

“Natural Products and Microbes for Health and Sustainability”



College of Biochemists of Sri Lanka

Proceedings of the 3rd Conference

24th July 2021

Online conference

College of Biochemists of Sri Lanka (CBSL)
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Proceedings of the 3rd Conference of the Collage of Biochemists of Sri Lanka (CBSL)-24th July 2021

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3rd CBSL Conference: GLANCE Program 2021.

Theme: Natural Products and Microbes for Health and Sustainability

Time (India Standard Time)	Event detail	Resource Person	Chair / Moderator
8.30 am		<i>Participants log in</i>	
8.45 am	Inauguration	<i>National Anthem</i> <i>Welcome address by President/CBSL</i> <i>Address by the Chief Guest: Prof Chandrika Wijeyaratne, Vice Chancellor University of Colombo, Sri Lanka</i> <i>Address by the Guest of Honour : Prof. Vajira Dissanayake, Dean, Faculty of Medicine University of Colombo, Sri Lanka</i>	
9.10 am		Presidential address: Sharmila Jayasena (20 min)	Chair: Prof. Lohini Athiththan
9.30 am		Keynote address: Prof. Angelo Azzi, (Tufts University, Boston, USA) (45 min)	Chair: Prof. Sharmila Jayasena
		<i>Vote of thanks: Dr. Niroshima Withanage (joint secretary CBSL)</i>	
10.15 am	Symposium 1: Natural products as anti-cancer agents	Prof. Iqbal Choudhary University of Karachi, Pakistan (20 min)	Chair: Dr. Swarna Hapuarachchi
		Dr. Imalka Munaweera University of Sri Jayewardenepura, Sri Lanka (20 min)	
		Prof. Preethi Udagama University of Colombo, Sri Lanka (20 min)	
		Q & A	
11.45 am	Oral Presentations I		Chair: Prof Rasika Perera
1.00 pm	Break / Viewing of Posters: https://www.collegeofbiochemists.lk/		
1.20 pm	Orals Presentation II		Chair: Dr. Anoja Attanayake

“Natural Products and Microbes for Health and Sustainability”

2.30 pm	Symposium 2: Microbiota for sustainable living	Dr. Thilini Jayasinghe Sydney Dental School and the Charles Perkins Centre, University of Sydney, Australia (20 min) Dr. Aparna Banerjee CIEAM, Universidad Catolica del Maule, Chile (20 min)	Chair: Prof Sharmila Jayasena
		Q & A	
3.30 pm	Panel Discussion Natural products for COVID-19	Prof. Arunee Thitithanyanont FS, Mahidol University, Thailand Dr. Kamal Jayasinghe CEO, National Medicines Regulatory Authority, Sri Lanka Dr. MWSJ Kumari IIM, University of Colombo, Sri Lanka Prof. Sugandhika Suresh FMS, University of Sri Jayewardenepura, Sri Lanka	Moderator: Dr. Sanath Mahawithanage
		Audience Q & A	
4.30 pm	Awards and closing		Dr. Tharanga Thoradeniya (Vice President CBSL)
4.45 pm	Streaming ends		

Welcome Message from the President of CBSL



Prof. Sharmila Jayasena

President,
College of Biochemists of Sri Lanka.

It is my privilege to welcome you to the 3rd conference of the College of Biochemists of Sri Lanka (CBSL). The current pandemic has imposed an unprecedented burden on researchers worldwide and Sri Lanka has had its fair share! However, this conference with over 30 abstract presentations, stands as evidence of the resilience and resourcefulness of local biochemists and molecular biologists, who have, despite all odds, found ways to carry on with their research. To all of you, well done and congratulations!

The theme of this year's conference is “Natural products and microbes for health and sustainability”. It was selected to emphasize the importance of the investigation of natural products as well as microbes in the exploration of sustainable solutions for human and planetary health. Plants, especially local and endemic plants in Sri Lanka are a popular choice as sources of natural products among local investigators. The microbes, less so. However, the importance of microbes as a bioresource, from microbial cell factories producing metabolites to functional consortia, cannot be over-emphasized. They provide an extensive arena with a multitude of avenues for research.

This conference will provide a platform to bring together several investigators working in these fields to exchange ideas and share expertise. In the keynote address and symposia, our distinguished invited speakers will highlight some of these areas. The panel discussion will deliberate on the use of natural products for COVID-19.

I acknowledge with gratitude the IT support provided by the Faculty of Medicine, Colombo as well as the contributions of our sponsors, without whom this conference would not be a possibility. I extend my appreciation to the members of the organizing committee who have worked tirelessly amid their busy schedules to make this conference a success.



Keynote speaker
Aging and Natural Products

Professor Angelo Azzi

School of Graduate Biomedical Pharmacology and Drug

Development Programme, Tufts University, Boston, MA, USA.

Professor Angelo Azzi is a faculty member of the School of Graduate Biomedical Pharmacology and Drug Development Programme, Tufts University, Boston, MA, USA. He is also a Professor Emeritus of the Universität Bern, Switzerland. Angelo has two doctoral degrees, one in Pathophysiology and the other in Biochemistry, along with an MD which he has earned from the University of Padua.

He has over 350 publications to his credit in peer-reviewed international journals, which has yielded 14495 citations together with an author h-index of 66. Furthermore, Angelo is the Editor-in-Chief of two leading scientific journals, namely, *Molecular Aspects of Medicine* and *BioFactors* and the Associate Editor of *Mechanisms of Ageing and Development*.

He holds a number of memberships in international organizations; President of the International Union of Biochemistry and Molecular Biology, IUBMB from 2006-2012 and the member of the advisory board of UNESCO (ISAB/IBSP) from 1997-2005; to name a few.

His subjects of research have been bioenergetics, mitochondrial structural studies, membrane structure and function and mechanisms of action of natural compounds, especially vitamin E, carotenoids and curcumin.

Aging and Natural Products

Professor Angelo Azzi

School of Graduate Biomedical Pharmacology and Drug Development Programme, Tufts

University, Boston, MA, USA

Efforts to uncover the age-related processes at the basis of chronic diseases to promote “healthy longevity” or prolong the “health span” is indispensable to reduce both healthcare and financial burdens imposed by an increasingly aged population. According to the free radical hypothesis of aging, aging of organisms is caused by the accumulation of oxidative damage. Studies have shown that oxidative damage increases with age and that its reduction increases the lifespan of several model organisms (yeast, nematodes, fruit flies, mice, etc.). However, a growing number of studies have contradicted the oxidative damage hypothesis. In particular, an increase of life span by vitamin E, considered by some scientists the most powerful natural antioxidant, has not been experimentally supported although vitamin E may be important in protecting against some age-related diseases. The role of natural compounds working as sirtuin activators has been also considered as a way of increasing life span. A recent understanding of the aging phenomenon has been described by which selective elimination of senescent cells could be obtained by compounds like dasatinib in combination with quercetin, or by the flavone fisetin a treatment that also prevented the appearance of the senescent mice phenotype. Modulation of Nuclear factor-erythroid 2 p45-related factor 2 (Nrf2) by natural products, has been shown to facilitate repair or degradation of damaged macromolecules, to modulate intermediary metabolism and to improve mitochondrial function in animal models of aging.

Presidential Address

Sharmila Jayasena PhD

Dept. of Biochemistry and Molecular Biology, Faculty of Medicine, University of Colombo

Sharmila Jayasena is a professor in the Department of Biochemistry and Molecular Biology, Faculty of Medicine, University of Colombo. She holds a BSc (Hons) in Microbiology from University College, London, and a PhD in molecular biology from the University of Colombo having investigated the ‘Heat shock protein 70 genes of the filarial nematode *Setaria digitata*’. She subsequently received training at Cold Spring Harbor Laboratory, New York, in ‘*Caenorhabditis elegans*’ techniques. She is currently investigating molecular pathways involved in microbial bioremediation of petroleum hydrocarbons. She is also a team member of a collaborative project investigating protein profiles and effects of plant extracts in plasma cells of multiple myeloma patients. Her research work is supported by the University of Colombo, National Science Foundation and World Bank AHEAD, grant schemes. She is a recipient of the President’s research awards and the IUBMB young scientists travel fellowship.

She has served the College of Biochemists of Sri Lanka in various capacities since its inception and is the current president. She is a delegate to the council of the Federation of Asian and Oceanian Biochemists and Molecular Biologists (FAOBMB), a member of the FAOBMB Fellowships Committee for 2020 and 2021 and a Co-chair of the IUBMB-FAOBMB-CBSL Education Symposium to be held on the 30th of July this year.

Microbial Bioremediation of Petroleum Hydrocarbon Pollution

Prof. Sharmila Jayasena

Dept. of Biochemistry and Molecular Biology, Faculty of Medicine, University of Colombo

Numerous products from plasticware to gasoline, paints, and dyes, used in domestic, transport as well as the industrial sectors are derived from crude petroleum oil. Accidental spillage of crude oil may occur during storage or transport of petroleum hydrocarbons. Ecosystem damage resulting from pollution of the environment with these toxic recalcitrant compounds is an increasing global concern. Microbial bioremediation of the polluted environment has increasingly generated interest in recent years as a green technology. This presentation will describe the isolation and characterization of a naturally occurring *Bacillus-Aspergillus* consortium that rapidly and synergistically degrades crude oil under laboratory conditions. Further it will highlight the identification of novel fungal homologs of key alkane monooxygenases, including those that have the capacity to degrade long-chain alkanes. *In silico* protein structure prediction and molecular docking analyses have been used to predict enzyme-substrate binding of the newly identified fungal homologs. It is envisaged that this knowledge will provide the foundation for process development in microbial bioremediation of petroleum hydrocarbon pollution.

Symposium 1: Natural products as anti-cancer agents



Prof. Iqbal Choudrey

International Center for Chemical and Biological Sciences

University of Karachi, Pakistan

Dr. M. Iqbal Choudhary is Director and Professor of Bioorganic and Natural Product Chemistry at the International Center for Chemical and Biological Sciences (H. E. J. Research Institute of Chemistry and Dr. Panjwani Center for Molecular Medicine and Drug Research) and Coordinator General COMSTECH (Organization of Islamic Cooperation Standing Committee on Scientific and Technological Cooperation). Prof. Choudhary has, since 1990, been among the world leaders in the field of natural product chemistry, and has made pioneering contributions in the discovery of novel natural products. He has discovered many potent anti-epileptic and anti-leishmanial compounds from indigenous medicinal plants that are under clinical trials. His contributions to reverse bacterial resistance to antibiotics represent seminal contributions in this important field. He leads the developing world's finest research center of natural product chemistry (H. E. J. Research Institute of Chemistry) since 2002, and has trained hundreds of young researchers, especially women, from across the Afro-Asian region in natural product chemistry. He has established several research centers in Pakistan, and helped to setup research units in Africa, and South and Central Asia. His scientific, and capacity building contributions have been recognized by prestigious national and international awards and honors, and fellowships of several academies of science. Prof. Choudhary has 1,154 publications (Citations 29,850, *h* index 70) in the fields of organic and bioorganic chemistry, along with 74 international patents (56 US Patents), 87 books and 40 chapters in books, published by major U.S. and European presses. On the basis of his researches, 98 students have been awarded Ph.D. degrees in various areas of natural product and bioorganic chemistry.

**Natural Products –As a Potential Therapeutic Source for the Discovery of
Anti-Cancer Molecules**

M. Iqbal Choudhary, and Atta-ur-Rahman

International Center for Chemical and Biological Sciences

*H. E. J. Research Institute of Chemistry, Dr. Panjwani Center for Molecular Medicine and
Drug Research), University of Karachi, Karachi-75270, Pakistan*

Sciences at the interface of chemistry and biology have led to increased opportunities for the identification of lead molecules against various therapeutic targets. For centuries, natural products have served as key sources of therapeutic agents, and still many of current drugs are derived from medicinal plants. However, synthesis of natural products is still a challenging task due to various reasons, including structural and stereochemical complexities, and long yields. These problems can be overcome with the help of biocatalysis and combinatorial biosynthesis, as enzymes have high selectivity and specificity, and can work under mild conditions in both organic and aqueous media.

During last four decades, our research group has focused on the discovery of chemical constituents from medicinal plants used in traditional medicines, as well as development of new biotransformed products with therapeutic potential. This has resulted in the identification of several novel lead molecules as therapeutic agents for cancer treatment.

Breast cancer is the most common cancer in women worldwide, with nearly 2.3 million new cases diagnosed in 2020 (second most common cancer overall). Approximately one-third of all breast cancer patients and two-thirds of postmenopausal breast cancer patients have hormone-dependent (estrogen-dependent) breast cancers, which express estrogen receptors and require estrogen for tumor growth. Aromatase inhibitors based-drugs are currently being used as primary prevention therapy, such as exemestane and tamoxifen, are in clinical practices for the treatment of estrogen sensitive breast cancers. Therefore, there is a need to identify new structural analogues of available drugs and evaluate their anticancer potential. In this study, our research group have implemented new approaches to synthesize new derivatives of existing aromatase inhibiting drugs through biotransformation, and use some molecules of natural sources to evaluate their potential against aromatase enzyme. This has provided an efficient method for the synthesis of new analogues of existing aromatase inhibitors. The new analogues of the mentioned drugs and some natural products were found to be moderate to potent inhibitors of aromatase enzyme, as compared to standard drugs letrozole, and exemestane.

During this presentation, several recent examples of our studies highlighting the translational potential of bioactive natural products, and biotransformation against various cancers will be presented.



The application of nanotechnology in medicine/cancer drug delivery

Dr. Imalka Munaweera

Senior Lecturer

Department of Chemistry, University of Sri Jayewardenepura

Dr. Munaweera graduated from University of Peradeniya with a Bachelor of Science Degree in Chemistry (Special) in 2007. She received her PhD in Chemistry from University of Texas at Dallas, Texas in USA in 2015. She was a postdoctoral fellow at Department of Radiology, University of Texas Southwestern Medical School, Texas in USA from 2015-2018. She served as an Assistant Professor in Chemistry at A&M University, Prairie View, Texas, USA from 2018-2019. She has developed, published and patented several nanoparticle drug delivery systems for cancer therapy, infectious diseases and other biological applications during her graduate and postdoctoral period. Currently, Dr. Munaweera is a Senior Lecturer at Department of Chemistry, University of Sri Jayewardenepura in Sri Lanka.

The application of nanotechnology in medicine/cancer drug delivery

Dr. Imalka Munaweera

Senior Lecturer,

Department of Chemistry, University of Sri Jayewardenepura.

According to the World Health Organization, cases of cancer are growing at a rapid rate and 22 million new cases and 13 million deaths are estimated each year by the year 2032. Therefore, new therapeutic systems are essential for cancer treatment in order to tackle this global issue effectively. Major drawbacks in current drug delivery systems are inefficient drug delivery with less payload and inefficient eradication of malignant cells. Targeting drug delivery increases efficacy, decreases side effects, and reduces systemic drug exposures. Engineered nanoparticles drug delivery systems have recently been developed for cancer therapy and other biomedical applications. An ideal controlled drug delivery vehicle should be equipped with the factors such as prolonged circulation of drug molecules in the blood, targeting into the tumor sites, response to local stimuli such as pH/temperature, etc., and efficient delivering of drugs into the tumor sites. Therefore, it is of great significance to develop a drug delivery vehicle combining several of the useful properties in one particle system. It is also essential to develop nanoparticle drug delivery systems with capability to distinguish between cancer and healthy cells to drug release.



**Cancer chemopreventive activity of mature leaf juice of
Carica papaya Sri Lankan wild type cultivar**

Prof. Preethi Udagama

Cadre Chair & Senior Professor,
Department of Zoology and Environment Sciences,
University of Colombo, Sri Lanka.

Senior Professor Preethi Udagama holds the Chair, and is the Head of the Department of Zoology & Environment Sciences, Faculty of Science, University of Colombo, where she has served for over 27 years as a teacher and a researcher.

Professor Udagama obtained both her BSc. Honours degree in Biological Sciences and her Masters' degree in Nuclear Science, from the Faculty of Science of the University of Colombo. She obtained her PhD in Immuno-parasitology in 1990 from the Faculty of Medicine, also from the University of Colombo. Her research training was further honed by a Post doctoral fellowship (1990) followed by a visiting fellowship (1992), awarded by the National Science and Engineering Research Council of Canada, which were tenured at the University of New Brunswick in Fredericton.

An Immunologist by training, Professor Udagama has numerous publications and communications to her credit and has received many awards for her research contributions. Professor Udagama is actively involved in capacity building of young scientists by training undergraduates and postgraduates in biomedical research. She functions as a reviewer of several national and international journals. Professor Udagama is a Fellow of the National Academy of Sciences, Sri Lanka.

Cancer chemopreventive activity of mature leaf juice of *Carica papaya* Sri Lankan wild type cultivar

Prof. Preethi Udagama,

Department of Zoology, Faculty of Science, University of Colombo.

Cancer chemopreventive activity of natural products reduces the incidence of tumorigenesis by intervening at one or more stages of carcinogenesis. Carcinogenesis is a complex and multistep process in which oxidative stress and inflammation plays a crucial role. Currently, a broad spectrum of plant preparations with antioxidant and anti-inflammatory properties has gained much attention as cancer chemopreventive or therapeutic agents. The leaf juice of *Carica papaya* has been traditionally claimed as a remedy against cancer; nonetheless this claimed chemopreventive activity was yet to be scientifically investigated. In the present study, the mature leaf juice of the Sri Lankan wild type cultivar of *Carica papaya* (MLJC) was evaluated for cancer chemopreventive activity in terms of anticancer, antioxidant and anti-inflammatory activities. Anticancer activity of the MLJC and positive drug cyclophosphamide was investigated using the MTT *in vitro* cell proliferation assay using Hep-2 (human laryngeal carcinoma) cells. Antioxidant and anti-inflammatory activities of the oral gavage of the MLJC (low: 0.18, mid:0.36 and high:0.72 ml/100g BW) were evaluated using murine models with CCl₄ induced oxidative stress and carrageenan induced peritonitis, respectively. The MLJC established a marked anticancer (cell cytotoxicity) activity with an EC₅₀ of 56.43±4.5 µg/ml against Hep-2 cells. Oral gavage of the mid and high doses of the MLJC significantly enhanced the enzymatic antioxidants, superoxide dismutase (SOD), glutathione peroxidase (GPx) and glutathione reductase (GR) compared with the control oxidative stressed rats ($p < 0.05$). Significant inhibition of leukocyte migration was observed with oral gavage of mid and high doses of MLJC in mice with carrageenan induced peritonitis ($p < 0.05$). Moreover significant inhibition of prostaglandin E₂ was observed with oral administration of the MLJC ($p < 0.05$). This study established that the MLJC possesses cancer chemopreventive activity in terms of anticancer, antioxidant and anti-inflammatory activities, and suggests that the MLJC warrants investigations as a potential cancer chemopreventive agent.

Symposium 2: Microbiota for Sustainable Living



Dr. Thilini Vidanelage

Postdoctoral Research Fellow in the Charles Perkins Centre,
University of Sydney, Australia.

Dr Thilini Vidanelage is an emerging medical researcher in the area of Microbiome research. Thilini received the "University of Auckland Prestigious Doctoral Scholarship" award in 2014. During her PhD program, she worked extensively in gut microbiome research in the Liggins Institute of The University of Auckland. She completed her PhD in Health Sciences after producing an exceptionally high-quality thesis in 2019 and published highly impacting pivotal first author peer-reviewed research papers and many collaborative research papers in the growing area of microbiome research. During her PhD, she played an integral role in one of the world-first clinical trials that tested novel treatments for treating obesity in adolescents. This study attracted an abundance of scientific and public interest worldwide. Thilini was one of the leading four researchers of the project featured in the TV documentary called *The Good Sh*t* in 2018.

Furthermore, Thilini conducted another unique research of analysing bacterial components of one of New Zealand's most well-known artists, Billy Apple's 46 years old stool sample. During this time, she presented those exciting research outcomes internationally and was awarded the Best Presentation in one of the highly distinguished conferences in the microbiome field, i.e. Welcome Trust Genome Conference, Cambridge UK. She was awarded in "Nutrition Research Methods at Bangalore Boston Nutrition Collaborative" and "Health Research Methods and Evidence-Based Medicine" at St. John's Medical College and Research Institute, Bangalore, India, in collaboration with McMaster University Canada.

Thilini completed her BSc in Food Science and Nutrition at Wayamba University of Sri Lanka and awarded the Professor HMP Gunasena Award for the Best Academic Performances in 2008. After that, she completed a master degree and a master by research degree in the Post Graduate Institute of Agriculture, Sri Lanka and The University of Sydney, respectively. Currently, Dr Vidanelage works as a Postdoctoral Research Fellow in the field of "Diet and periodontitis".

Short- and long-term dynamics of gut microbiome

Dr. Thilini Vidanelage

*Postdoctoral Research Fellow in the Charles Perkins Centre, University of Sydney,
Australia.*

There are 100 trillion microbes living in and on our human body. At the gene level, humans have 100 times more microbial genes than their own gene. Research has shown that the microbiome changes throughout life. Thus, we explored short and long-term fluctuations in the human gut microbiome that are associated with early life-events, ageing, and an intervention. Firstly, we hypothesised that adverse early life events in preterm children change the microbial composition, functions and metabolic products of the gut microbiome in mid-childhood. Our results identified different active microbial species as classifiers of the preterm condition together with functional changes, altered profiles of plasma and faecal amino acids, faecal volatiles and faecal calprotectin levels. We speculate that the preterm-specific changes observed in the active gut microbiome were established in early infancy and are associated with on-going low-grade gut inflammation. Secondly, characterisation of the gut microbial composition of artist Billy Apple® from stool contaminated toilet tissues collected in 1970 and 2016 showed that the microbial composition in 2016 represents 45% of the microbial species in 1970. Moreover, components of Apple’s microbiome were associated with the allele frequency at seven single nucleotide polymorphisms in his genome confirming that genetics contribute to the selection and maintenance of the microbiome over the artist’s lifetime. Thirdly, we studied the short- and medium-term effects of lean donor faecal microbiota transplantation on the gut microbiota composition in a group of female adolescents with severe obesity. Our results showed that the faecal microbiome transplantation can shift the recipient microbiota successfully at 6 weeks, post-faecal microbiota transplantation. The engrafted microbiota remained unchanged for half of a year despite subtle dietary and environmental changes.



**Bacterial polysaccharides from extreme environments:
Realms of possibilities**

Dr. Aparna Banerjee

Assistant Professor CIEAM, Universidad , Catolica del Maule,
Chile.

Dr. Aparna Banerjee is PhD Botany from University of Burdwan, India. She has Bachelors & Master's in biotechnology. She has done her postdoctorate from Universidad Católica del Maule, Chile where she is presently working as Assistant Professor. In her 7 years of research experience, she worked on extremophilic microbiology. Her research interest also includes biodiversity, conservation and bioinformatics. She is presently the joint secretary of India's Bioinformatics Society, Biolcues.

She is currently running three international projects and has more than 25 publications in nationally and internationally reputed journals.

Furthermore, she has been featured in the documentary podcast in UK, featured in the world campaign “1 million women in STEM” in 2020, also featured in Unicef-India programs on climate change. Dr.Banerjee received Indian Science Congress Association (ISCA) Young Scientist Awardee in 2018, National Academy of Sciences, India (NASI) Swarna Jayanti Puraskar in 2017, and has been selected in Young Researcher Forum at the 10th International Conference on “Nanomedicine and Nanotechnology in Health Care” 2016 in Bangkok, Thailand

Bacterial polysaccharides from extreme environments: Realms of possibilities

Dr. Aparna Banerjee

Centro de Investigación de Estudios Avanzados del Maule, Vicerrectoría de Investigación y Postgrado, Universidad Católica del Maule, Talca, Chile

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Microbes in extreme environment are uniquely adapted to great temperature variations by different cellular and molecular modifications, such as exopolysaccharide production, genomic changes, regulation of gene expression or production of pigments and many more. In an era of ever increasing climate change impacts, it is more important to address the extremophiles before we lose them. Extremophiles often harbour industrially important secondary metabolites like enzymes, pigments or polysaccharides that have promising industrial applicability. In northern hemisphere regions, including India, the Indian Himalayas and the Deccan region, there are several mining zones where geothermal springs have formed with high surface temperature. While in high altitude region of Himalayas, the psychrophiles thrive. In the southern hemisphere, especially in Chile, polyextremophiles thrive in hot springs of the Andes Mountains, or in Southern Ocean / Antarctica. The main shared feature of these extremophiles from different parts of the globe is production of exopolysaccharides as a part of protection strategy. While the thermotolerant microorganisms in a relatively unexplored hot spring of India produce glucan- and rhamnoglucan-type of exopolysaccharides, the ones from natural hot springs of the Transitional Southern Volcanic Zone of Chile show the presence of complex α - and β -glycosidic bonds. The psychrophiles in Southern Ocean, Antarctica has also exhibited complex network of polysaccharides so that of the thermophiles. Our study highlights the occurrence of highly thermostable polysaccharides that may be used as antioxidant, emulsifiers or viscosifiers in the food industry or as mediators for the synthesis of green nanoparticles. Overall, the compositional dynamics of polyextremophiles in different parts of the world reveals excellent potential for industrial applications.

Keywords: Extreme environment, Hot springs, Southern Ocean, Exopolysaccharides, Microbial diversity, Microbial conservation

Panel Discussion: Natural products for COVID-19



Prof. Arunee Thitithanyanont

Department of Microbiology, Faculty of Science, Mahidol University, Thailand

I am a faculty member of the Department of Microbiology, Faculty of Science, Mahidol University, since 1997. After my Pediatric residency and clinical fellow training at Ramathibodi Hospital Faculty of Medicine, I joined the international Fogarty program at Harvard AIDS Institute, Harvard School of Public Health, for three years. At the Faculty of Science, Mahidol University, my foremost curiosity is to understand why H5N1 causes extrapulmonary viral dissemination and the interplay between the virus and the immune system. I also serve as the Director of the BSL-3 Core Facility at her faculty, leading a team of skilled personnel who operates the facility and performs in vitro and ex vivo work with risk group 3 (RG3) pathogens. Since 2015, the facility has been annually certified by international agents to comply with the WHO requirements and CDC/NIH Guidelines for BSL-3 laboratory facilities. Currently, I collaborate and support many researchers from Mahidol University and other institutions in Thailand to conduct their research on COVID-19. Our activities include viral isolation, neutralization test, vaccine development, and drug discovery.



Dr. Kamal Jayasinghe

MBBS, DFM, MSc-Med. Admin, MCMA, MBA, DIPPCA

Chief Executive Officer,
National Medicines Regulatory Authority, Sri Lanka

Dr. Kamal Jayasinghe is the Chief Executive Officer of the National Medicines Regulatory Authority. He holds postgraduate qualifications in Family Medicine, Medical Administration, Business Administration, Public Procurement and Contract Administration. He has held many senior positions in a career spanning 28 years in medical administration.

Dr Jayasinghe functions as the secretary to the Authority and is responsible for the execution of all its decisions.



Dr. (Ms.) M. W. S. J. KUMARI

Senior Lecturer of the Institute of Indigenous Medicine, University of Colombo, Rajagiriya, Sri Lanka

Dr. (Ms.) M. W. S. J. KUMARI is a Senior Lecturer of the Institute of Indigenous Medicine, University of Colombo, Rajagiriya, Sri Lanka with 29 years of service. She also works as a Consultant to the National Ayurveda Teaching Hospital, Borella. She is a Bachelor of Ayurveda Medicine and Surgery (BAMS) (First Class) Degree holder, graduated in 1990 from the University of Colombo. In 1996, she has obtained Doctor of Medicine, (MD) (Ayurveda) with the specialization of Basic Principles of Ayurveda from the Gujarat Ayurved University, India. She earned PhD (Doctor of Philosophy) from the Dr. Sarvepalli Radhakrishnan Rajasthan Ayurved University, India (2014). She is holding Postgraduate Diploma in Education (2001), Certificate in Teaching in Higher Education (1998), Diploma in Dangerous Drug Abuse Management Studies (Distinction Pass) (2005) and Diploma in Health Promotion (Distinction Pass) (2008) from the University of Colombo. She also obtained special training certificates in Yoga, Panchakarma and Massage therapy from India. She has received Australia Awards Fellowship from the Department of Foreign Affairs and Trade, Australian Government, for Developing a Medical Research Governance Strategy for Sri Lanka (2017). She has served in many positions as Department Head (Department of Basic Principles), Sectional Head (Ayurveda Section) and Acting Director of the Institute of Indigenous Medicine. She engaged in research specially in joint diseases, osteo-arthritis, diabetes, stress, mental disorders and geriatric diseases. She has presented her research work at national and international forums. She has conducted lectures in various Institute and Universities as an external lecturer and a resource person. She is a Life Member of All India Ayurvedic Specialist (P.G.) Association, Global Ayurveda Society of India, Society of United Life Sciences (SOULS) of India., Sri Lanka Association for Advancement of Science, Senior Scientists Forum (National Science & Technology Commission), Institute of Biology (Sri Lanka) and Royal Asiatic Society of Sri Lanka. She was also awarded Certificate of appreciation and gold medal to the dedicated service to Ayurveda on 20th March 2019, presented by Sri Lanka-United Nations Friendship Organization for World Women’s Day. Presently she serves as a member of National Health Research Council of Sri Lanka and a member of Ethics Review committee of IIM. She is the current President of the Teachers Association of the Institute of Indigenous Medicine, University of Colombo and the Secretary of the Population Association of Sri Lanka. She was recently appointed as a committee member to recommend Drug Protocol for COVID- 19 by the State Ministry of Indigenous Medicine, Rural and Ayurveda Hospitals Development and Community Health.



Professor Sugandhika Suresh

Department of Biochemistry,

Faculty of Medical Sciences, University of Sri Jayewardenepura

Professor Sugandhika Suresh obtained BSc Honours in Human Biology from the Faculty of Medical Sciences, University of Sri Jayewardenepura in 1998 and obtained her PhD in Biochemistry in 2002. She was attached to the Department of Biochemistry and Clinical Chemistry, Faculty of Medicine, University of Kelaniya (2001-2006) as a Lecturer, prior to joining University of Sri Jayewardenepura in 2006 as a Senior Lecturer. Presently she is a Professor in Biochemistry (since 2014) at the Department of Biochemistry, Faculty of Medical Sciences, University of Sri Jayewardenepura. She has been extensively involved in undergraduate teaching, curriculum development and revision as well as research.

Professor Suresh has a keen interest in research on traditional medicine and herbal extracts and effects on chronic diseases such as diabetes and arthritis. She has contributed to the scientific validation of traditional preparations during the past 2 decades. She is a pioneer in conducting registered clinical trials for traditional herbal medicines. Professor Suresh has been involved in the development and commercialization of novel products from indigenous medicines, where she has conducted collaborative studies with local and international centres of excellence which provided value-addition to herbal drugs. She is also a qualified Laboratory Animal scientist and is the Coordinator of the Animal House of the university.

Professor Suresh has authored more than 35 indexed and peer-reviewed papers and 100+ communications and won numerous awards and prizes; among which are the presidential, institute of Chemistry, SLAAS and NSF awards. She has successfully supervised several PhD, MPhil and MSc students. Professor Suresh delivered the Faculty Oration at the annual research conference of the Faculty of Medical Sciences, USJ in 2016.

Professor Sugandhika Suresh was the Founder Treasurer of CBSL and she was instrumental in getting the affiliation of CBSL to the FAOBMB. She was the President of CBSL from 2017-2019. She was a Council Member of FAOBMB and is presently a member of the Education Committee of FAOBMB. Professor Suresh was also the Founder Secretary (2012-2014) and President (2015-2016) of SLALAS. Presently she is the President of OWSD-Sri Lanka National Chapter.

“Natural Products and Microbes for Health and Sustainability”

Oral Presentations

Assessment of cytotoxicity and apoptotic activity of different fractions of *Gracilaria edulis* against human rhabdomyosarcoma (RMS) cell line

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Marine seaweeds are a rich source of bioactive metabolites that can be used as an alternative source for the development of the anti-cancer drug. The present study aimed to evaluate the cytotoxicity and apoptotic activity of different fractions of *Gracilaria edulis* against the human rhabdomyosarcoma (RMS) cell line. De-polysaccharide polyphenol-rich methanol extract of *G. edulis* was sequentially partitioned with hexane, chloroform, and ethyl acetate to determine the cytotoxic and apoptotic effects. The cytotoxic activity was assessed by MTT and neutral red assays while apoptotic activity was examined by cellular morphology, DNA fragmentation, and caspase 3/7 assays. The results of the cytotoxicity assay showed that the decrease in the percentage of cell viability in a dose-dependent manner as signified by cell death. According to the MTT assay, the hexane (HF; $IC_{50Hexane}: 32.5 \pm 2.2 \mu g/ml$) and chloroform fractions (CF; $IC_{50Chloroform}: 77.1 \pm 1.6 \mu g/ml$) exhibited potent cytotoxic activity compared to the standard cycloheximide ($IC_{50}: 36.2 \pm 1.8 \mu g/ml$). Further, the neutral red assay confirmed the cytotoxic activity of hexane fraction ($IC_{50Hexane}: 33.5 \pm 2.3 \mu g/ml$) compared to the standard cycloheximide ($IC_{50}: 32.8 \pm 0.9 \mu g/ml$). The morphological assessment of apoptosis was confirmed using Hoechst 33342 staining and crystal violet staining. The prominent activation of Caspase 3/7 was observed in the RMS cells treated with hexane and chloroform fractions of *G. edulis* compared to the standard staurosporine and cycloheximide. Similarly, the typical DNA ladder pattern was observed in HF and standard cycloheximide-treated RMS cells. It can be concluded that the HF of *G. edulis* has the ability to suppress cellular proliferation and induce apoptosis-mediated cell death in RMS cells via a caspase-dependent pathway. Thus, the HF of *G. edulis* can be a potent candidate to isolate the new anti-cancer compounds.

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Key Words: Cytotoxicity, Apoptotic activity, *Gracilaria edulis*, Human rhabdomyosarcoma

Nephroprotective effects of *Abelmoschus moschatus* against Adriamycin-induced nephrotoxicity model in Wistar rats: A biochemical, histopathological and immunohistochemical assessment

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Abelmoschus moschatus Medik (family; Malvaceae) is widely being used in the treatment of kidney diseases in traditional Ayurvedic practice. The objectives of the present study was to evaluate the nephroprotective effects of the hexane, ethyl acetate, butanol and aqueous leaf extracts of *A. moschatus* against Adriamycin-induced nephrotoxicity model in Wistar rats. Healthy male Wistar rats were randomly divided into seven groups (n=6/group). Experimental rats of Group 1 and 2 were considered healthy and nephrotoxic (adriamycin; 5 mg/kg, ip) controls respectively and administered distilled water. The nephrotoxic rats in group 3-7 were orally administered with hexane; 55 mg/kg, ethyl acetate; 75 mg/kg, butanol; 60 mg/kg, and aqueous; 140 mg/kg extracts of *A. moschatus* and the standard drug (0.09 mg/kg) respectively for a period of 28 days. A sample of urine (24 hour), blood and kidney tissues were collected at the end for the biochemical, histopathological and immunohistochemical assessments. Ethical clearance was granted from the Ethical Review Committee, Faculty of Medicine, University of Ruhuna (14.12.2015:3.1). Treatment with the selected extracts of *A. moschatus* significantly decreased the elevation of serum creatinine (22%, 26%, 21%, 18%), blood urea nitrogen (37%, 43%, 27%, 38%), cystatin-C (59%, 59%, 55%, 56%) and decreased the reduction of serum albumin (32%, 7%, 51%, 27%) and total protein (53%, 25%, 54%, 53%) (p<0.05). Serum β_2 -microglobulin was reduced with significant changes in hexane (18%) and ethyl acetate (20%) extracts. A reduction in proteinuria was observed only with the butanol (37%) and aqueous (27%) extracts (p>0.05). The semi-quantitative assessment of histopathology in the H and E stained kidney sections revealed attenuation of the features of acute kidney injury in plant extract treated rats. The immunohistochemical expression of COX-2 and Bax was decreased and the expression of BCL-2 was increased following the treatments. The results revealed that hexane, ethyl acetate, butanol and aqueous leaf extracts of *A. moschatus* possess significant nephroprotective activity, probably mediated via anti-inflammatory and antiapoptotic mechanisms in Adriamycin-induced nephrotoxicity.

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Key Words: Nephroprotective, *Abelmoschus moschatus*, Nephrotoxicity, Wistar rats

***In vitro* antidiabetic activity of aqueous leaf extract of *Coccinia grandis* L. encapsulated alginate nanoformulation**

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Development of alginate nanoparticles loaded with extract of *Coccinia grandis* L. (Ivy Gourd, Family: Cucurbitaceae) is a prospective approach to enhance the antidiabetic activity of the aqueous leaf extract of *C. grandis*. The objective of the present was to assess *in vitro* antidiabetic activity of aqueous leaf extract of *C. grandis* encapsulated alginate nanoformulation (CNF). Aqueous leaf extract of *C. grandis* (0.06 mg/mL) was prepared using ultrasonication (40°C, 30 min, 40 kHz) followed by refluxing (100°C, 2 ½ h). CNF was synthesized via ionic gelation method. The encapsulation efficiency (EE) and loading capacity (LC) of the nanoformulation was calculated with respect to the total phenol content in the supernatant. Resultant pellet was subjected to the analysis of particle size and zeta potential. CNF was evaluated for α - amylase and α - glucosidase inhibitory activities with the reference compound of acarbose. Glucose uptake and glucose adsorption assays were performed on CNF using metronidazole as the reference compound. The optimum concentration of aqueous leaf extract of *C. grandis* for encapsulation was determined as 4 mg/mL with the highest EE (56.7 \pm 0.6%), LC (3.1), lowest particle size (298.9 nm) and agreeable zeta potential ((-) 21.0 mV). Both α - amylase and α - glucosidase inhibitory activity of CNF was improved as 60.8% and 19.1 % with respect to the crude *C. grandis* leaf extract. CNF gained higher glucose adsorption capacity of 16.6% (200 mM) than the crude extract. Glucose uptake percentage of CNF was increased by 48.28% in 5 mM and 1.6% in 25 mM glucose concentration when compared to the crude *C. grandis* leaf extract. The results revealed that the *in vitro* antidiabetic activity in terms of α - amylase, α - glucosidase inhibitory potential, glucose uptake and glucose adsorption capacity of CNF was enhanced compared to the crude aqueous leaf extract of *C. grandis*.

Acknowledgement: Financial assistance AHEAD DOR-15

Key Words: Antidiabetic activity, *Coccinia grandis*, aqueous extract, alginate nanoformulation

Bioconversion of bioethanol from coconut palm products using *Saccharomyces cerevisiae*

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This study was aimed to determine the efficient coconut palm products for bioethanol production and to optimize the conditions for fermentation to enhance the yield. Different coconut palm products, such as mature leaves (old leaves), young leaves (green leaves), young fruit fibre (kurumba), roots and husk fiber were used as substrates. These coconut palm products were inoculated with *Saccharomyces cerevisiae* (baker's yeast- 2g/L) in the fermentation media (100ml, 8° Brix, Waste extract: distilled water = 1:3) composed of 10 g/L yeast extract, 10 g/L KH₂PO₄, 2 g/L (NH₄)₂SO₄, 2g/L peptone and 0.5 g/L MgSO₄•7H₂O and allowed for fermentation for 24h at 30°C and 100 rpm. Significantly higher quantities of bioethanol were produced by young leaves (green leaves) and husk fibres. However, coconut husk fiber was selected as a bioethanol source, for further studies due to its abundance and availability in the farms and the lengthy period of natural degradation in the farms that leads to the development of different diseases caused by mosquitoes and emission of methane gas. The conditions for fermentation of coconut husk fibre using *Saccharomyces cerevisiae* were optimized sequentially by changing one factor at a time while keeping the other variables constant. After the optimization of fermentation time (3 days), amount of coconut husk fiber (12.5g /100ml), pH of the media (4.8), solution (V): air space (V) ratio (1:1.3), rotation speed (100rpm) and temperature (30°C), bioethanol yield was significantly increased by 6.6 times than the non-optimized conditions.

Key Words: Bioconversion, Bioethanol, Coconut palm products, *Saccharomyces cerevisiae*

Evaluation of antibacterial activity of polyphenol rich fraction of *Moringa oleifera* Lam at flowering stage against bacterial strains causing wound infections

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Moringa oleifera (Moringaceae) is a medicinally important plant found in tropical countries. Antimicrobial, antioxidant and antidiabetic activities of polyphenols of *M. oleifera* have been reported previously. However, the antibacterial activity of different polyphenol extracts of *M. oleifera* at flowering stage has not been determined previously. This study investigates the antibacterial activity of polyphenol rich fractions of *M. oleifera* leaves at the flowering stage against bacterial strains causing wound infections. Fresh leaves of *M. oleifera* were extracted to methanol (FM) and dry leaves were extracted to methanol (DM), distilled water (DDW) and hydro-alcoholic (DHA) solution. Total polyphenol and flavonoid content of each fraction were quantified using gallic acid (GA) and quercetin (QE) as the standards respectively. Different concentrations of crude aqueous extract and the polyphenol rich fractions (FM, DM, DDW and DHA) were evaluated for antibacterial activity using agar well diffusion method against ATCC reference strains of *Streptococcus aureus* (25923) and *Staphylococcus pyogenes* (19615). Each concentration performed in triplicates. Gentamycin (0.3%w/v) and distilled water were used as the positive and negative controls respectively. Total polyphenol contents and flavonoid contents of crude aqueous extract and the polyphenol rich fractions (FM, DM, DDW and DHA) were 0.14, 10.21, 11.23, 2.56 and 3.09 mg GAE/g and 4.65, 21.98, 42.26, 8.79 and 27.05 mg QE/g respectively. All four polyphenol enriched fractions at concentrations of 500 and 1000 mg/g were demonstrated antibacterial activity against *S. aureus* and *S. pyogenes*. The FM showed highest antibacterial activity against both *S. aureus* (25.8mm±0.2) and *S. pyogenes* (12.4mm±0.2). *S. aureus* was sensitive to FM at concentrations ranged from 62.5-1000 mg/ml. DM had highest values for total polyphenol (11.23mg GAE/g) and flavonoid content (42.26 mg QE/g). In conclusion, different polyphenol extracts obtained at flowering stage of *M. oleifera* possesses antibacterial activity against *S. aureus* and *S. pyogenes*.

Keywords: Antibacterial activity, *Moringa oleifera*, polyphenol content, flowering stage

Total phenolic, flavonoid contents and *in-vitro* antioxidant activity of a novel formulation incorporating *Cinnamomum zeylanicum* Blume and *Elettaria cardamomum* L

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Cinnamomum zeylanicum (Kurundu) and *Elettaria cardamomum* (Enasal) have many medicinal uses in Ayurveda. Study aimed to develop a novel formulation with cinnamon and cardamom and evaluate total phenolic (TP), flavonoid (TF) contents and *in-vitro* antioxidant activity. Cinnamon leaves, barks and cardamom seeds were collected, oven dried and powdered. Each was incorporated in 1:1:1 proportion. Aqueous extracts of each ingredient and product were obtained by hot Soxhlet extraction and cold maceration and freeze-dried. TP and TF contents of each were determined using Folin-Ciocalteu assay and aluminium chloride colorimetric method respectively. Antioxidant activity was evaluated using 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS) assays. Results were analyzed with one sample t-test using SPSS 25. Tests were done in triplicate and results were expressed as average standard deviation. It was revealed that TP contents of hot extracts of cinnamon leaves, barks, cardamom and formulation were 0.28±0.11, 0.24±0.02, 0.25±0.12 and 0.42±0.03 while TP values of cold extracts were 0.17±0.01, 0.12±0.00, 0.16±0.10 and 0.41±0.23 Gallic acid equivalents/g of extract. TF contents of hot extracts of cinnamon leaves, barks, cardamom and formulation were 0.06±0.02, 0.04±0.02, 0.02±0.01 and 0.08±0.06 while TF values of cold extracts were 0.01±0.00, 0.01±0.01, 0.02±0.00 and 0.02±0.01 Quercetin equivalents/g. DPPH radical scavenging percentages of hot extracts of cinnamon leaves, barks, cardamom and formulation were 220.90±0.65, 205.49±0.13, 192.02±0.21 and 251.15±6.21 while values of cold extracts were 213.50±0.21, 192.11±0.06, 160.19±0.03 and 234.89±5.13. ABTS radical scavenging percentages of hot extracts of cinnamon leaves, barks, cardamom and formulation were 55.33±1.29, 75.90±0.24, 62.11±4.11 and 80.38±3.35 while values of cold extracts were 42.00±0.37, 56.47±1.11, 60.22±0.76 and 59.67±2.86. Results indicated that formulation has high TP, TF and antioxidant contents than individual ingredients. It was also revealed that hot extracts have high TP, TF contents and antioxidant capacity compared to cold extracts (p<0.05). It was concluded that the hot infusions of the novel formulation have promising antioxidant activity to use against pathological states of oxidative stress.

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Key Words: Antioxidant activity, Formulation, *C. zeylanicum*, *E. cardamomum*

Comparison of Starch Degradation Ability of Recombinant *Escherchia coli* Containing Candidate Genes for Alpha Amylase Enzyme

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Starch hydrolysing enzymes have attracted great attention in starch related industries. They are widely used to produce numerous products which have high demand in the current world. *Thielaviopsis ethacetica* is a soil born fungi which has been identified to exhibit high enzyme activity on starch substrates and would be an ideal candidate for production of alpha amylases. Therefore, an attempt was made to clone alpha amylase gene from *Thielaviopsis ethacetica*. cDNA clones containing putative amylase gene in pH6HTN His6HaloTag® T7 expression vector system have been isolated and transformed into *E. coli* JM109 as the compatible host for protein expression. The starch hydrolysing ability of recombinant clones were compared with native fungi of *T. ethacetica*. Starch hydrolysing ability of selected clones were detected by treating plates with 10 mL of lugose Iodine solution (5 mM I₂ and 5 mM KI). Five recombinant clones (C3, C5, C6, C7 and C8) were selected and analysed for total starch hydrolysing assay. Enzyme activities were calculated in milli units (mU) and compared with the native fungal source. Native fungi have shown its maximum enzyme activity of 58.82 mU at 72hrs incubation time. Clones C3, C5, C6 and C8 have obtained their maximum enzyme activities of 38.65, 39.22, 28.01 and 10.08 in mU after 168hrs and Clone C7 has shown a maximum activity of 133.33 after 192hrs. At 168hrs, C7 has also shown 80.11 mU of total enzyme activity. Therefore, C7 has shown their capability in high production of starch hydrolysing enzymes over other recombinant clones and feasibility studies will be carried out to optimize all necessary conditions in the production of starch hydrolysing enzyme in C7 clone to meet the industrial demand effectively and efficiently.

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Keywords: Enzyme activity, Recombinant *E. coli*, Starch hydrolysing enzymes, *Thielaviopsis ethacetica*

Antibacterial activity of green synthesized silver nanoparticles using aqueous leaf extract of *Ophiorriza mungos*

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An eco-friendly mosquito repellent fabric using silver nanoparticles (AgNPs) biosynthesis by natural ingredients recently made considerable attention. Two commensal bacteria *Staphylococcus epidermidis* and *Staphylococcus hominis* are reacting with sweat to produce body odour, leading to the attraction of mosquitoes. The present study was on green synthesis and characterization of AgNPs using *Ophiorriza mungos* and the study of antibacterial activity. The preparation of AgNPs was carried out by adding 100 ml aqueous leaf extract of *O. mungos* dropwise to a 100 mL of silver nitrate solution (20 mM). Characterization of AgNPs was carried out by UV–Visible spectroscopy, Fourier Transform–infrared spectral analysis (FTIR), scanning electron microscopy (SEM), and Energy-dispersive spectra (EDS). The antimicrobial activity of AgNPs was investigated using the well diffusion method against *S. epidermidis* and *S. hominis*. Distilled water was used as the negative control. The formation of AgNPs was evident by the change of colourless solution into yellow colour. Further, the absorption band appeared at 448 nm due to surface plasmon resonance of AgNPs (UV spectra) confirmed the formation of AgNPs. The spherical morphology of AgNPs is indicated by the SEM images. The reduction of functional groups –OH, –C=C, C–N, and =CH observed on the FTIR spectra was indicative of the formation of AgNPs. The EDS revealed that AgNPs were in their pure form. The well diffusion method showed that AgNPs have a marked antibacterial activity with the zone of inhibition of 21±1 mm for *S. hominis* and 20.33±0.58 mm for *S. epidermidis* (100% v/v original solution). There was no antibacterial activity observed for the aqueous leaf extracts against *S. hominis* and *S. epidermidis*. The green synthesized AgNPs using aqueous leaf extract of *O. mungos* showed considerable antibacterial activity over aqueous leaf extract of *O. mungos*, against *S. hominis* and *S. epidermidis*.

Key Words: Antibacterial activity, Green synthesized, Silver nanoparticles, *Ophiorriza mungos*

Association of serum leptin with BMI, body fat percentage and Q223R leptin receptor gene polymorphism in an adult female population

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Elevated serum leptin level is present in most obese people and is considered as a state of leptin resistance. Genetic variations of leptin receptor (LEPR) gene including Q223R single nucleotide polymorphism (SNP) is considered as one cause of leptin resistance. The aim of this descriptive cross-sectional study was to determine the association between serum leptin levels with obesity and Q223R SNP in a healthy female population. Two hundred and forty female subjects were recruited (26 ±4 years old). Anthropometry and bio-impedance analysis methods were used to determine the body mass index (BMI) and body fat percentage (%BF). Serum leptin level was determined using enzyme-linked immunosorbent assay (ng/ml). Restriction fragment length polymorphism-polymerase chain reaction was used for genotyping (QQ- wild type homozygous, QR- heterozygous, RR- mutated homozygous). Student t-test and Analysis of variance statistical tests were used where appropriate. Binary logistic regression analysis was used to determine the associations [dependent variable– serum leptin, independent variables– genotypes, BMI and %BF, odds ratio (OR), 95% confidence interval (CI)]. Mean serum leptin level of obese group (13.88 ±6.31) was significantly higher compared to the non-obese group (7.69 ±5.52) (p=0.00). Serum leptin level between 3 genotypes were not significantly different (p=0.43) (QQ= 11.09 ±6.38, QR= 8.68 ±6.43, RR= 8.82± 6.04). According to the binary logistic regression, having BMI ≥25kgm⁻² (OR= 1.74, 95% CI= 1.06 – 2.86, p= 0.03), BF%≥ 35% (OR= 1.35, 95% CI= 1.04 – 1.76, p= 0.03) and the presence of R allele (QR and RR genotypes) (OR= 2.64, 95% CI= 1.07- 6.52, p= 0.04) are associated with increased risk of elevated serum leptin. Based on the findings of the current study, obesity and Q223R SNP are associated with increased risk of elevated serum leptin levels.

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Key Words: Serum leptin, BMI, Body fat percentage, Q223R

Association of Anthropometric Measurements with Echocardiographic Parameters of Chronic Heart Failure Patients in Sri Lanka – Preliminary Study

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Obesity is an important contributor to many cardiovascular diseases and has been associated with abnormalities in cardiac contractile function. The aim of this study was to assess the association of anthropometric measurements with echocardiographic parameters of chronic heart failure (CHF) patients among a selected population of Sri Lanka. An analytical cross sectional study was carried out with 76 consecutive CHF patients who came to the cardiology clinic at district hospital Kalutara. Ethical clearance was obtained from the Ethics Review Committee of the Faculty of Medical Sciences, University of Sri Jayewardenepura. Their body mass index (BMI), waist circumference (WC), waist-to-hip ratio (WHR), mid upper arm circumference (MUAC) and mid-thigh circumference (MTC) was measured as per WHO guidelines and the data analyzed using SPSS. 2D echocardiography was performed in all patients and obtained values for ejection fraction (EF), left ventricular end diastolic diameter (LVEDD) and left ventricular end systolic diameter (LVESD). Both genders showed increased risk of CHF with significant correlations ($P < 0.0001$) in BMI and WC. WHR correlated significantly ($r=0.47$, $p=0.002$) with LVEDD only in males whereas it correlated significantly with WC in both genders. When receiver operating curves (ROC) were plotted, it showed optimal sensitivity and specificity for BMI and WC with severity of CHF. The NT pro BNP levels positively correlated with the MUAC and with EF and LVEDD. MUAC, MTC and WC positively correlated with severity of CHF. Study showed an increased risk of having CHF in obese individuals when compared to individuals with normal BMI and increased WC, WHR and MUAC are highly significantly associated with higher risk of CHF. Therefore WC and WHR have the potential to be used as markers to evaluate the CHF risk levels.

Keywords: Anthropometric parameters, Ejection fraction, Ventricular function, Left ventricular end diastolic diameter

Selected markers of gluconeogenesis and its association with the severity of organophosphate and carbamate poisoning

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Acute anticholinesterase pesticide poisoning leads to hyperglycaemia, where gluconeogenesis is postulated as a key mechanism. Aim of this study was to assess the correlation between selected gluconeogenic makers Phosphoenol pyruvate carboxy kinase (PEPCK) and glucagon] and severity of poisoning. This cross-sectional study was carried out in 124 acute organophosphates (50) and carbamate (74) poisoned patients (age: 18- 60 years), who were admitted to Teaching Hospital Anuradhapura, within 24 hours of poisoning. Severity of poisoning was measured on admission using Peradeniya organophosphorus poisoning scale (POP) and RBC cholinesterase level. Blood samples were collected at the time of discharge after 8 – 10 hours fasting. Fasting blood glucose (FBS), glucagon and PEPCK were measured. Data analysis was done using SPSS. Mean age of the total population was 33(±13) years and the majority was males (68.5%). Mean glucagon of mild moderate and severe groups were 15.4(±9.4) pg/mL, 39.2(±30.2) pg/mL and 114.9(±86.7) pg/mL. It was 1.79(±1.6) pg/mL, 2.5(±2.3) pg/mL and 2.0(±1.7) pg/mL for PEPCK respectively. Mean FBS and glucagon at discharge were significantly different between mild - moderate groups, moderate- severe and mild – severe groups (p<0.05). Significant mean difference was not observed between any of the groups based on severity with regard to PEPCK. However, PEPCK were elevated in mild (85%), moderate (94 %) and in all severely poisoned patients. Severity of poisoning measured according to both methods was significantly correlated with FBS and glucagon while PEPCK was significantly correlated with the severity of poisoning according to RBC cholinesterase method. Analysis of additional gluconeogenic enzymes is recommended to confirm the present finding.

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Key Words: Gluconeogenesis, Organophosphate, Carbamate, Poisoning

“Natural Products and Microbes for Health and Sustainability”

Poster presentations

Prevalence of dysglycemia and its associations with age and body mass index among semi urban community dwelling adults in Galle, Sri Lanka

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Dysglycemia includes prediabetes and diabetes. The gender wise association of dysglycemia with increasing age and BMI has not been explored previously in the Sri Lankan population. The objectives of the present study was to study the gender wise prevalence of glycemic status, its associations with age and body mass index (BMI) and to determine an optimal cutoff of BMI to assess the risk of dysglycemia in semi urban community dwelling adults in Galle, Sri Lanka. This was a community based cross-sectional study, involving 1120 healthy individuals (females; 803) of age 30-60 years in randomly selected divisional secretariat areas of semi urban localities in Galle, Sri Lanka. Prevalence of dysglycemic state (FPG>100 mg/dL) and its associations with age and BMI in both genders were estimated. The association between gender and glycemic status in different BMI and age groups were estimated. Receiver operating characteristic (ROC) curves were developed for BMI values of both genders separately as a risk factor of dysglycemia and the area under the curve (AUC) values were obtained for both genders. The optimal cut-off point of BMI was determined using Youden index. Prevalence of prediabetes and diabetes of females were 25.3% and 16.4% and of males were 26.2% and 17.4% respectively. Dysglycemia showed a significant positive correlation with age in both genders and significant positive correlation with BMI in males ($p<0.05$). Aging (OR=1.05, CI 1.02–1.08, $p<0.001$) and increasing BMI (OR=1.10, CI 1.05–1.15, $p<0.001$) of males and aging (OR=1.04, CI 1.02–1.06, $p<0.001$) of females are significantly associated with dysglycemia. The optimal cut-off point of BMI for males was 22.86 kg/m² (AUC 0.651, sensitivity 76.6%, specificity 53.9%) to determine the risk of dysglycemia. The prevalence of dysglycemia either in the form of prediabetes or diabetes in male population is higher than that of the female population in this cohort. An increase in age in both genders and BMI in males are significantly associated with dysglycemia. The cut-off value of BMI>22.86 kg/m² could be used as an acceptable clinical parameter to determine the risk of dysglycemia by means of overweight among male population in Galle, Sri Lanka. Since the AUC value of BMI for females is almost 0.5 (0.51), it was not used for further to estimate sensitivity or specificity.

Acknowledgement: Financial assistance by NRC grant (17-029)

Key words: Prevalence, Dysglycemia, Semi urban community, Adults

Relationship between Antioxidant and Antibacterial Properties of Flowers, Leaves and Stems of Tanner’s Cassia (*Senna auriculata*) grown in Sri Lanka.

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Medicinal plants have been widely used around the world to cure many diseases because their constituents show satisfactory efficacies. For example, the South-Asian herbal plant, Tanner’s Cassia(*Senna auriculata*) contains many antioxidants/phytochemicals such as flavonoids and phenols which cure diabetes & conjunctivitis. While unstable free radicals abstract electrons from other stable compounds to attain stability, those compounds become free radicals and develop an oxidative stress which can cause cell death/diseases. But plant antioxidants with enough electrons can stabilize the unstable free radicals present in our body to avoid oxidative-stress while being stable. According to studies, Tanner’s Cassia expresses broad-spectrum antimicrobial activity against standard-strains. This research was conducted to determine any relationship between antioxidant capacity and antibacterial properties of Tanner’s Cassia extracted to a series of solvents. Initially, phytochemicals in powdered Tanner’s Cassia plant parts such as leaves, flowers & stems from Negombo-Sri Lanka, were extracted using 3-polar-solvents; 80% ethanol, 80% methanol and distilled water. After the roller-mixer solvent-extraction by cold-maceration, to determine the antioxidant capacities of extractions, antioxidant assays such as total-flavonoid-content (TFC), total-phenolic-content (TPC), total-antioxidant-content(TAC), ferric-reducing-antioxidant-power (FRAP) assays were carried out. Additionally, chemical compounds such as ABTS (2,2’-azino-bis(3-ethylbenzothiazoline-6-sulfonic-acid)) and DPPH (2,2-diphenyl-1-picrylhydrazyl) were used for radical scavenging assays. Spectrophotometric methods were used in these tests to determine the unknown phytochemical concentrations. Antibacterial properties against standard strains; *Staphylococcus aureus* and *Escherichia coli* were measured by Minimum Inhibitory Concentration(MIC). Statistical analysis was carried out using IBM-SPSS-Statistics-21 and Microsoft-Excel softwares. It was confirmed that strong positive correlations of $r=+0.870$ (leaves), $r=+0.896$ (flowers) and $r=+0.987$ (stems) present between antioxidant capacities of TFC & TAC. Moreover, strong positive correlations of $r=+0.830$ (leaves), $r=+0.335$ (flowers) and $r=+0.984$ (stems) present between TPC & TAC of the solvent extracts indicating the abundance of flavonoids and phenols in this plant. A negative correlation of $r=-0.428$ between TAC & MIC shows an indirect proportionality. Studies show that MIC is indirectly proportional to antibacterial activity. Therefore it was confirmed that antioxidant-capacity (TAC) is directly proportional to the antibacterial activity of Tanner’s Cassia plant. Moreover 80%-methanol flower-extracts showed a significant impact on above correlations compared to other extracts. Further improvements such as, antimicrobial-gradient-method to test the combined effect of multiple plant extracts, can bring this research into a turning point of a new drug development against common diseases.

Keywords: Tanner’s Cassia, Phytochemicals, Antioxidant capacity, Antibacterial properties, Flavonoids, *Escherichia coli*.

Isolation of cytochrome monooxygenase (*cyp52*) gene from *Aspergillus* sp. MM1, *in-silico* protein modelling and molecular docking simulation of hexadecane binding

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Cytochrome P450 proteins (Cyp52) have been proven to degrade *n*-hexadecane in yeasts. It was hypothesized that *Aspergillus* sp. MM1 may also utilize Cyp52 for *n*-hexadecane degradation. Therefore, its putative *cyp52* gene was isolated and the encoded protein was verified for *n*-hexadecane binding. A BLAST search of NCBI non redundant protein database with *Candida maltosa* Cyp52A3, resulted in 47 Cyp52 sequences (>40% sequence identity; lowest E score =2e-100) from *A. flavus* and *A. oryzae*. Of these, seven sequences having 100% amino acid identity to the Cyp52A3 signature motifs were utilized for PCR primer designing. The PCR-amplified *cyp52* gene (1689 bp) (GenBank Acc. MW995970) encoded a protein of 509 amino acids. Computational 3D model of the deduced amino acid sequence of *Aspergillus* sp. MM1 Cyp52 and *C. maltosa* CYP52A3 were predicted using I-TASSER server. Best models were selected based on protein structure validation tools, VERIFY3D, PROCHECK and ERRAT. The structures for haem (enzyme cofactor) and *n*-hexadecane were retrieved from Protein Data Bank and geometrically optimized using ORCA 4.2.1 program. The docking simulations were carried out using AutoDock Vina and visualized by BIOVIA Discovery Studio. Haem was observed to covalently bind to the invariant cysteine and other residues of the heme pocket. Cyp52:heme complex was then bound to the terminal carbon atom of hexadecane via pi-alkyl interaction to the heme. Twentynine active site residues were shared *Aspergillus* MM1 Cyp52 and *C. maltosa* CYP52A3 suggesting that the *Aspergillus* sp. MM1 Cyp52 is a potential medium-chain alkane monooxygenase. This finding can support future biotechnological applications in bioremediation of petroleum hydrocarbon pollution.

Acknowledgments: Financial assistance grant No: AP/3/2/2014/RG/12 University of Colombo.

Key words: Cytochrome monooxygenase, *Aspergillus*, *In-silico* protein modeling, Molecular docking

Effect of roasting on antioxidant activity of methanol extracts of seeds of *Vigna mungo* seeds cultivated in Sri Lanka

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Antioxidants present in plant materials are reported to reduce the risk of oxidative stress and development of diseases including diabetes, cardiovascular diseases and cancers. In Sri Lanka seeds of *Vigna mungo* (black gram/Undu) are commonly consumed after processing in different methods such as roasting. Aim of the present study was to compare the antioxidant potential of methanol extract of roasted and raw black gram cultivated in Sri Lanka. *Vigna mungo* seeds of Anuradha variety, were collected from field crops research and development institute, Mahailuppallama. Methanol extracts (20%) of finely ground raw and roasted (160 °C, 30 minutes) unpolished black gram were analyzed in triplicates for DPPH free radical scavenging, nitric oxide scavenging activity, total phenolic content (TPC) and total flavonoid content (TFC). Thin layer chromatography was performed to separate different phenolics. DPPH scavenging activity of raw seed sample ($23.8 \pm 0.2 \mu\text{g/mL}$) was slightly higher ($p > 0.05$) than the roasted seed sample ($23.78 \pm 0.2 \mu\text{g/mL}$). The nitric oxide scavenging activity of raw seed samples ($3.7 \pm 0.04 \text{ mg GAE/g}$) was significantly higher ($P < 0.001$) than the roasted seed sample ($2.4 \pm 0.01 \text{ mg GAE/g}$). TPC of the raw sample was $1.1 \pm 0.01 \text{ mg GAE/g}$ and for the roasted sample it was $0.8 \pm 0.02 \text{ mg GAE/g}$. TFC for raw and roasted sample were $0.058 \pm 0.009 \text{ mg QE/g}$ and $0.001 \pm 0.0 \text{ mg QE/g}$ respectively. A significant difference ($P < 0.05$) between raw and roasted samples was observed in relation to TPF and TFC values. Thin layer chromatography results indicated that the methanol extracts of the both raw and roasted seed sample contain low amounts of gallic acid and quercetin. The present study reveals that the roasting affects the content of phenols and flavonoids in black gram and therefore reduces the total antioxidant potential in roasted seeds.

Key Words: Roasting, Antioxidant activity, Methanol extracts *Vigna mungo*

Lactate dehydrogenase (LDH) enzyme assay in cytotoxicity testing and the interferences caused by Triphala

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Measuring the activity of cytoplasmic Lactate dehydrogenase (LDH) enzyme released by damaged cells to the cell culture supernatant is a widely accepted method of evaluating the cytotoxicity of pharmacological compounds. The principle underlying relies on two hypotheses: the testing drug has zero interference with the enzyme activity, and the test drug itself has no UV absorbance ability. Triphala (TPL) is a renowned polyherbal formulation used in ayurvedic medicine. The objective of the study is to identify interferences caused by TPL in LDH assay. LDH assay was conducted after incubating rhabdomyosarcoma cells (RD) with TPL (10-1000 μ g/ml) for 24 hours and the UV absorbance of same concentrations of TPL before and after incubating at 37°C for 24 hours, were measured. The Impact of TPL on LDH activity was determined using fresh human serum after incubating 15 minutes with a concentration series (100-750 μ g/ml) of TPL. Assays were done in technical triplicates. EC50 value was calculated considering the percentage cell viability and TPL concentration. The LDH activity declined in both cell lysate and supernatant in a dose-dependent manner. TPL showed a concentration-dependent increment in UV absorbance (absorbance of 500 μ g/mL of TPL was 1.144) which inclined significantly after incubation ($p < 0.05$). Negative control (without TPL) showed the highest enzyme activity compared to TPL treated serum. A percentage inhibition of 29.54 \pm 8.66 (mean \pm SD) observed in serum incubated with 750 μ g/mL of TPL. To minimize the interferences, percentage cell viability was determined by considering the enzyme activity in the cell lysates relative to the negative control, and the EC50 value was 871.0 \pm 57.9 μ g/mL. TPL significantly affects the measurement of LDH activity due to its high UV absorbance and ability to inhibit LDH activity. Therefore, before conducting LDH assay, interference by the testing compound should be evaluated.

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Keywords: LDH, Triphala, RD cells, UV absorbance

Biodegradation of selected agro wastes using an oyster mushroom, *Pleurotus ostreatus*

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This study was aimed to determine the biodegrading capacity of oyster mushroom *Pleurotus ostreatus* on corn husk, coffee husk and peanut hull naturally from 0 to 45 days by assessing lignocellulosic compounds, extracellular enzyme production, protein content, reducing sugar content, pH, degraded weight and ash content on every 15 days interval. Lignin content significantly decreased from 14.28 to $2.92\% \pm 0.36$ in corn husk, from 29.7 to $16.69\% \pm 0.005$ in coffee husk and from 32.89 to $28.61\% \pm 0.200$ in peanut hull. Cellulose content showed a significant decrease in corn husk from 44.543 to $18.967\% \pm 0.072$ in coffee husk, from 25.84 to 8.399% and in peanut hull from 35.768 to $25.11\% \pm 0.006$, in 45 days. Hemicellulose content was decreased from 39.65 to $23.65\% \pm 0.500$ in corn, from 23.78 to $12.36\% \pm 0.100$ in coffee and from 19.67 to 12.56 % in peanut hulls, in 45 days. MnPase enzyme activity was optimum on the 30th day in all the three substrates. Endo-1,4-β-D- Glucanase activity and Exo-1,4-β-D- Glucanase activity was significantly higher on the 45th day in all three substrates. Percentage of protein content was increased from 0.688 to $2.92\% \pm 0.002$ in corn husks, 1.02 to $2.67\% \pm 0.005$ in coffee husks and 2.83 to $3.92\% \pm 0.001$ in peanut hulls in 45 days. Reducing sugar content was decreased from 0.22 to 0.005% in corn husks, from 0.046 to 0.006% in coffee husks and from 0.008 to -0.012% in peanut hulls. pH showed varying trends in all the 3 substrates used. Ash content of the substrates significantly decreased from 4.5 to $0.876\% \pm 0.17$ in corn husk, from 6.8 to 1.78% in coffee husk and from 6.1 to 4.91% in peanut hull, in 45 days. Degraded weight of all the substrates decreased significantly from the beginning towards the end. The degradation capacity of *P.ostreatus* was significantly higher in corn husks (61%) in 45 days, than the other waste.—Therefore, *P.ostreatus* mushroom could be recommended as a natural degrader of corn husk left over.

Key words: Agro-wastes, Biodegradation, Corn husk, Glucanase, *Pleurotus ostreatus*

Prevalence of hypertension and high body mass index among rural non-endemic residents in Sri Lanka

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Although non-communicable diseases (NCDs) are more prevalent in urban areas, now it is increasing among rural populations too. Hypertension and obesity is identified as main risk factors to increase NCDs. Since average literature is presented regarding the hypertension and obesity in urban areas, related research studies among the rural Sri Lankan communities are still limited. The aim of this study was to assess the prevalence of hypertension and obesity among adult rural residents in Sri Lanka. A descriptive cross-sectional study conducted among 131 previously healthy residents in Hanguranketha Divisional Secretariat of Central Province in Sri Lanka. We used convenient sample technique and informed consent was obtained prior to the collection of the data. For the assessment of BMI, height and weight measurements were taken using standard protocols and Omron HEM 7120 digital BP apparatus was used to measure the BP. The mean age of the cohort was 47.5713.433 (21-78) years, 58.8% (n=77) of the participants were female. According to body mass index (BMI) categorization prevalence of overweight (25-29.9 Kg^m-²) and obese (30Kg^m-²<) were 31.3% (n=41) and 11.5% (n=15) respectively. Around 22.9% (n=30) had raised BP (Systolic BP [SBP] 140mmHg or Diastolic BP [DBP] 90mmHg) than the normal level. Although significant difference was not shown between BMI and the hypertension group, occurrence of hypertension is 1.3 times greater among overweight/obese adults rather than adults with normal/under-weight BMI. BMI is a considerable health factor related to the developing of hypertension among in adult rural residents.

Key Words: Prevalence, Hypertension, High body mass index, non-endemic residents

***In vitro* antibacterial activity of selected marine weeds on selected bacteria**

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Certain marine products have been found to exhibit significant antibacterial effects against number of pathogens. *In vitro* antibacterial activity of ethanolic extracts of fresh and dried material of *Enhalus* spp., *Sargassum* spp., *Turbinaria* spp. and *Halimeda* spp. were evaluated. The marine weeds were collected from North sea of Jaffna District, Sri Lanka. Fresh and dried ethanol extracts of the four marine weeds were evaluated for activity against 4 bacterial species namely *Staphylococcus aureus* ATCC 29213, *Enterococcus faecalis* ATCC 29212, *Pseudomonas aeruginosa* ATCC 27853 and *Escherichia coli* ATCC 25922 by nutrient agar well diffusion method. Twenty grams of properly washed fresh and dried and milled leaves of were soaked in 150 mL of absolute ethanol (99.98 %) for 5 successive days separately at room temperature (31 ± 3 °C). Solvent was removed by rotating evaporator and crude extracts were used for evaluation of antimicrobial activity. Plates were incubated for 48h at 37 °C and the inhibition zone that formed around the well were measured (mm). Triplicates were maintained for each experiment. Fresh ethanol extracts of *Turbinaria* spp. showed antibacterial activity against all four types of bacteria; *Staphylococcus aureus* (13.2 ± 0.249 mm), *Enterococcus faecalis* (12.5 ± 0.166 mm), *Pseudomonas aeruginosa* (11.5 ± 0.221 mm) and *Escherichia coli* (13.5 ± 0.166 mm) while fresh ethanol extracts of *Enhalus* sp showed activity against *Staphylococcus aureus* (14.1 ± 0.179 mm), *Enterococcus faecalis* (14.4 ± 0.163 mm) and *Escherichia coli* (14.5 ± 0.166 cm). Ethanol extract of *Enhalus* sp showed more antimicrobial activity than *Turbinaria* spp. Dried ethanol extracts of all plant samples did not show antibacterial activity. Streptomycin ($100 \mu\text{g mL}^{-1}$) and sterile distilled water were as the positive and negative control respectively. The results indicate that there is scope for using these marine weeds as a source of antimicrobial substances.

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Key Words: *In vitro*, Antibacterial activity, Marine weeds, Bacteria

Assessment of risk Factors of Gestational Diabetes Mellitus among Pregnant Mothers attending Antenatal Clinics in Matugama MOH Area

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Gestational diabetes mellitus (GDM) is a subtype of diabetes that occurs for the first time during pregnancy as a result of insulin resistance produced by the placental hormones. GDM is associated with serious life threatening complications both in the mother and the offspring and, the prevalence of GDM in 2014 was 13.9% in Sri Lanka. Thus, the present study aimed at assessing the presence of GDM related risk factors among a selected population of pregnant mothers. This was a descriptive cross sectional study conducted in year 2018 among eight antenatal clinics in Matugama MOH area. Total of 150 pregnant mothers < 20 weeks of gestation and the clinics were selected using convenience sampling. Ethical approval was obtained from the Ethics Review Committee, FMS, USJ. All the data were collected using an interviewer administered questionnaire and analyzed using SPSS version 23.0. Among the participants, 34.0% (N=51) were primigravida. Mean gestational age (\pm SD) was 12 \pm 5 weeks. Maternal overweight (booking BMI \geq 25 Kgm⁻²) was the most prevalent risk factor for GDM identified among the participants (28.7%, N=43) followed by age \geq 35 years (20.7%, N=31). Further, maternal obesity (booking BMI \geq 30 Kgm⁻²) was observed in 6.7% (N=10), history of delivering large babies (birth weight \geq 3.5Kg) in 10.7% (N=16), family history of diabetes in first degree relatives in 24.0% (N=36), history of GDM in 4.7% (N=7), past history of pregnancy induced hypertension in 4.0% (N=6), history of miscarriages/ still births in 15.3% (N=23) and long term use of Antipsychotics/ Antiepileptic drugs in 3.3% (N=5) were also seen among the participants. At least one risk factor was evident in 28.0% (N=42) while two in 20.7% (N=31) and \geq 3 risk factors were evident in 14.0% (N=21). Early identification and proper education of GDM related risk factors is necessary to overcome development and complications associated with GDM in current and index pregnancies.

Key words: Pregnant mothers, gestational diabetes mellitus, risk factors

***In vitro* study on skin whitening and anti-aging potential of leaves of *Moringa oleifera* Lam. (Murunga)**

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Moringa oleifera which is known as the “Miracle Tree” is one of the most useful tropical trees. The leaves of this plant are reported to have many biological activities. However, its skin whitening and anti-aging potential are not well documented to date. This study evaluated skin whitening and anti-aging potential of leaf of *M. oleifera*. Water extract of dried leaf powder of *M. oleifera* (2 g in 100 ml boiling water for 10 mins) was used in this study. Skin whitening and anti-aging properties were evaluated using anti-tyrosinase (AT), anti-elastase (AE), anti-collagenase (AC) and anti-hyaluronidase (AH) activities *in vitro* (n=3 each). Quercetin (AT activity) and epigallocatechin gallate (EGCG: AC and AH activities) were used as the reference drugs. Further, selected phenolic compounds [(PC): arbutin, 4-hydroxybenzoic acid, epicatechin, quercetin, gallic acid, kaempferol, ferulic acid and catechin n=3 each] were quantified using High Performance Liquid Chromatography (HPLC). Results clearly showed that leaf of *M. oleifera* possesses skin whitening (AT: IC₅₀ 1.88±0.08 mg/ml) and anti-aging activities (AC: IC₅₀ 0.79±0.01 mg/ml; AH: 4.96±1.67% inhibition at 1.5 mg/ml) (except AE activity). Observed activities were moderate compared to the reference drugs (AT: IC₅₀ Quercetin 29.38±0.49 µg/ml; IC₅₀ EGCG: for and AC & AH: and 112.12±0.93 & 90.00±0.00 µg/ml respectively) used in the study. Further, it contains varying quantities of PCs (except Epicatechin, Quercetin and Ferulic acid) where arbutin was the highest (67154±295 µg/g of extract) and 4-hydroxybenzoic acid was the lowest (37±0.8 µg/g of extract) among studied. In conclusion, leaves of *M. oleifera* possess marked skin whitening and anti-aging properties with significantly high quantity of arbutin, a known skin whitening agent used in the cosmetic industry. Therefore, leaves of *M. oleifera* may have the potential in utilizing effectively in development of novel skin whitening and anti-aging cosmaceuticals.

Keywords: *Moringa oleifera*, Skin whitening, Anti-aging, Phenolic compounds

Bioethanol production from selected marine algae species

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Algae were identified as third generation carbon sources for bioethanol production. Hence, four different types of marine algae, *Sargassum* sp, *Turbinaria* sp, *Padina* sp and *Halimeda* sp were evaluated for its bioethanol production. The marine algae were collected from North sea of Jaffna District, Sri Lanka. Samples were cleaned half of the each plant species was dried and crushed to powder and another half was kept as fresh in refrigerator at 4°C. Samples were (30g) subjected to autoclave with distilled water (150mL) then removed the water and followed by 1% H₂SO₄ (150mL) to release the sugar. Fifty milliliter of fermentation medium contained (L⁻¹) 4g (NH₄)₂SO₄, 4g MgSO₄ and 8g KH₂PO₄ was added to treated samples and pH was adjusted to 5.0 with 0.1N NaOH. Fermentation medium was inoculated with 10mL of activated baker's yeast (50g yeast in 50g L⁻¹ sucrose solution) separately and incubated at room temperature (31±3 °C) and 100rpm in orbital shaker. Samples (50mL) were taken at different time intervals (24, 48 and 72h) and bioethanol activity was measured by ebulliometer. There was no bioethanol activity in all fresh algal samples but dried powdered samples of *Sargassum* sp (0.2%) and *Halimeda* sp (0.2) showed bioethanol activity at 48h of incubation. This study shows that above mentioned two seaweeds are potential sources for bioethanol production and these marine algae were selected for further analysis of bioethanol production.

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Key words: Bioethanol, Marine algae

Abstract No PP 12

Determination of antimicrobial activity of some selected plant species in Rubiaceae family

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Investigation of new antimicrobial agents has become one of the key aspects of today's world due to the continuous increase in resistance of pathogenic microbes. Sri Lanka being a biodiverse hot spot, has a rich collection of plants that can play a huge therapeutic role. The objective of this study was to identify such indigenous plants belong to the family Rubiaceae to investigate their undisclosed antimicrobial activities. The information on the Rubiaceae plants use in Ayurvedic treatments were collected from Ayurvedic doctors and fresh parts (leaves, roots, stem, and whole plant) of five such plant species *Knoxia zeylanica* (Ela ratmal), *Ophiorrhiza mungos* (Dathketiya), *Oldenlandia herbacea* (Wal Kottamalli), *Wendlandia bicuspidata* (Rawan Idala) and *Morinda umbellata* (Kiriwel) were collected and authenticated. The collected plant parts were cleaned, dried and crude extracts were prepared in a solvent mixture of dichloromethane and methanol (1:1, v/v). An antifungal activity of crude extracts was screened against *Aspergillus spp.*, *Rhizopus spp.* and *Penicillium spp.* by poison food technique. Also, an antibacterial assay was conducted against two Gram-positive bacteria *Staphylococcus aureus* (ATCC 25928), *Bacillus cereus* (ATCC 11778), and two Gram negative bacteria *Pseudomonas aeruginosa* (ATCC 9027) and *Escherichia coli* (ATCC 35218) using the agar well diffusion method. Among all the fungi, *Penicillium spp.* showed the significant antifungal activity for many crude extracts including, *K. zeylanica*, *O. mungos*, and *O. herbacea*. However, the crude extract of *O. mungos* (whole plant) has almost completely inhibited (98.56%) the growth of *Aspergillus spp.* at the concentration of 1 mg/ml giving a promising indication of the availability of strong antifungal compound/s in *O. mungos*. The crude extract (1mg per well) of *K. zeylanica* (root) showed the highest antibacterial activity, compared to the previously reported antibacterial activities of all selected plant parts, against *B. cereus* (16.2 ± 0.3 mm) and *S. aureus* (16.9 ± 0.3 mm). The results of this study showed that *K. zeylanica* and *O. mungos* could be potential candidates to search for anti-bacterial and antifungal compounds, respectively.

Key words: Antimicrobial activity, Rubiaceae family

Comparison of linearity and slope of the standard curves of two different chondroitin sulfate reference materials in dimethylmethylene blue assay

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Dimethylmethylene blue assay is used to quantify sulfated glycosaminoglycans in urine. Chondroitin sulfate polysaccharides have been routinely used as the reference materials for the assay, although majority of urinary glycosaminoglycans are oligosaccharides. The objectives of the present study was to compare the linearity and slope of the standard curves of two different standards in dimethylmethylene blue assay; chondroitin sulfate from bovine trachea with high degree of polymerization vs. chondroitin sulfate oligosaccharide with degree of polymerization of 12. Standard stock solutions [100 µg/ml] and serial dilutions were prepared using the two types of chondroitin sulfate standards. The assay was conducted in duplicates, using three standard-to-dye volume (µl) ratios (50:150, 20:180 and 10:190) on 96-well microplates. The absorbance was read at 520 nm, using a microplate photometer (Multiskan FC). At 20:180 standard-to-dye ratio, the standard curve of chondroitin sulfate from bovine trachea was linear up to standard concentration of 100 µg/ml, while the oligosaccharide showed linearity only up to 25 µg/ml. At 10:190 standard-to-dye ratio, both standard curves were linear up 100 µg/ml, but the slope of the standard curve of bovine chondroitin sulfate (0.0287 [g/100ml]⁻¹) was slightly lower than that of the chondroitin sulfate oligosaccharide (0.0312 [g/100ml]⁻¹). The results of the present study revealed that the degree of polymerization and source of the glycosaminoglycan reference materials may affect linearity and slope of the standard curves and hence parallelism and commutability of the reference materials in dimethylmethylene blue assay.

Acknowledgments: Financial assistance by University Research Grants of University of Sri Jayewardenepura (Grant Number: ASP/01/RE/MED/2019/44).

Key words: Standard curves, Chondroitin sulfate, Reference materials, Dimethylmethylene blue assay

Abstract No PP 14

Tea Catechin Incorporated Graphene based Bio-nanocomposites: *In Vitro* Analysis of their Antimicrobial Properties

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Tea catechins (TC) are polyphenols found in tea leaf extracts which exhibit protective effects against diseases such as cancer, diabetes, obesity, hepatitis, neurodegenerative diseases, and tooth decay. Their usage in the native form is limited due to low rate of absorption, low solubility, low bioavailability, and unstable nature. To overcome these problems several studies are underway to synthesize composites by modifying the properties of TC to develop effective therapeutic products. This study aims to synthesize novel bio-nano composites using TC and graphene-based nanomaterials and to assess their antimicrobial activity against some selected bacteria and fungi causing superficial skin infections. Composites were synthesized via the adsorption method. The adsorption capacity of each material was determined by measuring the remaining concentration of TC in the supernatant using UV-Visible Spectroscopy. The synthesized composites were characterized with Fourier-Transform infrared spectroscopy (FTIR) and X-Ray Diffraction (XRD). Antimicrobial activity of composites was assessed against *Staphylococcus aureus* (ATCC25923), *Escherichia coli* (ATCC25923) and clinically isolated *Candida albicans* using well diffusion method. Commercially available antimicrobial discs Clindamycin, Ciprofloxacin, and Fluconazole were used as positive controls. DMSO was used as the negative control. After incubation at 37°C for 24 hours, inhibition zone diameters were measured. Adsorption capacities of composites synthesized using graphene oxide (GO), reduced graphene oxide (RGO), and expanded graphite (EG) were 250 mg/g, 240 mg/g, and 158 mg/g, respectively. XRD peak enhancement around 20-24° and stretching of O-H at 3400 cm⁻¹ in FTIR confirm successful incorporation of TC. In diluted DMSO, all composites showed inhibitory activity against *S.aurries* and *E.coli* giving average zone diameters of 30mm and 29mm for the positive control and 20mm and 19mm for TC. No zones were observed for the negative control. GO, RGO and EG composites gave an average zone diameter of 16mm against *S.aurries* and 15mm, 15mm and 14mm against *E.coli*. This confirms slow & sustained release of TC. Composites were not active against the *Candida* species. TC/graphene bio-nano composites have a potential antibacterial activity against *S. aureus*, and *E. Coli*. The observed slow release of TC suggests the potential applications of them in pharmaceutical industry.

Key Words: Tea Catechin, Graphene based, Bio-nanocomposites, Antimicrobial Properties

Glucose-lowering activity of *Coccinia grandis* (L.) Voigt leaf extract encapsulated nanoliposomes in Wistar rats induced with diabetes mellitus

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Glucose-lowering activity of polyphenol rich aqueous extracts of *Coccinia grandis* (L.) Voigt (Family: Cucurbitaceae) has been scientifically proven. However, it was reported that the *C. grandis* extract reduced oral absorption, bioavailability and stability. The application of nanotechnology to encapsulate *C. grandis* leaf extract into nanoformulations provides an attractive strategy to overcome the aforementioned limitations leading to the development of novel nutraceuticals against hyperglycaemia. The objective of the present study was to determine glucose-lowering activity of *C. grandis* leaf extract encapsulated nanoliposomes using Wistar rats with diabetes mellitus induced with high-fat diet/low-dose streptozotocin. A high-fat diet (60% calories from fat) followed by intraperitoneal injection of streptozotocin was used to induce type 2 diabetes mellitus in Wistar rats. The diabetic rats (fasting blood glucose ≥ 200 mg/dL) were administered with ethanol (70% v/v) extract of *C. grandis* (65, 190; human equivalent therapeutic dose, 570 mg/kg), encapsulated nanoliposomes (65, 185; equivalent therapeutic dose, 555 mg/kg), metformin (positive control, 300 mg/kg) and blank nanoliposomes. The glucose-lowering activity was assessed via total area under the oral glucose tolerance curve (TAUC). Ethical clearance was obtained from the Ethical Review Committee, University of Ruhuna [2020.P.004 (21.01.2020)]. A significant ($p < 0.05$) reduction of TAUC were observed in nanoliposome treated diabetic rats at the doses of 185 and 555 mg/kg. The percentage of glucose-lowering effect at the doses of 185 and 555 mg/kg was 28.2% and 34.8%, respectively with respect to the diabetic control ($p = 0.002$ and 0.000) and the glucose-lowering activity was increased by 7% ($p = 0.997$) and 7.5% ($p = 0.997$) in encapsulated nanoliposomes with respect to the crude extract of *C. grandis* at the therapeutic and high dose levels. The blank nanoliposomes did not show any glucose-lowering effect in diabetic rats. The results revealed that *C. grandis* encapsulated nanoliposomes possess increased dose-dependent glucose-lowering activity in Wistar rats with diabetes mellitus.

Acknowledgements: Financial assistance by AHEAD DOR-15

Key words: Glucose-lowering activity, *Coccinia grandis*, Nanoliposomes, Wistar rats

Evaluation of *in-vitro* anti-inflammatory activity of different extracts of *Carica papaya* leaves

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Carica papaya L. is a perennial large herbaceous plant and belongs to the family Caricaceae. The plant exhibits many medicinal properties such as anti-inflammatory, anti-cancer, anti-thrombolytic, anti-microbial and antioxidant. *In-vitro* anti-inflammatory activity of *C. papaya* leaves has not been evaluated using egg albumin denaturation assay in previous studies. Hence, this study was conducted to identify the activity of different extracts of *C. papaya* leaves against inflammatory conditions using *in-vitro* methods. Ethanol extract of *C. papaya* leaves was obtained by maceration and evaporation of the solvent. The aqueous extract was obtained from crushing fresh leaves (900g) with 300ml of distilled water. A part of the filtered aqueous extract was freeze-dried. The remaining part of the aqueous extract was subjected to sequential fractionation. The resulted hexane, dichloromethane, ethyl-acetate and residual aqueous fractions were evaporated using a rotary vacuum evaporator. The extracts were assessed for *in-vitro* anti-inflammatory activity using egg albumin denaturation assay using diclofenac sodium as the reference drug. All the tests were triplicated. The IC₅₀ values of the reference drug, diclofenac sodium, aqueous extract and ethanol extract of *C. papaya* leaves were 1228.0 mcg/ml, 643.7 mcg/ml, and 829.0 mcg/ml respectively. The IC₅₀ values of aqueous and ethanol extracts of *C. papaya* leaves were less than the IC₅₀ value of the reference drug. Among the fractions of the aqueous extract, only the fractions; ethyl acetate (IC₅₀ =126.5 mcg/ml) and residual aqueous fraction (IC₅₀ =1287.0 mcg/ml) showed *in-vitro* anti-inflammatory activity. No anti-inflammatory activity was observed with the hexane and dichloromethane fractions. In conclusion, the aqueous extract of *C. papaya* leaves possessed higher *in-vitro* anti-inflammatory activity than the ethanol extract. Moreover, the activity was retained in the ethyl acetate fraction of the aqueous extract of *C. papaya* leaves. Isolation and characterization of anti-inflammatory active compounds from ethyl acetate fraction of aqueous extract are recommended in future studies.

Keywords: Anti-inflammatory activity, *Carica papaya*, Egg albumin denaturation assay, Solvent extracts

Metabolome of *Eucalyptus* oil glands; Phytochemical screening of glandular extracts of *Eucalyptus pumila*, *E. gillenii* and *E. parvula*

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The species from the Genus *Eucalyptus* (Myrtaceae) synthesize defensive chemicals some of which are widely used as therapeutics. Eucalypts have been used in traditional medicines mainly for respiratory illnesses. The presence of sub-dermal glands rich in volatile terpene essential oils is characteristic of the trees of this genus. The aim of this study is to characterise the essential oil components and non-volatile phytochemicals localised to foliar oil glands of three *Eucalyptus* species; *Eucalyptus pumila*, *E. gillenii* and *E. parvula*. Non-volatile compounds (NVCs) were extracted from enzymatically isolated foliar glands and analysed using high-performance liquid chromatography (HPLC) and mass spectrometry (LC-MS). NVCs were identified based on the diagnostic mass spectral fragmentation patterns. Volatile oil components were analysed using gas chromatography with flame ionisation detection and mass spectrometry (GC-FID and GC-MS). Monoterpenes and sesquiterpenes were identified and quantified for each species. The gland isolation technique was successfully used to extract NVCs directly from oil glands. This protocol facilitates the extraction of compounds exclusively localised to glands. Several monoterpene acid glucose esters (MAGEs) were identified using the unique mass fragmentation pattern. Cuniloside B, cypellocarpin C and froggattiside A were present in glandular extracts of all three species. Several novel MAGEs were present in the extracts of *E. pumila* and *E. parvula*. Leaf extracts from all three species contained monoterpenes and sesquiterpenes. The most abundant oil component was 1,8-cineole. Percentage 1,8-cineole was highest in *E. pumila* (75.5 %). Limonene, α -pinene, *p*-cymene, β -pinene and α -terpineol were present in comparatively low levels. The results of the present study revealed that foliar glands of *Eucalyptus* species co-house NVCs and volatile oil components suggesting a possible biosynthetic/physiological relationship between the two groups. The extracts may be potential sources of new pharmaceutically important phytochemicals.

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Key word: Metabolome, Phytochemical screening, glandular extracts, *Eucalyptus*

In silico* identification and molecular docking analysis of long chain alkane monooxygenase (LadA) in filamentous fungus *Aspergillus flavus

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The filamentous fungus, *Aspergillus* sp. MM1 completely removes n-alkanes (C12-C31) in crude oil, indicating the presence of enzymes responsible for the initial oxidation of n-alkanes. Therefore, *in silico* identification and functional characterization of a LadA homolog was carried out in the closely related *A. flavus* (NRRL 3357). A similarity search using PSI-BLAST (NCBI) against *Geobacillus* LadA (PDB ID: 3B9O) revealed the presence of a putative LadA homolog in *A. flavus* (49.13% amino acid identity; 98% query cover). The 3D-structure of the LadA homolog was predicted using SWISS-MODEL server. A variety of validation tools confirmed the reliability of the predicted structure [VERIFY3D (average 3D-1D score ≥ 0.2 for 94.37 % of the residues), PROCHECK (> 90% residues located in most favoured regions) and ERRAT (84.55)]. Flavin mononucleotide (FMN) and alkane structures were retrieved from the Protein Data Bank / PubChem and geometrically optimized using the ORCA4.2.1 program. Docking simulations were performed by AutoDock Vina software. Structures of enzyme:cofactor:substrate complexes were validated by comparison of conformations and binding energies obtained between crystal structures of *Geobacillus* LadA and predicted *A. flavus* models. Binding energies obtained with hexadecane (C16) (-4.5 and -4.9 kcal/mol), eicosane (C20) (-5.2 and -4.9 kcal/mol), pentacosane (C25) (-5.6 and -5.1 kcal/mol), triacontane (C30) (-5.5 and -6.1 kcal/mol) and hexatriacontane (C36) (-5.8 and -4.5 kcal/mol) indicated reliable docking confirmations with sixteen shared active site residues. This study reports the presence of a potential LadA in *A. flavus*. This finding can support future biotechnological applications in bioremediation of petroleum hydrocarbon pollution.

Acknowledgments: Financial assistance grant AP/3/2/2014/RG/12 University of Colombo.

Key words: *In silico*, Molecular docking, Monooxygenase, *Aspergillus flavus*

Assessment of acute toxicity of *Averrhoa carambola* (starfruit) juice in Wistar rats

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Averrhoa carambola L. (starfruit) is a commonly consumed fruit with a high nutritive value. It is popular as a fruit juice. However, it has been reported that the fruit contains the neurotoxin, caramboxin and a large concentration of oxalic acid, which may exert harmful effects on kidneys. The present study was carried out to assess the possible acute toxicity effects of the high doses of starfruit juice in Wistar rats. Ethical clearance was obtained from the Ethics Review committee of University of Sri Jayewardenepura. Twelve male Wistar rats were divided randomly into two groups (n=6) and each rat in the test group was fed with a single high dose of 35 ml/kg of body weight according to the calculations done using standard dose conversion guidelines. Following overnight fasting, the control group and the test group was fed with 7ml of distilled water and star fruit juice respectively, which were orally administered as four discrete doses. After 24 h, urine and blood were collected and subjected to serum biochemical, haematological and urine analysis. Red blood cell, total white blood cell, monocyte, granulocyte, lymphocyte and platelet counts and values of mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration and red cell distribution width showed no significant ($p>0.05$) difference between the two groups. A significant reduction in the haemoglobin level ($p=0.022$) and Haematocrit ($p=0.019$) of the test group was observed when compared to the control group. There was no significant difference ($p>0.05$) in fasting blood glucose level and serum creatinine levels between the groups. While serum urea level showed a significance difference ($p=0.037$), the urine output, urine glucose, protein, pH, specific gravity and creatinine clearance showed no significant changes compared to the control group. Serum AST levels ($p=0.001$) showed a significant difference while ALT level did not show any significant difference ($p>0.05$) compared to the control group. The results suggest that the consumption of starfruits in high doses may lead to development of an acute anaemic condition and a degree of acute renal impairment.

Key words: Acute toxicity, *Averrhoa carambola*, Starfruit juice, Wistar rats

***In silico* identification of a bacterial AlmA-like protein in *Aspergillus flavus* NRRL 3357**

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Alkane degradation is an integral part of the bioremediation process of crude oil spills. AlmA is a bacterial flavin-binding monooxygenase which has been shown to have the capacity for the oxidation of long chain alkanes (>C32). Degradation of crude oil by *Aspergillus flavus* has been shown previously, but there are limited studies on the responsible enzymes. A verified AlmA protein sequence from *Acinetobacter* sp. was identified from the UniProtKB database. This was used to conduct a Position-Specific Iterative BLAST (PSI-BLAST) search of the NCBI non-redundant protein database. Among the 20,000 sequences retrieved in the first iteration, eight sequences of *A. flavus* NRRL3357 were identified. A domain search of the eight sequences using Pfam database revealed six of them carried the FMO-like domain, which was also present in the bacterial AlmA. The *A. flavus* sequence with the highest similarity, and also having the FMO-like domain was selected for further analysis. Three-dimensional models of the *Acinetobacter* AlmA (5 models) and the selected *A. flavus* sequence (4 models) were predicted using I-TASSER. The 3D model of the bacterial AlmA having the higher c-score was superimposed with all four models predicted for the *A. flavus* sequence, using UCSF Chimera. Superimposition with model 1 gave the lowest RMSD value (0.547 Å). The validity of model 1, was further evaluated using ERRAT (87.7895,) VERIFY3D (83.02 %) and PROCHECK (92.1% residues in most favourable and additionally allowed regions). The validation scores obtained demonstrate that the model 1 is reliable. Together, these results indicate that the selected *A. flavus* sequence represents an AlmA-like monooxygenase, suggesting that AlmA-like enzymes present in *A. flavus* may play a role in degradation of long chain alkanes.

Key Words: *In silico*, Bacteria, AlmA-like protein, *Aspergillus flavus*

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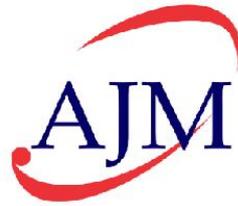
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