volts, and a dynamic resistance ranging from 676k&! to 3M&!. The maximum reverse saturation current of the semiconductor diodes are in the order of 10^{-9}A. When the diodes were exposed to UV photons, dynamic resistance values decreased while the current increased linearly in the reverse bias region which means that the fabricated PANI/ZnO composite films are photosensitive devices. Signals from these samples show a rise time in the order of 10^{-9} to 10^{-6} seconds when exposed to UV illumination. The two peaks observed in the XRD patterns of all the PANI samples prepared at different concentrations correspond to the oligo-substituted PANI of high cyrstallinity. Also, the XRD result of ZnO film

semiconductor, with a peak of about 33^0, showed a good crystalline structure as expected.

Keywords: PANI, CBD, XRD, diodes, conductivity, forward bias, reverse bias, ideality factors

CMPSD NO. 37
SPATIAL RESOLUTION AND DRIFT VELOCITY MEASUREMENTS USING MICROMEGAS EQUIPPED TIME PROJECTION CHAMBER

Rosario L. Reserva[1]*, Dennis C. Arogancia[1], Angelina M. Bacala[1], K. Boudjemline[3], P. Colas[4], Keisuke Fujii[2], D. Fujishima[5], Masahiro Habu[6], Hermogenes C. Gooc, Jr[1], T. Higashi[5], M. Kobayashi[2], H. Kuroiwa[11], V. Lepeltier[4], Takeshi Matsuda[2], S. Matsushita[5], K. Nakamura[6] T. Watanabe[7], A. Yamaguchi[8], H. Yamaoka[2], O. Nitoh[6], R. Settles[9], A. Sugiyama[5], Y. Kato[10], and T. Zerguerras[4]

[1]High Energy Physics Laboratory, Department of Physics
MSU-Iligan Institute of Technology 9200 Iligan City, Philippines,
[2]IPNS, High Energy Accelerator Research Organization, Tsukuba, Japan,
[3]Carleton University, Ontario, Canada,
[4]Saclay, Orsay/LAL/IPN/CEDEX, France,
[5]Saga University, Japan,
[6] Tokyo University of Agricultural Technology, Japan,
[7]Kogakuin University, Japan,
[8] Tsukuba University, Japan,
[9] Max Planck Institute, Munich, Germany,
[10]Kinki University, Japan &
[11]Hiroshima University, Japan
Email: rose@physics.msuiit.edu.ph

Time Projection Chamber (TPC) technology is a promising central tracker for the proposed construction of energy frontier e^+-e- International Linear Collider (ILC). The scientific goals of the ILC are to develop spin-off technologies toward critical discoveries that lead to new fundamental principles in nature, medical science, and cosmology among others.

R & D studies on the performance of the micro mesh gaseous (micromegas) time projection chamber readout sensor as well as the gas properties used in a prototype Max Planck Institute (MPI)-built TPC were carried out using a 4 GeV/c pion beam in a magnetic field from 0 to 1 Tesla at the Proton Synchrotron 4p2 beam line of the East Experimental Hall, High Energy Accelerator Research Organization, Tsukuba, Japan.

Time Projection Chamber (TPC), a three-dimensional tracking device, was filled with 95% argon and 5% isobutane gas. TPC is a double–stage parallel plate chamber with 3mm conversion gap and 100 mm amplification region separated by micromesh structure. Voltage (350 volts) was applied on the microgrid that defines the cathode element of the amplification gap. A mixture of argon and isobutane gas was ionized inside the detector that leaves a trail of electrons in the gas drifting towards the conversion gap. Avalanche of electrons were produced at the anode strips under the action of the high electric field. Data were then read out by Aleph Electronics and analyzed using DoubleFit Program which reads run data files written in LCIO format.

Preliminary analysis of data reveals a spatial resolution $s_o = 127 \pm 2.0$ mm. With TPC drift length of 26.08 cm, recent measurement yielded an average electron drift velocity of 4.181 ± 3.73 cm/msec at a field of 220 V/cm, P = 1002 hPa at 27°C and magnetic field of 0.5 Tesla. These results are in very good agreement with the Magbolts simulation.

Keywords: time projection chamber (TPC), DoubleFit Program, micromesh structure and International Linear Collider (ILC)

CMPSD NO. 38

SIMTOOLS: PHYSICS/DETECTOR SIMULATION SUITE FOR THE INTERNATIONAL LINEAR COLLIDER

Allister Levi C. Sanchez[1*], Akiya Miyamoto[2], Keisuke Fujii[2], Ming-chuan Chang[3] Tomoaki Fujikawa[3], Hiroaki Ono[4], Atsushi Yamaguchi[5], Yoshihiro Yamaguchi[6] Sumie Yamamoto[7], and Tamaki Yoshioka[8]
(Active ACFA-SIM-J Developers)

[1]Department of Physics, MSU – Iligan Institute of Technology, 9200 Iligan City
[2]High Energy Accelerator Research Organization (KEK) Tsukuba 3050801, Japan
[3]Research Center for Neutrino Science, Tohoku University Sendai, Japan
[4]Graduate School of Science and Technology, Niigata University, Japan
[5]Institute of Applied Physics, Tsukuba University, Tsukuba 305-8573, Japan
[6]Institute of Physics, Tsukuba University, Tsukuba 305-8571, Japan
[7]Graduate University for Advanced Studies, Hayama 240-0193, Japan
[8]International Center for Elementary Particle Physics, Tokyo University
Tokyo 113-8654, Japan

The international high energy physics (HEP) community is full of excitement in anticipation of the completion of, and fresh experimental data from, the Large Hadron Collider (LHC) which is under construction at CERN in Europe. The LHC

will collide protons against antiprotons at a center-of-mass energy of 14 TeV, and is expected to discover the elusive Higgs boson, the particle believed to be responsible for the masses of all the other particles. This much-anticipated discovery, however, will only be the first step in our understanding of the physics beyond the Standard Model. And the LHC, limited in precision by the noise generated by the multiple quark interactions between the protons and antiprotons, will leave many important physics regions unexplored. Thus, there is now an international consensus on the necessity of an electron-positron collider.

The International Linear Collider (ILC) project is being proposed by the global HEP community to complement the LHC. In contrast to the noisy proton-antiproton collisions at LHC, the electron-positron collisions at the ILC are expected to produce a clean environment for the precise measurement of the final states observed. In preparation for the ILC, HEP institutions worldwide are now conducting R&D on the physics requirements on, as well as the technological viability of, the particle detector devices. One important component in this effort is the availability of reliable computer simulation tools to help optimize the detector design and performance. It is in this context that we develop SimTools.

SimTools is a collection of software packages for the Monte Carlo event generation of physical phenomena, the simulation of detector response to incident particles, and the analysis of simulated as well as real experimental data. The included software are JSF, LCLIB, Physsim, Leda, Jupiter, Uranus, and Satellites. SimTools contains event generators such as Pythia, Bases/Spring, and Herwig, among others. ILC detector response can be studied with QuickSim (for fast simulation) and Jupiter (for detailed and realistic detector simulation). SimTools is based on two powerful physics software: ROOT, an object-oriented data analysis framework, and Geant4, a toolkit for the simulation of the passage of particles through matter.

In this report we present the status and maturity of SimTools and demonstrate some of its capabilities. We hope to promote ILC-related R&D in the country – to increase the country's contribution to the ground-breaking field of high energy physics and to hasten the development of spin-off technologies that will prove useful to the nation.

Keywords: linear collider, detector simulation, software, high energy physics

ENGINEERING SCIENCE AND TECHNOLOGY

ESTD No. 1
DEVELOPING ENVIRONMENTAL MANAGEMENT ZONES USING GIS TECHNOLOGY

Nathaniel C. Bantayan

Institute of Renewable Natural Resources
College of Forestry and Natural Resources
University of the Philippines Los Baños, College, Laguna 4031
Email: ncbantayan@gmail.com

An environmental management plan (EMP) was prepared for the province of Lanao del Sur with focus on the Lake Lanao watershed. This study was conducted with support from The Netherlands and implemented through collaborative efforts among different agencies like FAO, UNDP, ARMM and other stakeholders.

The scope of the EMP is extensive in terms of geographic coverage and purpose considering the state of human welfare and environmental condition in the area. While Lake Lanao Watershed reportedly provides 70% of the power needs of Mindanao, ironically, the Meranaos have not sufficiently benefited from being stewards of this vital resource. The preparation of the EMP was guided by this fact including the roles and responsibilities that must be played and shared in order to ensure the success of the Plan and thus sustain the benefits which can be derived from the watershed.

The planning exercise revealed important insights into the process of plan preparation: (1) The development of baseline information (e.g. basemaps in digital (GIS) format) is an important advantage for Lanao del Sur. The necessary next step is to provide a mechanism for the operationalization of the EMP and the implementation of the identified proposals contained in the plan. Thus, the zones were expressed according to the distribution in each local government unit (LGU) to allow them (the LGUs) to make comparisons and integrate these with their own respective land use plans. (2) It is crucial that the EMP be validated on the ground and integrated into the individual management plans of the municipalities of Lanao del Sur. (3) DENR-ARMM should consider this dataset as an important reference material in designing land use interventions in the province. However, validation and its continued and regular updating should be pursued as part of the monitoring and evaluation activities of the province.

Keywords: environmental management plan; geographic information system; management zones

ESTD No. 2

IDENTIFICATION OF EROSION-PRONE AREAS IN THE PROVINCE OF AKLAN USING GEOGRAPHIC INFORMATION SYSTEM

Alan Dino Moscoso*

School of Technology
University of the Philippines in the Visayas
5023 Miagao, Iloilo
Email: admoscoso@hotmail.com

This study used the Universal Soil Loss Equation (USLE) model to compute the rate of soil erosion in the 17 municipalities of the province of Aklan, Philippines. About half of the land area in this 181,790-hectare province is allotted to agriculture, thus, proper management of its land resources is of great importance.

The erosion factors in the USLE model (rainfall erosivity, soil erodibility, slope length and steepness and land cover and management) were computed from data gathered from various agencies and institutions. Data on vegetation cover were interpreted from a Landsat 7 image acquired on July 30, 2001. Geographic Information System was then used to integrate these factors to come up with the erosion potential map in terms of mass of soil loss per area of land. Digitizing of paper maps was done through Arcview v8.3 while interpolation and overlaying of values were done using IDRISI v14. The results of the study show that the average soil loss in the province was 5.8 ton/ha/yr, a bit higher than the national average of 2.69 ton/ha/yr. The erosion map was further classified into five different classes of erosion intensity. Results further showed that 82% of the province had a low soil loss rate (less than 7.4 ton/ha/yr). Areas with severe erosion rate (>37 ton/ha/yr) comprised 2.5% and were located on steep (>30% slope) hills and mountains, and on grassy areas. Thirty-nine percent of these areas are found in the municipalities of Madalag and Libacao. In terms of vegetation cover, grasslands had the highest average soil loss of 19.8 ton/ha/yr. This study concluded that, for Aklan province, the slope length and steepness factor had the highest correlation with soil loss.

Keywords: geographic information system, erosion, soil, remote sensing, USLE

ESTD No. 3

COMPARISON OF TECHNIQUES FOR BIODIVERSITY ASSESSMENT

Naty Catotal[1], Leonardo Co[2], Daniel Lagunzad[3] and Nathaniel C. Bantayan[4]*

[1]Environment and Natural Resources Office, Province of Masbate,
[2]Conservation International – Philippines
[3]Institute of Biology, College of Science
University of the Philippines Diliman
[4]Institute of Renewable Natural Resources
College of Forestry and Natural Resources
University of the Philippines Los Baños
Email: ncbantayan@gmail.com

A study of landuse necessitates a careful consideration of the factors that determine its appropriateness and sustainability. The key then is to assess the state and identify areas that are susceptible to agents of land degradation. The analysis of land capability is implemented by comparing an estimate of land degradation with a tolerance limit. The tolerance limit prescribes the threshold for agricultural productivity above which necessitates leaving the area for forest-use purposes. To operationalize the analytical procedure, a system for geodatabasing the general characteristics of the land for assessment, monitoring and evaluation was developed. The description of the landscape is carried out using the vecter approach – a new method for comprehensively describing all aspects about the landscape. The vecter approach enriches the description of the landscape through the various levels of the GAME model – GIS-based assessment, monitoring and evaluation. The geodatabase component of GAME allows a comprehensive description of geographic phenomena. The mapping component, on the other hand, enables visualization of the landscape using GIS. Thus, the status of natural resources (e.g. forest cover) can be readily assessed; the progress of relevant projects (e.g. forest renewal) can be monitored; and the quality of natural resources and the impact of interventions (e.g. forest conversion) can be evaluated.

As a technique for biodiversity assessment, the GAME Model was compared with the line plot and strip methods using actual census data from a 16-ha permanent plot in Palanan, Isabela. The results of the study show that species abundance is dependent on the sampling intensity used in a particular inventory technique. Among the three inventory techniques, the GAME Model proved the most economical and practical.

Keywords: geographic information system; biodiversity; geodatabase

ESTD No. 4

IMAGE ANALYSIS OF LEAF SHAPE VARIATIONS IN ENDEMIC, INDIGENOUS AND NEW SPECIES OF HOYAS FROM THE PHILIPPINES USING ELLIPTIC FOURIER SHAPE DESCRIPTORS

[1]Mark Anthony J. Torres and Cesar G. Demayo and [2]Simeona V. Siar

[1]Department of Biological Sciences, College of Science and Mathematics
Mindanao State University-Iligan Institute of Technology, Iligan City
[2]Institute of Plant Breeding, College of Agriculture,
University of the Philippines Los BañosCollege, Laguna, Philippines
Email: c_gdemayo@yahoo.com

Applying technological advances in image analysis have contributed much in the advancement of understanding sources of variations not only in non-living objects but also in living organisms. Modeling shapes and understanding developing organisms including their relationships have already been possible by combining image analysis of the geometry of shapes coupled with appropriate statistical tools have increased our knowledge in understanding complexities in living organisms. In this study, we studied leaf shape variations in eleven (11) endemic, two (2) indigenous and three (3) new species of Philippine Hoyas by leaf contours summarized as increments of x and y coordinates extracted from scanned images (600 dpi) of leaves using image analysis and processing software. Elliptic Fourier (EFA) shapes descriptors, which are the cosine and sine components of the x and y increments along the leaf outline for the first ten harmonics were then computed. These descriptors were then normalized to avoid variations related to size, rotation and starting point of the contour traces. This procedure has allowed for the analysis of leaf shape variations only. Then, the leaf outlines were reconstructed using all ten harmonics. The information contained in the coefficients of the EFA descriptors were summarized through principal component analysis based on a variance-covariance matrix of the coefficients. The scores of the components were used in the subsequent analysis of leaf shape characteristics. Results showed variations in leaf shapes among the different species of Hoyas. Also, the first few components accounted for the total leaf variation and were good measures of the ratio of the length to width, the position of the centroid, and the curvature of the leaf wing size. It is very interesting to report that this methodology is very effective in illustrating graphically and statistically the variations in leaf shapes among the different species of Hoyas and can also be applicable in analyzing not only other biological shapes but also images of inanimate objects.

Keywords: image analysis, leaf variations, Hoyas, Elliptic Fourier Analysis

ESTD No. 5

ODOR EMISSION REDUCTION FROM ENCLOSED GROWING-FINISHING PIG HOUSE USING DIFFERENT BIOFILTER MEDIA

J.I. Song[1], Y.H. Yoo[1], T.I. Kim[1], Antonio J. Barroga[2], H.C. Choi[1], J.W. Jeong[1] K.Y. Yeon[1], C.B. Yang[1], D.H. Kim[2], and J.W. Lee[3]

[1]RDA – National Livestock Research Institute, Suwon, 441-350, Korea
[2]Central Luzon State University, Science City of Munoz
Nueva Ecija, Philippines
[3]Department of Animal Resources Technology
Chinju National.University, Korea;
[4]Gyeongnam Province Advanced Swine Research Institute, Korea
Email: antonio_barroga@yahoo.com

This study was conducted to determine the odor reduction efficiency of a biofilter design using different filter materials. The summary of results are as follows: (1) The airflow penetration rate of the different filter materials namely; rice straw, woodchips, rice hulls and sawdust were 0.72 m/s, 0.64 m/s, 0.48 m/s and 0.17 m/s, respectively. (2) Compared to a pig barn with no biofilter, NH_3 emission was reduced by 77% using a biofilter media of sawdust and wood chip followed by rice hull and rice straw with a removal efficiency of 69% and 46%, respectively. Based on a two bi-weekly monitoring of NH_3 emission, wood chip as a biofilter media proved to be the most superior and consistent in reducing this gas due to removal efficiencies of 76%, 55% and 76% for days 7, 14 and 36, respectively. On the other hand, rice hull was the most effective among biofilter media in reducing H_2S with a 5.76% removal efficiency. The above findings indicated that NH_3 can be easily trapped/ absorbed effectively by all the biofilter media than H_2S. Finally, the airflow penetration rate of the different biofilter media tended to be related to odor elimination efficiency with sawdust having the slowest penetration rate of 0.17m/s.

Keywords: biofilters, odor reduction efficiency.

ESTD No. 6

POTENTIAL OF LIGNOCELLULOSIC MAT AS FILTER FOR INDUSTRIAL WASTE WATER

Shirley C. Agrupis[1] and Bernard Benjamin P. Albano

[1]Biology Department , College of Arts and Sciences
Mariano Marcos State University, Batac 2906 Ilocos Norte
Email: shirleyagrupis@yahoo.com

The study was conducted primarily to develop a lignocellulosic mat from waste banana trunks for use as filter for industrial waste water effluent from a secondary fiber-based paper industry. The banana fibers were prepared by retting the material in four different retting times: 1, 2, 3 and 4 weeks. Activated carbon and unretted banana fibers were used as controls. The different treatments were fitted separately in an improvised water column for the run through trials. The filtration capacity of the lignocellulosic mats including the controls were used twice in 48 hours to evaluate their re-usabilities.

Retting time affected significantly the removal of extractives without considerably affecting the other biomass components of the material. The significant removal of extractives with prolonged retting time correspondingly favored defibrillation of the individual banana fibers for a much improved mat formation. In effect, the freeness (flow of water, mL/sec) of the mat decreased, favoring the filtration efficiency of the lignocellulosic mat.

The filtration performance of the banana fiber lignocellulosic mat (BCFM) is generally comparable with that of the activated carbon and even showed better filtration capacity than the latter during the second time run through trials. Data analysis using the orthogonal polynomial contrast showed that prolonged retting time had a direct effect on the ability of the BCFM to reduce or remove the water pollutants. Except for the microbial load, all the other pollution indicators: Total dissolved solid (TSS); Total suspended solids, (TSS); biological oxygen demand (BOD); chemical oxygen demand; Calcium and calcium carbonate were reduced significantly by the BCFM filter.

Key words : lignocellulosic mat, filter, waste water effluent, banana fiber

ESTD No. 7

STUDIES ON THE PREPARATION OF BANANA PULP FOR VULCANIZED FIBER

Shirley C. Agrupis

Biology Department , College of Arts and Sciences
Mariano Marcos State University, Batac 2906 Ilocos Norte
Email: shirleyagrupis@yahoo.com

Banana pulps were prepared by soda, soda AQ and retting process with two stage bleaching processes following the TAPPI standard procedures with some modifications to suit the fiber material. The pulps produced showed superior properties even before bleaching and beating. However, only those prepared by retting process met the criteria for vulcanization; e.g. low air permeability (40.70 sec/100ml) and superior strength properties. Hence, only those sheets prepared from retted pulp were used for the vulcanization trials.

Sufficient number of sheets at two different basis weights: 90 and 120 gsm were prepared and conditioned for the vulcanization. Two-ply sheet vulcanization was done by placing the two sheets on top of the other and passed them together in the vulcanization series. Their adjacent surfaces penetrated by the $ZnCl_2$ swelled and gelatinized making the two sheets to fuse homogenously. The same process was done for the one-ply sheet. Percent (%) $ZnCl_2$ absorption by the sheets was found higher in the 90 gsm- than the 120 gsm-sheets for both the one- and two- ply samples. Correspondingly, the plybond strength of the two-ply sheets was remarkable with the 90 gsm sheet giving higher strength (3.33 lb/inch^2) than the 120 gsm (2.86 lb/inch^2). The values obtained were within the ranges obtained in previous studies using other materials like tobacco, kenaf, commercial softwood unbleached and bleached kraft pulps (Agrupis, 2000).

The physical appearance of the banana vulcanized fiber sheets closely resemble those of the commercial grade vulcanized fiber, but obviously showed the need to further improve the quality. Vulcanized fiber is a high grade cellulosic sheet, which is water- and heat-resistant. It is used in many industrial applications like electrical washers, heat insulators and the like.

Keywords : Vulcanized fiber, TAPPI(Technological Asso. of Pulp and Paper Industries) AQ (Anthraquinone), banana fiber

ESTD No. 8

CHARACTERIZATION AND CEMENTITIOUS SOLIDIFICATION/ STABILIZATION OF A SLUDGE GENERATED BY A METAL COATING/PLATING PLANT USING RICE HULL ASH AS ADDITIVE

Rex B. Demafelis, Renacel P. Promentilla, Ronald R. Navarro and Albert A. Samuela

Dept. of Chemical Engineering, College of Engineering and AgroTechnology, University of the Philippines Los Baños, College, Laguna 4031, Philippines
Email: rbdema@yahoo.com

The semiconductor industry is the top exporting industry in the Philippines. Sludges containing heavy metals from semi-conductor and metal coating/plating plants pose a grave threat to the environment that need to be given attention.

A treatability study was conducted to determine the feasibility of employing solidification/stabilization (S/S) technology to treat a heavy metal-contaminated sludge generated by the wastewater treatment facility of a metal coating/plating plant and evaluated the performance by conducting the Toxicity Characteristic Leaching Procedure (TCLP), compression and durability tests. The use of rice hull ash (RHA) as partial substitute to ordinary Portland cement (OPC) that will act as the S/S binder or reagent was also investigated.

Seven treatments of OPC, RHA and sludge were designed for the relative amounts of mixture components. Seven duplicate concrete blocks were molded and cured for 28 days. Three blocks were tested for unconfined compressive strength (UCS) using the universal testing machine (UTM). Two blocks were subjected to wetting and drying cycles to test the durability. TCLP was employed to determine the percent reduction in the nickel that leached after extraction.

The results of the study clearly show that the mobility of heavy metals can be effectively reduced by the S/S process. Leaching of nickel can be minimized or retarded to meet standards by blending the waste with siliceous materials such as Portland cement and rice hull ash. Immobilization is more effective when the OPC is available at higher amounts as in higher formulations.

From the general comparison made on the seven mixture formulations, OPC-RHA-SLU ratio of 0.3/0.35/0.35 was rated the most outstanding composition after passing the leachability, compression, and durability test, having the highest OPC substitution with RHA hence highest binder cost reduction, and highest amount of sludge treated.

Keywords: metal coating-plating plants, nickel, heavy metals, solidification/ stabilization technology, sludge, rice hull ash

ESTD No. 9

RHEOLOGICAL AND CASTING PROPERTIES OF SANITARY WARE SLIPS WITH RICE HULL ASH

Samuel S. Franco and Rowelyn C. Aganus

Department of Ceramic Engineering, College of Engineering
Mariano Marcos State University, Batac, 2906, Ilocos Norte
Email: samuelsfranco@yahoo.com

The manufacture of ceramic sanitary ware utilizes white clay, feldspar and quartz and these are usually imported. Rice hull ash is high grade amorphous silica when subjected to heat. We evaluated the progressive replacement of quartz by active silica obtained from rice hull ash for ceramic casting slips to determine the level of substitution to be used without affecting its rheological property.

Calcined rice hull ash was used to gradually replace the quartz content of the slip at 3%, 6%, 9% 12% and 15%. Sodium silicate (NA_2SiO_3) as the electrolyte and deflocculant was added at a varying rate of 1.3%, 1.6% and 1.9%. Rheological properties such as viscosity, specific gravity and casting rate were determined

Results of the experiments indicate that the addition of rice hull ash significantly increased the viscosity (decrease in fluidity). Using 1.9% deffloculant, all the formulations flowed while with 1.3% of deffloculant, only up to 6% quartz substitution was possible. Deflocculation level above 1.3% resulted in high viscosity that resisted flow. Increased percentage of rice hull ash also increased the thickness of the hollow cast specimen. However, increasing the percentages of deflocculant decreased the cast rate for all formulations.

Furthermore, results of the study revealed that the utilization of the calcined rice hull ash could be utilized for ceramic manufacturing particularly for slip casting but should not to exceed 3% to maintain an acceptable rheological characteristics and proper degree of deffloculation for ceramic sanitary ware casting slips.

Keywords: calcined rice hull, ceramic, deflocculant, feldspar, quartz, rheological properties, specific gravity, sodium silicate, viscosity, white clay

ESTD No. 10

REMOVAL OF LEAD FROM ELECTROPLATING WASTEWATER USING PHOSPHONOMETHYLATED POLYETHYLENEIMINE (PPEI)-Ca^{2+}

Jovita l. Movillon, Ronald R. Navarro, Casiano S. Abrigo Jr., Myra G. Borines, Elson R. Montibon, and Myriam L. Perez

Dept. of Chemical Engineering, CEAT-UPLB
College, Laguna, 4031 Philippines
Email: mgborines@yahoo.com

Lead contamination in the environment has long been recognized as a serious health problem. Lead, despite its toxic effect, is still being used by industries. This study aimed to evaluate the performance of phosphonomethylated polyethyleneimine (PPEI) as a chelating flocculant for the removal of lead from an actual electroplating wastewater. The effect of equilibrium pH conditions and the amount of PPEI used on lead flocculation using PPEI-Ca^{2+} as chelating flocculant was studied. The regenerative property of the PPEI polymer was tested and an absorption isotherm was established for Pb^{2+}.

Even at a low equilibrium pH condition (pH 3), the polymer was able to remove 66.93% of the initial 2.06 ppm Pb^{2+} concentration. At high equilibrium pH (pH 8, 9, and 10), almost 100% removal was obtained. The results showed that increasing equilibrium pH condition increased lead removal. This may be due to the improved affinity between PPEI and Pb^{2+} at high pH, where less hydronium ions would be available to compete with Pb^{2+} and Ca^{2+}. The same trend was observed at increasing PPEI amounts. At equilibrium pH 10, even a small amount of 0.05 mL of PPEI can already remove more than half (59.08%) of Pb^{2+} in a 40 mL electroplating wastewater and 0.50 mL PPEI can already remove almost 100% of Pb^{2+} in the solution.

This study showed that in lead flocculation using PPEI as chelating flocculant, the PPEI can be used up to the second cycle with an 85.99% regeneration and would still show satisfying results. It also showed that Pb^{2+} follows the Freundlich absorption isotherm with a 36.69037 mg/g absorptive capacity and 1.95413 L/mg stability constant. Therefore, lead flocculation using PPEI-Ca^{2+} as chelating flocculant is a good method for lead removal in wastewater.

Key words: lead, PPEI-Ca^{2+}, phosphonomethylated polyethyleneimine, flocculation

ESTD No. 11

BIOMASS RESOURCE RECOVERY SYSTEMS FOR SUSTAINABLE AGRICULTURE

Bernardo D. Tadeo, Jocel C. Cordero*, Rizal G. Corales*, Dexter L. Orpilla, Larry A. Santos, Ailon V. Capistrano

Philippine Rice Research Institute
Maligaya, Science City of Muñoz
3119 Nueva Ecija
E-mail: jcordero@philrice.gov.ph

Through effective and practical conversion or transformation of wastes into resources such as biomass residues that contain abundant organic matter, nutrients and other components can be recycled beneficially into the soil as amendments or nutrient sources.

A close loop rice resource recovery system or 3RS in the agricultural production that considered turning waste into resource was developed. Stopgap technology using a carbonizer with smokestack can convert 182 kg rice husk into 119 kg carbonized rice hull (CRH) in 3 to 4 h with 65% conversion efficiency. On the other hand, fermentation-decomposition pathway using effective microorganism or EM Technology reduced decomposition of biomass in 2 to 4 weeks.

The 3Rs technology was introduced to more than 10,000 farmers nationwide wherein 7 People's Organization engaged in the production and marketing of CRH and organic fertilizer. These resulted in additional profit and income of about 30%. Likewise, the utilization of organic fertilizers reduced the farm input cost of about 30 to 40%.

The technologies are simple and appropriate and can encourage the general public to increase the utilization of biomass to a level that would equal to less generation of wastes.

Keywords: Biomass, conversion, resource recovery system, carbonizer, EM technology

ESTD No. 12

DEVELOPMENT OF TRANSPARENT LEADLESS GLAZE FOR LOW-FIRED CERAMIC WARE UTILIZING LOCAL RAW MATERIALS IN ILOCOS NORTE

Jime C. Portugal

Department of Materials Science and Engineering, College of Engineering
Mariano Marcos State University, Batac, 2906, Ilocos Norte
Email: jimeportugal@yahoo.com

The ceramic industry in the Ilocos Region relies on glazes that are prepared by a company specializing in supplying industry. For this reason, potters are investing on expensive commercial (imported) glazes, since glazes are at one and the same time the area of most fascination and most difficult for the potters. Additionally, the producers do not usually have access to raw materials and its chemical analysis at reasonable prices.

The study focused on the development of transparent leadless glaze for low firing ceramic products utilizing local raw materials such as Solsona white clay, Ventura feldspar and red silica with the addition of commercial carbonates and boron compounds (borax). The compatibility of such materials as alternative replacement for commercial glaze materials was determined.

Raw materials were subjected to preliminary drying, grinding and screening to achieve even particle size of the glaze materials. The prepared slip glaze batched was applied on both red clay and earthenware (white ware) bodies made from slabbing and fired in gas fired (LPG) kiln at 1050°C based on the recording pyrometer of the kiln.

Results obtained from the formulated glaze show a clear transparent glassy surface appearance in the test ceramic clay bodies. The clay bodies and glaze formulation were found to be acceptable with some minor defect such as pin holes but these gave a unique artistic surface appearance especially for decorative artwares.

Keywords: transparent leadless glaze, slip glaze, red clay, earthenware, gas fired, recording pyrometer, slabbing.

ESTD No. 13

PRODUCTION OF HYDROGEN FROM ETHANOL THROUGH STEAM REFORMING USING A FABRICATED CATALYTIC REACTOR

Sixto A. Valencia , Jovita L. Movillon
Catalino G. Alfafara and Erli Eros D. Lee

Dept. of Chemical Engineering,
College of Engineering and Agro-Technology,
University of the Philippines Los Baños
College, Laguna, 4031 Philippines
Email: rbdema@yahoo.com

Developmental work on renewable energy source is imperative in the face of the threatening energy crisis. Hydrogen is a clean and good substitute to fossil-based fuel. This study focused on producing hydrogen from ethanol using a fabricated catalytic reactor with $CuO/ZnO/Al_2O_3$ catalyst. The effect of reaction temperature and reaction time on percent recovery of hydrogen were also tested. The catalyst used was prepared by first precipitating oxides of copper, zinc and aluminum. The precipitated oxides were then filtered, dried in an oven and calcined in a furnace at 400°C. The calcined precipitate was granulated after which a binder (graphite) was added and lastly it was pelletized.

During the ethanol reforming experiment, the catalyst packed in the reactor was divided into half to ensure good surface area of contact. An ethanol to steam mole ratio of 1:45.5 mole were passed-on to the reactor at certain set of reaction temperature and reaction time. At each set of reaction temperature and reaction time, samples were collected by water displacement method. The samples were collected at reaction time at 200°C and 400°C. At each reaction temperature, reaction times were set at 10 seconds and 30 seconds. The samples collected were analyzed for its components by gas chromatography (GC).

Results of the GC analysis showed that the highest percent volume and percent recovery of hydrogen were obtained in the sample collected at reaction temperature of 400°C and reaction time of 10 seconds. The said sample contained 66.09% volume hydrogen giving a 34.30% recovery of hydrogen. The sample taken at 400°C too, but at longer reaction time, set at 30 seconds, had a lower percent recovery of 27.06%. Those samples gathered at a lower temperature of 200°C had a lower percent recovery of 16.40% and 4.48%. Percent recovery was calculated on the assumption that all the hydrogen in the ethanol prepared was totally recovered.

It was observed that the production of hydrogen was directly related to reaction temperature and inversely related to reaction time. Further, as the reaction temperature increased, percent recovery of hydrogen also increased. ZnO (an

active agent of the catalyst used) activity at a higher temperature predominated because of its redox property, which promoted the steam reforming reaction, thus producing more hydrogen. On the other hand, percent recovery of hydrogen was observed to decrease as reaction time increased. At a longer reaction time, H_2 was given a chance to react with CO_2 via the reverse water gas shift reaction that resulted in the production of CO and H_2O; thus the recovery of hydrogen became smaller.

Keywords: hydrogen gas production, catalytic reactor,

ESTD No. 14

FABRICATION AND PHYSICAL PROPERTIES OF PROTOTYPE CERAMIC WATER FILTER ELEMENT PREPARED FROM MIXTURES OF RICE HULL ASH, KAOLINITE CLAY, AND PULVERIZED CHARCOAL

Dionesio C. Pondoc and Jennifer L. Bautista

Department of Ceramic Engineering
College of Engineering, Mariano Marcos State University
Batac, 2906 Ilocos Norte
Email: dondon_pondoc@yahoo.com

Fabrication of prototype ceramic water filter element was conducted utilizing calcined rice hull ash, kaolinite clay from Solsona, Ilocos Norte, pulverized charcoal and organic binder. Five different mixtures (F1, F2, F3, F4, and F5) were formulated based on the batch composition of 8.65% clay material, 67.31% calcined rice hull ash, 19.23% combustible material and 4.81% starch as organic binder. Solid cylindrical test specimens with a dimension of 9.19 cm long and 3.18 cm diameter were fabricated from the formulated mixtures using semi-plastic pressing process in a steel mold. The forming process was done by manual pressing operation using Nagasaki NSP-5 hydraulic press with a pressure load of approximately 150kg/ cm^2 and holding time interval of 3
–5 minutes per test specimen. The formed test specimens were subjected to both natural drying for 24 h and oven drying for 8 h at 110°C. The test specimens were sintered at temperature of 1100°C with varying sintering time of 2, 3, and 4 h respectively, under oxidizing condition.

Physical properties of the sintered test specimens like apparent porosity, bulk density, linear shrinkage, water absorption, and water permeability were determined and compared with the properties of commercial ceramic water filter element in order to evaluate if the fabricated element will pass the acceptable standards.

Results show that test specimen of F1, F3, and F4 sintered at 1100°C for 3 and 4 h with an apparent porosity of 70.59%, 69.49% and 69.30% respectively, were acceptable based on the requirements for the commercial water filter element with a minimum value of 68.84%. The bulk densities of all test specimens from the five mixtures were within the range of 0.60–0.67 g/cc which were also acceptable compared with the minimum value of 0.63 g/cc in the commercial. All mixtures have an acceptable result of good filterability within the range of 94–95 % efficiency based on their water absorption and water permeability test.

Keywords: ceramic water filter element, semi plastic pressing, and physical properties

ESTD No. 15

ALPHA-PSO: DEVELOPING A SWARM OPTIMIZER AND ITS APPLICATION IN PROCESS INTEGRATION

Kristine Joy Col-long and Raymond Girard Tan

Chemical Engineering Department
De La Salle University, Manila
Email: tanr_a@dlsu.edu.ph

Due to rapid industrialization, water use has increased six-fold over the last century. Freshwater resources have been rising in cost and end-of-pipe wastewater treatments have become expensive too. Process Integration aims to optimize interrelationships of the different units in a plant to meet performance criteria in plants, such as maximum profit or minimum cost, energy efficiency or good operability. Evolutionary-based algorithms have been the subject of extensive researches to approach these types of problems and particle swarm optimization (PSO) is the most recent development in this field. Several variations have been introduced since PSO was presented in 1995. ALPHA-PSO was developed to enhance the search performance of the basic PSO and is based on the concept of having a leader in a group. The leader is called the alpha-particle. Only the leader knows the best performing follower in his swarm. On the other hand, the follower particles have no global vision. Thus, they can only access g_{best} indirectly through the alpha-particle. A set of ten benchmark functions was used to compare its computational efficiency against five other PSO variants. Statistical tests were done and showed significant improvement over them.

Keywords: swarm intelligence, process integration

ESTD No. 16

OPTIMIZATION OF BIOFUEL LIFE CYCLE GREENHOUSE GAS EMISSIONS USING PARTICLE SWARM COMPUTATION

Liezzel P. Aquino and Raymond R. Tan

Chemical Engineering Department, De La Salle University-Manila
2401 Taft Ave., 1004 Manila, Philippines
Email: liezzelpaquino@yahoo.com; liezzel.pascual@dlsu.edu.ph

Coconut methyl ester or CME is one of the promising fuel alternatives used in the Philippines. It is derived from coconut oil by transesterification. The use of CME in the Philippines was supported when the national government in 2004 issued an order directing all government departments to incorporate the use of one percent (1%) CME in their diesel-fueled vehicles. Along with the production of CME, during farming, considerable amount of coconut residues are produced and disposed. In fact, according to the report of the Philippine Department of Energy, in 1997 the total available shell and husk are 1.79 and 4 million metric tons, respectively. In connection with this supply, the United Nations Development Program and the World Bank estimated a total of 20 MW potential power source based from these residues alone. Furthermore, during the pressing of the coconut oil from coconut meat or copra, substantial amount of copra meal is generated. Total life cycle greenhouse gas (GHG) emissions from four different biofuels: methanol, ethanol, coconut methyl ester and coconut ethyl ester were quantified. Life cycle assessment or LCA is a process to evaluate the environmental burdens associated with a product system, or activity by identifying and quantitatively describing the energy and materials used, and wastes released to the environment, and to assess the impacts of those energy and material uses and releases to the environment. Different fuel pathways were considered to form the biofuels superstructure. This consists of options for cogeneration and cofiring using agricultural wastes and coconut meals as well as utilization of agricultural wastes to produce raw materials such as ethanol and methanol. The minimum life cycle GHG emissions were computed based on the optimum fuel paths using swarm intelligence for the optimization. Particle swarm optimization (PSO) is an algorithm developed by Kennedy and Eberhart in 1995, which is based on the social behavior of animals. PSO was first applied in unconstrained optimization problems with continuous variables. This study explores the different pathways on how these agricultural wastes can be utilized to obtain the minimum total GHG emissions on a life cycle basis. Convergence characteristics of the different variants of the algorithm were studied.

Keywords: greenhouse gas, life cycle and particle swarm optimization

ESTD No. 17

SYNTHESIS OF OPTIMAL TOPOLOGICALLY-CONSTRAINED WATER REUSE NETWORK USING SWARM INTELLIGENCE

Hul Seingheng[1], Raymond R. Tan[1], Joseph Auresenia[1], Tetsuo Fuchino[2], Dominic C. Y. Foo[3]

[1] Chemical Engineering Department,
De la Salle University-Manila, Philippines,
2401 Taft Avenue, 1004 Manila, Philippines
[2] Tokyo Institute of Technology, 2-12-1 Ookayama
Meguro-ku, Tokyo 152-8550, Japan
[3] School of Chemical and Environmental Engineering, University of Notthingham Malaysia, Jalan Broga , 43500 Semenyih, Selangor, Malaysia
Email: hulsengheng@yahoo.com

Industries are finding ways to minimize the water costs and to reduce the water pollution through efficient use of water supplies. Process water integration achieves minimum water usage was by establishing the reuse scheme, or interconnections, from the sources to feed the demands in an industrial plant. A procedure for designing reuse networks with topological, network complexity and stream matching constraints has been developed. The procedure uses particle swarm optimization (PSO) which is an evolutionary algorithm based on the behavior of social animals developed by Kennedy and Eberhart in 1995. In this study, a prototype software coded using Visual Basic was developed. The case study of the acrilonitrile plant from El-Halwagi (1997) was used to demonstrate the algorithm. A 60% reduction of freshwater consumption was achieved for just one recycle stream, while the result by linear programming gave about 70% reduction with two recycle streams. This methodology thus provides flexibility to plant designers to balance the freshwater usage and the retrofitting capital cost.

Keywords: Process water integration, interconnection, particle swarm optimization

References

1. El-Halwagi, M. M. (1997). Pollution prevention through process integration: Systematic design tools. Academic Press, Amsterdam.
2. Kennedy, J. and Eberhart, R. (1995). Particle swarm optimization. Proceeding of the IEEE international conference on neural networks (Perth, Australia). 1942 – 1948.

ESTD No. 18

APPLYING ARTIFICIAL NEURAL NETWORK FOR VAPOR LIQUID EQUILIBRIUM PREDICTION

Nguyen Viet Dinh[1], Raymond R. Tan[1], Yolanda Brondial[1], Tetsuo Fuchino[2]

[1]Department of Chemical Engineering, College of Engineering
De La Salle University-Manila
[2]Department of Chemical Engineering, Tokyo Institute of Technology
Email: nguyendinhvietbk@yahoo.com

Most solvents used in the semiconductor industry are toxic and costly. Thus, the solvents should be recovered for re-use in these processes by distillation methods, and vapor liquid equilibrium (VLE) data are necessary for the design and operation distillation columns. VLE data can be estimated using thermodynamic models (Wilson and Tan-Wilson) based on calculation activity coefficients. In this work, artificial neural networks (ANNs) developed using MatlabÒ software were applied to predict and estimate VLE data for ternary systems saturated salts. The databases taken from Tan et al. (2004 and 2005) were split into training, validating and testing data and the best architecture was 8-7-5-4 network. The mean deviations were 0.01279, 0.009336 and 0.499 K for vapor mole fraction of ethanol, 1-propanol and bubble points, respectively. The ANNs showed better agreement with published experimental data than the thermodynamic models.

Key words: ANNs, VLE, ternary system, salts.

References

1. Demuth, H.and Beale, M. (1998). Neural network Toolbox, Version 3. The Math Works Inc.
2. Hagan, M. T., Demuth, H. B. and Beale, M. (1996). Neural Network Design. PWS Publishing Co.
3. Tan, T. C., Chai, C. M., Tok, A.T. and Ho, K.W. (2004). Prediction and experimental verification of the salt effect on the vapor liquid equilibrium of water- ethanol- 2-propanol mixture. Fluid phase equilibrium 218, 113-121.
4. Tan, T.C., Tan, R. and Soon, L.H., Ong S.H.P. (2005). Prediction and experimental verification of the effect of salt on the vapour- liquid equilibrium of ethanol/1-propanol/water mixture. Fluid Phase Equilibria 234, 84–93.

ESTD No. 19

DETERMINATION OF THE FUEL OIL (BUNKER) PROPERTIES OF 1,2 BENZENEDICARBOXYLIC ACID, DIISOOCTYL ESTER FROM THE ETHYL ACETATE EXTRACT OF *Euphorbia tirucalli L.* (POBRENG KAHOY)

Carolyn

Lorna T. Enerva, Jhoann M. Sapa and Caroilina L. Uy

College of Science and SciTech R and D Center
Polytechnic University of the Philippines
Sta. Mesa, Manila
Email: lornaenerva@yahoo.com

This study was conducted to determine the bunker fuel properties of *pobreng-kahoy* crude extract using the standard test methods used in bunker fuel oil and identify the presence of 1,2 benzenedicarboxylic acid, diisooctyl ester by instrumentation analysis.

Ground stems of *pobreng kahoy* were percolated with ethyl acetate as solvent and the extract was concentrated in vacuum using a rotary evaporator. The crude extract was purified using column chromatography. Fractions A_6 to A_7 were recolumned and fractions B_2 and B_3analyzed using gas chromatography-mass spectrometry. The gc-mass spectrum of the compound showed the presence of 1,2 benzenedicarboxylic acid, diisooctyl ester in the retention time of 24.40 and 24.40 and its mass spectrum showed the fragmentation peaks m/z at 149 as its base peak and m/z at 279 as its molecular ion peaks of the said compound. Fractions A_8 to A_9 were used for the infrared and ultraviolet spectroscopy. The infrared spectrum showed C-H stretch, C=O stretch, C-C stretch, aromatic ring and C-O as the functional groups present. The ultraviolet spectrum gave a ε_{max} at 283 nm.

The *pobreng kahoy* crude extract gave 9,928 BTU/lb for gross heating value, specific gravity of 0.927, 0.03% sulfur, kinematic viscosity at 40°C. These values were compared to the standard limits for the said fuel. The study of the bunker fuel oil properties of *pobreng kahoy* to determine an acceptable fuel-source will be of great help to lessen the economic problem regarding the increasing crude oil demand of the country.

Keywords: fuel, bunker fuel, gross heating value, kinematic viscosity, flash point

ESTD No. 20

NON-CHLOROFLUOROCARBON (CFC) REFRIGERANT HEAT PUMP DRYER

[1]Samuel S. Franco, [1]Lorceli B. Taclan, [2] Robeo B. Gavino, [2]Francisco D. Cuaresma, and [2]Teresito G. Aguinaldo

[1]Mariano Marcos State University
Batac, 2906, Ilocos Norte
[2]Central Luzon State University
Science City of Muñoz, Nueva Ecija
Email: ssfranco@digitelone.com, samuelsfranco@yahoo.com

Drying is one of the major aspects of agricultural whether at the farm or in the industrial level. It is an energy intensive operation and it is estimated to be between 20 to 30% of the total energy required before the product reaches the consumers. A heat pump dryer was developed to ensure a relatively clean drying atmosphere for fruits and vegetables. The heat pump is an air-conditioning machine using non-chlorofluorocarbons (non-CFC) as refrigerant that extracts heat from one environment and discharge it to the other. It will collect heat from the condenser and discharges it to the air handler and with the help of the expansion valve, the flow of the refrigerant moves in the opposite direction and heat is extracted from the air handler and discharged to the drying chamber.

The heat pump was observed to have a thermal efficiency of 97% and dehumidifying efficiency of 67%. It can be used to dry mango, pine apple, papaya, banana, onions and other fruits and vegetables.

The heat pump has the advantage of drying agricultural products and at the same time cooling perishable fruits and vegetables.

The heat pump can be manufactured in modules or designed and installed on-site depending upon the drying requirements.

Keywords: dryer, heat pump, Drying, refrigerant, non-chlorofluorocarbon, fruits, vegetables

ESTD No. 21

ONLINE MEDICAL TRAINING SYSTEM FOR SIMULATED CATARACT SURGERY

Richard Bryann L. Chua and Pamela Rica M. Florentino*

DPSM, Rm. 102, Rizal Hall, CAS,
University of the Philippines Manila
Padre Faura St., Ermita Manila, 1000
Email: rbchua@mail.upm.edu.ph

Majority of the eyes used in the laboratory for the training of medical students for cataract surgery often lacks the diseased state necessary for surgical training. As such, the Online Medical Training System for Simulated Cataract Surgery was developed to provide training for students on cataract surgery. The system was developed using Java and Java 3D API. The surgical procedure training provided by this system includes incision in the junction of clear and white outer parts of the eye, suction of the damaged lens, and insertion of an intraocular lens (I.O.L.) in place of the damaged lens. Aside from the surgical procedures training provided, it also evaluates and grades the performance of the medical student in cataract surgery. This system will be able to help the medical student to have a more realistic look at a cataract eye for their cataract surgery training and will allow the students to undergo cataract surgery for a number of times without using a lot of eyes from the eye bank.

Keywords: medical training system, simulation, eye surgery, cataract

ESTD No. 22

SURVEILLANCE SYSTEM OF HOSPITAL-ACQUIRED INFECTION IN THE PHILIPPINES

Avegail D. Carpio, Edwin Joseph D. Ortile*

Department of Physical Sciences and Mathematics
College of Arts and Sciences, University of the Philippines Manila,
Padre Faura, Ermita Metro Manila
Email: avegail.carpio@up.edu.ph; ave_carpio@yahoo.com

The Surveillance of Hospital-Acquired Infection in the Philippines (SHIP) was established in 1999 with an aim to create a national database of nosocomial infections and improve surveillance methods of hospitals in the Philippines.

Nosocomial or hospital-acquired infections are defined as those that were neither present nor incubating at the time the patient came for care.

The Surveillance System of Hospital-Acquired Infection in the Philippines (SSHIP) is a web-based computer program that would aid infection control doctors and nurses on the data collection and retrieval of important nosocomial infection information. SSHIP has the capability to allow infection control nurses to update the Infection Worksheet, Laboratory Data Form and the ICU Monthly Report Form; and to permit infection control doctors and nurses to generate rates and graphs of the ICU Monthly Report Form. It also has the interface to permit online users to submit online SHIP Application Forms, and sent via email of notification of application to Philippine hospitals who submitted their online SHIP Application Forms.

Keywords: surveillance system, hospital-acquired infections in the Philippines, nosocomial infections in the Philippines, SHIP, Philippine surveillance system

ESTD No. 24

PEDIGREE AND CANINE MANAGEMENT SYSTEM

Avegail D. Carpio, Johanne C. Pirante*

Department of Phusical Sciences and Mathematics
College of Arts and Sciences
University of the Philippines Manila
Padre Faura, Ermita Metro Manila
Email: avegail.carpio@up.edu.ph; ave_carpio@yahoo.com

The Pedigree and Canine Management System (PCMS) manages the data used by the Philippine Canine Club (PCCI) and generates pedigree of 3rd to 5th generation of each registered dog. PCCI was formed for the protection and advancement of pure-bred dogs. PCCI implements standardized regulations administering dog competitions and field trials; safeguard its members from fraud information; and to further improve the study, breeding, exhibiting, administration and maintenance of the breed veracity of pure-bred dogs.

PCMS has a user-friendly interface to let the PCCI members view requested dog information, search and submit dog activities and advertisement and view summary reports on dog mortality. Additionally, the system is capable of notifying the member with dog activities through short messaging services (SMS). Furthermore, the system is able to allow online guests to view summary reports on dog mortality, dog advertisements and post dog competition information. Lastly, a

module was created for the PCCI administrator to manage the system with his/her ability to update dog information and PCMS user's profile.

Keywords: *dogs, PCCI, pedigree, dog information management system

ESTD No. 25

TRAUMAP: A GEOGRAPHIC INFORMATION SYSTEM

Aldrich Colin K. Co, Gregorio Baes*

Department of Physical Sciences and Mathematics
College of Arts and Sciences
University of the Philippines Manila
P. Faura St., Ermita, Manila.
Telefax: 5265858
Email: aldrich.co@gmail.com, gbaes@yahoo.com

Traumap is a decision-support tool that allows medical practitioners to graphically track trauma-related cases by showing them reported occurrences alongside a scalable (zoomable) view of the cityscape, which contains street-level detail.

Traumap is a Geographical Information System (GIS) based on the Scalable Vector Graphics standard. The system allows users to report and view trauma cases directly on the maps through a point-and-click interface. The following features were implemented: entering, editing, and querying information, generating reports with regard to the cases, locating streets in the map using regular-expression search, and additional controls to easily navigate the vector map.

Keywords: geographic information systems, Scalable Vector Graphics (SVG),decision support systems, trauma

ESTD No. 26

ONLINE TRAINING SYSTEM FOR PIT AND FISSURE DENTAL CARIES

James Cecilio B. Tuano*, Vincent Peter Magboo**

Department of Physical Sciences and Mathematics
College of Arts and Sciences
University of the Philippines Manila
Padre Faura, Ermita, Manila
Email: jamestuano@yahoo.com*;petermagboo@yahoo.com**

The Online Training System for Pit and Fissure Dental Caries (OTSPFDC) is a web-based application that allows student/users to view a variety of learning materials. They are also allowed to assess their knowledge online through the system's evaluation tools.

Through OTSPFDC, student/users are able to view online lectures about pit and fissure dental caries in plain text or in different file formats such as word documents or powerpoint files. They are also able to view animations about amalgam dental restoration for pit and fissure dental caries. The student/users are presented with step-by-step procedure on amalgam restoration. Objective type quizzes give student/users a good way to test their knowledge about the online lectures on pit and fissures. A special feature of this system is the simulation exam on amalgam restoration for pit and fissure dental caries. The simulation exam offers the student/user a more challenging test as they are administered via 'hands-on' practical exam type. Additionally, error messages regarding the hands-on exam are displayed whenever a user makes a mistake. The dental expert acting as system administrator modifies the database entries enabling him to update necessary lecture files and lessons. He is allowed to add, edit or delete lectures, files and objective-type quizzes.

OTSPFDC is a tool that addresses the need to reinforce the objective of dentistry – which is to restore teeth as much as possible. This tool helps students to further improve their knowledge regarding the topic and provides them with lessons in restorative techniques which can be repeatedly viewed for better understanding.

Keywords: training system, pit and fissure dental caries, simulation

ESTD No. 27

THE MAKINGS OF AN INTERNET-BASED RICE INFORMATION SERVICE (IRIS): PILOTING IN THE PHILIPPINES

Cheryll B. Casiwan[1]*, Suan Peng Kam[2], Holecz, Francesco.[3] Eric van Valkengoed[4], Massimo Barbieri[3], Sonia L. Asilo[1], Larry A. Santos[1], Rowena G Manalili[1], Wilfredo B. Collado[1], Sherwin A. Adriano[1] and Aileen Maunahan[2]

[1]Philippine Rice Research Institute
Munoz, Nueva Ecija, Philippines
[2]International Rice Research Institute, DAPO Box 7777
Metro Manila,
[3]sarmap s.a. Cascine di Barico
CH 6989 Purasca, Switzerland
[4]Synoptics Remote Sensing & GIS Applications, Wageningen, The Netherlands
*Email: cbcasiwan@philrice.gov.ph

A collaborative activity involving a private consortium, an international and a national rice research institution was conducted to validate the design of an internet-based information system that would provide more timely and objective data on rice area and production. This system consists of two components that make use of geospatial tools including remote sensing, GIS and GPS technologies. The remote sensing component comprises a largely automated protocol using multi-date SAR imagery for mapping and estimating rice area and planting dates. These outputs are fed into a production estimation component comprising a crop growth model that predicts harvest dates and crop yield using meteorological data. Rice area and yield estimates are summarized by administrative boundaries and are offered through a web-based service to subscribers.

Pilot testing of the data generation component of the system was carried out in 81 municipalities covering Nueva Ecija, Isabela and Pangasinan for the dry season rice crop of 2003"2004. Acquisition of RADARSAT and ENVISAT ASAR data at various dates throughout the rice-growing season permitted evaluation of the capacity of the automated SAR processing system to handle multi-platform data. Investigations were made on the minimum number of acquisition dates that would provide reliable rice area estimates. A field campaign was conducted covering 68 municipalities and 667 geo-referenced survey points to collect ground truth information. Daily weather data were collected from 5 surrounding weather stations for at least the past 10 years; data for the 2003"2004 season were available for 3 stations. Processing of the SAR data provided rice area and planting date estimates, which were fed together with the weather data into the crop model. The predicted yields and rice area are reported by municipality. The implications of operationalizing

such an information system for rice is discussed based on the experience from this pilot study.

Keywords: IRIS, rice monitoring, crop growth model, rice area mapping

ESTD No. 28

EVALUATION OF PARTIALLY PURIFIED PROTEIN FROM MALUNGGAY (*Moringa oleifera*) SEED EXTRACT AS A CATIONIC POLYMERIC FLOCCULANT FOR THE DECOLORIZATION OF DISTILLERY BIODIGESTER EFFLUENT

Billy Ray P. Matanguihan[2], Catalino G. Alfafara[2], Ronald R. Navarro[3], and Fidel Rey P. Nayve Jr.[1*]

[1]National Institute of Molecular Biology and Biotechnology (BIOTECH),
[2]Department of Chemical Engineering,
College of Engineering and Agro-Industrial Technology,
University of the Philippines Los Baños, College, Laguna 4031, Philippines
[3] *c/o* Institute of Applied Biochemistry, University of Tsukuba, Tsukuba, Japan
Email: linojp@yahoo.com

The ability of partially purified protein from malunggay (*Moringa oleifera*) seed extract as a cationic polymeric flocculant for the decolorization of distillery biodigester effluent was experimentally evaluated. Screening and selection of the most appropriate partial protein purification method (ammonium sulfate precipitation, ethanol precipitation, and acetone precipitation) based on the amount of protein precipitated, cost of salt/solvent used per gram of protein precipitated, and percent decolorization was also done. The optimum initial pH of the distillery effluent and optimum dosage of malunggay protein for decolorization were determined. Ammonium sulfate precipitation was found to be the most appropriate partial protein purification method in terms of protein recovery (34.7%), cost (Php 1.11 per gram protein precipitated), and decolorization efficiency (81.6%). Addition of partially purified malunggay protein solution in the distillery effluent resulted in a dramatic change in color of the effluent along with the formation of brownish flocs. At the optimum pH of the effluent (pH 8), 88.2% decolorization was achieved at an optimum dosage of 2.707 mg/ml malunggay protein. The addition of EPS improved the ability of the flocs to settle.

The color removal efficiency of partially purified proteins from malunggay seed extract was found to be similar to crude malunggay seed extract. The main advantage of using the former flocculant was the reduction in the COD concentration of the treated effluents by as much as 40% (relative to the COD

concentration of the treated effluents using crude MOSE). However, further treatment may be necessary to reduce the COD of the treated decolorized effluents to conform with DENR effluent standards.

Keywords: *Moringa oleifera*, *Rhizobium*, exopolysaccharide, color removal, flocculation

ESTD No. 29

PARTIALLY PURIFIED PROTEIN FROM MALUNGGAY (*Moringa oleifera*) SEED EXTRACT FOR USE AS SECONDARY CATIONIC POLYMERIC FLOCCULANT FOR COPPER REMOVAL FROM WASTEWATER EFFLUENTS

Danvir Mark dlC Farnazo[2], Catalino G. Alfafara[2]
Ronald R. Navarro[3] and Fidel Rey P. Nayve Jr.[1*]

[1]National Institute of Molecular Biology and Biotechnology (BIOTECH),
[2]Department of Chemical Engineering,
College of Engineering and Agro-Industrial Technology
University of the Philippines Los Baños
College, Laguna 4031, Philippines
[3] *c/o* Institute of Applied Biochemistry, University of Tsukuba, Tsukuba, Japan
Email: linojp@yahoo.com

The efficiency of the partially isolated/purified *Moringa oleifera* seed extract as a secondary cationic polymeric flocculant to *Rhizobium sp.* BJVr12 EPS for copper removal in simulated wastewater was investigated. Selection of the best purification process by precipitation (ammonium sulfate precipitation, acetone precipitation, and ethanol precipitation) in terms of protein yield, cost of precipitant and percent copper removal was also done. The role of pH and the mass ratio of the EPS to the protein isolates were determined. Moreover, evaluation of the capacity of the EPS-partially isolated/purified *Moringa oleifera* seed extracts flocculation system and the recovery of the copper from the flocs formed after treatment were made. Ammonium sulfate precipitation was taken as the recommended protein purification method with a protein yield of 8.15 mg/ml, a raw material cost of Php 0.24/ g protein produced, and had the highest percent mean percent copper removal among the three methods. At the optimum workable pH of 6, the percent copper removal had a mean average of 90.86% along with the formation of firm flocs, clear and easily filterable supernatant after each treatment. Optimum EPS-ammonium sulfate protein isolates mass ratio was at 0.767 with a mean copper removal efficiency of 93.30%. Decreasing copper removal capacity of

the EPS-partially isolated/purified *Moringa oleifera* flocculation system was observed with increasing amounts of EPS and ammonium sulfate protein isolates employed during treatment.

The EPS-*Moringa oleifera* flocculant system which utilized the $(NH_4)_2SO_4$ precipitated seed extract performed better than the crude (unprecipitated) one. Copper removal efficiency was 88.7% or a final copper concentration of 1.10 ppm which approximates the allowable copper concentration in wastewater effluents set by the DAO 35. COD was reduced by 75.8 %(relative to the COD of effluents treated using EPS-crude *Moringa oleifera* seed extract flocculation system).

Keywords: *Moringa oleifera*, *Rhizobium* exopolysaccharide, protein purification, copper removal, flocculation

ESTD No. 30

EVALUATION OF THERMO-TOLERANT YEAST ISOLATES FOR ETHANOL PRODUCTION IN FLASK AND BIOREACTOR FERMENTATIONS.

Ian Anthony dC Amon[2], Catalino G. Alfafara[2], Ronald R. Navarro[3] and Fidel Rey P. Nayve Jr.[1*]

[1]National Institute of Molecular Biology and Biotechnology (BIOTECH),
[2]Department of Chemical Engineering
College of Engineering and Agro-Industrial Technology
University of the Philippines Los Baños, College, Laguna 4031, Philippines
[3] *c/o* Institute of Applied Biochemistry, University of Tsukuba, Tsukuba, Japan
Email: linojp@yahoo.com

Ten yeast strains which were isolated locally from various sites around the Philippines were screened for their ability to ferment sugarcane molasses at high temperatures. From among the strains used, the most promising strain was selected and was used in the bioreactor experiments.

All strains were found capable of fermentation up to 50 °C although no apparent cell growth was seen at the end of 24 hr at 50 °C; only six strains were able to remain viable up to 47 °C. The maximum ethanol concentration for all strains was reached at 45°C for which five strains produced about 5% (v/v) or more. Based on the amount of ethanol produced as well as on cell viability, strain 2015 was selected as the best strain from among the strains used. The performance of the selected strain was studied in a worst-case scenario of fermentation under uncontrolled as well as under controlled (30 °C) temperature condition. This strain was able to produce a maximum ethanol concentration of 8.73% (v/v) after 20 hr under

uncontrolled temperature condition and 9.83% (v/v) after 31 hr under controlled temperature condition.

Strain 2015 was found to be comparable with other strains used in industries which under controlled temperature conditions, usually yield about 8 to 10% (v/v) ethanol. Furthermore, fermentation under uncontrolled temperature condition using this strain was comparable with fermentation under controlled (30 °C) temperature condition. Thus, strain 2015 has a potential for use in industries having the advantage of maintaining high ethanol production while decreasing energy costs for cooling.

Keywords: thermo-tolerant, yeast, ethanol, bioreactor

ESTD No. 31

DESIGN, FABRICATION AND TESTING OF A BENCH-SCALE AND CONTINUOUS FLOCCULATION SYSTEM USING A MICROBIAL EXOPOLYSACCHARIDE (EPS) BIOPOLYMER AND MALUNGGAY (*Moringa oleifera*) SEED EXTRACT FOR TREATMENT OF COPPER CONTAINING WASTEWATER

Clarito B. Geron, Jr.[1]*, Fidel Rey Nayve, Jr.[2], Catalino G. Alfafara[1], and Ronald R. Navarro[1]

[1]Department of Chemical Engineering
[2]National Institute of Molecular Biology and Biotechnology (BIOTECH)
University of the Philippines Los Baños, College, Laguna 4031
Email: linojp@yahoo.com

A continuous bench scale flocculation system for the treatment of copper containing wastewater was designed, fabricated and tested. The flocculation system involved the use of an exopolysaccharide (EPS) as primary agent for copper sequestration, and Malunggay (*Moringa oleifera*) (MO) seed extract as secondary flocculant to separate the copper-EPS complex from the solution. Jar test experiments were also done to obtain a workable velocity gradient (G) to be used for design calculations of the flocculator.

Laboratory flocculation experiments to optimize the pH and flocculant dose conditions yielded the following results: an optimum initial pH of 5, a 0.01 ml EPS solution/mL of synthetic copper wastewater, and a ratio of 1.65 g EPS/g protein in the MO seed extract. Jar test experiments using these conditions obtained a workable velocity gradient (G) of 63.5/s which was used as an scale up factor of the setup.

The above process information was used to design, fabricate and test a continuous bench scale flocculation system consisting of a rapid mixing tank, flocculator and settler. At steady state, a copper removal efficiency of 80% was obtained, with an effluent copper content approaching the DENR effluent standard of 1 ppm. Although the performance was considered satisfactory, further improvements (such as optimization of the EPS and MO seed extract flowrates) were recommended to increase copper removal.

Keywords: flocculation, copper wastewater, exopolysaccharide (EPS), Malunggay

ESTD No. 32

ELECTROLYTIC DESTRUCTION OF AZO-DISPERSE DYE IN A MODEL SYSTEM

Rommel B. Santos[1], Catalino Alfafara[1*], Veronica Migo[2], Jovita Movillon[1] Elson R. Montibon[1], and Masatoshi Matsumura[3]

[1] Department of Chemical Engineering, University of the Philippines Los Banos, College, Laguna 4031
[2] National Institute of Molecular Biology and Biotechnology (BIOTECH) University of the Philippines Los Banos, College, Laguna 4031
[3] Institute of Applied Biochemistry, University of Tsukuba, Tennodai 1-1-1 Tsukuba City 305-0006, Japan
Email: linojp@yahoo.com; Email: acer1@laguna.net

Decolorization, through electrolytic oxidation of azo-disperse dye (C.I. Disperse Red 1) was evaluated in a model-system-wastewater simulating the dye concentrations in textile effluent, and supplemented with sodium chloride. The electro-oxidation system involved the use of chlorine-based oxidants generated by the electrochemical oxidation of chloride ions in the wastewater. The objective was to obtain the best conditions of electrolyte (sodium chloride) supplementation and charge dose which would give the highest decolorization efficiency. The charge dose (in the units of coulombs/unit amount of pollutant removed) is an empirical factor which could be useful in the scale-up of an electrochemical process for wastewater treatment. Nuclear magnetic resonance (NMR) spectroscopy was also used to check molecular changes in the dye after electrolytic treatment.

Results showed that the initial rate of decolorization increased with operating current, up to a saturation value at constant level of NaCl. The same phenomenon was also observed when NaCl supplementation was increased at constant operating current. The saturation value was attributed to the limitation of oxidant formation imposed by the operating current or concentration of chloride ions. The following

optimum electrolytic conditions were obtained from experiments: a charge dose of 714.6 coulombs/Abs.L color removed, with a sodium chloride supplementation of 2 g/L. The maximum decolorization efficiency was 83%. The cost of electrical energy for electrolysis was computed to be PhP 14.76/m3 of 200 ppm C.I. Disperse Red 1. Significant changes in the specific signals in NMR spectra also confirmed partial oxidation of the dye molecule during electrolysis.

Keywords: electrooxidation, electrolysis, azo dye, charge dose

ESTD No. 33

XRD AND SEM CHARACTERIZATION OF SINTERED CORDIERITE PREPARED FROM MIXTURES OF KAOLIN CLAY, TALC AND OTHER ADDITIVES

DIONESIO C. PONDOC
Department of Ceramic Engineering
College of Engineering
Mariano Marcos State University, Batac, 2906 Ilocos Norte
Tel.: +63 77 792 2573 Fax: +63 77 792 2633
Email: dondon_pondoc@yahoo.com

Stoichiometric composition of cordierite was prepared from the mixtures of kaolin clay, talc powder, and magnesium carbonate with a percentage composition of 72.06, 13.23, and 14.71 respectively via solid state sintering reaction. Test specimens in the form of pelletized circular disk of 20 mm diameter by 2.5 mm thickness were prepared from the mixtures by powder pressing in a metal mold using hydraulic press with a pressing load of 80-100 MPa. The sintering process was conducted using Linberg Blue M box furnace with four segment temperature control program. The sintering time was set to 2 and 3 hours respectively within the sintering temperature range of 1000°C-1100°C. The sintered cordierite test specimens were characterized using Philips PW 1830 x-ray diffractometer to determine the formation of crystalline phases and Leica S440 scanning electron microscope to determine the morphology and microstructures.

X-ray analysis revealed that at 1000°C sintering temperature within the sintering time of 2 and 3 hours, there were two types of cordierite phase that can be identified in the test specimen at almost equal amount based from the relative intensity of the x-ray diffraction pattern. The crystalline phases were identified as μ-cordierite and Ü-cordierite. At 1100°C sintering temperature with the same sintering time, it was noticed that the formation of Ü-cordierite was increasing while μ-cordierite phase was decreasing. This phenomenon was due to the polymorphic transformation reaction of μ-cordierite starting at 1050°C as described from earlier

study in the formation of cordierite.

SEM observation and analysis of the micrographs taken at magnifications of 1,000X, 5,000X, 10,000X and 15,000X of the test specimen sintered at 1000°C for 2 and 3 hours revealed a microstructures consisting mostly of euhedral and subhedral particles with sizes approximately in the range of 1-10 μm. As the sintering temperature increases to 1100°C of the same sintering time, the micrographs revealed a microstructures consisting of partially melted euhedral grain particles and was started to transform into spherical shape. Open pores between particles could also be observed in the micrographs. The presence of pores between particles can be concluded that the sintering reaction was not completely achieved at the sintering temperature range of 1000-1100°C and the individual particles did not formed a dense body.

Keywords: solid-state sintering, x-ray analysis, scanning electron microscopy, crystalline phases, and microstructures

HEALTH SCIENCES

HSD No. 1

MICROBIOLOGICAL ASSESSMENT OF DRINKING WATER QUALITY OF SELECTED BARANGAYS IN ILIGAN CITY

Peter D. Blanco* and Lucilyn D. Lahoylahoy

Department of Biological Sciences, College of Science and Mathematics
Mindanao State University-Iligan Institute of Technology , Iligan City
Email: petdel2orb@yahoo.com

Microbiological drinking water quality using Most Probable Number (MPN) Method and Heterotrophic Plate Count (HPC) was assessed from two water sources in each of three selected barangays of Iligan City from April to June 2005. Two pumping stations distribute treated water to these barangays: Pumping Station 1 for Barangays Palao and Hinaplanon, and Pumping Station 2 for Barangay San Miguel with a water source from a farther and higher elevation.

MPN test showed that 28% of 108 water samples yielded total coliforms and only 10% were confirmed to have faecal coliform. Abrupt increase in MPN coliform was observed in Sites A and B from Palao on the second of six samplings, and stresses the unpredictability of possible faecal contamination which might pose a potential health risk. Water samples from Sites C and D of San Miguel displayed irregular and varying MPN coliform levels which can be attributed to a probable

breach in the piping system since the source is farther from the point of use and may have resulted to an increase in assimilable organic carbon. Sites E and F in Hinaplanon yielded water samples which consistently showed very minimal levels of coliform thus are considered to be microbiologically safe to drink.

Coliforms are not the only potentially pathogenic bacteria which may be isolated from drinking water. HPC is mainly used as an operational tool to screen the efficacy of disinfection procedures. International standards for acceptable HPC for microbiologically safe drinking water vary from less than 100 to 500 cfu/ml. Fifty-nine of the samples exhibited values greater than 100 cfu/ml, and thirty-five of them exceeded 500 cfu/ml. Only forty-nine of the total collected water samples had less than 100 cfu/ml. HPC levels exceeding acceptable standards were obtained from San Miguel as the farthest from the water source, whereas Hinaplanon holds the lowest number of colony counts .

Keywords: Most Probable Number (MPN), Heterotrophic Plate Count (HPC), faecal coliform

HSD No. 2

HEALTH STATUS OF SELECTED FACULTY AND STUDENTS OF ILIGAN CITY ASSESSED THROUGH "THE 20-MINUTE HEALTH CHECK" SCREENING PROGRAM

Cesar G. Demayo, Mark Anthony J. Torres, Sydney Corpin, Crystelle Capa and Corilyn C. Vena

Department of Biolological Sciences
College of Science and Mathematics,
Mindanao State University-Iligan Institute of Technology
Iligan City
Email: c_gdemayo@yahoo.com

A comprehensive screening program known as "The Twenty Minute Health Check" designed to test every aspect of one's lifestyle—from eating habits to susceptibility to stress developed by Dr. Vernon Coleman in 2004 was used in evaluating the health status of the members of the faculty and students of MSU-IIT, Iligan City. In this survey, one is required to answer the 120 questions where points will be collected for the answers given and the total points received for the 120 questions will give one an automatic health check. Every score has a corresponding health status – excellent health, heart, respiratory, digestive, joints, stress, alcohol problems and predisposition to cancer. A separate survey on monozygotic and dizygotic twins was also done to evaluate possible concordance of the scores.

A total of six hundred (600) students, two-hundred ninety-four (294) members of the faculty and staff, sixty (60) twins were the respondents of the study. Results of the study showed similar patterns in both faculty and student respondents. Both groups have similar results for scores under the very unacceptable and unacceptable range and these were high for heart, digestive and stress problems and relatively low for joint, respiratory, cancer and alcoholism. The results on twins (pooled) showed scores of the same range with the faculty and students and concordance patterns cannot be established as yet as the number of twin pairs who participated in the research are still very few. Nevertheless, it is interesting to note in this study that both faculty and students alike have similar patterns in terms of susceptibility to certain health problems. An on-going study is being done to evaluate the health status of those individuals working in other schools and those not working in the academic community to see what particular health problems are common to these groups.

Keywords: health status, screening

HSD No. 3

PREVALENCE AND SUSCEPTIBILITY PROFILES OF COMMUNITY-ACQUIRED METHICILLIN-RESISTANT *Staphylococcus aureus* ISOLATES FROM TERTIARY STUDENTS OF ILIGAN CITY

Lucilyn D. Lahoylahoy* and Primrose C. Makinano

Department of Biological Sciences,
College of Science and Mathematics
Mindanao State University-Iligan Institute of Technology
Iligan City
E-mail: luzlin@yahoo.com

Methicillin-resistant *Staphylococcus aureus* (MRSA) is a pathogen that has been recovered from and associated with hospitals and health care facilities. However, there has been increasing number of studies which has indicated that MRSA is prevalent in the community, thus, making community-acquired methicillin-resistant *Staphylococcus aureus* (CA-MRSA) an emerging threat worldwide. This community-based study was undertaken to assess the prevalence of CA-MRSA among healthy college students with no apparent risk factors for MRSA.

Sixty-six methicillin-resistant *Staphylococcus aureus* isolates were detected from the facial swab cultures of 116 individuals included in the study. The *S. aureus* isolates were identified by traditional methods: gram positive staphylococci, mannitol fermentor, catalase positive, coagulase positive and oxacillin-resistant. Stan-

dard disc diffusion method as proposed by the Clinical Laboratory Standards Institute (CLSI) was employed to determine the susceptibility profiles of the MRSA isolates to clindamycin, erythromycin and tetracycline. Seventeen percent of the MRSA isolates (11/66) were found to be resistant to erythromycin, 14% (9/66) were resistant to clindamycin and 5% (3/66) were resistant to tetracycline. Only one isolate exhibited resistance to all three antibiotics. The susceptibility profiles of the CA-MRSA are consistent with the observation that CA-MRSA strains are more susceptible to antibiotics than hospital-acquired MRSA strains. The detection of significant number of CA-MRSA isolates supports the postulation that healthy individuals of the society are the highest reservoir of antimicrobial resistant bacteria.

Keywords: *Staphylococcus aureus*, *Antibiotic* Resistance, Community-Acquired Methicillin-Resistant *Staphylococcus aureus* (CA-MRSA), Antibiotic Disc Diffusion Method (Kirby-Bauer)

HSD No. 4

MULTI-DRUG RESISTANT *Staphylococcus aureus* FROM WASTE PICKERS OF ILIGAN CITY

Lucilyn D. Lahoylahoy, Ermee Gild S. Calumba and Leonell Albert L. Quitos*

Department of Biological Sciences, College of Science and Mathematics
Mindanao State University-Iligan Institute of Technology , Iligan City
E-mail: leonell_730@yahoo.com

An estimated twenty-five percent of the Philippine population is urban poor who reside in slums near open garbage dumps. Majority of these individuals resort to waste-picking as means of livelihood often ignoring the significant occupational risk of exposure to hospital wastes. One possible origin of community-acquired multidrug-resistant *Staphylococcus aureus* (CA-MDRSA) are hospital strains that may be carried into the community where then they are spread from person to person. This study sought to investigate the prevalence of MDRSA among local waste pickers.

Forty-six *S. aureus* isolates were detected from one hundred seven waste-pickers of Iligan City. Susceptibility profiles of all *S. aureus* isolates to antimicrobial agents were determined and interpreted according to Clinical Laboratory Standards Institute (formerly NCCLS) published guidelines. The resistance of the isolates were 100%, 44%, 9% and 7% to oxacillin, ampicillin, clindamycin and tetracycline, respectively. The results suggest that all isolates are methicillin-resistant *S. aureus* (MRSA) and supports the increasing reports of detecting CA-MRSA.

However, the CA-MRSA isolates from the respondents could not be categorized as true CA-MRSA due to the possibility that it might have originated from the individuals' direct contact with infected hospital wastes during scavenging. Hospital-acquired MRSA are different from CA-MRSA in the sense that these isolates are multi-drug resistant.

Fifty percent of the CA-MRSA from the waste-pickers exhibited multi-drug resistance (MDR) against two to three (oxacillin, clindamycin and ampicillin) of the four chosen antibiotics and are considered MDR-MRSA. There is then a high possibility that the original MRSA isolate of this group may have come from hospitals or health-care facilities. However, the other 50% of the MRSA isolates are non-multidrug resistant (NMDR-MRSA) which exhibited the characteristic trait of a true CA-MRSA which is still susceptible to most antibiotics. The detection of significant numbers of MRSA and MDRSA is of great concern since these individuals are asymptomatic and may further spread these resistant strains in the community.

Keywords: Waste pickers, *Staphylococcus aureus*, Methicillin-resistant *S. aureus* (MRSA), Community-Acquired MRSA (CA-MRSA), Multi-drug Resistant MRSA

HSD No. 5

THE INCIDENCE OF METHICILLIN-RESISTANT *Staphylococcus aureus* (MRSA) IN BATANGAS REGIONAL HOSPITAL (BRH)

Carina R. Magbojos

College of Medical Technology, Lyceum of Batangas
Email: karen_rm2002@yahoo.com

Methicillin-resistant *Staphylococcus aureus* (MRSA) is one of the major concerns in the healthcare system. The rise in MRSA infections was associated with epidemics in large teaching hospitals and in general hospitals. Scientific studies have shown the increasing incidence and the persistence of MRSA cases in different hospitals in our country. This study was conducted to determine the incidence of Methicillin-Resistant *Staphylococcus aureus* (MRSA) in Batangas Regional Hospital (BRH) where the identification of MRSA is not performed. From November 2003 to July 2004, four hundred one (401) *S. aureus* clinical isolates were identified based on morphological and biochemical characteristics. Using the NCCLS oxacillin-salt agar screening test and agar dilution method, 32.17% (129/401) of the isolates were confirmed to be MRSA in which the minimum inhibitory concentration (MIC) is 8 mg/mL up to >64 mg/mL. According to the demographic and clinical characteristics of patients with *Staphylococcus aureus* infections and MRSA infections, many of the patients were newborns and children below 2 years old;

admitted in Pediatrics and Neonatal Intensive Care Unit (NICU); have stayed long in the hospital; and have recovered well from the infections. Half of the patients have one or more co-morbidities. The *S. aureus* and MRSA infections were acquired in the hospital but most infections were acquired from the community. The length of stay of the patients in BRH was found to be correlated with the spread of MRSA; however, the age, sex location of confinement and underlying conditions of the patients failed to correlate with the spread of MRSA in BRH due to insufficient number of MRSA isolates (129/401). Most of the clinical isolates were obtained from the blood specimens.

Keywords: Methicillin-resistant *Staphylococcus aureus*, MRSA, Incidence, MIC

HSD No. 6

ASTHMATIC CHILDREN HOME MONITORING SYSTEM

Vincent Peter C. Magboo ,* Cheza Marie R. Vianzon

Dept. of Physical Sciences and Mathematics, College of Arts and Sciences
University of the Philippines Manila
Padre Faura, Ermita, Manila
E-mail: petermagboo@yahoo.com

In the Philippines, asthma affects over six million children and is ranked 32nd in "self-reported asthma" in the world. Recent studies have shown that monitoring of asthma severity at home combined with adequate patient education can reduce the incidence of asthma exacerbation and subsequent hospitalization. Asthmatic Children Home Monitoring System (ASCHOMS) is created with the objective of allowing parent or guardian to manage his child's asthma at home. It is a web-based application which allows physicians to maintain the medical records of their asthmatic patients, create an action or treatment plan for the asthma self-management of the patient. It also allows patient's guardian or parent to monitor and to inform the physician online or via short messaging system (SMS) the clinical symptoms, spirometry and peak flow measurements. In return, the physician can assess the condition of the patient and create an appropriate self-treatment plan for the patient. Interaction among active valid users of the system can be done through online messaging and/or SMS. Additionally, different asthma related information, activities, services and organizations are likewise maintained in the system for information dissemination about asthma. There is also a report generation on asthma prevalence according to asthma type and other clinical parameters within Metro Manila. An automated asthma home monitoring is beneficial to over six million Filipino asthmatic children. With adequate guidance, self-management can provide

proper health care and a way to minimize medication cost but maximize health progress without the physical supervision of the primary physician.

Keywords: asthma, home monitoring, health information system, short messaging system

HSD No. 7

ONLINE HEARING LOSS SCREENING SYSTEM

Vincent Peter C. Magboo,* Michael Vincent A. Capulong

Dept. of Physical Sciences and Mathematics
College of Arts and Sciences University of the Philippines Manila
Padre Faura, Ermita, Manila
E-mail: petermagboo@yahoo.com

Hearing loss is a relevant problem in the Philippines as 20 % of Filipino children and more than half of the population have serious hearing problems. Due to the increasing noise pollution in our environment, early detection of hearing loss is recommended as the effects of hearing loss are often recognized in the advanced stage of hearing loss. The Online Hearing Loss Screening System (OHLSS) is created with the objective of allowing users to take tests for hearing loss detection as well as obtain information on loss of hearing. The screening tests used and included in the system are the hearing loss questionnaire, tone audiometry test and speech audiometry test. The system begins with instruction on the proper calibration of the sound-related hardware such as sound cards and earphone and the software configuration (volume control) An electronic questionnaire is to be answered followed by tone audiometry which test each ear for sensitivity to normal sound levels. The tone audiometry plays three tones at 20 db HL (for pre-school and school-age users) and 25 db HL (for adults) in 1000 Hz, 2000 Hz, and 4000 Hz and silent intervals for each ear. Then the screening application runs the speech audiometry which test the user's speech recognition. Speech audiometry randomly plays 20 sounds of words with noisy background and allows users to select from four possible choices the appropriate picture and/or word that matches what the user heard. For each screening test, results are shown to the user and advice is given. Additionally, OHLSS also provides information about hearing loss, classification, causes, cure as well as preventive measures. OHLSS can be utilized in areas where there are few experts or unavailable audiometric equipment.

Keywords: hearing loss, online screening system, audiometry, health information system

HSD No. 8

CHILD IMMUNIZATION MONITORING SYSTEM

Ma. Sheila A. Magboo*, Apple Peach Coloyan

Dept. of Physical Sciences and Mathematics
College of Arts and Sciences
University of the Philippines Manila
Padre Faura, Ermita, Manila
E-mail: sheilaabad@yahoo.com

Childhood immunization is significant in preventing occurrence of several diseases. Failure to immunize a child on time can lead to new outbreaks of various diseases that are often deadly. Thus, the Department of Health implemented the Under Five Clinic and the Expanded Program on Immunization (EPI). The Under Five Clinic monitors immunization as well as growth patterns of children under five years of age while the Expanded Program on Immunization reduces infant mortality and morbidity through decreasing the prevalence of six immunizable diseases namely TB, diphtheria, pertussis, tetanus, polio and measles. At present there is no automated system that handles these records thus it is difficult to consolidate the results at the city, regional and much more on the national level which are necessary in order to come up with a more effective plan to reach out to families. As a response, a prototype system called "Child Immunization Monitoring System" was created. This is a is a web-based application that manages immunization records as well as the child's growth and weight and patterned after the Department of Health's Under Five Clinic and EPI. It has four major modules namely: a) vaccination coverage, b) vaccine inventory management, c) reminder and recall notification, and d) growth and weight. The vaccination coverage module can generate summary reports such as i) number of patients scheduled to have immunizations on a given month, ii) number of patients who showed up for follow-up on a given month, iii) number of patients who have not returned since their last follow-up date, iv) percentage of fully immunized child (FIC), etc.. The vaccine inventory management module can show the number of vaccines used in a specific clinic, city, or region over a given period. The reminder and recall notification module uses short messaging system (SMS) for parents with mobile phones to remind them of their child's upcoming vaccination. The growth and weight module enables health workers to update the child's age, height, and weight as well as view the growth chart. The usefulness of this system can be appreciated more especially when linked to a patient's medical record.

Keywords: child immunization, Expanded Program On Immunization, Under Five Clinic, monitoring system, health-care delivery system

HSD No. 9

PREVALENCE OF *Staphylococcus aureus* NASAL COLONIZATION AMONG GERIATRIC POPULATION OF ILIGAN CITY

Edgar Quitos*, Niño Marohom, Muriel Alejo and Lucilyn Lahoylahoy

Department of Biological Sciences, College of Science and Mathematics
Mindanao State University-Iligan Institute of Technology , Iligan City
E-mail: ed_quitos@yahoo.com

Staphylococcus aureus is considered to be one of the major human pathogens causing a wide range of infections and diseases and its colonization is documented to be one of the significant risk factor for subsequent infection. Elderly adults, 60 years and above, are a high risk of nosocomial and community-acquired staphylococcal infections due to impaired host defenses. The study was undertaken to determine the carriage rates of staphylococcus among healthy geriatric individuals and to determine the possible predisposing factors of *S. aureus* colonization. Fifty healthy geriatric individuals were recruited for the study in December 2005 to January 2006. Nasal swabs were taken from each respondent and were directly inoculated onto mannitol salt agar (MSA) for the detection and isolation of staphylococcal strains. Eighty-six percent (43/50) of the study population yielded positive nasal swab cultures.

One hundred fifty-three presumptively identified *Staphylococcus* isolates were randomly picked with 74 (48%) *S. aureus* isolates (mannitol fermentor, catalase and coagulase positive) and 79 (52%) were coagulase-negative *Staphylococcus* (conS). Forty-four percent of the plates (19/43) exhibited growth of both *S. aureus* and conS, 33% (14/43) with *S. aureus* alone and 19% (10/43) exclusive for conS. Individuals with history of antibiotic use yielded the most number of *S. aureus* isolates (17/43) followed by those with current illness (15/43), smokers (14/43), users of nasal sprays (9/43), users of herbal supplements (6/43) and with skin lesions (1/43).

The significantly high percentage of staphylococcal carriage rate (86%) in the study population is beyond the commonly reported colonization rates of normal individuals which would range from 20–40% of the population. The aged respondents generally would have lowered immunity and thus may have contributed to the higher *S. aureus* nasal colonization which may facilitate subsequent infection.

Keywords: Geriatric population, ***Staphylococcus aureus*** carriage rates, coagulase-negative staphylococci (conS)

HSD No. 10

BEHAVIORAL TRAITS IN MONOZYGOTIC AND DIZYGOTIC TWINS

Maritess Bonachita and Cesar G. Demayo

Department of Biological Sciences
College of Science and Mathematics
MSU-Iligan Institute of Technology
Iligan City
Email: c_gdemayo@yahoo.com

It is argued that behavior is the most complex phenotype reflecting the whole functioning of the organism. It is very dynamic and changes in response to its environment. How can behavior be studied then? Are behaviors inbred, written indelibly in our genes as immutable imperatives? Or, is the environment more important in shaping our thoughts and actions? Twin studies can assess the extent of relatedness to which resemblance is due to shared genes and the extent to which is due to shared environments and have been traditionally used in the assessment of hereditary and environmental influences on behavioral development. In this study, we determined the prevalence of behavioral traits in twins and estimate twin similarity using concordance and discordance rates. The following traits were studied: handedness (combing, toothbrushing, folding of things, picking of things, handwriting, change of hand preference in doing heavy tasks), hair whorl (clockwise, counterclockwise), tongue abilities (rolling, curling, rolling/curling), hand clasping (right over left, left over right), arm folding (right over left, left over right), stuttering (yes, no). One hundred eighty two (182) twin pairs, 149 monozygotic and 33 dizygotic were the subjects of this study. Left hand clasping, arm folding and toothbrushing were found to occur more frequently in monozygotic twins. On the other hand, holding of things by the left hand was found prevalent in dizygotic twin pairs. Non-stuttering was frequently observed in monozygotic twins. Chi-squared test showed no sex-difference in prevalence of behavioral traits. Twin similarity for ordinal data calculated as concordance rates (pairwise and probandwise) according to sex and zygosity revealed high probandwise and pairwise concordance rates in monozygotic twins. Tongue ability (curling), combing (left) and stuttering (yes) and left hand clasping were found to have high probandwise and pairwise concordance rates in dizygotic twin pairs. For monozygotics, pairwise and probandwise concordance rates were found in high frequencies for hairwhorl (counterclockwise), toothbrushing (left), holding of things (left), handwriting (left), preferential hand use when doing heavy tasks (left). Arm folding (left) was found to have equal concordance rates in both monozygotics and dizygotic twin pairs. We are still in the process of increasing the number of twin pairs being studied especially the dizygotics to be able to present phenotypic correlations of behavioral traits and

show the source of the influences in behavioral traits through structural equation modeling to accommodate the analysis of gender differences in heritability estimates.

Keywords: behavior, heritability, monozygotic twins, dizygotic twins

HSD No. 11

AN INTEGRATED SYSTEM FOR DNA ANALYSIS OF SEXUAL ASSAULT EVIDENCE

Frederick C. Delfin, Gayvelline C. Calacal, Kristina A. Tabbada, Henry B. Perdigon, Miriam Ruth M. Dalet, Minerva S. Sagum, Lilian.P. Villamor, Jazelyne M. Salvador, Saturnina C. Halos and Maria Corazon A. De Ungria*

DNA Analysis Laboratory, Natural Sciences Research Institute
University of the Philippines Diliman
Quezon City 1101, Philippines
Email: forengen@yahoo.com

The strength of DNA evidence depends on the effectiveness and reliability of the procedures used to collect, transport, store and analyze biological samples from which DNA evidence is generated. A system ensuring the proper documentation and preservation of evidence during collection and transport to the forensic laboratory and the subsequent storage and DNA analysis of the evidence at the laboratory would facilitate the resolution of sexual assault cases.

An evidence collection kit designed and assembled by our laboratory for sexual assault cases was used by Child Protection Units (CPUs) across the country (Baguio, Cebu, Davao and Manila) to collect, document and preserve biological evidence. Transport of evidence from the CPUs to our laboratory was handled by Air21, a local courier service provider. Upon receipt at our laboratory, evidence packages and appropriate documentation were inspected; the evidentiary materials inventoried and placed under appropriate storage conditions. For DNA analysis, a standard DNA extraction protocol (Phenol-chlorofom-isoamyl alcohol) and a commercial DNA extraction kit (Qiagen™, QIAamp® DNA Micro Kit) were used and compared. DNA extracted from sexual assault evidence was analyzed using Y-chromosome and autosomal short tandem repeat (STR) typing.

We report here our efforts to validate (1) sexual assault evidence collection using our evidence collection kit; (2) transport of evidence via a local courier system (3) A more efficient and cost effective protocol for DNA extraction from sexual assault evidence; (4) STR (autosomal and Y-chromosome) screening of biological

evidence and (5) the overall system of appropriate procedures for handling of sexual assault evidence from collection to DNA analysis.

Keywords: sexual assault, DNA analysis, STRs, autosomal, Y-chromosome

HSD No. 12

CYTOGENETIC AND MOLECULAR DIAGNOSIS AND MONITORING OF RESIDUAL DISEASE IN FILIPINO PATIENTS WITH CHRONIC MYELOGENOUS LEUKEMIA

Ma. Luisa D. Enriquez [1,5] Mignonette Hipolito[1], Michael Ernesto R. Arnante[1], Honorata Baylon[2], Priscilla B. Caguioa[3], Carmen T. Narciso[4], and The SLMC Leukemia Study Group

[1] Molecular Cytogenetics Laboratory, Research and Biotechnology Division,
[2] Bone Marrow and Transplant Unit,
[3] Cancer Institute, [4] Institute of Pathology,
St. Luke's Medical Center, Quezon City and
[5] Physics Department, College of Science, De La Salle University, Taft Ave. M.M.
Email: ftnatividad@stluke.com.ph

Methods for monitoring response to treatment in chronic myelogenous leukemia (CML) have changed considerably in recent years. In addition to routine analysis of Ph chromosome in G-banded metaphase chromosomes, molecular techniques like reverse transcriptase polymerase chain reaction (RT-PCR) and fluorescence in situ hybridization (FISH) are currently being used. These techniques have proved extremely available not only for diagnosis of CML but also for assessing and monitoring minimal residual in patients who have achieved Ph negativity after chemotherapy.

RT-PCR is considered to be the most sensitive method to date in detecting low leukemic burden. The technique can be carried out using peripheral blood (instead of bone marrow aspirate) and does not require the presence of dividing cells. It has the unique ability of determining specifically the type of fusion gene encoding either p190 or p210.

Ninety-five samples were submitted both for routine cytogenetic analysis and RT-PCR (nested PCR). Of these, 46 (48%) were newly diagnosed CML cases while 49 (51%) were from patients undergoing chemotherapy, many of whom are treated with ABL tyrosine kinase inhibitor imatinib mesylate or GLIVEC. In 15 (15.78%) of these patients, cultures failed to produce mitotic cells so cytogenetic analysis was not done, but RT-PCR detected the presence of the BCR-ABL fusion gene. In fourteen samples (14.73%), cytogenetic study showed normal karyotype

but RT-PCR revealed these were BCR-ABL-positive. The use of these molecular methods provided vital information that helped clinicians plan the therapeutic management of these patients, especially in recognizing early signs of relapse. The awareness of clinicians of the availability of these tests in the country will have a great impact on healthcare delivery for leukemia patients.

Keywords: Chronic myelogenous leukemia, residual disease, imatinib mesylate RT-PCR, FISH

HSD No. 13

ONLINE B-CELL EPITOPE PREDICTION APPLICATION

Vincent Peter C. Magboo ,* Pauline Donna P. Dizon

Department of Physical Sciences and Mathematics, College of Arts and Sciences
University of the Philippines Manila, Padre Faura, Ermita, Manila
E-mail: petermagboo@yahoo.com

Epitopes are the regions of a peptide antigen which interact with antibodies. Locating these epitopes is of particular interest to areas of vaccine research particularly designing subunit or recombinant vaccines that contain only the components necessary to induce production of memory cell. The Online B-Cell Epitope Prediction Application aims to predict the location of epitopes on an antigen by constructing the sequence scaling profile of the antigen's linear amino acid sequence. It provides a module that uses multiple scales to produce a composite profile that allow user to enter a linear sequence of amino acids from a protein antigen, select propensity scales to compute propensity scores for the sequence using Jameson-Wolf model, outputs the profiles representing the composite score in graphical format. In addition, there is a tutorial module which provides static and animated lectures to aid the users in understanding the concepts behind the construction of scaling profiles. An automatically generated test is likewise provided to the users. The application provides an efficient means of comparing which combination of sequence scales predict with high degrees of correctness the set of possible antigenic sites for a given antigen and therefore predict epitope portions. Hence, it can be used to identify potential vaccine candidates for human pathogens such as viruses. It can also be used to in testing genetically modified food for possible allergens. Lastly, it can be also be used as a teaching supplement in B-cell epitope prediction in molecular biology classes.

Keywords: immunology, antigen, epitope prediction, propensity scale, Jameson-Wolf model

HSD No. 14

CHROMagar *Candida* AS PRESUMPTIVE IDENTIFICATION MEDIUM FOR *Candida albicans*

Audrey A. Putong*, Lady C. Fanuncio, Johanna. L. Cuadra, Lucilyn D. Lahoylahoy

Department of Biological Sciences
College of Science and Mathematics
Mindanao State University-Iligan Institute of Technology
Iligan City
Email: audrey365@yahoo.com

CHROMagar *Candida* (CA) as a chromogenic differential medium has been reported to have 100 percent sensitivity and specificity in the identification of *Candida albicans*. This study was performed to presumptively identify *C. albicans* and non-*C. albicans* from 34 self-collected vaginal swabs of healthy women of Iligan City and to determine if there is any significant difference in the carriage rates between contraceptive users and non-contraceptive users. Fifty-nine percent (20/34) of the vaginal swab cultures yielded positive yeast-like growth on the primary isolation medium Sabouraud Dextrose Agar (SDA). Of the 20 plates with yeast-like growth, thirteen vaginal swab cultures came from individuals currently using contraceptive methods.

Generally, three isolates per colony type per plate were randomly picked. A total of seventy-nine isolates were collected and subcultured twice onto SDA to ensure purity. All 79 isolates grew on CA and the results were read by two different people according to the color, morphology of colonies and the existence of halo around them after 24 and 48 hours. Only thirteen percent of the total number of isolates (10/79) exhibited green colonies on CA and were presumptively identified as *C. albicans*. The remaining 69 non-*C. albicans* isolates had varying colony colors: cream (42%), violet (31%) and pink (27%).

Keywords: *Candida albicans*, CHROMagar *Candida* (CA), vaginal swab cultures, Sabouraud Dextrose Agar (SDA)

HSD No. 15

MODULATION OF MMP EXPRESSION OF NORMAL HUMAN DERMAL FIBROBLAST VIA PARACRINE FACTOR(S) GENERATED BY SQUAMOUS CELL CARCINOMA IS MEDIATED BY ERK1/2 AND p38

Maria Cristina Ramos [a*], Holger Steinbrenner, Dominik Stuhlmann, Helmut Sies and Peter Brenneisen

Institute for Biochemistry and Molecular Biology I, Heinrich-Heine-University Duesseldorf, Universitaetstrasse 1, 40225 Duesseldorf, Germany
[a]Research Center for Natural Sciences, University of Santo Tomas Espana Blvd., Manila, Philippines
Email: macrramo@yahoo.com; crramos@mnl.ust.edu.ph

Enhanced secretion of several types of matrix metalloproteinases (MMPs) is a characteristic of metastasis. Since a coordinated interaction of epithelial cells with stromal cells is a prerequisite for tumor invasion and metastasis, this study was undertaken in order to determine if paracrine factor(s) secreted by cancer cells modulate(s) the expression of MMPs in stromal cells and to study the molecular mechanism of the modulation of SCL-1 induced MMP expression in fibroblasts. Incubation of confluent monolayer of normal human dermal fibroblasts (NHDF) with supernatant taken from the tumor cell squamous cell carcinoma cell line (SCL-1) resulted in 5 and 60-fold increase of MMP-1 and MMP-10 expression, respectively compared to untreated NHDF as measured by RT-PCR. Time course analysis of MMP expressions in SCL-1 supernatant-treated NHDF revealed that the upregulation starts 4 h after treatment. ELISA assay revealed that MMP protein secretion is also increased when NHDF was treated with SCL-1 supernatant. Furthermore, treatment of NHDF with SCL-1 supernatant resulted in the activation of the 3 MAPK pathways (ERK-1/2, p38 and JNK). Incubating NHDF with inhibitors against MEK1/2 (U 0126) or p38 (SB 202190) before and during incubation with SCL-1 supernatant resulted in the inhibition of enhanced MMP expression of NHDF as induced by SCL-1 supernatant. Taken together, this study indicates that the activation of the ERK1/2 and p38 pathways plays a crucial role in the regulation of MMP activity of NHDF induced by the paracrine factors secreted by SCL-1 cells.

Keywords: MMPs, NHDF, SCL-1, ERK1/2, p38, metalloproteinases, metastasis, paracrine

HSD No. 16

GENETIC POLYMORPHISM OF gstm1 AND nqo1 IN FILIPINO PEDIATRIC ACUTE LYMPHOBLASTIC LEUKEMIA (ALL)

Marilyn G. Rimando1* and Gloria De Castro-Bernas, Ph.D.2

1Department of Biological Sciences, College of Science
University of Santo Tomas Espana, Manila
Email: mrimando7@yahoo.com
731-31-01loc. 4056
2 Research Center for the Natural Sciences
University of Santo Tomas, Espana, Manila

Leukemia is the leading type of cancer among Filipino children and being a complex disease is associated with hereditary susceptibility to a number of carcinogenic and biological agents. Gene polymorphisms coding for carcinogenic detoxifying enzymes such as Glutathione S-transferases (GSTT1 and GSTM1) and NAD(P)H:Quinone Oxidoreductase1 (NQO1) are reported to have an association with the risk of developing various cancer types. Frequency distribution of these biomarkers varies among Asians, Caucasians and other ethic groups, and has not been identified among blood related cancer types such as Leukemia among Filipino cases.

Screening of GSTT1, GSTM1and NQO1 Genotypes was conducted using Multiplex Polymerase Chain Reaction (PCR) from 180 DNA samples (60 randomly selected Acute Lymphoblastic Leukemia patients, 60 normal individuals, and 60 cases with other hematological diseases) isolated from peripheral blood of pediatric individuals (d"1 to 18 yrs old) diagnosed at the University of Santo Tomas Hospital and The Children's Medical Center, Banawe Manila. Optimization of PCR conditions using varying Magnesium chloride concentrations , annealing temperatures and DNA concentration was conducted. Pertinent clinical information was also gathered and analyzed.

Results showed that Multiplex amplification of the "positive" and "null" genotypes for GSTT1 (459bp) and GSTM1(219 bp) using Albumin as internal control (350 bp) was optimized at 3.0 mM Magnesium chloride concentration, 66°C annealing temperature and 100 ng/ul DNA concentration. NQO1 detection of the C (161 bp) and T (283 bp) alleles for the C/C, C/T and T/T genotypes is recommended at 3.5 mM Magnesium chloride concentration, 60°C annealing temperature and 100 ng/ul DNA concentration.

Based on the preliminary screening conducted among 180 Filipino Pediatrics , it was observed that most individuals carry the GSTT1 "positive" allele (65%) with almost equal distribution between leukemia, normal and other cancer individuals (65%,67%,50% respectively) . Most pediatrics however possess the "null" GSTM1

genotype (60%) with 43 out of 60 leukemia patients (72%) lacking the allele, 52% of normal individuals and 57% with other hematological disease. For the NQO1 genotypes, most normal and those individuals with hematological diseases are heterozygotes (C/T) with 75% and 98% respectively, and interestingly, most Leukemia patients posses the wild type (C/C) genotype (60%).

These results conform with reported frequencies from different race and confirms the lack of the GSTTM1 allele in most Asians including Filipinos. Comparison of the Leukemia genotypes with normal individuals and with other hematological can assist clinicians in providing insights on leukemia susceptibility, possible measures of avoidance or prevention and additional information for clinical management.

Keyworks: Gene Polymorphisms, Multiplex PCR, GSTT1,GSTM1, NQO1

HSD No. 17

PHYLOGENETIC ANALYSIS OF HEMOLYSIN GENE FAMILIES IN *Vibrio* SPECIES

Boris B. San Luis*, April B. Cabang and Cynthia T. Hedreyda

National Institute of Molecular Biology and Biotechnology
University of the Philippines Diliman, Quezon City
Email: bbsanluis@up.edu.ph

Bacterial hemolysins are considered important virulence factors of pathogenic *Vibrio* species, causing erythrocyte membrane lysis in the host. *Vibrio* species express a range of hemolysins that can be grouped into four representative families namely: TDH (thermostable direct hemolysin) family, HlyA (El Tor hemolysin) family, the TLH (thermolabile hemolysin) family and the *d*-VPH (thermostable hemolysin) family. The genes encoding hemolysins have been isolated, cloned, sequenced and characterized in several pathogenic and non-pathogenic *Vibrio* species. In this study, the phylogenetic relationship of hemolysins among *Vibrio* species is determined first by multiple sequence alignment of available hemolysin nucleotide and amino acid sequences using CLUSTALW/MULTALIN 5.4.1 and phylogenetic tree construction using MEGA software. Results show separate clusters for each of the hemolysin gene families, with the hemolysin gene of *Vibrio harveyi* and the recently isolated and sequenced hemolysin gene of *Vibrio campbellii* forming a close group with thermolabile hemolysins. The thermostable direct hemolysin and HlyA families each form a separate cluster. The phylogenetic analysis was extended to include the related proteins phospholipases and lecithinases, and results reveal close relatedness of phospholipases to the thermolabile hemolysins of

Vibrio while the lecithinases group was closer to the thermostable direct hemolysins.

Keywords: *Vibrio,* hemolysin genes, phylogenetic analysis

HSD No. 18

SEQUENCE ANALYSIS OF 16S rRNA GENE IDENTIFIES UNKNOWN MYCOBACTERIAL PATHOGENS OF THE EYE AFTER CATARACT SURGERY

Brian Carlmichael L. Torres[1], Ma. Luisa G. Daroy[1]
Ruben Lim Bon Siong[2], and Ronald R. Matias[1]

[1]Research and Biotechnology Division and
[2]Institute of Ophthalmology, St. Luke's Medical Center, Philippines
Email: mldaroy@yahoo.com

Gene sequence analysis of 16S rRNA provides more rapid, unambiguous identification of difficult bacterial isolates compared to conventional methods. Accurate identification can translate to improved clinical outcomes through the application of appropriate treatment. This study reports on the use of 16S rRNA gene analysis to identify unknown mycobacteria isolated from eyes which have undergone cataract removal. Lowenstein-Jensen agar cultures of mycobacteria from infected corneal tissues and lens implant were studied. Biochemical tests and species identification by 16S rRNA gene sequence analysis were done. The sequences were compared to those in the NCBI GenBank database and Ribosomal Differentiation of Medical Micro-organisms Database (RIDOM).

The partial 16S rRNA gene sequences of 3 isolates were found to have 99%, 90%, and 95% homologies to that of *Mycobacterium massiliense*, sp. nov. based on a comparison with sequences in NCBI GenBank. Comparison of the same with those in the RIDOM database showed 91.92% identity with both *M. abscessus* and *M. chelonae*. . Four biochemical tests capable of differentiating *M. massiliense* from *M. abscessus* and *M. chelonae*, tryptophan desaminase, b-glucuronidase, N-acetyl-b-glucosaminidase, and b-galactosidase, were performed on two of the isolates. Both were positive for tryptophan desaminase, and negative for the other three enzymes. This profile differed in one test from that reported for *M. abscessus* and *M. chelonae*, and 2 tests from that of *M. massiliense*. In this study, 16S rRNA gene analysis and biochemical profiling showed that the three isolates are closely related to the *M. abscessus-M. chelonae* group.

Keywords: 16S rRNA gene, nontuberculous mycobacteria, eye infection

HSD No. 19
CARRAGEENAN-COCOA BUTTER BASED SUPPOSITORIES FOR FEVER

Annabelle V. Briones, Wilhelmina O. Ambal, Ronald Domingo, Romeo Banal
Romulo R. Estrella, Norvina S. Deocampo, Josefina R. Celorico
Teresita S. Bonifacio and Merle A. Villanueva

Industrial Technology Development Institute
Department of Science and Technology
Gen. Santos Avenue, Bicutan, Taguig City
Email: abriones2003@yahoo.com

A carrageenan-cocoa butter based suppository for fever was produced. Acetaminophen was used as the active ingredient. This was done by dissolving the carragenan with water then mixed thoroughly to obtain a thick homogenous mixture. Afterward the mixture was added to melted cocoa butter previously mixed with stearic acid and preservatives followed by constant stirring while slowly adding the acetaminophen. After obtaining a dough-like consistency, the mixture was then poured into the molder and allowed to congeal at 4 °C. Then removed from the molder and packed. The suppositories are prepared in 250 mg dose for children and 500 mg dose for adult. They melt at body temperature (37 °C) and liquefy within 3–7 minutes. Bioavailability assay of the carrageenan-cocoa butter based suppositories in rabbits showed that acetaminophen was absorbed from the suppository and becomes available at the site of drug action based on the presence of acetaminophen at various concentration levels in the blood. The blend of carrageenan-cocoa butter is a good suppository base for acetaminophen. Carrageenan increases the release and absorption rate of acetaminophen from the suppository. It is a good drug-carrier for acetaminophen in suppository form via rectal administration.

The use of carrageenan in the production of suppositories is an innovative technology that will expand the application of carrageenan in the areas of pharmaceuticals and medicine. Thus, it will indirectly help boost the Philippine carrageenan industry in the world market with these other fields of development.

Keywords: Carrageenan, Suppository, Acetaminophen, Cocoa-butter

HSD No. 20

INHIBITORY EFFECTS OF ANTIBIOTIC-LADEN CEMENT STICKS ON MICROBIAL PATHOGENS: POTENTIAL TREATMENT OF POST-SURGICAL INFECTION

Blanquita B. de Guzman[1], Andrew Gabriel Tabberrah[2], Cristine R. Casingal[1], Lindsay Sydney N. Fajardo[1], Maria Adelwisa Belen[2], Antonio Tanchuling[2] and Filipinas F. Natividad[1]

[1]Research & Biotechnology Division and
[2]Institute of Orthopaedics & Sports Medicine,
St. Luke's Medical Center, Quezon City, Philippines
Email: ftnatividad@stluke.com.ph

Infection, one of the most feared complications in prosthetic joint surgery, is of great concern because it may result in total loss of joint function. The use of antibiotic-laden cement sticks is a new modality to manage such bone infections. These are implanted into areas where periprosthetic infections persist. This study aims to determine the inhibitory activity of antibiotics released from the bone cement sticks on three different pathogens - *Staphylococcus epidermidis, S. aureus* and *Enterococcus faecalis.*

CM3 cements, pre-loaded with Gentamicin (*Gen*), Vancomycin (*Van*) and a fusion of Vancomycin and Fucidin (*Van-Fuc*) and bone cement without antibiotic, were tested. They were immersed in 5 ml 0.9% normal saline solution (NSS) at room temperature. The eluate was collected once a day for a week, after which fresh NSS was added. Thereafter, collection was done every week for a period of one month. Agar-diffusion technique was used to determine the inhibitory effects of the eluates on the 3 test bacteria. The biggest zones of inhibition (25.25, 20.33 and 15.50 mm for *Van-Fuc, Van,* and *Gen* respectively) were observed with *S. epidermidis* using the 24-hour eluates. The zone diameter decreased for the rest of the week, but started to increase up to the 2nd month. The decrease in the size of the inhibition zones was 55% for *Gen*, 38% for *Van,* and 23% for *Van-Fuc* at the end of the 1-week period. In terms of effectivity, *Van* and *Van-Fuc* inhibited all the microorganisms tested, while *Gen* inhibited only *S. epidermidis.*

In this study, we found that regardless of the kind of antibiotic loaded in the cement, the highest elution occurred during the first 24 hours. The agar plate diffusion method is a useful and reproducible technique for *in vitro* measurements of inhibitory activity of the antibiotics released from the bone cement sticks.

Keywords: Antibiotic-laden cement sticks, elution, diameter of inhibition zone, Vancomycin, Gentamycin, Vancomycin-Fucidin, prosthetic joint surgery, post-surgery infection

HSD No. 21

SIMPLE TESTS PREDICTED MULTIPLE DRUG RESISTANT *STAPHYLOCOCCUS AUREUS* IN A TERTIARY HOSPITAL, 2001-2005

***Delia C Ontengco[1,2], Leilani A Alcaraz[1], Roberta S Santiago[1], Wilfredo M Verar Jr[1], and Alexander O Tuazon[1]**

[1]Medical Affairs Division, United Laboratories, Inc., Mandaluyong City 1501 and [2]Graduate School, University of Sto. Tomas, España St., Manila
Email: dcontengco@unilab.com.ph

Staphylococcus aureus (Sa) causes boils, toxic shock syndrome, and diseases of the lung, heart, and bone. Penicillins easily treated these infections. In 1950, Sa began producing beta-lactamases that destroyed the beta-lactam ring and rendered these agents inactive. In 1960, Sa became resistant to the newer beta-lactams, BLs, (methicillin, oxacillin) and cephalosporins. This new strain, known as methicillin-resistant Sa (MRSA), harbors the *mecA* gene, is resistant to ALL BLs and BL/BL-inhibitor combinations, and is now causing immense problems in treatment leading to morbidity and mortality. Our study monitored multiple-drug resistant Sa in a tertiary hospital from 2001-2005 by using oxacillin disk as preliminary indicator, and confirmed methicillin resistance with cefoxitin disk. *MecA* is expressed at high levels in the presence of cefoxitin. Fifty-three MRSAs were identified out of 297 (17.8%) Sa isolated from clinical specimens in 2001-2005: 22.5% (13/40), 15.5% (11/71), 21.4% (15/70), 11.8% (10/85), and 12.9% (4/31), respectively. The antibiogram profile of the MRSAs showed resistance to ALL BLs and BL/BL-inhibitor combinations. Although vancomycin is the agent of choice for MRSAs, results indicate this pathogen is still susceptible to the following: sulfamethoxazole/trimethoprim (98%), gentamicin (96%), tetracycline (94%), and rifampicin (96%). An important observation in this study is the emergence of another threat: macrolide-lincosamide-streptogramin (MLS) resistance which was noted in 2003 (1.4%, 1/70) and 2004 (1.1%, 1/85). MLS-resistance is suspected in erythromycin-resistant and clindamycin-susceptible isolates. The D test was performed to assess resistance to clindamycin. Positive D test results indicate that the clinical isolates in these cases should be reported as resistant to clindamycin. Without supplementary tests the patient runs the risk of treatment failures. Results further suggest the presence of two resistance mechanisms in these isolates: *mecA*-encoded methicillin-resistance and *erm*-encoded MLS-resistance. We, therefore, conclude that for every Sa strain three simple tests be done: the oxacillin, cefoxitin, and D tests.

Keywords: *Staphylococcus aureus,* MRSA, MLS resistance, D test

HSD No. 22

ANALYSIS OF ETHANOL IN BLOOD USING HEADSPACE GAS CHROMATOGRAPHY WITH FLAME IONIZATION DETECTOR

Ma. Cristina B. Portilla[1], Johanna Paula N. Ebarvia[2] and Cherrie B. Pascual[1,2]

[1]Research and Biotechnology Division, St. Luke's Medical Center
279 E. Rodriguez Sr. Blvd, Cathedral Heights, Quezon City 1102
[2] Institute of Chemistry, University of the Philippines, Diliman, Quezon City 1101
Email: ffnatividad@stluke.com.ph

Blood alcohol concentration is an important measure of alcohol consumption. The alcohol level in the blood at a certain time may affect a court decision resolving a criminal act such as murder, rape and other reckless acts related to high levels of alcohol in the blood. Currently, there is no known forensic lab in the Philippines that tests alcohol in blood or urine.

Gas chromatography with a flame ionization detector, together with headspace analysis was utilized for the determination of ethanol. One mL of the headspace gas in vials containing varying concentrations of ethanol, which were heated at 70 to 80°C for 5 minutes, was injected into the GC column. A DB-WAX column with an isothermal temperature of 120°C was used. In this method, ethanol had a retention time of approximately 2 minutes. The coefficient of variation for the retention times (n=5) was 13.6% while that of peak areas was 6.9% ethanol gave linear response with the concentration range of 1 to 5 g/L both in aqueous solution and in spiked blood sample. The limit detection was 0.008% ethanol (w/v).

Healthy volunteers were made to consume alcohol with informed consent and their blood samples were analyzed for ethanol using this chromatographic method. The chromatogram of blood sample from a non-drinker showed the absence of the ethanol peak. The calibration plot of ethanol spiked in blood was used to determine the ethanol concentrations in the blood samples.

This method can be used for the analysis of ethanol in blood for forensic analysis, and more importantly, the diagnosis of patients suffering from alcoholism. It is recommended for use in routine tests for employment screening and driving license application.

Keywords: ethanol, blood, headspace gas chromatography, flame ionization detector, alcoholism

HSD No. 23

RECOMBINANT BLO T 11 PEPTIDE FOR THE DIAGNOSIS OF HOUSE DUST MITE ALLERGY AMONG FILIPINO ALLERGIC PATIENTS

[1,2]John Donnie A. Ramos*, [2]Tomas Marion Falconitin, [2]Reynaldo Lazo, [2]Dim Ralyn Medina, [2]Minerva Nicolas, and [2]Christine Ysabelle Roman

[1]Research Center for the Natural Sciences and
[2]Biological Sciences Department
College of Science, University of Santo Tomas
España St., 1008, Manila
Email: ramosjda@yahoo.com

The paramyosin-homolog allergen Blo t 11 from the house dust mite (HDM) *Blomia tropicalis* (Bt) is an important allergen causing sensitization of more than 50% of allergic patients in tropical countries. Recombinant peptide coding for the immunodominant epitope of Blo t 11 is an invaluable reagent for HDM allergy diagnosis and immunotherapy. The 666 bp immunodominant fragment of Blo t 11 was amplified by PCR, cloned into pGEX-4T-1 vector, expressed as a GST-fusion peptide in *Escherichia coli*, purified by affinity chromatography using glutathione agarose column, and was used in an IgE ELISA using a panel of sera from 100 Filipino allergic patients. The 222 aa immunodominant peptide fD was highly expressed in soluble form with an average yield of 5.7 mg recombinant peptide per liter of E. coli culture. Recombinant fD migrated to ~50 kD in a 12% Tris-Glycine SDS-PAGE gel in accordance to its predicted molecular weight (24 kD + 26 kD GST). The allergen reacted positively to 87% of Filipino allergic patients' sera tested. An inhibition assay indicated that fD is responsible for more than 50% IgE reactivity observed in Bt extract. The results presented in this study showed the utilization of recombinant Blo t 11 peptide fD as diagnostic reagent for HDM allergy. The inclusion of this peptide in the panel of allergens used in the diagnosis and immunotherapy of HDM allergy is highly recommended.

Keywords: Allergy, recombinant peptide, *Blomia tropicalis*, immunoglobulin E

HSD No. 24

THE ANTIDIABETIC ACTIVITY OF *Tinospora rumphii boearl* (MAKABUHAI) LEAVES

Lorna T. Enerva, Ferdinand B. Batoon, Cesario V. Ong Jr. and Mary Rose Ricafrente

College of Science and SciTech R and D Center
Polytechnic University of the Philippines, Sta. Mesa, Manila
Email: lornaenerva@yahoo.com

Leaves of makabuhai (1500 g) were soaked in 3 liters of ethylacetate for five days and the crude extract was obtained. The crude extract exhibited a bluish dark green color and was very sticky. The filtrate was concentrated using a rotary evaporator and purified by column chromatography. The structure of the purified extracts were elucidated using infrared spectra which revealed the presence of phenolic OH,C-H and ester functional groups, UV-Vis analysis indicated the possible presence of polyenes and multiple bonds with a maximum absorbance of ë 434 cm^{-1}. The presence of the following compounds were identified using gas chromatography-mass spectrometry: 1,2-benzenedicarboxylic acid, diisooctyl ester; 1-oxaspiral (2.5) octane, 5,5-dimethyl-4-(3-methyl-1,3-butadienyl); cedran-diol, 8S,14; spiculesporic acid; retinoic acid, methyl ester; 2-(4-methyl-6(2,6,6-trimethyl.cyclohex-1-en-1-enyl)hexa-1,3,5-trenyl)cyclohex-1-en-1-carboxaldehyde;4.8,13-cyclotetradecatriene-1.3diol,1,5.9trimethyl12-(1-methyl ethyl).

Albino Swiss Webster Mice of 4–5 weeks-old, male were used as the test specimen for the bioassay using Tail Venipuncture Method. Diabetes was induced in the mice which were divided into four groups. The purified and crude extract of makabuhai showed 19.65% and 19.52% decrease in blood glucose level and was close to the standard medicine decrease of 20.41%. The % decrease is relative to the hyperglycemic blood glucose level. Based on the results obtained, the leaves of makabuhai exhibits hypoglycemic properties.

Keywords: antidiabetic activity, hyperglycemia, bioassay, infrared spectra, ultraviolet spectra, gas chromatography, mass spectra.

HSD No. 25

ANTITUMOR POTENTIAL OF THE LEAF EXTRACT OF SOME HEALTH FOODS

Annabelle A. Herrera[1], Esperanza Ong and Irene Villasenor[2]

[1]Institute of Biology and [2]Institute of Chemistry
College of Science, University of the Philippines Diliman
Quezon City 1101;
Email: anabelle.a.herrera@gmail.com

Cancer is one of the leading causes of death all over the world. In this study, histopathological techniques were used to determine the antitumor potential of lettuce, coriander, leek and strawberry in the heart and aorta of Swiss albino mice. Dimethylbenzanthracene was the tumor initiation agent while croton oil was the promoter. The leaf extract was given before initiation or after tumor promotion for 24 weeks. The heart and aorta were dissected out and processed after the 24th week. *Lactuca sativa* or lettuce showed potential as antitumor initiator while *Coriandrum sativum* or coriander had antitumor promotion potential.

Keywords: Antitumor potential, leaf extract, coriander, lettuce, leek, strawberry

HSD No. 26

BIOACTIVE COMPOUND FROM ACTINOMYCETE FOR HEALTH AND AGRICULTURE APPLICATIONS

Teofila O. Zulaybar*, Irene A. Papa, Eufrocinio DC. Marfori, Edwin P. Alcantara and Bernardo C. Mercado

National Institute of Molecular Biology and Biotechnology
University of the Philippines Los Baños, College, Laguna 4031
*E-mail: fezu@laguna.net

Philippine Actinomycetes are untapped microbial resources which can be exploited for the discovery of new bioactive compounds against existing antibiotic resistant organisms and pests in agricultural crops.

The worldwide occurrence of Methicillin Resistant *Staphylococcus aureus* (MRSA) and *Candida albicans* are the most common causes of fatal infections and diseases in hospitals. In 2002, emergence of Vancomycin-resistant *S. aureus* and enterococci is raising serious public health concerns. *Candida albicans* is an equally significant organism causing Candidiasis in women. With the threat of antibiotic

resistant organisms, it is imperative to continue search for organisms producing novel antibiotics.

Out of 388 Actinomycetes assayed, A5 from garbage soil in Pakil, Laguna showed antimicrobial activity against 9 clinical strains of MRSA (9.1-13.3mm) and 2 strains of *C. albicans,* Ca1 (13.15mm) and Ca4 (11.95mm). Minimum inhibitory concentration (MIC) (Marfori et. al., 2002) of ethyl acetate extract using *C. albicans* as test organism showed the same MIC as the positive control (Nystatin) 7.8 ppm. Bioautography (Marfori, 2002) produced 1 active spot against *C. albicans*. The same extract showed 4 active spots against MRSA. Fractionation of the active compound was done using solvents of increasing polarity.

Ethyl acetate extract of A5 was highly toxic to larvae of diamondback moth, *Plutella xylostella* and Asian corn borer, *Ostrinia furnacalis*, major insect pests of crucifers and corn, respectively.

Actinomycete isolate A5 is a potential source of active compound against antibiotic resistant organisms and pests in agricultural crops.

Keywords: Actinomycete, Methicillin resistant *Staphylococcus aureus*, MRSA, *Candida albicans*, bioautography

SOCIAL SCIENCES

SSD No. 1

ATTENTION DEFICIT HYPERACTIVITY DISORDER (ADHD) AMONG TEENAGERS AND IN SCHOOLCHILDREN AGES 3–8 IN ILIGAN CITY

Cesar G. Demayo, Mark Anthony J. Torres, Concepcion Apao and Ianni Bea Garcia

Department of Biological Sciences, College of Science and Mathematics
MSU-Iligan Institute of Technology, Iligan City
Email: c_demayo@yahoo.com

Attention Deficit Hyperactivity Disorder (ADHD) is a developmental disability that usually affects children and is usually characterized by behavioral and learning disorders. Some of commonly cited characteristics in children with ADHD are hyperactivity, distractibility and impulsivity. Children with ADHD find it difficult to perform a task assigned to them and focus on some of the important aspects of conversations. We used the SNAP-IV Teacher and Parent Rating Scale to get the information needed from the child in selected schools from Iligan City. In this

study, we allowed only the teachers to fill-out the survey form based on his observations of the children under his/her class. The SNAP-IV Rating Scale consisting of ninety (90) behavioral descriptions of the child is a revision of the Swanson, Nolan and Pelham (SNAP) Questionnaire (Swanson et al, 1983). We paid attention to four subtypes of ADHDs in the survey–the two subsets of symptoms: inattention (ADHD-1) and hyperactivity/ impulsivity items (ADHD-H/ Im), items from the DSM-IV criteria for Oppositional Defiant Disorder (ODD) and items from the Conners Index Questionnaire (Conners, 1968) where the items which loaded highest on the multiple factors of the Conners Questionnaire, represents a general index of childhood problems. Our preliminary results on 251 schoolchildren ages 3–8 years old showed none of those surveyed have symptoms of ADHD based on the total 90 descriptors. However, when the four subtypes were assessed based on selected descriptors of SNAP-IV, results showed none has symptoms of ADHD-I (inattention), 2 females (0.8%) and 10 males (4%) showed ADHD-H/Im (inattention and impulsivity), 2 males (0.8%) and none for the females for ADHD-C. Thirteen (13) females (5.18%) and 32 males (12.75%) have the ODD descriptions.

A study to determine ADHD was also conducted on young adults. We now know that ADHD symptoms frequently persist in the adult lives of people who had ADHD as a child. Because it is not always practical, or possible, to obtain information from an informant such as a parent, employer or clinicians, we relied on the student's own account of his or her current symptoms and the subject's recollection of childhood symptoms. We used the Wender-Utah Rating Scale developed by Dr. Paul Wender in 1995 to determine adult ADHD among the 739 students in six colleges of MSU-IIT. Of the 739 students, 509 or 68.87% were observed negative for ADHD behavior, 151 or 20.43% probably had ADHD and 79 or 10.69% were observed positive. The result of this study is comparable to published reports that showed at least 10% of adult populations manifest this disorder.

Keywords: Attention Deficit Hyperactivity Disorder, ADHD, SNAP-IV, Conner's Index

SSD No. 2

VALIDATED TRADITIONAL WEATHER FORECASTING METHODS IN ILOCOS NORTE, PHILIPPINES

Evangeline S. Galacgac and Criselda M. Balisacan

Research and Development Directorate
Mariano Marcos State University Batac, 2906 Ilocos Norte
rddirectorate@yahoo.com

Traditional weather forecasting knowledge and methods (weatherlore), which guide Ilocano folks in their farming, fishing activities and for disaster preparedness, were documented and validated from year 2002-2004.

Indicators which are reliable to predict the onset of rainy season and upcoming rain are: ants storing foods, herons migrating to the mountain, lesser caucal (Centropus bengalensis) uttering its call "kok-kok", and frogs croaking near swampy areas. When dragonflies fly low, loose dogs defecate in the middle of the road, plaintive cuckoo (Cocomantis merulinus) utter its call "pee-to-peet", and Himalayan swiftlet (Aerondramus brevirostris) fly low chasing for insects, rain is imminent. Typhoon/storm will occur when a long parallel band of feathery clouds, moon with ring (lunar corona), giant seawaves, and sea roaring or visible seawater evaporation are observed. With Bristol, weather-wise folks could tell the seasonal outlook for the incoming year based on the prevailing weather condition of the last 12 days of December of the current year.

Animal behavior and appearance of clouds, moon and sea have scientific basis. They are highly reliable to Ilocano folks and are useful for agriculture, fishery planning and operation, and to prepare for adverse weather condition to supplement PAGASA forecasts.

Key words: weatherlore, forecasting, weather, rainy season, traditional

SSD No. 3

MEASURING THE ECOLOGICAL FOOTPRINT OF LOYOLA SCHOOLS (2001)

Chloe Jean M. Mojica and Teresita R. Perez

Department of Environmental Science
School of Science and Engineering
Ateneo de Manila University
Loyola Heights, Quezon City, Philippines
Email: chloemojica@yahoo.com

Ecological Footprint Analysis (EFA) is developed by Mathis Wackernagel and William Rees (1990s) to respond to the issues of carrying capacity and sustainability. It serves as an accounting tool that estimates the resource consumption and waste assimilation requirements of a defined human population in a defined productive land area. It is rapidly carving its niche in the scientific world as scientists continuously contribute to its broadening concepts and has been used in evaluating academic institutions.

The EFA of Ateneo de Manila University has been studied with the following objectives : (1) to measure the spatial impact (footprint) and the ecological footprint of Loyola Schools in terms of power and water consumption and solid waste generation during 2001; (2) to assess the availability, accessibility and quality of data necessary for this type of analysis; and (3) to present results that identify significant environmental impacts that will be instrumental in setting up future environmental management systems applicable inside Loyola Schools.

The calculated footprint from the three components studied is 139.79 hectares. This is four times larger than the estimated area of Loyola Schools which is 32 hectares. Loyola Schools had a population of 25,999 during 2001 thus per capita footprint is 0.005 global hectare. The footprint associated with power consumption is 101.11 hectares which is 72.33% of the total footprint – the highest contributing component. The footprint associated with water consumption is 37.76 hectares which 27.01% of the total footprint. The footprint associated with solid waste generation is 0.92 hectare which is only 0.66% of the total footprint – the smallest contributing component.

In terms of ecological footprint, Loyola Schools has a total ecological footprint of 226.17 global hectares which is three times larger than its potential productive area (70.40 global hectares). Per capita ecological footprint is 0.008 global hectare.

EFA offers a simplistic way of identifying and addressing activities (resource consumption and waste generation) with large spatial impacts to the biophysical environment. Data required for this analysis were available but were mostly in

basic form which required conversions and several assumptions to fill information gaps to reconcile incomplete data.

EFA is a viable tool for analyzing the sustainability of a university unit similar to Loyola Schools since it has clearly identified that resource consumption and waste generation have definite ecological and economic consequences. This confirms that EFA is a practical method to measure ecological footprints of universities.

Keywords : accounting tool, carrying capacity, ecological footprint analysis, sustainability

SSD No. 4

EFFECTIVENESS OF FOUR COMMUNICATION STRATEGIES IN CONVEYING BIODIVERSITY INFORMATION FROM THE UPLB MUSEUM OF NATURAL HISTORY TO SELECTED GRADE V PUPILS IN LOS BAÑOS, LAGUNA

Graciosa B. Romero,*[1] Rosa Pilipinas J. Fadri,[1] Aimee Lynn A. Barrion[2] and Ireneo L. Lit, Jr.[2,3]

[1]Department of Development Journalism
College of Development Communication,
[2]UPLB Museum of Natural History, and
[3]Department of Forest Biological Sciences
College of Forestry and Natural Resources
University of the Philippines Los Baños College Laguna 4031
*E-mail: graciousromero@yahoo.com

Extension and communication of biodiversity information is virtually an unexplored area of study in the Philippines. Previous efforts along this line focused mainly on conveying agricultural knowledge and technological innovations. Eliciting appreciation for biological diversity and its conservation, especially to the younger sectors of the population, is of paramount importance considering the fast decline and seemingly irreversible loss of our natural biological heritage. The University of the Philippines Los Baños Museum of Natural History is one of the informal learning institutions, that is considered also as a unique educational environment that impart life-long learning, particularly in the field of biodiversity and its conservation. To determine the effect of its new insect biodiversity exhibit on the cognitive learning scores of students, 76 Grade V pupils from Lopez Elementary School were randomly selected. The study adopted a pre-test and post-test scheme composed of four treatments, namely: T1 – exhibit viewing only,

T2 – exhibit with lecture / guided tour, T3 – exhibit viewing with brochure and T4 – exhibit viewing with lecture / guided tour and brochure provided. Mean cognitive learning pre- and post-tests scores were recorded, tallied and averaged. Ranked mean of the pre-test cognitive learning scores were: T2 (6.53) = T3 (6.53) > T4 (6.37) > T1 (6.16) whereas those of the post test were: T2 (10.26) > T4 (9.58) > T3 (8.84) > T1 (8.05). Results suggest that, by itself, viewing the insect biodiversity exhibit had improved the mean cognitive learning scores. However, combining the exhibit with a lecture or guided tour or with both a lecture guide and brochure generated the highest scores. These two treatments did not differ significantly suggesting that the pupils appreciated or learned more from listening to a guided tour where there is greater interaction than from mere reading of a brochure. Nonetheless, the higher scores for T3 compared to those for T1 suggest that compared to plain viewing of exhibits, an attractive yellow brochure helps a lot in cognitive learning. Despite limitations, this study yielded interesting results as far as communicating scientific information on biodiversity is concerned. More extensive and more detailed studies should be conducted to further refine biodiversity extension strategies and more efficiently communicate information on our remaining natural heritage.

Keywords: museums, biodiversity extension, insects, science communication